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Effects of Selective Sleep Deprivation on Sleep-Linked Prolactin and Growth Hormone Secretion

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Summary. 1. The secretion of prolactin and growth-hormone (hGH) was investigated during sleep in 10 healthy volunteers (8 males and 2 females): The comparison of one baseline night, one night after daytime physical exercise, and one night with selective deprivation of sleep stages 3 and 4 and paradoxical sleep showed clear differences of prolactin and hGH secretion during sleep.

2. Prolactin secretion is entrained into the sleep cycle of Non-REM and REM periods. A maximum of plasma hormone elevations occurs during the first quarter of sleep cycles, i.e., during Non-REM periods and less frequent rises at the end of the cycles, mainly during REM periods.

3. In contrast to growth hormone, concentrations of prolactin remain high also during later cycles occurring toward morning. This shows that high prolactin, but not high concentrations of hGH, regularly occur during sleep cycles with small amounts of slow-wave sleep.

4. Maximal prolactin concentrations during sleep are affected neither by preceding daytime physical exercise nor by selective deprivation of slow sleep stages 3 and 4. This is further evidence that slow-wave sleep stages are not necessary for the development of high plasma prolactin concentrations. However, peak values of growth hormone in the first and second cycle are significantly diminished after selective deprivation of sleep stages 3 and 4.

5. In abnormally long sleep cycles with artificial delay of the first REM period, the cyclical rhythmicity of prolactin release seems disturbed. This is further evidence for the sleep-dependent rhythmicity in the secretion of this hormone.

Key words: Sleep – Growth-Hormone and Prolactin Secretion – Sleep Cycle – Selective Deprivation of Sleep – Man.

Zusammenfassung. 1. Bei 8 männlichen und 2 weiblichen Versuchspersonen wurde die Ausschüttung von *Prolaktin* und *Wachstumshormon* (hGH) im

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Schlaf untersucht. Eine Basisnacht, eine Nacht nach körperlichem Streß und eine mit selektivem Tiefschlaf- und REM-Entzug wurden verglichen mit dem Ergebnis unterschiedlicher Prolaktin- und Wachstumshormonsekretion in den Schlafcyclen.

2. Die Prolaktinsekretion zeigt Zeitbeziehungen zu den Schlafcyclen mit häufigem Prolaktinanstieg im ersten Cyclusviertel. Dagegen sind während des letzten Cyclusviertels, d. h. vorwiegend im paradoxen Schlaf, Anstiege selten.

3. Die Prolaktinspiegel bleiben auch während späterer Schlafcyclen gegen Morgen hoch im Gegensatz zum Abfall des Wachstumshormons. Dies zeigt, daß hohe Prolaktinspiegel auch in Cyclen mit geringen oder fehlenden Tiefschlafanteilen vorkommen.

4. Weder körperlicher Streß noch Entzug von Tiefschlaf (DE Stadien 3 und 4) beeinflussen die Prolaktinmaximalwerte. Daher ist Tiefschlaf mit langsamen Wellen keine Vorbedingung für hohe Prolaktinspiegel. Dagegen sind die Wachstumshormongipfelwerte in den ersten beiden Schlafcyclen nach selektivem Tiefschlafentzug deutlich vermindert.

5. In abnorm langen Schlafcyclen mit künstlichem Aufschub der ersten REM-Phase erscheint auch die cyclische Rhythmik der Prolaktinsekretion gestört. Trotz Unabhängigkeit von Tiefschlafstadien besteht eine schlafabhängige Rhythmik der Prolaktinausschüttung.

Schlüsselwörter: Schlaf – Wachstumshormon- und Prolaktinausschüttung – Schlafcyclus – Selektiver Schlafentzug – Mensch.

Introduction

After the investigations of Nokin et al. (1972), Sassin et al. (1972), and Parker et al. (1974) it is known that prolactin exhibits a sleep-dependent secretion pattern. Human growth hormone, another anterior pituitary hormone, is secreted in temporal relationship to sleep onset (Takahashi et al., 1968; Honda et al., 1969; Pawel et al., 1972) and highest plasma levels are found during slow-wave stages 3 and 4 in the first or second cycle after onset of sleep (Parker et al., 1969; Takahashi et al., 1968; Honda et al., 1969; Beck et al., 1975). Awakening will abolish this secretory rhythm which is restored when sleep is resumed (Beck et al., 1975). The secretion of growth hormone during night sleep can also be altered by squash (Ogunremi et al., 1973) and by changing the body temperature (Beck et al., 1976).

While growth hormone secretion has been extensively studied during night sleep there are comparatively few reports on prolactin secretion. Parker et al. (1974) hypothesized an entrainment of prolactin secretion into the NREM-REM sleep cycle; thus, decrease of plasma concentrations seemed to occur at the beginning of paradoxical sleep and cyclical elevations were recorded during NREM sleep stages. These findings could reflect serotoninergic and adrenergic control of prolactin secretion during sleep, as it has been shown for growth hormone in rats (Collu et al., 1972).

In this study more data will be presented concerning the rhythmicity of prolactin secretion during NREM-REM sleep cycles. A part of these investi-

gations will deal with the influence of selective sleep deprivation on prolactin secretion. A similar study involving growth hormone secretion has been published (Sassin et al., 1969). The authors demonstrated a fall of growth hormone after selective deprivation of sleep stages 3 and 4.

The questions to be answered in this study are:

1. Is the prolactin secretion affected by qualitative changes of sleep, i.e., selective deprivation of NREM stages 3 and 4 and paradoxical sleep as it was shown for growth hormone?

2. If there is a periodical increase of prolactin secretion during NREM sleep stages and a decrease at the onset of or during paradoxical sleep, can this pattern be disturbed by altering the rhythmicity of sleep?

3. Does physical daytime exercise affect the secretion of prolactin during sleep similarly as the sleep secretion of human growth hormone?

Subjects and Methods

We have investigated eight male and two female healthy volunteers with an average age of 25 years (range 19—35 years) during four nights on randomized distribution. An adaptation night preceded the investigational period. During the baseline-night blood was taken at 20 min intervals by means of an indwelling catheter till the subject awakened in the morning (Ogunremi et al., 1973). The experimental design had foreseen a similar protocol for the following nights including standardized ergometer exercise (3 times 120 Watt × h) during daytime (in the morning, early afternoon, and at 6 a.m.) preceding the first sleep period. An attempt to delay the onset of the first REM period as long as possible was performed during another investigational night. Purpose of this experiment was to induce an abnormally long sleep cycle by simultaneously depriving the subjects of slow-wave stages 3 and 4 and paradoxical sleep. As soon as the first slow-waves appeared, painful electrical stimulation was applied to the forehead or to one foot. The same procedure was repeated as soon as signs of paradoxical sleep appeared either in the EEG, electromyogram or rapid eye movements. It was emphasized not to wake up the subjects during these deprivation nights. Monitoring was startet at 10:30 p.m. and continued till 7:30 a.m. No food was allowed after 7 p.m.

Sleep EEG records were scored according to the international fixed criteria of Rechtschaffen and Kales (1968). Measurements of plasma prolactin and growth hormone concentrations were performed by previously reported methods (Hwang et al., 1971; Molinatti et al., 1969; del Pozo et al., 1976). The mean maximal plasma concentrations of both hormones were calculated in each sleep cycle during the different experimental conditions and also compared intraindividually. T-test was used for statistical evaluation. During normal sleep and the nights after physical stress application, the mean values of prolactin were calculated 20 min before, at the onset, at the end of REM periods, and 20 min thereafter. These values were taken from graphs. In the artificially prolonged sleep cycles, REM durations and intervals were hypothesized in an approximate 90 min rhythm (Hartmann, 1968; Globus, 1970). Onset and duration of these hypothetical REM periods were calculated as individual mean values from the baseline night and normal night sleep pattern after physical exercise. Mean prolactin plasma concentrations were obtained from curves for "hypothetical" and "real" NREM-REM cycles.

Results

Polygraphic EEG Data

1. Total Duration of Sleep. The total duration of sleep during baseline nights and nights after physical stress was about 7 h. A statistical comparison between baseline nights and nights after physical stress did not show significant differ-

ences. During deprivation nights total duration of sleep was reduced to 330.3 ± 60.9 min compared to 422.5 ± 40.05 min recorded in the baseline night ($t = 3.94$, $P < 0.025$).

2. Waking Periods and Stage 1 Sleep. Baseline nights and nights after physical stress did not show significant differences when the duration of waking periods and stage 1 sleep were compared. However, in deprivation nights the amount of waking periods and stage 1 sleep were significantly increased.

3. Stage 2 Sleep. The duration of stage 2 sleep did not vary between the baseline and nights after physical stress, where a difference of only 5 min was found. However, in nights with deprivation of stages 3 and 4 and REM sleep, the

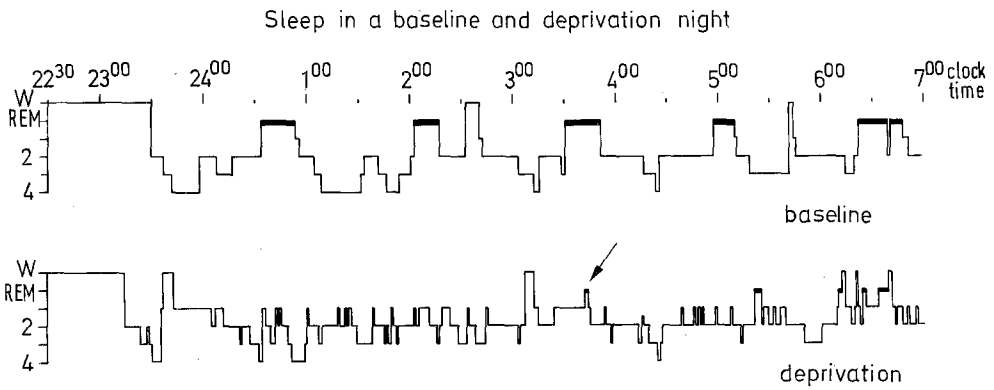

Fig. 1. Loomis scheme of normal night sleep (upper part of figure) and deprivation night with artifical delay of first REM period (REM period is marked by arrow). Frequent changes in sleep stages can be seen. Periods of slow-wave sleep are very short

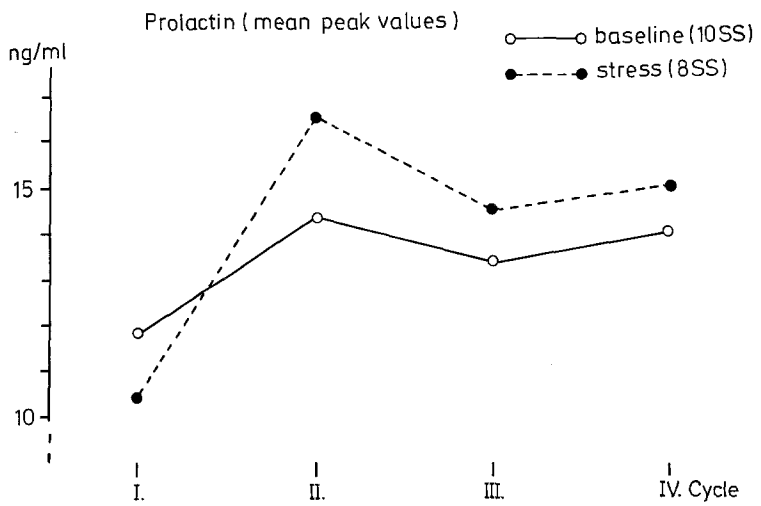

Fig. 2. Mean maximal plasma concentration of prolactin in four successive NREM-REM cycles during baseline nights and nights following physical stress. High plasma concentrations are also found during later cycles (III. and IV.)

amount of stage 2 averaged 276.12 ± 41.24 min in comparison to 199.4 ± 33.08 min recorded during baseline nights ($t = 4.4213$, $P<0.025$).

4. Slow Wave Sleep (Stages 3 and 4). There were no differences between baseline nights and nights after physical stress as it was described in the literature (Hobson et al., 1969). The selective deprivation of sleep stages 3 and 4 in this study was very effective: in these nights 16.9 min were measured in contrast with 170.20 ± 150.59 min recorded during baseline nights ($t = 4.8059$, $P<0.0005$).

5. Paradoxical Sleep. Paradoxical sleep did not differ between baseline nights and nights after physical stress. In deprivation nights paradoxical sleep was diminished to 25.5 ± 12.57 min in comparison to 104.6 ± 25.7 min in baseline condition ($t = 4.179$, $P<0.0005$).

6. Duration of Prolonged Sleep Cycles (see Fig. 1). In deprivation nights the average duration of the prolonged "first cycle" was 233 ± 92.64 min, while in baseline nights the duration of the first cycle was 114.1 ± 81.86 min. The difference was statistically significant ($t = 3.18$, $P<0.01$).

Endocrinological Findings

Prolactin Secretion

1. Maximal Plasma Concentrations. Figure 2 shows the maximal plasma concentration of prolactin in 4 successive NREM-REM cycles during the baseline nights and the nights following physical stress. It can be seen that the highest concentrations are found in the second sleep cycle. During later cycles (3 and 4) there is no obvious decrease in maximal plasma concentrations as observed during night sleep with growth hormone investigations.

A statistical comparison between maximal plasma hormone concentrations during baseline nights and during nights following physical stress does not show significant differences.

2. Temporal Distribution of Rises in Prolactin Plasma Concentration within the NREM-REM Cycles (see Fig. 3). An analysis of 72 cycles of normal sleep from 10 subjects in 18 nights showed most frequent rises of prolactin plasma concentrations during the first quarter of the cycle and the lowest number of rises during the last quarter. This shows that rises occur mainly during NREM sleep stages and very seldom during paradoxical sleep which occurs in the last quarter of the cycle. Analysis shows that stages 3 and 4 are mainly encountered in the second quarter of the sleep cycle.

3. Concentrations of Prolactin in Temporal Relationship to Paradoxical Sleep (Baseline Nights and Nights Following Physical Stress) (see Fig. 4). The investigations of prolactin plasma concentrations were performed 20 min before the start of the REM period, immediately at the beginning of paradoxical sleep, at the end of the REM period, and 20 min after the end of the REM period. Significant differences in plasma prolactin concentrations before, during, and at the end of paradoxical sleep were not found. However, there was a rather constant decrease in prolactin plasma levels at the beginning of paradoxical sleep

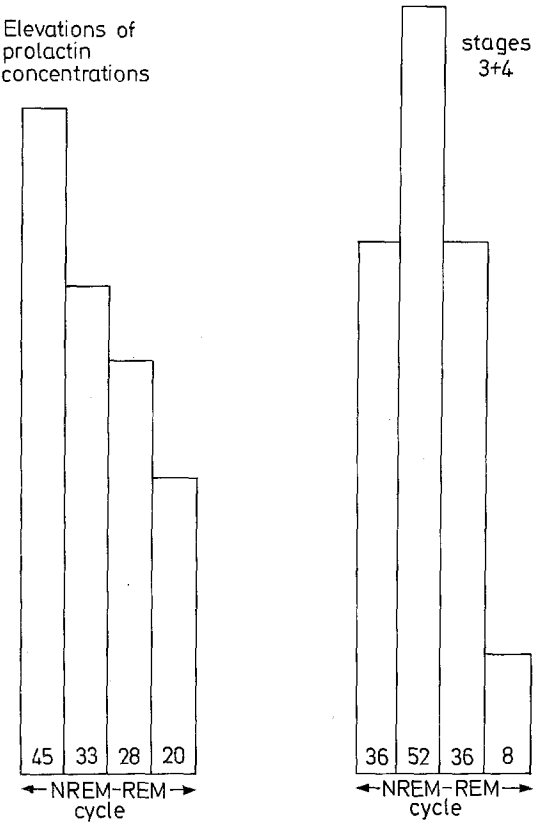

Fig. 3. Analysis of elevations of prolactin plasma concentrations in 72 sleep cycles of healthy subjects. Columns on left symbolize frequency of plasma hormone rises in comparison to preceding quarters of cycles. Columns on right demonstrate distribution of slow-wave sleep stages 3 and 4 across cycles. It can be seen that they mainly occur in second part of the cycle, while prolactin rises are mainly encountered in first quarter of NREM-REM sleep cycle and much less frequently at end of cycles

as compared to NREM sleep stages. More significant is the increase in prolactin concentrations during NREM sleep that followed paradoxical phases. Only after the fourth sleep cycle does this increase disappear.

Deprivation Experiments and the Secretion of Prolactin and hGH

1. The Secretion of Prolactin after Selective Deprivation of Slow Wave and Paradoxical Sleep. Maximal concentrations of prolactin following selective deprivation of stages 3 and 4 and paradoxical sleep did not show significant differences when compared with baseline levels. However, only the first two NREM cycles can be compared because of the small number of later cycles.

The normal rhythmicity of prolactin secretion exhibited by decrease in plasma concentrations before and during paradoxical sleep and elevation after the end of

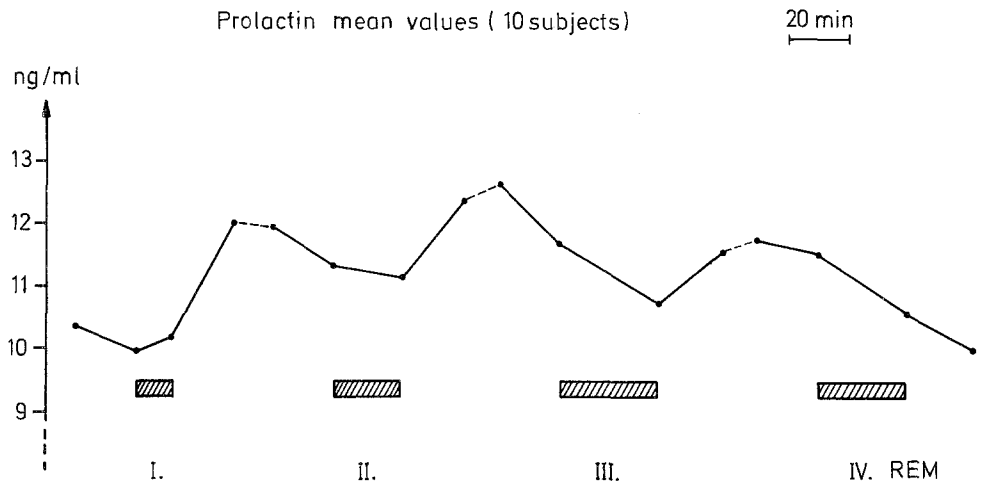

Fig. 4. Plasma prolactin concentrations in temporal relation to paradoxical phases of sleep in four successive sleep cycles of 10 subjects. An analysis of plasma hormone concentrations was performed 20 min before, at onset, at end of REM period, and 20 min after end of REM period. It can be seen that plasma hormone concentrations decrease before onset of paradoxical sleep and elevations are found after end of REM period, with single exception of fifth cycle

REM periods is disturbed in the deprivation nights. This disturbance is shown in Figure 6.

2. hGH Maximal Plasma Concentrations after Selective Deprivation of Slow Wave Sleep. Figure 6 shows that after selective deprivation of sleep, stages 3 and 4 maximal plasma concentrations are significantly diminished during the first two cycles. During baseline nights the mean value of highest plasma concentrations was 21.8 ± 12.8 ng/ml. This difference is not statistically significant. In the second cycle, however, the difference was 16.8 ± 12.6 ng/ml during baseline conditions compared to 3.59 ± 2.64 ng/ml plasma after selective deprivation of the sleep stages 3 and 4 ($t=4.3$, $P<0.005$).

Discussion

Like the findings of Parker et al. (1974), our observations show that the secretion of prolactin is entrained into the NREM-REM sleep cycle. Most frequently elevations of plasma hormone concentrations occur during the first quarter of sleep cycles and rarely at the end of the NREM-REM cycle. This means that there is a temporal relationship between increases in hormonal secretion and NREM sleep and nadirs and paradoxical sleep. A similar relationship is shown in the sleep-linked hGH release. Serotoninergic, dopaminergic, or noradrenergic mechanisms may be involved in the control of these two anterior pituitary hormones during sleep. A striking difference between hGH and prolactin sleep levels is shown in the "spiking" of prolactin concentration found even in the late cycles, which

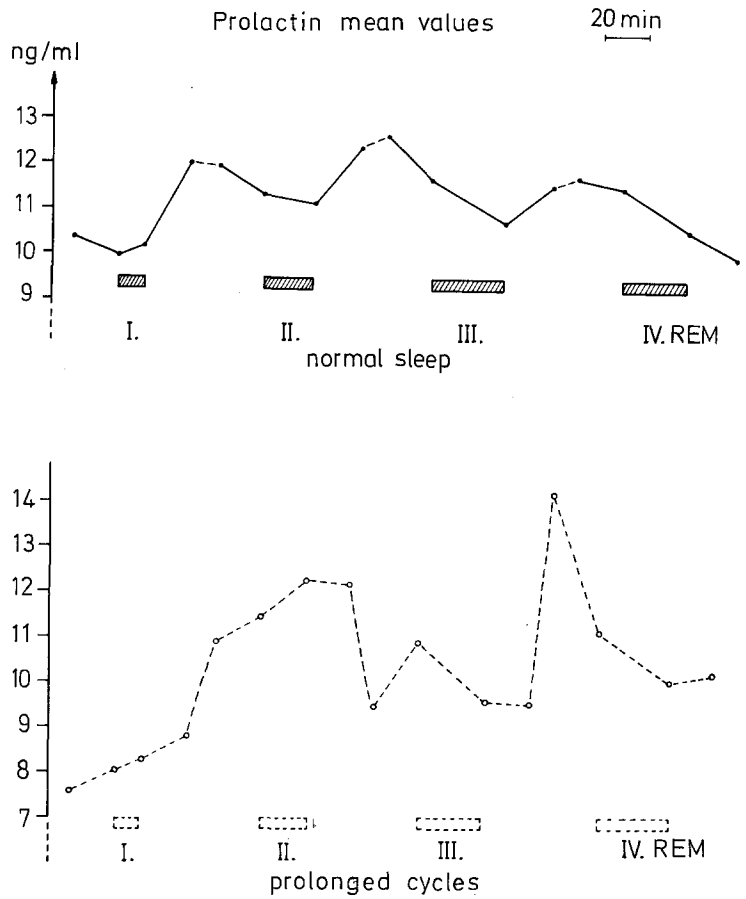

Fig. 5. Rhythmicity of prolactin secretion during normal sleep (upper part) and deprivation nights with artificially prolonged first sleep cycles (lower part). Mean intraindividual plasma concentrations were calculated from curves 20 min before onset of real or hypothetical REM, at their onset, end, and 20 min after their end. Usual decrease of prolactin plasma levels before onset of paradoxical sleep is not seen in first three epochs of hypothetical cycles in deprivation nights

is rarely seen in hGH in normal night sleep. Possibly there is no antagonism between prolactin and circadian release of ACTH which may account for hGH inhibition during sleep in the early morning hours. Another explanation could be an independence from slow-wave stages 3 and 4 which occur mainly in temporal relation to sleep onset at night. Another difference is the lack of increased release of prolactin after daytime physical exercise (described for growth hormone by Adamson et al., 1974), and a significantly elevated hGH peak after the onset of sleep. Although prolactin is also a stress hormone, its long term alterations by stress seem to be less pronounced than those of growth hormone.

Our findings of a diminished secretion of hGH in nights after selective deprivation of sleep stages 3 and 4 are consistent with the findings of Sassin et al. (1973) who found no significant growth hormone plasma elevations or very

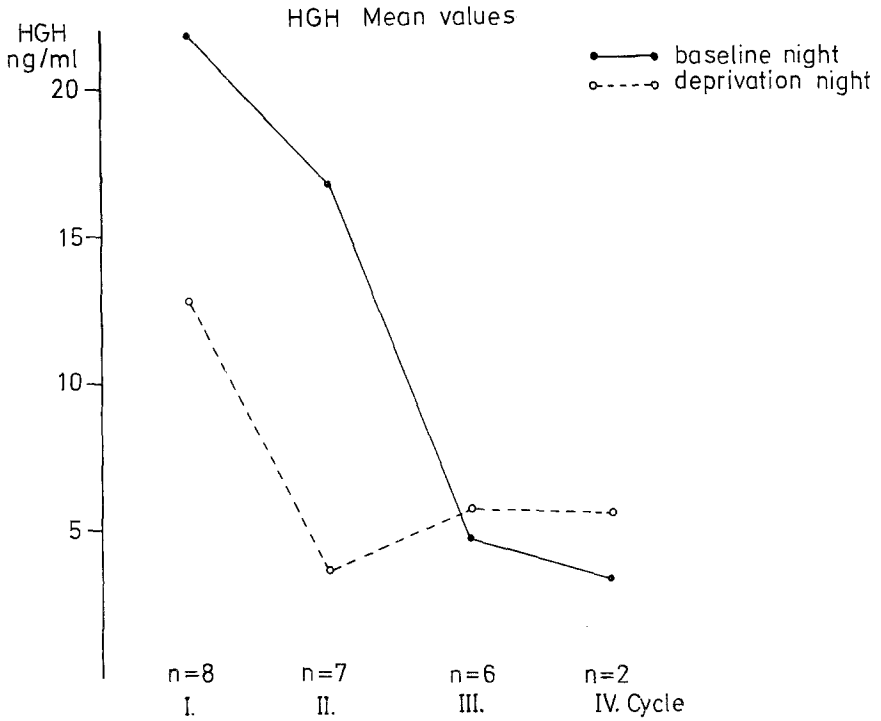

Fig. 6. hGH maximal plasma concentrations during selective deprivation of sleep stages 3 and 4. It can be seen that maximal plasma concentrations during first and second cycle are significantly diminished

delayed hGH peaks in deprivation nights. The lack of influence on prolactin secretion during deprivation nights is a further difference from growth hormone which is influenced considerably by the quality of sleep. The decrease in prolactin plasma concentrations before or during periods of paradoxical sleep is probably based on dopaminergic mechanisms and the appearance of paradoxical sleep. Our observations during periodicity disturbed sleep in which the cyclical increase and decrease in prolactin secretion was not found, demonstrate this, and provide further evidence that the fluctuations in plasma prolactin concentration are not due to a sleep-independent, 90-min rhythm.

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