OXYGEN CONSUMPTION, CIRCADIAN RHYTHMICITY AND SLEEP

OXYGEN CONSUMPTION, CIRCADIAN RHYTHMICITY AND SLEEP

by

John

Geoffrey J. Fraser, B.Sc., B.A.(Hons)

in the Department of Psychology

Submitted in fulfilment of the requirements

of the degree of Master of Arts

University of Tasmania

August, 1989.

I certify that this thesis contains no material which has been accepted for the award of any other higher degree or graduate diploma in any university, and that to the best of my knowledge and belief the thesis contains no copy or paraphrase of material previously published or written by another person, except where due reference is made in the text of the thesis.

ABSTRACT

Oxygen consumption is lower during sleep than relaxed wakefulness. However, there is disagreement as to the particular metabolic changes which produce the difference. The present study assessed

(i) the contribution of sleep, circadian cycle and the specific dynamic action effect of the evening meal, to the fall in metabolic rate during the sleep period.

(ii) the effects of sleep stage on oxygen consumption which had been suggested by previous researchers, and

(iii) the effect of body movement arousals on oxygen consumption.

Five subjects were tested for a total of nine nights under three conditions in a repeated measures design. Subjects were confined to bed throughout their usual sleep period, but were allowed to go to sleep 0, 3 or 6 hours following their usual time for lights out. Oxygen consumption was measured in all conditions for the half hour before and after each of the times for lights out and then throughout the sleep period following lights out. The results demonstrated that changes in energy expenditure during the sleep period are a function of both sleep and circadian cycle. In this study the contribution of the components was approximately equal. However, the effect of sleep was rapid, with oxygen consumption values reaching an asymptote within fifteeen minutes of sleep onset, while the effect of circadian cycle was constant over the assessment period. No evidence was found implicating the specific dynamic action effect of the evening meal in the reduction in sleep period metabolic rate. The results of previous studies can be interpreted as being due to the combined effect of circadian and sleep influences, and not to specific differences in metabolic rate between separate sleep stages. These would appear to be artifactual. In addition, the period of metabolic disturbance following a movement arousal was shown to be longer than that suggested by previous researchers.

ACKNOWLEDGEMENTS

To Iain Montgomery, my supervisor I extend my thanks for continued encouragement and advice. I am greatly indebted to my former supervisor John Trinder for his guidance, support and perseverance.

To my colleague Ian Colrain I express my gratitude. He has been a source of great wisdom and amusement and a fine example of a healthy and successful attitude toward research and life.

Enthusiastic help was provided by Joanne, Amanda, Donna, Felicity and Melissa, who all helped brighten a seldom boring laboratory. I thank Carol for the opportunity to test the capabilities of her computer and for just being herself. I also thank my flatmate Jason for reading and not always understanding.

The data collection was made easier by the excellent support of the technical staff, John, Brian, Steve, Bob and Richard, and the talented programmers Peter and the afore-mentioned Ian.

I wish to thank my subjects, who endured long for small reward, but provided much to the study.

Finally I would like to thank my family who continued to support me in my chosen career, although at times even they must have wondered if the end was in sight.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER ONE

INTRODUCTION

CHAPTER ONE

INTRODUCTION

Whilst there is no disputing the benefit derived from a good night's sleep, there is considerable debate as to the function of sleep. Although initial attempts to collect data used observational techniques, the development of more accurate measures of brain activity via the electroencephalogram (EEG), provided new avenues in sleep research. As a result, the discovery that sleep was not an homogeneous state, but rather comprised several distinct EEG-defined phases, led to the postulation of specific functions for the separate sleep stages. Two major theories which have been proposed, view the function of sleep in terms of overall energy balance with the fall in energy expenditure during sleep being a critical component of both theories. The present investigation is not a comparative test of these theories, because the reduction in oxygen consumption observed during sleep can be used as support for both theories. Rather, an examination of these theories provides a framework for a test of the claim that the level of oxygen consumption is reduced during sleep, and that slow wave sleep (SWS) has some particular property which results in it having the lowest level of oxygen consumption observed during sleep.

First, energy conservation theorists contend that sleep is a means of conserving energy above that gained through relaxed wakefulness. While sleep generally is thought to be energy-conserving, SWS is hypothesised to be particularly so. Several lines of evidence have been used in support of the theory. States such as daily torpor and hibernation, which clearly have an energy conservation function, show characteristics similar to those of sleep, particularly SWS (Walker, Garber, Berger & Heller, 1979; Walker, Haskell, Berger, & Heller, 1981). Torpor and hibernation are thought to have evolved in association with the development of homeothermy (Berger, 1975). This evolution of SWS in parallel with homeothermy in higher

animals (Allison & van Twyver, 1970), further suggests a conservation function for SWS (Berger, 1984). Other evidence in support of the notion that sleep is energy conserving has come from the finding of lower metabolic rate (MR) during sleep, the lowest MR being observed in SWS (Brebbia & Altschuler, 1965; Haskell, Palca, Walker, Berger & Heller, 1981a; Shapiro, Goll, Cohen & Oswald, 1984). An inverse relationship between daily energy expenditure and both the amount of SWS (Baekland & Lasky, 1966; Griffin & Trinder, 1978) and total sleep time (TST) (Montgomery, Trinder & Paxton, 1982) is also consistent with this hypothesis. However, there is also evidence which conflicts with the view that increased daytime energy expenditure results in an increase in the amount of SWS and TST during the subsequent sleep period (e.g. Haskell, Palca, Walker, Berger & Heller, 1981b; Paxton, Trinder, Shapiro, Adam, Oswald & Graf, 1984).

Second, restorative theorists argue that there is a need for restoration following periods of catabolic activity. It is proposed that the major function of sleep is protein synthesis following protein catabolism, as a result of activity during the previous period of wakefulness (Adam, 1980; Hartman, 1973; Oswald, 1970). The mechanism of restoration may ultimately be reduced to a cellular level with the notion of an Energy Charge (EC) providing an explanation of cellular regulation of energy transfer (Adam & Oswald, 1977). At an organismic level, an increase in energy expenditure during awake periods with its associated increase in catabolism should lead to greater restoration during inactive periods. Therefore, during sleep following a period of greater than usual daytime catabolism, there should be a resultant increase in the amount of SWS and TST. The longer than usual period of low MR during sleep would enable greater restoration as energy would become available for protein synthesis. The effect on sleep of a number of factors known to increase daytime catabolism has also been investigated. For example, patients suffering from hyperthyroidism (Oswald, Dunleavy & Strong, 1972), and anorexic patients upon

gaining weight (Lacey, Crisp, Kalucy, Hartmann & Chen, 1975), both exhibit increased amounts of SWS. In addition, sleep deprivation leads to an increase in SWS on recovery nights (Williams Agnew & Webb, 1964) and daytime exercise results in an increase in SWS (Baekland & Lasky, 1966; Zloty, Burdick & Adamson, 1973). There is also evidence that sleep, particularly SWS, is associated with a pattern of physiological activity which is conducive to synthetic processes. The level of human growth hormone (hGH), a substance known to promote protein synthesis (Korner, 1965), increases during sleep (Adamson, Hunter, Ogunremi, Oswald & Percy-Robb, 1974; Honda, Takahashi, Takahashi, Azumi, Irie, Sakuma, Tsushima & Shizume, 1979). Moreover, administration of amphetamine, which increases metabolic rate, also leads to an increase in the level of hGH (Besser, Butler, Landon &Rees, 1969; Dunleavy, Oswald & Strong, 1973). Thus, proponents of both theories view the reduction in energy expenditure observed during sleep as leading to benefits. Chapter Two discusses the energy conservation and restorative theories in more detail.

Central to both theories is the belief that the reduction in energy expenditure observed during sleep is a unique property of sleep. Further, it is argued that energy expenditure is particularly low during SWS. However, while it is clear that there is a reduction in MR during sleep (Buskirk, Thompson, Moore & Whedon, 1960; Colrain, Trinder, Fraser & Wilson, 1987; White,Weil & Zwillich, 1985), there remains speculation as to the factors involved. Thus it is unclear if the reduction is due to sleep itself, to circadian variation in MR or to factors which are temporally associated with sleep, such as metabolism associated with the evening meal. These factors are discussed in Chapter Three. Further, there is some doubt as to the validity of the belief that MR is particularly low during SWS. Several studies have measured MR during specific stages of sleep. The findings, although inconsistent, have been interpreted as showing low MR during SWS and are used in support of both the

energy conservation and restoration theories. These studies are discussed in Chapter Three and alternative explanations for their findings are forwarded.

The present study is then described in detail in Chapter Four. The results of the experiment are presented in Chapter Five. The findings are discussed in Chapter Six, which concludes with comments on the implications of the findings for future research.

CHAPTER TWO

THEORIES OF SLEEP:
ENERGY CONSERVATION AND RESTORATION

CHAPTER TWO
THEORIES OF SLEEP:
ENERGY CONSERVATION AND RESTORATION

The present chapter consists of an examination of energy conservation and restorative theories as they pertain to the function of sleep. Although not mutually exclusive, the theories will be presented separately. There will be an analysis of the underlying assumptions and arguments for and against each theory followed by a brief summary. The chapter concludes with a brief statement of the relevance of the theories to the present study. It should be noted that the results of some of the studies investigating MR during sleep following daytime exercise have been used as evidence for both the conservation and restorative theories. Therefore, both sections of this chapter include the relevant supporting and contradictory evidence drawn from the literature relating to the effects of exercise on subsequent sleep.

2.1 Energy conservation theory.

When an organism's energy store is depleted, and it faces a demand which may result in a threat to life, conservation of energy resources becomes paramount to survival. Until the organism replenishes its energy store, it must attempt to overcome the negative consequences of energy expenditure. It has been proposed that during periods of energy stress, the organism needs to conserve energy. Energy stress may be caused endogenously by normal daily energy expenditure or exogenously due to limited availability of food, change in ambient temperature etc.

With respect to sleep, energy conservation theorists argue that sleep itself is energy conserving. Energy conservation is identified by a low MR during sleep and this reduction in sleep period MR is associated with an increase in SWS and TST. There is no question that MR is low during sleep (refer to Chapter Three). However,

it is argued that SWS is characterized by a particularly low MR, a situation which makes it especially energy conserving. Whether MR is low during SWS relative to the rest of sleep is open to debate. In addition, the question is not only whether MR falls during sleep, but also whether the magnitude and the duration of the fall, is systematically related to energy requirements, i.e. to energy balance. This question can be assessed at several levels; phylogenetically, across members of a species, and within an individual member of a species.

There is considerable evidence which is used in support of the energy conservation theory. Sleep appears to be a characteristic of animals with high cortical development, such as birds and mammals. Both sleep with rapid eye movements (REM) and sleep characterised by an electrophysiological pattern of large slow waves, have been found in birds and all higher mammals studied in the laboratory (Allison & Van Twyver, 1970). Neither amphibians nor reptiles appear to exhibit sleep as it is defined for higher animals, but rather alternate between periods of quiet and active wakefulness as a function of ambient temperature and circadian rhythmicity. This pattern of behaviour arises because both amphibians and reptiles are poikilothermic, their body temperature adjusting to ambient temperature as they are unable to produce enough heat through their own metabolic processes. In contrast, birds and mammals are homeothermic, maintaining a relatively constant body temperature, ranging from 36 to 40^{o}C, via the generation of heat through high rates of metabolism (Berger, 1984).

Poikilotherms are able to reduce energy expenditure in situations of low ambient temperature and limited availability of food by reverting to a condition of relaxed inactivity with an associated reduction in body temperature. In contrast, during inactivity homeotherms still expend considerable energy in maintaining body temperature at a euthermic level. Berger (1984) argues that during evolution, the

energy demands of homeothermy were compensated for by the development of a low energy state. Phylogenetic studies of mammals and birds suggest a close interrelationship between the development of SWS and homeothermy (Allison & Van Twyver, 1970; Berger, 1975). Therefore SWS may be the major factor in providing a low energy state during sleep, to offset the energy demands of homeothermy.

Some homeotherms enter a state of torpor during times of food shortage or low ambient temperature as a means of conserving energy (Berger, 1984). This represents a reversion to a type of partial poikilothermia during which core body temperature equilibrates with ambient temperature down to a certain point, with the set point characteristic to the type of mammal. Shallow torpor is exhibited in some birds and many small mammals and may follow a diurnal cycle, in which case it is referred to as daily torpor (Berger, 1984). In contrast, deep torpor (more commonly referred to as hibernation) occurs in only a few mammalian species, in response to extreme seasonal reductions in temperature and availability of food (Berger, 1984). The main feature of both types of torpor is a decline in deep body temperature associated with a reduction in metabolic rate and consequent conservation of energy (Heller, Walker, Florant, Glotzbach & Berger, 1978).

In many cases, these states of reduced metabolism appear to be entered via sleep (Heller & Glotzbach, 1977; Heller et al., 1978; Shimizu & Himwich, 1968; Walker & Berger, 1980; Walker, Glotzbach, Berger & Heller, 1977) and there may be a continuum in both electrophysiological activity and thermoregulatory adjustment from sleep to hibernation (Heller, 1979; Heller et al., 1978). In fact, during daily torpor and hibernation, brain electrical activity is characterised by the presence of slow waves which are isomorphic with those present in normal SWS (Walker et al., 1979; Walker et al., 1981). These findings support the notion that SWS may provide a daily source of energy conservation by performing a compensatory function similar to

the processes of daily torpor and hibernation (Berger, 1975; Berger, 1984; Heller et al., 1978; Walker & Berger, 1980; Walker et al., 1979; Walker et al., 1981).

Support for the conceptualization of sleep as an energy conserving state has come from a correlational study which reported that species whose waking activity is relatively expensive in terms of energy expenditure, tend to spend a greater proportion of their time asleep (Zepelin & Rechtschaffen, 1974). It has also been found that nocturnal sleepers with longer TST have higher daytime core body temperatures, and an increased daytime MR, in contrast to sleepers with shorter TST (Taub & Berger, 1976). These findings suggest that in animals which have a higher MR, there exists a greater need for energy conservation. It has also been found that body weight has a strong negative correlation with MR (Alison & Cicchetti, 1976). In addition, body weight and brain weight both have a strong negative correlation with SWS, and MR has a strong positive correlation with SWS (Alison & Cicchetti, 1976). Those animals with increased cortical development (usually larger animals) spend periods of time in relaxed wakefulness and would therefore, in terms of energy conservation, derive less benefit from sleep, particularly SWS (Horne, 1977). However, Zepelin and Rechtschaffen (1974) argue that SWS provides a mechanism for enforcing rest and hence conserving energy.

Across members of a species, the relationship between MR and sleep can be seen in the ontogenetic development of the two variables. Human babies, in contrast to adults, have an high daily MR followed by long periods of sleep (Berger, 1975). Also, the rate of both body and brain metabolism has been found to decline with increasing age (Feinberg & Carlson, 1968) and thus there may be an associated decrease in the need for sleep, particularly stage 4 sleep in older humans (Berger, 1975). Indeed, older humans have exhibited reduced stage 4 sleep and TST (Agnew, Webb & Williams, 1967b; Feinberg, 1974; Feinberg, Koresko & Heller, 1967; Kahn

& Fisher, 1969; Kales, Wilson, Kales, Jacobsen, Paulson, Kollar & Walter, 1967, Webb & Agnew, 1971).

Within a member of a species, changes in sleep and SWS can be observed following changes in daytime energy expenditure, and exercise has been used as a means of eliciting this change in energy expenditure. An investigation of the sleep of aerobically fit subjects showed that the subjects had increased SWS subsequent to daytime exercise, when compared with no exercise days (Baekland & Lasky, 1966; Shapiro, Griesel, Bartel & Jooste, 1975; Shapiro & Verschoor, 1979). Also, three recent studies which looked at sleep in highly trained young athletes following exercise at high rates of energy expenditure, found increased amounts of SWS following daytime exercise compared with no exercise days (Horne & Moore, 1985; Horne & Staff, 1983; Shapiro, Bortz, Mitchell, Bartel & Jooste, 1981). Further, starvation, a condition resulting in greater pressure to conserve energy, lead to an increase in SWS (Karacan, Rosenbloom, Londono, Salis, Thornby & Williams, 1973; MacFadyen, Oswald & Lewis, 1973; Parker, Rossman & VanderLaan, 1972).

However, while the evidence, from between and within species comparisons, for an energy conservation role for sleep is compelling, the evidence from within individual studies, particularly in humans, is more contradictory. Some studies have found no increase in SWS following daytime exercise (Adamson et al., 1974; Browman & Tepas, 1976; Desjardins, Healey & Broughton, 1974; Hauri, 1968; Walker, Floyd, Cavness, Lualhati & Feinberg, 1978; Zir et al., 1971). Also, two studies which combined sleep deprivation with exercise failed to find an exercise effect (Webb & Agnew, 1973; Webb & Agnew, 1974). Trinder, Montgomery & Paxton (1988) report that a number of studies produced ambiguous results and suggest, as did Bunnell, Bevier & Horvath (1984) that possibly this reflected a temporal redistribution of SWS components, or exercise altering the timing of the

sleep cycles rather than potentiating SWS. In addition, when ambient temperature is increased or decreased relative to thermoneutrality, producing an increase in MR, there is a reduction in the amount of SWS obtained during sleep under these conditions (Haskell et al., 1981b).

Despite the conflicting evidence, energy conservation theorists maintain that the primary function of sleep is to conserve energy and that the main vehicle for this process is SWS. The immobilisation of the organism during sleep results in a reduction of metabolic requirements and this, combined with the associated reduction in body temperature decreases the energy demands of homeothermy, thus conserving energy. This process is possible because sleep, in particular SWS, has the property of low MR and thus in conditions where there exists a greater pressure to conserve energy, more SWS occurs.

2.2 Restorative theory.

Proponents of the restorative theory argue that following periods of catabolism, an organism needs to replenish its energy store via the buildup, through a process of synthesis, of a store of usable metabolites. Animals experience cycles of activity and inactivity and associated degradative/synthetic rhythms. The differing energy requirements of the activity/inactivity cycle mainly determine the relative positioning of the degradative/synthetic rhythm such that the synthetic period inevitably coincides with inactivity or rest (Adam, 1980). This period of synthesis allows the animal to rebuild its energy supplies. Whereas in wakefulness the metabolic conditions associated with activity ensure degradation has priority, in sleep the balance shifts towards synthetic processes, with sleep providing more suitable conditions for restoration than relaxed wakefulness (Adam & Oswald, 1977).

The study of energy expenditure during sleep is relevant to the restorative theory because a low MR during sleep has been interpreted as support for the theory. It is proposed that the periodic reduction in metabolic demand provided by sleep facilitates bodily restoration by allowing energy to be diverted to synthetic processes to produce a nett gain of usuable metabolites (Adam, 1980; Hartman, 1973; Horne, 1983; Oswald, 1970, 1976, 1980). SWS is believed to be the most effective component of sleep for restorative purposes because it is thought to have a low MR relative to other sleep stages and thus provides greater energy resources for protein synthesis.

What evidence is there to support the restorative theory? First, sleep appears to be associated with conditions of high cellular energy. The concept of a cellular 'energy charge' (EC) being an indicator of the energy potential of a cell, was introduced by Atkinson (1968) and the EC is believed to play a fundamental role in the coordination of cellular metabolism (Adam & Oswald, 1983). Energy transfer within the cell is effected through reactions involving adenosine triphosphate (ATP) and protein synthesis via ribosomal RNA. The EC is an index of ATP levels and therefore, available energy. Synthetic processes depend on ATP to energize them and are thus promoted by high levels of EC, which occur as the rate of cellular work decreases. High levels of EC act to limit cellular degradation, whilst low levels act to promote degradation and reduce synthesis, stabilizing the EC level (Adam & Oswald, 1977). Cellular oxygen consumption (and ultimately total oxygen consumption) is determined by the energy state of the cell and thus by intracellular ATP levels. Rest reduces the rate of ATP depletion, so oxygen consumption and hence MR, falls (Adam & Oswald, 1977). Indeed, it has been reported that sleep is associated with high cellular ATP levels (Van den Noort & Brine, 1970). In addition, nett protein synthesis occurs at times when low cellular work allows ATP and EC to rise to high levels and thus reduce degradation. Protein synthesis requires energy, but the absolute amounts are small compared with the cost of muscular work. Therefore, it

has been argued that low levels of MR during sleep enable a nett increase in protein synthesis due to both an increase in synthesis and a reduction in protein degradation, during sleep as compared to wakefulness (Adam, 1980).

Second, evidence has been found that sleep, particularly SWS, is associated with a pattern of physiological activity which is conducive to synthetic processes. Human growth hormone (hGH) has been found to promote protein synthesis (Korner, 1965). Secretion of hGH has been found to increase during sleep, in contrast to wakefulness, in both children (Honda et al., 1979) and adults (Adamson et al., 1974; Karacan, Rosenbloom, Williams, Finley & Hursch, 1971). In addition, relaxed wakefulness was found to induce only a small increase of approximately 17% in plasma hGH levels, whereas during sleep the levels increased by about 250% (Alford, Baker & Burger, 1973). Moreover, a significant increase in hGH secretion was observed to coincide with the onset of deep sleep (SWS) (Honda et al., 1979; Parker, Sassin, Mace, Gotlin & Rossman, 1969; Sassin, Parker, Mace, Gotlin, Johnson & Rossman, 1969; Takahashi, Kipnis & Daughaday, 1968) and hGH level appears to be at a maximum during SWS (Honda et al., 1979). SWS therefore appears to stimulate the production of hGH, which is known to promote protein synthesis.

Further support for the restorative theory is derived from the findings that:

(i) acute starvation leads to an increase in SWS (Adamson et al., 1974; Crisp, Stonehill, Fenton & Fenwick, 1973; MacFadyen et al., 1973) and a corresponding increase in hGH (Parker et al., 1972) and

(ii) amphetamine administration, which is associated with stimulation of metabolic activity, leads to an increase in hGH (Besser et al., 1969; Dunleavy et al., 1973) and an increase in SWS (Lewis, Oswald & Dunleavy, 1971; Rechtschaffen & Maron, 1964) during the following sleep period. However, the claim that hGH

levels implicate SWS as restorative, should be approached with caution as hGH is involved in a number of other physiological processes.

What evidence is there that protein synthesis is elevated during sleep? In rats, a higher rate of cerebral protein synthesis has been found to be associated with SWS (Ramm & Smith, 1987). Synthetic activity shows an increase during sleep, as indexed by, for example, an increase in the rate of mitosis (Adam & Oswald, 1977). Although most studies of this type have used infrahuman species as subjects, in man the rate of mitosis has been found to increase during sleep, in skin tissue (Fisher, 1968) and bone marrow (Mauer, 1965).

However, it can be argued that the association with sleep of peaks in mitotic activity, may be coincidental and not causal. The circadian peaks in mitosis may be time of day phenomena which appear even in the absence of sleep (Scheving, 1959). These peaks may even occur in the evening, prior to the sleep period (Fisher, 1968). The peaks may also be present as a consequence of activity, or be a reflection of the sleep-independent low levels of corticosteriod output (Horne, 1979). It is therefore conceivable that hGH might not play any major role in determining mitotic levels.

Third, the assumption of a high need for restoration following an increase in the duration, or intensity, of daily activity implies an increase in the amount of sleep.

(i) Sleep deprivation studies have assessed the extent of compensation for increased wakefulness. An increase in the amount of time spent awake results in an increase in catabolism above that of a normal length awake period and should therefore result in an increase in SWS during recovery sleep. In fact, sleep deprivation leads to an increase in total sleep duration and SWS on recovery nights, with increased deprivation resulting in larger SWS rebound (Berger & Oswald, 1962; Johnson, Slye & Dement, 1965; Rechtschaffen & Maron, 1964; Williams et al.,

1964). In addition, specific SWS deprivation leads to a SWS rebound, lethargy and physical discomfort (Agnew, Webb & Williams, 1967a). One hour of extra wakefulness during the night leads to an increase in SWS and hGH levels later in the night (Beck, Brezinova, Hunter & Oswald, 1975). However, this increase in SWS may only be a mechanism for obtaining a quota required under an homeostatic system (Agnew, Webb & Williams, 1964; Tilley, Donohoe & Hensby, 1987).

(ii) Studies with various patient groups suggest that high wake time catabolism gives more impetus to protein synthesis and hence resulting in more SWS. Patients with hyperthyroidism exhibit increased basal metabolic activity due to their overproduction of thyroxin (Terjung & Winder, 1975). They also demonstrate increased amounts of SWS as well as elevated nocturnal plasma hGH levels. These abnormalities gradually return to normal following treatment with, for example, the drug carbimazole (Oswald et al., 1972; Dunleavy, Oswald, Brown & Strong, 1974). The opposite pathological condition, hypothyroidism, is associated with significantly less SWS which returns to normal with the administration of thyroid hormone and the subsequent increase in the rate of energy expenditure and cellular degradation (Kales, Heuser, Jacobsen, Kales, Hanley, Zweizig & Paulson, 1967). Also, the increase in MR following weight gain in female anorexic patients is associated with an initial increase in SWS which diminishes on reaching a stable weight (Lacey et al., 1975).

(iii) The restorative theory has been supported by the claim that daytime exercise and hence an increase in catabolic activity, leads to an increase in SWS. A number of studies which investigated sleep in aerobically fit subjects following daytime exercise reported an increase in SWS relative to no exercise days (Baekland & Lasky, 1966; Horne & Moore, 1985; Horne & Staff, 1983; Shapiro et al., 1975; Shapiro et al., 1981; Shapiro & Verschoor, 1979). Also, two studies showed a positive redistribution of SWS with an increase in stage 4 at the expense of stage 3 (Bonnet,

1980; Maloletnev et al., 1977). A positive effect was also reported in two studies which used subjects who were either moderately fit or unfit (Bunnell et al., 1983; Matsumoto, Saito, Abe & Furumi, 1984).

It is unclear as to whether the effect of exercise on SWS is causal or is due to other factors. A number of studies have failed to show significant exercise effects, in both younger (Paxton, Montgomery, Trinder, Newman & Bowling, 1982; Kupfer, Sewitch, Epstein, Bulik, McGowen & Robertson, 1985; Paxton, Trinder & Montgomery, 1983; Trinder, Bruck, Paxton, Montgomery & Bowling, 1982; Montgomery, Trinder, Paxton & Fraser, 1987) and older subjects (Montgomery, Trinder, Paxton, Fraser, Meaney & Koebin, 1985; Montgomery et al., 1987; Torsval, Akerstedt & Lindbeck, 1984). It was previously thought that the positive effect of exercise on SWS occurred only when exercise was of relatively long duration at high rates of energy expenditure (Griffin & Trinder, 1978; Horne, 1981).

The systematic variation of a number of potentially relevant factors has failed to show an exercise effect on SWS in any of the experimental conditions or subject samples. These factors include the amount and rate of energy expenditure (Horne & Staff, 1983; Kupfer et al., 1985; Paxton et al., 1982), the nature of the exercise (Montgomery et al., 1988), and the physical fitness (Paxton et al., 1983), the age (Trinder et al., 1982; Montgomery et al., 1987) and sex (Montgomery et al., 1988) of the subjects.

It has been suggested by Horne (1981) that the critical factor in producing the exercise effect on sleep is the presence of an associated increase in body temperature which mediates an increase in brain temperature, stimulating brain metabolism and resulting in a compensatory increase in SWS. Studies in which an elevation of body temperature was produced by passive heating of the body have shown an increase in

SWS (Horne & Staff, 1983; Horne & Reid, 1985), and exercise with associated body heating produced an increase in SWS (Horne & Moore, 1985). Consistent with the body heating hypothesis, cooling of the subject's head during the exercise produced no increase in SWS (Horne & Moore, 1985). It is unclear whether an increase in the amount of SWS is brought about by either prolonged or acute exercise or perhaps mediation through body heating or a combination of the two.

2.3 Conclusions.

Both the energy conservation and restorative theories view sleep as serving a compensatory function. They predict that the relative amount of time spent in each stage during sleep will change in response to an increase in wake period energy expenditure (energy conservation) or catabolic activity (restoration). Each theory regards the reduction in MR during sleep as evidence of that response. Whether SWS has a lower MR which makes it particularly energy conserving or bodily restoring has not yet been resolved. This issue is discussed in Chapter Three.

CHAPTER THREE

METABOLIC RATE AND SLEEP

CHAPTER THREE

METABOLIC RATE AND SLEEP

3.1 Metabolic rate during sleep.

As stated previously, metabolic rate as indexed by oxygen consumption, has been shown to be lower during sleep than relaxed wakefulness (Brebbia & Altschuler, 1965; Buskirk et al., 1960; Colrain et al., 1987; Kreider & Iampietro, 1959; Robin , Whaley, Crump & Travis, 1958; Shapiro et al., 1984; White et al., 1985; Yamashiro, Fukushima, Okudaira, Suzuku & Nishi, 1987). Studies which have measured MR across the sleep period have found that MR falls gradually during sleep, reaches a minimum late in the night and then rises slightly prior to awakening (Buskirk et al., 1960; Haskell et al., 1981a; Kreider, Buskirk & Bass, 1958; Kreider & Iampietro, 1959; Shapiro et al., 1984; Webb & Hiestand, 1975; White et al., 1985). Some studies have emphasised the rapidity with which MR falls during the first hour of sleep (Kreider & Iampietro, 1959; White et al., 1985) or following sleep onset (Colrain et al., 1987). However, with the exception of one study (Colrain et al., 1987), in which the ten minutes following sleep onset were analysed, the time unit used for analysis has been too large to accurately determine the rapidity of the fall in MR in the early part of the sleep period.

Whilst there has been agreement as to the empirical relationship between MR and sleep, there have been three main suggestions as to the particular metabolic changes which might contribute to the observed effect. First, the fall in MR during sleep may be due to sleep itself, either because of a general reduction in metabolic activity (Shapiro et al., 1984) or a change in a specific mechanism, such as muscle activity (Kleitman, 1963; Kreider et al., 1958; Mason & Benedict, 1934), or thermoregulatory set point (Kreider & Buskirk, 1957; Kreider et al., 1958; Milan & Evonuk, 1967). A second view is that the changes during sleep are a consequence of

circadian variations in general metabolic activity (Grollman, 1930; White et al., 1985), or body temperature rhythm (Kleitman, 1963; Webb & Hiestand, 1975). Finally, it has been suggested that the reduction in MR during sleep may be due to the specific dynamic action effect (SDA) of food associated with the evening meal (Buskirk et al., 1960; Kreider et al., 1958) producing an initial increase in MR following the meal and then a gradual reduction over the sleep period.

3.2 Core body temperature during sleep.

It has been shown that deep body temperature exhibits a circadian rhythm. The existence of a diurnal rise and nocturnal fall in body temperature has been accepted since the nineteenth century, although researchers have differed in the temporal positioning of the maximum and minimum temperature (Conroy & Mills,1970). This variability was probably due to a difference in the phase relationship between the body temperature rhythm and the individual's sleep/wake cycle. Indeed, the characteristic pattern of body temperature for a given individual has been emphasised by Kleitman and Ramsaroop (1948). The time and duration of the maximum has been found to vary from person to person, but tend to be constant in any one individual (Horne, 1977).

Although activity levels, food intake and sleep have specific effects on temperature regulation, they do not generate the body temperature rhythm, but rather act to modulate the regulated temperature (Fuller & Sulzman, 1982). It has been postulated that the endogenous component of the body temperature cycle is one of two coupled oscillators (the other is the sleep/wake cycle), which form the basis of the overall circadian rhythm (Gander & Kronauer, 1984). Thus, the overt temperature rhythm is influenced by the alternation between sleep and wakefulness but is mainly controlled by the endogenous oscillator (Zulley & Wever, 1982). The circadian rhythm for temperature is superficially similar to that suspected for MR,

suggesting that variations in MR may depend on variations in core body temperature.

Consistent with this are the changes which occur in body temperature at sleep onset. Under normal conditions, body temperature decreases following sleep onset. Thereafter, there is a gradual reduction over the sleep period and a rise towards morning (Gillberg & Akerstedt, 1982). There appears to be a strong sleep evoked decrease in body temperature (Barrett, Morris & Lack, 1987; Lack, Balfour & Kalucy, 1985) and it has been found that this initial decrease in temperature occurs with sleep onset at different times of the day or night (Gillberg & Akerstedt, 1982). Also, the decrease in body temperature over the sleep period does not differ from the change observed during wakefulness (Gillberg & Akerstedt, 1982). Thus there appears to be a sleep evoked component and an endogenous component determining the body temperature rhythm (Barrett et al., 1987). As the pattern of MR across sleep appears to parallel that of the body temperature rhythm, it may be that there is a sleep evoked component and an endogenous component in the MR rhythm, two factors which may account for the observed reduction in MR during sleep.

3.3 The effect of sleep state on metabolic rate.

In addition to the possibility that the fall in MR during sleep is due to mechanisms specific to sleep, circadian variation, or an SDA effect, it has been suggested that the stages of sleep have specific effects on MR. Thus it is claimed that MR reaches its lowest level during SWS and is relatively elevated during REM sleep. However, examination of studies which recorded EEG data and MR during a night's sleep, revealed that the evidence relating MR to sleep stage, particularly SWS, is inconsistent. The relevant results of these studies are presented in Table 3.1.

Table 3.1: Results of previous studies measuring MR and sleep stage.

	METABOLIC RATE COMPARISON			
STUDY	S < W	SWS < 2	SWS < REM	NREM < REM
Brebbia & Altschuler (1965)	YES	YES*	YES*	YES*
Shapiro et al. (1984)	YES	YES	YES	NO
Haskell et al. (1981a)	#	YES	YES	YES
Yamashiro et al. (1986)	YES*	NO	YES	YES
Webb & Hiestand (1975)	#	NO	NO	NO
White et al. (1985)	YES	NO	NO	NO
Duron et al. (1968)	YES	NO	NO	NO

'S' refers to sleep and 'W' to wakefulness. Where significance levels were reported, these are indicated by * ($p<.05$). '#' signifies that results were not reported.

In some studies, MR was found to be lower in SWS than stage 2 (Brebbia & Altschuler, 1965; Shapiro et al., 1984), lower in SWS than REM (Brebbia & Altschuler, 1965; Shapiro et al., 1984; Yamashiro et al., 1986) and lower in NREM than REM (Brebbia & Altschuler, 1965; Haskell et al., 1981a; Yamashiro et al., 1986). The remaining studies found no change in MR between selected stages of sleep (Duron, Andrac & Laval, 1968; Webb & Hiestand, 1975; White et al., 1985). As noted previously (refer Chapter 2), the data from the studies which did find differences between stages have been used as support for both the restorative and conservation theories of sleep, such that it has been argued that SWS has specific characteristics which make it more beneficial than relaxed wakefulness or sleep generally. However, the effect of stage of sleep on MR is complicated by a number of methodological factors.

3.4 Methodological factors in the measurement of metabolic rate, as a function of sleep state.

The major problem in determining the metabolic rate associated with the various sleep stages has been that MR varies systematically over the sleep period, whereas the sleep stages themselves are not uniformly distributed over the night (Kahn & Fisher, 1969; Williams, Agnew & Webb, 1964, 1966).

Slow wave sleep occurs predominantly in the early part of the sleep period and REM predominates in the later part of the night. It has therefore been thought inappropriate to simply average MR across total minutes of a given sleep stage during the night. To overcome this problem, some studies have attempted by the use of various analysis procedures, to control for time of night effects. The methods of data analysis used by the studies are summarized in Table 3.2.

Table 3.2: Methods of data analysis used in previous studies

	STAGE COMPARISON			
STUDY	All Minutes	Contiguous Stages	Length of stage	Control measure exclusion criteria
Brebbia & Altschuler	Y	Y	> 15 min continuous	> 2 stages in 5 min epoch
Haskell et al.	Y	Y	> 10 min continuous	for 3 min after mvt/ch > 10 min W between sleep stages
Shapiro et al.	Y	Y	> 6 min continuous	for 2 min after mvt & where > 50% mvt
Yamashiro et al.	Y	N	> 2 min continuous	where mvt in epoch
Webb & Hiestand	Y	Y	15 minute period	> 7 min W in period
White et al.	Y	N	up to 25 min period	for 3 min after mvt/ch
Duron et al.	Method not reported.			

'Y' denotes yes and 'N' denotes no. 'W' refers to wakefulness and 'mvt' refers to body movement. 'ch' is the abbreviation for change.

The simplest method to compare sleep stages is to average MR for each stage over the entire night. Only two of seven studies which have investigated MR as a function of sleep stage, using all minutes of a stage during the night, have reported MR lowest during SWS (Brebbia & Altschuler, 1965; Shapiro et al., 1984). Conversely, the remainder of the studies failed to observe this result (Haskell et al., 1981a; Webb & Hiestand, 1975; White et al., 1985; Yamashiro et al., 1986). In order to avoid the problem outlined above, comparisons were made using contiguous stages, that is, temporally adjacent stages, including counterbalanced contiguous stages (Brebbia and Altschuler, 1965; Haskell et al., 1981a; Shapiro et al., 1984; and Webb and Hiestand, 1975). Three studies which used this method to control for time of night effects reported MR lowest during SWS (Brebbia and Altschuler, 1965; Haskell et al., 1981a; Shapiro et al., 1984).

The view was expressed that body movement could contribute significantly to artifact in psychophysiological studies of sleep (Altschuler and Brebbia, 1967). Of the studies which report their criteria for exclusion of periods of sleep contaminated by body movement, only Webb and Hiestand (1975) did not exclude the minutes during which the movement occurred. Yamashiro et al. (1986) excluded only the epoch contaminated by the movement, whilst Shapiro et al. (1984) excluded two minutes following the movement and Haskell et al. (1981a) excluded three minutes following the movement, however one minute of this exclusion period was included to cover the time lag involved in using a hood for collection of expired air. It is not clear for how long following a movement, MR is affected by the movement, but at least the epoch containing the movement should be excluded. Only one study commented on the effect of movement on MR, reporting that MR always returned to baseline within three minutes after a body movement (Haskell et al., 1981a). In addition to the duration of movement, the distribution of body movements has not been considered. Rather than occurring randomly, they occur frequently at the

transition from a stable period of stage 4 to stage 2 (Muzet, Naitoh, Townsend & Johnson, 1972). This could result in a higher MR in stage 2 which immediately follows stage 4. Thus even counterbalanced contiguous stages would be in danger of finding a difference between stages where there was none. In addition to this effect due to movement, a lack of movement could cause MR to be lower in stage 4 following stage 2. If, in comparisons, the length of time in undisturbed sleep was not controlled, then differences in MR may become apparent Therefore the effect of time of night and undisturbed sleep would favour a lower MR during stage 4 as would movement at the end of a period of stage 4. Therefore, for these reasons, and not because of some property of SWS, a lower MR could be observed in SWS than in stage 2.

There exist other methodological factors which differed between the studies, but which on the balance of the literature, are unlikely to be critically related to stage differences although they may introduce additional variability into the data. They are briefly discussed and are listed in Table 3.3.

Table 3.3: Methods of data collection and number and sex of subjects used in previous studies.

	MEASUREMENT CONDITIONS			SUBJECTS	
STUDY	Oxygen Device	MR Episode(min)	EEG Epoch	Age (yrs)	Number and sex
Brebbia & Altschuler.	Hood (C)	5	5 min	19-35	5 M
Haskell et al.	Hood (C)	#	30 sec	18-30	6 M
Shapiro et al.	Hood (A)	2	20 sec	19-29	4 M
Yamashiro et al.	Mask (C)	>2	20 sec	21-23	8 M
Webb & Hiestand.	Mask (C)	15	30 sec	19-63	20 M
White et al.	Mask (C)	#	20 sec	21-77	11M,10F
Duron et al.	Mask (C)	#	#	20-26	2 M

'C' refers to continuous collection of expired air samples and 'A' automatic collection every two minutes. 'M' refers to male subjects and 'F' to female. Where measurement conditions were not reported, these are signified by '#'.

(a) measurement device - the breathing apparatus used was either a ventilated hood or close-fitting face mask. The use of a hood entails some mixing of ambient air with expired air, leading to lower VO_2 values than would be expected with a mask system (Brebbia & Altschuler, 1965; Shapiro et al., 1984). However neither system is likely to differentially affect measurement for different sleep stages. Sampling of expired air was continuous in all cases except for one study where a sample was taken automatically once every two minutes (Shapiro et al., 1984). This method of sampling could also result in less accurate measurement of VO_2 if the time of sampling coincided with some disturbance or instability in the system. A finding of MR lower in SWS than in stage 2 occurred in all three studies using a hood (Brebbia & Altschuler, 1965; Haskell et al., 1981a; Shapiro et al., 1984). Only one of the four studies which used the mask method found stage differences (Yamashiro et al., 1986).

(b) time unit of measurement - the studies varied as to the unit of time over which MR was averaged, or collected. Of those studies which reported the time period over which MR was calculated, the methods of Webb and Hiestand (1975), Brebbia and Altschuler (1965) and to a lesser extent Shapiro et al. (1984) could have produced less accurate measurement of VO_2 due to the intrusion of other stages into the supposed pure periods of sleep. The method of Webb and Hiestand (1975) could have resulted in contamination of pure sleep by periods of wakefulness, intrusion of other sleep stages or presence of body movement. A similar problem may have existed in relation to the five minute EEG scoring period of Brebbia and Altschuler (1965). In fact, Haskell et al. (1981a) recognize that long sampling intervals increase the likelihood that body movements, periods of wakefulness or multiple stages were included, increasing the variability of the sample.

(c) unit of EEG scoring - the use of 20 or 30 second epochs for the scoring of EEG data is standard (Rechtschaffen & Kales, 1968). The 5 minute epoch used by Brebbia & Altschuler (1965) is unusual and as outlined above, problems may have arisen due to the intrusion of different sleep stages. Duron et al. (1968) did not report their method of EEG scoring.

(d) sex of subjects - this is unlikely to be a problem. All studies used male subjects, however, White et al. (1985) also used female subjects reporting no sex differences.

(e) age of subjects - there is unlikely to be a problem with respect to differences in MR between sleep stages, although both studies using older subjects did not get stage effects. The ages of the subjects ranged from 18 to 77 years, however only two studies used subjects older than 35 years. The results in these two studies differ, with Webb & Hiestand (1975) reporting a negative correlation of VO_2 with age and White et al. (1985) reporting no relationship.

3.5 Conclusions.

In conclusion therefore, although it has been shown that MR is lower during sleep than wakefulness, it is unclear as to the particular factors which may be contributing to the effect. The evidence from studies which have investigated MR in relation to sleep stage is inconclusive as to the existence of specific stage effects in MR during sleep. The finding of a circadian rhythm in body temperature which appears to mirror the night-time pattern of MR during sleep suggests the possibility of a circadian factor affecting MR. It is also possible that the findings of previous studies were the result of methodological artifact. Thus, those studies which did report stage differences may have done so because their methods did not appropriately take into account time of night, and movement effects.

Accordingly, the present investigation will measure oxygen consumption in order to,

1. determine if the pattern of energy expenditure observed across the night is a function of sleep processes, circadian rhythmicity or factors temporally associated with sleep.

2. determine if stage differences previously reported by other investigators are genuinely related to stage or are an artifact of time of night, or body movement during sleep.

The method of an experiment to test these aims and hypotheses is described in Chapter Four.

CHAPTER FOUR

METHOD

CHAPTER FOUR

METHOD

4.1 Subjects and design.

Subjects were five healthy males, ranging in age from 19 to 24 years, recruited from the University community. All were non-smokers and free from respiratory and sleep pathology. In addition, during a preliminary interview it was established that the subjects had normal and regular sleep habits. A sixth subject was discarded due to the presence of periodic breathing. The average weight of the subjects was 81.9 kg and the average height was 184.8 cm. The age, weight and height of individual subjects are shown in Table 4.1.

All subjects were tested in the laboratory on nine non-consecutive nights following one adaptation night. The adaptation night was included to remove any first night effects (Agnew, Webb & Williams, 1966). However, as a function of equipment failure, one subject (I.B.) was run on an additional seven nights. In all, the study included 74 subject nights. Of these, 24 were excluded due to equipment failure and the discarded sixth subject. On all but 3 nights the laboratory was managed by the author, although on 17 other nights 2-3 hours assistance was obtained, from experienced sleep laboratory personnel, in monitoring the subject and the equipment.

Table 4.1: Relevant subject variables

	VARIABLE		
	Age (years)	Weight (Kg)	Height (cm)
SUBJECT			
I.B.	24	78.0	180
A.C.	21	82.4	185
D.L.	20	82.5	195
J.M.	19	71.1	180
J.V.	20	95.3	184
Mean	20.8	81.9	184.8
(s)	1.9	8.8	6.1

The nine experimental nights were distributed between three experimental conditions. The design is presented in Figure 4.1. In all conditions, subjects went to bed thirty minutes before their usual bedtime and remained in bed for at least seven hours. The conditions differed as to when the lights were turned out and when the subject was instructed to go to sleep. In condition 1 this was at the subjects' usual bedtime and in conditions 2 and 3 it was three and six hours later respectively. The order of conditions was counterbalanced within each subject with the particular order varying between subjects.

In all conditions recording of oxygen consumption and arousal state began when the subject was first put to bed, thirty minutes prior to their usual lights out time. In condition 1, recordings were taken throughout the period the subject was in bed. In condition 2, recording was terminated after one hour and reinitiated thirty minutes before lights out (three hours after the subject's bedtime) and then continued for the remainder of the night. In condition 3, data were collected during the first and fourth hours and then beginning thirty minutes before lights out (six hours after the subject's bedtime). Recording continued until the subject awoke, but was for at least one hour.

Figure 4.1 indicates that the three conditions could be compared at three points across the night: the 1st, 4th and 7th hours that the subject was in bed. Since the conditions differed as to whether the subject was awake, or asleep, during the intervening periods, the contribution of sleep as opposed to circadian variability could be assessed. In addition, the effect of sleep onset on O_2 consumption could be assessed at three points in time. Comparison between sleep stages was determined by analysis within recording session. The effect of body movement on O_2 consumption was determined by analysis within condition, then across subjects.

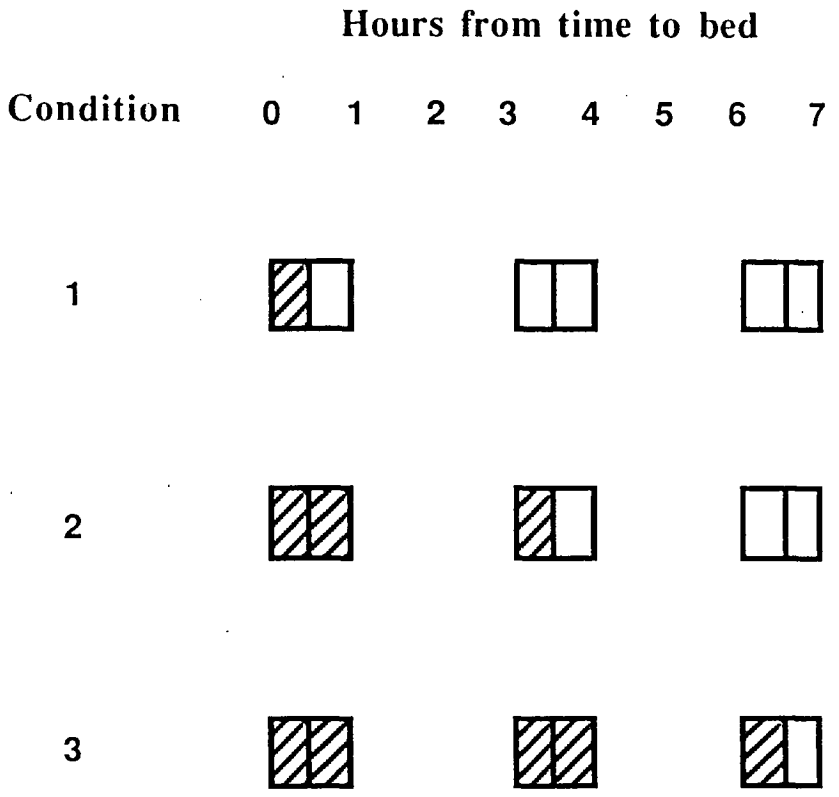

Figure 4.1 A schematic representation of the design showing the critical half hour recording periods. The closed boxes represent periods of wakefulness while the open boxes indicate periods when the subject was asleep. Data collection was continuous following lights out, which occurred at 0.5, 3.5 and 6.5 hours in conditions 1, 2 and 3 respectively.

4.2 Procedures.

4.2.1 Measurement of metabolic rate.

MR was measured by indirect calorimetry and was expressed in terms of O_2 consumption. Concurrent CO_2 production analysis was not available for this experiment. Each subject wore a facemask (Commonwealth Industrial Gases antistatic anaesthetic mask) attached to a Rudolph no. 2600 or 2700 two-way breathing valve. The mask was fitted with an inflatable cuff and was held tightly in place with a head strap. The mask was used in preference to a hood as its use enabled a more accurate breath by breath analysis of ventilation and oxygen consumption. The system also introduced the possibility of error due to the effect of instability in breathing, however close monitoring of the equipment enabled data loss to be minimized (see below). Before beginning the experiment, subjects were given experience in wearing the mask. Two of the subjects had participated in other experiments using the same equipment, and the remaining three subjects came to the laboratory for two afternoon sessions in addition to the adaptation night. The effect of these experiences was that the subjects were able to tolerate the mask for long periods without major disruption to their sleep. Further, the subjects were trained to detect air leaks and were instructed to check for them during periods of wakefulness. The ventilation record was closely monitored during data collection to detect changes due to air leaks.

The valve deadspace was 33.3 ml or 118.8 ml, for the no. 2600 and 2700 valves respectively, and as the mask deadspace (which was measured by water displacement with the valve removed), was dependent on facial topography, the total equipment deadspace varied between 115.3 and 233.8 ml. Air was collected for O_2 analysis from an outlet in the breathing valve, located where the mask joined the valve. The

air was transported via 110 cm of polyurethane tubing (1 mm internal diameter) to an Applied Electrochemistry S-3A Oxygen Analyser, where it was heated to body temperature before it entered the analyser.

The oxygen analyser was calibrated to fresh air at the beginning of each session and periodically to tank air of known oxygen concentration. The experimental bedroom was ventilated with fresh air throughout the period of each session and the room temperature was maintained within the range 20-24^{o}C via the use of a thermostatically controlled, oil-filled electric heater. Expiratory airflow was measured by a Fleisch pneumotachograph (connected to the major outlet of the breathing valve) and a Gould PM 15E pressure transducer.

The output signals of both the oxygen analyser and pressure transducer were amplified using a Beckman R411 polygraph recorder and input to a D.E.C. PDP-11/23 computer via an eight-bit analogue to digital converter. The raw data were stored for subsequent analysis on paper chart and a thirty megabyte Winchester hard disk. The sampling rate was 20Hz for both variables. Software running under RT-11 Pascal was developed in the laboratory to enable the online display, via a high resolution graphics screen, of data entering the computer. In addition, the integrity of the data being collected was checked throughout each experimental session via paper chart, a variety of digital voltmeters and computer alarms. Thus the experimenter was able to identify the source of data collection problems. When these occurred the equipment was checked, and adjusted or repaired if necessary.

The accurate measurement of MR by indirect calorimetry may be affected by ventilatory instability. Rapid shifts in ventilation, which are unrelated to metabolic stimuli, are associated with less rapid shifts in arterial O_2 levels and thus in expired O_2. The delay in shift in O_2 levels is approximately thirty seconds. In these

circumstances oxygen consumption, when measured on a breath by breath basis, will be underestimated when ventilation falls and overestimated when it rises. Because ventilation falls at sleep onset (Colrain et al., 1987) oxygen consumption during this transition period will tend to be underestimated. Under these conditions, in which ventilation is continuously falling, actual oxygen consumption values would be expected to lag approximately thirty to sixty seconds behind the measured values.

Oxygen consumption was determined for each breath by integration of the cross product of O_2 level and rate of expired air flow. Thus O_2 consumption was available for each breath, each minute, or minute values extrapolated from individual breaths. Minute O_2 consumption (VO_2) was used for this study. In addition to VO_2, minute ventilation (VE) and fractional end tidal oxygen level ($FetO_2$) were analysed.

4.2.2 Measurement of wakefulness and sleep.

To discriminate wakefulness from sleep and to identify stages of sleep, an electroencephalogram (EEG), electromyogram (EMG) and electrooculogram (EOG) were collected using standardised procedures (Rechtschaffen & Kales, 1968). The EEG was recorded using disk electrodes applied to positions C3/A2 (International 10-20 system), the EMG by electrodes located bilaterally over the submental muscle and the EOG by electrodes vertically displaced on the outer canthi of the eyes. The three measures were amplified and recorded on paper chart by a Beckman R411 polygraph. The records were subsequently scored in 30 second epochs using standardised procedures (Rechtschaffen & Kales, 1968). Due to the nature of the study and the difference in size of the records, it was not possible to have the scorer blind to condition. However this should not present a problem as the analysis for the EEG variables was completed separately from that of oxygen consumption. The EEG data were later combined into 60 second epochs to equate the duration of the

sleep-wake scoring unit with that of the metabolic and respiratory variables. Initially, the sleep records were scored into the stages: wake (W), movement time (MT), stages 1, 2, 3, 4 and REM. Stages 3 and 4 were combined into SWS for MR comparison. The sleep variables analysed were: sleep onset latency (SOL); the duration of the lights out period (TIB); the total sleep time (TST); the total time awake (TTA) during TIB; W, MT and 1 combined in TIB; time in each of the sleep stages (MT, 1, 2, 3, 4, SWS; 2,3 & 4 [NREM], and REM); the percentage of stage 2 (%Stage2), SWS (%SWS) and REM (%REM) in TST; percentage of (W + MT + 1) in TIB (%DIST); and sleep efficiency (SEFF). However %SWS, SEFF, %DIST, %REM, and %Stage2 were of particular interest.

In addition to providing information on the sleep state during the lights out period, the recordings were used to ensure that subjects remained awake during the awake measurements when the lights were on. There are two reasons why it was critical to ensure that subjects remained completely awake during resting metabolic rate measurements. First, the loss of alpha activity is associated with substantial reductions in ventilation (Colrain et al., 1987). The subsequent adjustment in arterial O_2 level is less rapid and as a consequence, O_2 consumption values are underestimated. Second, it has been suggested that the early period of sleep is associated with a real reduction in MR (Colrain et al., 1987). For both of these reasons, waking oxygen consumption is likely to be underestimated if subjects become drowsy and enter stage 1 sleep.

4.2.3 General Laboratory Procedures.

Subjects arrived at the laboratory one hour before the time scheduled for them to go to bed, to allow for preparation and adjustment to the temperature of the laboratory. After they had prepared for bed, the electrodes were attached and the facemask was put in place. The necessary tubing was connected to the mask after the subject was in bed. The subjects slept in night attire under bedclothes with the room temperature maintained at 20 - 24°C. A previous study has shown that with the subject wearing pyjamas and sleeping under bedclothes, and with room temperature in the range 16 to 25°C, the temperature in the bed microclimate remained within the range 28.6 to 30.9°C (Candas et al., 1979). The temperature of the air surrounding the subject was therefore in this study maintained close to the recognized thermoneutral temperature of 29°C (Haskell et al, 1981a).

During awake recording sessions, the lights were left on, but dimmed, and the subjects could watch television or listen to music whilst in a supine position. Supine positioning was used to control for any masking, by upright posture or activity, of the hypothesised effects on MR, as both upright posture (Kleitman & Doktorsky, 1933) and activity (Gander, Graeber & Connell, 1985) have been found to affect MR.

During sleep periods the lights, television and music were switched off. Recording was initiated after the subjects had settled in bed and continued all night in condition 1 and for one hour in conditions 2 and 3. In the latter conditions, following one hour of recording, the facemask was removed and the subject was allowed to read, watch television or listen to music, while remaining supine in bed until the next recording period. The subjects were monitored by the experimenter to ensure that they remained awake. These procedures were repeated in subsequent awake periods.

Subjects were interviewed to establish their usual sleep pattern and were instructed to conform to that pattern throughout the period of the experiment. They were required to refrain from exercising and from drinking alcohol or coffee on experimental days. Food was not allowed after the evening meal, which was at the subjects' usual meal time each experimental day. The time between the evening meal and the commencement of recording ranged from 3.5 to 5.0 hours, but was consistent for each subject. The amount eaten at the evening meal was kept constant and subjects were required to keep food intake diaries for the evening meal.

4.2.4 Preparation of data for statistical analysis.

Software running under RT-11 Pascal developed in the laboratory enabled the derivation of a number of variables from the stored digitised data. These data were analysed using a breath by breath procedure from which minute values were then calculated. The software-based determination of what constituted each breath was subject to verification by the experimenter. During periods when body movements were absent, or were small in magnitude, the program was essentially 100% accurate, as determined by the experimenter. However, during gross body movements the program often failed to identify breaths accurately. Indeed, it was often difficult for the experimenter. Each epoch of one minute was included in subsequent analyses only if breaths covering at least thirty seconds of that minute were accurately identified. Minute values for the time recorded for each condition were then calculated for each variable.

In all conditions, measurements were available from the first, fourth and seventh hours of the period the subjects were confined to bed. Lights out occurred half way through hours one, four, and seven for conditions 1, 2 and 3 respectively. Thus, each of these hours was divided into two thirty minute periods, giving six measurements across the night. As subjects may take some minutes to adapt to the

recording conditions, the data from the first fifteen minutes of each awake recording period were discarded. To ensure that the timing of measurement periods remained constant over conditions, the data from the first fifteen minutes of the equivalent time periods during which the subjects were asleep were also discarded. Thus, MR estimates for the intervals 0 to 0.5 hour, 3 to 3.5 hours and 6 to 6.5 hours in all conditions were based on the second fifteen minutes of recording from the half hour periods. The measurement of sleep MR in the interval immediately following lights out was based on all minutes of stage 2 sleep within the half hour interval. This procedure provided at least fifteen minutes of data in all but four of the sleep onsets, because the subjects were all well adapted to the laboratory and typically fell asleep in less than fifteen minutes. The remaining half hour periods were based on approximately thirty minutes of data with small adjustments being made to control for the average sleep onset latency for a particular subject at a particular time of night.

CHAPTER FIVE

RESULTS

CHAPTER FIVE

RESULTS

Data analysis included first, a comparison of standard sleep variables for individual subjects with the results of a number of previous studies. For each night of Condition 1, the time spent in each stage was totalled and using these totals, sleep variables were calculated. The values were then averaged within subjects and then averaged across subjects. Second, analyses were performed to test for the existence of sleep, circadian or dietary factors influencing sleep period oxygen consumption. Third, differences in oxygen consumption between sleep stages were investigated using the same comparisons as had produced positive outcomes in other studies. Finally, an assessment was carried out of the extent to which the results found in the third analysis may have been due to methodological artifacts.

It is important to note that a small amount of data were lost during data collection. Of the 270 half hour sessions recorded across all subjects, 28 were excluded due to contamination by air leakage. During the total recording time, some data were lost due to movement (6.6%) and due to the presence of an inappropriate stage (sleep in wakefulness or wakefulness in sleep)(7.6%). Importantly, only 4% of the time (approximately half of the 7.6%), was there an intrusion of Stage 1 or 2 sleep into wakefulness recording periods. These intervals were discarded in the computation of O_2 consumption. Some additional data were lost due to contamination by air leakage, during recording outside the half hour sessions. Two complete subject nights were excluded from analysis, and some data were excluded for the same reason from 11 further nights. As mentioned in Chapter Four, the integrity of the data being collected was checked regularly during experimental sessions, and this procedure enabled the experimenter to correct the majority of equipment problems at the moment they arose.

5.1 Assessment of standard sleep variables.

Standard sleep variables (based on all nights in Condition 1), from individual subjects are shown in Table 5.1. Despite the presence of the mask and other monitoring equipment, all subjects slept well with a mean sleep efficiency (TST/TIB) of 0.93 ± 0.17. This value is considerably higher than the mean value of 0.771 reported by White et al. (1985) but is comparable to values reported by a number of other studies (Akerstedt & Gillberg, 1986; Bonnet, 1986; Shapiro et al., 1984; Webb & Hiestand, 1975; Williams, Karacan & Hursch, 1974). Standard sleep variables reported in these studies are presented in Table 5.2. However, in comparison with the other studies, subjects in this experiment had a higher percentage of stage 2 sleep, and three of the subjects (I.B., J.M., & J.V.) had a higher percentage of stage 1 sleep. The lowest levels of disturbed sleep (Wake + MT + Stage 1), were recorded by the two subjects (A.C. & D.L.) who were most experienced in wearing the mask during sleep. The percentage of time spent in SWS was markedly lower in one subject (J.M.). However across subjects, the mean percentage of time spent in SWS was of the same magnitude (Akerstedt & Gillberg, 1986; Webb & Hiestand, 1975; White et al., 1985) or lower (Bonnet, 1986; Shapiro et al., 1984; Williams et al., 1974) than the value recorded in other studies. Sleep onset latency (SOL) varied greatly in the other studies, ranging from 2.8 to 26.0 minutes, however in this study, SOL ranged from 1.8 to 15.2 with a mean of 8.8 minutes. SWS latency (SWSL) was shorter (14.8 minutes), REM latency (REML) was longer (145.0 minutes), and the percentage of REM (REM%) of 14.4 was less than that reported in the other studies. However, in general terms, the sleep of the subjects in this study was similar to that reported in the other studies in the literature.

Table 5.1: Sleep variables for individual subjects.

	SUBJECT					
	I.B.	A.C.	D.L.	J.M.	J.V.	
VARIABLE						MEAN
TIB	436.5	451.5	418.5	414.5	441.2	432.4
TST	384.3	439.7	385.7	402.0	415.5	405.4
TTA%	11.8	3.6	8.7	3.1	8.1	7.1
STAGE1%	13.6	6.5	6.8	19.0	14.2	12.0
STAGE2%	60.9	58.8	63.6	59.3	59.7	60.5
SWS%	13.1	13.4	11.8	7.2	15.1	12.1
REM%	12.3	20.3	16.7	12.4	10.4	14.4
SOL	15.2	1.8	4.7	13.0	6.0	8.8
SWSL	22.0	14.0	15.3	12.2	10.3	14.8
REML	138.0	156.8	79.8	156.8	193.8	145.0
SEFF	0.88	0.97	0.91	0.97	0.92	0.93
DIST%	23.7	9.8	15.4	23.6	21.6	18.8

SEFF refers to sleep efficiency (TST/TIB) and DIST to disturbed sleep (wakefulness + movement time + stage 1). TTA and DIST are shown as a % of TIB, and SWS, REM, STAGE1 and STAGE2 are shown as a % of TST. For each subject, the values are the average of the three nights of condition 1.

Table 5.2: Sleep variables from previous studies.

	STUDY					
	1	2	3	4	5	6
VARIABLE						
TIB	442.2	-	437.3	-	452.0	404.0
TST	419.3	408.8	423.1	-	424.0	396.0
TTA %	1.3	2.4	3.3	-	6.9	-
STAGE1 %	4.4	-	8.4	5.6	13.0	-
STAGE2 %	45.5	-	40.5	64.6	53.0	50.5
SWS %	20.8	13.3	29.9	12.9	12.0	23.2
REM %	28.0	22.3	17.9	16.7	20.0	23.2
SOL	14.6	-	2.8	4.7	26.0	14.0
SWSL	20.9	-	19.0	-	28.0	-
REML	88.3	-	121.1	-	92.0	-
SEFF	0.95	-	0.97	0.77	0.94	0.98

SEFF refers to sleep efficiency (TST/TIB). TTA is shown as a % of TIB, and SWS, REM, STAGE1 and STAGE2 are shown as a % of TST.

Key for studies.

1 - Williams et al., 1974

2 - Webb & Hiestand, 1975

3 - Shapiro et al., 1984

4 - White et al., 1985

5 - Akerstedt & Gillberg, 1986

6 - Bonnet 1986

5.2 The effect of sleep as opposed to circadian rhythm on the sleep period fall in oxygen consumption.

The mean oxygen consumption for each of the six half hour periods, based on three nights in each of the three conditions, for each of the five subjects, is shown in Table 5.3 and in Figure 5.1. From the results it can be seen that MR changes during the first six and a half hours of the sleep period are the product of two factors. The effect of the first factor can be seen to be rapid following sleep onset with no further contribution following the first 15 to 30 minutes of sleep. Furthermore, the change at sleep onset was independent of the timing of sleep onset within the night. The changes during the remainder of the night were independent of whether or not the subjects were asleep and appear to be due to a second factor.

The statistical analysis of the sleep onset data compared the period immediately before lights out with the sleep period immediately after sleep onset, for each of three conditions (lights out at 0, 3 and 6 hours after each subjects' usual bedtime) in a 2 x 3 repeated measures analysis of variance (ANOVA). The results showed a significant main effect of sleep onset ($F = 29.9$ $(1,4)$, $p < 0.01$), but no significant interaction effect ($F = 1.6$ $(2,8)$, $p > 0.05$). This indicated that the size of the effect at sleep onset was independent of the time of night. This analysis also revealed a significant main effect of condition ($F = 16.0$ $(2,8)$, $p < 0.01$) reflecting the reduction in MR across the night (the full statistical analysis is presented in Appendix 2).

Table 5.3: Oxygen consumption values in ml./min. averaged over subjects and replications within subjects for the half hour periods identified in the design (see Figure 4.1).

	HOURS FROM TIME TO BED					
	0.0	0.5	3.0	3.5	6.0	6.5
CONDITION						
1	330.7	276.9 *	250.0 *	239.6 *	228.7 *	231.4 *
2	302.1	298.7	285.4	248.7 *	222.6 *	226.4 *
3	299.1	300.2	289.1	284.5	273.7	241.1 *

Sleep periods are denoted by ' * ', the remainder are values for wakefulness.

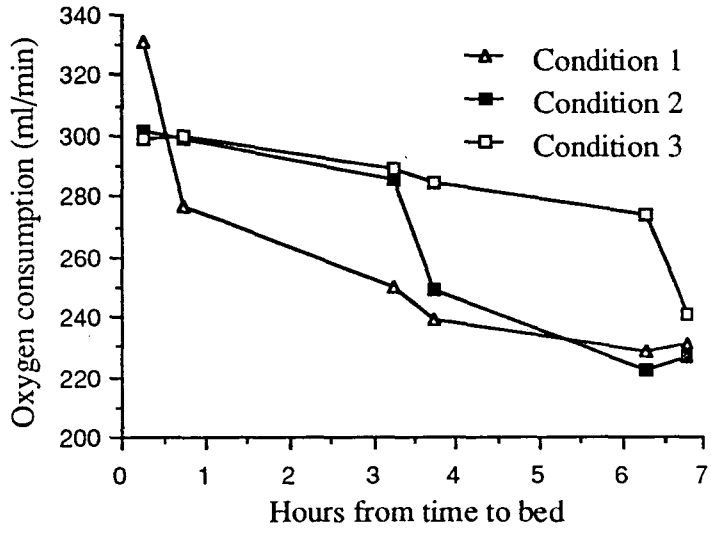

Figure 5.1: Average oxygen consumption values for the half hour periods before and after the time for lights out in each of the three conditions.

In order to demonstrate that the change in MR at sleep onset was significantly greater than change due to time over the same interval, two 2 x 2 repeated measures ANOVA were conducted, one on the data from hour one and the other on the data from hour seven. These analyses compared the first half hour with the second and the sleep onset condition with the average of the two constant conditions. Thus in the analysis of hour one, conditon 1 (awake - sleep) was compared with the average of conditions 2 and 3 (awake - awake). In the analysis of hour seven, condition 3 (awake - sleep) was compared with the average of conditions 1 and 2 (sleep - sleep). In each case, the interaction was statistically significant ($F = 11.7$ (1,4), $p < 0.05$; $F = 11.8$ (1,4), $p < 0.05$, for hours one and seven respectively), indicating that over this period the combined effect of sleep onset and time was greater than that of time alone.

As reported above, the comparison of the sleep onset period at hours one, four and seven in conditions 1 to 3 respectively, indicated a significant effect of time over the seven hours of recording. A comparison of conditions 1 and 3 allowed the effects of the passage of time, in wakefulness as opposed to sleep, to be assessed. The statistical analysis consisted of a 2 x 3 repeated measures ANOVA. The first factor was condition (1 versus 3) and the second was time (first, fourth and seventh hour). In both conditions, the MR measurement for the first hour was taken from the second half hour whereas for the seventh hour the measurement used was the first half hour. At the fourth hour, the two half hour measurements were averaged. Although there was a significant main effect of both condition ($F = 24.4$ (1,4), $p < 0.01$), and time ($F = 21.5$ (2,8), $p < 0.001$), the interaction was not significant ($F = 3.87$ (2,8), $p > 0.05$)., although it did approach significance. However, it should be noted that the first sleep period in condition 1 was not a true estimate of the sleep value as it was the average of approximately the first twenty-one minutes following sleep onset (average sleep onset latency was 8.8 minutes). Because the asymptote following sleep onset

was not achieved for approximately fifteen minutes, the estimate was not independent of the sleep onset effect. In a subsequent analysis in which the asymptotic value was used, the interaction between state and time did not approach significance ($F = 1.17$ $(2,8)$, $p > 0.05$). Thus the rate of fall of MR was approximately the same in wakefulness as in sleep (see Table 5.4 and Figure 5.2). Anova summary tables are included in Appendix 3.

Table 5.4: Oxygen consumption values in ml./min. averaged over subjects and replications within subjects for the half hour periods identified in the design (see Figure 4.1). These are asymptotic values.

	HOURS FROM TIME TO BED					
	0.0	0.5	3.0	3.5	6.0	6.5
CONDITION						
1	330.7	267.9 *	250.0 *	239.6 *	228.7 *	231.4 *
2	302.1	298.7	285.4	244.6 *	222.6 *	226.4 *
3	299.1	300.2	289.1	284.5	273.7	233.4 *

Sleep periods are denoted by ' * ' while the remainder are values for wakefulness. Includes asymptotic values, which are underlined, for the three onset periods.

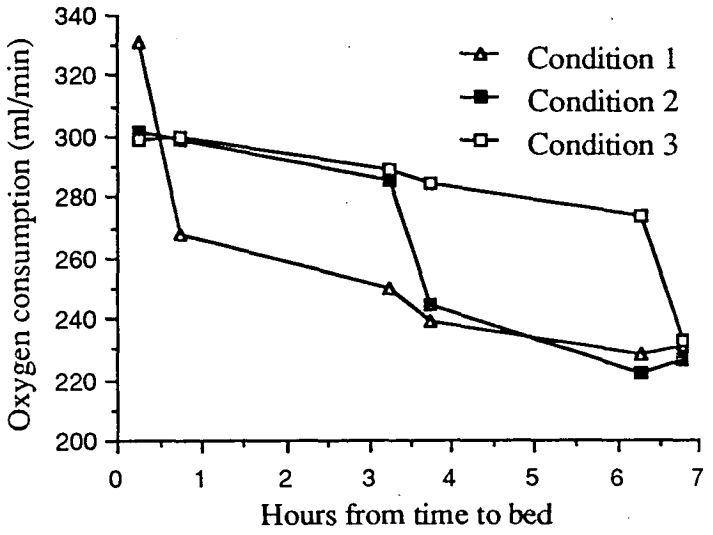

Figure 5.2: Average oxygen consumption values for the half hour periods before and after the time for lights out in each of the three conditions (asymptotic values).

The fall in oxygen consumption at sleep onset was investigated further by averaging minute values over the nine sleep onsets for each subject, with subjects being treated as independent samples. The time period averaged was the last 25 minutes before lights out, the transition period between continuous wakefulness and stage 2 sleep, and the first 60 minutes of sleep. The transition period, which may consist of rapid alternations of wakefulness and stage 1 sleep, is of variable length. As a consequence, it was divided into quartiles for each sleep onset and the quartiles were then averaged over sleep onsets. A comparison of the data from the three conditions using this form of analysis did not reveal any systematic differences. Thus, only the data averaged over all sleep onsets has been presented. These data are illustrated in Figure 5.3. The most notable feature was the rapidity with which oxygen consumption fell during sleep onset. Oxygen consumption had become relatively stable within 15 minutes of the onset of stage 2 sleep. The mean rate of fall of oxygen consumption across all subjects over the first fifteen minutes following sleep onset was 3.3 ml/min and 0.9 ml/min over the first hour. However, the rate of fall was only 0.1 ml/min from minutes fifteen to sixty. The mean rate of fall of oxygen consumption over the sleep onset period for each subject is shown in Table 5.5.

Finally, as discussed in Chapter Four, as changes in arterial O_2 lag behind changes in ventilation, the reported O_2 consumption underestimates actual values. However, the lag would not be expected to be more than about a minute in the current data. Values reported in Table 5.3 and shown in Figure 5.1 would not be affected, although the functions shown in Figure 5.3 slightly overestimate the fall in MR.

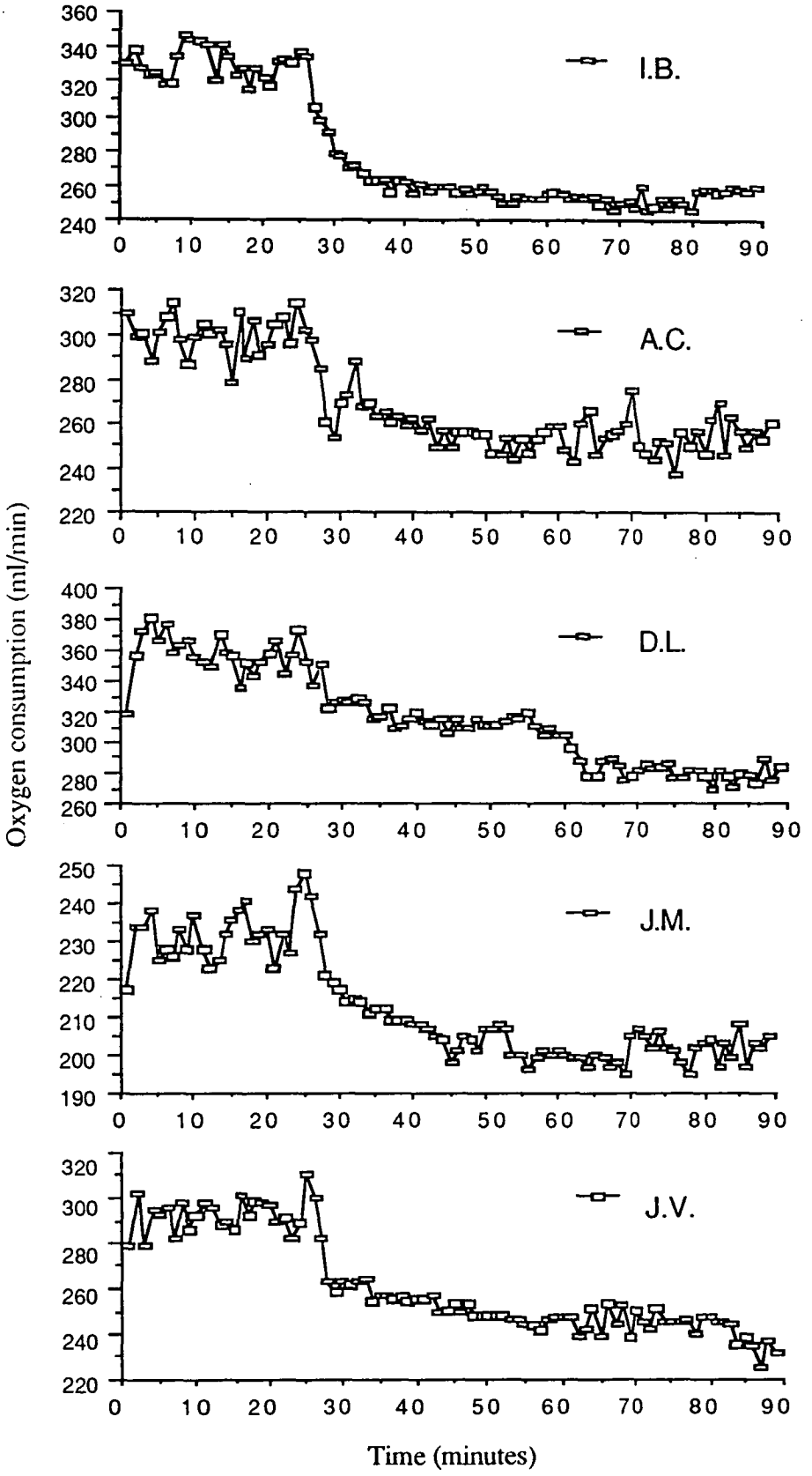

Figure 5.3 Minute oxygen consumption for each subject averaged over all sleep onsets for 25 minutes prior to lights out to 60 minutes after sleep onset

Tbale 5.5: Rate of fall of oxygen consumption (ml./min) following sleep onset.

	PERIOD OF FALL		
	0 to 15 min.	0 to 60 min.	15 to 60 min.
SUBJECT			
I.B.	4.7	1.2	0.0
A.C.	3.2	0.7	-0.9
D.L.	3.5	1.3	0.5
J.M.	2.1	0.5	-0.2
J.V.	2.9	0.9	0.2
Mean	3.3	0.9	0.1
(s)	0.9	0.3	0.5

Minute ventilation and $FetO_2$ were also analysed over the sleep onset period. Minute ventilation fell rapidly following the onset of the transitional phase of wakefulness/stage 1, and in four of the five subjects had reached a relatively stable level by the onset of stage 2 sleep. The fifth subject showed a further small reduction over the first ten minutes of stage 2 sleep. The average reduction was 28%. In all subjects, $FetO_2$ levels fell following the loss of continuous wakefulness and then rose again during the first 50 minutes of sleep. However, values did not return to awake levels. The latter result indicates that the fall in ventilation is greater than that required by the fall in MR. Furthermore, it suggests that the reduction in MR is not as rapid as that of ventilation. These results replicate previous data on the effects of sleep onset on respiratory variables (Colrain et al., 1987).

5.3 Assessment of differences in oxygen consumption between sleep stages.

As shown by the results presented in section 5.2, oxygen consumption during sleep is influenced by sleep itself, and by a circadian factor. It remained to be determined if stage of sleep also affected oxygen consumption. Accordingly, to assess this possibility, the data from this study were initially analysed using the same analytic techniques as were used in the other previously reported studies.

The first analysis involved averaging of total minutes of each sleep stage in each night spent in condition 1. The fifteen subject nights from condition 1 were used in the analysis, however, as two nights were unusable due to mask leakage, the oxygen consumption data from 13 nights were finally included in the analysis. The results of this analysis are presented in Table 5.6.

Table 5.6: Mean oxygen consumption in ml/min for different sleep stages for each of thirteen subject nights in condition 1.

	SLEEP STAGE		
SUBJECT	2	SWS	REM
I.B.	269.8	293.2 *	283.9 *
	285.7	283.2	285.0
	180.3	178.9	202.8 *
A.C.	257.5	263.5	254.6
D.L.	305.8	327.7 *	298.3
	275.4	303.0 *	274.2
	296.5	323.6 *	304.6
J.M.	209.9	258.4 *	209.3
	167.4	198.5 *	169.7
	206.4	211.6	#
J.V.	237.2	243.0	244.7
	212.1	212.3	213.8
	276.8	294.5 *	#
MEAN	244.7	260.9	249.2

Values which are signified ' * ' were significantly different from the value for stage 2 using Student's t - test ($p = 0.05$). The symbol ' # ' signifies that a value was not available for that night.

The mean value for oxygen consumption was lowest in stage 2, intermediate in REM, and highest during SWS. In only 2 of the 13 available within subject comparisons using mean values averaged across all minutes of a particular stage, was oxygen consumption lower in SWS than in stage 2 sleep, and in neither case was the difference significant. In fact, in seven cases oxygen consumption was significantly greater in SWS than in Stage 2 sleep. Of the 11 comparisons involving REM and Stage 2 sleep, in 6 cases oxygen consumption was greater in REM than Stage 2, however in only two cases was the difference significant. This information is presented in Table 5.7.

Table 5.7: Comparison of oxygen consumption between sleep stages for all subjects.

	DIRECTION OF DIFFERENCE IN OXYGEN CONSUMPTION			
	2 > SWS	SWS > 2	2 > REM	REM > 2
COMPARISONS				
Number	2	11	5	6
Number significant	0	7	0	2

($p < 0.05$).

From these results it can be seen that when comparisons are made between sleep stages independent of time of night, then oxygen consumption during SWS is not low compared to stage 2 sleep. In an attempt to control for time of night effects, comparisons were made between contiguous stages: that is stages which are temporally adjacent during the night. The data used was taken from the 13 usable nights from condition 1, and from the 15 nights from condition 2. Data from the first fifteen minutes following sleep onset was excluded from the analysis as this period is not free of the sleep onset fall in oxygen consumption. The results of the comparison of oxygen consumption between contiguous sleep stages are presented in Table 5.8.

Table 5.8: Comparison of oxygen consumption between contiguous sleep stages for all subjects.

	TEMPORAL POSITIONING OF SLEEP STAGES	
DIRECTION OF CHANGE	2 -> SWS	SWS -> 2
2 > SWS	22 (10)	7 (3)
No Change	0	1
SWS > 2	5 (2)	12 (6)

Table 5.8 shows the number of comparisons and direction of change of oxygen consumption between contiguous periods of SWS and stage 2. The values in parentheses show the number of comparisons where there was a significant difference in the level of oxygen consumption between SWS and stage 2 (Student's t test, $p = 0.05$).

It is clear that when SWS followed stage 2, oxygen consumption was usually lower during SWS, however, in only 10 of the 22 cases was this difference significant. In fact, in two cases, oxygen consumption during SWS was significantly higher than during stage 2. When SWS preceded stage 2, in 12 of 19 cases the level of oxygen consumption during SWS was higher than that for stage 2, and in six of these cases the difference in oxygen consumption was significant. However, in three of seven cases, oxygen consumption was significantly higher during stage 2 than during SWS. It can be seen from these results, that when comparisons are made of contiguous or close stages, then oxygen consumption is generally lower during SWS as contrasted to stage 2.

5.4 The effect of movement arousals on oxygen consumption during sleep.

As movement arousals almost invariably follow, but rarely precede SWS, they could contribute to oxygen consumption during SWS being lower in contrast to other sleep stages if they result in an elevation of oxygen consumption following the arousal. To assess the possible effect of movement arousals on sleep period MR, a comparison was made between a period containing an arousal and a period of undisturbed sleep of the same number of minutes. A movement arousal was defined as a period of EMG disturbance which lasted for greater than five seconds and which resulted in a change of sleep stage. An arousal may have lasted for longer than one minute and was not necessarily continuous. The mean length of the EMG disturbance during an arousal period ranged from 19.0 to 39.4 seconds across subjects with an overall mean of 32.2 seconds and the mean number of minutes affected by an arousal was two.

To identify the effect of an arousal, sequences were identified of ten minutes of undisturbed sleep followed by an arousal which was then followed by a further ten

minutes of undisturbed sleep. To eliminate the possibility of contamination by the rapid reduction in MR following sleep onset, the first 15 minutes following sleep onset were excluded from this analysis. To control for the circadian factor, a period of undisturbed sleep was taken as closely as possible from the same time of night as a period containing an arousal, but from one of the other two nights of the same condition, for the same subject. Minute oxygen consumption values were obtained for each of the ten minutes prior to and the ten minutes following an arousal. As the mean length of an arousal was two minutes, a matching period consisted of 22 minutes of undisturbed sleep. In no case had an arousal occurred in the three minutes prior to the first minute of the selected arousal or no arousal period. A total of 37 periods containing movement arousals were identified from the 45 subject nights and 37 periods of undisturbed sleep were identified for comparison. The data were averaged across the 37 periods and are shown in Figure 5.4.

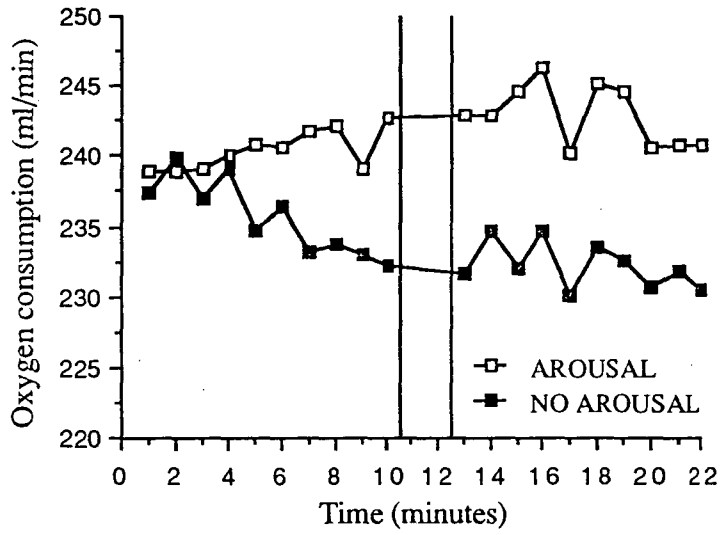

Figure 5.4 Minute oxygen consumption values averaged across all subjects for the arousal and no arousal conditions. The vertical lines signify the limits of the arousal in that condition.

Upon inspection of Figure 5.4, it can be seen that in the no arousal condition, there is a gradual decline in oxygen consumption across time. In contrast, in the arousal condition there is a slight increase in oxygen consumption prior to the movement arousal and a continued increase following the arousal. In addition, in the arousal condition, the oxygen consumption does not begin to consistently decline until six minutes following the arousal.

A (2 x 2 x 10) analysis of variance was conducted on these data with the factors being condition (arousal v no arousal), period (pre v post arousal) and time (minute within period) respectively. The results showed a significant main effect of time ($F = 2.39$ (9,648), $p < .05$), and a significant interaction effect of condition x period ($F = 5.02$ (1,72), $p < .05$). All other effects were not significant. Anova summary tables are included in Appendix 4.

The arousal condition in these data consists of periods where the arousal marks a transition from both stage 4 to stage 2 and also stage 2 to stage 2. These two subsets of data, with their matched no arousal conditions, were analysed separately using the same analysis of variance design as was used with the combined data. The data from periods involving transitions from stage 2 to stage 2 are presented in Figure 5.5.

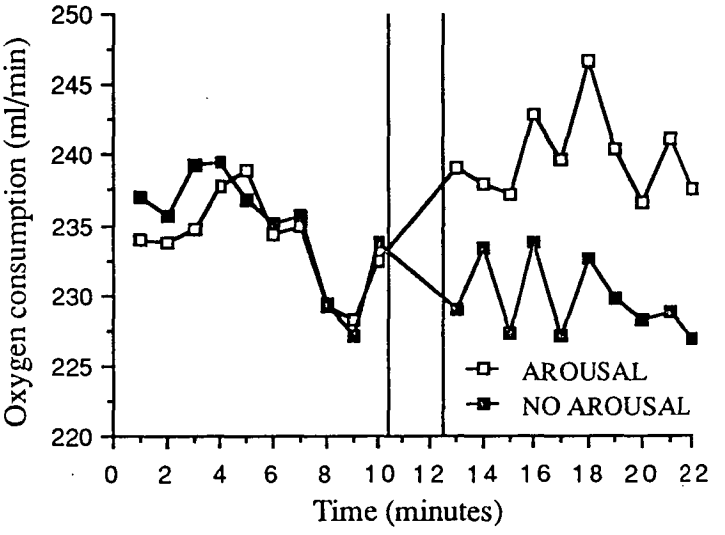

Figure 5.5 Minute oxygen consumption values averaged across all subjects for the arousal and no arousal conditions with data from the stage 2 to stage 2 transitions only. The vertical lines signify the limits of the arousal in that condition.

Inspection of Figure 5.5 shows again a gradual decrease in oxygen consumption across time in the no arousal condition. In contrast however, in the arousal condition, there is an increase in oxygen consumption following the arousal. The statistical analysis of these data showed a significant main effect of time ($F = 2.46$ (9,162), $p < .05$). However, the interaction between condition and period was not significant, although it approached significance ($F= 3.81$ (1,18), $p = .07$).

The data for periods involving transitions from stage 4 to stage 2 are presented in Figure 5.6.

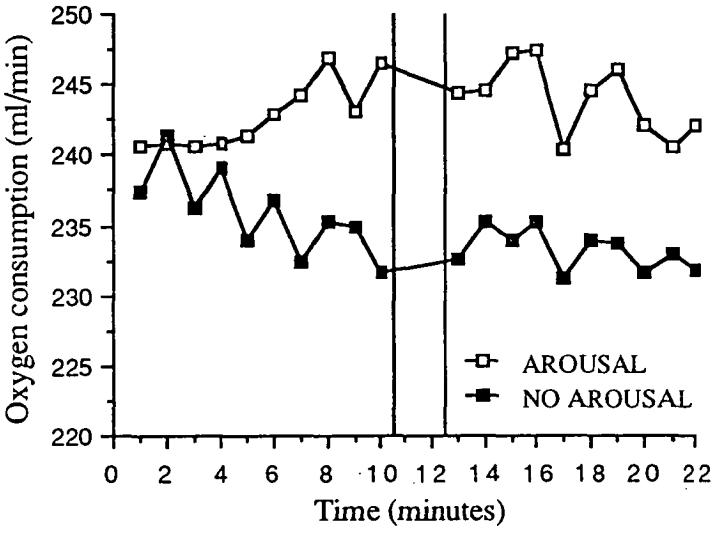

Figure 5.6 Minute oxygen consumption values averaged across all subjects for the arousal and no arousal conditions with data from the stage 4 to stage 2 transitions only. The vertical lines signify the limits of the arousal in that condition.

As with periods involving transitions from stage 2 to stage 2, the data in Figure 5.6 show a gradual reduction in oxygen consumption across time in the no arousal condition. In addition there is an increase in oxygen consumption in the arousal condition prior to the arousal and a further small increase post arousal. However, the statistical analysis of these data showed no significant effects.

It is clear from these data that oxygen consumption increases following an arousal, and that there exists a period of obvious disturbance following the arousal. This perturbation continues for at least seven minutes before the oxygen consumption appears to return to the gradual reduction across time which is observed during periods without the presence of an arousal.

CHAPTER SIX

DISCUSSION AND CONCLUSIONS

CHAPTER SIX
DISCUSSION AND CONCLUSIONS

This study had two main aims. These were to determine if the pattern of energy expenditure observed across the night is a function of sleep processes, circadian rhythmicity or the specific dynamic action (SDA) effect of the evening meal (a factor temporally associated with sleep), and to determine if differences in MR between sleep stages previously reported by other investigators are genuinely related to sleep stage or are an artifact of time of night, or body movement arousals. The results show that sleep period MR is influenced by circadian rhythmicity and sleep itself, however no evidence was found implicating the SDA effect of the evening meal in sleep period MR. In addition, the differences in MR between sleep stages reported by other investigators appear to be artifacts of time of night and body movement effects. These findings will now be considered in more detail.

6.1 Sleep and circadian factors.

The results indicate that sleep and circadian cycle determine the course of MR over the sleep period. The data do not support the view that the fall in MR during the sleep period is due to a diminishing SDA effect following the evening meal. Two observations support the latter conclusion. First, the rapid change in oxygen consumption at sleep onset was found to be independent of the evening meal. Second, the rate of fall of MR was constant across the night and continued for longer than would have been expected on the basis of a SDA effect alone. The results of one of the few studies to assess the time course of the change in MR following a meal show that the major change in MR due to the SDA effect occurs in the three hours immediately following the meal, with a further small change occurring in the following one to one and a half hours (Schwartz, Ravussin, Massari, O'Connell & Robbins, 1985). Although an evening meal consumed immediately prior to sleep

might influence MR during the sleep period, the present data clearly show that the fall in MR across the sleep period is not due to, or even influenced by, a SDA effect associated with the evening meal consumed approximately four hours prior to sleep onset.

Informal observations and comments in the literature suggest that the change in MR at sleep onset is rapid. However, these earlier studies typically reported MR values which were averaged over relatively long time periods, such as one hour. One previous study has demonstrated that MR falls substantially from wakefulness to the first ten minutes of stage 2 sleep (Colrain et al., 1987). The present study describes the shape of the metabolic function, confirms its rapidity and demonstrates that the change at sleep onset is specifically related to sleep and is not due to the SDA effect of the evening meal. The rate of fall was 3.3 ml O_2/min to the end of the first 15 minutes and 0.9 ml O_2/min across the first sixty minutes of sleep. The rate of fall from 15 minutes to one hour was 0.1 ml O_2/min. Calculations from the data of previous papers suggest a fall, across the first hour, of from 0.15 to 0.5 ml O_2/min with a mean of 0.3 ml O_2/min (Kreider & Buskirk, 1957; Kreider et al., 1958; Kreider & Iampietro, 1959; Shapiro et al., 1984; White et al., 1985).

A common explanation of the fall in MR during the sleep period has been that it is due to sleep itself. The present results show that the sleep-related effect is largely limited to the period immediately after sleep onset. Suggestions as to the specific mechanisms involved have included a reduction in muscle activity (Kleitman, 1963; Kreider et al., 1958; Mason & Benedict, 1934) and a reduction in thermoregulatory control (Kreider & Buskirk, 1957; Kreider et al., 1958; Milan & Evonuk, 1967). Changes in muscle tension are unlikely candidates for the changes reported in this study. Although it has been known for some time that muscle activity is reduced

during sleep (Kleitman, 1963), marked changes have not been reported as being systematically associated with sleep onset. The reduction in MR at sleep onset is also unlikely to be due to a reduction in thermogenesis as a consequence of the downward regulation of the thermoregulatory set point, because the subjects in this and several other experiments were maintained at thermoneutrality. Thus they were presumably not expending energy on thermoregulatory heat production before the adjustment.

However, a general reduction in metabolic activity secondary to a fall in body temperature may contribute to the fall in metabolic rate. O_2 consumption is thought to vary by two- to threefold for a 10°C change in body temperature. If this change (Q_{10}) is assumed to be 2.5 and the fall in body temperature at sleep onset is assumed to be 0.4°C, then the fall in O_2 consumption would be approximately 3.6% (Schmidt-Neilson, 1979). Similarly, if the total sleep period fall in body temperature was 1°C, then the expected fall in O_2 consumption would be 8.8%. Although body temperature was not measured in the present study and the Q_{10} factor is only a rough estimate of the effect of body temperature on O_2 consumption, the magnitude of the effect is such that it is possible that it accounts for some of the reduction in MR, both at sleep onset and during the subsequent sleep period. However, a further cautionary note should be added with respect to the changes at sleep onset. It is not clear that the fall in deep body temperature would be sufficiently rapid to produce the relatively rapid changes in MR observed in the present study. A simultaneous analysis of the time course of the two variables at sleep onset is clearly required. Thus the particular metabolic activities that change to produce the rapid reduction in MR at sleep onset remain uncertain, although a close link with body temperature is likely. The possibility of an active adjustment in metabolic processes should also be considered.

The results indicate that, in addition to a rapid change at sleep onset, MR varies over the night as a function of circadian cycle. The circadian variability in MR clearly parallels the body temperature rhythm. However, for reasons stated above, the changes in MR are unlikely to reflect reduced thermogenesis, because the subjects were run under thermoneutral conditions and should not have been expending energy to maintain body temperature. Nevertheless, as for sleep onset, the changes in body temperature and MR are likely to be linked.

The average reduction in MR over the night was 30% of the initial wake level. Approximately 16.3% was due to changes in the first half hour following sleep onset and the remaining 13.7% to changes during the remainder of the sleep period. The total fall in MR was greater than has been reported previously. Robin et al. (1958) found a reduction of 22%, but other papers have reported falls of close to 10% (Mason & Benedict, 1934; Shapiro et al., 1984; White et al., 1985).

Two major factors may have contributed to the larger reduction in oxygen consumption observed in this study. Perhaps the most critical was that particular efforts were made to ensure that subjects were awake during wakefulness measurements. Although this may appear obvious, what has not been recognised until recently is that even brief periods of drowsiness can be associated with dramatic falls in MR (Colrain et al., 1987). However, subjects are unaware that they have drifted into stage 1 sleep and if aroused, vehemently deny they were asleep. Thus, to ensure that subjects do not enter stage 1 sleep, it is critical that they be monitored electrophysiologically. Failure to do so is likely to result in artifactually low estimates of awake MR. In the present study, 4.9% of the wake recordings was scored as stage 1 sleep and discarded from the analysis.

A related issue is that a procedure frequently used in metabolic studies of sleep, is measurement of the awake MR during the period between lights out and the attainment of sleep. This is likely to produce low estimates of MR in the awake state because it is inevitably going to be contaminated by periods of stage 1 sleep. In addition, MR is known to be lower with eyes closed and in the absence of visual stimulation (Asmussen, 1977; Shea, Walter, Pelley, Murphy & Guz, 1987). The measurements in this study will have been elevated by having the lights on and the subjects' having their eyes open.

The increase in MR during the seventh and eighth hours of sleep reported in some studies (Haskell et al., 1981a; Kreider et al., 1958; Kreider & Iampietro, 1959; Shapiro et al., 1984: White et al., 1985) was not observed in the present study. This result was most likely because data collection was not continued for sufficient time. In this study, recording generally ceased 7 hours after subjects had first gone to bed Inspection of Figure 5.1 suggests that in conditions 1 and 2, MR had reached asymptotal values by the end of the recording period.

Carbon dioxide production was not measured in the present study, and thus MR was inferred from O_2 consumption. However, it is unlikely that CO_2 measurements would have altered the conclusions. One study (Robin et al., 1958) has reported that the respiratory quotient (RQ) remains unchanged from wakefulness to sleep, whereas another reported a change from 0.85 to 0.82 (White et al., 1985). Thus if a change did occur in the present study, it would be likely that it involved increased fat utilization and a reduction in energy expenditure for each litre of O_2. This suggests that the present results may, in fact underestimate slightly the magnitude of the change in MR from wakefulness to sleep. However, it should also be noted that the magnitude of the effects observed in this study is such that the consequences of a change in RQ would be negligible.

6.2 Differences in oxygen consumption between sleep stages.

6.2.1 The effect of contiguous stages methodology.

Two methods of comparison of oxygen consumption were used: total minutes in a particular sleep stage and contiguous periods of particular sleep stages. The method of comparison using total minutes per night in each sleep stage revealed that oxygen consumption was not significantly lower in SWS than in stage 2 sleep. In fact, in the majority of comparisons, SWS had a mean oxygen consumption significantly greater than that of stage 2. The usual explanation of this result is that it is because of a failure to control for the circadian variation in MR.

The typical methodology to overcome the problem of circadian variation has been to compare stages which are contiguous, and in contrast to the first comparison, using this method the results showed a greater proportion of comparisons having a lower oxygen consumption in SWS than stage 2. However, when SWS follows stage 2, the results can be explained by a circadian factor. In addition, when the subject moved from SWS to Stage 2, there was a tendency for oxygen consumption to be less in the following Stage 2, but this trend was not as strong as the decrease in oxygen consumption observed in the change from Stage 2 to SWS. This result cannot be explained on the basis of a circadian factor and suggests a specific stage effect. However, as will be explained in section 6.2.2, body movements exert a considerable elevating effect on oxygen consumption following the arousal, and in this study, 91.7% of the time the transition from SWS to stage 2 involved a body movement. This suggests an explanation why stage 2 following SWS has a higher oxygen consumption.

6.2.2 The effect of movement arousals.

The assessment of the effect of body movement arousals on sleep period oxygen consumption suggests that the stage effects found by previous studies are artifactual. Comparison of periods of sleep with and without an embedded arousal shows different patterns of oxygen consumption for the two conditions. In the no arousal condition, there was a gradual and relatively consistent decrease in oxygen consumption across the period. In the arousal period however, there was a gradual increase in oxygen consumption prior to the arousal, and a further increase post arousal. In addition, a period of instability was evident for up to seven minutes post arousal before a consistent downward trend in oxygen consumption was resumed.

When only periods involving transitions from stage 2 to stage 2 were considered, there was no evidence of the gradual increase in oxygen consumption prior to the arousal and the increase from pre to post arousal was more evident. The pattern in periods involving stage 4 to stage 2 transitions appears to mirror the pattern in the combined data suggesting the slight increase in oxygen consumption in the arousal condition prior to the arousal is associated with this type of transition. It should be noted that in these transitions, the subjects remained in Stage 4 with no decrease in SWS density up until the movement arousal. The slight increase in oxygen consumption observed prior to the arousal is not due to a change to Stage 2 sleep, but rather appears to be some anticipation of the movement arousal itself.

Since each individual period with an arousal was matched with a no arousal period, any difference between the two cannot be attributed to circadian variation. Also, to eliminate the possibility of contamination from the sleep onset effect, no period was selected which included the fifteen minutes following sleep onset.

It appears from the present results, that movement arousals have a significant elevating effect on oxygen consumption during sleep, and that the period of metabolic disturbance following a movement arousal is noticeably longer than previous researchers have suggested. An explanation of the gradual increase in oxygen consumption pre arousal when the subject is in stage 4 sleep is not available at present, however one may speculate that perhaps there is some change which occurs prior to an arousal which takes a few minutes to manifest itself in an observable body movement and EEG disruption.

6.3 Methodological implications.

In investigations which rely on measurements of metabolism, the need to account for individual differences cannot be overemphasized. Considerable differences may occur in the subjects' response under different experimental conditions if there is no control for differences in factors such as weight, height, calorific intake and energy expenditure. Age is also an important factor, especially in view of the changes in amount and depth of SWS as a person ages. Also, given what is known about the differences in metabolism between males and females, control for sex differences should be a standard procedure in future experiments.

It is of paramount importance that subjects be trained extensively in the use of the mask. This is evident given the results observed in this study, where the lowest amount of disturbed sleep was found in the two subjects who were most experienced in wearing the mask. Both these subjects had participated in other experiments prior to the commencement of this study and this experience was reflected in the better quality of their sleep.

It now appears that oxygen consumption does not intrinsically differ between sleep stages, but is influenced by sleep onset, a circadian factor and by movement

arousals during the sleep period. As there is some evidence of a similar nightime pattern of MR and body temperature, future research should be directed towards a closer investigation of the relationship between metabolic rate and body temperature. Research in this area would enable a determination of how closely the two rhythms are aligned, and the extent of their interrelationship during sleep. In addition, the question of metabolic rate response to changes in peripheral temperature relative to core body temperature could be investigated.

6.4 Theoretical implications.

Whilst it has been stated previously in this thesis that the study was not a test of the Restorative and Conservative theories of the function of sleep, the results of the experiment throw some doubt on one of their major points. It would appear on the basis of these results, that the level of metabolic rate observed during SWS may not be a result of a particular property of that sleep state, but may rather be a result of the combination of the placement of SWS within the total sleep period, and the influence on sleep period metabolic rate of a circadian and a sleep onset effect.

6.5 Conclusions.

The circadian factor produced a gradual reduction in MR over the sleep period, while sleep was associated with a rapid reduction in MR at the point of sleep onset and a slower change over the first ten to thirty minutes of sleep. The results of previous studies can be interpreted as being due to the combined effect of circadian and sleep influences, and not due to specific differences in MR between separate sleep stages. These would appear to be artifactual. In addition, present results indicate that the period of metabolic disturbance following a movement arousal is longer than that suggested by previous researchers.

REFERENCES

REFERENCES

Adam, K. (1980). Sleep as a restorative process and a theory to explain why. Progress in Brain Research, 53, 289-305.

Adam, K. and Oswald, I. (1977). Sleep is for tissue restoration. Journal of the Royal College of Physicians, 11, 376-388.

Adam, K. and Oswald, I. (1983). Protein synthesis, bodily renewal and the sleep-wake cycle. Clinical Science, 65, 561-567.

Adamson, L., Hunter, W.M., Ogunremi, O.O., Oswald, I. and Percy-Robb, I.W. (1974). Growth hormone increase during sleep after daytime exercise. Journal of Endocrinology, 62, 473-478.

Agnew, H.W.Jr., Webb, W.B. and Williams, R.L. (1964). The effects of stage four sleep deprivation. Electroencephalography and Clinical Neurophysiology, 17, 68-70.

Agnew, H.W.Jr., Webb, W.B. and Williams, R.L. (1966). The first night effect: An EEG study of sleep. Psychophysiology, 2(3), 263-266.

Agnew, H.W.Jr., Webb, W.B. and Williams, R.L. (1967a). Comparison of stage four and 1-REM sleep deprivation. Perceptual and Motor Skills, 24, 851-858.

Agnew, H.W.Jr., Webb, W.B. and Williams, R.L. (1967b). Sleep patterns in late middle age males: An EEG study. Electroencephalography and Clinical Neurophysiology, 23, 168-171.

Akerstedt, T. and Gillberg, M. (1986). A dose-response study of sleep loss and spontaneous sleep termination. Psychophysiology, 23, 293-297.

Alford, F.P., Baker, H.W.G. and Burger, H.G. (1973). The secretion rate of human growth hormone. I. Daily secretion rates, effect of posture and sleep. Journal of Clinical Endocrinology and Metabolism, 37(4), 515-520.

Allison, T. and Cicchetti, D.V. (1976). Sleep in mammals: Ecological and constitutional correlates. Science, 194, 732-734.

Allison, T. and Van Twyver, H. (1970). The evolution of sleep. Natural History, 79(2), 57-65.

Altschuler, K.Z. and Brebbia, D.R. (1967). Body movement artifact as a contaminant in psychophysiological studies of sleep. Psychophysiology, 3, 327-335.

Asmussen, E. (1977). Regulation of respiration: 'The blackbox'. Acta Physiologica Scandinavia, 99, 85-90.

Atkinson, D.E. (1968). The energy charge of the adenylate pool as a regulatory parameter. Interaction with feedback modifiers. Biochemistry, 7, 4030-4034.

Baekland, F. and Lasky, R. (1966). Exercise and sleep patterns in college athletes. Perceptual and Motor Skills, 23, 1203-1207.

Barrett, J., Morris, M. and Lack, L. (1987). The sleep evoked decrease of body temperature. Sleep Research, 16, 596. (Abstract)

Beck, U., Brezinova, V., Hunter, W.M.,and Oswald, I. (1975).Plasma growth hormone and slow-wave sleep increase after interruption of sleep. Journal of Clinical Endocrinology, 40, 812-815.

Berger, R. (1975). Bioenergetic functions of sleep and activity rhythms and their possible relevance to aging. Federation Proceedings, 34(1), 97-102.

Berger, R. (1984). Slow wave sleep, shallow torpor and hibernation: Homologous states of diminished metabolism and body temperature. Biological Psychology, 19, 305-326.

Berger, R.J. and Oswald, I. (1962). Effects of sleep deprivation on behaviour, subsequent sleep, and dreaming. Journal of Mental Science, 108, 457-465.

Besser, G.M., Butler, P.W.P., Landon, J. and Rees, L. (1969). Influence of amphetamines on plasma corticosteroid and growth hormone levels. British Medical Journal, 4, 528-530.

Bonnet, M.H. (1980). Sleep performance and mood after the energy expenditure equivalent of 48 hours of sleep deprivation. Psychophysiology, 17, 56-63.

Bonnet, M.H. (1986). Performance and sleepiness as a function of frequency and placement of sleep disruption. Psychophysiology, 23, 263-271.

Brebbia, D.R. and Altschuler, K.Z. (1965). Oxygen consumption rate and electroencephalographic stage of sleep. Science, 150, 1621-1623.

Browman, C.P. and Tepas, D.I. (1976). The effects of presleep activity on all night sleep. Psychophysiology, 13, 536-540.

Bunnell, D.E., Bevier, W.C. and Horvath, S.M. (1983). Nocturnal sleep, cardiovascular function, and adrenal activity following maximum-capacity exercise. Electroencephalography and Clinical Neurophysiology, 56,186-189.

Buskirk, E.R., Thompson, R.H., Moore, R. and Whedon, G.D. (1960). Human energy expenditure studies in the National Institute of Arthritis and Metabolic Diseases Metabolic Chamber. I. Interaction of cold environment and specific dynamic action effect. II. Sleep. American Journal of Clinical Nutrition, 8, 602-613.

Candas, V., Libert, J.P., Vogt, J.J., Ehrhart, J. and Muzet, A. (1979) Body temperature during sleep under different thermal conditions. In P.O. Fanger and O. Valbjorn (Eds.), Indoor climate: effects on human comfort, performance and health. Copenhague: Danish Building Research Institute, 763-775.

Colrain, I.M., Trinder, J., Fraser, G. and Wilson, G.V. (1987). Ventilation during sleep onset. Journal of Applied Physiology, 63, 2067-2074.

Conroy, R.T.W.L. and Mills, J.N. (1970). Human circadian rhythms. London: J. & A. Churchill.

Crisp, A.H., Stonehill, E., Fenton, G.W. and Fenwick, P.B.C. (1973). Sleep patterns in obese patients during weight reduction. Psychotherapy and Psychosomatics, 22, 159-165.

Desjardins, J., Healey, T. and Broughton, R. (1974). Early evening exercise and sleep. Sleep Research, 3, 31. Abstract

Dunleavy, D.L.F., Oswald, I., Brown, P. and Strong, J.A. (1974). Hyperthyroidism, sleep and growth hormone. Electroencephalography and Clinical Neurophysiology, 36, 259-263.

Dunleavy, D.L.F., Oswald, I. and Strong, J.A. (1973). Fenfluramine and growth hormone release. British Medical Journal, iii, 48.

Duron,B., Andrac,C. and Laval,P. (1968). Ventilation pulmonaire globale, CO2 alveolaire et consommation d'oxygene, au cours du sommeil chez l'Homme normal. Comptes Rendus, Societe de Biologie, 162, 139-145.

Feinberg, I. (1974). Changes in sleep cycle patterns with age. Journal of Psychiatric Research, 10, 283-306.

Feinberg, I. and Carlson, V.R. (1968). Sleep variables as a function of age in man. Archives of General Psychiatry, 18, 239-250.

Feinberg, I., Koresko, R.L. and Heller, H.C. (1967). EEG sleep patterns as a function of normal and pathological aging in man. Journal of Psychiatric Research, 5, 107-144.

Fisher, L.B. (1968). The diurnal mitotic rhythm in the human epidermis. British Journal of Dermatology, 80, 75-80.

Fuller, C.A. and Sulzman, F.M. (1982). Circadian control of body temperature in primates. In J. Aschoff, S. Daan and G. Groos (Eds.), Vertebrate Circadian Systems, (pp. 224-236), Berlin: Springer-Verlag.

Gander, P.H., Graeber, R.C. and Connell, L.J. (1985). Masking of the circadian rhythms of heart rate and core temperature by the rest/activity rhythm in man. Sleep Research, 14, 298. Abstract.

Gander, P.H. and Kronauer, R.E. (1984). Phase shifting two coupled circadian pacemakers: implications for jet lag. American Journal of Physiology, 249, 704-709.

Gillberg, M. and Akerstedt, M. (1982). Body temperature and sleep at different times of day. Sleep, 5(4), 378-388.

Griffin, S.J. and Trinder, J. (1978). Physical fitness, exercise and human sleep. Psychophysiology, 15, 447-450.

Grollman, A. (1930). Physiological variations in the cardiac output of man. XI The pulse rate, blood pressure, oxygen consumption, arterio-venous oxygen differences, and cardiac output during normal sleep. American Journal of Physiology, 95, 274-284.

Hartman, E.L. (1973). The functions of sleep. New Haven: Yale University Press.

Haskell, E.H., Palca, J.W., Walker, J.M., Berger, R.J. and Heller, H.C. (1981a). Metabolism and thermoregulation during stages of sleep in humans exposed to heat and cold. Journal of Applied Physiology, 51, 948-954.

Haskell, E.H., Palca, J.W., Walker, J.M., Berger, R.J. and Heller, H.C. (1981b). The effects of high and low ambient temperatures on human sleep stages. Electroencephalography and Clinical Neurophysiology, 51, 494-501.

Hauri, P. (1968). Effects of evening activity on early night sleep. Psychophysiology, 4, 267-277.

Heller, H.C. (1979). Hibernation: Neural aspects. Annual Review of Physiology, 41, 305-321.

Heller, H.C. and Glotzbach, S.F. (1977). Thermoregulation during sleep and hibernation. Environmental Physiology II, International Review of Physiology, 15, 147-188.

Heller, H.C., Walker, J.M., Florant, G.L., Glotzbach, S.F. and Berger, R. (1978). Sleep and hibernation: Electrophysiological and thermoregulatory homologies. In L. Wang and J.W. Hudson (Eds.), Strategies in Cold: Natural Torpidity and Thermogenesis, (pp. 225-265), New York: Academic Press.

Honda, Y., Takahashi, K., Takahashi, S., Azumi, K., Irie, M., Sakuma, M., Tsushima, T. and Shizume, K. (1979). Growth hormone secretion during nocturnal sleep in normal subjects. Journal of Clinical Endocrinology and Metabolism, 29, 20-29.

Horne, J.A. (1977). Factors relating to energy conservation during sleep in mammals. Physiological Psychology, 5(4), 403-408.

Horne, J.A. (1979). Restitution and human sleep: A critical review. Physiological Psychology, 7, 115-125.

Horne, J.A. (1981). The effect of exercise upon sleep: a critical review. Biological Psychology, 12, 241-290.

Horne, J.A. (1983). Human sleep and tissue restoration: some qualifications and doubts. Clinical Science, 65, 569-578.

Horne, J.A. and Moore, V.J. (1985). Sleep EEG effects of exercise with and without additional body cooling. Electroencephalography and Clinical Neurophysiology, 60, 33-38.

Horne, J.A. and Staff, L.H.E. (1983). Exercise and sleep: Body-heating effects. Sleep, 6, 36-41.

Johnson, L.C., Slye, E.S. and Dement, W.C. (1965). Electroencephalographic and autonomic activity during and after prolonged sleep deprivation. Psychosomatic Medicine, 27, 415-423.

Kahn, E. and Fisher, C. (1969). The sleep characteristics of the normal aged male. The Journal of Nervous and Mental Disease, 148(5), 477-494.

Kales, A., Heuser, G., Jacobson, A., Kales, J.D. Hanley, J., Zweizig, J.R. and Paulson, M.J. (1967). All night sleep studies in hypothyroid patients, before and after treatment. Journal of Clinical Endocrinology and Metabolism, 27, 1593-1599.

Kales, A.T., Wilson, J.D., Kales, A., Jacobson, A., Paulson, M.J., Kollar, E. and Walter, D. (1967). Measurement of all-night sleep in normal elderly persons: effect of aging. Journal of the American Geriatric Society, 15, 405.

Karacan, I., Rosenbloom, A.L., Londono, J.H., Salis, P.J., Thornby, J.I.,and Williams, R.L. (1973).Sleep and the sleep-growth hormone (GH) response during acute fasting. Sleep Research, 2, 197.

Karacan, I., Rosenbloom, A.L., Williams, R.L., Finley, W.W. and Hursch, C.J. (1971). Slow wave sleep deprivation in relation to plasma growth hormone concentration. Behavioural Neuropsychiatry, 2, 11-14.

Kleitman, N. (1963). Sleep and wakefulness. Chicago: University of Chicago Press.

Kleitman, N. and Doktorsky, A. (1933). The effects of the position of the body and sleep on rectal temperature in man. Studies on the Physiology of Sleep VII, 104, 341-343.

Kleitman, N. and Ramsaroop, A. (1948). Periodicity in body temperature and heart rate. Endocrinology, 43, 1-20.

Korner, A. (1965). Growth hormone control of biosynthesis of protein and ribonucleic acid. Recent Progress in Hormone Research, 21, 205-236.

Kreider, M.B. and Buskirk, E.R. (1957). Supplemental feeding and thermal comfort during sleep in the cold. Journal of Applied Physiology, 11(3), 339-343.

Kreider, M.B., Buskirk, E.R. and Bass, D.E. (1958). Oxygen consumption and body temperatures during the night. Journal of Applied Physiology, 12(3), 361-366.

Kreider, M.B. and Iampietro, P.F. (1959). Oxygen consumption and body temperature during sleep in cold environments. Journal of Applied Physiology, 14, 765-767.

Kupfer, D, Sewitch, D.E., Epstein, L.H., Bulik, C.R., McGowen, C.R. and Robertson,R.J. (1985) Exercise and subsequent sleep in male runners: failure to support the SWS-mood-exercise hypothesis. Neuropsychobiology, 14, 5-12.

Lacey, J.H., Crisp, A.H., Kalucy, R.S., Hartmann, M.K. and Chen, C.N. (1975). Weight gain and the sleeping electroencephalogram: Study of 10 patients with Anorexia Nervosa. British Medical Journal , 4, 556-558.

Lack, L.C., Balfour, R. and Kalucy, R. (1985). The circadian rhythm of body temperature in poor sleepers. Sleep Research, 14, 301. Abstract

Lewis, S.A., Oswald, I. and Dunleavy, D.L.F. (1971). Chronic fenfluramine administration: some cerebral effects. British Medical Journal, 3, 67-70.

MacFadyen, U.M., Oswald, I. and Lewis, S.A. (1973). Starvation and human slow-wave sleep. Journal of Applied Physiology, 35(3), 391-394.

Maloletnev, V.I., Telia, Z.A. and Tchatchanashvili, M.G. (1977). Changes in the structure of night sleep in man after intensive muscular work. Sech Physiol J USSR, 63, 11-20.

Mason, E.D. and Benedict, F.G. (1934). The effect of sleep on human basal metabolism, with particular reference to South Indian Women. American Journal of Physiology, 108, 372-382.

Matsumoto, K., Saito, Y., Abe, M. and Furumi, K. (1984) The effects of daytime exercise on nightsleep. Journal of Human Ergology, 13, 31-36.

Mauer, A.M. (1965). Diurnal variation of proliferative activity in human bone marrow. Blood, 26, 1-7.

Milan, F.A. and Evonuk, E. (1967). Oxygen consumption and body temperature of Eskimos during sleep. Journal of Applied Physiology, 22, 565-567.

Montgomery, I., Trinder, J., Harriss, D. and Fraser, G. (1988). Physical exercise and sleep: the effect of age and sex of the subjects and type of exercise. Acta Physiologica Scandinavica, 133, Supp 574, 36-40.

Montgomery, I., Trinder, J. and Paxton, S.J. (1982). Energy expenditure and total sleep time: The effect of physical exercise. Sleep, 5, 159-168.

Montgomery, I., Trinder, J., Paxton, S.J. and Fraser, G. (1987). Aerobic fitness and exercise: Effect on the sleep of young and older adults. Australian Journal of Psychology, 39, 259-272.

Montgomery, I., Trinder, J., Paxton, S.J., Fraser, G., Meaney, M. and Koebin, G. (1985). Sleep disruption following a marathon. Journal of Sports Medicine and Physical Fitness, 25, 69-74.

Muzet, A., Naitoh, P., Townsend, R.E. and Johnson, L.C. (1972). Body movements during sleep as a predictor of stage change. Psychonomic Science, 29, 7-10.

Oswald, I. (1970). Sleep, the great restorer. New Scientist, 46, 170-172.

Oswald, I. (1976). The function of sleep. Postgraduate Medical Journal, 52, 15-18.

Oswald, I. (1980). Sleep as a restorative process: Human clues. Progress in Brain Research, 53, 279-288.

Oswald, I., Dunleavy, D.L.F. and Strong, J.A. (1972). Hyperthyroidism and the sleeping brain. Hormone, 3, 278.

Parker, D.C., Rossman, L.G. and VanderLaan, E.F. (1972). Persistence of rhythmic human growth hormone release during sleep in fasted and nonisocalorically fed normal subjects. Metabolism, 21, 241-252.

Parker, D.C., Sassin, J.F., Mace, J.W., Gotlin, R.W. and Rossman, L.G. (1969). Human growth hormone release during sleep: Electroencephalographic correlation. Journal of Clinical Endocrinology and Metabolism, 29, 871-874.

Paxton, S.J., Montgomery, I., Trinder, J., Newman, J. and Bowling, A. (1982). Sleep after exercise of variable intensity in fit and unfit subjects. Australian Journal of Psychology, 34, 289-296.

Paxton, S.J., Trinder, J. and Montgomery, I. (1983). Does aerobic fitness affect sleep? Psychophysiology, 20, 320-324.

Paxton, S.J., Trinder, J., Shapiro, C.M., Adam, K., Oswald, I. and Graf, K.J. (1984). The effect of physical fitness and body composition on sleep and sleep related hormone concentrations. Sleep, 7, 339-346.

Ramm, P. and Smith, C.T. (1987). Cerebral protein synthesis during slow wave and REM sleep in the rat. Sleep Research, 16, 67.

Rechtschaffen, A. and Kales, A. (1968). A manual of standardized terminology, techniques and scoring system for sleep stages of human subjects. Washington, D.C.: US Government Printing Office.

Rechtschaffen, A. and Maron, A.B. (1964). The effect of amphetamine on the sleep cycle. Electroencephalography and Clinical Neurophysiology, 16, 438-445.

Robin, E.D., Whaley, R.D., Crump, C.H. and Travis, D.M. (1958). Alveolar gas tensions, pulmonary ventilation and blood pH during physiologic sleep in normal subjects. Journal of Clinical Investigation, 37, 981-989.

Sassin, J.F., Parker, D.C., Mace, J.C., Gotlin, R.W., Johnson, L.C. and Rossman, L.G. (1969). Human growth hormone release: Relation to slow-wave sleep and sleep-waking cycles. Science, 165, 513-515.

Scheving, L.E. (1959). Mitotic rhythm in the human epidermis. Anatomical Record, 135, 7-20.

Schmidt-Neilson, K. (1979). Animal Physiology: Adaptation and Environment (2nd ed.) Cambridge, U.K.: Cambridge University Press.

Schwartz, R.S., Ravussin, E., Massari, M., O'Connell, M. and Robbins, D.C. (1985). The thermic effect of carbohydrate versus fat feeding in man. Metabolism, 34(3), 285-293.

Shapiro, C.M., Bortz, R., Mitchell, D., Bartel, P., and Jooste, P.L. (1981). Slow wave sleep: A recovery period after exercise. Science, 214, 1253-1254.

Shapiro, C.M., Goll, C.C., Cohen, G.R. and Oswald, I. (1984). Heat production during sleep. Journal of Applied Physiology: Respiration, Environment, Exercise Physiology, 56(3), 671-677.

Shapiro, C.M., Griesel, R.D., Bartel, P.R. and Jooste, P.L. (1975). Sleep patterns after graded exercise. Journal of Applied Physiology, 39, 187-190.

Shapiro, C.M. and Verschoor, G.J. (1979). Sleep patterns after a marathon. South African Journal of Science, 75, 415-416.

Shea, S.A., Walter, J., Pelley, C., Murphy, K. & Guz, A. (1987). The effect of visual and auditory stimuli upon resting ventilation in man. Respiratory Physiology, 68, 345-357.

Shimizu, A. and Himwich, H.E. (1968). The ontogeny of sleep in kittens and young rabbits. Electroencephalography and Clinical Neurophysiology, 24, 307.

Takahashi, Y., Kipnis, D.M. and Daughaday, W.H. (1968). Growth hormone secretion during sleep. The Journal of Clinical Investigation, 47, 2079-2090.

Taub, J.M. and Berger, R.J. (1976). Effects of acute sleep pattern alteration depend upon sleep duration. Physiological Psychology, 4(4), 412-420.

Terjung, R.L. and Winder, W.W. (1975). Exercise and thyroid function. Medicine and Science in Sports, 7, 20-26.

Tilley, A., Donohoe, F. and Hensby, S. (1987). The homeostasis of slow wave sleep. Sleep Research, 16, 539.

Torsvall, L., Akerstedt, T. and Lindbeck, G. (1984). Effects on sleep stages and EEG power density of different degrees of exercise in fit subjects. Electroencephalography and Clinical Neurophysiology, 57, 347-353.

Trinder, J., Bruck, D., Paxton, S.J., Montgomery, I. and Bowling, A. (1982). Physical fitness, exercise, age and human sleep. Australian Journal of Psychology, 34, 131-138.

Trinder, J., Montgomery, I. and Paxton, S.J. (1988). The effect of exercise on sleep: the negative view. Acta Physiologica Scandinavica, 133, Supp 574, 14-20.

Van den Noort, S. and Brine, K. (1970). Effect of sleep on brain labile phosphates and metabolic rate. American Journal of Physiology, 218, 1434-1439.

Walker, J.M. and Berger, R. (1980). Sleep as an adaptation for energy conservation functionally related to hibernation and shallow torpor. Progress in Brain Research, 53, 255-278.

Walker, J.M., Floyd, T.C., Fein, G., Cavness, C., Lualhati, R. and Feinberg, I. (1978). Effects of exercise on sleep. Journal of Applied Physiolgy, 44, 945-951.

Walker, J.M., Garber, A., Berger, R. and Heller, H.C. (1979). Sleep and estivation (shallow torpor): Continuous processes of energy conservation. Science, 204, 1098-1100.

Walker, J.M., Glotzbach, S.F., Berger, R.J. and Heller, H.C. (1977). Sleep and hibernation in ground squirrels (Citellus spp.): Electrophysiological observations. American Journal of Physiology, 233, R231-R221.

Walker, J.M., Haskell, E.H., Berger, R.J. and Heller, H.C. (1981). Hibernation at moderate temperatures: a continuation of slow wave sleep. Experientia, 37, 726-728.

Webb, W.B. and Agnew, H.W.Jr. (1971). Stage 4 sleep: Influence of time course variables. Science, 174, 1354-1356.

Webb, W.B. and Agnew, H.W. (1973). Effects on performance of high and low energy expenditure during sleep deprivation. Perceptual and Motor Skills, 37, 511-514.

Webb, W.B. and Agnew, H.W. (1974). Sleep and waking in a time free environment. Aviation and Space Environmental Medicine, 45, 617-622.

Webb, P. and Hiestand, M. (1975). Sleep, metabolism and age. Journal of Applied Physiology, 38, 257-262.

White, D.P., Weil, J.V. and Zwillich, C.W. (1985). Metabolic rate and breathing during sleep. Journal of Applied Physiology, 59(2), 384-391.

Williams, R.L., Agnew, H.W.Jr. and Webb, W.B. (1964). Sleep patterns in young adults: An EEG study. Electroencephalography and Clinical Neurophysiology, 17, 376-381.

Williams, R.L., Agnew, H.W.Jr. and Webb, W.B. (1966). Sleep patterns in the young adult female: An EEG study. Electroencephalography and Clinical Neurophysiology, 20, 264-266.

Williams, R.L., Karacan, I. and Hursch, C.J. (1974). EEG of Human Sleep. New York: Wiley.

Yamashiro, Y., Fukushima, Y., Okudaira, N., Suzuku, Y. and Nishi, I. (1987). Gas exchange ratio during sleep. Sleep Research, 16, 60.

Zepelin, H. and Rechtschaffen, A. (1974). Mammalian sleep, longevity and energy metabolism. Brain, Behaviour and Evolution, 10, 425-470.

Zir, L.M., Smith, R.A. and Parker, D.C. (1971). Human growth hormone release in sleep: effect of daytime exercise. Journal of Clinical Endocrinology, 32, 662-665.

Zloty, R.B., Burdick, J.A. and Adamson, J.D. (1973). Sleep of long distance runners. Activitas Nervosa Superior, 15, 217-221.

Zulley, J. and Wever, R.A. (1982). Interaction between the sleep-wake cycle and the rhythm of rectal temperature. In J. Aschoff, S. Daan and G. Groos (Eds.), Vertebrate Circadian Systems, (pp. 224-236), Berlin: Springer-Verlag.

APPENDICES

APPENDIX 1

List of Papers from the Thesis.

Fraser, G., Trinder, J., Colrain, I.M. & Montgomery, I. (1989). Effect of sleep and circadian cycle on sleep period energy expenditure. Journal of Applied Physiology, 66(2), 830-836.

APPENDIX 2

Anova summary table for sleep onset analysis (p. 44).

Source of Variation	df	Sum of Squares	Mean Square	F	p
Subjects	4	39232.055	9808.014		
C	2	11853.429	5926.714	16.034	.0016
Error	8	2957.085	369.636		
O	1	12423.675	12423.675	29.949	.0054
Error	4	1659.297	414.824		
CO	2	603.446	301.723	1.568	.2663
Error	8	1539.167	192.396		

Anova summary table for Hour 1 analysis (p. 47).

Source of Variation	df	Sum of Squares	Mean Square	F	p
Subjects	4	25825.068	6456.267		
S	1	35.378	35.378	.195	.6817
Error	4	726.312	181.578		
C	1	3650.402	3650.402	11.180	.0287
Error	4	1306.068	326.517		
SC	1	3474.248	3474.248	11.700	.0268
Error	4	1187.752	296.938		

Anova summary table for Hour 7 analysis (p. 47).

Source of Variation	df	Sum of Squares	Mean Square	F	p
Subjects	4	20879.468	5219.867		
S	1	4345.352	4345.352	18.860	.0122
Error	4	921.588	230.397		
C	1	1241.888	1241.888	17.430	.0140
Error	4	285.002	71.251		
SC	1	1827.872	1827.872	11.768	.0265
Error	4	621.278	155.320		

APPENDIX 3

Anova summary table for circadian analysis (p. 47).

Source of Variation	df	Sum of Squares	Mean Square	F	p
Subjects	4	37209.129	9302.282		
C	1	10300.827	10300.827	24.400	.0078
Error	4	1688.685	422.171		
T	2	7347.843	3673.921	21.482	.0006
Error	8	1368.187	171.023		
CT	2	733.752	366.876	3.872	.0667
Error	8	758.091	94.761		

Anova summary table for adjusted circadian analysis (p. 48).

Source of Variation	df	Sum of Squares	Mean Square	F	p
Subjects	4	38609.370	9652.343		
C	1	12048.048	12048.048	38.000	.0035
Error	4	1268.222	317.056		
T	2	5647.885	2823.942	12.828	.0032
Error	8	1761.172	220.147		
CT	2	245.418	122.709	1.165	.3598
Error	8	842.812	105.352		

APPENDIX 4

Anova summary table for combined arousal data (p. 63).

Source of Variation	df	Sum of Squares	Mean Square	F	p
A	1	21914.206	21914.206	.456	.5015
Error	72	3457403.286	48019.490		
P	1	71.368	71.368	.112	.7389
AP	1	3196.368	3196.368	5.015	.0282
Error	72	45893.114	637.404		
T	9	2331.784	259.087	2.393	.0114
AT	9	1396.530	155.170	1.433	.1700
Error	648	70164.335	108.278		
PT	9	612.314	68.035	.519	.8612
APT	9	1368.882	152.098	1.161	.3176
Error	648	84908.454	131.032		

Anova summary table for Stage 2 to Stage 2 transitions only (p. 65).

Source of Variation	df	Sum of Squares	Mean Square	F	p
A	1	2125.210	2125.210	.066	.8001
Error	18	579499.590	32194.422		
P	1	19.360	19.360	.023	.8813
AP	1	3214.890	3214.890	3.807	.0668
Error	18	15199.150	844.397		
T	9	2078.400	230.933	2.459	.0119
AT	9	336.290	37.366	.398	.9347
Error	162	15214.510	93.917		
PT	9	1336.840	148.538	1.078	.3819
APT	9	140.810	15.646	.114	.9993
Error	162	22324.950	137.808		

Anova summary table for Stage 4 to Stage 2 transitions only (p. 67).

Source of Variation	df	Sum of Squares	Mean Square	F	p
A	1	21094.008	21094.008	.382	.5392
Error	52	2870930.996	55210.211		
P	1	157.934	157.934	.278	.6004
AP	1	1003.408	1003.408	1.765	.1898
Error	52	29566.107	568.579		
T	9	1434.353	159.373	1.399	.1856
AT	9	1522.242	169.138	1.485	.1505
Error	468	53306.856	113.904		
PT	9	963.538	107.060	.831	.5882
APT	9	1798.582	199.842	1.550	.1277
Error	468	60324.930	128.899		