From Prohibition to Prescription: The Role of Cannabinoids in Sleep

Anastasia Suraev

A thesis submitted to fulfil requirements for the degree of Doctor of Philosophy

School of Psychology
Faculty of Science
University of Sydney

November 2022

Table of Contents

Acknowledgements

I would first like to thank my supervisor and mentor, Dr Camilla Hoyos. I am grateful for your ongoing support and guidance, your words of encouragement, and for teaching me invaluable skills that I continue to use each day!

To Prof Iain McGregor, it would be impossible to count all the ways you have helped me in my career. It has been a real privilege working alongside you all these years. Your intellectual curiosity, creativity, and quick wit makes work an absolute joy. You inspire me to think big and to make the most of the opportunities that come to me, and for that I thank you.

A big thank you to Prof Ron Grunstein for your continuous support and direction. I look forward to working with you on many more projects. Your sense of humour is always appreciated! To A/Prof Nathaniel Marshall, thank you for being a great mentor and teaching me everything I need to know about good quality clinical trial design and statistics.

To the Sleep and Chronobiology Team at the Woolcock Institute, thank you for giving me a home these past few years and for your support and encouragement in making the 'CANSLEEP' trial come to life. Thank you to Ben Zhang, Dr Carla Evans, Gosia Bronisz, Kyle Kremerskothen, Fraser Lowrie, and Garry Cho for all your support with setting up and trouble-shooting the high-density EEG sleep studies. To all the study doctors (Dr Tom Altree, Dr Keith Wong, Dr Brendon Yee, and Dr Sheila Sivam), thank you for all your assistance with screening and monitoring the participants. A big thanks to Dr Aaron Lam for waking up early and helping me complete those early morning treatment sessions! I also wish to thank Isabella Valenzuela for assisting me in completing the lengthy post-processing of the high-density EEG data.

To the Lambert Initiative team at the University of Sydney, you are a dream team! It has been a privilege to work alongside you and I can't wait to see what we achieve in 2023 and beyond. Thank you to Dr Danielle McCartney for being an incredible source of knowledge and guidance, and to Dr Richard Kevin and Dr Rebecca Gordon for their analytical chemistry assistance.

I would also like to Barry & Joy Lambert for their generous support without whom this research would not be possible. You have changed the landscape of medicinal cannabis research in Australia, and we are honoured to be on this journey with you to advance cannabinoid science.

A very special thank you goes out to my family for their continued support and encouragement. You have taught me to be resilient, to stay curious, and be adaptable to change, and I believe these have played a big part in getting me here today. To my amazing partner, Jake, thank you for your love and support, and for keeping me calm over these past few years. I can't wait to sit on a beach and relax with you in few weeks' time.

Statement of Originality

This is to certify that, to the best of my knowledge, the intellectual content of this thesis is the product of my own work and that all the assistance received in preparing this thesis and sources have been acknowledged.

I hereby declare that I have not submitted this material, either in full or in part, for a degree at this or any other institution.

Signature:

Name: Anastasia Suraev

Authorship Attribution Statement

I, Anastasia Suraev, am the primary author of the work featured in Chapters 2, 3, and 4.1-4.3.

Chapter 2 was published in *Sleep Medicine Reviews*.

Chapter 3 is under review in *Nature and Science of Sleep*.

Chapter 4.1 was published in *BMJ Open*

Chapter 4.2 is ready for submission to *Nature Communications*.

Chapter 4.3 is ready for submission to *Neuropsychopharmacology*.

For the work feature in Chapter 3, I took the lead role in the data analysis and interpretation and the writing and appraisal of the manuscript. For the work in featured in Chapters 2 and 4.1–4.3, I took the lead role in the conception and design of the research, conducting the research, data analysis and interpretation, and the writing and appraisal of manuscripts.

In addition to the statements above, in cases where I am not the corresponding author of a published item, permission to include the published material has been granted by the corresponding author.

Anastasia Suraev

Signature:

Date: 25-Nov-2022

As supervisor for the candidature upon which this thesis is based, I can confirm that the authorship attribution statements above are correct.

Dr Camilla M. Hoyos

Signature:

Date: 25-Nov-2022

Supervisor Attestation

I, Dr Camilla Hoyos, certify that Anastasia Suraev's thesis:

- is sufficiently well presented to be examined and,

- does not exceed the prescribed word limit for which approval has been granted.

Name: Dr Camilla Hoyos

Signature:

Date: 25-Nov-2022

Publications and Presentations

Publications

Suraev A., Grunstein R.R., Marshall N.S., D'Rozario A.L., Gordon C., Bartlett D., Wong K., Yee B.J., Vandrey R., Irwin C., Arnold J.C., McGregor I.S., and Hoyos C.M. (2020) Cannabidiol (CBD) and Δ^9-tetrahydrocannabinol (THC) for chronic insomnia disorder ('CANSLEEP' trial): protocol for a randomised, placebo-controlled, double-blinded, proof of concept trial. *BMJ Open*, *10*(5), e034421. http://dx.doi.org/10.1136/bmjopen-2019-034421

Suraev A., Marshall N.S., Vandrey R., McCartney D., Benson M., McGregor I.S., Grunstein R.R., and Hoyos C.M. (2020) Cannabinoid therapies in the management of sleep disorders: a systematic review of preclinical and clinical studies. *Sleep Medicine Reviews*, 53, 101339 https://doi.org/10.1016/j.smrv.2020.101339

Suraev A., Mills L., Abelev S.V., Arkell T.R., Lintzeris N., and McGregor I.S. (2021). Cannabis use patterns for sleep disorders in Australia: A subanalysis of an online 2020-2021 cross-sectional survey. *Revision submitted to Nature and Science of Sleep on 21-Nov-2022.*

Suraev A., McGregor I.S., Marshall N.S., Kao T., D'Rozario A.L., Grunstein R.R., Irwin C., Gordon C., Bartlett D., Wong K., Yee B.J., Vandrey R., Arnold J.C., and Hoyos C.M. (2023) Acute effects of combined cannabidiol (CBD) and δ9-tetrahydrocannabinol (THC) on sleep in insomnia disorder: a randomised, placebo-controlled, trial using high-density EEG. *In preparation for submission to Nature Communications.*

Suraev A., McCartney D., McGregor I.S., Grunstein R.R., Marshall N.S., D'Rozario A.L., Gordon C., Bartlett D., Wong K., Yee B.J., Vandrey R., Irwin C., Arnold J.C., and Hoyos C.M. (2023) The 'morning after' effects of combined cannabidiol (CBD) and δ9-tetrahydrocannabinol (THC) in insomnia disorder: a randomised, placebo-controlled, trial. *In preparation for submission to Neuropsychopharmacology.*

Supporting Publications

The following publications are derived from research projects which extend on the scope of work presented in the current thesis:

I. McCartney D., **Suraev A.,** McGregor I.S., (2022). The 'next-day' effects of cannabis use: A systematic review. *Cannabis and Cannabinoid Research.* Accepted 22-Oct-2022.

II. McCartney D., **Suraev A.,** et al (2022). Effects of cannabidiol (CBD) on simulated driving and cognitive performance: a dose-ranging randomised controlled trial. *Journal of Psychopharmacology.* https://doi.org/10.1177/02698811221095356

III. Arnold J., McCartney D., **Suraev A.,** and McGregor I.S. (2022) The safety and efficacy of low oral doses of cannabidiol: an evaluation of the evidence. *Clinical and Translational Science.* Accepted 17-Sep-2022. https://doi.org/10.1111/cts.13425

IV. Lavender I., McGregor I.S., **Suraev A.,** Grunstein R.R. and Hoyos C.M. (2022) Cannabinoids, insomnia, and other sleep disorders. *CHEST.* https://doi.org/10.1016/j.chest.2022.04.151

V. Lintzeris N., Mills L., Abelev S., **Suraev A.,** Arnold J.C., McGregor I.S. (2022) Medical cannabis use in Australia: consumer experiences from the online Cannabis as Medicine Survey 2020 (CAMS-20). *Harm Reduction Journal.* 19, 88. https://doi.org/10.1186/s12954-022-00666-w

VI. McCartney, D., Kevin, R. C., **Suraev, A.,** Irwin, C., Grunstein, R. R., Hoyos, C. M., & McGregor, I. S. (2021). Orally administered cannabidiol (CBD) does not produce false-positive tests for THC on the Securetec DrugWipe® 5S or Dräger Drug Test® 5000. *Drug Testing and Analysis.* doi: 10.1002/dta.3153

VII. McCartney, D., Benson, M. J., **Suraev, A.,** Irwin, C., Arkell, T. R., Grunstein, R. R., ... & McGregor, I. S. (2020) The effect of cannabidiol (CBD) on simulated car driving performance: A randomised, double-blind, placebo-controlled, crossover, dose-ranging clinical trial protocol. *Human Psychopharmacology: Clinical and Experimental.* 35 (5). https://doi.org/10.1002/hup.2749

VIII. Arkell, T. R., Lintzeris, N., Mills, L., **Suraev, A.,** Arnold, J. C., & McGregor, I. S. (2020). Driving-related behaviours, attitudes, and perceptions among Australian medical cannabis users: results from the CAMS 18-19 survey. *Accident Analysis & Prevention*, *148*, 105784. https://doi.org/10.1016/j.aap.2020.105784

Conference Abstracts

Suraev A., Grunstein R.R., McGregor I.S., et al (2022) (November 2022). Poster Presentation. Effects of Cannabidiol (CBD) And Δ9-Tetrahydrocannabinol (THC) on Sleep and Daytime Function in Insomnia Disorder: A Randomised, Double-Blinded, Placebo-Controlled, Trial using High-Density Electroencephalography (HdEEG). *Brain and Mind Centre Symposium 2022*, Sydney, Australia.

Suraev A., Grunstein R.R., McGregor I.S., et al (2022) Oral Presentation. Effects of Cannabidiol (CBD) And Δ9-Tetrahydrocannabinol (THC) on Sleep and Daytime Function in Insomnia Disorder: A Randomised, Double-Blinded, Placebo-Controlled, Trial using High-Density Electroencephalography (HdEEG). *Sydney Postgraduate Psychology Conference 2022*, Sydney, Australia.

Suraev A., Grunstein R.R., McGregor I.S., et al (2022) New Investigator Oral Presentation. Effects of Cannabidiol (CBD) And Δ9-Tetrahydrocannabinol (THC) on Sleep and

Daytime Function in Insomnia Disorder: A Randomised, Double-Blinded, Placebo-Controlled, Trial using High-Density Electroencephalography (HdEEG). *Australasian Sleep Association, Sleep DownUnder Conference,* Brisbane, Australia.

Suraev A., Grunstein R.R., McGregor I.S., et al (2022) Oral Presentation. Effects of Cannabidiol (CBD) And Δ9-Tetrahydrocannabinol (THC) on Sleep and Daytime Function in Insomnia Disorder: A Randomised, Double-Blinded, Placebo-Controlled, Proof-Of-Concept Trial. *International Cannabinoid Research Society 32nd Annual Conference,* Galway, Ireland.

Suraev A., Mills L., Abelev S.V., et al (December 2021). Oral Presentation. Medicinal cannabis use patterns for sleep disorders in the Australian community: results from the CAMS20-21 Survey. *Sydney Postgraduate Psychology Conference 2021*, Sydney, Australia.

Suraev A., Mills L., Abelev S.V., et al (October 2021), Oral Presentation. "Medicinal cannabis use patterns for sleep disorders in the Australian community: results from the CAMS20-21 Survey" at the *Woolcock Institute Postgraduate Student Symposium 2021*.

Suraev A., Mills L., Abelev S.V., et al (June 2020). Oral Presentation. Cannabis use patterns for sleep disorders in Australia: a subanalysis of the Cannabis as Medicine (CAMS) 2018 survey. *International Cannabinoid Research Society 32nd Annual Conference,* Galway, Ireland. (*Abstract peer-reviewed and accepted but conference was cancelled due to COVID-19*).

Suraev A., Hoyos, C., Mills, L., McGregor, I., Bravo, M., Arkell, T., M. Benson & Lintzeris, N. (October 2019). Oral Presentation. Medicinal use of cannabis for sleep disorders in Australia. *Australasian Sleep Association, Sleep DownUnder Conference,* Sydney, Australia.

Suraev A., (October 2019). Poster Presentation. Good-quality restful sleep: what does cannabis have to do with it? *Brain and Mind Symposium 2020*, Sydney, Australia.

Invited Presentations

I. Invited speaker (August 2022), "Oral Presentation. Effects of Cannabidiol (CBD) And Δ9-Tetrahydrocannabinol (THC) on Sleep and Daytime Function in Insomnia Disorder: A Randomised, Double-Blinded, Placebo-Controlled, Trial using High-Density Electroencephalography (HdEEG)" at the *CogSleep Symposium*, Sydney, Australia

II. Oral presentation (June 2021), "Cannabinoids, sleep and daytime function" at the *ECR Think Tank Symposium*, Sydney, Australia.

III. Invited speaker (November 2019), "Sleep and ageing: What do cannabinoids have to do with it?" *ForeFront 11th Scientific Meeting* at the Refectory, University of Sydney, Australia.

Media Engagement

I. **The New York Times** | **"**Does CBD Help with Insomnia?" 31 August 2022, Interviewed by NYT journalist Rachel Peachman [https://www.nytimes.com/2022/08/30/well/live/does-cbd-help-with-insomnia.html]

II. **Cannabiz** | "Meet the women blazing a trail in cannabis research" 16 December 2021, Interviewed by freelance writer Hannah Adler [https://www.cannabiz.com.au/meet-the-women-blazing-a-trail-in-cannabis-research/]

III. **SleepJunkies Blog** | "Cannabis sleep aids are flooding the market, but what does the science say?" 28 October 2020, Interviewed by freelance writer Eren Heger: [https://sleepjunkies.com/cannabis-sleep-aids-are-flooding-the-market-but-what-does-the-science-say/]

IV. **SleepJunkies Podcast** | "Cannabis and Sleep: From Prehistory to the 21st Century" 23 June 2020, Interviewed by Sleep Junkies Founder Jeff Mann [https://sleepjunkies.com/podcast/]

V. **Channel 9 News** | "Researchers are testing a cannabis-based medicine as a more 'natural' remedy to help fight insomnia" 13 March 2020 [https://www.youtube.com/watch?v=SpHEEeKfbaE]

Awards and Scholarships

2022	Australasian Sleep Association (ASA) New Investigator Award 2022
2022	ASA Student Travel Grant for Sleep DownUnder Conference in Brisbane, Australia
2022	International Cannabinoid Research Society (ICRS) Ethan Russo Trainee Award
2022	Best Predoctoral Oral Presentation at the 32nd Annual ICRS Conference in Galway, Ireland
2021	Best Presentation (2nd Prize) at the Sydney Postgraduate Psychology Conference
2021	Sleep Health Foundation Emerging Sleep Hero Award
2021	Winner of the University of Sydney's 'Visual Your Thesis' Competition (watch here)
2020	School of Psychology Publication Prize
2018-2021	Vice Chancellor's Research Scholarship (VCRS) ($35,000 top-up over 3.5 years)
2018-2021	Research Training Program (RTP) scholarship ($28,000 for 3.5 years)

List of Figures

List of Tables

List of Abbreviations

AHI	apnea-hypopnea index
CB_1	cannabinoid receptor type 1
CB_2	cannabinoid receptor type 2
CBD	cannabidiol
CBD/THC	investigational product containing 200 mg CBD and 10 mg THC
DT5000	DrugTest 5000
DW5s	DrugWipe 5s
EEG	electroencephalography
FTT	Finger Tapping Task
ISI	Insomnia Severity Index
KSS	Karolinksa Sleepiness Scale
LC-MS/MS	liquid chromatography with tandem mass spectrometry
MWT	Maintenance of Wakefulness Test
NREM	non-rapid eye movement sleep
POCT	point-of-collection testing
PSG	polysomnography
PSQI	Pittsburgh Sleep Quality Index
PVT	Psychomotor Vigilance Task
RCT	randomised controlled trial
REM	rapid eye movement sleep
SOL	sleep onset latency
SWS	slow wave sleep
THC	Δ^9-tetrahydrocannabinol
THC-COOH	11-nor-9-carboxy-Δ^9-tetrahydrocannabinol
TGA	Therapeutic Goods Administration
TST	total sleep time
WASO	wake after sleep onset

Thesis Abstract

Insomnia disorder is the most common sleep disorder and is characterised by self-reported difficulties with falling asleep and/or staying asleep and is associated with significant daytime distress. Despite significant advances in the understanding and treatment of insomnia and the availability of effective treatment options, insomnia management remains suboptimal, posing a significant challenge to public health. Emerging research also indicates that insomnia is not a benign condition given its association with a range of negative health outcomes and risks, highlighting a strong need for novel treatment options. Anecdotally, cannabinoids such as cannabidiol (CBD) and Δ9-tetrahydrocannabinol (THC) are being touted as sleep-promoting drugs. However, evidence for the therapeutic utility of cannabinoids in the treatment of sleep disorders is limited, thus making it challenging for clinicians to make evidence-based decisions regarding efficacy and safety. The extant literature on cannabinoids and sleep is complicated by a lack of objective measures of sleep, and other factors that are likely to confound effects on sleep outcomes such as administering cannabis extracts of unknown cannabinoid concentrations and recruiting participants with a history of chronic and/or heavy (non-medicinal) cannabis use, and/or non-clinical insomnia populations. No study to-date has explored the effects of cannabinoids on *next day* function including cognitive function and driving performance. This precludes any definitive conclusions regarding the safety and efficacy of cannabinoids on sleep in clinician-diagnosed insomnia disorder.

With increasing consumer interest and uptake of medicinal cannabis globally for the treatment of sleep disorders, it is important that we develop a better understanding of how cannabinoids affect sleep and 'next day' function before it becomes routine clinical practice. This thesis centers arounds a series of investigations designed to address gaps in our understanding and build on existing knowledge of the effects of cannabis on sleep. It aims to (1) examine the acute effects of a commonly used ratio of CBD and THC on objective sleep outcomes using

polysomnography with high-density EEG in insomnia disorder, and (2) determine the 'next day' effects of evening administration of CBD and THC on cognitive function, alertness, and driving performance. It also explores the behaviours and patterns of medicinal cannabis use among a sample of Australians with a self-reported sleep disorder and examines the utility of point-of-collection testing (POCT) in detecting individuals who may be under the influence of cannabis.

Chapter 1 begins with a brief introduction to insomnia disorder, including its pathophysiology and management, and cannabinoids, including cannabinoid pharmacology and pharmacokinetics and the complex interplay between cannabinoids and the sleep-wake cycle. It then goes on to review the literature around the effects of cannabis on sleep architecture with a specific focus on several key sleep parameters. The chapter subsequently provides a comprehensive review of studies examining the 'next day' (i.e., >8 hour) residual effects of cannabis or THC use on cognitive function and safety-sensitive tasks.

Chapter 2 (published in *Sleep Medicine Reviews*, 2020) presents the results of the first systematic review to synthesise all published clinical and preclinical studies that administered a cannabinoid in an attempt to treat an underlying sleep disorder. The review identified limited evidence to support the clinical use of cannabinoids for the treatment of any sleep disorder with a moderate-to-high risk of bias identified within the majority of studies published to-date. Future research directives were identified and included (1) the use of validated objective and subjective measures of sleep-related outcomes to assess therapeutic efficacy of cannabinoids, (2) the use of robustly designed randomised, controlled trial designs with an adequate comparator (e.g., placebo), and (3) exploring the effects of THC that confers clinical efficacy without causing 'next day' impairment (e.g., daytime drowsiness). The recent publication of two pivotal clinicals trials (one in insomnia disorder and one in REM sleep behaviour disorder) are described in an addendum in Section 2.1.

Chapter 3 (submitted to *Nature and Science of Sleep*; 2022) presents the results of a subanalysis of Australian consumers who self-reported using medicinal cannabis, prescribed and/or illicit, to

treat a sleep disorder recruited as part of the larger 'Cannabis as Medicine 2020-2021 Survey' (CAMS20-21) (n=1,600). Of the 1600 respondents who completed the survey, the majority (64.4%) self-reported using medicinal cannabis to treat a sleep disorder, but only 16.8% of respondents chose a sleep disorder as their main condition. This suggests that sleep disorders were commonly being treated *secondary* to a primary medicinal condition such as chronic pain or a mental health disorder. We also identified that use of inhaled methods (i.e., smoking or vaping), THC-dominant products, and illicit sources of medicinal cannabis were common among people with sleep disorders; an important finding that can help to guide future policy and research in this area.

Chapter 4.1 (published in *BMJ Open*; 2020) shares the clinical trial protocol of our randomised, placebo-controlled, crossover trial examining cannabinoids on sleep and daytime function in insomnia disorder using high-density EEG. In complement to the trial registration, the aim of this publication was to facilitate critical appraisal of the clinical trial design and to encourage transparency in the reporting of outcomes via public access to pre-specified study methods. This will hopefully allow for adequate assessment of the risk of bias as well as ensure clarity around the role of the sponsor, funding body, and the supplier of the investigational product in the trial design, conduct, and reporting.

Chapter 4.2 describes a randomised, double-blind, placebo-controlled, crossover trial that was designed to assess the acute effects of an oral formulation containing a 20:1 ratio of CBD to THC ('CBD/THC') on objective sleep outcomes using in-laboratory polysomnography with high-density EEG in chronic insomnia disorder. Contrary to expectations, a single dose of CBD/THC significantly reduced total sleep time (TST) and time spent in REM sleep while increasing latency to REM sleep with no change in subjective sleep outcomes. High-density EEG analysis revealed paradoxical effects with decreased fast activity during N2 sleep suggesting deeper sleep with decreased delta activity during N3 sleep indicating reduced sleep depth. Increased fast activity during REM sleep is also consistent with heightened arousal. This study shows, for the first time,

an acute REM suppressing effect and sleep-reducing effect of cannabinoids in a clinical insomnia population.

Chapter 4.3 follows on from the previous chapter and describes the acute effects of evening administration of CBD/THC on 'next day' function. Given the current legal framework for driving under the influence of cannabis in Australia (i.e., detection of THC in saliva with no functional assessment), we also examined the accuracy and reliability of two commonly used POCT devices (Securetec DrugWipe 5s and Dräger Drug Test 5000) in detecting THC in oral fluid the morning after evening administration. Apart from a possible (subtle) increase in subjective measures of sleepiness, no reliable changes in 'next day' function including cognitive function, objective measures of alertness, and driving performance were observed. Accuracy on the POCT devices was lowest at 0.5 h post-drug administration yielding the highest number of false positive and false negative tests with improved performance the next day. Overall, it appears that a single, oral dose of combined 200 mg CBD and 10 mg THC does not substantially impair 'next day' function in individuals with insomnia disorder.

Chapter 5 provides a general discussion of the work described in this thesis and considers the real-world implications and relevance of these findings. Several remaining knowledge gaps and priorities for future research are discussed. These include: (1) identifying whether repeated dosing and gradual up-titration from a lower dose of THC (i.e., <10 mg starting dose; alone or in combination with CBD) will improve clinical efficacy without the propensity to suppress REM sleep; (2) explore whether the lack of 'next day' impairment following acute evening administration of CBD/THC remains with repeated dosing; and (3) achieving a better understanding of the clinical significance of the observed changes on high-density EEG, particularly the dynamic changes over the entire night. It is hoped that the work contained in this thesis will advance our understanding of how cannabis impacts sleep and help to guide future research directives and clinical decision making.

1. General Introduction and Literature Review

1.1 Prologue

Cannabis sativa has been long cultivated for fibre, food, and medicine, as well as its sedating and relaxing properties since ancient times. The medicinal properties of cannabis were first described in ancient Indian *Ayurveda* tradition[1] and introduced to Western medicine in the 19th Century.[2] Use of cannabis for medicinal purposes flourished throughout the 19th Century and through the first decades of the 20th Century. However, in response to sociocultural and political pressure, the use of cannabis were removed from the United States Pharmacopeia in 1941 and outlawed in the United Nations Single Convention on Narcotic Drugs in 1961.

Decades later, shifting social attitudes and more lenient cannabis laws have seen the status of cannabis undergo a rapid global change. Uruguay became the first country to legalise cannabis in 2013, while in Australia, cannabis was legalised for medicinal purposes in November 2016. At the time of writing, more than 300,000 approvals for medicinal cannabis products have been issued to Australian patients.[3] In December 2020, the UN Commission on Narcotic Drugs reclassified cannabis to recognise its therapeutic uses, from Schedule IV (the most restrictive category e.g., fentanyl) to Schedule I (the least-controlled schedule) in response to evidence reviews and associated propositions for easing restrictions issued by the World Health Organisation in 2019.

Sleep disorders are one of the most common reasons people report using illicit and licit medicinal cannabis, after pain and mental health.[4-6] However, despite the increasing use of cannabis to treat sleep disorders, the clinical evidence to support the use of cannabis and its constituents in the management of sleep disorders is limited. As this introductory chapter will highlight, this is an important issue with several critical knowledge gaps. Specifically, there is considerable uncertainty about how cannabis affects the brain during sleep and the magnitude of possible residual 'next day' effects on daytime function. Addressing this knowledge gap is a crucial first step in developing evidence-based guidelines to inform health professionals of the appropriate prescription, dosing, and safety of using cannabinoid-based therapies for sleep disorders.

1.2 Sleep Health

Sleep is a fundamental physiological process that plays a vital role in restorative functions that are essential for normal daytime function[7]. It is increasingly acknowledged as a 'vital sign';[8] a signal of one's general physical and mental health. Taking up around one third of a human lifetime, sleep is hypothesised to provide a dedicated time window for neuronal plasticity, regulate brain chemicals and remove toxic by-products, and allow for adaptive processing of emotional memories.[9] Healthy adults need between 7 – 9 hours of sleep per night, while babies, young children and teens need even more to enable their growth and development. Optimal sleep health involves multiple factors, including adequate duration, timing, efficiency, and a sense of having restorative sleep that leaves the individual feeling alert and functional throughout the day.[10] The *National Sleep Foundation's* consensus regarding indicators of good sleep quality included sleep continuity (i.e., uninterrupted sleep), shorter sleep latencies, and fewer awakenings.[11]

Insomnia symptoms are reported in approximately 30-35% of the general population at any given time,[12] which may be partly due to some sort of interruption in a sleep schedule (e.g., jet lag, acute medical illness, shift work) or a stressful life event (e.g., job loss, relationship or family problems). However, a subset will display persistent insomnia symptoms which can often present independently or comorbidly with another medical or psychiatric disorder.[13] Insomnia disorder is the most common sleep disorder that is characterised by subjective complaints of poor sleep and daytime symptoms such as fatigue which, if left untreated, can increase the risk of developing depression and cardiovascular disease; highlighting a strong need for clinical intervention.[13]

1.3 Insomnia

1.3.1 Clinical Definition and Prevalence

Insomnia disorder is a highly prevalent, complex and heterogeneous disorder characterised by chronic dissatisfaction with sleep occurring at least three nights per week for at least three

months, and is associated with difficulties in falling asleep, maintaining sleep or early-morning awakening with inability to return to sleep.[14] These sleep difficulties must be associated with clinically significant distress or impairment, and cannot be explained by inadequate opportunity or cirumstance for sleep. It is often the daytime impairment that predominately drives treatment-seeking behaviour.[15] The *International Classification of Sleep Disorders Version 3 (*ICSD-3) categorises insomnia disorder into three categories: 'chronic insomnia disorder', 'short-term insomnia disorder' (e.g. a short-term stressor such as work or marital stress affecting sleep) and 'other insomnia disorder'.[16] The *Diagnostic and Statistical Manual of Mental Disorders Fifth Edition* (DSM-5), on the other hand, advocates for a unifying concept of insomnia disorder with the pathway of causality no longer the core diagnostic focus.[14] Both the ICSD-3 and DSM-5 no longer use the terms 'primary' and 'secondary' or 'comorbid' insomnia because they do not improve diagnostic accuracy or assist with treatment options. The term 'comorbid insomnia' is misleading because it implies that adequate treatment of the primary condition (e.g., depression) will resolve the insomnia complaints which is not always the case.[17]

In terms of prevalence, insomnia disorder is the most common sleep disorder and the second most common neuropsychiatric disorder. The worldwide prevalence of insomnia symptoms is approximately 30 – 35% with different countries yielding similar prevalence estimates.[18 19] Depending on the diagnostic criteria applied, prevalence rates for insomnia disorder range from 4% to 22% using the DSM-4 criteria.[20] The course of insomnia disorder is often persistent, with one longitudinal study showing that 74% of individuals reported insomnia for at least 1-year and 46% reported insomnia persisting over the 3-year follow-up period.[21] Insomnia generally does not resolve spontaneously, with a remission rate of 54%; however, half of those (27%) with remission of insomnia eventually relapsed at subsequent follow-up.[21]

A diagnosis of insomnia disorder is strictly based on *subjective* sleep complaints and not objective sleep outcomes such as in-laboratory polysomnography (PSG). PSG is a multi-parameter sleep study that measures electroencephalography (EEG; brain activity), electromyography (EMG;

muscle activity), electrooculography (EOG; eye movements), and blood oxygen levels overnight, and is the gold standard for measuring sleep. PSGs are not used in the routine evaluation of insomnia for several reasons. First, insomnia symptoms wax and wane making it difficult for the sleep complaint to be fully captured in a single night PSG. Second, insomnia diagnosis is often complicated by sleep discrepancy (i.e., the difference between subjective and objective sleep parameters).[22] This often presents as an underestimation of total sleep time and an overestimation of sleep onset latency and number of awakenings known as 'paradoxical insomnia' (i.e., patients perceive and self-report inadequate sleep despite normal objective sleep parameters).[22] Although previously defined as a misperception of sleep as wakefulness, recent research suggests this may represent a lack of precision in detecting subtle EEG changes using traditional sleep analyses.[22] Therefore, the use of PSGs, within an insomnia context, are generally limited to cases who are refractory to standard treatments and where there is the need to rule out another underlying sleep disorder such as sleep apnea, periodic limb movement disorders, or unexplained daytime sleepiness.[23]

1.3.2 Pathophysiology of Insomnia

The pathophysiology of insomnia is complicated by its heterogeneity presenting as a primary disorder and as a condition co-existing with numerous medical and psychiatric disorders.[24] While there is still no universally accepted model, several models of insomnia aetiology have been proposed, most of which encompass both external stressors as well as internal genetic, physiological and psychological factors.[25]

The '3P' model describes predisposing, precipitating, and perpetuating factors relevant to the development and maintenance of insomnia symptomology.[26] *Predisposing* factors include genetics, personality traits (e.g., neuroticism) and stress-reactivity that increase a person's risk for insomnia symptoms. Genetic factors have shown to contribute to the regulation of sleep-wake traits (e.g., sleep duration and timing of sleep),[27] with family-based heritability estimates suggesting that insomnia has a substantial genetic component (38% in males and 59% in females).[28] Female

gender is a risk factor for insomnia as is advancing age.[21] *Precipitating* factors, on the other hand, involve a biopsychosocial trigger (e.g., job loss/stress, death of a loved one, end of a long-term relationship etc) that 'push' an individual over the threshold and into a clinically significant insomnia disorder. Maladaptive behaviours and/or thinking patterns that attempt to cope with or compensate for the stressor act as *perpetuating* factors. These include restructuring sleep-wake cycle (e.g., taking naps during the day, excessive caffeine intake, spending more time in bed or sleeping in) or engaging in negative thinking styles (e.g., rumination, catastrophising, all or nothing thinking).[29] Together, this leads to the dysregulation of sleep homeostasis.

A subpopulation of people will develop chronic insomnia without the presence of maladaptive behaviours or thinking patterns (i.e., perpetuating factors). The hyperarousal model of insomnia suggests that the main etiological factor in the onset and the maintenance of insomnia is cognitive, emotional, and physiological hyperarousal present at night and during the day.[30] Hyperarousal can be seen as sympathetic nervous system overactivation, with increased basal metabolic rate[31] and body temperature,[32] altered heart rate variability,[33] and elevated cortical activation on EEG[34]. While studies often fail to demonstrate consistent PSG-derived sleep changes that correspond to the subjective complaints of patients with insomnia, a meta-analysis has shown that patients with insomnia present with a disruption of sleep continuity (the amount and distribution of sleep versus wakefulness) and significant reduction in slow wave sleep (SWS) and REM sleep.[35] Evidence suggests that patients with insomnia exhibit an abnormal amount of beta EEG activity (14-35 Hz range; associated with attention, perception, and cognition in humans) during NREM sleep relative to good sleepers.[36] This is in line with another study showing that patients with insomnia had more high-frequency EEG activity during all-night NREM compared to good sleepers.[37] This is consistent with psychological studies suggesting that patients with insomnia are hypervigilant and/or prone to excessive rumination at sleep onset and during sleep.[38]

More recently, high-density EEG, a novel technology that combines the superior temporal resolution of EEG recordings with high temporal resolution, has been utilised to explore the

cortical sources of EEG activity observed at the scalp level.[39] A study using 256 channel high-density EEG showed that those with insomnia had more high-frequency EEG activity compared to good sleeping controls across the sensory and sensorimotor brain regions during NREM sleep.[40] This suggests that, even during deep sleep, parts of the brain are still relatively 'awake' (or at least not totally asleep) in patients with insomnia. This lends support to the model of insomnia as a disorder of sleep-wake regulation characterised by simultaneous sleep and wake-like activation patterns in specific brain regions.[41] This concept suggests that arousal in insomnia need not be viewed as a global construct but may be viewed a dysfunction in specific neural circuits.

The pathophysiology of insomnia is complex and multi-faceted and is not within the scope of the current thesis to adequately cover. **Figure 1**, adapted from [25], highlights a plausible model in which insomnia is most likely to develop.

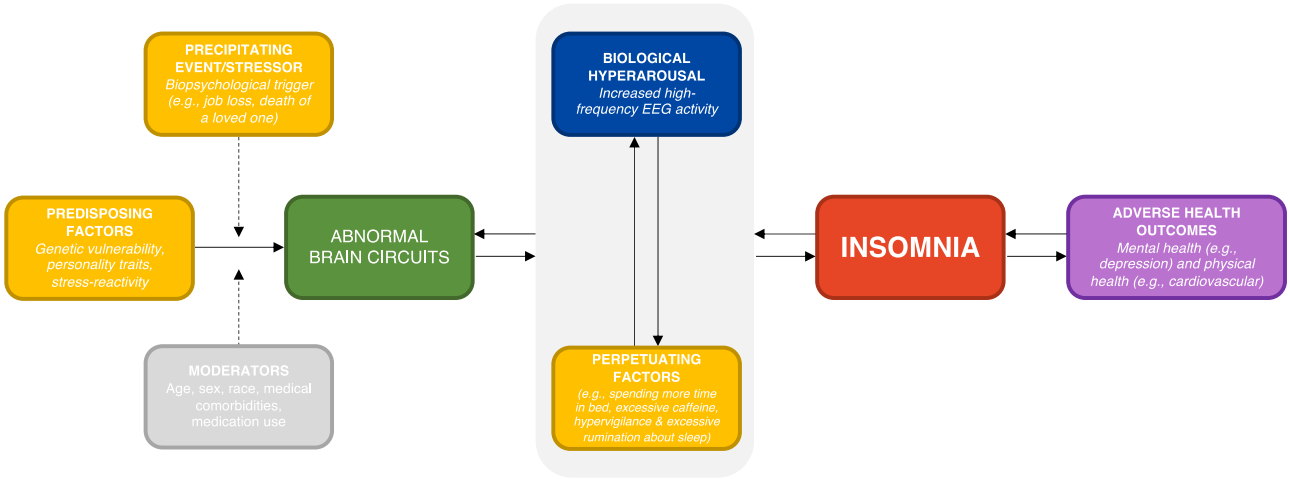

Figure 1 Model of the pathophysiology of insomnia adapted from Levenson et al., 2015.[25]

1.3.3 Consequences of Insomnia

Poor sleep is often associated with clinically significant daytime impairments in social life, educational attainment and/or occupational function. This is associated with higher incidence of absenteeism, poorer workplace productivity, and motor vehicle/workplace accidents.[42] Recent health economic analyses suggest that more than 90% of insomnia-related costs result from these occupational consequences.[43] Insomnia is also linked to higher healthcare utilisation and costs,

particularly in those with co-existing medical or psychiatric disorders.[44] Annual costs arising from chronic insomnia disorder are estimated at approximately $30 - $107 billion in the USA.[45]

The most concerning consequences of poor sleep are on the body's key regulatory systems, acting as a multi-system biological risk.[46] Although insomnia may occur as a primary condition, it is more commonly associated with at least one comorbid disorder with studies showing that insomnia severity is associated with increased chronic medical and psychiatric illnesses.[47] For example, chronic poor sleep is associated with higher incidence of cardiovascular disease,[48 49] metabolic disorders,[50 51] and neurodegenerative disease such as dementia,[52] with or without an independent diagnosis of a sleep disorder. A recent meta-analysis revealed that insufficient (<4 hours per night or totally daily) sleep duration was associated with an elevated risk of all-cause cognitive disorders or Alzheimer's disease dementia.[53] Poor sleep can significantly negatively impact disease development, relapse or worsening of disease symptoms across several therapeutic areas[54 55] often in a bi-directional manner. It is well understood that insomnia is related to both the onset and course of several psychiatric disorders (for a review, see[56] and [57]). Indeed, a recent meta-analysis indicated that insomnia was a significant predictor for the onset of depression, anxiety, and alcohol abuse.[57] People with chronic insomnia have a twofold risk for developing depression compared to good sleepers (for meta-analysis, see[58]), with poor sleep considered to be a transdiagnostic process in depression (i.e., it co-occurs with depression and is related to the onset and maintenance of depression).[59] This highlights the role of sleep as an important biological function essential to optimal living and that quality of sleep can significantly impact disease development and/or worsening of disease course across several therapeutic areas,[54 55] often in a bi-directional manner.

1.3.4 Management of Insomnia

Regardless of the type of therapy used, the treatment of insomnia has two primary objectives: improve sleep quality and quantity, and ameliorate daytime impairments.[60] Treatment broadly falls under two approaches – psychological therapy and short-term pharmacological

treatment. First-line treatment is cognitive behavioural therapy for insomnia (CBTi) which yields moderate-to-large, immediate and lasting improvements in sleep quality and quantity.[61] However, there are several barriers to treatment including access to a therapist and substantial time commitment and cost.[62] There are now increasing efforts to improve access via innovative digital CBTi approaches.[63] Regardless, the perceived benefits from these therapies are typically delayed. Thus, patients with chronic symptoms often seek short-term strategies such as pharmacological therapy to obtain relief from insomnia symptoms and maintain normal daytime functioning.

At present, benzodiazepines, antidepressants, orexin antagonists (e.g., lemborexant), and non-benzodiazepine hypnotics such as Z-drugs (e.g., zolpidem) remain the most common forms of pharmacological treatment for insomnia disorder.[60] Some can be effective in the short-term treatment of insomnia, but are either associated with poor tolerability, or lacking in information about long-term effects.[64] A recent meta-analysis published in the *Lancet* concluded that eszopiclone and lemborexant had the best profile in terms of efficacy and tolerability; however, the former can cause significant adverse events while safety data for the latter are inconclusive.[64] There was insufficient evidence to support the prescription of benzodiazepines and zolpidem for long-term treatment, and melatonin and ramelteon showed no overall material benefits. Undesirable side effects such as next-day hangover effects, cognitive or memory impairment, rapid development of tolerance, and car accidents or falls, due to their "off-target" effects at various binding sites in the central nervous system, are a major concern.[65-68] Moreover, most of the drugs currently used as hypnotics – in particular benzodiazepines, but also Z-drugs to a lesser extent – disturb sleep architecture.[69] Such disturbances can result in a sense of having had non-restorative sleep and can be associated with next-day impairments in conducting daily activities.[69] This has led to a rise in interest in alternative treatments such medicinal cannabis to target the unmet needs in individuals with insomnia disorder.

1.4 Use of Medicinal Cannabis for Insomnia

The pace and scale of the interest and uptake of medicinal cannabis in Australia and worldwide are unprecedented. Sleep disorders are one of the most common reasons that individuals self-report using medicinal cannabis in the community, after chronic pain and mental health-related disorders.[70-74] This is consistent with a recent analysis of medicinal cannabis prescribing trends in Australia with sleep disorders being the third most common indication after pain and anxiety for approval via the Therapeutic Goods Administration (TGA), the Australian federal regulator.[75] **Figure 2A** shows a steady increase in medicinal cannabis approvals for sleep disorders over time with a more rapid increase from April 2019. 'Sleep disorder' as an indication has by far the largest number of approvals per month (see **Figure 2B**) with a sharp drop in prescriptions for 'insomnia' from January 2021. This may reflect a major limitation of the Australian application process which does not require prescribers to specific strict diagnostic criteria, resulting in ambiguous or generic classifications.

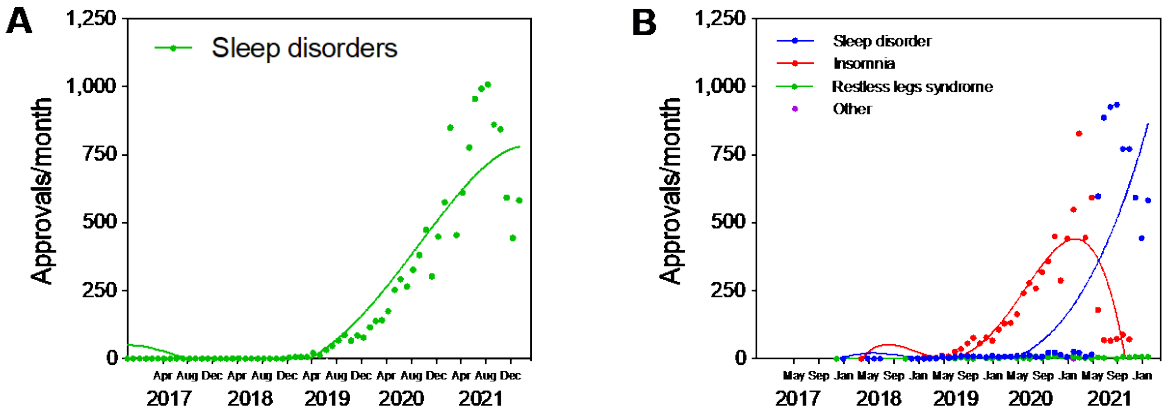

Figure 2 *Approvals for medicinal cannabis in Australia (via SAS-B pathway) since 2016. (A) Number of approvals per month for 'sleep disorders'; (B) Number of approvals per month for each different sleep disorder category. Solid lines represent the best fit with shaded standard error of the mean (SEM).*

Oral ingestion tends to be the preferred method among individuals using medicinal cannabis due to its discrete nature and lack of respiratory side effects compared with inhaled methods.[71 76] This is reflected in the recent analysis of prescribing trends in Australia with oil-based

products comprising an average of 80% of applications each month. Most approvals (75%) for sleep disorders were Schedule 8 products (i.e., containing >2% THC),[75] which is not unexpected given that THC is known to increase subjective feelings or drowsiness and sedation.[77,78]

Recent legislative changes have allowed increased access to cannabinoid products in many jurisdictions (e.g., US, Canada, Germany, and Australia) which includes online access to products containing CBD.[79] In the US, the online marketing of CBD products, typically oil formulations or alcohol-based tinctures containing CBD and combined with other ingredients such as melatonin or CBN (see **Figure 3**), are becoming increasingly widespread. However, the increasing patient access to and fascination with medicinal cannabis as a remedy for sleep disturbances has put the cart before the horse given the limited clinical evidence to support the use of cannabinoids for sleep disorders.

Figure 3 *Examples of CBD products marketed towards people with insomnia in the USA. Left: Terra Vita Sleep CBD oil with each dose formulated with 45 mg CBD and 2.5 mg melatonin; Right: CBDistillery 'Sleep Synergy' with each dose formulated with 900 mg CBD and 300 mg CBN.*

Surveys and observational studies of cannabis consumers have found that individuals commonly report using cannabis for sleep. Medicinal cannabis consumers reported using cannabis with higher CBD concentrations, with self-reported decreased sleep latency.[80] In the US, a recent survey showed that 74% of people accessing cannabis through adult-use markets in Colorado reported effective treatment for sleep, with a concomitant reduction in the use of prescription sleep aids.[81] In Canada, 92.6% of patients using prescribed medicinal cannabis reported a

significant improvement in their sleep after six weeks of treatment as assessed using the Pittsburgh Sleep Quality Index (PSQI).[82] A longitudinal, web-based survey of 1276 self-reported medicinal cannabis consumers found a significant improvement in sleep quality on the PSQI.[74] Similarly, in 593 older veterans enrolled in an US medicinal cannabis program, 77.1% self-reported a positive impact of medicinal cannabis use on their sleep quality as well as their pain (86%) and quality of life (89.4%). In a survey of 383 individuals with fibromyalgia in Israel, medicinal cannabis use was associated with improvements in pain (94%), sleep quality (93%), and depression (87%); with medicinal cannabis described as a "versatile remedy" in this population.

In addition to the aforementioned studies, more rigorously controlled studies using nabiximols (*Sativex)*, an oromucosal spray delivering equal parts THC and CBD, have examined efficacy for sleep but only as a secondary outcome using subjective outcome measures. Indeed, nabiximols improved subjective sleep quality across multiple clinical trials in the treatment of chronic pain (e.g., multiple sclerosis, neuropathic pain, cancer pain, and rheumatoid arthritis)[83]. There was moderate evidence for the use of Sativex in improving short-term sleep outcomes in individuals with sleep disturbances secondary to a pain condition,[83] however, it remains unclear whether this is due to an improvement in sleep *per se* or an improvement in the associated underlying condition. No studies have assessed the effects of *Sativex* in individuals with a sleep disorder as the primary condition. Nabilone, a synthetic THC analogue, has also been examined within this context. Specifically, Ware and colleagues conducted a study investigating the effectiveness of nabilone, compared with amitriptyline, in improving sleep among patients with fibromyalgia.[84] The researchers found that patients in both conditions evidenced improvements in sleep; however, treatment with nabilone was associated with greater improvements in sleep compared to amitriptyline.

Despite increased interest and uptake of prescribed and unregulated medicinal cannabis to treat insomnia and other sleep disorders, the evidence supporting therapeutic utility of cannabinoid therapies in sleep disorders is unclear and will be addressed in the current thesis.

1.5 The Phytocannabinoids

Cannabis is a chemically diverse plant containing more than 120 phytocannabinoids (the term used to emphasise the botanical origin of these cannabinoids; hereafter referred to as 'cannabinoids') as well as terpenes/terpenoids, flavonoids, phenolic compounds, and alkaloids.[85] Cannabinoids are synthesised within the glandular trichomes present in the flowers, leaves, and branches of the female cannabis plant. The first step in the cannabinoid biosynthesis pathway is the formation of cannabigerolic acid (CBGA) through the coupling of olivetolic acid and geranyl diphosphate (**Figure 4**). Through an enzymatic interaction, CBGA is then converted in the plant into the other acidic cannabinoids such as Δ^{9}-tetrahydrocannabinolic acid (THCA) and cannabidiolic acid (CBDA). When heated, THCA and CBDA undergo decarboxylation to produce the neutral cannabinoids, THC and CBD.[86] Partial decarboxylation can occur more naturally under room temperature under the influence of time (i.e. drying or curing) or exposure to light.[87]

Both THC and CBD interact with the endogenous cannabinoid (endocannabinoid) system, a complex and ubiquitous neuromodulatory network that includes cannabinoid 1 (CB_1) and 2 (CB_2) receptors, the endogenous ligands for these receptors such as anandamide (AEA) and 2-arachidonoylglyercol (2-AG), and the enzymes responsible for the biosynthesis and inactivation of these ligands including fatty acid amide hydrolase (FAAH).[88] THC, the most studied cannabinoid, has well-known characteristic psychoactive effects including euphoria, time distortion, and intensification of the sensory experiences.[89] The lack of notable activity of most other cannabinoids at CB_1 receptors explains why THC is usually described as the 'primary psychoactive component' in cannabis. THC is a partial agonist of the CB_1 receptor, found primarily within the central nervous system (CNS), and the CB_2 receptor, found primarily within the immune system and on peripheral organs. CBD, on the other hand, has weak binding affinity for the CB1 receptor and instead, acts predominantly as a negative allosteric modulator at CB1 (i.e., it can reduce the potency and/or efficacy of other ligands such as THC but does not

activate the receptor itself).[90] CBD exhibits promiscuous pharmacological activity at a range of receptor targets including ligand-gated ion channels (e.g., $GABA_A$), transient receptor potential channels (e.g., TRPV1), enzymes (e.g., FAAH, CYP450), and nuclear receptors (e.g., PPARγ),[91] which may explain the wide range of claimed therapeutic applications of CBD.

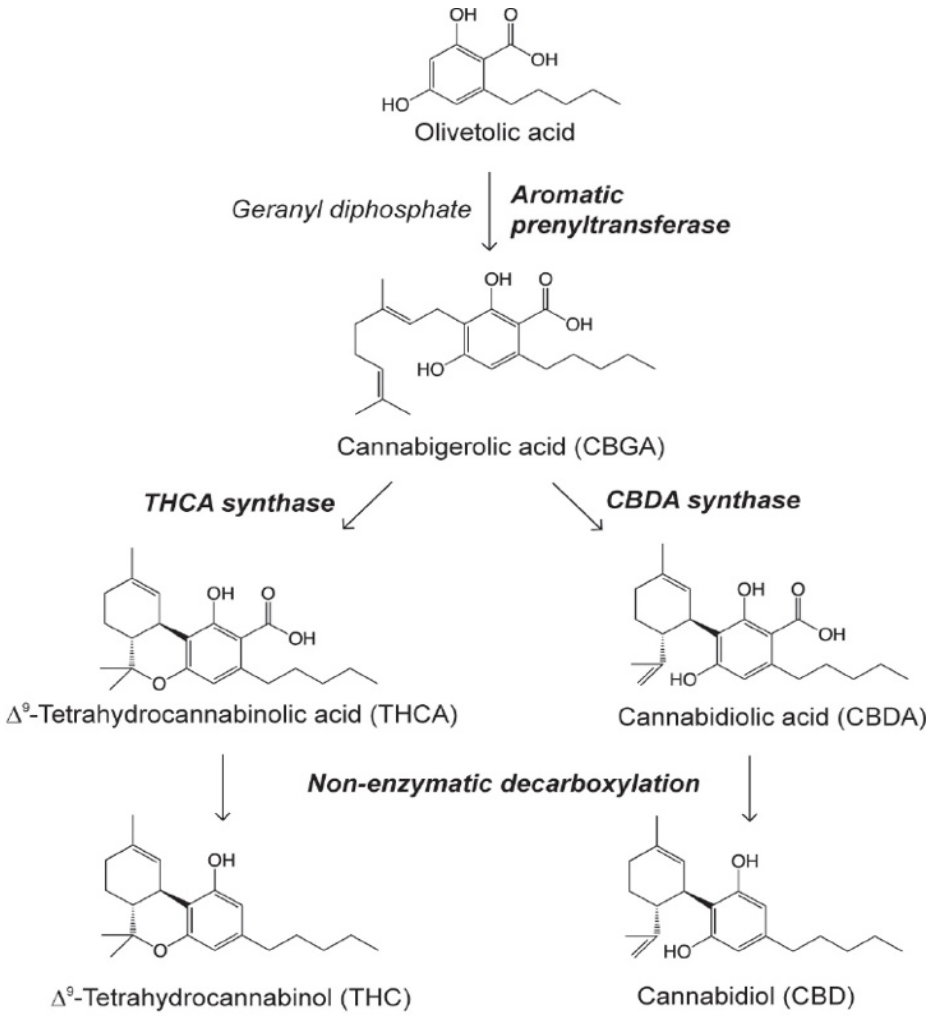

Figure 4 *Biosynthetic pathway of the main phytocannabinoids, THC and CBD.*

1.6 Pharmacokinetics of Cannabis and Cannabinoids

Cannabis used for medicinal purposes is typically taken orally as an oil or in capsule form, smoked, or vaporised, or ingested in the form of food (e.g., edibles). Less common routes of

administration include sublingual, topical and rectal. Depending on the route of administration, frequency and magnitude of drug exposure, the onset and duration of therapeutic drug effects can vary. Smoking or vaping produces a rapid and transient peak in THC blood and oral fluid concentrations within minutes of exposure.[92] Oral administration is the most common route for therapeutic application among medicinal cannabis users due its discrete nature and lack of respiratory side effects.[76] Compared with inhalation, oral administration has a later time of onset and longer duration of subjective drug effects due to slow absorption through the gastrointestinal tract, producing lower blood concentrations of THC and CBD with a more delayed peak plasma concentration occurring at 2-4 hours.[93]

Cannabinoids rapidly distribute into well-vascularised organs such as the lung, heart, brain and liver, with distribution affected by body size and composition.[94] Bioavailability of orally administered cannabinoids is generally low (~6%)[95] and highly variable due to significant first-pass metabolism by the liver via cytochrome P450 isozymes CYP2C9, CYP2C19 and CYP3A4.[96] THC is broken down to its pharmacologically active primary metabolite, 11-hydroxy-THC (11-OH-THC), by CYP2C9, and its direct oxidation produces a pharmacologically inactive metabolite, 11-carboxy-THC (11-COOH-THC).[94] CBD is metabolised by CYP3A4 and CYP2C19 to the pharmacologically active 7-hydroxy-CBD (7-OHCBD), which is subsequently oxidized to pharmacologically inactive 7-carboxy-CBD (7-COOH-CBD).[94] There is some evidence to suggest that 11-COOH-THC does not elicit subjective or physiological effects,[97] however, little is understood about the pharmacological activity of the metabolites of CBD in humans.[98]

1.7 Subjective Effects of Cannabis and Cannabinoids

THC is known to produce characteristic subjective drug effects including euphoria/elation, intensification of the sensory experiences, sedation, dry mouth, and increased appetite.[89] Indeed, oral THC administration dose-dependently increases subjective drug effects such as "good drug

effect", "sleepy/tired", "restless", and "hungry" 30-60 minutes after ingestion with peak effects occurring at 1.5 – 3 h post-drug administration, but does not tend to impair cognitive or psychomotor performance in infrequent cannabis users.[93] Of note, in Canada and several US states, 10 mg dose of THC has been set as the standard unit dose or "serving size" for cannabis edibles sold in the retail stores.[99] Higher doses of oral THC (i.e., 25 mg and 50 mg) produce more pronounced subjective effects and markedly impaired cognitive and psychomotor function relative to placebo in line with previous studies.[77] Preclinical studies consistently demonstrate sex differences in the response to acute cannabis effects with females exhibiting greater sensitivity to cannabinoid effects than males.[78] However, evidence in humans is less consistent with some studies showing significantly higher self-ratings of subjective drug effects (e.g., "heart racing", "anxiety/nervous")[94] [100] in females than in males while others show no evidence of sex differences.[93] [101]

In contrast, CBD is non-intoxicating and does not produce any overt subjective effects or impairment of cognitive or psychomotor function.[102-104] In an experimental study involving healthy volunteers, 100 mg CBD administered orally and via vaporization did not impact subjective ratings of alertness and sleepiness.[105] In clinical trials of high dose CBD (up to 25 mg/kg/day) in treatment-resistant paediatric epilepsy, increased somnolence and sedation was sometimes observed.[106] However, in these studies, CBD was found to be a potent metabolic inhibitor of concurrently-administered anticonvulsant medications such as clobazam and/or sodium valproate, which may have driven the sedating effects reported in these trials.[107] [108] In a Phase I ascending dose trial of CBD in healthy volunteers, drowsiness was reported as the fourth most common side effect, but the incidence did not differ from placebo and the greatest frequency of drowsiness observed was with an acute dose of 6000 mg, which far exceeds even the highest dose therapeutically indicated in humans (for example, the unit dose of CBD in Epidiolex is 100 mg).[109]

Some hypothesise CBD may have a 'calming' effect, although clinical evidence is limited.[110] Prior research has shown that CBD (oral, dose ranging from 300-400 mg) reduced anxiety in individuals with social anxiety disorder and healthy volunteers placed in stress-provoking situations

(e.g., simulated public speaking).[111] A recent 12-week open-label trial of CBD (oral; 800 mg) significantly reduced anxiety in young people with treatment-resistant anxiety disorder.[112] This suggests a possible anti-anxiety effect of CBD whereby a reduction in stress and/or anxiety may in turn improve sleep disturbances in people with insomnia. However, there are no published studies using validated subjective and/or objective measures investigating the effects of CBD on sleep in patients with anxiety disorders. Interestingly, there is emerging preclinical evidence describing the potential 'alerting' properties of CBD,[113] with one preclinical study showing that CBD partially blocked excessive sleepiness in hypocretin-deficient rats, an animal model of narcolepsy.[114] However, compelling clinical evidence is lacking.

1.8 Interactions between THC and CBD

Therapeutic doses of THC (e.g., 2.5-10 mg) tend to be considerably lower than for CBD (e.g., 50 – 1500 mg). Many combined products therefore contain CBD:THC ratios of 10:1, 20:1 or 50:1 whereby the CBD content is dominant, although 1:1 products are also very common.[115] There is emerging evidence that co-administration of THC and CBD may produce pharmacokinetic and pharmacodynamic interactions. It has been hypothesised that CBD may 'reverse' some of the adverse effects of THC such as anxiety or paranoia.[116] As mentioned previously, CBD is a negative allosteric modulator at CB1 receptors which means it could hypothetically "lessen' the partial agonist action of THC at these receptors thereby attenuating THC psychoactive effects. In human studies, CBD has sometimes attenuated some of the adverse effects of THC (e.g. on emotion recognition,[117] next-day memory performance,[118] appetitive effects,[119] and acute psychotic symptoms[120 121]). Naturalistic studies suggest that CBD-dominant cannabis was associated with significantly lower psychotic symptoms in regular cannabis users suggesting a potential role in modifying the impact of THC on the risk of psychotic outcomes.[121] An early review paper commented that the rationale for combining THC and CBD in a 1:1 ratio

in nabiximols (*Sativex*), an oromucosal spray approved in many countries for the treatment of spasticity associated with multiple sclerosis (MS), was to diminish the undesirable effects of THC.[122] Nabiximols, in doses of up to 43.2 mg THC and 40 mg CBD, does not appear to impair cognition in cannabis-naïve patients with MS[123-125]. Moreover, post-marketing data from the UK, Germany, and Switzerland suggests long-term use of prescribed nabiximols does not appear to be associated with cognitive decline or driving impairment in patients with treatment-resistant spasticity in MS.[126] Of note, a recent study showed that experienced cannabis users who were using a combined CBD and THC (oral; 5 mg each) product reported similar levels of positive and psychotomimetic effects compared to those who consumed a THC-only product (oral; 10 mg), despite consuming less THC and displaying lower plasma THC concentrations.[127] This suggests that co-administration with CBD may improve tolerability by lowering levels of THC exposure.

However, findings have not always been consistent with some studies failing to detect any CBD attenuation of THC-related subjective drug effects following inhaled and oral administration.[128-130] One study comparing the effects of vaporised THC-dominant (11% THC; < 1% CBD), THC/CBD equivalent (11% THC, 11% CBD), or placebo (< 1% THC/CBD) cannabis showed that CBD did not prevent THC-induced impairment on simulated driving and cognitive performance, and in some circumstances, CBD *exacerbated* THC-induced impairment.[131] These findings were subsequently validated in an on-road driving study where CBD did not mitigate the impairing effects of THC on driving and cognition when co-administered via cannabis vaporisation in a 1:1 ratio.[132] A recent study showed inhaled vaporised cannabis containing 10 mg THC and either 0, 10 mg, 20 mg, or 30 mg CBD did not protect against the acute adverse effects of cannabis.[133] However, most of these studies involve inhaled vaporised cannabis where higher CBD:THC ratios are impractical (compared to oral administration).

1.9 Cannabinoids and the Sleep-Wake Cycle

As noted above, both THC and CBD interact with the endogenous cannabinoid (endocannabinoid) system, a complex and ubiquitous neuromodulatory network that includes CB_1 and CB_2 receptors, the endogenous ligands for these receptors such as anandamide (AEA) and 2-arachidonoylglyercol (2-AG), and the enzymes responsible for the biosynthesis and inactivation of these ligands.[134] Our understanding of the influence of the endocannabinoid system on the circadian sleep-wake cycle is still evolving.[135] Clinical and preclinical studies describe a circadian rhythm in circulating endocannabinoid concentrations,[136-138] with plasma 2-AG levels increasing from mid-sleep to early afternoon in humans; an effect amplified by sleep restriction.[139] This effect is implicated in the complex relationship between sleep and appetite, whereby sleep deprivation caused afternoon elevations in 2-AG levels which coincided with self-reported increases in hunger and appetite in one study.[136] Pharmacological inhibition of monoacylglycerol lipase (MAGL), the rate-limiting enzyme responsible for the degradation of 2-AG, leads to elevated brain 2-AG concentrations and wake-promoting effects in rats, including reductions in both NREM and REM sleep.[140]

In contrast to 2-AG, AEA is associated with sleep-promoting effects: increasing endogenous AEA, via pharmacological inhibition of the degradative enzyme fatty acid amide hydrolase (FAAH), normalised deficits in stage N3 or 'slow wave sleep' (SWS) in cannabis-dependent males undergoing cannabis withdrawal.[141] Preclinical data similarly indicates that AEA promotes slow wave sleep, possibly via increases in extracellular adenosine concentrations.[142-144] Systemic injection of AEA increased extracellular concentrations of adenosine in the basal forebrain of a rat which increased sleep (decreased wakefulness and increased time in SWS),[142] while a CB_1 receptor antagonist, SR141716A, significantly reduced adenosine levels. This suggests a possible endocannabinoid-adenosine interaction mediated by the CB_1 receptor in sleep induction. Santucci and colleagues (1996) conducted one of first studies to understand the physiological role of

cannabinoid receptors in sleep, which showed that the CB_1 receptor inverse agonist, SR141716A (rimonabant), dose-dependently increased time awake and decreased time in slow wave sleep (SWS) and rapid eye movement (REM) sleep.[145]

In preclinical models, the sleep-promoting effects of AEA are blocked by rimonabant, indicating a CB_1-specific mechanism of action of AEA on sleep although this may also reflect the intrinsic actions of rimonabant itself.[146] In human clinical trials, insomnia and other sleep disorders were common with rimonabant treatment (observed in up to 10% of participants) and occurred more frequently than placebo.[147-150] Like AEA, THC is a partial agonist at the CB_1 receptor, and, thus, may exert sleep promoting effects via this direct pharmacological action on cholinergic neurons located in the basal forebrain and pons, assisting in the induction of sleep.[151]

CBD, on the other hand, has a weak binding affinity for the CB_1 receptor and, as noted above, acts predominantly as a negative allosteric modulator. [90] It may therefore sometimes attenuate the pharmacological effects of THC and anandamide.[152] [153] CBD, however, is a promiscuous molecule that exhibits activity on a wide array of molecular targets beyond CB_1 and CB_2 receptors including benzodiazepine-like effects on inhibitory $GABA_A$ receptors,[154] which may also influence sleep. CBD can also increase AEA concentrations via inhibition of fatty acid binding proteins (FABPs) and FAAH,[155-157] which provides an alternative pharmacological mechanism by which CBD may promote sleep. Overall, this highlights a complex modulatory role for the endocannabinoid system, and potential mechanisms for THC and CBD, in regulating the sleep-wake cycle.

1.10 Effects of Cannabis on Sleep Architecture

1.10.1 Introduction

Acute and prolonged cannabis use can have a broad range of effects on the structure of sleep, which can have important implications for optimal daytime function. Sleep architecture refers to the basic structural organisation of sleep and is measured using overnight polysomnography or actigraphy. A typical night involves 4 – 6 sleep cycles each lasting approximately 90 minutes each. There are two categories of sleep: non-rapid eye movement (NREM) sleep and rapid eye movement (REM) sleep. NREM sleep constitutes about 75-80% of total time spent in sleep, while REM sleep constitutes the remaining 20-25%. NREM occurs first and consists of three stages: N1, N2 and N3, representing a continuum of relative depth of sleep.[158] Stage N1 or 'light sleep' serves a transitional role in sleep occurring immediately upon falling asleep and is typically very short (<10 mins). This quickly progresses to stage N2 sleep during which the body enters a more subdued state including a decrease in body temperature, relaxed muscles, and slowed breathing and heart rate.[158] EEG shows relatively low-voltage, mixed-frequency activity hallmarked by the ubiquitous presence of K-complexes and sleep spindles.[159] Stage N2 lasts approximately 10-25 minutes in the initial cycle and lengthens with each successive cycle, constituting 45-55% of each sleep cycle.[160] The next stage of sleep, N3 or 'slow wave sleep' or 'deep sleep', is characterised by low-frequency, high-amplitude oscillatory EEG activity that plays an important role in sleep-dependent memory consolidation.[160] The final stage of the sleep cycle is REM sleep (when dreams occur) which is characterised by the presence of desynchronised low-voltage, mixed-frequency brain activity, muscle atonia and bursts of rapid eye movements, features that are remarkably similar to that of the awake state.[160] In the initial cycle, REM sleep lasts for only 1-5 minutes, but becomes progressively prolonged with each new cycle. Sleep architecture is known to change with age,[161] medication use (e.g., antidepressants reduce REM sleep),[162] and certain psychiatric and neurological conditions (e.g., Parkinson's disease).[163]

The current section reviews the literature on the effects of cannabis on sleep architecture in relation to five key objective sleep outcomes. These are: (1) sleep onset latency, (2) total sleep time, (3) wake after sleep onset, (4) slow wave sleep, and (5) REM sleep. **Table 1** provides a summary of key sleep terminology for reference.

Table 1. Sleep terminology

Objective sleep outcome	Description
Sleep onset latency (SOL)	Amount of time it takes to initiate the first period of any sleep stage
Total sleep time (TST)	Total amount of sleep time from onset of sleep to final wakening
Wake after sleep onset (WASO)	Total amount of time spent awake after sleep onset until final wakening
Slow wave sleep (SWS)	Stage N3 sleep characterised by low frequency and high amplitude waves
REM sleep	Rapid eye movement (REM) sleep

Table 2 summarises the outcomes of studies to-date that examined the effects of cannabis on sleep architecture using objective measures such as polysomnography or actigraphy. These studies were identified through a comprehensive literature search that included key words relating to each of the sleep stages described above as well as "sleep cycle", "sleep stage", "sleep architecture", "polysomnography", "actigraphy", "cannabi*", "THC", "CBD", and related terms (e.g., "marijuana"; "dronabinol"; "nabilone"; "Sativex"; "Epidiolex").

The following studies were included: (a) involved administration of cannabis, a cannabinoid, or a modulator of the CB1 and/or CB2 receptors at a clearly defined or estimable concentration; (b) assessed and reported the above listed objective sleep parameters (i.e., studies that reported on objective sleep parameters ('nocturnal motor activity'[164]) other than the above listed were excluded).

Table 2 Clinical studies investigating the effects of cannabis and cannabinoids on sleep architecture using objective sleep measures

Author, year [citation]	Study Design	Participants	Cannabis Use History	Treatment Duration	Intervention, dose, and timing	Objective sleep measures	SOL (mins)	WASO (mins)	TST (mins)	SWS (mins)	REM sleep (mins)	Other effect(s)
Oral administration												
Walsh et al., (2021)[165]	DB, PC Crossover	*N*=23 (3 M 20 F) 52 ± 9 y *Insomnia*	P	2 weeks	10 mg THC, 1 mg CBN, and 0.5 mg CBD Dose-escalation to double the dose permitted from the 4th night	ACT	x	↓	↑	-	x	↑ SE (+2.9%)
						PSG	x	x	x	x	x	↑ REM latency (+54.2 min)
Linares et al., (2018)[166]	DB, PC Crossover	N=27 (12 M 14 F) 29.3 ± 8.5 y *Healthy volunteers*	N	Acute	CBD 300 mg	PSG	x	x	x	x	x	
Carley et al., (2018)[167]	DB, PC Parallel	N=73 (52 M) I: 52.7 ± 7.7 y II: 54.7 ± 7 y *OSA*	N	6 weeks	Dronabinol 2.5 mg/day	PSG	x	x	x	x	↑	
					Dronabinol 10 mg/day		x	x	x	x	↑	
Zalai et al., (2015)[168]	DB, PC Crossover	*n*=11 (age not specified) *Insomnia & pain*	P	4 weeks	Nabilone (dose unspecified)	PSG	↑	x	x	x	x	
Farabi et al., (2014)[169]	Open label Compared to baseline	N=15 (6 M 9 F) 51.7 ± 7.9 y *OSA*	N	1 week	Dronabinol W1: 2.5 mg/day (n=2) W2: 5 mg/day (n=5) W3: 10 mg/day (n=8)	PSG	x	x	x	x	x	↓ Stage N2 sleep
Prasad et al., (2013)[170]	Open label Compared to baseline	*n*=17 (6 M) I: 51.6 ± 7.9 y *OSA*	N	3 weeks	Dronabinol 10 mg/day	PSG	x	x	x	x	x	
Nicholson et al., (2004)[118]	PC Crossover	N=8 (4 M, 4 F) 28.8 y (24-34 y) *Healthy volunteers*	O	Acute	15 mg THC	PSG	x	x	x	x	x	↑ increased drowsiness
					5 mg THC + 5 mg CBD		x	x	x	↓	x	
					15 mg THC + 15 mg CBD		x	↑	x	↓	x	
Haney et al., (2007)[171]	DB, PC Crossover	N=10 (all M; 7 with sleep data) 40.1 ± 1.9 y *HIV positive*	Fr	4 days	Daily dose of THC (2% and 3.9%) taken over 4 d and then daily dose of dronabinol (5 mg or 10 mg) over 4 d	'Nightcap' sleep monitor	-	-	-	-	-	No effect on objective sleep outcomes
Bedi et al., (2010)[172]	PC Crossover	N=14 (all M; 7 with sleep data) 36.6 ± 1.3 y *HIV positive*	Fr	16 days	Dronabinol 40 mg/day for 16 d	'Nightcap' sleep monitor	-	↓	-	-	-	↑ Sleep efficiency on day 1-8 only (due to ↑ NREM sleep and ↓ time spent awake)
Feinberg et al., (1975)[173]	DB, PC Crossover	N=7 (all M) 25 y (22-27 y) *Chronic cannabis users*	Fr	~ 16 days	Initial dose: 70 mg/day THC	PSG	x	x	x	x	↓	
					Titrated to 210 mg/day THC		x	x	x	x	↓	
Feinberg et al., (1976)[174]	PC Crossover	N=4 (all M) 21.7 – 31.2 y	Fr	3 nights	Initial dose: 70 mg/day THC	PSG	x	x	x	-	x	↑ average duration of awakenings with 210 mg/d ($p<0.01$)
					Titrated to 210 mg/day THC		x	x	x	-	x	

Table 2 Clinical studies investigating the effects of cannabis and cannabinoids on sleep architecture using objective sleep measures

Author, year [citation]	Study Design	Participants	Cannabis Use History	Treatment Duration	Intervention, dose, and timing	Objective sleep measures	SOL (mins)	WASO (mins)	TST (mins)	SWS (mins)	REM sleep (mins)	Other effect(s)
		Chronic cannabis users										
Tassinari et al., (1999)[175]	Open label Compared to baseline	N=7 21 – 25 y *Healthy volunteers*	N	Single dose following 1-2 nights no drug	0.7 – 1.4 mg/kg THC	PSG	-	-	-	↓	↓	↑ Stage N2 sleep
Pivik et al., (1972)[176]	Open label Compared to baseline	N=6 (all M) Age not specified *Healthy volunteers*	N	Acute	13- 17 mg THC before bed (n=4) 20 mg THC the morning after two nights of sleep deprivation (n=2)	PSG	x x	↓ x	x x	x x	↓ ↓	Recovery: ↓ REM latency, SOL and Stage N1
Inhaled administration												
Freemon et al., (1972)[177]	Open label Compared to baseline	N=2 (all F) *Healthy volunteers*	O	4 nights	THC 20 mg	PSG	-	-	-	-	↓	Recovery: ↓ REM latency & ↑ wakefulness
Barratt et al., (1974)[178]	Compared to drug-naïve group	N=12 (all M) 21- 26 y *Chronic cannabis users*	Fr	10 d with 2 joints/day	0.2 mg/kg of THC per joint	PSG	x	x	x	↕	x	Acute: ↑ SWS Chronic: ↓ SWS
Freemon et al., (1982)[179]	PC Crossover	N=2 (all M) *Healthy volunteers*	O	2 weeks	THC 30 mg before bed	PSG	x	x	x	↓	x	Withdrawal: ↓ SWS & ↑ wakefulness
Hosko et al., (1973)[180]	DB, PC Crossover	N=7 (all M) 24 – 28 y Mixed type cannabis users	N (n=2) Fr (n=4) 0 (n=1)	Acute Acute	THC 0.2 mg/kg THC 0.3 mg.kg	PSG PSG	x x	x x	x x	x x	x x	↑ SWS in 28.6% ↓ REM sleep (n=2 only)
Pranikoff et al., (1973)[181]	Compared to drug naïve (no blinding)	N=20 (all M) 20 - 25 y *Cannabis-naïve and frequency users*	N (n=10) Fr (n=10)	Acute	THC (unclear dose) – use until "reaching a subjective high"	PSG	x	x	x	x	x	
Karacan et al., (1976)[182]	Open label Compared to drug-naive	N=32 (all M) 20 - 48 y *Chronic cannabis users*	Fr	8 nights	'Usual pattern of cannabis use' (~ 9.2 joints per day)	PSG	↑	x	x	x	↑	↑ Length of REM period in chronic cannabis users relative to drug-naïve controls

ACT, actigraphy; DB, double blind; F, female; Fr, frequent cannabis user; M, male; N, cannabis-naïve; O, occasional cannabis user; OSA, obstructive sleep apnea, P, possible prior cannabis use (frequency not specified); PC, placebo-controlled; REM, rapid eye movement sleep; SOL, sleep onset latency; SWS, slow wave sleep; TST, total sleep time; WASO, wake after sleep onset; y, year

1.10.2 Sleep Onset Latency

No study identified an improvement (i.e., reduction) in the time taken to fall asleep (sleep onset latency; SOL) following cannabis administration. Two studies showed an increase in SOL following ingestion of oral nabilone (synthetic THC-like compound), and smoked cannabis, respectively.[168 182] In the former, 4-week treatment with nabilone (oral) in 11 patients with chronic pain and insomnia produced a clinically significant delay in SOL of 31.8 min relative to placebo.[168] This effect occurred despite pain improving by an average of 3 points on the McGill Pain Questionnaire in all participants.[169] The latter study, conducted in 1976, showed a significant increase in SOL by 10.3 min in 32 male participants following eight consecutive nights of 'usual pattern of cannabis use' (i.e., smoked; dose unspecified).[182] In this study, all participants regularly used cannabis making it difficult to make any conclusions regarding the effects of cannabinoids in cannabis-naïve individuals. No other study included in this review showed a significant change in SOL following cannabinoid administration.

1.10.3 Wake After Sleep Onset

Four studies reported opposing effects of cannabinoids on time spent awake after sleep onset (i.e., wake after sleep onset; WASO), with three studies showing a decrease in WASO, and one showing an increase.[165] Of the three studies that showed a decrease in WASO, the first was a randomised, placebo-controlled trial of a 2-week treatment with 'ZTL-101' (containing 10 mg THC, 1 mg cannabinol (CBN), and 0.5 mg CBD) in 23 individuals with clinician-diagnosed insomnia disorder.[165] Relative to placebo, this intervention produced a significant actigraphy-derived reduction in WASO of 10.2 min, although this was not corroborated with polysomnography.[165] Second, in a 1972 study, six cannabis-naïve males showed a reduction in WASO following an acute dose of both 13 and 17 mg THC; however, adequate statistical analysis was not performed.[176] In the third study, oral consumption of 40 mg/day dronabinol (THC) for 16 days in seven males who frequently used cannabis resulted in a reduction in WASO; however, similar to the above study, statistical analysis was lacking.[172]

In a study involving eight cannabis-naïve healthy volunteers, an acute dose of 15 mg THC and 15 mg CBD combined produced a significant increase in WASO of 23.8 min relative to placebo.[118] This effect was not observed with a lower dose (i.e., 5 mg THC and 5 mg CBD combined) or with 15 mg THC alone, with the authors concluding that co-administering CBD with THC at the higher dose of 15 mg each produced an 'alerting' effect.

1.10.4 Total Sleep Time

Only one study showed a change with cannabinoids in the total amount of time spent asleep (total sleep time; TST). In this randomised, placebo-controlled trial of 'ZTL-101' (containing 10 mg THC, 1 mg cannabinol (CBN), and 0.5 mg CBD), TST was significantly increased by 33.4 min on actigraphy relative to placebo in 23 cannabis-naïve participants with chronic insomnia.[165] This was accompanied by an improvement in the Insomnia Severity Index (ISI) of 5.1 points and significant improvements in self-reported sleep quality and 'feeling more rested/refreshed on waking'. Participants also self-reported that average TST increased by 64.6 min as measured on the sleep diary, double the change measured on actigraphy. However, no changes to TST with cannabinoid treatment were observed with polysomnography (-3.5 min, $p>0.05$). No other study observed a change in TST.

1.10.5 Slow Wave Sleep

Four studies reported a reduction in stage N3 sleep (slow wave sleep; SWS) following administration of cannabis or cannabinoids. In one of these studies, both low and high acute doses of combined THC and CBD (i.e., 5 mg and 15 mg each, respectively) produced a significant reduction in SWS in eight healthy volunteers who used cannabis occassionally.[118] Another study showed that 1-week oral administration of 0.7 – 1.4 mg/kg THC in seven cannabis-naïve healthy volunteers similarly produced a reduction in SWS.[175] A study from 1982 involving two cannabis-naïve males found a reduction in SWS follow 2-week treatment with high dose THC (oral; 30 mg), with a sustained reduction in SWS following withdrawal.[179] Another study showed that 0.2 mg/kg THC ingested via smoking in 12 males who were frequent cannabis users reduced SWS following

10 nights of dosing.[173] All four studies were underpowered, with three studies involving <8 cannabis-naïve volunteers and one study involving 12 chronic cannabis users. This precludes any definitive conclusions on the impact of cannabis on stage N3 sleep.

1.10.6 REM Sleep

Five studies have investigated effects of cannabinoids (all THC) on REM sleep, with mixed results. A 6-week randomised, placebo-controlled trial of dronabinol (THC) in 73 individuals with moderate-to-severe obstructive sleep apnea demonstrated a significant increase in the percentage of time spent in REM sleep at 2.5 mg/day (+4.4%) and 10 mg/day (+4.3%). No other significant effects on EEG outcomes were identified in this study. A second study conducted in 1976 showed that *ad libitum* smoked cannabis (9.2 joints per day) increase the length of the REM period in 32 males who were frequent cannabis users relative to drug-naïve controls.[165]

The three remaining studies (all conducted in the 1970s) found a reduction in REM sleep following THC administration. One study showed that 'round-the-clock' THC dosing in four individuals who were frequent cannabis users resulted in a significant reduction in REM sleep at both dosages (70 mg/day and 210 mg/day).[173] Another study reported a signification reduction in REM sleep in cannabis-naïve individuals following acute administration of 13-17 mg THC and following administration of 20 mg THC the morning after two nights of sleep deprivation (i.e., THC prevented REM rebound following sleep deprivation).[176] The third study showed a reduction in REM sleep following four nights of 20 mg THC in four individuals who used cannabis occasionally.[177] Upon cessation of dosing (i.e., 'recovery'), the researchers noted a decrease in the latency to REM sleep and increased wakefulness. Of note, the recently published randomised, placebo-controlled trial of 'ZTL-101' (containing 10 mg THC, 1 mg CBN, and 0.5 mg CBD) in individuals with chronic insomnia found a non-significant trend towards a reduction in REM sleep (mean difference 3.5%, p=0.055).

1.10.7 Cannabis Withdrawal and Sleep

THC displays minimal toxicity and lethality,[183] inferring a safety advantage over hypnotic medications. However, abrupt discontinuation of daily, or near daily cannabis use may lead to abstinence-induced insomnia[184] with sleep difficulty a commonly reported symptom during cannabis withdrawal in frequent cannabis users (e.g. at least 25 days/month).[185] Poor sleep quality is also a risk factor for lapse following a cannabis quit attempt in cannabis dependent users.[186] Laboratory studies have shown that cannabis abstinence-induced sleep disturbance is specific to THC exposure, as it can be reversed with administration of dronabinol (THC) or by a return to cannabis use.[187] [188] Most of the extant literature has focused on cannabis withdrawal syndrome in recreational (non- medicinal) cannabis users. PSG studies of cannabis withdrawal in daily cannabis users have demonstrated increases in sleep-onset latency and wakefulness, and decreased TST, SWS and sleep efficiency.[189] REM sleep rebound (i.e., increase in REM sleep and decreased REM onset latency following a period of REM sleep suppression) has also been reported.[189] Nightmares and/or strange dreams are common but tend to cause relatively little associated distress.[190] Changes in sleep architecture typically persist for two weeks post-abstinence[191] and self-ratings of sleep difficulty (including strange dreams) have been observed up to 1.5 months.[192]

In contrast to inhaled cannabis, controlled studies examining withdrawal from *Sativex*, a regulated oromucosal spray delivering 2.7 mg THC and 2.5 CBD per spray, did not observe a clear withdrawal syndrome (the average dose in short- and long-term clinical trials was 8 sprays/day in divided doses).[193] In two clinical trials, *Sativex* treatment was abruptly stopped to assess the possibility of a withdrawal syndrome occurring.[194] [195] The first study randomly allocated 36 patients with multiple sclerosis (MS) maintained on Sativex for an average of 3.6 years to continue with Sativex (n = 18) or to change to identical placebo (n = 18).[195] No withdrawal syndrome was observed. A second study reported details of 25 patients with MS who interrupted treatment for 2 weeks during long-term therapy and again no consistent withdrawal syndrome was observed with an average of 11 sprays daily (equivalent to 30 mg THC and 28 mg CBD).[194] However, 11/25

(46%) reported at least one of the following symptoms during the withdrawal period: tiredness (4/25; 16%), interrupted sleep (4/25; 16%), hot and cold flushes (4/25; 16%), mood alteration (3/25; 12%), reduced appetite (2/25; 8%), emotional lability (2/25; 8%), and vivid dreams. However, no control group was used for comparison in either of the aforementioned studies. CBD, on the other hand, has not been associated with dependency or a withdrawal syndrome of any kind following abrupt cessation.[196]

1.10.8 Summary

There is only very limited research examining the effects of cannabis on sleep, with a conspicuous lack of modern studies that have employed rigorous measures of sleep architecture. Much of the extant literature was conducted using small sample sizes with considerable heterogeneity with respect to dosage, timing, and route of administration and, importantly, patient characteristics including their prior cannabis use history. Three studies involved healthy volunteers (who were cannabis-naïve) while the remaining involved heterogenous patient populations (i.e., sleep disorders, immunocompromised individuals, or individuals who used cannabis regularly) which introduces a range of confounding factors that may conceivably influence sleep architecture. Thirteen out of the 19 included studies recruited participants with a history of possible, occasional, or frequent cannabis use, who tend to have poorer sleep at baseline compared to non-users.[189] Results may therefore reflect sleep architecture associated with chronic heavy cannabis use and/or withdrawal, which can be significant. The majority of studies have focused on the effects of THC or a THC-like compound (e.g., nabilone) although one study examined two doses of a balanced THC:CBD formulation and another study examined a formulation of combined THC, CBD and CBN. One study investigated the acute effects of CBD alone. Therefore, much of our current understanding of how cannabis affects sleep architecture is specific to the effects of THC only despite the fact that many patients currently use products that combine THC and CBD.[115] Some involved smoked whole cannabis/flower or 'cannabis extracts' with unknown concentration of

cannabinoids and other constituents (e.g., minor cannabinoids or terpenes/terpenoids), precluding any definitive conclusions regarding the effects of THC alone on sleep architecture.

Despite these limitations, there may be a possible association between cannabis and a reduction in REM sleep and SWS, however, controlled studies using high-quality trial design in cannabis-naïve individuals are necessary to confirm these findings. Only one study examined the effects of CBD alone (oral; 300 mg) on sleep architecture in healthy volunteers and found no significant effects. Another study alluded to the possibility of CBD having wakefulness-promoting properties based on the observation that an acute dose of combined 15 mg THC and 15 mg CBD produced a significant increase in WASO. This effect, however, was not seen with a lower dose (i.e., combined 5 mg THC and 5 mg CBD) or with 15 mg THC alone.[118] The effects observed in this small study (n=8) have not been replicated to-date. While this adds to a larger body of existing preclinical work describing the potential 'alerting' properties of CBD,[113] compelling clinical evidence is lacking.

In summary, we have limited understanding of the effects of THC, CBD, and their combination on sleep architecture. Only one study to-date has explored the effects of cannabinoids on sleep architecture in insomnia disorder. These findings of this work will be extended on in the current thesis.

1.11 Next-day Residual Effects of Cannabis

1.11.1 Introduction

The therapeutic utility of an insomnia medication cannot be solely determined by its ability to induce and maintain sleep. Residual daytime sleepiness and associated impairment of psychomotor and cognitive functioning is a major problem with some hypnotic drugs.[197] Similarly, one ongoing concern around cannabis is that the major psychoactive constituent, Δ^9-tetrahydrocanninol (THC), can induce intoxication and impair cognitive and psychomotor

performance (e.g., reaction time, working memory, divided attention).[198] This can potentially increase the risk of error, accident and injury when operating a motor vehicle or equipment or engaging in other safety-sensitive tasks. The duration of such impairment is the critical issue, particularly for those using a THC-based medication at night for sleep. A recent systematic review and meta-regression analysis concluded there was a 'window of impairment' extending after THC use for between ~3- and 10-hours, with the exact duration dependent on dose, route of administration and whether regular or occasional users were being assessed.[198] However, it did not include outcomes involving performance assessment >12-hours after THC use which is relevant to the 'next day' or 'next-morning' effects of medications taken at night by people with insomnia disorder.

Several governmental agencies, and various experts in occupational safety, have cautioned that THC-induced impairment may last for >24-hours and recommend individuals avoid performing safety-sensitive tasks for at least this long after THC use.[199 200] Such prohibition is problematic for those using cannabis in the evenings to treat a sleep disorder but does not appear to have been informed by a comprehensive review of the scientific evidence. Therefore, the current section reviews the extant literature to better understand the 'next day' (i.e., >8 hour) effects of THC use on cognitive function and safety-sensitive tasks. Studies that measured performance on 'safety-sensitive' tasks (e.g., simulated or on-road driving, simulated aeroplane flying) and/or discrete neuropsychological tests >8-hours post-THC (or cannabis) administration using an interventional design were eligible for inclusion (**Table 3**). Studies were excluded if THC was co-administered with another treatment (e.g., alcohol). Risk of bias in included studies was evaluated by two independent assessors using: (1) the Revised Cochrane Risk of Bias tool (RoB 2.0)[201]; and (2) the RoB 2.0 for crossover trials[202], as appropriate (see **Figure 5**).

Table 3 Clinical studies that measured performance on 'safety-sensitive' tasks and/or neuropsychological tests >8 hours post-THC (or cannabis) administration

Author, year [citation]	Study Design	Participants	Cannabis Use History	Treatment	THC Dose (mg)	Performance Test	Time Since Last THC Use	Effect of THC (compared to placebo unless otherwise stated)
Matheson et al., (2020)[203]	Randomised; DB; PC (BSD)	C: 30 (21 M); 22±2 y I: 31 (18 M); 22±2 y	O/F	Smoked Cannabis Cigarettes (562±170 mg; 12.5% THC) (<0.5% CBD)	70.3±21.3[a]	Grooved Pegboard Task DSST CPT HVLT-R	24 & 48 h	**THC ↑ Number of Correct Trials at 48 h on DSST at both doses**
		C: 30 (21 M); 22±2 y I: 30 (26 M); 22±2 y		Smoked Cannabis Cigarettes (752±131 mg; 12.5% THC) (<0.5% CBD)	94.0±16.4[a]			
Brands et al., (2019)[204]	Randomised; DB; PC (BSD)	C: 30 (21 M); 22±2 y I: 31 (18 M); 22±2 y	O/F	Smoked Cannabis Cigarettes (562±170 mg; 12.5% THC) (<0.5% CBD)	70.3±21.3[a]	Simulated Driving	24 & 48 h	No significant effects
		C: 30 (21 M); 22±2 y I: 30 (26 M); 22±2 y		Smoked Cannabis Cigarettes (752±131 mg; 12.5% THC) (<0.5% CBD)	94.0±16.4[a]	Simulated Driving	24 & 48 h	**THC ↓ SDLP at 48 h**
Hartley et al., (2019)[205]	Randomised; DB; PC (WSD)	15 M; 22±3 y	O O F F	Smoked Cannabis Cigarettes (9.8% THC; 1 g tobacco) (<0.1% CBD and CBN)	10 30 10 30	Simulated Driving PVT	12 & 24 h	No effect[b]
Schoedel et al., (2018)[103]	Randomised; DB; PC (WSD)[c]	43 (31 M)[d]; 38±9 y	O/F	THC Capsules	10 30	Divided Attention Task HVLT-R DSST	12 & 24 h	No significant effect No relevant analyses[e]
Ronen et al., (2008)[206]	DB; PC (WSD)	14 (10 M); 22±2 y	O	Smoked THC Cigarettes	17	Simulated Driving	24 h	No significant effects[f]
Ménétrey et al., (2005)[207]	Randomised; DB; PC (WSD)	8 M[g]; Range: 22–30 y	Unclear	Hemp Milk Decoction Hemp Milk Decoction THC Capsules	16.5 45.7 20	Road Sign Test Divided Attention Task	10 & 25 h	Ambiguous[h]
Nicholson et al., (2004)[118]	DB; PC (WSD)	8 (4 M); 21–34 y	N	Oromucosal Spray	15	Word Memory Recall Digit Memory Recall 6-Letter Memory Recall	10 h	**THC ↓ Immediate & Delayed Recall**
				Oromucosal Spray (+5 mg CBD)	5	DSST Multi-attribute Task		**THC ↑ Reaction Time**
				Oromucosal Spray (+15 mg CBD)	15	Choice Reaction Time Sustained Attention Task		No significant effects
Curran et al., (2002)[208]	Randomised; DB; PC (WSD)	15 M; 24±2 y	Unclear	THC Capsules	7.5	Buschke Selective Reminding RVIPT Baddeley Reasoning Task Subtract Serial Sevens Task Choice Reaction Time Task	24 & 48 h	Ambiguous No significant effects
					15	Digit Cancellation Task Simple Reaction Time Task		

Author, year [citation]	Study Design	Participants	Cannabis Use History	Treatment	THC Dose (mg)	Performance Test	Time Since Last THC Use	Effect of THC (compared to placebo unless otherwise stated)
Fant et al., (1998)[209]	Randomised; DB; PC (WSD)	10 M; 27 y, Range: 24–31 y	O	Smoked Cannabis Cigarettes (1.8% THC) Smoked Cannabis Cigarettes (3.6% THC)	"Eight Puffs"	Smooth-Pursuit Eye Movements Circular Lights Task Serial Addition and Subtraction Digit Recall Task Logical Reasoning Task Mannequin Task	23, 24 & 25 h	Ambiguous [i] No significant effect
Chait and Perry (1994)[210]	DB; PC (WSD)	14 (10 M); 25 y, Range: 21–34 y	O/F	Smoked Cannabis Cigarettes (3.6% THC)	"Eight Puffs"	Time Production Task Standing Steadiness Task DSST Backward Digit Span Task Logical Reasoning Task Visual Divided Attention Free Recall Task	11 & 18 h	No significant effects
Leirer et al., (1991)[211]	"Blinded"[j]; PC (WSD)	9 (Sex NS); 31 y, Range: 24–40 y	Unclear	Smoked Cannabis Cigarettes	20	Simulated Flying	24 & 48 h	**THC ↓ performance at 24 h**
Chait (1990)[212]	DB; PC (WSD)	12 (9 M); 21 y, Range: 18–26 y	O/F	Smoked Cannabis Cigarettes (800–900 mg; 2.1% THC)	"Eight Puffs"[k]	Time Production Task Simple Reaction Time Task Forward Digit Span Task Visual Divided Attention Choice Reaction Time Task Backward Digit Span Task DSST Buschkel Selective Reminding	12, 12 & 12 h[k]	**THC ↓ Time Interval (all days)[l, m]** **THC ↑ Reaction Time (all days)[l]** **THC ↓ Digit Span on Day 1**
Heishman et al., (1990) [213]	Randomised; DB; PC (WSD)	3 M; Range 27–29 y 3 M; Range 27–29 y 2 M; Range 27–29 y	Unclear	Smoked Cannabis Cigarettes (2.57% THC)	"1 x Cigarette" "2 x Cigarette" "4 x Cigarette"	Two Letter Search Task Logical Reasoning Task Digit Recall Task Serial Addition and Subtraction Task Circular Lights Task	23, 25, 27, 29 & 31 h 19, 21, 23, 25 & 27 h	Results not adequately reported
Leirer et al., (1989)[214]	Randomised; DB; PC (WSD)	9 (Sex NS); 26 y, Range: 18–29 y 9 (Sex NS); 38 y, Range: 30–48 y	Unclear	Smoked Cannabis Cigarettes	10 20	Simulated Flying	24 & 48 h	No significant effects
Barnett et al (1985)[215]	"Blinded"[j]; PC (WSD)	8 M; Range: 22–33 y	Unclear	Smoked Cannabis Cigarettes (700 mg; 1% THC)	100 µg·kg^{-} (6.8–7.3 mg) 200 µg·kg (14–15 mg) 250 µg·kg (17–18 mg)	Visual Search Task Divided Attention Task Critical Tracking Task	10, 12 & 23 h	No effect[b]

Author, year [citation]	Study Design	Participants	Cannabis Use History	Treatment	THC Dose (mg)	Performance Test	Time Since Last THC Use	Effect of THC (compared to placebo unless otherwise stated)
Chait et al., (1985)[216]	"Blinded"[j]; PC (WSD)	13 M; 25 y, Range: 21–35 y	O/F	Smoked Cannabis Cigarettes (1 g; 2.9% THC)	"Ten Puffs" (Dose Unknown)	Card Sorting Task Free Recall Task DSST Time Production Task	9.5 h	**THC ↑ Time Interval (10 & 30 s) at 9.5 h compared to Target**
		6 M; age NS	Unclear	Smoked Cannabis Cigarettes (1 g; 2.9% THC)	"Five Puffs" (Dose Unknown)			Results not reported
Yesavage et al., (1985)[217]	Pre/Post Trial	10 (Sex NS); 29 y	Unclear	Smoked Cannabis Cigarettes	19	Simulated Flying	24 h	**THC ↑ Distance Off-Centre on Landing, Lateral Deviation, Aileron (Number of Changes)** **Aileron (Mean Size) and Elevations (Mean Size) at 24 h compared to Baseline**
Rafaelsen et al., (1973)[218]	Randomised; DB; PC (WSD)	8 M; Range 21–29 y	Unclear	Oral Cannabis (Baked into Cake)	8 12 16	Simulated Driving	~15 h	No significant effects[p]
Rafaelsen et al., (1973)[219]	Randomised; DB; PC (WSD)	8 M; Range 21–29 y	Unclear	Oral Cannabis (Baked into Cake)	8 12 16	Digit Span Task Addition Test Subtract Serial Sevens Task Finger Labyrinths Task Bourdon's Cancellation	~15 h	No significant effects[p]
Kielholz et al., (1973)[220]	DB; PC (BSD)	54[q] (Sex NS); 34 y	Unclear	THC Capsules	350 µg·kg^{-} (~24.5 mg[r]) 400 µg·kg (~28 mg[r]) 450 µg·kg (~31.5 mg[r])	Tapping Task Spiral Rotor Task The Compensation Apparatus The Tracking Apparatus	17.5 h	Results not adequately reported

BSD: Between Subject Design; C: Control Group; CBD: Cannabidiol; CBN: Cannabinol; CPT: Continuous Performance Test; DB: Double Blind; DH: Dominant Hand; DSST: Digit Symbol Substitution Test; DT: Double Target; F: frequent cannabis user; HVLT-R: Hopkins Verbal Learning Test Revised; I: Intervention Group; L: Left; M: Male Participants; N: naïve cannabis user; NS: Not Specified; O: occasional cannabis user; PC: Placebo Controlled; PVT: Psychomotor Vigilance Task; R: Right; RA: Response Accuracy; RT: Reaction Time; RVIPT: Rapid Visual Information Processing Task; SB: Single Blind; SDLP: Standard Deviation of Lane Position; ST: Single Target; WSD: Within Subject Design. Significant effects are in **bold text**.

a: Cigarettes were smoked *ad libitum*;
b: The authors modelled the 'behavioural pharmacokinetics' of THC rather than investigating its effect at specific times post-treatment; however, their modelling still suggests impairment resolves within 8-hours;
c: Though 'double-blinded', participants had to demonstrate a capacity to distinguish between THC and placebo (in a 'Quantification Phase') to be eligible for inclusion;
d: Only 35 of these participants were included in the analyses investigating THC's effects on cognitive function;
e: Only the 'minimum' and 'maximum' performance scores were presented and subjected to statistical analysis;
f: Compared to '20-minutes post-placebo' (as performance was not assessed 24-hours post-placebo);
g: It is unclear whether six or eight participants completed the cognitive function tests;
h: It is unclear how the time parameter was handled in these statistical analyses (see also Sect. 3.4 'Next Day Effects of THC');
i: The authors indicate that THC decreased pursuit speeds at 1.75-hours but do not clearly describe its effects at the other time points;
j: The authors do not state whether a single or double-blind design was used;
k: Participants completed a total of five smoking periods involving "eight puffs" each: (1) 9 PM Friday; (2) 3 PM Saturday; (3) 9 PM Saturday; (4) 3 PM Sunday; (5) 9 PM Sunday; cognitive function was assessed 12-hours after each evening (9 PM) smoking period;
l: Main effect of treatment across all three days;
m: This effect is described as 'negative' in the current paper (since any change in time production could indicate 'impairment'); however, it is worth noting that participants were closer to the target time on THC than placebo;
n: the first cigarette was administered 4 hours before the second;
o: the first two cigarettes were administered 4 hours before the second two;
p: We presume these comparisons are against placebo;
q: Total number across all four treatment groups;
r: Value estimated at a body weight of 70 kg.

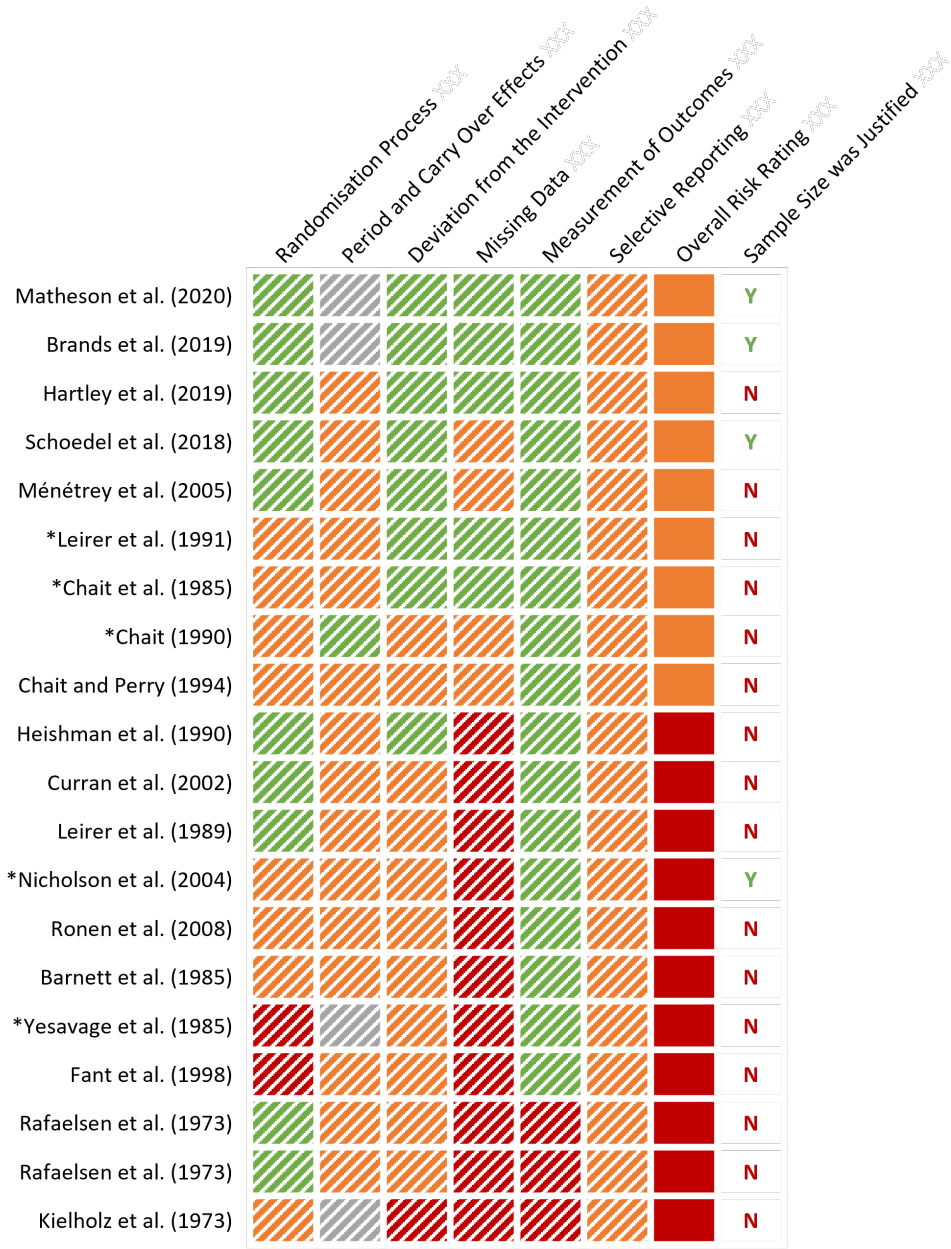

Figure 5 *Risk of bias as assessed using the Revised Cochrane Risk of Bias tool (RoB 2.0) and the RoB 2.0 for crossover trials (as appropriate). Green: Low risk of bias; Orange: Some concerns; Red: High risk of bias; Grey: Not applicable (not a crossover trial); N: No; Y: Yes. *Studies that detected significant detrimental effects of THC on 'next day' performance.*

1.11.2 No Next-Day Effects Findings

Of the 20 included studies (n=458), 16 studies found no 'next day' residual effects of THC (these studies involved a total of 180 neuropsychological tests and 29 'safety-sensitive tasks' such as simulated driving and simulated flying). Most of the neuropsychological tests and safety-sensitive tasks were conducted >12-24 h post-drug administration (82 and 17 tasks, respectively).

Most studies administered a single dose of THC (median [interquartile range]: 15 [10-20] mg), where reported. Most studies also used randomised, double-blind, placebo-controlled designs (nine studies), involved occasional cannabis users (12 studies), and administered THC via inhaled methods (i.e., smoking; 11 studies).

In terms of risk of bias, half of these 16 studies were found to have 'some concerns' (eight studies)[103 203-205 207 210-212] and, half, a 'high risk' of bias (also eight studies)[118 206 208 209 214 215 217-220] (see **Figure 5**). Two studies with lowest risk rating of all 20 studies (i.e., received 'low risk' rating on four of the five RoB domains assessed) found no 'next day' effects of THC.[204,205] Both justified their chosen sample size and employed robust standardisation procedures (i.e., methods used to control participant pre-trial and within-trial sleep behaviour and cannabis, alcohol, caffeine, and psychoactive drug use).

Some studies may have been underpowered to detect a significant effect with only three studies justifying their chosen sample size.[203 204 210] Of note, 42% of the tests showing no residual 'next day' effects of THC failed to demonstrate 'acute' impairment (i.e., < 8 h post THC use). It is therefore unlikely that *residual* THC effects would be observed in the absence of initial impairment (e.g., at lower THC doses or on tests that are relatively insensitive to the effects of THC).

1.11.3 Negative Next-Day Effects Findings

Five studies identified negative (i.e., impairing) effects of THC across 10 neuropsychological tests (N=3 studies; learning and/or memory, perception, working memory, and divided attention) conducted between >8-12 h post-treatment and two safety-sensitive tasks (N=2 studies; both simulated flying tasks) conducted 24 h post drug administration.[118 211 212 216 217] None of the five studies used randomised double-blind, placebo-controlled designs and all were published >18 years ago (four, >30 years ago). Most administered THC via smoking (four studies) and THC doses were 5, 15, 19, and 20 mg (where reported; N=3 studies only). In terms of risk of

bias, three of these five studies were found to have 'some concerns'[211 212 216] and two, a 'high risk' of bias.[118 217]

In the two studies involving safety-sensitive tasks, both administered ~20 mg THC via smoking (cannabis) and detected impairment persisting beyond 24 h.[211 217] However, these negative effects were not replicated in a third flight simulator study (using a randomised, double-blind, placebo-controlled design) conducted by the same authors.[214]

1.11.4 Positive Next-Day Effects Findings

Positive (i.e., enhancing) effects of THC on performance were observed in two randomised, double-blind, placebo-controlled trials of regular cannabis users who smoked either 70.3±21.3 or 94.0±16.4 mg THC ad libitum.[203 204] These effects were observed on the Digital Symbol Substitution Test (DSST; a test of speed of information processing) and one safety-sensitive task (simulated driving); both observed at 48 h post THC use. Both studies had 'some concerns' regarding risk of bias but received 'low risk' ratings on four of the five RoB domains assessed, as previously mentioned.

1.11.5 Summary

Overall, there appears to be limited published evidence to-date to support the assertion that THC impairs 'next day' performance. Five studies included some tests where THC worsened performance on cognitive tasks (namely, learning and/or memory, perception, working memory, and divided attention) and on two safety-sensitive tasks (i.e., simulated flying).[118 211 212 216 217] All were published >18 years ago (four, >30 years ago) and none used randomised double-blind, placebo-controlled designs. However, 16 studies showed no next-day residual effects of THC of which nine employed randomised, double-blind, placebo-controlled designs, including two studies that had the lowest risk rating of all 20 studies (i.e., received 'low risk' rating on four of the five RoB domains).[203 204] Despite this, half of the studies reporting no next-day residual THC effects had a 'high risk' of bias. Further, just under half (42%) of the tests showing no residual next day effects of THC failed to demonstrate acute impairment (i.e., THC-related impairment occurring

<8 h post-treatment). Seven studies were observed to have insufficient information provided to determine the findings resulting in 'unclear' next day effects of THC.[103 207-209 213 216 220] Further studies using improved methodologies involving occasional or cannabis-naïve medicinal cannabis users and oral THC administration are needed. Whether evening THC consumption results in residual next-day impairment in cognitive function, alertness and/or safety-sensitive tasks is a critical question that will be of growing relevance as the prescriptions for medicinal cannabis continue to increase. This will be addressed in the current thesis.

The duration of THC impairment is a critical issue given the rise in medicinal cannabis prescription in Australia and globally. While CBD is non-intoxicating and poses no restrictions around driving or operating heavy machinery while taking CBD-only products in Australia, there is currently no exemption for people with a legitimate prescription for THC (except for in Tasmania). Of note, contamination with THC, particularly in illicit (unregulated) CBD-containing products, is a major issue worldwide and requires monitoring and oversight (e.g., increased regulation for testing and provision of certificate of analysis).[221] Given the current legal framework for driving under the influence of cannabis in Australia (i.e., detection of THC in saliva with no functional assessment), point-of-collection testing (POCT) devices are a frequently used method for detection of recent cannabis use and cannabis-impaired driving. However, the accuracy and reliability of these devices have been previously criticised in previous studies of inhaled cannabis and usability limited to very recent cannabis use.[222] The implications of this for patients who need to drive (i.e., for employment, family life) and are prescribed a THC-based medicine in the evening to help them sleep is yet unknown and will be addressed in the current thesis.

1.12 Aims and Overview of Chapters

This thesis involves a series of investigations that are designed to address the key knowledge gaps and controversies around the use of cannabinoids as a treatment for sleep disorders, as outlined in this introductory chapter. The aims of these studies are as follows:

1. Systematically review and evaluate the preclinical and clinical evidence for the use of cannabinoid therapies in the treatment of a defined sleep disorder (Chapter 2).
2. Characterise individual characteristics and use patterns of medicinal cannabis for the treatment of sleep disorders in the Australian community (Chapter 3).
3. Describe a high-quality, randomised, placebo-controlled, crossover trial protocol that aims to examine the acute effects of combined CBD/THC in a clinical insomnia population (Chapter 4.1).
4. Characterise the acute effects of combined CBD/THC on objective and subjective sleep quality in a clinical insomnia population relative to placebo (Chapter 4.2).
5. Explore the effects of combined CBD/THC on average spectral power during sleep using high-density EEG (Chapter 4.2).
6. Determine the safety profile of acutely administered combined CBD/THC in a clinical insomnia population (Chapter 4.2).
7. Establish the duration of impairment produced by combined CBD/THC by assessing cognition, alertness, and simulated driving performance at multiple time points: prior to bedtime (0.5 h post drug administration), upon waking (10 h post drug administration), and throughout the next day (up until 16 h post drug administration) (Chapter 4.3).
8. Determine the plasma concentration of CBD and THC upon waking (10 h post drug administration) relative to the placebo arm (Chapter 4.2).
9. Establish whether the DW and DT5000 salivary drug test devices detect the presence of THC following oral cannabinoids at multiple time points: baseline, prior to bedtime (0.5 h

post drug administration), upon waking (10 h post drug administration), and upon departure of session (16 h post drug administration) (Chapter 4.3).

10. Characterise oral fluid THC and CBD pharmacokinetics following oral ingestion of a combined CBD/THC product (Chapter 4.3).

Thus, Chapter 2 presents a systematic review that synthesises and evaluates the preclinical and clinical evidence for the use of cannabinoid therapies in the treatment of a defined sleep disorder. Conducted in accordance with the PRISMA statement, the review aims to synthesise the extant research on cannabinoids as therapeutics for sleep in a manner that informs policy, research priorities, and clinical decision-making.

Chapter 3 presents results of a subanalysis of Australian consumers (n=1030) who self-reported using medicinal cannabis to treat a sleep disorder from the 'Cannabis as Medicine Survey' 2020-2021 (CAMS-20). This chapter aims to provides a snapshot of medicinal cannabis use characteristics, types of sleep disorders treated with medicinal cannabis, and perceived efficacy in the period following the introduction of legal medicinal cannabis in Australia in 2016. It also explores the influence of respondent characteristics, cannabis use patterns, and prescription medication use with the aim of discerning factors that may increase the likelihood of using medicinal cannabis to treat a sleep disorder.

Chapter 4.1 describes a clinical trial protocol for a randomised, placebo-controlled, crossover trial examining the acute effects of an orally administered oil consisting of combined 200 mg CBD and 10 mg THC (hereinafter referred to as 'CBD/THC') on sleep and daytime function in patients with clinician-diagnosed insomnia disorder. In this study, the acute effects of CBD/THC are examined over a 24-hour period in a controlled in-laboratory environments using polysomnography and neurobehavioural assessment including a simulated driving performance task and various cognitive tests. This protocol is the first of its kind to investigate the impact of cannabinoids on objective sleep outcomes using 256-electrode high-density EEG; a novel

technology used to comprehensively examine and localise differences in brain activation during sleep and wake periods.

Chapter 4.2 reports on the primary outcomes of the aforementioned randomised clinical trial including the effects of CBD/THC on objective sleep outcomes as measured by polysomnography and high-density EEG spectral power analysis. It also explores adverse events and next-day impairment on a clinical test of alertness, the Maintenance of Wakefulness Test.

Chapter 4.3 reports on the next-day residual effects of CBD/THC on cognitive function and simulated driving performance. It also examines the reliability and accuracy of DW5s and DT5000 oral fluid collection devices that are used for roadside drug testing in NSW and other Australian jurisdictions. It does so by comparing observed test results against LC-MS/MS quantified oral fluid THC concentrations. It describes oral fluid cannabinoid concentrations over multiple time points to answer the question: are current roadside drug tests likely to produce a positive result after use of an oral oil cannabinoid product?

Chapter 5 is a general discussion of the research that has been conducted as part of this thesis. The work presented in Chapters 2, 3 and 4.1-4.3 is synthesized and discussed in relation to the extant literature that has been reviewed here in this introductory chapter. Chapter 5 also highlights potential avenues for future research.

1.13 References

1. Russo E. Cannabis in India: ancient lore and modern medicine. Cannabinoids as Therapeutics: Springer; 2005. p 1-22.
2. Kogan M, Sexton M. Medical Cannabis: A New Old Tool for Palliative Care. *J Altern Complement Med* 2020;26(9):778-80.
3. Therapeutic Goods Administration. Access to Medicinal Cannabis Products 2019. Accessed 01 June 2020. https://www.tga.gov.au/access-medicinal-cannabis-products-1
4. Hazekamp A, Ware MA, Muller-Vahl KR, et al. The medicinal use of cannabis and cannabinoids—an international cross-sectional survey on administration forms. *J Psychoactive Drugs* 2013;45(3):199-210.
5. Lintzeris N, Driels J, Elias N, et al. Medicinal cannabis in Australia, 2016: the Cannabis as Medicine Survey (CAMS-16). *Med J Aust* 2018;209(5):211-16.
6. Pledger MJ, Martin G, Cumming J. New Zealand Health Survey 2012/13: characteristics of medicinal cannabis users. *N Z Med J (Online)* 2016;129(1433):29.
7. Ramar K, Malhotra RK, Carden KA, et al. Sleep is essential to health: an American Academy of Sleep Medicine position statement. *J Clin Sleep Med* 2021;17(10):2115-19.
8. Ojile J. National Sleep Foundation sets the standard for sleep as a vital sign of health. *Sleep Health: Journal of the National Sleep Foundation* 2017;3(4):226.
9. Van Someren EJ. Brain mechanisms of insomnia: new perspectives on causes and consequences. *Physiol Rev* 2021;101(3):995-1046.
10. Murawski B, Wade L, Plotnikoff RC, et al. A systematic review and meta-analysis of cognitive and behavioral interventions to improve sleep health in adults without sleep disorders. *Sleep Med Rev* 2018;40:160-69.
11. Ohayon M, Wickwire EM, Hirshkowitz M, et al. National Sleep Foundation's sleep quality recommendations: first report. *Sleep Health* 2017;3(1):6-19.

12. Liu Y, Wheaton AG, Chapman DP, et al. Prevalence of healthy sleep duration among adults—United States, 2014. *Morb Mortal Wkly Rep* 2016;65(6):137-41.

13. Morin CM, Benca R. Chronic insomnia. *Lancet* 2012;379(9821):1129-41.

14. The Diagnostic and Statistical Manual of Mental Disorders (5th ed.; DSM–5; American Psychiatric Association, 2013).

15. Morin CM, LeBlanc M, Daley M, et al. Epidemiology of insomnia: prevalence, self-help treatments, consultations, and determinants of help-seeking behaviors. *Sleep Med* 2006;7(2):123-30. doi: 10.1016/j.sleep.2005.08.008

16. American Academy of Sleep Medicine. International classification of sleep disorders, 3rd ed. Darien, IL: American Academy of Sleep Medicine, 2014.

17. Reynolds III CF, O'Hara R. DSM-5 sleep-wake disorders classification: overview for use in clinical practice. *Am J Psychiatry* 2013;170(10):1099-101.

18. Daley M, Morin CM, LeBlanc M, et al. The economic burden of insomnia: direct and indirect costs for individuals with insomnia syndrome, insomnia symptoms, and good sleepers. *Sleep* 2009;32(1):55-64.

19. Kyle SD, Espie CA, Morgan K. "...Not just a minor thing, it is something major, which stops you from functioning daily": quality of life and daytime functioning in insomnia. *Behav Sleep Med* 2010;8(3):123-40. doi: 10.1080/15402002.2010.487450

20. Ohayon MM. Epidemiology of insomnia: what we know and what we still need to learn. *Sleep Med Rev* 2002;6(2):97-111.

21. Morin CM, Bélanger L, LeBlanc M, et al. The natural history of insomnia: a population-based 3-year longitudinal study. *Arch Intern Med* 2009;169(5):447-53.

22. Rezaie L, Fobian AD, McCall WV, et al. Paradoxical insomnia and subjective–objective sleep discrepancy: A review. *Sleep Med Rev* 2018;40:196-202.

23. Crönlein T, Geisler P, Langguth B, et al. Polysomnography reveals unexpectedly high rates of organic sleep disorders in patients with prediagnosed primary insomnia. *Sleep Breath* 2012;16(4):1097-103.

24. Roth T, Roehrs T, Pies R. Insomnia: pathophysiology and implications for treatment. *Sleep Med Rev* 2007;11(1):71-79.

25. Levenson JC, Kay DB, Buysse DJ. The pathophysiology of insomnia. *Chest* 2015;147(4):1179-92.

26. Spielman AJ, Caruso LS, Glovinsky PB. A behavioral perspective on insomnia treatment. *Psychiatr Clin North Am* 1987;10(4):541-53.

27. Taheri S, Mignot E. The genetics of sleep disorders. *The Lancet Neurology* 2002;1(4):242-50.

28. Lind MJ, Aggen SH, Kirkpatrick RM, et al. A longitudinal twin study of insomnia symptoms in adults. *Sleep* 2015;38(9):1423-30.

29. Perlis M, Shaw PJ, Cano G, et al. Models of insomnia. Principles and practice of sleep medicine 2011;5(1):850-50.

30. Bonnet MH, Arand DL. Hyperarousal and insomnia: state of the science. *Sleep Med Rev* 2010;14(1):9-15.

31. Bonnet MH, Arand D. 24-Hour metabolic rate in insomniacs and matched normal sleepers. *Sleep* 1995;18(7):581-88.

32. Lack LC, Gradisar M, Van Someren EJ, et al. The relationship between insomnia and body temperatures. *Sleep Med Rev* 2008;12(4):307-17.

33. Farina B, Dittoni S, Colicchio S, et al. Heart rate and heart rate variability modification in chronic insomnia patients. *Behav Sleep Med* 2014;12(4):290-306.

34. Fernandez-Mendoza J, Li Y, Vgontzas AN, et al. Insomnia is associated with cortical hyperarousal as early as adolescence. *Sleep* 2016;39(5):1029-36.

35. Baglioni C, Regen W, Teghen A, et al. Sleep changes in the disorder of insomnia: a meta-analysis of polysomnographic studies. *Sleep Med Rev* 2014;18(3):195-213.

36. Perlis ML, Smith MT, Andrews PJ, et al. Beta/Gamma EEG activity in patients with primary and secondary insomnia and good sleeper controls. *Sleep* 2001;24(1):110-17.

37. Spiegelhalder K, Regen W, Feige B, et al. Increased EEG sigma and beta power during NREM sleep in primary insomnia. *Biol Psychol* 2012;91(3):329-33.

38. Carney CE, Harris AL, Falco A, et al. The relation between insomnia symptoms, mood, and rumination about insomnia symptoms. *J Clin Sleep Med* 2013;9(6):567-75.

39. Lustenberger C, Huber R. High density electroencephalography in sleep research: potential, problems, future perspective. *Front Neurol* 2012;3:77.

40. Riedner BA, Goldstein MR, Plante DT, et al. Regional patterns of elevated alpha and high-frequency electroencephalographic activity during nonrapid eye movement sleep in chronic insomnia: a pilot study. *Sleep* 2016;39(4):801-12.

41. Buysse DJ, Germain A, Hall M, et al. A neurobiological model of insomnia. *Drug Discov Today: Dis Models* 2011;8(4):129-37.

42. Daley M, Morin CM, LeBlanc M, et al. Insomnia and its relationship to health-care utilization, work absenteeism, productivity and accidents. *Sleep Med* 2009;10(4):427-38.

43. Wickwire EM, Shaya FT, Scharf SM. Health economics of insomnia treatments: the return on investment for a good night's sleep. *Sleep Med Rev* 2016;30:72-82.

44. Dopheide JA. Insomnia overview: epidemiology, pathophysiology, diagnosis and monitoring, and nonpharmacologic therapy. *Am J Manag Care* 2020;26(4 Suppl):S76-S84.

45. Hillman D, Mitchell S, Streatfeild J, et al. The economic cost of inadequate sleep. *Sleep* 2018;41(8):zsy083.

46. Carroll JE, Irwin MR, Merkin SS, et al. Sleep and multisystem biological risk: a population-based study. *PloS One* 2015;10(2):e0118467.

47. Sarsour K, Morin CM, Foley K, et al. Association of insomnia severity and comorbid medical and psychiatric disorders in a health plan-based sample: insomnia severity and comorbidities. *Sleep Med* 2010;11(1):69-74.

48. Bradley TD, Floras JS. Obstructive sleep apnoea and its cardiovascular consequences. *Lancet* 2009;373(9657):82-93.

49. Tobaldini E, Fiorelli EM, Solbiati M, et al. Short sleep duration and cardiometabolic risk: from pathophysiology to clinical evidence. *Nat Rev Cardiol* 2019;16(4):213-24.

50. Wang F, Xiong X, Xu H, et al. The association between obstructive sleep apnea syndrome and metabolic syndrome: a confirmatory factor analysis. *Sleep Breath* 2019;23(3):1011-19.

51. Lian Y, Yuan Q, Wang G, et al. Association between sleep quality and metabolic syndrome: A systematic review and meta-analysis. *Psychiatry Res* 2019;274:66-74.

52. Irwin MR, Vitiello MV. Implications of sleep disturbance and inflammation for Alzheimer's disease dementia. *Lancet Neurol* 2019;18(3):296-306.

53. Xu W, Tan C-C, Zou J-J, et al. Sleep problems and risk of all-cause cognitive decline or dementia: an updated systematic review and meta-analysis. *Journal of Neurology, Neurosurgery & Psychiatry* 2020;91(3):236-44.

54. Haack M, Simpson N, Sethna N, et al. Sleep deficiency and chronic pain: potential underlying mechanisms and clinical implications. *Neuropsychopharmacology* 2020;45(1):205-16.

55. Freeman D, Sheaves B, Waite F, et al. Sleep disturbance and psychiatric disorders. *Lancet Psychiatry* 2020;7(7):628-37.

56. Riemann D. Insomnia and comorbid psychiatric disorders. *Sleep Med* 2007;8:S15-S20.

57. Hertenstein E, Feige B, Gmeiner T, et al. Insomnia as a predictor of mental disorders: a systematic review and meta-analysis. *Sleep Med Rev* 2019;43:96-105.

58. Baglioni C, Battagliese G, Feige B, et al. Insomnia as a predictor of depression: a meta-analytic evaluation of longitudinal epidemiological studies. *J Affect Disord* 2011;135(1-3):10-19.

59. Riemann D, Krone LB, Wulff K, et al. Sleep, insomnia, and depression. *Neuropsychopharmacology* 2020;45(1):74-89.

60. Lie JD, Tu KN, Shen DD, et al. Pharmacological treatment of insomnia. *Pharmacy Ther* 2015;40(11):759.

61. Curtis F, Laparidou D, Bridle C, et al. Effects of cognitive behavioural therapy on insomnia in adults with tinnitus: Systematic review and meta-analysis of randomised controlled trials. *Sleep Med Rev* 2021;56:101405.

62. Cheung JM, Bartlett DJ, Armour CL, et al. To drug or not to drug: a qualitative study of patients' decision-making processes for managing insomnia. *Behav Sleep Med* 2018;16(1):1-26.

63. Hasan F, Tu Y-K, Yang C-M, et al. Comparative efficacy of digital cognitive behavioral therapy for insomnia: A systematic review and network meta-analysis. *Sleep Med Rev* 2022;61:101567.

64. De Crescenzo F, D'Alò GL, Ostinelli EG, et al. Comparative effects of pharmacological interventions for the acute and long-term management of insomnia disorder in adults: a systematic review and network meta-analysis. *Lancet* 2022;400(10347):170-84.

65. Zammit G. Comparative tolerability of newer agents for insomnia. *Drug Safety* 2009;32(9):735-48.

66. Stranks EK, Crowe SF. The acute cognitive effects of zopiclone, zolpidem, zaleplon, and eszopiclone: a systematic review and meta-analysis. *J Clin Exp Neuropsychol* 2014;36(7):691-700.

67. O'Hanlon JF. Residual effects on memory and psychomotor performance of zaleplon and other hypnotic drugs. *Prim Care Companion J Clin Psychiatry* 2002;4(Suppl 1):38-44.

68. Verster JC, Volkerts ER, Schreuder AH, et al. Residual effects of middle-of-the-night administration of zaleplon and zolpidem on driving ability, memory functions, and psychomotor performance. *J Clin Psychopharmacol* 2002;22(6):576-83.

69. Atkin T, Comai S, Gobbi G. Drugs for insomnia beyond benzodiazepines: pharmacology, clinical applications, and discovery. *Pharmacol Rev* 2018;70(2):197-245.

70. Lintzeris N DJ, Elias N, Arnold JC, McGregor IS, Allsop DJ. Findings from the Cannabis as Medicine Survey (CAMS-16): an online survey of medical cannabis use in Australia. *Med J Aust* 2018

71. Lintzeris N, Mills L, Suraev A, et al. Medical cannabis use in the Australian community following introduction of legal access: The 2018-2019 Online Cross-Sectional Cannabis as Medicine Survey (CAMS-18). 2020 *Harm Reduct J, 17*(1), pp.1-12.

72. Lintzeris N, Mills L, Abelev SV, et al. Medical cannabis use in Australia: consumer experiences from the online cannabis as medicine survey 2020 (CAMS-20). *Harm Reduct J* 2022;19(1):1-10.

73. Cahill SP, Lunn SE, Diaz P, et al. Evaluation of patient reported safety and efficacy of cannabis from a survey of medical cannabis patients in Canada. *Front Public Health* 2021;9:626853.

74. Schlienz NJ, Scalsky R, Martin EL, et al. A cross-sectional and prospective comparison of medicinal cannabis users and controls on self-reported health. *Cannabis Cannabinoinds Res* 2021;6(6):548-58.

75. MacPhail S, Bedoya-Perez MA, Cohen R, et al. Medicinal Cannabis Prescribing in Australia: An analysis of trends over the first five years. *Front Pharmacol*:1368.

76. Russell C, Rueda S, Room R, et al. Routes of administration for cannabis use–basic prevalence and related health outcomes: A scoping review and synthesis. *Int J Drug Policy* 2018;52:87-96.

77. Spindle TR, Martin EL, Grabenauer M, et al. Assessment of cognitive and psychomotor impairment, subjective effects, and blood THC concentrations following acute administration of oral and vaporized cannabis. *J Psychopharmacol* 2021;35(7):786-803.

78. Spindle TR, Cone EJ, Schlienz NJ, et al. Acute effects of smoked and vaporized cannabis in healthy adults who infrequently use cannabis: a crossover trial. *JAMA Netw Open* 2018;1(7):e184841-e41.

79. McGregor IS, Cairns EA, Abelev S, et al. Access to cannabidiol without a prescription: A cross-country comparison and analysis. *Int J Drug Policy* 2020;85:102935.

80. Belendiuk KA, Babson KA, Vandrey R, et al. Cannabis species and cannabinoid concentration preference among sleep-disturbed medicinal cannabis users. *Addict Behav* 2015;50:178-81.

81. Bachhuber M, Arnsten JH, Wurm G. Use of cannabis to relieve pain and promote sleep by customers at an adult use dispensary. *J Psychoactive Drugs* 2019;51(5):400-04.

82. Cahill SP, Lunn SE, Diaz P, et al. Evaluation of patient reported safety and efficacy of cannabis from a survey of medical cannabis patients in Canada. *Front Public Health* 2021;9

83. Russo EB, Guy GW, Robson PJ. Cannabis, Pain, and Sleep: Lessons from Therapeutic Clinical Trials of Sativex®, a Cannabis-Based Medicine. *Chem Biodivers* 2007;4(8):1729-43.

84. Ware MA, Fitzcharles M-A, Joseph L, et al. The effects of nabilone on sleep in fibromyalgia: results of a randomized controlled trial. *Anesth Analg* 2010;110(2):604-10.

85. Morales P, Hurst DP, Reggio PH. Molecular targets of the phytocannabinoids: a complex picture. *Phytocannabinoids* 2017:103-31.

86. Baker P, Taylor B, Gough T. The tetrahydrocannabinol and tetrahydrocannabinolic acid content of cannabis products. *J Pharm Pharmacol* 1981;33(1):369-72.

87. Grotenhermen F. Pharmacokinetics and pharmacodynamics of cannabinoids. *Clin Pharmacokinet* 2003;42(4):327-60.

88. Di Marzo V, Bifulco M, De Petrocellis L. The endocannabinoid system and its therapeutic exploitation. *Nat Rev Drug Discov* 2004;3(9):771.

89. Grotenhermen F. Pharmacokinetics and pharmacodynamics of cannabinoids. *Clin Pharmacokinet* 2003;42(4):327-60.

90. Tham M, Yilmaz O, Alaverdashvili M, et al. Allosteric and orthosteric pharmacology of cannabidiol and cannabidiol-dimethylheptyl at the type 1 and type 2 cannabinoid receptors. *Br J Pharmacol* 2019;176(10):1455-69.

91. Nelson KM, Bisson J, Singh G, et al. The essential medicinal chemistry of cannabidiol (CBD). *J Med Chem* 2020;63(21):12137-55.

92. Huestis MA. Human cannabinoid pharmacokinetics. *Chemistry & biodiversity* 2007;4(8):1770.

93. Schlienz NJ, Spindle TR, Cone EJ, et al. Pharmacodynamic dose effects of oral cannabis ingestion in healthy adults who infrequently use cannabis. *Drug Alcohol Depend* 2020;211:107969.

94. Lucas CJ, Galettis P, Schneider J. The pharmacokinetics and the pharmacodynamics of cannabinoids. *Br J Clin Pharmacol* 2018;84(11):2477-82.

95. Perucca E, Bialer M. Critical aspects affecting cannabidiol oral bioavailability and metabolic elimination, and related clinical implications. *CNS Drugs* 2020;34(8):795-800.

96. Agurell S, Carlsson S, Lindgren J, et al. Interactions ofΔ 11-tetrahydrocannabinol with cannabinol and cannabidiol following oral administration in man. Assay of cannabinol and cannabidiol by mass fragmentographywith cannabinol and cannabidiol following oral administration in man. Assay of cannabinol and cannabidiol by mass fragmentography. *Experientia* 1981;37(10):1090-92.

97. Ujváry I, Grotenhermen F. 11-Nor-9-carboxy-Δ 9-tetrahydrocannabinol–a ubiquitous yet underresearched cannabinoid. A re-view of the literature. *Cannabinoids* 2014;9:1-8.

98. Ujváry I, Hanuš L. Human metabolites of cannabidiol: a review on their formation, biological activity, and relevance in therapy. *Cannabis Cannabinoid Res* 2016;1(1):90-101.

99. Goodman S, Hammond D. THC labeling on cannabis products: an experimental study of approaches for labeling THC servings on cannabis edibles. *J Cannabis Res* 2022;4(1):1-9.

100. Sholler DJ, Strickland JC, Spindle TR, et al. Sex differences in the acute effects of oral and vaporized cannabis among healthy adults. *Addict Biol* 2021;26(4):e12968.

101. Arkell TR, Kevin RC, Vinckenbosch F, et al. Sex differences in acute cannabis effects revisited: Results from two randomized, controlled trials. *Addict Biol* 2022;27(2):e13125.

102. Arout CA, Haney M, Herrmann ES, et al. A placebo-controlled investigation of the analgesic effects, abuse liability, safety and tolerability of a range of oral cannabidiol doses in healthy humans. *Br J Clin Pharmacol* 2022;88(1):347-55.

103. Schoedel KA, Szeto I, Setnik B, et al. Abuse potential assessment of cannabidiol (CBD) in recreational polydrug users: a randomized, double-blind, controlled trial. *Epilepsy Behav* 2018;88:162-71.

104. McCartney D, Suraev AS, Doohan PT, et al. Effects of cannabidiol on simulated driving and cognitive performance: A dose-ranging randomised controlled trial. *J Psychopharmacol* 2022:02698811221095356.

105. Spindle TR, Cone EJ, Goffi E, et al. Pharmacodynamic effects of vaporized and oral cannabidiol (CBD) and vaporized CBD-dominant cannabis in infrequent cannabis users. . *Drug Alcohol Depend* 2020;In Press

106. Chesney E, Oliver D, Green A, et al. Adverse effects of cannabidiol: a systematic review and meta-analysis of randomized clinical trials. *Neuropsychopharmacology* 2020;45(11):1799-806.

107. Geffrey AL, Pollack SF, Bruno PL, et al. Drug–drug interaction between clobazam and cannabidiol in children with refractory epilepsy. *Epilepsia* 2015;56(8):1246-51.

108. Gaston TE, Bebin EM, Cutter GR, et al. Interactions between cannabidiol and commonly used antiepileptic drugs. *Epilepsia* 2017;58(9):1586-92.

109. Taylor L, Gidal B, Blakey G, et al. A phase I, randomized, double-blind, placebo-controlled, single ascending dose, multiple dose, and food effect trial of the safety, tolerability and pharmacokinetics of highly purified cannabidiol in healthy subjects. *CNS Drugs* 2018;32(11):1053-67.

110. Shannon S, Lewis N, Lee H, et al. Cannabidiol in anxiety and sleep: a large case series. *Perm J* 2019;23

111. Skelley JW, Deas CM, Curren Z, et al. Use of cannabidiol in anxiety and anxiety-related disorders. *J Am Pharm Assoc* 2020;60(1):253-61.

112. Berger M, Li E, Rice S, et al. Cannabidiol for Treatment-Resistant Anxiety Disorders in Young People: An Open-Label Trial. *J Clin Psychiatry* 2022;83(5):42111.

113. Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, et al. The nonpsychoactive Cannabis constituent cannabidiol is a wake-inducing agent. *Behav Neurosci* 2008;122(6):1378.

114. Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, et al. Cannabidiol Partially Blocks the Excessive Sleepiness in Hypocretin-deficient Rats: Preliminary Data. *CNS Neurol Disord Drug Targets* 2019;18(9):705-12.

115. Arnold JC, Nation T, McGregor IS. Prescribing medicinal cannabis. *Aust Prescr* 2020;43(5):152.

116. Boggs DL, Nguyen JD, Morgenson D, et al. Clinical and preclinical evidence for functional interactions of cannabidiol and Δ9-tetrahydrocannabinol. *Neuropsychopharmacology* 2018;43(1):142-54.

117. Hindocha C, Freeman TP, Schafer G, et al. Acute effects of delta-9-tetrahydrocannabinol, cannabidiol and their combination on facial emotion recognition: a randomised, double-blind, placebo-controlled study in cannabis users. *Eur Neuropsychopharmacol* 2015;25(3):325-34.

118. Nicholson AN, Turner C, Stone BM, et al. Effect of Δ-9-tetrahydrocannabinol and cannabidiol on nocturnal sleep and early-morning behavior in young adults. *J Clin Psychopharmacol* 2004;24(3):305-13.

119. Morgan CJ, Freeman TP, Schafer GL, et al. Cannabidiol attenuates the appetitive effects of Δ 9-tetrahydrocannabinol in humans smoking their chosen cannabis. *Neuropsychopharmacology* 2010;35(9):1879.

120. Bhattacharyya S, Morrison PD, Fusar-Poli P, et al. Opposite effects of Δ-9-tetrahydrocannabinol and cannabidiol on human brain function and psychopathology. *Neuropsychopharmacology* 2010;35(3):764.

121. Schubart CD, Sommer IE, van Gastel WA, et al. Cannabis with high cannabidiol content is associated with fewer psychotic experiences. *Schizophr Res* 2011;130(1-3):216-21.

122. Russo E, Guy GW. A tale of two cannabinoids: the therapeutic rationale for combining tetrahydrocannabinol and cannabidiol. *Med Hypotheses* 2006;66(2):234-46.

123. Alessandria G, Meli R, Infante MT, et al. Long-term assessment of the cognitive effects of nabiximols in patients with multiple sclerosis: a pilot study. *Clin Neurol Neurosurg* 2020;196:105990.

124. Russo M, De Luca R, Torrisi M, et al. Should we care about sativex-induced neurobehavioral effects? A 6-month follow-up study. *Age (y)* 2016;42(8.9):43-9.

125. Aragona M, Onesti E, Tomassini V, et al. Psychopathological and cognitive effects of therapeutic cannabinoids in multiple sclerosis: a double-blind, placebo controlled, crossover study. *Clin Neuropharmacol* 2009;32(1):41-47.

126. Etges T, Karolia K, Grint T, et al. An observational postmarketing safety registry of patients in the UK, Germany, and Switzerland who have been prescribed Sativex®(THC: CBD, nabiximols) oromucosal spray. *Ther Clin Risk Manag* 2016;12:1667.

127. Gibson LP, Mueller RL, Winiger EA, et al. Cannabinoid Exposure and Subjective Effects of THC and CBD in Edible Cannabis Products. *Cannabis Cannabinoid Res* 2022

128. Dalton WS, Martz R, Lemberger L, et al. Influence of cannabidiol on delta-9-tetrahydrocannabinol effects. *ClinPharm Ther* 1976;19(3):300-09.

129. Ilan A, Gevins A, Coleman M, et al. Neurophysiological and subjective profile of marijuana with varying concentrations of cannabinoids. *Behav Pharmacol* 2005;16(5-6):487-96.

130. Haney M, Malcolm RJ, Babalonis S, et al. Oral cannabidiol does not alter the subjective, reinforcing or cardiovascular effects of smoked cannabis. *Neuropsychopharmacology* 2016;41(8):1974-82.

131. Arkell TR, Lintzeris N, Kevin RC, et al. Cannabidiol (CBD) content in vaporized cannabis does not prevent tetrahydrocannabinol (THC)-induced impairment of driving and cognition. *Psychopharmacology* 2019:1-12.

132. Arkell TR, Vinckenbosch F, Kevin RC, et al. Effect of cannabidiol and Δ9-tetrahydrocannabinol on driving performance: a randomized clinical trial. *JAMA* 2020;324(21):2177-86.

133. Englund A, Oliver D, Chesney E, et al. Does cannabidiol make cannabis safer? A randomised, double-blind, cross-over trial of cannabis with four different CBD: THC ratios. *Neuropsychopharmacology* 2022:1-8.

134. Di Marzo V, Bifulco M, De Petrocellis L. The endocannabinoid system and its therapeutic exploitation. *Nat Rev Drug Discov* 2004;3(9):771-84.

135. Pava MJ, Makriyannis A, Lovinger DM. Endocannabinoid signaling regulates sleep stability. *PLoS One* 2016;11(3):e0152473.

136. Hanlon EC, Tasali E, Leproult R, et al. Sleep restriction enhances the daily rhythm of circulating levels of endocannabinoid 2-arachidonoylglycerol. *Sleep* 2016;39(3):653-64.

137. Vaughn LK, Denning G, Stuhr KL, et al. Endocannabinoid signalling: has it got rhythm? *Br J Pharmacol* 2010;160(3):530-43.

138. Hanlon EC. Impact of circadian rhythmicity and sleep restriction on circulating endocannabinoid (eCB) N-arachidonoylethanolamine (anandamide). *Psychoneuroendocrinology* 2020;111:104471.

139. Hanlon EC, Tasali E, Leproult R, et al. Circadian rhythm of circulating levels of the endocannabinoid 2-arachidonoylglycerol. *J Clin Endocr Metab* 2015;100(1):220-26.

140. Wyatt RM, Fraser I, Welty N, et al. Pharmacologic Characterization of JNJ-42226314, [1-(4-Fluorophenyl)indol-5-yl]-[3-[4-(thiazole-2-carbonyl)piperazin-1-yl]azetidin -1-yl]methanone, a Reversible, Selective, and Potent Monoacylglycerol Lipase Inhibitor. *J Pharmacol Exp Ther* 2020;372(3):339-53.

141. D'Souza DC, Cortes-Briones J, Creatura G, et al. Efficacy and safety of a fatty acid amide hydrolase inhibitor (PF-04457845) in the treatment of cannabis withdrawal and dependence in men: a double-blind, placebo-controlled, parallel group, phase 2a single-site randomised controlled trial. *Lancet Psychiatry* 2019;6(1):35-45.

142. Murillo-Rodriguez E, Blanco-Centurion C, Sanchez C, et al. Anandamide enhances extracellular levels of adenosine and induces sleep: an in vivo microdialysis study. *Sleep* 2003;26(8):943-47.

143. Mechoulam R, Fride E, Hanu L, et al. Anandamide may mediate sleep induction. *Nature* 1997;389(6646):25-26.

144. Murillo-Rodrıguez E, Sanchez-Alavez M, Navarro L, et al. Anandamide modulates sleep and memory in rats. *Brain Res* 1998;812(1-2):270-74.

145. Santucci V, Storme J-j, Soubrié P, et al. Arousal-enhancing properties of the CB1 cannabinoid receptor antagonist SR 141716A in rats as assessed by electroencephalographic spectral and sleep-waking cycle analysis. *Life Sci* 1996;58(6):PL103-PL10.

146. Murillo-Rodríguez E, Cabeza R, Méndez-Díaz M, et al. Anandamide-induced sleep is blocked by SR141716A, a CB1 receptor antagonist and by U73122, a phospholipase C inhibitor. *Neuroreport* 2001;12(10):2131-36.

147. Després J-P, Golay A, Sjöström L. Effects of rimonabant on metabolic risk factors in overweight patients with dyslipidemia. *N Engl J Med* 2005;353(20):2121-34.

148. Pi-Sunyer FX, Aronne LJ, Heshmati HM, et al. Effect of rimonabant, a cannabinoid-1 receptor blocker, on weight and cardiometabolic risk factors in overweight or obese patients: RIO-North America: a randomized controlled trial. *JAMA* 2006;295(7):761-75.

149. Scheen AJ, Finer N, Hollander P, et al. Efficacy and tolerability of rimonabant in overweight or obese patients with type 2 diabetes: a randomised controlled study. *Lancet* 2006;368(9548):1660-72.

150. Van Gaal LF, Rissanen AM, Scheen AJ, et al. Effects of the cannabinoid-1 receptor blocker rimonabant on weight reduction and cardiovascular risk factors in overweight patients: 1-year experience from the RIO-Europe study. *Lancet* 2005;365(9468):1389-97.

151. Murillo-Rodríguez E. The role of the CB1 receptor in the regulation of sleep. *Prog Neuro-Psychopharmacol Biol Psychiatry* 2008;32(6):1420-27.

152. Laprairie R, Bagher A, Kelly M, et al. Cannabidiol is a negative allosteric modulator of the cannabinoid CB1 receptor. *Br J Pharmacol* 2015;172(20):4790-805.

153. Todd S, Arnold J. Neural correlates of interactions between cannabidiol and Δ9-tetrahydrocannabinol in mice: implications for medical cannabis. *Br J Pharmacol* 2016;173(1):53-65.

154. Bakas T, Van Nieuwenhuijzen P, Devenish S, et al. The direct actions of cannabidiol and 2-arachidonoyl glycerol at GABAA receptors. *Pharmacol Res* 2017;119:358-70.

155. McPartland JM, Duncan M, Di Marzo V, et al. Are cannabidiol and Δ9-tetrahydrocannabivarin negative modulators of the endocannabinoid system? A systematic review. *Br J Pharmacol* 2015;172(3):737-53.

156. Deutsch DG. A personal retrospective: elevating anandamide (AEA) by targeting fatty acid amide hydrolase (FAAH) and the fatty acid binding proteins (FABPs). *Front Pharmacol* 2016;7:370.

157. Elmes MW, Kaczocha M, Berger WT, et al. Fatty acid-binding proteins (FABPs) are intracellular carriers for Δ9-tetrahydrocannabinol (THC) and cannabidiol (CBD). *J Biol Chem* 2015;290(14):8711-21.

158. Harding K, Feldman M. Sleep disorders and sleep deprivation: An unmet public health problem. *J Am Acad Child Adolesc Psychiatry* 2008;4(47):473-74.

159. Amzica F, Steriade M. The functional significance of K-complexes. *Sleep Med Rev* 2002;6(2):139-49.

160. Chokroverty S. Overview of normal sleep. Sleep disorders medicine: Springer 2017:5-27.

161. Djonlagic I, Mariani S, Fitzpatrick AL, et al. Macro and micro sleep architecture and cognitive performance in older adults. *Nat Hum Behav* 2021;5(1):123-45.

162. Hutka P, Krivosova M, Muchova Z, et al. Association of sleep architecture and physiology with depressive disorder and antidepressants treatment. *Int J Mol Sci* 2021;22(3):1333.

163. Zhang Y, Ren R, Sanford LD, et al. Sleep in Parkinson's disease: a systematic review and meta-analysis of polysomnographic findings. *Sleep Med Rev* 2020;51:101281.

164. Walther S, Schüpbach B, Seifritz E, et al. Randomized, controlled crossover trial of dronabinol, 2.5 mg, for agitation in 2 patients with dementia. *J Clin Psychopharmacol* 2011;31(2):256-58.

165. Walsh JH, Maddison KJ, Rankin T, et al. Treating insomnia symptoms with medicinal cannabis: a randomized, crossover trial of the efficacy of a cannabinoid medicine compared with placebo. *Sleep* 2021;44(11):zsab149.

166. Linares IM, Guimaraes FS, Eckeli A, et al. No acute effects of cannabidiol on the sleep-wake cycle of healthy subjects: a randomized, double-blind, placebo-controlled, crossover study. *Front Pharmacol* 2018;9:315.

167. Carley DW, Prasad B, Reid KJ, et al. Pharmacotherapy of apnea by cannabimimetic enhancement, the PACE clinical trial: effects of dronabinol in obstructive sleep apnea. *Sleep* 2018;41(1)

168. Zalai D., Chung SA., Hussain, N., e al. Does cannabinoid really improve sleep? Testing the sleep effects of nabilone in chronic pain patients: a placebo-controlled, randomized, pilot study. *Psychother Psychosom*; 2015

169. Farabi SS, Prasad B, Quinn L, et al. Impact of dronabinol on quantitative electroencephalogram (qEEG) measures of sleep in obstructive sleep apnea syndrome. *J Clin Sleep Med* 2014;10(1):49-56.

170. Prasad B, Radulovacki MG, Carley DW. Proof of concept trial of dronabinol in obstructive sleep apnea. *Front Psychiatry* 2013;4:1.

171. Haney M, Gunderson EW, Rabkin J, et al. Dronabinol and marijuana in HIV-positive marijuana smokers: caloric intake, mood, and sleep. *J Acquir Immune Defic Syndr* 2007;45(5):545-54.

172. Bedi G, Foltin RW, Gunderson EW, et al. Efficacy and tolerability of high-dose dronabinol maintenance in HIV-positive marijuana smokers: a controlled laboratory study. *Psychopharmacology* 2010;212(4):675-86.

173. Feinberg I, Jones R, Walker JM, et al. Effects of high dosage delta-9-tetrahydrocannabinol on sleep patterns in man. *Clin Pharm Therap* 1975;17(4):458-66.

174. Feinberg I, Jones R, Walker J, et al. Effects of marijuana extract and tetrahydrocannabinol on electroencephalographic sleep patterns. *Clin Pharm Therap* 1976;19(6):782-94.

175. Tassinari C, Ambrosetto G, Peraita-Adrado M, et al. The Neuropsychiatric Syndrome of Δ 9-Tetrahydrocannabinol and Cannabis Intoxication in Naive Subjects. Marihuana and medicine: Springer 1999:649-64.

176. Pivik R, Zarcone V, Dement W, et al. Delta-9-tetrahydrocannabinol and synhexl: Effects on human sleep patterns. *Clin Pharm Therap* 1972;13(3):426-35.

177. Freemon FR. Effects of marihuana on sleeping states. *JAMA* 1972;220(10):1364-65.

178. Barratt ES, Beaver W, White R. The effects of marijuana on human sleep patterns. *Biol Psychiatry* 1974; *8*(1), 47–54.

179. Freemon FR. The effect of chronically administered delta-9-tetrahydrocannabinol upon the polygraphically monitored sleep of normal volunteers. *Drug Alcohol Depend* 1982;10(4):345-53.

180. Hosko M, Kochar M, Wang R. Effects of orally administered delta-9-tetrahydrocannabinol in man. *Clin Pharm Therap* 1973;14(3):344-52.

181. Pranikoff K, Karacan I, Larson E, et al. Effects of marijuana smoking on the sleep EEG. Preliminary studies. *J Fla Med Assoc* 1973;60(3):28-31.

182. Karacan I, Fernández-Salas A, Coggins W, et al. Sleep electroencephalographic-electrooculographic characteristics of chronic marijuana users: Part I. *Ann N Y Acad Sci* 1976;282(1):348-74.

183. World Health Organisation. WHO Expert Committee on Drug Dependence: Critical Review -Delta-9-tetrahydrocannabinol 2018. Accessed 25 Nov 2019. https://www.who.int/medicines/access/controlled-substances/THCv1.pdf?ua=1

184. Gates P, Albertella L, Copeland J. Cannabis withdrawal and sleep: A systematic review of human studies. *Subst Abus* 2016;37(1):255-69.

185. Vandrey R, Smith MT, McCann UD, et al. Sleep disturbance and the effects of extended-release zolpidem during cannabis withdrawal. *Drug Alcoh Depend* 2011;117(1):38-44.

186. Babson KA, Boden MT, Harris AH, et al. Poor sleep quality as a risk factor for lapse following a cannabis quit attempt. *J Subst Abuse Treat* 2013;44(4):438-43.

187. Budney AJ, Vandrey RG, Hughes JR, et al. Oral delta-9-tetrahydrocannabinol suppresses cannabis withdrawal symptoms. *Drug Alcoh Depend* 2007;86(1):22-29.

188. Vandrey R, Stitzer ML, Mintzer MZ, et al. The dose effects of short-term dronabinol (oral THC) maintenance in daily cannabis users. *Drug Alcoh Depend e* 2013;128(1-2):64-70.

189. Angarita GA, Emadi N, Hodges S, et al. Sleep abnormalities associated with alcohol, cannabis, cocaine, and opiate use: a comprehensive review. *Addict Sci Clin Pract* 2016;11(1):1-17.

190. Allsop D, Norberg MM, Copeland J, et al. The cannabis withdrawal scale development: patterns and predictors of cannabis withdrawal and distress. *Drug Alcoh Depend* 2011;119:123-29.

191. Bolla KI, Lesage SR, Gamaldo CE, et al. Polysomnogram changes in marijuana users who report sleep disturbances during prior abstinence. *Sleep Med* 2010;11(9):882-89.

192. Budney AJ, Moore BA, Vandrey RG, et al. The time course and significance of cannabis withdrawal. *J Abnorm Psychol* 2003;112(3):393.

193. Robson P. Abuse potential and psychoactive effects of δ-9-tetrahydrocannabinol and cannabidiol oromucosal spray (Sativex), a new cannabinoid medicine. *Expert Opin Drug Saf* 2011;10(5):675-85.

194. Wade DT, Makela P, House H, et al. Long-term use of a cannabis-based medicine in the treatment of spasticity and other symptoms in multiple sclerosis. *Mult Scler* 2006;12(5):639-45.

195. Notcutt W, Langford R, Davies P, et al. Results of a randomised withdrawal study of subjects with spasticity due to multiple sclerosis who were receiving long-term Sativex. *Mult Scler* 2009. 18(2):219-28.

196. Taylor L, Crockett J, Tayo B, et al. Abrupt withdrawal of cannabidiol (CBD): a randomized trial. *Epilepsy Behav* 2020;104:106938.

197. Verster JC, Veldhuijzen DS, Volkerts ER. Residual effects of sleep medication on driving ability. *Sleep Med Rev* 2004;8(4):309-25.

198. McCartney D, Arkell TR, Irwin C, et al. Determining the magnitude and duration of acute Δ9-tetrahydrocannabinol (Δ9-THC)-induced driving and cognitive impairment: a systematic and meta-analytic review. *Neurosci Biobehav Rev* 2021;126:175-93.

199. Els C, Jackson T, Aidoo H, et al. Position statement on the implications of cannabis use for safety-sensitive work: Winnipeg, MB: Occupational and Environmental Medical Association of Canada, 2018.

200. Beckson M, Hagtvedt R, Els C. Cannabis use before safety-sensitive work: what delay is prudent? *Neurosci Biobehav Rev* 2022;133:104488.

201. Higgins, J., et al., *Chapter 8: Assessing risk of bias in a randomized trial*, in *Cochrane Handbook for Systematic Reviews of Interventions Version 6.2 (updated February 2021)*, J. Higgins, et al., Editors. 2021, Cochran.

202. Higgins J, Eldridge, and T. Li, *Chapter 23: Including variants on randomized trials*, in *Cochrane Handbook for Systematic Reviews of Interventions Version 6.2 (updated February 2021)*, J. Higgins, et al., Editors. 2021, Cochran.

203. Matheson J, Mann RE, Sproule B, et al. Acute and residual mood and cognitive performance of young adults following smoked cannabis. *Pharmacol Biochem Behav* 2020;194:172937.

204. Brands B, Mann RE, Wickens CM, et al. Acute and residual effects of smoked cannabis: Impact on driving speed and lateral control, heart rate, and self-reported drug effects. *Drug Alcohol Depend* 2019;205:107641.

205. Hartley S, Simon N, Larabi A, et al. Effect of smoked cannabis on vigilance and accident risk using simulated driving in occasional and chronic users and the pharmacokinetic–pharmacodynamic relationship. *Clin Chem* 2019;65(5):684-93.

206. Ronen A, Gershon P, Drobiner H, et al. Effects of THC on driving performance, physiological state and subjective feelings relative to alcohol. *Accid Anal Prev* 2008;40(3):926-34.

207. Ménétrey A, Augsburger M, Favrat B, et al. Assessment of driving capability through the use of clinical and psychomotor tests in relation to blood cannabinoids levels following oral administration of 20 mg dronabinol or of a cannabis decoction made with 20 or 60 mg Δ9-THC. *J Anal Toxicol* 2005;29(5):327-38.

208. Curran VH, Brignell C, Fletcher S, et al. Cognitive and subjective dose-response effects of acute oral Δ9-tetrahydrocannabinol (THC) in infrequent cannabis users. *Psychopharmacology* 2002;164(1):61-70.

209. Fant RV, Heishman SJ, Bunker EB, et al. Acute and residual effects of marijuana in humans. *Pharmacol Biochem Behav* 1998;60(4):777-84.

210. Chait L, Perry J. Acute and residual effects of alcohol and marijuana, alone and in combination, on mood and performance. *Psychopharmacology* 1994;115(3):340-49.

211. Leirer VO, Yesavage JA, Morrow DG. Marijuana carry-over effects on aircraft pilot performance. *Aviat Space Environ Med* 1991; 62(3): 221-7.

212. Chait L. Subjective and behavioral effects of marijuana the morning after smoking. *Psychopharmacology* 1990;100(3):328-33.

213. Heishman SJ, Huestis MA, Henningfield JE, et al. Acute and residual effects of marijuana: profiles of plasma THC levels, physiological, subjective, and performance measures. *Pharmacol Biochem Behav* 1990;37(3):561-65.

214. Leirer V, Yesavage J, Morrow D. Marijuana, aging, and task difficulty effects on pilot performance. *Aviat Space Environ Med* 1989;60(12):1145-52.

215. Barnett G, Licko V, Thompson T. Behavioral pharmacokinetics of marijuana. *Psychopharmacology* 1985;85(1):51-56.

216. Chait L, Fischman MW, Schuster CR. 'Hangover'effects the morning after marijuana smoking. *Drug Alcohol Depend* 1985;15(3):229-38.

217. Yesavage JA, Leirer VO, Denari M, et al. Carry-over effects of marijuana intoxication on aircraft pilot performance: a preliminary report. *Am J Psychiatry* 1985;142(11):1325-9.

218. Rafaelsen OJ, Bech P, Christiansen J, et al. Cannabis and alcohol: effects on simulated car driving. *Science* 1973;179(4076):920-23.

219. Rafaelsen L, Christrup H, Bech P, et al. Effects of cannabis and alcohol on psychological tests. *Nature* 1973;242(5393):117-18.

220. Kielholz P, Hobi V, Ladewig D, et al. An experimental investigation about the effect of cannabis on car driving behaviour. *Pharmacopsychiatry* 1973;6(01):91-103.

221. Johnson E, Kilgore M, Babalonis S. Label accuracy of unregulated cannabidiol (CBD) products: measured concentration vs. label claim. *J Cannabis Res* 2022;4(1):1-7.

222. Arkell TR, Kevin RC, Stuart J, et al. Detection of Δ9 THC in oral fluid following vaporized cannabis with varied cannabidiol (CBD) content: An evaluation of two point-of-collection testing devices. *Drug Test Anal* 2019;11(10):1486-97.

2. Cannabinoid Therapies in the Management of Sleep Disorders: A Systematic Review of Preclinical and Clinical Studies

Sleep Medicine Reviews 53 (2020) 101339

Contents lists available at ScienceDirect

Sleep Medicine Reviews

journal homepage: www.elsevier.com/locate/smrv

CLINICAL REVIEW

Cannabinoid therapies in the management of sleep disorders: A systematic review of preclinical and clinical studies

Anastasia S. Suraev [a, b, c, d], Nathaniel S. Marshall [b, e], Ryan Vandrey [f], Danielle McCartney [a, c, d], Melissa J. Benson [a, c, d], Iain S. McGregor [a, c, d], Ronald R. Grunstein [b, g], Camilla M. Hoyos [a, b, d, *]

[a] The University of Sydney, Faculty of Science, School of Psychology, Sydney, New South Wales, Australia
[b] Centre for Sleep and Chronobiology, Woolcock Institute of Medical Research, University of Sydney, Camperdown, New South Wales, Australia
[c] The University of Sydney, Lambert Initiative for Cannabinoid Therapeutics, Sydney, New South Wales, Australia
[d] The University of Sydney, Brain and Mind Centre, Sydney, New South Wales, Australia
[e] The University of Sydney, Faculty of Medicine and Health, Susan Wakil School of Nursing and Midwifery, Sydney, New South Wales, Australia
[f] Johns Hopkins University, School of Medicine, Baltimore, MD, USA
[g] RPA-Charles Perkins Centre, Royal Prince Alfred Hospital, Sydney, New South Wales, Australia

ARTICLE INFO

Article history:
Received 16 January 2020
Received in revised form
13 April 2020
Accepted 14 April 2020
Available online 16 May 2020

Keywords:
Sleep disorders
Insomnia
Obstructive sleep apnea
Cannabinoids
Cannabis
THC
Cannabidiol

SUMMARY

Cannabinoids, including the two main phytocannabinoids Δ^9-tetrahydrocannabinol (THC) and cannabidiol (CBD), are being increasingly utilised as pharmacological interventions for sleep disorders. THC and CBD are known to interact with the endocannabinoid and other neurochemical systems to influence anxiety, mood, autonomic function, and circadian sleep/wake cycle. However, their therapeutic efficacy and safety as treatments for sleep disorders are unclear. The current systematic review assessed the available evidence base using PubMed, Scopus, Web of Science, Embase, CINAHL and PsycInfo databases. A total of 14 preclinical studies and 12 clinical studies met inclusion criteria. Results indicated that there is insufficient evidence to support routine clinical use of cannabinoid therapies for the treatment of any sleep disorder given the lack of published research and the moderate-to-high risk of bias identified within the majority of preclinical and clinical studies completed to-date. Promising preliminary evidence provides the rationale for future randomised controlled trials of cannabinoid therapies in individuals with sleep apnea, insomnia, post-traumatic stress disorder-related nightmares, restless legs syndrome, rapid eye movement sleep behaviour disorder, and narcolepsy. There is a clear need for further investigations on the safety and efficacy of cannabinoid therapies for treating sleep disorders using larger, rigorously controlled, longer-term trials.

© 2020 Elsevier Ltd. All rights reserved.

Introduction

Sleep is a vital physiological process that plays an important role in restorative functions that are essential for normal daytime function [1]. Optimal sleep health involves multiple factors, including adequate duration, timing, efficiency, and a sense of having restorative sleep that leaves the individual feeling alert and functional throughout the day [2]. Inadequate sleep is reported in approximately 30–35% of the general population [3], which may be partly due to lifestyle choices, employment, or other demands, and partly attributable to untreated sleep disorders [4]. Sleep disorders such as insomnia and obstructive sleep apnea (OSA) are associated with an increased risk of depression [5,6], cardiovascular disease [7,8], and dementia [9,10]. The direct and indirect financial costs of sleep disorders, such as those attributable to health care, lost productivity and road traffic accidents, are substantial. Annual costs arising from chronic insomnia disorder are estimated at approximately $30 - $107 billion in the USA [11]; indicating a strong need for clinical intervention.

Abbreviations: AHI, apnea hypopnea index; CBD, cannabidiol; CB_1, cannabinoid receptor 1; CB_2, cannabinoid receptor 2; EEG, electroencephalography; OSA, obstructive sleep apnea; PSG, polysomnography; PTSD, post-traumatic stress disorder; RCT, randomised controlled trial; REM, rapid eye movement; RBD, rapid eye movement sleep behaviour disorder; RLS, restless legs syndrome; THC, Δ^9-tetrahydrocannabinol.

* Corresponding author. PO Box M77, Missenden Road, Camperdown, 2050, NSW, Australia. Fax: +61 2 9114 0010.
E-mail address: camilla.hoyos@sydney.edu.au (C.M. Hoyos).

https://doi.org/10.1016/j.smrv.2020.101339
1087-0792/© 2020 Elsevier Ltd. All rights reserved.

Cannabis sativa has been used for its pain-relieving and soporific effects since ancient times [12]. Sleep disorders are one of the most common reasons individuals report using cannabis for medicinal purposes, alongside chronic pain and anxiety [13–15]. The growing legal availability of medicinal cannabis around the world is prompting an upswing of research into the effects of cannabinoids such as Δ^9-tetrahydrocannabinol (THC) and cannabidiol (CBD) as novel treatments for a variety of sleep disorders [16]. Both THC and CBD interact with the endogenous cannabinoid (endocannabinoid) system, a complex and ubiquitous neuromodulatory network that includes cannabinoid 1 (CB_1) and 2 (CB_2) receptors, the endogenous ligands for these receptors such as anandamide (AEA) and 2-arachidonoylglycerol (2-AG), and the enzymes responsible for the biosynthesis and inactivation of these ligands [17]. Our understanding of the pharmacological influence of the endocannabinoid system on the circadian sleep–wake cycle is gradually evolving [18]. Clinical and preclinical studies describe a circadian rhythm in circulating endocannabinoid concentrations [19–21], with plasma 2-AG levels increasing from mid-sleep to early afternoon in humans; an effect amplified by sleep restriction [22]. Pharmacological inhibition of monoacylglycerol lipase (MAGL), the rate-limiting enzyme responsible for the degradation of 2-AG, leads to elevated brain 2-AG concentrations and wake-promoting effects in rats, including reductions in both NREM and REM sleep [23].

In contrast to 2-AG, AEA is associated with sleep-promoting effects: increasing endogenous AEA, via pharmacological inhibition of the degradative enzyme fatty acid amide hydrolase (FAAH), normalised deficits in stage N3 (or "slow wave") sleep in cannabis-dependent males undergoing cannabis withdrawal [24]. Preclinical data similarly show that AEA promotes slow wave sleep, possibly via increases in extracellular adenosine concentrations [25–27]. In preclinical models, the sleep-promoting effects of AEA are blocked by co-administration of the CB_1 inverse-agonist, rimonabant, indicating a CB_1-specific mechanism of action for AEA on sleep [28]. In human clinical trials, insomnia and other sleep disorders were common with rimonabant treatment and occurred more frequently than placebo [29–32]. Like AEA, THC is a partial agonist at the CB_1 receptor, and, thus, may exert sleep promoting effects via this direct pharmacological action [33]. CBD, on the other hand, has a weak binding affinity for the CB_1 receptor and instead, acts predominantly as a negative allosteric modulator at CB_1 (i.e., it can reduce the potency and/or efficacy of other ligands such as THC but does not activate the receptor itself) [34]. CBD has also shown to increase AEA concentrations via FAAH inhibition [35] and also via action on fatty acid-binding proteins (FABPs) [36], which provides an alternative pharmacological mechanism by which CBD may promote sleep. Overall, this highlights a complex modulatory role for the endocannabinoid system, and potential mechanisms for THC and CBD, in regulating the sleep–wake cycle.

A recent authoritative review concluded that there was moderate evidence that exogenously administered cannabinoids (primarily nabiximols, a buccal spray containing equal parts of THC and CBD) were effective for improving short-term sleep outcomes in individuals with sleep disturbance secondary to pain conditions such as multiple sclerosis and fibromyalgia [37]. However, it remains unclear whether this is due to an improvement in sleep *per se* or an improvement in the associated underlying condition (i.e., pain). Despite increased use of medicinal cannabis to treat insomnia and other sleep disorders, the evidence supporting therapeutic utility of cannabinoid therapies in sleep disorders is unclear. This systematic review presents a synthesis and evaluation of the preclinical and clinical evidence for cannabinoid therapies for the treatment of defined sleep disorders. To extend prior reviews on this topic [38–40] using a more specific focus, both preclinical and clinical studies that involved: (a) patients with a sleep disorder (or a preclinical model of a sleep disorder); and (b) the administration of any cannabinoid in an attempt to treat or manage an underlying sleep disorder were considered in this review. The aim was to synthesise the extant research on cannabinoids as therapeutics for sleep in a manner that informs policy, research priorities, and clinical decision-making.

Methods

Search strategy and data sources

Relevant preclinical and clinical studies were identified by searching the electronic databases PubMed, Embase, PsycINFO, Scopus, Web of Science, and CINAHL from inception until the 26th June 2019 using the following Boolean expression: (*cannabis* OR *cannabinoid* OR *marijuana* OR *tetrahydrocannabinol* OR *THC* OR *cannabidiol* OR *CBD* OR *nabilone* OR *sativex* OR *nabiximols* OR *dronabinol* OR *marinol* OR *namisol*) AND (*sleep* OR *sleep disorder* OR *sleep apnea* OR *insomnia* OR *narcolepsy* OR *idiopathic hypersomnolence* OR *excessive daytime sleepiness* OR *REM sleep behaviour disorder* OR *restless legs syndrome* OR *parasomnias* OR *night terrors* OR *circadian rhythm sleep disorder* OR *shift work sleep disorder* OR *sleep phase syndrome* OR *bruxism*). The search was restricted to English-language articles only, and terms were adapted as needed to meet the specific requirements of each database. The primary literature search was undertaken by one reviewer (AS) who imported the articles into reference management software (EndNote, Clarivate Analytics, PA, USA) where duplicates were removed. Two independent reviewers (AS, DM) then systematically screened each article against the eligibility criteria, first by title and abstract, and subsequently, by full text, to identify relevant studies. Disagreements were resolved by consensus through discussion with a third independent reviewer (CMH). The search was updated in November 2019 to capture any recent publications. One reviewer (AS) also searched the reference lists of all included studies and prior major reviews for missing publications and several major clinical trial registries (ClinicalTrials.gov, The Australian New Zealand Clinical Trials Registry, and The European Union Clinical Trials Register) for ongoing or unpublished investigations. This systematic literature review was conducted in accordance with the Preferred Reported Items for Systematic Reviews and Meta-analyses (PRISMA) statement [41].

Studies were evaluated against the following inclusion criteria:

1. Presented original data (i.e., not a review).
2. Conference abstracts were excluded if data had been published in an article that was already included in the review.
3. Population: sleep disorder or self-reported symptoms of a sleep disorder.
4. Intervention: involved administration of cannabis, a cannabinoid or a modulator of the CB_1 and/or CB_2 receptors (e.g., CB_1 receptor inverse agonist such as rimonabant) at any dose, via any route of administration, in an attempt to treat or manage the underlying sleep disorder in a controlled setting.
5. Primary outcome assessed changes in sleep-related clinical outcomes via any method.
6. Research did not involve participants with a sleep disorder secondary to a primary condition (e.g., insomnia secondary to chronic pain or cannabis withdrawal syndrome) *except* if the primary outcome was measuring a sleep-related outcome OR the sleep disorder was secondary to a psychiatric condition (e.g., anxiety or post-traumatic stress disorder).
7. Research did not involve participants who were subjected to an experimental condition that modelled a sleep disorder (e.g.,

simulated night shift work or sleep deprivation in healthy volunteers).
8. Research could be either observational or interventional.

For preclinical studies, criterion (3) was adapted to include preclinical models of sleep disorders (e.g., serotonin-induced reflex apnea to model OSA in humans), but excluded models in which sleep behaviour was manipulated (e.g., REM sleep deprivation) to induce a phenotype other than a sleep disorder (e.g., aggressiveness [42]). The study characteristics, methods, and measurement of any and all sleep-related outcomes of the included studies were extracted in duplicate (AS, MJB) into a template spreadsheet.

Risk of bias

The SYRCLE tool was used to assess risk of bias in preclinical studies [43]. It comprised 10 domains covering six types of biases: sequence generation, baseline characteristics, allocation concealment, random housing, researcher blinding, random outcome assessment, outcome assessor blinding, incomplete outcome data, selective outcome reporting, and "other sources of bias". Data on the timing and/or phases of the light/dark cycle were also extracted as an additional indicator of study quality. All clinical studies were assessed for risk of bias using the revised Cochrane Risk of Bias tool (RoB 2.0) [44]. The RoB 2.0 comprises five domains, including the randomisation process, deviation from intended interventions, missing data, measurement of the outcome, selective outcome reporting, and "other sources of bias". Two independent assessors performed the risk of bias assessments for preclinical (AS and MB) and clinical (AS and NSM) studies, with any disagreement resolved by consensus.

Results

Characteristics of the included studies

The primary search identified 4342 records from 6 databases: PubMed (743), Embase (1137), PsycINFO (420), Scopus (826), Web of Science (958), CINAHL (258) (see Fig. 1). After removing duplicates, there were 1689 records for title and abstract screening. Following this, the full-texts of 16 preclinical studies and 20 clinical studies were checked for eligibility with a further 10 excluded (three preclinical and seven clinical studies - see Fig. 1 for reasons). The study characteristics and outcomes for each of the 14 preclinical studies (11 full-text articles and three abstracts) and the 12 clinical studies (10 full-text articles, one abstract, and one Letter to the Editor) are summarised in Tables 1 and 2, respectively. All clinical studies involved oral cannabinoid administration aside from one case study reporting on five individuals with severe restless legs syndrome (RLS) who smoked illicit cannabis to manage symptoms [45] – an exception for inclusion in the current review due to the specific focus of the research on a sleep disorder.

Nine preclinical studies (all conducted by the same research group) investigated the therapeutic effects of dronabinol (synthetic THC), AM251 and SR141716A (CB_1 receptor antagonists), AM630 (CB_2 receptor antagonist), chromenopyrazole 13a (CB_1 receptor agonist), and HU-308 (CB_2 receptor agonist) in a variety of animal models of OSA: four studies using acute serotonin (5-HT)-induced reflex apnea (intravenous administration of 5-HT can reduce upper airway muscle tone and increase apnea susceptibility in anesthetized rats) [46–49], four studies using adult rats as natural models of central sleep apnea [50–53], and one study using mechanical airway obstruction [54]. All nine studies used male rats. Two clinical trials used dronabinol in patients with OSA: a 3-wk open-label trial [55] and a 6-wk randomised controlled trial (RCT) [56]. Two ongoing pre-registered clinical trials in patients with OSA were also identified, with the first an open-label trial assessing the effects of 10 mg dronabinol and palmitoylethanolamide [57], and the second, an RCT investigating the effects of 10 mg THC with 200 mg of a proprietary mineral supplement [58] (see Table S1).

Four preclinical studies investigated the effects of cannabinoid treatment including CBD, oleamide, and AM251 and SR141716A in different animal models of disordered sleep, mainly stress-induced. Two studies utilised maternal separation [59,60]. One study used the 'flowerpot technique' to induce REM sleep deprivation; this method involves housing the rat on a small platform where a loss of muscle tone (e.g., during REM sleep) causes it to fall into water [61]. One study used a model of persistent stress in which the animals were repeatedly exposed to anxiety-provoking tests such as the open-field test (50 min) and a subsequent elevated plus-maze test (10 min) over four consecutive days [62]. All of these studies used male rats. No RCTs administered cannabinoids to participants with clinician-diagnosed insomnia. Six studies were identified in which cannabinoids were administered to participants with self-reported insomnia or sleep difficulties. Of those, three studies evaluated CBD formulations [63–65], two evaluated nabilone, a synthetic analogue of THC [66,67], and one evaluated a 95% pure THC product in dehydrated alcohol [68]. Four ongoing pre-registered clinical trials in chronic insomnia disorder were identified: three studies are using different ratios of THC and CBD [69–71] with one study also co-administering cannabinol (CBN) alongside THC and CBD [71], and one study administering 200 mg CBD/night [72] (see Table S1). One preclinical study administered CBD to examine effects on excessive sleepiness in an animal model of narcolepsy using hypocretin-deficient rats [73]. No clinical studies of cannabinoid treatment in patients with narcolepsy or excessive daytime sleepiness were identified. Two studies were identified assessing the effects of nabilone in males with PTSD-related nightmares [74,75]. One case series reported on the effects of CBD as an adjunct to standard treatment in four participants with REM sleep behaviour disorder (RBD) and Parkinson's disease [76]. Another case series reported on six patients with severe RLS who self-medicated with cannabis of varying composition to manage RLS symptoms [45].

Risk of bias in individual studies

The results of the risk of bias assessment for preclinical and clinical studies are reported in Tables 3 and 4, respectively. All preclinical studies, except two, exhibited high risk of bias in at least four of the domains. Ten out of 13 preclinical studies adequately reported their outcomes in an unbiased manner. No study reported using techniques of random housing, sequence generation, allocation concealment, or blinded caregivers/investigators. The timing of drug administration during the light/dark phase was only reported in three (23%) studies.

No clinical study was deemed to have an overall low risk of bias, with three studies identified as having 'some concerns' and all others a high risk of bias. The most frequent problems were bias arising from the randomisation process and selection of the reported results. Of the eight prospective studies conducted after 2005 (when trial pre-registration was mandated [77]), only three studies were pre-registered. A decision around the interpretability of the available evidence was made by categorising preclinical and clinical studies by the research question and rating them based on their quality (as per the relevant risk of bias assessment) (see Table 5).

Discussion

This review identified 12 clinical studies examining the therapeutic effects of cannabinoid therapies across a range of sleep

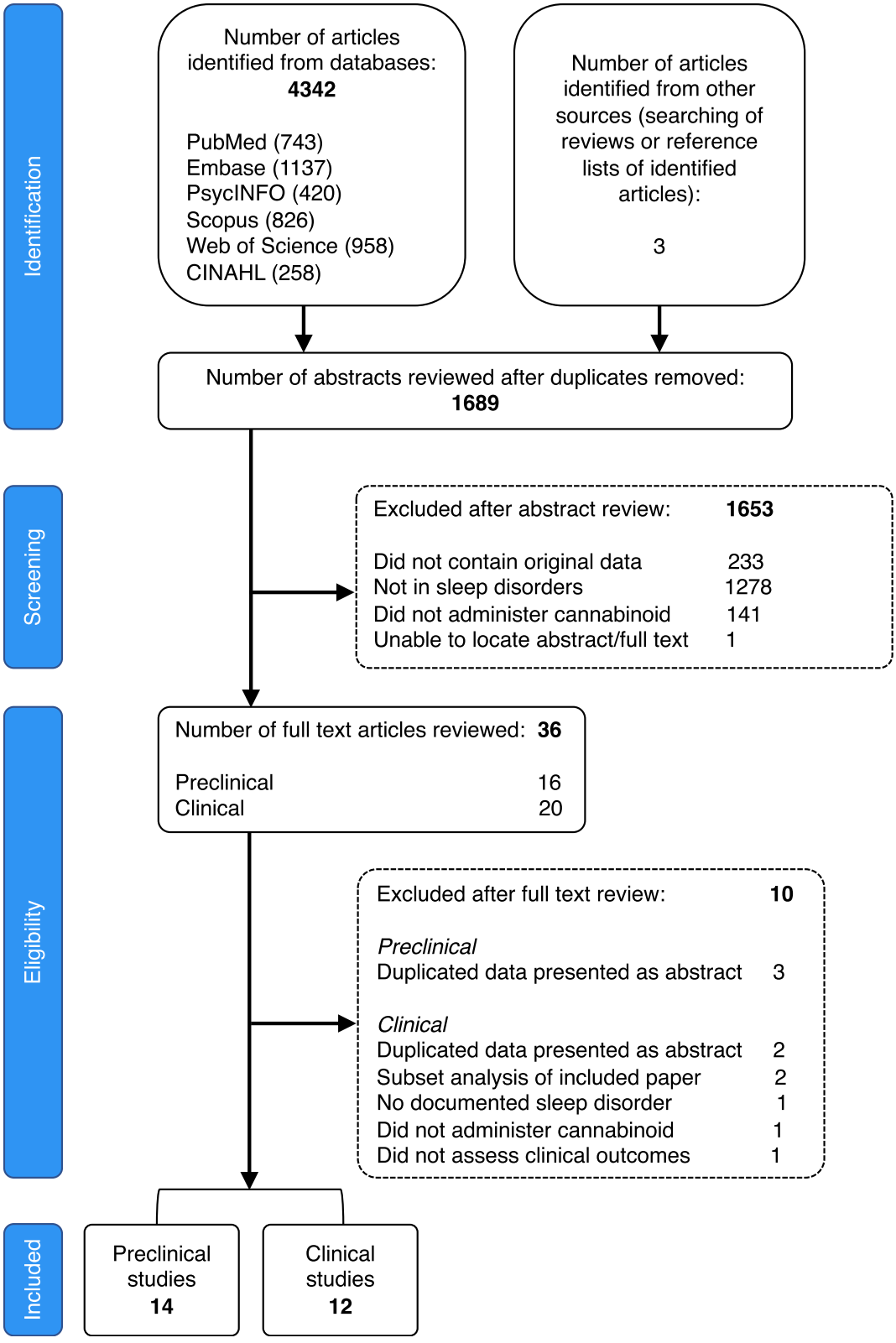

Fig. 1. PRISMA diagram illustrating the procedure used for identifying the eligibility of studies for review inclusion.

Table 1
Preclinical studies investigating the effects of cannabinoids or compounds that target the endocannabinoid system in models of sleep disorders.

Authors, year [reference number]	Country of origin	Animal species (n)	Animal model	Intervention	Measure(s)	Results
Models of obstructive sleep apnea						
Calik & Carley 2019 [53]	USA	Sprague Dawley rats ($n = 12$; all M)	Natural model of central sleep apnea	Each rat received each of the eight IP injections exactly one time in random order: I: Vehicle alone (25% DMSO; one mL) II: Dronabinol alone (10 mg/kg) III: AM251 alone (5 mg/kg) IV: AM630 alone (5 mg/kg) V: (III) + (IV) VI: (II) + (III) VII: (II) + (IV) VIII: (II) + (V)	PSG (EEG and EMG)	No significant change in sleep apnea or sleep efficiency with dronabinol (dissolved in 25% DMSO) when compared to vehicle.
Calik & Carley 2017 [50]	USA	Sprague Dawley rats ($n = 22$; all M)	Natural model of central sleep apnea	Each rat received each of the eight IP injections exactly one time in random order: I: Vehicle alone (DMSO; one mL) II: Dronabinol alone (10.0 mg/kg) III: AM251 alone (5 mg/kg) IV: AM630 alone (5 mg/kg) V: (III) + (IV) VI: (II) + (III) VII: (II) + (IV) VIII: (II) + (V)	PSG (EEG and EMG)	Compared to vehicle, dronabinol: ↓ apnea events ($p < 0.01$) ↓ post-sigh apneas ($p < 0.01$) ↓ sleep efficiency ($p < 0.05$) ↓ REM sleep ($p = 0.02$) with no changes in REM bouts or REM bout duration Pre-treatment with CB_1, but not CB_2, receptor antagonists blocked apnea suppression by dronabinol.
Calik & Carley 2016 [47]	USA	Sprague Dawley rats ($n = 30$; all M)	Acute 5-HT-induced reflex apneas	Rats (n = 6/group) were administered the following treatments via ICV injection: I-IV: Dronabinol (100, 10, 1, or 0.1μg/3μL DMSO V: Vehicle (3μLDMSO)	EMGgg and respiratory responses	No significant change in apnea durations, average breath duration, or tonic/phasic EMGgg with dronabinol (all doses) when compared to vehicle.
*Carley & Topchiy 2015 [54]	USA	Sprague Dawley rats ($n = 6$; all M)	Brief airway occlusions	Rats were injected with 1 mg/100 μL dronabinol directly into nodose ganglia	EMGgg and respiratory responses	Compared to baseline, dronabinol: ↑ phasic EMGgg ($p = 0.04$)
*Topchiy et al., 2015 [52]	USA	Sprague Dawley rats (n = not specified; gender not specified)	Natural model of central sleep apnea	Each rat received each of the seven IP injections exactly one time in random order:	EEG, nuchal EMG and respiratory responses	Compared to vehicle, chromenopyrazole 13a: ↓ apneas ($p < 0.05$) at both

(*continued on next page*)

Table 1 (*continued*)

Authors, year [reference number]	Country of origin	Animal species (*n*)	Animal model	Intervention	Measure(s)	Results
				I: Vehicle alone (undefined) II-IV: Chromenopyrazole 13a (0.1, 1 and 10 mg/kg) V-VII: HU-308 (0.1, one or 10 mg/kg)		doses HU-308 did not significantly change in apnea index at any dose.
Calik & Carley 2014 [46]	USA	Sprague Dawley rats ($n = 36$; all M)	Acute 5-HT-induced reflex apneas	Rats (n = 6/group) received the following pre-treatments via IP injection: I-II: AM251 (0.5 or 5 mg/kg) III-IV: AM630 (0.5 or 5 mg/kg) V: AM251 + AM630 (5 mg/kg each) VI: Vehicle (15% DMSO) Dronabinol (100μg/5 μL sesame oil) was injected into nodose ganglia to all groups directly followed by 5-HT infusion.	EMGgg and respiratory responses	Compared to baseline, dronabinol: ↓ apneas ($p = 0.03$) ↑ phasic and tonic EMGgg ($p < 0.05$) Both AM251 and AM630 pre-treatment reversed dronabinol's reduction in reflex apneas.
Calik et al., 2014 [48]	USA	Sprague Dawley rats ($n = 24$; all M)	Acute 5-HT-induced reflex apneas	Rats (n = 6/group) received the following treatment via direct injection into the nodose ganglia: I: Dronabinol (100μg/5μL sesame oil) II: Dronabinol (10μg/5 μL sesame oil) III: Vehicle (5 μL sesame oil) IV: Sham group (no treatment)	EMGgg and respiratory responses	Compared to baseline, dronabinol: ↓ 5-HT-induced apnea (n.s.) ↓ apnea duration at both doses ($p < 0.05$) ↑ phasic EMGgg ($p < 0.01$) No effect of dronabinol on tonic EMGgg.
*Topchiy et al., 2012 [49]	USA	Sprague Dawley rats ($n = 6$; all M)	Brief airway occlusion and acute 5-HT-induced reflex apneas	Dronabinol (dose not specified) injected directly into nodose ganglia	EMGgg and respiratory responses	Compared to baseline, dronabinol: • Reduced 5-HT-induced apneas ($p = 0.04$) • Did not reverse the effects of airway occlusion on EMGgg and EMGgg pre-activation time) (n.s.)
Carley et al., 2002 [51]	USA	Sprague Dawley rats ($n = 11$; all M)	Natural model of central sleep apnea	Each rat received each of the 12 IP injections exactly one time in random order: I-III) Vehicle (saline, DMSO, or peanut oil) IV-VI) THC alone (0.1, 1.0, or 10 mg/kg) VII-IX: OLE alone (0.1, 1.0, or 10 mg/kg) X: 5-HT alone (0.79 mg/kg) XI: THC (0.1 mg/kg) → 5-	PSG (EEG and EMG)	Compared to vehicle, Δ9THC: ↓ frequency of apneas during NREM ($p = 0.03$ for 1.0 and 10 mg/kg) ↓ frequency of apneas during REM ($p = 0.03$ for 10 mg/kg only) Compared to

				HT (0.79 mg/kg) XII: THC (0.1 mg/kg)→OLE (0.1 mg/kg)→5-HT (0.79 mg/kg)		vehicle, oleamide: ↓ frequency of apneas during NREM ($p < 0.05$ all doses)
Models of disordered sleep						
Perez-Morales et al., 2014 [59]	Mexico	Wistar rats ($n = 24$; all M)	Maternal separation	Two groups of rats (MS or no-MS, n = 6/group) received the following treatments via ICV injection into lateral hypothalamus: I: Vehicle (100% DMSO) II: AM251 (0.01 μg) III: 2-AG (0.01 μg) IV: (II) + (III)	Implanted EEG/EMG	2-AG restored sleep (i.e., ↓wakefulness and ↑NREM and REM) in MS rats ($p < 0.05$). AM251 blocked these effects ($p = 0.001$).
Prieto et al., 2012 [60]	Mexico	Wistar rats ($n = 40$; all M)	Maternal separation	Rats (n = 10/group) received the following treatments via IP injection: I: OLE (1 mg/kg) II: AM251 (1.6 mg/kg) III: (I) + (II) IV: Vehicle (30% DMSO)	Implanted EEG/EMG	Compared to vehicle, OLE normalised sleep (i.e., ↓wakefulness and ↑NREM and REM) in MS rats ($p < 0.05$).
Hsiao et al., 2012 [62]	Taiwan	Wistar rats ($n = 28$; all M)	Repeated combination tests (50 min of open field test and 10 min of EPM for four consecutive days)	Rats (n = 7/group) were subjected to either RCT or SD before receiving microinjections into the CeA with the following treatments: I: RCT + Vehicle (2% DMSO) II: RCT + CBD (0.5μg/1 μl) III: RCT + CBD (1μg/1 μl) IV: SD + Vehicle (2% DMSO)	Implanted EEG	Compared to vehicle, high dose CBD (1μg) blocked anxiety-induced REM sleep suppression during hours 4–10 of the light period ($p < 0.05$).
Navarro et al., 2003 [61]	Mexico	Wistar rats ($n = 24$; all M)	REM sleep deprivation using the 'flowerpot technique'	Rats (n = 6/group) were subjected to SD or no-SD before receiving the following ICV injections: I: SD + Vehicle (saline, 5μL) II: SD + SR141716A (3μg) III: No-SD + ICV saline (5 μL) IV: No-SD + SR141716A (3μg)	Implanted EEG/EMG	Compared to vehicle, CB_1 receptor antagonism partially prevented stress-induced REM sleep rebound ($p < 0.05$).
Model of narcolepsy						
Murillo-Rodriguez et al., 2019 [73]	Mexico	Wistar rats (n = 15, all M)	HCRT2/SAP-lesioned rats	Rats (n = 5/group) received the following treatments via IP injection: I: Control + Vehicle II: HCRT2/SAP + Vehicle III: HCRT2/SAP + CBD (5 mg/kg)	Implanted EEG	Compared to vehicle, CBD ↓excessive somnolence in hypocretin-deficient rats over a 5-h period only ($p < 0.05$).

2-AG = 2-Arachidonoylglycerol; 5-HT = serotonin; CB_1 = cannabinoid receptor 1; CB_2 = cannabinoid receptor 2; CBD = cannabidiol; CeA = central nucleus of amygdala; DMSO = dimethyl sulfoxide; EEG = electroencephalography; EMG = electromyography; EMGgg = genioglossus electromyography; EPM = elevated plus maze; HCRT2/SAP = hypocretin-2-saporin; ICV = intracerebroventricular; IP = intraperitoneal; M = male; MS = maternal separation; n.s. = not significant; OF = open field; OLE = oleamide; PSG = polysomnography; RCT = repeated combination tests (to provoke anxiety); REM = rapid eye movement; THC = tetrahydrocannabinol; SD = sleep deprivation; *Conference abstract.

For ease of reference: AM251 = CB_1 receptor antagonist; AM630 = CB_2 receptor antagonist; Chromenopyrazole 13a = CB_1 receptor agonist; HU-308 = CB_2 receptor agonist; SR141716A = CB_1 receptor antagonist.

Table 2
Clinical studies investigating the effect of cannabinoid therapies in the treatment of sleep disorders.

Authors, year [reference number]	Country of origin	Overall risk rating	Study design	Participant details (*n*, gender, mean ± SD age)	Treatment period	Intervention, dose, and timing	Primary outcome (measure)	Primary outcome result	Adverse events
Obstructive sleep apnea									
Carley et al., 2018 [56]	USA	*Some concerns*	DB, PC Parallel	$n = 73$ (52 M) I: 52.7 ± 7.7 y II: 54.7 ± seven y C: 58.8 ± 6.1 y	6 weeks	Dronabinol (oral capsules) I: 2.5 mg/day II: 10 mg/day C: Placebo capsule Self-administered 1 h before bedtime	Change in AHI relative to placebo at W6	Baseline AHI: I: 28.2 ± 12.5 II: 26.0 ± 11.9 C: 23.9 ± 9.6 Compared to placebo, change in AHI at 6 weeks: *Note:* Adjusted values in brackets I: ↓ 2 (6.6 ± 5.9*) II: ↓ 4 (8.5 ± 5.2**) C: ↑ 8.5 (4.1 ± 5.5) Unadjusted values extrapolated from Tabl 2 in Carley et al. (2018)	• AE rate: 96.3% • Common: sleepiness/drowsiness (63%), headache (48%), nausea/vomiting (33%) • Two SAEs (one related to dronabinol treatment − diarrhea and vomiting requiring hospitalisation) • Four withdrew due to treatment-related AEs
Prasad et al., 2013 [55]	USA	*High risk*	UB, open-label Comparison to baseline	$n = 17$ (6 M) I: 51.6 ± 7.9 y	3 weeks	Dronabinol (oral capsules) I: 10 mg/day Self-administered 0.5 h before bedtime	Change in AHI relative to baseline at W3	Baseline AHI: 48.4 ± 17.6 I: ↓ 14.1 ± 17.5**	• AE rate: 75% • Common: somnolence (50%) • No SAEs • Two withdrew due to treatment-related AEs
Insomnia/individuals with sleep difficulties									
Shannon et al., 2019 [64]	USA	*High risk*	UB, medical chart review Comparison to baseline	$n = 25$ (9 M) I: 36.5 y	3 months	CBD (oral capsules) I: 25 mg/day *Note*: Some patients received 50 or 75 mg/day Self-administered "after dinner"	PSQI	↓ PSQI Baseline: 13.08 ± 3.03 3-months: 9.33 ± 4.63	• AE rate: not stated • Common: fatigue (n = 2), mild sedation (n = 3), abnormal behaviour (n = 1), and dry eyes (n = 1)
Shannon et al., 2016 [65]	USA	*High risk*	UB, open-label Comparison to baseline	$n = 1$ (1 F) 10 y *PTSD-related insomnia disorder and anxiety*	5 months (ongoing)	CBD (25 mg capsule)/night and CBD (12 mg sublingual spray)/day Self-administered CBD capsule "before bed" and CBD sublingual spray "during the day"	Sleep Disturbance Scale for Children	↓ 21 points relative to baseline	None observed
#Zalai et al., 2015 [67]	Canada	*High risk*	DB, PC Crossover	$n = 11$ (age not specified) *Insomnia disorder and chronic pain*	2 × 4 weeks	Nabilone (oral capsules) I: Dose unspecified C: Placebo Timing of drug administration not reported	Sleep parameters (PSG, MWT, MSLT at baseline, W4, and W8)	Sleep efficiency: ↑ +3.8% (n.s.) Total sleep time: ↑ +3.8% (n.s.) Arousal index: ↓ −24.3% (n.s.) Sleep onset latency: ↑+31.8 min ($p < 0.05$)	Not reported
Ware et al., 2010 [66]	Canada	*Some concerns*	DB, AC Crossover	$n = 32$ (5 M) 49.5 ± 11.2 y *Insomnia disorder and fibromyalgia*	2 × 2 weeks separated by 2-week WO	Nabilone (oral capsules) I: 0.5−1 mg/day II: 10 mg amitriptyline Dose-escalation to 1 mg nabilone or 20 mg amitriptyline for W2 Self-administered "before bed"	PSQI & ISI	Compared to amitriptyline: PSQI ↓ 3.25 points (n.s.) ISI ↓ −3.25 adjusted difference (CI, −5.26 to − 1.24) (n.s.)	• 91 AEs possibly or probably related to nabilone • Common: dizziness, nausea, and dry mouth • No SAEs

Carlini et al., 1981 [63]	Brazil	*Some concerns*	DB, PC Crossover	$n = 15$ *Age/gender not specified* *Relatives of research staff with subjective complaints of sleep difficulties*	Single dose	CBD (oral capsules) I: 40 mg CBD II: 80 mg CBD III: 160 mg CBD CI: Placebo CII: 5 mg Nitrazepam Self-administered 0.5 h before bedtime	Non-validated 10-point sleep questionnaire	All CBD doses ↓ remembering dreams* 160 mg CBD ↑duration of sleep*	AE rate not specified Four participants reported experiencing somnolence
Cousens et al., 1973 [68]	USA	*High risk*	DB, PC Crossover	$n = 9$ (9 M) 21–40 y	Single dose	95% THC (oral liquid) I: 10 mg THC II: 20 mg THC III: 30 mg THC C: Placebo Administered 1.5 h before the "average sleep time of the group"	Sleep latency ("experienced sleep observer-rater")	I: ↓137 min* II: ↓118 min** III: ↓126 min* C: 180 min	Pre-sleep AEs: perceptual/cognitive distortions, loss of control and judgement, dry mouth Post-sleep AEs: dizziness/grogginess, dry mouth, funny taste in mouth.
REM sleep behaviour disorder (RBD)									
Chagas et al., 2014 [76]	Brazil	*High risk*	Case series, UB Comparison to baseline	$n = 4$ (4 M) 63.5 ± 5.3 y *RBD and Parkinson's disease*	6 weeks	CBD (oral capsules) Three patients received 75 mg/day while one received 300 mg/day Self-administered "at night under supervision of relatives/caretakers"	Frequency of RBD events (patient & carer self-report)	"Prompt, substantial and persistent reduction in RBD events after 6 weeks"	None observed
Restless legs syndrome									
ˆMegelin & Ghorayeb 2017 [45]	France	*High risk*	Case series, UB Comparison to baseline	$n = 6$ (2 M) 43.2 ± 11.3 y	Self-reported acute use	Smoked cannabis ($n = 5$) Sublingual CBD ($n = 1$) Not reported	Frequency of RLS symptoms (patient self-report)	"All patients reported spontaneous relief of RLS symptoms"	Not reported
PTSD-related nightmares									
Jetly et al., 2015 [75]	Canada	*Some concerns*	DB, PC, Crossover	$n = 10$ (10 M) 43.6 ± 8.2 y	2 × 7 weeks separated by 2-week WO	Nabilone (oral capsules) I: 0.5 mg/day up to 3 mg C: Placebo Self-administered 1 h before bedtime	Change in nightmare frequency (CAPS Recurring and Distressing Dream score)	CAPS Frequency: I: −1.9 ± 1.3* C: −0.4 ± 1.4 CAPS Intensity: I: −1.7 ± 1.3 (n.s.) C: −0.6 ± 1.1	AE rate: I: 50% C: 60% Common: dry mouth and headache
Fraser 2009 [74]	Canada	*High risk*	UB, open-label Comparison to baseline	$n = 47$ (20 M) 44 ± 9 y	4–12 months	Nabilone (oral capsules) I: 0.5 mg/day up to 6 mg Self-administered 1 h before bedtime	Subjective self-report of nightmare frequency	"Total cessation of nightmares," n = 28, 60% "Satisfactory reduction", n = 6, 12%)	AE rate: 28% (all 28% withdrew) Common: light-headedness, memory impairment, dizziness, and headache

AC = active control; AE = adverse events; AHI = apnea hypoxia index; C = control; CAPS = clinician-administered post-traumatic stress scale; CBD = cannabidiol; CI = confidence interval; DB = double-blind; I = intervention; ISI=Insomnia Severity Index; M = males; MSLT = Multiple Sleep Latency Test; MWT = Maintenance of Wakefulness Test; OSA = obstructive sleep apnea; PC = placebo-controlled; PTSD = post-traumatic stress disorder; PSG = polysomnography; PSQI=Pittsburgh Sleep Quality Index; RBD = rapid eye movement sleep behaviour disorder; REM = rapid eye movement; RLS = restless legs syndrome; SAE = serious adverse events; THC = tetrahydrocannabinol; UB = unblinded; W = week; WO = wash-out; #Conference abstract; ˆLetter to the Editor *p < 0.05 relative to placebo **p < 0.01 relative to placebo.

Table 3
Risk of bias of individual preclinical studies using the SYRCLE risk of bias tool.

	Selection bias			Performance bias		Detection bias		Attrition bias	Reporting bias	Other
Authors, year [reference number]	***Sequence generation***	***Baseline characteristics***	***Allocation concealment***	***Random housing***	***Investigator blinding***	***Random outcome assessment***	***Blinded outcome assessment***	***Incomplete outcome data***	***Selective outcome reporting***	***Other risk of bias***
Models of obstructive sleep apnea										
Calik & Carley (2019) [53]	High risk	Unclear	High risk	High risk	High risk	Low risk	Low risk	Low risk	Low risk	High risk
Calik & Carley (2017) [50]	Unclear	Low risk	High risk	High risk	High risk	Low risk	Low risk	Unclear	High risk	Unclear
Calik & Carley (2016) [47]	High risk	High risk	High risk	High risk	High risk	High risk	High risk	Unclear	Low risk	Unclear
*Carley & Topchiy (2015) [54]	Unclear	Unclear	Unclear	Unclear	Unclear	Unclear	Unclear	Unclear	Low risk	Unclear
*Topchiy et al., (2015) [52]	Unclear	Unclear	Unclear	Unclear	Unclear	Low risk	Unclear	Unclear	Low risk	Low risk
Calik et al., (2014) [48]	High risk	High risk	High risk	High risk	High risk	High risk	High risk	Unclear	Low risk	Unclear
Calik & Carley (2014) [46]	High risk	Low risk	High risk	High risk	High risk	High risk	High risk	Unclear	Low risk	Unclear
*Topchiy et al., (2012) [49]	High risk	Unclear	High risk	Unclear	High risk	Unclear	High risk	Low risk	Low risk	Unclear
Carley et al., (2002) [51]	Unclear	High risk	High risk	High risk	High risk	High risk	High risk	Unclear	Low risk	High risk
Models of sleep disturbances										
Pérez-Morales et al., (2014) [59]	High risk	Low risk	High risk	High risk	High risk	High risk	High risk	Low risk	Low risk	Low risk
Hsias et al., (2012) [62]	High risk	High risk	High risk	High risk	High risk	High risk	High risk	Low risk	Low risk	Low risk
Prieto et al., (2012) [60]	High risk	Low risk	High risk	High risk	High risk	High risk	High risk	Unclear	Low risk	Unclear
Navarro et al., (2003) [61]	High risk	High risk	High risk	Unclear	High risk	High risk	High risk	Unclear	High risk	High risk
Models of narcolepsy										
Murillo-Rodríguez et al., 2019 [73]	High risk	High risk	High risk	High risk	High risk	High risk	Low risk	Low risk	Unclear	Low risk

*Conference abstracts are difficult to assess for risk of bias due to restricted word limits. For more information on each SYRCLE risk of bias domain, see: https://doi.org/10.1186/1471-2288-14-43

Table 4
Risk of bias of individual clinical studies using revised Cochrane Risk of Bias (RoB 2.0) tool.

Authors, year [reference number]	**Randomisation process**	**Deviations from intended intervention**	**Missing data**	**Measurement of outcomes**	**Selection of the reported results**	**Overall risk of bias**
Obstructive sleep apnea						
Carley et al., (2018) [56]	Some concerns	Low risk	Low risk	Low risk	Low risk	**Some concerns**
Prasad et al., (2013) [55]	High risk	Some concerns	Low risk	Low risk	Some concerns	**High risk**
Insomnia or sleep disturbances						
Shannon et al., (2019) [64]	High risk	High risk	High risk	High risk	Some concerns	**High risk**
Shannon et al., (2016) [65]	High risk	High risk	Low risk	High risk	Some concerns	**High risk**
*Zalai et al., (2015) [67]	Some concerns	High risk	High risk	Some concerns	Some concerns	**High risk**
Ware et al., (2010) [67]	Low risk	Low risk	Low risk	Low risk	Some concerns	**Some concerns**
Carlini et al., (1981) [63]	Some concerns	Low risk	Low risk	Low risk	Some concerns	**Some concerns**
Cousens et al., (1973) [68]	Some concerns	Low risk	Low risk	High risk	Some concerns	**High risk**
PTSD-related nightmares						
Jetly et al., (2015) [75]	Some concerns	Low risk	Low risk	Low risk	Some concerns	**Some concerns**
Fraser (2009) [74]	High risk	High risk	High risk	High risk	High risk	**High risk**
REM sleep behaviour disorder						
Chagas et al., (2014) [76]	Some concerns	Low risk	Low risk	High risk	Some concerns	**High risk**
Restless legs syndrome						
Megelin et al., (2017) [45]	High risk	High risk	High risk	High risk	High risk	**High risk**

*Conference abstracts are difficult to assess for risk of bias due to restricted word limits.

disorders and 14 preclinical studies involving animal models of these disorders. The majority of these studies carried a substantial risk of bias such that the conclusions from this review are only tentative. The discussion will focus on synthesis of the existing data for each research question.

Do cannabinoids improve sleep-related breathing outcomes in obstructive sleep apnea?

The current evidence for the use of THC (dronabinol) in individuals with OSA is weak, but the potential therapeutic benefits

Table 5
Synthesis of the available preclinical and clinical studies based on their interpretability (availability and quality), categorised by the research question.

Research question	Preclinical studies	Clinical studies	Evidence for Use
Do cannabinoids improve…			
1. Sleep-related breathing outcomes in obstructive sleep apnea?	Interpretable	Interpretable	Weak
2. Sleep-related outcomes in insomnia disorder?	Interpretable	Interpretable	None
3. Sleep-related outcomes in PTSD-related nightmares?	–	Interpretable	Weak
4. Sleep-related outcomes in REM sleep behaviour disorder?	–	Not interpretable	None
5. Sleep-related movement outcomes in restless legs syndrome?	–	Not interpretable	None
6. Sleep/wake-related outcomes in narcolepsy?	Interpretable	–	None

Dash (–) signifies no studies identified; 'Interpretable' signifies studies were identified and deemed to have a low-moderate risk of bias; 'Not interpretable' signifies studies were identified but deemed to have a high risk of bias. REM = rapid eye movement; PTSD = post-traumatic stress disorder.

warrant further investigation. All but two preclinical studies [47,53] found that THC reduced apneic events in rats when administered intraperitoneally but not when administered via intracerebroventricular injection [47]. This finding, and the results of other studies, suggest that the effects of THC on reflex apnea may be peripherally mediated via suppression of vagal nerve activity by the endocannabinoid system. Specifically, one study showed that THC inhibition of reflex apnea could be reversed with administration of CB_1 and CB_2 receptor antagonists [46]. Another study indicated that THC may dampen afferent vagal feedback to the medulla via actions on the nodose ganglia, a component of the vagus nerve that expresses excitatory serotonin type 3 ($5\text{-}HT_3$) and inhibitory CB_1 receptors [47]. This is a plausible mechanism through which THC could stabilise respiratory patterns and increase activation of upper airway dilating muscles during sleep in a manner largely independent of cannabinoid receptors located in the central nervous system [78].

Two clinical studies of oral dronabinol as a potential treatment for OSA showed reductions in the apnea-hypopnea index (AHI) after treatment. In their initial open-label, pre-post, proof of concept trial in individuals with moderate to severe OSA ($n = 15$), Prasad et al. (2013) reported a significant 14.1 ± 17.5-point reduction in AHI relative to baseline (AHI = 48.4 ± 17.6) after three weeks of dronabinol treatment [55]. The subsequent 6-wk RCT by Carley et al. (2018) in individuals with moderate to severe ($n = 73$) also identified a positive effect [55]. However, findings from this trial should be interpreted with caution as the statistically significant reduction in the adjusted AHI of 12.9 ± 4.3-points with 10 mg/d dronabinol treatment was at least partly attributable to a potentially clinically meaningful baseline imbalance in AHI (placebo = 23.9 ± 9.6 vs. 10 mg/d dronabinol = 26.2 ± 11.9) and a significant 8.5-point increase in AHI in the placebo-treated group after six weeks. Baseline AHI was statistically controlled for in the analyses along with age, race, and ethnicity as additional covariates. After adjustment, the increase in AHI from baseline in the placebo group was smaller (−4.1 ± 5.5-points) and not statistically significant. This resulted in a 12.9-point difference relative to placebo for the 10 mg/d dronabinol-treated group. The authors hypothesised the worsening AHI in the placebo group may have been due to the participant's discontinuation of other interventions (e.g., continuous positive airway pressure (CPAP)) one month prior to the trial, although, this was not objectively confirmed. Neither dose of dronabinol showed a statistically significant reduction in AHI relative to baseline. Furthermore, of the 39 participants who received dronabinol treatment, only six (15.4%) met the trial's responder criteria (i.e., AHI≤15 and AHI reduction of 50% or more from baseline). Treatment-related adverse events occurred in 75% and 96% of participants receiving 10 mg/d in the Prasad et al. (2013) and Carley et al. (2018) trial, respectively. The most common adverse event in the Carley et al. (2018) RCT was drowsiness (63% vs. 0% in placebo group) followed by headache (48% vs. 15% in placebo group) and nausea/vomiting (33% vs. 4% in placebo group) [79]. Four participants withdrew due to treatment-related adverse events (dizziness and vision changes; vertigo; ECG arrhythmias; headache, dizziness, and vomiting). One serious adverse event related to dronabinol treatment was reported (diarrhea and vomiting requiring hospitalisation).

It is worthwhile noting that a 2018 position statement from the *American Academic of Sleep Medicine* warned clinicians against prescribing dronabinol as a treatment for OSA and argued that OSA should not be a certifiable health condition for medical cannabis programs due to unknown short- and long-term side effects of dronabinol in patients with OSA [80]. Additional research in this area is recommended, especially studies that evaluate clinical response in specific OSA phenotypes [81]. Thus, despite a positive signal, dronabinol is not currently recommended for the treatment of OSA. Well-designed randomised controlled short-term trials as well as longer-term studies are needed to further determine the efficacy and safety of dronabinol in individuals with OSA. Indeed, one pre-registered Phase IIa placebo-controlled clinical trial will test the effects of 10 mg THC with a 200 mg proprietary mineral supplement over a 6-week period [58], while a second open-label Phase IIa clinical trial will assess the effects of an oral formulation containing 10 mg dronabinol in combination with 800 mg palmitoylethanolamide (PEA), an endogenous fatty acid amide, in 30 patients with OSA over a 4-wk period [57] (see Table S1). The primary outcome for both trials is AHI index post-treatment as compared to baseline using overnight polysomnography. The Phase IIa trial of dronabinol in combination with PEA recently announced top line findings for 10 patients in a press release [82]. No serious adverse events were reported, with one patient withdrawing due to treatment-related dizziness. Of the remaining nine participants, just over half showed a significant reduction in average AHI from 24.2 ± 5.0 at baseline to 11.2 ± 6.8 at four weeks. The press release cited the "encouraging tendency of the results" as the main reason for early study recruitment closure.

Do cannabinoids treatment improve sleep-related outcomes in insomnia disorder?

There were no published RCTs investigating the effects of cannabinoid therapies in patients with clinician-diagnosed insomnia, limiting any conclusions regarding their utility for insomnia disorder. THC displays minimal toxicity and lethality [83], inferring a safety advantage over hypnotic medications. However, abrupt discontinuation of daily, or near daily cannabis use may lead to abstinence-induced insomnia [84] and sleep difficulty is a commonly reported symptom of cannabis withdrawal among frequent cannabis users (e.g., at least 25 days/mo) [85]. Poor sleep quality has also been shown to be a risk factor for lapse following a cannabis quit attempt in cannabis dependent users [86]. Laboratory studies have shown that cannabis abstinence-induced sleep

disturbance is pharmacologically specific to THC exposure, as it can be reversed with administration of dronabinol or by a return to cannabis use [87,88]. It is important to note that this research has mainly focused on heavy recreational (non-medicinal) use of cannabis. *Sativex*, an oromucosal spray delivering equal parts THC and CBD, improved self-reported sleep across multiple clinical trials in the treatment of pain conditions (such as multiple sclerosis, peripheral neuropathic pain, intractable cancer pain, and rheumatoid arthritis) [89]. Although not covered in the present review, there is moderate evidence for the use of Sativex in improving short-term sleep outcomes in individuals with sleep disturbances secondary to a pain condition [89], however, no studies have assessed its effects in individuals with a primary sleep disorder.

In an animal model of sleep disturbances induced by repeated exposure to anxiety-provoking environments, CBD microinjected into the central nucleus of the amygdala reversed stress-induced REM suppression, with little effect on NREM sleep [62]. This suggests improvements in sleep via an anxiolytic mechanism not yet fully understood, but which may involve serotonin 1A (5-HT_{1A}) receptor activation [90] and/or enhancement of AEA signalling by inhibition of FAAH and FABPs [91]. Other preclinical evidence suggests that CB1 receptor activation via endocannabinoids, oleamide and 2-AG, normalised maternal separation-associated sleep disturbances such as increased wakefulness and decreased NREM and REM sleep duration [59,60]. Other preclinical evidence suggests endocannabinoids may also play a role in modulating REM sleep generation following sleep deprivation in rats [61]. This is in line with previous work suggesting that endocannabinoid signalling is necessary to promote sleep stability [18].

The acute and chronic effects of CBD on sleep are poorly understood currently, and there is a lack of empirical data in which CBD has been evaluated among individuals with disordered sleep. In clinical trials involving 25/mg/kg CBD (Epidiolex) in children with severe epilepsies, increased somnolence and sedation was observed. However, in these studies, CBD was found to be a potent metabolic inhibitor of concurrently-administered anticonvulsant medications, which may have driven the sedating effects reported in these trials [92,93]. Drowsiness was reported as the fourth most common side effect in an ascending dose Phase 1 trial of CBD in healthy volunteers, but the incidence did not differ from placebo and the greatest frequency of somnolence observed was with an acute dose of 6000 mg, which far exceeds the typical dose found in retail CBD products (for example, the unit dose of CBD in Epidiolex is 100 mg) [94]. In another laboratory study of healthy adults, 100 mg CBD administered orally and via vaporization did not impact subjective ratings of alertness and sleepiness [95]. Because CBD can act as a negative allosteric modulator of the CB_1 receptor [34], it is feasible that CBD could exhibit stimulating properties at certain doses, however, this has not been clearly demonstrated in controlled studies.

In summary, there is no published evidence to-date assessing the effects of cannabinoid therapies in individuals with clinician-diagnosed insomnia disorder. Future studies should use validated objective measures to assess the therapeutic impact of pharmaceutical-grade cannabinoid therapies in individuals with clinician-diagnosed insomnia in both the short- and long-term. Four pre-registered randomised placebo-controlled trials (ranging from one night to 9-wk treatment periods) are currently underway administering either CBD alone [72] or proprietary combinations of CBD, THC and/or CBN [69–71] in individuals with chronic insomnia (see Table S1). Two studies will use objective primary outcome measures (overnight polysomnography to assess various sleep metrics such as total sleep time and wake after sleep onset) [69,70]. The other two studies will use the Insomnia Severity Index (ISI) [71,72] with one study also co-administering an additional standardised self-report questionnaire [72]. One of these studies, a 2-wk Phase Ib/IIa RCT of a proprietary combination of THC, CBD, and CBN ('ZLT-101') [71], recently announced top line findings in a press release, recruiting 23 of its intended 30 participant target [96]. ISI scores significantly decreased from 18.0 ± 3.7 at baseline to 12.9 ± 5.3 at two weeks post-treatment. Dry mouth, dizziness, and headache were the most common adverse events, with no serious adverse events reported. These new clinical trials represent a useful step in advancing our understanding of the potential therapeutic effects of cannabinoids in insomnia disorder.

Do cannabinoids improve sleep-related outcomes in PTSD-related nightmares?

There is accumulating evidence that the synthetic THC analogue nabilone may be effective in the management of nightmares among individuals with PTSD. In both an RCT and an open label study, nabilone (0.5 mg/d up to a maximum of 3 mg/d) significantly reduced the frequency of nightmares. Mild adverse effects were reported among 50% of patients, the most common being dry mouth, headache and dizziness. The small sample size in the RCT and open-label design of the study by Fraser et al. [74] are notable limitations and further well-designed trials with larger, more diverse clinical populations (i.e., inclusion of females and individuals with non-trauma-related nightmare disorders), along with longer-term follow-up, are needed to consolidate these findings. While the limited available evidence indicates that nabilone reduces PTSD-related nightmares, the long-term safety of CB_1 receptor agonists in this population is unclear, particularly given their complex comorbidities and increased risk of substance abuse.

Do cannabinoids improve sleep-related outcomes in REM sleep behaviour disorder?

No RCTs assessing the effects of cannabinoid therapies in REM sleep behaviour disorder (RBD) in Parkinson's disease were identified. In a subset of patients with Parkinson's disease and RBD, a low-to-moderate dose of CBD resulted in a rapid and substantial reduction in the frequency of RBD-related events with nil adverse events reported [76]. This clearly requires further placebo-controlled investigation to identify potentially more effective and safer therapies for patients with this neurodegenerative disease.

Do cannabinoids improve sleep-related movement outcomes in restless legs syndrome?

The case series of patients with treatment-resistant RLS involved spontaneous self-report to their clinical team of an instant and complete reduction in RLS symptoms with illicit cannabis products (including one patient using sublingual CBD). These observations are promising and warrant further investigation using a RCT design. This is particularly important given that standard treatment with dopaminergic agents is often associated with severe side effects such as augmentation, a worsening of RLS symptoms after starting dopaminergic medication [97] or dopamine agonist-related impulse control [98], limiting their long-term usefulness. Despite the promising patient self-reports, there is no available evidence from studies using prospective study design that would warrant clinical use of cannabinoids for RLS at present.

Do cannabinoids improve sleep/wake-related outcomes in narcolepsy?

A similarly difficult-to-treat disorder is narcolepsy, with the characteristic symptom of excessive daytime somnolence. While no clinical studies administering cannabinoid therapies were identified, a recent preclinical study showed that peripheral injection of CBD partially blocked excessive sleepiness in hypocretin-deficient rats, an animal model of narcolepsy [73]. This adds to a larger body of existing preclinical work from this group describing the potential wakefulness-promoting properties of CBD [99–101]. Indeed, a recent study indicated that CBD (i.e., 30 mg/kg *i.p.*) enhances alertness, and decreases slow wave sleep and REM sleep during the 'lights-on' period in rats [102]. These effects were associated with increases in neuronal activation of lateral hypothalamus and dorsal raphe nuclei (areas implicated in alertness control) and elevated extracellular dopamine concentrations [99,100], consistent with the well-documented role for dopamine in mediating wakefulness and arousal [103]. Given the favourable safety profile of CBD in humans, this initial evidence provides impetus to translate the findings into a properly designed RCT of CBD as an adjunctive treatment in individuals with excessive daytime sleepiness such as in patients with narcolepsy, OSA, and idiopathic hypersomnolence.

Safety considerations

The short- and long-term safety risks of cannabinoid therapies for individuals with sleep disorders are still being determined. Although THC displays minimal toxicity and lethality [83], moderate doses of THC (>10 mg) in naïve or occasional cannabis users can produce significant intoxication and/or impair cognitive performance (e.g., reaction time tasks) [68,104]. THC can also impair driving performance [105,106]. While the majority of clinical studies identified in the current review administered the cannabinoid treatment prior to bedtime (acute impairment was less of a concern), residual next-day effects of cannabinoid treatment on cognition, alertness, and driving performance should also be explored for safety. For instance, somnolence was the most common side-effect of dronabinol in the OSA trials. However, Carley et al. (2018) reported that participants receiving 10 mg/d dronabinol showed significantly decreased self-reported daytime sleepiness relative to baseline as measured on the Epworth Sleepiness Scale at 6-weeks [56]. Nonetheless, in places where cannabis products are legally accessible, use of THC-containing products for sleep disorders and the potential adverse events (e.g., daytime sleepiness) must be carefully considered and managed, particularly when unregulated products are being accessed.

CBD is non-intoxicating and has shown to be safe and well-tolerated in humans [107] – even at very high doses (e.g., 1500 mg twice daily for six days or as an acute dose of 6000 mg) [94]. No evidence of a withdrawal syndrome was evident following abrupt cessation of 4-wk treatment with 750 mg CBD twice daily in healthy volunteers [108]. THC and CBD are potent substrates and inhibitors of the cytochrome P450 enzymatic pathways which is involved in the biotransformation of many commonly prescribed medications [109]. Potential drug–drug interactions with cannabinoids are theoretically possible, as now shown between CBD and the anticonvulsant drug, clobazam, in children with severe epilepsy [92]. Few specific directives can be made at this stage due to the limited data on drug–drug interactions with CBD and THC, therefore, healthcare professionals should familiarise themselves with potential drug–drug interactions relevant to the patient's medication history and hepatic function [109,110]. Finally, the comparative safety and efficacy of cannabinoid medications relative to conventional treatment approaches is also a worthy area of investigation.

Strengths and limitations

This is the first systematic review focused specifically on cannabinoid therapies for sleep disorders that covers both the preclinical and clinical literature. This systematic review has several limitations. Firstly, only English-language articles were included. Second, despite our systematic approach, we cannot be completely certain that all relevant articles were retrieved via our literature search. Finally, most of the included clinical studies had small sample sizes and poor methodological quality including most studies with a high-risk of bias, limiting the strength of the conclusions that can be drawn from this review.

Conclusion

At present, there is limited evidence to support the clinical use of cannabinoid therapies for the treatment of any sleep disorder given the dearth of published research and the moderate-to-high risk of bias identified within the majority of clinical and preclinical studies completed to-date. Nonetheless, there are promising signs in a number of therapeutic applications that warrant additional study and there is a clear need for intensification of high-quality research into the safety and efficacy of cannabinoid therapies for treating sleep disorders. Research should utilize well-defined cannabinoid products, validated assessments, and include measures of next-day function (i.e., cognition and driving performance). Additional scientific endeavour is required to define the mechanisms through which the endocannabinoid system affects sleep and sleep-related physiology and the pharmacological actions of various cannabinoid agents in relation to sleep. Currently ongoing clinical trials of cannabinoids in obstructive sleep apnea and insomnia are a positive step to a better understanding of the role of cannabinoid therapies in the treatment of sleep disorders.

Practice points

1) Individuals who use medicinal cannabis often do so to treat or manage sleep disorders such as insomnia, with many self-reporting that is highly effective in managing their symptoms.
2) Our systematic review deemed the available clinical and preclinical evidence to have a moderate-to-high risk of bias precluding any definitive conclusions regarding their therapeutic efficacy of cannabinoids in sleep disorders.
3) Promising preliminary evidence from preclinical and clinical studies provide the rationale for future randomised, controlled trials of cannabinoid therapies in individuals with sleep apnea, insomnia, post-traumatic stress disorder-related nightmares, REM sleep behaviour disorder, restless legs syndrome, and narcolepsy.
4) The safety profile of acute and chronic treatment with cannabinoids in sleep disorders is not yet well understood.
5) In places where cannabis products are legally accessible, use of THC-containing products for sleep disorders and the potential adverse effects on next-day function such as driving must be carefully considered and managed, particularly when unregulated products are being used.

Research agenda

To further examine the therapeutic utility of cannabinoid therapies on sleep disorders, future research should:

1) utilise validated objective and subjective measures of sleep-related outcomes to assess therapeutic efficacy of cannabinoids;
2) utilise robustly designed randomised, controlled trial designs, employing properly powered sample sizes with an adequate comparator (placebo and active treatment, where available);
3) consider the ecological validity of administering cannabinoids as an adjunctive treatment as opposed to a stand-alone treatment given the potential implications of drug−drug interactions on safety;
4) explore dose-dependent effects of THC in order to identify the optimal dose that confers clinical efficacy without causing next-day impairment such as drowsiness;
5) further explore the opposing effects of CBD on the sleep/wake cycle and to prioritise research on CBD given its non-intoxicating properties and nil potential for abuse or dependence.

Conflicts of interest

RRG and NSM have received discounted investigational products for an unrelated clinical trial from Neurim Pharmaceuticals Inc. RRG and NSM have also received investigational product and matched placebo from Teva Pharmaceutical in unrelated clinical trials. ISM is a consultant for Kinoxis Therapeutics, and is an inventor on several patents relating to novel cannabinoid therapeutics. RV has received income as a consultant or advisory board member from Zynerba Pharmaceuticals, Canopy Health Innovations Inc., and FSD Pharma. The other authors have no conflicts of interest to disclose.

Acknowledgements

This work was supported by the Lambert Initiative for Cannabinoid Therapeutics, a philanthropically-funded center at the University of Sydney. AS was supported by the Australian Government Research Training Program (RTP) and the Vice-Chancellor's Research Scholarship (VCRS) at the University of Sydney. DM, MJB, and ISM were supported by the Lambert Initiative for Cannabinoid Therapeutics at the University of Sydney. CMH was supported by a Dementia Research Development Fellowship of the Australian National Health and Medical Research Council-Australian Research Council (NHMRC-ARC) (APP1104003). RRG was supported by an NHMRC Senior Principal Research Fellowship (APP1106974).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.smrv.2020.101339.

References

[1] Buxton OM, Broussard JL, Zahl AK, Hall M. Effects of sleep deficiency on hormones, cytokines, and metabolism. Impact of sleep and sleep disturbances on obesity and cancer. Springer; 2014. p. 25−50.
[2] Murawski B, Wade L, Plotnikoff RC, Lubans DR, Duncan MJ. A systematic review and meta-analysis of cognitive and behavioral interventions to improve sleep health in adults without sleep disorders. Sleep Med Rev 2018;40:160−9.
[3] Liu Y. Prevalence of healthy sleep duration among adults—United States, 2014, vol. 65. Morbidity and Mortality Weekly Report; 2016.
[4] Hillman D, Mitchell S, Streatfeild J, Burns C, Bruck D, Pezzullo L. The economic cost of inadequate sleep. Sleep 2018;41(8):zsy083.
[5] Baglioni C, Battagliese G, Feige B, Spiegelhalder K, Nissen C, Voderholzer U, et al. Insomnia as a predictor of depression: a meta-analytic evaluation of longitudinal epidemiological studies. J Affect Disord 2011;135(1−3):10−9.
[6] Harris M, Glozier N, Ratnavadivel R, Grunstein RR. Obstructive sleep apnea and depression. Sleep Med Rev 2009;13(6):437−44.
[7] Bertisch SM, Pollock BD, Mittleman MA, Buysse DJ, Bazzano LA, Gottlieb DJ, et al. Insomnia with objective short sleep duration and risk of incident cardiovascular disease and all-cause mortality: sleep Heart Health Study. Sleep 2018;41(6):zsy047.
[8] Golbin JM, Somers VK, Caples SM. Obstructive sleep apnea, cardiovascular disease, and pulmonary hypertension. Proc Am Thorac Soc 2008;5(2):200−6.
[9] Buratti L, Luzzi S, Petrelli C, Baldinelli S, Viticchi G, Provinciali L, et al. Obstructive sleep apnea syndrome: an emerging risk factor for dementia. CNS Neurol Disord - Drug Targets 2016;15(6):678−82.
[10] Hung C-M, Li Y-C, Chen H-J, Lu K, Liang C-L, Liliang P-C, et al. Risk of dementia in patients with primary insomnia: a nationwide population-based case-control study. BMC Psychiatr 2018;18(1):38.
[11] Kraus SS, Rabin LA. Sleep America: managing the crisis of adult chronic insomnia and associated conditions. J Affect Disord 2012;138(3):192−212.
[12] Bonini SA, Premoli M, Tambaro S, Kumar A, Maccarinelli G, Memo M, et al. Cannabis sativa: a comprehensive ethnopharmacological review of a medicinal plant with a long history. J Ethnopharmacol 2018;227:300−15.
[13] Hazekamp A, Ware MA, Muller-Vahl KR, Abrams D, Grotenhermen F. The medicinal use of cannabis and cannabinoids—an international cross-sectional survey on administration forms. J Psychoact Drugs 2013;45(3):199−210.
[14] Lintzeris N, Driels J, Elias N, Arnold JC, McGregor IS, Allsop DJ. Medicinal cannabis in Australia, 2016: the cannabis as medicine survey (CAMS-16). Med J Aust 2018;209(5):211−6.
[15] Pledger MJ, Martin G, Cumming J. New Zealand Health Survey 2012/13: characteristics of medicinal cannabis users. N Z Med J 2016;129(1433):29.
[16] Choi S, Huang BC, Gamaldo CE. Therapeutic uses of cannabis on sleep disorders and related conditions. J Clin Neurophysiol 2020;37(1):39−49.
[17] Di Marzo V, Bifulco M, De Petrocellis L. The endocannabinoid system and its therapeutic exploitation. Nat Rev Drug Discov 2004;3(9):771.
[18] Pava MJ, Makriyannis A, Lovinger DM. Endocannabinoid signaling regulates sleep stability. PloS One 2016;11(3):e0152473.
[19] Hanlon EC, Tasali E, Leproult R, Stuhr KL, Doncheck E, De Wit H, et al. Sleep restriction enhances the daily rhythm of circulating levels of endocannabinoid 2-arachidonoylglycerol. Sleep 2016;39(3):653−64.
[20] Vaughn LK, Denning G, Stuhr KL, De Wit H, Hill MN, Hillard CJ. Endocannabinoid signalling: has it got rhythm? Br J Pharmacol 2010;160(3):530−43.
[21] Hanlon EC. Impact of circadian rhythmicity and sleep restriction on circulating endocannabinoid (eCB) N-arachidonoylethanolamine (anandamide). Psychoneuroendocrinology 2020;111:104471.
[22] Hanlon EC, Tasali E, Leproult R, Stuhr K, Doncheck E, De Wit H, et al. Circadian rhythm of circulating levels of the endocannabinoid 2-arachidonoylglycerol. J Clin Endocrinol Metabol 2015;100(1):220−6.
[23] Wyatt RM, Fraser I, Welty N, Lord B, Wennerholm M, Sutton S, et al. Pharmacologic characterization of JNJ-42226314, [1-(4-Fluorophenyl)indol-5-yl]-[3-[4-(thiazole-2-carbonyl)piperazin-1-yl]azetidin -1-yl]methanone, a reversible, selective, and potent monoacylglycerol lipase inhibitor. J Pharmacol Exp Therapeut 2020;372(3):339−53.
[24] D'Souza DC, Cortes-Briones J, Creatura G, Bluez G, Thurnauer H, Deaso E, et al. Efficacy and safety of a fatty acid amide hydrolase inhibitor (PF-04457845) in the treatment of cannabis withdrawal and dependence in men: a double-blind, placebo-controlled, parallel group, phase 2a single-site randomised controlled trial. The Lancet Psychiatry 2019;6(1):35−45.
[25] Murillo-Rodriguez E, Blanco-Centurion C, Sanchez C, Daniele P, Shiromani PJ. Anandamide enhances extracellular levels of adenosine and induces sleep: an in vivo microdialysis study. Sleep 2003;26(8):943−7.
[26] Mechoulam R, Fride E, Hanu L, Sheskin T, Bisogno T, Di Marzo V, et al. Anandamide may mediate sleep induction. Nature 1997;389(6646):25−6.
[27] Murillo-Rodrıguez E, Sanchez-Alavez M, Navarro L, Martınez-Gonzalez D, Drucker-Colın R, Prospero-Garcıa O. Anandamide modulates sleep and memory in rats. Brain Res 1998;812(1−2):270−4.
[28] Murillo-Rodríguez E, Cabeza R, Méndez-Díaz M, Navarro L, Prospéro-García O. Anandamide-induced sleep is blocked by SR141716A, a CB1 receptor antagonist and by U73122, a phospholipase C inhibitor. Neuroreport 2001;12(10):2131−6.
[29] Després J-P, Golay A, Sjöström L. Effects of rimonabant on metabolic risk factors in overweight patients with dyslipidemia. N Engl J Med 2005;353(20):2121−34.
[30] Pi-Sunyer FX, Aronne LJ, Heshmati HM, Devin J, Rosenstock J, Group R-NAS. Effect of rimonabant, a cannabinoid-1 receptor blocker, on weight and cardiometabolic risk factors in overweight or obese patients: RIO-North

America: a randomized controlled trial. J Am Med Assoc 2006;295(7):761–75.
[31] Scheen AJ, Finer N, Hollander P, Jensen MD, Van Gaal LF, Group R-DS. Efficacy and tolerability of rimonabant in overweight or obese patients with type 2 diabetes: a randomised controlled study. Lancet 2006;368(9548):1660–72.
[32] Van Gaal LF, Rissanen AM, Scheen AJ, Ziegler O, Rössner S, Group R-ES. Effects of the cannabinoid-1 receptor blocker rimonabant on weight reduction and cardiovascular risk factors in overweight patients: 1-year experience from the RIO-Europe study. Lancet 2005;365(9468):1389–97.
[33] Murillo-Rodríguez E. The role of the CB1 receptor in the regulation of sleep. Prog Neuro Psychopharmacol Biol Psychiatr 2008;32(6):1420–7.
[34] Tham M, Yilmaz O, Alaverdashvili M, Kelly ME, Denovan-Wright EM, Laprairie RB. Allosteric and orthosteric pharmacology of cannabidiol and cannabidiol-dimethylheptyl at the type 1 and type 2 cannabinoid receptors. Br J Pharmacol 2019;176(10):1455–69.
[35] McPartland JM, Duncan M, Di Marzo V, Pertwee RG. Are cannabidiol and Δ9-tetrahydrocannabivarin negative modulators of the endocannabinoid system? A systematic review. Br J Pharmacol 2015;172(3):737–53.
[36] Elmes MW, Kaczocha M, Berger WT, Leung K, Ralph BP, Wang L, et al. Fatty acid-binding proteins (FABPs) are intracellular carriers for Δ9-tetrahydrocannabinol (THC) and cannabidiol (CBD). J Biol Chem. 2015;290(14):8711–21.
[37] Abrams DI. The therapeutic effects of cannabis and cannabinoids: an update from the national academies of sciences, engineering and medicine report. Eur J Intern Med 2018;48:7–11.
[38] Babson KA, Sottile J, Morabito D. Cannabis, cannabinoids, and sleep: a review of the literature. Curr Psychiatr Rep 2017;19(4):23.
[39] Gates PJ, Albertella L, Copeland J. The effects of cannabinoid administration on sleep: a systematic review of human studies. Sleep Med Rev 2014;18(6):477–87.
[40] Kuhathasan N, Dufort A, MacKillop J, Gottschalk R, Minuzzi L, Frey BN. The use of cannabinoids for sleep: a critical review on clinical trials. Exp Clin Psychopharmacol 2019;27(4):383–401.
[41] Moher D, Liberati A, Tetzlaff J, Altman DG, Group P. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Ann Intern Med 2010;151(4):264–9.
[42] Hicks RA, Moore JD, Hayes C, Phillips N, Hawkins J. REM sleep deprivation increases aggressiveness in male rats. Physiol Behav 1979;22(6):1097–100.
[43] Hooijmans CR, Rovers MM, De Vries RB, Leenaars M, Ritskes-Hoitinga M, Langendam MW. SYRCLE's risk of bias tool for animal studies. BMC Med Res Methodol 2014;14(1):43.
[44] Sterne JA, Savović J, Page MJ, Elbers RG, Blencowe NS, Boutron I, et al. RoB 2: a revised tool for assessing risk of bias in randomised trials. BMJ 2019;366:l4898.
[45] Megelin T, Ghorayeb I. Cannabis for restless legs syndrome: a report of six patients. Sleep Med 2017;36:182.
[46] Calik MW, Carley DW. Cannabinoid type 1 and type 2 receptor antagonists prevent attenuation of serotonin-induced reflex apneas by dronabinol in Sprague-Dawley rats. PloS One 2014;9(10):e111412.
[47] Calik MW, Carley DW. Intracerebroventricular injections of dronabinol, a cannabinoid receptor agonist, does not attenuate serotonin-induced apnea in Sprague-Dawley rats. J Negat Results Biomed 2016;15(1):8.
[48] Calik MW, Radulovacki M, Carley DW. Intranodose ganglion injections of dronabinol attenuate serotonin-induced apnea in Sprague-Dawley rat. Respir Physiol Neurobiol 2014;190:20–4.
[49] Topchiy I, Waxman J, Radulovacki M, Carley D, editors. Effects of dronabinol on vagally mediated respiratory reflexes and upper airway motor output. Sleep; 2012 (Vol. 35, pp. A55-A55). One Westbrook Corporate Ctr, Ste 920, Westchester, IL 60154 USA: Amer Acad Sleep Medicine.
[50] Calik MW, Carley DW. Effects of cannabinoid agonists and antagonists on sleep and breathing in Sprague-Dawley rats. Sleep 2017;40(9).
[51] Carley DW, Pavlovic S, Janelidze M, Radulovacki M. Functional role for cannabinoids in respiratory stability during sleep. Sleep 2002;25(4):388–95.
[52] Topchiy I, Petukhov P, Petukhova V, Hickok J, Thatcher G, Carley DW. Peripheral cannabinoid receptor agonists chromenopyrazole 13a and HU-308 attenuate sleep apneas in rats. Sleep 2015;38.
[53] Calik MW, Carley DW. DMSO potentiates the suppressive effect of dronabinol, a cannabinoid, on sleep apnea and REM sleep. bioRxiv 2019:769463.
[54] Carley DW, Topchiy I. Activation of nodose ganglion cannabinoid receptors potentiates upper airway muscle activation. Barcelona: 3rd Sleep and Breathing Conference; 2015.
[55] Prasad B, Radulovacki MG, Carley DW. Proof of concept trial of dronabinol in obstructive sleep apnea. Front Psychiatr 2013;4:1.
[56] Carley DW, Prasad B, Reid KJ, Malkani R, Attarian H, Abbott SM, et al. Pharmacotherapy of apnea by cannabimimetic enhancement, the PACE clinical trial: effects of dronabinol in obstructive sleep apnea. Sleep 2018;41(1).
[57] ClinicalTrialsgov. A study to examine the efficacy of a therapeutic THX-110 for obstructive sleep apnea. 2018 [updated 22 Nov 2019. Available from: https://clinicaltrials.gov/ct2/show/NCT03646552.
[58] ANZCTR. The effects of novel medicinal cannabis formulation IHL-42X on apnoea hypopnea index in adults with suspected or diagnosed mild to moderate obstructive sleep apnoea [updated 22 Nov 2019. Available from: https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=378058&isReview=true; 2019.
[59] Pérez-Morales M, Fajardo-Valdez A, Méndez-Díaz M, Ruiz-Contreras AE, Prospéro-García O. 2-Arachidonoylglycerol into the lateral hypothalamus improves reduced sleep in adult rats subjected to maternal separation. Neuroreport 2014;25(18):1437–41.
[60] Prieto NMR, López AR, Morales MP, Pech O, Méndez-Díaz M, Contreras AER, et al. Oleamide restores sleep in adult rats that were subjected to maternal separation. Pharmacol Biochem Behav 2012;103(2):308–12.
[61] Navarro L, Martínez-Vargas M, Murillo-Rodríguez E, Landa A, Méndez-Díaz M, Prospéro-García O. Potential role of the cannabinoid receptor CB1 in rapid eye movement sleep rebound. Neuroscience 2003;120(3):855–9.
[62] Hsiao Y-T, Yi P-L, Li C-L, Chang F-C. Effect of cannabidiol on sleep disruption induced by the repeated combination tests consisting of open field and elevated plus-maze in rats. Neuropharmacology 2012;62(1):373–84.
[63] Carlini EA, Cunha JM. Hypnotic and antiepileptic effects of cannabidiol. J Clin Pharmacol 1981;21(S1). 417S-27S.
[64] Shannon S, Lewis N, Lee H, Hughes S. Cannabidiol in anxiety and sleep: a large case series. Perm J 2019;23.
[65] Shannon S, Opila-Lehman J. Effectiveness of cannabidiol oil for pediatric anxiety and insomnia as part of posttraumatic stress disorder: a case report. Perm J 2016;20(4):108.
[66] Ware MA, Fitzcharles M-A, Joseph L, Shir Y. The effects of nabilone on sleep in fibromyalgia: results of a randomized controlled trial. Anesth Analg 2010;110(2):604–10.
[67] Zalai D, Chung S, Hussain N, Shapiro C. Does cannabinoid really improve sleep? Testing the sleep effects of nabilone in chronic pain patients: a placebo-controlled, randomized, pilot study. Psychother Psychosom 2015;84.
[68] Cousens K, DiMascio A. (−) δ 9 THC as an hypnotic. Psychopharmacologia 1973;33(4):355–64.
[69] ANZCTR. A single-dose, double-blind, placebo-controlled, randomised, crossover study of an oral cannabis-based medicine (ETC120) on sleep, cognition, and next-day function in adults with chronic insomnia disorder Australian New Zealand Clinical Trials Registry. Available from: https://anzctr.org.au/Trial/Registration/TrialReview.aspx?id=377330; 2019.
[70] ClinicalTrialsgov. Investigation of the efficacy and safety of CHI-921 in insomnia. 2019. Available from: https://clinicaltrials.gov/ct2/show/NCT03984604.
[71] ANZCTR. A trial to evaluate an oral medicinal cannabis extract compared with placebo for the treatment of insomnia Australian New Zealand Clinical Trials Registry. 2019. Available from: https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=373959.
[72] ANZCTR. Cannabidiol (CBD) treatment for insomnia: a three-week randomised controlled pilot trial with a placebo run-in period (CBD-INS) Australian New Zealand Clinical Trials Registry. Available from: https://www.anzctr.org.au/Trial/Registration/TrialReview.aspx?id=379003&isReview=true; 2020.
[73] Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, Morales-Lara D, Mechoulam R, Drucker-Colín R. Cannabidiol partially blocks the sleepiness in hypocretin-deficient rats. Preliminary data. CNS & Neurological Disorders Drug Targets 2019;18(9):705–12.
[74] Fraser GA. The use of a synthetic cannabinoid in the management of treatment-resistant nightmares in posttraumatic stress disorder (PTSD). CNS Neurosci Ther 2009;15(1):84–8.
[75] Jetly R, Heber A, Fraser G, Boisvert D. The efficacy of nabilone, a synthetic cannabinoid, in the treatment of PTSD-associated nightmares: a preliminary randomized, double-blind, placebo-controlled cross-over design study. Psychoneuroendocrinology 2015;51:585–8.
[76] Chagas MH, Eckeli A, Zuardi AW, Pena-Pereira MA, Sobreira-Neto MA, Sobreira E, et al. Cannabidiol can improve complex sleep-related behaviours associated with rapid eye movement sleep behaviour disorder in Parkinson's disease patients: a case series. J Clin Pharm Therapeut 2014;39(5):564–6.
[77] De Angelis C, Drazen JM, Frizelle FA, Haug C, Hoey J, Horton R, et al. Clinical trial registration: a statement from the international committee of medical journal. Circulation 2005;111(10):1337–8.
[78] Carley DW, Radulovacki M. Pharmacology of vagal afferent influences on disordered breathing during sleep. Respir Physiol Neurobiol 2008;164(1–2):197–203.
[79] ClinicalTrialsgov. Safety and efficacy study of dronabinol to treat obstructive sleep apnea (PACE). Available from: https://clinicaltrials.gov/ct2/show/results/NCT01755091?term=PACE; 2017.
[80] Ramar K, Rosen IM, Kirsch DB, Chervin RD, Carden KA, Aurora RN, et al. Medical cannabis and the treatment of obstructive sleep apnea: an American Academy of Sleep Medicine position statement. J Clin Sleep Med 2018;14(4):679–81.
[81] Zinchuk AV, Gentry MJ, Concato J, Yaggi HK. Phenotypes in obstructive sleep apnea: a definition, examples and evolution of approaches. Sleep Med Rev 2017;35:113–23.
[82] Therapix biosciences announces positive topline results from phase IIa clinical trial of THX-110 for obstructive sleep apnea program therapix biosciences. Available from: http://therapix.investorroom.com/2019-11-13-Therapix-Biosciences-Announces-Positive-Topline-Results-From-Phase-IIa-Clinical-Trial-of-THX-110-for-Obstructive-Sleep-Apnea-Program; 2019.

[83] Organisation Wh. WHO expert committee on drug dependence: critical review - delta-9-tetrahydrocannabinol 2018. Available from: https://www.who.int/medicines/access/controlled-substances/THCv1.pdf?ua=1; 25 Nov 2019.
[84] Gates P, Albertella L, Copeland J. Cannabis withdrawal and sleep: a systematic review of human studies. Subst Abuse 2016;37(1):255–69.
[85] Vandrey R, Smith MT, McCann UD, Budney AJ, Curran EM. Sleep disturbance and the effects of extended-release zolpidem during cannabis withdrawal. Drug Alcohol Depend 2011;117(1):38–44.
[86] Babson KA, Boden MT, Harris AH, Stickle TR, Bonn-Miller MO. Poor sleep quality as a risk factor for lapse following a cannabis quit attempt. J Subst Abuse Treat 2013;44(4):438–43.
[87] Budney AJ, Vandrey RG, Hughes JR, Moore BA, Bahrenburg B. Oral delta-9-tetrahydrocannabinol suppresses cannabis withdrawal symptoms. Drug Alcohol Depend 2007;86(1):22–9.
[88] Vandrey R, Stitzer ML, Mintzer MZ, Huestis MA, Murray JA, Lee D. The dose effects of short-term dronabinol (oral THC) maintenance in daily cannabis users. Drug Alcohol Depend 2013;128(1–2):64–70.
[89] Russo EB, Guy GW, Robson PJ. Cannabis, pain, and sleep: lessons from therapeutic clinical trials of Sativex®, a cannabis-based medicine. Chem Biodivers 2007;4(8):1729–43.
[90] Fogaça MV, Reis F, Campos A, Guimarães FS. Effects of intra-prelimbic prefrontal cortex injection of cannabidiol on anxiety-like behavior: involvement of 5HT1A receptors and previous stressful experience. Eur Neuropsychopharmacol 2014;24(3):410–9.
[91] Lutz B, Marsicano G, Maldonado R, Hillard CJ. The endocannabinoid system in guarding against fear, anxiety and stress. Nat Rev Neurosci 2015;16(12):705.
[92] Geffrey AL, Pollack SF, Bruno PL, Thiele EA. Drug–drug interaction between clobazam and cannabidiol in children with refractory epilepsy. Epilepsia 2015;56(8):1246–51.
[93] Gaston TE, Bebin EM, Cutter GR, Liu Y, Szaflarski JP, Program UC. Interactions between cannabidiol and commonly used antiepileptic drugs. Epilepsia 2017;58(9):1586–92.
[94] Taylor L, Gidal B, Blakey G, Tayo B, Morrison G. A phase I, randomized, double-blind, placebo-controlled, single ascending dose, multiple dose, and food effect trial of the safety, tolerability and pharmacokinetics of highly purified cannabidiol in healthy subjects. CNS Drugs 2018;32(11):1053–67.
[95] Spindle TR, Cone EJ, Goffi E, Weerts EM, Mitchell JM, Winecker RE, et al. Pharmacodynamic effects of vaporized and oral cannabidiol (CBD) and vaporized CBD-dominant cannabis in infrequent cannabis users. Drug and Alcohol Dependence 2020;211:107937.
[96] ProactiveInvestors. Zelira Therapeutics achieves primary endpoints of medicinal cannabis trial for insomnia. 2020. Available from: https://www.proactiveinvestors.com.au/companies/news/913189/zelira-therapeutics-achieves-primary-endpoints-of-medicinal-cannabis-trial-for-insomnia-913189.html.
[97] Comella CL. Restless legs syndrome: treatment with dopaminergic agents. Neurology 2002;58(suppl 1):S87–92.
[98] Cornelius JR, Tippmann-Peikert M, Slocumb NL, Frerichs CF, Silber MH. Impulse control disorders with the use of dopaminergic agents in restless legs syndrome: a case-control study. Sleep 2010;33(1):81–7.
[99] Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, Mechoulam R, Drucker-Colín R. Cannabidiol, a constituent of Cannabis sativa, modulates sleep in rats. FEBS (Fed Eur Biochem Soc) Lett 2006;580(18):4337–45.
[100] Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, Mechoulam R, Drucker-Colín R. The nonpsychoactive Cannabis constituent cannabidiol is a wake-inducing agent. Behav Neurosci 2008;122(6):1378.
[101] Murillo-Rodríguez E, Sarro-Ramírez A, Sánchez D, Mijangos-Moreno S, Tejeda-Padrón A, Poot-Aké A, et al. Potential effects of cannabidiol as a wake-promoting agent. Curr Neuropharmacol 2014;12(3):269–72.
[102] Murillo-Rodríguez E, Di Marzo V, Machado S, Rocha NB, Veras AB, Neto GA, et al. Role of N-Arachidonoyl-Serotonin (AA-5-HT) in sleep-wake cycle architecture, sleep homeostasis, and neurotransmitters regulation. Front Mol Neurosci 2017;10:152.
[103] Wisor JP. Dopamine and wakefulness: pharmacology, genetics, and circuitry. In: Handbook of experimental pharmacology. Springer International Publishing; 2018 [Internet].
[104] Vandrey R, Herrmann ES, Mitchell JM, Bigelow GE, Flegel R, LoDico C, et al. Pharmacokinetic profile of oral cannabis in humans: blood and oral fluid disposition and relation to pharmacodynamic outcomes. J Anal Toxicol 2017;41(2):83–99.
[105] Rogeberg O, Elvik R. The effects of cannabis intoxication on motor vehicle collision revisited and revised. Addiction 2016;111(8):1348–59.
[106] Arkell TR, Lintzeris N, Kevin RC, Ramaekers JG, Vandrey R, Irwin C, et al. Cannabidiol (CBD) content in vaporized cannabis does not prevent tetrahydrocannabinol (THC)-induced impairment of driving and cognition. Psychopharmacology 2019;236(9):2713–24.
[107] Machado Bergamaschi M, Helena Costa Queiroz R, Waldo Zuardi A, Crippa AS. Safety and side effects of cannabidiol, a Cannabis sativa constituent. Curr Drug Saf 2011;6(4):237–49.
[108] Taylor L, Crockett J, Tayo B, Checketts D, Sommerville K. Abrupt withdrawal of cannabidiol (CBD): a randomized trial. Epilepsy Behav 2020;104:106938.
[109] Rong C, Carmona NE, Lee YL, Ragguett R-M, Pan Z, Rosenblat JD, et al. Drug-drug interactions as a result of co-administering Δ9-THC and CBD with other psychotropic agents. Expet Opin Drug Saf 2018;17(1):51–4.
[110] Taylor L, Crockett J, Tayo B, Morrison G. A phase 1, open-label, parallel-group, single-dose trial of the pharmacokinetics and safety of cannabidiol (CBD) in subjects with mild to severe hepatic impairment. J Clin Pharmacol 2019;59(8):1110–9.

2.7 Addendum

Since publication of this systematic review, two key clinical studies exploring the effects of cannabinoids in the treatment of a sleep disorder: a 10:1:0.5 ratio of THC:CBN:CBD for insomnia disorder[1] and CBD for REM sleep behaviour disorder.[2]

First, a randomised, placebo-controlled, crossover trial examined the effects of 'ZTL-101' (containing 10 mg THC, 1 mg CBN and 0.5 mg CBD) nightly for two weeks in 23 participants with insomnia disorder.[1] Over half (52%) doubled the dose at the start of week 2. A significant improvement of 5.1 points on the ISI was observed with ZTL-101 treatment with self-reported improvements in sleep quality and 'feeling more rested/refreshed on waking' relative to placebo. On actigraphy, there was a 33.4 min increase in TST and 10.2 min decrease in WASO relative to placebo. No significant differences were observed on polysomnography except for a non-significant reduction in REM sleep and a significantly longer latency to REM sleep for ZTL-101 (124 min) compared with placebo. The latter may be at least partly attributed to a -56 min decrease in REM sleep latency from baseline (127 min) in the placebo group (71 min). Further, a relatively short washout period (one week) may have led to possible carryover effects[3 4] and contributed to an order effect, although this was not discussed. All participants correctly guessed the order in which they had received the active medication (not uncommon for cannabinoid trials).

Second, a 12-week randomised, placebo-controlled, parallel trial examined the effects of CBD (oral, 300 mg) in 33 individuals with PSG-confirmed REM sleep behaviour disorders and Parkinson's disease.[2] Primary outcomes included the difference in the average total number of nights with events suggestive of RBD per week on sleep diary and changes in the Clinical Global Impression (CGI) scale. No significant effect of CBD treatment was observed either primary outcome, however, an improvement in sleep satisfaction was observed in Weeks 4 and 8 compared to placebo. No improvements in anxiety or depressive symptoms were observed at this dose and there was no evidence of sleep architecture alterations in line with a previous study in healthy volunteers.[5]

References

1. Walsh JH, Maddison KJ, Rankin T, et al. Treating insomnia symptoms with medicinal cannabis: a randomized, crossover trial of the efficacy of a cannabinoid medicine compared with placebo. *Sleep* 2021;44(11):zsab149.
2. de Almeida CM, Brito MM, Bosaipo NB, et al. Cannabidiol for rapid eye movement sleep behavior disorder. *Mov Disord* 2021;36(7):1711-15.
3. Taylor L, Gidal B, Blakey G, et al. A phase I, randomized, double-blind, placebo-controlled, single ascending dose, multiple dose, and food effect trial of the safety, tolerability and pharmacokinetics of highly purified cannabidiol in healthy subjects. *CNS Drugs* 2018;32(11):1053-67.
4. Taylor L, Crockett J, Tayo B, et al. Abrupt withdrawal of cannabidiol (CBD): a randomized trial. *Epilepsy Behav* 2020;104:106938.
5. Linares IM, Guimaraes FS, Eckeli A, et al. No acute effects of cannabidiol on the sleep-wake cycle of healthy subjects: a randomized, double-blind, placebo-controlled, crossover study. *Front Pharmacol* 2018;9:315.

3. Medicinal Cannabis Use Patterns for Sleep Disorders in Australia: Results of the CAMS-20 Survey

Medicinal Cannabis Use Patterns for Sleep Disorders in Australia: Results of the CAMS-20 Survey

Anastasia Suraev[1,2,3], Llewellyn Mills[5,6], Sarah V. Abelev[1], Thomas R. Arkell[7], Nicholas Lintzeris[5,6]*, and Iain S. McGregor[1,2,3]*

**Co-senior authorship*

[1] The University of Sydney, Lambert Initiative for Cannabinoid Therapeutics, Sydney, NSW, Australia

[2] The University of Sydney, Faculty of Science, School of Psychology, Sydney, NSW, Australia

[4] The University of Sydney, Brain and Mind Centre, Sydney, NSW, Australia

[5] Drug and Alcohol Services, South East Sydney Local Health District, Sydney, NSW, Australia

[6] Department of Addiction Medicine, Faculty Medicine and Health, University of Sydney, NSW, Australia

[7] Centre for Human Psychopharmacology, Swinburne University of Technology, Hawthorn, VIC, Australia

3.1 Abstract

The use of cannabis for medicinal purposes is growing in Australia, but consumer behaviours and patterns of use, particularly for sleep disorders, are poorly understood. Here we present the results of a subanalysis of Australian consumers (n=1600) who self-reported using medicinal cannabis to treat a sleep disorder from the 'Cannabis as Medicine Survey' 2020-2021 (CAMS-20). When asked to specify up to seven different conditions they were treating with medicinal cannabis, a total of 1030 (64%) respondents (median [IQR] age, 44 [21] years) selected a sleep disorder, with 'insomnia disorder' (85.5%), 'sleep-related movement disorders' (26%) and 'sleep-related breathing disorders' (11.1%) being the most common. Only 165 (16.8%) respondents selected a sleep disorder as the main health condition being treated. Those using medicinal cannabis for a sleep disorder were significantly younger and more likely to use both prescribed and illicit forms of medicinal cannabis, inhaled routes of administration (i.e., smoking or vaping), and THC-dominant products compared to those using medicinal cannabis for any other indication. Most respondents using cannabis for sleep reported a concomitant reduction in the use of benzodiazepines and alcohol. Binary logistic regression showed that respondents who predominantly used inhaled routes of administration, and concomitant use of medicinal cannabis for pain, mental health and/or substance use disorder, or a gastrointestinal disorder were significantly more likely to also use medicinal cannabis to treat a sleep disorder. Overall, these results suggest that sleep disorders are often being treated secondary to another primary condition (i.e., other medicinal or psychiatric disorder) and that use of inhaled methods, THC-dominant products, and illicit sources of medicinal cannabis are common among people with sleep disorders.

3.2 Introduction

Sleep is a biological necessity that enables restorative functions that are essential for normal daytime function.[1] Approximately 30% of the general population report poor sleep, which may be attributed to lifestyle choices, environmental factors, and/or the presence of an untreated sleep disorder or other medicinal complaints such as pain.[2] Treatment typically involves both pharmacological and behavioural approaches for optimal management. While behavioural interventions such as cognitive behaviour therapy for insomnia (CBT-I) are the mainstay of treatment,[3] patients often seek short-term strategies to maintain normal daytime function; this frequently includes adjunctive use of pharmaceutical sleep aids such as benzodiazepines and Z-drugs. However, such drugs are often associated with undesirable side effects and should not be used long-term,[4] leading to a rise in the popularity of alternative treatments.

Despite limited robust clinical evidence for the use of cannabis and its constituents in the treatment of sleep disorders[5], medicinal cannabis is becoming an increasingly popular alternative to common sleep aids.[6 7] In the US, a recent survey showed that 74% of people accessing cannabis through adult-use markets in Colorado reported effective treatment for sleep, with a concomitant reduction in the use of prescription sleep aids.[6] In Canada, 92.6% of patients using prescribed medicinal cannabis reported a significant improvement in their sleep after six weeks of treatment as assessed using the Pittsburgh Sleep Quality Index.[8] In Australia, consumers surveys in 2016[9] and 2018[10] showed that sleep disorders were the third most common primary indication treated with medicinal cannabis, after pain and mental health conditions. This mirrors approval rates for prescription medicinal cannabis via the Australian Therapeutic Goods Administration (TGA), the federal regulator, with sleep disorders the third most common indication after pain and anxiety.[11]

Despite increasing access to medicinal cannabis in Australia, consumer behaviours and patterns of use of both prescribed and illicit medicinal cannabis for sleep disorders are poorly understood. Here we describe the results of a subanalysis of Australians who self-reported using

cannabis, prescribed and/or illicit, for a sleep disorder in an online consumer survey, 'Cannabis as Medicine Survey' 2020-2021 (CAMS-20).

The term *medicinal cannabis* used in this paper refers to any licit or illicit cannabis-based product (including plant matter) used to treat or alleviate the symptoms of a self-identified health condition. In the Australian context, pharmaceutical-grade cannabis products are strictly regulated, federally approved, and quality-assured products that are only available on prescription via a medicinal doctor. All other cannabis products are illicit and unregulated (i.e., of unknown composition).

3.3 Methods

Study design

The current investigation was conducted using data collected within the 'Cannabis as Medicine Survey' 2020-2021 (CAMS-20); a web-based, cross-sectional survey of Australians who self-reported using cannabis for medicinal reasons conducted every two years since 2016 (see CAMS-16[9], CAMS-18[10] and CAMS-20 survey[12]). The full methodology and main findings of the CAMS-20 survey are published elsewhere.[12] The study was approved by the University of Sydney Human Research Ethics Committee (2018/544).

Recruitment and eligibility

Respondents were eligible to participate if they: (a) provided informed consent, (b) were aged ≥18 years, (c) resided in Australia, and (d) self-identified as a user of licit or illicit cannabis or a cannabis-based product for a medicinal purpose within the previous 12 months. The CAMS-20 survey was available online over a 5-month period between September 2020 – January 2021 and advertised via social media and consumer group pages, at consumer and professional forums, and through several private medicinal cannabis clinics.

Survey design

The original survey was developed by the investigators and updated with new questions to reflect the evolving regulatory changes in Australia.[13] Data were collected and managed using Research Electronic Data Capture (REDcap), a secure web-based platform.[14] Individuals were asked about their (a) demographic characteristics and general health (including alcohol and tobacco use); (b) current and lifetime patterns of medicinal cannabis use including how they accessed their medicinal cannabis products ('Prescribed Only', 'Illicit Only' or 'Prescribed and Illicit') as well as specific questions relating to those products such as route of administration and perceived cannabinoid composition; (c) medicinal conditions for which they were using medicinal cannabis; (d) perceived change in their sleep problems after starting use of medicinal cannabis products on a 7-point Likert scale ranging from 'very much worse' to 'very much better'; and (e) change in benzodiazepine and alcohol use since starting medicinal cannabis (the full survey is available in the **Supplementary Materials**).

Data analysis

Data were analysed using SPSS version 26 (IMB Corp., Armonk, NY) and figures were created using GraphPad Prism version 9 (GraphPad Inc., San Diego, CA). Only valid responses were reported. Responses relating to perceived cannabinoid composition were collapsed into: 'THC-dominant, 'CBD-dominant, 'THC/CBD-equivalent, and 'Unknown'. Similarly, responses relating to route of administration were collapsed into: 'Oral Only', 'Oral & Inhaled', 'Inhaled Only' and 'Other' (e.g., topical, suppository). Independent t-tests were used to compare normally distributed (i.e., Shapiro–Wilk test, $p>0.05$) demographics, cannabis use patterns, and psychosocial characteristics between those using medicinal cannabis to treat a sleep disorder versus all other reasons. Non-normally distributed data were analysed using the Mann–Whitney U test. Categorical variables were compared using Fisher's exact test or Chi-square test. Chi-square post-hoc analysis using adjusted standardised residuals with Bonferroni correction was conducted for variables with

more than two categories.[15] Adjusted p values for these variables are described in **Table 1**. Binary logistic regression models were used to explore the influence of demographics (age and gender), cannabis use characteristics (cannabis user type, route of administration, cannabinoid composition), other health conditions treated with medicinal cannabis (pain, mental health and/or substance abuse, gastrointestinal, cancer, neurological, other), alcohol use, and benzodiazepine use on the odds of using medicinal cannabis to treat a sleep disorder. Only statistically significant odds ratios are reported in-text (p <0.05).

3.4 Results

In total, 1600 respondents completed and provided valid responses to the larger CAMS-20 survey. Of these, 1030/1600 (64.4%) respondents self-reported using medicinal cannabis to treat a sleep disorder when asked to specify up to seven different conditions they were medicating with cannabis. **Figure 1** shows the types of sleep disorders being treated with medicinal cannabis. 'Insomnia disorder' (881/1030, 85.5%) was the most common sleep disorder followed by 'sleep-related movement disorders' (268/1030, 26%), 'sleep-related breathing disorders' (114/1030, 11.1%), 'circadian rhythm sleep disorders' (109/1030, 10.6%), 'parasomnias' (84/1030, 8.2%), 'narcolepsy' (15/1030, 1.5%), and 'other sleep disorder' (7/1030, 0.7%). Only 165/982 (16.8%) reported using medicinal cannabis to treat a sleep disorder as their main health condition (**Supplementary Table S1**). Main indications selected by those with a sleep disorder were as follows: 'pain' (412/982, 42%), 'mental health and/or substance use disorder' (319/982, 32.5%), 'neurological disorder' (68/982, 6.9%), 'cancer' (23/982, 2.3%), 'gastrointestinal condition' (22/982, 2.2%), and 'other' (55/982, 5.6%). Post-traumatic stress disorder (PTSD) was the second most common mental health disorder condition treated with medicinal cannabis (54/984, 5.5%).

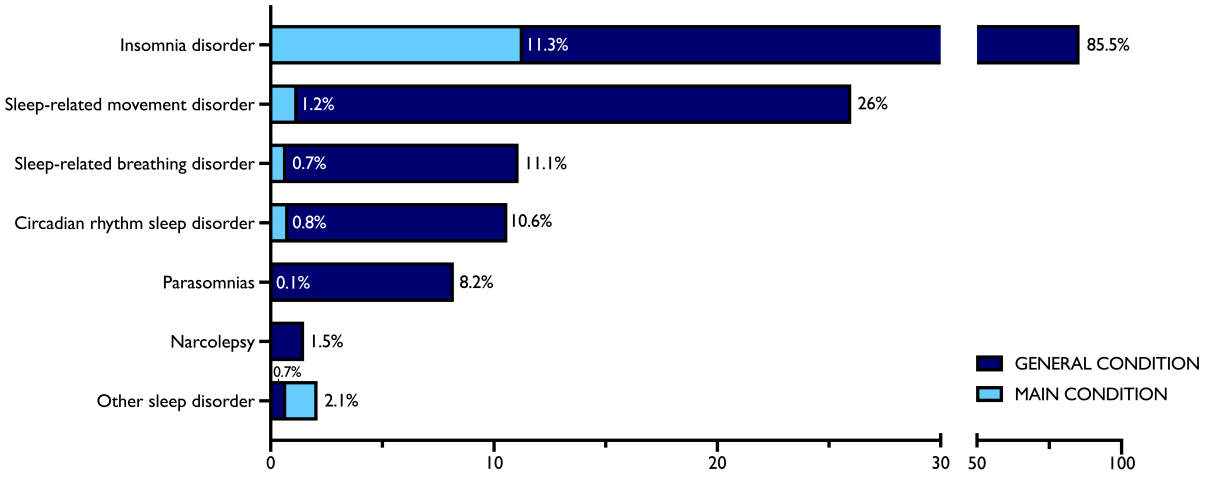

Figure 1 *Types of sleep disorders being treated with medicinal cannabis as a main health condition (n=165) or general health condition (n=1,030). For main condition, respondents could only select one main condition that they treated with medicinal cannabis. For general condition, respondents could select up to seven conditions from a structured list of following: 'sleep', 'pain', 'cancer', 'mental health and/or substance use', 'neurological', 'gastrointestinal', and 'other'.*

As **Table 1** indicates, the majority of respondents treating sleep disorder were male with a median [interquartile range, IQR] age of 44 [21]. Most of these respondents (62.8%) accessed their medicinal cannabis via illicit sources ('Illicit Only'; 647/1030), 10.7% were using medicinal cannabis products prescribed by a medicinal doctor ('Prescribed Only'; 110/1030), and 26.5% had used both in the preceding 12 months ('Prescribed and Illicit'; 273/1030). Those using medicinal cannabis for a sleep disorder were younger compared to those using medicinal cannabis for all other reasons (median [IQR]; 44 [21] vs 48 [23], $p<0.001$). Respondents using medicinal cannabis for a sleep disorder were also younger when they first started using cannabis (for any reason and for a medicinal reason) compared to those using medicinal cannabis for all other reasons (16 [6] vs 19 [10] and 35 [24]) vs 40 [24]) respectively), both $p<0.001$. More respondents using medicinal cannabis to treat a sleep disorder were using both prescribed and illicit forms of medicinal cannabis (26.5% versus 20.2%, $p=0.005$), inhaled routes of administration (52.5% vs 40.2%, $p<0.001$), and THC-dominant medicinal cannabis (32.1% vs 24.7%, $p=0.002$) than respondents using medicinal cannabis for all other reasons.

Table 1 Demographic and other substance use characteristics of the CAMS-20 survey respondents who self-reported using cannabis to treat a sleep disorder vs all other reasons.

Characteristic	Valid n	Sleep Disorders	Valid n	All Other Reasons	*p*
Number of respondents, n (%)		1030		570	
Age (y), Md [IQR]	1030	44 [21]	570	48 [23]	.001
Gender, n (%)	1030		570		.951
Male		539 (52.3%)		303 (53.3%)	
Female		478 (46.4%)		260 (45.6%)	
Unspecified		13 (1.3%)		7 (1.2%)	
Attained tertiary qualification, n (%)	1030	825 (80.1%)	570	431 (75.6%)	.037
Current employment, n (%)	1030	509 (49.4%)	570	272 (47.7%)	.515
Age (y), first used cannabis (any reason), Md [IQR]	910	16 [6)]	495	18 [10]	.001
Age (y), first used cannabis (medicinal reason), Md [IQR]	911	35 [24]	495	40 [24]	.001
MC use as % of total cannabis use, Md, range	906	87.1, 13-100	490	89.3, 4-100	.013
[a]Cannabis use type, n (%)	1030		570		
Prescribed Only		110 (10.7%)		103 (18.1%)	<.001
Prescribed & Illicit		273 (26.5%)		115 (20.2%)	.005
Illicit Only		647 (62.8%)		352 (61.8%)	.689
[b]Route of administration, n (%)	1030		527		
Oral Only		347 (36%)		280 (53.1%)	<.001
Oral & Inhaled		95 (9.9%)		25 (4.7%)	<.001
Inhaled Only		506 (52.4%)		212 (40.3%)	<.001
Other		16 (1.7%)		10 (1.9%)	.764
[c]Cannabinoid type, n (%)	943		511		
CBD-dominant		175 (18.6%)		151 (29.5%)	<.001
THC/CBD-equivalent		165 (17.5%)		107 (20.9%)	.107
THC-dominant		303 (32.1%)		126 (24.7%)	.002
Unknown		300 (31.8%)		127 (24.9%)	<.001
Alcohol use in last 28 days, M (SD) [IQR]	862	5.6 (7.9) [8]	469	6.1 (8.4) [9]	.153
Standard drinks per day, M (SD) [IQR]	514	3.9 (4.2) [3]	305	3.5 (3.5) [2]	.287
Cigarette (tobacco) use in last 28 days, M (SD) [IQR]	863	7.0 (11.6) [15]	469	6.3 (11.4) [2]	.130
Cigarettes (tobacco) per day, M (SD) [IQR]	261	9.1 (7.9) [13]	119	11.5 (9.6) [10]	.095

IQR, interquartile range M, mean; Md, median; MC, medicinal cannabis; SD, standard deviation; y, year
Nb 'Inhaled' includes 'smoking' and 'vaporising'
Adjusted p-value: [a]p=.008; [b]p=.006, [c]p=.006

Figure 2 shows the route of administration and cannabinoid composition for illicit and prescribed medicinal cannabis products. Prescribed medicinal cannabis products were consumed predominantly by oral routes (246/361, 68.1%) whereas illicit products were mostly consumed via inhaled routes (585/863, 67.8%; i.e., smoking or vaporising). Cannabinoid composition of prescribed medicinal cannabis was predominantly 'THC/CBD equivalent' (128/322, 39.7%) or 'THC-dominant' (97/322, 30.1%) while illicit medicinal cannabis was largely 'THC-dominant' (285/833, 34.3%) or 'Unknown' (280/831, 33.7%).

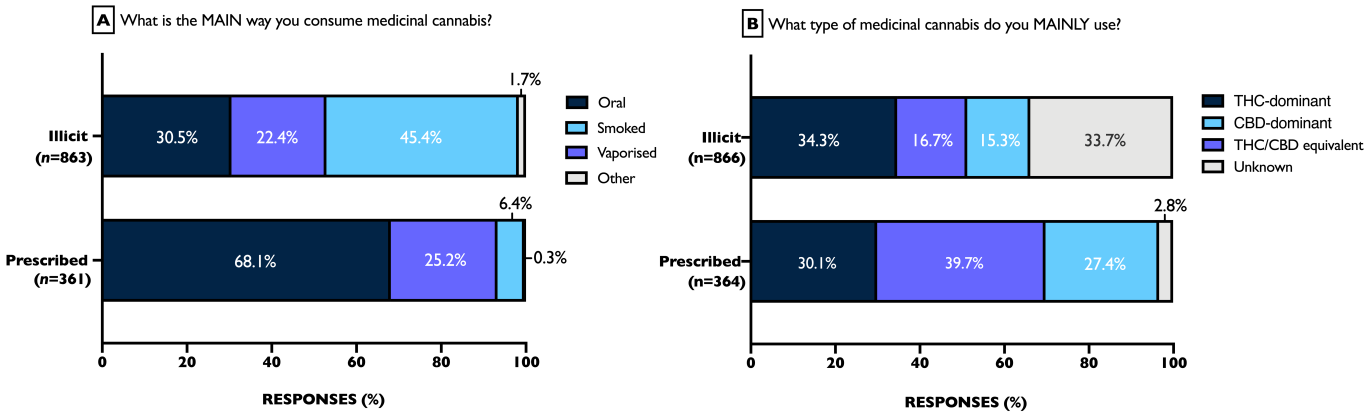

Figure 2 *(A) Main route of administration respondents used to consume prescribed (n=361) and illicit (n=863) medicinal cannabis products to treat a sleep disorder. (B) Cannabinoid composition of the prescribed (n=364) and illicit (n=866) medicinal cannabis products used to treat a sleep disorder.*

Figure 3 shows that the majority of participants who self-reported using medicinal cannabis to treat a sleep disorder reported an improvement in sleep since commencing medicinal cannabis, as reported on the Patient Global Impression of Change Scale (PGIC)[16] (93.5% for 'Prescribed' product, 96.4% for 'Illicit' product). An overwhelming proportion also reduced their use of benzodiazepines (391/414, 95%) while alcohol intake decreased in 62.8% (321/514), remained unchanged in 37.2% (191/514), and increased in 0.4% (2/514) of respondents (**Supplemental Figure S1**).

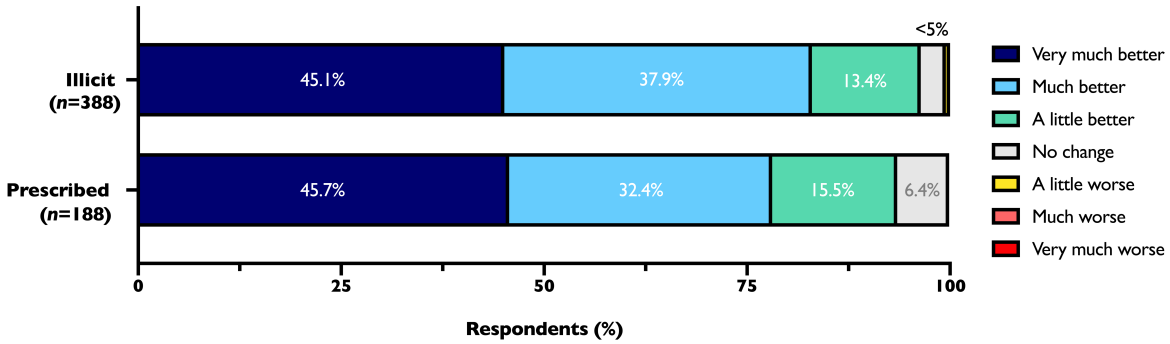

Figure 3 *Self-reported change in the symptom, 'sleep problems', since starting prescribed (n=188) and illicit (n=388) medicinal cannabis products.*

Table 2 shows the results of a binary logistic regression model that assessed the relationship between respondent characteristics and the odds of using medicinal cannabis to treat a sleep disorder. Those using illicitly sourced medicinal cannabis products were twice as likely to use medicinal cannabis to treat a sleep disorder (OR=2.04, 95% CI: 1.06 – 3.92, p=.032). Concomitant use of medicinal cannabis for pain (OR=2.15, 95% CI: 1.49 – 3.09, p<0.001), mental health and/or substance use disorder (OR=2.51, 95% CI: 1.78 – 3.55, p<0.001), and gastrointestinal disorder (OR=2.30, 95% CI: 1.38 – 3.85, p=0.001) significantly increased the odds of also using medicinal cannabis to treat a sleep disorder.

Table 2 Associations between respondent characteristics and using medicinal cannabis to treat a sleep disorder

Variable	Odds ratio (95% CI)	*p*-value
Age	-0.99 (0.98 – 1.01)	.379
Gender		
Male (vs female)	-0.88 (0.62 – 1.25)	.472
Unspecified (vs female)	-0.53 (0.14 – 2.07)	.365
User type		
Prescribed + Illicit (vs Prescribed Only)	1.57 (0.93 – 2.63)	.090
Illicit Only (vs Prescribed Only)	**2.04 (1.06 – 3.92)**	**.032**
Using MC for other condition		
Pain	**2.15 (1.49 – 3.09)**	**<.001**
Mental health and/or substance abuse	**2.51 (1.78 – 3.55)**	**<.001**
Gastrointestinal	**2.3 (1.38 – 3.85)**	**.001**
Cancer	1.28 (0.69 – 2.38)	.436
Neurological	-0.98 (0.61 – 1.60)	.993
Other	-0.86 (0.54 – 1.36)	.524
Route of administration		
Inhaled & Oral (vs Oral Only)	2.40 (0.94 – 6.21)	.068
Inhaled Only (vs Oral Only)	1.17 (0.74 – 1.87)	.500
Other (vs Oral Only)	1.00 (0.24 – 4.22)	.998
Main type of cannabis used		
THC/CBD-equivalent (vs. CBD-dominant)	-0.98 (0.60 – 1.63)	.953
THC-dominant (vs. CBD-dominant)	1.12 (0.67 – 1.90)	.665
Unknown (vs. CBD-dominant)	1.09 (0.64 – 1.85)	.765
Benzodiazepine use	1.33 (0.93 – 1.82)	.124
Alcohol use (in the last 28 days)	1.01 (0.99 – 1.03)	.289
Alcohol use (number of standard drinks)	1.00 (0.97 – 1.05)	.764

3.5 Discussion

The present study examined the characteristics and cannabis use patterns among a sample of Australian medicinal cannabis users with a self-reported sleep disorder, recruited as part of our larger CAMS-20 survey.[12] Results showed just under 65% of respondents who completed the survey were using medicinal cannabis to treat a sleep disorder, with insomnia disorder and restless legs syndrome (RLS) being the most common. Although evidence for the use of cannabinoids to treat a sleep disorder is limited, a recent randomised, controlled trial showed that 2-week treatment with combined 10 mg THC, 1 mg cannabinol (CBN), and 0.5 mg CBD significantly improved insomnia symptoms in patients with chronic insomnia as assessed on the Insomnia Severity Index.[17] Three pre-registered randomised, placebo-controlled trials are currently underway administering either CBD alone[18-20], CBD-terpene combination[21] or combined THC and CBD formulation[22] in patients with insomnia; all oral formulations. Even fewer studies have examined the utility of cannabinoids for RLS. In two case series combining 18 patients with treatment-resistant RLS, smoked cannabis (i.e., THC-dominant products) was self-rated as more efficacious in improving RLS symptoms than sublingual CBD.[23 24] The evidence base for cannabis in treating other sleep disorders has been recently reviewed elsewhere.[25]

Our survey data showed that Australians using medicinal cannabis for sleep disorders are more likely to use a mix of both illicit and prescribed forms of medicinal cannabis, inhaled forms of administration (i.e., smoking or vaping), and THC-dominant products compared to those using medicinal cannabis for other indications. A recent analysis of medicinal cannabis prescriptions in Australia showed approvals for sleep disorders were typically for flower products (i.e., consumed via inhalation) and predominantly Schedule 8 products (i.e., containing >2% THC).[26] THC is known to increase subjective ratings of feeling 'drowsy' or 'sleepy/tired' after oral and smoked/vaporised ingestion.[27,28] Inhalation produces much faster onset and shorter duration of

subjective effects with greater bioavailability (2%-56%) and higher peak concentrations of THC in blood, all within minutes of exposure.[29] This allows individuals to self-titrate to the desired effect with multiple smaller doses that have a rapid effect. In contrast, oral ingestion produces slower and less predictable onset and longer duration of subjective effects (peak concentration occurring at 2-4 hours),[30] compared to inhaled methods. Low bioavailability (4%-19%) and inter- and intra-individual variability in absorption of orally ingested cannabis make dose titration difficult.[31] Nonetheless, oral ingestion tends to be the preferred method among medicinal cannabis users due to its discrete nature and lack of respiratory side effects associated with smoking and vaping.[10 32] As noted above, all recently published and currently ongoing clinical trials examining the use of cannabinoid treatments for insomnia utilise oral formulations.

Our survey also found that only 16.8% of respondents selected a sleep disorder as their main condition suggesting that sleep disorders were commonly being treated secondary to a primary health condition such as chronic pain or psychiatric disorder (e.g., PTSD). This may explain the high rate of medicinal cannabis prescription for sleep disorders;[11] aimed to improve sleep amidst a range of other symptoms with a primary condition such as pain, anxiety, or depression. For example, nabiximols (*Sativex*), an oromucosal spray delivering equal parts THC and CBD, improves short-term outcomes in individuals with sleep disturbances secondary to chronic pain (e.g., neuropathic pain, spasticity in multiple sclerosis, rheumatoid arthritis).[33] Sleep problems are often exacerbated by comorbid illness, often in a bi-directional manner. Sleep disturbance is a common symptom of chronic pain and/or a psychiatric condition,[34 35] and is associated with negative daytime consequences such as fatigue, poor concentration, and mood disturbance.[36,37] In turn, poor sleep is hypothesised to promote pain amplification via its impact on various neurobiological systems influencing nociceptive processing[34] and increase the risk of developing depression.[38] Of note, sleep disturbance is a known risk factor for relapse in addictions (e.g., alcohol)[39] and a common residual symptom following withdrawal from long-term use of benzodiazepines for insomnia.[40] Sleep disturbance is also a key symptom during cannabis

withdrawal,[41] and it is therefore possible that a small proportion of respondents were using medicinal cannabis to treat a sleep disorder arising *from* cannabis dependence and withdrawal. Further research efforts are needed to explore the short- and long-term safety of medicinal cannabis in the treatment of sleep disorders.

Despite the overwhelming majority of participants reporting improvement in their sleep problems after starting medicinal cannabis (93.5% for prescribed products; 96.4% for illicit products), the continued reliance on illicit cannabis raises some safety concerns. Illicit (unregulated) cannabis products are often of unknown strength, composition and quality posing unpredictable risks to the consumer.[42] The overall CAMS-20 survey findings highlighted some advantages in prescribed over illicit cannabis use including safer routes of administration, access to quality-assured products of known composition, and better communication with and safety monitoring from healthcare providers.[12]

Limitations include use of self-report data that may be associated with inaccurate information such as incorrect diagnosis and/or misinterpretation of efficacy and side effects which may not be generalisable to those with a formal diagnosis. Convenience sampling in a survey may produce selection bias whereby those who are more likely to report favourable experiences with medicinal cannabis complete the survey. Responder fatigue due to the length of the survey also resulted in higher rates of missing data. Despite this, this survey provided useful and novel insights into the patterns of both licit and illicit use of medicinal cannabis for sleep disorders in Australia four years following legislation allowing access to medicinal cannabis.

Conclusions

Sleep disorders are one of the top three leading indications for approvals for MC in Australia after pain and anxiety. The present study provides a snapshot of medicinal cannabis use for sleep disorders among a sample of individuals in Australia four years following major regulatory changes that allowed patient access to legal medicinal cannabis. These results suggest that sleep

disorders are often being treated secondary to a primary condition such as a chronic pain, or a mental health or substance abuse problem. The use of inhaled routes of administration, THC-dominant products, and illicit sources of medicinal cannabis are common among people with sleep disorders. As prescribing rates for medicinal cannabis rise dramatically each year in Australia, it is imperative that rigorously controlled trials using quality-controlled products are conducted to better explore the efficacy and safety of cannabis use in patients with sleep disorders

Acknowledgements

This work was supported by the Division of Addiction Medicine, University of Sydney, and the Lambert Initiative for Cannabinoid Therapeutics, a philanthropically funded center at the University of Sydney, NSW, Australia. We would like to acknowledge the important contribution of the late Associate Professor David Allsop in the creation of the original CAMS-16 questionnaire, upon which the CAMS-20 questionnaire was based. The authors gratefully acknowledge the contributions of Dr Melissa Benson, Rhys Cohen, and Dr Dilara Bahceci in this work, and the services and forums that distributed information regarding the survey. We would also like to thank the participants who gave their time in completing the survey.

Conflict of Interest

NL has received research funding from Camurus and Indivior for unrelated research. ISM is Academic Director of the Lambert Initiative for Cannabinoid Therapeutics, a philanthropically funded research program at the University of Sydney. He has served as an expert witness in various medicolegal cases involving cannabis and has received consulting fees from Medicinal Cannabis Industry Australia (MCIA) and Janssen. He currently acts as an advisor/consultant to Kinoxis Therapeutics, Psylo and Emyria. He reports research grants and salary support from the Australian National Health and Medicinal Research Council (NHMRC) and from Lambert Initiative for Cannabinoid Therapeutics. He is an inventor on patents WO2018107216A1 and WO2017004674A1, licensed to Kinoxis Therapeutics involving use of novel small molecules (non-cannabinoid) to treat addictions and social deficits. ISM also has patents WO2020102857A1 and WO2021042178A1 related to use of small molecules (non-cannabinoid) for treating weight gain and opioid withdrawal, as well as patents WO2019227167 and WO2019071302 issued, which relate to cannabinoid therapeutics. AS has received consulting fees from the Medicinal Cannabis Industry Australia (MCIA). All other authors have no competing financial or non-financial interests to declare.

3.6 References

1. Ramar K, Malhotra RK, Carden KA, et al. Sleep is essential to health: an American Academy of Sleep Medicine position statement. *J Clin Sleep Med* 2021;17(10):2115-19.
2. Liu Y, Wheaton AG, Chapman DP, et al. Prevalence of healthy sleep duration among adults—United States, 2014. *Morb Mortal Wkly Rep* 2016;65(6):137-41.
3. Baglioni C, Altena E, Bjorvatn B, et al. The European Academy for Cognitive Behavioural Therapy for Insomnia: An initiative of the European Insomnia Network to promote implementation and dissemination of treatment. *J Sleep Res* 2020;29(2):e12967.
4. De Crescenzo F, D'Alò GL, Ostinelli EG, et al. Comparative effects of pharmacological interventions for the acute and long-term management of insomnia disorder in adults: a systematic review and network meta-analysis. *Lancet* 2022;400(10347):170-84.
5. Suraev AS, Marshall NS, Vandrey R, et al. Cannabinoid therapies in the management of sleep disorders: a systematic review of preclinical and clinical studies. *Sleep Med Rev* 2020;53:101339.
6. Bachhuber M, Arnsten JH, Wurm G. Use of cannabis to relieve pain and promote sleep by customers at an adult use dispensary. *J Psychoactive Drugs* 2019;51(5):400-04.
7. Piper BJ, DeKeuster RM, Beals ML, et al. Substitution of medicinal cannabis for pharmaceutical agents for pain, anxiety, and sleep. *J Psychopharmacol* 2017;31(5):569-75.
8. Cahill SP, Lunn SE, Diaz P, et al. Evaluation of patient reported safety and efficacy of cannabis from a survey of medicinal cannabis patients in Canada. *Front Public Health* 2021;9
9. Lintzeris N DJ, Elias N, Arnold JC, McGregor IS, Allsop DJ. Findings from the Cannabis as Medicine Survey (CAMS-16): an online survey of medicinal cannabis use in Australia. *Med J Aust* 2018; 209, 211-216.

10. Lintzeris N, Mills L, Suraev A, et al. Medicinal cannabis use in the Australian community following introduction of legal access: The 2018-2019 Online Cross-Sectional Cannabis as Medicine Survey (CAMS-18). *Harm Reduct J* 2020; 1-12.2020.

11. MacPhail S, Bedoya-Perez MA, Cohen R, et al. Medicinal Cannabis Prescribing in Australia: An analysis of trends over the first five years. *Front Pharmacol*:1368.

12. Lintzeris N, Mills L, Abelev SV, et al. Medicinal cannabis use in Australia: consumer experiences from the online cannabis as medicine survey 2020 (CAMS-20). *Harm Reduct J* 2022;19(1):1-10.

13. Arnold JC, Nation T, McGregor IS. Prescribing medicinal cannabis. *Aust Prescr* 2020;43(5):152.

14. Harris P.A., Taylor R., Thielke R., et al. Research electronic data capture (REDCap) – A metadata-driven methodology and workflow process for providing translational research informatics support. *J Biomed Inform* 2009;42(2):377-81.

15. Beasley TM, Schumacker RE. Multiple regression approach to analyzing contingency tables: Post hoc and planned comparison procedures. *J Exp Educ* 1995;64(1):79-93.

16. Hurst H, Bolton J. Assessing the clinical significance of change scores recorded on subjective outcome measures. *J Manipulative Physiol Ther* 2004;27(1):26-35.

17. Walsh JH, Maddison KJ, Rankin T, et al. Treating insomnia symptoms with medicinal cannabis: a randomized, crossover trial of the efficacy of a cannabinoid medicine compared with placebo. *Sleep* 2021;44(11):zsab149.

18. Australian New Zealand Clinical Trials Registry. A phase IIb double-blind, randomised placebo-controlled clinical trial to evaluate the efficacy and safety of a botanical cannabidiol (CBD) for sleep disturbances in a healthy population. ACTRN12621000632897. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12621000632897

19. National Institutes of Health Clinical Center. The Cannabidiol use for relief of short term insomnia (CANREST). NCT05253417. ClinicalTrials.gov. National Institutes of Health; 2022. Accessed 19 September, 2022. https://clinicaltrials.gov/ct2/show/NCT05253417

20. Australian New Zealand Clinical Trials Registry. Cannabidiol (CBD) treatment for insomnia. ACTRN12620000070932. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12620000070932.

21. National Institutes of Health Clinical Center. Evaluation of an oral cannabidiol (CBD)-terpene formulation on sleep physiology in participants with insomnia. NCT05233761. ClinicalTrials.gov. National Institutes of Health; 2022. Accessed 19 September, 2022. https://clinicaltrials.gov/ct2/show/NCT05233761

22. Australian New Zealand Clinical Trials Registry. A single-dose, double-blind, placebocontrolled, randomised, crossover study of an oral cannabis-based medicine (ETC120) on sleep, cognition, and next-day function in adults with chronic insomnia disorder. ACTRN12619000714189. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12619000714189.

23. Current R. Cannabis for restless legs syndrome: a report of six patients. *Sleep Med.* 2017;36:182e83.

24. Ghorayeb I. More evidence of cannabis efficacy in restless legs syndrome. *Sleep Breath.* 2020;24(1):277-79.

25. Lavender I, McGregor IS, Suraev A, et al. Cannabinoids, insomnia, and other sleep disorders. *Chest* 2022

26. Therapeutic Goods Administration. Freedom of Information Disclosure Log. FOI 3732. Australian Government. Accessed 19 September, 2022. https://www.tga.gov.au/foi-disclosure-log-0

27. Spindle TR, Martin EL, Grabenauer M, et al. Assessment of cognitive and psychomotor impairment, subjective effects, and blood THC concentrations following acute administration of oral and vaporized cannabis. *J Psychopharmacol* 2021;35(7):786-803.

28. Spindle TR, Cone EJ, Schlienz NJ, et al. Acute effects of smoked and vaporized cannabis in healthy adults who infrequently use cannabis: a crossover trial. *JAMA Netw Open* 2018;1(7):e184841-e41.

29. Huestis MA. Human cannabinoid pharmacokinetics. *Chem Biodivers* 2007;4(8):1770.

30. Schlienz NJ, Spindle TR, Cone EJ, et al. Pharmacodynamic dose effects of oral cannabis ingestion in healthy adults who infrequently use cannabis. *Drug Alcohol Depend* 2020;211:107969.

31. Perucca E, Bialer M. Critical aspects affecting cannabidiol oral bioavailability and metabolic elimination, and related clinical implications. *CNS Drugs* 2020;34(8):795-800.

32. Russell C, Rueda S, Room R, et al. Routes of administration for cannabis use–basic prevalence and related health outcomes: A scoping review and synthesis. *Int J Drug Policy* 2018;52:87-96.

33. Russo EB, Guy GW, Robson PJ. Cannabis, Pain, and Sleep: Lessons from Therapeutic Clinical Trials of Sativex®, a Cannabis-Based Medicine. *Chem Biodivers* 2007;4(8):1729-43.

34. Haack M, Simpson N, Sethna N, et al. Sleep deficiency and chronic pain: potential underlying mechanisms and clinical implications. *Neuropsychopharmacology* 2020;45(1):205-16.

35. Freeman D, Sheaves B, Waite F, et al. Sleep disturbance and psychiatric disorders. *Lancet Psychiat* 2020;7(7):628-37.

36. Bjorvatn B, Jernelöv S, Pallesen S. Insomnia–a heterogenic disorder often comorbid with psychological and somatic disorders and diseases: a narrative review with focus on diagnostic and treatment challenges. *Front Psychol* 2021:289.

37. Husak AJ, Bair MJ. Chronic pain and sleep disturbances: A pragmatic review of their relationships, comorbidities, and treatments. *Pain Med* 2020;21(6):1142-52.

38. Franzen PL, Buysse DJ. Sleep disturbances and depression: risk relationships for subsequent depression and therapeutic implications. *Dialogues Clin Neurosci* 2022

39. Gillin JC, Smith TL, Irwin M, et al. Increased pressure for rapid eye movement sleep at time of hospital admission predicts relapse in nondepressed patients with primary alcoholism at 3-month follow-up. *Arch Gen Psychiatry* 1994;51(3):189-97.

40. Morin CM, Bélanger L, Bastien C, et al. Long-term outcome after discontinuation of benzodiazepines for insomnia: a survival analysis of relapse. *Behav Res Ther* 2005;43(1):1-14.

41. Allsop D, Norberg MM, Copeland J, et al. The cannabis withdrawal scale development: patterns and predictors of cannabis withdrawal and distress. *Drug Alcohol Depend* 2011;119:123-29.

42. Suraev A, Benson MJ, Martin L, et al. Determination of contaminants in artisanal cannabis products used for childhood epilepsy in the Australian community: A sub-analysis of the 'PELICAN'study. *Epilepsy Behav* 2022;127:108496.

4. Investigating the Therapeutic Effects of Cannabidiol and Δ9-Tetrahydrocannabinol for Insomnia Disorder

4.1 A Trial Protocol for a Randomised, Placebo-Controlled, Double-blinded, Crossover Trial Exploring the Effects of Cannabidiol and Δ9-tetrahydrocannabinol for Insomnia Disorder

BMJ Open

Cannabidiol (CBD) and Δ^9-tetrahydrocannabinol (THC) for chronic insomnia disorder ('CANSLEEP' trial): protocol for a randomised, placebo-controlled, double-blinded, proof-of-concept trial

Anastasia Suraev ,[1,2,3,4] Ronald R Grunstein,[1,5] Nathaniel S Marshall,[1,6] Angela L D'Rozario,[1,3] Christopher J Gordon,[1,6] Delwyn J Bartlett,[1] Keith Wong,[1,5] Brendon J Yee,[1,5] Ryan Vandrey,[7] Chris Irwin,[8] Jonathon C Arnold,[2,4,9] Iain S McGregor,[2,3,4] Camilla M Hoyos[1,3,4]

To cite: Suraev A, Grunstein RR, Marshall NS, *et al*. Cannabidiol (CBD) and Δ^9-tetrahydrocannabinol (THC) for chronic insomnia disorder ('CANSLEEP' trial): protocol for a randomised, placebo-controlled, double-blinded, proof-of-concept trial. *BMJ Open* 2020;**0**:e034421. doi:10.1136/bmjopen-2019-034421

► Prepublication history and additional material for this paper are available online. To view these files, please visit the journal online (http://dx.doi.org/10.1136/bmjopen-2019-034421).

Received 19 September 2019
Revised 06 January 2020
Accepted 11 March 2020

Check for updates

© Author(s) (or their employer(s)) 2020. Re-use permitted under CC BY-NC. No commercial re-use. See rights and permissions. Published by BMJ.

For numbered affiliations see end of article.

Correspondence to
Dr Camilla M Hoyos;
camilla.hoyos@sydney.edu.au

ABSTRACT

Introduction Insomnia is a highly prevalent and costly condition that is associated with increased health risks and healthcare utilisation. Anecdotally, cannabis use is frequently reported by consumers to promote sleep. However, there is limited research on the effects of cannabis on sleep and daytime function in people with insomnia disorder using objective measures. This proof-of-concept study will evaluate the effects of a single dose of an oral cannabis-based medicine on sleep and daytime function in participants with chronic insomnia disorder.

Methods and analysis A randomised, crossover, placebo-controlled, single-dose study design will be used to test the safety and efficacy of an oral oil solution ('ETC120') containing 10 mg Δ^9-tetrahydrocannabinol (THC) and 200 mg cannabidiol (CBD) in 20 participants diagnosed with chronic insomnia disorder. Participants aged 35–60 years will be recruited over an 18-month period commencing August 2019. Each participant will receive both the active drug and matched placebo, in a counterbalanced order, during two overnight study assessment visits, with at least a 1-week washout period between each visit. The primary outcomes are total sleep time and wake after sleep onset assessed via polysomnography. In addition, 256-channel high-density electroencephalography and source modelling using structural brain MRI will be used to comprehensively examine brain activation during sleep and wake periods on ETC120 versus placebo. Next-day cognitive function, alertness and simulated driving performance will also be investigated.

Ethics and dissemination Ethics approval was received from Bellberry Human Research Ethics Committee (2018-04-284). The findings will be disseminated in a peer-reviewed open-access journal and at academic conferences.

Trial registration number ANZCTRN12619000714189.

Strengths and limitations of this study

- This is the first study to use novel assessment techniques including high-density electroencephalography (EEG) with structural brain MRI to comprehensively examine and localise differences in brain activation during sleep and wake periods in participants with insomnia disorder.
- This study uses a randomised, controlled trial design to investigate the effects of a pharmaceutical-grade oral oil solution containing 10mg Δ^9-tetrahydrocannabinol (THC) and 200mg cannabidiol (CBD) on sleep and daytime function in a clinical population.
- Participants will have sleep physician-confirmed insomnia disorder and will be thoroughly screened to rule out other sleep disorders.
- This study is a single-dose design investigating only the acute effects of a cannabis-based medicine over a 24-hour period in a controlled in-laboratory environment.
- This study cannot assess the individual contribution of THC and CBD.

INTRODUCTION

Insomnia is a common sleep disorder that can present in isolation or comorbid to other medical or psychiatric conditions.[1] Despite often emerging as a transient response to stress or change to one's normal sleep–wake cycle,[2] approximately 30% of individuals with insomnia display chronic symptoms.[3] Chronic insomnia is characterised by sleep disturbances (difficulties with falling asleep, maintaining sleep, or inability to return to sleep on awakening) occurring at least

AUTHOR PROOF

three nights per week and for at least 3 months.[4 5] The sleep disturbance is often coupled with clinically significant daytime impairments in social life, occupational function and/or educational achievement.[4 5] It is often the perceived daytime impairments, as opposed to the noctural insomnia symptoms *per se*, that prompt patients to seek treatment.[6] Chronic insomnia tends to either 'wax and wane' or persist over a lifetime, with the latter course predicted by more severe insomnia symptoms at baseline, female gender and older age.[7 8]There are emerging associations between chronic insomnia and increased health risks such as cardiovascular disease,[9] depression[10–12] and dementia,[13 14] as well as high rates of absenteeism[15 16] and healthcare utilisation.[17] Indeed, longitudinal studies with follow-up period ranging from 1 to 34 years have found a substantial risk for developing depression (both first onset and recurrent major depressive disorder) in patients with insomnia, an association that is bidirectional.[18] As such, there is a strong need for early clinical intervention.

The goal of treatment for insomnia is to improve sleep (both duration and quality) and alleviate daytime impairments. Psychological therapies such as cognitive behavioural therapy for insomnia (CBT-I) and psychoeducation regarding sleep hygiene can be effective.[19] However, these often require access to a therapist and can involve substantial effort, time and financial commitment.[20] Furthermore, the perceived benefits from these approaches are typically delayed. Thus, patients with persistent symptoms often seek strategies offering short-term relief to maintain normal daytime functioning; highlighting a specific role for adjunctive use of pharmacological treatments such as benzodiazepines, sedating antidepressants, and Z-drugs.[21] However, these are associated with undesirable side effects such as cognitive impairment, tolerance/dependence and impaired driving due to sedative effects that can persist into the following day.[22] Moreover, many of these medications disturb sleep architecture; increasing sleep fragmentation and one's sense of having non-restorative sleep and ultimately impair the ability to undertake normal daily activities.[23] Thus, novel approaches are needed to address the needs of people with chronic insomnia disorder.

Anecdotally, consumers of cannabis commonly report that the drug promotes uninterrupted sleep.[24] The plant *Cannabis sativa* contains >100 different cannabinoids—the most abundant of which are the main psychoactive component, Δ^9-tetrahydrocannabinol (THC), and the non-intoxicating cannabinoid, cannabidiol (CBD).[25] Both CBD and THC affect components of the endogenous cannabinoid system which are involved in the regulation of the circadian sleep–wake cycle, including the maintenance and promotion of sleep.[26 27] THC is a partial agonist of the cannabinoid 1 (CB_1) receptor, found primarily within the central nervous system[28] and the cannabinoid 2 (CB_2) receptor, found primarily in the immune system and on peripheral organs.[29] THC is known to have sedating properties via its action at the CB_1 receptor, which is notably dense in areas of the central nervous system such as the thalamus, hypothalamus, hippocampus, basal ganglia and cortex, suggesting a diverse role in the modulation of physiological functions including sleep.[30 31] CBD is an indirect CB_1 and CB_2 receptor agonist, and has shown to increase concentrations of the major endogenous cannabinoid, anandamide, by inhibiting its degradative enzyme, fatty acid amid hydrolase (FAAH).[32 33] Increasing endogenous anandamide via FAAH inhibition normalised deficits in stage N3 sleep in cannabis-dependent men experiencing withdrawal,[34] consistent with preclinical data showing that anandamide promotes slow wave sleep, possibly through increases in extracellular adenosine concentrations.[35–37] This effect can be blocked by administration of the CB_1 antagonist, rimonabant.[38] Indeed, clinical trials of rimonabant have reported an increased risk of sleep disturbances,[39] suggesting a role for the CB_1 receptor in mediating sleep. CBD is also a negative allosteric modulator of CB_1 receptor[40] and may reduce the effects of THC and anandamide on the brain.[41 42] There is an emerging viewpoint that coadministration of CBD with THC may enhance therapeutic outcomes by attenuating the adverse effects of THC (e.g., on emotion recognition,[43] next-day memory performance,[44] appetitive effects[45] and acute psychotic symptoms[46 47]); however, findings are inconsistent with a recent study showing CBD exacerbating THC-induced impairment on driving and cognition, possibly via a pharmacokinetic interaction.[48] Furthermore, CBD is a promiscuous molecule that exhibits activity on a wide array of molecular targets beyond CB_1 and CB_2 receptors such as inhibitory $GABA_A$ receptors,[49] which may also influence sleep. Administration of THC alone (15 mg) in the evening was associated with next-day changes in mood, sleepiness and memory in healthy adults,[44] emphasising the need for careful consideration of dose and ratio of cannabinoids when administered in clinical insomnia populations.

To date, there have been no well-designed randomised controlled trials employing objective measures assessing the effects of cannabis on sleep duration and quality in a clinical insomnia population.[50–52] Previous studies have shown potential benefits in the therapeutic use of nabiximols (*Sativex*), an oromucosal spray containing equal parts THC and CBD, in the relief of pain and other chronic symptoms including improved sleep, with the latter only assessed as a secondary outcome using subjective rating scales.[53] Other studies using synthetic THC (nabilone) showed improvements in subjective sleep quality in patients with post-traumatic stress disorder (PTSD)[54 55] and fibromyalgia,[56] while CBD was found to be effective in reducing the frequency of rapid eye movement sleep behaviour disorder events in Parkinson's disease.[57] One case study showed that 25 mg CBD daily reduced anxiety symptoms and improved sleep disturbances in a young child with PTSD.[58] Indeed, preclinical evidence has demonstrated anxiolytic effects of CBD, likely dependant on CB_1 and $5\text{-}HT_{1A}$ receptor action, with early human experimental evidence supporting

AUTHOR PROOF

preclinical findings.[59] To address the lack of studies in a clinical insomnia population, we will investigate the acute effects of a plant-derived, pharmaceutical-grade, oral formulation containing 10 mg THC and 200 mg CBD relative to placebo on sleep and next-day function (cognitive function, alertness, simulated driving performance) in participants with physician-confirmed chronic insomnia disorder. This study will be the first to employ 256-channel high-density electroencephalography (EEG) coupled with structural MRI brain scans to examine and localise differences in sleep depth and brain activation during both sleep and wakefulness in this clinical population.

METHODS AND ANALYSIS

Study design

A double-blind, randomised, placebo-controlled, crossover study design will be used to evaluate the effects of 10 mg THC and 200 mg CBD on sleep and daytime function in participants diagnosed with chronic insomnia disorder. Participants will be recruited over an 18-month period commencing August 2019. The recruitment target is 20 participants, which will provide the proof-of-concept evaluation of the study drug to determine whether future larger studies in insomnia disorder are warranted. The study site and sponsor is the Woolcock Institute of Medical Research; a research institute and specialist sleep clinic in inner suburban Sydney, Australia. Participants will undergo two separate overnight study assessment visits. Each study assessment visit will be scheduled at least one week apart to avoid any carryover effects, as informed by previous studies of this nature.[60 61] The protocol (Version 2.3, July 2019) has been prepared in accordance with the SPIRIT statement (see online supplementary file 1).[62]

Recruitment and enrolment

The study population will be adults aged 35–60 years with chronic insomnia disorder as per International Classification of Sleep Disorders–Third Edition (ICSD-3) criteria.[63] This age range was chosen to limit age-related variability in sleep architecture for better interpretation of EEG changes.[64] Participation will be voluntary under conditions of informed consent. A list of the inclusion and exclusion criteria is presented in box 1. Participants will be recruited through the following strategies: (1) referral from sleep physicians and psychologists at the Woolcock Institute of Medical Research, Australia; (2) via two databases that host the details of people who have provided consent to be contacted about future clinical trials ("Woolcock Volunteer Database" and the "Lambert Initiative for Cannabinoid Therapeutics Expression of Interest database"); (3) physical study advertisements displayed around the local University area; and (4) study advertisements posted on social and news media. All participants will receive financial compensation for their time commitment to the study.

Study intervention

The investigational product ('ETC120') is a plant-derived oral formulation containing a 1:20 ratio of THC to CBD suspended in medium-chain triglycerides (MCT) oil. ETC120 will be purchased from Linnea (Ticino, Switzerland). Participants will be administered a single fixed dose of ETC120 (2 mL containing 10 mg THC and 200 mg CBD) or matched placebo (2 mL containing no cannabinoids). The 1:20 ratio of THC to CBD was chosen to harness the sedating properties of THC while including some of the potential anti-anxiety properties of CBD,[65] given that anxiety is a very common comorbidity in

Box 1 Inclusion and exclusion criteria

Inclusion criteria

- Aged 35–60 years
- Diagnosis of insomnia disorder made by a physician or a psychologist
- Insomnia Severity Index (ISI) score≥15
- Insomnia symptoms for >3 times per week and present longer than 3 months

Exclusion criteria

- Shift worker
- Medical condition (e.g., chronic pain) or medication that is the cause of the insomnia
- Sleep apnoea (defined as Apnoea Hypopnoea Index (AHI)>15 and Oxygen Desaturation Index (ODI)>10) or sleep-related movement disorder based on in-laboratory polysomnography
- Advanced or delayed sleep–wake phase disorder based on actigraphy
- Used any modality of treatment for insomnia, including cognitive–behavioural therapy (CBT) and CNS-active drugs, within 3 months before screening or at the medical doctor's discretion
- Transmeridian travel (two time zones) in the past month
- Use of medications that may influence cannabinoid metabolism (e.g. inhibitors/inducers of the CYP450 pathway)
- Clinically relevant cardiovascular abnormalities (as determined by 12-lead ECG at screening)
- Pregnancy or lactation (females)
- History of a major psychiatric disorder within the past 12 months (except clinically-managed mild depression or anxiety) as per the Diagnostic and Statistical Manual of Mental Disorders (DSM)-5 or at the medical doctor's discretion
- History of attempted suicide or current suicidal ideation as determined by a score >0 on Q9 of the Patient Health Questionnaire (PHQ)-9
- History of drug or alcohol abuse/dependency within the past 2 years
- Urinary drug screen positive for drugs (benzodiazepines, opiates, cannabis, amphetamines, cocaine)
- Known hypersensitivity to cannabis
- Cannabis use within the past 3 months (confirmed by negative urine drug screen)
- Unable to undergo brain MRI due to implanted device or other reason
- Excessive caffeine use that, in the opinion of the medical doctor contributes to the participant's insomnia, or is unable to abstain from caffeine use ≥24 hours prior to each overnight study assessment visit
- Inability to refrain from alcohol consumption ≥24 hours prior to each overnight study assessment visit
- Medical conditions that result in frequent need to get out of bed (e.g., nocturia)
- Required to complete mandatory drug testing for cannabis (e.g., workplace testing, court order)

AUTHOR PROOF

people with insomnia disorder.[66 67] As noted above, there is also possibility that this dose of CBD might reduce some of the possible adverse effects of THC (e.g., anxiety, memory impairment). The chosen ratio also mimics naturalistic findings in recent surveys where individuals reported using cannabis with higher CBD concentrations in addition to THC to effectively manage insomnia symptoms.[68 69] The THC dose (10 mg) was chosen as being the maximum dose that is likely to induce subjective drug effects of feeling 'sleepy/tired' without impairing cognitive performance (e.g., reaction time tasks) or producing significant intoxication in naïve or occasional cannabis users.[60] A significant intoxicating effect might inadvertently cause a stimulatory response and interfere with sleep induction.[60 70]

Randomisation and allocation concealment

Each participant will be randomly allocated to one of two treatment sequences: (1) ETC120–placebo, or (2) placebo–ETC120. As this is a blinded study, the participant, the study staff (including the medical doctor) and the outcome assessors will not be aware of which treatment order participants have been allocated to. Method of allocation concealment will involve central randomisation by computer prepared by the trial statistician (NSM) and identical containers numbered according to the randomisation sequence prepared by the drug distributor. Neither the drug distributer or the trial statistician will meet any prospective or enrolled participants or be involved in any day-to-day trial process. The sequence will be computer-generated using a simple 1:1 randomisation ratio by the trial statistician, and by the order of participant enrolment. The sequence will be stored in a password-protected data management system and cannot be accessed by blinded study staff who have contact with participants. The order of treatment will only be known by the drug distributer and the trial statistician. In the event of a serious adverse event (SAE) or reaction, the allocation list will be retrieved from the unblinded trial statistician or drug distributer to reveal the participant's allocated treatment during the trial.

Study objectives

The primary outcome of the study is to assess the effect of 10 mg THC and 200 mg CBD on sleep continuity (wake after sleep onset) and quantity (total sleep time) assessed using attended overnight full polysomnography in participants with chronic insomnia disorder.

Secondary objectives include:

- To determine changes in sleep microarchitecture metrics measured using high-density EEG and source modelling in participants with chronic insomnia disorder treated with ETC120 relative to placebo.
- To assess next-day neurobehavioural functioning (cognition, alertness and simulated driving performance) in participants with chronic insomnia disorder treated with ETC120 relative to placebo.
- To demonstrate feasibility of a cannabinoid study in chronic insomnia and establish clinical trial procedures for future trials in this area.

The ANZCTR trial registry has a comprehensive list of the trial's primary and secondary outcomes. See online supplementary file 2 for WHO Trial Registration Data Set.

Study visits and procedures

Screening

A flowchart of the study is depicted in figure 1. Initial suitability assessment via a brief online questionnaire and a follow-up telephone screen will be conducted by the study investigator. Written informed consent will be obtained by the study medical doctor before conducting an interview to ascertain sleep difficulties and diagnose ICSD-3 chronic insomnia disorder (Visits 1–2). Individuals will then undergo comprehensive screening to be completed within one month of study entry, which will include a diagnostic sleep study at the Woolcock Clinic to exclude sleep disorders other than insomnia disorder (unless one has already been conducted in the past 12 months). The Insomnia Severity Index[71] (ISI; measure of nature, severity, and impact of insomnia), the Pittsburgh Sleep Quality Inventory[72] (PSQI; measure of sleep quality and sleep habits), the Epworth Sleepiness Scale[73] (ESS; measure of daytime sleepiness), the Hospital Anxiety Depression Scale[74] (HADS; measure of anxiety and depression) and the Patient Health Questionnaire[75] (PHQ-9; multi-purpose tool for assessing severity of depression) will be administered to phenotype insomnia symptoms and to assess suitability for study inclusion. All participants will be screened for prior cannabis use history (ie, whether they have consumed cannabis in the past, the form(s) in which it was consumed, and frequency of use) as well as for past or present cannabis use disorder as per the ICD-10 criteria.[76] A urine specimen will be screened (DrugCheck NxStep OnSite Drug Test, Minnesota, USA) to rule out recent drug use. Participants testing positive for any drug (cannabis, cocaine, benzodiazepines, opiates or amphetamines/MDMA/methamphetamines) will result in exclusion or rescheduled at the studymedical doctor's discretion. A standard 12-lead electrocardiogram (ECG) will be recorded to screen for any clinically relevant cardiovascular abnormalities. Rapid urine pregnancy test (Alere HCG Combo Cassette, Massachusetts, USA) will be administered to female participants, and identification of pregnancy will result in exclusion. Participants will then be instructed to maintain a sleep diary and wear a wrist-worn commercially available device (Actiwatch 2, Philips Respironics) to monitor sleep and wake periods for one week. These data will allow the study team to estimate the participant's individual typical sleep-onset and wake-onset times for the study assessment visits as well as rule out advanced or delayed sleep-wake phase syndrome.

Following screening, the participants will undergo a structural brain MRI at a medical imaging clinic (Visit

AUTHOR PROOF

3). Then, participants will attend the sleep clinic for an orientation session (Visit 4) to practise wearing the high-density EEG sensor cap during a short nap opportunity as well as complete a familiarisation and practice drive on the driving simulator. Participants will then be asked to maintain consistent sleep-onset and wake-onset times, confirmed by at-home sleep diary and actigraphy for one week prior to each study assessment visit. Participants will be instructed to abstain from illicit drug use for the duration of the study (ie, from pre-enrolment until after the final study assessment visit) and to refrain from consuming alcohol and caffeine for 24 hours prior to and during the study assessment visits, but to continue use of any regular prescribed medications (except those listed in the exclusion criteria). Standardised meals and snacks will be provided for participants at each study assessment visit. Table 1 depicts the schedule of visits and procedures from pre-enrolment to study completion.

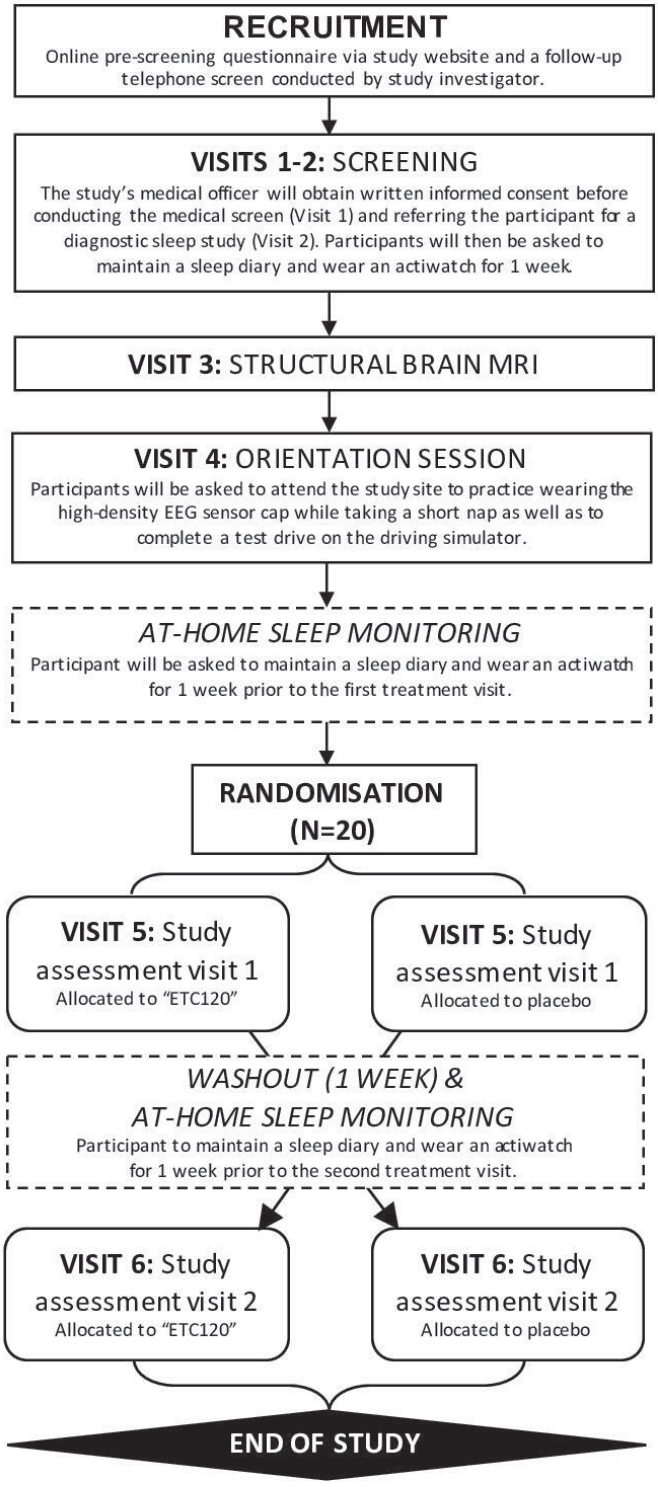

Figure 1 Study flow diagram.

Study assessment visits

Participants will arrive at the clinic at approximately 16:00 hours for each study assessment visit (Visit 5 and 6; see figure 2). Drug-free and alcohol-free status and pregnancy status will be confirmed as described earlier.

Memory consolidation

To assess the next-day effects of ETC120 on memory consolidation, participants will be required to learn the Word Pairs Task (WPT; a declarative memory task) and the Finger Tapping Task (FTT; a procedural memory task) prior to drug administration at approximately 21:00 hours on the night of each study assessment visit and will be re-tested the following morning at approximately 07:30 hours.

Study drug administration

The study's medical doctor will prepare the study drug on the same day of the study assessment visit by drawing 2 mL of the active drug or matched placebo in an amber plastic syringe secured with a tip cap. To mask smell and taste, participants will be instructed to consume one peppermint lozenge (Fisherman's Friend Mint; Lofthouse of Fleetwood, England) immediately prior to drug administration. One hour prior to the participant's typical sleep-onset time, the study investigator will then instruct and directly observe the participant to orally ingest the fixed dose of the study drug. This timeframe was chosen to represent the THC T_{max} following a single oral dose of 10 mg THC.[60] All participants will be given an 8-hour sleep opportunity. Time of drug administration is relative to the participant's typical sleep-onset time, which may vary up to a maximum of 45 min.

Assessment of sleepiness

Participants will be administered the Karolinska Drowsiness Test (KDT) in conjunction with the Karolinska Sleepiness Scale (KSS), two measures that assess physiological and subjective sleepiness respectively,[77 78] immediately before bedtime, immediately upon awakening, and at 10:00, 12:00, 14:00, and 16:00 hours coinciding with the Maintenance of Wakefulness Tests (MWT) described later. The main outcome measure is resting wake EEG power during the KDT before and after polysomnography.

Polysomnography with high-density EEG

Participants will undergo an in-laboratory 256-electrode high-density EEG (Electrical Geodesics, Oregon, USA) full polysomnography which includes electrooculogram

AUTHOR PROOF

AUTHOR PROOF

Table 1 Schedule of study visits and procedures

Measure	Visit 1: Medical assessment	Visit 2: Diagnostic sleep study	At-home sleep monitoring	Visit 3: Brain MRI	Visit 4: Practice session	At-home sleep monitoring	Visit 5: Study assessment visit 1		Washout & at-home sleep monitoring	Visit 6: Study assessment visit 2	
							Evening	Daytime		Evening	Daytime
Informed consent	●										
Physical assessment (vital signs)	●						●			●	
ECG	●										
Baseline questionnaires (ISI, PSQI, ESS, HADS, PHQ-9)	●										
Urinary drug/alcohol screen	●						●			●	
Pregnancy test	●						●			●	
Overnight PSG (standard clinical EEG)		●									
Brain MRI				●							
Actigraphy			●			●			●		
Karolinska sleep diary			●			●			●		
Salivary drug screen (Securetec DrugWipe & Dräger 5000)							●	●		●	●
Saliva collection (Quantisal)							●	●		●	●
Mood POMS abbreviated							●	●		●	●
Memory Tasks – Consolidation (WPT & FT)							●	●		●	●
Study drug administration							●			●	
DEQ							●	●		●	●
KSS & KDT							●	●		●	●
KSS							●	●		●	●
Overnight PSG (Research high-density EEG)							●			●	
Blood plasma collection								●			●
Memory Tasks - Test (WPT & FT)								●			●
Simulated driving performance task								●			●
DISRS								●			●
LSEQ								●			●
Cognitive test battery (DSST, DAT, PASAT, PVT, Stroop test and 1 and 2 n-back task)											●
MWT								●			●

DAT, Divided Attention Task; DEQ, Drug Effects Questionnaire; DISRS, Daytime Insomnia Symptoms Response Scale; DSST, Digit Symbol Substitution Test; ECG, electrocardiogram; EEG, electroencephalography; ESS, Epworth Sleepiness Scale; FTT, Finger Tapping Task; HADS, Hospital Anxiety Depression Scale; ISI, Insomnia Severity Index; KDT, Karolinska Drowsiness Test; KSS, Karolinska Sleepiness Scale; LSEQ, Leeds Sleep Evaluation Questionnaire; MRI, magnetic resonance imaging; MWT, Maintenance of Wakefulness Test; PASAT, Paced Auditory Serial Addition Task; PHQ-9, Patient Health Questionnaire; POMS, Profile of Mood States; PSG, polysomnography; PSQI, Pittsburgh Sleep Quality Inventory; PVT, Psychomotor Vigilance Task; WPT, Word Pairs Task.

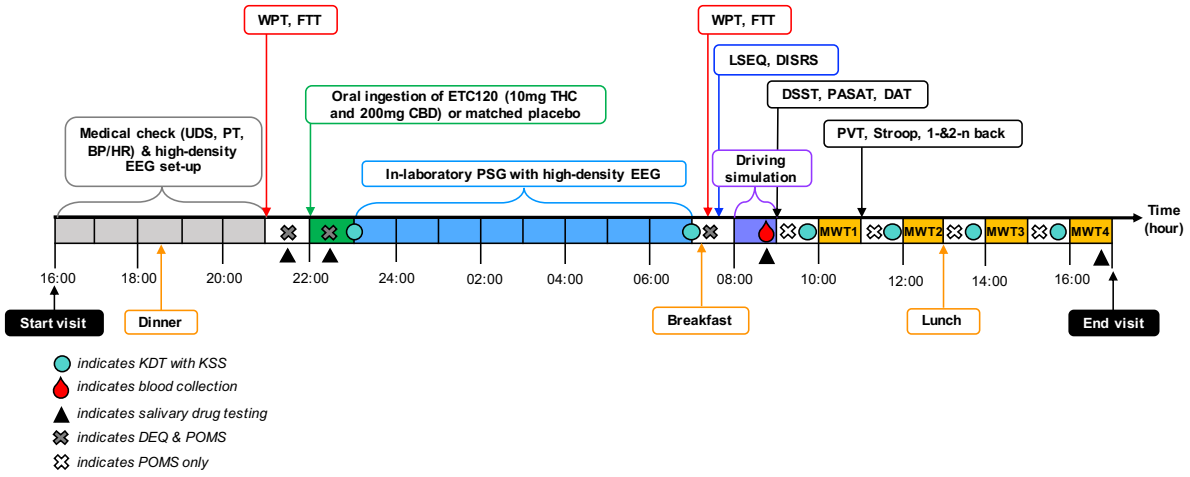

Figure 2 Schedule of events during study assessment visits. BP/HR, blood pressure/heart rate; CBD, cannabidiol; DAT, Divided Attention Task; DEQ, Drug Effects Questionnaire; DISRS, Daytime Insomnia Symptom Response Scale; DSST, Digit Symbol Substitution Task; EEG, electroencephalography; FTT, Finger Tapping Task; KDT, Karolinska Drowsiness Test; KSS, Karolinska Sleepiness Scale; LSEQ, Leeds Sleep Evaluation Questionnaire; MWT, Maintenance of Wakefulness Test; PASAT, Paced Auditory Serial Addition Task; POM, Profile of Mood States; PT, pregnancy test; PSG, polysomnography; PVT, Psychomotor Vigilance Task; THC, delta-9-tetrahydrocannabinol; UDS, urinary drug screen; WPT, Word Pairs Task.

(EOG), electromyogram (EMG), ECG, pulse oximetry and a position sensor. Sleep recordings will be scored by a sleep technologist specialising in high-density EEG and reviewed by a certified sleep physician. The GeoScan device will be used to measure, identify and create a three-dimensional coordinate file of the 256-electrode locations on the high-density EEG sensor cap. This will be combined with each individual participant's structural brain MRI scan to localise the source of brain activity to specific brain regions.

Subjective measurements

Mood will be assessed using the Profile of Mood States (POMS) abbreviated version[79] at baseline, 60 min post-drug administration, and the next-day at approximately 08:00, 10:00, 12:00, 14:00 and 16:00 hours. Subjective drug effects will be assessed using the Drug Effects Questionnaire (DEQ) which includes a series of visual analogue scales at baseline, 60 min post-drug administration and the next-day at approximately 08:00. Measurements will stop after the 08:00 timepoint because subjective drug effects following a single acute dose are not expected to persist beyond this time. On the VAS, participants will rate on a 100 mm line their responses to the statements: 'Strength of drug effect', 'Liking of drug effect', 'Feeling stoned' and 'Feeling sedated', with all scales unipolar. Perceived changes in sleep and daytime function will be assessed using the Leeds Sleep Evaluation Questionnaire (LSEQ)[80] and the Daytime Insomnia Symptom Response Scale (DISRS)[81] at approximately 07:30 hours the morning post-drug administration.

Driving performance

At 08:00 hours the next day, participants will be asked to complete a 30 min simulated driving task using a custom-built fixed-base computerised driving simulator (Hyperdrive, Adelaide, Australia) equipped with original vehicle controls (steering wheel, indicators, seat, safety belt), hi-resolution Fanatec pedals, and a servo motor wheel base (Endor AG, Landshut, Germany) linked to four networked computers running the SCANeR Studio simulation engine software (V.1.6, AVSimulation, Paris, France). The driving scenario is identical to that previously employed in a study examining the effects of vaporised cannabis on driving performance in healthy volunteers.[61] Outcome measures include standard deviation of lateral position (SDLP) and number of lane crossings as measures of lateral vehicle control (ie, lane swerving behaviour), and average speed and standard deviation of speed (SDSP) as measures of longitudinal vehicle control. These outcome parameters have previously demonstrated sensitivity to the impairing effects of sleep disturbance,[82] hypnotic medication[83] and cannabis administration.[84]

Salivary drug testing

Given the current legal framework for driving under the influence of cannabis in Australia (ie, detection of THC in saliva with no functional assessment),[85] all participants will undergo salivary drug testing to test for the presence of THC. Oral fluid samples will be collected using Quantisal collection devices (Immunalysis, Pomona, California, USA) at baseline, 30 min post-drug administration, and the next-day after completing the driving simulation (approximately 09:00 hours) and at completion of the study visit (17:00 hours). Samples will be kept at −80°C prior to analysis for THC and CBD using liquid chromatography-tandem mass spectrometry (LC-MS/MS). Oral fluid will also be screened for

AUTHOR PROOF

cannabis (THC) using two devices: DrugWipe 5s (Securetec, Neubiberg, Germany) and Dräger Drug Test 5000 (Drägerwerk AG & Co., Lübeck, Germany) at four timepoints: baseline (30 min prior to drug administration), T1 (30 min after drug administration), T2 (08:30 hours; the next day immediately after completing the driving task) and T3 (17:00 hours; prior to leaving the study site). Both devices have a manufacturer-specified detection limit of 10 ng/mL THC. Participants will be provided with taxi vouchers to and from the study site at both study assessment visits as they will not be permitted to drive. Participants will be given explicit instruction not to drive for at least 24 hours after leaving the study site to allow adequate time for drug washout following a single dose.

Blood collection and plasma cannabinoid levels

Blood will be collected once via venepuncture into EDTA vacutainer tubes (Becton, Dickinson and Company, New Jersey, USA) at approximately 08:45 hours, immediately after the driving performance task, to measure levels of THC and other cannabinoids the morning post-drug administration. Blood will be centrifuged at 1500×*g* for 10 min at 4°C with the supernatant plasma aliquoted and stored in 1.8 mL cryotubes at −80°C until subsequent analysis. Plasma will be analysed via LC-MS/MS according to previously published methods[85 86] for cannabinoids (CBD, THC) and their metabolites (11-OH-THC, THC-COOH; 7-COOH-CBD, 7-OH-CBD and 6-OH-CBD) as well as a range of endocannabinoid and related molecules (anandamide, 2-AG, 1-AG, oleoylethanolamide (OEA), palmitoylethanolamide (PEA), linoyl-ethanolamide (LEA) and oleamide).

Cognitive performance

Cognitive assessment will take place the morning post-drug administration (see table 1), to explore the functional consequences of 10 mg THC and 200 mg CBD on next-day daytime function. This will be measured from approximately 09:00 hours using the following battery of computerised cognitive/psychomotor tasks known to be sensitive to the impairing effects of THC[60 70]: Digit Symbol Substitution Test (DSST; measure of processing speed, working memory and attention), Divided Attention Task (DAT; measure of processing speed, working memory and attention) and Paced Auditory Serial Addition Task (PASAT; measure of processing speed and sustained attention). Other cognitive tasks to be administered in conjunction include the Psychomotor Vigilance Task (PVT; simple reaction time task measuring sustained attention), Stroop test (a measure of executive functioning), and the 1- and 2-n back test (a measure of working memory and information processing).

Maintenance of Wakefulness Test

The Maintenance of Wakefulness Test (MWT) is a validated, objective measure of an individual's ability to stay awake in a room with low levels of stimulation that will test for drowsiness the next-day post-drug administration.[86] As recommended by American Academy of Sleep Medicine (AASM) practice parameters,[87] four 40 min MWT trials will be administered at 10:00, 12:00, 14:00 and 16:00 hours on the day post-drug administration. Participants will be instructed to lay semi-recumbent on a bed (above the covers) in a darkened room and try to remain awake for 40 min. An experienced sleep technician will record the polysomnography using high-density EEG. Trials will end after 40 min if no sleep occurs, or after unequivocal sleep, defined as three consecutive epochs of non-rapid eye movement stage 1 (N1) sleep or one epoch of any other sleep stage (N2, N3, N4 or REM). The main outcome measure is the mean sleep latency of the four MWT trials.

Patient and public involvement

The present trial was developed by the investigators based on previous clinical experience and gaps identified in the existing literature. Patients were not involved in the design of the study. The outcomes are commonly used assessments of insomnia in research. The cost of interventions and outcome measurements are covered by the study funding. All participants will be offered a clinical follow-up appointment with a sleep physician on conclusion of the study. Participants will receive a summary of the study results once published.

Data collection and management

All clinical data and information obtained for the purpose of this research that could identify participants will be treated as confidential and securely stored, adhering to the University regulations and the Australian Code for the Responsible Conduct of Research. Participant data will be identified by a unique code number that will be allocated after the participant gives consent to participate in the study. The unique code linking the participant's identity/personal details (e.g., name, date of birth) will be stored in a password-encrypted file that will not be accessible from the internet. All data will be stored at the Woolcock Institute of Medical Research in written and computerised formats. Participant information will reside on a secure server that is regularly backed up. All data will be stored securely for at least 15 years. Only researchers affiliated with the study will have access to participant data. Study progress and safety will be monitored and evaluated internally in an ongoing fashion by the Trial Management Group consisting of the principal investigators, trial coordinator, research assistants, trial statistician, data manager and sleep clinic manager. There are no planned interim analyses. The final decision to terminate the trial lies with the principal investigators and will be based on (1) safety data and (2) target recruitment number. The investigator team will conduct an internal 3-monthly review of all adverse events and reactions and if after discussion, the rate of such events is deemed unacceptable then the study will be stopped and the human research ethics committee will be advised of the decision.

AUTHOR PROOF

Sample size and statistical analyses

This protocol was designed to be a single dose, proof-of-concept study to ascertain initial safety and efficacy of the study drug in participants with chronic insomnia disorder. As there is no commercially available power calculation software for mixed-model analyses available at present, using a simple paired t-test, a crossover trial of 20 participants is adequately powered to detect an effect size of 0.67 with 80% power at an alpha level of 0.05 (two-tailed). Data obtained will guide future studies by providing 95% confidence limits for sensitivity analyses for power calculations of a larger trial if warranted. Data will be analysed using mixed-model analyses of variance in SAS (SAS Institute, V.9.4) to test whether either of the treatments are different from the other. Order and treatment will be fixed effects and the patient code will be used as a random effect.[88] Treatment by order effect will not be tested. All variables are suitable for mixed-model analyses except for the adverse event profile which will be tabulated but not statistically tested. The least-squares means procedure will be used in the mixed-model analyses to handle missing data. All participants will be analysed in the groups they have been randomised to. Primary outcomes will be interpreted as affected if either are significant at 0.05.

Significance

Cannabis is commonly believed to be a useful sleep aid. However, there are no published studies to-date assessing its effects on sleep in people with physician-confirmed chronic insomnia disorder. Given the increased consumer interest and expansion of legal prescription for cannabis globally, it is important to better understand how cannabis-based medicines affect sleep and next-day function prior to becoming a routine intervention in clinical practice. This is particularly important as sleep disturbances are fiercely comorbid in many chronic health conditions such as pain; key indications for the prescription of cannabis-based medicines around the world.[89] Of note, this is a proof-of-concept trial that is limited by its small sample size and single-dose design, precluding examination of the long-term effects of this cannabis-based medicine in this clinical population. Moreover, the study cannot assess the individual contribution of THC and CBD. Nonetheless, the current study is a rigorous double-blinded, placebo-controlled, within-subjects, crossover design that will provide a preliminary signal on the efficacy and safety of a pharmaceutical-grade cannabis-based medicine in people with chronic insomnia disorder, and will hopefully help inform the development of future longer-term research trials.

Ethics and dissemination

Ethics approval was received from Bellberry Human Research Ethics Committee (2018-04-284). The findings of this trial will be disseminated in peer-reviewed journal publications and at academic conferences. The sponsor controls the final decision regarding all aspects of the trial including dissemination of results. The study investigator is responsible for communicating important protocol modifications to relevant parties.

Author affiliations
[1]Woolcock Institute of Medical Research, Centre for Sleep and Chronobiology, Sydney, New South Wales, Australia
[2]The University of Sydney, Lambert Initiative for Cannabinoid Therapeutics, Sydney, New South Wales, Australia
[3]The University of Sydney, Faculty of Science, School of Psychology, Sydney, New South Wales, Australia
[4]The University of Sydney, Brain and Mind Centre, Sydney, New South Wales, Australia
[5]RPA-Charles Perkins Centre, Royal Prince Alfred Hospital, Sydney, New South Wales, Australia
[6]The University of Sydney, Faculty of Medicine and Health, Susan Wakil School of Nursing and Midwifery, Sydney, New South Wales, Australia
[7]Behavioral Pharmacology Research Unit, Johns Hopkins University, Baltimore, Maryland, USA
[8]Menzies Health Institute Queensland, School Allied Health Sciences, Griffith University, Gold Coast, Queensland, Australia
[9]The University of Sydney, Faculty of Medicine and Health, Discipline of Pharmacology, Sydney, New South Wales, Australia

Twitter Nathaniel S Marshall @NatSleep

Contributors AS, RRG, NSM, ALD, CJG, DJB, KW, BJY, RV, CI, JCA, ISM and CMH were involved in the methodological design and drafting of the trial protocol. RRG and CMH are the medical and non-medical principal investigator, respectively, who have overall responsibility for the design, conduct and decision to submit for publication. NSM is the trial statistician who designed and wrote the analysis plan. AS is the trial coordinator responsible for collecting trial data. AS drafted the manuscript. All authors read and approved the final manuscript.

Funding This study is funded by the Lambert Initiative for Cannabinoid Therapeutics, a philanthropically funded centre for cannabinoid research at the University of Sydney. AS was supported by the Australian Government Research Training Program (RTP) and the Vice-Chancellor's Research Scholarship (VCRS) at the University of Sydney. CMH (APP1104003) and ALD (APP1107716) were supported by Dementia Research Development Fellowships of the Australian National Health and Medical Research Council-Australian Research Council (NHMRC-ARC). RRG was supported by an NHMRC Senior Principal Research Fellowship (APP1106974). The investigational product was purchased from Linnea (Ticino, Switzerland) who were not involved in the conception or design of this study.

Competing interests ISM is Academic Director of the Lambert Initiative for Cannabinoid Therapeutics. He has served as an expert witness in various medicolegal cases involving cannabis, has received honoraria from Janssen, is currently a consultant to Kinoxis Therapeutics, and has received research funding and fellowship support from the Lambert Initiative for Cannabinoid Therapeutics, National Health and Medical Research Council (NHMRC) and Australian Research Council. He holds a variety of patents for cannabinoid and non-cannabinoid therapeutics. RV has received financial compensation from Zynerba Pharmaceuticals, Canopy Health Innovations and Brain Solutions. JCA is Deputy Academic Director of the Lambert Initiative for Cannabinoid Therapeutics. He has served as an expert witness in various medicolegal cases involving cannabis and recently served as a temporary advisor to the WHO on their review of cannabis and the cannabinoids. His research is funded by the NHMRC, Canopy Growth Corporation and the Lambert Initiative for Cannabinoid Therapeutics. JCA also holds several patents on novel cannabinoid therapies. All other authors have no conflicts of interest to disclose.

Patient and public involvement Patients and/or the public were not involved in the design, or conduct, or reporting or dissemination plans of this research.

Patient consent for publication Not required.

Provenance and peer review Not commissioned; externally peer reviewed.

Open access This is an open access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited, appropriate credit is given, any changes made

AUTHOR PROOF

indicated, and the use is non-commercial. See: http://creativecommons.org/licenses/by-nc/4.0/.

ORCID iD
Anastasia Suraev http://orcid.org/0000-0002-5993-2246

REFERENCES

1 Morin CM, Benca R. Chronic insomnia. *The Lancet* 2012;379:1129–41.
2 Van Reeth O, Weibel L, Spiegel K, *et al*. Interactions between stress and sleep: from basic research to clinical situations. *Sleep Med Rev* 2000;4:201–20.
3 Roth T. Insomnia: definition, prevalence, etiology, and consequences. *J Clin Sleep Med* 2007;3:S7.
4 American Psychiatric Association (ASA). *Diagnostic and statistical manual of mental disorders*. 5th Edition. Arlington, VA, 2013.
5 American Academy of Sleep Medicine. *International classification of sleep disorders*. 3rd Edition. Darien: IL, 2014.
6 Morin CM, LeBlanc M, Daley M, *et al*. Epidemiology of insomnia: prevalence, self-help treatments, consultations, and determinants of help-seeking behaviors. *Sleep Med* 2006;7:123–30.
7 Morin CM, Bélanger L, LeBlanc M, *et al*. The natural history of insomnia: a population-based 3-year longitudinal study. *Arch Intern Med* 2009;169:447–53.
8 Ji X, Ivers H, Savard J, *et al*. Residual symptoms after natural remission of insomnia: associations with relapse over 4 years. *Sleep* 2019;42:zsz122.
9 Bertisch SM, Pollock BD, Mittleman MA, *et al*. Insomnia with objective short sleep duration and risk of incident cardiovascular disease and all-cause mortality: sleep heart health study. *Sleep* 2018;41:zsy047.
10 Baglioni C, Battagliese G, Feige B, *et al*. Insomnia as a predictor of depression: a meta-analytic evaluation of longitudinal epidemiological studies. *J Affect Disord* 2011;135:10–19.
11 Leggett AN, Sonnega AJ, Lohman MC. The association of insomnia and depressive symptoms with all-cause mortality among middle-aged and old adults. *Int J Geriatr Psychiatry* 2018;33:1265–70.
12 Riemann D, Voderholzer U. Primary insomnia: a risk factor to develop depression? *J Affect Disord* 2003;76:255–9.
13 de Almondes KM, Costa MV, Malloy-Diniz LF, *et al*. Insomnia and risk of dementia in older adults: systematic review and meta-analysis. *J Psychiatr Res* 2016;77:109–15.
14 Hung C-M, Li Y-C, Chen H-J, *et al*. Risk of dementia in patients with primary insomnia: a nationwide population-based case-control study. *BMC Psychiatry* 2018;18:38.
15 Sivertsen B, Øverland S, Pallesen S, *et al*. Insomnia and long sleep duration are risk factors for later work disability. The Hordaland health study. *J Sleep Res* 2009;18:122–8.
16 Lallukka T, Haaramo P, Rahkonen O, *et al*. Joint associations of sleep duration and insomnia symptoms with subsequent sickness absence: the Helsinki health study. *Scand J Public Health* 2013;41:516–23.
17 Bin YS, Marshall NS, Glozier N. The burden of insomnia on individual function and healthcare consumption in Australia. *Aust N Z J Public Health* 2012;36:462–8.
18 Franzen PL, Buysse DJ. Sleep disturbances and depression: risk relationships for subsequent depression and therapeutic implications. *Dialogues Clin Neurosci* 2008;10:473.
19 Okajima I, Komada Y, INOUE Y. A meta-analysis on the treatment effectiveness of cognitive behavioral therapy for primary insomnia. *Sleep Biol Rhythms* 2011;9:24–34.
20 Cheung JMY, Bartlett DJ, Armour CL, *et al*. To drug or not to drug: a qualitative study of patients' decision-making processes for managing insomnia. *Behav Sleep Med* 2018;16:1–26.
21 Miller CB, Valenti L, Harrison CM, *et al*. Time trends in the family physician management of insomnia: the Australian experience (2000-2015). *J Clin Sleep Med* 2017;13:785–90.
22 Mendelson WB, Roth T, Cassella J, *et al*. The treatment of chronic insomnia: drug indications, chronic use and abuse liability. summary of a 2001 new clinical drug evaluation unit meeting symposium. *Sleep Med Rev* 2004;8:7–17.
23 Bastien CH, LeBlanc M, Carrier J, *et al*. Sleep EEG power spectra, insomnia, and chronic use of benzodiazepines. *Sleep* 2003;26:313–7.
24 Bachhuber M, Arnsten JH, Wurm G. Use of cannabis to relieve pain and promote sleep by customers at an adult use dispensary. *J Psychoactive Drugs* 2019;51:400–4.
25 Pertwee RG. The pharmacology of cannabinoid receptors and their ligands: an overview. *Int J Obes* 2006;30:S13–18.
26 Sanford AE, Castillo E, Gannon RL. Cannabinoids and hamster circadian activity rhythms. *Brain Res* 2008;1222:141–8.
27 Vaughn LK, Denning G, Stuhr KL, *et al*. Endocannabinoid signalling: has it got rhythm? *Br J Pharmacol* 2010;160:530–43.
28 Pertwee R. The diverse CB1 and CB2 receptor pharmacology of three plant cannabinoids: Δ9-tetrahydrocannabinol, cannabidiol and Δ9-tetrahydrocannabivarin. *Br J Clin Pharmacol* 2008;153:199–215.
29 Herkenham M, Lynn AB, Little MD, *et al*. Cannabinoid receptor localization in brain. *Proc Natl Acad Sci U S A* 1990;87:1932–6.
30 Murillo-Rodríguez E. The role of the CB1 receptor in the regulation of sleep. *Prog Neuropsychopharmacol Biol Psychiatry* 2008;32:1420–7.
31 Pava MJ, Makriyannis A, Lovinger DM. Endocannabinoid signaling regulates sleep stability. *PLoS One* 2016;11:e0152473.
32 Elmes MW, Kaczocha M, Berger WT, *et al*. Fatty acid-binding proteins (FABPs) are intracellular carriers for Δ9-tetrahydrocannabinol (THC) and cannabidiol (CBD). *J Biol Chem* 2015;290:8711–21.
33 McPartland JM, Duncan M, Di Marzo V, *et al*. Are cannabidiol and Δ(9) -tetrahydrocannabivarin negative modulators of the endocannabinoid system? A systematic review. *Br J Pharmacol* 2015;172:737–53.
34 D'Souza DC, Cortes-Briones J, Creatura G, *et al*. Efficacy and safety of a fatty acid amide hydrolase inhibitor (PF-04457845) in the treatment of cannabis withdrawal and dependence in men: a double-blind, placebo-controlled, parallel group, phase 2A single-site randomised controlled trial. *Lancet Psychiatry* 2019;6:35–45.
35 Murillo-Rodriguez E, Blanco-Centurion C, Sanchez C, *et al*. Anandamide enhances extracellular levels of adenosine and induces sleep: an in vivo microdialysis study. *Sleep* 2003;26:943–7.
36 Mechoulam R, Fride E, Hanus L, *et al*. Anandamide may mediate sleep induction. *Nature* 1997;389:25–6.
37 Murillo-Rodríguez E, Sánchez-Alavez M, Navarro L, *et al*. Anandamide modulates sleep and memory in rats. *Brain Res* 1998;812:270–4.
38 Murillo-Rodríguez E, Cabeza R, Méndez-Díaz M, *et al*. Anandamide-Induced sleep is blocked by SR141716A, a CB1 receptor antagonist and by U73122, a phospholipase C inhibitor. *Neuroreport* 2001;12:2131–6.
39 Padwal RS, Majumdar SR. Drug treatments for obesity: orlistat, sibutramine, and rimonabant. *Lancet* 2007;369:71–7.
40 Tham M, Yilmaz O, Alaverdashvili M, *et al*. Allosteric and orthosteric pharmacology of cannabidiol and cannabidiol-dimethylheptyl at the type 1 and type 2 cannabinoid receptors. *Br J Pharmacol* 2019;176:1455–69.
41 Laprairie RB, Bagher AM, Kelly MEM, *et al*. Cannabidiol is a negative allosteric modulator of the cannabinoid CB1 receptor. *Br J Pharmacol* 2015;172:4790–805.
42 Todd SM, Arnold JC. Neural correlates of interactions between cannabidiol and Δ(9) -tetrahydrocannabinol in mice: implications for medical cannabis. *Br J Pharmacol* 2016;173:53–65.
43 Hindocha C, Freeman TP, Schafer G, *et al*. Acute effects of delta-9-tetrahydrocannabinol, cannabidiol and their combination on facial emotion recognition: a randomised, double-blind, placebo-controlled study in cannabis users. *Eur Neuropsychopharmacol* 2015;25:325–34.
44 Nicholson AN, Turner C, Stone BM, *et al*. Effect of delta-9-tetrahydrocannabinol and cannabidiol on nocturnal sleep and early-morning behavior in young adults. *J Clin Psychopharmacol* 2004;24:305–13.
45 Morgan CJA, Freeman TP, Schafer GL, *et al*. Cannabidiol attenuates the appetitive effects of delta 9-tetrahydrocannabinol in humans smoking their chosen cannabis. *Neuropsychopharmacology* 2010;35:1879–85.
46 Bhattacharyya S, Morrison PD, Fusar-Poli P, *et al*. Opposite effects of delta-9-tetrahydrocannabinol and cannabidiol on human brain function and psychopathology. *Neuropsychopharmacology* 2010;35:764–74.
47 Schubart CD, Sommer IEC, van Gastel WA, *et al*. Cannabis with high cannabidiol content is associated with fewer psychotic experiences. *Schizophr Res* 2011;130:216–21.
48 Arkell TR, Lintzeris N, Kevin RC, *et al*. Cannabidiol (CBD) content in vaporized cannabis does not prevent tetrahydrocannabinol (THC)-induced impairment of driving and cognition. *Psychopharmacology* 2019;236:2713–24.
49 Bakas T, van Nieuwenhuijzen PS, Devenish SO, *et al*. The direct actions of cannabidiol and 2-arachidonoyl glycerol at GAB_{AA} receptors. *Pharmacol Res* 2017;119:358–70.
50 Kuhathasan N, Dufort A, MacKillop J, *et al*. The use of cannabinoids for sleep: a critical review on clinical trials. *Exp Clin Psychopharmacol* 2019;27:383–401.
51 Babson KA, Sottile J, Morabito D. Cannabis, cannabinoids, and sleep: a review of the literature. *Curr Psychiatry Rep* 2017;19:23.

AUTHOR PROOF

52 Gates PJ, Albertella L, Copeland J. The effects of cannabinoid administration on sleep: a systematic review of human studies. *Sleep Med Rev* 2014;18:477–87.
53 Russo EB, Guy GW, Robson PJ. Cannabis, pain, and sleep: lessons from therapeutic clinical trials of Sativex, a cannabis-based medicine. *Chem Biodivers* 2007;4:1729–43.
54 Cameron C, Watson D, Robinson J. Use of a synthetic cannabinoid in a correctional population for posttraumatic stress disorder-related insomnia and nightmares, chronic pain, harm reduction, and other indications: a retrospective evaluation. *J Clin Psychopharmacol* 2014;34:559–64.
55 Fraser GA. The use of a synthetic cannabinoid in the management of treatment-resistant nightmares in posttraumatic stress disorder (PTSD). *CNS Neurosci Ther* 2009;15:84–8.
56 Ware MA, Fitzcharles M-A, Joseph L, *et al*. The effects of nabilone on sleep in fibromyalgia: results of a randomized controlled trial. *Anesth Analg* 2010;110:604–10.
57 Chagas MHN, Eckeli AL, Zuardi AW, *et al*. Cannabidiol can improve complex sleep-related behaviours associated with rapid eye movement sleep behaviour disorder in Parkinson's disease patients: a case series. *J Clin Pharm Ther* 2014;39:564–6.
58 Shannon S, Opila-Lehman J. Effectiveness of cannabidiol oil for pediatric anxiety and insomnia as part of posttraumatic stress disorder: a case report. *Perm J* 2016;20:108.
59 Blessing EM, Steenkamp MM, Manzanares J, *et al*. Cannabidiol as a potential treatment for anxiety disorders. *Neurotherapeutics* 2015;12:825–36.
60 Vandrey R, Herrmann ES, Mitchell JM, *et al*. Pharmacokinetic profile of oral cannabis in humans: blood and oral fluid disposition and relation to pharmacodynamic outcomes. *J Anal Toxicol* 2017;41:83–99.
61 Arkell TR, Kevin R, Stuart J, *et al*. Detection of ∆9THC in oral fluid following vaporised cannabis with varied cannabidiol (CBD) content: an evaluation of two point-of-collection testing devices. *Drug Test Anal* 2019.
62 Chan A-W, Tetzlaff JM, Gøtzsche PC, *et al*. Spirit 2013 explanation and elaboration: guidance for protocols of clinical trials. *BMJ* 2013;346:e7586.
63 American Academy of Sleep Medicine. Diagnostic and Coding Manual. In: *International classification of sleep disorders*, 2005: 51–5.
64 Sprecher KE, Riedner BA, Smith RF, *et al*. High resolution topography of age-related changes in non-rapid eye movement sleep electroencephalography. *PLoS One* 2016;11:e0149770.
65 Linares IM, Zuardi AW, Pereira LC, *et al*. Cannabidiol presents an inverted U-shaped dose-response curve in a simulated public speaking test. *Braz J Psychiatry* 2019;41:9–14.
66 Taylor DJ, Lichstein KL, Durrence HH, *et al*. Epidemiology of insomnia, depression, and anxiety. *Sleep* 2005;28:1457–64.
67 Belleville G, Cousineau H, Levrier K, *et al*. Meta-Analytic review of the impact of cognitive-behavior therapy for insomnia on concomitant anxiety. *Clin Psychol Rev* 2011;31:638–52.
68 Belendiuk KA, Babson KA, Vandrey R, *et al*. Cannabis species and cannabinoid concentration preference among sleep-disturbed medicinal cannabis users. *Addict Behav* 2015;50:178–81.
69 Pearce DD, Mitsouras K, Irizarry KJ. Discriminating the effects of *cannabis sativa* and *cannabis indica:* a web survey of medical cannabis users. *J Altern Complement Med* 2014;20:787–91.
70 Cousens K, DiMascio A. (–) δ 9 THC as an hypnotic. *Psychopharmacologia* 1973;33:355–64.
71 Bastien CH, Vallières A, Morin CM. Validation of the insomnia severity index as an outcome measure for insomnia research. *Sleep Med* 2001;2:297–307.
72 Buysse DJ, Reynolds CF, Monk TH, *et al*. The Pittsburgh sleep quality index: a new instrument for psychiatric practice and research. *Psychiatry Res* 1989;28:193–213.
73 Johns MW. A new method for measuring daytime sleepiness: the Epworth Sleepiness scale. *Sleep* 1991;14:540–5.
74 Zigmond AS, Snaith RP. The hospital anxiety and depression scale. *Acta Psychiatr Scand* 1983;67:361–70.
75 Martin A, Rief W, Klaiberg A, *et al*. Validity of the brief patient health questionnaire mood scale (PHQ-9) in the general population. *Gen Hosp Psychiatry* 2006;28:71–7.
76 World Health Organisation. *The ICD-10 classification of mental and behavioural disorders: clinical descriptions and diagnostic guidelines*. Geneva: World Health Organization, 1992.
77 Putilov AA, Donskaya OG. Construction and validation of the EEG analogues of the Karolinska sleepiness scale based on the Karolinska drowsiness test. *Clin Neurophysiol* 2013;124:1346–52.
78 Kaida K, Takahashi M, Åkerstedt T, *et al*. Validation of the Karolinska sleepiness scale against performance and EEG variables. *Clinical Neurophysiology* 2006;117:1574–81.
79 Curran SL, Andrykowski MA, Studts JL. Short form of the profile of mood states (POMS-SF): psychometric information. *Psychol Assess* 1995;7:80–3.
80 Parrott AC, Hindmarch I. The Leeds Sleep Evaluation Questionnaire in psychopharmacological investigations - a review. *Psychopharmacology* 1980;71:173–9.
81 Carney CE, Harris AL, Falco A, *et al*. The relation between insomnia symptoms, mood, and rumination about insomnia symptoms. *J Clin Sleep Med* 2013;9:567–75.
82 Perrier J, Bertran F, Marie S, *et al*. Impaired driving performance associated with effect of time duration in patients with primary insomnia. *Sleep* 2014;37:1565–73.
83 Verster JC, Veldhuijzen DS, Volkerts ER. Residual effects of sleep medication on driving ability. *Sleep Med Rev* 2004;8:309–25.
84 Kevin RC, Allsop DJ, Lintzeris N, *et al*. Urinary cannabinoid levels during nabiximols (Sativex®)-medicated inpatient cannabis withdrawal. *Forensic Toxicol* 2017;35:33–44.
85 Schwope DM, Scheidweiler KB, Huestis MA. Direct quantification of cannabinoids and cannabinoid glucuronides in whole blood by liquid chromatography-tandem mass spectrometry. *Anal Bioanal Chem* 2011;401:1273–83.
86 Baiardi S, Mondini S. Inside the clinical evaluation of sleepiness: subjective and objective tools. *Sleep Breath* 2019:1–9.
87 Littner MR, Kushida C, Wise M, *et al*. Practice parameters for clinical use of the multiple sleep latency test and the maintenance of wakefulness test. *Sleep* 2005;28:113–21.
88 Marshall NS, Neill AM, Campbell AJ, *et al*. Randomised controlled crossover trial of humidified continuous positive airway pressure in mild obstructive sleep apnoea. *Thorax* 2005;60:427–32.
89 Aguilar S, Gutierrez V, Sanchez L, *et al*. *Medicinal cannabis policies and practices around the world*. International drug policy Consortium, 2018.

AUTHOR PROOF

4.2 A Randomised Controlled Trial of Cannabidiol and Δ9-Tetrahydrocannabinol in Insomnia Disorder using High-Density EEG

A Randomised Controlled Trial of Combined Cannabidiol and Δ9-Tetrahydrocannabinol in Insomnia Disorder using High-Density EEG

As prepared for submission to Nature Communications, November 2022

Anastasia Suraev[1,2,3,4], Iain S McGregor[1,3,4], Danielle McCartney[1,3,4], Tancy Kao,[1] Angela L D'Rozario.[1,2,4], Nathaniel S. Marshall[1,5], Keith Wong [6], Brendon J Yee [6], Christopher Gordon [1,5], Delwyn Bartlett[1], Chris Irwin[7], Ryan Vandrey[8], Ronald R. Grunstein[1,6], Camilla M Hoyos[1,2,4]

[1] Centre for Sleep and Chronobiology, Woolcock Institute of Medical Research, Sydney, Australia

[2] University of Sydney, Faculty of Science, School of Psychology, Sydney, Australia

[3] University of Sydney, Lambert Initiative for Cannabinoid Therapeutics, Sydney, Australia

[4] University of Sydney, Brain and Mind Centre, Sydney, Australia

[5] University of Sydney, Faculty of Medicine and Health, Sydney, Australia

[6] Royal Prince Alfred Hospital, Department of Respiratory and Sleep Medicine, Sydney, Australia

[7] Menzies Health Institute Queensland, Griffith University, Queensland, Australia

[8] Johns Hopkins University, School of Medicine, Baltimore, USA

4.2.1 Abstract

Medical cannabis is an emerging popular alternative to common sleep aids, however clinical evidence is limited. We conducted a randomised, placebo-controlled, crossover trial using high-density electroencephalography (EEG) in 20 adults with chronic insomnia disorder to determine the acute effects of combined 200 mg cannabidiol (CBD) and 10 mg Δ9-tetrahydrocannabinol (THC) ('CBD/THC') on sleep. Compared to placebo, CBD/THC significantly decreased total sleep time (-24.5 min, p=0.047) and time spent in REM sleep (-8.1%, p<0.001) and increased REM sleep latency (+65.6 min, p=0.008). No significant changes to subjective sleep outcomes were identified. High-density EEG analysis revealed paradoxical effects with decreased fast activity during N2 sleep (deeper sleep) with decreased delta activity during N3 sleep indicating reduced sleep depth. Increased fast activity during REM sleep is also consistent with heightened arousal. This study shows, for the first time, an acute REM suppressing effect and sleep-reducing effect of cannabinoids in a clinical insomnia population. (Registration: ACTRN12619000714189).

4.2.2 Introduction

Insomnia disorder is a highly prevalent sleep condition affecting around 10-30% of the general population depending on defining criteria used.[1] It is characterised by persistent difficulty initiating and/or maintaining sleep for ≥3 nights per week for ≥3 months and is associated with significant daytime impairment and distress.[2] Insomnia is a key risk factor for psychopathology such as depression, anxiety, and alcohol dependence,[3] cardiovascular disease mortality,[4] and poorer quality of life;[5] highlighting a strong need for clinical intervention. Cognitive behaviour therapy for insomnia (CBTi) is the first-line treatment and can be effective. CBTi implementation and success, however, is limited by barriers to access, cost, and delays in perceived benefits.[6] Short-term ("as-needed") pharmacological interventions including benzodiazepines, orexin antagonists (e.g., lemborexant), and Z-drugs (e.g., zolpidem) are widely prescribed.[7] However, undesirable side effects such as daytime somnolence, cognitive and/or memory impairment, and increased risk of falls and fractures,[7] necessitates caution in prescribing, and drives research into novel alternative therapies.

Cannabis products, primarily those containing the cannabinoids Δ^9-tetrahydrocannabinol (THC) and cannabidiol (CBD), are increasingly used in the community to treat insomnia.[8] Although some evidence supports cannabinoid efficacy in insomnia disorder[9] and sleep apnea,[10] most studies have been limited by small sample sizes, inadequate designs, and high risk of bias.[11] Further, few studies have characterised potential therapeutic effects of cannabinoids using validated objective assessments such as polysomnography (PSG). Advanced techniques such as power spectral analysis can provide fine-grained and sophisticated analyses of EEGs during sleep.[12] A recent systematic review and meta-analysis of studies using EEG spectral analysis showed that insomnia disorder was associated with increased beta power during sleep and wakefulness, in support of the 'round-the-clock' hyperarousal phenomenon of insomnia.[13] Novel techniques such as high-density EEG combine the superior temporal resolution of EEG recordings with high spatial resolution to explore the topographic distribution of these alterations.[14] A study using 256

channel high-density EEG showed that patients with insomnia had more high-frequency EEG activity across the sensory and sensorimotor brain regions than normal sleepers during NREM sleep.[15] This suggests that, even during deep sleep, some brain regions in patients with insomnia are still 'awake' lending support to the local sleep/wakefulness dysregulation hypothesis in insomnia.[16] However, no study to-date has applied such techniques to explore the physiological mechanisms underlying pharmacological intervention in insomnia disorder.

Here, we employed a randomised, placebo-controlled, crossover design to explore the acute effects of an oral cannabinoid treatment on sleep architecture in individuals with chronic insomnia disorder using polysomnography with high-density EEG.

4.2.3 Methods

Trial Design

This randomised, double-blinded, placebo-controlled, crossover trial was approved by Bellberry Human Research Ethics Committee (2018-04-284) and conducted in accordance with the guidelines of the International Council for Harmonisation, principles of the Declaration of Helsinki, and local regulations. The trial was registered with the Australian New Zealand Clinical Trials Registry (ACTRN12619000714189) and the trial protocol was published elsewhere.[17] The full trial protocol is available upon request. The Woolcock Institute of Medical Research, Sydney, Australia was the site and the sponsor.

Participants

Twenty adults aged 35-65 years presenting with chronic insomnia, determined clinically as self-reported difficulty initiating and/or maintaining sleep on ≥3 nights per week and for ≥3 months and an Insomnia Severity Index (ISI) score >15 were recruited. Participants were recruited via referral from sleep specialists or psychologists, social media, and television advertisement. Exclusion criteria were as follows: (1) a clinically significant prior adverse response to cannabis,

cannabinoid products or synthetic cannabinoids; (2) reported use of cannabis within the past three months (abstinence confirmed with a urinary drug screen); (3) past or present history of alcohol and/or drug (including cannabis) dependence; (4) shift work or trans-meridian travel (two time zones) over the past month; (5) use of any modality of treatment for insomnia (including cognitive behaviour therapy) within three months; (6) history of major psychiatric disorder in the last 12 months (except clinically-managed mild depression or anxiety); (7) past or present history of suicidal ideation; (8) known clinically relevant cardiovascular abnormalities; (9) pregnancy or lactation; (10) current use of medications that (a) affect the central nervous system (CNS) (e.g., hypnotics, antidepressants) or (b) induce or inhibit cytochrome (CYP) 450 enzyme system, or (c) are metabolised by CYP enzymes that are inhibited by CBD; (11) medical conditions that result in frequent need to get out of bed (e.g., nocturia); (12) required to complete mandatory workplace or court-ordered drug testing.

Trial Procedures

All participants were informed about the nature and risk of experimental procedures by a sleep specialist and the trial coordinator before their written informed consent was obtained. Initial eligibility was ascertained during a clinical interview at the screening visit which included an electrocardiogram (ECG) to confirm absence of any cardiac abnormalities, urinary drug test (DrugCheck® NxStep Onsite Urine Drug Test) to verify abstinence from alcohol, cannabis and illicit drugs, and a pregnancy test (as applicable; Human Chorionic Gonadotrophin Cassette, Alere™) to rule out pregnancy. Participants then completed an overnight in-laboratory diagnostic sleep study to rule out presence of other sleep disorders (i.e., sleep apnea, periodic limb movements). All participants attended a separate familiarization visit where they wore the high-density EEG sensor cap during a short 'nap' (20 minutes). Participants were instructed to abstain from caffeine and alcohol (≥24 h) prior to each treatment session and to avoid using illicit drugs (including cannabis) and all CNS-active medications including hypnotics for the duration of the

trial. One week prior to each treatment session, participants were encouraged to maintain regular sleep- and wake-onset times as best as possible and complete a sleep diary. These data were used to determine each participant's individual sleep-onset time for the treatment sessions.

On the day of the treatment session, a brief medical screen with the study doctor and repeat urinary drug and pregnancy tests (as applicable) were completed. Participants then completed two 24-hour overnight treatment sessions during which they were administered CBD/THC or placebo in random order with each treatment session separated by a minimum ≥7 days washout period. In both treatment sessions, participants slept in the same bedroom allocated to them at the diagnostic sleep study to provide familiarity with the testing environment.

Randomisation

Each participant was randomly allocated to one of two treatment sequences: (1) CBD/THC–placebo, or (2) placebo–CBD/THC. The sequences were computer-generated using a simple 1:1 randomisation prepared by the trial epidemiologist (NSM) and sequentially numbered using identical containers according to the randomisation sequence were prepared by the drug distributor; neither the statistician nor the distributor had any contact with any prospective or enrolled participants. The sequence was held in a central location and only accessible to the trial epidemiologist, drug distributer, and the principal investigator (in the event of a serious adverse event). All participants, trial personnel (including study doctors), and the outcome assessors were blind to the treatment allocation. The study doctor (KW, BY, SS) made the decision to randomise the participant. To assess blinding success, after each treatment session, participants were asked to guess the treatment they had received the night before (i.e., "CBD/THC', 'Placebo', or 'Not sure') and their certainty of the guess (assessed on a 4-point Likert scale: 'Not at all', 'Somewhat', 'Moderately', and 'Extremely').

Investigational Product

The investigational product was a plant-derived oral formulation containing a 20:1 ratio of CBD to THC i.e., 100 mg/mL CBD and 5 mg/mL THC in medium-chain triglyceride (MCT) oil; manufactured at a GMP-certified facility (Linnea SA, Lavertezzo, Switzerland). Neither the placebo nor active treatment contained any other cannabinoids or cannabis constituents (e.g., minor phytocannabinoids, flavonoids, mono- or sesquiterpenes). The matched placebo consisted of MCT oil (only). Each product was administered as a fixed dose of 2 mL (the acute active treatment was 200 mg CBD and 10 mg THC henceforth referred to as 'CBD/THC'). The 20:1 ratio has been extensively studied in paediatric populations with comorbid insomnia symptoms (e.g., intractable epilepsy,[18 19] autism,[20] complex motor disorders[21]) and is currently available on prescription in Australia.[22] Prior naturalistic studies have also suggested that cannabis containing higher concentrations of CBD relative to THC were associated with improved sleep.[23 24] The dose was selected based on a tolerable dose limit for THC (i.e., 10 mg THC dose produced discriminable subjective drug effects such as "drowsiness" without altering cognitive/psychomotor performance among infrequent cannabis users).[25]

The active and placebo treatments did not differ in their visual appearance. Participants were instructed to ingest one peppermint lozenge (Fisherman's Friend Mint; Lofthouse of Fleetwood, England) to mask any possible differences in taste/smell. The trial physician prepared the study drug on the day of each treatment session by drawing 2 mL of the oil solution into a pre-labelled amber single-use plastic syringe secured with a tip cap. The trial coordinator instructed and observed the participant self-administer the fixed dose ~1 h prior to the participant's typical bedtime (as determined on the sleep diary).

Co-primary and secondary outcomes

The two primary outcomes were total sleep time (TST) and wake after sleep onset (WASO) measured (in minutes) as determined by in-laboratory polysomnography. The main secondary outcomes included: (a) sleep architecture metrics as measured on polysomnography; (b) global

EEG power spectral analysis measured using high-density EEG; (c) subjective ratings of changes to sleep-wake behaviour as assessed on the Leeds Sleep Evaluation Questionnaire (LSEQ), and (d) adverse event profile. No changes were made to the trial outcomes after the study commenced.

Data collection

Polysomnography

Participants underwent in-laboratory high-density EEG polysomnography using 256-channels (Electrical Geodesics Inc., Eugene, OR), as well as standard monitoring with electrooculogram (EOG), submental electromyogram (EMG), electrocardiogram (ECG), bilateral tibial EMG, respiratory inductance plethysmography, pulse oximetry, and a position sensor. Participants were allowed to go to bed at their habitual bedtime and sleep undisturbed in the laboratory for 8 hours. Recordings were scored according to the AASM criteria[26] by an experienced sleep scientist using ProFusion PSG v4 software (Compumedics®, Abbotsford, Australia). Sleep staging (including wake, N1, N2, N3 and REM) was performed in 30-sec epochs based on the six high-density EEG channels (F3, F4, C3, C4, O1, O2) re-referenced to the mastoids.

High-density EEG Recordings

Overnight high-density EEG signals were recorded on NetStation Software (Electrical Geodesics Inc.) with sampling ratio 500 Hz. During NetStation acquisition, impedances were less than 150 kW and a 50 Hz notch filter was used to minimize power line noise. A first-order high-pass filter (0.1 Hz) was initially applied in NetStation to mimic common hardware analog filters and eliminate low frequency drift. The data were then band-pass filtered (Kaiser type, 0.3 – 50 Hz) in NetStation.

High-density EEG Pre-Processing Analysis

The raw EEG signals were analysed in MATLAB using custom-built functions based on the EEGLAB toolbox (The MathWorks Inc., Natick, MA). The quality of channels and artifact epochs were determined through semi-automatic identification and visualized inspection. The data were average-referenced to the mean voltage across all good channels after the bad channels and artifact epochs were removed. The excluded channels were interpolated using spherical splines. The data quality was visually confirmed before the spectral power computation. Power spectral density was calculated using Welch's method in 6s data segments (Hamming windows and 50% overlap) for six frequency ranges (low-delta: 0.5-1.5 Hz; delta: 1.5–4.5 Hz; theta: 4.5–8 Hz; alpha: 8–12 Hz; sigma: 12–16 Hz; beta: 16–30 Hz; and gamma: 30-45 Hz) in each sleep stage. We identified 164 channels within a radius of 0.57 from the vertex (Cz), excluding channels overlying the neck, ears, and forehead. The topographic analysis will be constrained to 164 good channels.

Subjective Sleep Assessment

Perceived changes in sleep quality were assessed using the Leeds Sleep Evaluation Questionnaire (LSEQ)[27], a validated 10-item questionnaire exploring four aspects of sleep: getting to sleep (GTS), quality of sleep (QOS), awakening from sleep (AFS), and behaviour following wakefulness (BFW) using a 100 mm visual analog scale assessed at approximately 9.5 h post-drug administration (within an hour of waking).

Plasma Cannabinoids

Blood was collected via venepuncture into EDTA vacutainer tubes (Becton, Dickinson and Company, New Jersey, USA) at approximately 09:00 hours, immediately after the driving performance task. Blood was centrifuged at 1500×*g* for 10 min at 4°C with the supernatant plasma aliquoted and stored in 1.8 mL cryotubes at −80°C until analysis. Plasma was analysed via LC-MS/MS according to previously published methods for cannabinoids (CBD, THC) and their metabolites (11-OH-THC, THC-COOH; 7-COOH-CBD, and 7-OH-CBD).[28]

Sample Size and Statistical Analyses

This protocol was designed to be a single dose, proof-of-concept study to ascertain preliminary efficacy and safety of the study drug in insomnia disorder. As there was no commercially available power calculation software for mixed-model analyses available at present, using a simple paired t-test, a crossover trial of 20 participants was adequately powered to detect an effect size of 0.67 with 80% power at an alpha level of 0.05 (two-tailed). No interim analyses were planned or undertaken, and there were no stopping guidelines.

Data were analysed under the intention to treat principle by the blinded trial coordinator (AS) under supervision (NSM and CH) using SPSS version 26 (IMB Corp., Armonk, NY). Figures were created using GraphPad Prism version 9 (GraphPad Inc., San Diego, CA). Descriptive statistics were calculated for demographic variables and adverse event profile. Linear mixed-model analyses were used to determine differences between the two treatment arms, with order and treatment (CBD/THC vs placebo) as fixed effects and the participant as a random effect. The least-squares means procedure was used in the mixed-model analyses to handle missing data.

Absolute spectral power between two groups were initially using electrode-to-electrode paired t-test (uncorrected $p<0.05$). The topographical differences were assessed using statistical non-parametric mapping (SnPM) with suprathreshold cluster analysis to identify significant channel clusters.[29] This was done in order to correct for type I error in multiple testing. After establishing the critical t value ($t=1.73$) with alpha=0.05, all possible combinations of topographic power maps are randomly shuffled between conditions (10,000 times). For each reshuffling, the size of the largest cluster above the threshold was used to generate a maximal cluster size distribution. The *p*-value for the suprathreshold cluster is then calculated by comparing the actual cluster size to the maximal cluster size distribution. To account for interindividual variability in absolute data, normalized topographic maps were created by taking the z-score across all good channels for each participant. All statistical analyses were performed using MATLAB (The MathWorks Inc.).

4.2.4 Results

Patient Characteristics

Between August 2019 and October 2021, a total of 857 individuals were pre-screened for eligibility with reasons for exclusion outlined in **Figure 1**. Of the 38 individuals considered for inclusion, 18 were ineligible due to failure to meet enrolment criteria, which included a diagnosis of a sleep disorder other than insomnia (n=8), self-exclusion for unknown reason (n=4), current major psychiatric disorder (n=3), clinically relevant cardiovascular abnormality (n=2; i.e., hypertension), and improvement in insomnia symptoms (n=1). The trial was stopped once the predetermined sample size was met. Twenty participants (16 female; mean (SD) age, 47.1 (8.7) years) were randomised, and all completed the trial (**Table 1**). Participant's insomnia symptoms were of moderate severity (20.8 ± 2.5) with no elevated levels of anxiety or depression on the HADS questionnaire, although evidence of mild depression was identified on the PHQ-9. Participants had an average body mass index (BMI) of 25.1 ± 3.7 $kg.m^2$. All participants were analysed by the group they were randomised to, and complete primary outcome data was available in all 20 participants.

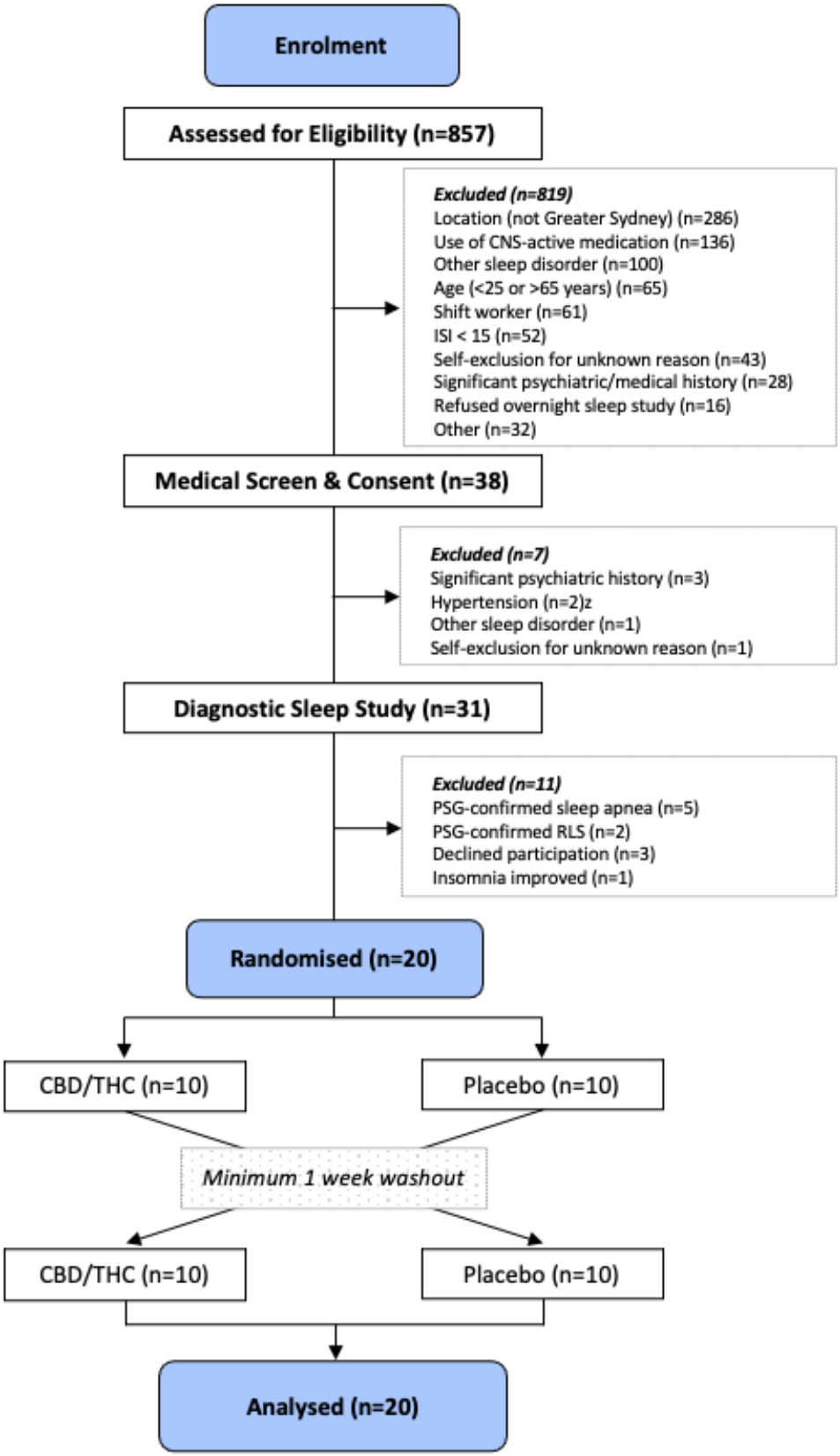

Figure 1 *CONSORT flow diagram.*

Table 1 Participant demographics and characteristics

Characteristic	
Number of participants, *n*	20
Sex (M / F), *n*	4 / 16
Age, mean (SD), *y*	47.1 (8.7)
Weight, *kg*	70.6 (14.7)
BMI, mean (SD), kg/m^2	25.1 (3.7)
Participants with at least some tertiary education, *n*	18
Participants with current employment, *n*	15
Weekly standard drinks, IQR (SD)	1.6 (1.7)
Sleep scales	
ISI total score, mean (SD)	20.8 (2.5)
PSQI total score, mean (SD)	12.6 (3.2)
ESS total score, mean (SD)	4.4 (4.2)
Psychiatric scales	
HADS total score, mean (SD)	
Anxiety	5.3 (3.5)
Depression	3.8 (3.5)
PHQ-9 total score, mean (SD)	7.5 (4.1)

AHI Apnea-Hypopnea Index; *BMI* Body mass index; *ESS* Epworth Sleepiness Scale; *HADS* Hospital Anxiety and Depression Scale; *ISI* Insomnia Severity Index; *PHQ-9* Patient Health Questionnaire 9; *PSQI* Pittsburgh Sleep Quality Inventory

Objective Sleep Outcomes

Compared to placebo, CBD/THC significantly decreased TST (-24.5 min [95%CI 0.01 to 0.13], p=0.047, d=-0.49) with no significant change to WASO (+10.7 min [95%CI -0.99 to 0.94], p=0.422, d=0.19) (**Table 2**). CBD/THC significantly increased time spent in stage N2 sleep (+5.3% [95%CI -0.98 to -0.10], p=0.019, d=0.58) while reducing time spent in REM sleep (-8.1% [95%CI 0.73 to 1.39], p<0.001, d=-1.53) relative to placebo and increased latency to REM sleep (+65.6 min [95%CI -1.34 to -0.23], p=0.008, d=0.68). Post-hoc analysis showed that CBD/THC consistently decreased percentage of time spent in REM sleep during the first (-4.7% [95%CI 0.09 to 1.39], p=0.028, d=-0.55), second (-9.5% [95%CI 0.30 to 1.44], p=0.005, d=-0.70), and third tertile (-11.4% [95%CI 0.34 to 1.17], p=0.001, d=-0.81) of sleep (see **Supplementary Table S1** in

Appendix D). No other significant differences in objective sleep parameters were identified (all p>0.05). No significant treatment order effects were observed for any of the objective sleep outcomes (or any other outcomes analysed in the present study) (all p's>0.05). There were no subgroup or adjusted analyses.

Table 2 Group mean (SD) objective sleep measures during CBD/THC and placebo (n=20)

	CBD/THC (n=20)	Placebo (n= 20)	p value[a]	Cohen's d [95% CI]
Sleep efficiency (%)	78.6 (11.0)	82.8 (9.9)	0.119	-0.37 [-0.83, 0.08]
Sleep onset latency, min	28.8 (25.6)	20.7 (19.3)	0.189	0.31 [-0.14, 0.76]
Total sleep time, min	371.8 (62.7)	396.3 (48.0)	0.047	-0.49 [-0.95, -0.03]
Wake after sleep onset, min	72.2 (46.4)	61.5 (39.6)	0.422	0.19 [-0.25, 0.63]
N1 sleep (%)	18.1 (9.8)	15.1 (6.4)	0.110	0.37 [-0.09, 0.82]
N2 sleep (%)	45.2 (8.4)	39.9 (7.7)	0.019	0.59 [0.11, 1.06]
N3 sleep (%)	22.5 (9.9)	22.7 (8.5)	0.917	-0.02 [-0.46, 0.42]
NREM, min	317.4 (48.4)	308 (41.4)	0.377	0.21 [-0.24, 0.65]
REM sleep (%)	14.2 (5.4)	22.3 (6.1)	<0.001	-1.53 [-2.18, -0.88]
REM latency, min	193.8 (74.8)	128.2 (58.7)	0.008	0.68 [0.19, 1.16]
Arousal index (TST), n (%)	27.6 (13.5)	24.5 (14.6)	0.203	0.29 [-0.16, 0.74]

N3 Stage 3 non-REM sleep (slow wave sleep); *NREM* non-rapid eye movement sleep; *REM* rapid eye movement sleep; *SOL* sleep onset latency; *WASO* wake after sleep onset

Subjective Sleep Outcomes

There were no significant differences between CBD/THC and placebo on any LSEQ domain (all p>0.05) (**Figure 2**).

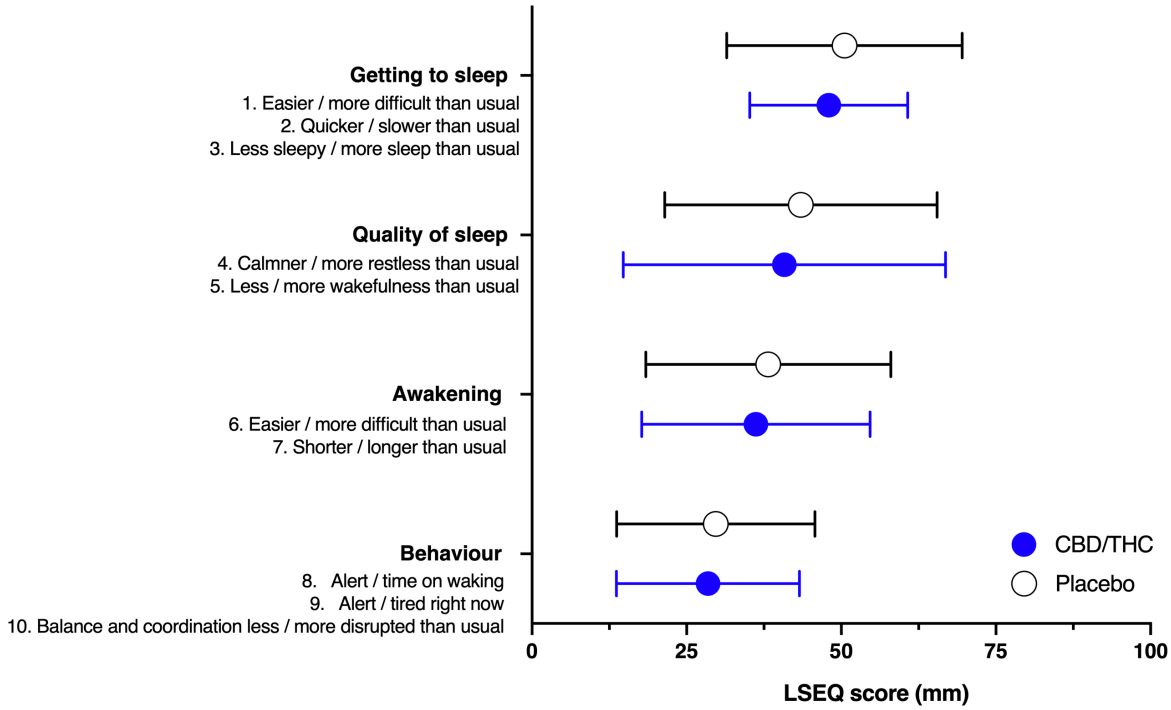

Figure 2 *Participants self-ratings across four domains of the Leeds Sleep Evaluation Questionnaire (LSEQ) as assessed the morning after CBD/THC and placebo treatment. Numbers are expressed as mean and standard deviation (SD).*

Global Power Spectral Analysis using High-Density EEG

Figure 3 shows the high-density EEG global spectral power *t*-value plots across frequency and sleep stages($t(18)=1.73$, all $p<0.05$). Relative to placebo, CBD/THC significantly decreased gamma and beta EEG activity overlying the posterior and frontal cortex, respectively, and increased sigma activity in the frontal cortex during N2 sleep During N3 sleep, there was a significant decrease in delta activity in the posterior region with CBD/THC treatment relative to placebo. CBD/THC produced a significance increase in alpha and beta activity during REM sleep in the parietal region relative to placebo.

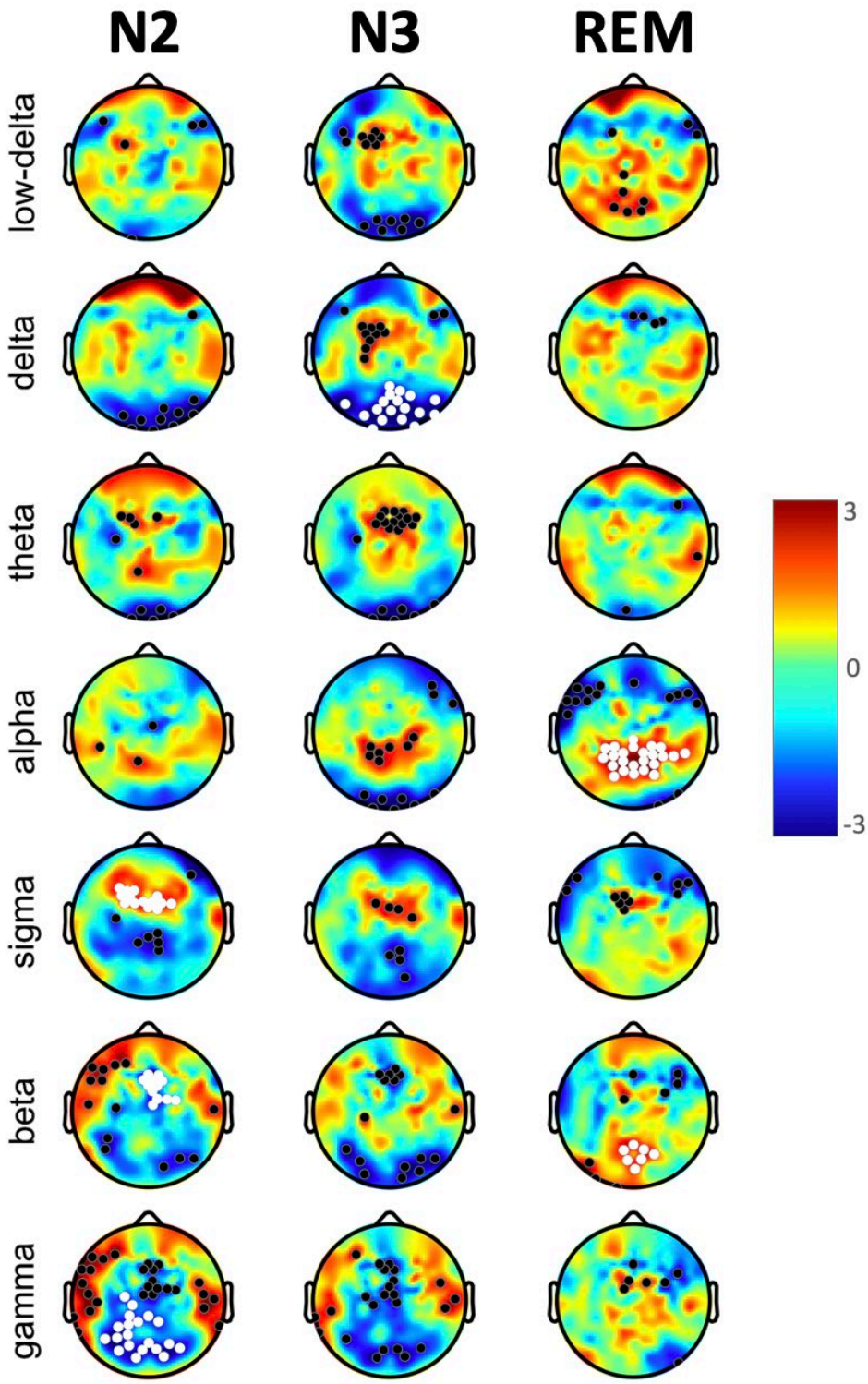

Figure 3 *High-density EEG t-value plots arranged vertically by sleep stage, and horizontally by frequency band. White dots represent channels where CBD/THC treatment was significantly different to placebo (paired t-test; corrected SnPM, p<0.05). Cooler (blue) values represent a decrease in absolute EEG power in CBD/THC treatment relative to controls (CBD/THC < placebo) and warmer (red) colours represent an increase (CBD/THC>placebo).*

Plasma Cannabinoid Concentration

The median (IQR) length of time between CBD/THC administration and blood sampling (washout; days) and the proportion of participants with detectable concentrations of CBD, THC, and their major phase-I metabolites in plasma are reported in **Supplementary Table S2** in Appendix D; the actual concentrations are reported in **Supplementary Table S3.** Overall, 60%, 70%, 40% and 0% of participants were found to have detectable concentrations of CBD, 7-COOH-CBD, 7-OH-CBD, and 6-OH-CBD respectively in plasma ≥7 post-drug administration. In contrast, residual THC was never observed. However, residual 11-COOH-THC and 11-OH-THC were observed in plasma in 40% and 50% of participants.

Adverse Events and Success of Blinding

No serious adverse events were reported, and no participant withdrew from the trial. Eighty-five mild, non-serious, adverse events were reported: 55 during CBD/THC treatment recorded from 16 participants and 30 during placebo recorded from 13 participants. The most common side effects related to CBD/THC were dry mouth and drowsiness/sedation (**Table 3**). All adverse events had either resolved overnight or soon after waking. In terms of blinding, 14 of 20 participants (70%) correctly guessed they were receiving CBD/THC. When receiving placebo, 12 of 20 participants (60%) correctly guessed they were receiving placebo and two participants (10%) were 'not sure' (see **Supplementary Table S4** in Appendix D).

Table 3 Adverse events reported during CBD/THC and placebo

	CBD/THC (*n*=20)	Placebo (*n*=20)
Participants with any AE, n (%)	16 (80%)	13 (65%)
Total numbers of AEs	55	30
Adverse event, n (%)		
Dry mouth	10 (50%)	1 (5%)
Drowsiness/sedation	10 (50%)	6 (30%)
Fatigue	8 (40%)	7 (35%)
Disorientation/confusion	4 (20%)	1 (5%)
Lethargy	4 (20%)	5 (25%)
Dizziness	3 (15%)	1 (5%)
Nausea	3 (15%)	3 (15%)
Feeling intoxicated	3 (15%)	1 (5%)
Visual disturbance	2 (10%)	0
Lightheadedness	1 (5%)	0
Headache	1 (5%)	3 (15%)
Effortful breathing	1 (5%)	0
Vivid dreams	1 (5%)	0
Anxiety	1 (5%)	2 (10%)
Heart palpitations	1 (5%)	0
Urinary retention	1 (5%)	0
Heartburn	1 (5%)	0

4.2.5 Discussion

In the present study, we investigated the effects of a single oral dose of CBD/THC (containing 200 mg CBD and 10 mg THC) on sleep in chronic insomnia disorder using overnight polysomnography with high-density EEG. Our findings showed a reduction in total sleep time and time spent in REM sleep with CBD/THC with no effect on WASO or subjective sleep outcomes. This is the first study to show clear acute REM suppressing effects of cannabinoids in people with insomnia disorder. We found that CBD/THC paradoxically decreased high-frequency EEG activity during N2 sleep suggestive of deeper sleep and decreased delta activity during N3 sleep indicating reduced sleep depth. CBD/THC also increase high frequency EEG activity during REM sleep suggestive of heightened arousal. Overall, this important preliminary study suggests caution in assuming cannabinoids are effective in insomnia disorder.

The reduction in TST with CBD/THC may in part relate to the dose used, the cannabis use history of participants, and the use of acute rather than repeated dosing. It is possible that a single dose of oral 10 mg THC dose was not optimal for sedative effects and may have inadvertently caused stimulatory effects,[30] particularly in infrequent cannabis users. In one study, THC (oral; 10 mg) significantly increased heart rate, 'stimulant-like' subjective effects, and anxiety relative to placebo in 16 infrequent cannabis users (lifetime use <15 times).[31] Infrequent or cannabis-naïve users tend to exhibit greater sensitivity to the acute pharmacodynamic effects of cannabis compared to regular users who typically develop tolerance to the adverse effects of THC.[32] One study showed that THC (vaporised; 8 mg) significantly increased heart rate and subjective measures of intoxication relative to placebo, with a relatively greater degree of intoxication in infrequent cannabis users compared to frequent cannabis users.[33] It is known that the first day of treatment with THC is often associated with the highest number of treatment-related adverse events and that treatment becomes better tolerated with repeated dosing.[34] Notably, a recent 2-week trial with a THC-dominant product (oral; 10 and 20 mg per night)

significantly improved ISI by 5.1 points relative to placebo in insomnia disorder with only minor and self-limiting side effects.[9]

A single oral dose of 200 mg and 10 mg THC was well tolerated with no withdrawals due to an adverse event. Sixteen out of 20 participants (80%) experience at least one adverse event after a single dose of CBD/THC, with dry mouth, drowsiness/sedation, and fatigue being most frequently reported, comparable to other studies administering THC-based formulations.[35] All adverse events were mild and had either resolved overnight or upon waking. Future studies using repeated dosing designs with cannabinoids should explore the frequency and severity of possible withdrawal symptoms of which sleep disturbances are a hallmark feature.[36]

Prior literature has indicated that cannabis, specifically THC, can sometimes supress REM sleep,[37-40] although findings are mixed[10 41] while in another study CBD (oral; 300 mg) had no significant effect on sleep architecture in healthy individuals.[42] Here, we showed that acute CBD/THC increased latency to REM sleep and suppressed REM sleep with no evidence of REM sleep fragmentation (i.e., increased arousals during REM sleep). Our findings somewhat converge with a recent study showing a significant increase in REM sleep latency (+54.2 min) and a non-significant trend towards reduced REM sleep (-3.5%, p=0.055) following 2-week treatment with a THC-dominant formulation (oral; 10-20 mg THC/night) in patients with insomnia.[9] However, the former may be at least partly attributed to a substantial decrease in REM sleep latency from baseline (127 min) to end of week 2 in the placebo group (71 min). This suggests possible tolerance to the effects of cannabinoids on REM sleep with repeated dosing and/or differences between study participant's prior cannabis use history. Of interest, antidepressant drugs inhibit REM sleep,[43] and some believe this is critical to the beneficial therapeutic effects of these drugs on affect.[44] REM sleep alterations (i.e., shortened REM sleep latency and increased REM sleep) are the most prominent feature of sleep architecture in individuals with depression,[43] which may explain why some consumers report antidepressant effects with cannabis use.[45-47] REM sleep suppression, if maintained with repeated dosing, may have clinical relevance for other sleep

disorders such as trauma-associated nightmares (i.e., post-traumatic sleep disorder) and REM sleep behaviour disorder (RBD).[48] Future studies might usefully explore whether lower doses of THC (<10 mg) may be more effective in inducing and maintaining sleep in insomnia disorder without the propensity for suppressing REM sleep.

The reduction in high frequency (i.e., gamma and beta activity) during N2 sleep in the posterior and frontal region following CBD/THC treatment suggests deeper sleep. A pilot study using high-density EEG recordings revealed that insomnia patients had more high frequency EEG activity during NREM sleep relative to good sleepers and that these changes were widespread across the scalp.[15] High frequency EEG activity is one of the most commonly reported physiological correlates of insomnia, and is regarded as a sign of cortical hyperarousal,[13 49] that can be ameliorated by CBT-I.[50 51] Thus, CBD/THC may act upon CNS hyperarousal during sleep in insomnia. Somewhat paradoxical effects of CBD/THC were evident with an increase in delta EEG activity in the posterior cortex during N3 sleep. A reduction in delta activity was unexpected for a drug meant to improve sleep with a meta-analysis showing that patients with insomnia disorder display decreases in delta power during NREM sleep.[13 52] Indeed, CBT-I treatment was associated with a decrease in delta EEG power during NREM sleep compared to a placebo intervention, and this predicted a greater therapeutic effect.[53] The observed increase in sigma EEG activity in the frontal cortex may represent a sleep-protective mechanism (i.e., against external stimuli such as noise),[54] as previously described in patients with insomnia.[55] This may point to a bolstered attempt to protect against sleep disruption from CBD/THC treatment. The significant increase in high frequency (i.e., alpha and beta) activity in the central-posterior region during REM sleep also suggests heightened arousal as previously described.[13] The clinical significance of the more comprehensive findings using high-density power spectral analyses are yet to be determined, particularly with repeated dosing study designs.

A strength of this study is examining effects of cannabinoids using a rigorous randomised, placebo-controlled trial design, reducing the risk of possible confounding factors inherent in observational studies. The application of a regulated and quality-assured cannabinoid formulation to patients with clinician-confirmed insomnia is also a strength. Diagnostic sleep studies were used to rule out other sleep disorders which were common (21%; 8/38 screened participants). Habituation to the testing environment was also provided with participants staying in the same bedroom across all three overnight stays (i.e., diagnostic sleep study and two treatment visits). Each participant also completed of a separate in-person visit to practice wearing the high-density EEG sensor cap during a 20-minute 'nap'. No significant treatment order effects were observed suggesting that ample habituation was achieved. Further, although most participants correctly guessed their treatment order (14/20 for active arm and 12/20 for placebo arm), no improvements in subjective sleep outcomes were observed nor, as mentioned, was there any significant effect of treatment order across any outcome measures.

A limitation of the trial design included the inability to assess the individual contribution of THC and CBD to observed effects. There is emerging evidence that co-administration of CBD with THC may attenuate the adverse effects of THC such as next day drowsiness,[56] however, findings are mixed.[57] Further, cannabinoids, in particular THC, are lipophilic and tend to 'linger' in plasma for a prolonged period.[58] Here, we showed that of the participants who received CBD/THC on the first treatment session and placebo on the second (n=10), residual CBD and its metabolites concentrations were observed in the second arm (placebo) despite a minimum 1-week washout indicating inadequate washout. This novel finding indicates that crossover studies involving cannabinoids should be conducted with caution, particularly when higher doses and/or repeating dosing regimens are used. Despite carryover effects, we did not observe any treatment order effects across any of our primary or secondary outcome variables as previously mentioned. The pharmacological significance of low residual plasma of 7-COOH-CBD is yet unclear

(although there is some evidence to suggest the analogous metabolite of THC, 11-COOH-THC, does not elicit subjective or physiological effects[59]).

Conclusions

This randomised, double-blind, placebo-controlled trial showed that acute oral administration of 200 mg CBD and 10 mg THC reduced total sleep time and REM sleep in chronic insomnia disorder with no effect on subjective sleep outcomes. This was the first study to show a clear acute effect of cannabinoids on REM sleep suppression in a clinical insomnia population. High-density EEG analysis revealed paradoxical effects of CBD/THC treatment that varied in frequency and cortical topography. Cannabinoids and their metabolites were shown to linger in blood plasma longer than anticipated, urging caution in using crossover trial designs with cannabinoids. Future studies should explore (a) whether lower doses of THC (<10 mg) may be more effective in inducing and maintaining sleep in insomnia disorder, and (b) the impact of repeated dosing with cannabinoids on objective sleep outcomes in insomnia disorder.

Funding Statement

This study is funded by the Lambert Initiative for Cannabinoid Therapeutics, a philanthropically funded centre for cannabinoid research at the University of Sydney. Lambert Initiative research staff were co-investigators and played an active role in the trial design and conduct, data analysis, and reporting. AS was supported by the Australian Government Research Training Program (RTP) and the Vice-Chancellor's Research Scholarship (VCRS) at the University of Sydney. CMH (APP1104003) and ALD (APP1107716) were supported by Dementia Research Development Fellowships of the Australian National Health and Medical Research Council-Australian Research Council (NHMRC-ARC). RRG was supported by an NHMRC Senior Principal Research Fellowship (APP1106974). The investigational product was purchased from BOD Australia who were not involved in the conception or design of this study, data analysis (with no access to the data) or the decision to publish. All other commercially available equipment was purchased.

4.2.6 References

1. Aernout E, Benradia I, Hazo J-B, et al. International study of the prevalence and factors associated with insomnia in the general population. *Sleep Med* 2021;82:186-92.
2. The Diagnostic and Statistical Manual of Mental Disorders (5th ed.; DSM–5; American Psychiatric Association, 2013).
3. Hertenstein E, Feige B, Gmeiner T, et al. Insomnia as a predictor of mental disorders: a systematic review and meta-analysis. *Sleep Med Rev* 2019;43:96-105.
4. Ge L, Guyatt G, Tian J, et al. Insomnia and risk of mortality from all-cause, cardiovascular disease, and cancer: Systematic review and meta-analysis of prospective cohort studies. *Sleep Med Rev* 2019;48:101215.
5. Darchia N, Oniani N, Sakhelashvili I, et al. Relationship between sleep disorders and health related quality of life—results from the Georgia SOMNUS study. *Int J of Environ Res and Public Health* 2018;15(8):1588.
6. Koffel E, Bramoweth AD, Ulmer CS. Increasing access to and utilization of cognitive behavioral therapy for insomnia (CBT-I): a narrative review. *J Gen Intern Med* 2018;33(6):955-62.
7. De Crescenzo F, D'Alò GL, Ostinelli EG, et al. Comparative effects of pharmacological interventions for the acute and long-term management of insomnia disorder in adults: a systematic review and network meta-analysis. *Lancet* 2022;400(10347):170-84.
8. Lavender I, McGregor IS, Suraev A, et al. Cannabinoids, insomnia, and other sleep disorders. *Chest* 2022, 162(2):452-465.
9. Walsh JH, Maddison KJ, Rankin T, et al. Treating insomnia symptoms with medicinal cannabis: a randomized, crossover trial of the efficacy of a cannabinoid medicine compared with placebo. *Sleep* 2021;44(11):zsab149.

10. Carley DW, Prasad B, Reid KJ, et al. Pharmacotherapy of apnea by cannabimimetic enhancement, the PACE clinical trial: effects of dronabinol in obstructive sleep apnea. *Sleep* 2018;41(1).
11. Suraev AS, Marshall NS, Vandrey R, et al. Cannabinoid therapies in the management of sleep disorders: a systematic review of preclinical and clinical studies. *Sleep Med Rev* 2020;53:101339.
12. Bastien CH, LeBlanc M, Carrier J, et al. Sleep EEG power spectra, insomnia, and chronic use of benzodiazepines. *Sleep* 2003;26(3):313-17.
13. Zhao W, Van Someren EJ, Li C, et al. EEG spectral analysis in insomnia disorder: A systematic review and meta-analysis. *Sleep Med Rev* 2021;59:101457.
14. Lustenberger C, Huber R. High density electroencephalography in sleep research: potential, problems, future perspective. *Front Neurol* 2012;3:77.
15. Riedner BA, Goldstein MR, Plante DT, et al. Regional patterns of elevated alpha and high-frequency electroencephalographic activity during nonrapid eye movement sleep in chronic insomnia: a pilot study. *Sleep* 2016;39(4):801-12.
16. Buysse DJ, Germain A, Hall M, et al. A neurobiological model of insomnia. *Drug Discov Today: Dis Models* 2011;8(4):129-37.
17. Suraev A, Grunstein RR, Marshall NS, et al. Cannabidiol (CBD) and Δ9-tetrahydrocannabinol (THC) for chronic insomnia disorder ('CANSLEEP'trial): protocol for a randomised, placebo-controlled, double-blinded, proof-of-concept trial. *BMJ Open* 2020;10(5):e034421.
18. Hausman-Kedem M, Menascu S, Kramer U. Efficacy of CBD-enriched medical cannabis for treatment of refractory epilepsy in children and adolescents–An observational, longitudinal study. *Brain Dev* 2018;40(7):544-51.
19. Tzadok M, Uliel-Siboni S, Linder I, et al. CBD-enriched medical cannabis for intractable pediatric epilepsy: the current Israeli experience. *Seizure* 2016;35:41-44.
20. Barchel D, Stolar O, De-Haan T, et al. Oral cannabidiol use in children with autism spectrum disorder to treat related symptoms and co-morbidities. *Front Pharmacol* 2019;9:1521.

21. Libzon S, Schleider LB-L, Saban N, et al. Medical cannabis for pediatric moderate to severe complex motor disorders. *J Child Neurol* 2018;33(9):565-71.

22. Therapeutic Goods Administration. Medicinal cannabis products by active ingredients. Australian Government. Department of Health. Accessed 20 November, 2022. https://www.tga.gov.au/medicinal-cannabis-products-active-ingredients

23. Belendiuk KA, Babson KA, Vandrey R, et al. Cannabis species and cannabinoid concentration preference among sleep-disturbed medicinal cannabis users. *Addict Behav* 2015;50:178-81.

24. Winiger EA, Hitchcock LN, Bryan AD, et al. Cannabis use and sleep: expectations, outcomes, and the role of age. *Addict Behav* 2021;112:106642.

25. Schlienz NJ, Spindle TR, Cone EJ, et al. Pharmacodynamic dose effects of oral cannabis ingestion in healthy adults who infrequently use cannabis. *Drug Alcohol Depend* 2020;211:107969.

26. Berry, R., Quan, S. and Abreu, A. (2020) The AASM Manual for the Scoring of Sleep and Associated Events: Rules, Terminology and Technical Specifications, Version 2.6. American Academy of Sleep Medicine.

27. Parrott A, Hindmarch I. The Leeds Sleep Evaluation Questionnaire in psychopharmacological investigations—a review. *Psychopharmacol* 1980;71(2):173-79.

28. Kevin R, Vogel R, Doohan P, et al. A validated method for the simultaneous quantification of CBD, THC, and their metabolites in human plasma, and application to plasma samples from an oral CBD open label trial. *Drug Test Anal* 2020

29. Jones SG, Riedner BA, Smith RF, et al. Regional reductions in sleep electroencephalography power in obstructive sleep apnea: a high-density EEG study. *Sleep* 2014;37(2):399-407.

30. Cousens K, DiMascio A. (−) δ 9 THC as an hypnotic. *Psychopharmacol* 1973;33(4):355-64.

31. Martin-Santos R, a Crippa J, Batalla A, et al. Acute effects of a single, oral dose of d9-tetrahydrocannabinol (THC) and cannabidiol (CBD) administration in healthy volunteers. *Curr Pharm Des* 2012;18(32):4966-79.

32. Colizzi M, Bhattacharyya S. Cannabis use and the development of tolerance: a systematic review of human evidence. *Neurosci Biobehav Rev* 2018;93:1-25.

33. Solowij N, Broyd S, Greenwood L-m, et al. A randomised controlled trial of vaporised Δ9-tetrahydrocannabinol and cannabidiol alone and in combination in frequent and infrequent cannabis users: acute intoxication effects. *Eur Arch Psychiatry Clin Neurosci* 2019;269(1):17-35.

34. Peters EN, Mosesova I, MacNair L, et al. Safety, pharmacokinetics and pharmacodynamics of Spectrum yellow oil in healthy participants. *J Anal Toxicol* 2022;46(4):393-407.

35. Robson P. Therapeutic aspects of cannabis and cannabinoids. *Br J Psychiatry* 2001;178(2):107-15.

36. Babson KA, Sottile J, Morabito D. Cannabis, cannabinoids, and sleep: a review of the literature. *Curr Psychiatry Rep* 2017;19(4):1-12.

37. Feinberg I, Jones R, Walker JM, et al. Effects of high dosage delta-9-tetrahydrocannabinol on sleep patterns in man. *Clin Pharm Therap* 1975;17(4):458-66.

38. Freemon FR. Effects of marihuana on sleeping states. *JAMA* 1972;220(10):1364-65.

39. Pivik R, Zarcone V, Dement W, et al. Delta-9-tetrahydrocannabinol and synhexl: Effects on human sleep patterns. *Clin Pharm Therap* 1972;13(3):426-35.

40. Tassinari C, Ambrosetto G, Peraita-Adrado M, et al. The Neuropsychiatric Syndrome of Δ 9-Tetrahydrocannabinol and Cannabis Intoxication in Naive Subjects. Marihuana and Medicine: Springer 1999:649-64.

41. Barratt ES, Beaver W, White R. The effects of marijuana on human sleep patterns. *Biol Psychiatry* 1974; *8*(1), 47–54.

42. Palagini L, Baglioni C, Ciapparelli A, et al. REM sleep dysregulation in depression: state of the art. *Sleep Med Rev* 2013;17(5):377-90.

43. Vogel GW, Vogel F, McAbee RS, et al. Improvement of depression by REM sleep deprivation: new findings and a theory. *Arch Gen Psychiatry* 1980;37(3):247-53.

44. Lintzeris N, Driels J, Elias N, et al. Medicinal cannabis in Australia, 2016: the cannabis as medicine survey (CAMS-16). *Med J Aust* 2018;209(5):211-16.

45. Lintzeris N, Mills L, Abelev SV, et al. Medical cannabis use in Australia: consumer experiences from the online cannabis as medicine survey 2020 (CAMS-20). *Harm Reduct J* 2022;19(1):1-10.

46. Lintzeris N, Mills L, Suraev A, et al. Medical cannabis use in the Australian community following introduction of legal access: the 2018–2019 Online Cross-Sectional Cannabis as Medicine Survey (CAMS-18). *Harm Reduct J* 2020;17(1):1-12.

47. Maddison KJ, Kosky C, Walsh JH. Is There a Place for Medicinal Cannabis in Treating Patients with Sleep Disorders? What We Know so Far. *Nat Sci Sleep* 2022;14:957.

48. Merica H, Blois R, Gaillard JM. Spectral characteristics of sleep EEG in chronic insomnia. *Eur J Neurosci* 1998;10(5):1826-34.

49. Cervena K, Dauvilliers Y, Espa F, et al. Effect of cognitive behavioural therapy for insomnia on sleep architecture and sleep EEG power spectra in psychophysiological insomnia. *J Sleep Res* 2004;13(4):385-93.

50. Jacobs GD, Benson H, Friedman R. Home-based central nervous system assessment of a multifactor behavioral intervention for chronic sleep-onset insomnia. *Behav Ther* 1993;24(1):159-74.

51. Krystal AD, Edinger JD, Wohlgemuth WK, et al. NREM sleep EEG frequency spectral correlates of sleep complaints in primary insomnia subtypes. *Sleep* 2002;25(6):626-36.

52. Krystal AD, Edinger JD. Sleep EEG predictors and correlates of the response to cognitive behavioral therapy for insomnia. *Sleep* 2010;33(5):669-77.

53. Dang-Vu TT, McKinney SM, Buxton OM, et al. Spontaneous brain rhythms predict sleep stability in the face of noise. *Curr Biol* 2010;20(15):R626-R27.

54. Spiegelhalder K, Regen W, Feige B, et al. Increased EEG sigma and beta power during NREM sleep in primary insomnia. *Biol Psychol* 2012;91(3):329-33.

55. Nicholson AN, Turner C, Stone BM, et al. Effect of Δ-9-tetrahydrocannabinol and cannabidiol on nocturnal sleep and early-morning behavior in young adults. *J Clin Psychopharmacol* 2004;24(3):305-13.

56. Boggs DL, Nguyen JD, Morgenson D, et al. Clinical and preclinical evidence for functional interactions of cannabidiol and Δ9-tetrahydrocannabinol. *Neuropsychopharmacol* 2018;43(1):142-54.

57. Johansson E, Halldin M, Agurell S, et al. Terminal elimination plasma half-life of Δ1-tetrahydrocannabinol (Δ1-THC) in heavy users of marijuana. *Eur J Clin Pharmacol* 1989;37(3):273-77.

58. Ujváry I, Grotenhermen F. 11-Nor-9-carboxy-Δ 9-tetrahydrocannabinol–a ubiquitous yet underresearched cannabinoid. A re-view of the literature. *Cannabinoids* 2014;9:1-8.

4.3 The ‘next day’ effects of combined cannabidiol and Δ9-tetrahydrocannabinol in insomnia disorder: a randomised, placebo-controlled trial

The 'next day' effects of combined cannabidiol and Δ9-tetrahydrocannabinol in insomnia disorder: a randomised, placebo-controlled trial

As prepared for submission to Neuropsychopharmacology, November 2022

Anastasia Suraev[1,2,3,4], Iain S McGregor[1,3,4], Danielle McCartney[1,3,4], Angela L D'Rozario.[1,2,4], Nathaniel S. Marshall[1,5], Keith Wong [6], Brendon J Yee [6], Christopher Gordon [1,5], Delwyn Bartlett[1], Chris Irwin[7], Ryan Vandrey[8], Ronald R. Grunstein[1,6], Camilla M Hoyos[1,2,4]

[1] Centre for Sleep and Chronobiology, Woolcock Institute of Medical Research, Sydney, Australia

[2] University of Sydney, Faculty of Science, School of Psychology, Sydney, Australia

[3] University of Sydney, Lambert Initiative for Cannabinoid Therapeutics, Sydney, Australia

[4] University of Sydney, Brain and Mind Centre, Sydney, Australia

[5] University of Sydney, Faculty of Medicine and Health, Sydney, Australia

[6] Royal Prince Alfred Hospital, Department of Respiratory and Sleep Medicine, Sydney, Australia

[7] Menzies Health Institute Queensland, Griffith University, Queensland, Australia

[8] Johns Hopkins University, School of Medicine, Baltimore, USA

4.3.1 Abstract

Cannabis and its major cannabinoid constituents, Δ^9-tetrahydrocannabinol (THC) and cannabidiol (CBD), are increasingly used as an alternative to common sleep aids. However, THC is intoxicating and can cause cognitive and psychomotor impairment. It is unclear whether impairment is present the day after evening cannabinoid use (i.e., a 'hangover' effect). Here, we report the 'next day' effects following acute administration of an oral cannabinoid formulation containing a 200 mg CBD and 10 mg THC ('CBD/THC') in patients with insomnia disorder. Twenty participants [16 female; mean (SD) age, 47.1 (8.7) years] with clinician-diagnosed insomnia disorder completed two 24-hour in-laboratory treatment sessions during which they were randomised to receive CBD/THC or placebo. Next-day (12 h+ post-treatment) neurocognitive function, alertness, and simulated driving performance was assessed. The reliability and accuracy of two point-of-collection testing (POCT) oral fluid testing devices, Securetec DrugWipe 5s (DW5s) and Dräger DrugTest 5000 (DT5000), to detect THC the morning after administration was also examined. Apart from a possible (subtle) increase in subjective measure of sleepiness, no reliable changes in 'next day' function including cognitive function, driving performance, and objective measures of alertness were observed. Accuracy on the POCT devices was lowest at 0.5 h post-drug administration yielding the highest number of false positive and false negative tests but performed better the following day. Overall, it appears that a single, oral dose of combined 200 mg CBD and 10 mg does not substantially impair 'next day' function in individuals with insomnia disorder (Registration: ACTRN12619000714189).

4.3.2 Introduction

Insomnia disorder is the most common sleep disorder that affects up to 30% of the general population at any given time.[1] It is characterised by subjective difficulties with falling asleep and/or staying asleep and is associated with significant daytime impairment.[2] It is often the daytime impairment (e.g., fatigue and psychological distress) that prompted help-seeking behaviour.[3] First-line treatment includes cognitive behaviour therapy for insomnia (CBT-I) which can be effective[4] [5], however, certain barriers limit its success such as cost, access to a therapist, and delayed perceived benefits.[6] Short-term use of pharmacological therapies such as benzodiazepines, Z-drugs (e.g., zolpidem), and orexin antagonists (e.g., lemborexant) can be useful, however, undesirable side effects of these drugs such as daytime somnolence, cognitive impairment, and increased falls and fractures,[4] restrict their use; igniting interest in novel alternative therapies.

Cannabinoids such as Δ^9-tetrahydrocannabinol (THC) and cannabidiol (CBD), alone or in combination, are becoming increasingly popular alternative to common sleep aids. Sleep disorders are one of the most common indications treated with medical cannabis, after pain and anxiety.[7-11] Although clinical evidence to support the routine use of cannabinoids in the treatment of sleep disorders is limited,[12] there is an increasing number of pre-registered trials exploring the effects of CBD, THC, and their combination in the treatment of insomnia.[13-15] Indeed, a recent 2-week randomised controlled trial of a THC-dominant oral formulation showed an improvement in subjective sleep outcomes (i.e., Insomnia Severity Index) in patients with insomnia disorder.[16] However, no study to-date has explore the 'next day' effects of cannabinoids on daytime function in individuals with insomnia disorder.

Cannabis and THC (even for therapeutic purposes) can impair cognitive and psychomotor performance. Although CBD is non-intoxicating and does not appear to induce impairment,[17] THC is a well-known intoxicant that can cause sedation, sensory changes, and cognitive and psychomotor impairment. This is significant and can potentially increase the risk of error, accident and injury when operating a motor vehicle or equipment or engaging in other safety-sensitive tasks.

Epidemiological studies indicate that cannabis (i.e., THC) intoxication is associated with increased crash risk and culpability.[18 19] Acute cannabis intoxication increases standard deviation of lateral position (SDLP), an index of lane weaving and a validated measure of alcohol- and drug-induced driving impairment.[20] It is therefore unclear whether impairment is present the *next day* in individuals using cannabis and THC-containing cannabis products in the evening to treat a sleep disorder (e.g., insomnia).

A recent meta-analysis confirmed that acute THC administration (i.e., <12 h post-drug administration) impairs aspects of driving performance (e.g., SDLP, tracking, divided attention) with most drivers predicted to recover within ~5 hours (all recovered by ~7 hours; i.e., likely within the usual overnight period) of inhaling 20 mg THC.[21] However, a subsequent systematic review noted that rigorous studies investigating the next-day effects of THC (i.e., >8 hours after use) are lacking.[22] However, most included studies involved non-medicinal and inhaled methods of administration, and half of the studies that reported no 'next day' residual THC effects failed to demonstrate acute impairment (i.e., THC-related impairment occurring <8 h post-treatment). Oral THC-induced impairment may take longer to subside due to its slower and often unpredictable rate of absorption and delayed peak plasma concentrations relative to inhaled methods.[23]

The reliability of common methods of identifying cannabis-impaired individuals at the roadside and in the workplace (e.g., oral fluid drug tests) also warrants consideration. Fast and non-invasive techniques that can be used at the roadside such as point-of-collection testing (POCT) devices are commonly used in Europe[24-27] and Australia[28]. POCT devices detect the presence of THC at or above a given concentration in oral fluid.[29] The accuracy and reliability of these devices have been previously criticised in previous studies of inhaled cannabis[30] and usability limited to very recent cannabis use (i.e., <60 minutes). However, no study to-date has examined the performance characteristics of POCT devices the *next day* following oral administration of an oil containing THC.

Therefore, the aim of this study was to explore the 'next day' effects (>10 hours post-drug administration) of an oral formulation containing 200 mg CBD and 10 mg THC oral on cognition function, alertness, and simulated driving performance in patients with chronic insomnia disorder. Given the current legal framework for driving under the influence of cannabis in Australia (i.e., detection of THC in saliva with no functional assessment), we also examined the accuracy and reliability of two commonly used POCT devices (Securetec DrugWipe 5s and Dräger Drug Test 5000) in detecting THC in oral fluid the morning after evening administration.

4.3.3 Methods

Trial Design

This randomised, double-blind, placebo-controlled, crossover trial was approved by Bellberry Human Research Ethics Committee (2018-04-284) and conducted in accordance with the guidelines of the International Council for Harmonisation, principles of the Declaration of Helsinki, and local regulations. The Woolcock Institute of Medical Research, Sydney, Australia, a research institute and specialist sleep clinic in inner suburban Sydney, Australia, was the study site and sponsor. The trial protocol is published elsewhere[31] and registered with the Australian New Zealand Clinical Trials Registry (ACTRN12619000714189).

Participants

Twenty adults aged between 35-65 years presenting with chronic insomnia, determined clinically as self-reported difficulty initiating and/or maintaining sleep on >3 nights per week and for >3 months and an Insomnia Severity Index (ISI) score >15, were recruited. Participants were recruited via referral from sleep specialists or psychologists, social media, and a television advertisement. The main exclusion criteria were as follows: (1) reported use of cannabis within the past three months (abstinence confirmed with a urinary drug screen); (2) shift work or trans-meridian travel (two time zones) over the past month; (3) use of any modality of treatment for insomnia (including cognitive behaviour therapy) within three months; (4) current use of

medications that (a) affect the central nervous system (CNS) (e.g., hypnotics, antidepressants) or (b) induce or inhibit cytochrome (CYP) 450 enzyme system, or (c) are metabolised by CYP enzymes that are inhibited by CBD; (5) required to complete mandatory workplace or court-ordered drug testing. For full exclusion criteria refer to the trial registration or Chapter 3b.

Trial Procedures

All participants were informed about the nature and risk of experimental procedures by a sleep specialist and the trial coordinator before their written informed consent was obtained. Initial eligibility was ascertained during a clinical interview at the screening visit which included a urinary drug test (DrugCheck® NxStep Onsite Urine Drug Test) to verify abstinence from alcohol, cannabis and illicit drugs, and a pregnancy test (as applicable; Human Chorionic Gonadotrophin Cassette, Alere™) to rule out pregnancy. Participants then completed two 24-hour overnight treatment sessions during which they were administered CBD/THC or placebo in random order with each treatment session separated by a minimum ≥7 days washout period. Participants were instructed to abstain from caffeine and alcohol (≥24 h) prior to each treatment session and to avoid using illicit drugs (including cannabis) and all CNS-active medications including hypnotics for the duration of the trial. One week prior to each treatment session, participants were also encouraged to maintain regular sleep- and wake-onset times as best as possible.

Participants arrived at the research clinic at ~16:30 before a urinary drug test (DrugCheck® NxStep Onsite Urine Drug Test) and pregnancy test (as applicable; Human Chorionic Gonadotrophin Cassette, Alere™) was completed. Approximately 1 hour prior to the participant's habitual bedtime (as determined using a 7-day sleep diary), participants were administered a fixed 2 mL dose of either placebo or active treatment). Participants were allowed to sleep undisturbed in the research clinic for 8 hours before a sleep technician gently woke them. Participants were administered standardised meals (evening prior: 18:30 dinner; next day: ~07:00 breakfast and ~12:45 lunch) and light snacks (e.g., popcorn, fruit). The next day, participants

completed a range of assessments starting from ~07:30 (i.e., approximately 10 h post-drug administration). The trial procedures are summarised in **Figure 1.**

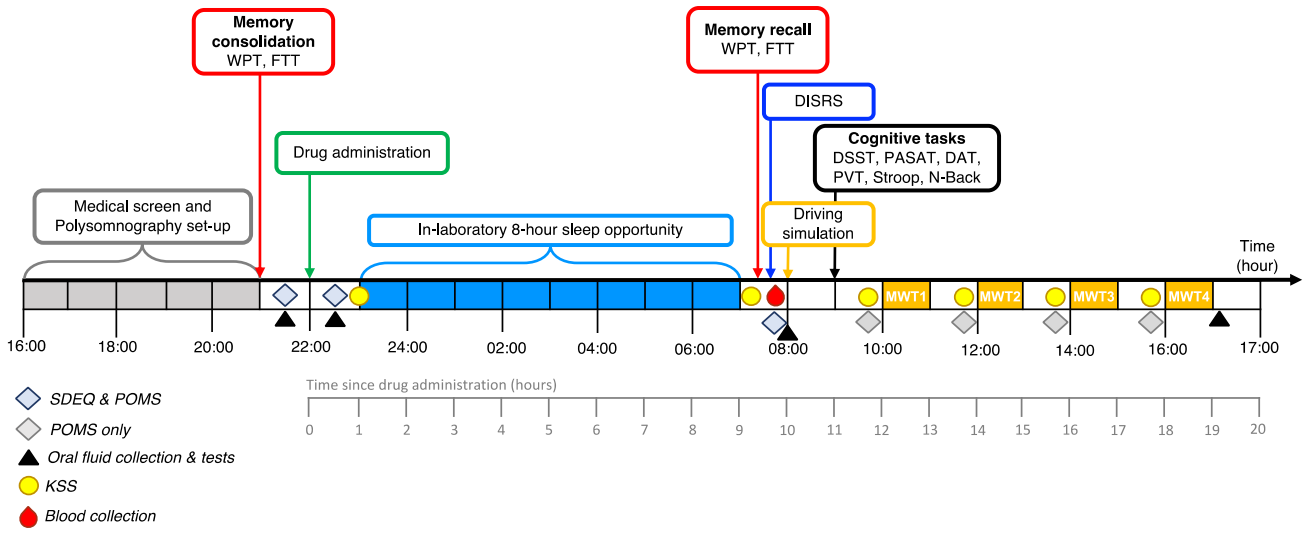

Figure 1 *Study procedures and timeline. DAT Divided Attention Task, DISRS Daytime Insomnia Symptom Response Scale, DSST Digit Symbol Substitution Task, FTT Finger Tapping Task, KSS Karolinksa Sleepiness Scale, MWT Maintenance of Wakefulness Test, POMS Profile of Mood States, PSAT Paced Serial Addition Task; PVT Psychomotor Vigilance Task; WPT Word Pairs Task.*

Investigational Product

The investigational product was a plant-derived oral formulation containing a 20:1 ratio of CBD to THC i.e., 100 mg/mL CBD and 5 mg/mL THC in medium-chain triglyceride (MCT) oil (hereafter referred to as 'CBD/THC'); manufactured at a GMP-certified facility (Linnea SA, Lavertezzo, Switzerland). Neither the placebo nor active treatment contained any other cannabinoids or cannabis constituents (e.g., minor phytocannabinoids, flavonoids, mono- or sesquiterpenes). The matched placebo consisted of MCT oil (only). The active and placebo treatments did not differ in their visual appearance. Participants were instructed to ingest one peppermint lozenge (Fisherman's Friend Mint; Lofthouse of Fleetwood, England) to mask any possible differences in taste/smell.

Randomisation

Each participant was randomly allocated to one of two treatment sequences: (1) CBD/THC–placebo, or (2) placebo–CBD/THC. The sequences were computer-generated using a simple 1:1 randomisation prepared by the trial epidemiologist (NSM) and sequentially numbered using identical containers according to the randomisation sequence were prepared by the drug distributor; neither the statistician nor the distributor had any contact with any prospective or enrolled participants. The sequence was held in a central location and only accessible to the trial epidemiologist, drug distributer, and the principal investigator (in the event of a serious adverse event). All participants, trial personnel (including study doctors), and the outcome assessors were blind to the treatment allocation.

Next-day Outcome Measures

Subjective Drug Effects Questionnaire (SDEQ)

Subjective drug effects were assessed at baseline, 0.5 h post-drug administration and the next morning at approximately 08:00 (10 h post-drug administration). Measurements stopped after the 08:00 timepoint because subjective drug effects following a single, oral dose of THC were not expected to persist beyond this time. The SDEQ requires participants to rate how 'Stoned, 'Sedated, 'Alert', 'Anxious' and 'Sleepy' they feel using a series of visual analogue scales (VAS), where zero represents "not at all" and 100 represents "extremely".

Daytime Insomnia Symptom Response Scale (DISRS)

Daytime sleep-related rumination was assessed the morning after drug administration at approximately 07:30 using the DISRS[32]. The DISRS is a self-rated 20-item questionnaire in which patients are asked how frequently they engage in certain behaviours when feeling tired (e.g., 'Think: 'I won't be able to do work because I feel so bad').

Maintenance of Wakefulness Test (MWT)

The MWT is a validated, objective measure of an individual's ability to stay awake during a defined period (i.e., a measure of daytime drowsiness). 40 min trials was administered at 10:00, 12:00, 14:00 and 16:00 hours starting on the morning and afternoon post-drug administration; in line with American Academy of Sleep Medicine (AASM) recommended protocols.[33] Participants were instructed to lay semi-recumbent on a bed (above the covers) in a room with low levels of stimulation and try to remain awake for the entire 40 min period. An experienced sleep technician recorded polysomnography throughout. Trials ended after 40 min if no sleep occurred, or after unequivocal sleep, defined as three consecutive epochs of non-rapid eye movement stage 1 (N1) sleep or one epoch of any other sleep stage (N2, N3, N4 or REM). The main outcome measure was mean sleep latency of the four MWT trials.

Profile of Mood States (POMS)

Mood was assessed at seven timepoints: baseline, 0.5 h post drug-administration, upon waking (10 h post-drug administration), and prior to the start of each MWT at 10:00, 12:00, 14:00, and 16:00 using the 40-item POMS abbreviated version[34]. The abbreviated POMS has seven subscales: 'tension', 'anger', 'fatigue', 'depression', 'esteem-related affect', 'vigour', and 'confusion'. *Total mood disturbance* was calculated by summing the negative subscales and subtracting the positive subscales (i.e., vigour and esteem-related affect). A constant (i.e., 100) was added to the TMD formula to eliminate negative scores.

Karolinska Sleepiness Scale (KSS)

The KSS was used to assess subjective sleepiness and administered at six timepoints: immediately prior to lights off (~1 h post-drug administration), upon waking at 07:30 (9 h post-drug administration), and prior to the start of each MWT at 10:00, 12:00, 14:00, and 16:00. Participants self-rated their level of sleepiness/alertness in the past half an hour using a 10-point

scale where '1' represented "extremely alert" and '10' represented "extremely sleepy, can't keep awake".

Driving Simulation Task

Participants completed a 30 min simulated driving task at approximately 08:00 (10 h post-drug administration) using a fixed-base driving simulator (Hyperdrive, Adelaide, Australia) equipped with standard vehicle controls (steering wheel, indicators, seat, safety belt), hi-resolution Fanatec pedals, and a servo motor wheelbase (Endor AG, Landshut, Germany) and linked to four networked computers running the SCANeR Studio simulation engine software (V.1.6, AVSimulation, Paris, France). The driving scenario was custom-built and identical to that previously employed in a study examining the effects of vaporised cannabis on driving performance in healthy volunteers.[35] The outcome measures include standard deviation of lateral position (SDLP), average headway and standard deviation of headway (i.e., distance to the lead vehicle), average speed and standard deviation of speed (measures of longitudinal vehicle control).

Neurocognitive Test Battery

Unless otherwise stated, all neurocognitive tests were administered between 11-12 h post-drug administration the next morning.

Word Pairs Task

The Word Pairs task (WPT) measures sleep-dependent declarative memory (procedural memory task) consolidation in adults. The encoding phase was administered prior to drug administration at approximately 21:00 (1 h prior to drug administration) on the night of each treatment session. Participants are presented with 32 pairs, one at a time for 5 seconds each, and asked to memorize the pairs. Easy (i.e., semantically related, e.g., fork-knife) and difficult (semantically unrelated, e.g., syrup-feet) word pairs were randomly interspersed, and the order of

presentation was randomized for each participant at the first treatment session. The recall (re-test) and recognition phase were administered the following morning at approximately 07:30 (9.5 h post-drug administration). The outcome measured was the percentage of evening scores correctly recalled in the morning.

Finger Tapping Task (FTT)

The FTT is a psychomotor sequence learning task that assesses procedural memory in which participants are asked to tap a 5-digits sequence (for example 4-1-3-2-4) with their non-dominant hand as rapidly and accurately as possible using the numeric key-buttons of a computer keyboard.[36] To reduce working memory load, the numeric sequence is displayed at the centre of the screen throughout the task. During a training session completed at approximately 21:30 (0.5 h prior to drug administration), participants completed 12 blocks, each consisting of a 30 sec task followed by 30 sec rest. The next morning (at approximately 07:30 or 9.5 h post-drug administration), participants completed a recall session composed of 6 blocks, each consisting of a 30 sec task followed by 30 sec rest. The outcome measures included: (a) pre-training learning (number of correct sequences averaged across the first three trials prior to sleep), (b) post-training learning (number of correct sequences averaged across the last three trials prior to sleep); (c) early retest learning (number of correct sequences averaged across the last three trials following sleep); (d) late retest learning (number of correct sequences averaged across the last three trials following sleep), (e) overnight early improvement (the percentage overnight improvement in motor skill defined as the early retest learning score/post-training learning score x100), also termed offline memory consolidation; (f) overnight late improvement (the percentage overnight improvement in motor skill defined as the late retest learning score/post-training learning score x100), as previously defined.[37]

Psychomotor Vigilance Task (PVT)

The PVT is a 10 min simple reaction-time task of sustained attention that is sensitive to sleep loss.[38] The device is a hand-held box with a red light-emitting diode display of a three-digit millisecond counter (PVT-192, Ambulatory Monitoring, Inc., Ardsley, NY, USA). Visual stimuli appeared at random intervals between 2 to 10 s. Participants were instructed to press the response button as quickly as possible each time the stimulus appears. The time taken to respond to the stimulus was displayed in milliseconds (ms). Variables analysed were: (a) mean reaction time (RT); (b) number of lapses (response time >500 ms).

Stroop Test

The Stroop test assesses the inhibition of dominant responses and reflects the "higher-order" executive functions.[39] It assesses reaction time to colours (Stroop-Colour) and words (Stroop-Word) displayed and cognitive interference due to presentation of simultaneous conflicting information. Words (red, green, or blue) and three different coloured squares (red, green, or blue) were displayed on the computer screen. Participants were required to click on the coloured square that matched either the colour (Stroop-Colour) or the meaning (Stroop-Word) of the word presented. Each part of the test was 45 s in duration and involved multiple trials. The outcome measures included the percentage of correct responses and the average response latency.

N-Back Task

The N-Back assesses working memory, encompassing short-term memory storage and information processing. For this visuospatial test, the 1-Back and 2-Back were used. The participant was asked to compare the position of a letter displayed on the screen to the position of the letter presented two or three trials previously. For example, for 2-back, the position of the 3rd letter is compared to the position of the 1st letter and the position of the 4th letter to the 2nd letter, and so on. If the position of the letters matched, the participant pressed "M" on the

keyboard for "Match" as quickly as possible. If the position of the letters did not match, the participant pressed "N" for "No Match" as quickly as possible. Each N-back task was 4 min in duration and consisted of 50 trials with a stimulus presented every 4.5 sec. Percentage accuracy was calculated for both tasks.

Digit Symbol Substitution Task (DSST)

The DSST measures a range of cognitive skills including speed, attention, working memory, and visuospatial function[40] and has demonstrated sensitivity to the impairing effects of THC.[41] Participants were presented with a series of geometric patterns labeled from 1 to 9, each consisting of an array of filled and blank squares in a 3 x 3 grid. When a number appeared in the middle of the screen, participants were instructed to replicate the pattern corresponding to that array using the numeric keypad of a computer keyboard. Participants had 90 sec to replicate as many patterns as possible. The outcome measures included the number of patterns correct and accuracy (number of patterns correct/number of patterns attempted).

Divided Attention Task (DAT)

The DAT assesses working memory and ability to allocate attention to different aspects of a task.[42] Participants were required to track a horizontally moving stimulus on the screen using their mouse while simultaneously responding to visual stimuli in the periphery by clicking the left mouse button whenever a number in any corner of the screen matched a target number presented at the bottom of the screen. The outcome measures included the mean distance of the cursor from the target in pixels (tracking error), the number of target numbers correctly identified (/24), and average response time (msec).

Paced Serial Addition Task (PSAT)

The PSAT measures working memory, attention, and simple arithmetic problem-solving.[43] Participants observed single digits appear on the screen and were instructed to summate each new digit with the preceding one. Participants responded by clicking on the correct answer from a list of numbers (1–10) presented on the screen. The outcome measures included average response time on correct trials and the total number of correct trials (/90).

Salivary Drug Tests

Oral fluid samples were collected at baseline and at 0.5 h, 10 h, and 18 h post drug administration using Quantisal™ collection devices (Immunalysis, Pomona, California, USA). Devices were placed under the tongue until indicators turned blue, or for a maximum of 10 min, before being placed into the stabilising buffer. Samples were kept at +4°C for a maximum of 30 days prior to analysis using liquid chromatography-tandem mass spectrometry (LC-MS/MS) (full methods in Appendix E). Oral fluid tests were also performed at the same timepoints using two devices: DrugWipe 5s (DW-5S; Securetec, Neubiberg, Germany), Dräger Drug Test 5000 (DT5000; Drägerwerk AG & Co., Lübeck, Germany). Both devices had a manufacturer-specified detection limit of 10 ng/mL THC.

The DW5s device has two small sampling pads which collect oral fluid from the tongue (about 10–20 μL). Participants were instructed to run their tongue around the inside of their mouth in a circular motion three times before slowly gliding the sampling pads down their tongue. Once sufficient volume is collected as indicated by a change in colour of the sampling pads, the researcher places the sample collector back on the test cassette and breaks the ampoule containing liquid. The test is held vertically for 10 seconds before being laid horizontally and results are visible within 10 minutes. A positive test is indicated by the appearance of a red line. Test results where the DW5s red 'positive' line was considered too ambiguous were excluded.

The DT5000 test consists of a test cassette (with a sampling pad) and an analytical instrument. Participants were instructed to wipe the sampling pad around the inside of their cheeks and across their gums until sufficient oral fluid had been collected (indicated by the appearance of a blue line or after 4 minutes). The test cassette was then inserted into the analysing instrument. The results (negative, non-negative, or invalid) can be printed using an attached printer. Test results for both devices were read and filed by an independent observer who had no direct contact with the participant. The results were only made available to the researchers upon completion of the study.

Results of the DW-5S and DT5000 drug tests were classified as previously described[30]: (a) a *true positive* (TP) was a positive test result that was subsequently confirmed by LC–MS/MS (i.e., confirmed value on LC-MS/MS ≥ confirmatory cut-off AND positive result obtained); (b) a *true negative* was a negative test result which was confirmed by LC–MS/MS (i.e., confirmed value ≤ confirmatory cut-off AND negative result obtained); (c) a *false positive* was a positive test result which was not confirmed by LC–MS/MS (i.e., confirmed value ≤ confirmatory cut-off AND positive result obtained); and (d) a *false negative* was a negative test result that was not confirmed by LC–MS/MS (i.e., confirmed value ≥ confirmatory cut-off AND negative result obtained). Analytical methods for LC-MS/MS plasma cannabinoid analysis available in Appendix E. Based on these classifications, sensitivity [TP/(TP + FN)], specificity [TN/TN + FP)], and accuracy [(TP + TN)/(TP + TN + FP+ FN)] were calculated at a confirmatory cut-off of 10 ng/mL THC (equivalent to the screening cut-off for both devices). Given that the cut-offs used for confirmatory analysis is typically lower than the screening cut-off in practice, these parameters were also calculated relative to THC cut-offs of 2 ng/mL (THC LOQ) and 1 ng/mL (THC LOD).

Statistical Analysis

All data were analysed using SPSS version 26 (IMB Corp., Armonk, NY). Figures were created using GraphPad Prism version 9 (GraphPad Inc., San Diego, CA). Linear mixed-model

analyses were used to determine differences between treatments. Fixed factors included treatment (2 levels), time (3, 6, and 7 levels for subjective drug effects, KSS, and POMS, respectively), order (2 levels), and the treatment by time interaction, and the participant as a random effect. The least-squares means procedure was used in the mixed-model analyses to handle missing data. If a significant main effect of treatment or a significant treatment × time interaction was observed, two-sided pairwise comparisons compared means across conditions at each level of time. The statistical significance level was set at $p < 0.05$.

4.3.4 Results

Participants

Twenty participants with insomnia disorder (16 female; median [IQR] age, 47 [13.8] years) were recruited and randomised between August 2019 and October 2021 (**Table 1**). All 20 randomised participants completed the trial. Insomnia symptoms were of moderate severity (20.8 ± 2.5), with no evidence of sleep apnea (AHI 1.6 ± 1.7 events/h) and an average body mass index (BMI) of 25.1 ± 3.7 kg.m^2. None of the 20 participants reported regular use of any CNS-active medications, and all participants provided negative urinary drug screens (including THC) on the afternoon of each treatment session.

Table 1 Participant demographics and characteristics

Characteristic	
Number of participants	20
Sex (M / F)	4 / 16
Age, *years*	46.1 (8.6)
BMI, kg/m^2	25.1 (3.7)
Participants with at least some tertiary education	18 (90%)
Participants with current employment	15 (75%)
Weekly standard drinks, IQR (SD)	1.6 (1.7)
Lifetime cannabis exposure, *n* (%)	
Never tried	4 (20%)
≤10 uses	11 (55%)
>10 uses	5 (25%)

Data are shown as mean (SD) or as frequency. BMI Body mass index

Subjective Drug Effects

VAS ratings of stoned, sedated, alert and sleepy did not indicate effect of Treatment or a Treatment x Time interaction, however, there was a main effect of Time with subjective ratings for *Sedated* were significantly higher with CBD/THC than placebo at 10 h post drug administration only (i.e., the next morning) (8.57 [95% CI, 0.56 to 16.73]; p=0.036, d=0.349) (**Figure 2**). No other significant effects were observed at any time point.

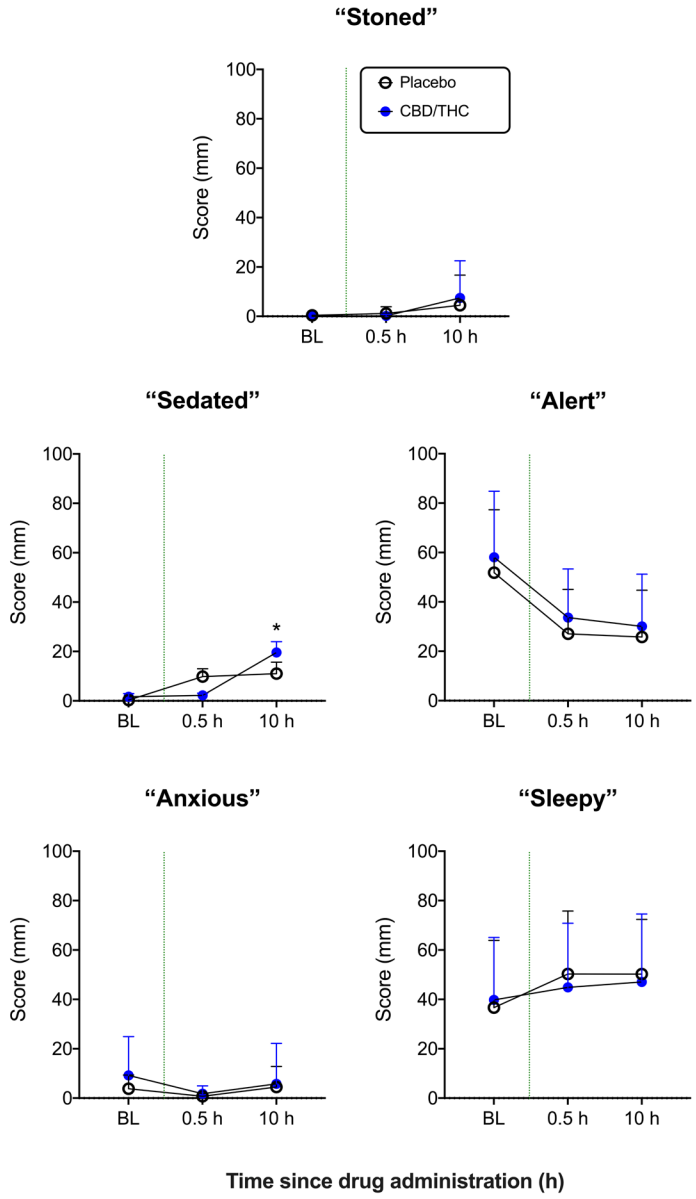

Figure 2 *Mean (SEM) participant ratings of "Stoned", "Sedated", "Alert", "Anxious", and "Sleepy" assessed using 0-100 mm visual analog scales after oral consumption of CBD/THC and placebo. Green dotted line indicates time of drug administration (i.e., 1 h before participant's habitual bedtime). Time as shown on the x-axis indicates time elapse since drug administration (h). *p<0.05. BL baseline (~0.5 h prior to drug administration).*

Mood and Daytime Sleep-Related Rumination

There was no significant main effect of Treatment or a Treatment x Time interaction on the total mood disturbance (TMD) score of the POMS (**Figure S1** in Appendix E). There was also no significant difference in daytime sleep-related rumination as measured on the DISRS between treatments (mean difference 0.85 [95% CI, -4.02 to 2.32]; p=0.581, d=0.127).

Next-day Subjective Sleepiness and Objective Alertness

There was a main effect of Treatment on self-ratings on the KSS with a small, albeit significant, increase with CBD/THC relative to placebo (mean difference 0.42 [95%CI 0.07 to 0.77]; p=0.01, d=0.219) (**Figure 3**). No other significant differences were observed. There was no effect of Treatment on the average latency to sleep (minutes) on the MWT (mean difference 1.98 [95%CI -6.15–2.19], p=0.331, d=0.227) (**Figure 4**).

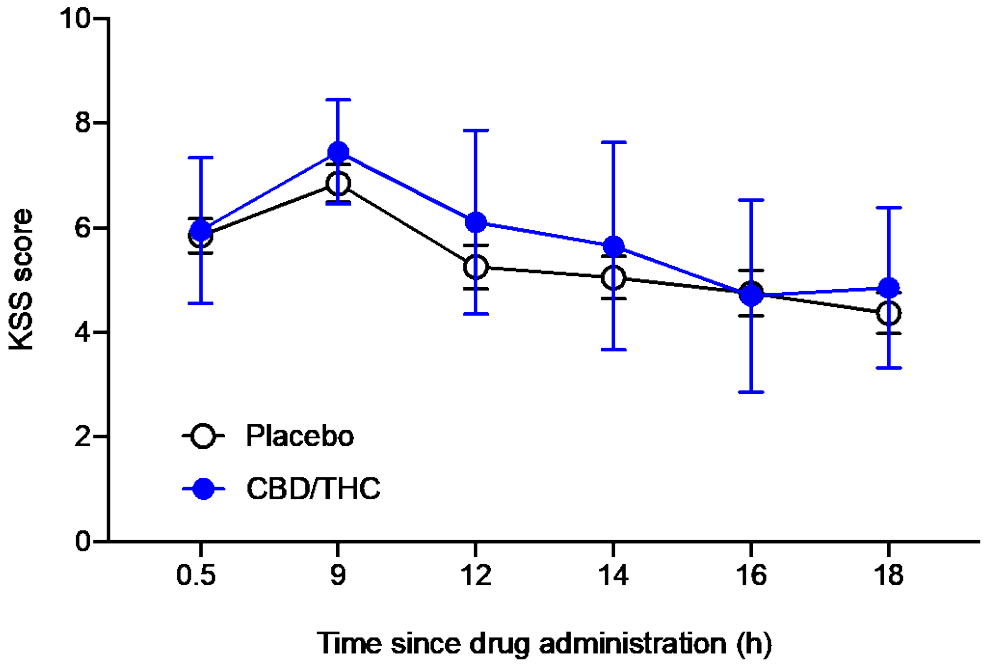

Figure 3 *Participants self-rated level of sleepiness/alertness on the Karolinska Sleepiness Scale (KSS) after oral consumption of CBD/THC and placebo as assessed at 0.5 h (prior to sleep), 9 h (upon waking) and immediately prior to the start of each MWT at approximately 12 h, 14 h, 16, and 18 h post-drug administration.*

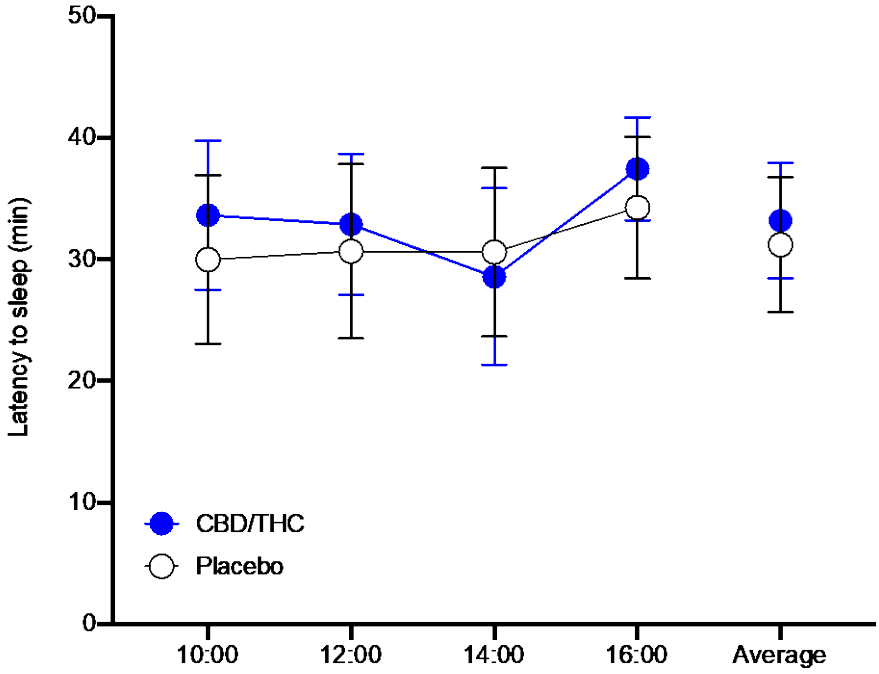

Figure 4 *Meant latency to sleep (minutes) across all four trials of the Maintenance of Wakefulness Test (10:00, 12:00, 14:00, 16:00) and the average sleep latency of all four trial the next-day post-treatment with CBD/THC and placebo.*

Cognitive Performance

Table 2 presents the mean (SD) values of cognitive task performance measures. There was a significant reduction in percentage accuracy on the Stoop-Colour test with CBD/THC compared to placebo (mean difference 1.4% [95%CI 95.9–99.8], p=0.016, d=-0.602). No other significant differences were observed for any of the other cognitive tasks (all p's>0.05).

Table 2 Results [means (SD)] of the cognitive assessment the morning after evening administration with CBD/THC and placebo (*n*=20)

	Placebo	CBD/THC	*p* value	Cohen's *d* [95% CI]
DSST				
Number correct	29.3 (10.5)	29.5 (7.1)	0.901	-0.025 [-0.463, 0.414]
Response accuracy (%)	91.8 (8.9)	90.7 (8.7)	0.723	-0.079 [-0.518, 0.360]
DAT				
Tracking errors, pixels	30.0 (11.0)	34.2 (25.1)	0.447	0.169 [-0.273, 0.610]
Response time, msec	1288.5 (316.5)	1213.6 (268.1)	0.317	-0.228 [-0.683, 0.228]
Number correct	21.6 (2.3)	21.9 (2.1)	0.597	0.129 [-0.322, 0.581]
PSAT				
Number correct	43.6 (12.2)	42.0 (16.1)	0.550	-0.135 [-0.575, 0.305]
Response time, msec	1632.1 (126.6)	1653.0 (156.2)	0.477	0.163 [-0.278, 0.604]
WPT				
Retention (%)	90.4 (9.9)	92.3 (9.9)	0.377	0.162 [-0.279, 0.603]
FTT				
Pre-training learning (PreTLS)	18.2 (1.1)	18.1 (1.1)	0.839	-0.112 [-0.552, 0.328]
Post-training learning (PoTLS)	21.9 (4.0)	22.2 (4.0)	0.267	0.131 [-0.309, 0.571]
Early retest learning (ERLS)	20.0 (7.2)	20.4 (5.9)	0.739	0.074 [-0.365, 0.513]
Late retest learning (LRLS)	31.7 (18.4)	33.4 (17.4)	0.476	0.153 [-0.288, 0.594]
Overnight early improvement (OEI)	100.9 (15.8)	91.4 (19.3)	0.210	-0.253 [-0.698, 0.193]
Overnight late improvement (OLI)	152.2 (66.7)	150.0 (70.1)	0.695	-0.050 [-0.488, 0.389]
PVT				
Mean RT, msec	300.4 (70.0)	305.4 (65.1)	0.567	0.155 [-0.298, 0.607]
Lapses, n (%)	3.5 (6.5)	4.3 (8.9)	0.283	0.263 [-0.194, 0.721]
Stroop Test				
Colour accuracy (%)	**99.2 (1.8)**	**97.8 (2.3)**	**0.016**	**-0.602 [-1.079, -0.126]**
Colour RT, s	1.3 (0.3)	1.4 (0.3)	0.288	0.250 [-0.195, 0.695]
Word accuracy (%)	99.8 (0.8)	93.6 (20)	0.182	-0.309 [-0.758, 0.140]
Word RT, s	1.4 (0.2)	1.4 (0.3)	0.967	0.010 [-0.429, 0.448]
N-back				
1-Back accuracy (%)	87.6 (9.7)	86.5 (10.9)	0.472	-0.136 [-0.576, 0.305]
2-Back accuracy (%)	89.4 (7.8)	89.5 (7.8)	0.849	0.017 [-0.422, 0.455]

Simulated Driving Performance

Mean (SD) values of the simulated driving outcome measures are presented in **Table 3**. CBD/THC did not significantly affect vehicular control parameters including SDLP, mean headway (i.e., distance to the lead vehicle) and speed on the car-following and standard component of the simulated driving task (all *p*'s>0.05).

Table 3 Measures of next day simulated driving performance

	Placebo	CBD/THC	*p* value	Cohen's *d* [95% CI]
Car Following Component				
SDLP (cm)	21.6 (4.7)	22.5 (5.4)	0.358	0.219 [-0.225, 0.662]
Headway (m)	127.2 (103.6)	130.5 (80.6)	0.907	0.027 [-0.411, 0.465]
SD Headway (m)	34.6 (30.7)	34.8 (28.6)	0.982	0.005 [-0.433, 0.444]
Standard Component[a]				
SDLP (cm)	32.2 (4.9)	33.0 (4.8)	0.486	0.171 [-0.294, 0.637]
Speed ($km \cdot h^1$)	97.4 (3.5)	97.2 (4.4)	0.774	-0.070 [-0.532, 0.393]
SD Speed ($km \cdot h^1$)	13.1 (2.7)	13.8 (4.0)	0.392	0.210 [-0.257, 0.677]

Values are Mean ± SD. *SDLP* Standard Deviation of Lateral Position, *SD* Standard Deviation.
This task was completed ~12 hours post-drug administration. [a] Sample size was n=18 as two participants failed to complete the Standard Component on each occasion due to motion sickness.

Salivary Drug Tests

Table 4 presents the test results (TP, TN, FP, FN) for the DW5s and DT5000 and overall device performance (sensitivity, specificity, and accuracy) at a 10 ng/mL confirmatory cut-off. A total of 160 DW5s were performed with one positive test result at baseline (on placebo) and three positive test results at +0.5 h post-drug administration. Only 136 tests were evaluated against LC-MS/MS-verified oral fluid THC concentrations due to technical difficulties with the analysis. With a 10 ng/mL confirmatory cut-off applied, overall sensitivity, specificity, and accuracy were calculated as 10%, 98%, and 91%, respectively. Of the four test results that were positive, three false positives were detected with corresponding oral fluid THC concentrations ranging from 0 to 2.72 ng/mL. Of the 132 tests that were negative, eight false negatives were detected with

corresponding oral fluid THC concentrations ranging from 13.0 to 425.2 ng/mL. The occurrence of both false positives and false negatives was greatest at the +0.5 h timepoint. Fewer false positives and more false negatives were observed with confirmatory cut-offs of 2 ng/mL and 1 ng/mL (see **Table S1** in Appendix E).

A total of 152 DT5000 test were performed with four positive test results at 0.5 h post-treatment and one positive test at 10 h post-treatment i.e., the morning after drug administration (all in the CBD/THC group). Eight tests could not be completed due to technical difficulties (DT5000 device temporarily broke) and one test produced an invalid result. Of these, only 127 tests were evaluated against LC-MS/MS-verified oral fluid THC concentrations due to technical difficulties with the analysis. With a 10 ng/mL confirmatory cut-off applied, overall sensitivity, specificity, and accuracy were calculated as 38%, 100%, and 96%, respectively. No false positives were identified. Of the 121 tests that were negative, five false negatives were detected with corresponding oral fluid THC concentrations ranging from 11.5 to 44.5 ng/mL. The occurrence of false negatives was greatest at the +0.5 h timepoint. Increasing the confirmatory cut-off to 2 ng/mL and 1 ng/mL had no effect on the number of false positives but substantially increased the number of false negatives (see **Table S1** in Appendix E). Overall accuracy was greatest with a 10 ng/mL confirmatory cut-off for both devices.

Table 4 Performance characteristics of the Securetec DrugWipe® 5 s (DW5s) and Dräger DrugTest® 5000 (DT5000) POCT devices when verified against LC–MS/MS quantified oral fluid THC concentrations using a 10 ng/mL confirmatory cut-off

Device	Time relative to drug administration (min)	*N* of tests	True positives	True negatives	False positives	False negatives	Sensitivity (%)	Specificity (%)	Accuracy (%)
DW5s	- 0.5 h	34	0	33	1	0	-*	97	97
	+ 0.5 h	34	1	23	2	8	11	92	71
	+ 10 h	34	0	33	0	1	-*	100	97
	+ 18 h	34	0	34	0	0	-*	100	100
	Total	136	1	123	3	9	10	98	91
DT5000	- 0.5 h	32	0	32	0	0	-*	100	100
	+ 0.5 h	31	3	24	0	4	43	100	87
	+ 10 h	31	0	30	0	1	-*	100	97
	+ 18 h	32	0	32	0	0	-*	100	100
	Total	127	3	118	0	5	38	100	96

-* Sensitivity could not be ascertained as there were no true positives.

4.3.5 Discussion

This is the first study to explore the 'next day' effects of cannabis use in a clinical population; namely, individuals with chronic insomnia disorder. With the exception of a possible (subtle) increase in subjective measures of drowsiness, no reliable changes in 'next day' function including cognitive function, driving performance, and (objective) alertness were observed after a single, oral dose of 200 mg CBD and 10 mg THC. We also evaluated the performance of the DW5S and DT5000 oral fluid testing devices by comparing observed test results against confirmatory LC-MS/MS quantified oral fluid THC and CBD concentrations. Both POCT devices performed relatively poorly soon after drug administration (~30 minutes), yielding the highest number of false positive and false negative tests, but performed better the next day.

Cannabis and THC are known to increase subjective feelings of 'drowsy' and 'sleepy/tired' after oral and smoked/vaporised ingestion.[44] [45] We, likewise, found that a single dose of CBD/THC increased subjective feelings of "sedated" the next morning (i.e., 10 h post drug-administration) and increased drowsiness by 0.42 points on the KSS relative to placebo. However, both effects were small and the increase in drowsiness on the KSS is not considered to be clinically meaningful (i.e., monotonous tasks such as driving a train through long stretches of homogenous forest will increase KSS values by 1-2 units).[46] [47] Further, there was no evidence of impairment on the MWT, a validated *objective* measure of daytime drowsiness. This suggests that although there was a mild increase in 'next day' subjective drowsiness, this did not translate into significantly poorer performance on an objective test of alertness.

The vast majority of neurocognitive tests, spanning attention, working memory, speed of information processing, showed no 'next day' effects of CBD/THC. The one exception was the *Stoop-Colour Test* (i.e., the 'easy/congruent condition' where the participant must match the *colour* of the word presented) where a 1.4% reduction in percentage response accuracy was observed. However, a ceiling effect was evident with participants demonstrating a high degree of accuracy (i.e., >97% accuracy) on both treatments. In addition, no significant difference in accuracy was

observed on the more difficult *Stroop-Word Test* (i.e., the 'hard/incongruent condition' where the participant must match the *meaning* of the word presented, not the ink colour); a measure of executive function and ability to inhibit cognitive interference.[39] Further, there was no effect on the more ecologically valid driving simulator task. This suggests that the effect could have occurred by chance (given the large number of assessments performed).

The lack of substantial impairment to neurocognitive function and driving performance is in line with a recent systematic review, which concluded that current published data does not support the assertion that cannabis impairs 'next day' performance (i.e., >8-hours after use) on safety-sensitive tasks and/or discrete neuropsychological tests.[22] This is particularly salient given that the present study observed an acute worsening effect of treatment on objective sleep outcomes (i.e., reduction in total sleep time and rapid eye movement (REM) sleep), with no improvement in subjective sleep quality as outlined in **Chapter 4b**. This suggests that despite a reduction in total sleep time, there were no meaningful deterioration in 'next day' function with CBD/THC treatment in individuals with insomnia disorder.

In line with previous literature, oral fluid cannabinoid concentrations were maximal at the time point closest to consumption (0.5 h post-drug administration) and declining rapidly thereafter.[30] The occurrence of false positive and false negative tests on both POCT devices was greatest at 0.5 h post-drug administration with no true positive THC results observed at the 10 h and 18 h post-drug administration (where the levels of THC in oral fluid were well below the 10 ng/mL screening cut-off on both devices). These data show that there is a low chance of testing positive to THC the next day after oral administration of an oil containing CBD/THC on two POCT devices. It is understood that THC in oral fluid originates exclusively from contamination of the oral cavity upon ingestion, with no circulation back into saliva from the blood.[45 48] This is consistent with previous studies showing no detectable THC in oral fluid following oral administration of encapsulated THC (dronabinol).[49] Of note, a recent study has shown that CBD (isolate) does not appear to cross-interact with THC on POCT testing devices and does therefore

not pose a risk for consumers using CBD-only products.[50] Future studies should explore whether repeated dosing with (oral) cannabinoids yield different results on POCT devices.

There are several limitations to the study. The study design was unable to assess the individual contribution of THC and CBD to observed effects. There is emerging evidence that co-administration of THC and CBD may produce pharmacokinetic and pharmacodynamic interactions, however, findings are mixed.[51] One study showed that when administered concurrently, CBD (15 mg, oral) counteracted the sedative effects of THC (oral, 15 mg) in eight healthy volunteers.[52] Future studies should examine THC and CBD in isolation on objective sleep outcomes in people with insomnia disorder. Further, this study also examined acute effects only precluding any conclusions made regarding the effects of repeated dosing with cannabinoids on daytime function in insomnia disorder.

Conclusions

The 'next day' effects of cannabis use are increasingly scrutinised and are considered a risk factor for daytime impairment including to driving performance. The results of this study suggest that acute, oral treatment with combined 200 mg CBD and 10 mg THC does not substantially impair 'next day' cognitive function, alertness or driving performance in individuals with insomnia disorder. POCT devices were limited in their ability to detect THC in oral fluid the morning after drug administration. Future research is required to determine the impact of repeated oral dosing of cannabinoids on 'next day' function in insomnia disorder.

Funding Statement

This study is funded by the Lambert Initiative for Cannabinoid Therapeutics, a philanthropically funded centre for cannabinoid research at the University of Sydney. Lambert Initiative research staff were co-investigators and played an active role in the trial design and conduct, data analysis, and reporting. AS was supported by the Australian Government Research Training Program (RTP) and the Vice-Chancellor's Research Scholarship (VCRS) at the University of Sydney. CMH (APP1104003) and ALD (APP1107716) were supported by Dementia Research Development Fellowships of the Australian National Health and Medical Research Council-Australian Research Council (NHMRC-ARC). RRG was supported by an NHMRC Senior Principal Research Fellowship (APP1106974). The investigational product was purchased from BOD Australia who were not involved in the conception or design of this study, data analysis (with no access to the data) or the decision to publish. All other commercially available equipment was purchased.

4.3.6 References

1. Aernout E, Benradia I, Hazo J-B, et al. International study of the prevalence and factors associated with insomnia in the general population. *Sleep Med* 2021;82:186-92.
2. The Diagnostic and Statistical Manual of Mental Disorders (5th ed.; DSM–5; American Psychiatric Association, 2013).
3. Morin CM, LeBlanc M, Daley M, et al. Epidemiology of insomnia: prevalence, self-help treatments, consultations, and determinants of help-seeking behaviors. *Sleep Med* 2006;7(2):123-30.
4. De Crescenzo F, D'Alò GL, Ostinelli EG, et al. Comparative effects of pharmacological interventions for the acute and long-term management of insomnia disorder in adults: a systematic review and network meta-analysis. *Lancet* 2022;400(10347):170-84.
5. van der Zweerde T, Bisdounis L, Kyle SD, et al. Cognitive behavioral therapy for insomnia: a meta-analysis of long-term effects in controlled studies. *Sleep Med Rev* 2019;48:101208.
6. Koffel E, Bramoweth AD, Ulmer CS. Increasing access to and utilization of cognitive behavioral therapy for insomnia (CBT-I): a narrative review. *J Gen Intern Med* 2018;33(6):955-62.
7. Lintzeris N, Driels J, Elias N, et al. Medicinal cannabis in Australia, 2016: the cannabis as medicine survey (CAMS-16). *Med J Aust* 2018;209(5):211-16.
8. Lintzeris N, Mills L, Abelev SV, et al. Medical cannabis use in Australia: consumer experiences from the online cannabis as medicine survey 2020 (CAMS-20). *Harm Reduct J* 2022;19(1):1-10.
9. Lintzeris N, Mills L, Suraev A, et al. Medical cannabis use in the Australian community following introduction of legal access: the 2018–2019 Online Cross-Sectional Cannabis as Medicine Survey (CAMS-18). *Harm Reduct J* 2020;17(1):1-12.
10. Cahill SP, Lunn SE, Diaz P, et al. Evaluation of patient reported safety and efficacy of cannabis from a survey of medical cannabis patients in Canada. *Front Public Health* 2021;9:626853.
11. Therapeutic Goods Administration. Freedom of Information Disclosure Log. FOI 3732. Australian Government. Accessed 19 September, 2022. https://www.tga.gov.au/foi-disclosure-log-0

12. Suraev AS, Marshall NS, Vandrey R, et al. Cannabinoid therapies in the management of sleep disorders: a systematic review of preclinical and clinical studies. *Sleep Med Rev* 2020;53:101339.
13. Australian New Zealand Clinical Trials Registry. A phase IIb double-blind, randomised placebo-controlled clinical trial to evaluate the efficacy and safety of a botanical cannabidiol (CBD) for sleep disturbances in a healthy population. ACTRN12621000632897. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12621000632897
14. Australian New Zealand Clinical Trials Registry. A trial to evaluate an oral medicinal cannabis extract compared with placebo for the treatment of insomnia. ACTRN12618000078257. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12618000078257
15. Australian New Zealand Clinical Trials Registry. Cannabidiol (CBD) treatment for insomnia: a three-week randomised controlled pilot trial with a placebo run-in period (CBD-INS). ACTRN12620000070932. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12620000070932
16. Walsh JH, Maddison KJ, Rankin T, et al. Treating insomnia symptoms with medicinal cannabis: a randomized, crossover trial of the efficacy of a cannabinoid medicine compared with placebo. *Sleep* 2021;44(11):zsab149.
17. McCartney D, Suraev AS, Doohan PT, et al. Effects of cannabidiol on simulated driving and cognitive performance: A dose-ranging randomised controlled trial. *J Psychopharmacol* 2022:02698811221095356.
18. Rogeberg O. A meta-analysis of the crash risk of cannabis-positive drivers in culpability studies—avoiding interpretational bias. *Accid Anal Prev* 2019;123:69-78.

19. Rogeberg O, Elvik R. The effects of cannabis intoxication on motor vehicle collision revisited and revised. *Addiction* 2016;111(8):1348-59.

20. Ramaekers JG. Driving under the influence of cannabis: an increasing public health concern. *Jama* 2018;319(14):1433-34.

21. McCartney D, Arkell TR, Irwin C, et al. Determining the magnitude and duration of acute Δ9-tetrahydrocannabinol (Δ9-THC)-induced driving and cognitive impairment: a systematic and meta-analytic review. *Neurosci Biobehav Rev* 2021;126:175-93.

22. McCartney D, Suraev A., McGregor I.S. The "next day" effects of cannabis use: a systematic review. *Cannabis and Cannabinoid Res* 2022 [accepted for publication 14-Nov-2022].

23. Ohlsson A, Lindgren JE, Wahlen A, et al. Plasma delta-9-tetrahydrocannabinol concentrations and clinical effects after oral and intravenous administration and smoking. *Clin Pharmacol Therap* 1980;28(3):409-16.

24. Gjerde H, Clausen GB, Andreassen E, et al. Evaluation of Dräger DrugTest 5000 in a naturalistic setting. *J Anal Toxicol* 2018;42(4):248-54.

25. Musshoff F, Hokamp EG, Bott U, et al. Performance evaluation of on-site oral fluid drug screening devices in normal police procedure in Germany. *Forensic Sci Int* 2014;238:120-24.

26. Van der Linden T, Wille SM, Ramírez-Fernandez M, et al. Roadside drug testing: comparison of two legal approaches in Belgium. *Forensic Sci Int* 2015;249:148-55.

27. Herrera-Gómez F, García-Mingo M, Colás M, et al. Drivers who tested positive for cannabis in oral fluid: a longitudinal analysis of administrative data for Spain between 2011 and 2016. *BMJ Open* 2019;9(8):e026648.

28. Boorman M, Owens K. The Victorian legislative framework for the random testing drivers at the roadside for the presence of illicit drugs: an evaluation of the characteristics of drivers detected from 2004 to 2006. *Traffic Inj Prev* 2009;10(1):16-22.

29. Scherer JN, Fiorentin TR, Borille BT, et al. Reliability of point-of-collection testing devices for drugs of abuse in oral fluid: A systematic review and meta-analysis. *J Pharm Biomed Anal* 2017;143:77-85.

30. Arkell TR, Kevin RC, Stuart J, et al. Detection of Δ9 THC in oral fluid following vaporized cannabis with varied cannabidiol (CBD) content: An evaluation of two point-of-collection testing devices. *Drug Test Anal* 2019;11(10):1486-97.

31. Suraev A, Grunstein RR, Marshall NS, et al. Cannabidiol (CBD) and Δ9-tetrahydrocannabinol (THC) for chronic insomnia disorder ('CANSLEEP'trial): protocol for a randomised, placebo-controlled, double-blinded, proof-of-concept trial. *BMJ Open* 2020;10(5):e034421.

32. Carney CE, Harris AL, Falco A, et al. The relation between insomnia symptoms, mood, and rumination about insomnia symptoms. *J Clin Sleep Med* 2013;9(6):567-75.

33. Krahn LE, Arand DL, Avidan AY, et al. Recommended protocols for the multiple sleep latency test and maintenance of wakefulness test in adults: guidance from the American Academy of Sleep Medicine. *J Clin Sleep Med* 2021;17(12):2489-98.

34. Grove JR, Prapavessis H. Preliminary evidence for the reliability and validity of an abbreviated profile of mood states. *Int J Sport Psychol* 1992; 23(2), 93-109.

35. Arkell TR, Lintzeris N, Kevin RC, et al. Cannabidiol (CBD) content in vaporized cannabis does not prevent tetrahydrocannabinol (THC)-induced impairment of driving and cognition. *Psychopharmacology* 2019;236(9):2713-24.

36. Walker MP, Brakefield T, Seidman J, et al. Sleep and the time course of motor skill learning. *Learn Mem* 2003;10(4):275-84.

37. Terpening Z, Naismith S, Melehan K, et al. The contribution of nocturnal sleep to the consolidation of motor skill learning in healthy ageing and P arkinson's disease. *J Sleep Res.* 2013;22(4):398-405.

38. Dorrian J, Rogers NL, Dinges DF. Psychomotor vigilance performance: Neurocognitive assay sensitive to sleep loss: CRC Press 2004.

39. Lezak MD, Howieson DB, Loring DW, et al. Neuropsychological Assessment. Fourth Edition. Oxford University Press, New York, USA 2004, 1029pp.

40. McLeod DR, Griffiths RR, Bigelow GE, et al. An automated version of the digit symbol substitution test (DSST). *Behav Res Methods Instrum* 1982;14(5):463-66.

41. Vandrey R, Herrmann ES, Mitchell JM, et al. Pharmacokinetic profile of oral cannabis in humans: blood and oral fluid disposition and relation to pharmacodynamic outcomes. *J Anal Toxicol* 2017;41(2):83-99.

42. Kleykamp BA, Griffiths RR, Mintzer MZ. Dose effects of triazolam and alcohol on cognitive performance in healthy volunteers. *Exp Clin Psychopharmacol* 2010;18(1):1.

43. Herrmann ES, Cone EJ, Mitchell JM, et al. Non-smoker exposure to secondhand cannabis smoke II: effect of room ventilation on the physiological, subjective, and behavioral/cognitive effects. *Drug Alcohol Depend* 2015;151:194-202.

44. Spindle TR, Cone EJ, Schlienz NJ, et al. Acute effects of smoked and vaporized cannabis in healthy adults who infrequently use cannabis: a crossover trial. *JAMA Netw Open* 2018;1(7):e184841-e41.

45. Huestis MA. Human cannabinoid pharmacokinetics. *Chem Biodivers* 2007;4(8):1770.

46. Åkerstedt T, Anund A, Axelsson J, et al. Subjective sleepiness is a sensitive indicator of insufficient sleep and impaired waking function. *J Sleep Res* 2014;23(3):242-54.

47. Ingre M, Kecklund G, Åkerstedt T, et al. Variation in sleepiness during early morning shifts: a mixed model approach to an experimental field study of train drivers. *Chronobiol Int* 2004;21(6):973-90.

48. Lee D, Huestis MA. Current knowledge on cannabinoids in oral fluid. *Drug Test Anal* 2014;6(1-2):88-111.

49. Lee D, Karschner EL, Milman G, et al. Can oral fluid cannabinoid testing monitor medication compliance and/or cannabis smoking during oral THC and oromucosal Sativex administration? *Drug Alcohol Depend* 2013;130(1-3):68-76.

50. McCartney D, Kevin RC, Suraev AS, et al. Orally administered cannabidiol does not produce false-positive tests for Δ9-tetrahydrocannabinol on the Securetec DrugWipe® 5S or Dräger DrugTest® 5000. *Drug Test Anal* 2022;14(1):137-43.

51. Boggs DL, Nguyen JD, Morgenson D, et al. Clinical and preclinical evidence for functional interactions of cannabidiol and Δ9-tetrahydrocannabinol. *Neuropsychopharmacology* 2018;43(1):142-54.

52. Nicholson AN, Turner C, Stone BM, et al. Effect of Δ-9-tetrahydrocannabinol and cannabidiol on nocturnal sleep and early-morning behavior in young adults. *J Clin Psychopharmacol* 2004;24(3):305-13.

5. General Discussion

5.1 Chapter Overview

This thesis is comprised of a series of investigations designed to address key knowledge gaps and scientific understanding of the therapeutic influence of cannabinoids on sleep, as outlined in the introductory chapter. The aims of these investigations were as follows:

1. Systematically review and evaluate the preclinical and clinical evidence for the use of cannabinoid therapies in the treatment of defined sleep disorders (Chapter 2).
2. Characterise current user characteristics and use patterns for prescribed and illicit medicinal cannabis for the treatment of sleep disorders in the Australian community (Chapter 3).
3. Develop a high-quality, randomised, placebo-controlled, crossover trial protocol to examine the acute effects of CBD/THC in a clinical insomnia population (Chapter 4.1).
4. Characterise the acute effects of CBD/THC relative to placebo on objective and subjective sleep measures in a clinical population with chronic insomnia (Chapter 4.2).
5. Explore the effects of CBD/THC relative to placebo on global spectral power during sleep using high-density EEG (Chapter 4.2).
6. Determine the safety profile of an acute oral dose of a CBD/THC product in a clinical insomnia population (Chapter 4.2).
7. Examine possible 'next day' impairment following night time use of a CBD/THC product by assessing cognitive function, alertness, and simulated driving performance (Chapter 4.3).
8. Establish the accuracy of two commonly used point-of-collection-testing (POCT) devices, Securetec DrugWipe 5s and Dräger Drug Test 5000, in detecting THC in oral fluid the morning after evening drug administration. (Chapter 4.3).
9. Characterise plasma and oral fluid THC and CBD concentrations at various time points following use of the CBD/THC product *versus* placebo (Chapter 4.2 and Chapter 4.3).

Overall, the studies that make up Chapters 2, 3, and 4.1-4.3 successfully addressed each of these aims. The current final chapter presents a general discussion of the main findings of the

thesis and is comprised of two sections. The first section provides a summary, integration, and discussion of key findings from each experimental chapter as well as discussing possible limitations. The second section will go on to consider the wider implications of these findings and potential avenues for future research. **Table 1** presented at the end of this chapter outlines future research directives in the investigation of cannabinoids as treatment for insomnia disorder.

5.2 Summary of Findings

5.2.1 Chapter 2: Cannabinoid Therapies in the Management of Sleep Disorders: A Systematic Review of Preclinical and Clinical Studies

Chapter 2 presented a systematic review of preclinical and clinical studies investigating the therapeutic effects of cannabinoids in the management of sleep disorders (or a preclinical model of a sleep disorder). The search was conducted through five electronic databases (PubMed, Embase, PsycINFO, Scopus, Web of Science, and CINAHL) up to November 2019. Each study was assessed for bias using the SYRCLE tool for preclinical studies and the Cochrane Risk of Bias tool (RoB 2.0) for clinical studies. Key studies published between 2019 and 2022 were described in an addendum in Section 2.7. As of November 2022, 14 preclinical and 14 clinical studies met criteria for inclusion.

There were several interesting outcomes from this review. Overall, there was insufficient evidence to support the routine use of medicinal cannabis as a safe and effective treatment for any sleep disorder. Most included studies carried a substantial risk of bias, typically by failing to control for other substance use, using outcome measures that lacked psychometric validation, and often failing to blind study participants. We identified weak supporting evidence for the use of cannabinoids in the treatment of obstructive sleep apnea (OSA) with one open-label study and a subsequent 6-week RCT (from the same research team) showing reductions in apnea-hypopnea index after treatment with THC (dronabinol; oral, 2.5 or 10 mg/day).[1 2] However, findings from

the RCT should be interpreted with caution as the statistically significant reduction in the adjusted AHI with 10 mg/day dronabinol treatment was at least partly attributable to a potentially clinically meaningful baseline imbalance in AHI and a significant 8.5-point worsening in AHI in the placebo-treated group after six weeks.

There was similarly weak evidence that cannabinoids were effective in improving sleep-related outcomes in insomnia disorder. One exception is a recently published 2-week trial administering a nightly dose of a THC-dominant oral formulation 23 participants with insomnia disorder.[3] Participants started with a nightly dose of combined 10 mg THC, 1 mg CBN and 0.5 mg CBD with 52% of participants doubling the dose by end of week 2. This study showed a significant improvement in ISI of 5.1 points relative to placebo which was accompanied with improvements in actigraphy-derived TST and WASO, but not in polysomnography measures. Further, a relatively short washout (one week) may have led to potential carryover effects, although treatment order effects were not reported in the statistical analysis. All participants correctly guessed the order in which they had received the active medication (not uncommon for cannabinoid trials), with the authors concluding that blinding of treatment condition could not be readily achieved.

There are still no published RCTs investigating the effects of CBD in insomnia disorder despite increasing interest in CBD as a sleep-promoting drug.[4] However, there has been a recent uptick in new clinical trials exploring the effects of CBD, especially 'low dose' CBD (i.e., 150 mg/day), in insomnia disorder,[5-8]. This array of trials has been in response to a decision by the Australian federal regulator, the Therapeutic Goods Administration (TGA), to down-schedule CBD to Schedule 3 (*Pharmacist Only Medicine*).[9] There are three additional pre-registered trials currently underway examining the effects of cannabinol (CBN),[10] CBD-terpene formulation,[11] and combined THC and CBD[12] in patients with insomnia; all using oral formulations. There was a lack of good-quality evidence to support the use of cannabinoids for other sleep disorders such as restless legs syndrome, REM sleep behaviour disorder (RBD), narcolepsy, and PTSD-related

nightmares. Of note, a 12-week treatment with CBD (oral; 300mg) involving 33 patients with RBD showed no advantage of CBD over placebo in the frequency of nights with RBD symptoms using the Clinical Global Impressions Scale.[13]

There are some limitations to note with the systematic review in Chapter 2. First, the review included only English-language articles, and, despite best efforts, it is possible that other relevant articles were missed in the search. The current review was limited to clinical studies where cannabinoids were administered to treat a sleep disorder (only). However, a recent meta-analysis of randomised trials showed that sleep improved when cannabinoids were used to treat a comorbid condition (e.g., chronic pain), albeit the magnitude of benefit was considered small.[14]

In summary, the systematic review presented in Chapter 2:

1. Synthesised the existing preclinical and clinical evidence base of studies in which cannabinoids were administered to treat a sleep disorder.
2. Concluded that there was currently insufficient evidence to support the routine use of medicinal cannabis as a safe and effective treatment for any sleep disorder.
3. Showed that the available clinical evidence has a moderate-to-high risk of bias with frequent problems arising from randomisation processes and selective reporting of results.
4. Highlighted key safety considerations for the use of cannabinoids such as the effects of THC on cognitive performance and driving, and the potential for drug-drug interactions.
5. Outlined key research agendas for further examination of the therapeutic utility of cannabinoid therapies in sleep disorders such as using validated objective measures and measures of 'next day' function (i.e., cognition and driving performance).

5.2.2 *Chapter 3. Medicinal Cannabis Use Patterns for Sleep Disorders in Australia: Results of the CAMS-20 Survey*

Chapter 3 presented the results of a substudy from the Cannabis as Medicine 2020-2021 Survey (CAMS 20-21).[15] This probed current patterns of medicinal cannabis use in Australian respondents who self-reported using prescribed or illicit cannabis, or both, to treat a sleep disorder. This chapter also explored associations between respondent characteristics and cannabis use patterns with the aim of elucidating factors that may increase a person's likelihood of using medical cannabis to treat a sleep disorder.

There were several noteworthy outcomes from this study. First, of the 1600 respondents who completed the survey, the majority (64.4%) self-reported using medical cannabis to treat a sleep disorder, but only 16.8% of respondents endorsed a sleep disorder as the main condition they were treating. This suggests that sleep disorders are commonly being treated *secondary* to a primary medical condition such as chronic pain or a mental health disorder, as was highlighted in this survey. The high rate of medical cannabis prescription for sleep disorders in Australia;[16] may therefore be aimed to improve sleep disturbances amidst an array of other symptoms associated with the primary condition such as pain, anxiety, and/or depression. Poor sleep can significantly adversely impact disease symptoms, and the development, relapse, or exacerbation of many different disease states. Disruptions in the sleep-wake cycle are a core component of the pathophysiology and symptomatology of pain, mood and anxiety disorders[17] [18], and a reciprocal bi-directional manner can see sleep disturbance worsen pain and anxiety symptoms and *vice versa*.

Across several Phase 3 trials, nabiximols (*Sativex*) improved short-term sleep-related outcomes in individuals with sleep disturbances secondary to chronic non-cancer pain (e.g., neuropathic pain, spasticity in multiple sclerosis, rheumatoid arthritis).[19] In a Canadian retrospective cohort study, medical cannabis was perceived to be efficacious in improving insomnia symptoms in a naturalistic sample of people with anxiety (n=463), depression (n=100), and comorbid depression and anxiety (n=114), regardless of age and gender.[20] A retrospective

study examining patterns of medical use in 61,379 US patients showed that the average number of comorbid medical conditions being treated with medical cannabis were 2.7 (SD=2.6).[21] The most commonly reported comorbid conditions were anxiety (42%), followed by back and neck problems (30%), and insomnia (27%), similar to CAMS20-21 survey findings. Although outside the scope of the current thesis, future research might usefully focus on the extent to which cannabinoids directly influence sleep, or have a primary influence on the co-morbid conditions that are known to affect sleep (e.g., anxiety), or both.[22]

Relative to other indications, the use of medical cannabis for a sleep disorder was associated with younger age, inhaled routes of administration, use of THC-dominant products, and use of both illicit and prescribed forms of cannabis. This agrees with a recent analysis of medical cannabis prescribing in Australia showing that SAS-B approvals for sleep disorders were typically for flower products (i.e., consumed via inhalation) and predominantly for Schedule 8 products (i.e., containing >2% THC).[16] THC is known to increase subjective drug effects such as 'drowsy' or 'sleepy/tired' which are indicative of sedative properties[23 24] while inhaled methods of administration are associated with faster onset and shorter duration of drug effects[25]. These are a potentially useful combination for those seeking immediate relief from insomnia symptoms. However, there are risks associated with use of inhaled THC-dominant products such as tolerance, dependence, and a number of possible adverse drug effects. As explored in Chapter 4.3, the co-administration of CBD with THC may possibly prevent some of the adverse effects of THC such as anxiety,[26] acute psychotic symptoms (in regular non-medical cannabis users)[27 28], and even next-day memory impairment and drowsiness.[29] For instance, one study showed that co-administration of CBD and THC (oral, 15 mg each) was associated with less residual sedative activity than administration of THC alone (oral, 15 mg) in eight healthy volunteers.[29] However, the ability of CBD to prevent THC effects remains controversial, with some studies unable to replicate such effects.[30 31]

The vast majority of respondents in the CAMS20-21 survey perceived improvement in their sleep disorder after starting medical cannabis irrespective of whether it was sourced illicitly or prescribed (both >93%). Further, 95% of respondents also reported some reduction in their use of benzodiazepines (95%) and alcohol (63%). While undoubtedly positive, the extent to which this reflects treatment efficacy versus positive expectancies (i.e., placebo effects) and a self-selected sample of is difficult to ascertain. Convenience sampling may produce a selection bias whereby those who had more favourable experiences with medical cannabis are more likely to complete the survey. The lack of formal diagnoses in a self-reporting sample also means that information about the participant's medical history may not be accurate or verifiable. Despite these limitations, the findings presented in Chapter 2 provide a novel insight into medical cannabis use patterns for sleep disorders in Australia and the gradual transition to use of legal prescription products that is underway. The outcomes of the survey also highlight the need for high quality placebo-controlled trials (as exampled in Chapter 4.1) to better understand the potential efficacy of cannabinoids in managing sleep disorders.

In summary, the survey presented in Chapter 2 showed that:

1. More than 60% of survey respondents self-reported using medical cannabis to treat a sleep disorder, with insomnia disorder (86%) being the most common.
2. Only 16.8% of respondents chose a sleep disorder as the primary condition being treated suggesting that most sleep problems are treated *secondary* to another health condition.
3. The main co-morbid health conditions selected by those using medical cannabis to treat a sleep disorder were pain (42%) and mental health-related (33%).
4. The use of inhaled methods (i.e., smoking or vaping) and THC-dominant products, from illicit sources were common among survey respondents with sleep disorders.
5. Most respondents (>93%) reported perceived improvement in their sleep disorder after commencing medical cannabis with many reporting a reduction in their benzodiazepine use.

5.2.3 *Chapter 4.1-4.2. Investigating the Therapeutic Effects of Combined Cannabidiol and Δ^9-Tetrahydrocannabinol in Insomnia Disorder*

Chapters 4.1 and 4.2 presented the methodology and results, respectively, of a randomised, placebo-controlled trial investigating the effects of an acute orally administered cannabinoid product containing a ratio of 20:1 CBD and THC in the treatment of chronic insomnia disorder. This study used a rigorous clinical trial crossover design to determine the effects of the CBD/THC product on (1) objective sleep outcomes (specifically, TST and WASO); (2) subjective sleep outcomes as measured on the LSEQ; (3) global EEG power spectral analysis using high-density EEG; and (4) type and frequency of adverse events. Several aspects of this study were novel relative to the existing literature (Chapter 4.2).

Chapter 4.2 showed that a single acute dose of the CBD/THC product significantly reduced TST (-24.5 min) with no effect of WASO or subjective sleep outcomes. Our findings were in contrast to the Walsh et al., (2022) RCT which showed an improvement in subjective sleep quality of 5.07 units on the ISI relative to placebo, accompanied by a significant increase in actigraphy-derived TST (+33.4 min) and a decrease in actigraphy-derived WASO (-10.2 min),after 2-weeks of treatment with a THC-dominant product that also contained CBD and CBN (orally administered; up to 20 mg THC per night).[3] Differences in outcomes between the two studies may be explained in part to the dose used, the difference in cannabinoid profile between investigational products, the cannabis use history of participants, and, perhaps most importantly, the use of acute versus repeated dosing.

With respect to dose, it is possible that a single dose of oral 10 mg THC dose is not optimal for sedative effects and may have inadvertently caused some stimulatory effects,[32] particularly in infrequent cannabis users. In one study, THC (oral; 10 mg) significantly increased heart rate, 'stimulant-like' subjective effects, and anxiety relative to placebo in 16 infrequent cannabis users (lifetime use <15 times).[33] Infrequent or cannabis-naïve users tend to exhibit greater sensitivity to

the acute pharmacodynamic effects of cannabis compared to regular users who typically develop tolerance to the adverse effects of THC.[34] For example, one study showed that THC (vaporised; 8 mg) significantly increased heart rate and subjective measures of intoxication relative to placebo, with a relatively greater degree of intoxication in infrequent cannabis users compared to frequent cannabis users.[35] While some prior studies have reported dose-related increases in sedative effects with cannabis,[36] other studies have reported dose-related increases in 'stimulant-like' effects.[37 38] The latter may be related to higher doses of cannabis or THC[39] and pertain to the negative effects of intoxication such as increased heart rate, anxiety, tension, or decreased relaxation.[40] This may not be suitable for individuals with a condition marked by hyperarousal including physiological (e.g., increased body temperature, altered heart rate variability, increased cortical activation on EEG)[41-43] and psychological (e.g., hypervigilance and excessive rumination at sleep onset)[44] as described in the introductory chapter of this thesis. It has also been shown that the first day of treatment with THC is associated with the highest number of treatment-related adverse events and that the drug becomes much better tolerated with repeated dosing.[45] This suggests increased tolerability with repeated dosing and may explain the observed improvement in sleep outcomes in the Walsh et al., (2022) RCT.[3]

As highlighted in the introductory chapter of this thesis (see Section 1.10.6), prior studies of cannabis effects on sleep architecture show mixed and contradictory results. Some have shown that cannabis can supress REM sleep,[46-49] others the opposite[1 50] while one study showed that CBD (oral; 300 mg) had no significant effect on sleep architecture in healthy individuals.[51] As illustrated in Chapter 4.2, CBD/THC significantly suppressed REM sleep and increased REM sleep latency. This is a key finding in the current thesis. These findings converge somewhat with the Walsh et al., (2022) RCT described earlier which showed a significant increase in REM sleep latency (54.2 min) and a non-significant trend towards reduced REM sleep (-3.5%, p=0.055) in patients with insomnia following 2-weeks of treatment with a THC-dominant formulation (oral; 10-20 mg THC/night).[3] However, the former finding may be at least partly attributed to a substantial

decrease in REM sleep latency from baseline (127 min) to end of week 2 in the placebo group (71 min). Interestingly, the dose administered in this trial was the same or higher (10 or 20 mg THC/night) than the study presented in the current thesis suggesting possible tolerance to the effects of cannabinoids on REM sleep with repeating dosing and/or differences in study participant's prior cannabis exposure as described earlier.

High-density EEG power spectral analysis revealed complex changes in sleep architecture with somewhat paradoxical features. This included decreased fast activity during N2 sleep indicating deeper sleep but decreased delta activity during N3 sleep indicating reduced sleep depth. This coincided with an increase in time spent in N2 sleep. High frequency EEG activity is one of the most commonly reported physiological correlates of insomnia, and is regarded as a sign of cortical hyperarousal,[52 53] that is amenable to treatment with CBT-I.[54 55] For example, a previous study using high-density EEG recordings revealed that insomnia patients had more high frequency EEG activity during NREM sleep relative to normal sleepers and that these changes were widespread across the scalp.[56] Thus, the present results suggest that a CBD/THC intervention may help ameliorate CNS hyperarousal during sleep in insomnia. We observed that the CBD/THC product caused decreased delta activity during N3 sleep indicating reduced sleep depth. The reduction in delta activity was unexpected for a drug intervention that was intended to improve sleep.[57] We also observed increased fast activity (i.e., alpha and beta) during REM suggesting heightened arousal. Overall, this indicates that acute CBD/THC treatment produces a complex array of effects on brain electrical activity across frequency bands and cortical topography. The clinical significance of these novel findings involving the new technique of high-density power spectral analyses are uncertain and require replication and expansion, particularly with repeated dosing study designs.

As highlighted in Chapter 4.2, a total of 55 adverse events were reported from 16 (out of 20) participants after the single dose of the CBD/THC product compared to 30 adverse events from 13 participants during placebo treatment. The most common adverse events related to

CBD/THC were dry mouth, drowsiness/sedation, and fatigue; all were mild and had either resolved overnight or upon waking. There were no serious adverse events or participant dropouts due to an adverse event. By comparison, the Walsh et al., (2022) RCT which administered the same or higher dose of THC (oral; 10-20 mg THC per night) reported a total of 36 adverse events in 17 participants during active treatment, with the most common being dry mouth, dizziness, and headache/feeling abnormal.[3] All adverse events were classified as mild and self-limiting, however, one participant withdrew after the fourth night of active medication dosing due to non-serious adverse events (i.e., dry mouth, oral hypesthesia, swollen tongue and nausea). Four non-serious adverse events were recorded from four participants during dosing with the placebo medication; a much lower frequency of adverse events with placebo than the study presented in the current thesis that may be related to inadequate blinding.

There were several potential limitations to the clinical trial described in this thesis. The study excluded high risk individuals (e.g., participants with comorbid sleep disorders or using concomitant CNS-active medications) that are typical of patients with insomnia disorder, thereby limiting generalisability of results. Further, the use of a combination CBD/THC investigational product meant that the individual contribution of THC and CBD to observed effects could not be discerned. Inadequate duration of washout was identified as another potential problem whereby residual concentrations of CBD and major Phase-I metabolites of CBD and THC (but not THC itself) could be observed in participants allocated to the treatment sequence where placebo was given as the second crossover treatment *after* CBD/THC (n=10). This indicates that a 1-week washout period is inadequate for a single dose of the CBD/THC product (oral; combined 200 mg CBD and 10 mg THC) to clear completely from blood and that caution is necessary in crossover designs involving cannabinoid treatment, particularly when higher doses and/or repeated dosing regimens are used. Despite this, there was no evidence of treatment order effects influencing outcomes, as described in Chapter 4.2.

Strengths of the clinical trial was the use of a randomised, placebo-controlled trial design and a regulated and quality-assured cannabinoids formulation in patients with clinician-diagnosed insomnia disorder as well as verified abstinence from external cannabis use. Familiarity with the testing environment was also provided with participants staying in the same bedroom across all three overnight stays (i.e., diagnostic sleep study and two treatment visits). Participants also completed a separate in-person visit to practise wearing the high-density EEG sensor cap during a 20-minute 'nap'. No significant treatment order effects were observed suggesting ample habituation was achieved. With regards to blinding, 70% of participants correctly guessed they were receiving CBD/THC and 60% correctly guessed they were receiving placebo and two participants (10%) were 'not sure' indicating that effective blinding was not achieved. Despite this, no positive expectancy effects were observed given the lack of significant improvement in subjective sleep outcomes and no significant treatment order effects were observed as previously mentioned. Further, our primary outcomes were objective sleep outcomes which are believed to be less susceptible to positive expectancy effects.

In conclusion, the work presented in Chapters 4.1 and 4.2 showed:

1. The design of a high-quality, randomised, placebo-controlled, crossover trial protocol that explores the acute effects of combined CBD/THC in a clinical insomnia population.
2. That an acute, single oral dose of combined 200 mg CBD and 10 mg THC significantly reduced TST and had no effect on WASO or subjective sleep outcomes.
3. A clear, acute, REM suppressing effect of cannabinoids in a clinical insomnia population.
4. Paradoxical effects of the cannabinoid product on high-density EEG global power spectral analysis with decreased high-frequency activity during N2 sleep indicating deeper sleep and decreased delta activity during N3 sleep indicating reduced sleep depth. Increased high-frequency activity during REM also suggested heightened arousal.
5. That residual cannabinoids can persist in blood for at least a week following a single acute CBD/THC dose mandating caution crossover trial designs involving cannabinoids.

5.2.4 *Chapter 4.3. The 'Next Day' Effects of Combined Cannabidiol and Δ9-Tetrahydrocannabinol in Insomnia Disorder*

Chapter 4.3 investigated the effects of acute evening treatment with a CBD/THC product on 'next day' neurocognitive performance, alertness, and driving performance. It also examined whether two commonly used point-of-collection testing (POCT) devices for mobile drug testing - the DrugWipe 5s and Draeger DT5000 – could detect exposure to this product. With the exception of a possible (subtle) increase in subjective measures of drowsiness, no reliable changes in 'next day' function including cognitive function, driving performance, and (objective) alertness were observed after a single, oral dose of combined 200 mg CBD and 10 mg THC. We found that a single dose of CBD/THC increased subjective feelings of "sedated" the next morning (i.e., 10 h post drug-administration) and there was a statistically (although not clinically) significant increase in drowsiness of 0.42 points on a measure of daytime drowsiness (Karolinksa Sleepiness Scale) relative to placebo. However, both effects were small and did not translate into significantly poorer performance on cognitive function, driving performance, or an objective test of alertness.

This adds to a pre-existing body of work (summarised in our recent review)[58] showing that short-term use of cannabinoids does not impact 'next-day' cognitive function. Interestingly, as shown in Chapter 4.2, acute CBD/THC treatment reduced total sleep time and REM sleep and provided no improvement in subjective sleep quality in individuals with insomnia disorder. This suggests that, despite a reduction in total sleep time and REM sleep, there were no observed deterioration in 'next day' function following acute CBD/THC treatment. It would be of interest for future research to focus on exploring possible 'next day' effects following higher dose levels, or repeated dosing, with cannabinoids in individuals with insomnia disorder.

Chapter 4.3 also characterised the cannabinoid concentrations in oral fluid following controlled oral administration of a single dose containing 200 mg CBD and 10 mg THC and evaluated the performance of two POCT devices (DW5s and DT5000). The aim of this was to address the major concerns around roadside drug testing in medicinal cannabis patients (including

those with sleep-related conditions)[59] and to investigate the reliability and the accuracy of these two devices, which have also given rise to previous concerns.[60]

The latter aim was achieved by comparing observed test results using these devices against "gold standard" LC-MS/MS quantified THC concentrations in oral fluid. As anticipated, THC concentrations were not detectable in oral fluid at baseline (prior to drug administration) and across all timepoints with placebo treatment. One confirmed false positive test result was observed at baseline during placebo treatment on the DW5s in one participant which is a major concern.

The average (SD) THC concentrations (ng/mL) were 44.7 (101.2), 2.6 (3.9), and 'not detectable', at 0.5 h, 10 h, and 18 h post-drug administration, respectively. Despite the reasonably high concentration of oral THC fluid at 0.5 h post-drug administration (>10 ng/mL screening cut-off on both POCT devices), the significant variability in concentration led to only three (out of a possible 20) DW5s tests returning a positive THC result and four (out of a possible 20) DT5000 tests returning a positive THC result. The next morning (10h post-drug administration), no positive THC result were observed on the DW5s while one (out of a possible 20) positive THC result was observed with the DT5000. No positive THC results were observed on either device at 18 h post-drug administration. Overall, these findings indicate a low of obtaining a positive THC result the next day after evening use of an oral (oil) formulation containing 10 mg THC (in combination with 200 mg CBD).

The results revealed that neither device met the minimum performance standard suggested by the highly influential European Union-funded Driving Under the Influence of Drugs (DRUID) project (i.e., minimum 80% sensitivity, specificity, and accuracy).[61] At the 10 ng/mL screening cut-off, the DW5s sensitivity, specificity, and accuracy was 10%, 98%, and 91%, respectively. Overall, 3/136 (2.2%) tests were false positives, and 9/136 (6.6%) tests were false negatives. For the DT5000, the sensitivity, specificity, and accuracy were 38%, 100%, and 96%. Overall, there were no false positives and 4% were false negatives. Of note, the occurrence of false positive and false negative tests on both devices was greatest at 0.5 h post-drug administration. True positive tests

were rarely observed at the 10 h and 18 h timepoint. This indicates that the morning after drug administration, THC concentrations in oral fluid were lower than the screening 10 ng/mL cut-off on the POCT devices and therefore posed a low chance of producing a positive THC result.

In conclusion, the work presented in Chapter 4.3 showed:

1. With the exception of a possible (subtle) increase in subjective measures of sleepiness, there were no reliable changes in 'next day' function (~12 h post-drug administration) including cognitive function, driving performance, or objective alertness.
2. That the lack of 'next day' effects of CBD/THC occurred despite a reduction in total sleep time and no clear improvement in subjective sleep outcomes.
3. Revealed that evening administration with an oral cannabinoid oil (a) produced significant variability in oral THC fluid concentrations post-administration and (b) posed a low chance of obtaining a positive THC result on two POCT devices during testing the *next day*.
4. POCT devices showed poor sensitivity, even at 30 min post-drug administration, yielding many false negative results. True negatives were apparent the morning after drug administration (10 h post-drug administration).
5. Highlighted the need for future research to explore the effects of repeated dosing, and higher doses of cannabinoids, on 'next day' function in insomnia disorder.

5.3 Wider implications and future directions

5.3.1. Are cannabinoids effective and safe in individuals with insomnia disorder?

One of the major aims of this thesis was to determine whether oral administration of cannabinoids improved sleep in chronic insomnia disorder. Chapter 2 illustrated that current clinical evidence is limited, however, research on this topic is gradually expanding with one notable clinical trial recently published[3] and several other clinical trials currently underway, including several examining the efficacy of low-dose CBD (i.e., <150 mg/day) in the treatment of insomnia.

Since the publication of our systematic review, a surplus of other reviews on cannabinoids and sleep have been published[14 22 62-72] despite limited new original research emerging during that time. Chapter 3 underscored the substantial interest and uptake of medical cannabis in the Australian community with more than 60% of the 1600 surveyed respondents self-reporting using medical cannabis to treat a sleep disorder. Of these, an overwhelming proportion self-reported improvement in their sleep disorder after commencing medical cannabis irrespective of how it was sourced [i.e., illicit (96.4%) and prescribed (93.5%)]. However, in something of a contradiction to such use, Chapters 4.1-4.3 showed that oral ingestion of a 20:1 CBD/THC product, a common formulation thought to promote sleep,[73 74] significantly reduced TST with no apparent beneficial effect of WASO or subjective sleep outcomes in individuals with chronic insomnia disorder. CBD/THC treatment was associated with a complex array of effects on the sleeping brain with high-density EEG analyses revealed complex and seemingly paradoxical effects with sleep-promoting (i.e., reduced fast activity during N2 sleep) and sleep-reducing effects (decreased delta activity during N3 sleep and increased fast activity during REM sleep) observed. Overall, this highlights the need for caution in automatically assuming that cannabinoids are an effective treatment for insomnia disorder.

As outlined previously, these unexpected findings may have a number of possible explanations including the single oral dose level (200 mg CBD and 10 mg THC) that was assessed, the use of acute rather than repeating dosing, and the light or negligible cannabis use history of the participants. The dose chosen in the current study may not be optimal for sedative effects and may even have caused inadvertent stimulatory effects which relate to the negative effects of THC such as increased heart rate, anxiety/nervousness, or restlessness,[40] particularly in infrequent cannabis users (who exhibit greater sensitivity to the effects of cannabis).[34] This may not be suitable for individuals with a condition marked by hyperarousal including physiological (e.g., increased body temperature, altered heart rate variability, increased cortical activation on EEG)[41-43] and psychological (e.g., hypervigilance and excessive rumination at sleep onset).[44] Further, possible sex

differences in the response to acute cannabis effects have been described with females exhibiting greater sensitivity than males [75] and may have been a contributing factor to the current findings. Most participants (16/20, 80%) included the study presented in the current thesis were females and the representative of the female predisposition of insomnia disorder.[76]

We also showed that CBD/THC significantly suppressed REM sleep and increased REM sleep latency. This is a key finding in the current thesis. Most antidepressants and benzodiazepines (often prescribed for short-term insomnia) suppress REM sleep.[77] [78] Some believe this is critical to the therapeutic effects of antidepressant drugs on mood.[79] REM sleep alterations (i.e., increased REM sleep and reduced REM sleep latency) are the most prominent feature of sleep architecture in individuals with depression,[78] which may explain why some consumers report antidepressant effects with cannabis use and the increasing rates of prescribing of medicinal cannabis for depressive illness.[15] [80-82] However, disturbances in sleep architecture can result in a sense of having had non-restorative sleep and is associated with next-day impairment.[83] [84] A meta-analysis of polysomnographic studies showed that patients with insomnia present a disruption of sleep continuity and a significant reduction in slow wave sleep and REM sleep relative to good sleepers.[85] Further REM suppression with THC may therefore be contraindicated in insomnia disorder. Future studies using gradual up-titration (e.g., from 2.5 mg THC), repeating dosing schedules are needed to identify the optimal dose of THC (alone or in combination with CBD) that confers clinical efficacy without significantly disturbing sleep architecture. Indeed, the current clinical guidance around cannabinoid prescribing emphasizes the importance of precise THC dosing, using guided patient self-titration, starting with a low dose, and increasing slowly by small increments until reaching relief from symptoms while avoiding side effects (i.e., *start low, go slow*).[86]

The REM suppressant of THC may have short-term therapeutic utility for other sleep disorders such as parasomnias (e.g., trauma-associated sleep disorder) and REM sleep behaviour disorder (RBD) in Parkinson's disease.[67] One randomised, placebo-controlled trial found administration of low-dose nabilone (a synthetic THC analogue) led to improvement in self-

reported sleep quality and a decrease in the frequency of nightmares in 10 individuals with post-traumatic stress disorder.[87] No study to-date, however, has explored the use of THC for RBD and this would clearly be of interest given the results of the current thesis.

Non-intoxicating cannabinoids such as CBD are being explored as alternative sleep-promoting agents. In Australia, recent legislation down-scheduled low-dose oral CBD (maximum 150 mg/day) to Schedule 3 (*Pharmacist Only Medicine)* for over-the-counter sale of indications that require minimal medical oversight (e.g., 'subclinical' or short-term insomnia).[9] This aligns Australia with other countries such as USA, Canada, Germany, UK, Switzerland, and Japan where accessibility to CBD products over-the-counter and/or online are already available.[88] Such products typically contain low or "nutraceutical" doses of CBD (e.g., up to 100 mg/day) and are often marketed for sleep and pain.[89] However, the question remains as to whether non-prescription low oral doses of CBD can deliver therapeutic doses. A recent review of the evidence for low dose CBD showed that, while it is safe and tolerable, there was limited published evidence showing efficacy at doses of <300 mg CBD.[90] Therapeutic benefits of oral CBD became more apparent at doses greater than or equal to 300 mg, particularly with the respect to anti-anxiety effects.[90] No published study to-date has examined the therapeutic utility of CBD for insomnia disorder, however, several clinical trials are currently underway exploring the effects of low-dose CBD in insomnia disorder.[5-7 91] Other phytocannabinoids such as cannabinol (CBN), an oxidative by-product of THC present in relatively low concentrations in the plant, are also being explored as a sleep-promoting agent.[10]

Interestingly, there is an emerging preclinical evidence suggesting that CBD may have 'wake-promoting' or alerting properties.[92] One preclinical study showing that CBD partially blocked excessive sleepiness in hypocretin-deficient rats, an animal model of narcolepsy.[93] There are no published clinical studies of cannabinoids for the treatment of disorders of hypersomnolence such as narcolepsy or idiopathic hypersomnia. Some hypothesise that the REM-suppressing effect of THC and supposed "wake-promoting" effect of CBD could be harnessed to

treat patients with disorders of hypersomnolence.[67] Future studies should prioritise research on CBD, given its benign side-effect profile and limited abuse potential, to better understand the possible dose-dependent "alerting" versus "sedative" effects of CBD.

In terms of safety, Chapter 2 illustrated that mild adverse events are commonly reported with THC-containing medications, and these include somnolence, dry mouth, dizziness, and headache. On the other hand, CBD is generally well-tolerated with minimal adverse events. The most prominent concern with CBD is the possibility of drug-drug interactions, and these that are still subject to various investigations.[94] Of note, in the US and other jurisdictions, availability of non-prescription CBD-containing products via online or retail outlets lack regulatory oversight regarding manufacture, accurate labelling, and cannabinoid content with some studies showing THC contamination in "THC-free" products.[95 96] In contrast, Schedule 3 products in Australia must be registered and therefore undergo strict regulations to ensure quality and safety.

Chapters 4.1-4.3 showed that a single dose of combined 200 mg CBD and 10 mg THC (as a standalone treatment) was generally safe and well-tolerated in patients with insomnia disorder with the most common adverse events being dry mouth, drowsiness/sedation, and fatigue; all mild and self-limited. Similarly, in the Walsh *et al* clinical trial involving two weeks administration of a THC-dominant formulation (10 to 20 mg THC per night), the most common adverse events were not dissimilar to those in the work presented in the current thesis: dry mouth, dizziness, and headache/'feeling abnormal'.[3] Overall, this suggests that acute and short-term (2 weeks) treatment with THC-containing products are relatively safe and tolerable in individuals with insomnia disorder.

Of note, current products used in the pharmacological management of insomnia are not generally approved for use beyond 3 months duration, with no evidence for effective long-treatment with hypnotics beyond four weeks.[97] Like other pharmacological agents, cannabinoids, particularly THC, may not be a long-term solution to insomnia. Moreover, several safety concerns are still outstanding and warrant further investigation, these include: (a) the potential risk for

rebound insomnia following cessation of a THC-containing product; (b) the long-term impact of cannabinoid treatment on sleep architecture in individuals with insomnia disorder; (c) the short-term (i.e., next day) and long-term effects of repeated dosing with cannabinoids on memory, mood, and cognitive/psychomotor performance. The latter will be discussed in the next section.

In sum, the work presented in the current thesis indicates that cannabinoids have a relatively favourable safety profile and are well-tolerated, however, further research is necessary to identify the optimal dose and combination of cannabinoids to effectively treat insomnia disorder.

5.3.2. Do orally administered cannabinoids cause impairment in 'next day' function?

As highlighted in the introductory chapter of this thesis (see Section 1.11), prior research has only occasionally reported significant cognitive and/or psychomotor impairment at long durations (>8 hours) following controlled cannabis or cannabinoid administration. Studies have generally been of poor quality[58] and have often involved non-medical (i.e., recreational) users and inhaled methods of administration. Only a minority of studies have shown acute impairment (i.e., THC-related impairment occurring <8 h post-treatment) prior to assessing longer-term effects. Self-report has typically been used to determine drowsiness rather than objective measures. Very few studies have explored impairment following the use of oral cannabis products in clinical populations.

Therefore, a key aim of this thesis was to explore the possible 'next day' effects of an oral cannabinoid product in individuals with chronic insomnia who infrequently used cannabis. This provides important safety information for the many patients who currently use THC products by night to treat conditions such as insomnia, chronic pain, and anxiety. Chapter 4.3 illustrated that no reliable changes in 'next day' function including cognitive function, driving performance and objectiveness alertness were observed following acute dosing with the CBD/THC product, with the exception of a possible subtle increase in subjective measures of sleepiness. This result is

particularly salient given the apparent worsening effect of CBD/THC treatment on objective sleep outcomes, including a significant reduction in total sleep time and REM sleep, and the lack of improvement in subjective sleep quality in patients. Further high-quality studies investigating the 'next day' effects of THC in (real-world) medicinal cannabis users are needed, particularly with oral administration, to confirm these findings.

Another key issue for Australian patients using medicinal cannabis is roadside drug testing.[98] All jurisdictions in Australia currently enforce a zero-tolerance policy for driving under the influence of cannabis (DUIC), with Tasmania the only jurisdiction that provides an exemption for medicinal cannabis patients.[99] The capacity of current drug testing technologies to detect THC in oral fluid after evening administration of an oral oil formulation was explored in Chapter 4.3. The extent to which such devices give positive tests for THC the morning after evening administration of an oral cannabinoid product would be of interest to many patients who are concerned about driving-related issues.

The results were particularly notable to the extent that these devices largely failed to show recent use of an oral CBD/THC-containing product. Even at 30 mins following drug administration a large majority of results were negative. As in previous studies,[98] there was significant variability in oral THC fluid concentrations. Most participant's oral fluid THC concentrations were lower than the 10 ng/mL screening cut-off on the POCT devices at most timepoints and therefore posed a low chance of producing a positive THC result. THC in oral fluid is thought to originate exclusively from direct contamination of the oral cavity upon inhalation or ingestion, with no circulation of cannabinoids from the blood back into saliva.[25 100] Prior research indicates that concentrations of THC in oral fluid rapidly declined over 2 h after ingestion of cannabis.[60] In practice, this means that POCT devices are only able to detect very recent use of cannabis via routes of administration that result in direct contact with the oral cavity such as smoking, vaping, or edibles (but not capsules or tablets). In particular, previous studies

showing no detectable THC in oral fluid following oral administration of encapsulated THC (e.g. dronabinol).[101]

Many jurisdictions enforce a zero-tolerance policy for driving under the influence of cannabis. Previous work has suggested that the application of *per se* limits (i.e., a driver has committed an offense if THC is detected in blood or oral fluid at or above a pre-determined cut-off), as used in many overseas jurisdictions, can be problematic due to unpredictable inter- and intra-individual variability in blood and oral fluid THC concentrations, and the lack of strong association between the such concentrations and impairment.[102] Although out of the scope of the present thesis, this is a major area of research for drug policy and current practices around the detection of impairment. It also has significant real-world implications for patients such as termination of employment (with a positive drug test) or refraining from medicinal cannabis use for fear of the consequences of testing positive on a roadside drug test.

5.4 Conclusions

Sleep disorders are amongst the top indications attracting the use of medicinal cannabis, prescribed or illicit, in Australia and worldwide. However, despite the increasingly widespread utilisation of medical cannabis products in the treatment for sleep, the evidence supporting therapeutic utility remains patchy and unclear. It is therefore of great importance that rigorous clinical research examines the effects of cannabinoids on sleep in clinical insomnia populations, and associated side effects, including possible 'next day' impairment. This will help validate the existing use of medicinal cannabis for insomnia and also facilitate the widespread adoption of such interventions in mainstream medicine.

The work presented in this thesis characterised and compared, for the first time, the effects of cannabinoids and placebo on measures on sleep using high-density EEG with additional assessment of 'next day' function. It was established that a single dose of CBD/THC (containing

200 mg CBD with 10 mg THC) had an acute sleep-reducing effect with no improvement in subjective sleep outcomes in individuals with chronic insomnia disorder. High-density EEG analysis revealed complex and seemingly paradoxical effects of the CBD/THC product on sleep architecture, with decreased fast activity during N2 sleep (indicating deeper sleep) and decreased delta activity during N3 sleep (indicating reduced sleep depth). Given the growing community use of medical cannabis for sleep disorders and the rapidly expanding rate of medical cannabis prescriptions for sleep disorders, these findings have some significant clinical implications, while recognising that clinical use typically involves repeated rather than acute dosing with cannabinoids.

The absence of reliable 'next day' impairment with evening use of an oral THC-containing cannabis product is a novel finding that further informs clinicians and researchers around the safety of medical cannabis. This is the first study to explore 'next day' effects of medical cannabis on cognition and driving performance in a clinical insomnia population. This may have implications for patients who need to drive (i.e., for employment, family life) and are prescribed a THC-based medicine in the evening to help them sleep. Further research is needed to establish whether the lack of 'next day' impairment remains with higher and/or repeated dosing and whether co-administration of CBD with THC (versus THC alone) produces pharmacodynamic interactions that influence sleep and next-day function.

This thesis also highlights some of the key limitations of POCT devices as a method for detection of recent cannabis use and cannabis-impaired driving, particularly for patients using oral methods of administration. The observation from Chapter 4.3 that evening administration with an oral cannabinoid oil posed an extremely low chance of testing positive to THC on two POCT devices the *next day* is an important finding that will provide some reassurance to patients. This confirms prior work showing that POCT devices tend to only be reliable indicators for very recent cannabis use, with detectable levels of THC in oral fluid decline rapidly over two hours post-drug administration. Future studies are needed to explore whether repeated dosing may yield different results the next day.

Moving forward, high-quality randomised, controlled trial designs using validated subjective and objectives measures of sleep and 'next day' function are needed to better understand the role of cannabinoids in the treatment of insomnia disorder (see **Table 1**). It is hoped that this thesis made a novel and significant contribution to the evidence base around the use of cannabinoids in the treatment of sleep disorders and will stimulate future research in this area.

Table 1	**Summary of research directives in the investigation of cannabinoid treatment for sleep disorders**

- Utilise robustly designed randomised, controlled trial designs, employing properly powered sample sizes and validated objective and subjective measures of sleep-related outcomes to assess therapeutic efficacy of cannabinoids.
- Employ gradual up-titration, repeated dosing schedules to explore lower dose ranges of THC (alone or in combination with CBD) in order to identify the optimal dose that confers clinical efficacy without significantly disturbing sleep architecture.
- Explore the potential role of THC in managing REM sleep-state conditions such as REM sleep behaviour disorder.
- Investigate the 'next day' effects of an oral THC-containing product in occasional and medicinal cannabis users.
- Use caution in conducting crossover studies involving cannabinoids due to the long window of detection in plasma, particular when higher doses and/or repeating dosing regimens are used.

5.5 Rerefences

1. Carley DW, Prasad B, Reid KJ, et al. Pharmacotherapy of apnea by cannabimimetic enhancement, the PACE clinical trial: effects of dronabinol in obstructive sleep apnea. *Sleep* 2018;41(1)
2. Prasad B, Radulovacki MG, Carley DW. Proof of concept trial of dronabinol in obstructive sleep apnea. *Front Psychiatry* 2013;4:1.
3. Walsh JH, Maddison KJ, Rankin T, et al. Treating insomnia symptoms with medicinal cannabis: a randomized, crossover trial of the efficacy of a cannabinoid medicine compared with placebo. *Sleep* 2021;44(11):zsab149.
4. Peachman RR. Does CBD Help With Insomnia? The New York Times. Accessed 20 November 2022.https://www.nytimes.com/2022/08/30/well/live/does-cbd-help-with-insomnia.html
5. ClinicalTrials.gov. The CANabidiol Use for RElief of Short Term Insomnia (CANREST). (2022) NCT05253417. Accessed on 20 November 2022. https://beta.clinicaltrials.gov/study/NCT05253417
6. Australian New Zealand Clinical Trials Registry. A Phase III Double-Blind, Randomised Placebo-Controlled Clinical Trial to evaluate the efficacy and safety of a botanical cannabidiol (CBD) for sleep disturbances in a healthy population. ACTRN12621000632897. Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12621000632897
7. Australian New Zealand Clinical Trials Registry. Cannabidiol (CBD) treatment for insomnia: a three-week randomised controlled pilot trial with a placebo run-in period (CBD-INS). 2020. ACTRN12620000070932. Sydney (NSW): NHMRC Clinical Trials Centre, University of Sydney (Australia). Accessed 19 September, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12620000070932

8. The Market Herald. Cann Group (ASX:CAN) starts recruitment for trial of CBD treatment for sleep issues, 2022. Accessed 20 November, 2022. https://themarketherald.com.au/cann-group-asxcan-starts-recruitment-for-trial-of-cbd-treatment-for-sleep-issues-2022-02-15/

9. Australian Government. Over-the-counter access to low dose cannabidiol. Health Department. Therapeutic Goods Administration. Accessed 20 November, 2022. https://www.tga.gov.au/news/media-releases/over-counter-access-low-dose-cannabidiol

10. ClinicalTrials.gov. Cannabinol Use in Patients With Insomnia Disorder (CUPID). NCT05344170. Accessed 20 November, 2022. https://beta.clinicaltrials.gov/study/NCT05344170

11. ClinicalTrials.gov. Evaluation of an Oral Cannabidiol (CBD)-Terpene Formulation on Sleep Physiology in Participants With Insomnia. NCT05233761. Accessed 20 November, 2022. https://beta.clinicaltrials.gov/study/NCT05233761

12. Australian New Zealand Clinical Trials Registry. A single-dose, double-blind, placebo-controlled, randomised, crossover study of an oral cannabis-based medicine (ETC120) on sleep, cognition, and next-day function in adults with primary insomnia disorder. ACTRN12619000714189. Accessed 20 November, 2022. https://www.australianclinicaltrials.gov.au/anzctr/trial/ACTRN12619000714189

13. de Almeida CM, Brito MM, Bosaipo NB, et al. Cannabidiol for rapid eye movement sleep behavior disorder. *Mov Disord* 2021;36(7):1711-15.

14. AminiLari M, Wang L, Neumark S, et al. Medical cannabis and cannabinoids for impaired sleep: a systematic review and meta-analysis of randomized clinical trials. *Sleep* 2022;45(2):zsab234.

15. Lintzeris N, Mills L, Abelev SV, et al. Medical cannabis use in Australia: consumer experiences from the online cannabis as medicine survey 2020 (CAMS-20). *Harm Reduct J* 2022;19(1):1-10.

16. MacPhail SL, Bedoya-Pérez MA, Cohen R, et al. Medicinal cannabis prescribing in Australia: an analysis of trends over the first five years. *Front Pharmacol* 2022;13

17. Haack M, Simpson N, Sethna N, et al. Sleep deficiency and chronic pain: potential underlying mechanisms and clinical implications. *Neuropsychopharmacology* 2020;45(1):205-16.

18. Freeman D, Sheaves B, Waite F, et al. Sleep disturbance and psychiatric disorders. *Lancet Psychiatry* 2020;7(7):628-37.

19. Russo EB, Guy GW, Robson PJ. Cannabis, Pain, and Sleep: Lessons from Therapeutic Clinical Trials of Sativex®, a Cannabis-Based Medicine. *Chem Biodivers* 2007;4(8):1729-43.

20. Kuhathasan N, Minuzzi L, MacKillop J, et al. An investigation of cannabis use for insomnia in depression and anxiety in a naturalistic sample. *BMC Psychiatry* 2022;22(1):1-8.

21. Mahabir VK, Merchant JJ, Smith C, et al. Medical cannabis use in the United States: a retrospective database study. *J Cannabis Res* 2020;2(1):1-13.

22. Narayan A, Downey LA, Manning B, et al. Cannabinoid Treatments for Anxiety: A Systematic Review and Consideration of the Impact of Sleep Disturbance. *Neurosci Biobehav Rev* 2022:104941.

23. Spindle TR, Martin EL, Grabenauer M, et al. Assessment of cognitive and psychomotor impairment, subjective effects, and blood THC concentrations following acute administration of oral and vaporized cannabis. *J Psychopharmacol* 2021;35(7):786-803.

24. Spindle TR, Cone EJ, Schlienz NJ, et al. Acute effects of smoked and vaporized cannabis in healthy adults who infrequently use cannabis: a crossover trial. *JAMA Netw* 2018;1(7):e184841-e41.

25. Huestis MA. Human cannabinoid pharmacokinetics. *Chem Biodivers* 2007;4(8):1770.

26. Boggs DL, Nguyen JD, Morgenson D, et al. Clinical and preclinical evidence for functional interactions of cannabidiol and Δ9-tetrahydrocannabinol. *Neuropsychopharmacology* 2018;43(1):142-54.

27. Bhattacharyya S, Morrison PD, Fusar-Poli P, et al. Opposite effects of Δ-9-tetrahydrocannabinol and cannabidiol on human brain function and psychopathology. *Neuropsychopharmacology* 2010;35(3):764.

28. Schubart CD, Sommer IE, van Gastel WA, et al. Cannabis with high cannabidiol content is associated with fewer psychotic experiences. *Schizophr Res* 2011;130(1-3):216-21.

29. Nicholson AN, Turner C, Stone BM, et al. Effect of Δ-9-tetrahydrocannabinol and cannabidiol on nocturnal sleep and early-morning behavior in young adults. *J Clin Psychopharmacol* 2004;24(3):305-13.

30. Arkell TR, Lintzeris N, Kevin RC, et al. Cannabidiol (CBD) content in vaporized cannabis does not prevent tetrahydrocannabinol (THC)-induced impairment of driving and cognition. *Psychopharmacology* 2019:1-12.

31. Englund A, Oliver D, Chesney E, et al. Does cannabidiol make cannabis safer? A randomised, double-blind, cross-over trial of cannabis with four different CBD: THC ratios. *Neuropsychopharmacology* 2022:1-8.

32. Cousens K, DiMascio A. (−) δ 9 THC as an hypnotic. *Psychopharmacology* 1973;33(4):355-64.

33. Martin-Santos R, a Crippa J, Batalla A, et al. Acute effects of a single, oral dose of d9-tetrahydrocannabinol (THC) and cannabidiol (CBD) administration in healthy volunteers. *Curr Pharm Des* 2012;18(32):4966-79.

34. Colizzi M, Bhattacharyya S. Cannabis use and the development of tolerance: a systematic review of human evidence. *Neurosci Biobehav Rev* 2018;93:1-25.

35. Solowij N, Broyd S, Greenwood L-m, et al. A randomised controlled trial of vaporised Δ9-tetrahydrocannabinol and cannabidiol alone and in combination in frequent and infrequent cannabis users: acute intoxication effects. *Eur Arch Psychiatry Clin Neurosci* 2019;269(1):17-35.

36. Hart CL, Van Gorp W, Haney M, et al. Effects of acute smoked marijuana on complex cognitive performance. *Neuropsychopharmacology* 2001;25(5):757-65.

37. Chait L. Delta-9-tetrahydrocannabinol content and human marijuana self-administration. *Psychopharmacology* 1989;98(1):51-55.

38. Chait L, Burke K. Preference for high-versus low-potency marijuana. *Pharmacol Biochem Behav* 1994;49(3):643-47.

39. Zuurman L, Ippel AE, Moin E, et al. Biomarkers for the effects of cannabis and THC in healthy volunteers. *Br J Clin Pharmacol* 2009;67(1):5-21.

40. Block RI, Erwin WJ, Farinpour R, et al. Sedative, stimulant, and other subjective effects of marijuana: relationships to smoking techniques. *Pharmacol Biochem Behav* 1998;59(2):405-12.

41. Farina B, Dittoni S, Colicchio S, et al. Heart rate and heart rate variability modification in chronic insomnia patients. *Behav Sleep Med* 2014;12(4):290-306.

42. Lack LC, Gradisar M, Van Someren EJ, et al. The relationship between insomnia and body temperatures. *Sleep Med Rev* 2008;12(4):307-17.

43. Fernandez-Mendoza J, Li Y, Vgontzas AN, et al. Insomnia is associated with cortical hyperarousal as early as adolescence. *Sleep* 2016;39(5):1029-36.

44. Carney CE, Harris AL, Falco A, et al. The relation between insomnia symptoms, mood, and rumination about insomnia symptoms. *J Clin Sleep Med* 2013;9(6):567-75.

45. Peters EN, Mosesova I, MacNair L, et al. Safety, pharmacokinetics and pharmacodynamics of Spectrum yellow oil in healthy participants. *J Analytical Toxicol* 2022;46(4):393-407.

46. Feinberg I, Jones R, Walker JM, et al. Effects of high dosage delta-9-tetrahydrocannabinol on sleep patterns in man. *Clin Pharm Therap* 1975;17(4):458-66.

47. Freemon FR. Effects of marihuana on sleeping states. *JAMA* 1972;220(10):1364-65.

48. Pivik R, Zarcone V, Dement W, et al. Delta-9-tetrahydrocannabinol and synhexl: Effects on human sleep patterns. *Clin Pharm Therap* 1972;13(3):426-35.

49. Tassinari C, Ambrosetto G, Peraita-Adrado M, et al. The Neuropsychiatric Syndrome of Δ 9-Tetrahydrocannabinol and Cannabis Intoxication in Naive Subjects. Marihuana and medicine: Springer 1999:649-64.

50. Barratt ES, Beaver W, White R. The effects of marijuana on human sleep patterns. *Biol Psychiatry* 1974

51. Linares I, Guimarães F, Eckeli A, et al. Lack of cannabidiol effects on sleep-wake cycle in healthy patients. *Eur Neuropsychopharmacol* 2016;26:S369.

52. Zhao W, Van Someren EJ, Li C, et al. EEG spectral analysis in insomnia disorder: A systematic review and meta-analysis. *Sleep Med Rev* 2021;59:101457.

53. Merica H, Blois R, Gaillard JM. Spectral characteristics of sleep EEG in chronic insomnia. *Eur J Neurosci* 1998;10(5):1826-34.

54. Cervena K, Dauvilliers Y, Espa F, et al. Effect of cognitive behavioural therapy for insomnia on sleep architecture and sleep EEG power spectra in psychophysiological insomnia. *J Sleep Res*2004;13(4):385-93.

55. Jacobs GD, Benson H, Friedman R. Home-based central nervous system assessment of a multifactor behavioral intervention for chronic sleep-onset insomnia. *Behav Ther* 1993;24(1):159-74.

56. Riedner BA, Goldstein MR, Plante DT, et al. Regional patterns of elevated alpha and high-frequency electroencephalographic activity during nonrapid eye movement sleep in chronic insomnia: a pilot study. *Sleep* 2016;39(4):801-12.

57. Krystal AD, Edinger JD, Wohlgemuth WK, et al. NREM sleep EEG frequency spectral correlates of sleep complaints in primary insomnia subtypes. *Sleep* 2002;25(6):626-36.

58. McCartney D, Suraev, A., McGregor, I.S. The 'next day' effects of cannabis use: A systematic review. *Cannabis Cannabinoid Res* 2022

59. Arkell TR, Lintzeris N, Mills L, et al. Driving-related behaviours, attitudes and perceptions among Australian medical cannabis users: results from the CAMS 18-19 Survey. *Accid Anal Prev* 2020;148:105784.

60. Arkell TR, Kevin R, Stuart J, et al. Detection of Δ9THC in oral fluid following vaporised cannabis with varied cannabidiol (CBD) content: an evaluation of two point-of-collection testing devices. *Drug Test Anal* 2019

61. Schulze H, Schumacher M, Urmeew R, et al. Driving under the influence of drugs, alcohol and medicines in Europe—findings from the DRUID project. 2012; 58, 1725-5767.

62. Kwasnik A, Abreu A, Chediak A. Cannabinoids and Sleep: Helpful or Harmful? *Curr Pulmonol Rep* 2020;9(3):96-101.

63. Kaul M, Zee PC, Sahni AS. Effects of cannabinoids on sleep and their therapeutic potential for sleep disorders. *Neurotherapeutics* 2021;18(1):217-27.

64. Kesner AJ, Lovinger DM. Cannabinoids, endocannabinoids and sleep. *Front Mol Neurosci* 2020;13:125.

65. Kuhathasan N, Dufort A, MacKillop J, et al. The use of cannabinoids for sleep: A critical review on clinical trials. *Exp Clin Psychopharmacol* 2019;27(4):383.

66. Velzeboer R, Malas A, Boerkoel P, et al. Cannabis dosing and administration for sleep: a systematic review. *Sleep* 2022;45(11):zsac218.

67. Maddison KJ, Kosky C, Walsh JH. Is There a Place for Medicinal Cannabis in Treating Patients with Sleep Disorders? What We Know so Far. *Nat Sci Sleep* 2022;14:957.

68. Monti JM, Pandi-Perumal SR. Clinical management of sleep and sleep disorders with cannabis and cannabinoids: implications to practicing psychiatrists. *Clin Neuropharmacol* 2022;45(2):27-31.

69. Mondino A, Cavelli M, González J, et al. Effects of cannabis consumption on sleep. *Cannabinoids and Sleep* 2021:147-62.

70. Choi S, Huang BC, Gamaldo CE. Therapeutic uses of cannabis on sleep disorders and related conditions. *J Clin Neurophysiol* 2020;37(1):39-49.

71. Lavender I, McGregor IS, Suraev A, et al. Cannabinoids, insomnia, and other sleep disorders. *Chest* 2022; 162(2), 452-465.

72. Edwards D, Filbey FM. Are sweet dreams made of these? understanding the relationship between sleep and cannabis use. *Cannabis Cannabinoid Res* 2021;6(6):462-73.

73. Belendiuk KA, Babson KA, Vandrey R, et al. Cannabis species and cannabinoid concentration preference among sleep-disturbed medicinal cannabis users. *Addict Behav* 2015;50:178-81.

74. Winiger EA, Hitchcock LN, Bryan AD, et al. Cannabis use and sleep: expectations, outcomes, and the role of age. *Addict Behav* 2021;112:106642.

75. Sholler DJ, Strickland JC, Spindle TR, et al. Sex differences in the acute effects of oral and vaporized cannabis among healthy adults. *Addict Biol* 2021;26(4):e12968.

76. Zhang B, Wing Y-K. Sex differences in insomnia: a meta-analysis. *Sleep* 2006;29(1):85-93.

77. Pagel J, Parnes BL. Medications for the treatment of sleep disorders: an overview. *Prim Care Companion J Clin Psychiatry* 2001;3(3):118.

78. Palagini L, Baglioni C, Ciapparelli A, et al. REM sleep dysregulation in depression: state of the art. *Sleep Med Rev* 2013;17(5):377-90.

79. Vogel GW, Vogel F, McAbee RS, et al. Improvement of depression by REM sleep deprivation: new findings and a theory. *Arch Gen Psychiatry* 1980;37(3):247-53.

80. Lintzeris N, Driels J, Elias N, et al. Medicinal cannabis in Australia, 2016: the cannabis as medicine survey (CAMS-16). *Med J Aust* 2018;209(5):211-16.

81. Lintzeris N, Mills L, Suraev A, et al. Medical cannabis use in the Australian community following introduction of legal access: the 2018–2019 Online Cross-Sectional Cannabis as Medicine Survey (CAMS-18). *Harm Reduct J* 2020;17(1):1-12.

82. Kosiba JD, Maisto SA, Ditre JW. Patient-reported use of medical cannabis for pain, anxiety, and depression symptoms: systematic review and meta-analysis. *Soc Sci Med* 2019;233:181-92.

83. Atkin T, Comai S, Gobbi G. Drugs for insomnia beyond benzodiazepines: pharmacology, clinical applications, and discovery. *Pharmacol Rev* 2018;70(2):197-245.

84. Bastien CH, LeBlanc M, Carrier J, et al. Sleep EEG power spectra, insomnia, and chronic use of benzodiazepines. *Sleep* 2003;26(3):313-17.

85. Baglioni C, Regen W, Teghen A, et al. Sleep changes in the disorder of insomnia: a meta-analysis of polysomnographic studies. *Sleep Med Rev* 2014;18(3):195-213.

86. Arnold JC, Nation T, McGregor IS. Prescribing medicinal cannabis. *Aust Prescr* 2020;43(5):152.

87. Jetly R, Heber A, Fraser G, et al. The efficacy of nabilone, a synthetic cannabinoid, in the treatment of PTSD-associated nightmares: a preliminary randomized, double-blind, placebo-controlled cross-over design study. *Psychoneuroendocrinology* 2015;51:585-88.

88. McGregor IS, Cairns EA, Abelev S, et al. Access to cannabidiol without a prescription: A cross-country comparison and analysis. *Int J Drug Policy* 2020;85:102935.

89. Rubin R. Cannabidiol products are everywhere, but should people be using them? *JAMA* 2019;322(22):2156-58.

90. Arnold JC, McCartney D, Suraev A, et al. The safety and efficacy of low oral doses of cannabidiol: An evaluation of the evidence. *Clin Transl Sci* 2022

91. ClinicalTrials.gov. Evaluation of an Oral Cannabidiol (CBD)-Terpene Formulation on Sleep Physiology in Participants With Insomnia. NCT05233761. Accessed 20 November, 2022. https://beta.clinicaltrials.gov/study/NCT05233761

92. Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, et al. The nonpsychoactive Cannabis constituent cannabidiol is a wake-inducing agent. *Behav Neurosci* 2008;122(6):1378.

93. Murillo-Rodríguez E, Millán-Aldaco D, Palomero-Rivero M, et al. Cannabidiol Partially Blocks the Excessive Sleepiness in Hypocretin-deficient Rats: Preliminary Data. *CNS Neurol Disord Drug Targets.* 2019;18(9):705-12.

94. Nasrin S, Watson CJ, Perez-Paramo YX, et al. Cannabinoid metabolites as inhibitors of major hepatic CYP450 enzymes, with implications for cannabis-drug interactions. *Drug Metab Dispos* 2021;49(12):1070-80.

95. Johnson E, Kilgore M, Babalonis S. Cannabidiol (CBD) product contamination: Quantitative analysis of Δ9-tetrahydrocannabinol (Δ9-THC) concentrations found in commercially available CBD products. *Drug Alcohol Depend* 2022:109522.

96. Vandrey R, Raber JC, Raber ME, et al. Cannabinoid dose and label accuracy in edible medical cannabis products. *JAMA* 2015;313(24):2491-93.

97. Australian Journal of General Practice. Insomnia management. 2019. Acessed 13 November, 2022. https://www1.racgp.org.au/ajgp/2019/april/insomnia-management

98. Arkell TR, Kevin RC, Stuart J, et al. Detection of Δ9 THC in oral fluid following vaporized cannabis with varied cannabidiol (CBD) content: An evaluation of two point-of-collection testing devices. *Drug Test Anal* 2019;11(10):1486-97.

99. Alcohol and Drug Foundation. Can I drive on medicinal cannabis products? Accessed 20 November, 2022. https://adf.org.au/insights/medicinal-cannabis-driving/

100. Lee D, Huestis MA. Current knowledge on cannabinoids in oral fluid. *Drug Test Anal* 2014;6(1-2):88-111.

101. Lee D, Karschner EL, Milman G, et al. Can oral fluid cannabinoid testing monitor medication compliance and/or cannabis smoking during oral THC and oromucosal Sativex administration? *Drug Alcohol Depend* 2013;130(1-3):68-76.

102. Arkell TR, Spindle TR, Kevin RC, et al. The failings of per se limits to detect cannabis-induced driving impairment: Results from a simulated driving study. *Traffic Inj Prev* 2021;22(2):102-07.

Appendix A

Supplementary Material for Chapter 2

Supplementary Table 1 Pre-registered clinical trials investigating the effect of cannabinoid therapies in the management of sleep disorders (accessed August 2022)

Primary sponsor [Citation]	Clinical trial registry	Country of origin	Patient population (target *n*)	Trial status, registry ID	Study design	Treatment period	Intervention and dose	Primary outcome(s)	Method
Woolcock Institute of Medical Research	ANZCTR	Australia	Insomnia (*n*=20)	Completed ACTRN12619000714189	DB, PC, WSD Crossover	Acute	'ETC120' Combined 10 mg THC with 200 mg CBD	TST and WASO (minutes) post-treatment compared to placebo	Overnight PSG
Woolcock Institute of Medical Research	ClinicalTrials.gov	Australia	Insomnia (*n*=20)	Currently recruiting NCT05344170	DB, PC, WSD Crossover	Acute	30 mg CBN 300 mg CBN	WASO post-treatment compared to placebo	Overnight PSG
BOD Australia	ClinicalTrials.gov	Australia	Insomnia (*n*=198)	Currently recruiting NCT05253417	DB, PC, WSD Parallel	8 weeks	50 mg CBD 100 mg CBD	Change in sleep quality	ISI
Southern Cross University	ANZCTR	Australia	Insomnia (*n*=438)	Active, not recruiting ACTRN12621000632897	DB, PC, WSD Parallel	8 weeks	Up to 150 mg CBD per day	Change in sleep quality	PROMIS
Swinburne University of Technology	ANZCTR	Australia	Insomnia (*n*=30)	Recruiting ACTRN12620000070932	DB, PC, BSD Parallel	2 weeks	200 mg CBD in corn oil	Insomnia symptoms, sleep latency and sleep efficiency (%) compared to placebo	ISI and standardised self-report questionnaires
Entoura Pty	ANZCTR	Australia	Insomnia (*n*=30)	Completed ACTRN12620000220965	DB, PC, WSD Crossover	2 weeks	Combined 15 mg THC and 22.5 mg CBD	Change in sleep quality	ISI
Defined Research	ClinicalTrials.gov	USA	Insomnia (*n*=125)	Active, not recruiting NCT05233761	DB, PC, WSD Crossover	4 weeks	"(CBD)-terpene" Combined 300 mg CBD and 8 mg terpenes	Time spent in SWS and REM sleep	Actigraphy
Cerebra Medical	ClinicalTrials.gov	Canada	'Poor sleep quality' (*n*=34)	Recruiting NCT05237037	Observational Prospective, cohort	6 weeks	'THC:CBD treatment' Dosage and ratio unknown	Change in sleep quality	PSQI
Radicle Science	ClinicalTrials.gov	USA	'Sleep disturbance' (*n*=300)	Active, not recruiting NCT05511818	SB, PC, BSD Parallel	4 weeks	'Cannabinoids' Unknown	Change in subjective sleep disturbance	PROMIS Short Form 8A
Therapix Biosciences Ltd (Tel Aviv, Israel)	ClinicalTrials.gov	Israel	OSA (*n*=30)	Unknown NCT03646552	UB, open label Comparison to baseline	4 weeks	'THX-110' 10 mg dronabinol (THC) and PEA	AHI index post-treatment compared to baseline	Overnight PSG
Cannvalate (Swinburne University of Technology)	ANZCTR	Australia	OSA (*n*=30)	Completed ACTRN12619001103156	DB, PC, BSD Parallel	6 weeks	'IHL-42X' 10 mg THC with 200mg mineral supplement	AHI index post-treatment compared to placebo	Overnight PSG

ANZCTR=Australian New Zealand Clinical Trials Registry; BSD=between-subjects design; CBD=cannabidiol; CBN=cannabinol; DB=double-blind; ISI=Insomnia Severity Index; MCT=medium-chain triglycerides; MDD=major depressive disorder; OSA=obstructive sleep apnea; PEA=palmitoylethanolamide; PC=placebo-controlled; PROMIS=Patient-Reported Outcomes Measurement Information System**;** PSG=polysomnography; PSQI= Pittsburgh Sleep Quality Index; SB=single blind; SOL=sleep onset latency; THC=tetrahydrocannabinol; TST=total sleep time; WASO=wake after sleep onset; WSD=within-subjects design; UB=unblinded

Appendix B

Supplementary Material for Chapter 3

Cannabis as Medicine Survey (CAMS) 2020-2021

Access to the full questionnaire is available for download via:

Lintzeris, N., Mills, L., Abelev, S. V., Suraev, A., Arnold, J. C., & McGregor, I. S. (2022). Medical cannabis use in Australia: consumer experiences from the online cannabis as medicine survey 2020 (CAMS-20). *Harm Reduction Journal*, *19*(1), 1-10.

https://harmreductionjournal.biomedcentral.com/articles/10.1186/s12954-022-00666-w

See 'Supplementary Information: Additional File 1'

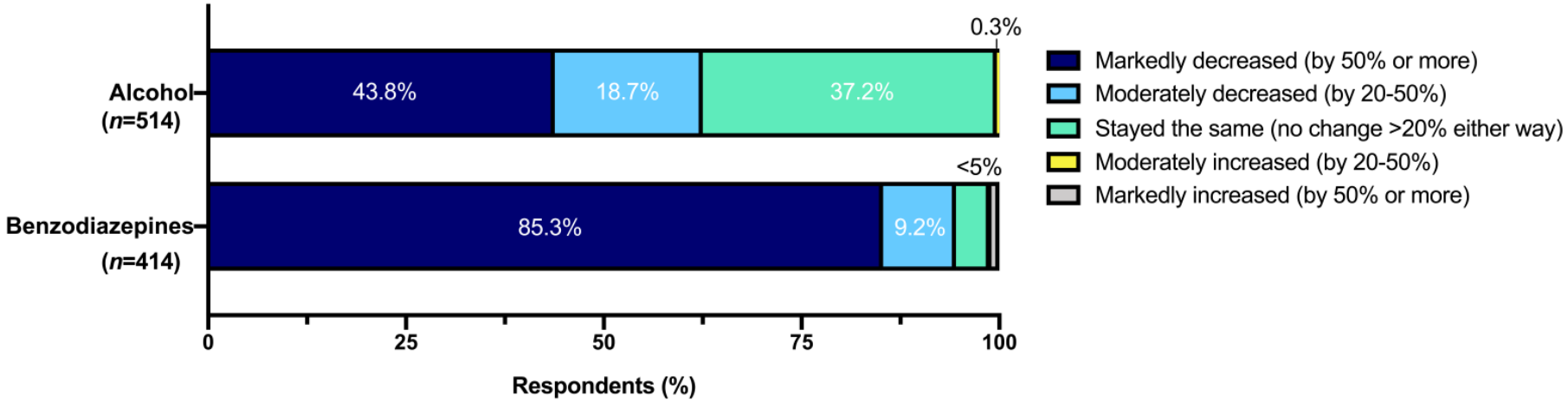

Figure S1 Self-reported change in alcohol (n=514) and benzodiazepine (n=414) use after commencing use of medical cannabis for the treatment of sleep disorders.

Table S1 Main conditions treated with medicinal cannabis by respondents who reported using medical cannabis to treat a sleep disorder as a general condition

	Rank	Condition	Respondents (n=982)
Pain		**Total**	**412 (42%)**
	1	Back pain	124 (12.6%)
	2	Arthritis	79 (8.0%)
	3	Fibromyalgia	64 (6.5%)
	4	Neuropathy	52 (5.3%)
		All others	93 (9.3%)
Mental Health/ Substance Use		**Total**	**319 (32.5%)**
	1	Anxiety	194 (19.8%)
	2	PTSD	54 (5.5%)
	3	Depression	46 (4.7%)
	4	ADHD	12 (1.2%)
		All others[b]	13 (1.3%)
Sleep		**Total**	**165 (16.8%)**
	1	Insomnia	116 (11.8%)
	2	Other[c]	22 (2.2%)
	3	Movement	12 (1.2%)
	4	Circadian	8 (0.8%)
		All others[b]	7 (0.7%)
Neurological		**Total**	**68 (6.9%)**
	1	Other[c]	30 (3.1%)
	2	MS	9 (0.9%)
	3	Epilepsy	8 (0.8%)
	4	Autism	5 (0.5%)
		All others[b]	16 (1.6%)
Gastrointestinal		**Total**	**22 (2.2%)**
	1	IBS	7 (0.7%)
	2	Other[c]	2 (0.2%)
	3	Crohn's	7 (0.7%)
	4	Ulc. colitis	6 (0.6%)
Cancer		**Total**	**23 (2.3%)**
	1	Blood	7 (0.7%)
	2	Brain	5 (0.5%)
	3	Gastro	4 (0.4%)
	4	Breast	3 (0.3%)
		All others[b]	4 (0.4%)
Other		**Total**	**55 (5.6%)**
	1	Gyn.	23 (2.3%)
	2	Other[c]	20 (2.0%)
	3	Immune	9 (0.9%)
	4	Diabetes	2 (0.2%)
		All others[b]	1 (0.1%)

***a**: percentages displayed represent the proportion each specific condition makes up of the entire group (i.e., 124 respondents reported 'back pain' as main condition, which represents 12.6% of the 982 respondents responded to this question) **b:** All others' refers to all the other specific conditions that were listed as a main condition, but which were not in the top 4 most commonly. **c**: 'Other' refers to other conditions that could be classed under the overall main condition, but which were not listed in the drop-down list of specific conditions (e.g., other neurological conditions not listed, other sleep conditions not listed). Note: Sum of respondents across all seven condition categories does not add up to n=982 due to 82 dual users choosing different main conditions for their prescribed and illicit medical cannabis product (e.g., main indication was 'pain' for prescribed product and 'sleep' for illicit product). ADHD = attentive deficit hyperactivity disorder; Circadian = Circadian rhythm disorder; Gyn. = gynaelogical condition; Immune = Auto-Immune condition; Movement = Sleep-related movement disorder; MS = Multiple sclerosis; PTSD= post-traumatic stress disorder; Ulc.Collitis = Ulcerative colitis.*

Appendix C

Supplementary Material for Chapter 4.1

Appendix 2 - SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	2
	2b	All items from the World Health Organization Trial Registration Data Set	Appendix 1
Protocol version	3	Date and version identifier	8
Funding	4	Sources and types of financial, material, and other support	26
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	26
	5b	Name and contact information for the trial sponsor	Appendix 1
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	20, 26

	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	18,19
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	4 - 7
	6b	Explanation for choice of comparators	9
Objectives	7	Specific objectives or hypotheses	10, 11
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	7, 8
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	8
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	21,22 (Table 1)
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	13, 14
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	n/a
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	13

	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	12
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	10, 11
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	11-18 & 23,24 (Table 2) & Fig 1
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	19,20
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	8
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	9,10
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	10
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	10
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	9,10,13
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	10

Methods: Data collection, management, and analysis

Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	11 - 18
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	n/a
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	18, 19
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	19, 20
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	19, 20
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	n/a

Methods: Monitoring

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	18, 19
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	19
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	19

Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	19
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	20
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	20
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	9
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	n/a
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	18, 19
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	26
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	18, 19
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	n/a
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	20, 21
	31b	Authorship eligibility guidelines and any intended use of professional writers	n/a
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	20, 21

Appendices

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	n/a
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	n/a

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

Supplementary File 2 World Health Organisation Trial Registration Data Set	
Primary registry and trial identifying number	Australian and New Zealand Clinical Trials Registry (ANZCTRN12619000714189)
Secondary identifying numbers	UTN: U1111-1231-0849
Date of registration in primary registry	13 May, 2019
Source(s) of monetary or material support	The Lambert Initiative for Cannabinoid Therapeutics, The University of Sydney
Primary sponsor	The Woolcock Institute of Medical Research 431 Glebe Point Road Glebe NSW 2037 Australia
Contact for public queries	Miss Anastasia Suraev (cansleep@woolcock.org.au)
Contact for scientific queries	Dr Camilla Hoyos (camilla.hoyos@sydney.edu.au)
Public title	A single-dose, double-blind, placebo-controlled, randomised, crossover study of an oral cannabis-based medicine (ETC120) on sleep, cognition, and next-day function in adults with chronic insomnia disorder
Scientific title	A single-dose, double-blind, placebo-controlled, randomised, crossover study of an oral cannabis-based medicine (ETC120) on sleep quality and quantity in adults with chronic insomnia disorder
Countries of recruitment	Australia
Health condition(s) or problem(s) studied	Insomnia disorder
Intervention(s)	*Active comparator*: oral solution containing 10 mg Δ^9-tetrahydrocannabinol (THC) and 200 mg cannabidiol (CBD) in medium-chain triglycerides (MCT) oil
	Placebo comparator: matching oil solution containing no active ingredients
Key inclusion and exclusion criteria	*Ages eligible for study*: 35 to 60 years inclusive *Sexes eligible for study*: Both *Accepts healthy volunteers*: No
	Inclusion criteria: Adult patient (35 to 60 years) diagnosed with chronic insomnia disorder
	Exclusion criteria: Shift worker, medical condition or medication that is the cause of the insomnia (including other sleep disorder), use of any modality of treatment for insomnia including CBT in the past 3 months, history of drug and alcohol abuse/dependency, history of major psychiatric disorder except clinically managed depression.
Study type	Interventional *Allocation*: randomized controlled trial. Crossover: double blind (participant, investigator, outcomes assessor) *Primary purpose*: Treatment Phase I/Phase 2
Date of first enrolment	August 2019
Target sample size	20
Recruitment status	Recruiting
Primary outcome(s)	Change in total sleep time (TST) and wake after sleep onset (WASO) measured in minutes from in-laboratory overnight PSG
Key secondary outcomes	Sleep microarchitecture metric measured using high-density EEG and source modelling; next-day neurobehavioural functioning (including cognition, alertness and simulated driving performance)

Appendix D

Supplementary Material for Chapter 4.2

Table S1 Breakdown of REM sleep (%) into tertiles

	REM sleep % (SD)			
	CBD/THC (*n*=20)	Placebo (*n*=20)	*p* value	Cohen's *d* [95% CI]
Tertiles				
1	2.4 (4.5)	7.0 (7.5)	.028	-0.55 [-1.02, -0.08]
2	11.9 (9.8)	21.4 (9.1)	.005	-0.69 [-1.18, -0.21]
3	24.4 (11.7)	35.8 (11.1)	.001	-0.81 [-1.31, -0.30]

Table S2 The median [IQR] length of time between CBD/THC use and blood sampling (washout; days) and the proportion (%) of participants with detectable concentrations of CBD, THC, and their major phase-I metabolites in plasma ≥7 days.

	Treatment Order 2 (CBD/THC → placebo) (*n*=10)
Washout[a]	12.0 [38.5]
CBD	6/10 (60%)
7-COOH-CBD	7/10 (70%)
7-OH-CBD	4/10 (40%)
6-OH-CBD	0
THC	0
11-COOH-THC	4/10 (40%)
11-OH-THC	5/10 (50%)

a: Median across all participants.

Table S3 The median (IQR) concentration (ng/mL) of CBD, THC, and their major phase-I metabolites in plasma ≥7 days for each treatment order.

	Treatment Order 1 (*n*=10)		**Treatment Order 2 (*n*=10)**	
	[1] Placebo	[2] CBD/THC	[1] CBD/THC	[2] Placebo
CBD	0 (0)	0 (0)	14.8 (12.5)	1.2 (2.1)
7-COOH-CBD	0 (0)	918.9 (532.7)	1331.6 (956.7)	10.9 (22.8)
7-OH-CBD	0 (0)	12.4 (4.5)	13.7 (13.0)	0.0 (1.9)
6-OH-CBD	0 (0)	0 (0)	0.0 (0.0)	0.0 (0.0)
THC	0 (0)	0 (0)	0.0 (0.0)	0.0 (0.0)
11-COOH-THC	0 (0)	23.1 (19.4)	24.5 (14.5)	0.0 (8.7)
11-OH-THC	0 (0)	1.7 (2.1)	2.46 (2.5)	0.3 (0.6)

Values are Median (IQR). Blood was collected 10.5 h post-drug administration (the morning after) on each treatment session. Median (IQR) washout period was 12.0 (38.5).

Table S4 Assessment of blinding success

	Participant's guess, *n* (%)			
Intervention	**Active**	**Placebo**	**Not sure**	**Total**
Active	14 (70%)	6 (30%)	0	20
Placebo	4 (30%)	12 (60%)	2 (10%)	20

Appendix E

Supplementary Material for Chapter 4.3

Plasma Cannabinoid Analysis

Chemicals and reagents

Acetonitrile, formic acid, methanol, dichloromethane, and methyl-*tert*-butyl ether were obtained from Fisher Scientific (Melbourne, VIC, Australia). Cannabinoid reference standards and deuterated internal standards were purchased from Cerilliant (Round Rock, TX, USA). All chemicals and solvents were at least American Chemical Society (ACS) or high-performance liquid chromatography grade, respectively.

Liquid chromatography-tandem mass spectrometry (LC-MS/MS) analysis

Cannabinoid analysis was performed as reported previously (Kevin et al., 2017) with minor modification. 200 µL of plasma samples were aliquoted in triplicate and spiked with a mixture of cannabinoid internal standards (THC-*d*3, CBD-*d3*, 11-OH-THC-*d3*, and THC-COOH-*d*3) in methanol. Calibrator and quality control samples of known cannabinoid concentrations were prepared by addition of reference standards to cannabinoid-free plasma which were treated identically to participant samples. 600 µL ice cold acetonitrile was added to all samples to precipitate protein, and the samples were centrifuged at 6000 x *g* for 10 min at 4 °C. The resultant supernatant was decanted into 96 well plates and dried under nitrogen.

The samples were reconstituted in 90 µL acetonitrile and 300 µL 0.1% formic acid and water and extracted using supported liquid extraction. The sample solutions were absorbed on Biotage Isolute SLE+ 400 µL capacity 96 well plates (Rydalmere, NSW, Australia), and the analytes were eluted with 700 µL dicholoromethane and 900 µL methyl-*tert*-butyl ether into a clean 96 well plate. The eluate was immediately evaporated to dryness under a gentle stream of nitrogen and reconstituted in 100 µL of 40:60 0.1% formic acid and methanol for immediate analysis via LC-MS/MS.

Cannabinoid quantification was performed using a Shimadzu Nexera LC-30AD ultra-high-performance liquid chromatograph (Shimadzu Corp., Kyoto, Japan) coupled to a Shimadzu LCMS-8040 triple quadrupole mass spectrometer. 20 µL injections of each sample, kept in an 8 °C autosampler, were chromatographically separated using an Agilent Zorbax XDB-C18 reverse-phased analytical column (50 x 2.1 mm i.d., particle size 3.5 µm; CA, USA). This was performed via gradient elution with 0.1% formic acid in water and methanol at a flow rate of 0.6 mL/min. The mass spectrometer was operated in positive electrospray ionization mode with multiple reaction monitoring to identify and quantify analytes against 7-point standard curves.

Oral fluid analysis via liquid chromatography with tandem mass spectrometry (LC-MS/MS)

Oral fluid samples were analyzed using LC–MS/MS. Duplicate 1 mL aliquots were fortified with an internal standard mixture containing *d*3- THC and *d*3-CBD. Duplicate calibrator samples were prepared using cannabinoid-free saliva (obtained from healthy volunteers using Quantisal™ collection devices and checked for cannabinoid content via LC–MS/MS), spiked with THC, CBD, and internal standards to generate a standard curve for each analyte and quality control samples. THC and CBD were isolated using supported liquid extraction (SLE), where each sample aliquot was absorbed onto a 1 mL capacity ISOLUTE® SLE+ column (Biotage, Sydney, Australia), and analytes were eluted with 1.6 mL DCM, 3.5 mL methyl *tert*-butyl ether (MTBE), and 1.6 mL 1:5 ethyl acetate and MTBE. The eluate was evaporated without heating under a gentle stream of nitrogen, and analytes were reconstituted in 200 µL of 1:1 acetonitrile and 0.1% formic acid in water, transferred to 2 mL autosampler vials fitted with 200 µL capacity glass inserts, and placed in the LC–MS/MS autosampler held at 4°C.

Chromatographic separation was achieved using an Eclipse XDB- C18 column (50 mm x 2.1 mm i.d., particle size 3.5 µm; Agilent Technologies, Singapore) using gradient elution with mobile phases 0.1% formic acid in water and acetonitrile, at a flow rate of 0.3 mL/min. This was coupled to a Shimadzu LCMS-8030 mass spectrometer for analyte identification and quantification.

The LC–MS/MS analysis was validated for selectivity, linearity, accuracy, precision, bench-top and autosampler stability, dilution integrity, limit of detection (LOD), and limit of quantification (LOQ) . (Table 2), following Food and Drug Administration (FDA) validation guidelines. Selectivity was verified by analyzing cannabinoid-free saliva samples for interferences. Linearity was assessed using calibrators at seven ascending concentration levels. Intra-assay accuracy and precision were determined using six replicate quality control (QC) samples at low, medium, and high concentrations relative to the concentration range on the same day. Inter-assay accuracy and precision were determined using similar QC samples three different days (three replicates per day). Repeat injections at 0-, 4-, and 8-hour timepoints were used to assess autosampler stability. Dilution integrity was assessed for 10x dilutions. The lower limit of quantification (LLOQ) was selected based on accuracy of calibrator samples (lowest calibrator within ±20% of the nominal value), while the LOD was set as the lowest calibrator concentration with signal-to-noise greater than 3. Samples that fell above the linear quantification range were diluted appropriately and re-analyzed.

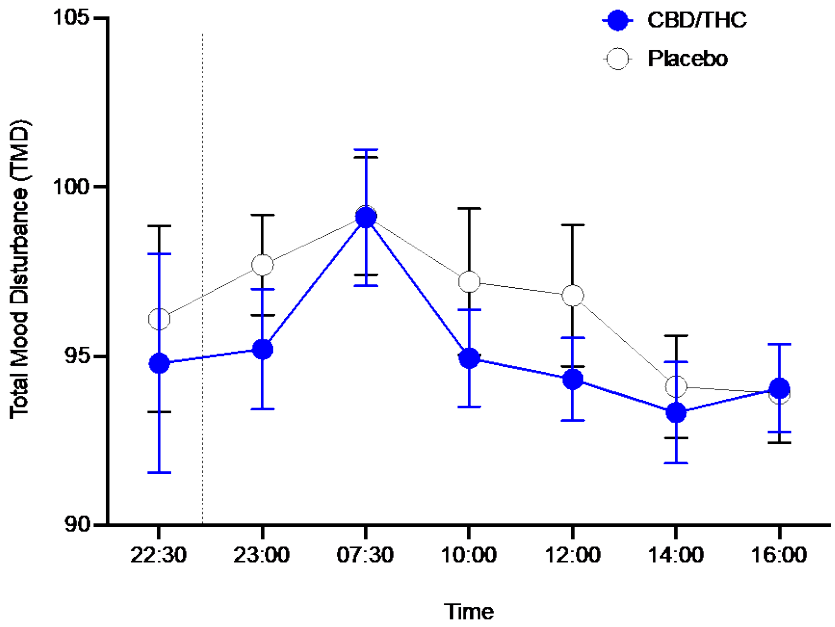

Figure S1 Total mood disturbance (TMD) as measured on the Profile of Mood States (POMS) questionnaire assessed immediately prior to sleep (~22:30), 0.5 h post drug-administration, upon waking (~07:30), and prior to the start of each Maintenance of Wakefulness Test trial at 10:00, 12:00, 14:00, and 16:00. Dotted line indicates timing of drug administration.

Table S1 Performance characteristics of the Securetec DrugWipe® 5 s (DW5s) and Dräger DrugTest® 5000 (DT5000) POCT devices when verified against LC–MS/MS quantified oral fluid THC concentrations using a 10 ng/mL confirmatory cut-off

Device	Cut-off	Time relative to drug administration (min)	*N* of tests	True positives	True negatives	False positives	False negatives	Sensitivity (%)	Specificity (%)	Accuracy (%)
DW5s	2 ng/mL	- 0.5 h	34	0	33	1	0	-*	97	97
		+ 0.5 h	34	2	20	1	11	15	95	65
		+ 10 h	34	0	26	0	8	-*	100	76
		+ 18 h	34	0	33	0	1	-*	100	97
		Total	136	2	112	2	20	9	98	84
	1 ng/mL	- 0.5 h	34	0	33	1	0	-*	97	97
		+ 0.5 h	34	3	20	0	11	21	100	68
		+ 10 h	34	0	25	0	9	0	100	74
		+ 18 h	34	0	33	0	1	0	100	97
		Total	136	3	111	1	21	13	99	84
DT5000	2 ng/mL	- 0.5 h	32	0	32	0	0	-*	100	100
		+ 0.5 h	31	3	20	0	8	27	100	74
		+ 10 h	32	0	24	0	8	0	100	75
		+ 18 h	32	0	31	0	1	0	100	97
		Total	127	3	107	0	17	15	100	87
	1 ng/mL	- 0.5 h	32	0	32	0	0	-*	100	100
		+ 0.5 h	31	3	19	0	9	25	100	71
		+ 10 h	32	0	23	0	9	0	100	72
		+ 18 h	32	0	31	0	1	0	100	97
		Total	127	3	105	0	19	14	100	85

-* Sensitivity could not be ascertained as there were no true positives.