AN EEG APPROACH

TO

ASPECTS OF INSOMNIA

by

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A Doctoral Thesis

Submitted in partial fulfillment of the requirements for the award of Doctor of Philosophy of the Loughborough University of Technology

August 1979

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ACKNOWLEDGEMENTS

I would like to express my sincere gratitude to my supervisor, Dr. J. A. Horne, for his advice and guidance throughout the research programme. I owe much to the active and personal interest which he showed in the research work.

I owe thanks to Dr. J. Traynor, Department of Chemistry, for his invaluable assistance in the biochemical areas of this thesis. I would also like to thank all my colleagues and friends who offered advice and encouragement on various aspects of the research, giving particular mention to: Mr. M. Sinclair, Dr. R. Meddis and Mr. M. Hunt who advised on some of the data analysis; Mr. J. M. Brittain, Miss. J. Martin and Mr. J. Youngman who read the draft manuscripts.

I am grateful to Fisons Pharmaceuticals Limited and the Niagara Therapy Company for providing funding for the research and to the Boots Company and Thomas Kerfoot & Sons for providing the placebo tablets.

A special word of thanks to my husband for his patience and understanding and for preparing the graphs for the thesis. Finally, I am indebted to all the subjects who took part in the studies.

ABSTRACT

The research programme was initiated by a study of possible non-drug "aids" to insomnia. Using a group of self-reported, sleep onset insomniacs, the efficacies of cycloid vibration, Horlicks and cocoa (placebo condition) were assessed by all-night EEG recordings. None of these "aids" were found to have significant effects upon either the quality or quantity of sleep. However, two interesting observations of this study were:

1. the unreliability of subjective reports of insomnia; the insomniacs, in general, tended to overestimate their sleep onset times,
2. the frequent use of Aspirin by the self-reported insomniacs to relieve their sleeping difficulties.

These observations served as the bases for further investigation.

With respect to the first observation, selecting a group of insomniacs who satisfy EEG criteria can be a very time consuming procedure. To partly overcome this difficulty, an EEG model of sleep onset insomnia was created, using daytime nap sleep onset times of good sleepers. The morning sleep onset times were found to simulate sleep onset insomnia. This model was then used to assess the sedative potential of a known tranquiliser (Temazepam); F.P., a drug supplied by Fisons Pharmaceuticals Limited, which was believed to have mild sedative properties; and a placebo. The results showed that Temazepam exerted a stronger sedative effect upon the model than either F.P. or placebo.

Concerning the second observation, from the literature it appeared that Aspirin (acetylsalicylic acid) would affect sleep. Initially, a pilot study was conducted with good sleepers who received a normal, therapeutic dose of Aspirin. A significant decline in stage 4 sleep

was found, which appeared to be more marked in the females. A larger, controlled study was then conducted with 16 females; eight received Aspirin and eight placebo, under double blind conditions. A significant increase in stage 2 sleep was found on the drug nights. Upon drug withdrawal, there was a significant decline in REM latency.

Two supplementary pilot studies were carried out; one to investigate the effects of Paracetamol upon sleep, the other to investigate the effects of Aspirin upon the urinary output of 5-HIAA. Neither study produced marked significant results.

CONTENTS

LIST OF ABBREVIATIONS

ANOVA	-	Analysis of Variance.
A.S.A.	-	Acetylsalicylic Acid (Aspirin)
D	-	Drowsy
d.f.	-	Degrees of Freedom
EEG	-	Electroencephalogram
EMG	-	Electromyogram
EOG	-	Electro-oculogram
F	-	Fisher's F-ratio
F.P.	-	The drug supplied by Fisons Pharmaceuticals Limited
gm.	-	gram
5-HIAA	-	5-Hydroxyindoleacetic acid
5-HT	-	5-Hydroxytryptamine (serotonin)
5-HTP	-	5-Hydroxytryptophan
Hz.	-	Hertz
I_{50}	-	The I_{50} value is a measure of the potency of the inhibitors of prostaglandin synthesis. This value is calculated from the curves plotted for the percentage inhibition versus the $\log_e$ concentration and is expressed as the concentration (μg/ml) required to produce 50% inhibition of the control synthesis.
i.p.	-	Intraperitoneal
kg.	-	Kilogram
kΩ	-	Kilohm
MAO	-	Monoamine Oxidase
MAOI	-	Monoamine Oxidase Inhibitor
mgm.	-	Milligram
ml.	-	Millilitre
mm.	-	Millimetre
ms.	-	Millisecond
MS	-	Mean Square (Variance Estimate)

mμ	-	Micrometre
P	-	Probability Level
PCPA	-	Parachlorophenylalanine
PG	-	Prostaglandin. The type designation of the primary PGs is calssified by structural differences in the cyclopentane ring, PGE, PGA and PGF. They are further subclassified by subscript according to the number of carbob-carbon double bonds (PGE_1, PGE_2 etc.) and according to cis (α) vs. trans (β) configurations, for example $PGF_{2\alpha}$.
r	-	Pearson's Product Moment Correlation Coefficient
REM	-	Rapid Eye Movement Sleep
SS	-	Sum of Squares
TST	-	Total Sleep Time
μg.	-	Microgram
μV.	-	Microvolt
W	-	Wakefulness

LIST OF APPENDICES

LIST OF GRAPHS

LIST OF TABLES

Table | Page

A.S.A. Main Study

Table | Page

Chapter Three - Part II Results Tables

Urine Analysis of 5-HIAA

Paracetamol Pilot Study

SCHEMA

SCHEMA

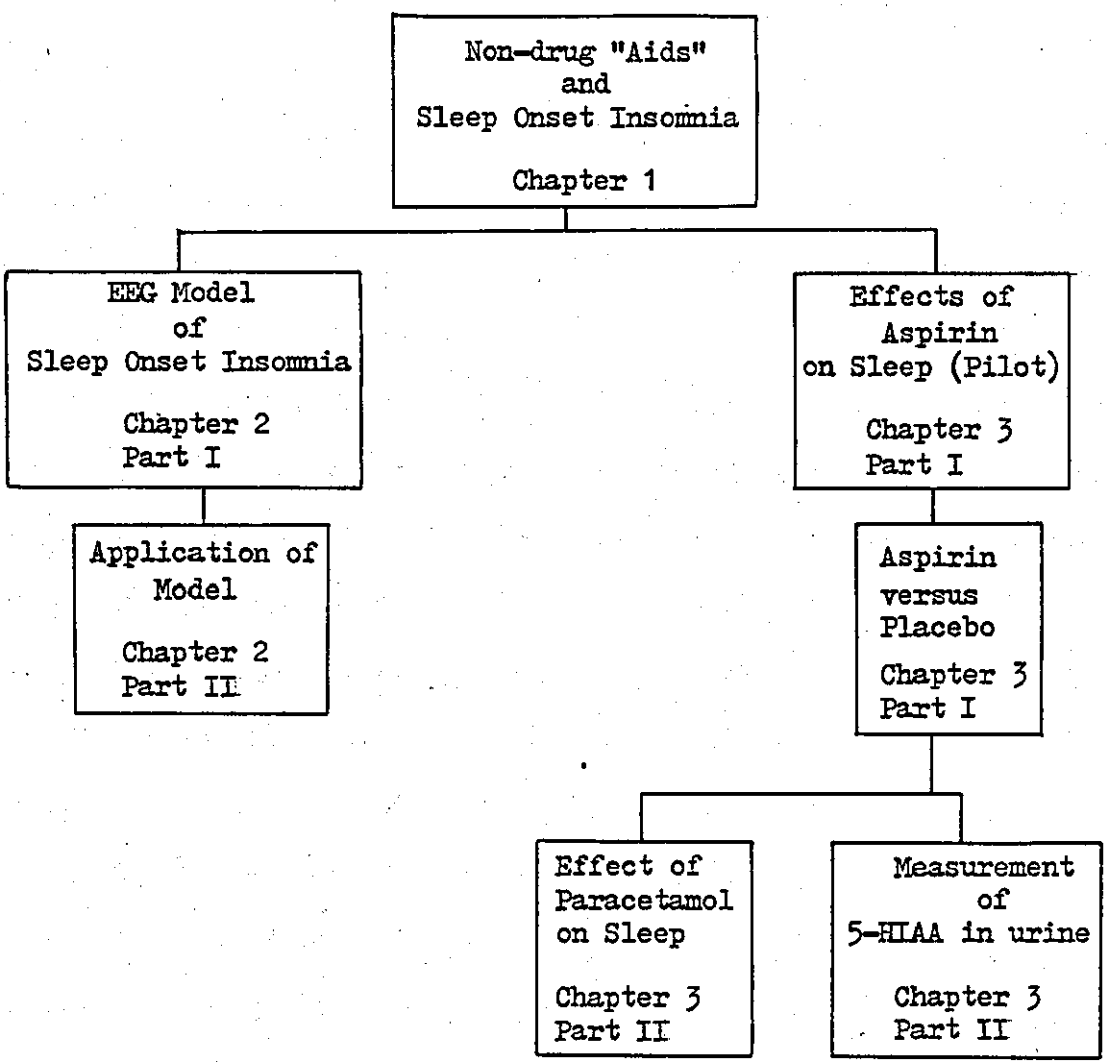
The research programme began with an EEG evaluation of two, commercial, non-drug "sleeping aids" using a group of self-reported, sleep onset insomniacs. The findings and observations of this initial study gave rise to two lines of further investigation. One was to create an EEG model of sleep onset insomnia, and the other was to investigate the effects of Aspirin (acetylsalicylic acid) upon sleep.

The stimulus for the former aspect of further research stemmed from the difficulties encountered in selecting from a group of self-reported, sleep onset insomniacs some who actually satisfied the EEG criteria. These difficulties arose primarily because the insomniacs tended to overestimate their sleep onset times. The aim of the EEG model of sleep onset insomnia was to provide a long sleep latency, which would form the basis of the initial evaluation procedure for various "sleeping aids". The approach adopted was to measure, by EEG criteria, the sleep onset times of good sleepers during daytime naps. This approach was successful. The model was then modified slightly and used to assess the sedative properties of two drugs.

The interest in the Aspirin aspect of insomnia arose from the comments made by many of the self-reported insomniacs that they frequently took Aspirin to relieve their sleeping difficulties. A literature search indicated that Aspirin (acetylsalicylic acid) might be expected to affect sleep, but the possible mechanisms involved were unclear and gave rise to much speculation. Nevertheless, sufficient interest had been aroused to warrant a pilot study. On the basis of the results of the pilot study, this aspect of insomnia was pursued further by means of a large, controlled study on the effects of acetylsalicylic acid upon sleep.

Two supplementary pilot studies were then conducted in an attempt to determine the mechanisms by which Aspirin may be exerting its effects upon sleep. One of these studies was concerned with the effect of Aspirin upon the daily urinary output of 5-hydroxyindoleacetic acid (5-HIAA), which is a breakdown product of 5-hydroxytryptamine. The other study examined the effects of Paracetamol (Acetaminophen) upon sleep.

Although all the studies in this research programme have a common starting point, they deal with quite different aspects of insomnia. Thus, rather than presenting an extensive introduction, covering a wide variety of topics, each chapter will have its own introductory sections explaining the background behind the aspect of insomnia under consideration. An overall view of the research programme is presented in the flow diagram below:



Not poppy nor mandragora,
Nor all the drowsy syrups of the world,
Shall ever medicine thee to that sweet sleep
Which thou owed'st yesterday.

SHAKESPEARE, Othello

Chapter One

NON-DRUG "SLEEPING AIDS"

1.0 INTRODUCTION

Sleep disorders can be classified into three, broad, general categories (Mauri, 1975):

1. Insomnia - An inability to obtain adequate sleep.
2. Hypersomnia - A need for excessive amounts of sleep, in some cases accompanied by short, irresistible sleep attacks during the day, known as narcolepsy.
3. Dysomnias - The exclusive occurrence of pathologies during sleep, such as somnambulism.

Insomnia is the sleep disorder with the highest incidence in the population. Johns (1972) stated that approximately 14 per cent of the people of the British Isles suffered from insomnia. It is also the sleep disorder which presents the largest problem with respect to treatment and management.

The literal translation of insomnia is, "no sleep". However, insomnia seldom results in total sleeplessness and the term usually refers to a sleeping difficulty. It is inadequate to define a sleeping difficulty simply in terms of hours of sleep, because of the wide range in individual sleep requirements. In the rare, extreme cases some individuals can sleep for as little as approximately three hours per night without complaining of insomnia (Jones & Oswald, 1968); whereas, others who feel they need ten hours sleep per night may complain of insomnia if they only have eight hours sleep. An individual is classified as an insomniac if he considers his sleep to be inadequate and if this sleeping difficulty is actually troubling him. Although most people at some time have experienced such difficulties, generally on a temporary basis, for many it is a chronic problem.

According to Worster-Drought (1927) there are three primary sleep disturbances making up the insomnia syndrome:

1. long sleep latencies - known as sleep onset insomnia,
2. frequent nocturnal awakenings,
3. early morning awakenings.

However, in practice these categories are seldom distinct and it is often very difficult to determine which is the major sleep disturbance for a particular insomniac. These sleep disturbances may occur singly or in combination, with varying degrees of severity. Kleitman (1963) reported that sleep onset insomnia was the commonest recorded and that this type of insomnia was the one usually referred to by a patient when he complains to the doctor that "he cannot sleep".

The most widely prescribed treatments for insomnia are hypnotic and sedative drugs. Johns (1972) reported that approximately three million sleeping tablets were consumed each night in the British Isles. Apart from the large number of sleeping tablets prescribed, many insomniacs, who do not seek medical attention, try various non-drug "sleeping aids" in an attempt to improve their sleep. These "sleeping aids" can range from various folk remedies such as counting sheep or a warm bath at night, to other commercial "aids". These latter "aids" include malted milk beverages, taped relaxation instructions and herbal pillows. Many claims are made by the manufacturers with regard to the efficacy of these various "aids"; however, as will be discussed in later sections, there is little experimental evidence to support these claims. In some of the studies which will be considered, various possible non-drug treatments of insomnia were found to be no more effective than a placebo condition. This might be expected, since it would appear from the literature survey that a major component of the

complaint of insomnia is psychological in origin.

Niagara Therapy (U.K.) Limited are a company who manufacture expensive "sleeping aids". Their products include cycloid vibrating cushions, chairs and bed units which they claim relieve sleeping difficulties, because these devices have been found to enhance muscle relaxation. At the request of this Company, and with their funding, an EEG evaluation of their products was carried out using a group of self-reported, sleep onset insomniacs. This type of sleep disorder was investigated since it would appear that sleep onset insomnia is one of the most commonly recorded sleep disorders and it was also considered that this group of insomniacs may try such devices. The Niagara Therapy cushions were evaluated against another commercial "sleeping aid", namely Horlicks, a milk beverage without the cereal component and the pre-treatment baseline condition.

2.0 INSOMNIA

2.1 Causes of Insomnia

There is no simple, straight forward answer to the question, "What causes insomnia?" An insomniac group is very heterogeneous. Although the majority of sufferers view their sleep disorder as the primary complaint, in most cases it is symptomatic of a wide variety of underlying causes, of which the following are some examples:

1. Somatic Disorders - for example, arthritic pain and various respiratory complaints.
2. Psychiatric Disorders and Mood Changes - In an extensive questionnaire study of referrals to a psychiatric clinic carried out by Stonehill, Crisp and Koval (1976), it was found that various moods and psychiatric states were associated with different sleep characteristics; anger

was associated with the greatest sleep disturbance throughout the night, and neurotic depression was characterised by reduced sleep length, with both sleep onset insomnia and early morning awakening. Kales, Caldwell, Preston, Healey and Kales (1976) reported similar findings although they used a different approach. The personality profiles of 128 self-reported insomniacs were examined; 85 per cent had personality profiles in the pathological range. The scale with the highest mean value was depression and the authors concluded that, "Overall this profile indicates neurotic depression and unhappiness and reflects internal reactions of stress that are handled by depression or inhibition of anger".

Although an association between psychiatric disturbance, mood changes and insomnia has been established, this does not indicate a direct cause-effect relationship.

3. Situational Phenomena - for example, worry and anxiety about a pressing problem or factors such as a hot, underventilated room or traffic noise.
4. Rhythm Disorders - for example, in the case of shift work.
5. Sleep Anomalies - for example, night terrors.
6. Drug Related Insomnia - this may be due to stimulants taken during the day or to drug withdrawal insomnia.
7. Pseudoinsomnia - the patient believes that he has insomnia, but in fact he is a good sleeper, according to the EEG.
8. Expectancy Effects - the patients often expect to sleep for eight hours and believe it to be detrimental if they do not.
9. Autonomic Arousal - Monroe's (1967) study formed much of the basis of the assumption that the insomniac is an "aroused" individual. This characteristic of insomnia will be discussed further in section 2.2, chapter one. Many of the non-drug treatments of insomnia,

such as relaxation therapy, are based on the assumption that the insomniac is an aroused individual whose sleeping difficulties may be alleviated if his anxiety is reduced and he is relaxed, but it will be argued that the claim that physiological arousal is a major cause of insomnia is inconclusive.

2.2 Characteristics of Insomniacs

Monroe's (1967) study of good and poor sleepers indicated that poor sleepers have higher levels of autonomic arousal than good sleepers, using the following measures: number of body movements per hour, number of peripheral vasoconstrictions and the group mean rectal temperature measured during the one recording night. The poor sleepers were also found to have significantly higher heart rates during a 30 minute pre-sleep period.

However, caution must be applied when generalising these findings to the sleep onset insomniac population. Although the poor sleepers had a self-reported group mean sleep onset time of 60 minutes, the group mean sleep onset time according to the EEG measure was only 15 minutes. Nevertheless, the poor sleepers did have a significantly shorter total sleep time and spent a longer time awake on the one recording night than good sleepers. Even the good sleep group's total sleep time (388 minutes) was lower than would be expected for these subjects, with a mean age of 26 years. According to the graph compiled by Roffwarg, Muzio and Dement (1966) the average range of total sleep time for the age range 19 - 30 years is 420 - 465 minutes. The fact that the subjects were not studied over consecutive nights may have contributed to this relatively low group mean total sleep time. After the subjects had been adapted to the laboratory, one or two nights elapsed before

they returned for the recording night. This may have resulted in some subjects not being fully adapted to the laboratory. The results of this study were based on only one night's EEG and physiological recordings.

Monroe's (1967) findings were supported in part by Johns, Masterton and Bruce (1971) who differentiated between good and poor sleepers on the basis of a sleep habits questionnaire, similar to that of Monroe's. Evidence of greater psychological distress in poor sleepers was seen both in their higher scores on psychiatric tests and in biochemical measures of stress, namely adrenocortical activity, in comparison with the good sleepers. Monroe (1967) has suggested that a high level of autonomic arousal may be a concomitant, if not causal factor in abnormal sleep patterns. Sleep is associated with low levels of autonomic arousal; individuals who are highly aroused may therefore experience difficulty in falling asleep. However, the studies discussed below would fail to support this suggestion.

Hauri and Good (1975) found a slight negative relationship between frontalis muscle tension and sleep onset time. They studied 15 subjects ranging from self-defined excellent to very poor sleepers; poor sleepers did not show higher EMG levels than good sleepers five minutes prior to "lights out", and throughout the first "sleep cycle". The authors found that there was no threshold below which frontalis muscle tension had to drop before sleep could occur. Freedman and Papsdorf (1976) and Browman and Tepas (1976), using a group of insomniacs and good sleepers respectively, were also unable to find any significant relationships between initial autonomic level and sleep onset times. The former measured heart rate and the EMG from the frontalis, masseter and forearm extensor muscles, and the latter heart rate and forearm

muscle tension. Furthermore, Borkovec and Fowles (1973) reported that reduction of physiological arousal as measured by skin conductance, forearm muscle tension and heart rate was unrelated to the insomniacs' self-reported reduction in sleep onset time. It has been suggested by Jacobson (1938) that insomnia might be caused by fluctuations of the EMG level rather than by the absolute EMG level.

In the studies discussed above, the EMG has been used as an indicator of body tension. However, it is debatable whether the EMG measured at a particular muscle site can be extrapolated to the muscle groups of the whole body. There is some disagreement as to which muscles, if any, are predictive of the overall state of muscle tension in the body. Nidever (1959) and Balshan (1962) concluded that a general muscle tension factor did exist. They found that the factor of general muscle tension was centred about the limb musculature in both males and females. Balshan (1962) found that in response to an auditory stimulus, the muscle tension pattern remained the same; but this was not true for all conditions, for when Nidever's subjects performed a serial learning task the muscle tension factor shifted to the neck and head muscles. This shift may be attributable to the head movements the subjects made in order to view the display. Voas (1952) reported that the arm muscles were the best indicators of muscle tension during mental work. However, during stress and frustration, tension was found to be most prominent in the trapezius and masseter muscles. These findings suggested that there may not be just one muscle tension factor for all individuals under all conditions. As stated by Goldstein (1972), "since there is no immediate answer to the problem of selection of muscle groups, it would be best for the investigator to sample several

muscles in different parts of the body". There is also the problem of comparing the EMGs from different muscle groups, as they may be at different depths which can cause distortion and attentuation of the signal. Of the studies previously considered, only the Freedman and Papsdorf (1976) study attempted to measure the EMG from more than one muscle group. Although the EMG can provide information about the state of tension at a particular muscle site, caution must be applied when extrapolating these findings to the whole body.

With regard to the psychological characteristics of self-reported insomniacs, Kales et al. (1976), Johns et al. (1971), Monroe (1967) and Stonehill et al. (1976) have all found a relationship between self-reported insomnia and psychological disorders associated with personality and mood. Overall, the self-reported insomniacs appeard to be anxious, depressed, tense and unhappy.

The literature does not indicate any strong causal relationship between physiological arousal and insomnia; it could be the psychological component which is all important with the "aroused mind" rather than the aroused sympathetic nervous system being the cause of many reports of insomnia. Thus, "psychological relaxation" might be the major component of non-drug treatments of insomnia and not "physiological relaxation". Physiological relaxation is defined as an absence of muscle tension, that is the muscle fibres lengthen. However, when referring to physiological relaxation in this context, it does not necessarily imply a total absence of muscle tension, but rather a reduction in its level accompanied by a reduction in the level of some other indicators of autonomic arousal, such as heart rate or respiratory rate. Paul (1969) found that training in relaxation techniques brought about a reduction in several parameters

associated with physiological arousal. Psychological relaxation is a more ambiguous term and refers to a "quietening of the mind". The aim of psychological relaxation is to prevent the individual from worrying and pondering about various issues and to encourage him to place his worries and anxieties on one side. Both forms of relaxation are seen here as contributing to the general "winding down" of the subject.

However, it is quite possible that a relationship between "psychological tension" and "bodily tension" does exist, but this is a debatable point, due to the varying results which have been obtained. These differences in results may be due to some of the following reasons:

1. the difficulties involved in defining anxiety itself,
2. some investigators have recorded from one muscle group only,
3. there are problems in determining whether the experimental conditions are free of stress during the resting condition.

In one investigation conducted by Malmstrom (1968), the Zuckerman Affect Adjective Check List (AACL), (Zuckerman & Lubin, 1965) and the Nowlis Mood Adjective List (MCL), (Nowlis & Nowlis, 1956) were utilised together. During a motion picture and during radio static noise, frontalis muscle tension was found to correlate positively with the AACL, but negatively with the MCL: apparently, these two scales have tapped different aspects of the psychophysiological state of anxiety. Balshan (1962) examined the EMG from 16 muscle groups of subjects selected because of their extreme scores on the Freeman (1953) and Taylor (1953) Anxiety Scales. There was no difference between the groups according to the EMG measure during the resting state; during an audio stimulus, however, the more anxious group responded with significantly greater activity in nine of the muscle groups. Sainsbury and Gibson (1954) found elevated muscle action potentials in the

frontalis and forearm extensors of anxious patients during a resting state. However, these subjects were selected on the basis of signs of tension and symptoms of anxiety.

There appeared to be no obvious relationship between EMG measures and anxiety, but the results were clearly dependent upon the definition of anxiety, the procedure for selecting subjects, the experimental setting, whether the subjects were studied at rest or whilst performing a task, and which muscle groups were measured. Nevertheless, the possibility still exists that if the insomniacs do suffer from high levels of anxiety which might be reflected in both psychological and physiological measures, then lowering anxiety, for example, by relaxation training might reduce their sleeping difficulty. Studies which have evaluated various methods of relaxation as a treatment for insomnia will be reviewed in later sections.

3.0 POSSIBLE NON-DRUG METHODS FOR THE TREATMENT OF INSOMNIA

3.1 Practical Difficulties in Comparing Studies

It is very difficult to make direct comparisons between studies using insomniacs for the following reasons:

1. Some studies are entirely based upon self-report questionnaire responses, for example, Borkovec and Fowles (1973), whilst others use the objective measure, the EEG, either alone or in combination with the questionnaires (Borkovec & Weerts, 1976). The subjective reports are prone to intervening variables such as response set, whereby the subjects might respond in accordance with the experimenters' expectations and to the problem of overestimation of sleep onset times by insomniacs (Monroe, 1967). However, as suggested by Hinkle and Lutker (1972), the subjective experience may be the critical

factor in the complaint of insomnia. Therefore, one cannot dismiss self-report studies.

2. There is a difference of opinion in some cases between those experimenters who use the EEG measure of sleep onset time; some use the stage 2 criterion for defining sleep onset time, some stage 1 and others fail to state their sleep onset criteria.
3. It is often not clear whether or not the insomniacs were receiving any treatment either prior to, or during the study. If they were, it is necessary to know the length and type of treatment.
4. The type of insomnia under investigation is seldom clearly defined, for example, it is often not stated whether the insomniac suffers primarily from a sleep onset disturbance, early morning awakening, or a combination of both. It is also useful to know whether the insomnia is chronic or acute.
5. Some studies are carried out on hospitalised patients suffering from either physical or mental disorders. Such patients are likely to be receiving various treatments which may confound any results obtained. Also, many of these patients may be spending the majority of their time in bed and therefore, it is to be expected that they would experience difficulties in falling asleep at night.

Taking these points into consideration, subsequent sections will survey and evaluate some of the insomnia studies relevant to the present research.

3.2 Relaxation Therapy

One possible method of reducing anxiety is by means of relaxation training. Jacobson (1938), using his progressive relaxation technique, was the first to report the use of relaxation therapy in the treatment of insomnia. However, it was not until the self-report study of Kahn,

Baker and Weiss (1968) that this method of treatment was further assessed. They employed a relaxation technique known as autogenic training, devised by Schultz and Luthe (1959). As with the progressive relaxation technique, the subjects were taught to relax voluntarily the main muscle groups; autogenic training simply places greater emphasis on the awareness of sensations than the progressive relaxation technique. Eleven of the thirteen subjects who completed the training programme reported a decline in their sleep onset times. This study was later criticised by Eisenman (1970) on the following grounds:

1. for confounding relaxation therapy with Rogerian interviewing, as all the subjects were interviewed prior to the study,
2. for their lack of an independent control group,
3. for failing to obtain behavioural or physiological measures to correct for possible biases of verbal report.

A variety of similar studies have since been conducted with slight modifications. Borkovec and Fowles (1973) attempted to control for possible placebo and demand effects. Their placebo group participated in self-relaxation which involved concentration on neutral and relaxing images. The other conditions consisted of a no treatment control group, a progressive relaxation group and an hypnotic relaxation group. In order to reduce demand effects, the experimenters told the subjects that the actual purpose of the study was to assess the effect of the techniques in reducing physiological arousal. The reason for this was that in some cases an individual may simply respond in accordance with the expectations and attitudes conveyed by the experimenter with respect to a particular treatment and not to the treatment alone. These expectations are normally conveyed by the instructions given to the subjects: for example, if an insomniac is told that drug X has strong hypnotic properties and

is expected to send him to sleep in minutes, then the experimenter is almost "demanding" the subject to respond favourably to the drug. Self-report studies are more susceptible to demand effects than studies employing objective measures. Demand effects can sometimes be reduced by appropriately adjusting the instructions given to the subjects. One may even give some counterdemand instructions; for example, the subjects could be instructed that the treatment would not be effective until after the fourth day. Borkovec and Fowles (1973) found that all three treatment conditions were almost equally effective in improving the reported time to fall asleep, number of awakenings and how rested the subjects felt in the morning in comparison with the no treatment control group. If these non-drug treatments exert their effects via physiological relaxation, then one would expect the progressive relaxation group to show significantly greater improvement than the other two groups. Paul (1969) has shown that an abbreviated form of relaxation training was significantly more effective than hypnotic suggestion and self-relaxation in producing decreased physiological arousal as determined by heart rate, muscle tension, respiratory rate and skin conductance.

In a similar study carried out by Haynes, Woodward, Moran and Alexander (1974), the placebo group, who discussed neutral topics with a therapist reported a reduced sleep onset time. This finding partly supported the results of the Borkovec and Fowles (1973) study. However, in the Haynes et al. (1974) study the relaxation therapy was significantly more effective than the placebo treatment. These authors suggested that the effectiveness of the group relaxation could be a function of cognitive control rather than decreased physiological arousal. Once again the issue was raised that "psychological relaxation" rather than "physiological relaxation" may be the important factor in reducing sleep onset times. It

is also interesting to note that the placebo conditions in these two studies were not matched. In the Borkovec and Fowles (1973) study, the placebo group focused upon neutral images, whereas, in the Haynes et al. (1974) study, the placebo group actually discussed sleep topics, as well as neutral topics with the therapists. Such a discussion may have caused the insomniacs to focus more upon their sleeping difficulties. If the psychological component is of major importance in the treatment of insomnia, as was suggested earlier, then the Borkovec and Fowles' (1973) placebo group would be expected to have had more therapeutic value than that of Haynes et al.'s (1974) study. Furthermore, since the placebo treatments in both the Borkovec and Fowles (1973) and the Haynes et al. (1974) studies were able to reduce sleep onset times, it suggested that placebo, expectation and demand effects all play an important role in the alleviation of self-reported insomnia. Another factor which may have contributed to the difference in results from these two studies is the scheduling of the relaxation training sessions. In the Haynes et al. (1974) study, the subjects were given 30 minute training periods, twice per week for three weeks. In the Borkovec and Fowles (1973) study the subjects were given three, one hour training sessions.

Nicassio and Bootzin (1974) found that both autogenic training and progressive relaxation were markedly superior to the placebo self-relaxation and the no treatment group. This result appears to be contrary to the findings of the Borkovec and Fowles' (1973) study, which showed that relaxation therapy did not result in significantly greater improvements than a form of self-relaxation. One explanation for this difference is that Borkovec and Fowles' (1973) subjects could be classified as moderate insomniacs, with a group mean sleep onset

time of 45 minutes, in contrast to Nicassio and Bootzin's (1974) severe, chronic insomniacs, with a mean reported sleep onset time of 120 minutes. The insomniacs of the Nicassio and Bootzin (1974) study were more comparable with those of the Haynes et al. (1974) study who had a mean reported sleep onset time of 94 minutes. A study conducted by Nicolis and Silvestri (1967) indicated that chronic, severe insomniacs may be more resistant to placebo therapy than moderate insomniacs. However, this study examined only hospitalised, psychiatric patients over two nights, with the sleep onset defined by observation. Also 17 of Nicassio and Bootzin's (1974) subjects had stopped taking their medications only one week prior to the commencement of the study. Therefore, some of these subjects may have been experiencing drug withdrawal effects. Another difference between this study and the majority of others is that the subjects were drawn from the general population, whereas, for the other studies they were chosen from the student population. Again the placebo conditions were not matched, for in the Nicassio and Bootzin (1974) study the placebo group was simply scheduled time to relax, but in the Borkovec and Fowles (1973) study the placebo group actually had contact with a therapist and concentrated upon neutral images which appeared to be of greater therapeutic value.

One important failure, common to the studies so far considered, has been the absence of suitable control groups to allow for the effects of demand characteristics, specific therapist effects and other treatment variables; for example, Borkovec and Fowles (1973), Haynes et al. (1974), Kahn et al. (1968) and Nicassio and Bootzin (1974) all failed to control for at least one of these variables. Of the self-report studies, the one conducted by Steinmark and Borkovec (1974) was well

controlled. The results of this study indicated that under conditions of counterdemand instructions, relaxation training was more effective than a placebo condition at reducing reported sleep onset times. But, this was not the case when positive demand instructions were given.

One objective study by Borkovec and Weerts (1976) indicated that relaxation training was only marginally more effective than the placebo condition at reducing the sleep onset time of insomniacs, as measured by the first appearance of stage 1 sleep, under positive demand conditions. This finding did not hold for the sleep onset times when measured to the time of the first appearance of stage 2 sleep. The findings of this study were weakened by the fact that both the placebo and the no treatment control groups showed a marked reduction in their sleep onset times under counterdemand instructions. The authors suggested that the laboratory setting itself may have contributed a major placebo component, thus accounting for the improvement of these groups. However, this explanation is not completely satisfactory as these groups showed an increased sleep onset time under positive demand conditions, in comparison with their sleep onset times under counterdemand instructions. If it was the laboratory setting which was bringing about their improvement, one would still expect its effects to be present, maybe even to a greater extent, under positive demand instruction conditions. Alternatively, these results might be due to the large amount of night-to-night variability reported by Karacan, Williams, Littell and Salis (1973) for insomniacs on various sleep parameters. They stated that a night of poor sleep tends to be followed by a better night's sleep, which would account for the non-systematic variation in sleep onset times.

From the literature reviewed, it is debatable whether relaxation training can be classified as a "sleeping aid". In many of the studies it appeared to have little advantage over the placebo condition, especially when the placebo was administered under positive demand conditions, with moderate insomniacs.

3.3 Systematic Desensitisation Therapy

In brief, systematic desensitisation therapy consists of neutralising the anxiety provoking stimuli by counter conditioning a relaxation response to the stimuli. This technique was devised by Wolpe (1958) and has been used in the field of psychotherapy. Gershman and Clouser (1974) applied this therapy to a group of insomniacs. They compared the techniques of muscle relaxation training and systematic desensitisation, using recorded instructions in a group setting. It was found that both treatments were equally effective and significantly superior to the no treatment control group in improving reported sleep quality. But no data were presented on the changes in the various sleep parameters. This study failed to control for possible placebo and demand effects on self-report questionnaire responses: it therefore provided only slight evidence for the efficacy of progressive relaxation and systematic desensitisation as possible treatments of insomnia. Both techniques appeared to have similar effects

Ribordy (1976) carried out a controlled, self-report study with insomniacs in order to compare the following treatments:

a) relaxation training,

b) systematic desensitisation,

c) distraction procedure, subjects in this group were trained with an imagery procedure involving "neutral scenes".

d) no treatment, control condition.

Counterdemand instructions were employed. It was found that all three treatment procedures produced a significant decline in reported sleep onset time, in comparison with the no treatment control condition.

Thus, neither of the studies discussed above has shown systematic desensitisation therapy to be superior to relaxation therapy in the treatment of insomnia. Moreover, neither of these treatments appeared to be significantly superior to the placebo condition, under counter-demand instructions.

3.4 Biofeedback

The role of biofeedback training as a possible treatment for insomnia has not been investigated to the same extent as the other techniques. The two main EEG studies were conducted by Hauri and Good (1975) and Freedman and Papsdorf (1976). The former found that biofeedback was ineffective as a treatment for insomnia, according to EEG criteria, but effective when the change was measured by questionnaire responses. The latter found it be effective, according to both EEG and subjective measures. Both studies employed frontalis muscle EMG biofeedback. The difference in results may be attributable to some of the reasons listed in section 3.1, chapter one. For example, Freedman and Papsdorf (1976) defined sleep onset as the time from "lights out" until the first two minutes of stage 1 sleep, whilst Hauri and Good (1975) did not even define their sleep onset criteria. The former used six, 30 minute biofeedback training sessions and the latter employed one hour biofeedback sessions until both patient and therapist felt that maximum benefits had been achieved (mean 22 sessions, range 13-67). The Freedman and Papsdorf (1976) study also included a control group

and a group who underwent progressive relaxation training. The Hauri and Good (1975) study did not employ a control group. It did appear from the results of the Freedman and Papsdorf (1976) study that biofeedback was no more effective than progressive relaxation in the treatment of insomnia, but both were markedly superior to the control condition in this study.

Other investigators, for example, Budzynski (1973) recommended a more elaborate biofeedback training sequence for the treatment of insomnia. This sequence includes:

1. frontalis muscle EMG training,
2. alpha EEG training, (for those individuals who still exhibit predominantly beta rhythms after EMG training),
3. theta EEG training.

However, the evaluation of the above procedure has been limited to self-report studies which in many cases lack a control group. Budzynski (1973) has evaluated the above sequence with six chronic insomnia clients and has reported that four moderately improved and one markedly improved.

3.5 Summary of Non-Drug Treatments

A major component in the treatment of insomnia appears to be anxiety reduction which can be achieved by a variety of methods, some of which were discussed in the previous sections. Overall, the literature reviewed suggested that there is some doubt as to the effectiveness of these possible non-drug treatments, namely progressive relaxation training, systematic desensitisation therapy and biofeedback as alleviators of insomnia. In some studies, for example, Borkovec and Fowles (1973) and Ribordy (1976), the non-drug "aids" appeared to be no more effective than the placebo condition, especially when administered under positive

demand conditions with moderate insomniacs. This may have been because the placebo conditions themselves, in some studies, appeared to have therapeutic value. In general, there was no significant difference between the three possible treatments, with respect to their "sleeping aid" potential (Freedman & Papsdorf, 1976; Ribordy, 1976).

An element common to all three possible treatments and some of the placebo conditions was that they involved the active participation of the insomniac. This may be an important component of these possible treatments, since the Davison and Valins' (1969) study indicated that when patients believed that they had contributed to their own treatment, their improvement was maintained. Since insomnia can be associated with "taking one's worries to bed", then the concentration involved in the above procedures would be beneficial. Storms and Nisbett (1970) have illustrated this point by describing the insomniac as being trapped in a vicious circle, consisting of three stages:

1. the occurrence of symptoms,
2. worry about symptoms,
3. consequent exacerbation of symptoms.

Thus, to alleviate the insomnia this cycle must be broken at some point. Any technique which distracts the insomniac's attention away from his sleeping difficulty may have beneficial effects; this was indicated by the favourable results produced by some of the placebo conditions, previously discussed. As already mentioned "psychological relaxation" may be necessary prior to the onset of sleep. Although no causal relationship between physiological arousal and insomnia has been established, the sensation of muscle relaxation, as suggested by the instructions of these behavioural techniques may bring about "psychological relaxation".

3.6 Drug versus Non-Drug Treatments

Although the previous sections have focused upon non-drug treatments for insomnia, it is the hypnotic and sedative drugs which are normally prescribed for the treatment of insomnia. Clearly, as new hypnotics and sedatives become available the prescribing trends will change. This can be seen from table 1, in Appendix I. These data refer to the United States, but Cohen and Blutt (1978) claimed that there have been similar trends of increasing benzodiazepine prescriptions in England. It is also noteworthy, that during the period investigated (1974 - 1976) there was a decline in the total number of hypnotics prescribed. This may indicate that doctors are now more aware of the underlying causes of the symptom insomnia.

Several recent review articles, for example Mathis (1978), discussed drug treatments of insomnia, but also emphasised that another important aspect in the treatment of some cases of insomnia was the establishment of a psychotherapeutic relationship with the patient in order to determine the underlying causes. There is an indication that not only are prescribing trends changing, but that non-drug methods are also being employed in conjunction with drug treatments. "What are the advantages and disadvantages of drug and non-drug treatments?" Some of the major points are listed below.

Drug Treatments

Advantages

1. Hypnotics and sedatives generally produce the desired effect, at least initially and can be very useful if used for a short time, for example, during times of emotional stress.
2. They are quick and easy to take.

Disadvantages

1. Tolerance to the intial hypnotic/sedative dosage can build up. This develops at different rates according to the hypnotic used, but a few weeks is sufficient for most drugs (Kales, Kales, Bixler & Slye, 1971). It is then necessary to increase progressively the drug dosage in order to maintain the improvement. In the case of the barbiturates the risk of self-poisoning and the likelihood of drug dependence are also increased. However, those insomniacs who fail to increase the dosage of the hypnotic/sedative may eventually have a longer sleep onset time, after having taken the drug for several weeks, than they did prior to the commencement of the treatment. It is more frustrating for an insomniac to be unable to fall asleep quickly when taking an hypnotic/sedative than when receiving no treatment. This is referred to as the reverse placebo effect or attribution process (Storms & Nisbett, 1970).
2. Hypnotics/sedatives interfere with the quality of the night's sleep. The barbiturates, in general, reduce the amount of rapid eye movement (REM) sleep. However, repeated drug administration can cause REM sleep to return to baseline values. These effects appear to be drug and dose related. For example, a low therapeutic dose of the benzodiazepine Flurazepam (15 mg.) has little effect on either REM or stage 4 sleep. At higher doses there may be reduced stage 4 sleep with inhibition of REM sleep (Kales et al., 1971). It is not clear at present what effects REM sleep suppression will have upon the individual, but it is apparent that the effects are not as dramatic as the early researchers had envisaged. Although this property of hypnotics/sedatives has been listed as a disadvantage, one cannot be certain

whether these changes in sleep patterns are detrimental.

3. There is also the problem of drug withdrawal insomnia. This is characterised by a rebound of REM sleep, often with disturbing dreams or nightmares accompanied by frequent awakenings (Oswald & Priest, 1965). The individual may then feel compelled to take a "sleeping pill" in an attempt to return to sleep. In other words a vicious circle can be set up.
4. They often leave one with a hangover the next day (Oswald, 1973). The commonest symptoms being fatigue, sluggishness, apathy, decreased motivation and clouded thought. The degree of this effect depends upon the individual, the tests used to assess this effect and the hypnotic/sedative type and dosage. These effects can lead to the insomniac taking counteracting stimulants during the day.
5. Hypnotics, upon interaction with other drugs, can produce dangerous side-effects.

Conclusions

The drug treatments of insomnia have many disadvantages, but they do appear to be beneficial if used for a short period under close supervision.

Non-Drug Treatments

Advantages

1. They are non-toxic.
2. It is unlikely that they will interfere with the stages of sleep. However, little work has been done in this area as many of the studies are based on self-report measures.
3. Some forms of non-drug treatments, such as relaxation therapy, may make the patient feel that he is "helping himself", which may be

a critical component in the treatment of insomnia. Also, these treatments involve a large amount of therapist-patient contact, so enhancing a placebo effect.

4. Tolerance to these treatments seems unlikely. Borkovec and Weerts (1976), in a one year follow up questionnaire study, found progressive relaxation therapy to be more effective than placebo in maintaining reported sleep improvement.

Disadvantages

1. The initial effects produced by these treatments are likely to be less marked than those produced by hypnotics. For example, relaxation training requires practice and is time consuming; so, although 20 minutes relaxation training may result in a 30 minute reduction of sleep onset time, the procedure may not be cost effective of time.
2. Since these procedures are often time consuming, the insomniacs may not continue with the treatment for any great length of time.

Conclusions

The non-drug treatments have several advantages over the drug treatments, but in the majority of cases they would not be cost effective of time. Furthermore, the majority of insomniacs may prefer their doctors to prescribe a drug rather than, for example, a relaxation training programme.

Although various claims are made by the manufacturers of "sleeping aids" concerning the hypnotic potential of their products, there has been little objective evaluation conducted in this field. Cycloid vibration is the "aid" which will be objectively evaluated in this research programme. This "aid" was chosen primarily because the

manufacturers were willing to provide the funding for the evaluation and also because it is a relatively new, expensive "aid" on the market. The rationale for the claims that cycloid vibration, provided by the Niagara Therapy cushions,"aids" sleep is presented in the next section.

4.0 THE RATIONALE BEHIND THE CYCLOID VIBRATION STUDIES

Bierman (1960) found cycloid vibration applied to the subjects' "back and posterior aspect of their lower extremities" to be effective in increasing trunk flexibility as measured by the finger to floor test. It would appear that this effect is brought about by muscle relaxation. Atha and Wheatley (1976) reported that cycloid vibration at a frequency of 44 Hz., provided by two Niagara Therapy vibrating cushions, under the thighs and lower back of the seated subject, was as effective as classical mobilising exercises in improving joint mobility. Joint mobility was measured by a modified sit-and-reach test which focused primarily upon hip flexion mobility. The authors tentatively suggested that "the primary mechanism involved in increasing short term mobility in joints limited by musculo-tendinous ties, is a re-setting, under conditions of excessive proprioceptor activity, of some centrally controlled level of muscle relaxation". Matheson, Edelson, Hiatrides, Newkirk, Twinem and Thurston (1976) were also able to demonstrate the "relaxing powers of vibration". Frontalis muscle activity, as measured by the EMG, was shown to decrease after vibration in comparison with a control group. The authors concluded that these results indicated that vibrotactile stimulation, especially in the lower back, is able to enhance muscle relaxation. In view of some of the comments made in section 2.2, chapter one, although the EMG was lowered in the frontalis muscle group, this does not necessarily imply that muscle tension throughout the body was reduced. The same holds for the muscle

relaxation measures used in other studies.

Nevertheless, since cycloid vibration has some muscle relaxing properties, it may be a useful "aid" for some psychotherapeutic treatments of, for example, tension and insomnia. However, the evidence cited in the previous sections for physiological arousal being one of the main causal factors of insomnia was very weak and seemed to indicate no necessary, causal relationship between the two. But, some of the relaxation techniques such as biofeedback, systematic desensitisation and progressive relaxation have produced some favourable, positive results in the treatment of insomnia, although in many cases the improvement was seldom greater than that produced by the placebo condition. It is therefore debatable whether the improved sleep is brought about by psychological rather than physiological means. Furthermore, of the 11 studies considered in the non-drug treatments of insomnia,(sections 3.2 - 3.4, chapter one), only three had used EEG criteria for measuring sleep onset times. Thus, there is a need for more objective studies in this particular field.

The "sleeping aid" to be investigated, cycloid vibration, is quite different from the techniques already examined in that it requires no training of the subject: he simply has to sit on the cushions. The relaxation is brought about by mechanical means, whereas, with the other techniques the subject had to learn to relax. It could be implied therefore, that vibration may have different properties with regard to the possible treatment of insomnia in comparison with the other techniques.

5.0 PILOT STUDIES - INTRODUCTION

Since the sensations provided by the vibrating cushions were rather novel and unusual, and because there appeared, to the subjects, to be no obvious connection between vibration and sleep, the experimenter was rather hesitant about initially using these devices with sleep onset insomniac subjects. It was therefore decided that, before using the vibrating cushions with insomniac subjects, these devices should be first evaluated with a group of good sleepers: if the vibration disrupted their sleep patterns, it would be unwise to test these "aids" on a group already suffering from sleeping difficulties. A further reason for this pilot study was to familiarise the experimenter with the laboratory techniques, the EEG and the experience of being awake all-night. Should the insomniac group be more anxious than the good sleepers, as the literature suggested, then it is essential that the experimenter should appear confident.

Initially nine subjects were brought into the laboratory in order to determine:

1. a suitable position for the two cushions,
2. a suitable frequency of vibration.

The cushions were tested in three different positions, as recommended by the manufacturer:

a) applied to the lower back and thighs of seated subjects (Atha & Wheatley, 1976),

b) applied to the calves and thighs of the recumbant subject, Matheson et al. (1976) found that vibration in the lower half of the body was more effective in producing muscle relaxation,

c) applied to the upper and lower back of the seated subject.

The frequencies of the cushions were set at 74Hz., 44Hz. and 14 Hz. The 44 Hz. setting was chosen on the basis of Atha and Wheatley's (1976) study, where this setting was found to enhance muscle relaxation. The other two frequencies were employed in order to provide a range above and below 44 Hz. A duration of 15 minutes was used, as in the Atha and Wheatley (1976) study, for it was considered that beyond this time limit the subjects may begin to feel bored. Subjects were asked to rate both the frequency and the position of the cushions on a five point rating scale. After only the first three subjects it became obvious that 74 Hz. was very unpleasant as was position (c), the reason being that vibration at the upper level caused the neck and head to vibrate. This set of conditions were therefore not applied to subsequent subjects.

One interesting observation was that the majority of the subjects found that the most pleasant frequency was 14 Hz. This raised the issue of whether this frequency of vibration was sufficient to produce physiological relaxation. This low frequency may simply have been producing a "relaxing sensation". Since the introductory sections indicated that "psychological arousal" may be one of the causes of inomnia, it would be very interesting to investigate the effects of this frequncy upon the sleep of insomniacs.

However, since the initial interest was in the muscle relaxing properties of cycloid vibration, it was decided to employ a frequency of 44 Hz., for it was this frequency and similar ones which brought about muscle relaxation in the Atha and Wheatley (1976) and Matheson et al. (1976) studies. Postion (a) was chosen as the subjects did not find this unpleasant in any way and this position has also been

previously used in the Atha and Wheatley (1976) study. It also has the advantage that some of the major muscle groups are situated in this area.

For the pilot study, it was therefore decided to use two cushions, applied to the lower back and thighs of the seated subject, at a frequency of 44 Hz., for a duration of 15 minutes.

6.0 METHOD

6.1 Subject Selection

The aim was to select six good sleepers, that is, individuals who have a sleep onset time of less than 30 minutes and who are not troubled by frequent nocturnal or early morning awakenings. They also had to satisfy the following criteria:

i) they had to be unaccustomed to daytime napping,

ii) they had to have no obvious signs of physical or psychiatric illness,

iii) they had to be drug free,

iv) they had to be non-smokers.

Advertisements were displayed on the University campus calling for paid volunteers. All those who wished to participate were required to complete a sleep questionnaire, a copy of which is in Appendix II. The responses to this questionnaire gave the experimenter an indication of their sleeping habits. The experimenter delivered the questionnaires personally, as this enabled the experimenter to chat with the prospective subjects and to explain in detail the full procedure. Before admitting subjects to the study they were asked to view the laboratory bedrooms and they were shown the equipment, so that they were familiar with the laboratory setting. The laboratory consists of

two, private, comfortably furnished bedrooms.

This subject selection procedure was applied to the majority of subsequent studies.

6.2 General Instructions to Subjects

The subjects were urged to comply with the following conditions during the course of the sleep study:

i) to refrain from alcohol,

ii) to refrain from taking daytime naps,

iii) to refrain from any strenuous, physical exercise to which they were unaccustomed,

iv) to refrain from taking medications, such as, analgesics and cough medicines,

v) to keep a fairly constant daily routine, as far as possible,

vi) to keep their daily dietary intake fairly constant.

These general instructions are applicable to all other subsequent studies.

6.3 General Laboratory Procedure

Subjects were studied in pairs. They arrived at the laboratory approximately one hour before retiring. As far as possible, subjects with similar retiring times came to the laboratory on the same nights to avoid unnecessary disturbance to the subjects.

A monopolar recording technique was employed. Electrode placements were made according to the 10 - 20 system, they were as follows; A1, A2, C3, C4 and lateral to each eye. The electrode placements are shown diagrammatically in Appendix VI. A ground electrode (C_z) was used.

Electrodes were silver/silver chloride discs attached to the scalp by "collodion" glue and to the face by means of adhesive electrode discs and micropore tape. Electrode gel was used to establish electrical contact with the skin. Electrode resistances were kept below 5kΩ and the electrodes were sealed. For each subject the following electrode montage was used: left eye (E1) - A1; right eye (E2) - A1; C4 - A1. A2 and C3 were not employed, but were held in reserve in case of loss of either A1 or C4 during the night. Once the electrodes were affixed, the subjects went about their normal preparations for bed. When the subjects were settled, the electrodes were plugged into the individual headboards. The electrode leads were long enough to allow the subjects freedom of movement.

Recordings were made at a paper speed of 10 mm. per sec. on a Grass model 78 - 12 channel EEG machine. The EEG gain was set at 10μV per mm.; the low frequency filter, on all channels was set at a ½ amplitude frequency of 0.3 Hz., with a fall time constant equal to 250 ms. The pen filter on all channels was set at 30 Hz. The 50 Hz. filters were not switched in.

Although the same procedure was carried out with each subject on every night, the first two nights in the laboratory were for adaptation purposes and EEG recordings were not made, (Agnew, Webb and Williams, 1966). The subjects were allowed to retire and rise at their usual times and these were kept to within ± 15 minutes on each night and morning.

This general laboratory procedure also applies to the subsequent studies.

6.4 Analysis of Sleep Records

Each sleep record was scored, using one minute epochs, according to the standardised criteria of Rechtschaffen and Kales (1968). The EEG measurement of sleep is explained in Appendix VII. The records were scored blind and independently by two, experienced scorers. If the disagreement between the scorers exceeded five per cent of the epochs, it was resolved by discussion. Sleep onset time was defined as the time from "lights out" to the first sign of stage 2, generally indicated by a spindle (12 - 14 Hz. activity) which was of at least 0.5 sec. duration. The duration of the stage 2 period had to exceed five minutes.

In qualifying the amounts of each sleep stage, the problem arose as to whether to use total sleep time, that is the period between sleep onset and the final awakening, including intervening wakefulness, or to use a fixed length of sleep time common to all subjects and nights. Taking total sleep time and calculating the percentage or absolute time in minutes of each sleep stage, has the disadvantage that REM sleep and the lighter stages are favoured. These stages of sleep predominate in the latter part of sleep, unlike stages 3 and 4. Thus, for example, stages 3 and 4 tend to be diluted by an increasing sleep length. It was therefore decided to express the stages of sleep in absolute minutes of a fixed length of sleep time, common to all subjects and nights.

6.5 Experimental Design

Six subjects, three males and females, mean age 19.6 years, were selected as described in section 6.1, chapter one. The general laboratory procedure of section 6.3, chapter one was carried out and

the "general instructions to subjects", as listed in section 6.2, chapter one, were given to the subjects. Each subject spent seven consecutive nights in the laboratory. The experimental protocol was:

Nights	1	2	3	4	5	6	7
Condition	Ad	Ad	B_1	B_2	V_1	V_2	R

Key:

Ad - Adaptation

B - Baseline

V - Vibration

R - Recovery

The cycloid vibration was applied to the lower back and thighs of the seated subject, at a frequency of 44 Hz. and for a duration of 15 minutes. The treatment was given 40 minutes before the subjects retired to bed.

6.6 Data Analysis

The first 450 minutes of the sleep records from sleep onset were analysed, this was the lowest measure of total sleep time for all subjects on all nights. Stages W+1, 2, 3, 4, (3+4) and REM were all expressed in minutes; sleep onset time and total sleep time were also calculated.

The average baseline value was calculated for each sleep parameter, for each subject, and expressed to the nearest whole number. The results are presented in Tables (2 - 9), section 14.0, chapter one. A two-way ANOVA for repeated measures was performed for each sleep parameter in order to determine if there were any significant differences between the nights (McNemar, 1969, page 338). F values yielding P levels equal to or less than .05 were considered to be statistically

significant. The summary ANOVA tables are presented in Appendix VIII.

7.0 RESULTS

No significant differences were found between the baseline, vibration and recovery nights, for any of the sleep parameters considered.

8.0 CONCLUSIONS

If the vibration was disruptive to sleep one might expect stage W+1 to show an increase on vibration nights, or sleep onset time to increase, or total sleep time to decrease. It would appear that cycloid vibration was having no statistically significant effect upon the sleep patterns of normal, healthy, young adults. Thus, the next step was to test the manufacturer's claims that cycloid vibration would be an effective "sleeping aid".

9.0 SLEEP ONSET INSOMNIA STUDY - INTRODUCTION

The aim of this study was to evaluate objectively cycloid vibration, provided by two Niagara Therapy cushions in the manner determined in the pilot study, as a possible "sleeping aid" for sleep onset insomniacs. The rationale for believing that cycloid vibration may be a possible "sleeping aid" was described in section 4.0, chapter one. It was decided to evaluate the cycloid vibration against:

i) a commercial "sleeping aid", such as Horlicks,

ii) a warm, milk beverage without the cereal component, control condition,

iii) a no treatment pre-condition, that is, two baseline nights.

The only EEG evaluation of the effects of Horlicks upon sleep was conducted by Brězinová and Oswald (1972). The subjects employed

in this study were not self-reported insomniacs. The results of this study indicated that restlessness during sleep, at the end of the night, was diminished in a group of young adults, mean age 22 years, after Horlicks. Restlessness was reported as being measured by "scoring all transitory increases of submental EMG when they rose to a level three times as high as the previous level and lasted more than two seconds, whether or not accompanied by movement artifact". In an older group of adults, mean age 55 years, sleep after Horlicks was of longer duration and less broken by periods of wakefulness. It would therefore be interesting to investigate if Horlicks does aid sleep in a group of self-reported sleep onset insomniacs.

Since Horlicks has a distinctive malt flavour, the lesser known and more unusual tasting chocolate flavoured Horlicks was used in this study, with the aim of reducing the placebo effect associated with Horlicks. As it is claimed by the manufacturers of Horlicks that it is primarily the malt component of the beverage which contributes to its beneficial effects, it was decided to administer a control condition of a milk and cocoa beverage. This control condition was more for means of comparison with the efficacy of Horlicks rather than with cycloid vibration. Thus, although this study was primarily concerned with the evaluation of cycloid vibration, it was thought that a concurrent evaluation of Horlicks would be interesting. However, the aim was not to determine whether it may be the milk, malt or cereal component of the beverages which may or may not improve sleep. This would need to be investigated in a series of separate, subsequent studies if an effect were to be found. The control condition in the Brězinová and Oswald (1972) study consisted of a yellow capsule which did not resemble Horlicks, the treatment under investigation. It might

be expected that a "pill" would elicit an entirely different placebo response in comparison with a milk beverage.

10.0 METHOD

10.1 Subject Selection

The aim was to select twelve subjects who were moderate sleep onset insomniacs. That is, individuals who took between approximately 30 minutes and one hour to fall asleep, according to EEG criteria, but who, when once asleep were not troubled by frequent nocturnal or early morning awakenings. Ideally, the individuals should have been experiencing this sleeping difficulty for approximately six months and have been completely drug free. A similar subject selection procedure to that in section 6.1, chapter one was adopted. As well as advertising on the campus for subjects an advertisement was also placed in the local paper. The heading of the advertisement being: "Does It Take You 45 Minutes or Longer to Fall Asleep ?" At no time, either during the initial advertising and interviewing, or during the study, were the subjects given the impression that they were being treated for their insomnia: this was made quite clear to the subjects before they agreed to take part.

Special care was taken in the insomniac subject selection procedure to avoid selecting those who were obviously physically or mentally ill, receiving treatment, or who relied upon their own sleep remedies, such as a sherry before bed. Out of the 45 subjects who applied, 14 were selected who appeared to satisfy the above criteria, according to their responses to the sleep questionnaire. The aim was to obtain 12 subjects; the two extra were to allow for drop outs due to illness or subjects failing to meet the EEG criteria for sleep onset insomnia. Freedman

and Papsdorf (1976) and Monroe (1967) had previously reported that insomniacs, in general, tended to overestimate their sleep onset times and it was anticipated that this problem would also be experienced in the present study.

Thirteen subjects, selected on the basis of their questionnaire responses (one had been taken ill), had their sleep onset times measured according to EEG criteria on their third adaptation night in the laboratory. In view of the fact that all 13 subjects were self-reported insomniacs, five had exceptionally short sleep onset times, (Table 10, section 14.0, chapter one): two had a sleep onset time of eight minutes, two fell asleep in eleven minutes and one in thirteen minutes. Although several other sleep onset times measured on the third adaptation night failed to reach the thirty minute sleep onset criteria, they were not as low as those previously mentioned. Subject C had a sleep onset time of 17 minutes which was low, but this subject actually commented that he considered this short sleep onset time a rare occurrence. Whereas, the other five subjects with short sleep onset times, during the morning conversation with the experimenter, all commented that they had a fairly typical night's sleep. This finding posed a difficult problem. "Should all 13 subjects be employed in the study or should the five suspect subjects be eliminated at the start, leaving only eight subjects for the evaluation?"

Although some subjects failed to satisfy the EEG definition of insomnia, that is a sleep onset time* of at least 30 minutes, they were

* According to EEG criteria, sleep onset time is defined as the time from lights out until the first appearance of a stage 2 sleep period of at least five minutes duration.

insomniacs according to subjective criteria. All the subjects volunteered to take part in the study claiming that it took them at least 30 minutes to fall asleep. However, the aim of the present study was to evaluate objectively two, commercial "sleeping aids" and with initial sleep onset times as low as eight minutes, one could hardly expect the "aid" to be able to bring about a reduction in the sleep onset time. One could argue that these insomniacs, since they had not sought medical attention for their sleeping difficulty may try such devices. Consideration must also be given to the fact that sleep onset times of both good and poor sleepers have been shown to vary from night-to-night (Agnew & Webb, 1971; Karacan et al., 1973). It could be therefore, that by chance the sleep onset time recording night coincided with a short sleep onset time for these five subjects; however, this idea was not borne out by the majority of their morning comments, but it did hold for subject C. Maybe if these five subjects were studied for a greater number of nights, a long sleep latency may be observed on some nights. This would be a rather costly procedure in order to obtain a few long sleep onset times. Furthermore, there would be no obvious method of determining which subject would experience a long sleep latency on a particular night.

After careful consideration, it was decided that these five subjects were not representative of the sleep onset insomniac population, according to EEG criteria on the third laboratory night. These subjects were told that they would not be suitable for the present study. Two of the subjects were so convinced that they had taken at least 45 minutes to fall asleep that the supervisor had to explain to the subjects the reasons for their being rejected from the study.

At the end of the selection procedure there were only eight subjects, three females and five males, average age 25.6 years, with a group mean baseline sleep onset time of 34 minutes. Although the experimenter was not entirely satisfied with this group of sleep onset insomniacs, with respect to the length of their sleep onset times according to the EEG criteria, the evaluation was conducted with them. It would appear that the selection of a group of sleep onset insomniacs which satisfies EEG criteria is a major problem in the objective evaluation of "sleeping aids".

The subjects were given the "general instructions to subjects", as listed in section 6.2, chapter one.

10.2 Experimental Design

The subjects attended the laboratory in pairs and the general laboratory procedure as described in section 6.3, chapter one was carried out. After much discussion with insomniacs not used in this study, and consideration of the fact that, in general, insomniacs tend to be rather anxious, it was decided not to use a post-sleep questionnaire. It was thought that the subjects' concern about having to complete the questionnaire may interfere with their recorded sleep onset times. However, the experimenter did informally and discretely question the subjects about their night's sleep and any important comments were noted later. No feedback was given to the subjects in the morning; if they asked how long it took them to fall asleep, they were told that the data had not been analysed. This study was to be as objective as possible.

Each subject underwent all three treatment conditions, vis:

i) Horlicks made up according to the manufacturer's recommendations

with 25 gms. chocolate flavoured Horlicks dissolved in 250 mls. of warm milk.

ii) 5 gms. cocoa powder dissolved in 250 mls. of warm milk. This mixture was chosen in order to produce a chocolate flavour almost equivalent to that of chocolate flavoured Horlicks. The subjects were allowed to add sugar to taste to both beverages. The maximum amount added was 7 gms.

iii) 15 minutes of 44 Hz. cycloid vibration, supplied by means of two Niagara Therapy cushions and applied as determined by the pilot study.

All experimental conditions were administered so as to terminate at least 15 minutes before the subjects retired. Each "aid" was given to all subjects twice over a period of six nights. No two subjects received the same treatment on the same night. The experimental conditions were administered in a detached, clinical manner, without the subjects receiving any obvious indication as to whether they may or may not aid sleep. The subjects were informed that it was simply an exploratory study and that little was known about the effects of the three conditions upon sleep. The beverages were labelled A and B and at no time were they given their brand names.

It was decided to allow the insomniacs three adaptation nights; it is reasonable to assume that if they tend to be rather anxious and tense, then it may take them longer to adapt to the laboratory setting than good sleepers. Two, baseline, no-treatment nights followed. Each subject spent eleven consecutive nights in the laboratory.

The experimental protocol was:

Night	1	2	3	4	5	6	7	8	9	10	11	
Condition	Ad	Ad	Ad_s	B_1	B_2	H	V	C	C	V	H	Subjects 1, 4, 7
	"	"	"	"	"	V	C	H	H	C	V	2, 5, 8
	"	"	"	"	"	C	H	V	V	H	C	3, 6

Key:

Ad – Adaptation

Ad_s – Sleep onset time recorded

B – Baseline

V – Vibration

H – Horlicks

C – Cocoa

10.3 Data Analysis

The first 380 minutes of the sleep records from sleep onset were analysed, in one minute epochs, according to the standardised criteria of Rechtschaffen and Kales (1968). A total sleep time of 380 minutes was chosen as this was the lowest common measure of total sleep time for all subjects, on all nights. The records were scored blind and independently by two, experienced scorers. If the difference between the scorers exceeded five per cent of the epochs, it was resolved by discussion.

1. The sleep parameters stage W+1, 2, 3, 4, (3+4), REM, sleep onset time, REM latency and total sleep time were all expressed in absolute minutes of the total sleep time. Sleep onset time was measured as the time from "lights out" until the first appearance of a stage 2 period, of at least five minutes duration. The number of stage changes were also counted, as they are thought to be a

measure of restlessness during sleep. The average score for the two baseline nights and for each pair of treatment nights, for each subject, on each sleep parameter was used for the data analysis. The results are presented in Tables (11-22),(section 14.0, chapter one).

2. In order to determine if there were any significant differences between the treatments a two-way ANOVA for repeated measures was performed for each sleep parameter (McNemar, 1969, page 338) Stage (W+1) was analysed for both the first and second halves of the night. The summary ANOVA table are presented in Appendix IX.
3. The sleep onset times of the insomniacs, over the recording nights, were analysed for trend effects to determine whether sleep onset times changed simply as a function of the number of nights the individual spent in the laboratory, regardless of the treatment (Table 23, section 14.0, chapter one).
4. It was decided to compare this group of eight insomniacs with a group of eight good sleepers, who are actually the placebo control group from chapter three. Although the two groups were not exactly matched for age and sex, a general comparison between the two groups was seen to be worthwhile. The good sleep group's total sleep time was reduced to 380 minutes and only the first eight recorded nights were used for the purpose of comparison with the insomniac group.
   a) To compare the night-to-night variability of the two groups, the standard deviations across all recording nights were calculated for each parameter and each subject. In order to be consistent with Karacan et al. (1973), the logarithmic standard deviation scores were used when comparing the group mean standard deviations by the method of unrelated t-tests.

b) To determine whether or not insomniacs differed from each other more than the good sleep group differed amongst themselves, the between subject variance estimate was calculated for each group of subjects, for each of the ten parameters, listed in 1. The results are presented in Table 25,(section 14.0, chapter one).

5. The correlation coefficient (Pearson's r) was computed between the actual sleep onset time, as measured by the EEG criteria on the third night in the laboratory, and the sleep onset time as estimated by the 13 subjects prior to the commencement of the study. The third laboratory night was considered for this comparison, as it was the only night on which the sleep onset times of all of the 13 self-reported insomniacs had been measured. For calculating purposes, the lower range of estimated values was used. For example, if the subject estimated 30-60 minutes, then 30 minutes was the time used. Logarithmically transformed sleep onset time scores were used, because the raw scores were not found to be normally distributed, but severely skewed to the right. This can be clearly seen from Graph A,(section 14.0, chapter one) which is a frequency plot of 35 independent sleep onset times, from the baseline data of good sleepers of the various studies in the research programme. There is a strong tendency for the majority of individuals to fall asleep in a relatively short time. In this case 24 subjects fell asleep in less than 24 minutes, which is the group mean sleep onset time. Graph B, (section 14.0, chapter one) illustrates that the logarithmic transformation performed on the data normalises the distribution. Skewness in a sleep onset time distribution was also reported by Johns (1977) and Agnew and Webb (1971). Johns' (1977) data showed a much

greater range of sleep onset times, of 2 to 186 minutes, compared with the scores of the present study which ranged from 5 to 82 minutes. As commented upon by Johns (1977), in order to be statistically correct the mean sleep onset time should be quoted as the geometric rather than the arithmetic mean. Throughout this text both are quoted in order to make comparisons with other studies easier.

Since the t and F tests have been shown to be robust (Boneau, 1960; Havlicek & Peterson, 1974), it was considered unnecessary to use the transformed sleep onset times for these statistical tests. Johns (1977) also performed the t test on the untransformed scores. However, the Pearson's r test does not appear to have been as rigorously tested to determine the effects of violating the assumptions of the test. Therefore, the Pearson's r was computed using both the transformed scores, in order to be consistent with Johns (1977), and the untransformed scores for means of comparison. The computational procedure was carried out according to the method of Meddis (1975, chapter eight).

6. Table 27 (section 14.0, chapter one) of the accumulation of the various sleep stages in 15 minute epochs over a period of one hour was drawn up for both the insomniac and good sleep group. The aim of this was to determine whether there were any distinguishing features between the two groups during the first hour of sleep. Any differences may account for the insomniacs feeling that they were still awake when, according to the EEG, they were in fact asleep. It was suggested by Monroe (1967) that poor sleepers require a longer time to reach the stages of deep sleep.

The sleep parameters of major interest when studying insomniacs are sleep onset time, total sleep time, stage (W+1) and the number of stage changes, as they are the most direct indicators of how well an individual is sleeping.

In all statistical tests a P level of less than or equal to .05 was considered to be statistically significant.

11.0 RESULTS

None of the treatment conditions, namely cycloid vibration, Horlicks or cocoa produced any significant changes with respect to the baseline for any sleep parameter. There were no significant differences between the treatments.

The insomniac group was found to exhibit significantly greater inter-subject variability than the good sleepers for sleep onset time, ($F = 4.7$, df 2, 7, $P \leq .05$). There was only one variable, the number of stage changes, for which the insomniacs exhibited greater night-to-night variability than the good sleepers, ($t = 2.4$, df 14, $P < .05$, two tailed test). No trend effects were found for sleep onset times.

There was no significant correlation between the prior estimated sleep onset times and the sleep onset times as measured objectively on the third night in the laboratory, using both the transformed ($r = 0.39$) and the raw scores ($r = 0.45$). The actual sleep onset time, as measured objectively for the initial 13 subjects, had an arithmetic mean of 25 minutes and a geometric mean of 21 minutes. The lower range of estimated sleep onset times had an arithmetic mean of 41 minutes and a geometric mean of 32 minutes.

12.0 DISCUSSION OF RESULTS

These non-drug "sleeping aids" were found to be ineffective in reducing the objectively measured sleep onset times in this group of sleep onset insomniacs. However, one could argue from the mean baseline sleep onset times given in Table 22 (section 14.0, chapter one), that there was little room for improvement in some cases, where the mean baseline sleep onset time was less than 30 minutes. Maybe these "aids" ought to have been evaluated upon a group of sleep onset insomniacs suffering from longer sleep onset times than the present group. It is generally believed that the majority of more severe insomniacs are likely to be receiving medical treatment for their sleeping difficulty, which would therefore eliminate them from the present study. Nevertheless, with reference to Table 22 (section 14.0, chapter one) subjects 2 and 4, with mean baseline sleep onset times of 59 and 50 minutes respectively, appeared to be suffering from relatively long sleep onset times. The only treatment which brought about a slight reduction in their sleep onset times was cycloid vibration, which reduced the sleep onset time of subject 4 by 10 minutes. Upon administration of Horlicks the sleep onset time of subject 4 increased by 11 minutes. Subjects 3 and 8 were also worthy of the title insomniacs, according to EEG criteria, with mean baseline sleep onset times of 37 and 46 minutes respectively. These two subjects showed a decline in sleep onset times upon administration of all the experimental conditions, but this was most marked for the cocoa control condition. Therefore, even by examining the responses of these four insomniacs no clear pattern emerges in response to the "aids".

Another interesting observation was that, as expected, the insomniacs of the present study, in general, were poor estimators of their sleep onset

time, even when the lower of the range of estimates was taken. Consideration must be given to the fact that the data used for computing the correlation coefficient were to some extent limited, as they were based on only one night's EEG recorded sleep onset time and upon the subjective estimate of sleep onset time prior to the commencement of the study. However, it did indicate this problem of overestimation of sleep onset times by insomniacs. Only three of the 13 subjects originally selected can be classified as insomniacs according to EEG criteria (Table 10, section 14.0, chapter one), that is, a sleep onset time of at least 30 minutes. Nevertheless, not all of the researchers who have studied insomnia objectively have reported that they experienced this problem of overestimation to the same extent. For example, in the Borkovec and Weerts (1976) study there was good agreement between the group mean reported sleep onset times of three groups of insomniacs and the group mean EEG recorded sleep onset times, as measured to the first occurrence of stage 2 sleep. There was no individual subject data provided, but the authors did comment that there was considerable variability between the lengths of the individual sleep onset times.

The present group of subjects, mean age 25.6 years, with an average sleep onset time of 34 minutes to some extent matched the young group of subjects in the Brězinová and Oswald (1972) study, mean age 22 years and average sleep onset time equal to 27 minutes. However, the subjects used in the Brězinová and Oswald (1972) study were not self-reported insomniacs. In neither the present study nor the Brězinová and Oswald (1972) study was Horlicks found to reduce the sleep onset times by a significant amount. Brězinová and Oswald (1972) did find that Horlicks reduced the amount of restlessness in the latter hours of sleep, when measured by an increase in submental EMG, but not according to the

number of stage changes. In view of section 2.2, chapter one, it would appear that muscle tension measured at only one muscle site is not necessarily a good indicator of general muscle tension throughout the body. Stage (W+1) and the number of stage changes alone were used as indicators of restlessness in the present study, because it seems reasonable to assume that the subjects may only be aware of the "tossing and turning" associated with wakefulness or a stage change, regardless of the submental EMG level. Although the ANOVA revealed no significant differences between the nights for these measures of restlessness, which supported the findings of Brězinová and Oswald (1972), the insomniacs were found to exhibit greater night-to-night variability for the number of stage changes than the good sleepers. This was the only parameter which yielded a significant difference in the night-to-night variability comparison between good sleepers and the self-reported insomniacs.

The group of self-reported, sleep onset insomniacs used in this study showed much less night-to-night variability when compared with the control group than Karacan et al.'s (1973) group of insomniacs. Furthermore, they only showed significantly higher inter-subject variability than the control group for sleep onset time. The reasons for this could be:

1. Karacan's group consisted of mixed types of insomniacs: some had long sleep latencies and others early morning awakenings. The present group reported difficulty in falling asleep, but some subjects on some nights failed to satisfy the EEG criteria for a sleep onset insomniac. Furthermore, the present group's claim that they did not suffer from nocturnal awakenings was supported by the EEG measures, with a group mean total sleep time of 428 minutes and the amount of stage (W+1) equal to 19 minutes on

the two baseline nights.

2. Only one adaptation night was used by Karacan et al. (1973), whilst the present study allowed three adaptation nights.
3. Karacan et al. (1973) expressed the various sleep parameters as percentages, which will vary as the total sleep time varies. In the present study the sleep parameters were expressed in absolute minutes of a fixed length of total sleep time which was common to all subjects on all nights.
4. Unlike the present study, Karacan et al.'s (1973) experimental and control groups were matched.
5. In comparison with the present study, Karacan et al. (1973) examined a wider range of sleep parameters, such as the ratio of minutes of REM sleep to non REM sleep.

There are two major shortcomings of this present insomnia study:

1. Due to the difference between the subjectively reported sleep onset times and the EEG measured sleep onset times, at the end of the subject selection procedure, there were insufficient subjects to study the number of experimental conditions in a balanced, well-designed manner.
2. According to the EEG criteria, apart from subjects 2, 3, 4 and 8 (Table 22, section 14.0, chapter one) it is debatable whether the other subjects should be classified as insomniacs. This group of eight insomniacs, as a whole, just satisfied the EEG criteria for a group of sleep onset insomniacs, with a group mean sleep onset time of 34 minutes. However, there was no significant difference between the group mean baseline sleep onset times of the insomniacs and good sleepers (Table 24, section 14.0, chapter one), when compared by an unrelated t test, ($t = 0.2$, df 14, two

tailed test).

One would have expected the self-reported, sleep onset insomniac group to have had a significantly greater group mean sleep onset time than the good sleep group. Also worthy of comment is the fact that five subjects from the good sleep group had a sleep onset time greater than 30 minutes, whereas, only four subjects from the self-reported, sleep onset insomniac group had a sleep onset time greater than 30 minutes. Two subjects in the good sleep group, subjects 1 and 6 (Table 24, section 14.0, chapter one), actually had longer sleep onset times than the insomniacs, yet they considered themselves to be good sleepers. The difference between the two groups appeared to be due to the subjects' expectations and attitudes concerning sleep and not solely to a time difference. Nevertheless, the experimenter was to some extent justified in using such a group of insomniacs because:

i) all the subjects did claim to be suffering from sleep onset times of at least 30 minutes,

ii) none of these subjects had sought medical attention for their sleeping difficulty and it is therefore highly probable that such a group would try various non-drug "sleeping aids".

Ideally, more advertisements should have been made calling for more "insomniacs". However, since 69 per cent of the subjects originally chosen had failed to satisfy the EEG criteria for sleep onset insomnia it was predicted that, with this failure rate, selecting more subjects by EEG methods would be a time consuming process. Due to time restraints, this proposal was not feasible. Furthermore, the majority of individuals

who claimed to have relatively long sleep onset times were receiving medical treatment for their sleeping difficulty, which therefore eliminated them from the present study.

"Why did all the subjects considered for this study initially present themselves as insomniacs?" "What makes the majority of them believe that they are awake, when according to the EEG criteria they are asleep?" The following points are put forward as possible explanations:

1. The sleep of the self-reported, sleep onset insomniacs may be more disturbed and broken than that of a good sleep group, during the first hour of sleep. This may be reflected by such measures as an increased amount of stage (W+1) or an increased number of stage changes. Table 27, (section 14.0, chapter one) which was compiled by calculating for each subject the accumulation of the various sleep parameters in 15 minute epochs during the first hour of sleep and then expressing them as group means, clearly shows a marked similarity between the two groups. However, the insomniacs did show more stage changes than the good sleepers during the first 15 minute epoch, ($t = 2.36$, df 14, $P \leq .05$, two tailed test). This may have created the feeling that they had been awake for the first 15 minute epoch.

2. The insomniacs might fall asleep, then awake after about 40 minutes and then quickly fall asleep again. Maybe the sleep onset insomniac measures his sleep onset time from the time of "lights out" until he falls asleep the second time. The lengths of such intervals were calculated for the two baseline nights and expressed as the average, (Table 26, section 14.0, chapter one). The group arithmetic mean sleep onset time increased to 243 minutes and the

geometric mean to 196 minutes. There was no significant correlation between the subjective and the recalculated objective sleep onset times. Subject 4 was the only individual whose subjective estimate was in better agreement with the recalculated EEG sleep onset time. In general, the time to the occurrence of the second sleep onset does not offer a satisfactory explanation for the problem of overestimation of sleep onset time by the insomniacs.

3. There is the possiblity that the change of sleeping environment may have alleviated the sleeping difficulty to some extent, without the subjects commenting upon it. In some cases the insomnia may have been conditioned to the subject's bedroom. Therefore, simply a change in the environment would tend to aid sleep. However, during the morning conversations with the subjects the general response was that they felt as though they had slept the same as they usually did at home, thus failing to support this idea. It could be that the insomniacs themselves were unaware of the improvement.

4. Another reason could be that the entire laboratory setting may have been acting as a placebo. A lot of attention was being paid to the insomniacs in the laboratory setting. Someone was actually interested in their complaints of a sleeping difficulty and the subjects were aware that the experimenter remained awake in the laboratory all-night, a factor which they may have found reassuring. Thus, it is possible that the insomniacs could sleep better in the laboratory than at home. However, as mentioned in (3), the subjects' claims did not support this idea.

5. The insomniacs, sometime previously, may have experienced a period of relatively long sleep onset times due, maybe, to some temporary anxieties or worries. As a result of this episode they may have come to label themselves as "insomniacs", even though they experience insomnia only on a very irregular basis. This point can be further illustrated by reference to the findings of the present study, (Table 23, section 14.0, chapter one). For example, subject 6 on the fourth treatment night experienced a long sleep onset time of 126 minutes, whereas, on every other laboratory night his sleep onset time had not exceeded 30 minutes, according to EEG criteria. If this self-reported insomniac had been the subject of an EEG assessment of sleep onset times on any night but the fourth treatment night, he would not have been classified as an insomniac, according to the EEG definition.* However, on the fourth treatment night he would have been classified as suffering from severe, sleep onset insomnia. It would therefore appear that an important feature of insomnia is the periodic occurrence of a long sleep onset time, on the basis of which the individual may label himself as an insomniac. Although this is a very interesting feature, it presents a problem for the objective study of sleep onset insomnia.

There is no clear indication as to which subjects may experinece a long sleep onset on a particular night. This makes it impossible for the experimenter to determine whether a sudden change in sleep onset time on a particular night is due to

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* The EEG definition of sleep onset insomnia is a sleep onset time of at least 30 minutes. The sleep onset time being defined as the time from "lights out" until the first occurrence of a stage 2 period of at least five minutes duration.

the effect of the treatment or due to the symptom insomnia itself. It is also likely to be a costly procedure, as several nights recording may be required before the occurrence of a long sleep onset time. This feature of insomnia was considered previously in the subject selection procedure, where five subjects were considered unsatisfactory on the basis of one night's EEG sleep onset time. The point was raised as to whether these subjects may have experienced a long sleep onset time if they had been studied for several nights. Even if they did, it would be a very inefficient and unsatisfactory approach to the objective evaluation of "sleeping aids", for the reasons given above.

6. Consideration must also be given to the fact that insomniacs who would satisfy the EEG criteria may not be inclined to volunteer to take part in laboratory studies; also many of them may be receiving treatment for their sleeping difficulty which would prevent them from taking part in the present study.

Finally, one of the most interesting observations made during the administration of the sleep questionnaire to the 45 self-reported insomniacs was that many reported frequently taking Aspirin, which they claimed relieved their insomnia. This is quite extraordinary, for Aspirin is widely known for its analgesic, antipyretic and anti-inflammatory actions, but not for any hypnotic activity. On the contrary, the gastric-irritation effect of Aspirin might be expected to disturb sleep rather than improve it. Clift (1975) listed analgesics amongst a number of treatments, such as antihistamines, cough suppressants and nasal decongestants, which he had prescribed for various complaints of

insomnia. Clift (1975) did not name the analgesics which he prescribed, but it is unlikely that Aspirin would be included, except, maybe, in some cases where the reported insomniac was suffering from rheumatoid arthritis. A doctor would normally prescribe more potent analgesics. However, there is the possiblity that on some occasions insomnia is symptomatic of an underlying cause such as a headache which could be alleviated upon administration of a non-prescription analgesic such as Aspirin. It seems unlikely that all those who reported taking Aspirin to relieve their insomnia were suffering from an underlying cause which could be treated by one dose of Aspirin. It is also possible that Aspirin may be acting as a placebo treatment in some cases, but it does seem an unusual choice of placebo treatment for one generally associates various bedtime beverages with sleep, rather than Aspirin tablets. Furthermore, there are no published studies upon the effects of Aspirin (acetylsalicylic acid) upon sleep. This observation therefore opened up a new line of inquiry.

13.0 CONCLUSIONS AND FURTHER RESEARCH

None of the "sleeping aids" were found to produce any significant improvement, either in the quality or quantity of sleep, in the sleep onset insomniac group studied. This conclusion cannot necessarily be applied to the population of sleep onset insomniacs, since, whilst the sleep onset insomniac group studied appeared to suffer from sleep onset insomnia as judged by subjective criteria, their sleep onset times, according to the EEG criteria, were not significantly different from the group of good sleepers. In fact, some of the self-reported good sleepers actually had longer sleep onset times than the sleep onset insomniac group, when measured by means of the EEG.

A major component of the complaint of insomnia appears to be the individuals' own attitudes and expectations with respect to sleep rather than an absolute time difference between insomniacs and good sleepers. Insomnia is therefore predominantly a subjective complaint of sleeping difficulty. This was supported by the fact that the majority of self-reported insomniacs overestimated their sleep onset times, according to the EEG measure. This phenomenon of overestimation has been reported by several investigators. Monroe (1967) found that his poor sleepers had a group mean estimated sleep onset time of 60 minutes and a group mean objective sleep onset time of 15 minutes. The term objective sleep onset time refers to the sleep onset time measured by means of the EEG. Freedman and Papsdorf's (1976) insomniac group had a group mean estimated sleep onset time of 81 minutes and a group mean objective sleep onset time of 43 minutes. Finally, Carskadon, Dement, Mitler, Guilleminault Zarcone and Spiegel (1976) reported that their insomniac group had a mean estimated sleep onset time of 62 minutes and a mean objective sleep onset time of 26 minutes. In the present study, the original insomniac group, prior to screening, had a mean estimated sleep onset time of 41 minutes and a group mean objective sleep onset time of 25 minutes, (Table 10, section 14.0, chapter one). There did not appear to be a common overestimation factor for the groups of insomniacs, as they overestimated by factors of 4, 2, $2\frac{1}{2}$ and $1\frac{1}{2}$ in each study respectively. It can be seen therefore that there is no easy solution for obtaining objective sleep onset times from subjective estimates.

Nevertheless, although the majority of insomniacs of the present and previous studies failed to satisfy the EEG criteria for a sleep onset insomniac, the fact that they reported sleeping difficulties still qualifies them as insomniacs from a subjective point of view.

These individuals do have a problem, for if a subject reports that he has a sleep onset time of 45 minutes and if this is actually troubling him, then he is effectively an insomniac regardless of the EEG measure.

Therefore, the subsequent, logical line of approach would be to embark upon a self-report questionnaire study which covered attitudes toward sleep, subject expectancies and the subjects' belief in the treatment. This would be an interesting line of investigation and a self-report study could be carried out with the "aids" considered in the present study, under both positive and counterdemand instruction conditions. If the subject reported that he fell asleep faster after, for example, a night-time beverage, he is effectively cured of his insomnia. However, this line of approach is somewhat outside the sphere of competence of the experimenter and although proficiency could be gained in this field, the question raised is whether this approach would solve the difficulties encountered in attempting to evaluate objectively various non-drug "sleeping aids". In short the answer is no. Although the subjective evaluation of the various "sleeping aids" would provide some useful information with respect to the efficacy of an "aid", a major failure of this approach is that it would fail to discriminate between those subjects who were actually overestimating their sleep onset times and those who were fairly good estimators.

In order to illustrate this criticism of the subjective approach the findings of the present study will be employed. Out of the 13 self-reported insomniacs only three were found to be fairly good estimators of their sleep onset times in comparison with the EEG measure, (Table 10, section 14.0, chapter one). It would appear that the

complaint of sleep onset insomnia by the other ten subjects is predominantly psychological, on the majority of occasions. These ten subjects might therefore be expected to report improvement in response to almost any "aid", whereas, the other three subjects, whose claim of long sleep onset times was confirmed by the EEG, may not report any change. Since sleep onset insomniacs who are good estimators of their sleep onset times are in the minority, their responses are likely to be weakened in any self-report group study. A "sleeping aid" could therefore be labelled as an effective treatment for sleep onset insomnia, when in fact, it may fail to help those who appear to be most in need.

As mentioned previously, these "aids" should ideally be evaluated objectively on a group of insomniacs with longer sleep onset times than those of the present study. However, similar problems to those encountered in the present study (some of which have been listed below) are likely to be experienced.

1. Difficulties were encountered in obtaining a group of sleep onset insomniacs who satisfied EEG criteria mainly because, in general, they tended to overestimate their sleep onset times. Furthermore, there does not appear to be a common overestimation factor, so there is no easy solution for obtaining objective sleep onset times from subjective estimates.

2. There is also the problem of obtaining drug-free insomniacs. Hypnotics and sedatives are the most widely prescribed treatment for insomnia and this treatment may lead to the problem of drug withdrawal insomnia, which may continue for several days or weeks depending on the type of drug, the dosage level and the duration of usage. It is not simply a matter of asking the subject to

refrain from taking the drug for a week prior to the commencement of the study.

3. The sleep of insomniacs has been shown to exhibit a high amount of night-to-night variability in comparison with a good sleep group (Karacan et al, 1973). However, the insomniac group of the present study only showed significantly higher night-to-night variability than a good sleep group on one sleep parameter, the number of stage changes. Various possible reasons for this difference are listed in section 12.0, chapter one.

4. Furthermore, psychological and somatic disorders have been found to be associated with some complaints of insomnia. It is therefore unlikely that a "sleeping aid" would be able to show its potential in such cases, where a treatment for the underlying cause of insomnia may be more effective.

In view of the difficulties which were encountered in the evaluation of non-drug "sleeping aids" using a group of self-reported, sleep onset insomniacs, it would be very useful if an EEG model of sleep onset insomnia could be created in the laboratory with good sleepers. Essentially, the aim of the model would be to provide a long sleep latency which would form the basis of the initial evaluation procedure for various "sleeping aids", be they drug or non-drug. It was decided to follow up this idea by means of a laboratory study.

One further line of investigation worth pursuing concerns the use of Aspirin by insomniacs to relieve their sleeping difficulty, as commented upon in the previous section. It was decided to investigate

the grounds, if any, for the claims that Aspirin aids sleep. It may appear rather contradictory that a further research proposal emanating from a study evaluating non-drug "sleeping aids" should be concerned with a possible drug "aid" for insomnia. However, these two approaches to the treatment of insomnia are not necessarily contradictory. As discussed in section 2.1, chapter one, the causes of insomnia are many and diverse, therefore, it is possible that drug "aids" may be beneficial to some insomniacs, whilst others may prefer non-drug "aids".

On the basis of the findings of this initial study the research programme will now proceed along two lines of inquiry:

1. to create an EEG model of sleep onset insomnia,
2. to investigate the effects of Aspirin (acetylsalicylic acid) upon sleep.

Section 14.0

RESULTS TABLES AND GRAPHS FOR CHAPTER ONE

14.1 Explanatory Notes for the Results Tables

All means in the results tables are expressed to the nearest whole number.

All figures in the results tables are expressed in minutes unless otherwise stated.

Key:

A.S.A - Analysed Sleep Time refers to the amount of each sleep record analysed from the time of sleep onset. A.S.T. is the lowest common measure of total sleep time for all subjects on all nights.

B - The average score for the two baseline nights.

C - The average score for the two cocoa nights.

H - The average score for the two Horlicks nights.

R - The score on the recovery night.

V_1 - The score on the first cycloid vibration night.

V_2 - The score on the second cycloid vibration night.

V - The average score for the two cycloid vibration nights.

Number of Stage Changes - A stage change was counted each time a subject passed from one stage of sleep into another, providing the length of time spent in any particular stage was at least two minutes.

Results Tables for the study investigating the effects of Cycloid Vibration upon the sleep of Good Sleepers.

Stage W+1 — Table 2

Treatment / Subject	B	V_1	V_2	R
1	8	15	9	6
2	41	20	21	8
3	27	37	21	14
4	35	31	24	42
5	19	13	22	22
6	8	11	8	5
Mean	23	21	18	16
S.D.	13.8	10.6	7.1	14.1

Stage 2 — Table 3

Treatment / Subject	B	V_1	V_2	R
1	205	222	204	222
2	226	224	222	216
3	194	181	180	207
4	188	222	183	172
5	254	245	219	232
6	210	191	222	243
Mean	213	214	205	215
S.D.	24.1	23.7	19.4	24.6

Stage 3 — Table 4

Treatment / Subject	B	V_1	V_2	R
1	44	30	35	36
2	28	45	42	28
3	35	35	33	33
4	23	31	33	30
5	35	38	35	37
6	25	21	27	30
Mean	32	33	34	32
S.D.	7.8	8.1	4.8	3.6

A.S.T. for the above three tables is equal to 450 minutes

Stage 4 — Table 5

Treatment / Subject	B	V_1	V_2	R
1	82	99	86	77
2	75	73	70	89
3	88	95	97	99
4	75	75	68	72
5	51	55	62	44
6	84	109	100	71
Mean	76	84	81	75
S.D.	13.2	20.1	16.1	18.8

Stage 3+4 — Table 6

Treatment / Subject	B	V_1	V_2	R
1	126	129	121	113
2	103	118	112	117
3	123	130	130	132
4	98	106	101	102
5	86	93	97	81
6	109	130	127	101
Mean	108	118	115	107
S.D.	15.2	15.3	13.7	17.3

Stage REM — Table 7

Treatment / Subject	B	V_1	V_2	R
1	111	84	116	109
2	80	88	95	109
3	106	102	119	97
4	129	91	142	134
5	91	99	112	115
6	123	118	93	101
Mean	107	97	113	111
S.D.	18.7	12.3	18.0	13.0

A.S.T. for the above three tables is equal to 450 minutes.

Sleep Onset Time — Table 3

Treatment / Subject	B	V_1	V_2	R
1	5	9	5	5
2	15	13	19	21
3	14	18	19	13
4	25	22	16	17
5	25	24	47	48
6	24	26	20	17
Mean	18	19	21	20
S.D.	8.1	6.6	13.9	14.7

Total Sleep Time — Table 9

Treatment / Subject	B	V_1	V_2	R
1	477	463	466	456
2	467	450	472	453
3	465	459	456	462
4	457	467	459	453
5	451	453	450	460
6	470	450	468	458
Mean	465	457	462	457
S.D.	9.3	7.1	8.3	3.7

Comparison Between the Sleep Onset Times as measured by EEG criteria on the Third Adaptation Night and the Subjects' Estimates prior to the commencement of the Study.

Table 10

Subjects	EEG Criteria	Estimate	
A	11	45 - 60 minutes	x
B	25	One hour or more	
C	17	1 - 2 hours	
D	26	30 - 45 minutes	
E	48	1 - 2 hours	
F	8	30 - 45 minutes	x
G	11	30 - 45 minutes	x
H	23	30 - 45 minutes	
I	26	30 - 60 minutes	
J	8	30 - 60 minutes	x
K	40	30 - 45 minutes	
L	63	About one hour	
M	13	45 minutes	x
Arithmetic Mean	25	41	
S.D.	16.8	13.9	
Geometric Mean	21	32	

$r = 0.45$ raw scores n.s.
$r = 0.39$ transformed scores ($\log_e$) n.s.

The subjects marked with a "x" were not allowed to take part in the study after the first EEG recording night.

Results Tables for the study investigating the effects of non-drug "sleeping aids" upon the sleep of self reported, sleep onset Insomniacs.

Stage W+1 — Table 11

Treatment / Subject	B	V	H	C
1	25	15	1	10
2	26	9	14	6
3	16	4	8	5
4	12	5	9	5
5	24	24	7	63
6	3	13	10	11
7	30	18	21	22
8	19	16	44	35
Mean	19	13	14	20
S.D.	8.8	6.8	13.3	20.4

Stage W+1 - First Half of Night — Table 12

Treatment / Subject	B	V	H	C
1	13	7	0	2
2	13	5	10	4
3	9	4	5	3
4	1	2	9	1
5	7	6	3	11
6	1	8	9	1
7	12	7	16	9
8	6	6	29	20
Mean	8	6	10	6
S.D.	4.9	1.9	9.1	6.6

Stage W+1 - Second Half of Night — Table 13

Treatment / Subject	B	V	H	C
1	12	8	1	8
2	13	4	4	2
3	7	0	3	2
4	11	3	0	4
5	17	18	4	52
6	2	5	1	10
7	18	11	5	13
8	13	10	15	15
Mean	12	7	4	13
S.D.	5.2	5.7	4.7	16.4

A.S.T. for the above three tables is equal to 380 minutes

Stage 2 — Table 14

Treatment / Subject	B	V	H	C
1	154	145	136	183
2	196	187	203	157
3	177	190	153	173
4	143	171	147	162
5	173	157	165	138
6	188	187	160	198
7	174	216	198	193
8	166	164	163	182
Mean	171	177	166	173
S.D.	17.2	22.4	23.5	20.0

Stage 3 — Table 15

Treatment / Subject	B	V	H	C
1	14	29	36	27
2	20	23	19	25
3	25	43	32	28
4	32	37	37	44
5	18	32	32	15
6	27	21	30	21
7	34	38	37	38
8	28	31	22	25
Mean	25	32	31	28
S.D.	6.9	7.5	6.8	9.2

Stage 4 — Table 16

Treatment / Subject	B	V	H	C
1	97	90	82	86
2	75	79	71	88
3	93	75	83	84
4	73	82	92	93
5	80	84	85	84
6	90	95	94	81
7	72	41	50	49
8	64	82	61	56
Mean	81	79	77	78
S.D.	11.6	16.4	15.4	16

A.S.T. for the above three tables is equal to 380 minutes.

Stage 3+4 — Table 17

Treatment / Subject	B	V	H	C
1	111	119	118	113
2	95	102	90	113
3	118	118	115	112
4	105	119	129	137
5	98	116	117	99
6	117	116	124	102
7	106	79	87	87
8	92	112	83	81
Mean	105	110	108	106
S.D.	9.7	13.7	18.1	17.5

Stage REM — Table 18

Treatment / Subject	B	V	H	C
1	91	102	125	75
2	64	83	73	104
3	70	70	105	92
4	118	86	95	76
5	85	84	91	80
6	73	65	86	69
7	70	68	72	77
8	103	88	91	82
Mean	84	81	92	82
S.D.	18.8	12.4	17.2	11.1

REM Latency — Table 19

Treatment / Subject	B	V	H	C
1	129	65	58	155
2	154	107	138	49
3	80	144	92	107
4	97	93	131	105
5	109	112	142	123
6	111	211	134	133
7	98	75	70	65
8	88	55	201	119
Mean	108	108	121	107
S.D.	23.8	50.1	46	34.9

A.S.T. for the above three tables is equal to 380 minutes.

Number of Stage Changes — Table 20

Treatment / Subject	B	V	H	C
1	28	31	28	39
2	46	32	47	50
3	50	45	34	26
4	54	47	43	46
5	50	40	55	32
6	29	46	43	42
7	82	66	49	59
8	51	40	39	52
Mean	49	43	42	43
S.D.	16.8	11	8.6	10.8

A.S.T. for the above table is equal to 380 minutes

Total Sleep Time — Table 21

Treatment / Subject	B	V	H	C
1	420	429	451	424
2	409	449	400	440
3	428	437	437	439
4	453	434	417	408
5	463	434	454	418
6	386	389	390	381
7	463	425	465	418
8	398	400	396	415
Mean	428	425	426	418
S.D.	29.7	20.1	29.3	18.7

Sleep Onset Time — Table 22

Treatment / Subject	B	V	H	C
1	22	10	8	22
2	59	56	60	63
3	37	23	19	17
4	50	40	61	56
5	22	32	16	23
6	12	11	18	70
7	22	21	21	21
8	46	32	32	13
Mean	34	28	29	36
S.D.	16.7	15.3	20.3	23.2
Geometric Mean	30	24	24	30

Sleep Onset Times for Each Treatment Night

Table 23

Nights / Subjects	B	1	2	3	4	5	6
1	22	18	10	11	8	5	26
2	59	63	60	63	61	49	64
3	37	26	18	15	22	26	20
4	50	56	41	43	37	61	18
5	22	38	26	12	19	20	25
6	12	23	9	12	126	13	11
7	22	28	14	17	25	13	20
8	46	12	32	23	39	32	11
Mean	34	33	26	25	42	27	24
S.D.	16.7	18.1	17.6	18.8	37.5	19.2	16.9
Geometric Mean	30	29	22	20	31	21	21

Mean Baseline Sleep Onset Times for Good Sleep Group and Insomniacs

Table 24

Subjects	Insomniacs	Good Sleepers
1	22	60
2	59	26
3	37	32
4	50	36
5	22	21
6	12	31
7	22	12
8	46	39
Mean	34	32
S.D.	16.9	14.2
Geometric Mean	30	29

$t = 0.2$, $df = 14$ n.s.

Table 25

Comparisons Between the Insomniac Group and the Good Sleep Group

a) The comparison of the inter-subject variability within the two groups by means of the between subject variance estimates, yielding an F ratio.

b) The comparison of the night-to-night variability by using an unrelated t test on the logarithmic mean standard deviations across the nights, yielding a t value.

Stage W+1

a) F_{max} = 1.1, df 2,7 , n.s.

b) t = 0.6, df 14 , n.s.

Stage 2

a) F_{max} = 1.1, df 2,7 , n.s.

b) t = 1.4, df 14 , n.s.

Stage 3

a) F_{max} = 1.9, df 2,7 , n.s.

b) t = 1.1, df 14 , n.s.

Stage 4

a) F_{max} = 1.1, df 2,7 , n.s.

b) t = 1.5, df 14 , n.s.

Stage 3+4

a) F_{max} = 2.1, df 2,7 , n.s.

b) t = 0.3, df 14 , n.s.

Stage REM

a) F_{max} = 1.1, df 2,7 , n.s.

b) t = 1.9, df 14 , n.s.

Total Sleep Time

a) F_{max} = 1.5, df 2,7 , n.s.

b) t = 1.2, df 14 , n.s.

REM Latency

a) F_{max} = 1.8, df 2,7 , n.s.

b) t = 1.0, df 14 , n.s.

Number of Stage Changes

a) F_{max} = 1.3, df 2,7 , n.s.

b) t = 2.4, df 14 , $P \leq .05$, two tailed test

Sleep Onset Time

a) F_{max} = 4.7, df 2,7 , $P \leq .05$

b) t = 1.6, df 14 , n.s.

Comparison of the Sleep Onset Time, as Measured from the Time of "Lights Out" until the Time to return to sleep after the First Awakening, and the Estimates of Sleep Onset Time.

Table 26

	EEG	Estimate
Subject		
1	195	30
2	328	60
3	49	30
4	74	60
5	460	30
6	202	60
7	332	30
8	303	60
Arithmetic Mean	243	45
Geometric Mean	196	42

The EEG time is the mean of the two baseline nights.

The estimated time is the lower of the range of the subjects' estimates of their sleep onset time prior to the commencement of the study.

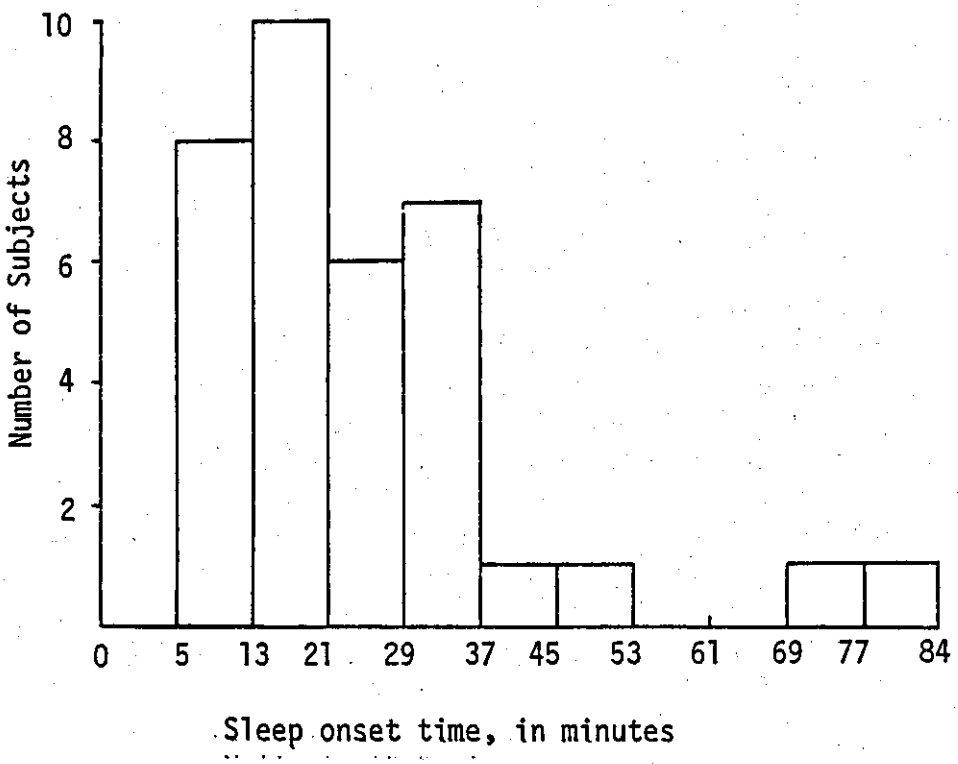
r = 0.12, n.s., raw scores.

$r = 1.7 \times 10^{-3}$, n.s., transformed scores, $(\log_e)$

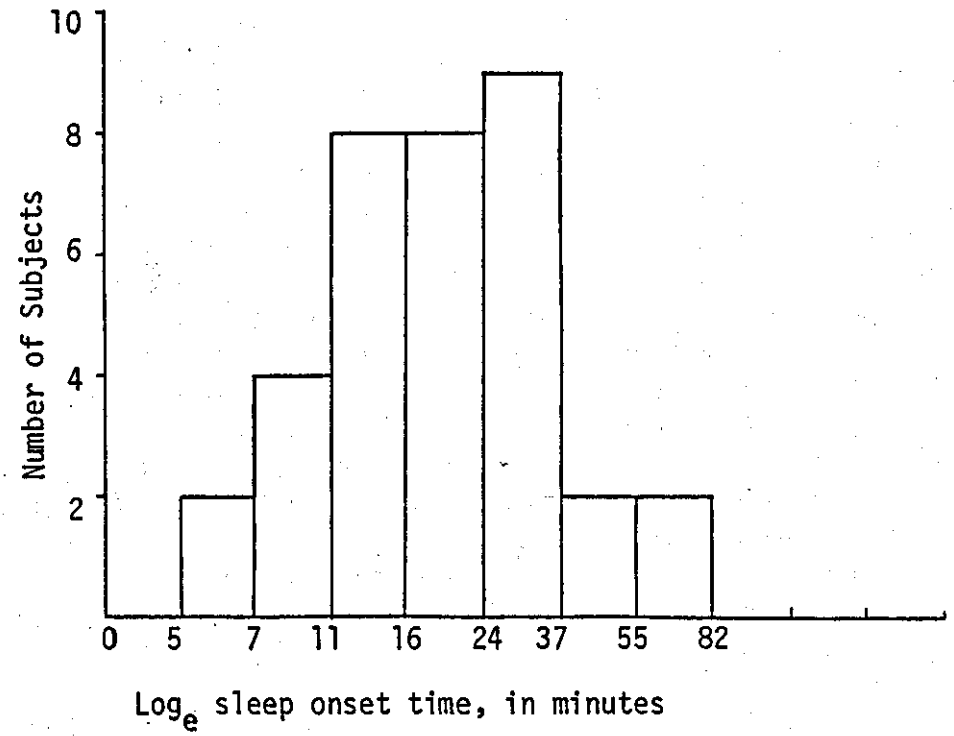
A Comparison Between the First Hour of Sleep of Good Sleepers and Insomniacs.

Table 27

	Good Sleepers		Insomniacs	
	Mean	S.D.	Mean	S.D.
First Epoch				
Stage W+1	0.1	0.4	0.3	0.4
2	7.8	2.4	8.9	2.9
3	3.9	1.4	2.5	2.1
4	3.2	2.9	3.3	2.5
No. Stage Changes	3.6	1.6	2.1	0.4
Second Epoch				
Stage W+1	0.1	0.4	0.3	0.5
2	1.1	1.6	1.1	1.8
3	1.5	1.4	1.3	1.3
4	12.3	2.6	12.3	2.7
No. Stage Changes	1.6	1.6	2.1	1.8
Third Epoch				
Stage W+1	0	0	0.6	1.1
2	2.1	2.9	2.3	3.5
3	1.5	1.1	0.6	0.7
4	11.4	3.3	11.5	4.7
No. Stage Changes	2.4	1.7	1.5	1.7
Fourth Epoch				
Stage W+1	0.9	0.8	0.6	0.9
2	4.8	4.0	5.3	5.1
3	1.3	0.9	1.6	1.4
4	6.3	4.7	7.5	4.9
REM	1.7	2.0	0	0
No. Stage Changes	2.6	1.5	2.3	1.1
Latency to Stage 4	13.8	5.5	14.4	5.4



Graph A: Frequency Distribution of Sleep Onset Times (Linear Scale)



Graph B: Frequency Distribution of Sleep Onset Times (Logarithmic Scale)

Chapter Two - Part I

AN EEG MODEL OF SLEEP ONSET INSOMNIA

1.0 INTRODUCTION

The previous study (chapter one), which was an objective evaluation of non-drug "sleeping aids" conducted with a group of self-reported, sleep onset insomniacs, revealed some of the many difficulties which can be encountered when using insomniac subjects. Briefly, the major problems encountered were:

1. the self-reported insomniacs frequently failed to satisfy the EEG criteria for sleep onset insomnia;
2. many insomniacs were receiving hypnotics or sedatives for their sleeping difficulty;
3. the sleep onset times of the insomniacs might be expected to show a large amount of night-to-night and inter-subject variability (Karacan et al., 1973); for example, if a reported sleep onset insomniac showed a long sleep latency, according to EEG criteria, on one laboratory night, it would not necessarily follow that a long sleep latency would be observed on every other night;
4. psychological and somatic disorders have been found to be associated with various complaints of insomnia.

These points were explained in more detail in section 13.0, chapter one.

For the reasons given above, it would be very advantageous if an EEG model of sleep onset insomnia could be created with good sleepers. The main aim of the model would be to provide a long sleep latency which would form the basis of an evaluation procedure for various possible treatments of insomnia. Although it is essential that any potential treatment for insomnia should be evaluated with a group representative of the population for which it is intended, an EEG model would be very useful in the initial evaluation procedure, be it for a possible drug or non-drug "aid". If a particular "aid" was found to reduce sleep

onset time within the model group, one would then be justified in going to the lengths of selecting a group of insomniacs which satisfied the EEG criteria. Karacan, Thornby, Booth, Okawa, Salis, Anch and Williams (1975) commented that there was a great need in sleep research for a model of insomnia. They proposed the possibility of administering pharmacological agents to good sleepers in order to produce model insomnia.

Okuma and Honda (1978) attempted to create such a model by using two approaches. One was to pipe white noise into the subjects' bedrooms, and the other was to give the subjects the drug methylphenidate, a central nervous stimulant. Although both methods increased the sleep onset time (baseline group mean of 20.2 minutes), the drug (group mean 40.8 minutes) being more effective than the white noise (group mean 31.2 minutes), neither increase was significant when compared with the baseline. This was rather unexpected, since the increase did appear to be high under the drug condition. It may have failed to reach statistical significance because of a large amount of inter-subject variability. However, the authors did not comment on this point and they did not present the standard deviations for the group mean sleep onset times: therefore, the idea of subject variability is only speculative. The authors claimed that both methods disturbed sleep, as measured by a decreased total sleep time, stage 4 and stage REM an increased stage W. They reported that this disturbance was reduced by the hypnotics Flurazepam and Triazolam. They were, however, unsuccessful in producing an EEG model of sleep onset insomnia, but their model did simulate, to some extent, the insomnia associated with early morning and nocturnal awakenings. One major disadvantage of using central nervous stimulants to increase sleep onset times, is that if one wished to use the model for the initial evaluation of a potential

hypnotic/sedative, then any results obtained may be due to the interaction effects of the hypnotic/sedative and the stimulant, not simply due to the hypnotic/sedative. Many drugs produce very uncharacteristic effects under interaction conditions.

The approach adopted in the present study, in an attempt to create an EEG model of sleep onset insomnia, was to measure the sleep onset times of the daytime naps of good sleepers. This approach was chosen on the grounds that as sleep is more difficult to achieve during the daytime, then a longer sleep onset time would be expected during the day than at night. Agnew and Webb (1971) found that sleep onset time, as measured to the first appearance of stage 1 sleep, was inversely related to the length of prior wakefulness. When subjects had been awake for only four hours prior to their bedtime, the sleep onset time was approximately three times greater than normal, with a value of 36 minutes; after 12 hours of wakefulness the mean sleep onset time decreased to 18 minutes. However, no clear details of the experimental procedure were given.

A literature search revealed that the majority of daytime sleep studies had been primarily concerned with sleep cycling during napping (Maron, Rechtschaffen & Wolpert, 1964; Webb & Agnew, 1967) and with the effects of naps on nocturnal sleep (Karacan, Williams, Finely & Hursch, 1970). Most studies employed subjects who were accustomed to napping. Napping sleep did not appear to have been used previously as an EEG model of sleep onset insomnia.

2.0 METHOD

2.1 Experimental Design

A total of six nights were to be spent in the laboratory by each of eight selected subjects. The first two nights were to allow for adaptation effects (Agnew et al., 1966). The remaining four nights were all associated with daytime naps. All-night EEG recordings were taken on these nights to assess whether or not each subject had a fairly typical, good night's sleep. It is to be expected that if a subject had a particularly poor night's sleep, this would greatly influence his sleep onset time during the day. As the subjects were sleeping in the laboratory at night, the experimenter was able to keep the subjects' retiring and rising times fairly constant from day to day.

It appeared from the findings of Agnew and Webb (1971) that sleep onset time during the day would be related to the length of prior wakefulness. For this reason it was decided to have two nap conditions, a morning nap and an afternoon nap. It was hypothesised that the sleep onset time of the morning nap would be greater than that of the afternoon nap. However, this hypothesis was not borne out by the findings of Karacan et al. (1970). In this study the group mean sleep onset times for one hour nap sessions were six minutes during the morning and seven minutes for the afternoon; both naps were taken on the same day. For the two hour nap sessions the group mean sleep onset times were 13 minutes during the morning and seven minutes during the afternoon. Overall, the morning nap sleep onset times did appear to be rather short. The reasons for this could be:

1. the subjects were awoken at 6.00 a.m. every morning and then they returned to the laboratory for their morning naps at 8.00 a.m. There is the possibility that the subjects were not sleep satiated

prior to their naps. Their group mean total sleep time at night was only 372 minutes which is rather low for the subjects' age range (21 - 28 years). According to Roffwarg et al. (1966) their total sleep time would be expected to lie within the range 420 - 465 minutes.

2. Karacan et al. (1970) did not state whether their subjects were habitual nappers. Evans, Cook and Cohen (1976) found, in a study of afternoon naps, that subjects who were accustomed to daytime napping fell asleep twice as quickly as those who were unaccustomed to daytime napping.

Since a sleep onset time of between 30 and 60 minutes (as defined by EEG criteria) is associated with moderate sleep onset insomnia, the napping sessions had to be of about 60 minutes duration. It was expected that nap sessions longer than 60 minutes would be very boring and monotonous for the subjects. The morning naps were scheduled from 10.30 - 11.30 a.m. This time was chosen in order to allow the subjects sufficient time to wake and organise themselves after rising; also, it was not too close to their lunchtime and approximately $2\frac{1}{2}$ - 3 hours had elapsed since they had arisen. The afternoon naps were scheduled from 4.00 - 5.00 p.m.; 8 - $8\frac{1}{2}$ hours having elapsed since they had arisen.

The subjects were allowed only five minutes of unbroken sleep during any nap session before being awakened by the experimenter. The reasons for this were:

1. if the subjects were allowed only five minutes of daytime sleep, it seemed unlikely that this would affect their night-time sleep;

2. the aim of the study was to create a long sleep onset time and not to examine the quality of the napping sleep;
3. if the duration of the nap was kept short, it may prevent the subjects from growing accustomed to napping.

Sleep onset time, both at night and during the nap sessions, was defined as the time from "lights out" until the first appearance of stage 2 sleep which was of at least five minutes unbroken duration. For the purpose of data analysis, it was decided that, if the subjects did not fall asleep at all during the nap period, this was to be expressed as a sleep onset time of 60 minutes. The most suitable experimental design which would correspond with the subjects' commitments was:

Night	1	2	3	4	5	6
Condition	Ad	Ad	B_1	B_2	B_3	B_4
Daytime						
2 males + 2 females			A_1	M_1	M_2	A_2
" "			M_1	A_1	A_2	M_2

Key:

Ad - Adaptation

B - Baseline

M - Morning nap

A - Afternoon nap

The subjects were also asked to complete the morning questionnaire upon awakening each morning (Appendix III) and the daytime questionnaire after each napping session (Appendix IV).

2.2 Subject Selection

The selection procedure was carried out as in section 6.1, chapter one. For this particular study it was essential that the subjects were good sleepers and not accustomed to daytime napping (Evans et al., 1976). Eight, healthy subjects (four males and four females), with a mean age of 20.4 years, were selected from the student population.

2.3 Instructions to Subjects

The subjects were given the "general instructions to subjects", as discussed in section 6.2, chapter one, with the following additional instructions:

1. On the days when the subjects had to return to the laboratory they were instructed to refrain from having tea, coffee, alcohol, or drinks such as Coca Cola and snacks after breakfast/lunch. They were also instructed to have only one weak tea or coffee with their breakfast/lunch and to keep the content of these meals fairly constant from day to day. Karacan et al. (1975) reported that subjects who drank one cup of coffee 30 minutes before retiring at night showed no significant changes in either the quality or quantity of their sleep. It was considered, therefore, that one cup of weak tea or coffee at breakfast or lunch would not affect the EEG recorded several hours later. The aim was to keep the daily routine of the subjects as normal as possible and if tea and coffee were completely banned some subjects may have been subject to stress.

2. The subjects were instructed to arrive at the laboratory approximately 30 minutes prior to the napping sessions.

3. At the commencement of the napping sessions the subjects were simply instructed to relax in the quiet, darkened bedrooms. They were told that the experimenter would enter the bedroom and turn on the light when a sufficient amount of a certain brain activity had been recorded. The idea of falling asleep was not suggested to the subjects.

4. The subjects were told that they would be in the bedroom for no longer than one hour.

5. The subjects were instructed to leave watches and clocks outside the bedrooms both at night and during the day.

2.4 Experimental Procedure

Each night the subjects arrived at the laboratory approximately one hour before their bedtime. The general laboratory procedure was carried out (section 6.3, chapter one), then the subjects prepared themselves for bed. The retiring and rising times were kept to within $\pm$ 15 minutes each night and morning. On awakening the subjects were asked to complete the morning questionnaire (Appendix III).

During the day the same procedure was employed as at night. The subjects retired to the quiet, darkened bedrooms at the scheduled nap time. At the end of the napping sessions, the subjects completed the daytime questionnaire (Appendix IV) and were then allowed to read until they were told to leave the bedroom, so that they would not disturb the other subjects.

2.5 Data Analysis

The EEG daytime sleep records were scored by the experimenter for the time of sleep onset and were checked by another experienced scorer. This scoring was done at the time of recording, because the subjects had to be awoken after five minutes of sleep. Sleep onset was defined as the time from "lights out" until the first appearance of a stage 2 sleep period, which was of at least five minutes unbroken duration. The results are presented in Tables 28 and 29 (section 11.0, chapter two).

The original aim was to compare the group mean of the individual average sleep onset times for both the morning and afternoon nap sessions with the group mean of the individual average sleep onset times over the four baseline nights. However, for unforseen reasons, which will be discussed later, it appeared that this would not be the most suitable method of analysis. Two comparisons were therefore made:

1. the group mean sleep onset time for the first night-time baseline was compared with the group mean of the individual average sleep onset times for both the morning and afternoon nap sessions;
2. the group mean of the individual average sleep onset times for the first and second baseline nights was compared with the group mean of the individual average sleep onset times for both the morning and afternoon nap sessions.

A related, two-tailed t-test was used for this comparison.

Pearson's r (correlation coefficient) was computed between the group mean objective and subjective sleep onset times of the individual average scores for each nap condition and the four night-time sleep onset times. This calculation was performed on both the raw and logarithmic transformed data, because sleep onset time scores are not

normally distributed (section 10.3, chapter one). The computation was performed according to the method of Meddis (1975, chapter eight).

With regard to the quality of the night-time sleep, the first 420 minutes of sleep time from sleep onset were analysed according to the standardised criteria of Rechtschaffen and Kales (1968). The records were scored blind and independently by two, experienced scorers; if the disagreement between them exceeded five per cent of the epochs, it was resolved by discussion. The five sleep stages (W+1), 2, 3, 4 and REM were all expressed in minutes of the total sleep time of 420 minutes. This length of time was chosen as it was the lowest common measure of total sleep time, for all subjects on all nights. The results are presented in Tables 30 - 36 (section 11.0, chapter two). Each sleep stage, over the four nights, was subjected to a two-way ANOVA for repeated measures to determine if there were any significant differences for the sleep stages over the four nights (McNemar, 1969, page 338). The ANOVA summary tables are presented in Appendix X. P values equal to or less than .05 were considered to be statistically significant.

3.0 RESULTS

The individual average sleep onset times of the morning nap condition, with a group arithmetic mean of 45 minutes and a geometric mean of 41 minutes, were significantly greater than the group mean sleep onset time for the first baseline night (arithmetic mean 26 minutes and geometric mean 23 minutes), ($t = 3.2$, df 7, $P \leq .02$, two-tailed test). If the group mean of the individual average sleep onset times for the first and second baseline night (arithmetic mean 28 minutes and geometric mean 24 minutes) had been used for this comparison, then the

increased sleep onset time during the morning was still significant ($t = 2.7$, df 7, $P \leq .05$, two-tailed test). By the same procedure no significant difference was found between the group mean sleep onset time on the first baseline night (arithmetic mean 26 minutes) and the group mean of the individual average sleep onset times for the afternoon nap, which had an arithmetic mean of 29 minutes and a geometric mean of 23 minutes. The group mean of the individual average sleep onset times during the morning was also significantly greater than the group mean of the individual average afternoon sleep onset times ($t = 4.8$, df 7, $P \leq .01$, two-tailed test). There were no significant differences between the group means for the first morning nap and second morning nap when compared by the related t-test; the same was true for the first and second afternoon nap group means.

On inspection of the results table 28 (section 11.0, chapter two), it was apparent that the group mean night-time sleep onset times were becoming progressively longer from baseline 1 to 4. The first baseline night had a group arithmetic mean of 26 minutes and a geometric mean of 23 minutes which was significantly lower than the fourth baseline night, with a group arithmetic mean of 42 minutes and a geometric mean of 36 minutes, using related t-tests ($t = 2.5$, df 7, $P \leq .05$, two-tailed test). The standard deviation of the sleep onset time group means also increased after the first baseline night.

The sleep onset time results (Tables 61, 72, 91, section 25.0, chapter three and Table 8, section 14.0, chapter one) from other studies with good sleepers in this research programme did not show such a progressive rise in sleep onset time over the laboratory nights. In general, the group mean sleep onset times showed only small, random

fluctuations over the recording nights. However, in the case of the placebo control group (Table 72, section 25.0, chapter three) there was a large increase in the group mean sleep onset time on the fourth laboratory night; this was the night when the placebo pill was first administered. After this one night, the group mean sleep onset times were approximately equal to the baseline value. Possible reasons for this sudden, transient increase will be discussed in section 11.0, chapter three. Caution must be applied when comparing these various sleep onset time results, as they may be related to the different conditions which each group underwent. Nevertheless, the overall trend of a progressively increasing group mean night-time sleep onset time was not shown by any other group studied; this indicated that the trend may be related to the daytime naps. However, this idea is only speculative at present.

Since the naps appeared to be influencing the night-time sleep onset times, it was considered that the first baseline night, which was not preceded by a nap, was the only night which could have escaped any possible interference from the naps. Therefore, the first baseline night is the only night which one can confidently say provided an indication of the subjects' normal sleep onset time. However, due to night-to-night variabiliy of sleep onset times (Agnew & Webb, 1971), it is usual to base results on the individual averages of several sleep onset times. The first and second baseline nights do have similar group mean sleep onset times, of 26 and 30 minutes respectively; the distinction appeared on the third and fourth baseline nights. If the group mean of the individual average sleep onset times of the first and second baseline nights had been used for the comparison with the nap conditions, it was unlikely that they would have altered the

results. This was borne out by the results previously reported.

There was a significant correlation between the individual averages of the four objective sleep onset times, measured by means of EEG criteria and the subjective estimates for the night-time sleep. For the raw scores ($r = 0.83$, $N = 8$, $P \leq .01$, one-tailed test) and for the logarithmic transformed scores ($r = 0.86$, $N = 8$, $P \leq .01$, one-tailed test). A one-tailed test was used in this case, because it was hypothesised that the good sleepers of the present study would be good estimators of their sleep onset times, on the basis of the findings of Johns (1977). However, for the nap sessions there were no significant correlations between the objective and subjective sleep onset times:

Morning nap, raw scores, $r = 0.41$, $N = 8$, n.s.

Morning nap, transformed scores, $r = 0.44$, $N = 8$, n.s.

Afternoon nap, raw scores, $r = 0.06$, $N = 8$, n.s.

Afternoon nap, transformed scores, $r = 0.14$, $N = 8$, n.s.

For the purpose of comparison with Johns' (1977) findings, graph C (section 11.0, chapter two) was drawn, on a logarithmic scale, of the individual average estimated (subjective) sleep onset times against the individual average objective sleep onset times over the four baseline nights. The regression line was also calculated.

The ANOVAs (Appendix X) showed that there were no significant differences over the four nights for any sleep stage.

4.0 DISCUSSION OF RESULTS

Since the group mean of the individual average sleep onset times for the morning naps was significantly greater than:

1. the group mean sleep onset time on the first baseline night, and
2. the mean of the individual average sleep onset times on the first and second baseline nights;

this approach to creating an EEG model of sleep onset insomnia appeared to be successful. The group mean sleep onset time for the morning naps was 45 minutes, which was in fact greater than the group mean sleep onset time of 34 minutes for the selected group of sleep onset insomniacs of chapter one. Furthermore, on the first morning nap session, six out of eight subjects had a sleep onset time greater than 30 minutes, whereas, on the first recording night for the self-reported insomniacs only three subjects out of thirteen had a sleep onset time greater than 30 minutes. The present approach, therefore provided a more economical method for obtaining long sleep latencies in terms of subject numbers.

As hypothesised, on the basis of the findings of Agnew and Webb (1971), the subjects, in general, fell asleep significantly faster during the afternoon nap sessions (group mean sleep onset time 29 minutes) in comparison with the morning nap (group mean sleep onset time 45 minutes). The afternoon nap, therefore, did not effectively represent an EEG model of sleep onset insomnia. There was no significant difference between the group mean sleep onset time for the first and second occasions of a particular nap session. This indicated that the subjects had not grown accustomed to napping over this short period. This is in fact a requirement of the model; if the subjects did quickly grow accustomed to napping this would limit the usage of the model.

The correlation between the objective and estimated sleep onset times, at night, for this group of good sleepers was high. Johns (1977) reported an r value of .69, using the transformed scores; in the present study this r value was .86. However, Johns (1977) used 28 subjects, whereas, in the present study only eight were used. From the regression line (Graph C, section 11.0, chapter two) it can be seen that up to an objective sleep onset time of approximately 30 minutes there was, in general, good agreement between the objective and estimated sleep onset times, with a tendency for some of the shorter sleep onset times to be underestimated. However, for the objective sleep onset times greater than about 30 minutes, the subjective estimates tended to be an overestimate. For example, on the basis of the regression line an actual sleep onset time of 37 minutes corresponded to a subjective estimate of 45 minutes. Similar trends were reported by Johns (1977).

When computing the daytime correlations between the objective and subjective sleep onset times, it was difficult to decide whether to include all the measures. In the case of the morning nap sessions only 44 per cent of the subject sessions resulted in sleep, whereas, in the case of the afternoon nap sessions 75 per cent of the subject sessions resulted in sleep; these percentages were higher than expected, considering that this group were not accustomed to napping at any time during the day. It was obviously easier to estimate the time if one did not fall asleep during the hour nap period than if one did. The subjects were also aided in their estimates by being told that they would be in the bedroom for no longer than one hour.

It was eventually decided to compute the daytime correlation coefficients using all the nap scores, but to take note of these points

when interpreting the data. Although no statistically significant correlations between the objective and subjective measures of sleep onset times were found for either nap condition, the morning nap session did have a higher correlation coefficient than the afternoon nap. This may have been due to a reason already mentioned, that is, it was easier to estimate time if one did not fall asleep during the nap session than to estimate the actual sleep onset time. This idea was not supported by subjects 2 and 8 (Table 29, section 11.0, chapter two), who both reported falling asleep in 15 minutes during their first morning session when, according to the EEG measure, they did not fall asleep at all during that session. When questionned about this at the end of the study both subjects reported that on that particular occasion they had lost all sense of time. It was unlikely that either daytime nap correlations between the actual and estimated sleep onset times would have been significant, because the subjects were habitually non-nappers and the experience of falling asleep during the day would have been novel.

During the afternoon naps the subjects tended to overestimate their sleep onset times. The group mean objective time was 29 minutes, whilst the group mean estimated sleep onset time was 42 minutes (Table 29, section 11.0, chapter two). It was found from the daytime questionnaire responses that during the afternoon nap sessions, on nine out of the 16 subject sessions, the subjects felt as though they had not fallen asleep, but on only three subject sessions were these claims supported by the EEG criteria for sleep onset. Similarily, during the morning nap periods on ten out of the 16 subject sessions, the subjects felt as though they had not fallen asleep; in this case, on seven subject sessions these claims were actually supported by the EEG measure.

One reason for this marked discrepancy between the objective and subjective sleep onset measures, for the afternoon nap sessions, may have been that the questionnaire responses were susceptible to expectancy effects. Since this group were non-nappers, they did not, in general, expect to fall asleep at anytime during the day. This expectation tended to hold more during the morning nap sessions when 44 per cent of the subject sessions resulted in sleep, than for the afternoon sessions when 75 per cent of the subject sessions resulted in sleep, due maybe to the increased length of prior wakefulness (Agnew & Webb, 1971).

On inspection of the results Table 29 (section 11.0, chapter two), it can be seen clearly that there was a wide variation in individual sleep onset times: in the morning, the range was from 16 to 60 minutes and in the afternoon, from 8 to 60 minutes. Eight minutes was a very short sleep onset time, in view of the fact that these subjects were habitually non-nappers; this was also the first napping session for this particular subject. One possible way of reducing the range in sleep latency for the morning nap condition may be also to employ the Morning Type - Evening Type questionnaire (Horne & Östberg, 1976) in the subject selection procedure. It might be expected that "morning types" who are at their peak early in the day would find it very difficult to fall asleep during the morning nap session. This idea did not come to mind until after the completion of the subject selection procedure for the subsequent study; so it has not yet been applied in an EEG model of sleep onset insomnia study.

There was only one subject who did not fall asleep during any of the four nap sessions. Excluding all the subject sessions which did not result in sleep, the longest sleep onset time was 42 minutes; in only four out of 19 subject sessions was the sleep onset time greater than 30 minutes. In conversation with the subjects after the study, the general feeling seemed to be that if they were not asleep within about 30 minutes, they felt bored, restless and wanted to get up. This appeared to simulate the feeling experienced by sleep onset insomniacs, when they simply abandon all hope of being able to fall asleep. However, caution must be applied when comparing the various psychological aspects of the EEG model with those of actual sleep onset insomnia. Firstly, there was no pressure upon the model subjects to fall asleep: they had the reassurance that they would be able to sleep that night, whereas, the insomniac feels almost compelled to try and fall asleep, otherwise he will feel tired the next day. This point was illustrated by the questionnaire responses, in that, on only two of 13 subject sessions which did not result in sleep, did the subjects report that they were worried or anxious about not having fallen asleep during the nap period. This aspect may be simulated to some extent by altering the subjects' response set: If they had been instructed that it was necessary for the success of the study that they fell asleep during the nap periods, then this may have caused feelings of anxiety if they had been unable to fall asleep. Secondly, the time of day is also an important difference between the model and actual insomnia; being unable to sleep at night must be a very different experience from that of lying in a laboratory bedroom, knowing that within one hour the study would be finished.

Also of interest is the point, commented upon in the results section, that the naps did appear to be lengthening the group mean night-time sleep onset times, with the fourth baseline night showing a significant increase in comparison with the first. However, this group trend of a progressively increasing sleep onset time over the four baseline nights did not necessarily hold for each individual subject. The naps did appear to be generally disrupting the night-time sleep onset times. On the fourth baseline night the standard deviation for the group mean sleep onset time had a value of 24.2 minutes, which was rather high in comparison with the first baseline score of 14.2 minutes. This may have been partly due to the sheer repetition of having to go through all the laboratory procedure twice on one day. There did not appear to be any obvious relationship between whether the subjects actually fell asleep during the nap period and the lengthening of the night-time sleep onset times. Nevertheless, it was an interesting finding in that it suggested another possible method of creating an EEG model of sleep onset insomnia.

Focusing now upon some of the questionnaire responses, the subjects' responses to question 8 on the morning questionnaire (Appendix III), "When you awoke this morning, how did you feel?", were 14 tired, 13 average and 5 rested. By the time the subjects returned to the laboratory for each morning session, on 10 out of 16 subject sessions they reported feeling alert, 3 normal, 2 tired and one anxious. Therefore, the time which had elapsed from awakening until the subjects returned to the laboratory had been sufficient to arouse and refresh the majority of them. Prior to the afternoon nap sessions a slightly different pattern of responses could be seen: on 6 out of 16 subject sessions they reported feeling alert, 2 normal, 6 tired, one relaxed and one anxious.

The trend, therefore, was for fewer of the subjects to feel alert and more to feel tired prior to the afternoon nap, in comparsion with the questionnaire responses before the morning naps. This trend was borne out by the objective findings, as the group mean sleep onset time for the afternoon naps was found to be significantly shorter than the group mean sleep onset time for the morning naps. After all four naps the general comments were that the subjects felt dozy, lethargic and tired. On only four out of the total of 32 subject sessions did the subjects report feeling alert after a nap period. These latter four responses were associated with nap periods that had not resulted in sleep; three in the morning and one in the afternoon.

When subjects were asked to estimate for how long they had been asleep, the group mean morning estimate was 21 minutes and the group mean afternoon estimate was 23 minutes. Both responses are very similar, yet in both cases the total sleep time allowed was only five minutes. The subjects did, therefore, tend to overestimate the length of time they were asleep. No statistical test was performed on this data, because of the small number of subjects under consideration, especially in the case of the morning naps, where only 44 per cent of the subject sessions resulted in sleep.

Unfortunately, question 8 on the daytime questionnaire (Appendix IV), which asked for an estimate of how long the subjects had been in the bedrooms, turned out to be ambiguous. Some subjects reported the time from the commencement of the nap period until the light was switched on, which was the response that had been originally intended. Others reported the time from the commencement of the nap session until they actually left the bedroom. The daytime questionnaire responses also revealed

that the subjects, as instructed, had attempted to keep the content of their meals fairly constant. All their breakfasts consisted of cereal, toast and a weak tea or coffee, (the majority of subjects had their breakfasts in the laboratory). At lunchtime the subjects had a light meal, usually a toasted sandwich or soup, biscuits or fruit and a weak tea or coffee.

A further interesting finding was that the nap sessions did not significantly change the stages of night-time sleep. Another point worthy of note is that for subject 3 the sleep onset for the morning nap was characterised by three minutes of stage 2 sleep followed by two minutes of stage REM. This sleep patterning is not unusual for this time of day: Webb, Agnew and Sternthal (1966) found that sleep during the early morning naps contained a high percentage of stage REM and stage 2 sleep, whilst stages 3 and 4 were virtually absent. They also reported that the latency to stage REM was only 6.8 minutes for a group of habituated subjects. The end of the night and the morning are known to be the most propitious hours for REM sleep. Although this pattern did not completely satisfy the sleep onset criteria, it was taken as indicating that the subject was asleep.

5.0 CONCLUSIONS AND FURTHER RESEARCH

The morning nap session provided a reasonable model of sleep onset insomnia, according to EEG criteria. This study opened up further possible lines of inquiry.

1. Of further interest was the observation that the napping sessions did appear to be lengthening the group mean night-time sleep onset time. This is another possible method of creating an EEG model of sleep onset insomnia. An area of further investigation would

be to conduct a similar study using only morning nap sessions, but to record the sleep onset time of both the nap and night-time sleep over a longer period of time. Such a study would have two main aims: firstly, to confirm whether an EEG model of sleep onset insomnia, at night, could be created by this method; secondly, to find out, on average, how many nap sessions a subject can undergo before he becomes accustomed to napping.

2. A further variation of the study proposed above would be to alter the instructions given to the subjects; for example, the subjects could be made to feel that they had to fall asleep. This would determine whether by instructing the subjects to fall asleep and stressing its importance, they would be subjected to concern about falling asleep, and hence extend the nap sleep onset time. Alternatively, the reverse could happen in some cases and a shorter sleep onset time result.

   The subjects could be allowed to remain ignorant of the fact that the nap session would terminate after 60 minutes, thereby removing a time cue which may be affecting the subjects' sleep onset times. However, this proposal was not feasible in the present study, because the subjects needed to know for how long they would be in the laboratory in order to arrange their daily commitments around the laboratory times.

3. Another aspect of further research would be to test the model with a variety of drug and non-drug "sleeping aids", some with known sedative properties, in order to determine the sensitivity and reliability of the model created in the present study. A set of

standards could then be drawn up against which the sedative properties of other drugs could be compared. It may also be beneficial, when using the EEG model of sleep onset insomnia to evaluate a possible treatment for insomnia, to increase the duration of the actual nap; only by a small amount, however, to avoid the risk of interfering with the quality of the night-time sleep. For example, the nap could be extended from five to ten minutes: thus, not only the sleep onset time could be measured, but some indication as to whether the nap could be sustained would also be gained.

It was this last research proposal which was pursued further, (chapter two, part II).

Chapter Two - Part II

AN APPLICATION OF THE EEG MODEL OF SLEEP ONSET INSOMNIA

6.0 INTRODUCTION

The first two research proposals, which were put forward at the end of chapter two, part I, would have provided an interesting line of investigation, but they were considered to be a deviation from the main path of the research programme. The Aspirin aspect of insomnia was still to be investigated and the time was limited. It was the third of these research proposals which was studied in this part of chapter two, mainly because an approach had been made by Fisons Pharmaceuticals Limited. Admittedly, the logical step, after having created this EEG model of sleep onset insomnia, would have been to use it for the evaluation of the hypnotic potential of the non-drug "sleeping aids" which were used in chapter one. The reason for this study would have been to determine whether the findings obtained with the model supported those previously obtained with the group of self-reported insomniacs. However, the Niagara Therapy Company, the manufacturers of the cycloid vibrating cushions, were unwilling to provide funding for such a study. Nevertheless, as previously mentioned, Fisons Pharmaceuticals Limited were interested in the model and were willing to provide both support and funding for an investigation to be conducted with the model and one of their compounds, which will be referred to as F.P. throughout the text. This offer provided an ideal opportunity for using the model that could not be missed. This Company were primarily interested in the potential of the model for testing the efficacy of various compounds which were believed to have sedative properties.

The Company believed that F.P. may have mild sedative properties. A known drug, with similar sedative properties, had to be chosen, for means of comparison with the unknown effect of F.P. After personal

communication with Max Fink, the drug eventually chosen was Temazepam, a member of the benzodiazepine group of compounds. Temazepam is listed as a tranquiliser (Martindale, 1977). The two main reasons for choosing this drug were:

1. it is relatively fast acting and has a biological half-life of 5.5 - 8.5 hours (Fuccella, Tosolini, Moro & Tamassia, 1972), similar to that of F.P.,
2. it was believed to have mild sedative properties.

Fuccella et al. (1972) reported that their subjects felt drowsy after a dose of Temazepam (30 mg.) and fell asleep for about one hour. However, only four subjects were employed in their study and the time of drug administration and whether the study was conducted under double or single blind conditions was not made clear in their report.

Temazepam is currently under investigation as a potential hypnotic. Two of the self-report studies conducted by Middleton (1978) and Fowler (1977) found that Temazepam (10 mg.) was reported as performing satisfactorily as an hypnotic. The former used hospitalised geriatric patients, and the latter used patients who were selected from the general practitioner's registers. However, there is some doubt as to the effectiveness of Temazepam as an hypnotic when evaluated by means of EEG criteria. In a study with six insomniac subjects, Bixler, Kales, Soldatos, Scharf and Kales (1978) claimed that Temazepam (30 mg.) was found to have no sleep inducing properties and was not effective at maintaining sleep, but the number of nightly awakenings were significantly reduced. Mitler, Phillips, Billiard, Spiegel, Zarcone and Dement (1975) found that Temazepam (30 mg.) produced a significant decrease in the amount of time spent awake after sleep onset and an increase in the total sleep time of a group of insomniacs. Again it was found that Temazepam had no

sleep inducing properties. However, in an earlier study, Maggini, Murri & Sacchetti (1969) reported that Temazepam produced an increase in the total sleep time and a decrease in the sleep onset time and duration of wake periods in a group of insomniacs. These authors concluded that Temazepam was effective in the treatment of both initial (sleep onset) and middle insomnia. Some possible reasons for these different findings are listed below.

1. The group mean sleep onset time for the Bixler et al. (1978) insomniac group was 27.8 minutes and for the Mitler et al. (1975) group only 20.7 minutes, therefore, it is unlikely that any hypnotic/sedative would be able to reduce these sleep onset times by a large amount. However, in the Maggini et al. (1968) study the group mean sleep onset time was 49.5 minutes; there was, therefore, more room for improvement in this study.

2. The Bixler et al. (1978) and the Mitler et al. (1975) insomniac groups were almost matched for the amount of wakefulness during the night, with a mean value of 38.6 minutes for the former and 40.4 minutes for the latter, but for the Maggini et al. (1968) insomniac group this parameter had a large group mean value equal to 93 minutes.

3. Bixler et al. (1978) kept the total time that the subjects' spent in bed constant from night-to-night, whereas, in the other two studies it was allowed to vary. The group mean baseline total sleep time for the Maggini et al. (1968) insomniac group was very short (238 minutes).

4. The subjects in these three studies were not matched.

Maggini et al. (1968) used eight male in-patients from a psychiatric clinic; five were suffering from neurosis and three from endogenous depression. Bixler et al. (1978) used six insomniacs from the sleep centre clinic and Mitler et al. (1975) claimed to have selected seven idiopathic insomniacs.

5. Maggini et al. (1968) used 20 second epochs when scoring the sleep records, whilst the other investigators used one minute epochs. However, in the results tables Maggini et al. (1968) expressed the stages of sleep in minutes.

6. The drug dosage employed is also a further important factor. Bixler et al. (1978) and Mitler et al. (1975) both used 30 mg. Temazepam. Maggini et al. (1968) simply stated that by the fourth night the total dosage administered was 300 mg. Temazepam; by the eighth night, 680 mg. Temazepam had been administered. It is therefore difficult to infer from these figures the actual dosage they employed, but it would appear to be higher than that used in the other two studies.

7. Both the Bixler et al. (1978) and Mitler et al. (1975) studies were relatively long term, lasting for 49 and 54 nights respectively, in comparison with the Maggini et al. (1968) study which continued for eight nights.

8. Both the Bixler et al. (1978) and the Mitler et al. (1975) studies incorporated a placebo condition into their experimental designs, whereas, Maggini et al. (1968) used the placebo days to screen for placebo responders prior to measuring the baseline. The

Maggini et al. (1968) results were therefore not compared with the placebo condition and the study was single-blind.

It would appear, from the reasons listed above, that Temazepam has not been satisfactorily evaluated as a sleep inducing agent. Although Maggini et al. (1968) claimed that Temazepam was effective in inducing sleep, this study has many shortcomings, as mentioned above, and no firm conclusions can be drawn from their findings. Nevertheless, it was hypothesised that Temazepam would reveal sedative properties when tested with the model, because there was prior indication that it had sedative propeties (Fuccella et al. 1972) and it is likely to have properties similar to other members of the benzodiazepine group, which are known to bring about sedation. F.P. was expected to reveal sedative properties similar to those of Temazepam when tested with the model, on the basis of the claims of Fisons Pharmaceuticals Limited that F.P. may have mild sedative properties.

7.0 METHOD

7.1 Experimental Design

It was necessary to change the original model slightly, for with drug studies it is important to examine the effects of the drug over time. This necessitates performing repeated measures on the "sedating potential" of the drugs during each nap session. It was commented in the previous discussion (section 4.0, chapter two), that after approximately 30 minutes in the bedrooms during the day, the subjects became bored and restless. For this drug study, it was therefore decided to reduce the length of the nap periods to 30 minutes each and to have four of these nap periods, during which EEG recordings were made. There was a 30 minute break between each EEG recording period

during which time the subjects were allowed to read some light material. The experimental protocol was:

Time	
0900	Dosed; electrodes applied
0930	Start of Period 1: 30 minutes of EEG recording
1000	30 minutes of reading
1030	Start of Period 2: 30 minutes of EEG recording
1100	30 minutes of reading
1130	Start of Period 3: 30 minutes of EEG recording
1200	30 minutes of reading
1230	Start of Period 4: 30 minutes of EEG recording
1300	End

The first nap period commenced at 9.30 a.m., which was approximately 2½ hours after the subjects had arisen from their night's sleep.

Another deviation from the original model was that due to various domestic commitments, the subjects were unable to sleep in the laboratory on the nights associated with the nap periods. However, in an attempt to compensate partly for this, the subjects were asked to complete the morning questionnaire (Appendix III), upon awakening each morning at home: the questionnaire responses would give some indication as to whether the subjects were sleep satiated prior to the nap periods. The subjects were also asked to complete this questionnaire on the mornings after they had been in the laboratory. As mentioned in the research proposal (section 5.0, chapter two), the subjects were to be allowed ten minutes sleep during any nap period, before being awoken by the experimenter. They were then allowed to return to sleep and the procedure repeated if they fell asleep again for more than ten minutes. The reason for this was to investigate whether the naps could be sustained

and to give some indication of the "pressure" forcing the subject to return to sleep after he had been awoken.

Each subject underwent all three conditions, namely, Temazepam, F.P. and placebo, on separate occasions, one week apart. The reasons for the laboratory sessions being one week apart was to allow a wash out period for the drug and to also prevent the subjects from acquiring the napping habit. The treatments were administered in a randomised order according to a replicated, latin square design. The treatments were all administered under double blind conditions. The compounds were presented in rice paper cachets, because the form of Temazepam used was like a syrup. However, it was impossible to distinguish between the three treatments. The dosage of Temazepam employed was 20 mg.; this was the mid-point of the recommended dosage, (10 - 30 mg.). The pattern of drug administration was:

Week	1st.	2nd.	3rd.
Subject			
1	B	A	C
2	C	B	A
3	A	C	B
4	B	A	C
5	C	B	A
6	A	C	B

Key:

Treatment A = F.P. (50 mg.)

Treatment B = Placebo

Treatment C = Temazepam (20 mg.)

The same pair of subjects attended the laboratory on the same day each week.

7.2 Subject Selection

The six subjects were selected from the Fisons Pharmaceuticals' volunteer panel, with the aid of the sleep questionnaire (Appendix II), according to the procedure of section 6.1, chapter one. The subjects had to satisfy the general requirements for subjects selected for a sleep study (section 6.1, chapter one) and they also had to be unaccustomed to daytime napping. The subjects, (average age 31.5 years) were all healthy males and self-reported good sleepers.

7.3 Instructions to Subjects

The subjects were given the "general instructions to subjects", section 6.2, chapter one. They were also instructed to have a light breakfast plus one weak tea or coffee and not to have any additional snacks or drinks between then and arriving at the laboratory. They were asked to keep the content of their breakfasts constant during the course of the study. For ethical reasons, the whole procedure was carefully explained to all the subjects and they knew which drugs were to be employed, but they did not know the order of presentation. As for the initial model study, the subjects were simply instructed to relax during the EEG recording periods and no suggestion of falling asleep was made.

7.4 Experimental Procedure

The subjects were studied in pairs. Each pair arrived at the laboratory between 8.45 a.m. - 9.00 a.m. The subjects were given the rice paper cachet, which contained the drug or placebo, at 9.00 a.m. The electrodes were then applied, according to the general laboratory procedure (section 6.3, chapter one). Each subject retired to his respective bedroom at 9.25 a.m., where he remained for $3\frac{1}{2}$ hours.

Any subject who fell asleep during the EEG recording periods was allowed ten minutes of unbroken sleep from the time of sleep onset and then, if still asleep, he was gently aroused by the experimenter until clearly awake. The experimenter then withdrew from the bedroom. If sleep returned, then the ten minute time restraint was again applied. If the subject awoke for more than one minute during any ten minute sleep period it, in effect, eliminated the preceding minutes of sleep from a scoring point of view. The ten minute time restraint was again applied from the time of the subjects' next sleep onset. The sleep onset criteria* were the same as for the model study in part I.

Subjects were not allowed to fall asleep during the reading periods. The experimenter periodically observed the subjects to ensure that they were still reading. If the subjects appeared as though they might have been about to fall asleep, then the experimenter chatted to them. In a few cases it was extremely difficult to keep the subjects awake. As these subjects were accustomed to having a morning tea break, they were given a drink of diluted orange squash and a plain biscuit at 11.00 a.m.

At the request of Fisons Pharmaceuticals Limited a check was made, by means of self-report data, on the general health and well-being of the subjects, at the end of the morning session and on the subsequent morning.

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* The time to sleep onset was defined as the time from "lights out" until the first appearance of a stage 2 period, which was of at least five minutes unbroken duration.

7.5 Data Analysis

The EEG records for each recording period were scored in one minute epochs and classified under the following headings: stage drowsy + 1 (D+1), 2, 3, 4 and REM. This was done according to the standardised criteria described by Rechtschaffen and Kales (1968). The EEG records were assessed blind and independently by two, experienced scorers. If the disagreement between the scorers exceeded five per cent of the epochs, it was resolved by discussion. Total sleep time, sleep onset time and the number of sleep onsets were also determined. Initially, the term "number of sleep onsets" was intended to refer to the number of times, during one recording period, that the subject, having been asleep for ten minutes and then awoken, returned to sleep again. However, considering the definition of sleep onset employed in this research programme, any amount of time greater than five minutes spent asleep should be classed as a sleep onset. For example, if a subject awoke after seven minutes of stage 2 sleep, it should still be classed as a sleep onset. This explanation accounts for subject 5 (Table 43, section 11.0, chapter two) having four sleep onsets during the first recording period of the placebo condition. Any further research with this model will have to make provision in the data analysis for a distinction to be made between these two types of sleep onset in order to aid the interpretation of the results.

The data were tabulated over all the four periods (Table 37, section 11.0, chapter two) and for periods 1 - 4 individually (Tables 38 - 41, chapter two). The individual subject data are presented in Tables 42 - 50 (section 11.0, chapter two). A three-way ANOVA, with repeated measures on two factors, was computed for the sleep stages (D+1), 2 and for the sleep onset time, total sleep time

and the number of sleep onsets (Winer, 1971, page 539). This analysis was not performed on stages 3, 4, (3 + 4) and REM, as it can be seen from Tables 47 - 50, (section 11.0, chapter two), that many of the scores for these parameters were equal to zero. The ANOVA would, therefore, have little meaning. A three-way ANOVA enables one to examine the drug and period effects simultaneously. If the ANOVA gave a significant F value, it was followed by a Newman-Keuls multiple comparison test (Winer, 1971, page 191), in order to determine which components of the factor contributed most to the significant F value. Summary ANOVA and Newman-Keuls tables are presented in Tables 42A - 46A (section 11.0, chapter two). Values of F which yieled a P value equal to or less than .05 were considered to be statistically significant.

8.0 RESULTS

A significant drug ($F = 7.1$, df 2,10, $P \leq .025$) and period ($F = 3.9$, df 3,15, $P \leq .05$) effect were found for sleep onset time (Table 42, section 11.0, chapter two). The Newman-Keuls multiple comparison test showed that Temazepam produced a significantly shorter group mean sleep onset time than both placebo and F.P., ($P \leq .05$). F.P. was not significantly different from placebo for this measure. The significant period effect indicated that regardless of the treatment condition there were some other factors operating. The multiple comparison test revealed that the second period produced a significantly shorter group mean sleep onset time than the first, third and fourth ($P \leq .05$), and that there were no significant differences between the other periods.

For total sleep time there was both a significant drug ($F = 15.3$, df 2,10, $P \leq .001$) and period ($F = 3.4$, df 3,15, $P \leq .05$) effect (Table 44,

section 11.0, chapter two). From the multiple comparison test, it was apparent that it was Temazepam which produced this significant result, because Temazepam produced a significantly longer group mean total sleep time than both F.P. and placebo ($P \leq .01$); there was no significant difference between F.P. and the placebo for this measure. With regard to the period effect, it was the second period which produced a significantly longer group mean total sleep time, regardless of the treatment condition, in comparison with the first and fourth periods ($P \leq .05$).

There was a significant drug effect for stage 2 ($F = 20.3$, df 2,10, $P < .001$), Table 46 (section 11.0, chapter two). The multiple comparison test showed that Temazepam produced a significantly greater group mean increase in stage 2 than did both placebo and F.P. ($P \leq .01$). However, for this sleep parameter there was also a borderline significant difference between F.P. and placebo ($P \leq .05$). A significant drug effect was also found for the number of sleep onsets ($F = 16.0$, df 2,10, $P \leq .001$), Table 43, (section 11.0, chapter two). Once again, it was found to be Temazepam which produced a greater number of sleep onsets than both F.P. and plcebo ($P \leq .01$).

9.0 DISCUSSION OF RESULTS

From the results section above it is apparent that almost all the significant drug effects were attributable to the action of Temazepam. Temazepam produced a significant increase in: total sleep time, number of sleep onsets and stage 2 sleep when compared with the placebo and the drug F.P. F.P. was only found to be significantly different from placebo for stage 2 sleep. There was a slight tendency for F.P. to increase total sleep time in comparison with the placebo, but this

did not reach statistical significance.

The question now raised is, what do these significant changes in the various sleep parameters, produced by the administration of these treatments, indicate? Since Temazepam produced a significant decrease in the group mean sleep onset time and an increase in the total sleep time, in comparison with the other treatments, it demonstrated that the model was sensitive to the sedative properties of this drug. It was hypothesised that Temazepam would bring about sedation in the model. This hypothesis was based on the findings of Fuccella et al. (1972) and the fact that this drug is a member of the benzodiazepine group of compounds. This sedative potential was also revealed by the finding that with Temazepam on only 6 out of the 24 subject sessions did the subjects fail to fall asleep, whereas, for the placebo and F.P. conditions these values were 15 and 13 respectively. This sedative potential was especially prevalent in the case of subjects 1 and 5 for the second period, when their sleep onset times were zero and one minute respectively (Table 42, section 11.0, chapter two). A significant increase in the total sleep time and the number of sleep onsets (Tables 44 and 43, section 11.0, chapter two) produced by Temazepam showed that even after being awoken after ten minutes sleep, there was a strong tendency for the subjects to return to sleep; much more so than with either F.P. or placebo. In some cases, when Temazepam had been administered, this tendency was so great that the subjects had difficulty in remaining awake during the reading periods.

Temazepam also produced a significant increase in stage 2 sleep, in comparison with the other treatments (Table 46, section 11.0, chapter two). This was to be expected due to the increase in total sleep time.

F.P. also brought about a significant increase in stage 2 sleep, in comparison with the placebo. Such an effect may be linked with the slight tendency of F.P. to increase total sleep time and is indicative of only a very slight sedative potential, particularly as F.P. did not produce a significant decrease in the parameter of major interest in this particular case, namely, sleep onset time.

Another interesting observation (Table 49, section 11.0, chapter two) was that Temazepam produced a marked increase in the amount of stage (3+4) during the second period. This may have been because this second period corresponded to the time of peak activity for Temazepam. The second period commenced at 10.30 a.m., one and a half hours after drug administration. Fuccella et al. (1972) found that 30 mg. Temazepam, when administered in a liquid form to four subjects, reached peak plasma levels approximately 40 - 60 minutes after administration. However, their subjects had been starved since the previous night. Therefore, even though a lower dose of Temazepam was employed in the present study, (20 mg.), because the subjects were not starved the peak plasma level would be expected to be delayed slightly. This effect may also be linked with the finding that the second period, regardless of the treatment condition, appeared to be more conducive to sleep than the others. However, this effect is only speculative, because no statistical analyses were performed on stage (3+4). The group mean amount of stage (3+4), during the second Temazepam period was 4.5 minutes, which on first consideration appeared rather small. However, it must be borne in mind that only ten minute sleep epochs were allowed during the EEG recording periods.

Also worthy of comment is the statistically significant period effect, which was found for sleep onset time and total sleep time. In both cases, it was the second period which produced the significant change, regardless of the treatment. One possible explanation for this effect may be associated with adaptation effects. The subjects might not have been adapted to the laboratory setting by the time of the first recording period, because only approximately 15 minutes prior to the first rest period they had had electrodes applied for the first time, and only five minutes prior had entered the darkened bedroom. Whereas, prior to the second recording period the subjects had been reading and they had been in the bedroom for one hour. In the previous EEG model insomnia study the subjects had slept in the laboratory for three nights prior to the first nap. These subjects were therefore adapted to the laboratory setting. In view of the present findings, the subjects of the present study should have attended the laboratory on four occasions, the first being for adaptation purposes. Alternatively, the first and second periods may have been made equally conducive to sleep by allowing the subjects to read for the first 30 minutes in the bedroom and then to have commenced the first recording period at 10.00 a.m. The disadvantage of this time scale was that any possible effects of the drug, evident after approximately 30 minutes, would be missed; therefore, the dosing would have to take place at 9.30 a.m. However, this alternative has its disadvantages as well, for in order to be able to examine the effects of drugs over four recording periods at 30 minute intervals the fourth period would then commence at 1.00 p.m. This would lead to another confounding effect, namely, that of boredom of the subjects and the feeling that their lunchtime was near. On the time scale used in the present study, there was an indication that even by 12.30 p.m. the subjects were anxious to be returning home for their lunch and were

bored with the experimental procedure.

This problem of boredom is very difficult to overcome in a repeated measures study such as this. One possible way to overcome this effect may be simply to record for three periods; but since the third period would then be the last, the same boredom factor may then apply to this period as it did to the fourth. However, the laboratory session would then finish earlier at 12.00 p.m. One disadvantage with recording for only three periods is that some of the effects of the drug may be missed. Another alternative may be to reduce the length of the reading periods between each recording period.

Another factor contributing to the significant period effect may have been that, if a subject fell asleep in one period, this may have interfered with his chances of falling asleep in a subsequent period. Since it appeared to be the second period which was conducive to sleep, regardless of the treatment, the third and fourth periods were the ones most likely to be affected by this factor. An alternative way to overcome this period effect may be to adopt a different experimental design. For example, a much larger sample of matched subjects, divided into four groups, could be used, each group of subjects representing a particular time period. All groups would be dosed at the same time, and each group would be allowed one hour of relaxation in the bedrooms, commencing at their time period. This design would enable the study of possible longer sleep onset times. However, this approach also introduces variablility due to the different groups of subjects employed.

It was interesting to note that 38 per cent of the subject sessions, for the placebo condition, resulted in sleep. For the previous model study the proportion was 44 per cent. These percentages are similiar, and both are higher than was initially expected. In consideration of the fact that these subjects were all unaccustomed to daytime napping, one would ideally expect these percentages to equal zero. The morning questionnaire responses by the subjects of the present study indicated that the majority of subjects felt "average" upon awakening (Table 51, section 11.0, chapter two). However, prior to all conditions the group mean estimated total sleep time was approximately 390 minutes, which was less than would be expected for this age group. A group mean total sleep time lying within the range 420 - 465 minutes (Roffwarg et al., 1966) would be expected. Only two subjects claimed that they felt as if they had not had enough sleep; subject 6, prior to the Temazepam and F.P. treatments and subject 1, prior to the F.P. treatment. It is possible that not all of the subjects were sleep satiated prior to the morning laboratory sessions. Although the experimental design may have randomised the effects of any prior sleep deprivation, this procedure would not have been entirely effective with this relatively small sample size. Following the experimental days, there was a slight tendency for an increased feeling of tiredness (Table 52, section 11.0, chapter two), but there was no obvious treatment associated trend. There was also indications of a slightly lengthened night-time sleep following Temazepam.

Scrutiny of the EEG traces for unusual or abnormal activity produced only one finding. This was an increase in duration and amplitude of 12-14 Hz. spindle activity in two subjects following the administration of F.P. in one subject and Temazepam in the other.

From Tables 53 and 54 (section 11.0, chapter two), it would appear that with regard to the subjects' general health and well-being, only subject 3 under the F.P. condition experienced any undesirable side-effects, namely, slight stomach discomfort, which did not persist into the next day.

Before attempting to draw any conclusions from the findings reported, it is necessary to be aware of some of the shortcomings of this study:

1. As mentioned previously, there was some indication that the subjects were not sleep satiated prior to each morning session in the laboratory. This could have been better controlled if the subjects' night-time sleep had been recorded in the laboratory.

2. There was a significant period effect, regardless of the treatment condition. It was the second period which was significantly more conducive to sleep than the others. Various suggestions were made to overcome this problem.

3. In order to carry out a repeated measures study of reasonable duration, the nap period was reduced to 30 minutes. Thus, the maximum sleep onset time which could possibly be achieved was 30 minutes. Ideally, a longer period time should have been used, for example, 45 minutes.

With the above points in mind, some conclusions will be drawn from the findings concerning the sedative potential of the two drugs.

10.0 CONCLUSIONS AND FURTHER RESEARCH

Temazepam, as hypothesised, clearly revealed its sedative properties when evaluated with the model, primarily through the significant decrease in sleep onset time and increase in total sleep time brought about by this drug. Temazepam was significantly more effective than both F.P. and the placebo at producing sedation in the model. F.P. showed only one effect which was significantly different from the placebo condition, namely an increase in stage 2 sleep. This effect of F.P. may be linked with the slight tendency for an increased total sleep time upon administration of this drug, but the increase was not statistically significant. F.P. did not bring about a significant reduction in sleep onset time in comparison with the placebo. This measure was of major importance in the present study. The evidence in support of the claim that F.P. may have mild sedative properties was very weak. With only one significant effect, one can only cautiously conclude that F.P. appeared to have very mild sedative properties in comparison with the placebo, but it failed to match the sedative potential shown by Temazepam.

The question now raised is, "Why is there such a marked difference between the two drugs when tested upon the model, when they were initially believed to have similar sedative properties?" At present, there appears to be two alternative, possible answers:

1. Temazepam may possess much stronger sedative powers than was initially believed. As mentioned in the introduction, the studies which have previously investigated this action of Temazepam are themselves open to criticism. There is also the possibility that F.P. has even less sedative potential than the Company believed.

2. The model itself may be "extra-sensitive" at discriminating between the sedative potential of various compounds. Therefore, there may only be slight differences between the sedative potential of these two drugs, but this evaluation procedure may be very sensitive, and as a result amplified this difference.

Although the second alternative is a possible explanation, there is little evidence to support it, since the present study is the first conducted with this model. The sensitivity of the model will have to be determined in a series of further studies with compounds of known sedative potential. Nevertheless, on the basis of the present findings, it would appear that Temazepam (20 mg.) has marked sedative properties.

In order to aid in the interpretation of the results from such model insomnia studies, a table of results must be drawn up of the action of known sedative compounds upon this model; this would then serve as a reference table against which drugs of unknown sedative potential could be compared. This is an important series of further studies which ought to be conducted before the model is again employed in studies with drugs of unknown sedative potential. These proposed studies will also give some indication of the sensitivity of the model.

It would also be interesting to use the model to examine drug interaction and multiple dosing effects. However, as mentioned in the previous discussion (section 9.0, chapter two), it is necessary to try to make all the recording periods equally conducive to sleep. Various suggestions were made, for example, to reduce the length of the reading periods which were interspersed between the recording periods and employing four groups of subjects, each corresponding to a particular time period. Some of these suggestions could be tested experimentally.

Any further research with this model should ideally require the subjects to attend the laboratory at night as well as during the day. If this is not possible, then a morning session should be allowed for adaptation purposes.

Another line of investigation would be to select a group of sleep onset insomniacs, according to EEG criteria, for the purpose of a double-blind assessment of Temazepam at night. The results of the present study indicated that Temazepam has stronger sedative properties than was initially expected: it may, therefore, be a useful "aid" for sleep onset insomniacs. Also, on the basis of the results of the present study one would feel justified in going to the lengths of selecting a group of sleep onset insomniacs to satisfy EEG criteria. The three studies already conducted in this area were discussed in the introduction and it was clear that, to date, Temazepam has not been satisfactorily evaluated by EEG criteria as a sleep inducing agent.

Although the EEG model of sleep onset insomnia study opened many avenues of further inquiry, they were not pursued at present, mainly because of time limitations. The experimenter was still interested in determining the rationale, if any, behind the claim made by many of the self-reported insomniacs that Aspirin helped them to sleep (chapter one). The following chapter will therefore focus upon this aspect of insomnia.

Section 11.0

RESULTS TABLES AND GRAPHS FOR CHAPTER TWO

11.1 Explanatory Notes for the Results Tables

All means in the results tables are expressed to the nearest whole number.

All figures in the results tables are expressed in minutes unless otherwise stated.

Key:

A - Afternoon Nap Session

A.S.T. - Analysed Sleep Time refers to the amount of each sleep record analysed from the time of sleep onset. A.S.T. is the lowest common measure of total sleep time for all subjects on all nights.

Av - Average

B - Baseline

M - Morning Nap Session

X - Sleep onset time as measured by the EEG criteria

Y - Sleep onset time as estimated by the subjects

Results Tables for Sleep Onset Times at Night and during the Day

Sleep Onset Time at Night — Table 28

Night	B_1		B_2		B_3		B_4	
	X	Y	X	Y	X	Y	X	Y
Subject								
1	19	20	17	20	15	15	18	20
2	21	20	32	30	32	25	33	15
3	20	30	9	30	61	60	24	30
4	36	60	15	45	21	30	86	120
5	14	15	18	15	28	20	32	30
6	12	10	27	10	46	30	21	30
7	35	30	81	80	57	60	64	45
8	54	30	37	60	47	45	55	90
Mean	26	27	30	36	38	36	42	48
S.D.	14.2	15.3	22.8	24	16.9	17.4	24.2	37.4
Geometric Mean	23	24	24	30	35	32	36	38

Sleep Onset Time during the Day — Table 29

Nap Session	M_1		M_2		A_1		A_2	
	X	Y	X	Y	X	Y	X	Y
Subject								
1	60	60	60	60	14	10	60	60
2	60	15	34	15	26	60	15	60
3	32	60	60	60	15	60	22	60
4	60	60	60	60	60	60	60	60
5	16	60	30	60	8	60	12	60
6	21	15	37	20	16	15	20	30
7	60	60	60	60	60	20	42	30
8	60	15	16	10	14	15	10	10
Mean	46	43	45	43	27	38	30	46
S.D.	19.6	23.3	17.5	23.4	21.2	24.2	20.9	20
Geometric Mean	41	36	41	35	21	30	24	41

Results Tables for the EEG Model of Sleep Onset

Insomnia Study

Stage W+1. Table 30

Night / Subject	B_1	B_2	B_3	B_4
1	22	9	15	12
2	23	9	19	25
3	12	29	2	27
4	4	10	39	13
5	14	8	16	17
6	5	5	5	3
7	11	15	7	22
8	19	13	18	14
Mean	14	12	15	16
S.D.	7.2	7.4	11.6	7.9

Stage 2 Table 31

Night / Subject	B_1	B_2	B_3	B_4
1	219	219	226	223
2	238	235	218	202
3	223	211	228	203
4	270	232	205	240
5	204	186	184	184
6	198	201	207	184
7	190	206	209	180
8	177	187	172	161
Mean	215	210	206	197
S.D.	29.6	18.5	19.6	25.4

Stage 3 Table 32

Night / Subject	B_1	B_2	B_3	B_4
1	32	35	24	27
2	10	13	13	16
3	32	29	22	29
4	29	29	18	35
5	27	37	29	20
6	19	22	21	22
7	18	22	24	27
8	32	35	33	25
Mean	25	28	23	25
S.D.	8.3	8.3	6.2	5.8

A.S.T. for the above three tables is equal to 420 minutes.

Stage 4 — Table 33

Night / Subject	B_1	B_2	B_3	B_4
1	60	76	70	76
2	75	62	74	73
3	51	62	59	52
4	30	60	54	30
5	65	62	61	76
6	115	101	98	118
7	76	72	73	72
8	89	72	74	98
Mean	70	71	70	74
S.D.	25.4	13.6	13.5	26.6

Stage 3+4 — Table 34

Night / Subject	B_1	B_2	B_3	B_4
1	92	111	94	103
2	85	75	87	89
3	83	91	81	81
4	59	89	72	65
5	92	99	90	96
6	134	123	119	140
7	94	94	97	99
8	121	107	107	123
Mean	95	99	93	100
S.D.	23.2	14.9	14.7	23.4

Stage REM — Table 35

Night / Subject	B_1	B_2	B_3	B_4
1	87	81	85	82
2	74	101	96	104
3	102	89	109	109
4	87	89	104	102
5	110	127	130	123
6	83	91	89	93
7	125	105	107	119
8	103	113	123	122
Mean	96	100	105	107
S.D.	16.6	15.1	15.6	14.6

A.S.T. for the above three tables is equal to 420 minutes.

Total Sleep Time — Table 36

Night / Subject	B_1	B_2	B_3	B_4
1	487	480	420	483
2	486	454	461	467
3	505	504	460	486
4	478	496	499	432
5	513	492	492	490
6	424	446	423	420
7	434	423	429	429
8	456	455	427	450
Mean	473	469	451	457
S.D.	32.1	28.5	31.6	28.1

Means and Standard Deviations of the Total Amounts of the Sleep Stages over all Four Half-Hour Recording Periods, in minutes.

Table 37

	Temazepam	F.P.	Placebo
Sleep Onset Time	18.6 (10.4)	23.5 (8.2)	25.0 (7.6)
Stage D+1	10.7 (6.9)	5.4 (4.5)	6.6 (6.6)
Stage 2	9.7 (6.8)	6.2 (7.1)	3.5 (5.9)
Stage 3	1.0 (1.6)	0.3 (1.0)	0.3 (0.7)
Stage 4	0.8 (2.7)	0 (0)	0 (0)
Stage 3+4	1.8 (3.7)	0.3 (1.2)	0.3 (0.7)
Stage REM	1.1 (3.6)	0.1 (0.6)	0 (0)
Total Sleep Time	12.3 (8.5)	6.7 (7.4)	3.8 (5.9)
Number of Sleep Onsets	1.5 (0.8)	0.9 (1.1)	0.6 (1.0)

The figures enclosed in brackets are the standard deviations

Means and Standard Deviations of the Sleep Stages in the First Half-Hour Recording Period, in minutes.

Table 38

	Temazepam	F.P.	Placebo
Sleep Onset Time	20.0 (11.1)	25.8 (5.9)	26.0 (7.6)
Stage D+1	12.2 (7.4)	6.7 (5.1)	6.8 (6.3)
Stage 2	8.7 (8.6)	5.2 (5.2)	3.5 (6.4)
Stage 3	0.8 (2.0)	0 (0)	0.2 (0.4)
Stage 4	0.5 (1.2)	0 (0)	0 (0)
Stage 3+4	1.3 (3.3)	0 (0)	0.2 (0.4)
Stage REM	0 (0)	0 (0)	0 (0)
Total Sleep Time	10.0 (9.9)	5.2 (5.2)	3.7 (6.5)
Number of Sleep Onsets	1.2 (0.8)	0.7 (0.8)	0.8 (1.6)

The figures enclosed in brackets are the standard deviations

Means and Standard Deviations of the Sleep Stages in the Second Half-Hour Recording Period, in minutes.

Table 39

	Temazepam	F.P.	Placebo
Sleep Onset Time	12.5 (11.5)	19.5 (9.6)	21.0 (10.8)
Stage D+1	10.2 (7.0)	6.3 (2.0)	6.8 (6.9)
Stage 2	11.3 (6.1)	9.5 (7.8)	6.2 (8.8)
Stage 3	2.0 (1.7)	0.8 (2.0)	0.3 (0.8)
Stage 4	2.5 (5.2)	0.2 (0.4)	0 (0)
Stage 3+4	4.5 (6.0)	1.0 (2.4)	0.3 (0.8)
Stage REM	1.3 (3.3)	0.5 (1.2)	0 (0)
Total Sleep Time	15.8 (8.1)	11.0 (7.4)	6.5 (8.7)
Number of Sleep Onsets	1.8 (0.8)	1.7 (1.2)	0.8 (1.0)

The figures enclosed in brackets are the standard deviations

Means and Standard Deviations of the Sleep Stages in the Third Half-Hour Recording Period, in minutes.

Table 40

	Temazepam	F.P.	Placebo
Sleep Onset Time	18.3 (11.1)	21.7 (9.3)	29.0 (2.4)
Stage D+1	9.7 (9.8)	5.5 (6.2)	4.8 (7.5)
Stage 2	9.0 (8.2)	7.0 (8.2)	1.3 (2.2)
Stage 3	0.7 (1.6)	0.2 (0.4)	0.3 (0.8)
Stage 4	0 (0)	0 (0)	0 (0)
Stage 3+4	0.7 (1.6)	0.2 (0.4)	0.3 (0.8)
Stage REM	3.2 (6.4)	0 (0)	0 (0)
Total Sleep Time	12.8 (10.5)	7.5 (8.7)	1.7 (2.9)
Number of Sleep Onsets	1.5 (0.8)	1.0 (1.3)	0.3 (0.5)

The figures enclosed in brackets are the standard deviations

Means and Standard Deviations of the Sleep Stages in the Fourth Half-Hour Recording Period, in minutes.

Table 41

	Temazepam	F.P.	Placebo
Sleep Onset Time	24.0 (6.3)	27.0 (7.3)	24.0 (7.5)
Stage D+1	10.7 (4.0)	3.2 (4.0)	8.0 (7.1)
Stage 2	9.7 (5.5)	3.0 (7.3)	3.0 (4.3)
Stage 3	0.6 (1.0)	0 (0)	0.5 (0.8)
Stage 4	0.2 (0.4)	0 (0)	0 (0)
Stage 3+4	0.8 (1.3)	0 (0)	0.5 (0.8)
Stage REM	0 (0)	0 (0)	0 (0)
Total Sleep Time	10.5 (5.9)	3.0 (7.3)	3.5 (4.4)
Number of Sleep Onsets	1.5 (0.8)	0.3 (0.8)	0.5 (0.6)

The figures enclosed in brackets are the standard deviations

Sleep Onset Time

Table 42

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	25	20	24	23	6	0	4	22	26	15	15	30
2	30	30	30	30	30	17	30	30	30	30	30	30
3	30	8	30	19	16	27	24	13	30	17	10	30
4	30	30	30	30	30	23	10	30	30	30	30	30
5	11	8	30	30	9	1	12	22	15	5	15	12
6	30	30	30	12	29	7	30	24	24	20	30	30
Mean	26.0	21.0	29.0	24.0	20.0	12.5	18.3	24.0	25.8	19.5	21.7	27.0
S.D.	7.6	10.8	2.4	7.5	11.1	11.5	11.1	6.3	5.9	9.6	9.3	7.3

Summary ANOVA Table

Table 42A

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Drug	540.8	2	270.4	7.1	2.5%	2,10
Period	559.2	3	186.4	3.9	5.0%	3,15
Subject	2034.4	5	406.9			
Drug x Period	246.6	6	41.1	0.8	n.s.	6,30
Drug x Subject	379.8	10	38.0			
Period x Subject	720.3	15	48.0			
Drug x Period x Subject	1471.5	30	49.1			
Total	5952.6	71				

Newman-Keuls Multiple Comparisons

Drug

	Temazepam	F.P.	Placebo
Temazepam		5%	5%
F.P.			n.s.

Period

	2nd	3rd	1st	4th
2nd		5%	5%	5%
3rd			n.s.	n.s.
1st				n.s.

Number of Sleep Onsets — Table 43

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	1	1	1	1	2	2	2	2	1	2	2	0
2	0	0	0	0	1	1	0	1	0	1	0	0
3	0	2	0	1	1	1	2	2	0	3	1	0
4	0	0	1	0	0	2	2	0	0	0	0	0
5	4	2	0	0	2	3	2	2	2	3	3	2
6	0	0	0	1	1	2	1	2	1	1	0	0
Mean	.8	.8	.3	.5	1.2	1.8	1.5	1.5	.7	1.7	1.0	.3
S.D.	1.6	1.0	.5	.6	.8	.8	.8	.8	.8	1.2	1.3	.8

Summary ANOVA Table — Table 43A

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Drug	9.5	2	4.8	16.0	0.1%	2,10
Period	4.7	3	1.6	2.8	n.s.	3,15
Subject	26.4	5	5.3			
Drug x Period	3.6	6	0.6	1.0	n.s.	6,30
Drug x Subject	2.8	10	0.3			
Period x Subject	8.6	15	0.6			
Drug x Period x Subject	17.4	30	0.6			
Total	73.0	71				

Newman-Keuls Multiple Comparisons

	Placebo	F.P.	Temazepam
Placebo		n.s.	1%
F.P.			1%

Total Sleep Time

Table 44

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	6	5	7	5	21	28	24	15	6	15	16	0
2	0	0	0	0	4	10	0	8	6	4	0	0
3	0	13	0	11	10	8	11	16	0	16	10	0
4	0	0	3	0	0	10	20	0	0	0	0	0
5	16	21	0	0	23	23	21	13	14	19	19	18
6	0	0	0	5	2	16	1	11	5	12	0	0
Mean	3.7	6.5	1.7	3.5	10.0	15.8	12.8	10.5	5.2	11.0	7.5	3.0
S.D.	6.5	8.7	2.9	4.4	9.9	8.1	10.5	5.9	5.2	7.4	8.7	7.3

Summary ANOVA Table

Table 44A

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Drug	889.7	2	444.9	15.3	0.1%	2,10
Period	321.9	3	107.3	3.4	5.0%	3,15
Subject	1737.2	5	347.4			
Drug x Period	88.6	6	14.8	0.5	n.s.	6,30
Drug x Subject	291.2	10	29.1			
Period x Subject	471.6	15	31.4			
Drug x Period x Subject	833.1	30	27.8			
Total	4633.3	71				

Newman-Keuls Multiple Comparisons

	Placebo	F.P.	Temazepam
Placebo		n.s.	1%
F.P.			1%

	4th	1st	3rd	2nd
4th		n.s.	n.s.	5%
1st			n.s.	5%
3rd				n.s.

Stage Drowsy+1

Table 45

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	4	13	3	6	5	2	5	10	14	5	4	0
2	0	3	0	0	10	13	0	9	9	5	1	0
3	14	16	0	16	18	22	15	7	2	10	17	3
4	0	0	7	0	13	11	7	12	0	6	1	0
5	12	9	19	15	4	7	4	8	8	5	8	9
6	11	0	0	11	23	6	27	18	7	7	2	7
Mean	6.8	6.8	4.8	8.0	12.2	10.2	9.7	10.7	6.7	6.3	5.5	3.2
S.D.	6.3	6.9	7.5	7.1	7.4	7.0	9.8	4.0	5.1	2.0	6.2	4.0

Summary ANOVA Table

Table 45A

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Drug	362.9	2	181.5	2.8	n.s.	2,10
Period	34.5	3	11.5	0.5	n.s.	3,15
Subject	559.2	5	111.8			
Drug x Period	62.4	6	10.4	0.3	n.s.	6,30
Drug x Subject	657.8	10	65.8			
Period x Subject	344.0	15	22.9			
Drug x Period x Subject	902.9	30	30.1			
Total	2923.7	71				

Stage 2

Table 46

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	5	3	5	3	13	12	8	12	6	9	16	0
2	0	0	0	0	4	8	0	8	6	1	0	0
3	0	13	0	11	10	8	8	16	0	16	9	0
4	0	0	3	0	0	6	16	0	0	0	0	0
5	16	21	0	0	23	23	21	13	14	19	17	18
6	0	0	0	4	2	11	1	9	5	12	0	0
Mean	3.5	6.2	1.3	3.0	8.7	11.3	9.0	9.7	5.2	9.5	7.0	3.0
S.D.	6.4	8.8	2.2	4.3	8.6	6.1	8.2	5.5	5.2	7.8	8.2	7.3

Summary ANOVA Table

Table 46A

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Drug	459.1	2	229.6	20.3	0.1%	2,10
Period	160.4	3	53.5	2.0	n.s.	3,15
Subject	1531.4	5	306.3			
Drug x Period	74.3	6	12.4	0.5	n.s.	6,30
Drug x Subject	113.3	10	11.3			
Period x Subject	410.6	15	27.4			
Drug x Period x Subject	742.7	30	24.8			
Total	3491.8	71				

Newman-Keuls Multiple Comparisons

	Placebo	F.P.	Temazepam
Placebo		5%	1%
F.P.			1%

Stage 3

Table 47

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	1	2	2	2	5	3	0	2	0	5	0	0
2	0	0	0	0	0	2	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	1	0
4	0	0	0	0	0	4	4	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
6	0	0	0	1	0	3	0	2	0	0	0	0
Mean	.2	.3	.3	.5	.8	2	.7	.6	0	.8	.2	0
S.D.	.4	.8	.8	.8	2.0	1.7	1.6	1.0	0	2.0	.4	0

Stage 4

Table 48

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	0	0	0	0	3	13	0	1	0	1	0	0
2	0	0	0	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
6	0	0	0	0	0	2	0	0	0	0	0	0
Mean	0	0	0	0	.5	2.5	0	.2	0	.2	0	0
S.D.	0	0	0	0	1.2	5.2	0	.4	0	.4	0	0

Stage 3+4

Table 49

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	1	2	2	2	8	16	0	3	0	6	0	0
2	0	0	0	0	0	2	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0	0	1	0
4	0	0	0	0	0	4	4	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
6	0	0	0	1	0	5	0	2	0	0	0	0
Mean	.2	.3	.3	.5	1.3	4.5	.7	.8	0	.3	.2	0
S.D.	.4	.8	.8	.8	3.3	6.0	1.6	1.4	0	.8	.4	0

Stage REM

Table 50

Drug	Placebo				Temazepam				F.P.			
Period	1	2	3	4	1	2	3	4	1	2	3	4
Subject												
1	0	0	0	0	0	0	16	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0	3	0	0
3	0	0	0	0	0	0	3	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0	0	0	0
6	0	0	0	0	0	8	0	0	0	8	0	0
Mean	0	0	0	0	0	1.3	3.2	0	0	.5	0	0
S.D.	0	0	0	0	0	3.3	6.4	0	0	1.2	0	0

Summary of some of the Morning Questionnaire Responses Prior to the Morning Naps.

Table 51

Subject Number	Temazepam	F.P.	Placebo
1	AV/6hr	Tired/ 5½hr	AV/6hr
2	AV/6½hr	AV/7½hr	AV/6½hr
3	AV/6hr	AV/7½hr	AV/6½hr
4	AV/6½hr	AV/6hr	AV/7hr
5	AV/7hr	AV/6hr	AV/6hr
6	Tired/ 5½hr	Tired/ 7hr	AV/8hr

Summary of some of the Morning Questionnaire Responses on the Morning Following the Nap Sessions.

Table 52

Subject Number	Temazepam	F.P.	Placebo
1	Tired/ 6½hr	Tired/ 6½hr	Tired/ 7hr
2	AV/7hr	AV/7½hr	AV/6hr
3	AV/7hr	AV/7hr	AV/7hr
4	AV/7hr	Tired/ 7hr	Tired/ 7½hr
5	Tired/ 7½hr	AV/7½hr	AV/7hr
6	Headachy/ 7hr	AV/4½hr	AV/5½hr

The General Health and Well-Being of the Subjects immediately Following the Nap sessions.

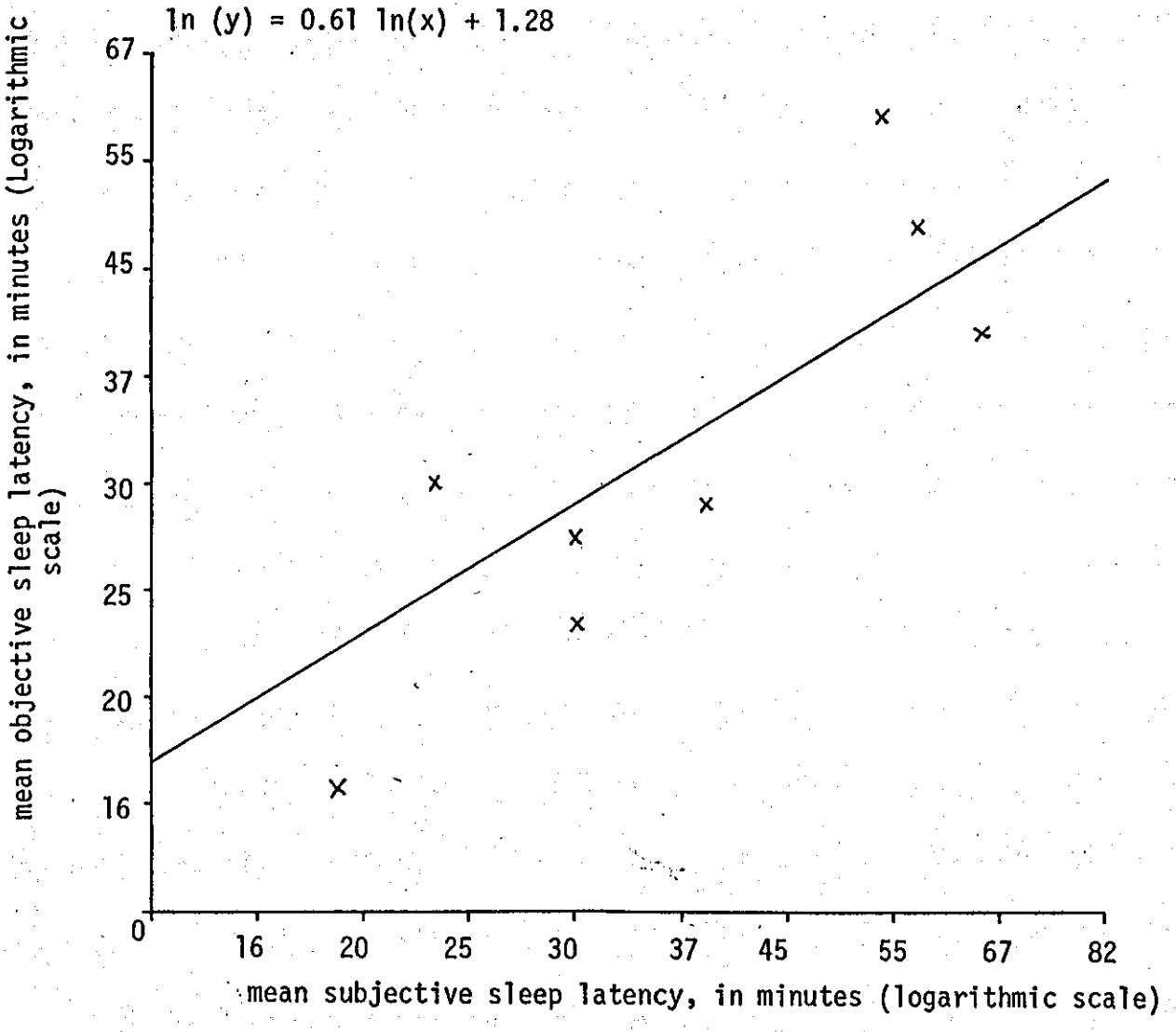
Table 53

Subject Number	Temazepam	F.P.	Placebo
1	OK	OK	OK
2	OK	OK	OK
3	OK	stomach discomfort	OK
4	OK	OK	OK
5	OK	OK	OK
6	OK	OK	OK

The General Health and Well-Being of the Subjects on the Morning after the Nap sessions.

Table 54

Subject Number	Temazepam	F.P.	Placebo
1	OK	OK	OK
2	OK	OK	OK
3	OK	OK	OK
4	OK	OK	OK
5	OK	OK	OK
6	OK	OK	OK



Graph C: Relationship Between Subjective and Objective Sleep Latencies

Chapter Three - Part I

THE EFFECTS OF ACETYLSALICYLIC ACID UPON SLEEP

1.0 INTRODUCTION

This aspect of the research programme emanated from the comment made by many of the self-reported, sleep onset insomniacs in chapter one, that they frequently took Aspirin* to relieve their insomnia. Initially, a literature search was carried out in order to determine if there was any evidence for the claims that Aspirin aids sleep: Only two studies of direct relevance were discovered.

Soldatos, Kales, Bixler, Scharf and Kales (1978) investigated the effects of sodium salicylamide (650 mg. and 1,300 mg.) upon the sleep of insomniacs; neither dose had a significant effect upon the sleep stages or latency to sleep onset, but with the higher dosage there was a declining trend in sleep latency upon drug administration, which was also reported by the subjects. This trend was indicative of a slight sedative effect. Some shortcomings of this study which are worthy of note are:

1. there were only four subjects in each dosage group,
2. the time of drug administration was not clearly stated, but simply referred to as "at night",
3. the total sleep time was not indicated in the results tables,
4. the percentage of stage 4 sleep, approximately zero for the groups of insomniacs, appeared to be rather low, but it is difficult to estimate by how much, since only the age range of the subjects was given (28 - 54 years) and not the group mean age,
5. no comments were made as to whether there was any inter-subject

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* It is necessary to explain the terminology to be used. Although the insomniacs reported taking Aspirin, very few, in fact knew what form of the drug they were taking. According to them Aspirin refers both to acetylsalicylic acid (A.S.A.) and its derivatives. However, throughout this text acetylsalicylic acid will be referred to as A.S.A. and its derivatives individually named.

variability in response to the drug; nor were the standard deviations presented in the results tables.

The second study by Pfeiffer, Goldstein, Murphee and Hopkins (1967), which examined the possible sedative properties of A.S.A. and its derivatives, was conducted during the day; ten minute EEG recordings were taken every hour over a period of seven hours. A.S.A. (1 gm.) was found to induce changes in the EEG, characterised by a decrease in the "mean energy content" and an increase in the "coefficient of variation" of the EEG signal. The authors claimed that upon visual inspection of the EEG records, their reported EEG changes would be represented by "EEG drowsiness", that is, "flattening of the amplitude of the alpha waves with sporadic occurrence of waves corresponding to light sleep and even some high amplitude slow waves", (Pfeiffer et al., 1967). They emphasised the similarity between their reported EEG changes brought about by A.S.A. and those brought about by some of the minor tranquilisers, such as chlorodiazepoxide. The authors suggested that A.S.A. may be effective in relieving some of the anxieties of everyday life. This study gave some indication that A.S.A. may have a sedative effect. Sodium salicylate, sodium salicylamide and acetaminophen were found to have negligible effects upon the EEG under the same experimental conditions. This study is open to the following criticisms.

1. The timing of the whole study was unclear; for example, whether all the drugs and placebo were administered to the subjects at the same time and whether all laboratory sessions commenced at the same time. Time of day effects are a very important variable when measuring sedation, as shown in the EEG model of insomnia study (chapter two). The subjects in the model study fell asleep significantly faster during the afternoon than during the morning.

2. No check was made as to whether the subjects were sleep satiated prior to the commencement of the study. This is another important variable which may have caused some subjects to show signs of drowsiness, regardless of the treatment. This may be reflected in the different "mean energy content" values for the subjects, which ranged from 38 - 84. It was not clear whether this initial difference in baseline values was taken into account during the data analysis.

3. The subjects were in the laboratory for seven hours, during which time they may have had refreshments. This is another variable which was not mentioned in the report.

It is also interesting to note that the EEG changes were observed approximately two hours after A.S.A. administration. If these changes are an indication of a sedative effect, it is unlikely that it would be beneficial to the sleep onset insomniac who takes the Aspirin just before retiring.

Soldatos et al. (1978) examined the effects of sodium salicylamide, an A.S.A. derivative, upon sleep. However, there were no published studies which had investigated the effects of A.S.A. upon sleep. It was therefore decided to continue the literature search further to determine whether any other properties of A.S.A. may be expected to affect sleep. Apart from its common effects, A.S.A. has a wide variety of lesser known effects at therapeutic doses, some of which are listed below:

1. Respiration - Full therapeutic doses of A.S.A. increase oxygen consumption and carbon dioxide production.

2. Acid-Base Balance and Electrolyte Pattern - In view of the previous point, equilibrium is maintained by the renal excretion of bicarbonate ions, accompanied by sodium and potassium ions.

3. Gastrointestinal Effects - The ingestion of A.S.A. may result in gastric bleeding, nausea, vomiting and gastric ulceration. Although some of these effects are experienced by some people at therapeutic dosages, in general, the dosage at which these effects are experienced varies widely from individual to individual.

4. Effect on Platelets - A.S.A., at a dose as low as 0.3 gm., can cause prolongation of bleeding time.
(Goodman & Gilman, 1970)

However, the aspects which were of particular interest were some of the proposed mechanisms by which A.S.A. is believed to exert some of its effects. One proposed mechanism is viā the inhibition of prostaglandin synthesis. The prostaglandins are believed to play an important role in neurotransmission. "They are thought to function postsynaptically by inhibition or facilitation of neurotransmission through cyclase inhibition or activation and by means of a negative feedback loop to inhibit further release of the neurotransmitter from the presynaptic nerve", (Gross, Dunner, Lafleur, Meltzer, Muhlbauer & Fieve, 1977).

1.1 Inhibition of Prostaglandin Synthesis by A.S.A.

Many of the studies demonstrating the inhibition of prostaglandin formation by A.S.A. have been conducted in vitro and with tissue preparations from animal species. For example, Vane (1971) found that

A.S.A. prevented the synthesis of prostaglandins in tissue homogenates from guinea pig lung.

Considering the human studies, Smith and Willis (1971) found that A.S.A. taken orally (600 mg., three times per day) inhibited the production of prostaglandins in the platelet system. However, they only experimented with three subjects. Koscis, Hernandovich, Silver, Smith and Ingerman (1973) in a similar study, found that A.S.A. (13 mg. per kg.) produced marked inhibition of prostaglandin formation in the platelet system and that the prostaglandin formation in the platelet system did not return to control levels for at least 72 hours. Hamberg (1972), by measuring the major urinary metabolites of prostaglandins in Man, found that A.S.A. (4 x 0.75 gm.) produced strong inhibition of the metabolites for 48 hours after cessation of A.S.A. administration.

This evidence indicated that A.S.A., at therapeutic doses, inhibited prostaglandin formation in Man and that this inhibition remained after the drug had been metabolised and removed from the body. However, reported I_{50} values for prostaglandin inhibition vary widely between different animal species and even between different enzyme preparations from the same species. This may be due to the following reasons:

1. Methods of Handling Tissue - Prostaglandin release, in some cases, can be brought about simply by handling the tissue.
2. Methods of Prostaglandin Analysis - There are a wide variety of assay techniques, for example, bioassay, radio-active tracers and gas-liquid chromatography. Furthermore, some experimenters examined the formation of the prostaglandins per se, whilst others looked at the prostaglandin metabolites. Koscis et al. (1973)

and Smith and Willis (1971) examined the formation of prostaglandins in the platelets by means of a bioassay, whereas, Hamberg (1972) examined the prostaglandin metabolites in the urine, using a radio-active tracer assay.

3. There is a whole family of prostaglandins; drugs may react differently to each subclass studied.

The inhibition of prostaglandin synthesis is not peculiar to the drug A.S.A.; Gross et al. (1977) compiled a list of some of the anti-depressant, anti-anxiety and anti-psychotic drugs which also inhibit prostaglandin synthesis to varying degrees.

1.2 Functions of Prostaglandins

Research in the prostaglandin field has advanced rapidly since 1962, when Bergström defined their chemical structure. Since then the prostaglandins have been implicated in many areas, for example, they are thought to play some rôle in pain, to contribute to the autoregulation of renal blood flow and to have various functions in the central nervous system. It is the latter function which is of major interest. The prostaglandins have been found to interact with brain neurotransmitter substances, such as, dopamine, noradrenaline and 5-hydroxytryptamine and they may well have an important role to play in the effectiveness of these neurotransmitters (Gross et al., 1977). For example, Bergström, Farnebo and Fuxe (1973) found that PGE_2 reduced the amounts of dopamine and norepinephrine that were released from the rat neostriatum and cerebral cortex following nerve stimulation.

1.3 Prostaglandins and Sedation

The prostaglandins have been found to bring about sedation in a wide variety of animal species. Horton (1964) observed in cats, that after intraventricular injection of prostaglandins of the E series (7 - 20 µg. per kg.), the cats showed signs of sedation and stupor. Horton and Main (1965) extended Horton's earlier work by demonstrating that $PGF_{2\alpha}$ did not bring about sedation in cats or chickens. PGE_1, E_2, $F_{1\alpha}$ and $F_{2\alpha}$ have all been identified in the dog brain and found to be distributed throughout all regions of the central nervous system (Holmes & Horton, 1968). On the basis of the findings of Horton and Main (1965) and Horton (1964), it would appear that it is the prostaglandins of the E series which are involved in sedation. Some investigators have qualified their behavioural observations with EEG and biochemical measures.

Haubrich, Perez-Cruet and Reid (1973), using rats injected with PGE_1 (1 mg. per kg., i.p.), found that sedation was evident within 15 minutes and persisted for about one hour. The period of sedation was reported to be characterised by a desynchronised EEG pattern, similar to that of the normal, waking rat, and a diminished muscular tone of the neck muscles; the rats could easily be aroused by gentle handling. The authors commented that these EEG and behavioural changes were indicative of REM sleep. The turnover rate of brain 5-hydroxytryptamine was found to increase following the administration of PGE_1. Furthermore, the behavioural effects of PGE_1 were blocked by drugs such as parachlorophenylalanine (PCPA), which lowers the levels of 5-hydroxyindole-acetic acid (5HIAA), a breakdown product of 5-hydroxytryptamine. Administration of PGE_1 (two doses of 1 mg. per kg., i.p., 45 minutes apart) was also found to increase the brain acetylcholine concentration

by 30 per cent. Prior administration of atropine (20 mg. per kg.) failed to block the sedation produced by PGE_1. The placebo group of rats who received four per cent ethanol in saline did not show any of these physiological or behavioural changes. The authors concluded therefore, that PGE_1 probably does not produce its sedative effect by increasing the brain acetylcholine concentration. Whereas, Perez-Cruet, Haubrich and Reid (1971) found that sedation in rats, produced by PGE_1 (1 mg. per kg., i.p.), was completely blocked by atropine (50 mg. per kg., i.p.). It would therefore appear that the dose of atropine used in their 1973 study was insufficient to block the production of acetylcholine completely. Prostaglandins may exert their sedative effects via both the 5-hydroxytryptamine and acetylcholine pathways.

Gilmore and Shaikh (1972) reported that PGE_1 and PGE_2 exerted a sedative effect when administered subcutaneously in the rat (approximately 2.5 mg. per kg.) and also caused flushing, increased respiration and diarrhoea. The authors found that the effects of the prostaglandins were not blocked by prior administration of Aspirin. This is to be expected, since Aspirin is believed to inhibit prostaglandin synthesis, but this does not necessarily imply that Aspirin will inhibit the action of administered prostaglandins. These results are therefore not contrary to the idea that the prostaglandins are involved in sedation. Gilmore and Shaikh (1972) concluded that the prostaglandins exerted their sedative effects by restricting blood flow to the brain (Yamamoto, Feindel, Wolfe, Katoh & Hodge, 1972).

Potts and East (1973) examined the sedative properties of the prostaglandins by another method, the response to a conditioned avoidance task. They also made EEG recordings. PGA_2, PGE_1, E_2 and arachidonic acid

(a precursor of prostaglandins) caused dose related decreases in avoidance responding, in the rat. The researchers reported that this behaviour was typical of that observed with general sedatives, such as, barbiturates. The EEG changes were less clear: PGE_2, administered intraventricularly, caused an increase in the low frequency activity of the power spectrum. Potts and East (1973) also reported that these changes were indicative of a "sleep like" state. When PGE_2 was administered either intraperitoneally or intragastrically, a decrease in the power of the EEG was evident, which indicates an alerting type of effect. These differences may be due to the various routes of administration employed, which results in difficulty in equating the dosages. There is also the possibility that the "alert type" of EEG, induced by PGE_2, was indicative of a REM like state of sleep, which is characterised by a low voltage, mixed frequency EEG, similar to that of the waking EEG. Haubrich et al. (1973) found that PGE_1 induced a state of sedation in the rat characterised by a desynchronised EEG pattern, which they claimed was indicative of REM sleep.

Nistico and Marley (1973), experimenting with adult fowls, concluded that PGE_1 (1 - 3 gms.), infused into the hypothalamus, resulted in behavioural and electrocortical sleep. Desiraju (1973) showed that PGE_1 (50 mg. per kg.), administered into the cerebral ventricle of an unanesthetised monkey, caused tremor, disturbance of posture and stupor for several minutes, followed later by somnolence; the EEG was desynchronised. Lower doses (25 mg. per kg.) caused drowsiness and facilitated sleep behaviour.

However, it is very difficult to compare the findings of these animal studies because:

1. different animal species were used,
2. not all of the studies employed EEG measures,
3. various routes of administration, a range of doses and different members of the prostaglandin family have been used.
4. Consideration must also be given to the fact that when the prostaglandins are injected directly into the brain areas they will result in abnormally high prostaglandin concentrations, since they are normally present in the brain in concentrations of the order of 10^{-9}gm.

It is difficult to extrapolate these animal findings to humans. However, there is sufficient evidence to suggest that prostaglandins in animals bring about sedation by means of interactions with brain neurotransmitter substances. It must be appreciated that the prostaglandins have a wide variety of effects and, at present, the sedative effect is being examined in isolation. Nevertheless, if the overall effects of prostaglandins in animals are taken as being indicative of their role in humans, then they might be expected to play some rôle in the sleep mechanisms of Man. This idea is admittedly rather speculative, but appears worthy of further investigation. Section 1.1, chapter three, provided evidence that therapeutic doses of A.S.A. administered orally to humans were able to inhibit the formation of prostaglandins in the platelets. If this inhibition can be assumed to hold for the brain areas as well, then A.S.A., rather than promoting sleep as the insomniacs believed, may instead have an arousing effect! It must be stressed that this proposed link between A.S.A., prostaglandins and sleep is only tentative and it is quite

possible that if A.S.A. is found to affect sleep in humans, it may be due to its actions on some entirely different mechanism.

1.4 The Relationship Between Aspirin and Tryptophan Binding

Another interesting property of A.S.A. is its capacity to bind to serum albumin, thereby causing the release of other bound molecules, such as tryptophan. Free, circulating tryptophan is the precursor of 5-hydroxytryptamine, which is believed to be one of the major neurotransmitter substances involved in sleep (Mendelson, Gillin & Wyatt, 1977). In rats, sodium salicylate (50 - 450 mg. per kg., i.p.) has been shown to increase serum free tryptophan, which then resulted in an increase in both the central nervous system uptake of tryptophan and the central nervous system turnover of 5-hydroxytryptamine (Tagliamonte, Biggio, Vargiu & Gessa, 1973). Guerinot, Poitou and Bohuon (1974) reported similar findings to those of Tagliamonte et al. (1973), but Guerinot et al. (1974) found that the lowest dose employed (50 mg. per kg., orally) did not increase brain tryptophan levels. This difference between the two studies may be due to:

1. Tagliamonte et al. (1973) used sodium salicylate, whereas Guerinot et al. (1974) used A.S.A.,
2. different routes of administration of the drugs were employed,
3. the drugs were administered at different times of day in the two studies,
4. different strains of rats were used,
5. tryptophan binding is very susceptible to changes in pH and temperature, therefore, any differences in handling techniques may have introduced another variable.

The comparable dose in humans to that employed in these two studies is approximately 17 times the recommended therapeutic dosage. The

equivalent study does not appear to have been performed on primates.

However, McArthur and Dawkins (1969) found that 10 mg. sodium salicylate in 50 mls. of human serum brought about a considerable release of tryptophan from its binding site. Smith and Lakatos (1971) found that in humans, after ingestion of A.S.A., the concentration of bound tryptophan was decreased and the mean free tryptophan concentration increased. A tryptophan loading study enabled the authors to demonstrate that a relatively high concentration of salicylate ion was required to produce a marked displacement of tryptophan. The increase in free tryptophan is therefore, likely to be short lived: it will only be elevated for as long as there is a sufficient salicylate ion concentration in the plasma to compete successfully with tryptophan for the binding site on albumin. Since tryptophan is the precursor of the neurotransmitter 5-hydroxytryptamine, which is believed to be involved in the sleep mechanisms (Mendelson et al. 1977), this action of A.S.A. was considered relevant. However, other drugs, such as the benzodiazepines, have also been shown to displace tryptophan from its binding site (Müller & Wollert, 1975). The pharmacological significance of this drug action is not yet fully understood.

1.5 Summary of the Literature

There has been no investigation into the effects of A.S.A. upon human sleep. Nevertheless, the literature suggested several A.S.A. actions which may affect sleep. However, caution must be applied when interpreting the experimental evidence for the various A.S.A. pathways, because the majority of studies have been conducted with different animal species or in vitro.

From the studies considered in the previous section, the link between A.S.A., prostaglandins and sleep is not absolutely clear. If A.S.A. does inhibit prostaglandin synthesis in the brain, one may have expected the results of the Guerinot et al. (1974) study to have shown the increased brain tryptophan content, but not the increased 5-hydroxytryptamine turnover rate. Haubrich et al. (1973) found that increased prostaglandin levels brought about an increased 5-hydroxytryptamine turnover rate. Subsequently, if the prostaglandins do play a major role in the pathways associated with the turnover of 5-hydroxytryptamine, when the prostaglandins are supposedly inhibited by, for example, A.S.A. administration as in the Guerinot et al. (1974) study, the turnover rate would be reduced. However, this discrepancy does not necessarily mean that the hypothesised link between prostaglandins, brain neurotransmitters and sleep no longer holds. It could be that the prostaglandins are exerting their sedative effects via the acetylcholine pathways, which were not examined by Guerinot et al. (1974). Haubrich et al. (1973) found, however, that prostaglandin administration was able to increase acetylcholine levels in rat brain.

There is also the possibility that the prostaglandins are involved in a regulatory rôle, maintaining a balance between the various neurotransmitters. In the Haubrich et al. (1973) study, the increased prostaglandin level may have indicated some neurochemical imbalance, which resulted in an increased turnover of 5-hydroxytryptamine and an increase in brain acetylcholine concentration. However, in the Guerinot et al. (1974) study this regulatory effect may have been lost due to prostaglandin inhibition by A.S.A., but the actual pathways for the conversion of tryptophan to 5-hydroxytryptamine may have been unaffected, resulting in an increased turnover rate. These ideas are

only speculative and much research is being conducted with the prostaglandins at present to determine the precise rôle of prostaglandins in neurotransmission.

There is sufficient evidence to suggest that prostaglandins play some rôle in sedation in animals. There is also evidence to suggest that A.S.A. inhibits prostaglandin production in both animals and Man. Caution must be applied here, because the evidence indicated prostaglandin inhibition in the periphery, but not necessarily in the brain areas. However, if brain prostaglandin synthesis is also inhibited, and if the prostaglandins are involved in the sleep mechanisms of Man, then one might expect A.S.A. to have an arousing effect upon sleep in Man. This idea is in fact contrary to the findings of the Pfeiffer et al. (1967) study, which indicated that A.S.A. has sedative properties; the results of this study were, however, inconclusive, because of the criticisms made in section 1.0, chapter three.

This proposed mechanism, involving the prostaglandins, does not eliminate the possibility that A.S.A. may exert an effect upon sleep by means of other pathways and actions. A.S.A. has a wide variety of effects, and it is possible that any one of them may produce a change in sleep, although the mechanism of action may not be clear at present. A.S.A. has also been shown to release tryptophan from its binding site, which, in turn, may bring about a change in sleep viā the 5-hydroxytryptamine pathway. However, it appeared that this A.S.A. action is likely to be short lived, effective only whilst the plasma concentration of A.S.A. could successfully compete with tryptophan for its binding site. The pharmacological significance of these two A.S.A. actions, tryptophan release and prostaglandin inhibition, is not yet clear.

The intial interest in the effects of A.S.A. upon human sleep stemmed from the comment made by the self-reported insomniacs of chapter one, that they frequently took Aspirin to relieve their sleeping difficulties. However, the literature search revealed that, apart from the findings of the Pfeiffer et al. (1967) study, which are open to criticism, A.S.A. is not reported as having sedative properties. Nevertheless, the literature search revealed other interesting aspects of the drug A.S.A., and although the evidence for A.S.A. having any effects upon sleep in Man was far from complete, it was decided that there were sufficient grounds to warrant a pilot study.

The aim of the pilot study was to investigate the effects of A.S.A. upon human sleep. It may seem that the most logical step would have been to evaluate the sedative potential of A.S.A. with a group of sleep onset insomniacs or by means of the EEG model of sleep onset insomnia (chapter two). Although both of these alternatives would have been very interesting lines of investigation, the literature revealed little evidence to support the claims that therapeutic doses of A.S.A. would have sedative properties: To the contrary, if some of the proposed mechanisms of A.S.A. action are correct, then one might expect A.S.A. to have an arousing effect upon sleep! It was therefore decided that priority ought to be given to the investigation of the effects of A.S.A. upon the sleep of normal, good sleepers before looking at its effects upon abnormal sleep.

2.0 PILOT STUDY

The aim of the pilot study was to investigate the effects of a normal, therapeutic dose of acetylsalicylic acid (A.S.A.) upon sleep.

3.0 METHOD

3.1 Experimental Design

The experimental protocol was:

Night	1	2	3	4	5	6	7	8	9
Treatment	Ad	Ad	B_1	B_2	B_3	A_1	A_2	R_1	R_2

Key:

Ad - Adaptation

B - Baseline

A - A.S.A. (2 x 300 mg., three times per day)

R - Recovery

Two adaptation nights, during which no EEG recordings were made, allowed for the adaptation effects described by Agnew et al. (1966). In this study three baseline nights were taken, in order to obtain a good, average measure of a typical night's sleep for each subject, for it was expected that any change brought about by A.S.A. may be slight. A.S.A., at a dosage of 2 x 300 mg., three times per day, was administered over a period of two days in order to determine if there was a cumulative effect of the drug. The recovery nights allowed for any changes in sleep upon drug withdrawal to be examined. The form of Aspirin chosen was A.S.A., because Pfeiffer et al. (1967) had found A.S.A. to be more effective at producing sedation than its derivatives, such as sodium salicylate.

For ethical reasons the dose of A.S.A. was limited to within the recommended range, 0.3 - 1.0 gm., taken orally every three or four

hours (Goodman & Gilman, 1970). The timing of A.S.A. administration for this study was 10.00 a.m., 4.00 p.m. and 10.00 p.m. Soldatos et al. (1978) appear to have given the total dose of sodium salicylamide just prior to the subjects' retiring time; however, it was considered that it may be rather disturbing for the subjects to have to take two tablets just as they were retiring. Therefore, 10.00 p.m. was the time chosen for the last dose, because this was only approximately one hour before the subjects retired; it also enabled the possible short term effects of A.S.A. to be observed. The dosage times were spaced at regular intervals during the day. No placebo control condition was incorporated into this experimental design, as it was simply a pilot study.

3.2 Subject Selection

The subject selection procedure was as described in section 6.1, chapter one. The subjects had to satisfy the general criteria of section 6.1, chapter one and in addition they had to be able to take A.S.A. without experiencing any unusual or marked side-effects. Eight subjects, (four males and four females), were selected from the student population. The females were all in the first half of their menstrual cycle at the commencement of the study.

3.3 Instructions to Subjects

The "general instructions to subjects" of section 6.2, chapter one, also applied to this group, along with the additional instruction that they must not take Aspirin or related drugs, such as Paracetamol and Disprin, for one week prior to the commencement of the study and during the course of the study, except when administered by the experimenter. The subjects were also instructed that if they had to take any medication for any reason during this time period, they were

to report it to the experimenter. The subjects were informed as to the dosage of A.S.A. to be used and of the timing of the dosages.

3.4 Experimental Procedure

The subjects were studied in pairs. They arrived at the laboratory approximately one hour before retiring. The general laboratory procedure was carried out, as described in section 6.3, chapter one. The retiring and rising times for each subject were kept constant ($\pm$ 15 minutes) from day to day.

A daily check was made, by means of self-report responses, on the general health and well being of each subject, to determine if they were experiencing any obvious adverse side-effects. The subjects were also instructed to report to the experimenter, if at any time during the course of the study they experienced gastric discomfort or nausea.

3.5 Data Analysis

The EEG records were scored, in one minute epochs, according to the standardised criteria of Rechtschaffen and Kales (1968). The records were scored blind and independently by two, experienced scorers. If disagreement between the scorers exceeded five per cent of the epochs, it was resolved by discussion.

The amounts of each sleep stage, (W+1), 2, 3, 4, (3+4) and REM, were all expressed in absolute minutes of 420 minutes, which was the lowest common measure of total sleep time for all subjects, on all nights. Total sleep times, sleep onset times and REM latency were also determined. The amount of each sleep stage, over the three baseline nights, for each subject was expressed as a mean. The results are presented in

Tables 55 - 63, section 25.0, chapter three.

A two way ANOVA with repeated measures on one factor was carried out for each sleep stage, total sleep time and sleep onset time, in order to determine if there were any significant differences between the nights for these parameters (McNemar, 1969, page 338). The Greenhouse-Geisser degrees of freedom correction factor was applied, (Winer, 1971, page 523): This correction factor lowers the degrees of freedom and makes the F test more conservative. An F value which yielded P levels of equal to or less than .05 was considered to be statistically significant. The significant ANOVA summary tables are presented in section 25.0, chapter three and the non-significant summary tables are presented in Appendix XI. If the ANOVA gave a significant F value it was followed by a Newman-Keuls multiple comparisons test, in order to determine which component of that factor contributed most to the significant F value (Winer, 1971, page 191).

Graphs D, E and F were drawn for both the whole group and the male and female groups independently, (section 25.0, chapter three). They are an illustration of nights against the group mean difference score for each sleep parameter. The difference scores were obtained by subtracting the value of a particular sleep parameter on each night from the baseline value. This enabled the changes from the baseline to be more clearly observed.

4.0 RESULTS

A significant F value for stage 4 sleep was found between the nights, ($F = 7.9$, df 1,7, $P \leq .05$, conservative test, Table 63A, section 25.0, chapter three). The Newman-Keuls multiple comparisons

test revealed that the group mean amount of stage 4 on nights A_1, R_1 and R_2 was significantly reduced in comparison with the baseline, but there was no significant difference between the baseline and A_2. On the second A.S.A. night, the group mean amount of stage 4 sleep returned to slightly above baseline values. Graph D (section 25.0, chapter three) clearly showed these trends. A trend test showed that a cubic trend was a very good fit for this data, ($P \leqslant .001$), as illustrated with graph G (section 25.0, chapter three), (Meddis, 1975, page 143).

5.0 DISCUSSION OF RESULTS

The major significant finding was the decline in stage 4, which reached its lowest value, compared with the baseline, on the first recovery night. There was also a marked decline in stage (3+4), which followed a similar pattern to stage 4, but failed to reach statistical significance. From graph E, for the male and female groups independently, it is interesting to note that for the males, by the second recovery night both stages 4 and (3+4) had returned to slightly beyond baseline values, whereas, in the case of the female group, both stages 4 and (3+4) were still markedly depressed in comparison with the baseline. Also of interest was the declining trend in stage 3 sleep for the female group, while the male group showed only a slight decline in stage 3 on the second A.S.A. night. Graph D (section 25.0, chapter three) revealed a marked increase in stage 2 sleep for the whole group on the first recovery night; as stage 4 was significantly reduced, it follows that one of the other stages would show an increase.

Since there were only four subjects in the male group and four in the female group, caution should be applied when interpreting the individual group findings. However, the general trend of a decline in

stages 4 and (3+4) did appear to be more marked in the female group, but, in fact, this general trend did not hold for all the females. With reference to Table 63 (section 25.0, chapter three), subject 5 did not show a decline in stage 4 on either A_1 or A_2, and only a slight decline on R_1 and R_2: there appeared to be a large amount of inter-subject variability in response to A.S.A. This might have been expected for a drug such as A.S.A., which has such a wide variety of effects, thereby increasing the number and degree of responses which each individual could show upon A.S.A. administration. The overall difference between the response of the male and female groups to A.S.A. may have been due to the differences in body size; or maybe, if A.S.A. is exerting its effects upon sleep via the prostaglandin pathway, higher doses of A.S.A. may be required to block prostaglandin synthesis in males, because men produce larger amounts of prostaglandins (50 - 330 μg per day) than females (20 - 40 μg per day), (Hamberg, 1972).

It is not clear why the effect of A.S.A. upon stage 4 sleep should show a cubic trend. One explanation may be that on the first A.S.A. night, the stage 4 decline is due to an initial "shock" response by the system. Perhaps by the second night of A.S.A. administration compensatory changes have been made, but on the recovery nights the system cannot cope, possibly due to a build up of some metabolites or due to some long lasting inhibitory effects of the drug. This is only a hypothetical explanation.

One may argue that the changes in sleep observed were simply due to placebo effects; however, the results of previous studies on the effects of placebos on sleep showed that this explanation seemed unlikely. Adam, Adamson, Brezinová and Oswald (1976); Touyz, Beumont, Saayman, Stern

and Zabow (1978) and Kales et al. (1971) all found that a placebo had no effects upon sleep. To the contrary, Hartmann and Cravens (1973) reported that a placebo did have a significant effect upon sleep. There was no dramatic effect when the placebo was first given, but several days after the discontinuation of the placebo, there was a distinct increase in REM sleep and total sleep time. Since total sleep time increased, one would expect REM sleep to increase, but the increase in REM sleep did appear to be greater than could be explained solely by an increased sleep length. This result is rather unusual and has not been reported by other researchers. The aim of the Hartmann and Cravens (1973) study was not primarily to evaluate the effects of placebos upon sleep; the placebo study was only one of six, 60 day drug studies with the same group of 14 subjects undergoing each condition. This may, in some way, have contributed to this unusual placebo response. Nevertheless, none of the placebo studies considered have reported effects similar to those of the present study. In fact, the group mean amount of REM sleep, for the present study, showed only minor fluctuations after A.S.A. administration: a slight decline on the second drug night and a small increase on the first recovery night. It is unusual that A.S.A. did not significantly reduce stage REM in the whole pilot study group, since this appears to be a common effect of many drugs upon sleep (Kay, Blackburn, Buckingham & Karacan, 1976). However, the male group did show a marked decline in stage REM, see graph F (section 25.0, chapter three), in comparsion with the female group.

A.S.A. did not have any significant effect upon sleep onset times or total sleep time. However, since these subjects were self-reported good sleepers, with a mean baseline sleep onset time of 21 minutes and

a group mean total sleep time equal to 455 minutes, it is unlikely that any drug would be able to either reduce the sleep latency or increase the total sleep time. Furthermore, A.S.A. did not prevent the subjects from falling asleep as quickly as they normally did nor did it cause them to wake earlier.

6.0 CONCLUSIONS

There was evidence from the pilot study to indicate that A.S.A. was having an effect upon sleep. Stage 4 was significantly depressed on nights R_1, R_2 and A_1, in comparison with the baseline. It also appeared that the general, overall declining trend in stage 4 was more marked in the female group. The stage 4 response to A.S.A. followed a cubic trend. There was also an increasing trend in stage 2 sleep for the whole group, reaching a peak on the first recovery night, but this trend did not reach statistical significance. There were no statistically significant changes in stage REM for the whole group, after A.S.A. administration.

7.0 MAIN A.S.A. STUDY

8.0 INTRODUCTION

The findings of the pilot study provided a sufficient basis for pursuing this line of investigation further. The next step was to investigate the effects of A.S.A. upon sleep by means of a larger, well controlled study.

9.0 METHOD

9.1 Experimental Design

The basic design was split plot with repeated measures on one factor (Winer, 1971, page 525), comprising of eight females in the A.S.A. group and eight in the placebo group. The study was conducted under double-blind conditions.

On the basis of the findings of the pilot study, it was decided to use only female subjects, because the overall trends did appear to be more marked in this group. Since there was quite a wide variation in response to A.S.A. in the pilot study, with respect to sleep patterns, an all female study may help to reduce some of this variability. It was decided to extend the period of A.S.A. administration from two to four consecutive days; the aim of this was to enable further investigation of the previous observations of an increasing trend in stage 4 sleep towards baseline values on the second A.S.A. night and to observe whether this increasing trend in stage 4 continues with repeated A.S.A. administration. Furthermore, since stages 3 and 4 had not returned to baseline values by the second recovery night, for the female group, it was decided to extend the recovery period to six nights, by which time these stages may have returned to baseline values.

All subjects were required to attend the laboratory for 15 consecutive nights. The experimental protocol was:

Night	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Treatment	Ad	Ad	B_1	B_2	B_3	A_1	A_2	A_3	A_4	R_1	R_2	R_3	R_4	R_5	R_6

Key:

Ad - Adaptation

B - Baseline

A - A.S.A. (2 x 300 mg., three times per day)

R - Recovery

The first two nights in the laboratory were for adaptation purposes (Agnew et al., 1966) and no EEG recordings were made. Similarly, on the third A.S.A. night and fourth recovery night no EEG recordings were made. It would have been very costly to have recorded on all 13 nights and it was considered that sufficient knowledge could be gained about the activity of A.S.A. even if nights A_3 and R_5 were not recorded. This also gave the experimenter time to relax. However, the subjects were led to believe that recordings were made on every night; the reason for this was to keep the subjects' set fairly constant from night to night and to avoid the risk of some of the subjects failing to observe some of the "instructions to subjects", if they had known that no recordings were being made.

The same dosage of A.S.A. (2 x 300 mg., three times per day) and the times of administration (10.00 a.m., 4.00 p.m. and 10.00 p.m.) were employed as in the pilot study. The placebo tablets were supplied by drug manufacturers and were made to match the A.S.A. tablets in shape, colour and consistency. They did, however, differ with respect to taste. This difference could only be detected if the tablets were broken down whilst still in the mouth; the subjects were therefore instructed to swallow the tablets whole. The contents of the placebo tablets are listed in Appendix XVII. The dosage of placebo administered was identical to that for A.S.A.

9.2 Subject Selection

The subjects were selected according to the general procedure of section 6.1, chapter one. The subjects had to satisfy the general criteria for good sleepers and they had to be able to take A.S.A. without experiencing any adverse side-effects. Sixteen females were selected, mean age 19.8 years. They were all in the first half of their menstrual cycle at the commencement of the study. The subjects were matched for age and body size; they were all drawn from the student population.

9.3 Instructions to Subjects

The subjects were given the "general instructions" of section 6.2, chapter one, plus the additional instruction that they must not take Aspirin or related drugs, such as Paracetamol and Disprin, for one week prior to the commencement of the study and also during the course of the study, except when administered by the experimenter. The subjects were also instructed that if they had to take any medication, for any reason during this time period, then they were to report it to the experimenter The subjects were informed as to the dosage of A.S.A. to be used and the timing of the dosages, but the subjects were not told that half of them would be on placebo. It was considered that this information may lessen any placebo response. This comment was also made by Kales et al. (1971).

9.4 Experimental Procedure

The subjects were studied in pairs, one of each pair receiving A.S.A. and one receiving placebo, under double-blind conditions. The subjects were allocated at random to either the drug or placebo group. The subjects who attended the laboratory on the same nights were matched

for similar retiring and rising times. They arrived at the laboratory approximately one hour before retiring. The general laboratory procedure was carried out as in section 6.3, chapter one. The retiring and rising times of each subject were kept constant to within ± 15 minutes over the 15 days.

A daily check was made, by means of self-report data, as to the general health and well being of the subjects. The subjects were instructed to report to the experimenter if, at any time during the study, they experienced gastric discomfort or nausea. The subjects were also asked to complete the questionnaire shown in Appendix V on the last morning of the study; the reason for this was to obtain some estimate of how frequently the subjects would normally take a drug such as A.S.A.

9.5 Data Analysis

The EEG records were scored, in one minute epochs, according to the standardised criteria of Rechtschaffen and Kales (1968). The records were scored blind and independently by two, experienced scorers. If the disagreement between the scorers exceeded five per cent of the epochs, it was resolved by discussion. Each sleep stage, (W+1), 2, 3, 4, (3+4) and REM, was expressed in absolute minutes out of a total sleep time of 400 minutes, this was the lowest common measure of total sleep time, for all subjects on all nights. The baseline values for each sleep parameter over the three nights were expressed as a mean. Total sleep time, sleep onset time, REM latency, REM periodicity and the number of stage changes were also determined for all subjects on all nights. The results are shown in Tables 64 - 74, (section 25.0, chapter three).

Initially, the two groups, A.S.A. and placebo, were tested for homogeneity of variance according to the method of Winer (1971, page 527). The F_{max} value was calculated for each sleep parameter, for both "subjects within groups" and "nights x subjects within groups", over the drug and recovery nights (Table 80, section 25.0, chapter three). Only 7 out of the 40 F_{max} values reached significance. Therefore, it is reasonable to assume that the two groups were drawn from populations with equal variances. Since the F test has been shown to be robust (Boneau, 1960; Havlicek & Peterson, 1974), it would be expected to accommodate the majority of these unequal variances. However, since sleep onset time scores had previously been shown to be severely skewed (section 10.3, chapter one), the significant F_{max} value for sleep onset times over the drug nights did give cause for concern. This parameter was, therefore, violating two of the underlying assumptions of the F test. Therefore, for this particular parameter it was decided to perform the ANOVA on both the raw and transformed scores for means of comparison.

Before the statistical analysis could be embarked upon, the problem of unmatched baseline scores for each sleep stage, for the two groups had to be resolved. Unrelated t-tests revealed that there were no significant differences between the baseline means for each sleep parameter, for the two groups. However, it was felt that this problem could not be overlooked simply because the difference was not significant, as any changes observed after drug administration may simply be due to the fact that the two groups had different values initially. The difference between the two groups was most marked for stage 4 sleep, with a value of 15 minutes. It is difficult to explain this large difference in baseline scores, as the subjects were matched for age,

sex and body size and were randomly assigned to either group. Furthermore, within each group there was a large amount of inter-subject variability for the baseline values of several sleep parameters, notably stages 2 and 4. In an attempt to overcome this problem of unmatched baseline scores for the two groups, it was decided to perform the main analysis on the difference scores, that is, the scores for each sleep parameter, on every night were expressed as the difference from the baseline.

The recommended statistical analysis for the experimental design employed in this study is a multivariate method (Thornby, 1976). However, after consultation with the program advisors, it was discovered that the package programs available would be unable to handle a multivariate analysis on this data, because of the large number of parameters. The best approach which they could recommend was to use the GENSTAT package (Nelder, 1977). This would, in effect, perform a univariate ANOVA, but would consider all the sleep stages at once. GENSTAT has only been available for a few months, therefore considerable difficulty was experienced in trying to run the program. Whilst sorting out the problems an alternative analysis was carried out.

A three factor, split plot ANOVA was performed on each sleep parameter's difference score, over the drug and recovery nights independently (Winer, 1971, page 525). The Geisser-Greenhouse correction factor for conservative degrees of freedom was applied where appropriate, according to the procedure in Winer (1971, page 523). The ANOVAs which gave significant results were examined, when applicable, by means of the Newman-Keuls multiple comparison test (Winer, 1971, page 191). P levels which were equal to or less than .05 were considered to be statistically significant. The significant ANOVA summary tables are

presented in section 25.0, Tables 75 - 79, and the non-significant ANOVA summary tables are presented in Appendix XII for the drugs nights and Appendix XIII for the recovery nights.

The standard deviations across the drug and recovery nights were computed for each subject, for each stage of sleep, and the geometric means of the standard deviations for the two groups were compared by means of an unrelated t-test. The reason for this procedure was to determine whether A.S.A. significantly increased the variability of the sleep stages across the nights, in comparison with the placebo.

Graphs (H, I, J and K) were drawn for the A.S.A. and the placebo groups of nights against the group mean difference scores, for each sleep stage. The cumulative graphs (L and M) were also drawn for the second drug night, because this appeared to be the night which showed the most prominent changes in stages 2 and 4. These graphs are a plot of half-hour epochs throughout the night against the accumulation of the stages in minutes. The reason for these graphs was to determine whether A.S.A. was affecting the distribution and build up of these sleep stages throughout the night. All these graphs are included in section 25.0, chapter three.

10.0 RESULTS

The three-way, split plot ANOVA gave the following significant results:

Drug Nights

Stage 2 - A significant difference between the A.S.A. and placebo conditions, over the drug nights: A.S.A. produced a significant increase in stage 2 ($F = 14.5$, df 1,14, $P \leq .01$, conservative test), (Tables 65 and 75, section 25.0).

Total Sleep Time - A significant difference between the three drug nights, which is independent of the treatment condition ($F = 9.4$, df 1,14, $P \leq .01$, conservative test): the second drug night showed a significantly lower total sleep time than either the fourth drug night ($P \leq .05$) or the first drug night ($P \leq .01$), (Tables 73 and 76, section 25.0).

Sleep Onset Time - A significant difference between the drug nights ($F = 10.8$, df 1,14, $P \leq .01$, conservative test, raw scores; $F = 6.4$, df 1,14, $P \leq .025$, conservative test, transformed scores), with a drug by night interaction effect ($F = 8.7$, df 1,14, $P \leq .01$, conservative test, raw scores; $F = 5.3$, df 1,14, $P \leq .05$, conservative test, transformed scores). The group mean sleep onset time was significantly increased on the first placebo night in comparison with the A.S.A. group (Tables 72, 77 and 78, section 25.0).

Recovery Nights

REM Latency - A significant difference between the two drug conditions, ($F = 5.5$, df 1,14, $P \leq .05$, conservative test): the A.S.A. group showed a significant decline in REM latency in comparison with the placebo group, (Tables 70 and 79, section 25.0).

A.S.A. was found to increase significantly the night-to-night variability of stage REM over the recovery nights ($t = 2.9$, df 14, $P \leq .02$) and of stage 3 over both the drug and recovery nights ($t = 2.6$, df 14, $P \leq .02$ and $t = 4.4$, df 14, $P \leq .01$, respectively), in comparison with the placebo group, using a two-tailed test.

The ANOVA summary tables from the GENSTAT analysis are presented in Appendix XIV for the drug nights and Appendix XV for the recovery

nights; this analysis revealed the same effects as were reported above.

11.0 DISCUSSION OF RESULTS

The significant increase in stage 2 sleep with A.S.A. administration can be clearly seen from graph H, the increase reaching a maximum on the second A.S.A. night. From the same graph it can also be seen that there was a slight declining trend in stage REM; this was not statistically significant. Also of interest is the declining trend in stage 4 sleep during A.S.A. administration (graph I). This trend was not statistically significant, but was greater that the decline in stage REM. In the pilot study a significant decline in stage 4 was found on nights A_1, R_1 and R_2, in comparison with the baseline. In the main study, however, stage 4 was found to return to baseline values on R_1 and R_2 and to increase slightly above on R_3 and R_4. Overall, the general trend in the stage 4 findings was similar to those of the pilot study, but they were not as marked; this may be due to the fact that, for the pilot study, the results were compared with the baseline and not with a placebo control group. Another possible reason for this difference in results may be the large amount of inter-subject variability shown by stage 4 sleep in response to A.S.A. administration: for example, on the second A.S.A. night the range in difference scores for stage 4 sleep was -51 minutes to +3 minutes. This wide variation in response may, in part, be attributable to:

1. the dosage level of A.S.A. used may not have been sufficiently high to bring about a consistent response in all subjects,
2. the range in individual tolerance levels to A.S.A.,
3. the degree of response of each subject to the large number of A.S.A. actions.

Another indication of the large amount of inter-subject variability in

response to A.S.A. can be seen on the recovery nights: subject 6 showed her lowest decline in stage 4 on R_3 and subject 3 on R_4.

Also worthy of comment is the fact that three of the placebo subjects, namely, 11, 12 and 16, showed a marked decline in stage 4 sleep on the second placebo night, but this decline was not revealed for the group as a whole, because two subjects showed a marked increase. It is difficult to explain these changes in the placebo group subjects' sleep over and above that due to nightly variation. This observation does weaken the findings of the A.S.A. pilot study with respect to stage 4 sleep: they may have been artefacts due to the lack of a placebo control group. In the pilot study data analysis, the sleep parameter scores on the drug and recovery nights were compared with the baseline scores and not with a placebo control group; since the subjects of the present placebo group showed a considerable amount of inter-subject variability in response to placebo administration, it appears necessary to compare all the results obtained upon A.S.A. administration with a placebo control group.

Upon withdrawal of A.S.A. a significant reduction in REM latency was found. This appeared to be most marked on the second recovery night and raises an interesting possibility: since this reduction in REM latency was not observed until the recovery nights, it indicated that the effect was not due to A.S.A. per se, but maybe to one of its metabolites which may have still been left in the body. This decrease in REM latency could be classed as a qualitative change in REM sleep and did not appear to be linked with a statistically significant quantitative change in REM sleep. However, the night-to-night variability of REM sleep was significantly increased for the A.S.A. group, in comparison

with the placebo group over the recovery nights. This increased variability may, in some way, be linked with the decline in REM latency: if the REM periods commenced earlier, there is the possibility that some subjects may have had lengthened REM periods on some nights, which may have been compensated for by a decrease on the next, thereby increasing the night-to-night variability. A possible explanation for the decrease in REM latency is that some A.S.A. metabolites may have interfered with a REM triggering mechanism, but mechanisms which controlled the absolute amounts of REM sleep were not significantly altered. These suggestions are only tentative.

Also worthy of comment is the fact that REM sleep underwent no significant changes during A.S.A. administration, in comparison with the placebo group. This was a rather unexpected result, as REM sleep is considered to be a fragile type of sleep which can easily be disrupted by a variety of factors, such as, drugs and anxiety. If A.S.A. was affecting one of the major neurotransmitter pathways or maybe, even if it was producing unpleasant side-effects, then one might expect REM sleep to decline.

The remaining significant findings were not simply due to the drug effects. Over the drug nights there was a significant night and night x drug interaction effect, for both the raw and transformed sleep onset time scores. Inspection of Table 72 (section 25.0, chapter three), clearly shows that it was the first placebo night which showed a significant increase in the group mean sleep onset time. The multiple comparison test revealed that the placebo group mean sleep onset time on A_1 was significantly higher than on A_2 and A_4. Only three of the eight subjects failed to show a large increase in sleep onset time on

this night. There was no obvious reason for this large increase on the first placebo night. Although the subjects were allocated at random to the groups, it may be that, by chance, subjects who were anxious about taking tablets were in the placebo group, but this idea was not borne out by the questionnaire responses. A significant night effect was also found for total sleep time; the second drug night for both A.S.A. and placebo groups showing a decreased total sleep time in comparison with the first and fourth drug nights. Once again there was no obvious reason why this effect should be observed, as the retiring and rising times of all subjects were kept to within $\pm$ 15 minutes of their normal times each day. These changes may be due to the fact that the second drug night was a Sunday night, which may have caused the subjects to wake earlier in anticipation of "Monday morning". The other Sunday night spent in the laboratory was R_5 which was not recorded, therefore, there is no other data available from this study to support this idea. However, the significant changes in total sleep time will not affect the results, because only the first 400 minutes of sleep time for each subject were analysed.

The cumulative graph (L) revealed little change in the distribution of stage 2 sleep over the second drug night. However, graph M revealed that the accumulation of stage 4 sleep levelled off earlier on the second A.S.A. night in comparison with the A.S.A. baseline night and the second placebo night. This levelling off occurred after approximately $2\frac{1}{2}$ hours. A.S.A. did, therefore, appear to be having a slight suppressing effect on stage 4 sleep after the first $2\frac{1}{2}$ hours of sleep.

On the basis of the overall questionnaire responses, neither group

could be classified as regular users of A.S.A. Only two subjects from the A.S.A. group and three from the placebo group reported taking Aspirin. Other compounds taken by the subjects, as determined by the questionnaire, included Paracetamol, Panadol and Phensic. Two weeks was the shortest time interval between a subject taking an analgesic and commencing the study, the longest was approximately six months (subject 5), the average time interval being 1 - 2 months. With regard to reported side-effects, only one subject in the A.S.A. group and two in the placebo group reported slight stomach discomfort. Three subjects (1, 4 and 6) in the A.S.A. group and one in the placebo group reported that A.S.A. made them feel drowsy and one subject (7) reported that A.S.A. made her feel lively. Five subjects reported being slightly anxious about taking A.S.A. for four days: three in the A.S.A. group and two in the placebo group. All subjects considered that they consumed the same amount or slightly less carbohydrate than the rest of the student population.

Having considered both the statistically significant findings and the trends observed from the graphs, it is necessary to determine whether there is a link between them. Although this study showed that A.S.A. did have an effect upon sleep, no indication was given as to why; but some speculations will be made. The only statistically significant finding for the sleep stages was the increase in stage 2 sleep shown by the A.S.A. group on the second drug night, in comparison with the placebo group. A change in stage 2 sleep is generally viewed as being secondary to a change in one of the other sleep stages which are believed to be associated with more important functional roles, such as, stage 4 and REM sleep. There is, however, the alternative view that the changes in stage 2 sleep may be the primary effect and the other changes

secondary to those observed for stage 2 sleep. Adopting the approach of the first view point, the overall impression gained from the results is that A.S.A. is having a slight disruptive effect upon sleep which is buffered by a significant increase in stage 2 sleep. This disruptive effect is reflected over the drug nights by the declining trend in stage 4 sleep, the slight decline in REM sleep, the significant increase in the night-to-night variability of stage 3 and the reduction in the accumulation of stage 4 after approximately 2½ hours of sleep on the second drug night. A.S.A. did not appear to have a marked disruptive effect upon sleep, otherwise REM sleep would have been expected to show a significant decline in comparison with the placebo.

One may speculate therefore, that A.S.A. is not acting viā one of the major neurotransmitter pathways associated with sleep, because a more dramatic change in the sleep stages would be expected. The effects of A.S.A. upon sleep appear to be rather subtle and may be brought about by the influence of A.S.A. upon subsidiary pathways which regulate the amounts of the various sleep stages. This might be achieved by the interactions of A.S.A. or its metabolites with various catalysts and co-enzymes, or maybe even viā prostaglandin inhibition. However, it must not be overlooked that A.S.A. may not be affecting sleep by its direct action on neural transmitter pathways associated with the sleep mechanisms, but by such actions as its effects upon respiration.

It was suggested in an earlier section (1.5, chapter three), that A.S.A. may have an arousing effect upon sleep. The reasoning behind this hypothesis stemmed from the evidence, revealed by the literature search, which indicated that prostaglandins produce sedation in animals. If the prostaglandins have a similar function in Man, and the dosage of

A.S.A. used was sufficient to block prostaglandin synthesis in the brain areas, then one might expect A.S.A. to exert an arousing effect. A.S.A. did not interfere with the sleep induction process, as the sleep onset times were not significantly increased upon A.S.A. administration. This hypothesised arousing effect was, therefore, not so strong as to delay sleep onset; neither did it bring about an increased amount of stage (W+1) or a decreased total sleep time. However, this idea was partly borne out by the significant increase in stage 2 sleep, which is a light stage of sleep: A.S.A. does, therefore, appear to have only a slight arousing effect upon sleep. It could be that A.S.A. is affecting some regulatory mechanism controlling the ratio of light to deep sleep during the course of the night. If this were the case, one might expect stage 2 sleep to show its largest increase during the first half of the night, when the deeper stages of sleep are most prominent, but this suggestion is not supported by graph L (section 25.0, chapter three), which shows that the increase in stage 2 sleep is fairly evenly ditributed throughout the night. Therefore, some control mechanism which may regulate the ratio of REM sleep to stage 2 may also have been affected by A.S.A. administration. These ideas are only speculative.

There is also the possibility that the action of A.S.A. upon respiration (increased oxygen consumption and carbon dioxide production) may have caused this significant shift to the lighter stage of sleep, (stage 2). The increased carbon dioxide production may have caused an unbalancing of the homeostasis, resulting in EEG arousal. However, it is unlikely that the total daily dosage of A.S.A. used in the present study would have a marked effect upon respiration: Goodman and Gilman (1970) reported that a full therapeutic dose of A.S.A. (4 gm.) produced these respiratory changes, which was more than twice the dose of A.S.A.

used in the present study (1.8 gm).

The decline in REM latency, observed upon withdrawal of A.S.A., was previously discussed in this section and it was tentatively suggested that this effect of A.S.A. may have been brought about by the action of some A.S.A. metabolites upon a REM triggering mechanism.

In the introduction to this chapter various A.S.A. actions were put forward as possible mechanisms by which A.S.A. may affect sleep. The tryptophan releasing effect does seem unlikely in this case, as there is evidence to suggest that high doses of A.S.A. are required to bring about a substantial release of tryptophan (Smith & Lakatos, 1971). There may be a transient rise in free tryptophan when the plasma concentration of A.S.A. is relatively high; this effect is likely to be short lived. With regard to the prostaglandin pathway, from the studies of Koscis et al. (1973) and Hamberg (1972), the dosage of A.S.A. used in the present study appears to be sufficient to block prostaglandin synthesis. The available evidence indicated inhibition of protaglandin synthesis in the periphery, but one cannot necessarily conclude that, in Man, oral administration of a therapeutic dose of A.S.A. blocks the synthesis of prostaglandins in the brain areas. Also, Hamberg (1972) claimed that A.S.A. produced marked inhibition of prostaglandin synthesis upto 48 hours after cessation of the drug. If the significant rise in stage 2 sleep were due primarily to the inhibition of prostaglandin synthesis, one would have expected it to have remained substantially elevated on the first two recovery nights; but only a slight increase was observed on R_1 and R_2. This may be due to a progressive decline in prostaglandin inhibition.

Another mechanism by which A.S.A. may be exerting its effects upon sleep has been suggested by Traynor (1978, personal communication). The acetyl moiety of A.S.A. may cause acetylation of important plasma proteins or may interfere with the amino-acid transport mechanism by which means tryptophan and related compounds gain entry to the brain. There is also the possibility that some side-effects of A.S.A., such as, gastro-intestinal bleeding may bring about changes in sleep.

Consideration must also be given to the fact that the results obtained from this study did not replicate those of Soldatos et al. (1978) who reported no statistically significant changes in the sleep stages of insomniac subjects, who had been administered sodium salicylamide (650 and 1,300 mg.), a derivative of A.S.A. There are several possible reasons for this:

1. Soldatos et al. (1978) investigated the effects of sodium salicylamide upon sleep, whereas, in the present study, A.S.A. was the drug used. It was reported by Goodman and Gilman (1970) that sodium salicylamide, under double blind conditions, did not demonstrate analgesic or anti-rheumatic properties even at high doses which caused a high percentage of gastro-intestinal and other side-effects. Sodium salicylamide does, therefore, appear to be a weaker analgesic and anti-rheumatic agent than A.S.A. It does not bind to plasma proteins, to any great extent, at therapeutic doses. These differences in drug action may bring about different effects upon sleep.

2. Soldatos et al. (1978) appear to have given a single dose of the drug prior to retiring on each of the three drug nights,

whereas, in the present study, the drug was administered three times per day, at six hourly intervals over a period of four days.

3. Soldatos et al. (1978) used insomniac subjects, whereas, good sleepers were used in the present study.

Furthermore, since the completion of this research programme, an abstract of a paper presented by Hauri and Silberfarb (1978) at the eighteenth Association of the Psychophysiological Study of Sleep meeting, at Stanford, California, has been published recently. These authors have studied the effects of A.S.A. on the sleep of insomniacs; they had also noted that A.S.A. is taken as an hypnotic by many insomniacs, which supports the claims made by the self-reported, sleep onset insomniacs of chapter one that Aspirin relieved their sleeping difficulties. They examined the effects of a single dose of A.S.A. (650 mgs.) upon the sleep of a group of eight insomniacs. They reported that the total sleep time was significantly increased and that the amount of wake time after sleep onset and the amount of total wake time were both significantly reduced in comparison with the placebo baseline, after A.S.A. administration. These changes were most marked during the first three days of A.S.A. administration. A.S.A. was reported as having no significant effect upon any of the sleep stages, REM latency, sleep onset time and the number of stage changes. The results of this study, therefore, did not support the idea put forward by the self-reported, sleep onset insomniacs of chapter one that A.S.A. aids sleep. However, the baseline group mean sleep onset time for the insomniacs of the Hauri and Silberfarb (1978) study was only 30.9 minutes; so A.S.A. may

not have been given sufficient opportunity to reveal its full sedative potential. Furthermore, no standard deviations were given in the abstract; so there is the possibility that only one or two subjects may have had a relatively long sleep latency. Nevertheless, the results of this study did indicate that A.S.A. may be a useful, non-prescription hypnotic for those insomniacs whose primary complaint is early morning or frequent nocturnal awakenings. Hauri and Silberfarb (1978) found A.S.A. to be as effective as some other prescription hypnotics, such as, chloral hydrate (Kales, Bixler, Kales & Scharf, 1977), on the variable of total wake time. The authors failed to define the type of insomnia they were investigating, that is, whether the subjects complaints were primarily a long sleep latency, early morning awakening or frequent nocturnal awakenings.

The results of the Hauri and Silberfarb (1978) study did not support those of the A.S.A. study conducted in this research programme. The reasons for this could be:

1. Hauri and Silberfarb (1978) used insomniac subjects, whereas, healthy females who were all good sleepers were used in the present study.

2. Hauri and Silberfarb (1978) examined the effects of a single dose of A.S.A. (650 mgs.) upon sleep. Although the time of drug administration was not stated, it is likely to have coincided with the subjects' retiring times, as this is the time when insomniacs would consider taking a "sleeping aid". The present study examined the effects of a normal, daily therapeutic dose of A.S.A. upon sleep, that is, 2 x 300 mg. tablets administered three times per day, at 10.00 a.m., 4.00 p.m. and 10.00 p.m., a

total daily dose of 1.8 gms.

3. A different experimental design was employed in both studies. Hauri and Silberfarb (1978) used each subject as their own placebo control. Their study lasted for 21 days, the A.S.A. being given for 14 days. The subjects slept at home from the fourth to the tenth drug nights. The present study used two groups of matched, subjects: one a placebo group, the other receiving A.S.A. under double blind conditions. The study continued for 14 consecutive laboratory nights, with no break. A.S.A. was taken for four days.

Regardless of these differences an interesting finding common to both studies was the large individual differences found in response to A.S.A. This may be due to the range in individual tolerance levels to the drug and how accustomed the subjects were to taking A.S.A. and related drugs. Also, A.S.A. is a drug with a wide variety of actions, and each subject may respond to different degrees to each A.S.A. action. Overall, the results of the Hauri and Silberfarb (1978) study indicated that A.S.A. may be a useful hypnotic for some insomniacs.

12.0 CONCLUSIONS AND FURTHER RESEARCH

The results showed that A.S.A., at a dosage of 2 x 300 mgs., three times per day, does affect sleep. The major finding was an increase in stage 2 sleep which reached statistical significance on the second drug night, in comparison with a placebo group. There was also a declining trend for stage 4 sleep during A.S.A. administration, but this did not reach statistical significance. The cumulative graph (M) revealed that on the second A.S.A. night the accumulation of stage 4 sleep was suppressed after 2½ hours of sleep, in comparison with the

placebo group. On withdrawal of A.S.A., there was a significant decline in REM latency and an increase in the night-to-night variability of stage REM and stage 3 sleep. Overall, these results were considered as being indicative of A.S.A. exerting a slight disruptive effect upon sleep.

Having found out that A.S.A. has an effect upon sleep the question now raised is "why?" Although various speculations have been made, there is little evidence for these various, possible mechanisms of A.S.A. action in humans, at the dosage employed in the present study. It was felt that some attempt ought to be made to determine a possible mechanism by which A.S.A. may be exerting its effects upon sleep. Two, general approaches to this problem were envisaged. One was a biochemical approach, with the aim of examining the effects of A.S.A. upon various neurotransmitter substances, for example 5-hydroxytryptamine and other neurochemical substances, such as the prostaglandins. 5-hydroxytryptamine is believed to be one of the major neurotransmitters involved in sleep (Mendelson et al. 1977). The prostaglandins are believed to play an important role in neurotransmission (Gross et al. 1977). The link between A.S.A. and 5-hydroxytryptamine is: if the dosage of A.S.A. used was sufficiently high to bring about the release of tryptophan, the precursor of 5-hydroxytryptamine, from its binding site, an increased 5-hydroxytryptamine concentration or turnover rate should result. Although this association appears quite straightforward when examined in isolation, when the body as a whole is considered the link is rather more complicated. Many other factors such as diet and free fatty acid concentrations can influence the ratio of free to bound tryptophan. One must be aware of these other factors which may be operating, for any assay technique for 5-hydroxytrytamine or its

metabolites can only serve as an indication of the effects of A.S.A.; it cannot necessarily show that it is A.S.A. alone which is bringing about a change in the levels of 5-hydroxytryptamine or its metabolites.

The assay techniques available were limited by the equipment and reagents at the experimenter's disposal and by the fact that it was not feasible to use a technique which involved invasive procedures. This, therefore, eliminated the possiblity of determining the concentrations of 5-hydroxytryptamine in the blood. The most suitable assay technique available at the time was the urinary analysis of 5-hydroxyindoleacetic acid, according to the method of Udenfriend, Weissbach and Brodie (1958). 5-Hydroxyindoleacetic acid (5HIAA) is the major end product of 5-hydroxytryptamine metabolism.

The other approach was to examine the effects of other analgesics upon sleep. The analgesic chosen was Paracetamol (acetaminophen), because:

1. it shares many common properties with A.S.A.. They are both effective analgesics and antipyretic agents at therapeutic doses.

2. Paracetamol has been found to inhibit prostaglandin synthesis from in vitro enzyme preparations (Flower & Vane, 1972). It has been found to be as effective as A.S.A. in the inhibition of prostaglandin synthesis in preparations from the dog, rabbit, mouse and gerbil brain, but less effective than A.S.A. against enzymes from the spleen.

3. Unlike A.S.A., Paracetamol, at normal, therapeutic doses does not release tryptophan from its binding site (Gazzard, Ford-Hutchinson,

Smith & Williams, 1973). If Paracetamol is found to have an effect upon sleep, different to that found for A.S.A., it may indicate that this pathway is involved.

4. Paracetamol has less overall toxicity and side-effects than A.S.A.: Paracetamol does not produce gastric irritation, erosion and bleeding which may occur after A.S.A. administration. Paracetamol will, therefore, serve also as a comparison to determine if the effects of A.S.A. upon sleep may simply be attributable to its adverse side-effects.

The reason for this second approach was to determine whether the effects produced by A.S.A. upon sleep are common to some related compounds or peculiar to A.S.A. An alternative choice of drug could have been to have used a drug such as indomethacin which, on a weight basis, is 23 times more potent than A.S.A. as an inhibitor of PGE_2 synthesis from an enzyme preparation from guinea pig lung (Vane, 1971). However, on ethical grounds, the use of indomethacin would not be allowed as it has severe side-effects, with complaints of headache, nausea, abdominal pain and diarrhoea (Goodman & Gilman, 1970).

The following two studies in this research programme will be pilot studies investigating:

1. the effects of a therapeutic dose of A.S.A. (2 x 300 mgs., three times per day, a total daily dose of 1.8 gms.) upon the amount of 5-hydroxyindoleacetic acid (5HIAA) excreted each day,

2. the effects of a therapeutic dose of Paracetamol (1 x 500 mgs., four times per day, a total daily dose of 2.0 gms.) upon human sleep.

Chapter Three - Part II

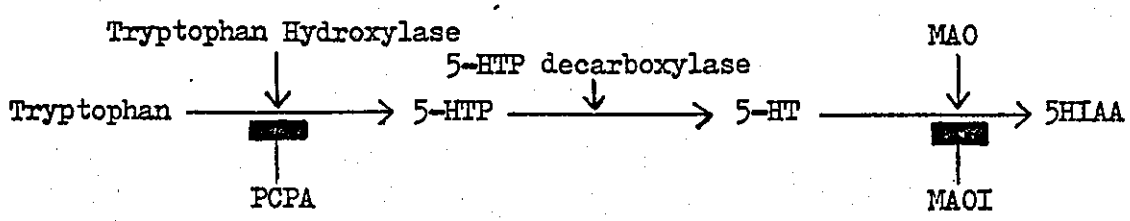
SUPPLEMENTARY PILOT STUDIES

13.0 URINE ANALYSIS OF 5-HYDROXYINDOLEACETIC ACID

14.0 INTRODUCTION

The aim of the present study was to obtain some indication as to whether A.S.A., at a dosage of 2 x 300 mgs., three times per day, was able to bring about changes in the urinary excretion of one of the metabolites of the tryptophan pathways. As considered in section 1.4 (chapter three), A.S.A. is known to compete with plasma tryptophan for its binding site to human serum albumin. However, Smith and Lakatos (1971) showed that the concentration of A.S.A. required to compete successfully with tryptophan for its binding site was high. Consideration must also be given to the fact that other factors such as diet may be operating to produce a change in the ratio of free to bound tryptophan: therefore, any changes found after A.S.A. administration does not necessarily show that A.S.A. is releasing tryptophan. The results of the assay will simply provide an overall view of the changes connected with the tryptophan pathway.

The pathway of particular interest is the formation of 5-hydroxytryptamine from tryptophan, as it is 5-hydroxytryptamine and its breakdown products which are believed to play a major rôle in sleep production and maintenance (Mendelson et al., 1977). 5-Hydroxytryptamine is oxidised to 5-hydroxyindoleacetic acid, which is excreted in the urine. The relationship between tryptophan and 5HIAA is represented below:



Key:

5-HTP - 5-Hydroxytryptophan

PCPA - Parachlorophenylalanine

MAOI - Monoamine Oxidase Inhibitor

5-HT - 5-Hydroxytryptamine (serotonin)

This tryptophan pathway is a minor one, as the bulk of tryptophan metabolised is acted upon by the enzyme tryptophan pyrrolase, eventually yielding such products as anthronilic acid, xanthurenic acid and nicotinic acid.

As mentioned in section 11.0, chapter three, the choice of assay techniques was limited by the equipment and reagents available at the time and the fact that any technique which involved invasive procedures was not feasible. In view of these points, the most suitable assay technique was the analysis of 5-HIAA in the urine by colormetric means. However, 80 to 90 per cent of the 5-HIAA excreted in the urine will be from the alimentary tract and only a small proportion from the brain areas. Therefore, urine analysis will only provide an overall view of the amount of 5-hydroxytryptamine in the body prior to, during and after A.S.A. administration, rather than giving specific details about the 5-hydroxytryptamine content in various areas. Nevertheless, it was considered that if A.S.A. was having a marked effect upon the formation of 5-HIAA, it would be detected in the urine.

15.0 EXPERIMENTAL DESIGN AND PROCEDURE

Three female subjects, who were able to take A.S.A. without experiencing any adverse side-effects were selected. The experimental

protocol was:

Day	1	2	3	4	5	6	7	8	9	10	11	12
Condition	B_1	B_2	A_1	A_2	A_3	A_4	R_1	R_2	R_3	R_4	R_5	R_6

Key:

B - Baseline

A - A.S.A. (2 x 300 mgs., three times per day)

R - Recovery

On days three to six the subjects were administered 2 x 300 mgs. of A.S.A., three times per day, at 10.00 a.m., 4.00 p.m. and 10.00 p.m., as for the previous study. The subjects were required to collect all the urine for each day. The volume of urine excreted upon awakening was combined with the urine of the previous day. The subjects were instructed to store their collecting bottles in a cool place. The total urinary output was collected each day apart from days R_2, R_4 and R_5, as it was considered that R_1, R_3 and R_6 would give sufficient indication of any recovery trends which may be present. The importance of keeping their daily dietary intake constant was stressed to the subjects.

In order to check the sensitivity of the assay technique, one subject, approximately four weeks after the study, supplemented her normal diet with two foods relatively high in tryptophan content, namely, the tomato and the banana. The approximate tryptophan contents of these foods are 9 mg. per 100 gms. and 13 mg. per 100 gms., respectively (Food and Agriculture Organisation, 1972). The subject ate 400 gms. of each food during the course of the day. The urine was collected and analysed as for the other days. The aim of this procedure was to determine whether the assay used was sufficiently sensitive to indicate the rise in dietary tryptophan.

For both parts of the study, the urine was analysed daily, according to the standard method of Udenfriend et al. (1958). This is a colormetric method: the extracted 5-HIAA yields a violet colour on interaction with nitrosonapthol reagent. After extraction and treatment with nitrosonapthol reagent, the final aqueous layer was transferred to a microcuvette, and the optical density was measured at 540 mμ. The reagent blank was prepared by treating 6 mls. of distilled water in the same manner as the 6 mls. urine sample. The ether used was washed with a dilute solution of ferrous sulphate to remove impurities. Each sample was assayed in triplicate.

The standard curve was compiled from the optical densities measured on 6 mls. of standard solutions containing 8, 14.5, 29, 44.5, 58 and 66 μg. of 5-HIAA treated in the same manner as the urine samples. The standards were assayed in duplicate. A standard was also assayed every day along with the samples as a check. The standard was stored in the refrigerator.

16.0 RESULTS

The standard curve was plotted, and the line of best fit calculated, (graph N, section 25.9, chapter three). The urinary excretion of 5-HIAA for each subject, for each day is shown in Tables 81 and 82 (section 25.0, chapter three). The 5-HIAA output is expressed both in mg. per day and per 1,000 mls. of urine, for ease of comparison with previous studies. Although N is too small for accurate statistical analysis, it can be clearly seen from Tables 81 and 82 that A.S.A. administration had little marked effect upon the urinary output of 5-HIAA. When the tryptophan load was given there was a marked increase in the amount of 5-HIAA in the urine. The 5-HIAA concentration increased from a baseline mean of

5.7 mg. per day to 17.9 mg. per day or when expressed per 1,000 mls. of urine from 3.1 to 7.0. Graph 0 (section 25.0, chapter three) has been plotted with days against the group mean 5-HIAA concentration, expressed both as mg. per day and per 1,000 mls. of urine.

17.0 DISCUSSION OF RESULTS

Firstly, mention must be made of the fact that there was variability in the optical density readings for the standard from day to day, the range was 0.15 ± 0.02. The error was thought to be due to the spectrophotometer. To allow for this error, the recorded excretion of 5-HIAA was adjusted according to the percentage error associated with the standard for that particular day. In applying this correction factor, the assumption was made that the standard had not undergone any change during the course of the study; this seems reasonable, as it was stored according to the drug company's instructions and the standard values did not simply show a progressive decline during the course of the study, but a random fluctuation. This indicated that the standard was not undergoing progressive degradation.

Tables 81 and 82 and graph 0 (section 25.0, chapter three) clearly show that there was no marked change in urinary output of 5-HIAA during A.S.A. administration. This may be because:

1. the dosage of A.S.A. used may have been insufficient to compete successfully with tryptophan for its binding site,
2. the effect of A.S.A. may be slight; more subjects or a more sensitive assay technique may be required in order to detect any change,
3. inaccuracies exist within the assay technique,

4. there is also the possibility that A.S.A. may be successfully competing with tryptophan for its binding site to human serum albumin, but another mechanism, such as a decrease in free fatty acid concentrations may be causing an increase in tryptophan binding, with the net result of no change in the free tryptophan concentration.

However, the assay technique was sufficiently sensitive to detect the increased urinary output of 5-HIAA due to the tryptophan supplemented diet. Wyatt, Engelman, Kupfer, Fram, Sjoerdsma and Snyder (1970) also found that this assay technique was sensitive to a tryptophan load (7.5 gms. per day). Wyatt et al. (1970) reported that the 24 hour urinary 5-HIAA concentration increased from a placebo mean of 6.2 mg. per day to 19.6 mg. per day. Worthy of comment is the fact that, although the amount of tryptophan administered in the diet, in the present study, was only approximately 90 mg., in comparison with the 7.5 gms. per day of the Wyatt et al. (1970) study, the increase in 5-HIAA concentration in the urine reported for both studies was almost equal. There are several reasons for this:

1. The conversion of tryptophan to 5-hydroxytryptamine and then to 5-HIAA is only a minor tryptophan pathway; there is the possibility that this pathway may quickly become saturated, resulting in the excess amounts of tryptophan passing along alternative pathways.

2. The tryptophan administered in the present study was of the order of that likely to be encountered in practice (90 mgs.). Whereas, in the Wyatt et al. (1970) study a very large tryptophan load was given, 7.5 gms. In the present study, the tryptophan was administered by means of high tryptophan foods, in the Wyatt et al. (1970) study the tryptophan was administered as a drug and it

is regarded as being very unpalatable. The extreme conditions of the Wyatt et al. (1970) study may have caused some of the bodily systems to react atypically.

3. In the Wyatt et al. (1970) study the urine was analysed over three days. In the present study the urine was only analysed on one day for the high tryptophan diet condition.

It is also interesting to note that the two graphs 0, one with 5-HIAA concentration expressed in mg. per day and the other per 1,000 mls. of urine, show slightly different trends. This may be due to the wide variation in the daily urinary volumes for each subject. For example, one subject had a range of daily volumes from 1,214 mls. to 3,065 mls. Also, with N = 3, variations in the urinary volume are likely to affect the group results. The subjects were not instructed to keep their daily fluid intake constant; this was a shortcoming in the instructions given to the subjects. The slight fluctuations in both graphs are likely to reflect normal daily changes in 5-HIAA excretion. The concentrations of 5-HIAA excreted were within the range quoted by Udenfriend et al. (1958) of 2 - 8 mg. per day.

18.0 CONCLUSIONS AND FURTHER RESEARCH

This pilot study indicated that A.S.A., at a dosage of 2 x 300 mg., administered three times daily, does not markedly effect the urinary excretion of 5-HIAA, as measured by the colormetric assay technique of Udenfriend et al. (1958). This finding suggests that the dosage of A.S.A. administered in the main study did not have a major effect upon tryptophan metabolism.

This study was essentially a pilot study, but further research in this area is not justified at present; firstly, because the fluctuations in the standard measurement from day to day throws suspicion on the reliability of this assay technique, indicating that another assay technique ought to be tried before examining the responses of more subjects to A.S.A. administration. A possible alternative assay is to use a flurometric technique (Udenfriend et al., 1958). However, the spectrophotoflurometer was under repair at the time. Secondly, this biochemical aspect of the research programme is a side-track and runs the risk of becoming divorced from the main theme.

19.0 PARACETAMOL (ACETAMINOPHEN) PILOT STUDY

20.0 INTRODUCTION

The aim of this pilot study was to determine whether a drug with similar analgesic and antipyretic activities to A.S.A. produced the same effect upon sleep. The drug chosen was Paracetamol (Acetaminophen). The reasons for choosing Paracetamol were listed in section 11.0, chapter three. In brief, the reasons were primarily that A.S.A. and Paracetamol are both effective analgesics and antipyretic agents at therapeutic doses and both drugs have been found to inhibit the synthesis of prostaglandins from in vitro enzyme preparations (Vane, 1971; Flower & Vane, 1972). They do differ in some respects: Paracetamol does not release tryptophan from its binding site to serum albumin, at therapeutic doses (Gazzard, Ford-Hutchinson, Smith & Williams, 1973). Furthermore, Paracetamol does not produce gastric irritation, erosion and bleeding that may occur after A.S.A. administration (Goodman & Gilman, 1970). Therefore, the comparison of the effects of these two drugs upon sleep, in view of their properties, both similar and dissimilar, may aid in

determining the pathway via which A.S.A. exerted its effects upon sleep.

21.0 METHOD

21.1 Experimental Design

Four female subjects were selected according to the procedure of section 6.1, chapter one. The subjects were given the same instructions as for the A.S.A. main study. The only changes being that the subjects had to be able to take Paracetamol without experiencing any adverse side-effects and the subjects were told the dosage of Paracetamol to be used. The dosage of Paracetamol administered was 1 x 500 mgs., four times per day, at 10.00 a.m., 2.00 p.m., 6.00 p.m. and 10.00 p.m., yielding a total daily dosage of 2.0 gms. This dosage was within the therapeutic range quoted by Goodman and Gilman (1970), 0.3 - 0.6 gms., every four hours.

The experimental protocol was the same as the A.S.A. main study:

Night	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Treatment	Ad	Ad	B_1	B_2	B_3	P_1	P_2	P_3	P_4	R_1	R_2	R_3	R_4	R_5	R_6

Key:

Ad - Adaptation

B - Baseline

P - Paracetamol (1 x 500 mgs., four times per day)

R - Recovery

Each subject attended the laboratory for a total of 15 nights. As for the A.S.A. study no EEG recordings were made on P_3 or R_5.

21.2 Experimental Procedure

The subjects attended the laboratory in pairs and arrived at the laboratory approximately one hour before retiring. The general laboratory procedure of section 6.3, chapter one was carried out. The retiring and rising times for each subject, on every night were kept to within ± 15 minutes. A daily check was made on the general health and well being of the subjects by means of self-report accounts.

21.3 Data Analysis

The EEG records were scored in one minute epochs, according to the standardised criteria of Rechtschaffen and Kales (1968). The records were scored blind and independently by two, experienced scorers; if the disagreement between them exceeded five per cent of the epochs, it was resolved by discussion. The stages of sleep, (W+1), 2, 3, 4, (3+4) and REM, were all expressed in minutes out of a total sleep time 400 minutes. This length of time was chosen in order that the results would be directly comparable with those of the A.S.A. study. Sleep onset time, total sleep time, REM latency and REM periodicity were also calculated. The values for each sleep parameter, over the three baseline nights, were expressed as a mean. The results are presented in Tables 83 - 92, section 25.0, chapter three.

Graphs P and Q (section 25.0, chapter three) are a plot of nights against the group mean difference score from the baseline for each sleep stage; the difference score was used so that any changes from the baseline could be clearly observed. From these graphs, it can be seen that the most pronounced changes from the baseline are an increase in stage 2 sleep and a decline in REM sleep on the fourth drug night. Inspection of results table 89 reveals that the REM latency declined

markedly on the fourth recovery night. In order to gain some indication as to whether these observed trends were significant, it was decided to compare the Paracetamol group with the placebo group from the A.S.A. study. The two groups were matched for age and sex, but the studies were conducted at different times.

There was also the problem of the two groups being of unequal size, with four subjects in the Paracetamol group and eight in the placebo group. Boneau (1960) and Havlicek and Peterson (1974) have shown that both the t and F tests are more susceptible to deviations from the underlying assumptions of the tests, such as non-normality and unequal group variances, when the group sizes are unequal. The groups which they assessed had group sizes with a ratio of approximately 1 to 5, whereas, in the present case the ratio is only 1 to 2, but there is also another problem: the Paracetamol group size is small. The shortcomings of performing statistical analysis upon such data was fully appreciated, but it was considered that it would aid in the overall interpretation of the results.

An ANOVA for repeated measures with unequal group sizes was performed according to the computational procedure described in Winer (1971, page 601). The ANOVA was performed only on those sleep stages which, from the graphs P and Q, appeared to show pronounced changes, namely, stage 2 and REM sleep over the drug nights. Inspection of the results tables 83 - 92, section 25.0, chapter three, revealed that REM latency, over the recovery nights, was the only other sleep parameter which showed any marked changes. The ANOVA summary tables for those parameters which approached statistical significance are presented in Tables 93 and 94, section 25.0, and the rest are in Appendix XVI.

22.0 RESULTS

The ANOVA revealed that the difference between the Paracetamol and placebo groups for stage 2 sleep, over the drug nights, was only significant at the ten per cent level ($F = 3.3$, df 1,10, $P \leq .1$). REM latency showed a drug by night interaction effect, over the recovery nights, also at the ten per cent level of significance ($F = 2.4$, df 4,40, $P \leq .1$). The changes observed in REM sleep did not even approach statistical significance. The null hypothesis has been rejected at the five per cent level of significance for all the other analyses conducted in this research programme. Since the results of this Paracetamol study were significant at only the ten per cent level, it is obviously not clear whether the trends which could be observed upon Paracetamol administration were pharmacologically significant. The ANOVA revealed that the results of the Paracetamol study were on the borders of statistical significance in comparison with the placebo group. More subjects will be required before the effects of Paracetamol upon sleep can be clarified, but the effects, if any, are not as great as those observed with A.S.A. administration.

From the graphs P and Q, it appeared that none of the other sleep stages were markedly effected by Paracetamol administration. Tables 90, 91 and 92 revealed that sleep onset time, total sleep time and REM periodicity showed no marked changes with Paracetamol administration.

23.0 DISCUSSION OF RESULTS

Overall, the results indicated that the effects of Paracetamol upon sleep are likened more to those of A.S.A. than placebo. The Paracetamol group findings are, in some respects, a part replication of the A.S.A. findings. Common to both drugs is the increase in stage 2 sleep

and the tendency for a slight decline in REM sleep upon drug administration. Upon comparison of the two graphs P and H, it can be seen that with Paracetamol administration stage REM returned to baseline on R_1 and and increased beyond it on R_2, but with A.S.A. stage REM continued to fluctuate below the baseline over the recovery nights. With regard to the changes in stage 2 sleep, the increase was more pronounced for the A.S.A. group. All eight subjects showed an increase in stage 2 on the second night of A.S.A. administration, but in the case of Paracetamol only one subject showed a marked increase in stage 2 on the second drug night and two subjects on the fourth drug night. Furthermore, the Paracetamol group did not show the declining trend in stage 4 sleep, which was found to be significant for the A.S.A. pilot study group, apart from on the second drug night. The A.S.A. main study group did show a declining trend in stage 4 sleep over the drug nights, reaching a minimum on the second drug night, but this was not statistically significant. As mentioned in section 11.0, chapter three, the observation that a few of the subjects in the placebo group also showed a decline in stage 4 sleep, on the second drug night, raised the possibility that the declining trend observed with A.S.A. administration may be due to other factors. The A.S.A. pilot study findings may be an artefact due to the absence of a control group.

There is also another confounding variable, which may have caused different trends to be observed with different groups of subjects, that of large individual variability in response to the drugs. This was noted for the A.S.A. group and for the Paracetamol group; for example, subject 2 on the second Paracetamol night showed a marked decline in stage 4 of 15 minutes, whereas, for the other three subjects there was little observed change in stage 4 on this particular night. With regard to

the decline in REM latency (night x drug interaction effect at $P \leq .1$), inspection of results table 89 indicated that it was the fourth recovery night which showed the marked decline in REM latency. For the A.S.A. group there was a significant decline in REM latency over all the recovery nights, when compared with the placebo.

The general, overall impression gained from the results was that Paracetamol was having a similar, but lesser effect upon sleep in comparison with A.S.A. This was reflected in the changes in stage 2, which were in the same direction for both drugs, but in the case of Paracetamol, the changes were less marked and did not peak until the fourth drug night. The trends in REM latency were similar for both drugs, but in the case of Paracetamol, the decline was most marked on the fourth recovery night. Paracetamol also appeared to be causing less disruption to stage 4 sleep. The reasons for these sleep changes may be similar to those speculated for A.S.A., that is, the increase in stage 2 sleep may be acting as a kind of buffer to compensate for the disruption in other sleep stages, such as REM sleep, in this case. Once again, a qualitative change in REM sleep was found on the recovery nights. As suggested for the A.S.A. study, this may be due to the action of some drug metabolites on the REM triggering mechanisms.

Paracetamol shares some common properties with A.S.A. Apart from its antipyretic and analgesic effects, it has been shown to inhibit the synthesis of prostaglandins from in vitro enzyme preparations, (Flower and Vane, 1972). Therefore, one may speculate that if both drugs were exerting some of their effects through the inhibition of prostaglandin synthesis, and since there is reason to believe that the prostaglandins may play some role in the sleep mechanisms, then these

drugs may have similar effects upon sleep. However, there is the distinct possibility that the drugs are acting via different pathways and the prostaglandins may not be the correct link between them.

There are slight pharmacological differences between the two drugs; for example, Gazzard et al. (1973) found that Paracetamol, at normal, therapeutic doses, did not release tryptophan from its binding site to human serum albumin. Paracetamol has less overall toxicity and side-effects than A.S.A.; Paracetamol does not produce gastric irritation, erosion and bleeding at therapeutic doses, which may occur after A.S.A. administration (Goodman & Gilman, 1970). It is unlikely that the slight differences in the effects of these two drugs upon sleep was due to the fact that Paracetamol does not bind to serum albumin, at therapeutic doses. It would appear that at the dosage of A.S.A. used in the present study, the increase in free tryptophan, if any, was small and short-lived. The results of the previous study supported this idea, because the therapeutic dose of A.S.A. used was insufficient to bring about an increase in the urinary output of 5-HIAA; 5-HIAA is a breakdown product of 5-hydroxytryptamine, which is produced from tryptophan. This is the pathway by which tryptophan is believed to exert its effects upon sleep.

Regardless of all the various actions and pathways previously considered, it must not be overlooked that the effect of these drugs upon sleep may be due to adverse side-effects. However, since Paracetamol has less adverse side-effects than A.S.A. at therapeutic doses (Goodman & Gilman, 1970) and appears to have similar effects to those of A.S.A. upon sleep, it would be reasonable to assume that the effects of these drugs upon sleep cannot be explained solely on the basis of side-effects.

There is also the possibility that the slight differences in the effects of these two drugs upon sleep may be due to their respective pharmokinetics. For example, Paracetamol is reported to reach peak plasma levels after 30 minutes - one hour after administration of a single therapeutic dose. Detectable levels of Paracetamol are reported to be present in the plasma for about five hours after a single therapeutic dose. A.S.A. has been shown to reach peak plasma levels after about two hours and then gradually decline (Goodman & Gilman, 1970).

Since both Paracetamol and A.S.A. are drugs with a wide variety of actions, it is possible that many more speculations could be made in connection with how these drugs bring about the observed changes in sleep. Support for some of these ideas previously put forward, such as the inhibition of prostaglandin synthesis, would involve entering further into the realms of biochemistry. Although an attempt was made along these lines with the urine analysis of 5-HIAA (chapter three, part II), it merely skimmed the surface of a complex field. Nevertheless, the results from the studies in this chapter revealed that A.S.A. does affect sleep. Its effect was classified as being slightly arousing, because of the significant increase in stage 2 sleep, a light stage of sleep. There are some indications from the results of the Paracetamol pilot study that it exerts a similar effect to that of A.S.A. upon sleep, but to a lesser degree. More subjects are required before the action of Paracetamol upon sleep can be clarified.

24.0 CONCLUSIONS AND FURTHER RESEARCH

Sleep, after the administration of a therapeutic dose of Paracetamol (1 x 500 mgs., four times per day), showed a trend toward an increase in stage 2 sleep and a decline in REM latency upon drug withdrawal.

These findings were similar to those observed upon A.S.A. administration. The next step would be to conduct a larger, controlled study with Paracetamol, similar to the A.S.A. main study, to determine if the results of this pilot study could be replicated.

In concluding one ought to go back and consider the reasons for investigating the effects of A.S.A. and Paracetamol upon sleep. These investigations stemmed from the comments made by many of the self-reported insomniacs of chapter one that they frequently took Aspirin to relieve their insomnia; therefore, the initial interest lay in the effects of A.S.A. upon insomnia. As a result of the literature search, it became apparent that no previous study had examined the effects of A.S.A. upon normal, human sleep. It was therefore considered that, within the time available, priority ought to be given to the investigation of the effects of A.S.A. upon normal sleep, rather than abnormal sleep. The results of the A.S.A. study indicated that it was having a slight arousing effect upon sleep, because of the significant increase in stage 2 sleep, a light stage of sleep. However, A.S.A. did not increase the sleep latency or decrease the total sleep time of the group of subjects to a significant degree. The subjects used in the A.S.A. studies were all self-reported good sleepers and therefore, one would not expect A.S.A. to be able to reveal any possible sleep inducing properties which it may possess. The findings of the daytime study by Pfeiffer et al. (1967) indicated that A.S.A. may have a slight sedative effect, but these results were not conclusive because of some of the points noted in section 1.0, chapter three. Once a table of standards has been drawn up for the EEG model of sleep onset insomnia (chapter two), the sedative potential of A.S.A. could be assessed by this method.

However, as discussed in section 11.0, of this chapter, since the completion of this research programme, the abstract of an investigation of the effects of a single dose of A.S.A. (650 mgs.) upon the sleep of a group of insomniacs has been published (Hauri & Silberfarb, 1978). Even in the light of this recent work, the further research proposal to investigate the sedative potential of A.S.A. by means of the EEG model of sleep onset insomnia is still feasible, because the group mean sleep onset time of Hauri and Silberfarb's (1978) insomniacs was only 30.9 minutes and no standard deviations were given. There is therefore the possibility that only one or two insomniacs had a long sleep onset time. One could argue that, as for the present study, A.S.A. had once again been unable to reveal its sedative potential.

Section 25.0

RESULTS TABLES AND GRAPHS FOR CHAPTER THREE

25.1 Explanatory Notes for the Results Tables

All means in the results tables are expressed to the nearest whole number.

All figures in the results tables are expressed in minutes unless otherwise stated.

Key:

A.S.A. - Acetylsalicylic Acid (Aspirin), 2 x 300 mg., three times per day.

A.S.T. - Analysed Sleep Time refers to the amount of each sleep record analysed from the time of sleep onset. A.S.T. is the lowest common measure of total sleep time for all subjects on all nights.

B - The average score for the three baseline nights.

B_x - The score on the xth baseline night.

P - Paracetamol, 1 x 500 mg., four times per day.

R - Recovery nights.

T - Diet supplemented with foods relatively high in tryptophan content.

Number of Stage Changes - A stage change was counted each time a subject passed from one stage of sleep into another providing the time spent in any particular stage was at least two minutes.

In the GENSTAT printout group 1 is the A.S.A. group and group 2 is the placebo group.

Results Tables for A.S.A. Pilot Study: Subjects 1 - 4 are males and subjects 5 - 8 are females.

Stage W+1 — Table 55

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	16	20	28	25	21
2	22	24	20	24	17
3	37	25	21	29	29
4	11	21	15	17	11
5	25	10	15	27	39
6	9	37	2	24	36
7	14	17	19	3	24
8	7	42	15	16	36
Mean	18	25	17	21	27
S.D.	9.9	10.4	7.4	8.4	10

Stage 2 — Table 56

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	190	199	190	180	177
2	166	167	159	179	202
3	148	170	171	184	173
4	177	167	207	184	177
5	197	192	197	203	215
6	165	173	160	200	165
7	185	193	214	191	187
8	163	168	167	169	167
Mean	174	179	183	186	182
S.D.	16.3	13.6	21.7	11.3	17.5

Stage 3 — Table 57

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	39	34	24	67	63
2	40	49	39	36	36
3	47	48	42	31	56
4	30	36	27	27	30
5	40	34	19	24	16
6	62	55	80	16	59
7	31	28	19	23	33
8	54	28	35	57	29
Mean	43	39	36	35	40
S.D.	11	10.3	20	17.8	16.9

A.S.T. for the above three tables is equal to 420 minutes.

For statistical analysis the above scores were expressed as a change from the baseline.

Stage 3+4 — Table 58

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	124	118	130	134	160
2	120	115	123	102	93
3	151	141	145	110	148
4	129	131	136	135	129
5	113	117	112	91	82
6	161	137	172	94	150
7	143	129	129	124	119
8	151	106	128	134	94
Mean	137	124	134	115	122
S.D.	17.3	12.1	17.9	18.6	29.7

Stage REM — Table 59

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	90	83	72	81	62
2	112	114	118	115	108
3	84	84	83	97	70
4	103	101	62	84	103
5	85	101	96	99	84
6	85	73	86	102	69
7	78	81	58	102	90
8	99	104	110	101	123
Mean	92	93	86	98	89
S.D.	11.5	14.1	21.6	10.8	21.5

REM Latency — Table 60

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	159	119	212	132	77
2	60	50	52	136	55
3	80	121	161	85	83
4	87	143	163	73	70
5	94	55	116	157	55
6	101	139	52	73	76
7	138	114	129	62	130
8	123	73	79	86	210
Mean	105	102	121	101	95
S.D.	32.6	37	57.3	35.6	52.2

A.S.T. for the above three tables is equal to 420 minutes.

For statistical analysis the above scores were expressed as a change from the baseline.

Sleep Onset Time — Table 61

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	14	12	11	16	7
2	22	18	30	51	20
3	25	16	16	14	12
4	13	12	10	8	10
5	18	15	13	19	32
6	10	18	20	26	18
7	27	45	20	34	30
8	35	39	23	27	33
Mean	21	22	18	24	20
S.D.	8.4	12.7	6.7	13.6	10.3

Total Sleep Time — Table 62

Treatment / Subject	B	A_1	A_2	R_1	R_2
1	426	422	420	424	430
2	455	453	447	421	449
3	423	447	429	455	454
4	453	432	458	420	460
5	427	428	441	435	426
6	527	520	503	472	507
7	424	426	421	435	420
8	505	529	538	535	513
Mean	455	457	457	450	457
S.D.	40.2	43.0	42.2	38.9	35.4

For statistical analysis the scores were expressed as a change from the baseline.

Stage 4 — Table 63

Treatment	B	A_1	A_2	R_1	R_2
Subject					
1	85	84	106	67	97
2	80	66	84	66	57
3	104	93	103	79	92
4	99	95	109	108	99
5	73	83	93	67	66
6	99	82	92	78	91
7	112	101	110	101	86
8	97	78	93	77	65
Mean	94	85	99	80	82
S.D.	13.1	11.0	9.5	15.9	16.4

A.S.T. for the above table is equal to 420 minutes

Table 63A

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	2029	4	507.0	7.9	5%	1,7
Subject	4521	7	645.9	10.1	5%	1,4
Residual	1805	28	64.5			
Total	8355	39				

Conservative Test

Newman-Keuls Multiple Comparison

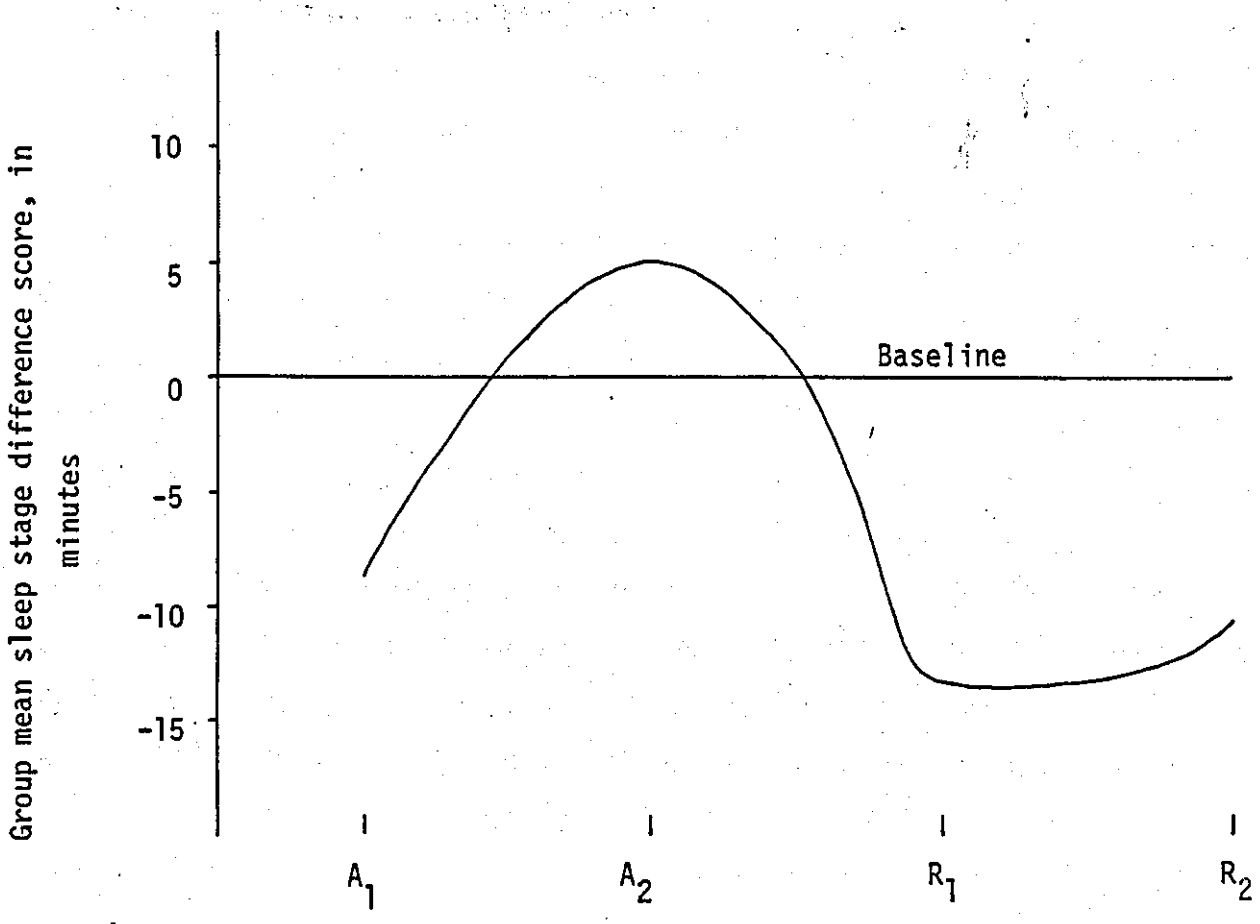
	R_1	R_2	A_1	B	A_2
R_1		ns	ns	1%	1%
R_2			ns	1%	1%
A_1				5%	1%
B					ns

For statistical analysis the scores were expressed as a change from the baseline.

Trend Test for Stage 4 sleep (A.S.A. Pilot Study)

Table 63B

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	1703.8	3	567.9	9.1	0.1%	3,21
Linear	342.2	1	342.2	5.5	5%	1,21
Quadratic	300.1	1	300.1	4.8	5%	1,21
Cubic	1060.9	1	1060.9	16.9	0.1%	1,21
Other	0.6	0				
Subject	2441.0	7	348.7	5.6	0.1%	7,21
Residual	1316.7	21	62.7			
Total	5461.5	31				



Graph G: To Illustrate the Cubic Trend for Stage 4 Sleep

Results Tables for the A.S.A. Main Study (Subjects numbered 1 - 8 are the A.S.A. group and subjects 9 - 16 are the Placebo Group.

Stage W+1 — Table 64

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	17	5	5	14	1	20	20	1	13
2	17	6	15	13	9	15	27	12	12
3	6	15	29	39	9	11	12	9	9
4	18	12	15	16	10	8	17	22	8
5	25	14	22	8	50	10	8	10	19
6	13	18	11	12	12	11	9	9	8
7	9	30	5	7	9	15	5	16	14
8	17	21	5	12	13	23	8	10	11
Mean	15	15	13	15	14	14	13	11	12
S.D.	5.8	8.1	8.8	10.1	14.9	5.2	7.5	6.1	3.7
9	4	12	19	7	8	12	18	13	10
10	33	24	44	16	12	36	19	22	18
11	28	42	10	27	22	33	43	33	28
12	21	29	21	38	29	32	11	22	30
13	16	20	23	23	27	12	16	14	14
14	3	2	3	0	7	1	7	9	4
15	14	13	13	1	7	12	22	13	9
16	7	8	26	3	4	12	10	7	10
Mean	16	19	20	14	15	19	18	17	15
S.D.	11.1	12.8	12.3	14	10	12.9	11.2	8.9	9.3

Stage 2 — Table 65

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	198	209	212	216	192	160	173	164	197
2	147	166	188	171	140	136	141	151	177
3	176	170	185	151	169	194	141	169	176
4	156	161	182	185	173	163	163	210	146
5	203	228	219	210	197	224	242	226	199
6	176	192	196	195	166	197	204	175	169
7	204	206	218	209	196	195	174	214	197
8	190	206	231	223	241	223	220	208	227
Mean	181	192	204	195	184	187	182	190	186
S.D.	21.4	24.2	18.4	24.7	29.8	31.1	36.6	27.9	24.4
9	186	187	163	185	191	191	180	187	191
10	184	181	152	192	194	185	177	184	190
11	164	161	197	156	159	158	162	160	158
12	175	176	168	166	161	159	194	171	160
13	171	149	168	164	200	166	172	188	160
14	192	177	180	169	189	187	167	191	167
15	185	164	199	204	180	199	178	182	157
16	130	139	139	128	128	156	115	154	158
Mean	173	167	171	171	175	175	168	177	168
S.D.	19.8	16.6	20.7	23.5	24.3	17.2	23.5	13.9	14.5

A.S.T. for the above tables is equal to 400 minutes.

For statistical analysis the above scores were expressed as a change from the baseline.

For statistical analysis the scores below were expressed as a change from the baseline.

Stage 3 — Table 66

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	22	17	33	23	20	29	16	26	21
2	27	23	24	27	44	36	12	21	30
3	27	18	27	18	21	19	37	26	23
4	27	27	49	31	36	40	38	31	33
5	42	40	24	55	35	48	42	22	38
6	31	22	23	29	42	45	24	39	21
7	34	27	41	21	31	19	28	38	28
8	24	20	38	12	8	26	8	8	26
Mean	29	24	32	27	30	33	26	26	28
S.D.	6.4	7.4	9.6	12.9	12.3	11.2	12.8	10	6
9	25	18	16	22	25	12	17	18	19
10	24	35	29	25	19	33	25	26	26
11	19	17	30	29	28	28	24	27	28
12	27	19	27	24	15	21	20	19	18
13	25	19	22	19	26	33	24	30	26
14	34	48	40	51	29	33	32	32	32
15	17	17	17	12	26	18	18	26	20
16	30	31	31	36	31	23	28	29	26
Mean	25	26	27	27	25	25	24	26	24
S.D.	5.5	11.4	7.9	11.9	5.3	7.9	5.1	5	5

Stage 4 — Table 67

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	79	78	80	69	75	102	110	116	89
2	93	72	42	65	84	97	105	94	83
3	65	61	53	74	70	70	71	47	66
4	106	97	84	102	110	102	98	88	131
5	42	20	35	33	36	32	35	37	44
6	60	66	63	52	51	37	30	83	38
7	61	62	56	47	73	75	90	64	52
8	50	54	28	43	53	41	59	64	44
Mean	70	64	55	61	69	70	75	74	68
S.D.	21.7	22	20.1	21.8	22.8	29.7	31.1	26	31.5
9	84	80	108	93	104	99	99	101	101
10	61	50	74	64	69	42	75	62	55
11	79	73	60	75	87	80	65	77	84
12	90	83	76	60	91	73	75	80	82
13	76	91	79	81	37	78	74	64	88
14	94	80	93	103	86	108	104	92	132
15	83	101	80	84	81	82	73	83	93
16	116	109	85	105	114	98	108	104	110
Mean	85	83	82	83	84	83	84	83	93
S.D.	15.9	18	14.2	16.7	23.3	20.4	16.7	15.6	22.5

A.S.T. for the above tables is equal to 400 minutes.

For statistical analysis the scores below were expressed as a change from the baseline.

Stage 3+4 — Table 68

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	101	95	113	92	95	131	126	142	110
2	120	95	66	92	128	133	117	115	113
3	92	79	80	92	91	89	108	73	89
4	133	124	133	133	146	142	136	119	164
5	84	60	59	88	71	80	77	59	82
6	91	88	86	81	93	82	54	122	59
7	95	89	97	68	104	94	118	102	80
8	74	74	66	55	61	67	67	72	70
Mean	99	88	88	88	99	102	100	101	96
S.D.	19.5	18.7	25.7	22.7	27.8	28.6	30.2	29.4	33.1
9	109	98	124	115	129	111	116	119	120
10	85	85	103	89	87	75	100	88	81
11	98	90	90	104	115	108	89	104	112
12	117	102	103	84	106	94	95	99	100
13	101	110	101	100	63	111	98	94	114
14	128	128	133	154	115	141	136	124	164
15	100	118	97	96	107	100	91	109	113
16	146	140	116	141	145	121	136	133	136
Mean	110	109	108	110	108	108	108	109	118
S.D.	19.4	18.9	14.6	25	25	19.4	19.3	15.5	24.5

Stage REM — Table 69

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	84	91	70	78	112	89	81	93	80
2	116	133	131	124	123	116	115	122	98
3	126	136	106	118	131	106	139	149	126
4	93	103	70	66	71	87	84	49	82
5	88	98	100	94	82	86	73	105	100
6	120	102	107	112	129	110	133	94	164
7	92	75	80	116	91	96	103	68	109
8	119	99	98	110	85	87	105	110	92
Mean	105	105	95	102	103	97	104	99	106
S.D.	17	20.5	20.9	20.8	23.5	11.9	24.1	31	27.6
9	101	103	94	93	72	86	86	81	79
10	98	110	101	103	106	104	104	106	111
11	110	107	103	111	104	101	106	103	102
12	87	93	108	113	105	115	100	108	110
13	112	121	108	113	110	111	113	104	112
14	77	93	84	77	89	71	90	76	65
15	101	105	91	99	106	89	109	96	121
16	117	113	119	128	123	111	139	106	96
Mean	100	106	101	105	102	99	106	98	100
S.D.	13.3	9.5	11.1	15.4	15.2	15.2	16.2	12.3	18.9

A.S.T. for the above tables is equal to 400 minutes.

For statistical analysis the scores below were expressed as a change from the baseline.

REM Latency

Table 70

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	147	123	153	130	143	80	167	165	141
2	89	67	51	42	63	67	67	59	51
3	70	67	73	78	77	64	68	68	62
4	116	121	118	133	132	74	110	140	149
5	129	59	61	129	227	148	159	124	66
6	131	124	134	158	51	38	142	130	28
7	157	99	143	152	80	76	143	163	128
8	126	108	111	107	141	100	112	131	131
Mean	121	96	106	116	114	81	121	123	95
S.D.	28.8	27.6	39	39.1	58.3	32.2	38.6	39.4	47.5
9	123	123	70	130	171	129	50	117	167
10	51	59	136	48	34	40	140	71	93
11	47	46	89	39	56	49	38	48	72
12	100	150	76	77	90	147	182	139	102
13	50	42	55	66	39	57	47	41	57
14	120	78	93	132	125	158	160	182	128
15	82	62	133	161	62	87	127	78	62
16	59	55	53	52	50	48	60	41	55
Mean	79	77	88	88	78	89	101	90	92
S.D.	31.9	39	31.9	46.2	47.8	48.5	57.8	51.6	39.5

REM Periodicity

Table 71

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	91	101	129	155	89	100	66	171	109
2	102	117	84	113	91	86	88	113	96
3	103	87	122	141	127	80	107	109	90
4	101	84	58	89	125	77	128	114	105
5	105	78	86	103	114	108	93	121	75
6	89	88	85	58	98	78	94	83	88
7	97	86	99	109	99	71	105	81	109
8	90	76	86	94	88	84	89	88	81
Mean	97	90	94	108	104	86	96	110	94
S.D.	6.4	13.4	22.8	30.3	16	12.5	17.9	29.1	12.9
9	69	69	103	81	86	119	90	98	90
10	86	98	64	107	92	81	79	84	81
11	78	70	73	82	78	90	70	79	80
12	83	94	97	87	92	113	54	103	100
13	94	104	115	100	95	102	103	94	102
14	93	102	90	81	80	80	96	102	94
15	97	89	92	108	109	102	117	109	101
16	90	84	67	81	89	90	92	88	80
Mean	86	89	88	91	90	97	88	95	91
S.D.	9.3	13.6	18.1	12.1	9.7	14.3	19.7	10.3	9.7

A.S.T. for the above tables is equal to 400 minutes.

For statistical analysis the scores below were expressed as a change from the baseline.

Sleep Onset Time

Table 72

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	17	16	13	13	27	12	11	9	13
2	11	18	20	15	27	8	13	32	16
3	19	25	9	15	9	12	22	28	12
4	30	10	10	12	30	9	14	10	11
5	18	30	19	10	18	16	14	13	14
6	12	10	16	18	20	27	38	19	37
7	11	6	23	13	2	10	6	14	9
8	51	34	29	41	31	40	12	19	22
Mean	21	19	17	17	21	17	16	19	17
S.D.	13.6	10.2	6.8	9.9	10.5	11.2	9.8	8.3	9.1
9	60	67	65	47	40	42	45	42	47
10	26	62	26	30	29	46	13	29	20
11	32	52	31	37	37	42	43	47	34
12	36	74	29	42	30	43	37	34	44
13	20	63	20	11	25	15	15	12	14
14	30	32	28	19	23	16	39	8	15
15	12	15	18	10	15	12	24	28	13
16	39	66	15	17	28	22	28	21	22
Mean	32	54	29	27	28	30	31	28	26
S.D.	14.2	20.2	15.6	14.3	7.9	14.7	12.4	13.6	13.7

Total Sleep Time

Table 73

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	403	456	415	415	410	405	421	413	417
2	400	400	401	405	400	402	400	402	403
3	431	419	419	417	419	420	419	418	417
4	401	455	417	413	400	415	420	421	418
5	404	404	405	404	410	404	427	404	405
6	420	422	411	407	402	402	447	401	403
7	443	421	400	434	423	477	406	423	406
8	411	429	400	400	402	400	400	404	406
Mean	414	426	409	412	408	415	418	411	409
S.D.	15.9	20.7	8	10.7	8.9	25.9	15.7	9.1	7
9	401	442	440	437	416	400	427	415	401
10	434	448	442	446	412	419	473	434	444
11	463	470	458	464	439	468	481	463	469
12	450	444	435	443	427	413	450	430	438
13	436	454	434	453	415	449	443	442	452
14	426	432	427	446	449	426	441	446	451
15	444	443	438	450	424	443	420	425	452
16	421	452	439	446	410	432	431	434	443
Mean	434	448	439	448	424	431	446	436	444
S.D.	19	11.1	8.9	8	13.9	21.7	21.6	14.5	19.6

Number of Stage Changes

Table 74

Night Subject	B	A_1	A_2	A_4	R_1	R_2	R_3	R_4	R_6
1	48	38	45	41	42	47	38	37	40
2	49	46	44	45	56	59	49	48	58
3	38	47	52	50	34	34	53	34	33
4	50	43	61	56	60	65	61	63	44
5	71	76	60	72	76	54	54	56	74
6	55	54	57	48	51	74	45	51	45
7	60	71	52	50	47	42	54	72	49
8	47	57	47	36	35	69	30	32	43
Mean	52	54	52	50	50	56	48	49	48
S.D.	9.8	13.5	6.6	10.9	14.0	13.9	10.0	14.3	12.6
9	33	41	37	35	43	34	37	36	38
10	42	52	41	56	41	57	50	46	49
11	52	44	41	54	54	53	53	46	52
12	61	57	62	42	47	60	47	65	55
13	64	52	57	59	64	69	61	64	59
14	50	63	48	57	52	51	58	53	53
15	41	43	37	30	51	36	39	51	41
16	62	61	75	53	60	49	45	49	47
Mean	51	52	50	48	52	51	49	51	49
S.D.	11.3	8.4	13.7	11.1	7.9	11.7	8.5	9.6	7.1

A.S.T. for the above table is equal to 400 minutes.

For statistical analysis the scores above were expressed as a change from the baseline.

Significant ANOVA Summary Tables for the A.S.A. Main Study

Stage 2 - Drug Nights — Table 75

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	4720.4	1	4720.4	14.5	1%	1,14
Subjects within treatments	4544.8	14	324.6			
Within Subjects						
Nights	493.0	2	246.5	1.5	ns	1,14
Nights x Treatments	178.6	2	89.3	0.5	ns	1,14
Residual	4672.4	28	166.8			
Total	14609.2	47				

Conservative Test

Total Sleep Time - Drug Nights — Table 76

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	602.1	1	602.1	0.9	ns	1,14
Subjects within treatments	9762.5	14	697.3			
Within Subjects						
Nights	1830.9	2	915.4	9.4	1%	1,14
Nights x Treatments	350.3	2	175.2	1.8	ns	1,14
Residual	2735.5	28	97.7			
Total	15281.3	47				

Conservative Test.

Newman-Keuls Multiple Comparison Test

	A_2	A_4	A_1
A_2		5%	1%
A_4			ns

Sleep Onset Time - Drug Nights

Table 77

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	892.7	1	892.7	3.5	ns	1,14
Subjects within treatments	3556.5	14	254.0			
Within Subjects						
Nights	2005.8	2	1002.9	10.8	1%	1,14
Nights x Treatments	1620.1	2	810.1	8.7	1%	1,14
Residual	2609.4	28	93.2			
Total	10684.5	47				

Conservative Test

Newman-Keuls Multiple Comparison Test

	A_4	A_2	A_1
A_4		ns	1%
A_2			1%

Logarithmic Transformation of Sleep Onset Times - Drug Nights

Table 78

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	0.5	1	0.5	0.9	ns	1,14
Subjects within treatments	8.7	14	0.6			
Within Subjects						
Nights	1.8	2	0.9	6.4	2.5%	1,14
Nights x Treatments	1.5	2	0.7	5.3	5.0%	1,14
Residual	4.0	28	0.1			
Total	16.5	47				

Conservative Test

REM Latency – Recovery Nights

Table 79

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	11472.1	1	11472.1	5.5	5%	1,14
Subjects within treatment	29332.8	14	2095.2			
Within Subjects						
Nights	7162.6	4	1790.7	1.7	ns	1,19
Nights x Treatments	5425.6	4	1356.4	1.3	ns	1,19
Residual	57813.0	56	1032.4			
Total	111206.1	79				

Conservative Test

Homogeneity of Error Variances

Table 80

Stage W+1

1. Subjects within groups

a) Drug nights	$F_{max} = 4.2$	df 2,7	ns
b) Recovery nights	$F_{max} = 1.9$	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	$F_{max} = 1.3$	df 2,14	ns
b) Recovery nights	$F_{max} = 1.8$	df 2,28	ns

Stage 2

1. Subjects within groups

a) Drug nights	$F_{max} = 1.6$	df 2,7	ns
b) Recovery nights	$F_{max} = 4.7$	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	$F_{max} = 3.0$	df 2,14	ns
b) Recovery nights	$F_{max} = 1.9$	df 2,28	ns

Stage 3

1. Subjects within groups

a) Drug nights	$F_{max} = 1.5$	df 2,7	ns
b) Recovery nights	$F_{max} = 1.0$	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	$F_{max} = 5.9$	df 2,14	Significant at 1% level
b) Recovery nights	$F_{max} = 5.9$	df 2,28	Significant at 1% level

Stage 4

1. Subjects within groups

a) Drug nights	$F_{max} = 1.0$	df 2,7	ns
b) Recovery nights	$F_{max} = 1.2$	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	$F_{max} = 1.6$	df 2,14	ns
b) Recovery nights	$F_{max} = 1.5$	df 2,28	ns

Stage 3+4

1. Subjects within groups

a) Drug nights	F_{max} = 1.0	df 2,7	ns
b) Recovery nights	F_{max} = 1.0	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	F_{max} = 1.1	df 2,14	ns
b) Recovery nights	F_{max} = 1.8	df 2,28	ns

Stage REM

1. Subjects within groups

a) Drug nights	F_{max} = 3.0	df 2,7	ns
b) Recovery nights	F_{max} = 1.0	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	F_{max} = 3.4	df 2,14	Significant at 5% level
b) Recovery nights	F_{max} = 3.4	df 2,28	Significant at 1% level

REM Latency

1. Subjects within groups

a) Drug nights	F_{max} = 1.4	df 2,7	ns
b) Recovery nights	F_{max} = 2.0	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	F_{max} = 3.8	df 2,14	Significant at 5% level
b) Recovery nights	F_{max} = 1.1	df 2,28	ns

REM Periodicity

1. Subjects within groups

a) Drug nights	F_{max} = 4.3	df 2,7	ns
b) Recovery nights	F_{max} = 1.7	df 2,7	ns

2. Nights x Subjects within groups

a) Drug nights	F_{max} = 1.6	df 2,14	ns
b) Recovery nights	F_{max} = 2.8	df 2,28	Significant at 1% level

Sleep Onset Time

1. Subjects within groups

a)	Drug nights	$F_{max} = 1.1$	df 2,7	ns
b)	Recovery nights	$F_{max} = 1.8$	df 2,7	ns

2. Nights x Subjects within groups

a)	Drug nights	$F_{max} = 3.4$	df 2,14	Significant at 5% level
b)	Recovery nights	$F_{max} = 1.2$	df 2,28	ns

Total Sleep Time

1. Subjects within groups

a)	Drug nights	$F_{max} = 2.2$	df 2,7	ns
b)	Recovery nights	$F_{max} = 1.1$	df 2,7	ns

2. Nights x Subjects within groups

a)	Drug nights	$F_{max} = 2.6$	df 2,14	ns
b)	Recovery nights	$F_{max} = 1.8$	df 2,28	ns

Results Tables for Urinary Analysis of 5-HIAA Study

Urinary Excretion of 5-Hydroxyindoleacetic Acid (5HIAA) in mg. per day

Table 81

Day / Subject	B_1	B_2	A_1	A_2	A_3	A_4	R_1	R_3	R_6	T
1	6.3	5	4.2	7.1	7	7	7.4	5.3	3.5	17.9
2	5.4	4.8	4.8	3.7	5.9	7 *	5.8*	5.5	5.9	
3	4	4.4	5	5	4.5	1.7	1.5	4	5.3	
Mean	5.2	4.7	4.7	5.3	5.8	5.2	4.9	5	4.9	
S.D.	1.2	.3	.4	1.7	1.3	3.1	3.1	.8	1.3	

Urinary Excretion of 5-Hydroxyindoleacetic Acid (5HIAA) per 1,000mls urine

Table 82

Day / Subject	B_1	B_2	A_1	A_2	A_3	A_4	R_1	R_3	R_6	T
1	3.4	2.8	3.5	2.5	3	2.3	3.3	2.4	1.3	7
2	3.9	4.2	4.3	4.5	3.4	2.1	2.6	3	5.3	
3	3.3	3.4	6	5.7	5.6	2.9	3.2	7.5	5.6	
Mean	3.5	3.5	4.6	4.2	4	2.4	3	4.3	4.1	
S.D.	.3	.7	1.3	1.6	1.4	.4	.4	2.8	2.4	

* Suspect as to whether total volume of urine voided was collected on these two days.

Results Tables for the Paracetamol Pilot Study

Stage W+1 — Table 83

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	13	30	31	19	25	29	15	33	24
2	22	11	21	14	9	17	31	11	14
3	10	8	7	27	7	13	4	4	19
4	10	8	2	8	2	7	10	3	9
Mean	14	14	15	17	11	17	15	13	17
S.D.	5.7	10.6	13.2	8	9.9	9.3	11.6	14	6.5

Stage 2 — Table 84

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	202	194	190	204	194	192	175	164	184
2	200	188	196	219	202	197	188	229	221
3	209	225	238	211	203	198	198	204	210
4	185	202	188	208	216	198	216	189	182
Mean	199	202	203	211	204	196	194	197	199
S.D.	10.1	16.2	23.6	6.4	9.1	2.9	17.3	27.2	19.3

Stage 3 — Table 85

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	25	30	22	50	25	18	27	14	23
2	29	33	22	30	18	25	26	20	18
3	34	23	31	17	32	33	28	36	30
4	38	33	46	30	42	30	19	43	33
Mean	32	30	30	32	29	27	25	28	26
S.D.	5.7	4.7	11.3	13.6	10.2	6.6	4.1	13.5	6.8

Stage 4 — Table 86

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	54	50	55	45	57	54	68	77	56
2	82	74	67	72	86	76	72	69	77
3	68	70	70	79	77	63	83	86	65
4	77	66	79	68	69	79	73	76	85
Mean	70	65	68	66	72	68	74	77	71
S.D.	12.3	10.5	9.9	14.7	12.3	11.6	6.4	7	12.8

A.S.T. for the above tables is equal to 400 minutes.

For statistical analysis the scores above were expressed as a change from the baseline.

Stage 3+4 — Table 87

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	79	80	77	95	82	72	95	91	79
2	110	107	89	102	104	101	98	89	95
3	102	93	101	96	109	96	111	122	95
4	115	99	125	98	111	109	92	119	118
Mean	102	95	98	98	102	95	99	105	97
S.D.	15.9	11.4	20.5	3.1	13.3	15.9	8.4	17.7	16

Stage REM — Table 88

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	107	96	102	82	99	107	115	112	113
2	74	94	94	65	85	85	83	71	70
3	65	74	54	66	81	93	87	70	76
4	91	91	85	86	71	86	82	89	91
Mean	84	89	84	75	84	93	92	86	88
S.D.	18.6	10	21	10.8	11.6	10.1	15.6	19.7	19.1

REM Latency — Table 89

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	86	119	100	125	117	51	46	50	55
2	96	107	102	242	157	116	104	53	120
3	99	123	123	52	135	171	127	46	130
4	133	54	143	70	60	126	129	60	54
Mean	104	101	117	122	117	116	102	52	90
S.D.	20.4	31.9	20.2	85.7	41.5	49.5	38.7	5.9	40.9

REM Periodicity — Table 90

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	88	84	75	91	59	80	90	76	85
2	101	78	85	96	103	89	94	94	98
3	91	95	94	95	109	99	78	89	90
4	96	102	92	98	78	90	109	83	82
Mean	94	90	86	95	87	90	93	85	89
S.D.	5.7	10.6	8.7	3	22.8	7.9	12.5	7.9	7

A.S.T. for the above tables is equal to 400 minutes.

For statistical analysis the scores above were expressed as a change from the baseline.

Sleep Onset Time — Table 91

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	8	10	6	3	9	9	15	6	5
2	30	9	6	22	15	18	25	24	9
3	15	26	17	30	10	18	13	9	13
4	5	9	10	18	13	16	7	17	9
Mean	15	14	10	18	12	15	15	14	9
S.D.	11.2	8.3	5.2	11.3	2.8	4.3	7.5	8.1	3.3

Total Sleep Time — Table 92

Night / Subject	B	P_1	P_2	P_4	R_1	R_2	R_3	R_4	R_6
1	426	430	428	431	422	422	428	440	440
2	419	430	440	421	414	426	415	432	451
3	460	446	465	444	458	450	468	467	446
4	470	460	471	455	464	451	474	433	449
Mean	444	442	451	438	440	437	446	443	447
S.D.	25	14.5	20.4	14.9	25.2	15.4	29.2	16.4	4.8

For statistical analysis the scores above were expressed as a change from the baseline.

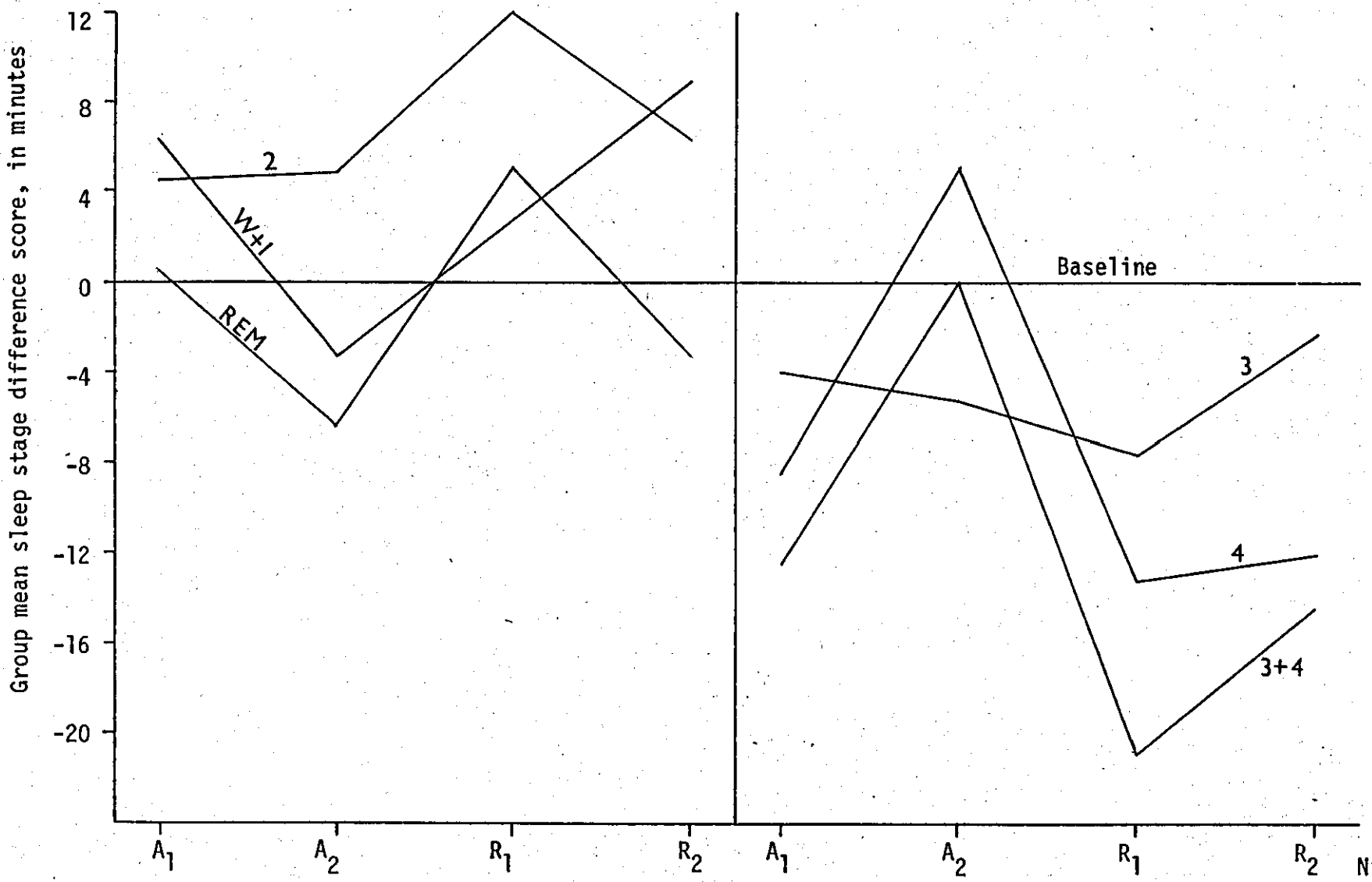
ANOVA Summary Tables for the Paracetamol Pilot Study

Stage 2 (drug nights) — Table 93

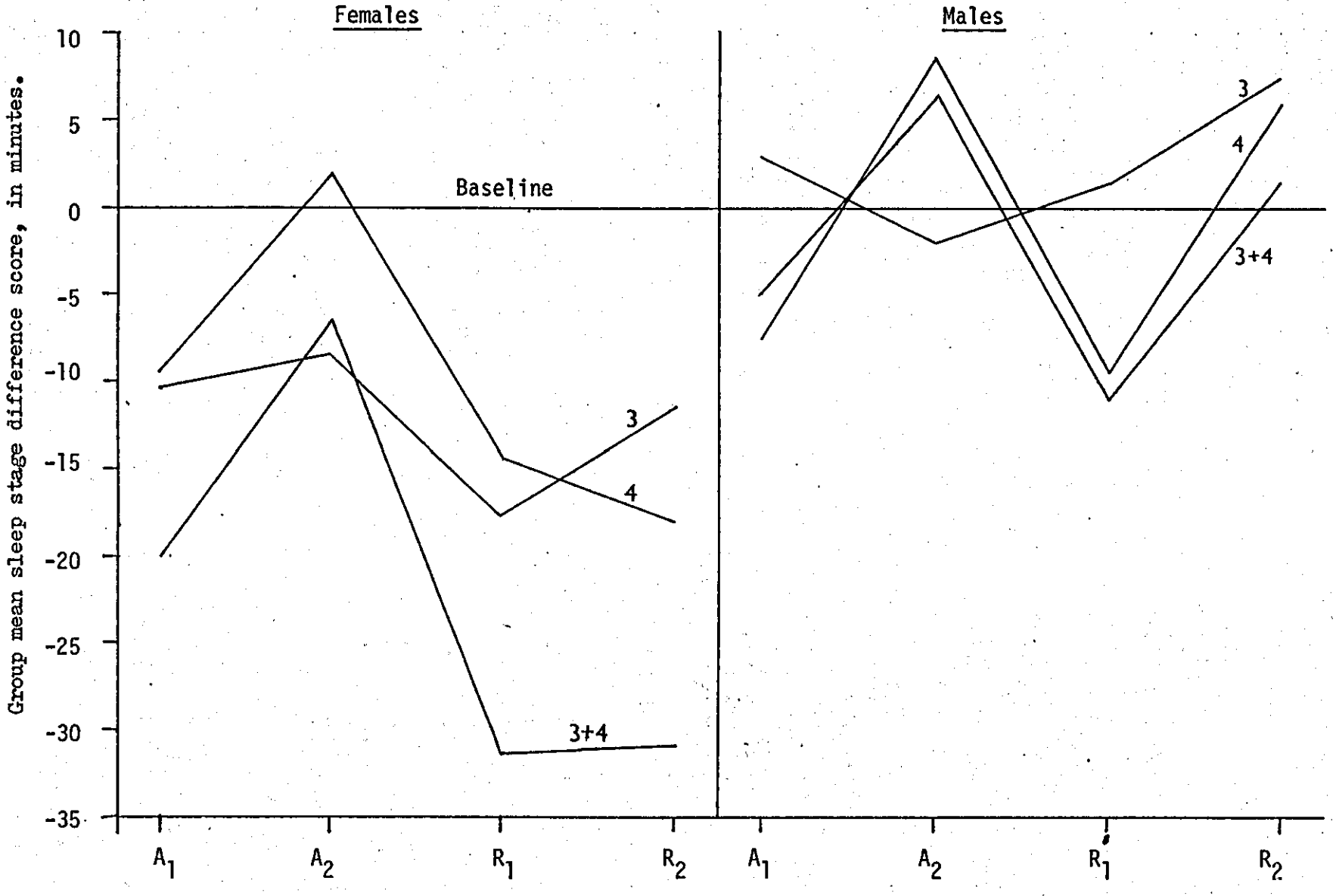
Source	SS	d.f.	MS	F	P	df_1, df_2
Between Subjects						
Treatment	847.6	1	847.6	3.3	10%	1,10
Subjects within treatments	2566.5	10	256.7			
Within Subjects						
Nights	194.7	2	97.3	0.4	ns	2,20
Nights x Treatments	80.8	2	40.4	0.2	ns	2,20
Residual	4511.8	20	225.6			

REM Latency (recovery nights) — Table 94

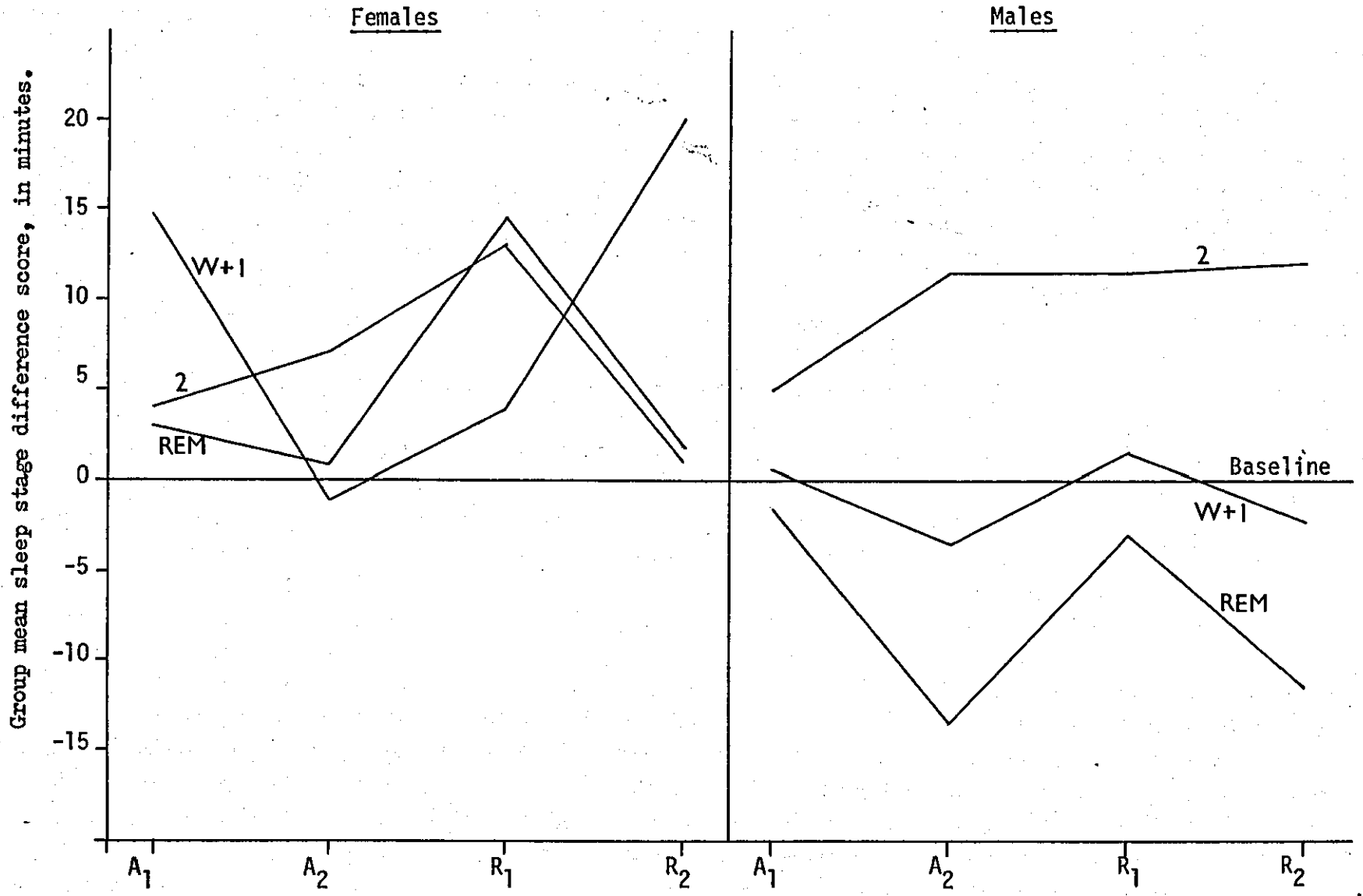
Source	SS	d.f.	MS	F	P	df_1,df_2
Between Subjects						
Treatment	4805.5	1	4805.5	2.0	ns	1,10
Subjects within treatments	24452.3	10	2445.2			
Within Subjects						
Nights	7124.8	4	1781.2	1.8	ns	4,40
Nights x Treatments	9304.6	4	2326.2	2.4	10%	4,40
Residual	38900.7	40	972.5			



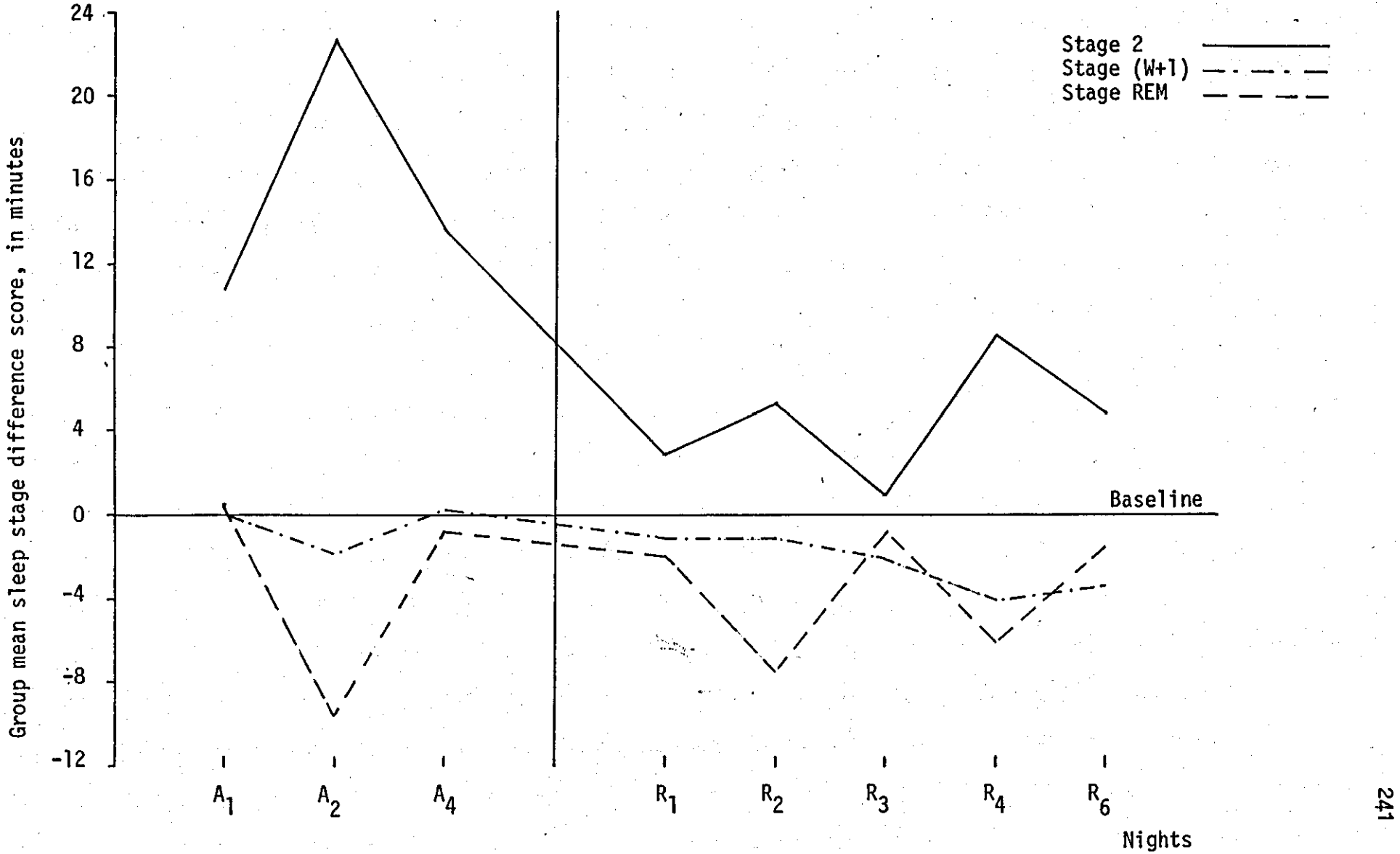
Graph D: A.S.A. Pilot Study (Whole Group)



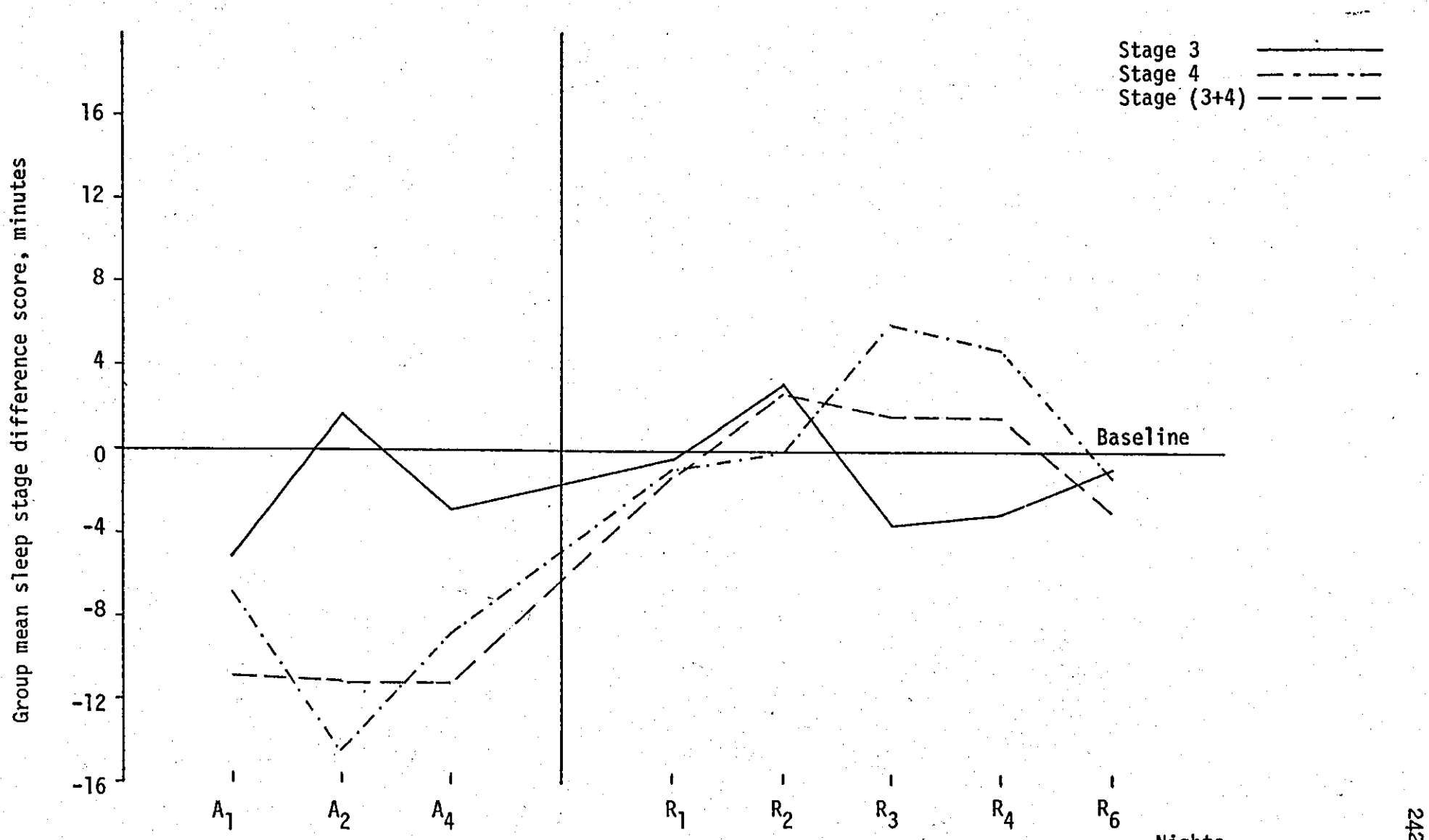
Group E: A.S.A. Pilot Study



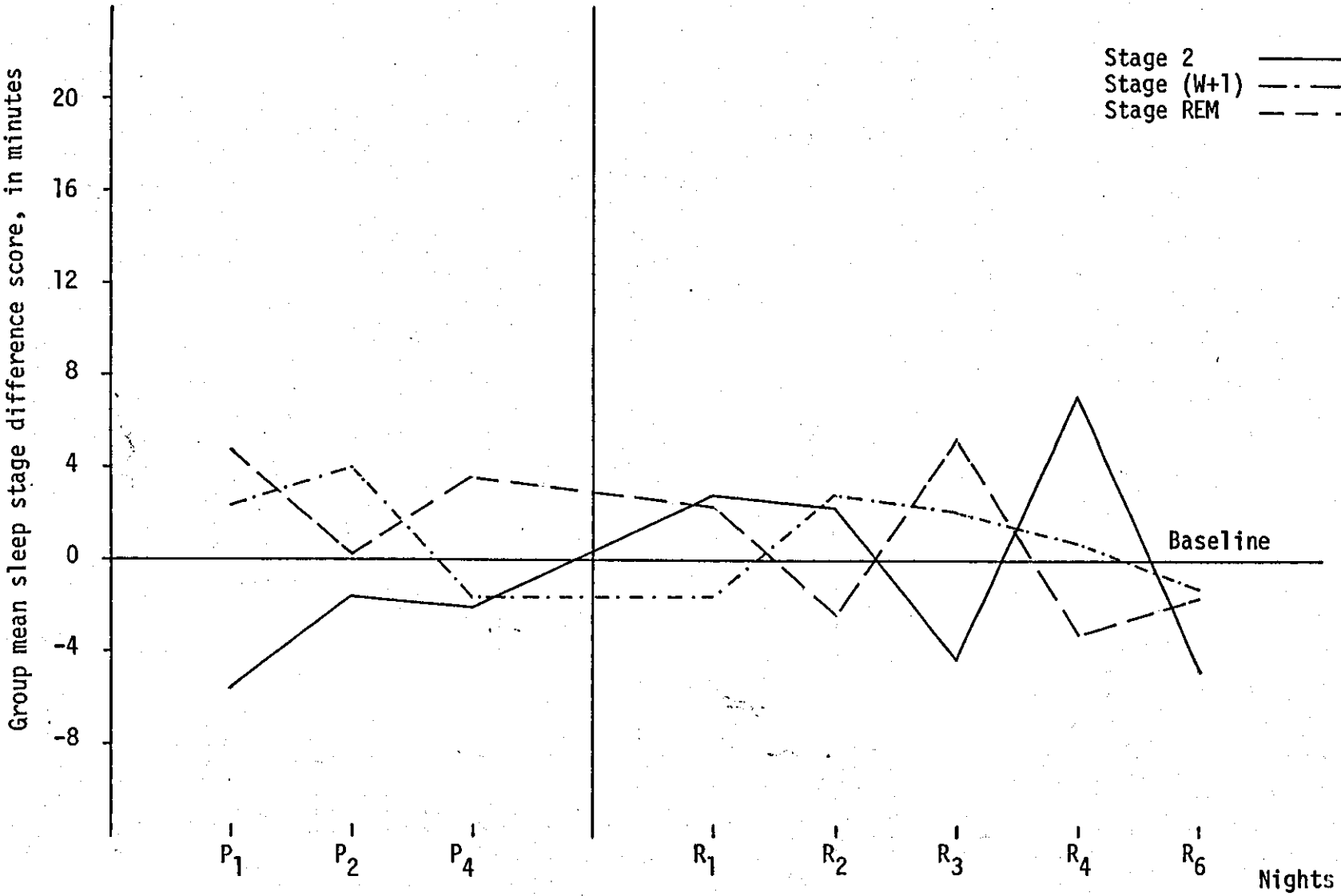
Graph F: A.S.A. Pilot Study



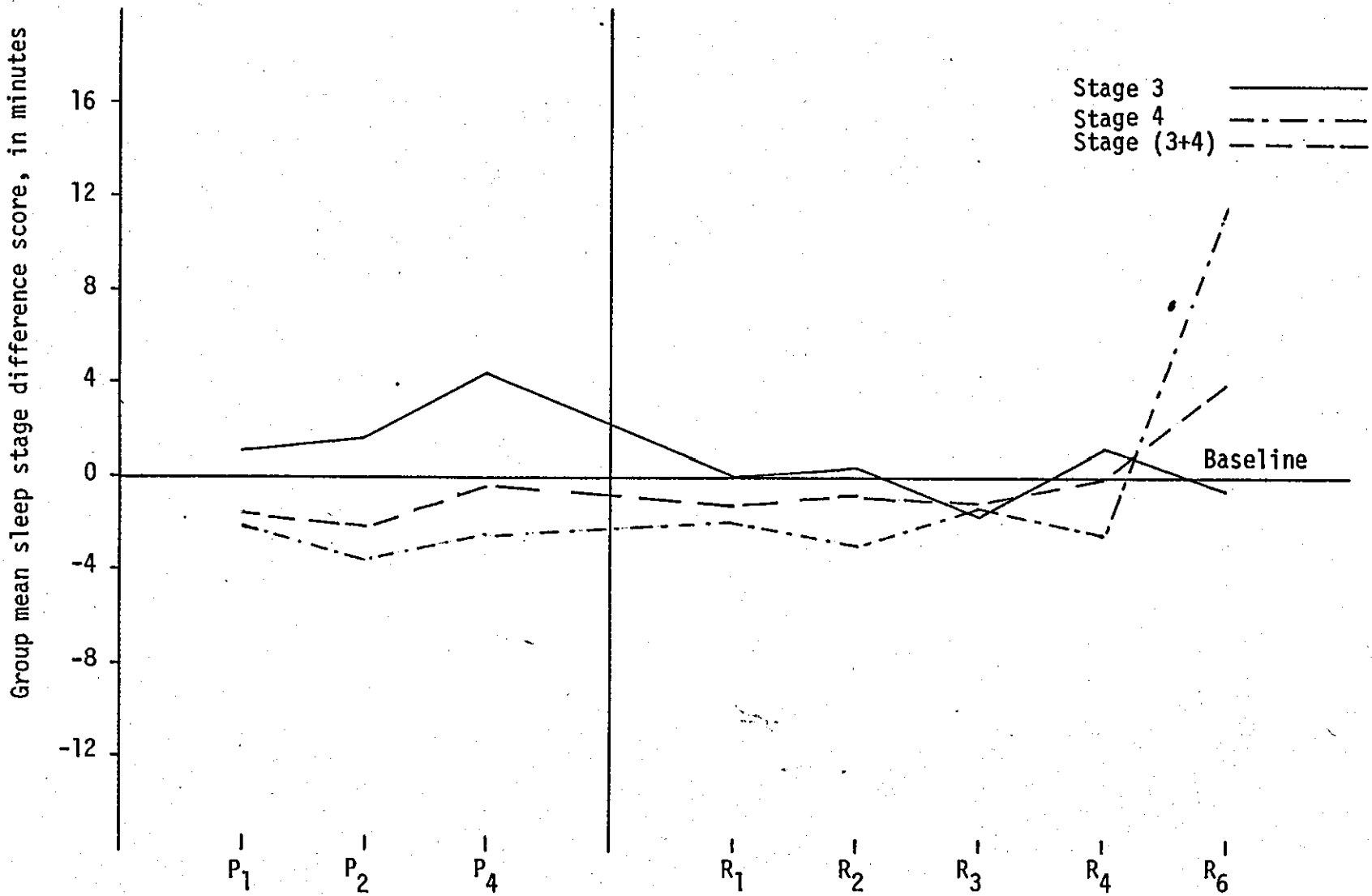
Graph H: A.S.A. Group (Main Study)



Graph I: A.S.A. Group (Main Study)



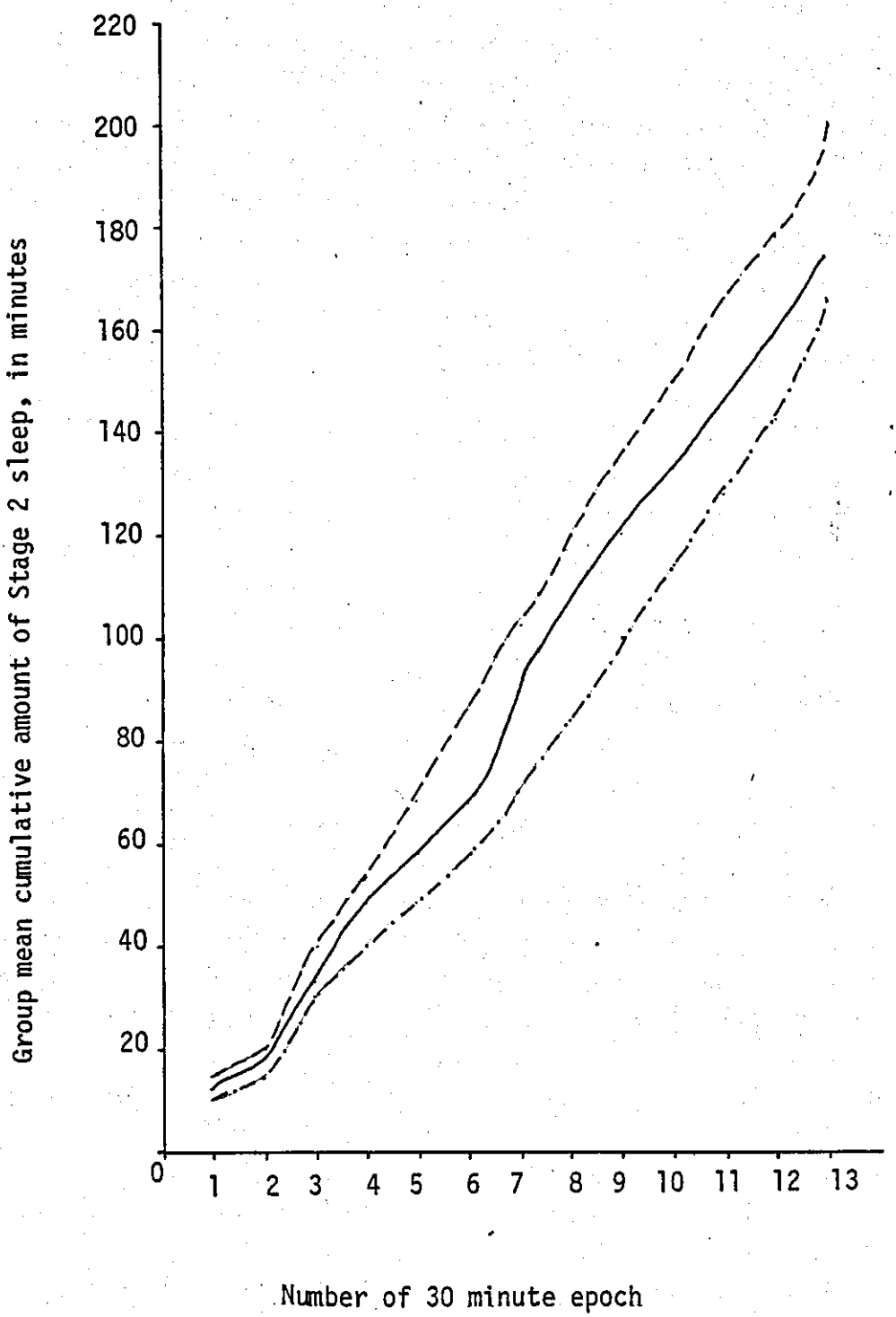
Graph J: Placebo Group



Graph K: Placebo Group

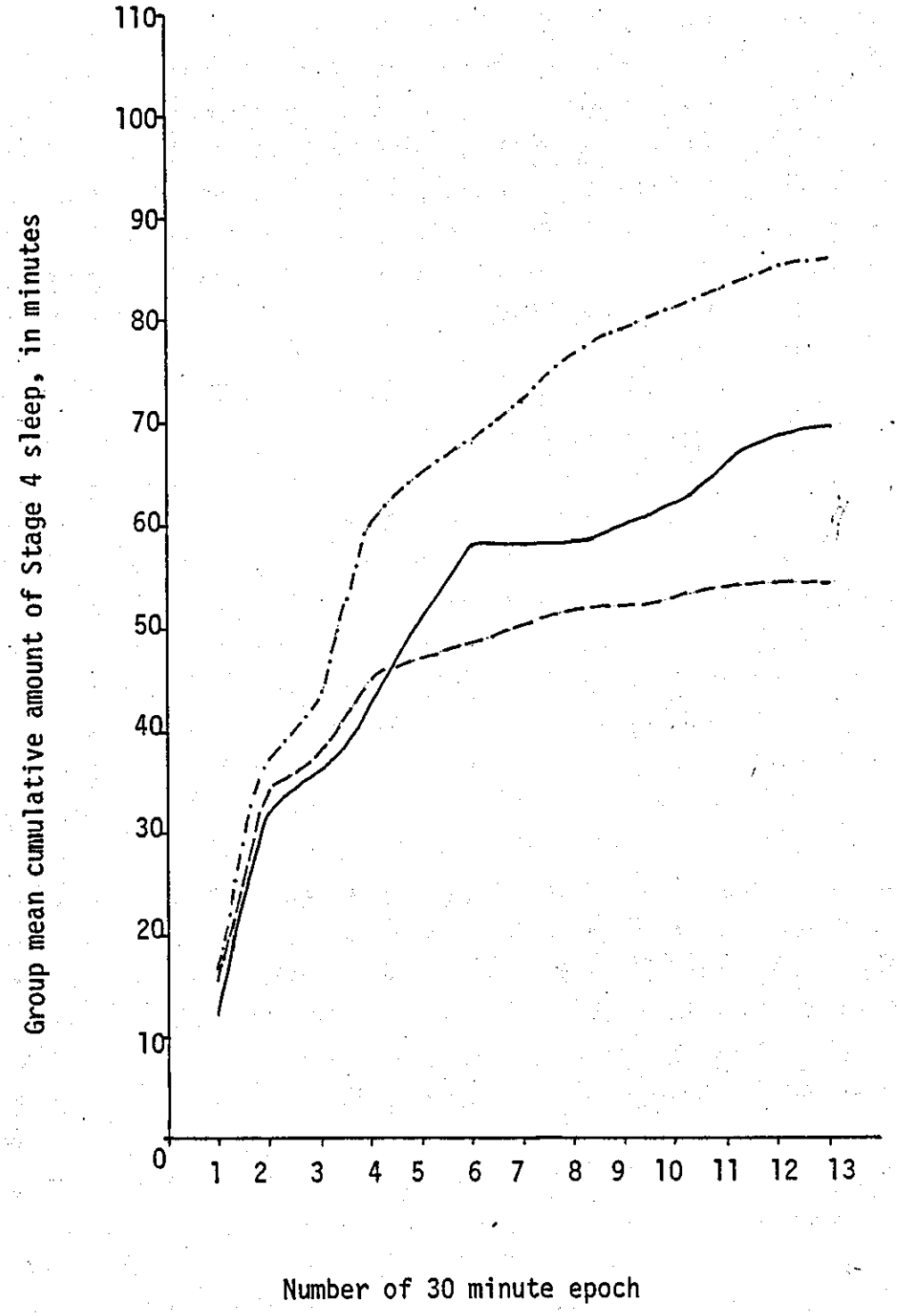
Graph L: The Accumulation of Stage 2 Sleep

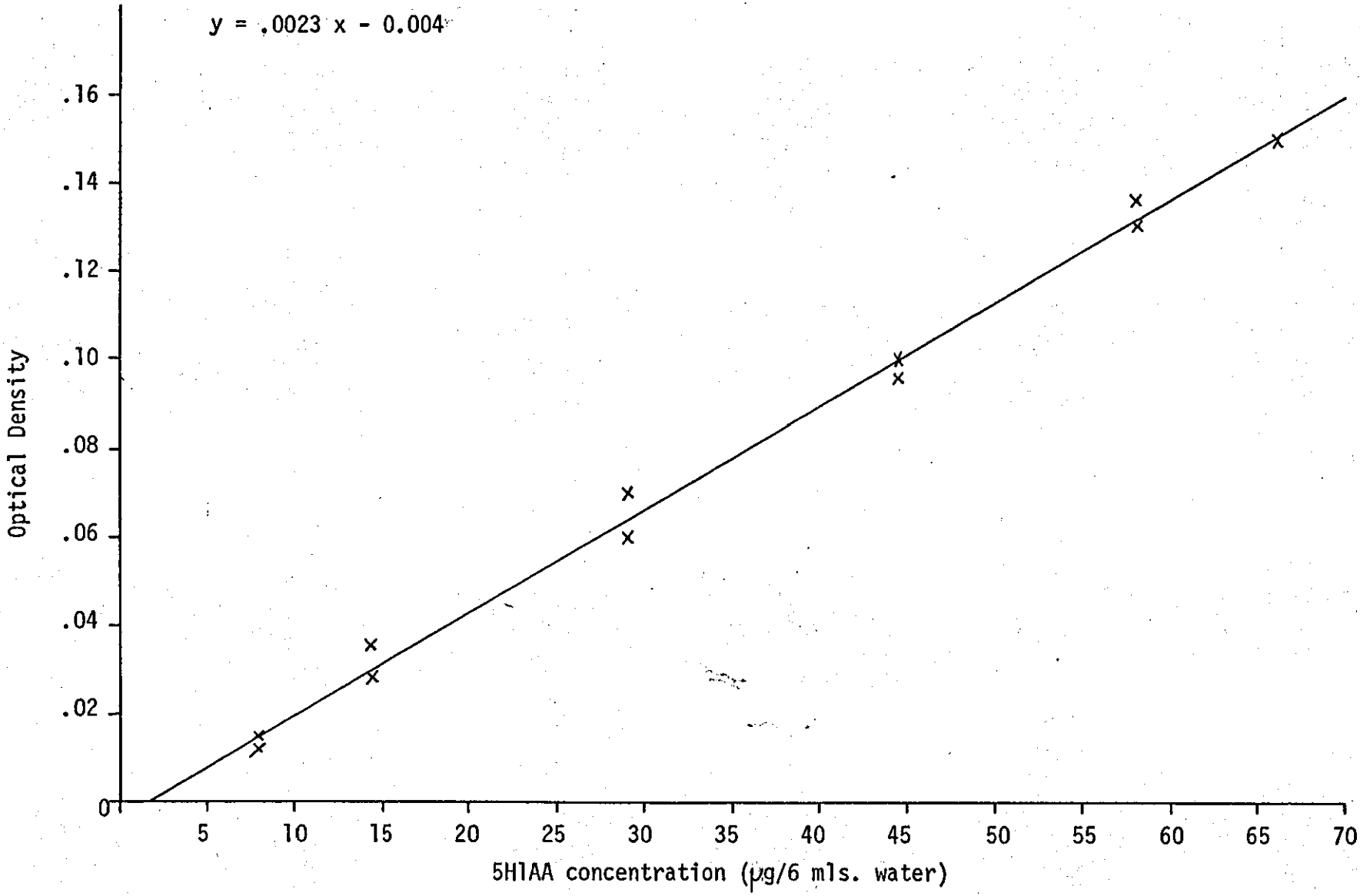
A.S.A. group, baseline ——————
A.S.A. group, second drug night – – – – – –
Placebo group, second drug night —·—·—·—



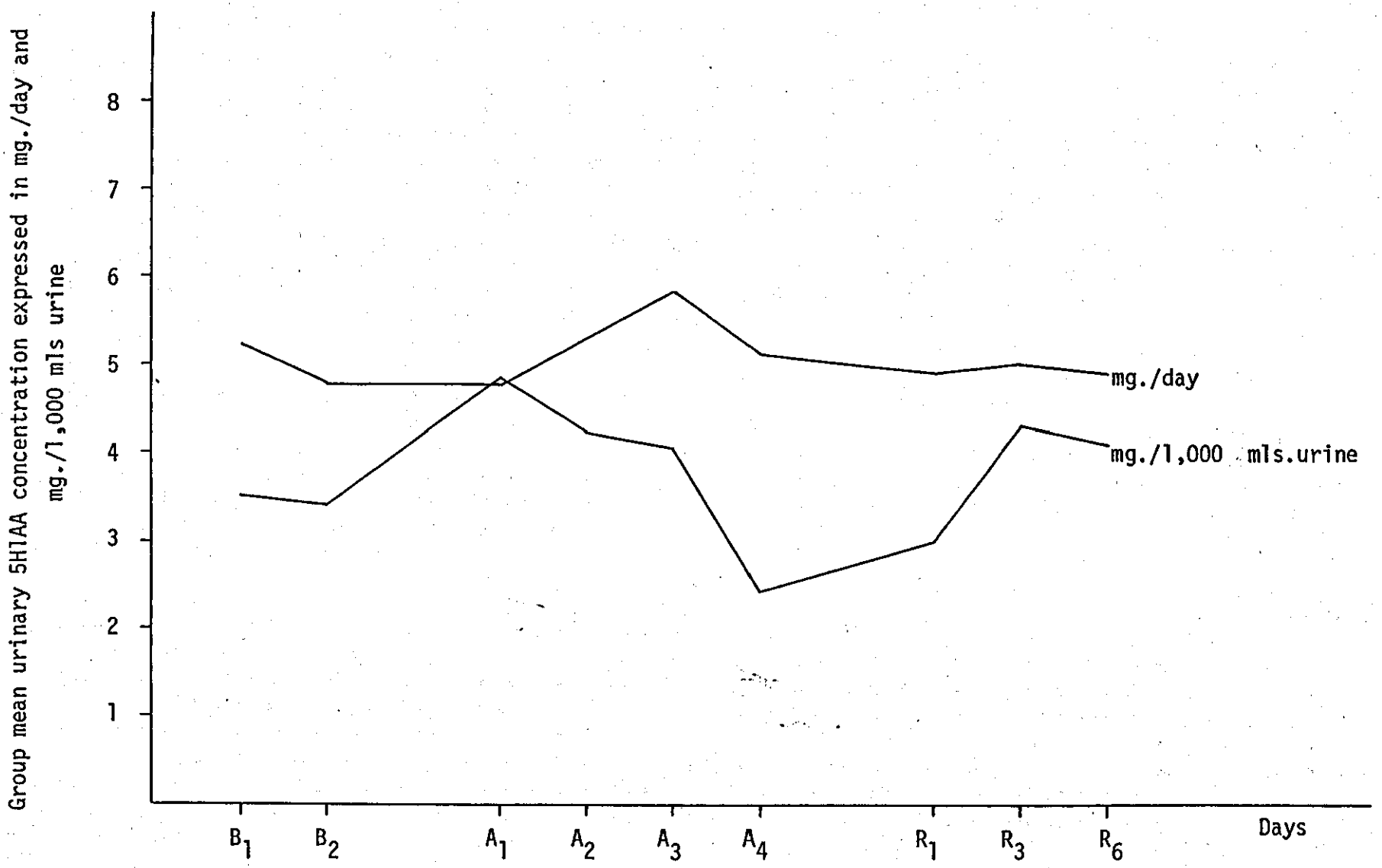
Graph M: The Accumulation of Stage 4 Sleep

A.S.A. group, baseline ——————
A.S.A. group, second drug night – – – – – –
Placebo group, second drug night —·—·—·—

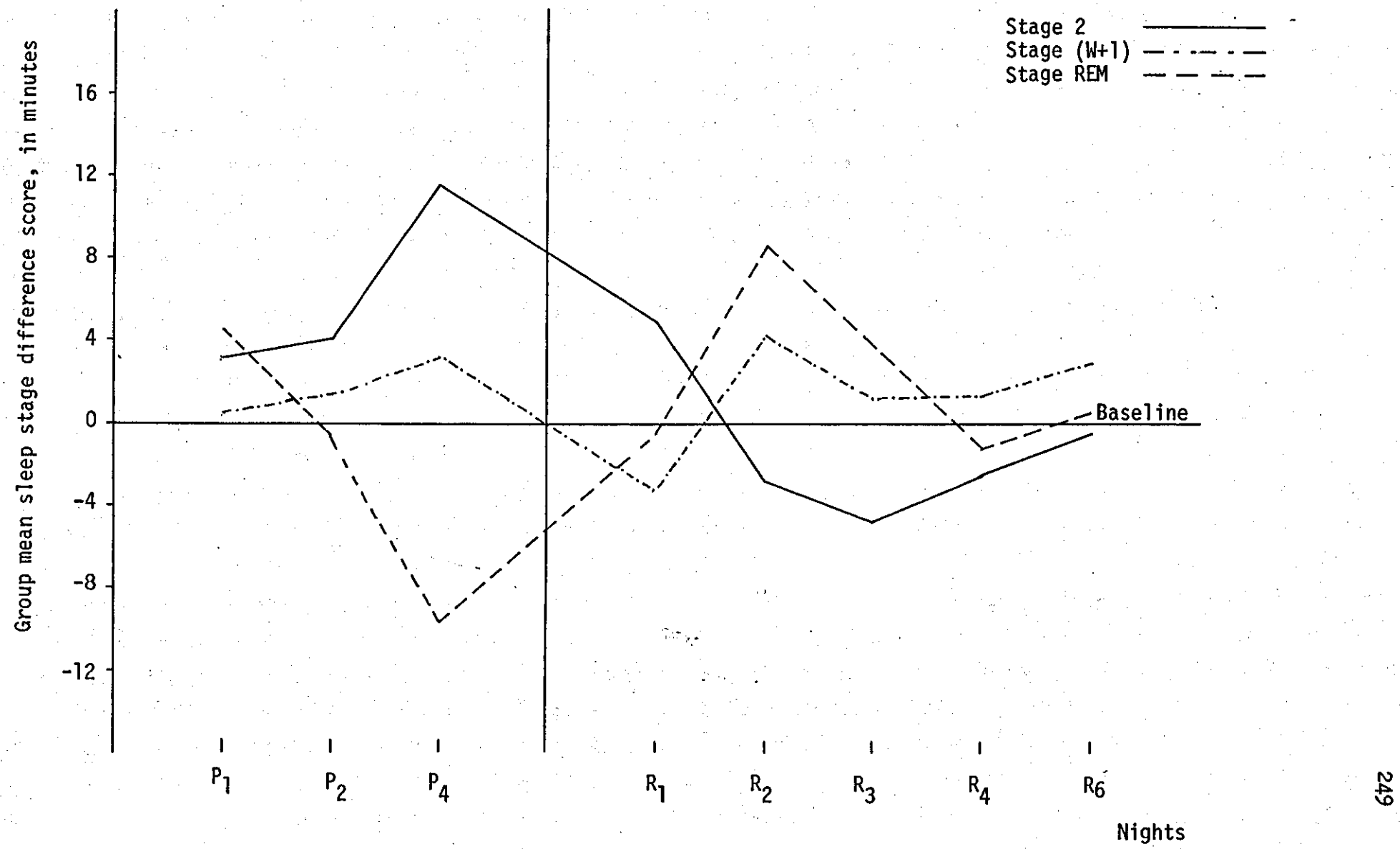




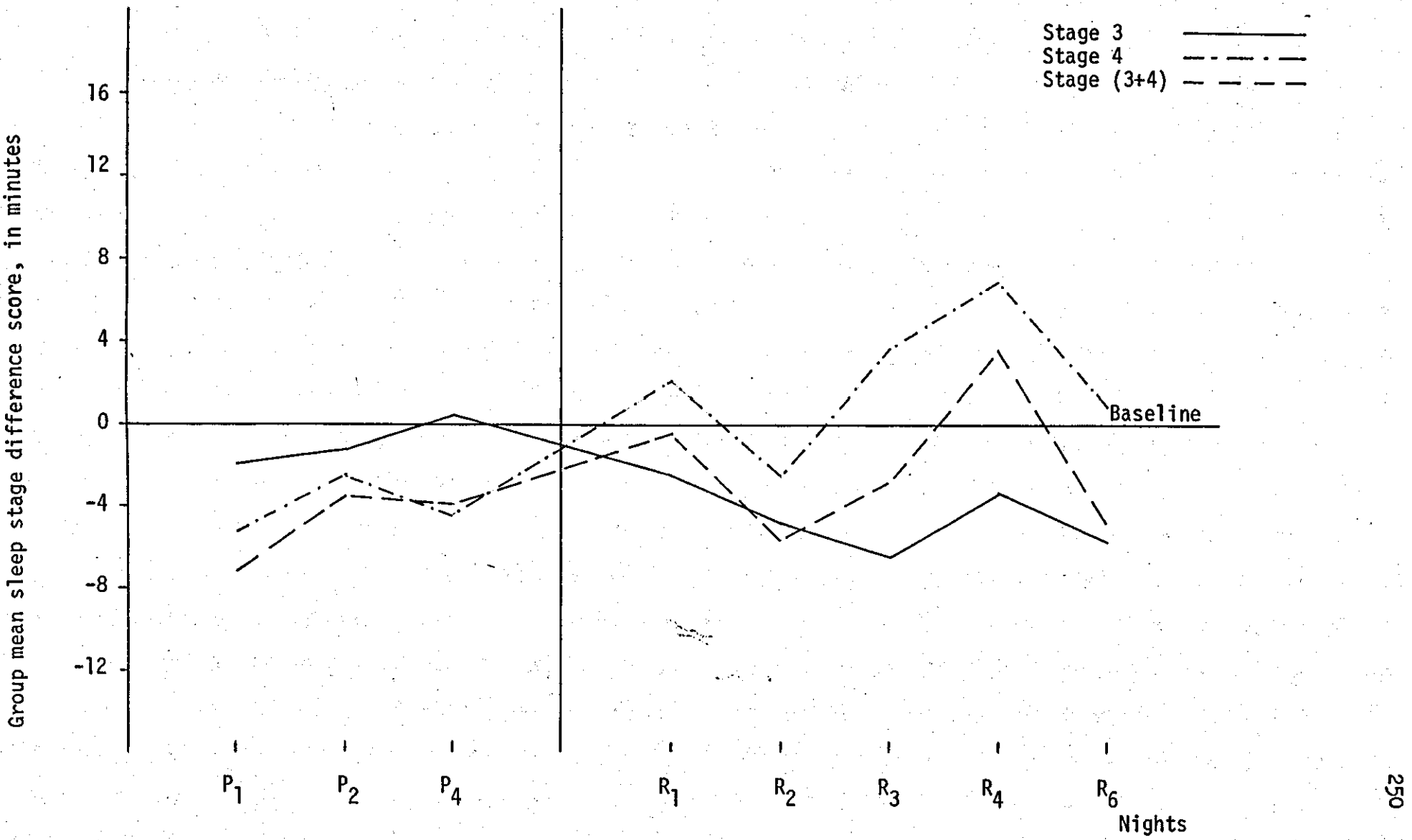
Graph N: Standard Curve for Urinary Analysis of 5HIAA



Graph O: Urine Analysis of 5HIAA



Graph P: Paracetamol Group



Graph Q: Paracetamol Study

Chapter Four

ENVOI

The non-drug, commercial "sleeping aids", namely, the cushions providing cycloid vibration, chocolate flavoured Horlicks and the placebo condition, cocoa, did not appear to have any marked effects upon the quality or quantity of sleep of a group of self-reported sleep onset insomniacs, according to EEG criteria (chapter one). However, some very interesting subsidiary findings and observations were made, which led to the subsequent studies in this research programme. One such finding was the difficulties encountered in obtaining a group of sleep onset insomniacs: out of the thirteen, self-reported sleep onset insomniacs who were originally selected on the basis of their responses to the sleep questionnaire (Appendix II), only three actually satisfied the EEG criteria for sleep onset insomnia* (Table 10, section 14.0, chapter one). This difficulty arose primarily from the majority of subjects overestimating their sleep onset times; this phenomenon has been reported by several other researchers, for example, Monroe (1967) and Freedman and Papsdorf (1976). However, this does not imply that all complaints of insomnia are merely "products of the imagination", for three of the subjects did estimate their sleep onset times quite accurately.

It would appear that after a period of sleeping difficulty, some people come to label themselves as "insomniacs", regardless of their present sleep onset times. It was also interesting to note that the mean sleep onset time of the insomniac group was not significantly different from that of a good sleep group (the placebo group from chapter

* Throughout this text a subject is classified as suffering from moderate sleep onset insomnia, if he has a sleep onset time of 30 - 60 minutes, when measured by the EEG. The EEG definition of sleep onset time employed here is the time from "lights out" until the first appearance of a stage 2 period which is of at least five minutes unbroken duration.

three). One of the self-reported good sleepers actually had a sleep onset time of 60 minutes, yet this subject still classified himself as a good sleeper. The difference between the two groups appeared to lie mainly within their attitudes and expectations with regard to sleep, rather than in an actual time difference. Johns (1977) reported that good sleepers were good estimators of their sleep onset times. This finding was supported by the results of the study conducted in chapter two, which also showed that good sleepers were good estimators of their sleep onset times. This indicates that the phenomenon of overestimation of sleep onset time is peculiar to the groups of self-reported sleep onset insomniacs.

In an attempt to determine why the insomniacs overestimated their sleep onset times, their first hour of sleep was divided into 15 minute epochs and compared with that of a good sleep group. There was a marked similarity between the two groups, but the insomniac group did show significantly more stage changes during the first 15 minutes, which may have left the impression that they had been awake for 15 minutes after their time of sleep onset, as measured by the EEG criteria. The time from "lights out" until the time that the insomniac returned to sleep after their first awakening was also calculated, in order to determine if this was the insomniacs' measure of sleep onset time. This measure did not agree with their estimates; so this was not a satisfactory explanation for the fact that the insomniacs overestimated their sleep onset times.

This study clearly showed that there are two main approaches to the study of insomnia, the subjective and the objective. Both have very important rôles to play in our understanding of insomnia. One of the

most fascinating aspects of the subjective approach is why so many insomnaics claims were not supported by the EEG measure. The subjective approach is very important in the treatment of insomnia, for if the subject claims that he is sleeping better after a particular treatment, then he is effectively cured. However, it is the EEG approach which will reveal the actual effects of the various treatments and whether they are altering the quality or quantity of sleep. It was the objective approach which was pursued further, since this was the initial approach adopted for the evaluation of the non-drug "aids".

Although three of the self-reported sleep onset insomniacs were fairly good estimators of their sleep onset time, there was no obvious feature which distinguished them from the rest of the group. Therefore, there appears to be no obvious way of determining the objective sleep onset time from the subjective estimate, apart from bringing the self-reported sleep onset insomniacs into the laboratory for several nights. Considering the number of subjects from the study in chapter one who failed to satisfy the EEG criteria for sleep onset insomnia, this procedure is likely to be very time consuming.

In an attempt to overcome this problem, to some extent, an EEG model of sleep onset insomnia was created in the laboratory by allowing good sleepers, who were unaccustomed to daytime napping, to nap during the morning and afternoon, and measuring their sleep onset times by means of the EEG. The group mean sleep onset time of the good sleepers during the morning nap session was 45 minutes, which was significantly greater than at night (26 minutes). Although this model will not eliminate the problem of selecting a group of insomniacs to satisfy EEG criteria, it will serve as a very useful, initial evaluation technique

for various "sleeping aids", be they drug or non-drug: if an "aid" were found to reduce sleep onset times in the model, one would then be justified in going to the lengths of obtaining a group of insomniacs who satisfied the EEG criteria. This model will yield more information concerning the sleep inducing potential of various "aids" than if they were administered at night to a group of good sleepers, for such a group would typically have short sleep onset times at the outset and the "aid" would have little opportunity to demonstrate its possible effectiveness as a sleep inducing agent. Similarly, if the "aid" was administered to good sleepers kept awake by artificial means, such as central nervous stimulants, little information would be gained about the sedative properties of the drug. One might be observing an interaction effect of the "aid" with the stimulant and not the typical, characteristic actions of the "aid". However, the model does possess one inherent disadvantage, that is, it looks at sleep latency during the day and not at night; therefore, there is a time of day difference between the EEG model of sleep onset insomnia and actual insomnia.

An interesting finding from the EEG model of sleep onset insomnia study was that the naps did appear to be influencing the night time sleep onset times: the fourth baseline night had a significantly greater group mean sleep onset time (42 minutes) than the first (26 minutes). This suggested another possible method of creating sleep onset insomnia, but further research is needed here. This would involve a similar study to the one previously conducted (chapter two), but it would continue for a longer period of time, to investigate whether the trends in increasing sleep onset time at night continued. If insomnia could be created at night this would eliminate the time of day difference. However, consideration must be given to the fact of whether the insomnia

created in the laboratory would be easily reversible. This study would also serve to give some indication as to how many naps it takes before an individual becomes accustomed to napping, at which point the sleep onset times would begin to decline. An unexpected finding was that slightly less than half (44%) of the subject sessions during the morning actually resulted in sleep; this percentage was considered rather high, as these subjects were habitually non-nappers. One possible method of reducing this percentage and thereby increasing the group mean morning sleep onset time may be to employ the "Morning Type/Evening Type" questionnaire (Horne & Östberg, 1976) in the subject selection procedure. It would be expected that subjects who were at their "peak" in the morning would be less likely to fall asleep than those who were at their "peak" in the evening. This is only a tentative suggestion.

The model was adapted slightly for a drug evaluation study, in order that repeated measures on the sedative potential of the drugs could be carried out. The results of the model study indicated that, as hypothesised, Temazepam had sedative properties. This effect had been hypothesised, because Temazepam is a member of the benzodiazepine group of compounds, is listed as a tranquiliser, and has been shown to bring about sedation (Fuccella et al., 1972). However, the drug F.P. did not appear to match Temazepam with respect to its sedative effects: it showed more resemblance to the placebo. The repeated measures design data analysis revealed that the second period, regardless of the treatment condition, was more conducive to sleep than the others. From this first application study of the EEG model of sleep onset insomnia a list of points and possible experimental design changes were drawn up for consideration before embarking upon another study with the model:

1. Ideally, all the subjects should sleep in the laboratory for at least two nights prior to the napping session and during the course of the study. If this is not possible, one nap session should be allowed for adaptation purposes and a check kept upon the night-time sleep by means of questionnaires.

2. The number of 30 minute recording periods could be reduced from four to three, in an attempt to reduce boredom effects. Likewise, the length of the interspersed reading periods could be reduced from 30 to 20 minutes. It was found that when the drug was exerting a sedative effect (Temazepam), it was very difficult for the subjects to keep awake during the 30 minute reading periods. Alternatively, if the drug was not exerting a sedative effect, the subjects became bored during the latter reading periods.

3. The model needs to be tested with more drugs of known sedative potential, such as other members of the benzodiazepine group and perhaps some barbiturates in order to produce a set of standards against which drugs of unkown sedative potential could be compared.

4. An alternative experimental design would be to have four groups of subjects, each group corresponding to a particular recording period. All subjects would be dosed at the same time and each group would come to the laboratory at the respective period recording times for a one hour nap session. This design would have two main aims, firstly, to reduce the amount of boredom, because the subjects would only be in the laboratory for one hour instead of 3½ hours and secondly to enable longer sleep onset times to be investigated. However, this design does introduce

the problem of inter-subject variability.

5. In the present model studies the subjects were not instructed to fall asleep during the EEG recording periods. It would be interesting to investigate whether altering the instructions given to the subjects would affect sleep latency.

6. On the basis of the results with Temazepam, it would be worthwhile selecting a group of sleep onset insomniacs and evaluating this drug upon them.

This EEG model of sleep onset insomnia shows potential as a useful evaluation technique in the drug field. There is also the possibility that the reverse aspect of the model may be useful, that is to allow subjects who are accustomed to napping to nap during the afternoon. This may result in short sleep onset times which might be useful in the evaluation of stimulants.

The final, main study examined the effects of acetylsalicylic acid (A.S.A.) upon sleep. This aspect of insomnia stemmed from the comments made by many of the self-reported sleep onset insomniacs that the frequently took Aspirin to relieve their insomnia. No previous studies have examined the effects of A.S.A. upon normal sleep; in fact, very few analgesics have been used in sleep research. The effects of some of these drugs upon sleep may help eventually in our understanding of the sleep mechanisms in Man. The A.S.A. study was somewhat unusual in that it examined the effects of a normal, therapeutic dose of the drug administered throughout the day, whereas, the majority

of drug studies simply evaluate the effects of a drug administered as the subject retires to bed.

The results of the A.S.A. study suggested that A.S.A. was exerting a slight arousing effect upon sleep, revealed by the significant increase in stage 2 sleep, which was accompanied by a decline in stage 4 and a slight decline in stage REM, neither of which reached statistical significance. The night-to-night variability of stage 3 sleep was significantly increased upon A.S.A. administration and the build up of stage 4 sleep was slightly depressed after the first 2½ hours of sleep. Upon withdrawal of A.S.A., there was a significant decline in REM latency and an increase in the night-to-night variability of stage REM. Various speculations were made in the discussion (section 11.0, chapter three) as to how A.S.A. might be exerting these effects.

There is also the possibility that A.S.A. could be producing changes in the sleep EEG which are undetectable by means of the visual scoring technique of Rechtschaffen and Kales (1968), which was employed throughout this research programme. A further aspect of research would be to analyse the EEG by computerised methods. However, at the time of the study this approach was not feasible. The visual scoring technique has the disadvantage that stage 2 sleep can contain from 0 - 20% of delta activity, (0.5 - 3.5 Hz., with an amplitude greater than 75 μV) and still be classed as stage 2. It is not until the amount of delta activity exceeds 20% of the activity of a one minute epcoh that it is classified as stage 3. It would therefore be interesting to investigate whether the increase in stage 2 brought about by A.S.A. administration contained a high percentage of delta activity. Feinberg, Fein, Walker, Price, Floyd

and March (1977), using a computerised EEG analysis technique, found that an increased amount of stage 2 produced by the drug Flurazepam contained a sufficient amount of delta activity to offset the loss of delta activity due to the significant decline in stage 4 sleep. It is possible that the increase in stage 2 sleep is the primary effect of A.S.A. and the decline in stage 4 could be secondary, due to the increased amount of delta activity in stage 2. This idea is in part borne out by the pattern of accumulation of stage 4 after A.S.A. administration. Graph M (section 25.0, chapter three)showed that the accumulation of stage 4 was depressed after the first 2½ hours of sleep on the second A.S.A. night. A possibility is that an increased amount of delta activity due to the rise in stage 2 sleep may have resulted in the "nightly quota of delta activity" being produced in the first 2½ hours of sleep; hence the depression in the build up of stage 4. This suggestion is only speculative at present.

A computer analysis technique may also be very useful in the evaluation of insomnia. Such a technique may reveal differences in the EEG between the self-reported sleep onset insomniacs of chapter one and the good sleepers. Any differences which may be found would provide a possible explanation of why the subjective reports of sleep onset insomnia were not always borne out by the EEG measure of sleep latency. As suggested by Karacan et al. (1973), "the meaning of insomnia to such patients must be sought in subtle EEG disturbances rather than in gross disturbances of sleep latency, total sleep time or sleep maintenance".

Another problem encountered in the studies of this research programme was the large amount of inter-subject variability for the various sleep stages and parameters. This can be observed from the ANOVA

summary tables. This problem was more evident in the A.S.A. study, as a comparison was being made between two groups of subjects. In an attempt to overcome the difference between the baseline scores for the the two groups, the scores for each sleep stage and each parameter, for each subject, on each night, were expressed as the difference score by subtracting them from the mean baseline value. An alternative approach to overcoming this problem may have been to have used a different experimental design. Each subject could have acted as his own placebo control, but this introduces another variable, that of a temporal factor, whereby a subject cannot receive both A.S.A. and placebo at once. The time interval between the two treatments may lead to variations which could affect comparisons. This problem of inter-subject variability appears to be an inherent problem which will have to be accommodated according to the study. The response to A.S.A. also showed a large amount of inter-subject variability. One of the reasons for this might have been that the dose of A.S.A. employed was too low to bring about a consistent response in all subjects. The next step, therefore, would be to increase the dose of A.S.A. used or to use a drug with similar but stronger actions, such as indomethacin. However, on ethical grounds neither of these approaches was feasible.

It was decided therefore, to investigate whether the effects of A.S.A. upon sleep were peculiar to this drug or whether related drugs, such as Paracetamol, also showed these effects. The results of the pilot study conducted with Paracetamol suggested that it was having an effect upon sleep similar to that shown by A.S.A., but the effects were less marked. However, more subjects are required before the Paracetamol findings can be confirmed. Nevertheless, these findings did suggest that the effect of A.S.A. upon sleep could not be explained simply on the

grounds of adverse side-effects, as Paracetamol, at therapeutic doses, has very few adverse side-effects (Goodman & Gilman, 1970).

A biochemical pilot study was also conducted with the aim of gaining some knowledge of how A.S.A. exerted its effects upon sleep. The choice of assay techniques was limited due to the facilities available and the fact that it was not feasible to use a technique which involved invasive procedures. The colormetric urine analysis of 5-HIAA was the most suitable at the time (Udenfriend et al., 1958). The link between A.S.A. and 5-HIAA is that A.S.A. has been shown to release tryptophan from its binding site (Smith & Lakatos, 1971), which may pass along the metabolic pathway which converts tryptophan to 5-hydroxytryptamine, which, in turn is broken down to 5-HIAA. Although the results of this analysis may reveal changes in 5-HIAA concentration due not only to A.S.A., but also other factors, such as diet and free fatty acid concentration in the plasma, it was considered that if A.S.A. was having any major effect upon this tryptophan pathway, then it would be detected by an increase in the concentration of urinary 5-HIAA. This analysis can, therefore, only give some indication as to the action of A.S.A. upon 5-HIAA output in the urine, as one cannot necessarily conclude that any changes observed are due solely to the action of A.S.A. The results of the urine analysis pilot study indicated that A.S.A. was having no marked effect upon the excretion of 5-HIAA. One problem encountered with this particular assay was that the standard readings fluctuated from day to day. However, this assay was sufficiently sensitive to detect a tryptophan load of approximately 90 mgs., administered via the diet. A flurometric assay may be more sensitive to slight changes in the concentration of 5-HIAA and ought to be considered if this aspect of the research programme were to be pursued further.

Overall, the effects of A.S.A. upon the sleep of good sleepers suggested that A.S.A. was exerting a slight arousing action rather than a sleep promoting one, as suggested by the self-reported, sleep onset insomniacs of chapter one. However, A.S.A. has not necessarily been given sufficient opportunity to reveal its full sedative potential. The recent work by Hauri and Silberfarb (1978) revealed that a single dose of A.S.A. (650 mgs.) brought about a significant increase in the total sleep time and a signficant reduction in the amount of wake time after sleep onset and the total amount of wake time in a group of insomniacs. The baseline group mean sleep onset time for this group of insomniacs was only 30.9 minutes; so A.S.A. may not have been able to reveal its sedating properties in this group, as only a few subjects may have had relatively long sleep latencies. Overall, the results of the Hauri and Silberfarb (1978) study indicated that A.S.A. may be a useful hypnotic for those insomniacs suffering from early morning or frequent nocturnal awakenings. In view of the findings of the Hauri and Silberfarb (1978) study, it would still be beneficial to pursue one of the further research proposals put forward in chapter three, that is, to assess the sedative potential of A.S.A. using the EEG model of sleep onset insomnia when a table of standards has been drawn up. The aim of this study would be to gain some indication of whether A.S.A. would be an effective "sleeping aid" for those insomniacs whose primary complaint is a long sleep latency. Hauri and Silberfarb (1978) reported that A.S.A. had no significant effects upon the stages of sleep. The present study revealed that A.S.A. produced a significant increase in stage 2 sleep, but that other stages were not significantly altered, whereas, the majority of hypnotics are known to suppress REM sleep. At present one cannot say whether these changes in the sleep stages brought about by drugs are detrimental to the subjects; therefore,

one cannot come to the conclusion that A.S.A. has the advantage over other hypnotics of not suppressing REM sleep. If A.S.A. is to be examined further as a potential hypnotic, consideration must be given to the fact that it is known to produce gastric irriation at varying doses in different individuals.

The range of possible drug and non-drug "aids" for insomnia appears to be almost as wide and diverse as the underlying causes of insomnia. This may reflect the insomniacs' limitless attempts to obtain a "good night's sleep". Since sleep is highly coveted by many, the insomniac feels almost compelled to adopt some "aid" to sleep. Sleep is held in high esteem and the insomniacs feel that they must achieve it regardless of the cost.

> "Do but consider what an excellent thing sleep is: it is so inestimable a jewel that, if a tyrant would give his crown for it, it cannot be bought:" (Thomas Dekker)

APPENDICES

APPENDIX I

New Hypnotic Prescriptions in the United States

1974 - 1976 * (in millions)

Table 1

	1974	1975	1976
Flurazepam (Dalmane)	4.45	5.86	6.76
All Benzodiazepines	6.51	8.52	8.49
All Hypnotics	8.29	6.42	5.32
Barbiturate - Benzodiazepine ratio	1.27:1	0.75:1	0.62:1

* Source: National Prescription Audit

From Cohen and Blutt (1978)

APPENDIX II

I N · S T R I C T C O N F I D E N C E

SLEEP QUESTIONNAIRE

Please complete this questionnaire as soon as possible. You may write your answers in the space provided immediately below the question. If there is insufficient space you may continue your answers in the blank space at the end of the questionnaire. You may also use this space to ask any questions which you may have about the study or to give us any additional information which you think may be useful.

1. Name:

2. Age:

3. Address:

4. At what time do you usually go to bed?

5. (a) How long does it take you to fall asleep?

   (b) Has it always taken you this long?

   (c) If not, since when has this been happening?

6. (a) Do you wake up during the night?

   (b) If so how often do you wake up?

   (c) How long does it take you to fall asleep again?

7. At what time do you wake up in the morning?

8. (a) Do you take naps during the day?

(b) If so, how long for?

(c) How often?

9. (a) Do you take anything to help you sleep?

(b) If so, what?

(c) Since when?

(d) Is your doctor prescribing this treatment?

10. (a) Do you smoke?

(b) If so, how many on average per day?

11. Do you suffer from bronchitis?

12. How many hours sleep per night do you need?

13. Do you normally get this amount?

14. Are you on a course of antibiotics or tranquilisers?

The above questions make up the basic questionnaire and the following questions i) - iv) were inserted at the end, according to the study.

i) Are you able to take Aspirin/Paracetamol without experiencing adverse side-effects?

ii) Would you be concerned about having to take a therapeutic dose of Aspirin/Paracetamol for four days?

iii) Do you suffer from menstrual pain to such an extent, that it is likely to affect your sleeping habits?

iv) It is important for us to avoid using you as a subject during menstrual onset, what was the date of your last menstrual period?

APPENDIX III

I N S T R I C T C O N F I D E N C E

MORNING QUESTIONNAIRE

Please complete this questionnaire upon awakening. Any further comments which you would like to make can be inserted at the end.

1. Name Date:

2. At what time did you go to bed last night?

3. How long did it take you to fall asleep?

4. (a) Did you awaken during the night?

   (b) If so how often?

   (c) How long did it take you to fall asleep again?

5. At what time did you awake this morning?

6. When you woke up this morning how did you feel?

   - tired
   - average
   - rested

7. Did you have a hard/easy/ average time awakening this morning?

8. Do you feel as though you had enough sleep last night?

9. When you went to bed last night how did you feel?

   - tired
   - average
   - alert

10. What were your dreams like last night?

- pleasant
- unpleasant
- cannot remember

11. When you went to bed last night how tense did you feel?

- tense
- average
- relaxed

APPENDIX IV

IN STRICT CONFIDENCE

DAYTIME QUESTIONNAIRE

1. Name ..... Date .....

2. Do you feel as though you fell asleep during the rest period?

   .....

3. If so, how long did it take you to fall asleep? .....

4. How long were you asleep ? .....

5. Would you please comment on how you felt at the outset of the rest period? e.g. tired, alert, tense, anxious?

6. How do you feel now?

7. If you were unable to fall asleep, did it worry you or play on your mind in anyway?

   .....

8. Would you please estimate for how long you have been in the bedroom?

   .....

9. Would you please list anything which you have had to eat or drink since awakening this morning, including your breakfast?

Thank-You

APPENDIX V

ASPIRIN QUESTIONNAIRE

IN CONFIDENCE

NAME:

1. Would you please give an estimate of how often you would normally take ASPIRIN tablets, for example, twice per week, once per month?

2. If you had a headache or pain, which drug would you normally take, for example, Disprin, Aspro, Aspirin, Anadin, Paracetamol, etc. ?

3. Can you remember the last time you took an ASPIRIN tablet or any other tablet prior to the study? Would you please indicate when and the name of the tablet?

4. Did the fact that you had to take ASPIRIN for four days worry you at all?

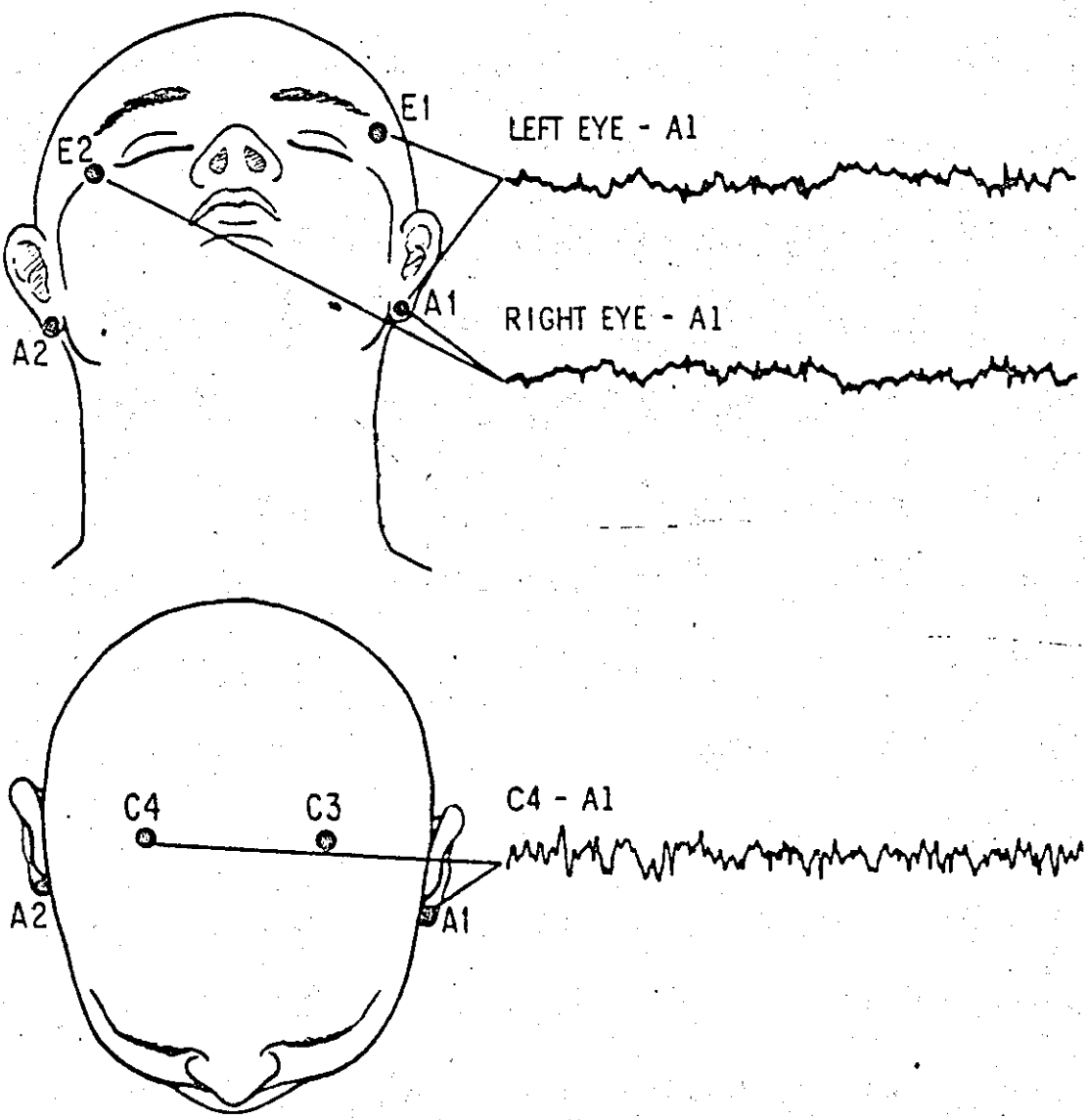
5. Did you experience any side-effects whilst taking ASPIRIN during the course of the study?

6. During the course of the study were you on any special diet, for example, vegetarian, sugar free, low carbohydrate?

7. Would you please indicate how you estimate your average, daily carbohydrate intake, compared with the student population?

1	2	3	4	5
more	slightly more	same	slightly less	less

Thank You



Electrode Placement for Sleep Stage Recording (after Rechtschaffen and Kales, 1968)

EOG - E1 1cm. above and out from outer canthus of left eye.
E2 1cm. below and out from outer canthus of right eye.

EEG - C3 of international 10/20 system for electrode placements.
C4 of international 10/20 system for electrode placements.

Refernece Electrodes - A1 left mastoid
A2 right mastoid

An earth electrode (C_z) was located between C3 and C4

APPENDIX VII

THE EEG MEASUREMENT OF SLEEP

EEG activity can be divided into four main kinds of EEG rhythms:

1. Alpha – The frequency of this rhythm varies between 8 and 12 Hz., with an amplitude of 25 to 100 microvolts. Alpha activity appears to be mainly localised in the occipital and parietal areas of the brain. It is the most prominent EEG activity to be found in the waking brain, although in some individuals it is virtually absent. Alpha activity typically appears when the eyes are closed and the subject is relaxed, with little on his mind.

2. Beta – There is some disagreement concerning the frequency range of this activity, although the consensus gives the range between 14 and 30 Hz. The amplitude is low and seldom exceeds 20 microvolts. Beta is normally found in the frontal and central areas of the brain, and it is considered to be a background activity which, in wakefulness, is prominent in the absence of alpha. Therefore, its presence is taken to indicate varying degrees of tension or excitement.

3. Theta – The frequency range of this activity is from 4 to 7 Hz., with an amplitude of about 30 microvolts. It is not normally found in much quantity in the adult waking EEG and is most evident during dozing or light sleep.

4. Delta – The frequency of delta ranges from 0.5 to 3.5 Hz., with an amplitude of up to 150 microvolts. It is not usually evident in the adult waking EEG, but is very prominent during deep sleep.

The Sleep Stages

Stage W (waking)

This corresponds to the waking state which is characterised by EEG alpha activity, with some beta activity. There is a high tonic EMG, rapid eye movements (REMs) and blinking are very evident.

Stage 1

Here there is a relatively low amplitude, mixed frequency EEG activity with a prominence in the theta range; vertex sharp waves may appear towards the end of the stage. The EOG (electro-oculogram) shows slow eye rolling, and there is a fairly high tonic EMG which is generally below that of wakefulness. Stage 1 generally occurs in the transition from wakefulness to other stages of sleep and tends to be of relatively short duration. This stage occupies approximately 5-10% of the total sleep time (TST) of the young adult.

Stage 2

The EEG shows clear signs of sleep spindles* and K complexes**. There is a quantitative absence of delta activity to preclude it from Stage 3. The EOG is quiescent and the EMG is lower than that for Stage 1. This stage occupies 40-50% of the TST of the young adult.

---

* The term sleep spindle refers to EEG activity between 12 and 14 Hz. The presence of a sleep spindle should not be defined unless it is of at least 0.5 seconds duration, that is, one should be able to count 6 or 7 distinct waves within the half-second period.

** K complexes are defined as EEG waveforms having a well delineated negative sharp wave which is immediately followed by a positive component. The total duration of the complex should exceed 0.5 seconds.

Stage 3

Delta activity is present in the EEG record for at least 20%, but not more than 50%, of the time. There may also be K complexes and spindles present in the EEG. EMG and EOG are of a similar level to Stage 2. Stage 3 is considered by many researchers to be a transitory stage between Stages 2 and 4, and it occupies between 5 and 10% of the TST of the young adult.

Stage 4

Delta activity is present for a minimum of 50% of the time and frequently the EEG record is completely dominated by this activity. Sleep spindles may be present. The EOG is quiescent and the EMG tends to be low, but still higher than that of the REM stage. Stage 4 occupies 15-20% of the TST of the young adult.

Stage REM

The EEG shows low voltage fast activity of mixed frequency, similar to that found in wakefulness. Episodic REMs are present and the EMG reaches the lowest level found in any stage. Stage REM occupies 20-25% of the TST.

(Davies & Horne, 1975).

APPENDIX VIII

Non-Significant ANOVA Summary Tables for the Study Investigating the Effects of Cycloid Vibration upon Good Sleepers

Sleep Onset Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	34.5	3	11.5	0.3	n.s.	3,15
Subjects	1914.8	5	383.0	8.5	0.1%	5,15
Residual	674.7	15	45.0			
Total	2624.0	23				

Stage W+1

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	181.5	3	60.5	0.9	n.s.	3,15
Subjects	1806.75	5	361.4	5.7	1.0%	5,15
Residual	949.75	15	63.3			
Total	2938.0	23				

Stage 2

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	392.3	3	130.7	0.5	n.s.	3,15
Subjects	6658.8	5	1331.8	5.0	1%	5,15
Residual	3976.2	15	265.1			
Total	11027.3	23				

Stage 3

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	22.4	3	7.5	0.3	n.s.	3,15
Subjects	386.4	5	77.3	2.7	n.s.	5,15
Residual	431.8	15	28.8			
Total	840.6	23				

Stage 4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	325.7	3	108.6	1.3	n.s.	3,15
Subjects	4659.0	5	931.8	10.9	0.1%	5,15
Residual	1277.3	15	85.2			
Total	6262.0	23				

Stage 3+4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	398	3	132.7	2.3	n.s.	3,15
Subjects	4484	5	896.8	15.8	0.1%	5,15
Residual	849	15	56.6			
Total	5731	23				

Stage REM

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	892.3	3	297.4	1.5	n.s.	3,15
Subjects	2001.8	5	400.4	2.0	n.s.	5,15
Residual	2955.2	15	197.0			
Total	5849.3	23				

Total Sleep Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	250	3	83.3	1.6	n.s.	3,15
Subjects	307	5	61.4	1.2	n.s.	5,15
Residual	787	15	52.5			
Total	1344	23				

APPENDIX IX

Non-Significant ANOVA Summary Tables for the Study investigating the Effects of Non-Drug "Sleeping Aids" upon the Sleep of a Group of Self-Reported, Sleep Onset Insomniacs.

Sleep Onset Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	302.9	3	101.0	0.6	n.s.	3,21
Subjects	6776.8	7	968.1	5.9	0.1%	7,21
Residual	3470.8	21	165.3			
Total	10550.5	31				

Stage W+1 - Whole Night

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	283.3	3	94.4	0.7	n.s.	3,21
Subjects	2285.5	7	326.5	2.5	5%	7,21
Residual	2725.2	21	129.8			
Total	5294.0	31				

Stage W+1 - First Half of Night

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	94.4	3	31.5	1.0	n.s.	3,21
Subjects	433.2	7	61.9	2.0	n.s.	7,21
Residual	642.4	21	30.6			
Total	1170.0	31				

Stage W+1 - Second Half of Night

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	411.0	3	137.0	2.3	n.s	3,21
Subjects	1221.0	7	174.4	3.0	2.5%	7,21
Residual	1228.7	21	58.5			
Total	2860.7	31				

Stage 2

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	550.5	3	183.5	0.7	n.s.	3,21
Subjects	6511.8	7	930.3	3.4	2.5%	7,21
Residual	5741.8	21	273.4			
Total	12804.1	31				

Stage 3

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	234.8	3	78.3	2.5	n.s.	3,21
Subjects	988.0	7	141.4	4.5	1%	7,21
Residual	659.2	21	31.4			
Total	1882.0					

Stage 4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	51.1	3	17.0	0.2	n.s.	3,21
Subjects	4572.3	7	653.2	8.0	0.1%	7,21
Residual	1708.6	21	81.4			
Total	6332.0	31				

Stage 3+4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	132.8	3	44.3	0.4	n.s.	3,21
Subjects	4063.5	7	580.5	5.2	1%	7,21
Residual	2365.2	21	112.6			
Total	6561.5	31				

Stage REM

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	646	3	215.3	1.1	n.s.	3,21
Subjects	2473	7	353.3	1.8	n.s.	7,21
Residual	4012	21	191.0			
Total	7131	31				

REM Latency

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	1034	3	344.7	0.2	n.s.	3,21
Subjects	10949	7	1564.0	1.0	n.s.	7,21
Residual	34237	21	1630.3			
Total	46220	31				

Total Sleep Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	442	3	147.0	0.5	n.s.	3,21
Subjects	11025	7	1575.0	5.2	1%	7,21
Residual	6397	21	304.6			
Total	17864	31				

Number of Stage Changes

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Treatment	207.8	3	69.3	0.9	n.s.	3,21
Subjects	2454.5	7	350.6	4.4	1%	7,21
Residual	1689.4	21	80.4			
Total	4351.7	31				

APPENDIX X

Non-Significant ANOVA Summary Tables for the EEG Model of Sleep Onset Insomnia Study.

Stage W+1

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	84.7	3	28.2	0.4	n.s.	3,21
Subject	548.4	7	78.3	1.05	n.s.	7,21
Residual	1568.8	21	74.7			
Total	2201.9	31				

Stage 2

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	1338.0	3	446.0	2.6	n.s.	3,21
Subject	12149.5	7	1735.6	10.3	0.1%	7,21
Residual	3542.5	21	168.7			
Total	17030.0	31				

Stage 3

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	92.3	3	30.8	1.4	n.s.	3,21
Subject	1007.0	7	143.6	6.6	0.1%	7,21
Residual	453.7	21	21.6			
Total	1553.0	31				

Stage 4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	94.5	3	31.5	0.3	n.s.	3,21
Subject	10116.5	7	1445.2	15.7	n.s.	7,21
Residual	1933.0	21	92.1			
Total	12144.0	31				

Stage REM

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	574.8	3	191.6	3.05	n.s.	3,21
Subject	5411.0	7	773.0	12.30	0.1%	7,21
Residual	1320.2	21	62.9			
Total	7306.0	31				

Total Sleep Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	2395.4	3	798.5	2.5	n.s.	3,21
Subject	18597.3	7	2656.8	8.2	0.1%	7,21
Residual	6799.3	21	323.8			
Total	27792.0	31				

APPENDIX XI

Non-Significant ANOVA Summary Tables for the Aspirin Pilot Study

Stage W+1

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	577.6	4	144.4	1.7	ns	4,28
Subject	651.1	7	93.0	1.1	ns	7,28
Residual	2400.8	28	85.7			
Total	3629.5	39				

Stage 2

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	736.5	4	184.1	1.2	ns	4,28
Subject	5270.7	7	753.0	5.0	1%	7,28
Residual	4206.7	28	150.2			
Total	10213.9	39				

Stage 3

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	338	4	84.5	0.5	ns	4,28
Subject	3489	7	498.0	2.7	2.5%	7,28
Residual	5099	28	182.0			
Total	8926	39				

Stage 3+4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	2476.4	4	619.1	2.4	ns	4,28
Subject	6614.8	7	945.0	3.6	1%	7,28
Residual	7354.8	28	262.7			
Total	16446.0	39				

APPENDIX XI ctd.

Stage REM

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	657	4	164.3	1.2	ns	4,28
Subject	5772	7	824.6	6.0	0.1%	7,28
Residual	3864	28	138.0			
Total	10293	39				

REM Latency

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	3042	4	760.5	0.4	ns	4,28
Subject	14749	7	2107.0	1.1	ns	7,28
Residual	53297	28	1903.5			
Total	71088	39				

Total Sleep Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	347	4	86.8	0.4	ns	4,28
Subject	50553	7	7221.9	36.6	0.1%	7,28
Residual	5524	28	197.3			
Total	56424	39				

Sleep Onset Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Night	182	4	45.5	0.8	ns	4,28
Subject	2411	7	344.4	6.2	0.1%	7,28
Residual	1568	28	56.0			
Total	4161	39				

APPENDIX XII

Non-Significant ANOVA Summary Tables for the Aspirin Main Study over the drug nights.

Stage W+1

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	46.1	1	46.1	0.2	ns	1,14
Subjects within treatments	2934.1	14	209.6			
Within Subjects						
Nights	47.3	2	23.7	0.3	ns	2,28
Nights x Treatments	114.0	2	57.0	0.7	ns	2,28
Residual	2300.0	28				
Total	5441.5	47				

Stage 3

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	143.5	1	143.5	1.4	ns	1,14
Subjects within treatments	1436.5	14	102.6			
Within Subjects						
Nights	124.1	2	62.1	0.8	ns	2,28
Nights x Treatments	83.3	2	41.7	0.6	ns	2,28
Residual	2083.9	28	74.4			
Total	3871.3	47				

APPENDIX XII ctd.

Stage 4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	660.1	1	660.1	2.0	ns	1,14
Subjects within treatments	4693.8	14	335.3			
Within Subjects						
Nights	179.3	2	89.7	0.8	ns	2,28
Nights x Treatments	78.8	2	39.4	0.3	ns	2,28
Residual	3263.9	28	116.6			
Total	8875.9	47				

Stage 3+4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	1250.5	1	1250.5	3.7	ns	1,14
Subjects within treatments	4775.0	14	341.1			
Within Subjects						
Nights	11.4	2	5.7	.04	ns	2,28
Nights x Treatments	12.0	2	6.0	.04	ns	2,28
Residual	4125.9	28	147.4			
Total	10174.8	47				

APPENDIX XII ctd.

Stage REM

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	414.2	1	414.2	1.3	ns	1,14
Subjects within treatments	4649.3	14	332.1			
Within Subjects						
Nights	513.5	2	256.8	2.3	ns	2,28
Nights x Treatments	90.5	2	45.3	0.4	ns	2,28
Residual	3035.3	28	108.4			
Total	8702.8	47				

REM latency

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	4860.2	1	4860.2	4.0	ns	1,14
Subjects within treatments	16965.5	14	1211.8			
Within Subjects						
Nights	2037.1	2	1018.6	1.2	ns	2,28
Nights x Treatments	259.6	2	129.8	0.2	ns	2,28
Residual	22981.9	28	820.8			
Total	47104.3					

REM Periodicity

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	117.2	1	117.1	2.2	ns	1,14
Subjects within treatments	9541.6	14	52.4			
Within Subjects						
Nights	947.8	2	473.9	2.1	ns	2,28
Nights x Treatments	525.1	2	262.6	1.2	ns	2,28
Residual	6411.8	28	229.0			
Total	17543.5	47				

Number of Stage Changes

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	3.0	1	3.0	.03	ns	1,14
Subjects within treatments	1503.7	14	107.4			
Within Subjects						
Nights	116.4	2	58.2	.97	ns	2,28
Nights x Treatments	2.4	2	1.2	.02	ns	2,28
Residual	1684.6	28	60.2			
Total	3310.0	47				

APPENDIX XIII

Non-Significant ANOVA Summary Tables for the Aspirin Main Study over the recovery nights.

Stage W+1

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	148.6	1	148.6	1.2	ns	1,14
Subjects within treatments	1677.8	14	119.8			
Within Subjects						
Nights	103.1	4	25.8	0.4	ns	4,56
Nights x Treatments	76.2	4	19.1	0.3	ns	4,56
Residual	3308.3	56				
Total	5314.0	79				

Stage 2

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	292.6	1	292.6	0.3	ns	1,14
Subjects within treatments	12179.5	14	870.0			
Within Subjects						
Nights	861.6	4	215.4	0.8	ns	4,56
Nights x Treatments	227.6	4	56.9	0.2	ns	4,56
Residual	14866.4	56	265.5			
Total	28427.7	79				

APPENDIX XIII ctd.

Stage 3

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	11.3	1	11.3	0.07	ns	1,14
Subjects within treatments	2166.8	14	154.8			
Within Subjects						
Nights	171.1	4	42.8	1.0	ns	4,56
Nights x Treatments	118.1	4	29.5	0.7	ns	4,56
Residual	2512.8	56	44.9			
Total	4980.0	79				

Stage 4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	63.0	1	63.0	0.1	ns	1,14
Subjects within treatments	6320.3	14	451.5			
Within Subjects						
Nights	264.4	4	66.1	0.4	ns	4,56
Nights x Treatments	663.4	4	165.9	1.0	ns	4,56
Residual	9283.4	56	165.8			
Total	16594.5	79				

Stage 3+4

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	12.8	1	12.8	0.03	ns	1,14
Subject within treatment	7088.6	14	506.3			
Within Subjects						
Nights	83.6	4	20.9	0.1	ns	4,56
Nights x Treatments	662.6	4	165.6	0.8	ns	4,56
Residual	11543.4	56	206.1			
Total	19391.0	79				

Stage REM

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	171.1	1	171.1	0.3	ns	1,14
Subject within treatment	9221.1	14	658.7			
Within Subjects						
Nights	670.3	4	167.6	1.0	ns	4,56
Nights x Treatments	197.3	4	49.3	0.3	ns	4,56
Residual	9807.6	56	176.1			
Total	20067.4	79				

APPENDIX XIII ctd.

REM Periodicity

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	515.1	1	515.1	1.2	ns	1,14
Subject within treatment	5840.5	14	417.2			
Within Subjects						
Nights	1360.4	4	340.1	1.5	ns	4,56
Nights x Treatments	1842.6	4	460.6	2.0	ns	4,56
Residual	12760.7	56	227.9			
Total	22319.2	79				

Total Sleep Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	277.5	1	277.5	0.4	ns	1,14
Subject within treatment	9693.4	14	689.7			
Within Subjects						
Nights	2106.7	4	526.7	2.9	ns	1,19
Nights x Treatments	1091.8	4	273.0	1.5	ns	1,19
Residual	10115.5	56	180.6			
Total	23284.9	79				

Conservative Test

Sleep Onset Time

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	0.6	1	0.6	0	ns	1,14
Subject within treatment	9168.7	14	654.9			
Within Subjects						
Nights	75.6	4	18.9	0.3	ns	4,56
Nights x Treatments	114.8	4	28.7	0.4	ns	4,56
Residual	3678.0	56				
Total	13037.7	79				

Number of Stage Changes

Source	S.S.	d.f.	M.S.	F	P	df_1, df_2
Between Subjects						
Treatment	64.8	1	64.8	0.3	ns	1,14
Subject within treatments	2661.0	14	190.1			
Within Subjects						
Nights	247.3	4	61.8	0.9	ns	4,56
Nights x Treatments	107.8	4	27.0	0.4	ns	4,56
Residual	3921.3	56	70.0			
Total	7002.2	79				

GENSTAT Summary Table - Drug Nights

***** ANALYSIS OF VARIANCE *****

VARIATE: TIME

SOURCE OF VARIATION	DF	SS	SSX	MS	VR	df_1, df_2	
SUBJECT STRATUM							
DRUG	1	0.0	0.00	0.0	0.000	1, 14	n.s.
RESIDUAL	14	45.1	0.10	3.2	0.024		
TOTAL	15	45.1	0.10	3.0	0.022		
SUBJECT.NIGHTS.STRATUM							
NIGHTS	2	10.2	0.02	5.1	1.536	2, 28	n.s.
NIGHTS.DRUG	2	9.3	0.02	4.6	1.396	2, 28	n.s.
RESIDUAL	28	93.3	0.20	3.3	0.024		
TOTAL	32	112.8	0.25	3.5	0.026		
SUBJECT.STAGES.STRATUM							
STAGES	4	4455.2	9.72	1113.8	3.345	4, 56	P≤.05
STAGES.DRUG	4	5457.8	11.91	1364.4	4.098	4, 56	P≤.01
RESIDUAL	56	18645.5	40.68	333.0	2.444		
TOTAL	64	28558.5	62.31	446.2	3.275		
SUBJECT.NIGHTS.STAGES.STRATUM							
NIGHTS.STAGES	8	1353.5	2.95	169.2	1.242	8, 112	n.s.
NIGHTS.STAGES.DRUG	8	504.8	1.10	63.1	0.463	8, 112	n.s.
RESIDUAL	112	15250.6	33.29	136.2			
TOTAL	128	17117.9	37.35	133.7			
GRAND TOTAL	239	45834.3	100.00				

GRAND MEAN 0.08

TOTAL NUMBER OF OBSERVATIONS 240

***** TABLES OF MEANS *****

VARIATE: TIME

GRAND MEAN 0.08

STAGES	S1	S2	S3	S4	REM
	0.98	6.35	-0.13	-7.17	0.38

NIGHTS	DRG1	DRG3	DRG4
	-0.08	-0.05	0.37

DRUG	1	2
	0.08	0.08

STAGES / NIGHTS	S1	S2	S3	S4	REM
DRG1	1.69	2.62	-2.19	-5.25	2.75
DRG3	1.50	10.56	1.75	-9.81	-4.25
DRG4	-0.25	5.87	0.06	-6.44	2.63

DRUG / STAGES	1	2
S1	-0.58	2.54
S2	15.79	-3.08
S3	-1.79	1.54
S4	-10.00	-4.33
REM	-3.00	3.75

DRUG / NIGHTS	1	2
DRG1	-0.18	0.02
DRG3	-0.23	0.12
DRG4	0.65	0.10

STAGES	S1		S2		S3		S4		REM	
DRUG	1	2	1	2	1	2	1	2	1	2
NIGHTS										
DRG1	-0.00	3.37	11.00	-5.75	-5.00	0.63	-6.75	-3.75	-0.13	5.63
DRG3	-1.75	4.75	22.62	-1.50	1.87	1.63	-14.38	-5.25	-9.50	1.00
DRG4	-0.00	-0.50	13.75	-2.00	-2.25	2.37	-8.88	-4.00	0.62	4.63

GENSTAT Summary Table – Recovery Nights

***** ANALYSIS OF VARIANCE *****

VARIATE: TIME

SOURCE OF VARIATION	DF	SS	SS%	MS	VR	df_1, df_2	
SUBJECT STRATUM							
DRUG	1	0.4	0.00	0.4	0.170	1, 14	3
RESIDUAL	14	34.9	0.05	2.5	0.014		
TOTAL	15	35.3	0.05	2.4	0.013		
SUBJECT.NIGHTS STRATUM							
NIGHTS	4	12.2	0.02	3.0	1.626	4, 56	33.
NIGHTS.DRUG	4	7.5	0.01	1.9	1.008	4, 56	
RESIDUAL	56	104.7	0.14	1.9	0.011		
TOTAL	64	124.4	0.16	1.9	0.011		
SUBJECT.STAGES STRATUM							
STAGES	4	591.1	0.77	147.8	0.261	4, 56	33.
STAGES.DRUG	4	1078.7	1.41	269.7	0.476	4, 56	
RESIDUAL	56	31727.5	41.40	566.6	3.204		
TOTAL	64	33397.2	43.58	521.8	2.951		
SUBJECT.NIGHTS.STAGES STRATUM							
NIGHTS.STAGES	16	2093.4	2.74	131.2	0.742	16, 112	33.
NIGHTS.STAGES.DRUG	16	1371.8	1.79	85.7	0.485	16, 112	
RESIDUAL	224	39607.3	51.68	176.8			
TOTAL	256	43077.6	56.21	168.3			
GRAND TOTAL	399	76634.5	100.00				

GRAND MEAN 0.13

TOTAL NUMBER OF OBSERVATIONS 400

Appendix XV ctd.

***** TABLES OF MEANS *****

VARIATE: TIME

GRAND MEAN 0.13

STAGES	S1	S2	S3	S4	REM
	-0.26	2.49	-0.50	-0.11	-0.97

NIGHTS	RECV1	RECV2	RECV3	RECV4	RECV6
	0.35	0.05	0.02	0.31	-0.10

DRUG	1	2
	0.10	0.16

STAGES / NIGHTS	S1	S2	S3	S4	REM
RECV1	-0.06	2.87	0.19	-2.00	0.75
RECV2	1.31	3.94	1.88	-2.31	-4.56
RECV3	0.56	1.69	-2.50	1.12	2.63
RECV4	-1.19	7.75	-0.94	0.19	-4.25
RECV6	-1.94	-0.44	-1.13	2.44	0.56

DRUG / STAGES	1	2
S1	-1.90	1.38
S2	4.47	0.50
S3	-0.87	-0.12
S4	1.65	-1.87
REM	-2.87	0.95

DRUG / NIGHTS	1	2
RECV1	0.38	0.33
RECV2	0.03	0.08
RECV3	0.03	0.05
RECV4	0.03	0.60
RECV6	0.03	-0.24

STAGES	S1		S2		S3		S4		REM	
DRUG / NIGHTS	1	2	1	2	1	2	1	2	1	2
RECV1	0.75	-0.87	5.00	2.75	0.38	0.00	-0.50	-3.50	-1.75	3.25
RECV2	-1.00	3.63	5.25	2.63	3.50	0.25	0.00	-4.63	-7.62	-1.50
RECV3	-1.88	3.00	1.00	-4.37	-3.63	-1.38	5.25	-3.00	-0.62	5.88
RECV4	-4.00	1.63	8.38	7.13	-2.88	1.00	4.63	-4.25	-6.00	-2.50
RECV6	-5.38	-0.50	4.75	-5.63	-1.75	-0.50	-1.13	6.00	1.63	-0.50

APPENDIX XVI

Non-Significant ANOVA Summary Table for the Paracetamol Pilot Study

Stage REM

Source	SS	d.f.	MS	F	P	df_1, df_2
Between Subjects						
Treatment	220.6	1	220.6	1.1	ns	1,10
Subjects within treatments	1997.3	10	199.7			
Within Subjects						
Nights	315.8	2	157.9	2.4	ns	2,20
Nights x Treatments	280.1	2	140.1	2.2	ns	2,20
Residual	1278.6	20	63.9			
Total	4092.4	35				

APPENDIX XVII

Placebo Tablet Composition

Maize Starch	51.85%
Lactose	47.25%
Aerosil 200*	0.225%
Magnesium Stearate B.P.	0.675%
	100.00%

Shape: Normal Concave

Weight: 315 mg.

* Aerosil 200 is a form of colloidal silicon dioxide permitted for human consumption.

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