2019

Evaluation of the impact of a low dose subcutaneous lignocaine and ketamine infusion utilising nerve excitability studies in a chronic migraine population.

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Evaluation of the impact of a low dose subcutaneous lignocaine and ketamine infusion utilising nerve excitability studies in a chronic migraine population.

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Bachelor of Science (Pharmacology)

Thesis submitted in accordance with the requirements for

Masters of Research (Medicine)

School of Medicine

Sydney Campus

August 2019

Acknowledgements

This work would not have been possible with the support and encouragement of many people.

I have had the privilege to be mentored by Associate Professor Ray Garrick and Associate Professor Susan Tomlinson over the past three years. They have provided valuable insights into all areas of this thesis. I sincerely thank them.

Dr James Howells provided invaluable assistance in modelling and analysis of nerve excitability studies.

I would like to acknowledge the support and funding that this project has received from both the St Vincent Clinic Foundation and the Brain Foundation.

I would also like to acknowledge the funding received from the Australian government of my student candidature under the RTP scheme.

Declaration

To the best of the candidate's knowledge, this thesis contains no material previously published by another person, except where due acknowledgement has been made.

This thesis is the candidate's own work and contains no material which has been accepted for the award of any other degree or diploma in any institution.

The research presented and reported in this thesis was conducted in accordance with the National Health and Medical Research Council National Statement on Ethical Conduct in Human Research (2007, updated 2018). The proposed research study received human research ethics approval from the University of Notre Dame Australia Human Research Ethics Committee (EC00418), Approval Number 017044S.

Signature:

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Date:

Abstract

Migraine is a common condition in which the diagnosis is based on clinical grounds. There is no clinically available biophysical marker that can evaluate migraine. Migraines are linked to functional brain changes in the absence of structural abnormalities. A clinically useful tool capable of evaluating functional changes in patients with migraine could be used to aid diagnosis and management.

Patients with chronic migraine have frequent or continuous headache which is accompanied by significant morbidity. There are limited data available regarding treatment options for curtailment of chronic migraine.

In this prospective observational study, patients suffering from chronic migraine underwent a prolonged subcutaneous lignocaine and ketamine infusion which has anecdotally been useful in management of chronic migraine. To determine if peripheral nerve excitability studies have a role in assessing patients with chronic migraine and their response to treatment, these studies were performed on patients before, during and after the infusion and at six months and compared to healthy age matched controls.

Most patients (13/14) had significant clinical benefit from the infusion. No changes in excitability studies were identified in patients at baseline, during or after intervention with low-dose lignocaine/ketamine infusion. The lack of detectable change in excitability measurements despite significant clinical improvement resulting from the infusion may implicate a central mechanism of action of the infusion.

Abbreviations

5HT	Serotonin
AP	Action potential
APB	Abductor pollicis brevis
ARP	Absolute refractory period
CM	Chronic Migraine
CNS	Central nervous system
CSD	Cortical spreading depression
CV	Conduction velocity
E_K	Potassium equilibrium potential
E_{Na}	Sodium equilibrium potential
FMH	Familial hemiplegic migraine
FASPS	Familial advanced sleep phase syndrome
K^+	Potassium ion
HCN	Hyperpolarisation-activated cyclic nucleotide-gated
I/V	Current threshold relationship
I_h	Inward rectification
IHS	International Headache Society
ICHD-3	International Classification of Headache Disorders Version 3
K^+	Potassium ion

MA	Migraine with aura
MO	Migraine without aura
Na^+	Sodium ion
Na_p	Persistent sodium channel
NCS	Nerve conduction studies
NDPH	New daily-persisting headache
NMDA	N-methyl-D-aspartate
NES	Nerve excitability studies
NSAIDs	Non-steroidal anti-inflammatory drug
RC	Recover cycle
RRP	Relative refractory period
SDTC	Strength duration time constant
SR	Stimulus response
TEd	Depolarising threshold electrotonus
TEh	hyperpolarising threshold electrotonus
TTH	Tension-type headache

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CHAPTER 1

1.1 Headache

Headache is a general term for the sensation of pain in the head region. The location of pain can vary greatly between individuals, with some headaches being isolated to certain regions while others are bilateral across the head. A headache may be described as a sharp pain, a throbbing sensation or a dull ache. Headaches can develop gradually or suddenly with the duration varying from less than an hour to several days.

Headache is a symptom, rather than a diagnosis. The clinical symptoms allow identification of underlying cause and direct treatment. Hence, an understanding of how underlying headache patterns are classified is imperative for patient care. While additional neurological tests may assist in the exclusion of some pathologies, headache requires clinical interpretation.

Different headache patterns may co-exist concurrently within an individual. For example it is common for frequent episodic tension-type headache to coexist with migraine without aura. It can be difficult to differentiate between some headache disorders.

The International Classification of Headache Disorders (ICHD-III) classifies headaches into either primary or secondary based on the pathophysiology (Headache classification Committee, 2018). The classification of the 14 types of headache is summarized in table 1.1.

A primary headache refers to a disorder generated by primary pathophysiology affecting the cranial structures which is not caused by other medical conditions. Secondary headache is the term given to headaches in which an underlying cause is found such as trauma, tumour, infection and metabolic disorders.

This thesis focuses on primary headache disorders, specifically those patients who have developed chronic migraine (CM) with or without medication overuse. This focus reflects the common presentation of migraine and CM patients in neurological practice.

Table 1.1: Summary of IHS Classification of headaches*

Primary Headache	Secondary Headaches	Other
• Migraine • Tension type headache (TTH) • Trigeminal autonomic cephalalgias • Other Primary headache disorders	• Headache attributed to trauma or injury • Headache attributed to cranial or cervical vascular disorder • Headache attributed to non-vascular intracranial disorder • Headache attributed to a substance or its withdrawal • Headache attributed to infection • Headache attributed to disorder of homoeostasis • Headache or facial pain attributed to disorder of the cranium/neck/eyes/ears/nose/ sinuses/ teeth/ mouth or other facial or cervical structure • Headache attributed to psychiatric disorder	• Cranial neuropathies, other facial pains • other headaches

*Headache classification Committee, 2018

1.1.1 Migraine diagnosis

Migraine is a recurrent disorder characterised by moderate to severe episodic headaches. Typical features are lateralised headache, 4-72 hours duration, and pulsating nature, aggravation by routine physical activity, generally associated with nausea, photophobia and phonophobia.

Migraine may be associated with aura (MA), which is a transient phenomenon of disturbed sensory perception. This may occur without headache (acephalic migraine). Aura symptoms are fully reversible symptoms and may include alterations to visual, sensory, speech, motor, brainstem and retinal vision. Up to 38% of patients with migraine have attacks with aura which usually occur before pain phase of the headache and may continue into the pain phase (Kelman, 2004). Visual aura is most common, occurring in over 90% of migraine with aura individuals.

Table 1.2 Diagnosis criteria for migraine per ICHD-III criteria *

	Migraine without Aura (MO)	Migraine with Aura
Diagnostic Criteria	1. At least 5 attacks meeting the criteria 2-4 2. Headache lasts between 4-72 hours 3. Headache has at least two of the following: - Lateralised - Pulsating quality - Moderate or severe pain - Aggravation by or avoidance of routine activity 4. During headache there is at least one of following - Nausea and or vomiting - Photophobia and phonophobia 5. Not better accounted by another ICHD-3 diagnosis	1. At least 2 attacks meeting the criteria 2-3 2. One or more of the following reversible changes in aura symptoms: - Visual - Sensory - Speech and or language - Motor - Brainstem - Retinal 3. At least three of the following aura symptom: - at least one aura symptom spreads gradually over ≥ 5 minutes - two or more aura symptoms occur in succession - each individual aura symptom lasts 5-60 minutes - at least one aura symptom is unilateral - at least one aura symptom is positive - the aura is accompanied, or followed within 60 minutes, by headache 4. Not better accounted by another ICHD-3 diagnosis

*Adapted from International Classification of Headache Disorders III, Headache Classification Committee, 2018

1.1.2 Medication overuse headache diagnosis

Medication overuse headache (MOH), previously known as “analgesic rebound” headache, is a recurring headache induced by repetitive and chronic overuse of acute headache medication. It is perpetuated by the frequent use of short acting analgesics where headache will develop after a short predicable time as medication levels fall.

MOH may escalate as a vicious cycle and develop when analgesics are taken an increasing frequency to alleviate the increased headache frequency.

The diagnostic criteria have updated in ICHD III definitions to be i) headaches that occurs at least 15 days per month in individuals with a pre-existing headache disorder while ii) regularly overusing medication for at least three consecutive months. ICHD III criteria brings MOH criteria into alignment with CM criteria and reflect the common dual clinical presentation.

MOH is typically seen in migraine and tension type headache (TTH) patients who use triptans, ergots, opioids and other analgesics where intake occurs on 10 or more days per month. Triptans tend to produce MOH more rapidly than either ergots or analgesics.

Management of the rebound cycle requires removal of the offending medication and withdrawal symptoms occurs with varying severity.

1.1.3 Chronic migraine

Chronic migraine (CM) also referred to as “transformed” migraine is defined by experiencing at least 15 headache days per month, which at least eight meet migraine diagnosis (Section 1.1.1), for at least three consecutive months.

In most cases, patients with CM have a history of occasional primary headache, increasing in frequency over months to years. This is common in MOH patients where overusing pain medications is a common behaviour in patients with CM. A patient can be classified as having CM together with MOH.

This sub-population of headache patients has a greater burden of disease and may be more refractory to conventional care, compared with other headache patients. The additional diagnosis

of CM in patients with MO or MA is important as it may reflect underlying pathophysiological changes.

1.1.4 Migraine epidemiology

Migraine is considered to be the world's third most prevalent disorder (Vos *et al*., 2010). Prevalence rates differ with age, gender and ethnicity (Bigal *et al*., 2010; Bigal *et al*., 2006).

Prevalence

The prevalence of migraine is assumed to be relatively stable over the last 3 decades (Table 1.3).

Table 1.3 Comparison of three largest US migraine populations over time

Study	Year	Migraine Prevalence % (Male/Female)
American Migraine Study (Steward *et al*., 1992)	1989	12.1 (5.7/ 17.6)
American Migraine Study II (Lipton *et al*., 2001)	1999	12.6 (6.5/ 18.2)
American Migraine Prevalence and Prevention (Lipton *et al*., 2007)	2004	11.7 (5.6/ 17.1)

Age

The prevalence of migraine changes with age. Migraine occurs in 3–10 % of pre-pubertal children, and the rates are similar among boys and girls. During adulthood prevalence increases to 11-13% with peak prevalence in both genders in the 30–39 age bracket. Prevalence declines in postmenopausal women. (Figure 1.1.)

Symptom patterns may vary with age and individuals under 18 years present with more bilateral pain than adults.

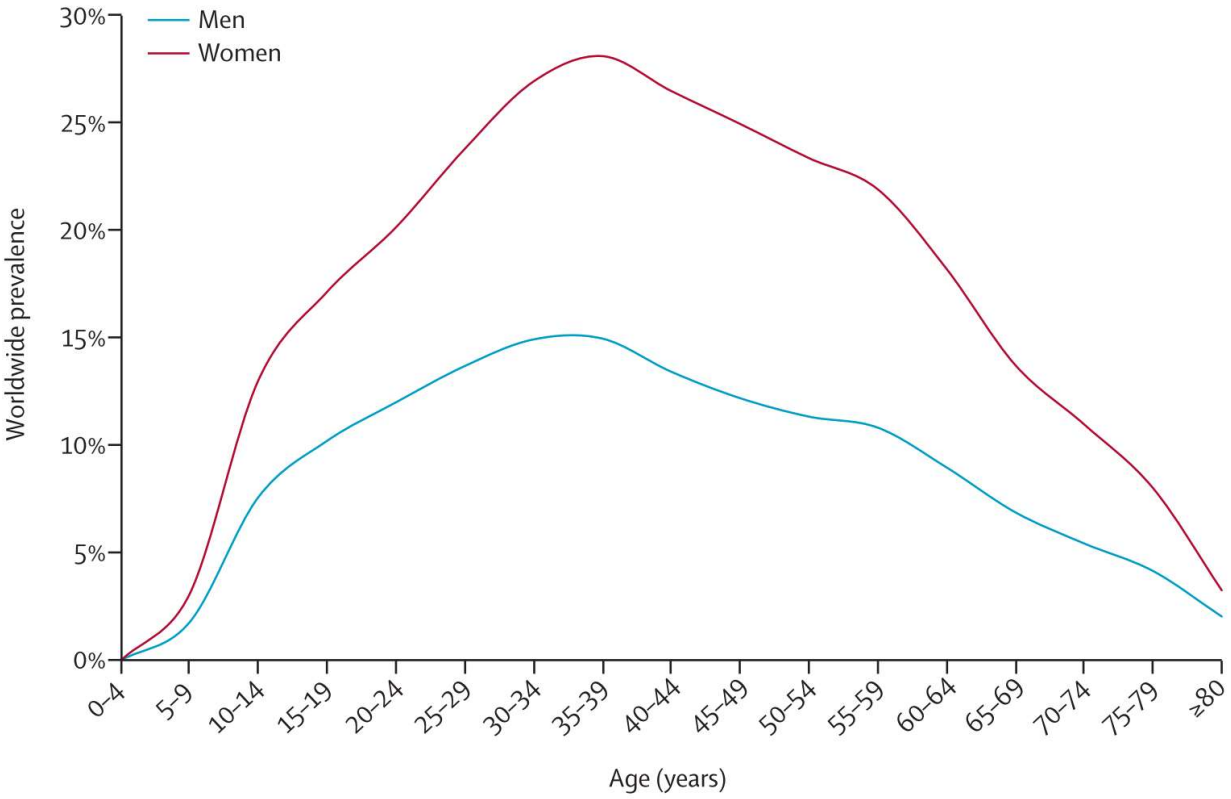
Gender

Worldwide prevalence data from the 2015 Global Burden of Disease Study show that migraine affects close to three times as many adult women (15–17 %) as adult men (6 %) with a strong correlation between childbearing age and prevalence (Global Burden of Disease, 2015).

Migraine severity is also affected by gender with women more likely to experience more intense migraine than men.

Figure 1.1 Global burden of disease 2015 point prevalence of migraine in men and women.

(Image adapted from Vetvik KG and MacGregor EA, 2016)



Ethnicity and Genetics

There are limited studies directly comparing ethnicity. However, there are strong genetic links in migraine. Approximately 70% of migraine patients have a first-degree relative with a history of migraine (Kors *et al*., 1999). The risk of migraine is increased 4-fold in relatives of people who have migraine with aura.

Table 1.4 Comparison of migraine point prevalence by region (Table adapted from Stovner et al., 2007)

Region	Overall Prevalence % (Number of studies)	Male Prevalence % (Number of studies)	Female Prevalence % (Number of studies)
Africa	5 (5)	3 (4)	6 (4)
Asia	9 (8)	6 (8)	11 (8)
Europe	15 (14)	7 (13)	18 (14)
North America	13 (9)	6 (7)	18 (7)
Central/ South America	9 (10)	4 (10)	12 (10)
Global	11 (41)	6 (41)	14 (43)

Incidence.

The American Migraine Prevalence and Prevention (AMPP) study estimated an overall migraine incidence of 8.1 per 1000 person–years (Lipton *et al*., 2001). A European study showed a peak incidence at 20- to 24-years in females (18.2 per 1000 person–years), and at 15- to 19-years in males (6.2 per 1000 person–years) (Lyngberg *et al*., 2005).

The number of new cases per year declines with age after a peak at 25- to 34-year-old females at 23 per 1000 person–years, and in males at about 10 per 1000 person–years. In the 55–64 years of age group, the incidence was less than 5 per 1000 person–years (Lipton *et al*., 2001).

Migraine severity is greater in patients with more frequent episodes. The AMPP and CaMEO studies have shown similar incidence and prevalence data comparing 2004 and 2014 (Lipton *et al*., 2016).

Essentially, incidence changes with age, incidence is similar between US and Europe and incidence rates have been stable over the last 20 years.

Economic burden/cost of disease

It has been reported that 90% of migraineurs have some headache-related disability, and approximately half become severely disabled or require bed rest during an event (Global Burden of Disease, 2015). Migraine can affect an individual's social, personal and professional

performance. There are also large direct costs to health system with the cost of medication and a significant investment of health care professional time to treat migraine.

Global Burden of Disease studies have classified migraine as the sixth highest cause of worldwide years lost due to disability recent studies have indicated migraine is the third cause of disability in under 50s (Global Burden of Disease, 2015). This estimates that migraine may reduce health-related quality of life to a similar degree as osteoarthritis or diabetes. The effects are augmented because migraine effects are greater during the most productive years of life (Steiner *et al*., 2016; Steiner *et al*., 2018).

In 2016, the economic burden of migraine in the US was estimated to an annual per-person cost of US $2649 for episodic migraine largely from absenteeism, decreased productivity and the cost of treatment (Messali *et al*., 2016). The indirect cost of migraine to US employers is estimated at $13 billion annually. These may be underestimates since they do not consider unemployment or underemployment related to migraine.

Socioeconomic Effects of migraine.

Some studies have shown an inverse relationship between the prevalence of migraine and socioeconomic status (measured by income or education). Stewart *et al*. (2013) reported a higher incidence in lower household income groups. However, other studies conflicting results and no clear consensus has been reached (Lipton *et al*., 2002; Buse *et al*., 2012). These differences may be a consequence of the barriers to good medical care in lower household income groups (defined as medical consultation, accurate diagnosis and appropriate pharmacological treatment). As migraine is undiagnosed or self-diagnosed and is largely self-treated. The barriers to good medical care may be larger in lower household income groups.

Comorbidity

Migraine is associated with multiple disease states and summarised in Table 1.5.

Table 1.5 Comorbidities of migraine (Adapted from Wang *et al*., 2010)

Epilepsy	Ischemic stroke
Chronic non headache pain	Coronary heart disease
Patent foramen ovale	Asthma/allergy
Mitral valve prolapse	Systemic lupus erythematosus
Sleep apnoea	Restless legs syndrome
Raynaud's phenomenon	Sub-clinical vascular brain lesions
Psychiatric diseases (depression, anxiety, bipolar disorder, panic disorder, and suicide)	Tourette syndrome

1.1.5 Migraine biomarker

There is no clinically useful migraine biomarker. This a challenge for clinicians as the sensation of pain associated with migraines is subjective.

1.2 Migraine Physiology

Migraine initiation probably depends upon a complex relationship between genetic, environmental, cognitive and emotive factors. The core underlying dysfunctions that ignite migraine attacks probably involves both neuronal and vascular components including the cerebral cortex, the brainstem, the thalamus and the peripheral and central components of the trigemino-cervicovascular complex. Functional MRI and PET scans have demonstrated that that the hypothalamus, the midbrain ventral tegmental area and the periaqueductal gray (PAG) are activated in migraineurs even in the absence of pain (Schulte *et al*., 2017; Schulte *et al*., 2016). The relative importance and the exact sequence of activation of these structures during a migraine attack are not fully understood and are under investigation.

There are likely pathophysiological differences between headache subtypes with peripheral pain mechanisms associated with episodic subtypes and central mechanism associated with the formation of chronic patterns. Structural changes including reduced gray matter in pain circuits have been reported in headache patients especially in the anterior cingulate, amygdala and operculum (Goadsby *et al*., 2017; Jia Z and Yu S, 2017; Goadsby PJ, 2015). Increased cortical thickness for somatotopical representation of the head and face in the cortex has been noted in high frequency chronic migraineurs compared to controls suggesting alterations in cellular structure which may render cortical cells more excitable. This increase in cortical thickness in migraine may result from a plastic reaction to repetitive pain processing (Hadjikhani N, 2008; Spenger T and Borsook D, 2012; Da Silva *et al*., 2007).

Functional MRI studies have identified significant hypothalamic involvement in the aura and acute pain phases of migraine. May (2017) identified a particular patient who was scanned on a daily basis over a month to monitor three spontaneous untreated headache attacks. He demonstrated hypothalamic activation in the prodromal phase (up to 24 hours before the onset of headache) compared with the interictal state. Pain related hypothalamic functional connections between the hypothalamus and the spinal trigeminal nuclei was significantly increased in the prodromal phase, strongly suggesting that the hypothalamus plays a generating role in the development of migraine symptoms.

The following section will outline four theories: neurotransmitter, neurovascular, cortical spreading depression and vascular.

1.2.1 Neurotransmitter hypotheses

This theory suggests that migraine originates from altered processing and release of neurotransmitters. Implicated in the pathogenesis of migraine are substance P, neurokinin A, calcitonin gene-related peptide, serotonin and nitric oxide which interact with the blood vessel wall to produce dilation, protein extravasation, and inflammation. Plasma extravasation may not be sufficient by itself to cause pain, in the presence of other stimulators it may be a critical step in migraine development.

Some medications that are effective for migraine inhibit neurogenic plasma extravasation, however, substance P antagonists and the endothelin antagonist bosentan inhibit neurogenic plasma extravasation but are ineffective as anti-migraine drugs. As well as activation of nociceptors in pain-producing intracranial structures the pain process also requires a reduction in the normal functioning of endogenous pain-control gate pathways.

1.2.2 Neurovascular theory

This theory postulates that migraine is primarily a neurogenic process where the release of neuropeptides from trigeminal nerve activation generates inflammation and pain. This produces sensitisation of primary afferent neurons that innervate the cranial meninges that further increases susceptibility to a future attack. This differs from previous theories where cranial vasodilation is a result of activation of trigeminal nerves and not the cause.

Pain associated with migraine is thought to be a result of the activation of the trigeminovascular system that consists of the neurons innervating the cerebral vessels whose cell bodies are located in the trigeminal ganglion. This system makes synaptic connections particularly with pain-producing large cranial vessels and dura and centrally projecting fibres synapsing on neurons in the caudal brain stem and high cervical cord. This will mediate the release of vasoactive peptides during a headache to activate pain pathways through a relay in the trigeminocervical complex. This produces the severe and throbbing nature of pain.

Transcranial magnetic stimulation and functional magnetic resonance imaging of the migraine patients at baseline confirms cortical activation as migraine evolves. This observation may explain the susceptibility of the migrainous brain to headache. Stimulation of the greater occipital nerve also causes neuronal activation in the same regions and enhances convergent inputs from the dural vasculature (Strassman *et al*., 1996).

It is suggested dysfunction of sensory processing plays a pivotal role for increased perception of pain and may explain the associated autonomic symptoms via ascending and descending pathways in the brain.

1.2.3 Cortical spreading depression

Cortical spreading depression (CSD) is a generally accepted theory to explain migraine aura. CSD a slowly propagating wave of depolarization followed by suppression of neuronal activity. It is initiated in the occipital region of the cerebral cortex and is propagated towards the front of the brain at 3-5 mm/ minute. CSD leads to the release of inflammatory mediators that alter nociceptors, irritate trigeminal nerve roots and change cerebral blood patterns.

Although CSD can be easily investigated in experimental animals and in humans, using functional magnetic resonance imaging, Hadjikhani (2008) was able to detect local increases in blood oxygen level dependent signals that spread through the visual cortex of a patient with MA which is similar to animal models.

The potential relationship between cortical spreading depression and migraine without aura remains controversial. It has been suggested that the long-term release of inflammatory mediators may structurally alter pathways and alter the processing of sensory inputs which alters disease progression.

1.2.4 Vascular spasm hypothesis

Willis first suggested that migraine is a vasospastic disorder of the cranial vessels (Willis T and Pordage S, 1683). Subsequently Wolff, supported that ischemia induced by intracranial vasoconstriction is responsible for the aura of migraine and that the subsequent vasodilation and

activation of perivascular nociceptive nerves resulted in headache. New imaging technologies have shown that intracranial blood flow patterns are inconsistent with this theory (Goadsby PJ, 2015). Furthermore, this theory does not explain why some effective migraine treatments do not affect blood vessels (Goadsby PJ, 2015).

1.2.5 Genetic causes of migraine (ion channel disorders)

Evidence for a genetic component in migraine comes from observational studies, which show that approximately half of migraineurs have an affected first-degree relative. While genetic determinants are seen as important, migraine risk is conferred by the complex interplay between predisposing and triggering factors.

Insights into the genetic and molecular pathophysiology of migraine have come from studies of rare monogenic subtypes of migraine, including dominantly inherited familial hemiplegic migraine (FHM) and migraine in familial advanced sleep phase syndrome FASPS). FHM is characterized by reversible hemiparesis plus other aura symptoms preceding or accompanying a migraine headache with at least one first-degree relative similarly affected. Many of the features of monogenic subtypes of migraine (e.g. hemiplegia during aura, progressive ataxia in FHM and FASP) are not found in common types of migraine (Montagna P, 2000).

1.2.6 Triggers

Migraine attacks are generally spontaneous but some individuals have known triggers which vary from individual to individual and will not always initiate a migraine. The mechanisms by which migraine triggers exert their effect is not clear despite a large number of trigger factors reported (Table 1.5). Furthermore, clinical studies investigating links between triggers migraine attacks have shown conflicting results (Hoffman J and Recober A, 2013; Lippi *et al*., 2014).

Table 1.6 List of common triggers

Stress	Dehydration
Emotion	Odours /Smoking
Hypoglycaemia	Alcohol
Altered sleep patterns	Caffeine
Physical exertion	Food chemicals (? Chocolate, MSG, nitrates)

1.3 Migraine Management

There are multiple approaches to manage the effects of migraine. It falls into two general types of approaches, a pharmacological and a non-pharmacological approach. There are wide array of pharmacological options that either aim to minimise the symptomatic effects or to act as a migraine prophylaxis.

1.3.1 Symptomatic

Table 1.7 outlines commonly used pharmacological agents used in the symptomatic treatment of migraines. The level of evidence varies greatly between commonly used agents and there are multiple physiological targets, highlighting the complex and heterogenous nature of migraines.

Table 1.7 Level of evidence of symptomatic migraine treatment

(Marmura *et al*., 2015; Schug *et al*., 2015).

Evidence	Pharmacological agent	Target
Level A	Triptans	Serotonin receptors
	dihydroergotamine (nasal spray)	Serotoninergic & adrenergic receptors 5-HT1D receptors
	NSAIDS	Cox 1 & 2
	opioids	μ receptors
	acetaminophen/aspirin/caffeine	Unknown. Postulated central effect and prostaglandin inhibition
Level B	Ergotamine	Serotoninergic & adrenergic receptors
	Ketoprofen	Cyclooxygenase inhibition
	Ketorolac (IV & IM)	Cyclooxygenase inhibition
	magnesium (IV)	Unknown. Postulated to interfere with substance P release
Level C	Dexamethasone IVI	Interleukin/CGRP & prostaglandin suppression
	Methadone IMI	μ receptor
	Codeine oral	μ receptor

1.3.2 Preventative therapy.

A number of guidelines have been established outlining the circumstances in which preventive treatment for migraine is recommended. These guidelines include:

- Recurring migraine attacks that significantly interfere with a patient's quality of life and daily routine despite trigger management, appropriate use of acute medications, and lifestyle modification strategies
- Frequent headaches (four or more attacks per month or eight or more headache days per month) because of the risk of chronic migraine
- Failure of, contraindication to, overuse of, or troublesome side effects from acute medications
- Patient preference, that is, the desire to have as few acute attacks as possible
- Presence of certain migraine conditions: hemiplegic migraine; basilar migraine (now called migraine with brainstem aura); frequent, prolonged, or uncomfortable aura symptoms; or migrainous infarction (Silberstein *et al*., 2012).

1.3.3 Non-pharmacological approach

There is evidence for self-care measures that help ease the frequency and intensity of migraine including:

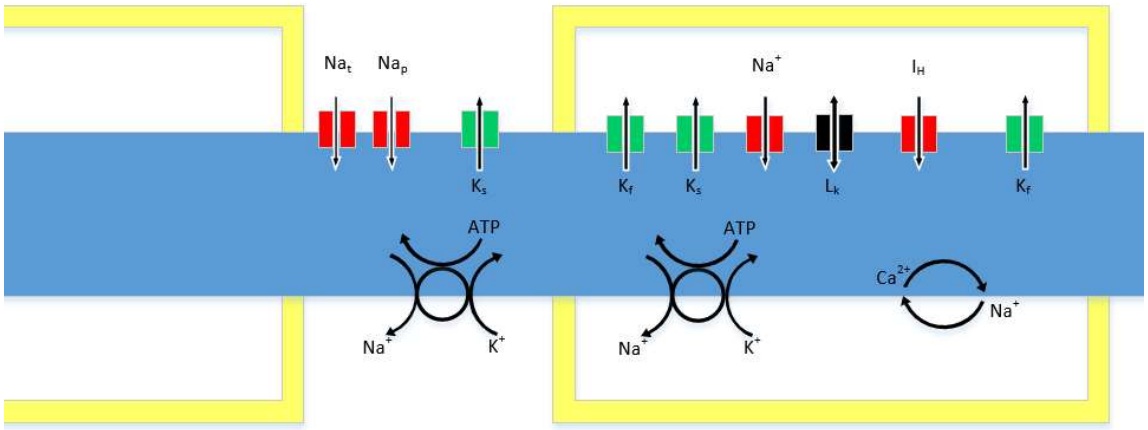
- Avoidance of provoking factors, particularly alcohol and dehydration.
- Physical therapy (including manual therapy, massage, muscle relaxation techniques, meditation and yoga)
- Sleep hygiene
- Appropriate rest at headache onset
- Maintenance of headache diary
- Sensible application of alternative medicine techniques including:
    - Acupuncture
    - Biofeedback
    - Cognitive behavioural therapy
    - Herbs and vitamins (Shaik MM and Gan SH, 2015) .

1.4 Nerve Excitability Studies

Physiological Background

The excitability of nerves is determined by the activity of a variety of ion channels, energy-dependent pumps, and ion exchange processes activated during the process of impulse conduction (figure 1.2). Clinical symptoms can result from disorder of function rather than structure. Therefore, tests of function are important investigatory tools for providing insights into disease states.

Figure 1.2 Diagram of Node of Ranvier with ion channels



Na_t = Transient Sodium
Na_p = Persistent Sodium
K_s = Slow Potassium
K_f = Fast Potassium
L_k = Leakage (Voltage Independent)
I_H = Hyperpolarisation Activated (Inward rectifier)

In myelinated nerves, salutatory impulse conduction occurs when the action potential (AP) jumps from one node of Ranvier to the next. The traditional view of impulse propagation is that most electrical activity develops at nodes of Ranvier, through specific Na^+ and K^+ channels and leakage currents, whereas the internodal axolemma and myelin function as a passive isolated cable. In

mammalian axons, the difference between internal to external voltages (resting membrane potential) is modelled as approximately -84 mV (Howells *et al*., 2012). A nerve impulse is generated as a result of the complex system of ionic pores changing between rest and activation.

Physiological states of depolarisation and hyperpolisation increase and decrease the ability of the cell to generate a signal, respectively.

The main generator of a resting membrane potential is permeability to K+ ions and impermeability to Na^+ ions. Hyperpolarisation-activated cyclic nucleotide-gated (HCN) channels allow for the passage of both Na^+ and K^+ ions and are most active at a range of -50 mV to -100 mV. This function may limit excessive hyperpolarisation mediated via Na^+/ K^+ pump activation from excessive impulses or from ischaemia.

Nerve Excitability Studies

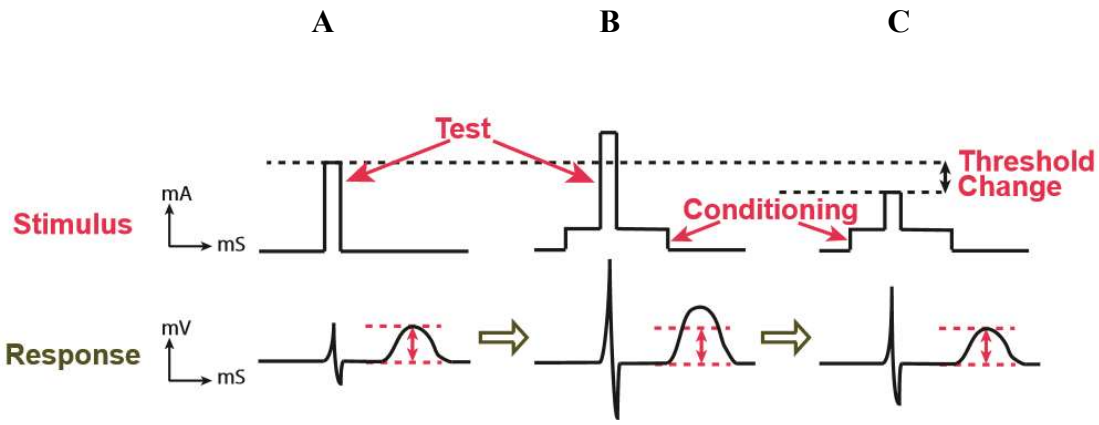
Currently, nerve conduction studies (NCS) are the mainstay of studying peripheral nerve function clinically. NCS use supramaximal stimuli to generate an action potential and measure velocity and amplitude of large myelinated motor or sensory fibre conduction, which are largely functions of nodal saltatory conduction. Nerve excitability studies (NES) are a non-invasive *in vivo* research tools used to investigate nerve function. In contrast to nerve conduction studies, NES use much smaller stimuli designed to *just* excite the nerve at its threshold. Nerve excitability studies involve applying a series of priming stimuli to the nerve before the test, and then track the resultant change in threshold to indirectly evaluate membrane potential and ion channel function. Hence nerve excitability studies provide complimentary information to NCS.

NES use a TROND protocol as described by Kiernan *et al*., (2000). The TROND protocol consists of a series of conditioning stimuli delivered via constant current stimulators and response signals displayed, analysed and recorded using QTRAC (copyright Institute of Neurology, London) software written by Professor Hugh Bostock. (Test stimulus combinations explained in sections 1.4.2 to 1.4.6.)

1.4.1 Threshold Tracking Principle

The principle of threshold tracking is illustrated in figure 1.3. This depicts a test pulse along a nerve and the elicited muscle response below (A). If a conditioning depolarising electronic pulse is added, the test pulse produces a supra maximal response (B). Test response A can be elicited by applying a conditioning stimulus with a reduced test pulse (C). Threshold change when the muscles response is the same is the difference between the control threshold and the conditioned threshold expressed as a percentage of initial stimulus.

Figure 1.3 Threshold tracking representation



(Adapted from TROND nerve excitability workshop Chicheley 2015)

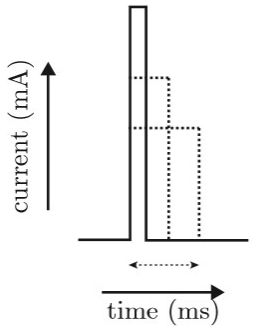
1.4.2 Stimulus response curve

A stimulus response curve is constructed from stimuli increasing in small increments to supra-maximal responses using a 0.2 ms pulse duration. A threshold current is then defined for tracking purposes as the stimulus strength required to elicit a 40% maximal response. The magnitude of changes in stimulus intensity is determined from the stimulus response curve and are automatically calculated by QTRAC-S software.

1.4.3 Strength duration relationship

The strength-duration relationship is plotted by adjusting the duration of the rectangular stimulating current pulse (Figure 1.4). The threshold or stimulus strength required to elicit the desired (40% maximal) response is obtained via threshold tracking for each time point. The threshold is measured at five different pulse widths from 0.2 to 1.0 ms, and the threshold charge is plotted against stimulus duration. The derived charge duration plots provide the measurements of strength-duration time constant (STDC) and the rheobase.

Figure 1.4 Multiple pulse widths used in Strength duration plots



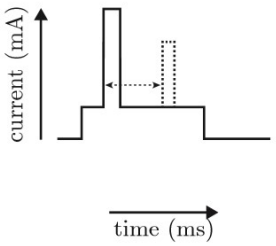
(Adapted from TROND nerve excitability workshop Chicheley 2015)

1.4.4 Threshold Electrotonus

Threshold electrotonus (TE) is a measurement of threshold changes as a result of sub-threshold conditioning stimuli (Figures 1.5 and 1.6).

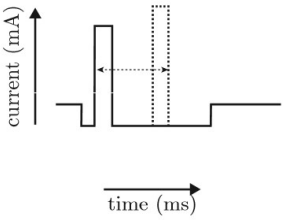
Conditioning currents of +20% and +40% (depolarising) and -20%, -40%, -70% and -100% (hyper-polarising) of control threshold were chosen. The threshold change was chosen at 26 time points that were before, during and after the conditioning stimuli.

Figure 1.5 Depolarising condition used in threshold electrotonus



(Adapted from TROND nerve excitability workshop Chicheley 2015)

Figure 1.6 Hyper-polarising condition used in threshold electrotonus



(Adapted from TROND nerve excitability workshop Chicheley 2015)

1.4.5 Recovery Cycle

The recovery cycle is measured by using a supramaximal conditioning stimulus followed by a test stimulus at varying conditioning-test intervals from 2 to 200 ms. This test creates three distinct periods: refractory, super-excitable and sub-excitable period.

The refractory period is determined by inactivation of the Na^+ channel gate on the internal aspect of fast Na^+ ion channels. The absolute refractory period corresponds to the closure of these inactivation gates and the relative refectory period is the period of time to recovery from inactivation to the opening of the in-activation gates.

The super-excitable period reflects the depolarising after potential, which is a result of capacitance charging at the internode. This is also known as Barrett and Barrett current. The Barrett and Barrett current discharges through or under the myelin sheath is dependent on membrane potential (Barrett and Barrett, 1982).

The sub-excitability period reflects hyper-polarising effects of inactivation of slow K^+ channels from the conditioning stimuli. This period is dependent on membrane potential and also the electrochemical gradient of K^+ ions.

1.4.6 Current Voltage Relationship

This relationship is analogous to threshold electrotonus while utilizing a fixed 200 ms conditioning current that varies in steps of 10% from +50% (depolarising) to -100% (hyperpolarising) of threshold. The change in threshold reflects the rectifying properties of the axon, specifically the properties of K^+ channels and hyperpolarisation-activated inwardly rectifying currents (I_h).

1.4.7 Nerve excitability studies and migraine

The scientific rational is described in detail in chapters three. A brief synopsis is outlined below.

Chronic migraine has a heterogenous pathophysiology theorised as aberrant peripheral and central hyperexcitability of pain pathways leading to a dysregulation of sensory perception. Many migraine treatments, including anti-epileptic agents act via alterations in resting membrane potential or possibly by altering central ion channel function.

In addition to documenting changes in membrane potential in a wide number of conditions affecting peripheral nerve, excitability studies have been able to identify changes in membrane potential in peripheral axons in selected CNS disorders (e.g. stroke, multiple sclerosis, spinal cord injury), probably reflecting compensatory altered regulation of ion channel expression in these

disorders (Krishnan *et al*., 2009; Tomlinson *et al*., 2018). Hence, in a heterogeneous population of chronic migraine patients, where the symptoms may be the nett effect of centrally and peripherally acting processes affecting nerve excitability, it could be hypothesised that changes in peripheral nerve excitability studies may reflect this nett effect of central and peripheral activity.

The use of NES to detect changes in peripheral nerve excitability reflecting disorders a central function is well established (Tomlinson et al., 2009; Tomlinson et al., 2016; Tomlinson et al., 2018; Krishnan et al., 2009). This has laid the groundwork to apply similar principles to common conditions such as chronic migraine in which the physiology is not completely understood.

To date, there are no studies that have specifically investigated nerve excitability in human migraine. Considering that migraine's pathophysiology is consistent with neurovascular theory with neuronal hyperexcitability, it is hypothesised that NES can be used as a research tool to provide insights into this disease state.

1.5 Evaluating a lignocaine and ketamine subcutaneous protocol in a chronic migraine population and search for a novel biomarker

The presentation of chronic migraine is a frequent occurrence in neurological practises. It is can be challenging to manage these patients as there are limited therapeutic options and an incomplete understanding of chronic migraine physiology. More neurophysiological research into chronic migraine is required to meet this unmet need.

1.5.1 Study Proposal

The studies in this thesis aim to evaluate the effectiveness of a protocol used in the treatment of chronic migraine and to investigate potential new objective markers for treatments. The specific research questions addressed are:

1. Does the use of subcutaneous lignocaine and ketamine infusion protocol in chronic migraine translate to improved clinical outcomes by subjective measures?

There is no reported information on the use of low dose combined subcutaneous lignocaine and ketamine infusion in a refractory chronic migraine population. Clinicians at St Vincent's Private Hospital (Sydney) have employed this protocol in the treatment of refractory chronic migraine based on similar reported protocols using intravenous lignocaine in headache patients. To date, the effect on subjective headache markers resulting from differences in i) combination with low dose ketamine and ii) administration through different routes, have been anecdotal.

2. Can nerve excitability studies be used as a clinical tool to provide *in vivo* assessment of the treatment?

The therapeutic action of some migraine treatments results from alteration of ion channel function and nerve excitability. Lignocaine's therapeutic benefits on pain is reported to relate to changes in sodium channel function. Therefore an *in vivo* assessment of ion channel function may reflect differences and provide an objective marker of this treatment. A biomarker of treatment would provide clinicians with greater information on how best to direct treatment.

Nerve excitability studies are research techniques that demonstrate *in vivo* peripheral excitability changes as result of ion channel mutations and from high dose lignocaine administration. Episodic Ataxia type 2, which is allelic with familial hemiplegic migraine, also has reported peripheral nerve excitability differences from normal subjects. Therefore, nerve excitability studies in chronic migraine may therefore identify a new biomarker where there are currently no clinically useful surrogate markers of migraine intensity or activity.

1.5.2 Aims

1. To evaluate the effectiveness of continuous (7-10 days) subcutaneous lignocaine and ketamine infusion for treatment of chronic migraine with regards to frequency and severity of migraine, lost days of productivity and amount of headache medication required.

2. To obtain peripheral nerve excitability studies in patients with chronic migraine before, during and after treatment with a lignocaine and ketamine infusion to develop an in vivo biophysical marker of change of neuronal hypersensitivity in these patients with treatment, as well as an objective measurement of lignocaine effect during infusion.

1.5.3 Hypothesis

We hypothesise that a continuous subcutaneous lignocaine and ketamine infusion will decrease the frequency and severity of migraines and improved productivity of participants.

We hypothesise that nerve excitability studies may detect changes in peripheral nerve ion channel function before and after treatment that may provide a useful predictive biomarker of migraine and treatment responses.

1.5.4 Study design

A prospective observational cohort study, designed to observe the outcome of a patient's management as determined by their treating neurologist. The study was not designed to direct or

alter therapy, rather follow the course of their individualised care before and after inpatient intervention.

Patients were considered eligible for inclusion if:
a) Chronic migraine diagnosis was confirmed by the treating neurologist according to ICHD-3 β criteria
b) patients were refractory to standard therapies
c) aged between 18-70.
Patients with known contraindications to the therapy including prolonged QT interval on ECG or malignant arrhythmia were excluded from the study.

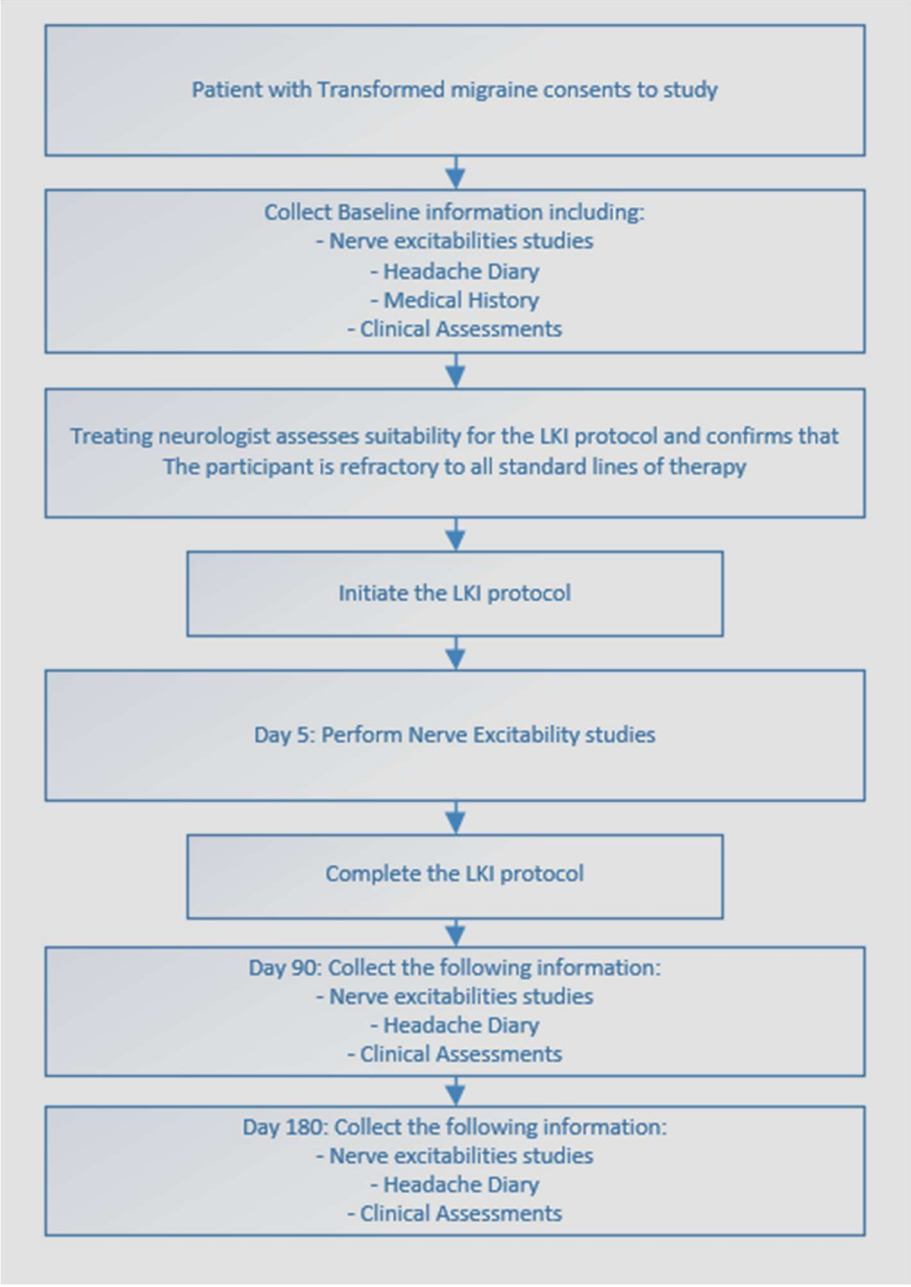
Patients underwent evaluation at four-time points including clinical assessment, headache diary review, MIDAS questionnaire (migraine disability assessment score), medication review and nerve excitability studies (See Figure 1.7 Summary of Procedures for Observational Study of Management of Chronic Migraine and Table 1.7 Schedule of Events).

More specific information is outlined in methodology section in Chapters two and three.

Table 1.8 Schedule of events

	Visit 1 (Baseline)	Visit 2 (admission)	Visit 3 (admission day 5)	Visit 4 (90 days)	Visit 5 (180 days)
Medical History and consent	X				
MIDAS		X		X	X
Medication review	X	X		X	X
Nerve excitability studies	X	X	X	X	X
Headache diary	Provided	X		X	X

Figure 1.7 Summary of Procedures for Observational Study of Management of Chronic Migraine



1.5.5 Study procedures

The following outlines the specific information that was collected:

Medical History

A clinical assessment and medical history were obtained (appendix 7). This included a comprehensive migraine history, medications used for migraine, clinical exam, social and family history.

MIDAS

Migraine disability assessment (MIDAS) is a scale that gives clinicians a measurement of impact of headaches on daily activity. See appendix 4.

Medication Review

Participants were asked to record medications used for migraine management.

Nerve Excitability Studies

Tests were performed on the participant's median nerve with six surface electrodes (per Figure 2 set up). Compound action potentials were recorded along the abductor pollicis brevis after stimulation of the median nerve near the wrist. Current was delivered from DS5 stimulators (Digitimer Ltd, UK) and QtracS stimulation software following the TRONDNF protocol. Nerve Excitability tests were performed prior, during and after the infusion**.**

Headache Diary

Each was requested to keep a detailed headache diary that included number of headache episodes, pain scores, medication used and other associated factors including menstrual periods. See appendix 5.

Infusion protocol

A preparation containing lignocaine, ketamine and saline was delivered subcutaneously to the patient via a syringe driver and butterfly cannula to the lateral abdominal wall or outer thigh. The

rate of infusion was slowly titrated over the first 24 hours and adjusted as clinically indicated by the headache response. The patients were regularly monitored for pain, sedation and adverse effects with rotation of subcutaneous infusion site. While in hospital, they engaged in regular consultation and create an appropriate management plan.

The infusion protocol was initiated on Visit 2 (admission) per scheduled of events (Table 1.8).

1.5.6 Treatment rationale

The scientific rational of the treatment is described in detail in chapters two. A brief synopsis of study rational is outlined below.

There are limited published data suggesting benefit from administration of intravenous lignocaine for treatment of MOH and CM (Hand and Stark, 2000; Rosen *et al*., 2009). Lignocaine blocks the activation of voltage-gated Na^+, preventing depolarisation of the post-synaptic membrane and propagation of the action potential. Its short half-life and duration of action necessitates continuous parenteral infusion. The efficacy of lignocaine in treatment of chronic migraine probably relates to reduction of neurally-driven pain in both the central nervous system and also in peripheral trigeminal nociceptive afferents.

The use of parenteral ketamine in chronic pain and neuropathic pain is well documented (Kvarnstrom *et al*., 2003; Campbell-Fleming *et al*., 2008), including some reports of response in chronic headache (Webster and Walker, 2006). Intranasal ketamine has been studied in acute migraine and may reduce severity but not duration of migrainous aura (Afridi *et al*., 2013). Short term improvement in chronic migraine severity has been shown with use of intravenous ketamine in a small case series of six (Lausisten *et al*.,2016).

Ketamine decreases central sensitization and allodynia (Sanchez-Porras R *et al*., 2014), possibly due to reduction activation of pain processing pathways including decreased activation of the secondary somatosensory cortex, insula and anterior cingulate cortex (Sprenger T *et al*., 2006). It is thus a suitable agent for chronic migraine treatment.

CHAPTER 2

Breaking the cycle of chronic migraine with a low-dose subcutaneous lignocaine and ketamine infusion: a case series.

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Abstract

The entrenchment of chronic migraine, often compounded by analgesic dependence and perpetuated by recognized barriers to treatment. This report describes an approach to treatment which includes admission to hospital for administration of a low dose subcutaneous lignocaine and ketamine infusion. The aim is to enable adequate analgesia and disruption of entrenched headache while patients undergo revision of oral medications and implementation of non-pharmacological strategies to treat chronic headache. Fourteen patients were recruited, nine of whom were female. Mean age was 43 years (range 27-61). The infusion was tolerated without significant side-effects. At six months, 13/14 patients had sustained benefit from admission. Three of 4 patients remained free of MOH headache. One patient remained headache-free at six month follow up. Conversion from chronic migraine to episodic migraine was seen in 6/14. Improvement in chronicity was reported by 6/14. Two of six patients unable to work because of chronic headache were able to return to work, and a third patient returned to studies. These findings suggest that a prolonged subcutaneous lignocaine and ketamine infusion is a useful adjunct to conventional management to enable breaking the entrenchment of chronic headache with.

Key words: chronic migraine, migraine, lignocaine, ketamine, medication overuse headache

Abbreviations: NSAIDS: Non steroidal anti inflammatory drugs

ICHD-3: International Classification of Headache Disorders version 3

Introduction

The management of chronic migraine includes correcting medication overuse headache and implementing suitable preventative agents and appropriate use of medications for acute episodes (May and Schulte, 2016). However, in many cases this management paradigm oversimplifies the complexity of chronic migraine and does not address the other factors that contribute to the cycle of headache, particularly the central pathways that perpetuate chronic migraine. Abrupt discontinuation of overused triptans and opioids may produce withdrawal symptoms (Kristoffersen and Lundqvist, 2014) and patients with chronic migraine may experience major escalation in headache while changing preventatives. The combination of headache intensification and/or withdrawal side effects may sabotage implementation of a management plan, particularly where the lead-in time of action of preventative medications may be days to weeks.

In an inpatient setting, chronic migraine patients are able to access adequate analgesia to minimize impact of medication withdrawal and be provided with support and education to implement a multifaceted management plan to address factors perpetuating their complex disability. In the long term, with reduction in both direct and indirect costs, this option may prove both cost-effective and more successful for those patients with recalcitrant headache.

While not first-line treatment, limited published data suggest benefit from inpatient administration of intravenous lignocaine for curtailment of medication overuse headache and chronic migraine(Hand and Stark, 2000; Rosen *et al*., 2009). Lignocaine blocks activation of voltage-gated sodium channels, preventing depolarisation of the post-synaptic membrane and propagation

of the action potential. Its short half-life and duration of action necessitates continuous parenteral infusion in this setting. The efficacy of lignocaine in the treatment of chronic migraine probably relates to reduction of neurally-driven pain in both the central nervous system and also in peripheral trigeminal nociceptive afferents. The mean duration for positive results appears to be 8.5 days (Lake *et al*., 1993;Rosen *et al*., 2009) implicating that treatment duration is a factor in 'resetting' entrenched patterns of neurally-driven pain.

Ketamine is an agonist of N-methyl-D-aspartate (NMDA) receptors acting in the central nervous system, and also has activity on opioid, monoaminergic, cholinergic, nicotinic, and muscarinic receptors (Craven, 2007). In the setting of transformed migraine, it provides short-term analgesia and enables reduction in central sensitisation of pain pathways, particularly in the setting of codeine and opioid overuse (Goldberg *et al*., 2005; Tawfic QA, 2013). The use of parenteral ketamine in chronic pain and neuropathic pain is well documented (Kvarnstrom *et al*., 2003; Campbell-Fleming *et al*., 2008), with some reports including chronic headache in their cohort (WebsterR and WalkerJ., 2006). Intranasal ketamine has been studied in acute migraine: it may reduce the severity but not the duration of migrainous aura in the acute setting (Afridi *et al*., 2013). Short term improvement in chronic migraine severity has been shown Pomeroy *et al.,* 2017). Ketamine decreases central sensitization and allodynia in pain conditions (Sanchez-Porras R *et al*., 2014), possibly due to reduced activation of areas involved in nociceptive signals, the secondary somatosensory cortex, insula and anterior cingulate cortex (Sprenger *et al*., 2006) thereby making it a suitable candidate for chronic migraine treatment.

Intravenous use of these agents has various limitations: intravenous doses of lignocaine may cause cardiac arrhythmias and administration may require cardiac monitoring. Ketamine may produce

obtundation, dysphoria or hallucinations in higher doses. There are no published data regarding the combination use of these medications in chronic migraine. This paper describes an inpatient approach to management of patients with chronic migraine that includes supportive care of symptoms with a prolonged subcutaneous lignocaine and ketamine infusion during implementation of appropriate medication, along with a management plan to address concurrent limiting comorbidities.

Methods

Study design

Ethics approval was obtained through St Vincent's Hospital Human Research Ethics Committee (HREC/15/SVH/356) and the University of Notre Dame Human Research Ethics Committee (017044S). Written informed consent was obtained from participants. A prospective observational cohort study was undertaken to document the outcome of a patient's management as determined by their treating neurologist. The study was not designed to direct or alter therapy; the aim was to follow the course of their individualised care as determined by their treating neurologist before and after inpatient intervention. Patients aged 18-70 were eligible for inclusion if they had suffered chronic migraine which had been refractory to standard migraine therapies. Exclusion criteria included pregnancy, breast feeding and known contraindications to the therapy including prolonged QT interval on ECG or malignant arrhythmia.

Patients underwent evaluation at four-time points: baseline assessment (before commencement of infusion), day 5 of infusion, 3 months after infusion and six months after infusion. Each assessment included clinical assessment, headache diary review and medication review. Prior to

commencement of the infusion, baseline ECG, full blood count, renal function and liver function were measured.

Infusion protocol

A preparation containing 1g lignocaine and 250 mg ketamine was diluted in 0.9% sodium chloride (normal saline) to a total volume of 24 ml. The infusion was administered by a registered nurse. Continuous infusion was delivered subcutaneously via a syringe driver (the NIKI T34™ Syringe Driver) and through a 22 gauge butterfly cannula to the subcutaneous tissue of the lateral abdominal wall or outer thigh and secured by a large transparent adhesive dressing. The infusion was commenced at a rate of 0.5 ml/hour and slowly titrated over the first 24 -48 hours according to clinical response. An infusion rate of 1.0ml/hour was regarded as optimal, based on the occasional development of dysphoria at higher doses but individual rates varied between 0.6ml/hour and 1.5ml/hour. Patients were regularly monitored for pain, sedation and adverse effects. The solution was replenished daily, and the, needle and insertion site were then changed. The Numerical Rating Scale (NRS) was used to score headache every four hours from 0 to 10 (10 being 'worst possible pain'). The infusion continued until adequate analgesia was reached or non-efficacy was established, as determined by patient report and evaluation of the treating clinician.

Inpatient Management

Analgesics including opioids and triptans that might have contributed to headache cycle were ceased. Expected rebound of severe headache during inpatient medication change was managed in the short term with judicious use of low dose subcutaneous midazolam, morphine and metoclopramide as required. All patients received education about chronic migraine management

with the importance of sleep, mood and fitness emphasized. Written management plans for acute headache and chronic headache were provided.

Results

Baseline characteristics

Fourteen patients were recruited from the clinical practices of headache neurologists over a 16 month period (Table 1). Nine patients were women. The age range was 27 – 61 years (mean = 43 years). Four had concurrent MOH at or immediately prior to admission attributed to triptans and/or codeine. Six patients had clinical depression and 3 had other pain syndromes. All patients had previously received extensive conventional outpatient headache management and had failed several first-line agents for prevention and acute headache. The most frequently prescribed analgesics for acute headache were triptans (4/14), non steroidal anti inflammatory drugs (NSAIDS) (4/14) and codeine (3/14). Several patients were not taking any abortive medications due to inefficacy. The frequency of analgesic use varied greatly and generally had limited benefit. All patients had been prescribed migraine prophylactic agents prior to treatment. Employment was directly affected in 8/14 patients. Six patients had stopped working entirely due to headache and 4 had reduced capacity to work. Five patients were not working for other reasons.

Table 1: Patient Demographics			
Patient	**Age; Sex**	**Diagnoses at enrolment**	**Relevant comorbidities**
1	57; M	Migraine Medication overuse headache (codeine)* Cervicogenic headache	Fungal sinusitis
2	51; M	Chronic migraine Medication overuseheadache (codeine)*	
3	42; M	Chronic migraine	Fibromyalgia Trigeminal neuralgia
4	41; F	Chronic migraine	Depression
5	29; M	Chronic migraine	Depression, anxiety
6	43; F	Chronic migraine Medication overuseheadache (triptan)	Chronic axial pain
7	27; F	Chronic migraine Medication overuseheadache (codeine, diazepam)*	Depression, post traumatic stress disorder, Anxiety,
8	48; F	Chronic migraine	Polycystic kidney disease Hypertension Alcohol excess
9	61; F	Chronic migraine Medication overuseheadache (triptan)	
10	58; M	Chronic migraine	Vertigo Non epileptic seizures
11	56; F	Chronic migraine	Hemifacial spasm, Stroke Epilepsy Depression
12	58; F	Chronic migraine Medication overuseheadache (codeine, triptan)	
13	55; F	Chronic migraine	Depression
14	29; F	Chronic migraine	Depression Fibromyalgia

*analgesic contributing to headache discontinued prior to admission to hospital

Outcomes during inpatient stay

The infusion was well tolerated in all patients. The duration of infusion was 6 - 22 days (mean 11 days). Minor subcutaneous infusion site reactions were seen in some patients characterized by erythema and mild induration. The reaction dissipated within a 1-2 days of re-siting the infusion. No patient experienced altered consciousness, hallucinations or arrhythmia during the infusion.

All patients underwent change of preventative medications during their inpatient stay. The most frequently prescribed preventatives were lamotrigine, botulinum toxin, gabapentin and topiramate.

Outcomes at six months

Thirteen of 14 patients in a population of previously refractory chronic migraine patients treated with subcutaneous lignocaine and ketamine infusion had improved headache and quality of life at discharge and follow up. Seven patients were no longer classified as having chronic migraine, with one being headache free (Table 2). Six patients had converted from chronic migraine to episodic migraine and 6/14 reported significant improvement in their chronic migraine at six months with subjective reduction in severity and frequency enabling increased circle of engagement (see Table 2). MOH was addressed where relevant and 3 of 4 patients remained free of MOH headache at six months. One patient had no improvement at six months and this patient had been unsuccessful at stopping daily triptan use (Patient 9). At six month follow up, only one patient used opiates (long acting) for headache control (Patient 13). This patient had a history of intolerance to tricyclic medications, and liver dysfunction precluded use of other alternatives. NSAIDs and triptans were the most frequently prescribed abortive agents at follow up. Suboccipital steroid/lignocaine injections were effective in aborting acute relapse headaches in four patients who had limited benefit from oral NSAIDs and triptans (Appendix 1).

Table 2: Outcome of treatment at 6 months follow up							
Patient	**Headache free**	**Episodic migraine only**	**Improved chronic migraine**	**Return to vocation**	**Lifestyle change**	**Opiate use at 6 months**	**Triptan overuse at six months**
1	No	No	Yes	Yes	Yes	No	No
2	No	Yes	N/A	N/A	Yes	No	No
3	No	No	Yes	N/A	Yes	N/A	N/A
4	No	Yes	N/A	Yes	Yes	N/A	N/A
5	No	No	Yes	No	Yes	N/A	N/A
6	No	Yes	N/A	N/A	Yes	No	No
7	No	Yes	N/A	Yes	Yes	N/A	N/A
8	No	Yes	N/A	No	Yes	N/A	N/A
9	No	No	No	N/A	No	No	Yes
10	Yes	N/A	N/A	Yes	Yes	N/A	N/A
11	No	No	Yes	No	Yes	N/A	N/A
12	No	No	Yes	Yes	Yes	N/A	N/A
13	No	Yes	N/A	No	No	Yes	N/A
14	No	No	Yes	Yes	Yes	No	N/A
N	1/14	6/14	6/14	6/14	13/14	1/14	1/14
%	7%	84%	84%	84%	93%	7%	7%

*N/A = not applicable

Work engagement or lifestyle significantly improved in 6 patients. Two of six who had stopped work for headache were able to return to work, with one other returning to studies. One patient returned to full time work after having had reduced hours. One further patient was able to undertake a strenuous holiday having been unable to for many years (reflecting improvement in activity/engagement).

Discussion

Chronic migraine is a complex, disabling disorder that at times requires intensive efforts from both the patient and the neurologist to manage. The cohort described in this paper reflects the experience described in headache literature with concurrent mood disorders and disengagement from work and other common activities. Chronic migraine was abolished in half the patients, with six converting to episodic migraine. Quality of life improved in 13 of the 14 patients as measured by return to vocational activities or increase engagement in lifestyle activities including regular exercise.

The positive outcomes observed may be in part due to a reduction of sensitized central pain pathways and peripheral trigeminal nociceptive afferent pathways (Kaube *et al*., 1994). Prior studies on chronic pain using intravenous lignocaine or ketamine reported sustained benefits when infusions were given for at least 4 days (Niesters *et al*., 2014; Lauritsen *et al*., 2016; Etchison *et al*., 2017) Allodynia scores, an indicator of central sensitisation has been reported to decrease following administration of intravenous ketamine (Sanchez-Porras R *et al*., 2014), . Ketamine is well recognized to have benefit for major depressive disorder (McGirr *et al*., 2014, Anrade, 2017) which is increased in prevalence in patients with chronic migraine. It is conceivable that improved mood and outlook with ketamine used in this protocol facilitates engagement with migraine

management. The mean length of stay in hospital was 11.5 days. This is similar to observations from other studies between 8.5 and 13 days (Rosen *et al*. 2009. Lake, Saper & Hamel, 2009). The length of time is suggestive that sustained pain reduction requires stabilization of the entrenched mechanisms that perpetuate pain.

There is good evidence that withdrawal of medications responsible for medication overuse headache is effective in reducing the frequency of chronic migraine headache frequency and improving quality of life (Diener & Limmroth, 2004). Inpatient and outpatient treatments as well as advice have each been shown to be beneficial to improving outcomes in chronic migraine (Rossi *et al*., 2006). However, randomised control studies have shown no significant differences when comparing inpatient versus outpatient management (Lai and Wang, 2016). Rossi *et al*. 2013 argued that inpatient withdrawal is more effective than outpatient management in complicated medication-overuse headache patients. The current patient cohort was selected after failure to respond to advice and outpatient management. If avoidance of admission to hospital for these patients is financially-driven, this may in fact be counter-productive as the long-term benefit with regards to reduction in direct and indirect costs with improved control may outweigh the cost of admission. Inpatient treatment allows for the constant monitoring of medication intake and for possible withdrawal symptoms. The hospital provides a safe environment for removal of offending medications and to re-educate patients on risk of medication overuse.

Prophylactic medication combinations are designed with the hope of synergistic effects from different mechanisms of action. The preventative medications used by this cohort are second-line agents (Appendix 8) reflecting that multiple first-line agents have been unsuccessful due to inefficacy or intolerability. In the current cohort, the preventative regimen was altered for each participant, often with combinations of migraine prophylactic agents including riboflavin,

magnesium and antidepressants. There is very limited evidence for the use of multiple versus single prophylactic agents in headache management. An improved response to preventative medications that had been previously tried post-infusion was identified in some patients and was presumably the result of multiple factors. It is difficult to determine whether the infusion protocol had a specific effect on regaining a response to medications and we assume that the observed restored response is primarily due to a sufficient period away from analgesic medication. This outcome suggests that there should be a greater role for planned drug rotation in the refractory chronic migraine populations to address tolerance and diminishing therapeutic responses.

While long term benefits will be compounded by a number of variables, reducing pain pathway sensitisation should be an initial step in changing the intractable pattern. By designing a treatment protocol that aims to reduce pain signals, the chance of providing headache treatments to benefit the patient will improve.

Lignocaine and ketamine do have potential for serious adverse side effects which therefore necessitate inpatient treatment. These risks are minimised through the protocol's use of minimal doses, subcutaneous administration to minimise risk of inadvertent bolus doses, gradual dose escalation based on participant response and constant monitoring. There were no serious adverse effects observed in this prospective cohort. The study was limited by population bias to a highly refractory migraine population who had received inadequate relief from standard treatments. Furthermore, the study participants were a heterogeneous population with multiple comorbidities and recruitment only occurred at one site. Lastly, there was no control group. However, this study was deliberately designed as a proof of principle to enable furthermore rigorous studies to be performed.

Conclusion

This study provides pilot data that support the use of low dose subcutaneous lignocaine and ketamine infusion in refractory chronic migraine populations. Future studies can use this as a platform for randomized placebo controlled trial and investigate the role of central sensitisation in the maintenance of chronic migraine, potentially allowing the development of novel treatments.

References

Afridi SK, Giffin NJ, Kaube H, Goadsby PJ. A randomized controlled trial of intranasal ketamine in migraine with prolonged aura. Neurology. 2013. 80:642-647.

Anrade C. Ketamine for Depression, 2: Diagnostic and Contextual Indications. J Clin Psychiatry 2017. 78:555-558.

Buse DC, Manack AN, Fanning KM, Serrano D, Reed ML, Turkel CC, Lipton RB. Chronic migraine prevalence, disability, and sociodemographic factors: Results from the American Migraine Prevalence and Prevention Study. Headache. 2012. 52:1456-1470.

Diener HC, Limmroth V. Medication-overuse headache: a worldwide problem. Lancet Neurol. 2004. 3(8):475-483

Etchison A, Manfredi L, Mohammed M, Phan V, McAllister KB, Ray M, Heitz C. Low-Dose Intravenous Ketamine for Acute Migraine in the Emergency Department: A Randomized Placebo-Controlled Trial. Annals of emergency medicine. 2017. 70:208

Garrick R. Intravenous and Subcutaneous delivery of Lignocaine and/or Ketamine for the Treatment of Pain. St Vincent's Private Hospital Policy and Procedure Manual 2014.

Fisher K., Coderre TJ, Hagen NA. Targeting the N-methyl-D-aspartate receptor for chronic pain management. Preclinical animal studies, recent clinical experience and future research directions. J Pain Symptom Manage. 2000. 20:358-373.

Goldberg ME, Domsky R, Scaringe D, Hirsh R, Dotson J, Sharaf I, Torjman MC, Schwartzman RJ. Multi-day low dose ketamine infusion for the treatment of complex regional pain syndrome. Pain Physician. 2005. 8:175–916.

Headache Classification Committee of the International Headache Society. The international classification of headache disorders, 3rd edition. Cephalalgia. 2018. 38:1-211.

Lake AE, 3rd, Saper JR, Madden SF, Kreeger C. Comprehensive Inpatient Treatment for Intractable Migraine: A Prospective Long-Term Outcome Study. Headache. 1993. 33(2): 55-62.

Lake, AE, 3rd, Saper JR, and Hamel RL. Comprehensive Inpatient Treatment of Refractory Chronic Daily Headache. Headache. 2009. 49(4): 555-62.

Lai TH, Wang SJ. Update of Inpatient Treatment for Refractory Chronic Daily Headache. Curr Pain Headache Rep. 2016. 20 (1):5

Lipton RB. Direct and Indirect Costs of Chronic and Episodic Migraine in the United States: A Web-Based Survey. Headache. 2016. 56:306-322.

Lauritsen C, Mazuera S, Lipton RB, Ashina S. Intravenous ketamine for subacute treatment of refractory chronic migraine: a case series. Headache. 2016. 17:106

Kaube H, Hoskin KL, and Goadsby PJ. Lignocaine and headache: an electrophysiological study in the cat with supporting clinical observations in man. J Neurol. 1994. 241:415-420

Kristoffersen ES, Lundqvist C. Medication-overuse headache: a review. J Pain Res. 2014. 26 (7): 367-78.

Marmura MJ, Goldberg SW. Inpatient management of migraine. Curr Neurol Neurosci Rep. 2015. 15 (4):13.

May A, Schulte LH. Chronic migraine: risk factors, mechanisms and treatment. Nat Rev Neurol. 2016 Aug;12(8): 455-64.

McGirr A, Berlim MT, Bond DJ, Fleck MP, Yatham LN, Lam RW. A systematic review and meta-analysis of randomized, double-blind, placebo-controlled trials of ketamine in the rapid treatment of major depressive episodes. Psychol Med. 2015. 45 (4): 693-704.

Messali A, Sanderson JC, Blumenfeld AM, Goadsby PJ, Buse DC, Varon SF, Stokes M, Lipton RB. Direct and Indirect Costs of Chronic and Episodic Migraine in the United States: A Web-Based Survey. Headache. 2016. 56 (2): 306-22

Niesters M, Martini C, Dahan A. Ketamine for chronic pain: risks and benefits. Br J Clin Pharmacol. 2014. 77 (2): 357–367.

Neurological Disorders Collaborator Group. Global, regional, and national burden of neurological disorders during 1990-2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet Neurol. 2017. 16 (11): 877-897.

Pomeroy JL1, Marmura MJ1, Nahas SJ1, Viscusi ER1. Ketamine Infusions for Treatment Refractory Headache. Headache. 2017 Feb; 57 (2): 276-282

Rosen N, Marmura M, Abbas M, Silberstein S. Intravenous lidocaine in the treatment of refractory headache: a retrospective case series. Headache. 2009. 49 (2): 286-91.

Rossi P, Di Lorenzo C, Faroni J, Cesarino F, Nappi G. Advice alone vs. structured detoxification programmes for medication overuse headache: a prospective, randomized, open-label trial in transformed migraine patients with low medical needs. Cephalalgia. 2006. 26 (9):1097-1105

Rossi P, Faroni JV, Tassorelli C, Nappi G. Advice alone versus structured detoxification programmes for complicated medication overuse headache (MOH): a prospective, randomized, open-label trial. J Headache Pain. 2013. 14: 10-17

Sanchez-Porras R, Santos E, Schöll M, Stock C, Zheng Z, Schiebel P, Orakcioglu B, Unterberg AW, Sakowitz OW. The effect of ketamine on optical and electrical characteristics of spreading depolarizations in gyrencephalic swine cortex. Neuropharmacology. 2014. 84:52–61

Scher AI, Stewart WF, Liberman J, Lipton RB. Prevalence of frequent headache in a population sample. Headache. 1998. 38:497-506.

Sprenger T, Valet M, Woltmann R, Zimmer C, Freynhagen R, Kochs EF, Tölle TR, Wagner KJ. Imaging pain modulation by subanesthetic S(+)-ketamine. Anesth Analg. 2006. 103 (3): 729–37

Tawfic QA. A review of the use of ketamine in pain management. J Opioid Manag. 2013. 9 (5): 379-88.

Consent

Written informed consent was obtained from the patients for publication of this Case report.

Acknowledgements: This research program has been supported by a grant from St Vincent's Clinic foundation and The Brain Foundation (Australia).

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CHAPTER 3

Subcutaneous lignocaine and ketamine infusion may act via central pathways in chronic migraine.

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Abstract

Pathophysiology of chronic migraine is postulated as sensitisation of trigemino-cervical pathways and entrenchment of central pathways involved in migraine generation. There are no readily available clinical biomarkers for migraine to serve as an objective marker of the condition. In this study, the hypothesis that nerve excitability studies may be useful in assessment of chronic migraine patients is explored. Peripheral nerve excitability studies are sensitive to changes in active and passive properties of the axonal membrane and have been used extensively as a marker of systemic alterations in nerve function. Fourteen patients with chronic migraine underwent nerve excitability studies of median nerve on four occasions over six months. During this time, their treatment included hospital admission for a subcutaneous lignocaine and ketamine infusion as part of headache containment. Studies performed before, during and after the infusion did not differ from control values despite therapeutic benefit during the infusion and afterwards. Lack of detectable change in peripheral axonal excitability has significance in that it could be inferred to suggest a more proximal mechanism of action of the lignocaine and ketamine infusion rather than via peripheral trigeminal afferents. It is noteworthy that medications used by this cohort that could potentially affect membrane potential do not affect peripheral axonal excitability studies.

Key words

Chronic migraine; lignocaine; ketamine; nerve excitability; threshold electrotonus

Introduction

The challenge in developing a biomarker for assessment and diagnosis of migraine partly lies in the heterogeneity of pathophysiology between individuals and within an individual, and the variable influence of triggers (including hormonal factors, sleep, mood, stressors etc). Both

central and peripheral pathways are implicated in the development of migraine and it has been hypothesised that a dysregulation of sensory processing involves activation and sensitisation of trigemino-vascular and upper cervical pathways relaying to the brain stem and diencephalic nuclei (Goadsby *et al*., 2017). Imaging, neurophysiologic and biochemical studies also implicate cortical dysfunction and hyperexcitability and release of proinflammatory and pain cytokines in the generation of migraine (Pietrobon, 2005; Pietrobon and Moskowitz, 2011). With repeated stimulation of trigeminal fibres, chronic migraine may lead to structural and functional changes which may include release of nociceptive neurotransmitters and upregulation of ion channels or sensory receptors on pain nerve endings. (Burstein *et al*., 2004; Aoki and Francis, 2011). As a result, peripheral afferents are sensitised and the lower threshold to firing promotes central sensitization, (Dodick and Silberstein, 2006) of which cutaneous allodynia, is a clinical marker (Burstein and Jakubowski, 2004).

Insight into the pathophysiology of migraine has been advanced by modalities that assess dynamic brain function during migraine and interictally. Functional assessment of brain or nerve activity in migraine would ideally lead to a useful biomarker of disease analogous to EEG in epilepsy or ECG in cardiac assessment. Tools for functional migraine evaluation have included functional MRI (fMRI), positron emission tomography (PET), blood oxygen level–dependent (BOLD) functional magnetic resonance imaging and neurophysiologic assessment of cortical excitability (magnetoencephalography (MEG)), magnetic suppression of perceptual accuracy (MSPA) and transcranial magnetic stimulation (TMS). These studies have given insight into the pathophysiology of migraine but have limited usefulness in the clinical sphere and have identified physiologic differences between acute and chronic migraine.

Activation of central pathways in acute migraine is different from that in chronic migraine. For example, PET studies show continuous overactivity in certain brain regions in chronic migraine compared to overactivity limited to attacks in episodic migraine (Weiller *et al*., 1995; Afridi *et al*., 2005). Functional MRI studies show a stronger connectivity in the pain matrix in chronic migraine patients than episodic migraine patients (Lee *et al*., 2019), and alterations in connectivity with the resting state with larger changes seen the higher the severity of the headache (Coppola *et al*., 2019). Results from studies using MSPA and MEG reflect increase in cortical excitability in patients with chronic migraine compared to those with episodic migraine (Aurora and Brin, 2017).

A role for nerve excitability studies in migraine?

In considering a relevant tool for clinical evaluation of migraine, the use of axonal excitability studies in peripheral nerve was explored in this study (the technique is described in the methods section below). Nerve excitability studies have been used in clinical research for over 20 years (Kiernan *et al*., 2000; Krishnan *et al*., 2009; Tomlinson *et al*., 2018). With relevance to this present study, nerve excitability studies have been shown to demonstrate changes in peripheral nerve that reflect reduction in calcium channel function in patients with Episodic Ataxia Type 2 (EA2) in whom mutations are found in the presynaptic calcium channel $Ca_v2.1$ (Tomlinson *et al*., 2016). EA2 is allelic with Familial Hemiplegic Migraine (FHM); the channel affected in EA2 and FHM is expressed both centrally and at the presynaptic neuromuscular junction. Although rare, FHM as a disease model for migraine implicates ion channel dysfunction and aberrant nerve excitability in the generation of migraine (Russell and Ducross, 2011). Nerve excitability studies in patients with EA2 show increased electrical threshold and increased response to hyperpolarisation and depolarising currents. This indicates an indirect effect of abnormal calcium current fluxes during development with the production of a calcium ion channel mutation. In the

heterogeneous population of chronic migraine patients studied in this paper, it would be expected that changes in CNS ion channel function may produce downstream effects that can be measured in ion channel populations in the peripheral nervous system but it would not be expected that excitability studies would identify single-channel dysfunction or a pathognomonic biomarker of chronic migraine. However, with the understanding that excitability studies have identified changes in other chronic CNS disorders (key findings summarised in Table 1), it is a reasonable expectation that peripheral nerve excitability studies may show the nett impact of a chronic disorder in which altered regulation of nerve excitability is a component of the pathophysiology, albeit a largely central effect.

Table 1. Nerve Excitability findings in central nervous system disorders		
Condition	**Key findings**	**Reference**
GEFS+ [1] due to SCN1B mutations	Alterations in peripheral motor axon excitability reflecting reduction in transient and persistent sodium channel conductance.	Kiernan MC *et al*., 2005b
Stroke	Modulation of HCN[2] channel activity with reduction of I_h[3] in motor nerves on the affected side	Jankelowitz *et al.,* 2007
Spinal cord injury	Changes in excitability may reflect changes in axonal structure and ion channel function. Changes were more pronounced when injuries were more clinically severe.	Lin *et al*., 2007
Multiple sclerosis	11% increase in slow K^+ channel activity in peripheral motor neurones	Ng K *et al*., 2008
Spinal cord injury	Acute changes in motor nerve excitability below the level of the lesion evolve over time. Brief improvement after stabilisation is noted before regression suggesting plasticity of expression or excitability as the injury evolves.	Boland *et al.,* 2009
Parkinson's disease	No change compared to control subjects	Jankelowitz SK and Burke D, 2012.
Episodic Ataxia Type 2	$Ca_v2.1$ dysfunction in episodic ataxia type 2 has effects on axon excitability, which may reflect an indirect effect of abnormal calcium current fluxes during development.	Tomlinson SE *et al*., 2016

[1]GEFS+ = generalised epilepsy with febrile seizures plus

[2] Hyperpolarisation activated, cyclin nucleotide gated ion channels

[3] I_h = hyperpolarisation activated conductance

Use of subcutaneous lignocaine and ketamine in chronic migraine

Treatment paradigms for acute episodic migraine are well established (Becker WJ, 2015). The benefits of preventative treatments for those with frequent episodic migraine is also well

documented (Silberstein, 2015). Despite this, at least 3% of patients with episodic migraine will convert to chronic migraine each year, with a prevalence of chronic migraine of 6.6% - 8.8% in the migraine population (Lipton *et al*., 2007; Adams *et al*., 2015) and of 1-2% in the general population (Buse *et al*., 2012); the latter figure being comparable to the prevalence of epilepsy in the general population. These patients have the highest morbidity and are the hardest to treat, with no consensus or clinical pathway for optimal treatment. The aim of treatment of chronic migraine is to convert the headache to a more manageable episodic profile, rather than aiming to 'cure' the patient of all headache. It is relevant therefore that the physiology of chronic migraine differs from acute migraine and involves entrenchment of central pathways and a lower threshold of trigeminovascular pathways to firing. With this in mind, the cohort of chronic migraine patients studied in this paper underwent a prolonged subcutaneous infusion of lignocaine and ketamine to arrest their chronic headache cycle.

Intravenous lignocaine has been shown to improve chronic migraine in patients with both migraine and analgesic overuse headache (Hand and Stark, 2000). Duration of the infusion is a key factor in long lasting benefit. Williams and Stark (2003) demonstrated a prolonged lignocaine infusion (mean 8.7 days) aborted chronic headache in 90% and removed medication overuse headache in 97% at the end of treatment with benefit enduring at six months in 70% of patients. Lignocaine is felt to stabilise excitable pathways and reduce the entrenchment of the headache cycle via sodium channel blockade.

With specific relevance to this present study, nerve excitability studies have been used to demonstrate sodium channel blockade *in vivo*. Moldovan *et al*. (2014), demonstrated a measurable effect of a locally-targeted lignocaine block of peripheral nerve *in vivo*. Lignocaine was injected

to cause local anaesthesia of the median nerve at the wrist and the nerve was then studied with nerve conduction and nerve excitability studies. The lignocaine caused rapid and complete block of motor axon conduction localized at the wrist. Within three hours, clinical assessment of power of the abductor pollicis brevis muscle had returned to normal, as had median motor nerve conduction. However, motor nerve excitability studies detected marked changes with only partial recovery at six hours and full recovery at 24 hours, illustrating the enhanced sensitivity of excitability studies in detecting changes of axonal excitability compared to nerve conduction studies. Mathematical modelling of the excitability measurements attributed the changes not only to reduction in the number of functioning voltage-gated sodium channels but also to a decrease of passive membrane resistance and an increase of capacitance. Furthermore, axonal excitability studies have been used to demonstrate sodium channel blockade in patients with acute tetrodotoxin poisoning after puffer fish ingestion (Kiernan *et al*., 2005a), defining a distinctive pattern of altered motor axons function with changes in nerve excitability reproduced in a mathematical model by a twofold reduction in sodium permeability. Thus it is reasonable to expect that a lignocaine infusion could produce a measurable effect on peripheral nerve.

The use of parenteral ketamine in chronic headache and migraine has shown at least short-term improvement (Webster and Walker 2006; Lauritsen *et al*.,2016). With reference to the reduction in central sensitization and allodynia with use of ketamine in pain conditions (Sanchez-Porras R *et al*., 2014), the mechanism is possibly due to reduced activation of affective areas of the pain processing pathways including decreased activation of the secondary somatosensory cortex, insula and anterior cingulate cortex (Sprenger *et al*., 2006). Ketamine is an agonist of N-methyl-D-aspartate (NMDA) receptors acting in the central nervous system (Craven, 2007) and in the protocol described below, ketamine provides adequate analgesia for the patient while modifying

oral medications (i.e. removing agents causative of medication overuse and introducing appropriate preventatives).

Methods

Study design

Approval for the study was obtained through St Vincent's Hospital Human Research Ethics Committee (HREC/15/SVH/356) and the University of Notre Dame Human Research Ethics Committee (017044S). Written informed consent was obtained from participants. A prospective observational cohort study was undertaken to assess peripheral axonal excitability in patients with chronic migraine before, during and after treatment with a subcutaneous lignocaine and ketamine infusion administered as part of a management plan as determined by the patient's treating neurologist. Inclusion and exclusion criteria are detailed in Table 2.

Table 2: Inclusion and Exclusion Criteria

Inclusion Criteria	Exclusion Criteria
Age 18-70	Pregnancy
Diagnosis of chronic migraine per IHS[1] criteria refractory to first line therapies	Breast feeding
Pre-treatment headache diary indicates diagnosis of transformed migraine in the 90 days prior to treatment.	Women of child bearing potential not willing to avoid pregnancy during the study timeframe
Headache refractory to conventional management	Prolonged QT or history of malignant arrhythmia
Clinician decision to prescribe infusion	Allergy to lignocaine or ketamine

[1]IHS = International Headache Society

Review of clinical state, headache diary and medications was undertaken in each assessment at four-time points: baseline (Day 0; immediately before infusion), day 5 of infusion, and at 3 and six months after infusion. Nerve excitability studies were also performed at these time points. The study was not designed to alter or direct treatment but to observe their course over time.

Infusion protocol

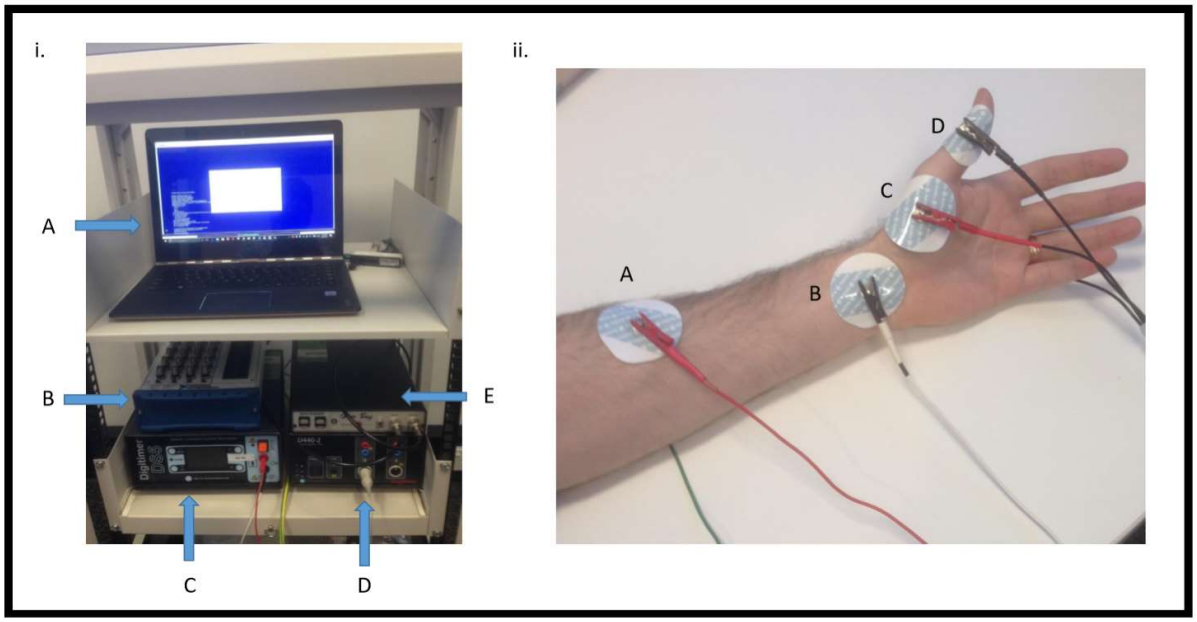
The infusion protocol is described in detail elsewhere (Rofe *et al*., 2019). To summarise, a preparation containing 1g lignocaine and 250mg ketamine was diluted in 0.9% normal saline to a total volume of 24 ml. Continuous subcutaneous infusion was delivered via a syringe driver through a 22 gauge butterfly cannula to the subcutaneous tissue of the lateral abdominal wall or outer thigh and secured by a large transparent adhesive dressing. The infusion was titrated over 24 -48 hours and continued until adequate analgesia was reached or non-efficacy was established for a mean duration of11 days (range 6 - 22 days)The target rate of infusion was 1.0 ml/hour with range of 0.5ml/hour to 1.5ml/hour depending on clinical response.

Nerve excitability studies: the TROND protocol

As with nerve conduction studies, nerve excitability studies involve stimulation of a peripheral nerve and recording of a compound muscle action potential (CMAP) or sensory nerve action potential (SNAP) in large myelinated fibres. Whereas nerve conduction studies use supramaximal stimuli to capture latency, velocity and maximal amplitude of the nerve, excitability studies use much weaker stimuli that excite a fixed fraction of axons to obtain a target response. Throughout the study, conditioning stimuli are applied to the nerve and these depolarising or hyperpolarising conditioning stimuli serve to change membrane potential. As a result, the test stimulus current required to activate the target response will be affected by polarisation and reflects the active and passive properties of the axonal membrane. . It is this change in stimulus that is then measured (Bostock *et al*., 1998; Kiernan *et al*., 2000a; Burke *et al.*, 2001) and from this measurement of change, excitability properties of the internodal membrane can be indirectly evaluated (Nodera and Kaji, 2006).

In this study, the semi-automated *TROND* protocol of axonal excitability studies, based on the principle of 'threshold tracking', was used for the assessment (Kiernan *et al*, 2000). A single study took 10-15 minutes to complete. At the start of each study, a target response was established using a stimulus-response curve the median nerve was stimulated at the wrist and the motor response recorded over the abductor pollicis brevis using non-polarizable Ag/AgCl electrodes (See Fig. 1). The *QTRAC* software (© Prof Hugh Bostock, UCL) delivered stimuli by a DS5 linear bipolar stimulator (Digitmer, UK), via a data acquisition system. The HumBug (Quest Scientific, Canada) eliminated 50 Hz interference.

Figure 1 Nerve Excitability Set up



Legend to Figure 1

i. Nerve excitability Equipment

A. Personal computer
B. Data acquisition system
C. Digitimer DS5 linear bipolar stimulator
D. D440 isolated preamplifier
E. Humbug for 50-Hz interference elimination

ii. Electrode position for median nerve motor study

A. Anode placed approx 10 cm proximal to wrist, positioned away from course of median nerve
B. Stimulating cathode at the wrist over median nerve, approximately 1cm proximal to palmar crease
C. Recording electrode over abductor pollicis brevis over the muscle belly
D. Reference electrode

The target response was 40% of the maximal response on the stimulus-response curve (identified at the steepest part of and therefore sensitive to change in threshold). The stimulus required to produce the target response is known as the 'threshold' and it is this threshold response that is tracked throughout the rest of the study. The TROND protocol obtains the following four measurements.

1. Strength–duration properties: determined by measuring the thresholds for unconditioned test stimuli of 0.2 - 1.0 ms duration. From this measurement, rheobase and the strength-duration time constant are derived using Weiss's law (Weiss, 1901; Bostock, 1983; Mogyoros *et al*., 1996). These properties are influenced by nodal persistent Na^+ currents which are active at resting membrane potential (Bostock and Rothwell., 1997).
2. Current–threshold relationship: the threshold for producing the test response is measured at the end of 200-ms conditioning currents which have strengths of between +50% (depolarising) and –100% (hyperpolarising) of the threshold stimulus. The change in threshold measured this way reflects the rectifying properties of the axon at both the nodal and internodal axolemmas. Specifically it measures outward rectification due to fast and slow K^+ channels activity induced by depolarising currents and hyperpolarisation-activated inwardly rectifying currents (I_H) activated by hyperpolarising currents.
3. Threshold electrotonus: measures the change in threshold in response to subthreshold depolarising and hyperpolarising conditioning stimuli of fixed strength (Bostock and Baker, 1988). A standard threshold electrotonus study measures the change in threshold before, during and after subthreshold conditioning stimuli of 100-ms duration which alter the potential difference across the axonal membrane. Threshold electrotonus provides insight into internodal conductances *in vivo* (Bostock *et al*., 1998; Burke *et al*., 2001)

including fast and slow K^+ channel activity, Na^+ channel activity and I_H. Subthreshold depolarising conditioning stimuli are applied at a fraction of the control threshold (+20% or +40%), increasing nerve excitability and thereby decreasing threshold. Hyperpolarising conditioning stimuli at strengths of -20%, -40%, of the target threshold serve to increase threshold and decrease the excitability of the nerve.

4. Recovery cycle: measured using a supramaximal conditioning stimulus followed by a test stimulus at varying conditioning-test intervals from 2 to 200 ms (Bostock *et al*., 1998; Kiernan *et al*., 2000). The relative refractory period and the subsequent measurements of superexcitability and late subexcitability during recovery following an action potential reflect internodal resistance pathways through and under the myelin sheath and internodal capacitance (Barrett & Barrett, 1982; Burke *et al*, 2001) Measurements are sensitive to juxta-paranodal fast K^+ channels and internodal slow K^+ channels.

Statistical analysis

Statistical analysis was performed using the QTRAC-P programme with statistical significance set at P value of less than 0.05. Data from 30 age-matched control subjects (Tomlinson *et al*., 2013) was used to perform a repeated-measures analysis of variance (ANOVA) between control data and mean data from each of three time points: baseline, Day 5 of infusion and at six months follow up. Unpaired T-tests were also performed comparing control data to each of these data. Plots of excitability measurements were generated using the QTRAC-P programme.

Results

Patient demographics and outcomes

Clinical outcome of the response to the subcutaneous lignocaine and ketamine infusion is reported elsewhere (Rofe *et al*., 2019). Fourteen patients were recruited (9 female) with a mean age of 43 years. At six months, 13 patients had sustained benefit from admission, characterised by conversion to episodic migraine rather than chronic migraine in 6/14 patients using ICHD criteria. One patient remained headache-free at the six month follow up. Improvement in chronic migraine was reported by 6.

Change in medications

Individualised care of all patients during the time frame included review of preventative and abortive medications. Medications at baseline and at six months are detailed in Appendix 1. A combination of antidepressant and anticonvulsant medications were used for headache control in 10 patients at six months. All patients had tried several first line and second line preventative medications in the past, with continuance precluded by inefficacy or intolerability. Reflecting this, second line preventative agents were often prescribed and the most frequently prescribed medications at six month follow up in addition to botulinum toxin (6) included lamotrigine (6) and the selective serotonin reuptake inhibitors (SSRI) or serotonin noradrenaline reuptake inhibitors (SNRI) antidepressants (6). Other commonly prescribed preventative agents at six months included gabapentin (4), topiramate (4) and tricyclic antidepressants (3/14). The importance of sleep restoration was reflected in prescription of quetiapine (2/14), agomelatine (2/14) and melatonin (1/14).

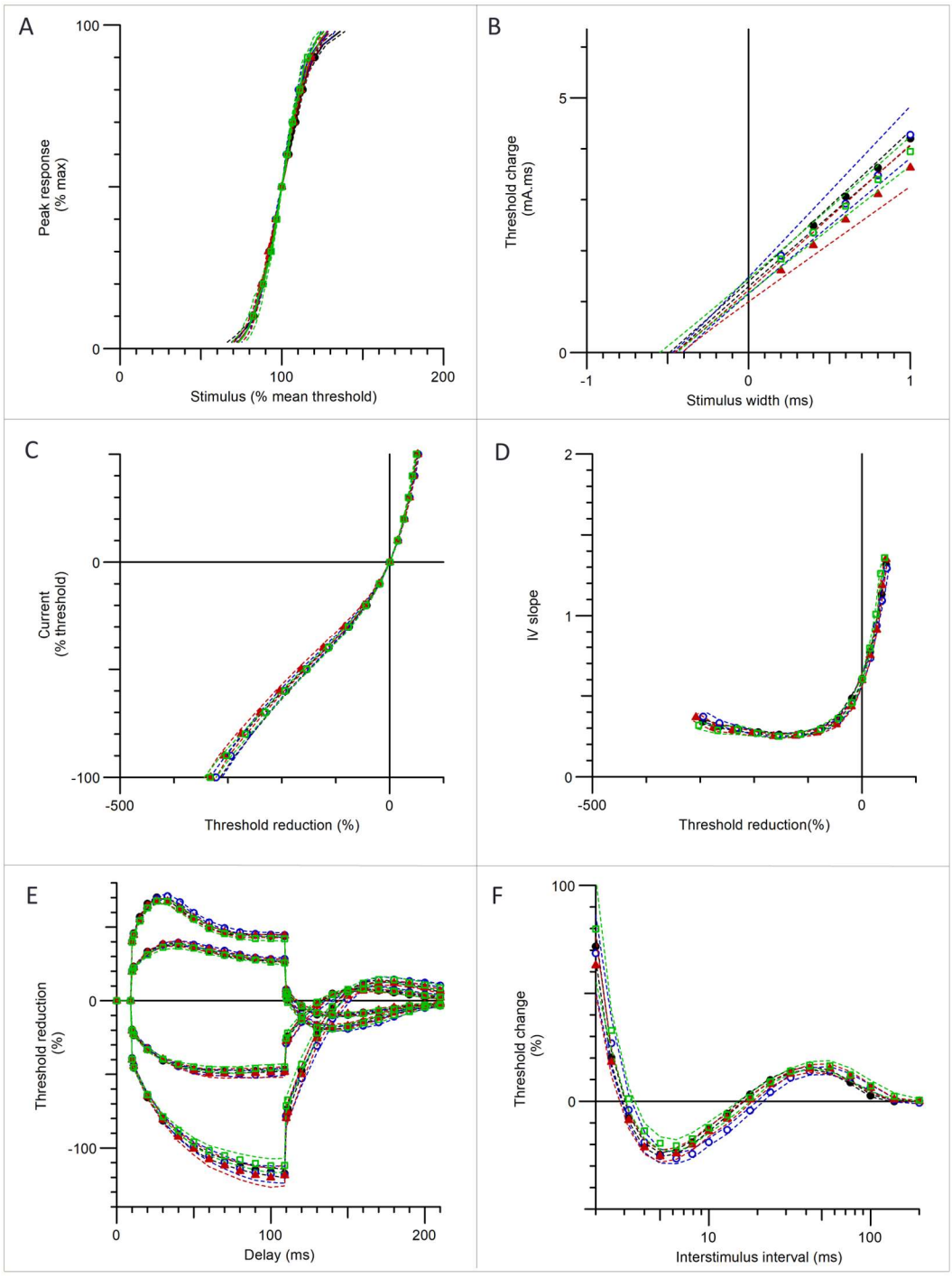
Nerve excitability studies

All 14 patients underwent nerve excitability studies at baseline (prior to infusion). Twelve patients completed excitability studies on Day 5 of infusion. Two were experiencing recalcitrant analgesic withdrawal headache on Day 5 and were disinclined to undergo studies at that time point. The study was not designed to alter patients' clinical course and, in this context, excitability studies were only performed on the 5 patients that attended at follow up at three months. Nine patients attended follow up at the six month mark and underwent studies at this time. Clinical data (including list of medications) were collected over the telephone or from chart review in those patients not attending at the three and six month time points. Statistical analysis was performed using the QTRAC-P programme with statistical significance set at P value of less than 0.05. ANOVA was used to compare differences in measurements of strength-duration, current threshold-relationship, threshold electrotonus and the recovery cycle between mean control data and mean data from each of three time points: baseline, Day 5 of infusion and at six months follow up (Appendix 2). Temperature, age and sex were also recorded. Data plotted from the 3 time points compared to normal controls are depicted in Figure 2. Unpaired T-tests were also performed comparing control data to each of these data (Appendix 2).

Controls were age matched (mean age controls 39.1 years; mean age patients 42.9 years). There was a greater proportion of women in the patient cohort (65% in patient cohort vs 50% in control group), and mean temperature was lower in the serial patient recordings (33.25 °C in control group vs 32.23°C -32.45 °C in patient groups). There was no statistically significant difference in measurements of membrane excitability attributable to chronic migraine, the impact of the lignocaine infusion, medications used to treat migraine or change in clinical state when compared to normal control data. ANOVA identified changes only in peak response and stimulus required to

produce a half-maximal response which were attributed to operator technique rather than based in physiologic differences. Occasional minor changes in single measurements seen were noted seen attributable to either operator technique (which produced variability in stimulus required for 50% CMAP and peak CMAP response) or temperature (which produced changes in rheobase and Ted10-20 in the baseline study; in Ted 40-60, accommodation half time and superexcitability at 7ms in the six month follow up study).

Figure 2: Nerve excitability Studies in Patients with Chronic Migraine



Legend to Figure 2

Black line = control (n=30); Green = patient baseline (n=14) Red = patient six months (n=9), Blue; Mean +/- standard error bars shown.

Discussion

This observational cohort study aimed to determine if nerve excitability studies could detect *in vivo* difference in chronic migraneurs compared to healthy volunteers and therefore provide a useful potential biomarker of disease. It was hypothesised that the excitability studies may detect the *in vivo* effect of the lignocaine and ketamine infusion and that a change in clinical state at six months may be reflected in change in peripheral axonal excitability. At baseline and throughout the study, all patients were taking medications that potentially impact axonal excitability via exerting effect on ion channel function or neurotransmitter activity. However, with consideration of clinical equipoise in this situation and the observational structure of the study, withdrawal of medication to study the patients at baseline off-treatment was felt beyond the scope of this project. There are no published data regarding the impact of oral anticonvulsant or antidepressant medications on peripheral axonal excitability studies.

The cohort of chronic migraine patients described here reflect the more severe end of the spectrum seen in by headache specialists, manifesting disabling symptoms and significant disruption of the normalcy of life. While not first line treatment, both lignocaine and ketamine have been described to be beneficial in migraine management and may have a role in curtailing chronic headache (Williams DJ and Stark RJ, 2003; Lauritsen *et al*.,2016). This study has not detected a change in peripheral nerve excitability in a chronic migraine population before, during and after treatment with a subcutaneous lignocaine and ketamine infusion despite clinical response in all but one patient. However, the present findings generate considerations of (i) applicability of nerve excitability studies in migraneurs, (ii) applicability of excitability studies in patients on medications which modify axonal excitability, (iii) mechanism of action of the lignocaine and

ketamine infusion and (iv) the bioavailability of lignocaine and ketamine at the level of the peripheral axon.

Nerve excitability studies in chronic migraine patients

The patients with heterogenous chronic migraine show no difference in excitability at baseline compared to healthy volunteers. Studies were performed while patients were taking neuromodulatory agents, and had been doing so for some time. It might be considered that the doses of the medications used (such as lamotrigine, sodium valproate, gabapentin and amitriptyline) were often used at lower doses than are prescribed for their other indications for their use (such as seizure disorders or depression), and that these medications might produce an effect on peripheral axonal excitability if given in larger doses. However, alterations in nerve conduction studies have been demonstrated with topiramate or sodium valproate (Freeman *et al*., 2007; Boylu *et al*., 2010) although Erdogan *et al*, 2012 did identify changes in nerve excitabilities studies attributable to topiramate. Alternatively, it might be considered that the chronic migraine patients could have a variation of peripheral axonal excitability at baseline if recorded off medication and the impact of the neuromodulatory agents prescribed for headache control serves to normalised those variations. The most likely explanation is that the peripheral axon is not a reliable biomarker of chronic migraine, in which the dominant mechanism of headache may be related to entrenchment of central pathways and is unlikely to affect peripheral nerve axonal excitability.

Applicability of excitability studies in patients on medications which modify axonal excitability

When considering the reports of the high sensitivity with which excitability studies identify detectable sodium channel blockade in nerves affected by lignocaine local injection (Moldovan *et*

al., 2014), the likely explanation for the normal studies in the patient cohort during the lignocaine infusion is that the doses used are too small when systemically distributed by subcutaneous infusion to exert impact *in vivo* in the peripheral nerve axon, rather than the lignocaine not modifying peripheral nerve excitability. The same could be postulated for the oral medications used by the patients. The lack of change in this cohort has important implications for using nerve excitability studies in the evaluation of patients with neurological disorders or medication effects, particularly where the disease mechanism or drug effect is postulated to act via dysfunction of membrane excitability (for example epilepsy, pain, neuromuscular disease). With reference to clinical equipoise, it may not be possible to withdraw neuromodulatory medications in these cohorts (especially, for example, in patients with epilepsy). However, this study finds that the oral medications prescribed (Appendix 1) do not impact nerve excitability study recordings *in vivo*. Therefore if a significant change in axonal excitability studies is identified in the study of a relevant disorder or medication, it might be better attributed to the disease process/study drug mechanism with the knowledge that the oral medications used in these patients do not translate to a significant effect.

Inference regarding mechanism of action of the lignocaine and ketamine infusion

All patients had clinical benefit from the lignocaine and ketamine infusion during the treatment in hospital with reduction of headache. However, no change was demonstrated in axonal excitability studies. While it may not be expected that ketamine produce a change in axonal excitability, demonstrable effect on nerve excitability studies with lignocaine has been documented (Moldovan *et al*., 2014). It could therefore be extrapolated that the prolonged infusion acts via a central mechanisms in stabilising the aberrant hyperexcitability in the entrenched central pathways that perpetuate chronic migraine and give patients a reduced threshold to trigger migraine.

Future directions

While this study has not demonstrated a biomarker in a heterogeneous population of chronic migraneurs on treatment, helpful observations regarding use of the nerve excitability studies in medicated patients has been documented which may be useful in future studies in migraine or other conditions. There may be a role for using axonal excitability studies in other headache cohorts in which peripheral nerve activity may be more relevant and where a more closely related nerve could be studied (e.g. trigeminal autonomic cephalgias, migraine due to genetic channelopathy). Further, there is potentially a role for use of excitability studies to be used to measure the in vivo impact of therapeutic agents if the mechanism of action is exerted by modulation of axonal excitability or membrane potential.

References

Adams AM, Serrano D, Buse DC, Reed ML, Marske V, Fanning KM, Lipton RB.

The impact of chronic migraine: the Chronic Migraine Epidemiology and Outcomes (CaMEO) Study methods and baseline results. Cephalalgia 2015; 35: 563-78

Afridi SK, Matharu MS, Lee L, Kaube H, Friston KJ, Frackowiak RS, Goadsby PJ. A PET study exploring the laterality of brainstem activation in migraine using glyceryl trinitrate. Brain 2005; 128: 932-939.

Aoki KR, Francis J. Updates on the antinociceptive mechanism hypothesis of botulinum toxin A. Parkinsonism Relat Disord. 2011; 17: S28-33.

Aurora SK, Brin MF. Chronic Migraine: An Update on Physiology, Imaging, and the Mechanism of Action of Two Available Pharmacologic Therapies. Headache 2017; 5: 109-125.

Barrett EF, Barrett JN. Intracellular recording from vertebrate myelinated axons: mechanism of the depolarising afterpotential. *J Physiol* 1982; 323: 117-144.

Becker WJ. Acute migraine treatment in adults. Headache 2015; 55: 778-793.

Boland, R.A., Bostock, H., Kiernan, M.C. Plasticity of lower limb motor axons after cervical cord injury. Clin. Neurophysiol 2009; 120: 204–209.

Bostock H, Baker M. Evidence for two types of potassium channel in human motor axons in vivo. Brain Res 1988; 462: 354-358.

Bostock H, Cikurel K, Burke D. Threshold tracking techniques in the study of human peripheral nerve. Muscle Nerve 1998; 21: 137-158.

Boylu E, Domac FM, Misirli H, *et al*.. Effects of the antiepileptic drugs on peripheral nerve function. Acta Neurol Scand 2010;121:7-10

Burstein R, Collins B, Jakubowski M. Defeating migraine pain with triptans: a race against the development of cutaneous allodynia. Ann Neurol 2004; 55: 19-26.

Burstein R, Jakubowski M. Analgesic triptan action in an animal model of intracranial pain: a race against the development of central sensitization. Ann Neurol 2004; 55: 27-36.

Burke D, Kiernan MC, Bostock H. Excitability of human axons. Clinical Neurophysiology 2001; 112: 1575-1585

Buse DC, Manack AN, Fanning KM, Serrano D, Reed ML, Turkel CC, Lipton RB. Chronic migraine prevalence, disability, and sociodemographic factors: results from the American Migraine Prevalence and Prevention Study. Headache 2012; 52: 1456-1470.

Coppola G, Di Renzo A, Petolicchio B, Tinelli E, Di Lorenzo C, Parisi V, Serrao M, Calistri V, Tardioli S, Cartocci G, Schoenen J, Caramia F, Di Piero V, Pierelli F. Aberrant interactions of cortical networks in chronic migraine: A resting-state fMRI study. Neurology 2019; 92: e2550-e2558.

Craven R. Ketamine. Journal of Anaesthesia 2007; 62: 48-53.

Dodick D, Silberstein S. Central sensitization theory of migraine: Clinical implications. Headache 2006; 46: S182-191.

Erdogan C, Yucel M, Akgun H, Kaskc T, Semai Bek V, Gokcil Z. Effect of Topiramate on peripheral nerve excitability. J Clin Neurophysiol. 2012. 29: 268–270

Freeman R, McIntosh KA, Vijapurkar U, Thienel U. Topiramate and physiologic measures of nerve function in polyneuropathy. Acta Neurol Scand 2007;115:222-231

Goadsby PJ, Holland PR, Martins-Oliveira M, Hoffmann J, Schankin C, Akerman S. Pathophysiology of Migraine: A Disorder of Sensory Processing. Physiol Rev 2017; 97: 553-622.

Hand PJ, Stark RJ. Intravenous lignocaine infusions for severe chronic daily headache. Med J Aust. 2000; 172: 157-159.

Jankelowitz SK, Howells J, Burke D. Plasticity of inwardly rectifying conductances following a corticospinal lesion in human subjects. J Physiol 2007; 581: 927-940.

Jankelowitz SK, Burke D. Do the motor manifestations of Parkinson disease alter motor axon excitability? Muscle Nerve 2012; 45: 43-47.

Kiernan MC, Burke D, Andersen KV, Bostock H. Multiple measures of axonal excitability: a new approach in clinical testing. Muscle Nerve 2000; 23: 399-409.

Kiernan, M.C., Isbister, G.K., Lin, C.S.Y., Burke, D., Bostock, H.. Acute tetrodotoxin-induced neurotoxicity after ingestion of puffer fish. Ann. Neurol 2005a; 57; 339–348.

Kiernan, M.C., Krishnan, A.V., Lin, C.S., Burke, D., Berkovic, S.F,. Mutation in the Na+ channel subunit SCN1B produces paradoxical changes in peripheral nerve excitability. Brain 2005b; 128: 1841–1846.

Krishnan AV, Lin CS, Park SB, Kiernan MC. Axonal ion channels from bench to bedside: a translational neuroscience perspective. Prog Neurobiol 2009; 89: 288-313.

Lauritsen C, Mazuera S, Lipton RB, Ashina S. Intravenous ketamine for subacute treatment of refractory chronic migraine: a case series. J Headache Pain 2016; 17: 106

Lee MJ, Park BY, Cho S, Kim ST, Park H, Chung CS. Increased connectivity of pain matrix in chronic migraine: a resting-state functional MRI study. J Headache Pain 2019; 20: 29.

Lin CS, Macefield VG, Elam M, Wallin BG, Engel S, Kiernan MC. Axonal changes in spinal cord injured patients distal to the site of injury. Brain 2007; 130: 985-994.

Lipton RB, Bigal ME, Diamond M, Freitag F, Reed ML, Stewart WF; AMPP Advisory Group. Migraine prevalence, disease burden, and the need for preventive therapy.

Neurology 2007; 68: 343-349.

Moldovan M, Lange KH, Aachmann-Andersen NJ, Kjær TW, Olsen NV, Krarup C. Transient impairment of the axolemma following regional anaesthesia by lidocaine in humans. J Physiol 2014; 592: 2735-50.

Mogyoros I, Lin CS, Kuwabara S, Cappelen-Smith C, Burke D. Strength-duration properties and their voltage dependence as measures of a threshold conductance at the node of Ranvier of single motor axons. Muscle Nerve. 2000; 23: 1719-1726.

Ng K, Howells J, Pollard JD, Burke D. Up-regulation of slow K(+) channels in peripheral motor axons: a transcriptional channelopathy in multiple sclerosis. Brain 2008; 131: 3062-3071.

Nodera H, Kaji R. Nerve excitability testing and its clinical application to neuromuscular diseases. 2006; 117: 1902-1916.

Pietrobon D. Migraine: New molecular mechanisms. Neuroscientist 2005; 11: 373-386.

Pietrobon D, Moskowitz MA. Pathophysiology of migraine. Annu Rev Physiol 2013; 75: 365-391.

Rofe CJ, Garrick R, Warhurst S, Burke D, Brew BJ, Tomlinson SE. Breaking the cycle of chronic daily headache with a low-dose subcutaneous lignocaine and ketamine infusion: a case series. Submitted to Cephalalgia August 2019, pending review.

Russell MB, Ducros A. Sporadic and familial hemiplegic migraine: pathophysiological mechanisms, clinical characteristics, diagnosis, and management. Lancet Neurol 2011; 10: 457-470.

Sanchez-Porras R, Santos E, Schöll M, Stock C, Zheng Z, Schiebel P, Orakcioglu B, Unterberg AW, Sakowitz OW. The effect of ketamine on optical and electrical characteristics of spreading depolarizations in gyrencephalic swine cortex. Neuropharmacology 2014; 84: 52–61

Silberstein SD. Preventive Migraine Treatment. Continuum 2015; 21: 973-989.

Sprenger T, Valet M, Woltmann R, Zimmer C, Freynhagen R, Kochs EF, Tölle TR, Wagner KJ. Imaging pain modulation by subanesthetic S(+)-ketamine. Anesth Analg 2006. 103: 729–737.

Tomlinson S, Burke D, Hanna M, Koltzenburg M, Bostock H. *In vivo* assessment of HCN channel current (I_h) in human motor axons. Muscle Nerve 2010; 41: 247-256.

Tomlinson SE, Tan SV, Burke D, Labrum RW, Haworth A, Gibbons VS, Sweeney MG, Griggs RC, Kullmann DM, Bostock H, Hanna MG. In vivo impact of presynaptic calcium channel dysfunction on motor axons in episodic ataxia type 2. Brain 2016; 139: 380-391.

Tomlinson SE, Howells J, Burke D. In vivo assessment of neurological channelopathies: Application of peripheral nerve excitability studies. Neuropharmacology 2018; 132: 98-107.

Webster LR, Walker MJ. Safety and efficacy of prolonged outpatient ketamine infusions for neuropathic pain. Am J Ther. 2006; 13: 300-305.

Weiller C, May A, Limmroth V Jüptner M, Kaube H, Schayck RV, Coenen HH, Diener HC. Brain stem activation in spontaneous human migraine attacks. Nat Med 1995; 1: 658-660.

Weiss G. Sur la possibilite de rendre comparables entre eux les appareils servant a l'excitation electrique. *Arch Ital Biol* 1901; 35: 413-446.

Williams DR, Stark RJ. Intravenous lignocaine (lidocaine) infusion for the treatment of chronic daily headache with substantial medication overuse. Cephalalgia 2003; 23: 963-971.

Consent

Written informed consent was obtained from the patients for publication.

Acknowledgements: This research program has been supported by a grant from St Vincent's Clinic foundation and The Brain Foundation (Australia).

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CHAPTER 4

Conclusion

The studies in this thesis have investigated in a prospective observational study whether patients suffering from severe chronic migraine and healthy age matched patients could be differentiated in terms of nerve membrane potentials via nerve excitability studies and longitudinally with therapeutic response.

A pilot study of clinical response to low dose lignocaine and ketamine subcutaneous infusion in refractory chronic migraine populations provided data that supports the use of this protocol to reduce long term migraine medication requirements. The pilot study established a high level of clinical safety and patient satisfaction with clinical outcomes although the study did not confirm that the infusions of lignocaine and ketamine were sole effective management in achieving the patient outcomes.

This outcome suggests that central desentisation may be achieved along with removal of medication overuse headache contributions to chronic migraine.

We hypothesised that peripheral nerve excitability studies that measure changes in the membrane potential of nerves may be potentially useful as biomarkers for migraine physiology and predict treatment response. We tested this hypothesis in a population of CM and compared nerve excitability responses to an age matched normal control population.

The nerve excitability studies did not identify significant alterations in peripheral ion channels following therapeutic intervention with low-dose lignocaine/ketamine infusion. Effective biomarkers in chronic migraine were not identified. Standard pain management modalities generally considered to act via sodium and calcium channel modification had no significant effect on

excitability parameters. However, significant clinical improvement did result from therapeutic interventions; the mechanism(s) for this improvement are uncertain but are likely to be independent of changes in nerve membrane potentials.

This study provides the first published data on NES in a chronic migraine cohort on medication. Thus, despite the results showing no significant differences to controls, it has important implications for other CNS diseases where differences have been found in participants, who are on medications that act via similar mechanisms.

Future studies investigating central sensitisation role in the maintenance of chronic migraine will allow new novel treatments to benefit this refractory population. There may be a role for using axonal excitability studies in other headache cohorts in which peripheral nerve activity may be more relevant and where a more closely related nerve could be studied (e.g. trigeminal autonomic cephalgias, migraine due to genetic channelopathy). Further, there is a potential role for excitability studies to measure the in-vivo impact of therapeutic agents if the mechanism of action is exerted by modulation of axonal excitability or membrane potential.

Key Points:

- This study confirms that a low does subcutaneous lignocaine and ketamine infusion is a safe management technique for patients with a severe refractory migraine and chronic migraine.
- The study fills a current gap in the literature and strengthens clinical evidence from other published data on NES's application as an investigatory tool in channelopathy disease states where unique patterns have been found.
- Presumed central desensitisation and removal of medication overuse factors in the chronic migraine can be achieved.
- Study shows the chronic migraine population on medication have similar nerve excitability profile to normal control population.
- In the presence of a clinical response, the lack of detectable change in peripheral nerve ion channel function suggest that lignocaine/ketamine infusion acts via central mechanisms in

stabilising the aberrant hyperexcitability in the entrenched central pathways that perpetuate chronic migraine and give patients a reduced threshold to trigger migraine.

- Study shows stability of nerve excitability in patients whose drug doses and drug types have been modified within standard therapeutic ranges.
- Nerve excitability studies are not suitable to be used as a biomarker for treatment responses at therapeutic doses.
- There may be a role for using axonal excitability studies in other headache cohorts in which peripheral nerve activity may be more relevant and where a more closely related nerve could be studied (e.g. trigeminal autonomic cephalgias, migraine due to genetic channelopathy).
- There is a potential role for excitability studies to measure the in-vivo impact of therapeutic agents if the mechanism of action is exerted by modulation of axonal excitability or membrane potential.

References

Adams AM, Serrano D, Buse DC, Reed ML, Marske V, Fanning KM, Lipton RB. The impact of chronic migraine: the Chronic Migraine Epidemiology and Outcomes(CaMEO) Study methods and baseline results. Cephalalgia 2015; 35: 563-78

Afridi SK, Giffin NJ, Kaube H, Goadsby PJ. A randomized controlled trial of intranasal ketamine in migraine with prolonged aura. Neurology. 2013. 80:642-647.

Afridi SK, Matharu MS, Lee L, Kaube H, Friston KJ, Frackowiak RS, Goadsby PJ. A PET study exploring the laterality of brainstem activation in migraine using glyceryl trinitrate. Brain 2005; 128: 932-939.

Anrade C. Ketamine for Depression, 2: Diagnostic and Contextual Indications. J Clin Psychiatry. 2017. 78:555-558.

Aoki KR, Francis J. Updates on the antinociceptive mechanism hypothesis of botulinum toxin A. Parkinsonism Relat Disord. 2011; 17: S28-33.

Aurora, SK and Brin, MF. Chronic Migraine: An Update on Physiology, Imaging, and the Mechanism of Action of Two Available Pharmacologic Therapies. Headache: The Journal of Head and Face Pain. 2017. 57: 109–125.

Barrett EF, Barrett JN. Intracellular recording from vertebrate myelinated axons: mechanism of the depolarising afterpotential. *J Physiol* 1982; 323: 117-144.

Becker WJ. Acute migraine treatment in adults. Headache 2015; 55: 778-793.

Bigal, M.E., J.N. Liberman, and R.B. Lipton, Age-dependent prevalence and clinical features of migraine. Neurology, 2006. 67(2): 246-51.

Bigal, M.E., *et al*., Migraine and cardiovascular disease: a population-based study. Neurology, 2010. 74(8): 628-35.

Boland, R.A., Bostock, H., Kiernan, M.C. Plasticity of lower limb motor axons after cervical cord injury. Clin. Neurophysiol 2009; 120: 204–209.

Bostock H, Baker M. Evidence for two types of potassium channel in human motor axons in vivo. Brain Res 1988; 462: 354-358.

Bostock H, Cikurel K, Burke D. Threshold tracking techniques in the study of human peripheral nerve. Muscle Nerve 1998; 21: 137-158.

Boylu E, Domac FM, Misirli H, *et al*.. Effects of the antiepileptic drugs on peripheral nerve function. Acta Neurol Scand 2010;121:7-10

Burke D, Kiernan MC, Bostock H. Excitability of human axons. Clinical Neurophysiology 2001; 112: 1575-1585

Burstein R, Collins B, Jakubowski M. Defeating migraine pain with triptans: a race against the development of cutaneous allodynia. Ann Neurol 2004; 55: 19-26.

Burstein R, Jakubowski M. Analgesic triptan action in an animal model of intracranial pain: a race against the development of central sensitization. Ann Neurol 2004; 55: 27-36.

Buse DC, Manack AN, Fanning KM, Serrano D, Reed ML, Turkel CC, Lipton RB. Chronic migraine prevalence, disability, and sociodemographic factors: Results from the American Migraine Prevalence and Prevention Study. Headache. 2012. 52:1456-1470.

Campbell-Fleming JM, Williams A. The use of ketamine as adjuvant therapy to control severe pain. Clin J Oncol Nurs. 2008 Feb;12(1): 102-7

Coppola G, Di Renzo A, Petolicchio B, Tinelli E, Di Lorenzo C, Parisi V, Serrao M, Calistri V, Tardioli S, Cartocci G, Schoenen J, Caramia F, Di Piero V, Pierelli F. Aberrant interactions of cortical networks in chronic migraine: A resting-state fMRI study. Neurology 2019; 92: e2550-e2558.

Craven R. Ketamine. Journal of Anaesthesia 2007; 62: 48-53.

DaSilva, A.F., *et al*., Thickening in the somatosensory cortex of patients with migraine. Neurology, 2007. 69(21): 1990-5

Diener HC, Limmroth V. Medication-overuse headache: a worldwide problem. Lancet Neurol. 2004. 3(8):475-483

Dodick D, Silberstein S. Central sensitization theory of migraine: Clinical implications. Headache 2006; 46: S182-191.

Erdogan C, Yucel M, Akgun H, Kaskc T, Semai Bek V, Gokcil Z. Effect of Topiramate on peripheral nerve excitability. J Clin Neurophysiol. 2012. 29: 268–270

Etchison A, Manfredi L, Mohammed M, Phan V, McAllister KB, Ray M, Heitz C. Low- Dose Intravenous Ketamine for Acute Migraine in the Emergency Department: A Randomized Placebo-Controlled Trial. Annals of emergency medicine. 2017. 70:208

Fisher K., Coderre TJ, HagenNA. Targeting the N-methyl-D-aspartate receptor for chronic pain management. Preclinical animal studies, recent clinical experience and future research directions. J Pain Symptom Manage. 2000. 20:358-373.

Freeman R, McIntosh KA, Vijapurkar U, Thienel U. Topiramate and physiologic measures of nerve function in polyneuropathy. Acta Neurol Scand 2007;115: 222-231

Garrick R., Intravenous and Subcutaneous delivery of Lignocaine and/or Ketamine for the treatment of pain. St Vincent's Private Hospital Policy and Procedure Manual 2014.

Global, regional, and national burden of neurological disorders during 1990-2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet Neurol, 2017. 16(11): 877-897.

Goadsby, P.J., Decade in review-migraine: incredible progress for an era of better migraine care. Nat Rev Neurol, 2015. 11(11): 621-2

Goadsby PJ, Holland PR, Martins-Oliveira M, Hoffmann J, Schankin C, Akerman S. Pathophysiology of Migraine: A Disorder of Sensory Processing. Physiol Rev. 2017. 97:553-622.

Goldberg ME, Domsky R, Scaringe D, Hirsh R, Dotson J, Sharaf I, Torjman MC, Schwartzman RJ. Multi-day low dose ketamine infusion for the treatment of complex regional pain syndrome. Pain Physician. 2005. 8:175–916.

Hadjikhani, N. Relevance of cortical thickness in migraine sufferers. Expert Rev

Neurother, 2008. 8(3): 327-9.

Hand PJ, Stark RJ. Intravenous lignocaine infusions for severe chronic daily headache. Med J Aust. 2000; 172: 157-159.

Headache Classification Committee of the International Headache Society. The international classification of headache disorders, 3rd edition. Cephalalgia. 2018. 38:1-211.

Hoffmann, J. and A. Recober, Migraine and triggers: post hoc ergo propter hoc? Curr Pain Headache Rep, 2013. 17(10): 370

Howells J, Trevillion L, Bostock H, Burke D. The voltage dependence of I(h) in human myelinated axons. J Physiol. 2012. 590 (7): 1625-1640

Jia, Z. and S. Yu, Grey matter alterations in migraine: A systematic review and meta-analysis. Neuroimage Clin, 2017. 14: 130-140.

Jankelowitz SK, Howells J, Burke D. Plasticity of inwardly rectifying conductances following a corticospinal lesion in human subjects. J Physiol 2007; 581: 927-940.

Jankelowitz SK, Burke D. Do the motor manifestations of Parkinson disease alter motor axon excitability? Muscle Nerve 2012; 45: 43-47.

Kaube H, Hoskin KL, and Goadsby PJ. Lignocaine and headache: an electrophysiological study in the cat with supporting clinical observations in man. J Neurol. 1994. 241:415-420

Kelman L The aura: a tertiary care study of 952 migraine patients. Cephalalgia. 2004 Sep; 24(9): 728-34.

Kiernan MC, Burke D, Andersen KV, Bostock H. Multiple measures of axonal excitability: a new approach in clinical testing. Muscle Nerve 2000; 23: 399-409.

Kiernan, M.C., Isbister, G.K., Lin, C.S.Y., Burke, D., Bostock, H.. Acute tetrodotoxin-induced neurotoxicity after ingestion of puffer fish. Ann. Neurol 2005a; 57; 339–348.

Kiernan, M.C., Krishnan, A.V., Lin, C.S., Burke, D., Berkovic, S.F,. Mutation in the Na+ channel subunit SCN1B produces paradoxical changes in peripheral nerve excitability. Brain

2005b; 128: 1841–1846.

Kors EE, Haan J, Ferrari MD. Genetics of primary headaches. Curr Opin Neurol. 1999. 12: 249–54

Krishnan AV, Lin CS, Park SB, Kiernan MC. Axonal ion channels from bench to bedside: a translational neuroscience perspective. Prog Neurobiol 2009; 89: 288-313.

Kristoffersen ES, Lundqvist C. Medication-overuse headache: a review. J Pain Res. 2014. 26 (7): 367-78.

Kvarnström A, Karlsten R, Quiding H, Emanuelsson BM, Gordh T. The effectiveness of intravenous ketamine and lidocaine on peripheral neuropathic pain. Acta Anaesthesiol Scand. 2003 Aug;47(7):868-77.

Lake AE, 3rd, Saper JR, Madden SF, Kreeger C. Comprehensive inpatient treatment for intractable migraine: a prospective long-term outcome study. Headache. 1993. 33(2): 55-62.

Lake, A.E., 3rd, J.R. Saper, and R.L. Hamel, Comprehensive inpatient treatment of refractory chronic daily headache. Headache. 2009. 49(4): 555-62.

Lai TH, Wang SJ. Update of Inpatient Treatment for Refractory Chronic Daily Headache. Curr Pain Headache Rep. 2016. 20 (1):5

Lauritsen C, Mazuera S, Lipton RB, Ashina S. Intravenous ketamine for subacute treatment of refractory chronic migraine: a case series. Headache. 2016. 17:106

Lee MJ, Park BY, Cho S, Kim ST, Park H, Chung CS. Increased connectivity of pain matrix in chronic migraine: a resting-state functional MRI study. J Headache Pain 2019; 20: 29.

Lin CS, Macefield VG, Elam M, Wallin BG, Engel S, Kiernan MC. Axonal changes in spinal cord injured patients distal to the site of injury. Brain 2007; 130: 985-994.

Lippi, G., C. Mattiuzzi, and G. Cervellin, Chocolate and migraine: the history of an ambiguous association. Acta Biomed, 2014. 85(3): 216-21

Lipton RB, Stewart WF, Diamond S, Diamond ML, Reed ML. Prevalence and burden of

migraine in the United States: Data from the American Migraine Study II. Headache. 2001; 41 (7): 646-657

Lipton RB, Scher AI, Kolodner K, *et al*. Migraine in the United States: epidemiology and patterns of health care use. Neurology 2002; 58(6): 885-894

Lipton RB, Bigal ME, Diamond M, Freitag F, Reed ML, Stewart WF. Migraine prevalence, disease burden, and the need for preventive therapy. Neurology. 2007; 68: 343-349.

Lipton RB. Direct and Indirect Costs of Chronic and Episodic Migraine in the United States: A Web-Based Survey. Headache. 2016. 56:306-322.

Lipton RB, Manack A, Buse DB, Ganning KM, Reed ML. A Comparison of the Chronic Migraine Epidemiology and Outcomes (CaMEO) Study and American Migraine Prevalence and Prevention (AMPP) study: Demographics and Headache related disability. Headache, 2016; 56 (8): 1280-1289

Lyngberg, A.C., *et al*., Incidence of primary headache: a Danish epidemiologic follow-up study. Am J Epidemiol, 2005. 161(11): 1066-73.

McGirr A, Berlim MT, Bond DJ, Fleck MP, Yatham LN, Lam RW. A systematic review and meta-analysis of randomized, double-blind, placebo-controlled trials of ketamine in the rapid treatment of major depressive episodes. Psychol Med. 2015. 45 (4): 693-704.

Marmura MJ, Goldberg SW. Inpatient management of migraine. Curr Neurol Neurosci Rep. 2015. 15 (4):13.

Marmura, M. J., Silberstein, S. D. and Schwedt, T. J., The Acute Treatment of Migraine in Adults: The American Headache Society Evidence Assessment of Migraine Pharmacotherapies. Headache: The Journal of Head and Face Pain. 2015. 55: 3-20.

May A, Schulte LH. Chronic migraine: risk factors, mechanisms and treatment. Nat Rev Neurol. 2016 Aug;12(8): 455-64.

May A. Understanding migraine as a cycling brain syndrome: reviewing the evidence from functional imaging. Ieuro Sci. 2017. 38(Supp 1): S125-S130.

Messali A, Sanderson JC, Blumenfeld AM, Goadsby PJ, Buse DC, Varon SF, Stokes M, Lipton RB. Direct and Indirect Costs of Chronic and Episodic Migraine in the United States: A Web-Based Survey. Headache. 2016. 56 (2):306-22

Moldovan M, Lange KH, Aachmann-Andersen NJ, Kjær TW, Olsen NV, Krarup C. Transient impairment of the axolemma following regional anaesthesia by lidocaine in humans. J Physiol 2014; 592: 2735-50.

Mogyoros I, Lin CS, Kuwabara S, Cappelen-Smith C, Burke D. Strength-duration properties and their voltage dependence as measures of a threshold conductance at the node of Ranvier of single motor axons. Muscle Nerve. 2000; 23: 1719-1726.

Montagna, P. Molecular Genetics of Migraine Headaches: A Review. Cephalalgia. 2000. 20 (1): 3–14.

Niesters M, Martini C, Dahan A. Ketamine for chronic pain: risks and benefits. Br J Clin Pharmacol. 2014. 77 (2): 357–367.

Neurological Disorders Collaborator Group. Global, regional, and national burden of neurological disorders during 1990-2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet Neurol. 2017. 16 (11):877-897.

Ng K, Howells J, Pollard JD, Burke D. Up-regulation of slow K(+) channels in peripheral motor axons: a transcriptional channelopathy in multiple sclerosis. Brain 2008; 131: 3062-3071.

Nodera H, Kaji R. Nerve excitability testing and its clinical application to neuromuscular diseases. 2006; 117: 1902-1916.

Pietrobon D. Migraine: New molecular mechanisms. Neuroscientist 2005; 11: 373-386.

Pietrobon D, Moskowitz MA. Pathophysiology of migraine. Annu Rev Physiol 2013; 75: 365-391.

Pomeroy JL1, Marmura MJ1, Nahas SJ1, Viscusi ER1. Ketamine Infusions for Treatment Refractory Headache. Headache. 2017 Feb; 57(2) :276-282

Rosen N, Marmura M, Abbas M, Silberstein S. Intravenous lidocaine in the treatment of refractory headache: a retrospective case series. Headache. 2009. 49 (2): 286-91.

Rossi P, Di Lorenzo C, Faroni J, Cesarino F, Nappi G. Advice alone vs. structured detoxification programmes for medication overuse headache: a prospective, randomized, open-label trial in transformed migraine patients with low medical needs. Cephalalgia. 2006. 26 (9):1097-1105

Rossi P, Faroni JV, Tassorelli C, Nappi G. Advice alone versus structured detoxification programmes for complicated medication overuse headache (MOH): a prospective, randomized, open-label trial. J Headache Pain. 2013. 14: 10-17

Russell MB, Ducros A. Sporadic and familial hemiplegic migraine: pathophysiological mechanisms, clinical characteristics, diagnosis, and management. Lancet Neurol 2011; 10: 457-470.

Sanchez-Porras R, Santos E, Schöll M, Stock C, Zheng Z, Schiebel P, Orakcioglu B, Unterberg AW, Sakowitz OW. The effect of ketamine on optical and electrical characteristics of spreading depolarizations in gyrencephalic swine cortex. Neuropharmacology. 2014. 84:52–61

Scher AI, Stewart WF, Liberman J, Lipton RB. Prevalence of frequent headache in a population sample. Headache. 1998. 38:497-506.

Schug SA, Palmer GM, Scot DA, Halliwell R, Trinca J; APM:SE Working Group of the Australian and New Zealand College of Anaesthetists and Faculty of Pain Medicine (2015), Acute Pain Management: Scientific Evidence (4th edition), ANZCA & FPM, Melbourne.

Schulte, L.H., A. Allers, and A. May, Hypothalamus as a mediator of chronic migraine: Evidence from high-resolution fMRI. Neurology, 2017. 88(21): 2011-2016.

Schulte, L.H., C. Sprenger, and A. May, Physiological brainstem mechanisms of trigeminal nociception: An fMRI study at 3T. Neuroimage, 2016. 124(Pt A): 518-525.

Shaik MM, Gan SH. Vitamin Supplementation as Possible Prophylactic Treatment against Migraine with Aura and Menstrual Migraine. BioMed Research International. 2015: 1-10

Silberstein, S.D., *et al*. Evidence-based guideline update: pharmacologic treatment for episodic migraine prevention in adults: report of the Quality Standards Subcommittee of the American Academy of Neurology and the American Headache Society. Neurology, 2012. 78(17): 1337-45

Silberstein SD. Preventive Migraine Treatment. Continuum 2015; 21: 973-989.

Sprenger, T. and D. Borsook, Migraine changes the brain: neuroimaging makes its mark. Curr Opin Neurol, 2012. 25(3): 252-62.

Sprenger T, Valet M, Woltmann R, Zimmer C, Freynhagen R, Kochs EF, Tölle TR, Wagner KJ. Imaging pain modulation by subanesthetic S(+)-ketamine. Anesth Analg. 2006. 103 (3):729–37 20.

Steiner, T.J., L.J. Stovner, and T. Vos, GBD 2015: migraine is the third cause of disability in under 50s. J Headache Pain, 2016. 17(1): 104.

Steiner, T.J., *et al*., Migraine is first cause of disability in under 50s: will health politicians now take notice? J Headache Pain, 2018. 19(1): 17.

Stewart W, Roy J, Lipton RB. Migraine prevalence, socioeconomic status, and social causation. Neurology. 2013. 81 (11): 948-955

Stewart WF. Lipton RB, Celentano DD, Reed ML. Prevalence of migraine headache in the United States. Relation to age, income, race and other sociodemographic factors. JAMA. 1992; 267: 64-69.

Stovner Lj1, Hagen K, Jensen R, Katsarava Z, Lipton R, Scher A, Steiner T, Zwart JA. The global burden of headache: a documentation of headache prevalence and disability worldwide. Cephalalgia. 2007 Mar; 27(3): 193-210.

Strassman, A.M., S.A. Raymond, and R. Burstein, Sensitization of meningeal sensory neurons and the origin of headaches. Nature, 1996. 384(6609): 560-4

Tawfic QA. A review of the use of ketamine in pain management. J Opioid Manag. 2013. 9 (5): 379-88.

Tomlinson SE, Hanna MG, Kullmann DM, Tan SV, Burke D. Clinical neurophysiology of the

episodic ataxias: Insights into ion channel dysfunction in vivo. Clinical Neurophysiology. 2009. 120 (10): 1768-1776.

Tomlinson SE, Tan SV, Burke D, Labrum RW, Haworth A, Gibbons VS, Sweeney MG, Griggs RC, Kullmann DM, Bostock H, Hanna MG. In vivo impact of presynaptic calcium channel dysfunction on motor axons in episodic ataxia type 2. Brain. 2016. 139: 380-391.

Tomlinson SE, Howells J, Burke D. In vivo assessment of neurological channelopathies: Application of peripheral nerve excitability studies. Neuropharmacology. 2018. 132: 98-107

Tomlinson S, Burke D, Hanna M, Koltzenburg M, Bostock H. *In vivo* assessment of HCN channel current (I_h) in human motor axons. Muscle Nerve 2010; 41: 247-256.

TROND Nerve Excitability Workshop. 2015 (Chicheley). H Bostock Convenor

Vetvik, K.G. and E.A. MacGregor, Sex differences in the epidemiology, clinical features, and pathophysiology of migraine. Lancet Neurol, 2017. 16: 76-87

Vos T, Flaxman AD, Naghavi M, Lozano R, Michaud C, *et al*. (2012) Years lived with disability (YLDs) for 1160 sequelae of 289 diseases and injuries 1990–2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet 380: 2163–2196

Wang SJ, Chen PK, Fuh LJ. Comorbidities of migraine. Frontiers in Neurology. 2010. 1(16): 1-9

Webster LR, Walker MJ. Safety and efficacy of prolonged outpatient ketamine infusions for neuropathic pain. Am J Ther. 2006; 13: 300-305.

Weiller C, May A, Limmroth V Jüptner M, Kaube H, Schayck RV, Coenen HH, Diener HC. Brain stem activation in spontaneous human migraine attacks. Nat Med 1995; 1: 658-660.

Weiss G. Sur la possibilite de rendre comparables entre eux les appareils servant a l'excitation electrique. *Arch Ital Biol* 1901; 35: 413-446.

Williams DR, Stark RJ. Intravenous lignocaine (lidocaine) infusion for the treatment of chronic daily headache with substantial medication overuse. Cephalalgia 2003; 23: 963-971.

Willis T, Pordage S. Two Discourses Concerning the Soul of Brutes, which Is that of the Vital and Sensitive of Man: The First Is Physiological, Shewing the Nature, Parts, Powers, And

Affections Of The Same; And The Other Is Pathological, Which Unfolds the Diseases Which Affect It and Its Primary Seat, to Wit, the Brain and Nervous Stock, and Treats of Their Cures: With Copper Cuts. London: Dring, Harper and Leigh, 1683

APPENDICES

1. St Vincent's Hospital ethical approval

St Vincent's Hospital

A facility of St Vincent's
& Mater Health Sydney

St Vincent's Hospital Sydney Ltd
ABN 77 054 038 872
390 Victoria Street
Darlinghurst NSW 2010
Australia

T + 61 2 8382 1111
F + 61 2 9332 4142
www.stvincents.com.au

11 April 2016

A/Prof Susan Tomlinson
Neurology Department
390 Victoria Street
Darlinghurst NSW 2010

Dear Susan,

SVH File Number: 15/236
Project Title: Prospective observational study examining the effectiveness of subcutaneous lignocaine and ketamine infusion in management of transformed migraine
Short Title: Management of Transformed Migraine.
HREC Reference Number: LNR/16/SVH/59

Thank you for your letter dated **25 February 2016** submitting a request to extend HREC approval to additional sites. St Vincent's Hospital HREC (EC00140) has been accredited by NSW Ministry of Health as a Lead HREC under the model for single ethical and scientific review and Certified by the NHMRC under the National Certification Scheme. This lead HREC is constituted and operates in accordance with the National Health and Medical Research Council's *National Statement on Ethical Conduct in Human Research* and the *CPMP/ICH Note for Guidance on Good Clinical Practice*. No HREC members with a conflict of interest were present for review of this project.

This project meets the requirements of the National Statement on Ethical Conduct in Human Research. I am pleased to advise that the HREC Executive at a meeting on **1 March 2016(study site approved approved following receipt of EEA on 11 April 2016)** approved this request. HREC approval has been extended to the following additional site:

- **Suite 703, Level 7, 438 Victoria Street Darlinghurst – Dr Susan Tomlinson**

Please note that only an electronic copy of this letter will be provided; if you require the original signed letter, please contact the Research Office and we will be happy to provide it.

Should you have any queries regarding this project please contact the Research Office, Tel: (02) 8382-2075, or by E-mail SVHS.Research@svha.org.au. The HREC Terms of Reference, Standard Operating Procedures, *National Statement on Ethical Conduct in Human Research* (2007) and the *CPMP/ICH Note for Guidance on Good Clinical Practice* and standard forms are available on the Research Office website that can be found at : https://svhs.org.au/home/research-education/research-office

Yours sincerely,

pp

Sarah Charlton
HREC Executive Officer
Research Office
St Vincent's Hospital
Level 6, de Lacy Building

TRIM REF: D/2016/11638

2. University of Notre Dame Cross institutional ethical approval

THE UNIVERSITY OF NOTRE DAME AUSTRALIA

19 Mouat Street (PO Box 1225) Fremantle WA 6959
+61 8 9433 0555 | enquiries@nd.edu.au

27 March 2017

Associate Professor Susan Tomlinson
& Mr Christopher Rofe

School of Medicine
The University of Notre Dame Australia
P.O Box 944
Broadway NSW 2007

Dear Susan and Christopher,

Reference Number: 017044S

Project title: "Prospective observational study examining the effectiveness of subcutaneous lignocaine and ketamine infusion in management of transformed migraine."

Thank you for submitting the above project for review. It is noted that you have ethics approval for this project from St Vincent's Hospital HREC, approval number LNR/16/SVH/59. Your application has been assessed as qualifying for a Cross-Institutional approval and is therefore exempt from HREC review. I am pleased to advise that ethical clearance has been granted for this proposed study.

Other researchers identified as working on this project are:

Name	School/Centre	Role
A/Prof Raymond Garrick	School of Medicine Sydney	Co-Supervisor
Prof Bruce Brew	School of Medicine Sydney - Adjunct	Co-Supervisor

All research projects are approved subject to standard conditions of approval. Please read the attached document for details of these conditions.

Should you have any queries about this project, please contact me at #2964 or Natalie.Giles@nd.edu.au.

Yours sincerely,

Dr Natalie Giles
Research Ethics Officer
Research Office

cc: Prof George Mendz, SRC Chair, School of Medicine Sydney

3. Patient information and consent form - Clean

Participant Information Sheet/Consent Form

Interventional Study - *Adult providing own consent*

[Insert site name]

Study Title: Prospective observational study examining the effectiveness of subcutaneous lignocaine and ketamine infusion in management of transformed migraine.

Short Title:	**Management of transformed migraine**
Protocol number:	**1**
Principal Investigator:	**A/Prof Susan Tomlinson**
Associate Investigator(s)	*[Invesitgator(s)]*
Location:	*[Location]*

Part 1: What does my participation involve?

You are being invited to take part in this research because you have a history of migraine. The study is designed to observe the effectiveness of your management. Participation in the study will not influence or direct the type of management you receive. Sometimes migraine management involves an admission to hospital for a subcutaneous infusion of medication that controls pain (lignocaine and ketamine). The research project aims to observe whether use of this infusion makes a difference in frequency or severity of migraine. If the appropriate individualized care of migraine involves admission for the infusion, your response will be measured. If you do not receive the infusion, your response will also be measured as a 'non-intervention 'subject (i.e. not receiving the treatment of interest).

This Participant Information Sheet/Consent Form tells you about the research project. It explains the tests and treatments involved. Knowing what is involved will help you decide if you want to take part in the research. Please read this information carefully. Ask questions about anything that you don't understand or want to know more about. Before deciding whether or not to take part, you might want to talk about it with a relative, friend or your local doctor.

Participation in this research is voluntary. If you don't wish to take part, you don't have to. You will receive the best possible care whether or not you take part.

If you decide you want to take part in the research project, you will be asked to sign the consent section. By signing it you are telling us that you:
• Understand what you have read
• Consent to take part in the research project
• Consent to have the tests and treatments that are described
• Consent to the use of your personal and health information as described.

You will be given a copy of this Participant Information and Consent Form to keep.

1. Introduction

Migraine is a common condition in women and can significantly affect a person's quality of life, relationships and financial situation. Over seven percent of patients can develop daily or near-daily migraine, described as chronic or transformed migraine. While not life-threatening, transformed migraine can be debilitating and the best treatment options are not clearly defined.

2. What is the purpose of this research?

Use of intravenous lignocaine has been shown to be effective in treatment of acute migraine. Ketamine is widely used for treatment of neuropathic headache. A protocol for the use of subcutaneous lignocaine and ketamine infusion for treatment of chronic pain including transformed migraine has been in place for many years at St Vincent's Private Hospital. Studies have shown that the medications can be given safely and effectively in low dose and can that a prolonged infusion (7 to 10 days) is more effective than a short infusion (ie single does) to abort the headache cycle. The protocol used at St Vincent's Private Hospital is based on treatments used in analogous pain units internationally. Anecdotally, the infusion renders great benefit for patients with transformed migraine. However, there are no published data to document this treatment as effective. Therefore, we aim to follow patients with transformed migraine to determine if the patients receiving the infusion have a better outcome.

This research has been initiated by the study doctor, A/Prof Susan Tomlinson (Neurologist, St. Vincent's Clinic), in collaboration with A/Prof Ray Garrick (St. Vincent's Clinic and Prof Bruce Brew).

3. What does participation in this research involve?

This study is aimed to observe the result of your migraine management. Your management will be tailored to your individual needs based on best practice ***and not determined by the study***. If you decide to participate in this study, we ask that you complete 9-month (270 day) period of headache monitoring under the Neurologist at *[Insert site name].*

During this time, as part of the management for migraine, your neurologist may discuss the appropriateness of an admission to St Vincent's Private Hospital for treatment with the subcutaneous lignocaine and ketamine infusion. Unfortunately this treatment is not currently available through the public health service. Therefore, only patients with adequate private health cover will be able to receive the infusion, which is currently the case in standard clinical care. Once the infusion has taken place, you will be asked to complete a further 180-day (6 month) headache diary so we can evaluate the outcome of your treatment.

If no infusion is advisable, you will be eligible to participate in the study as a non-intervention participant and will complete the 9 month surveillance period while using your standard migraine treatment.

As per standard practice, you will also require a follow-up appointment with your Neurologist approximately 3 months and 6 months after your infusion or after you commence participation in the non-intervention group. There is no additional cost to you for participation, other than that which would normally be incurred as part of standard management.

4. What do I have to do?

Your involvement will involve four short research visits. The information collected at each visit will include your medical history and investigation results, clinical examination findings and medication use. A short questionnaire will be used to standardize measurement of treatment response. Between visits, you will keep a simple headache diary which is frequently used in the clinical setting.
In addition to the clinical assessment and questionnaire, each study visit will involve a brief electrical test of the nerves in your forearm. This test is called a nerve excitability study and involves stimulating the median nerve in the wrist with short electrical pulses. The electrical pulses last milliseconds only. They may be mildly uncomfortable but there are not long term side effects. The test can be stopped at any time, should you require. Nerve excitability studies are a research tool that assesses how the infusion settles the nerve during treatment. The nerve excitability studies will be performed on 4 occasions during 9 months: at baseline, during the admission for infusion (Day 5) or at 90 days after initial assessment, then repeated at 3 and six months after the infusion or at 3 and six months after the second assessment.

5. Other relevant information

It is anticipated that approximately 40 people will complete this study. Information about your response to your treatment will be analysed. Two groups will be observed including are those whose standard care involves no infusion for their migraine, are those who receive the infusion. All participants will be seen by their Neurologist at *[Insert site name].*over a minimum of four appointments.

6. Do I have to take part in this project?

Participation in any research project is voluntary. If you do not wish to take part, you do not have to. If you decide to take part and later change your mind, you are free to withdraw from the project at any stage. If you do decide to take part, you will be given this Participant Information and Consent Form to sign and you will be given a copy to keep. Your decision whether to take part or not to take part, or to take part and then withdraw, will not affect your routine treatment, your relationship with those treating you or your relationship with *[Insert site name].*

7. What are the alternatives to participation

You will be offered the standard of care for your migraine treatment, including other migraine-preventing drugs, regardless of whether you participate in the study. Your study doctor will discuss these options of best practice with you before you decide whether or not to take part in this research project.

8. What are the possible benefits of taking part?

We cannot guarantee or promise that you will receive any direct benefits from this research.

9. What are the possible risks of taking part?

There are no risks associated with this study because the study is designed to observe your journey, not to prescribe specific treatments. Any risk of migraine management relates to the individual therapies, which will only be prescribed after full discussion with you of the relevant risks, benefits and alternatives, in keeping with best practice and standard clinical care. You will be provided will information regarding all the appropriate treatment modalities.

10. What if new information arises during this research project?

Sometimes during the course of a research project, new information becomes available about the treatment that is being studied. If this happens, your study doctor will tell you about it and discuss with you whether you want to continue in the research project. If you decide to withdraw, your study doctor will make arrangements for your regular health care to continue.

withdraw, your study doctor will make arrangements for your regular health care to continue. If you decide to continue in the research project you will be asked to sign an updated consent form. On receiving new information, your study doctor might consider it to be in your best interests to withdraw you from the research project. If this happens, he/ she will explain the reasons and arrange for your regular health care to continue.

11. Can I have other treatments during this research project?

This is an observational study only. Any limitations on other treatments will be directed by your neurologist, and according to the treatments chosen as best appropriate for you. Participation in this study will not prevent you from using medications that may help your migraine management when indicated. It is important to tell your study doctor and the study staff about any treatments or medications you may be taking, including over-the-counter medications, vitamins or herbal remedies, acupuncture or other alternative treatments. You should also tell your study doctor about any changes to these during your participation in the research project.

12. What if I withdraw from this research project?

If you decide to withdraw from the project, please notify a member of the research team. This notice will allow that person or the research supervisor to discuss any health risks or special requirements linked to withdrawing.

If you do withdraw your consent during the research project, the study doctor and relevant study staff will not collect additional personal information from you, although personal information already collected will be retained to ensure that the results of the research project can be measured properly and to comply with law. You should be aware that data collected by the investigators up to the time you withdraw will form part of the research project results. If you do not want them to do this, you must tell them before you join the research project.

13. Could this research project be stopped unexpectedly?

It is unlikely that this would happen. However, this will not impact your medical care. You will be informed if the study is stopped.

14. What happens when this research project ends?

You will continue to receive the appropriate management by your treating doctors as clinically indicated.

Part 2 How is this research project being conducted?

15. What will happen to information about me?

By signing the consent form you consent to the study doctor and relevant research staff collecting and using personal information about you for the research project. Any information obtained in connection with this research project that can identify you will remain confidential and be stored securely. Your information will only be used for the purpose of this research project and it will only be disclosed with your permission, except as required by law.

Information about you may be obtained from your health records held at this and other health services for the purpose of this research. By signing the consent form you agree to the study team accessing health records if they are relevant to your participation in this research project.

It is anticipated that the results of this research project will be published and/or presented in a variety of forums. In any publication and/or presentation, information will be provided in such a way that you cannot be identified, except with your permission. All information about

participants in the study will be presented as group means and descriptive statistics, such that it will be impossible to identify a particular participant in any way.

Information about your participation in this research project may be recorded in your health records.

In accordance with relevant Australian and NSW privacy and other relevant laws, you have the right to request access to your information collected and stored by the research team. You also have the right to request that any information with which you disagree be corrected. Please contact the study team member named at the end of this document if you would like to access your information.

Any information obtained for the purpose of this research project that can identify you will be treated as confidential and securely stored. It will be disclosed only with your permission, or as required by law.

16 Complaints and compensation

If you suffer any injuries or complications as a result of this research project, you should contact the study team as soon as possible and you will be assisted with arranging appropriate medical treatment. If you are eligible for Medicare, you can receive any medical treatment required to treat the injury or complication, free of charge, as a public patient in any Australian public hospital.

18 Who is organising and funding the research?

This research project is being conducted by A/Prof Susan Tomlinson (Neurologist, St. Vincent's Clinic). No member of the research team will receive a personal financial benefit from your involvement in this research project (other than their ordinary wages). The Neurologists involved in the study and St. Vincent's Clinic/Hospital have no conflicts of interest with regard to this research. The study is supported by a grant from the St Vincent's Clinic Research Foundation.

19 Who has reviewed the research project?

All research in Australia involving humans is reviewed by an independent group of people called a Human Research Ethics Committee (HREC). The ethical aspects of this research project have been approved by the HREC of St Vincent's Hospital, Sydney.

This project will be carried out according to the *National Statement on Ethical Conduct in Human Research (2007)*. This statement has been developed to protect the interests of people who agree to participate in human research studies.

The study was peer reviewed as part of the process of application for the St Vincent's Clinic Foundation Grant Application Process.

20 Further information and who to contact

The person you may need to contact will depend on the nature of your query. If you want any further information concerning this project or if you have any medical problems which may be related to your involvement in the project (for example, any side effects), you can contact the principal study doctor, A/Prof Susan Tomlinson on 8382 6712 or any of the following people:

Clinical contact person

Name	*A/Prof Susan Tomlinson*
Position	*Neurologist*
Telephone	*83826712*
Email	*sydheadache@svha.com.au*

For matters relating to research at the site at which you are participating, the details of the local site complaints person are:

Complaints contact person

Position	*Research Office Manager*
Telephone	02 8382 2075
Email	SVHS.Research@svha.org.au

If you have any complaints about any aspect of the project, the way it is being conducted or any questions about being a research participant in general, then you may contact:

Reviewing HREC approving this research and HREC Executive Officer details

Reviewing HREC name	St Vincent's Hospital HREC
HREC Executive Officer	Executive Officer
Telephone	02 8382 2075
Email	SVHS.Research@svha.org.au

Consent Form - *Adult providing own consent*

tle	**Prospective observational study examining the effectiveness of subcutaneous lignocaine and k infusion**
ıort Title	Management of Transformed Migraine
otocol Number	1
oject Sponsor	None
ordinating Principal vestigator/ Principal Investigator	A/Prof Susan Tomlinson
sociate Investigator(s)	*[Investigator(s)]*
cation	*[Location]*

Declaration by Participant

I have read the Participant Information Sheet or someone has read it to me in a language that I understand.

I understand the purposes, procedures and risks of the research described in the project.

I give permission for my doctors, other health professionals, hospitals or laboratories outside this hospital to release information to St Vincent's Clinic, concerning my disease and treatment for the purposes of this project. I understand that such information will remain confidential.

I have had an opportunity to ask questions and I am satisfied with the answers I have received.

I freely agree to participate in this research project as described and understand that I am free to withdraw at any time during the study without affecting my future health care.

I understand that I will be given a signed copy of this document to keep.

Name of Participant (please print) _____

Signature _____ Date _____

Declaration by Study Doctor/Senior Researcher†

I have given a verbal explanation of the research project, its procedures and risks and I believe that the participant has understood that explanation.

Name of Study Doctor/
Senior Researcher† (please print) _____

Signature _____ Date _____

† A senior member of the research team must provide the explanation of, and information concerning, the research project.

Note: All parties signing the consent section must date their own signature.

Form for Withdrawal of Participation - *Adult providing own consent*

Title	**Prospective observational study e the effectiveness of subcutaneous and ketamine infusion in management of transformed mi**
Short Title	Management of Transformed Migraine
Protocol Number	1
Project Sponsor	None
Coordinating Principal Investigator/ Principal Investigator	A/Prof Susan Tomlinson
Associate Investigator(s)	*[Investigator(s)]*
Location	*[Location]*

Declaration by Participant

I wish to withdraw from participation in the above research project and understand that such withdrawal will not affect my routine treatment, my relationship with those treating me or my relationship with St. Vincent's Clinic, St. Vincent's Hospital or my treating doctor.

Name of Participant (please print) _____

Signature _____ Date _____

Circumstances for withdrawal (if given verbally)

Declaration by Study Doctor/Senior Researcher†

I have given a verbal explanation of the implications of withdrawal from the research project and I believe that the participant has understood that explanation.

Name of Study Doctor/
Senior Researcher† (please print) _____

Signature _____ Date _____

† A senior member of the research team must provide the explanation of and information concerning withdrawal from the research project.

Note: All parties signing the consent section must date their own signature.

4. Migraine Disability and Assessment Score (MIDAS)

Appendix D: MIDAS Questionnaire

Participant Number: _____ Date: _____

Research Visit Number (1/2/3/4): _____

INSTRUCTIONS • Please answers the following questions about ALL your headaches you have had over the last 3 months. Select your answer in the box next to each question. If a single headache affects more than one area of your life (e.g., work and family life) it is counted more than once. Select zero if you did not have the activity in the last 3 months.

1. On how many days in the last 3 months did you miss work or school because of your headaches?	days
2. How many days in the last 3 months was your productivity at work or school reduced by half or more because of your headaches? (Do not include days you counted in question 1 where you missed work or school).	days
3. On how many days in the last 3 months did you not do household work because of your headaches?	days
4. How many days in the last 3 months was your productivity in household work reduced by half or more because of your headaches? (Do not include days you counted in question 3 where you did not do household work).	days
5. On how many days in the last 3 months did you miss family, social, or leisure activities because of your headaches?	days
A. On how many days in the last 3 months did you have any headache? (If a headache lasted more than 1 day, count each day.)	days
B. On a scale of 0-10, on average, how painful were these headaches?	days

From: Stewart, W. F., Lipton, R. B., Dowson, A. J., & Sawyer, J. (2001). Development and testing of the Migraine Disability Assessment (MIDAS) Questionnaire to assess headache-related disability. Neurology, 56(suppl 1), S20-S28.

5. Headache Diary

Insert Header with institution's name or institution's letterhead

HEADACHE DIARY

Participant Number: .....
Date Started: ..... Date Completed: .....

Contact syd.headache@svha.org.au if you have any questions or to submit this diary electronically.

Tick which time period this diary applies to:

- ☐ 90 days (3 months) before intervention
- ☐ 90 days (3 months) after intervention
- ☐ 90-180 days (3-6 months) after intervention

1	2	3	4	5	6	7
8	9	10	11	12	13	14
15	16	17	18	19	20	21
22	23	24	25	26	27	28
29	30	31				

Instructions: Write 'M' on the date of migraine;
'H' on the days of headaches other than migraine.
Circle the number on days of bleeding/menstruation (even if spotting/irregular).
Write in tablets taken for headache and migraine.

6. Nerve excitability studies configuration

Figure 6.1: Nerve Excitability equipment

Hardware:

16-bit data acquisition Analogue to digital system (National Scientific)

DS5 linear constant-current bipolar stimulator (Digitimer)

D440-2 amplifier (Digitimer)

Humbug 50/60 Hz eliminator (Quest scientific)

Laptop with QtracS stimulation software (© Professor H Bostock, University College London)

Peripheral cables and disposable electrodes

Tests were performed on the participant's median nerve with six surface electrodes (per Figure 6.2 set up). Compound action potentials were recorded along the abductor pollicis brevis after stimulation of the median nerve near the wrist. Current was delivered from DS5 stimulators and controlled through the QtracS stimulation software following the TRONDNF protocol. Recording of compound action potentials were measured through the D440 amplifier and then routed through a humbug to remove background noise.

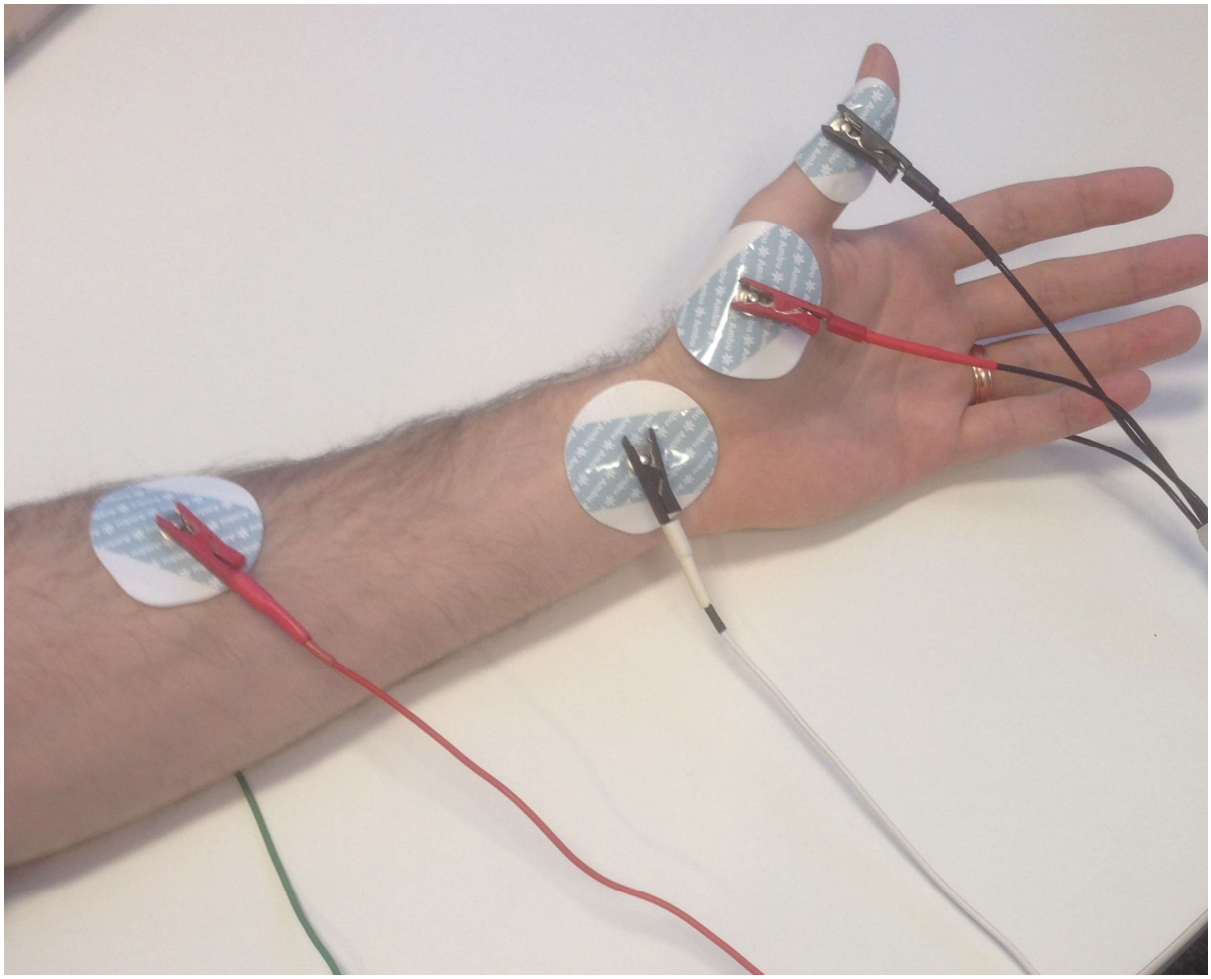

Figure 6.2: Electrode placement

(Note: Two electrodes are also located on the back of the hand and on the forearm and are attached with earth cables)

7. Medical History Worksheet

Insert Header with institution's name or institution's letterhead

Appendix B: Templates for Enrolment Visit and Follow-Up Visits

Checklist for Enrolment Visit History and Examination

Participant Number: Date Completed:

Migraine History

1. Age at onset
2. Accompanying symptoms i.e. nausea, vomiting, photophobia, phonophobia
3. Aura
4. Frequency
5. Lateralisation
6. Duration of headaches
7. Severity and quality of headache
8. Triggers e.g. stress, fatigue, odours, missing meal, weather, exertion, dehydration
9. Relationship with activities and movements
10. Relationship with hormonal cycle i.e. during periods, other times of cycle
11. Investigations performed for headache/migraine

Medications for Migraine

12. Medications used for migraine prophylaxis
13. Medications used for migraine symptomatic control

Concurrent Headache History

14. Analgesia rebound
15. Cervicogenic headache
16. Chronic daily headache or transformed migraine

Intercurrent Medical History

17. Other medications and doses
18. Any other medical conditions (e.g. cancer/IHD, auto-immune diseases, HIV)
19. Diabetes
20. History of other neurological conditions/stroke (Y/N)
21. Any history of head trauma?
22. Psychiatric history
23. Any systemic symptoms e.g. fevers, chills, anorexia and weight loss

Social History

24. Current Smoker (Y/N); Pack years
25. Drink alcohol currently? (Y/N); Standard drinks in average week
26. Other illicit drug use
27. Nutrition and physical activity

Family History

28. Family history of migraine and menstrual migraine (Y/N). If yes, please detail
29. Any Hx of coagulopathy or thromboembolitic events

Examination

Vitals : BP, RR, 02 Saturation, Temp

30. Cranial nerve examination I-XII (normal/abnormal)
31. Including fundoscopy (Normal/abnormal) If abnormal, detail abnormalities
32.

Nerve Excitability Studies

8. Medication summary of participants

Appendix 8 lists the headache medication that the participant was taking at each respective timepoint.

Table Summary of Medication changes		
Patient 1	**Baseline**	**6 months**
PREVENTIVE AGENTS	Amytriptyline 50 mg Sodium valproate 200mg bd	Amytriptyline 50mg Meloxicam 15mg Verapamil 40mg bd Gabapentin 600 bd Botulinum toxin
ABORTIVE AGENTS	Ibuprofen Codeine phosphate	
Patient 2	**Baseline**	**6 months**
PREVENTIVE AGENTS	Amytriptyline 50 Topiramate 50 Codeine phosphate	Sodium valproate 200mg bd
ABORTIVE AGENTS	--	--
Patient 3	**Baseline**	**6 months**
PREVENTIVE AGENTS	Gabapentin 200mg mane, 300mg nocte Lamotrigine 100mg bd Duloxetine 120mg Baclofen 5mg mane Botulinum toxin	Gabapentin 200mg bd Lamotrigine 50mg bd Duloxetine 120mg Botulinum toxin Topiramate 50mg bd
ABORTIVE AGENTS	Paracetemol Celecoxib Rizatriptan Diazepam	Paracetamol Celecoxib Rizatriptan Diazepam Sub occcipital blocks
Patient 4	**Baseline**	**6 months**
PREVENTIVE AGENTS	Zonisamide Botulinum toxin Lamotrigine 200mg daily Duloxetine 120mg	Propranolol 10mg bd Botulinum toxin Lamotrigine 400mg daily Duloxetine 180mg
ABORTIVE AGENTS	--	Meloxicam 15mg daily Sub occciptial blocks
Patient 5	**Baseline**	**6 months**

PREVENTIVE AGENTS	Duloxetine 90mg Agomelatine 25 mg Botulinum toxin	Venlafaxine 75mg mane Lamotrigine 75mg mane, 100mg nocte Quetiapine 25mg nocte Botulinum toxin
ABORTIVE AGENTS	Naproxyn 250mg	Naproxyn 250mg
Patient 6	**Baseline**	**6 months**
PREVENTIVE AGENTS	Topiramate 50 Verapamil 40mg bd Zolpidem	Zonisamide 25mg Doxepin 25mg nocte
ABORTIVE AGENTS	--	Subocciptal blocks
Patient 7	**Baseline**	**6 months**
PREVENTIVE AGENTS	Stopped diazepam and codeine prior to admission	Agomelatine 50mg nocte Duloxetine 120mg daily Topiramate 100mg nocte
ABORTIVE AGENTS	--	Naproxen 200mg
Patient 8	**Baseline**	**6 months**
PREVENTIVE AGENTS	--	Melatonin 2mg Gabapentin 300mg daily
ABORTIVE AGENTS	--	Metaclopramide 10mg Naproxen 200mg Rizatriptan 10mg
Patient 9	**Baseline**	**6 months**
PREVENTIVE AGENTS	--	Magnesium 300mg bd Quetiapine 25mg Riboflavin 400mg Zonisamide 50mg mane 100mg nocte Amitriptyline 37.5mg nocte Botulinum toxin
ABORTIVE AGENTS	Rizatriptan 3-4x/week	
Patient 10	**Baseline**	**6 months**
PREVENTIVE AGENTS	Sodium valproate 1g mane, 500mg nocte Lamotrigine 100mg mane Vitamin B2 400mg daily	Sodium valproate 1g mane, 500mg nocte Lamotrigine 100mg mane Vitamin B2 400mg daily Verapamil 40mg tds,
ABORTIVE AGENTS	Maxalt 10mg prn Ondansetron 4mg prn Clonazepam 0.5mg prn	
Patient 11	**Baseline**	**6 months**

PREVENTIVE AGENTS	Gabapentin 300mg tds Zonisamide Sub occipital blocks	Gabapentin 400mg bd Amitriptyline 150 Wean Zonisamide Sub occipital blocks
ABORTIVE AGENTS	--	--
Patient 12	**Baseline**	**6 months**
PREVENTIVE AGENTS	Topiramate 50mg mane and 100mg nocte Mexiletine 200mg bd Oxytocin 60units bd Magnesium vitamin B2 400mg daily	Topiramate 50mg nocte Lamotrigine 100mg bd Botulinum toxin
ABORTIVE AGENTS	Parecoxib IMI 40mg, Naratriptan Codeine phosphate 30mg daily	Parecoxib IMI 40mg, Occasional codeine phosphate Naratriptan
Patient 13	**Baseline**	**6 months**
PREVENTIVE AGENTS	Lamotrigine 100mg bd Agomelatine 50mg nocte Amitriptyline 50mg nocte Botulinum toxin	Lamotrigine 100mg bd Agomelatine 50mg nocte Vortioxetine 15mg mane Tapentadol SR 50mg prn Naproxyn 250mg prn Metaclopramide 10mg po
ABORTIVE AGENTS	--	--
Patient 14	**Baseline**	**6 months**
PREVENTIVE AGENTS	Topiramate 50mg Sertraline 50mg	Topiramate 25mg nocte Gabapentin 200mg tds
ABORTIVE AGENTS	--	--

9. Measurements used for analysis of Nerve Excitability Studies

Measurement	Definition
Strength-duration relationship (Figure 2B)	
Strength-duration time constant: SDTC	Estimated from the negative intercept on the X-axis of the plot of stimulus charge *v.* stimulus duration (Fig. 2B)
Current/threshold relationship (Figure 2C and 2D)	
Resting I/V slope	The slope of the current-threshold relationship in Fig2C. calculated from the polarising currents -10% and +10% of threshold (see Fig. 2D)
Minimum I/V slope	Minimal slope of the curve in Fig. 2C
Hyperpolarising I/V slope	The leftmost point in Fig. 2D
Threshold electrotonus (Figure 2E)	
Ted	Change in threshold in response to a subthreshold depolarising conditioning stimulus
TEd^{20}	Threshold electrotonus in response to a subthreshold depolarising conditioning stimulus which is 20% of threshold stimulus
TEd^{40}	Threshold electrotonus in response to a subthreshold depolarising conditioning stimulus which is 40% of threshold stimulus
TEd^{20}(peak)	Peak % reduction in threshold during depolarising currents set to 20% of the resting threshold ‡
TEd^{40}(peak)	Peak % reduction in threshold during depolarising currents set to 40% of the resting threshold ‡
TEd^{40}(90-100 ms)	Mean % threshold reductions between the specified latencies for the 40% depolarising current
TEd^{40}(undershoot)	Minimal % threshold reduction after the 100 ms depolarising current ‡
TEd^{40}(accom)	Maximal drop from TEd^{40}(peak) during 100 ms depolarisation ‡
TEh	Change in threshold in response to a subthreshold hyperpolarising conditioning stimulus
TEh^{40}(90-100 ms)	As TEd^{40}(90-100 ms) but hyperpolarising
TEh^{40}(90-100 ms)	As TEh^{40}(90-100 ms) but during 20% hyperpolarising current
TEh^{40}(overshoot)	Maximal % threshold reduction after the 100 ms hyperpolarisation ‡
Accommodation half time	Half-time of accommodative response to a 100 ms subthreshold depolarising conditioning stimulus
Recovery Cycle (Figure 2F)	
Relative refractory period (RRP)	Interstimulus interval at which threshold first returns to normal
Superexcitability	Maximal % threshold reduction †
Late Subexcitability	Maximal % threshold increase after 10 ms †

† = measurements averaged over 3 adjacent points; ‡ = measurements averaged over 20 ms

i. ANOVA comparing control data (n=30) with patient data at baseline (n=14); day 5 of the lignocaine and ketamine infusion (n=12); and at six months follow up (n=9).

1. Stimulus (mA) for 50% m	F=4.425(3, 61)	p=0.00712**
3. Strength-duration\time	F=0.562(3, 61)	p=0.6461
4. Rheobase (mA)	F=2.717(3, 61)	p=0.05152
5. Stimulus-response\slope	F=0.641(3, 58)	p=0.59568
6. Peak response\(mv)	F=3.172(3, 59)	p=0.03033*
7. Resting I/V slope	F=0.407(3, 60)	p=0.75206
8. Minimum I/V slope	F=0.176(3, 60)	p=0.91001
9. Temperature (C)	F=2.928(3, 59)	p=0.04039*
10. RRP (ms)	F=0.461(3, 60)	p=0.71454
11. TEh(90-100ms)	F=0.161(3, 61)	p=0.91909
12. TEd(10-20ms)	F=1.627(3, 61)	p=0.19103
13. Superexcitability (%)	F=1.238(3, 61)	p=0.30346
14. Subexcitability (%)	F=0.264(3, 61)	p=0.85147
17. Age (years)	F=1.132(3, 40)	p=0.34805
18. Sex (M=1, F=2)	F=0.738(3, 60)	p=0.53692
19. Latency (ms)	F=0.054(3, 61)	p=0.97766
20. TEd(40-60ms)	F=2.161(3, 61)	p=0.10043
21. TEd(90-100ms)	F=0.542(3, 61)	p=0.65952

22. TEh(10-20ms)	F=1.022(3, 61)	p=0.39035
23. TEd(undershoot)	F=0.417(3, 61)	p=0.74473
24. TEh(overshoot)	F=1.416(3, 61)	p=0.24573
25. TEd(peak)	F=1.753(3, 61)	p=0.1641
26. S2 accommodation	F=1.09(3, 61)	p=0.36096
27. Accommodation half-tim	F=2.637(3, 61)	p=0.0567
28. Hyperpol. I/V slope	F=0.939(3, 60)	p=0.42912
29. Refractoriness at 2.5m	F=0.382(3, 60)	p=0.7691
30. TEh(20-40ms)	F=0.465(3, 61)	p=0.71175
31. TEh(slope 101-140ms)	F=0.28(3, 61)	p=0.84062
32. Refractoriness at 2 ms	F=0.261(3, 51)	p=0.8536
33. Superexcitability at 7	F=1.436(3, 60)	p=0.24021
34. Superexcitability at 5	F=0.895(3, 60)	p=0.45105
35. TEd20(peak)	F=0.575(3, 61)	p=0.63742
36. TEd40(Accom)	F=0.404(3, 42)	p=0.75387

ii. Unpaired t-test: comparing control data to baseline study in 14 patients with chronic migraine.

Variable	Mean+/-SE(n)	Mean+/-SE(n)	t(df)	p
1. Stimulus (mA) for 50% m	4.287x|/1.04(30)	3.364x|/1.08(14	t=3.235(42)	p=0.00247**
3. Strength-duration\time	0.4807 ± 0.0184(30)	0.4439 ± 0.0151(14)	t=1.273(42)	p=0.2074
4. Rheobase (mA)	2.796x|/1.04(30)	2.266x|/1.09(14)	t=2.611(42)	p=0.01201*
5. Stimulus-response\slope	5.128x|/1.04(30)	5.371x|/1.07(14)	t=0.598(42)	p=0.56039
6. Peak response\(mv)	8.847x|/1.06(30)	7.249x|/1.12(14)	t=1.688(42)	p=0.09505
7. Resting I/V slope	0.6071 ± 0.0142(30)	0.6262 ± 0.0304(13)	t=0.652(41)	p=0.52533
8. Minimum I/V slope	0.2462 ± 0.008(30)	0.2448 ± 0.011(13)	t=0.103(41)	p=0.88212
9. Temperature (C)	33.25 ± 0.17(30)	32.45 ± 0.283(12)	t=2.48(40)	p=0.01666*
10. RRP (ms)	2.953x|/1.02(30)	3.017x|/1.04(13)	t=0.519(41)	p=0.61237
11. TEh(90-100ms)	-116.7 ± 2.77(30)	-114.2 ± 4.83(14)	t=0.486(42)	p=0.63436
12. TEd(10-20ms)	68.69 ± 0.744(30)	65.91 ± 1.14(14)	t=2.079(42)	p=0.04148*
13. Superexcitability (%)	-23.05 ± 0.926(30)	-20.96 ± 2.47(14)	t=0.969(42)	p=0.34032
14. Subexcitability (%)	14.4 ± 0.655(30)	14.24 ± 1.47(14)	t=0.118(42)	p=0.87344
17. Age (years)	39.1 ± 2.4(30)	42.89 ± 4.23(9)	t=0.763(37)	p=0.45629
18. Sex (M=1, F=2)	1.467 ± 0.0926(30)	1.615 ± 0.14(13)	t=0.883(41)	p=0.38619
19. Latency (ms)	6.468 ± 0.114(30)	6.419 ± 0.257(14)	t=0.203(42)	p=0.82147
20. TEd(40-60ms)	50.66 ± 0.667(30)	49.99 ± 0.884(14)	t=0.58(42)	p=0.57188
21. TEd(90-100ms)	43.96 ± 0.663(30)	43.18 ± 0.962(14)	t=0.662(42)	p=0.51842
22. TEh(10-20ms)	-73.55 ± 0.732(30)	-72.72 ± 1.18(14)	t=0.621(42)	p=0.54542
23. TEd(undershoot)	-18.78 ± 0.604(30)	-17.8 ± 1.23(14)	t=0.808(42)	p=0.42909
24. TEh(overshoot)	14.06 ± 0.597(30)	12.16 ± 1.33(14)	t=1.511(42)	p=0.13431

25. TEd(peak)	68.17 ± 0.696(30)	65.68 ± 1.08(14)	t=1.985(42)	p=0.05098
26. S2 accommodation	24.21 ± 0.528(30)	22.49 ± 1.17(14)	t=1.552(42)	p=0.12423
27. Accommodation half-tim	40.1 ± 0.777(30)	41.62 ± 0.998(14)	t=1.146(42)	p=0.25725
28. Hyperpol. I/V slope	0.3414 ± 0.0105(30)	0.3785 ± 0.023(13)	t=1.69(41)	p=0.09488
29. Refractoriness at 2.5m	20.2 ± 2.9(30)	22.36 ± 5.56(13)	t=0.378(41)	p=0.70729
30. TEh(20-40ms)	-91.11 ± 1.25(30)	-90.21 ± 2.15(14)	t=0.386(42)	p=0.70203
31. TEh(slope 101-140ms)	2.036 ± 0.0609(30)	2.03 ± 0.0925(14)	t=0.049(42)	p=0.91408
32. Refractoriness at 2 ms	71.69 ± 6.22(27)	64.7 ± 10(11)	t=0.599(36)	p=0.55998
33. Superexcitability at 7	-21.28 ± 0.914(30)	-20.82 ± 1.89(13)	t=0.245(41)	p=0.79433
34. Superexcitability at 5	-24.79 ± 0.879(30)	-22.94 ± 2.27(13)	t=0.925(41)	p=0.36355
35. TEd20(peak)	38.19 ± 0.525(30)	37.05 ± 1.11(14)	t=1.061(42)	p=0.29518
36. TEd40(Accom)	24.09 ± 0.527(30)	22.96 ± 1.17(14)	t=1.023(42)	p=0.31334
38. TEh(peak,-70%)	-250.1 ± 10.5(15)	-243.4 ± 8.14(12)	t=0.486(25)	p=0.63619

iii. Unpaired t-test: comparing control data to day 5 of lignocaine and ketamine infusion in 12 patients with chronic migraine.

Variable	Mean+/-SE(n)	Mean+/-SE(n)	t(df)	p
1. Stimulus (mA) for 50% m	4.287x|/1.04(30)	3.785x|/1.06(12)	t=1.864(40)	p=0.06651
3. Strength-duration\time	0.4807 ± 0.0184(30)	0.4807 ± 0.0412(12)	t=0.000(40)	p=0.9505
4. Rheobase (mA)	2.796x|/1.04(30)	2.583x|/1.07(12)	t=1.062(40)	p=0.29505
5. Stimulus-response\slope	5.128x|/1.04(30)	5.804x|/1.09(11)	t=1.397(39)	p=0.1667
6. Peak response\(mv)	8.847x|/1.06(30)	6.798x|/1.13(11)	t=2.091(39)	p=0.04084*
7. Resting I/V slope	0.6071 ± 0.0142(30)	0.5886 ± 0.0198(12)	t=0.719(40)	p=0.48286
8. Minimum I/V slope	0.2462 ± 0.008(30)	0.2364 ± 0.007(12)	t=0.726(40)	p=0.47884
9. Temperature (C)	33.25 ± 0.17(30)	32.35 ± 0.337(12)	t=2.633(40)	p=0.01155*
10. RRP (ms)	2.953x|/1.02(30)	3.163x|/1.04(12)	t=1.662(40)	p=0.10048
11. TEh(90-100ms)	-116.7 ± 2.77(30)	-115.2 ± 4.18(12)	t=0.299(40)	p=0.75943
12. TEd(10-20ms)	68.69 ± 0.744(30)	67.15 ± 1.34(12)	t=1.067(40)	p=0.29283
13. Superexcitability (%)	-23.05 ± 0.926(30)	-21.58 ± 2.09(12)	t=0.749(40)	p=0.46437
14. Subexcitability (%)	14.4 ± 0.655(30)	15.33 ± 1.74(12)	t=0.618(40)	p=0.54739
17. Age (years)	39.1 ± 2.4(30)	37.33 ± 4.18(3)	t=0.227(31)	p=0.8064
18. Sex (M=1, F=2)	1.467 ± 0.0926(30)	1.583 ± 0.149(12)	t=0.67(40)	p=0.51348
19. Latency (ms)	6.468 ± 0.114(30)	6.525 ± 0.209(12)	t=0.256(40)	p=0.78753
20. TEd(40-60ms)	50.66 ± 0.667(30)	50.9 ± 1.3(12)	t=0.18(40)	p=0.83563
21. TEd(90-100ms)	43.96 ± 0.663(30)	42.72 ± 1.19(12)	t=0.962(40)	p=0.34439
22. TEh(10-20ms)	-73.55 ± 0.732(30)	-71.67 ± 1.47(12)	t=1.27(40)	p=0.20907
23. TEd(undershoot)	-18.78 ± 0.604(30)	-19.04 ± 1.36(12)	t=0.199(40)	p=0.82376
24. TEh(overshoot)	14.06 ± 0.597(30)	15.2 ± 1.37(12)	t=0.891(40)	p=0.38226
25. TEd(peak)	68.17 ± 0.696(30)	67.14 ± 1.46(12)	t=0.723(40)	p=0.48039
26. S2 accommodation	24.21 ± 0.528(30)	24.41 ± 1.41(12)	t=0.166(40)	p=0.84387

27. Accommodation half-tim	40.1 ± 0.777(30)	42.51 ± 1.27(12)	t=1.643(40)	p=0.1043
28. Hyperpol. I/V slope	0.3414 ± 0.0105(30)	0.3426 ± 0.023(12)	t=0.054(40)	p=0.91138
29. Refractoriness at 2.5m	20.2 ± 2.9(30)	29.72 ± 7.13(12)	t=1.488(40)	p=0.14073
30. TEh(20-40ms)	-91.11 ± 1.25(30)	-88.64 ± 2.16(12)	t=1.031(40)	p=0.30988
31. TEh(slope 101-140ms)	2.036 ± 0.0609(30)	1.944 ± 0.0875(12)	t=0.821(40)	p=0.42165
32. Refractoriness at 2 ms	71.69 ± 6.22(27)	76.71 ± 16.1(9)	t=0.355(34)	p=0.72261
33. Superexcitability at 7	-21.28 ± 0.914(30)	-20.31 ± 2.28(12)	t=0.476(40)	p=0.64124
34. Superexcitability at 5	-24.79 ± 0.879(30)	-21.04 ± 2.38(12)	t=1.84(40)	p=0.06981
35. TEd20(peak)	38.19 ± 0.525(30)	37.64 ± 1.26(12)	t=0.487(40)	p=0.6337
36. TEd40(Accom)	24.09 ± 0.527(30)	24.43 ± 1.42(12)	t=0.274(40)	p=0.77579

iv. Unpaired t-test: comparing control data to patients with chronic migraine at 6 months follow up after lignocaine and ketamine infusion.

Variable	Mean+/-SE(n)	Mean+/-SE(n)	t(df)	p
1. Stimulus (mA) for 50% m	4.287x|/1.04(30)	3.979x|/1.09(9)	t=0.932(37)	p=0.36007
3. Strength-duration\time	0.4807 ± 0.0184(30)	0.4458 ± 0.0348(9)	t=0.905(37)	p=0.37482
4. Rheobase (mA)	2.796x|/1.04(30)	2.808x|/1.13(9)	t=0.045(37)	p=0.91692
5. Stimulus-response\slope	5.128x|/1.04(30)	5.385x|/1.08(7)	t=0.498(35)	p=0.62707
6. Peak response\(mv)	8.847x|/1.06(30)	3.087x|/2.35(8)	t=2.387(36)	p=0.02128*
7. Resting I/V slope	0.6071 ± 0.0142(30)	0.5991 ± 0.0364(9)	t=0.245(37)	p=0.79455
8. Minimum I/V slope	0.2462 ± 0.008(30)	0.2457 ± 0.0105(9)	t=0.036(37)	p=0.92231
9. Temperature (C)	33.25 ± 0.17(30)	32.23 ± 0.431(9)	t=2.623(37)	p=0.01212*
10. RRP (ms)	2.953x|/1.02(30)	3.027x|/1.07(9)	t=0.497(37)	p=0.62754
11. TEh(90-100ms)	-116.7 ± 2.77(30)	-118.2 ± 5.57(9)	t=0.248(37)	p=0.79298
12. TEd(10-20ms)	68.69 ± 0.744(30)	68.04 ± 1.39(9)	t=0.418(37)	p=0.68054
13. Superexcitability (%)	-23.05 ± 0.926(30)	-26.16 ± 2.23(9)	t=1.499(37)	p=0.13854
14. Subexcitability (%)	14.4 ± 0.655(30)	13.43 ± 1.7(9)	t=0.645(37)	p=0.53025
18. Sex (M=1, F=2)	1.467 ± 0.0926(30)	1.444 ± 0.176(9)	t=0.114(37)	p=0.87547
19. Latency (ms)	6.468 ± 0.114(30)	6.503 ± 0.182(9)	t=0.151(37)	p=0.85349
20. TEd(40-60ms)	50.66 ± 0.667(30)	53.89 ± 0.792(9)	t=2.493(37)	p=0.01652*
21. TEd(90-100ms)	43.96 ± 0.663(30)	44.74 ± 1.46(9)	t=0.536(37)	p=0.60129
22. TEh(10-20ms)	-73.55 ± 0.732(30)	-71.22 ± 1.79(9)	t=1.414(37)	p=0.16209
23. TEd(undershoot)	-18.78 ± 0.604(30)	-18.82 ± 1.55(9)	t=0.024(37)	p=0.93007
24. TEh(overshoot)	14.06 ± 0.597(30)	14.86 ± 1.67(9)	t=0.562(37)	p=0.58403
25. TEd(peak)	68.17 ± 0.696(30)	69.09 ± 1.29(9)	t=0.631(37)	p=0.53884
26. S2 accommodation	24.21 ± 0.528(30)	24.35 ± 1.8(9)	t=0.104(37)	p=0.8814
27. Accommodation half-tim	40.1 ± 0.777(30)	44.7 ± 1.1(9)	t=2.976(37)	p=0.0051**

28. Hyperpol. I/V slope	0.3414 ± 0.0105(30)	0.3717 ± 0.0327(9)	t=1.164(37)	p=0.25076
29. Refractoriness at 2.5m	20.2 ± 2.9(30)	26.88 ± 11.4(9)	t=0.828(37)	p=0.41787
30. TEh(20-40ms)	-91.11 ± 1.25(30)	-89.32 ± 2.91(9)	t=0.647(37)	p=0.52883
31. TEh(slope 101-140ms)	2.036 ± 0.0609(30)	1.989 ± 0.107(9)	t=0.37(37)	p=0.71286
32. Refractoriness at 2 ms	71.69 ± 6.22(27)	68.58 ± 17.6(8)	t=0.21(33)	p=0.81678
33. Superexcitability at 7	-21.28 ± 0.914(30)	-25.5 ± 2.18(9)	t=2.065(37)	p=0.04364*
34. Superexcitability at 5	-24.79 ± 0.879(30)	-25.39 ± 3.3(9)	t=0.254(37)	p=0.78888
35. TEd20(peak)	38.19 ± 0.525(30)	38.99 ± 1.23(9)	t=0.685(37)	p=0.50422
36. TEd40(Accom)	24.09 ± 0.527(30)	24.27 ± 1.81(9)	t=0.128(37)	p=0.86695

Legend

* Significant

**Highly significant (temperature is a major factor in the significance of stimulus response in baseline, day five and six month follow-up – therefore, significance mainly influenced by operational factors, particularly temperature rather than migraine patients having heightened stimulus sensitivity related to central sensitisation).