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CIRCADIAN, ENDOCRINE, AND METABOLIC EFFECTS OF PROLONGED BEDREST: TWO 56-DAY BEDREST STUDIES

by Joan Vernikos-Danellis, Charles M. Winget, Carolyn S. Leach, and Paul C. Rambaut

Ames Research Center
Moffett Field, Calif. 94035

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16. Abstract

Two bedrest studies of 56 days each, involving a total of 20 male subjects aged 20-26 (one subject aged 40), have been conducted to evaluate the effects of prolonged bedrest on circadian synchrony and endocrine and metabolic function. Measurements included the pituitary-adrenal, thyroid, parathyroid, insulin-glucose-growth hormones, catecholamine excretion, body temperature, and heart rate. The results indicate that a rigorous regimen of isotonic/isometric exercise did not prevent the endocrine and metabolic effects of prolonged bedrest. Changes in circadian, endocrine, and metabolic functions in bedrest appear to be due to changes in hydrostatic pressure and lack of postural cues rather than to inactivity, confinement, or the bleeding schedule. Changes in circulating metabolic and endocrine parameters are unreliable if measured once per day because their amplitude and time of peak of their diurnal fluctuations are altered during bedrest. Therefore, data should be expressed as units/24 hours. Recovery periods up to 20 days are insufficient for full recovery from 56 days of bedrest. Bedrest beyond 42 days results in periodic hypoglycemia, possibly in response to meals, which may warrant modification of meal composition.

Prolonged bedrest, particularly beyond 24 days, results in rhythm desynchronization in spite of well-regulated light/dark cycles, temperature, humidity, activity, and meal times and meal composition and in increased lability of all endocrine parameters measured.

It also results in an apparent insensitivity of the glucose response to insulin, of cortisol secretion to ACTH, and of growth hormone secretion to hypoglycemia. This may be due to an effect of bedrest on the number or sensitivity of target organ receptors; it may reflect a change in radioimmunoassayable levels of the peptide hormones, or it may result from an alteration of the central nervous system's input/feedback integrating mechanisms.

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TWO 56-DAY BEDREST STUDIES

Joan Vernikos-Danellis and Charles M. Winget
Ames Research Center

and

Carolyn S. Leach and Paul C. Rambaut
Johnson Spacecraft Center

SUMMARY

Two bedrest studies of 56 days each, involving a total of 20 male subjects aged 20-26 (one subject aged 40), have been conducted to evaluate the effects of prolonged bedrest on circadian synchrony and endocrine and metabolic function. In addition, the contribution to the observed results of factors inevitably associated with bedrest (i.e., lack of activity, confinement, and the blood sampling schedule) was assessed. Around the clock blood sampling was designed (a) to determine the effects of bedrest on circadian synchrony as a measure of central nervous system function and (b) to determine if changes in hormone levels are sustained or intermittent. Measurements included the pituitary-adrenal, thyroid, parathyroid, insulin-glucose-growth hormones, catecholamine excretion, body temperature, and heart rate.

Bedrest resulted in rhythm asynchrony in spite of a well-regulated L:D environment. The most drastic rephasing of heart-rate rhythms occurred suddenly on day 23 or 24 in all 14 bedrested subjects but not in ambulatory controls. Mean daily body temperature decreased about 1°C in all subjects by the end of 56 days of bedrest.

Hormone and glucose level changes during bedrest were not sustained throughout the day, but reflected changes in the amplitude of the fluctuations.

Glucose homeostasis was maintained for the first 30 days of bedrest accompanied by a 2.5-fold increase in circulating insulin levels. Beyond that, insulin levels fell as did glucose (by day 54, the glucose level at 4:00 p.m. was 62.2 mg/100 ml). The pituitary did not respond to hypoglycemia by a rise in growth hormone secretion.

Plasma cortisol concentration doubled during the first 20 days of bedrest but decreased subsequently to levels below controls by day 54. Plasma ACTH increased after 30 days. The diurnal rhythmicity of thyroid hormones was abolished by bedrest but levels were unchanged.

Parathormone levels showed an overall increase and greater fluctuations during bedrest. Sporadic urinary catecholamine measurements suggested increased excretion during bedrest.

1. INTRODUCTION

Weightlessness is one of the most important factors of space flight to which space crews are being exposed continuously. Since prolonged weightlessness cannot be reproduced on the ground, the pertinent literature discusses several experimental approaches that allow assessment of biological effects of prolonged weightlessness.

The hypokinetic state combined with the recumbent position of test subjects is the most appropriate ground-based method with which to study the effects of prolonged weightlessness. In the realm of prolonged bedrest, two 56-day studies designed to investigate prolonged weightlessness ulitizing the absolute bedrest technique have been conducted jointly by the Johnson Space Center and Ames Research Center. Both studies were conducted at the Texas Woman's University in Denton, Texas, under the leadership of Dr. Pauline Beery Mack. The two studies referred to as 56-Day Bedrest Study I and 56-Day Bedrest Study II involved 20 subjects and were carried out in the summer of 1969 and the spring of 1970, respectively.

The main outcome of these studies was an extensive evaluation of the endocrine and metabolic changes in man associated with prolonged bedrest and an investigation into the various factors associated with bedrest that may be bringing them about. For instance, by maintaining a strict control over the environmental conditions, it was possible to draw conclusions regarding the relative contribution to the results of the lack of activity or exercise, posture, relative confinement or the experimental sampling schedule.

Data were collected continuously throughout both studies. As a result, a large volume of information relating to many physiological systems has emerged, which has been reported in fragmented and abbreviated versions on a variety of occasions, thus causing confusion regarding the experimental origin of the information. This report therefore attempts to consolidate all that information and to summarize the most pertinent conclusions that can be drawn from these two prolonged bedrest studies. Since the primary purpose of these studies was to evaluate flight foods, the data relating to that and to the nutritional status of the subjects are not included here but can be found in the final report of contract NAS9-9755.

2. METHODS AND PROCEDURES

The experimental design of the two studies is schematically represented in figure 1. Study I included a 10-day prebedrest period during which all 8 subjects were ambulatory but confined to the ward. This period was followed by 56 days of absolute bedrest during which 4 subjects (group 1A) did not exercise while the other 4 subjects (group 1B) conformed to a rigorous 1-hr/day, isometric and isotonic exercise regimen. After 56 days of bedrest, there was a 10-day postbedrest recovery period.

Study II began with a 20-day prebedrest period during which all 12 subjects were ambulatory but confined to the ward. Following this period, 6 subjects (group IIA) were subjected to absolute bedrest for 56 days without exercise while the other 6 remained ambulatory but confined to the

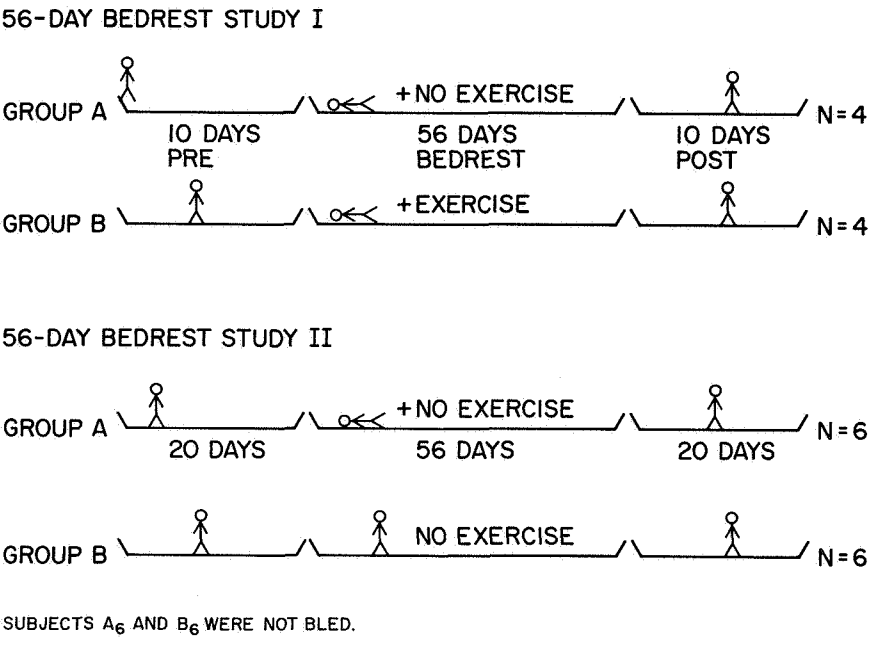


Figure 1.– Experimental design for 56-day bedrest studies I and II.

ward. After 56 days, group IIA began a 20-day postbedrest ambulatory recovery period and group IIB remained ambulatory for the next 20 days. Thus, group IIB served as ambulatory controls for the entire 96 days of the study – remaining confined to the metabolic ward in the same controlled environment and following the same daily schedule as all group IIA subjects. Blood and urine samples were collected and, body temperature and heart rate were measured throughout both studies. In study II, one subject from each group was not bled but was monitored only for body temperature and heart rate. This procedure was followed because there had been concern that some of the findings in study I could have been attributed to the bleeding schedule rather than to the bedrest per se. Group IIB were kept active and motivated by working in the laboratories.

Experimental Subjects

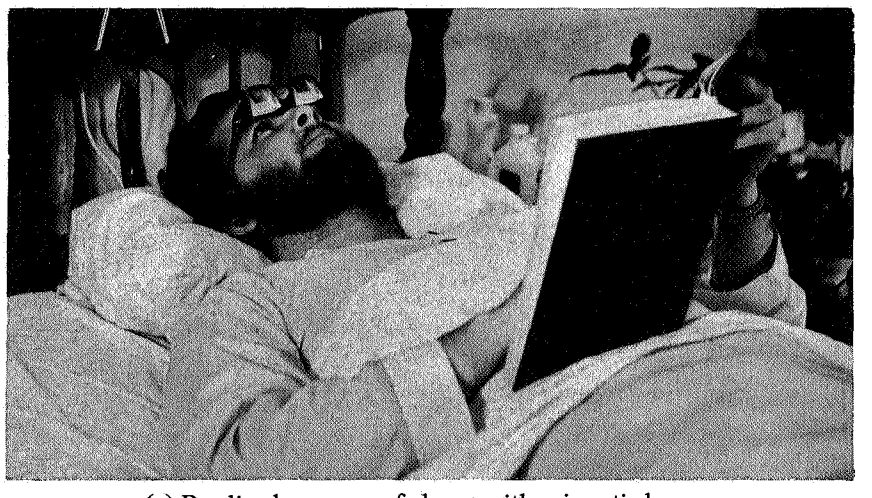
Young healthy males, 20-26 years old, weighing 61 to 89 kg and 167 to 185 cm tall, were selected from more than 60 applicants. In study I, one 40-year-old subject was also included. These men were screened psychologically and given physical examinations, which included electrocardiograms, chest x-rays, and clinical tests, both blood and urinary analyses.

Experimental Conditions

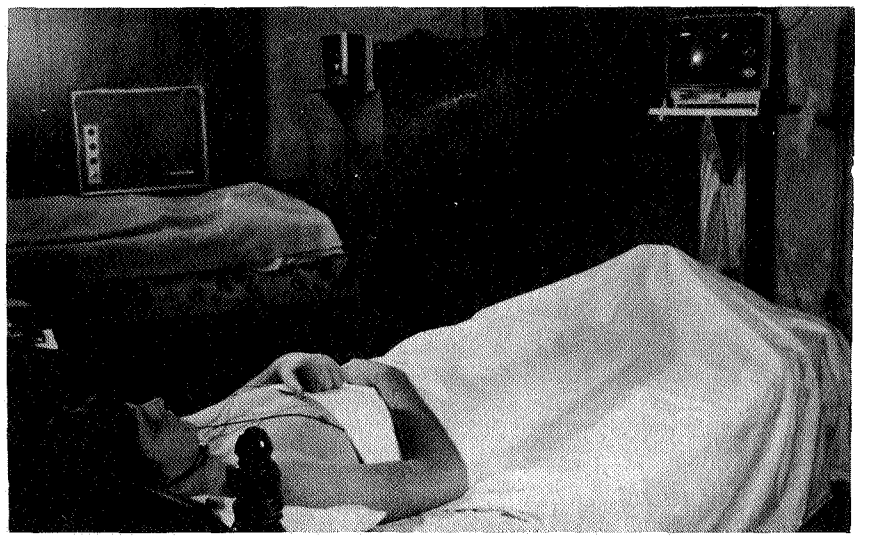
Two men shared a room in each study. Table 1 shows the daily schedules for the two studies. Bedrest was maintained as rigidly as possible. The subjects lay flat on the bed using only a thin head pillow. They were spoon fed by orderlies. They were provided with glasses with prismatic lenses so that they could read without raising their heads and televisions were also raised for individual viewing while lying flat (fig. 2).

TABLE 1.– DAILY SCHEDULE OF 56-DAY BEDREST STUDIES I AND II

Study I	Study II
0900 – Lights on, breakfast	0900 – Lights on, breakfast
1130 – Exercise (20 min)	
1300 – Lunch	1300 – Lunch
1530 – Exercise (minute)	
1730 – Dinner	1730 – Dinner
1930 – Exercise (20 min)	
2200 – Snack	2200 – Snack
2300 – Lights off	2300 – Lights off



(a) Reading by means of glasses with prismatic lenses.



(b) Watching individual TV with ear phones.

Figure 2.– Methods of passing the time.

All known exogenous influences on human circadian rhythms were maintained in an environment of 14L:10D (lights on at 0900 hr). Light intensity was 30 ft-c or greater at eye level, and the rooms were draped to minimize light leakage. Television was permitted only during the lights-on phase. Ambient temperature was maintained at 20° ±1°C.

Food consisted of a balanced diet of 2500 kcal/day. Subjects were hand fed by dietitians and orderlies. There was no significant ($P > 0.05$) loss or gain of body weight by the end of the study (see table 2).

TABLE 2.– 56-DAY BEDREST STUDY I: BODY WEIGHTS OF BEDREST SUBJECTS

	Subject	Prebedrest, kg	Postbedrest, kg
Group IA No exercise	AG	70.45	69.55
	CR	70.45	71.36
	GA	89.09	89.09
	BR	60.90	62.27
Group IB Exercise	SM	65.00	66.36
	BI	66.36	66.36
	KE	80.91	80.91
	JO	79.45	79.09

Exercise Program

The Exer-Genie, which had been used with modifications during the mission of Apollo 7, was further modified to include these improved hand straps and foot stirrups and padded foot coverings. Figure 3 shows the type of exercises that were performed in study I. The entire exercise program

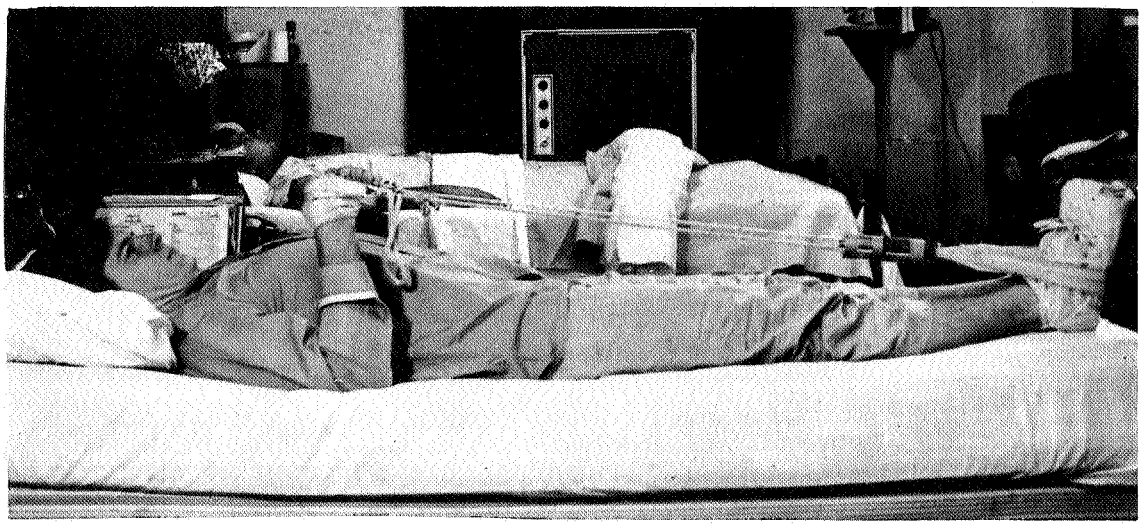


Figure 3.– Exercise regimen in study I.

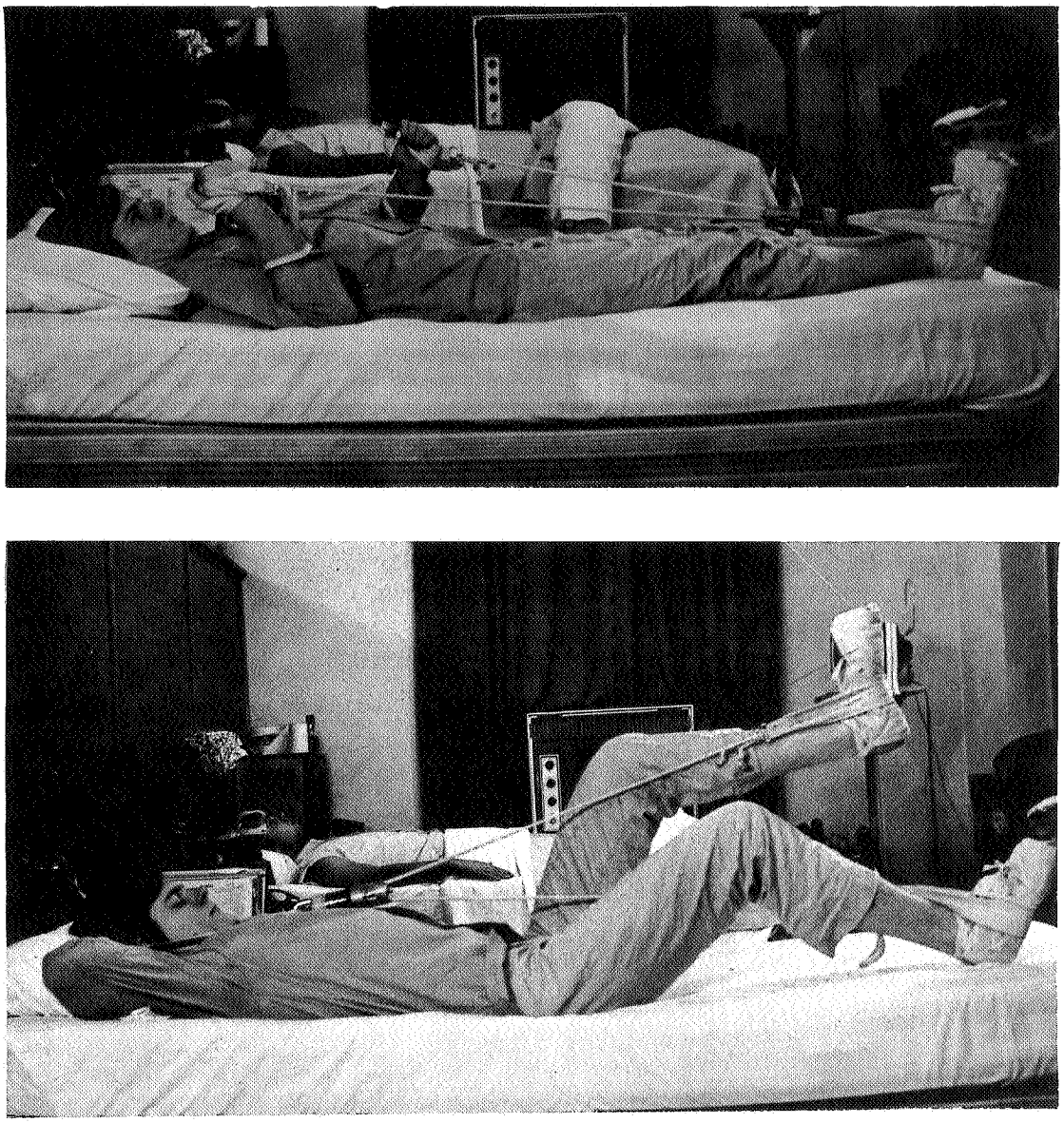


Figure 3.– Exercise regimen in study I – Concluded.

was performed daily during bedrest at 11:30 a.m., 3:30 p.m., and 7:30 p.m. The exercise routine is outlined in table 3.

Each exercise schedule covered 30 sec of isometric and 20 min of isotonic exercise. The three daily programs therefore involved a total of 150 min of programmed exercise. Records were kept on each subject as to the time expended during each step of the schedule.

During the 48-hr periods when blood was taken every 4 hours, the exercise was omitted.

TABLE 3.– EXERCISE SCHEDULE

(Cylinder on Exerciser set at 3.64 kg and metronome speed of 1 beat/sec)

Step	Activity		Time
1	Isometric (EXER–GENIE)		10 sec
2	Leg exercise (EXER–GENIE)		6 min
3	Rest		2 min
4	Hand - fingers (Gripper)		1 min
5	Rest		2 min
6	Isometric (EXER–GENIE)		10 sec
7	Arm exercise (EXER–GENIE)		6 min
8	Rest		2 min
9	Hand - fingers (Gripper)		1 min
10	Rest		2 min
11	Isometric (EXER–GENIE)		10 sec
12	Leg exercise (EXER–GENIE)		6 min
	Total	Isometric	30 sec
		Isotonic	20 min

Blood Samples

The subjects were bled by repetitive venous punctures every 4 hours for a 48-hr period at each of 9 points throughout the study; these included two 48-hr periods before bedrest, 10, 20, 30, 42, and 54 days after confinement to bed and 13 and 20 days after the subjects had again become ambulatory. Fifteen milliliters of blood was removed at each bleeding to obtain approximately 5 ml of plasma and 2.5 ml of serum. The blood samples were kept cold in crushed ice during collection and separation, then frozen promptly and stored.

Once each 48-hr bleeding period (8:00 a.m. of the second day), hemoglobin, hematocrit, RBC, and WBC, were determined in all subjects. No appreciable changes were observed throughout the study.

In study II, only 5 of the 6 subjects in each group were bled. The sixth acted as a control to determine the effects of blood sampling on other parameters. No such effects were noted.

Urine Samples

Urine collected as voided was pooled into 6-hr pools (study I) or 4-hr pools (study II). Aliquots from each pool were frozen and stored.

Physiological Measurements

The physiological measurements in the two studies are summarized in table 4. Deep-body temperature and heart rate were measured at 4-hr intervals in study I and at 2-hr intervals in

TABLE 4.– PHYSIOLOGICAL MEASUREMENTS

Study I	Study II
Body temperature (6 time/24 hr)	Body temperature (12 time/24 hr)
Heart rate (6 time/24 hr)	Heart rate (12 times/24 hr)
Plasma cortisol	Plasma cortisol
Plasma thyroxine	Plasma thyroxine
Plasma triiodothyronine	Plasma triiodothyronine
	Plasma parathormone
	Plasma insulin
	Plasma glucose
	Plasma growth hormone
	Plasma ACTH
	Urinary cortisol

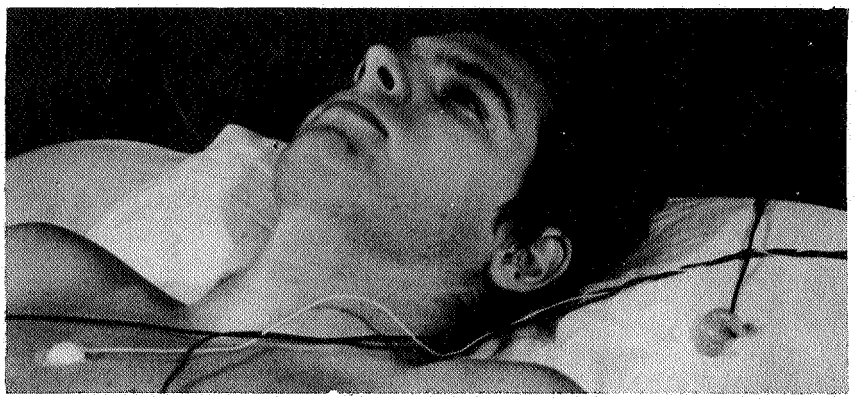
study II, on a 24 hr/day throughout each study. Body temperature data were obtained using ear probes containing a thermistor (Yellow Springs Model 402) (fig. 4). Heart rate was measured manually from the pulse rate and by Beckman EKG sensors connected to a cardiotachometer (fig. 5).

Plasma-free hydrocortisone and urinary hydrocortisone levels were determined by Murphy's competitive protein-binding radioassay (ref. 1) (expressed as μg/100 ml plasma or μg/24 hr). Serum total thyroxine was measured using Murphy's protein-binding assay (expressed as μg/ml plasma) (ref. 2). Serum triiodothyronine was estimated by determining the binding capacity of serum to the hormone (expressed as relative percent uptake) (ref. 3). Immunoreactive parathormone in the plasma was assayed by the method of Potts (refs. 4 and 5) (expressed as pg/ml). Immunoreactive insulin was measured by the method of Herbert *et al.* (ref. 6) (expressed as μg/ml of plasma). Immunoreactive growth hormone was also measured by radioimmunoassay (ref. 7) (expressed as mμg/ml plasma). Immunoreactive ACTH was measured by the method of Donald (ref. 8) (expressed as pg/ml plasma). Plasma glucose was estimated by the automated ferricyanide method (expressed as mg/100 ml plasma). Urinary catecholamines were determined by the method of Von Euler (ref. 9) (expressed as μg/24 hr).

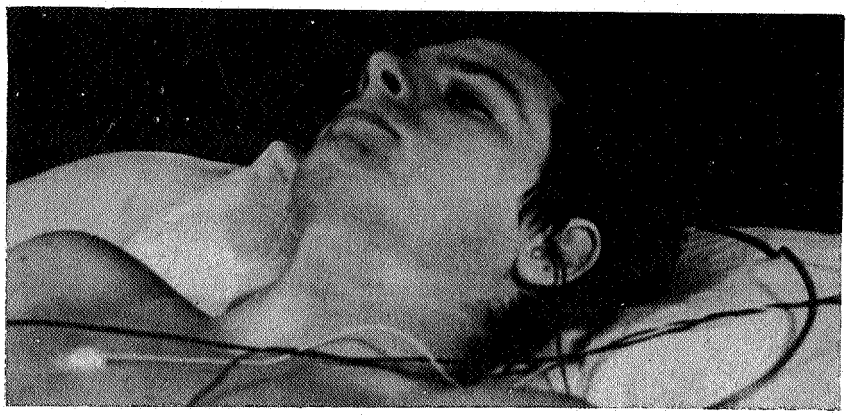
Data Analysis

The data were analyzed by standard statistical techniques (ref. 10). In addition, the results were expressed as integrated daily means representing the mean 24-hr concentration of any parameter taking into account the 6 four-hour samples obtained per day.

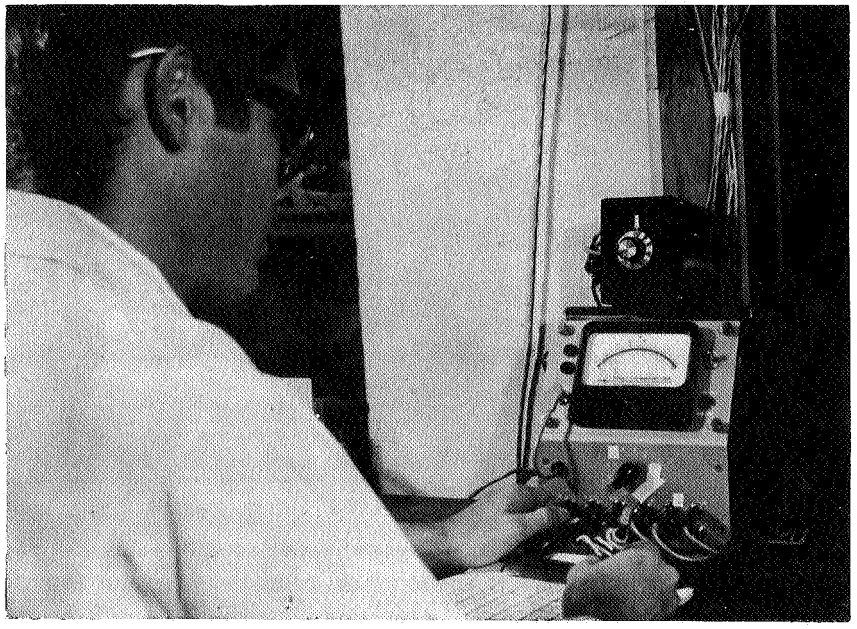
It was necessary to develop a method that expresses, in chart form, rhythmic data that may be nonstationary in time. The summation-dial method (ref. 11), developed specifically to meet these



(a) Ear probe on pillow.

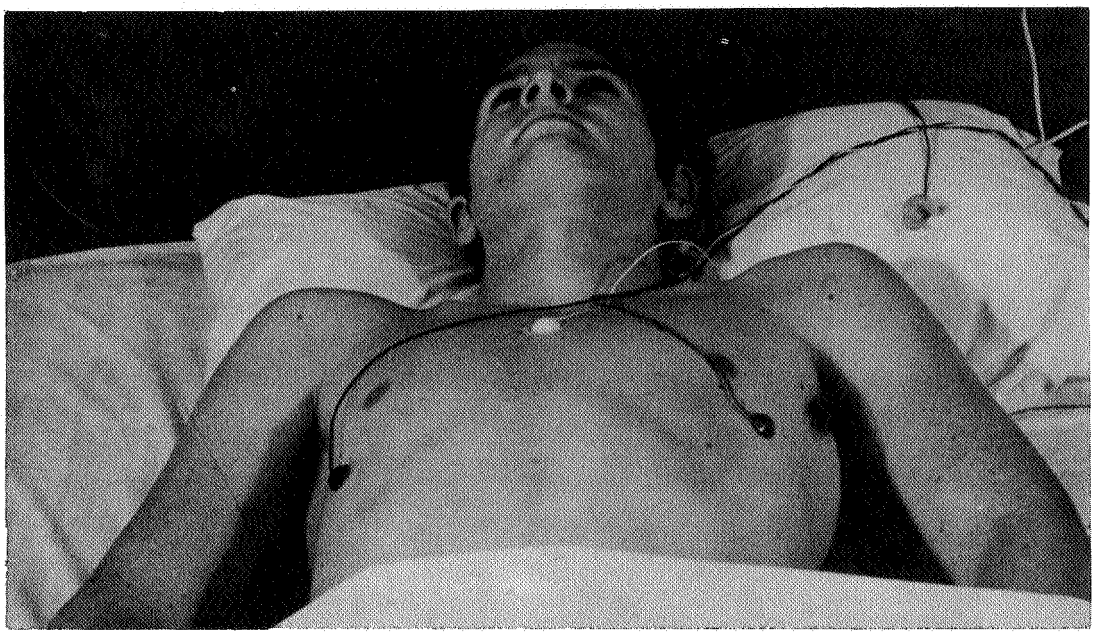


(b) Ear probe in place.

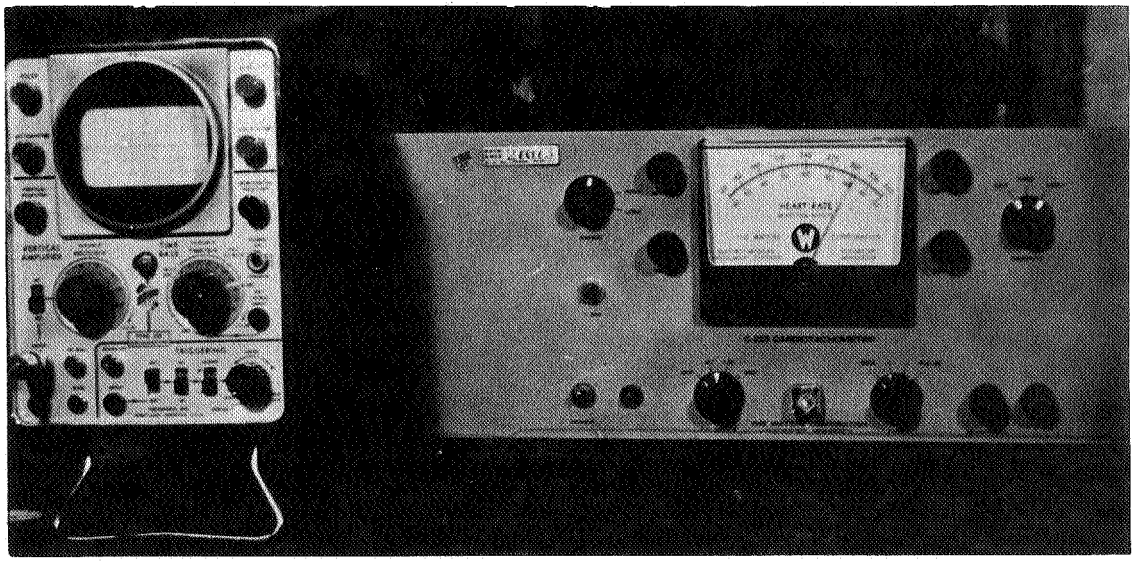


(c) Tele-thermometer with selector mounted above.

Figure 4.– Obtaining deep-body temperature readings.



(a) Subject in bed with electrocardiographic sensors connected with wires.



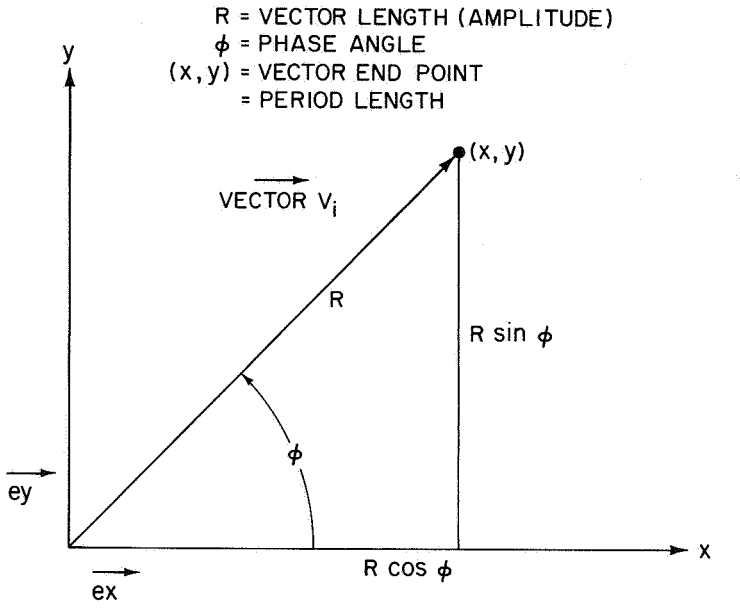
(b) Equipment on a table in the clinic with an oscilloscope (left) and a cardiotachometer (right).

Figure 5.– Obtaining heart-rate readings.

requirements, is one in which the curve that best fits the data is derived mathematically assuming a specified period (e.g., $\tau = 24$ hr). Each point on the curve represents the end of a vector, which has a certain magnitude and direction that describes the phase of the rhythm for that day (fig. 6). The summation of these vectors or train of vectors produces the summation dial (fig. 7). The direction of the vector train, when plotted on a 24-hr clock, indicates the hour of day at which the estimated peak activity of that parameter occurred. The length of the vector indicates the diurnal amplitude of the rhythm (fig. 7).

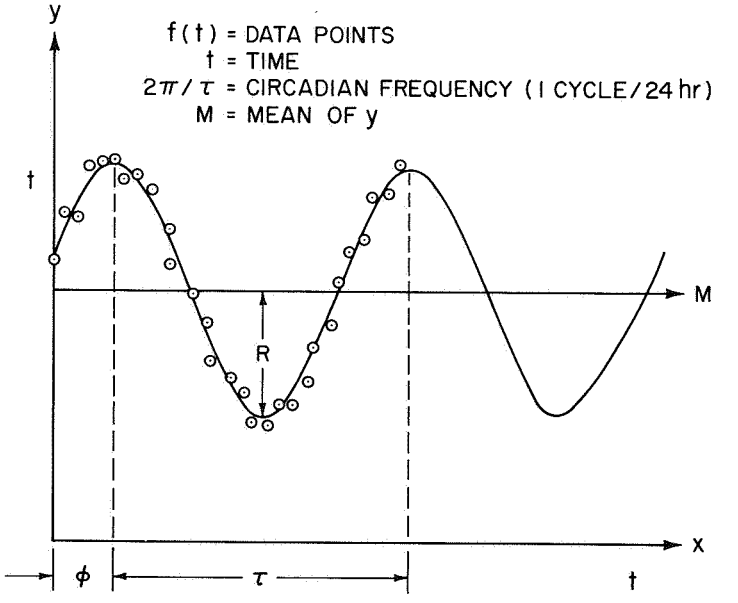
$$R = \sqrt{a^2 + b^2} = L$$

$$\phi \equiv \tan^{-1}\left(\frac{b}{a}\right) \text{ DIRECTION OF THE VECTOR}$$



(a) Graphic representation of the vector components.

$$f(t) = R\cos\left(\frac{2\pi}{\tau}t - \phi\right) + M$$



(b) Least-squares method applied to fit equally spaced discrete values.

Figure 6.– Summation-dial method, a vector representation of two components x and y.

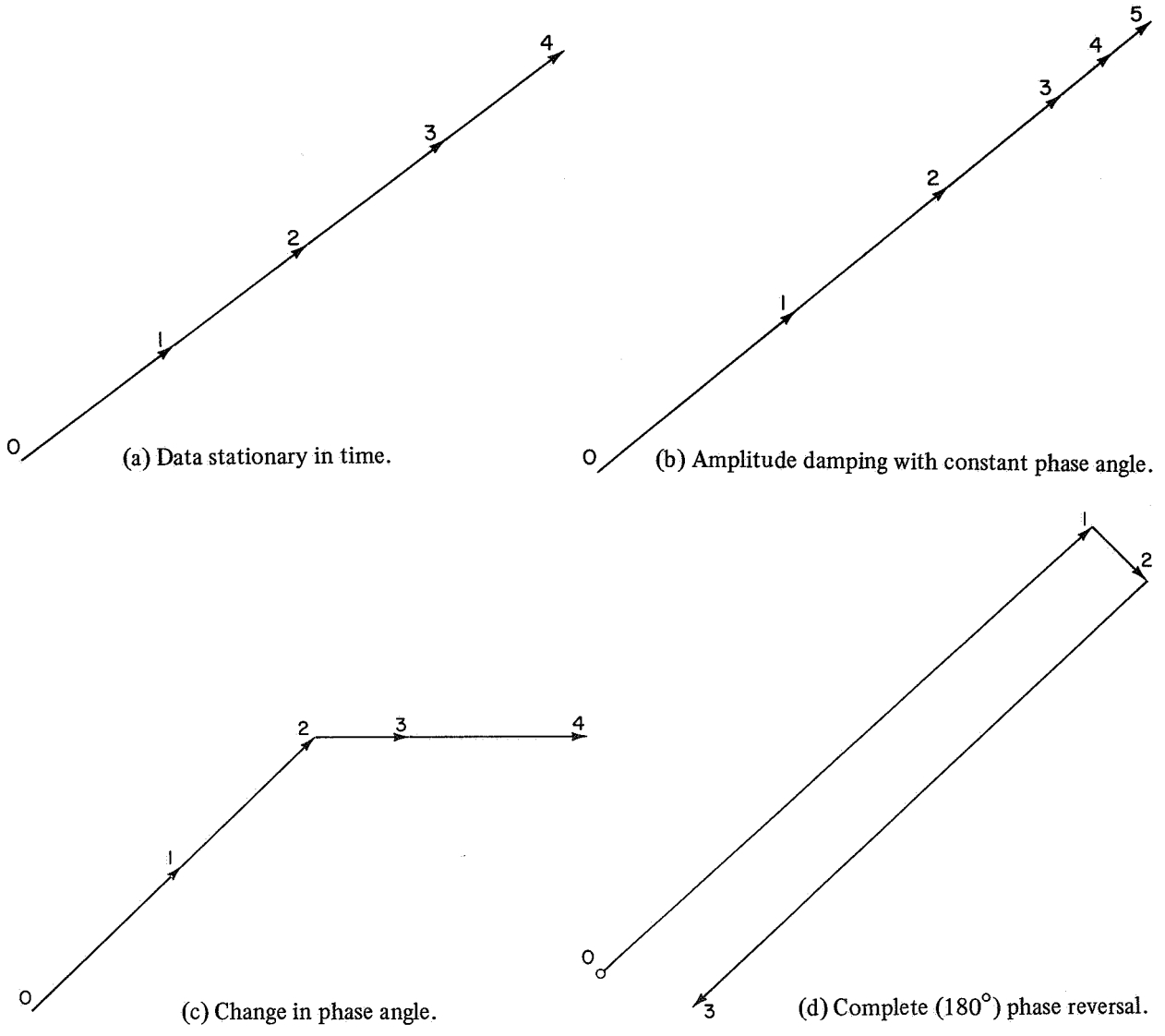


Figure 7.– Schematic illustrations of certain features encountered in the construction of summation dials.

3. EFFECTS OF BEDREST ON CIRCADIAN SYNCHRONY, BODY TEMPERATURE, AND HEART RATE

The two basic factors that act on the normal subject during bedrest are restriction of muscular activity (hypokinesis) and a characteristic redistribution of blood because of a change in hydrostatic pressure (ref. 12). Bedrest for varying periods with healthy individuals has been found to lead to venous thrombosis, hypostatic pneumonia, a reduction in heart volume, and increases in resting and work pulses (ref. 13). Other studies have revealed reductions in circulating blood volume, muscle atrophy, and negative nitrogen and mineral balances (refs. 14-16). Most of this work reported that

hypokinesis had a significant role in the etiology of the reported disorders. The characteristic time structure of these biological processes and their importance in regulatory mechanisms were not considered.

If the central nervous system plays any role in the development of the bedrest symptoms, then it might be expected that circadian synchrony, which is also regulated by central mechanisms (endogenous synchronizers), would be disturbed, and that it would be disturbed in spite of maintaining subjects in an environment with well-defined exogenous synchronizers (light, meals, etc.).

These two bedrest studies evaluated the influence on healthy human subjects of removing such factors as posture and activity on circadian waveform in a well-regulated environment with a fixed photoperiod.

Results

Study I indicated that all subjects showed a stable phase and amplitude during the 6 days before bedrest, with the maximum occurring in the latter half of the light period. Despite this homogeneity of data, continuation of the regular photoperiod of 14L:10D, and feeding at regular hours, there was a tendency, when the subjects were put to bed, for the ear canal temperature (BT) rhythm to become desynchronized with the environment although it remained circadian. An example of this desynchronization is the sequential information for subject 6A (fig. 8).

Bedrest also produced a depression in the mean body temperature (BT) that did not return to the prebedrest values (fig. 9). A change in the amplitude and time of peak of the heart rate (HR)

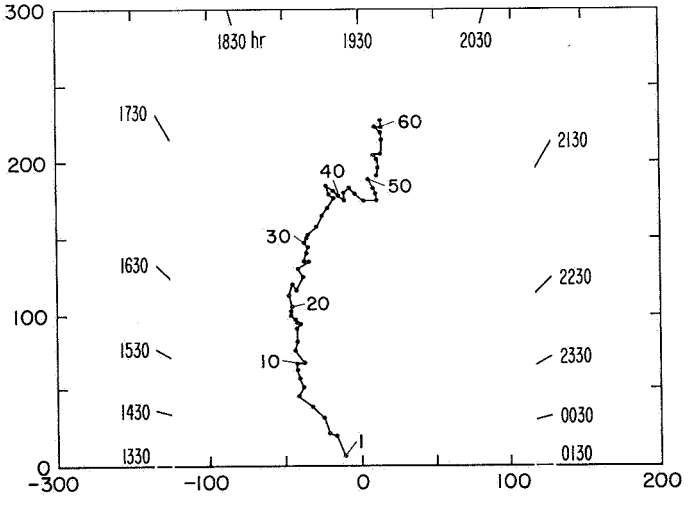


Figure 8.– Summation dial showing successive addition of body temperature daily vectors in order of day for *subject 6A.*

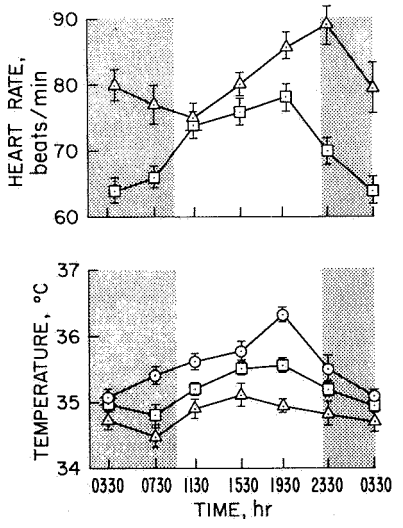


Figure 9.– Mean heart rate and body temperature values (± standard error) of eight subjects for experimental periods of 6 days before (⊙———⊙), 56 days during (⊡———⊡), and 10 days after (Δ———Δ) bed rest. Lights were turned on at 0900 hr and off at 2300 hr.

rhythm were also noted during the period after bedrest. The daily range of BT was about 1°C and the HR was about 15 beats/min. The nonexercised group had less stable circadian oscillations than the exercised group. Study II confirmed and expanded these findings.

Figures 10 and 11 demonstrate the usefulness of the summation dial method to depict concisely a large volume of sequential rhythmic data. Figure 10 shows the actual heart rate (HR) and figure 11, the body temperature (BT) data obtained from one representative ambulatory subject over the entire experimental period. The same data are also presented as summation dials. The main characteristic of the data obtained from the ambulatory subjects was their remarkable consistency. This was particularly true for HR, which showed a peak at about 1600 hours in all six subjects and did not deviate throughout the study. The straightness of the summation dial plot pointing toward 1600 hours demonstrated that feature. The BT of this subject was somewhat less stable – the peak occurred at about 2200 hours as indicated by the direction of the train of vectors of the summation dial.

Since the summation dial method for analyzing nonstationary data assumes a period of 24 hours, it was important to determine that this was indeed so under the present experimental conditions and that bedrest was not affecting the circadian period of these parameters. Figure 12 compares the individual periodograms of the ambulatory and bedrested subjects, which indicate the presence of a circadian period in both cases.

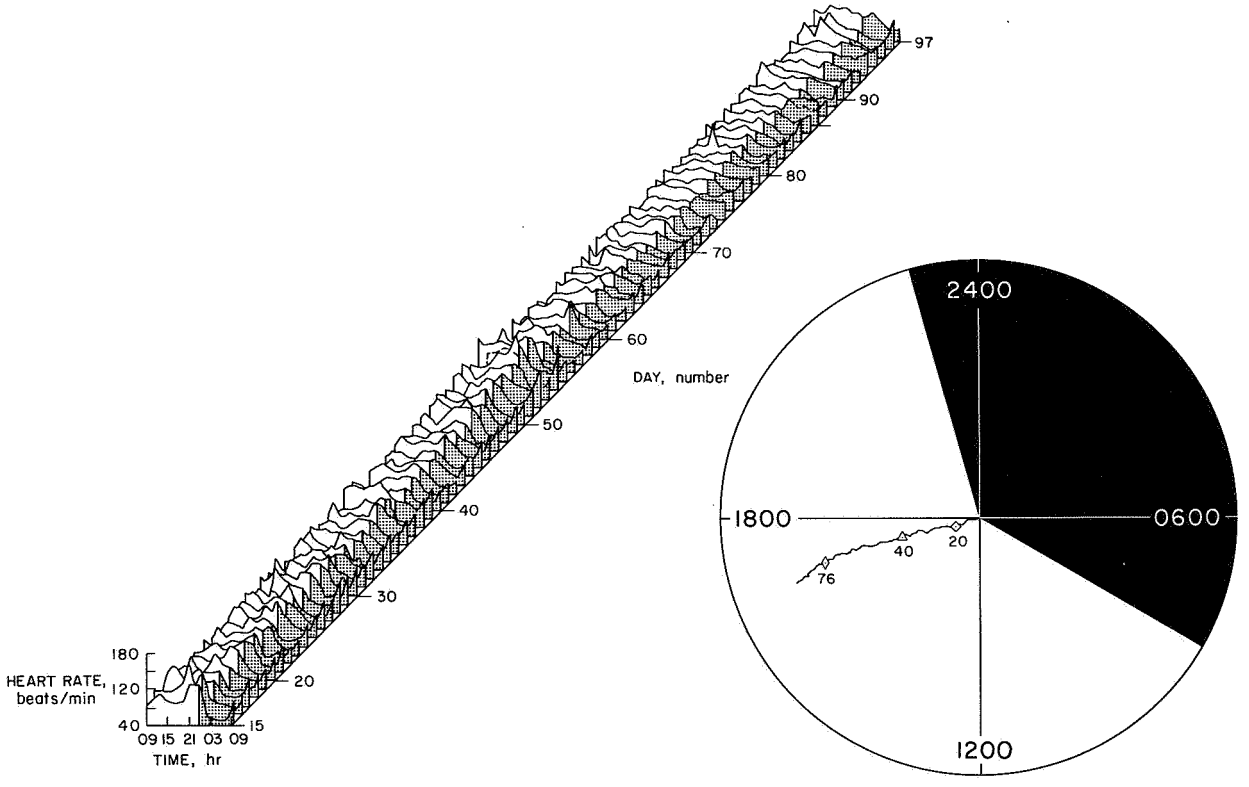


Figure 10.– Circadian oscillations in heart rate for an ambulatory subject over 96 days; actual data (left) and summation dial (right).

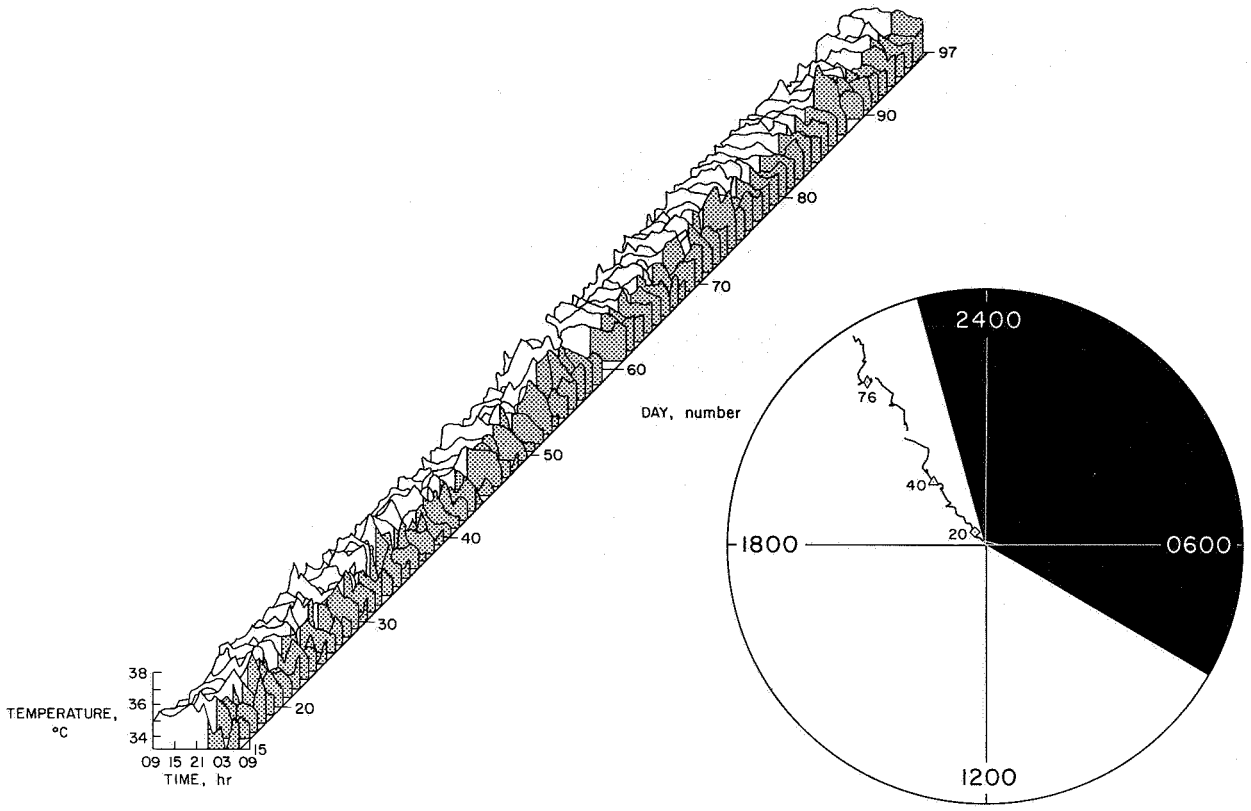


Figure 11.– Circadian oscillation in body temperature for an ambulatory subject over 96 days; actual data (left) and summation dial (right).

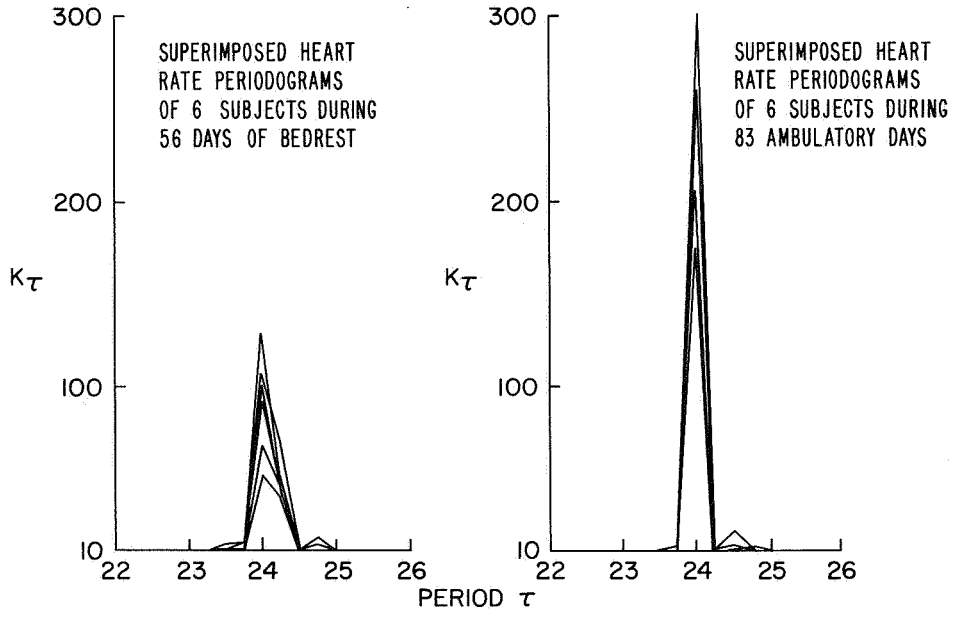


Figure 12.– Individual periodograms that indicate a circadian period in the ambulatory and bedrest subjects.

In the bedrested group (fig. 13), the HR data can be divided into four segments: a prebedrest period of stable rhythms similar to those in the ambulatory group (fig. 14). In the first 3 weeks of bedrest, there is a small phase shift (about 2 hr); a second phase shift of about 4 hours occurred

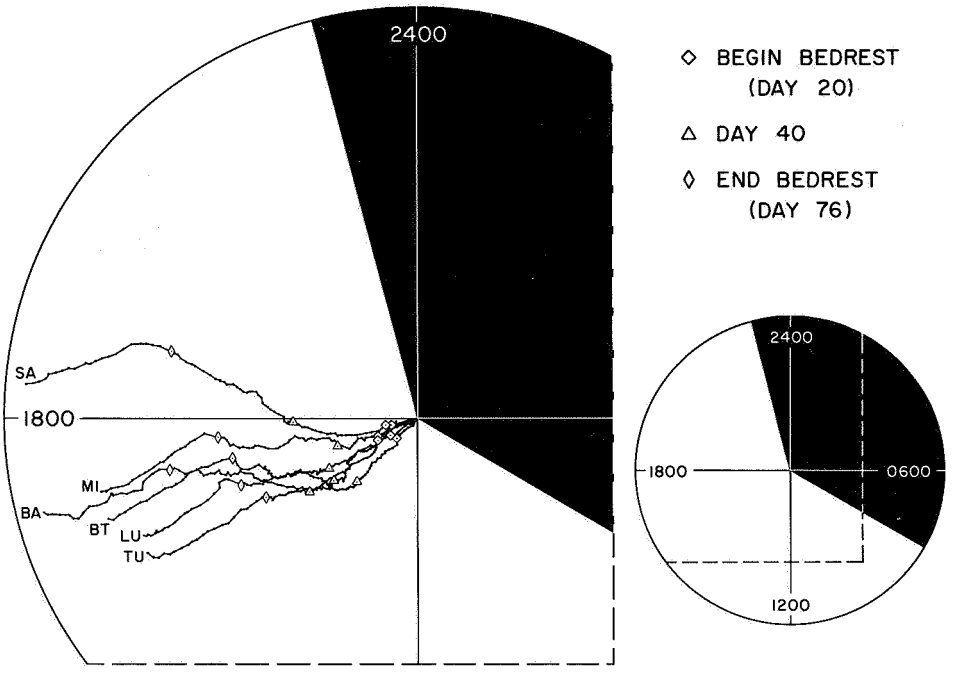


Figure 13.– Heart rate summation dials of the bedrested subjects.

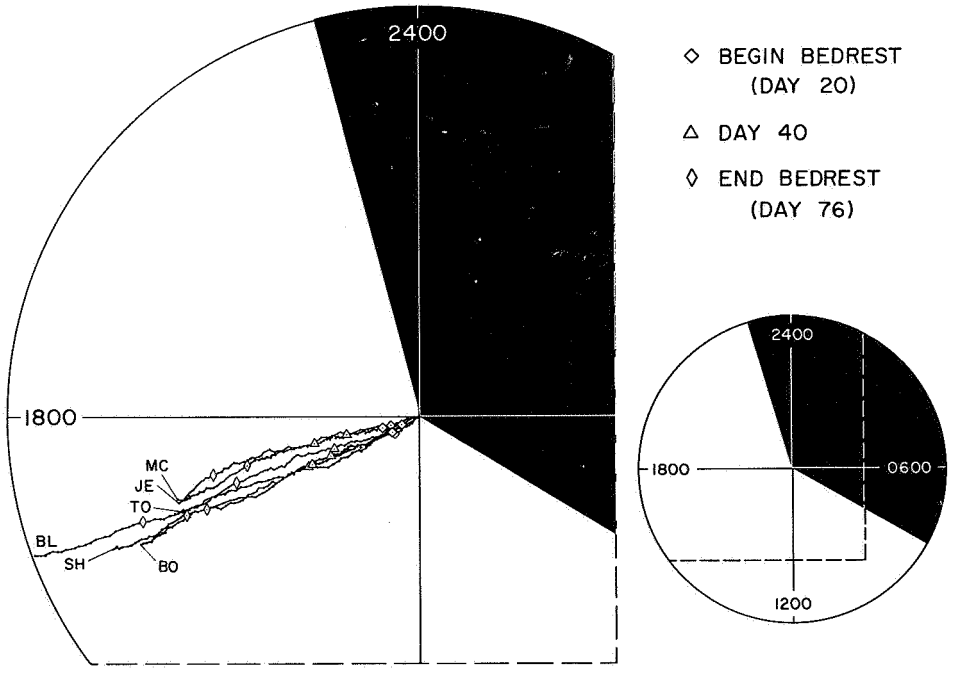


Figure 14.– Heart rate summation dials of the ambulatory subjects.

after about 20 days of bedrest (day 40 of the experiment) which lasted for the remaining 5 weeks of bedrest; postbedrest subjects almost immediately resynchronized with their original prebedrest rhythms.

The summation dials of the BT of the ambulatory and bedrested subjects are shown in figures 15 and 16, respectively. The BT of the ambulatory group was somewhat less stable that their HR rhythms. However, the peaks for all subjects occurred in the same time quadrant, that is, between 2045 and 0245 hours, as indicated by the direction of the train of vectors. During bedrest, the BT data varied even more. Three subjects peaked in the same quadrant as the ambulatory and the other three peaked around 0600 hours (60° out of phase). All subjects showed considerable phase changes throughout the bedrest; three of them showed random walks that represent rhythm asynchrony. Only two subjects resynchronized relative to base at the end of the 3-week postbedrest period.

Figure 17 compares the daily phase angle (ϕ), the daily integrated amplitude (area between the daily cycle and its mean), and the daily mean of the HR data for the ambulatory and bedrested subjects. The greatest phase-angle shift in the HR rhythm occurred about the 22-24th day of bedrest (43rd-45th day of study) and continued for the duration of the bedrest. The amplitude of the rhythm decreased sharply when the subjects got into bed and recovered promptly in the postbedrest period. Similarly, the mean HR dropped initially and then increased sharply when the subjects got out of bed and remained elevated during the 20 days postbedrest.

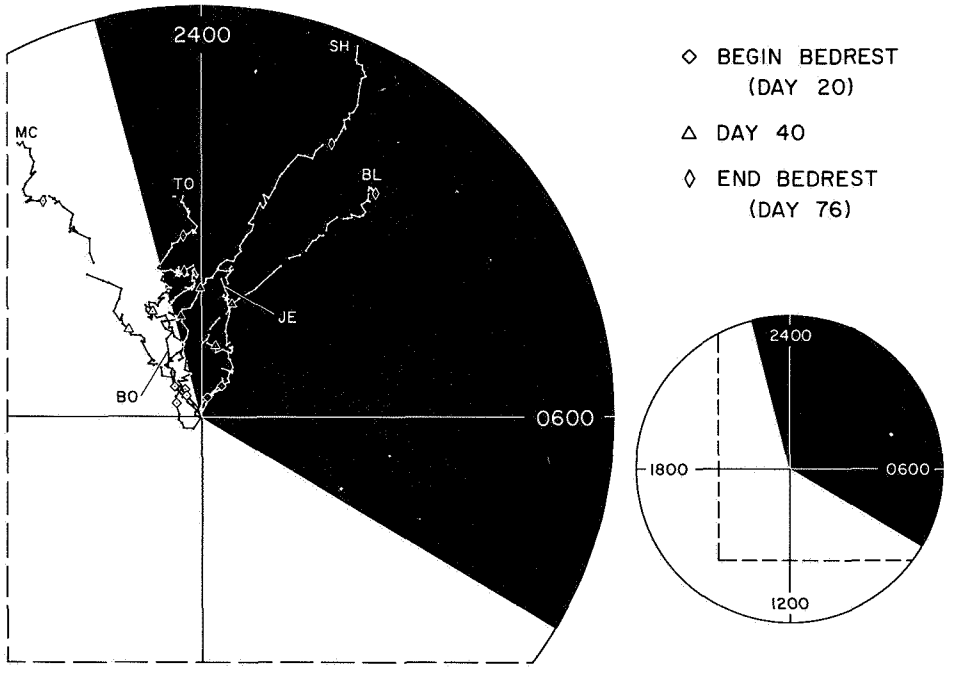


Figure 15.– Body temperature summation dials for the ambulatory subjects.

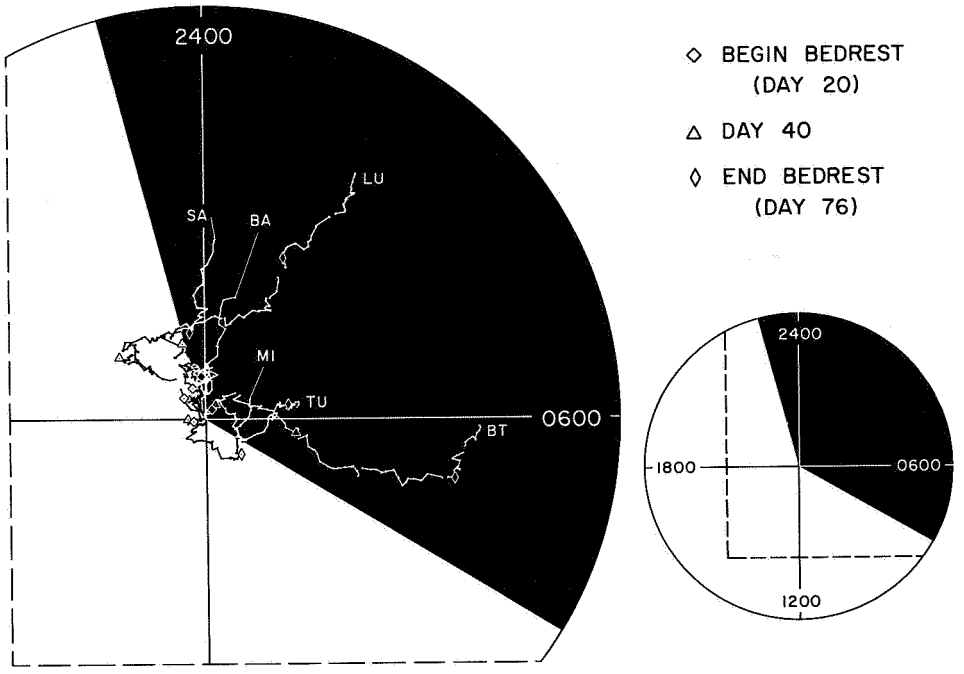


Figure 16.– Body temperature summation dials for the bedrested subjects.

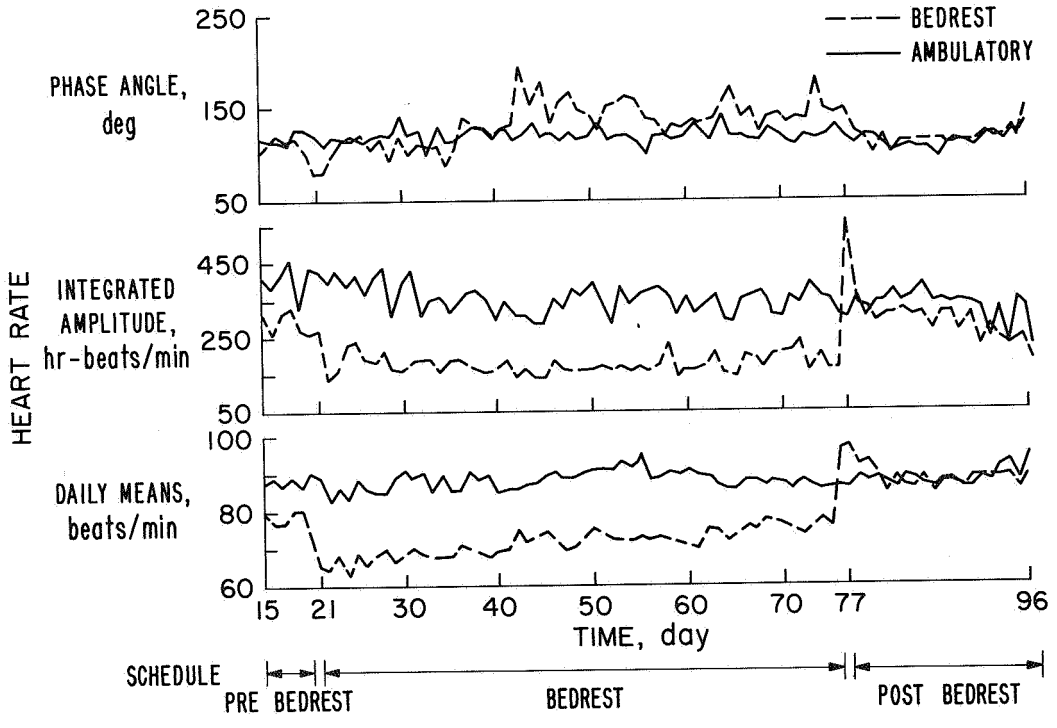


Figure 17.– A comparison of daily phase angle, integrated amplitude, and daily means of heart-rate rhythm in ambulatory and bedrested subjects.

Since the sharpest phase shift in the HR rhythm appeared to occur on about day 20 of bedrest, the data were scanned to determine more precisely the actual time and day at which it occurred (fig. 18). Four bedrested subjects showed their sharpest phase shift on day 23 of bedrest (day 43 of study) whereas the other two bedrested subjects showed a similar change on day 24 of bedrest (day 45 of study). On those days, the HR of these bedrested subjects increased sharply to 87 beats/min by 0100 hours and did not reach its lowest point (56 beats/min) until 0700 hours. In contrast, none of the ambulatory subjects showed an increase in HR at these hours; in fact, their HR decreased from 74 beats/min at 2300 to 62 beats/min at 0100 hours.

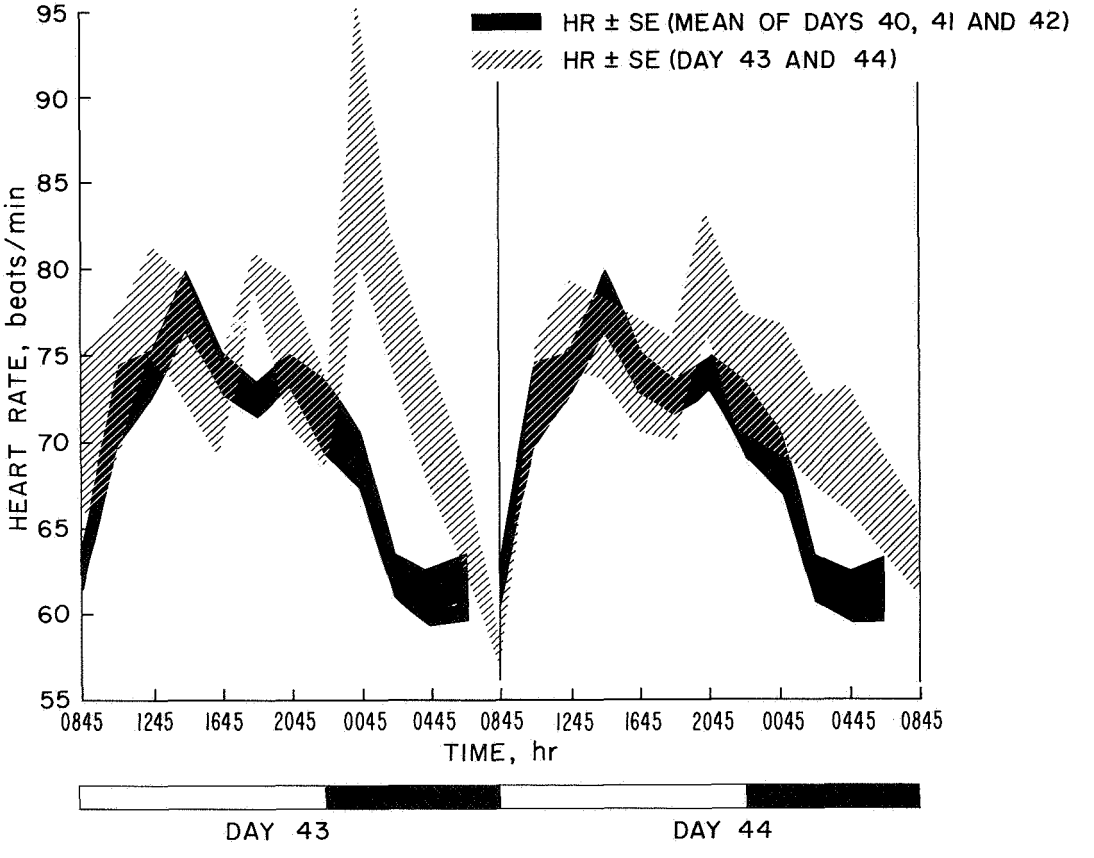


Figure 18.– Mean heart rate (±SE) of four subjects on days 23 and 24 during bedrest (days 43 and 44 of study) compared to that of the same subjects during the three previous days.

The BT rhythm did not show a sudden, single phase shift as did the HR. Figure 19 compares the daily phase angle (ϕ), the daily integrated amplitude, and the daily mean of the BT data of both groups. The results show that numerous phase shifts occurred in the bedrested subjects as compared to the relatively unchanging phase angle of the ambulatory controls and of these same subjects during their ambulatory periods. The mean daily BT decreased progressively in spite of an unchanged integrated amplitude, indicating that the BT rhythm fluctuated in bedrest with the same amplitude about a new lower mean level. The decrease in mean daily BT had not recovered by the end of the 21-day postbedrest period.

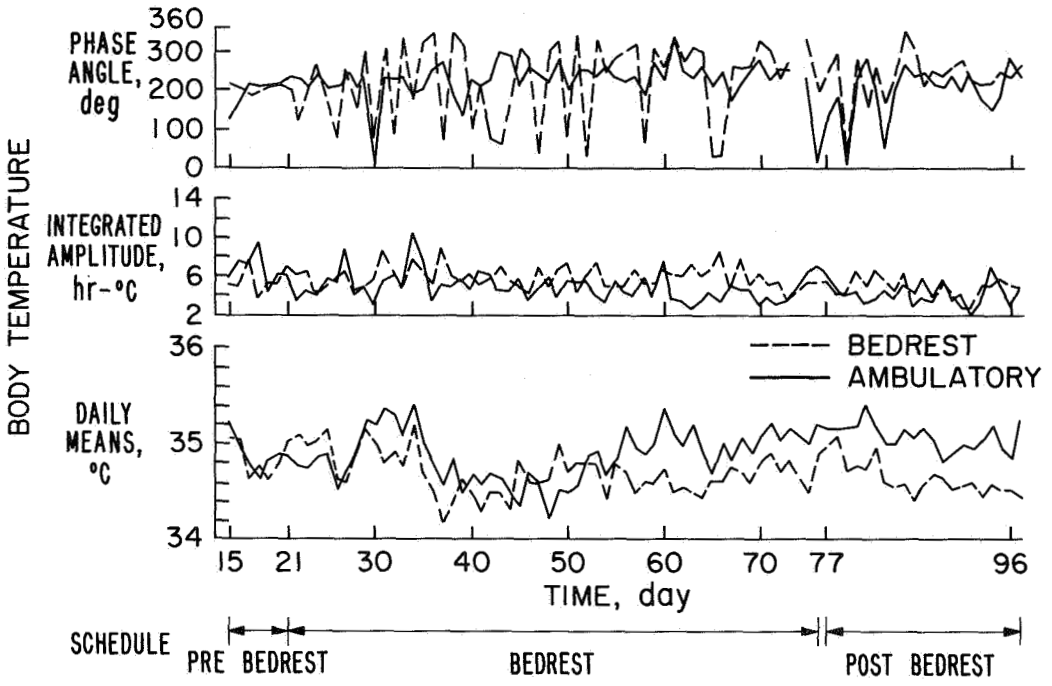


Figure 19.– A comparison of daily phase angle (ϕ), integrated amplitude, and daily means of body temperature in ambulatory and bedrested subjects.

Discussion

It has generally been agreed that light acts as the primary influence in maintaining synchrony of circadian rhythms (refs. 17-22). Research on the properties and characteristics of rhythms has concentrated on producing rhythm desynchronization by manipulating the photoperiod (refs. 23 and 24). Even the search for secondary synchronizers such as temperature and magnetic fields has involved analysis of the influence of these variables in the absence of light cues, that is, either continuous light or continuous dark environment (refs. 25-27). Study I, investigated the physiological changes that occur in man in response to prolonged bedrest; it was observed that desynchronization of some circadian rhythms occurred in spite of the fact that the subjects were maintained in a highly structured environment including a controlled photoperiod of 14L:10D. Since such desynchronization in the presence of a defined light environment has not been described previously in healthy subjects, it was important to first confirm this finding and, secondly, to determine if the change induced by bedrest was sufficiently powerful to cause rhythm asynchrony in spite of the unchanged photoperiod.

The results of study II confirmed our previous findings that the primary influence of bedrest on BT and HR rhythms is to reduce the amplitude and change their phase relationships. The normally entrained rhythms were altered *after approximately 20 days of bedrest*, when they lost their normal relationship to the photoperiod and to each other. In addition, bedrest induced a

depression of BT and an initial bradycardia. The possibility that this rhythm asynchrony may have been due to the inactivity associated with bedrest was ruled out in bedrest study I, where a moderately heavy exercise regimen did not prevent these changes. Study II also rules out the possibility that the prolonged confinement associated with this type of experiment may have been causing the observed phase shift since the ambulatory control subjects showed none of the changes seen in the bedrested individuals.

The dissociation of the HR and BT rhythms from each other and from the light schedule during bedrest and the prompt reassociation of the two rhythms in the postbedrest ambulatory period suggest that synchrony of these rhythms may be dependent on posture. Hence it seems reasonable that the postural change involved in bedrest or some physiological consequence of that, such as hydrostatic pressure changes or redistribution of body fluids and electrolytes, may be primarily responsible for the rhythm asynchrony.

Various rhythms in man have been reported to be dependent on different cues. For example, it has been suggested that the rhythm in aldosterone excretion is primarily posture-dependent (ref. 28), while the rhythmicity of other parameters such as plasma cortisol levels appear to be unaffected by bedrest and remain entrained to the light-dark cycle (ref. 29). On the other hand, HR and, to a lesser extent, BT appear to require both light and other (postural) cues to maintain their rhythm synchrony.

The level of the baseline about which the homeostatic mechanisms operate varies rhythmically, and this rhythmicity is controlled by exogenous and endogenous synchronizers (ref. 22). Since the time of Claude Bernard, physiologists have emphasized the study of the mechanisms by which organisms maintain the relative constancy of their internal environment in response to change in the external environment. Such change or stress has generally been considered as an increase in magnitude or duration of inputs to the system. With the exception of behavioral and biologic rhythm research, little attention has been given to the biological consequences of an absence of or reduction in input stimuli. The psychological consequences of isolation (ref. 30), the disturbance of circadian rhythms in an environment where light intensity was below a certain threshold (ref. 19), and the clinical consequences of prolonged bedrest in hospitalized patients have long been recognized. Studies on the effects of prolonged bedrest suggest that neither reduced activity alone nor relative confinement alone, both examples of low-input environments, result in HR and BT rhythm asynchrony. On the other hand, it appears that the reduction in input stimuli to proprioceptive receptors resulting from the postural change alone, or in addition to the confinement and inactivity inherent to prolonged bedrest, was responsible for the observed rhythm asynchrony. The results further support the hypothesis that maintaining circadian rhythm synchrony is not dependent on light alone as the environmental synchronizer. It is more likely that a variety of input stimuli is required to attain a certain threshold before synchrony of these rhythms with the environment and with each other can be maintained. If this hypothesis is true, it should be possible to maintain rhythm synchrony in bedrest by changing light intensity or other social and environmental stimuli.

4. CHANGES IN GLUCOSE, INSULIN, AND GROWTH HORMONE LEVELS ASSOCIATED WITH BEDREST

It was recently reported that the cardiovascular deconditioning and alterations in plasma volume that result following exposure to bedrest without exercise for 2 weeks are accompanied by impaired tolerance to a glucose load and excessive plasma insulin responses to glucose tolerance tests (ref. 31) (Dolkas, unpublished observations, 1972). Similarly, various degrees of physical inactivity have been reported to result in an apparent inefficient handling of glucose. Lutwak and Whedon (ref. 32) found that complete bedrest for 1 to 3 weeks resulted in decreased glucose utilization. Blotner (ref. 33) studied 70 nondiabetic adults and 16 children confined to bed for 1 month to 8 years and found excessive increases in blood glucose following a 100 g-oral glucose load; the effects were most pronounced in those confined for the longest duration. In addition, Naughton and Wulff (ref. 34) found that the insulin response to a glucose load in sedentary men is much greater than in active men, but the disappearance of glucose is the same. In none of these studies, however, were resting levels of plasma glucose or insulin reported to be altered. Our 56-day Bedrest Study II was designed to determine the changes in the circulating levels of insulin, glucose, and growth hormone (HGH) in young healthy male subjects exposed to bedrest for 56 days.

Results

Figure 20 shows the mean circulating growth hormone levels per 48-hr sampling period before, during, and after 56 days of bedrest. Each point represents the mean of twelve 4-hour samples per 48-hr period for the group of five subjects. Plasma growth hormone showed an initial drop at 10 days of bedrest, then rose significantly ($P < 0.05$) at 20 days (1.5-fold increase), and subsequently decreased gradually, reaching levels (2.5 mg/ml) well below prebedrest control levels (4.2 mg/ml) by day 54 of bedrest. The changes in mean daily plasma glucose and insulin levels are

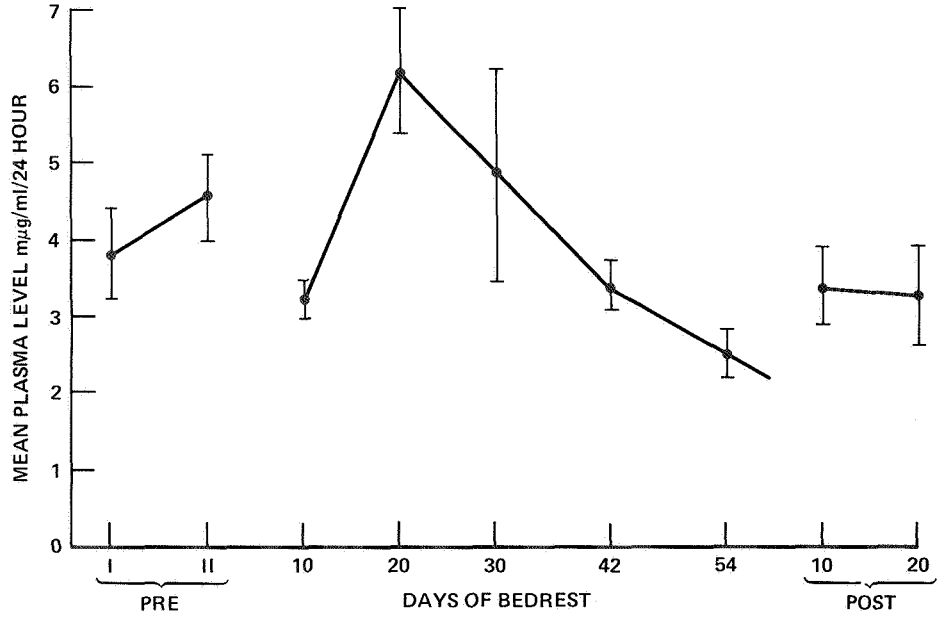


Figure 20.– Mean circulating growth hormone levels per 48-hr sampling period before, during, and after 56 days of bedrest; vertical lines represent standard errors ($N = 5$).

shown in figure 21. Glucose concentrations remained unchanged in spite of a marked increase in mean daily insulin levels during the first 30 days of bedrest. With continued exposure to bedrest, insulin began decreasing toward prebedrest levels and glucose followed with a similar reduction to below control levels. By day 54 of bedrest, glucose reduction was significant ($P < 0.05$), reaching a level of 75 mg/(100 ml • 24 hr) and recovering during the control period.

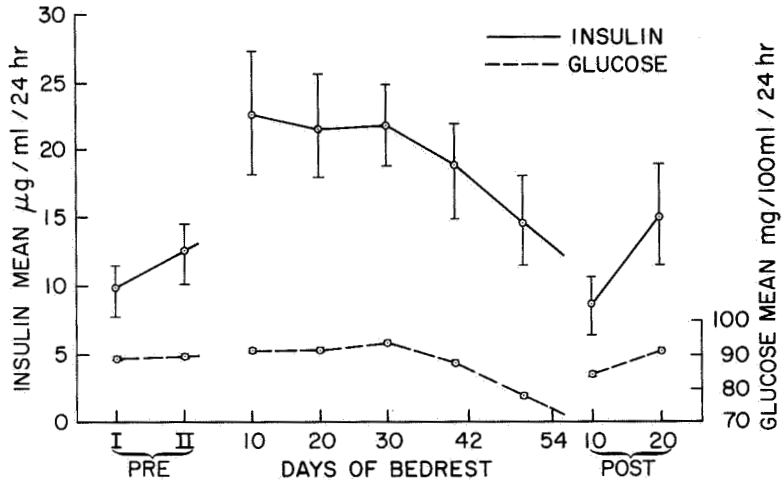


Figure 21.– Mean circulating insulin (solid line) and glucose (broken line) levels per 48-hr sampling period before, during, and after 56 days of bedrest; vertical lines represent standard errors ($N = 5$).

The daily mean changes generally do not result from an overall increase in hormone or glucose levels at all times of the day, but reflect a change in the amplitude of the diurnal variation. Figures 22, 23, and 24 show the diurnal rhythms in plasma growth hormone, insulin, and glucose,

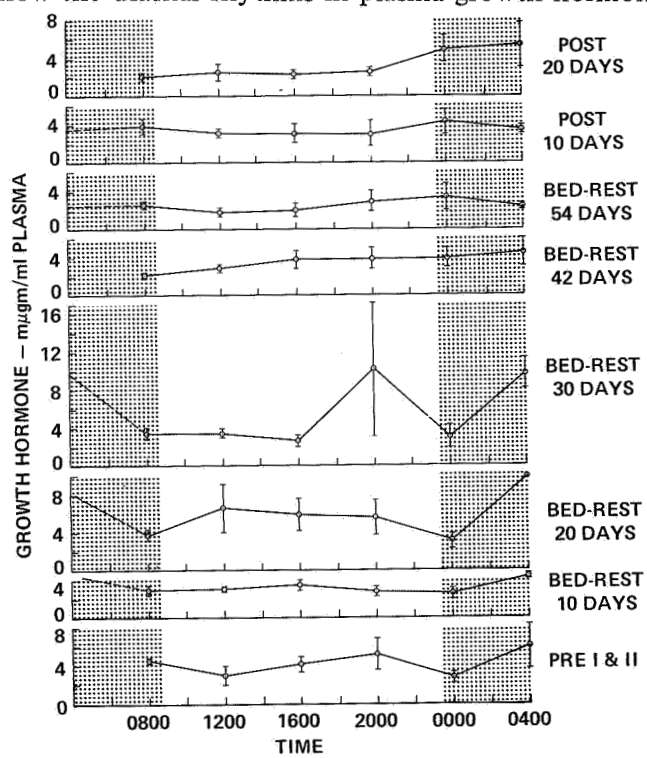


Figure 22.– Diurnal rhythms in mean circulating growth hormone levels before, during, and after 56 days of bedrest; vertical lines represent standard errors, stipled area represents lights-off period ($N = 5$).

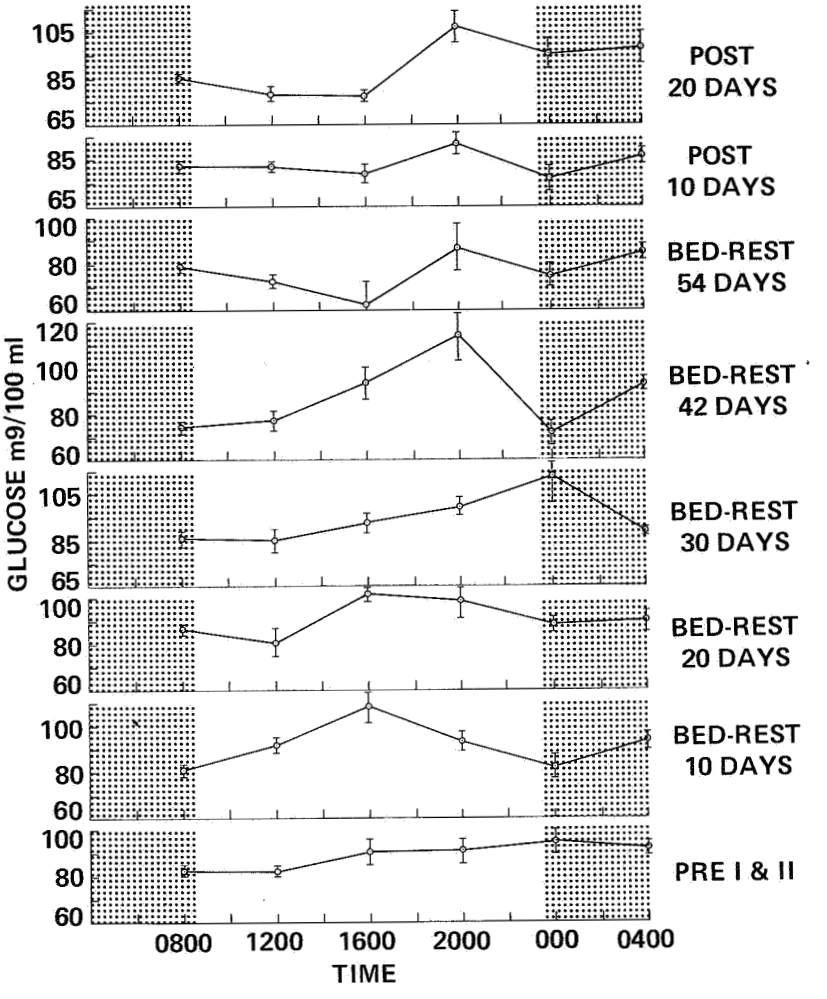


Figure 23.– Diurnal rhythms in mean circulating glucose levels before, during, and after 56 days of bedrest; vertical lines represent standard errors, stipled area represents lights-off period ($N = 5$).

respectively, at various intervals during the study. Each point represents the mean concentration in the five subjects. Growth hormone showed a significant daily fluctuation (fig. 22) with peak levels occurring at 2000 and 0400 hours. An initial reduction in amplitude at 10 days was followed by considerable increases in amplitude at 20 and 30 days of bedrest and subsequently almost complete disappearance of the rhythm on days 42 and 54 of bedrest. Although the time at which the peak occurred varied considerably, it was always associated with the late evening samples; the concentration of growth hormone was always lowest at 0730, just before lights on. This was also true for the plasma insulin and glucose rhythms (figs. 23 and 24). The amplitude of the glucose rhythm also varied throughout the study and on day 54, when the daily mean showed a significant decrease, the blood glucose concentration at 1600 hours was as low as 62.2 mg/100 ml. The mean increase in circulating insulin levels was due to the increased amplitude of its diurnal rhythm (fig. 24). The peak insulin level shifted to earlier in the day and was maintained at a higher level during the lights-on phase for the first 30 days of bedrest. After day 30, the amplitude began to decrease toward prebedrest levels.

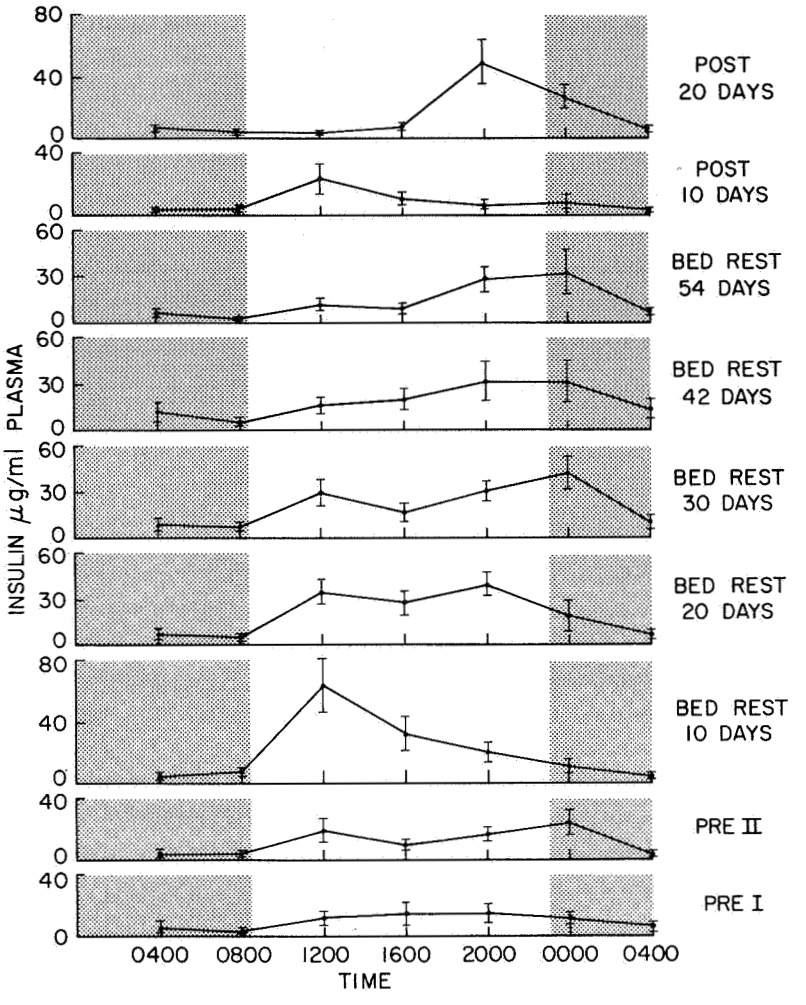


Figure 24.– Diurnal rhythms in mean circulating insulin levels before, during, and after 56 days of bedrest; vertical lines represent standard errors, stipled area represents lights-off period ($N = 5$).

Discussion

The effect of bedrest on the amplitude of diurnal rhythms has been observed for other hormones as well. Since exercise stimulates HGH secretion (ref. 35) without altering blood glucose levels, the initial drop in blood HGH during bedrest may be related to the relative inactivity of bedrest. The subsequent rise in HGH may be secondary to the increased insulin levels and may contribute to the impaired glucose tolerance reported by others (ref. 31), and to the relative ineffectiveness of the rising levels of insulin on blood glucose seen here. On the other hand, the increased levels of cortisol during the first 30 days of bedrest may be contributing to the decreased glucose tolerance and the raised plasma insulin levels by decreasing peripheral utilization of glucose by inhibiting glucose phosphorylation in muscle and adipose tissue. A further possibility is that the apparent insulin insensitivity could be attributed to increases in the radioimmunoassayable hormone

level and may not reflect biological activity. Levels of radioimmunoassayable and biological activity of various hormones have recently been found to differ, and it has been proposed that alterations in the molecular configuration of the hormone may account for these discrepancies (Nicoll, personal communication, 1973). It is also possible that bedrest brings about a reduction in the number of receptors in the target cells, as has been reported to occur in certain types of obesity (ref. 36).

Finally, the increased circulating levels of insulin required to maintain normal glucose levels, the wide diurnal fluctuations in growth hormone, insulin, and glucose levels and, particularly, the occurrence of periodic hypoglycemia, possibly in response to a meal, suggest that prolonged bedrest results in increased liability of the mechanisms that regulate glucose homeostasis and leaves open to speculation what the consequences of bedrest beyond 56 days would be.

5. CHANGES IN PLASMA ACTH AND CORTISOL CONCENTRATION IN BEDREST

Adaptability to virtually every type of environmental change remains one of man's outstanding characteristics. However, each of these changes has occurred with a constant gravitational force. In the presence of gravity, man has evolved the necessary anatomic structure and physiologic mechanism to permit him to live and function effectively. Of these, the pituitary-adrenal system, regulated by a sensitive target organ feedback mechanism, plays a key role in maintaining homeostasis by responding and adapting to the stressful or novel situations to which man is exposed. Since bedrest appears to be the closest approximation to weightlessness and since indirect evidence from the Gemini and Apollo flight data suggests that hyperactivation of this system was occurring, we determined the changes in circulating cortisol in study I and the changes in circulating cortisol and ACTH as well as urinary cortisol in study II. Since the results are very similar, they are combined for the purpose of this discussion.

Results

The mean daily hormone concentrations for the 12 samples analyzed each 48 hours are shown in figure 25. The mean 24-hr secretion of ACTH increases gradually from the beginning of bedrest but rises more sharply after 30 days. The increase measured by day 54 of bedrest was threefold. There was a gradual return postbedrest to control levels. In contrast, the mean cortisol level rose significantly during the early part of the study, almost doubling after 20 and 42 days of bedrest, but decreased sharply thereafter while ACTH was still rising.

These changes in the daily mean of circulating ACTH and cortisol concentrations reflect the change in the amplitude of their respective circadian rhythms.

Figure 26 shows these circadian rhythms in cortisol for the five subjects at various intervals during the study. Bedrest had little effect on the circadian rhythmicity of this hormone. A significant fluctuation in plasma cortisol was evident, with peak levels occurring around 7:30 a.m. throughout the experiment. However, progressive bedrest initially increased and, by day 54, reduced the amplitude of the steroid rhythm.

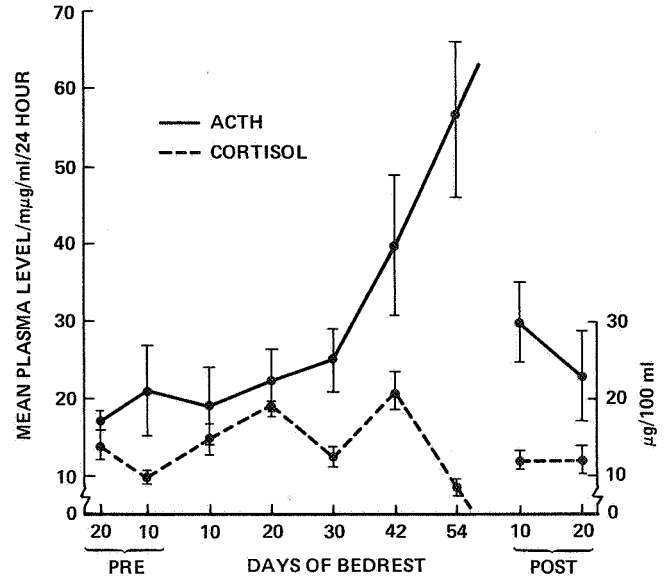


Figure 25.– Mean circulating cortisol and ACTH per 48-hr sampling period during 56-days of bedrest; vertical lines represent SE ($N = 5$).

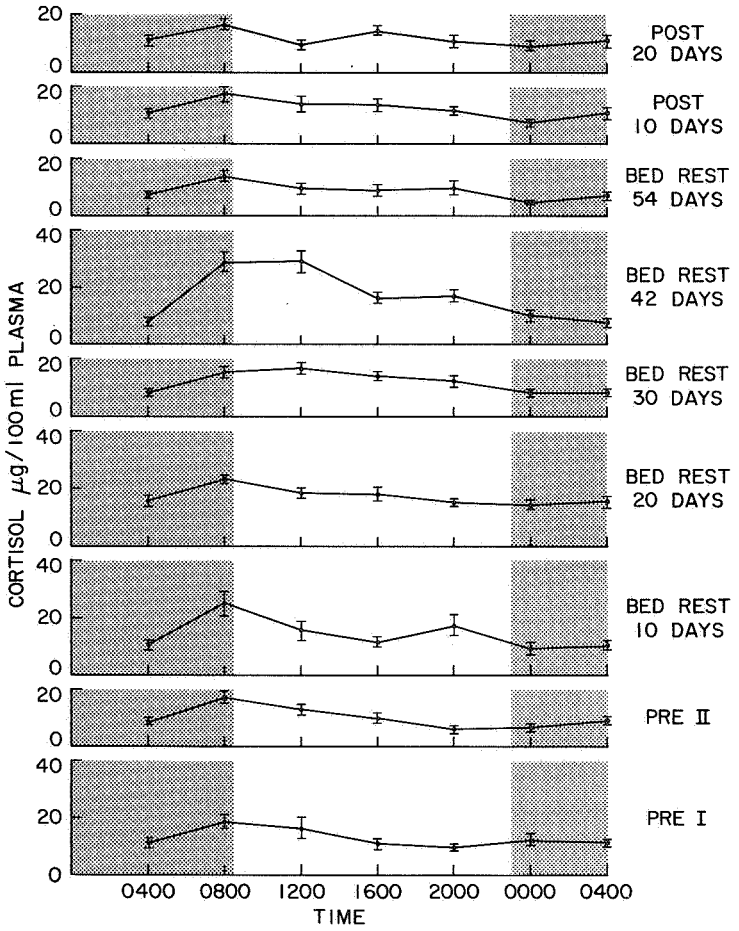


Figure 26.– Plasma cortisol rhythm at various intervals during 56 days of bedrest; vertical lines represent SE ($N = 5$), stipled area represents lights-off period.

The daily rhythm in circulating ACTH (fig. 27) was considerably less stable than the cortisol rhythm, showing several peaks and phase shifts as the bedrest progressed. These results confirm previous reports of significant diurnal fluctuations in ACTH and cortisol. The rhythmicity of both hormones is generally variable with evidence of several peaks. Numerous spikes in these hormonal secretions throughout the day were reported recently to occur approximately every 20 minutes (refs. 37 and 38). Since sampling was at four hourly intervals in this study, some of the variability in our results may be related to this phenomenon. Individual variability was considerably greater for the ACTH than the cortisol rhythms. However, progressive bedrest reduced the amplitude of the steriod rhythm and increased the amplitude of the ACTH secretion. During bedrest and in the postbedrest period, a secondary peak around 1600-2000 hours was observed in hydrocortisone. A similar second peak has been reported by Perkoff *et al.* in studies conducted on 51 normal subjects (ref. 39). As with cortisol, the most impressive change was also in the amplitude of the ACTH rhythm, which started to increase after day 30 and fluctuated from 10 to 120 pg/ml (a 12-fold change) over a 24 hour period by day 54 of bedrest. This large amplitude in the rhythm decreased promptly when the subjects got out of bed. Thus, there was an inverse relationship between the changes in the mean daily circulating levels of cortisol and ACTH throughout bedrest.

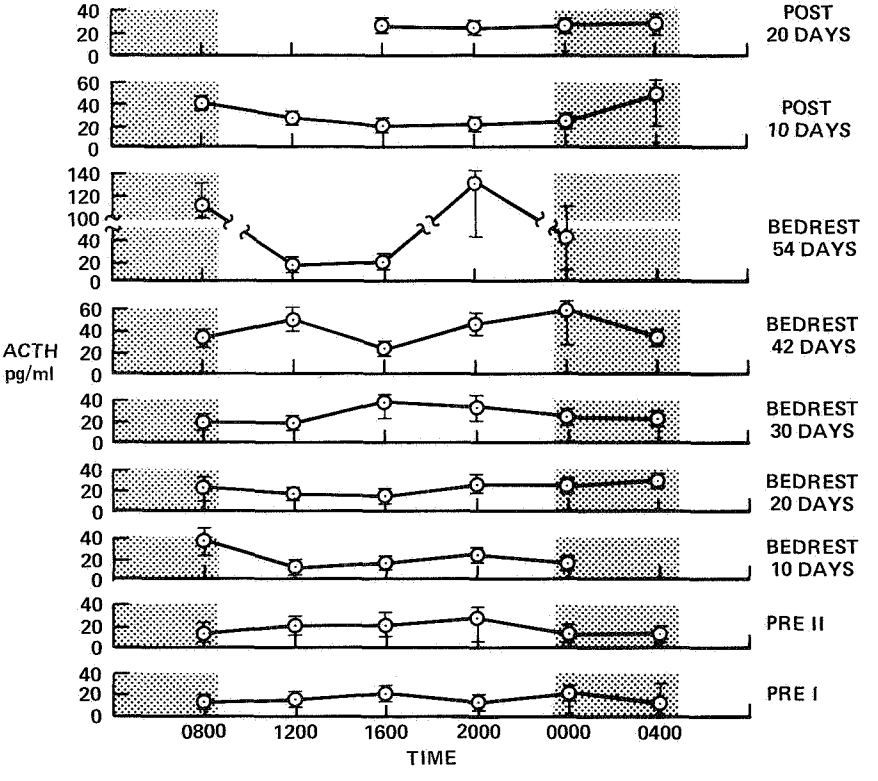


Figure 27.– Plasma ACTH rhythm at various intervals during 56 days of bedrest; vertical lines represent SE ($N = 5$), stipled area represents lights-off period.

That the well-known negative feedback mechanism regulating ACTH secretion was functional during bedrest is indicated by the data in figure 28, which shows a correlation between mean 24-hr prebedrest cortisol levels and the mean 24-hr level during bedrest for each of the five subjects. Those subjects with the lowest initial levels showed the greatest increase during bedrest and vice versa.

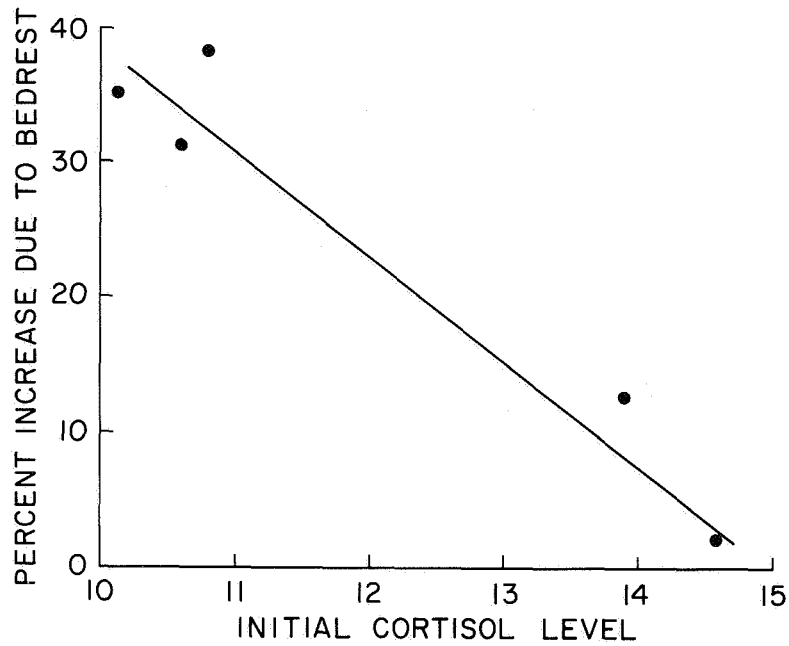


Figure 28.– Correlation between mean 24-hr prebedrest cortisol level with the mean 24-hr level during bedrest for each of the five subjects.

Discussion

The average plasma levels of cortisol found in these subjects during their ambulatory period and the circadian changes of these levels agree with past studies (refs. 40 and 41). This pattern consisted of a peak around 0730 just before lights on and a progressive decrease thereafter to midnight.

The diurnal rhythms of plasma ACTH and cortisol levels have been studied in depth (refs. 28 and 42). Besides documenting the existence of rhythms in these circulating hormones, studies have been conducted with reference to the effect of changes in work-rest or light-dark cycles on these rhythms (refs. 43 and 44). The response of the pituitary-adrenal system to prolonged bedrest showed an early rise in circulating cortisol with a very gradual rise in circulating ACTH followed by a decrease in steroid levels as bedrest progressed, but accompanied by a very marked increase in plasma ACTH. Exercise did not prevent these changes and the confined but ambulatory subjects did not show comparable effects.

The rise in daily cortisol levels early in the study may be partly attributed to the reduction in plasma volume known to occur within the first week of bedrest (ref. 45, 46) and partly to the slow rise in ACTH observed since relatively small changes in circulating ACTH are sufficient for maximal cortisol secretion. The decrease in measurable free cortisol in the plasma is thought to reflect an increase in the urinary excretion of the hormone. This is supported by previous studies in which an increase in 24-hr excretion of cortisol was noted in individuals bedrested for prolonged periods (ref. 47).

It is possible that the greater increase in circulating ACTH that occurs with progressive bedrest may be in response to the decreasing plasma cortisol levels. It is also possible that this large increase in ACTH measured by radioimmunoassay may not reflect levels of the biologically active hormone. These possibilities are being investigated. Alternatively, it is possible that the biphasic response seen with bedrest in man is similar to that characteristically seen in animals exposed to chronic stress for several weeks (ref. 47). In animals exposed chronically to such stresses as confinement, cold, centrifugation, or water restriction, the pituitary-adrenal response is characterized by an initial activation as evidenced by high circulating steroid levels followed by what has been considered to be "adaptation," that is, a return of steroid concentrations to prestress levels. Recently, we have shown that at the time when plasma steroids have decreased (or adapted) in spite of continuing exposure to the stressor, the pituitary-adrenal system is hyperreactive to additional stimuli, responding with a much greater and faster secretion of ACTH and corticosteroids than normal. The experimental evidence indicates that during such chronic or repeated stress the ACTH driving mechanism may compensate or override steroid feedback (refs. 48 and 49).

6. THYROID AND PARATHYROID HORMONE CONCENTRATIONS DURING BEDREST

Because of the reduced activity that necessarily accompanies bedrest and of the obvious loss of calcium from the bone, the changes that prolonged bedrest could produce on the secretion of the thyroid and parathyroid glands was one of the primary interests of these studies. Since most physiological systems vary diurnally, it was important to determine whether such rhythms existed in these parameters and how they were affected by bedrest. The possibility of a diurnal variation in thyroid activity has been investigated using only indirect indices without definite conclusions. For instance, fluctuations in protein-bound iodine have been reported to run parallel to adrenal activity, whereas the iodine-131 uptake of the thyroid gland showed the opposite pattern with its peak between 7:30 and 11:30 p.m. (ref. 50). Such a rhythm in parathormone has not been reported. By sampling around the clock, not only could the existence of such rhythmicity be established but any changes produced by bedrest could be better dissociated from those effects on rhythm periodicity, phase, and amplitude. These studies were therefore undertaken to determine the effect of continuous 56 days of bedrest with or without exercise on the diurnal rhythms in circulating levels of thyroxine, tri-iodothyronine, and parathormone.

Results

Figure 29 shows the relationship of the rhythm in circulating levels of thyroxine and tri-iodothyronine to cortisol in the eight subjects in study I during their ambulatory prebedrest control period. All three hormones showed a significant ($P < 0.05$) diurnal fluctuation, with maximal levels occurring at 7:30 a.m. anticipating lights-on. In addition, thyroxine showed a secondary peak at 3:30 p.m. The amplitude of both thyroid hormone rhythms was much smaller than that of the corticosteroid. Both thyroid hormones showed more variable rhythms during the bedrest period with a return to original rhythmicity at the postbedrest collection period. In fact, no significant rhythmicity ($P > 0.05$) (fig. 30) existed throughout the bedrest period because of the enormous individual variation among subjects in the occurrence of the daily peak in both parameters. Exercise did not appear to increase the stability of these rhythms.

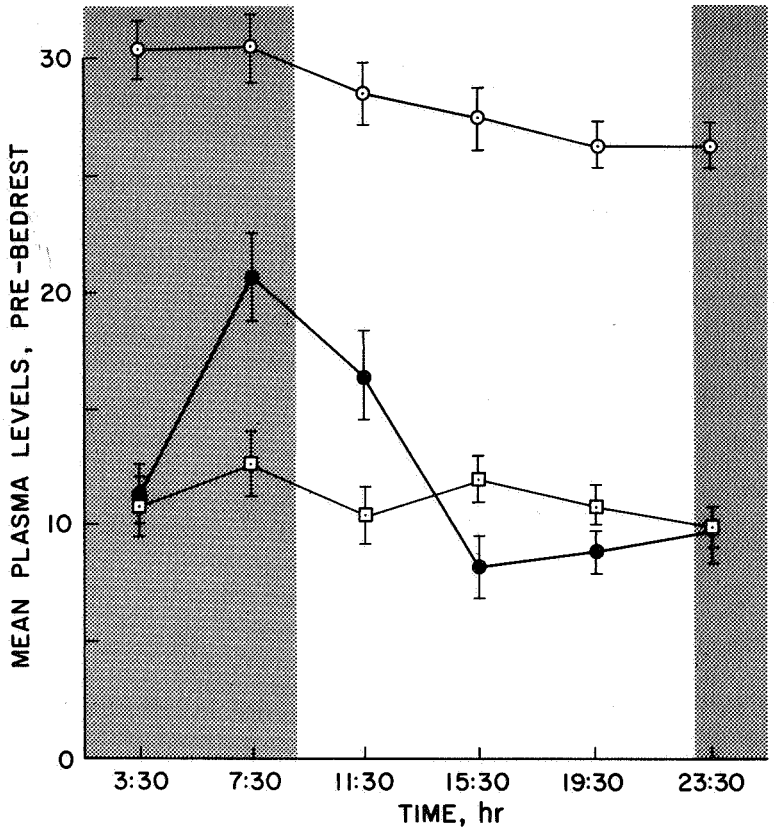


Figure 29.– Diurnal rhythm in mean circulating cortisol (●), Thyroxine (⊡), and triiodothyronine (⊙) in eight normal ambulatory subjects; vertical lines represent SE of the mean, stipled area represents lights-off period.

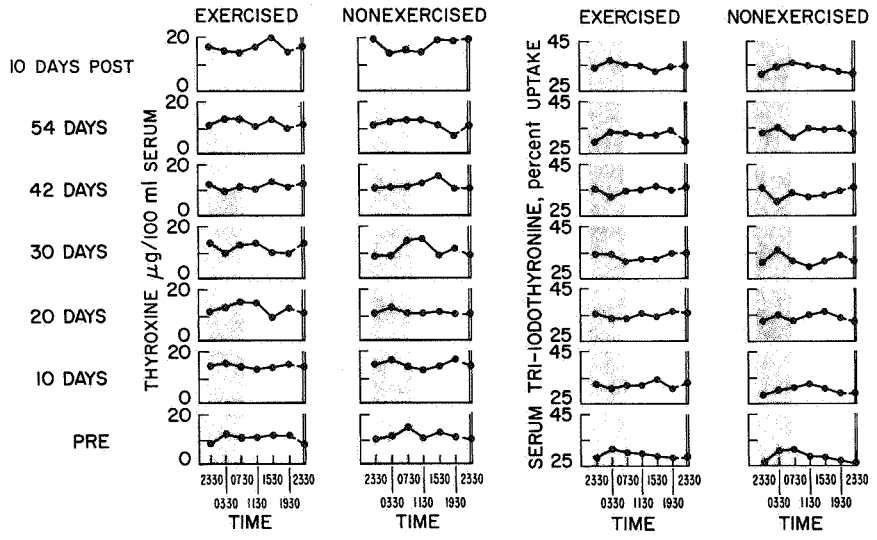


Figure 30.– Serum thyroxine and triiodothyronine in four exercised and four nonexercised subjects before, during, and after 56 days of bedrest; stipled area represents lights-off period.

Figure 31 illustrates the effects of continuous bedrest for 56 days and of exercise on the integrated daily mean thyroid hormone levels of these subjects. This was done by plotting the mean circulating levels of the three hormones per 48-hr sampling period as well as the maximal and minimal concentrations (range or amplitude) that occurred during that period, regardless of the hour of day they occurred.

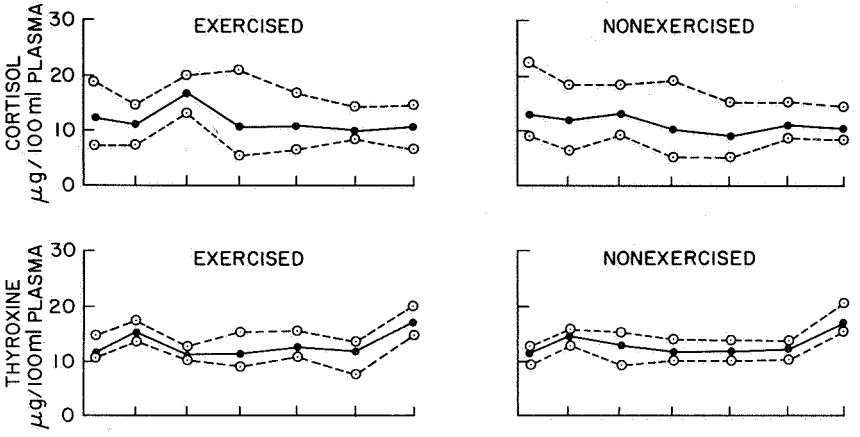


Figure 31.– Integrated daily mean of plasma thyroxine and triiodothyronine in subjects bedrested 56 days with or without exercise.

Both exercised and nonexercised subjects showed similar patterns in circulating thyroxine. The only effect of bedrest appeared to be a transient increase in the mean daily concentration during the first 10 days, with no change in amplitude throughout the experiment. However, in both groups of subjects, there was an obvious increase in mean thyroxine levels when the subjects got out of bed. On the other hand, the concentration of tri-iodothyronine markedly increased during the first few days of this study (study I) and remained elevated in both groups of subjects for the duration of the experiment as well as through the postbedrest period. In study II, a similar disappearance in the thyroid hormone rhythms was observed. However, in contrast to the first study, the daily mean level of the hormones (fig. 32) remained unchanged throughout 96 days of the experiment.

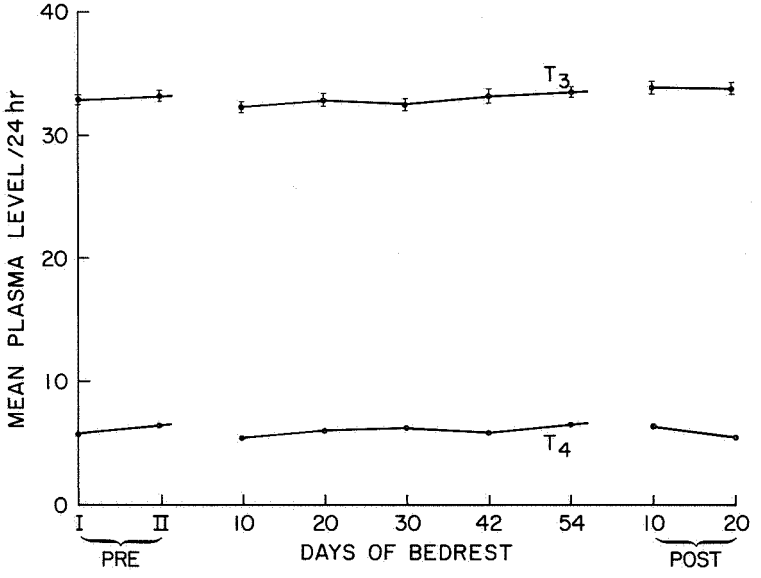


Figure 32.– Integrated daily mean of plasma thyroxine and triiodothyronine in subjects ($N = 5$) bedrested 56 days without exercise.

No obvious circadian rhythm in plasma parathormone levels was observed during the 20-days prebedrest ambulatory phase of study II (fig. 33). Levels ranged from 0.05 to 0.6 mg/ml plasma, but maximal and minimal levels did not consistently occur at any one time of day nor was there any uniformity between subjects. The period of bedrest was characterized by an increase in the number and size of the fluctuations throughout the day. This was particularly true around 20 days of

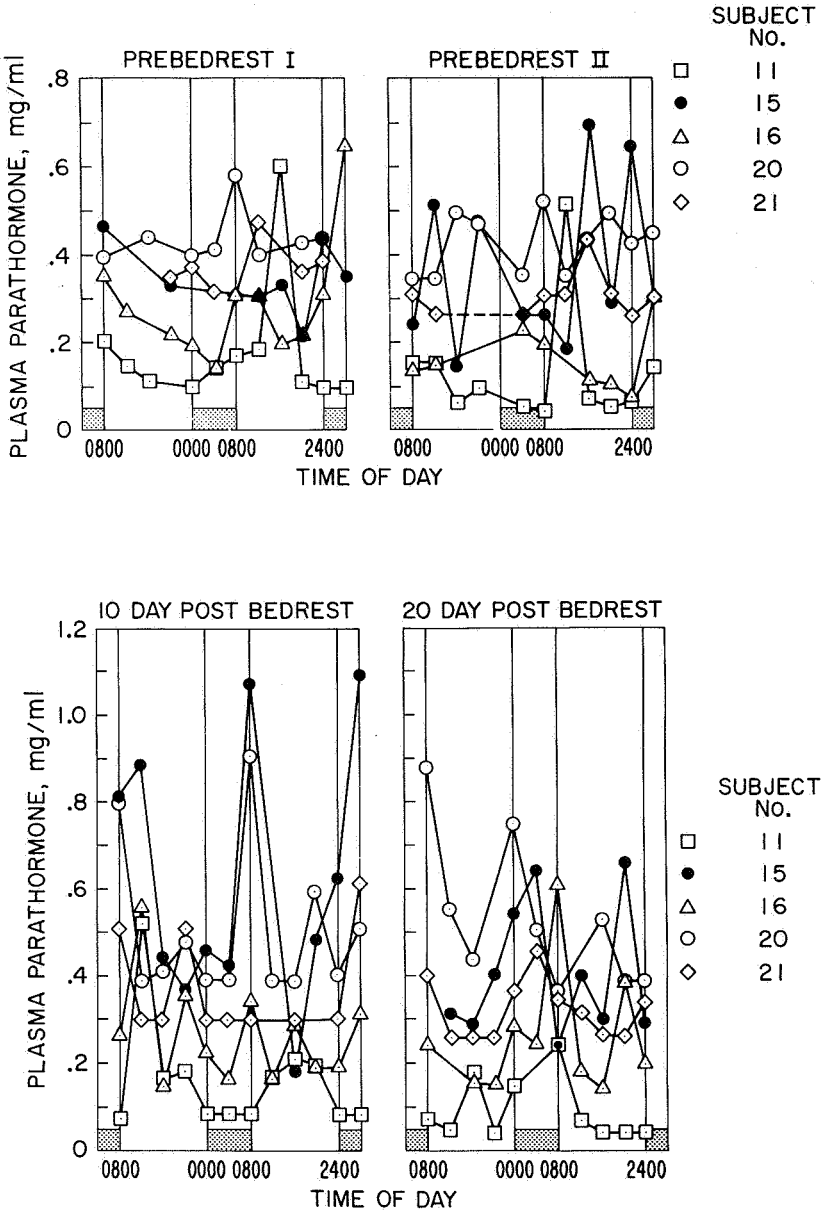


Figure 33.– Plasma parathormone levels in five ambulatory subjects.

bedrest. During the postbedrest period (fig. 34), there was again a greater degree of fluctuation within a day, the levels ranging from 0.05 to 1.1 mg/ml plasma. Figure 35 shows the integrated daily mean for plasma parathormone concentrations throughout the 96 days of the study for each of the five subjects. An overall increase in daily parathormone levels occurred during bedrest with a great deal of individual variation. Four out of five subjects showed an initial increase with bedrest. With three subjects, it was sustained throughout bedrest and was apparent during the postbedrest period. In the fifth subject (subject 16), parathormone levels remained unchanged.

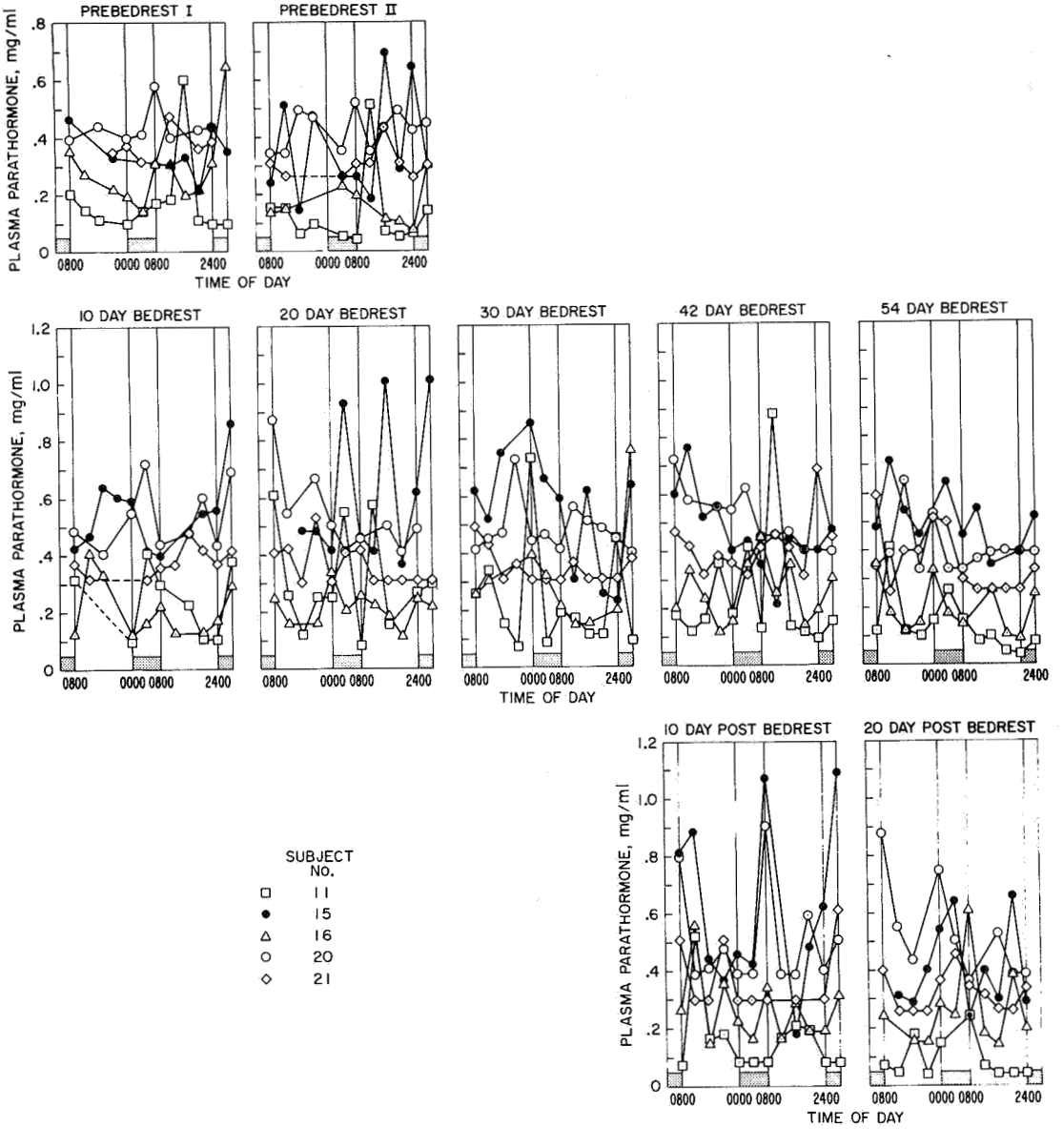


Figure 34.– Plasma parathormone in 56-day bedrest II.

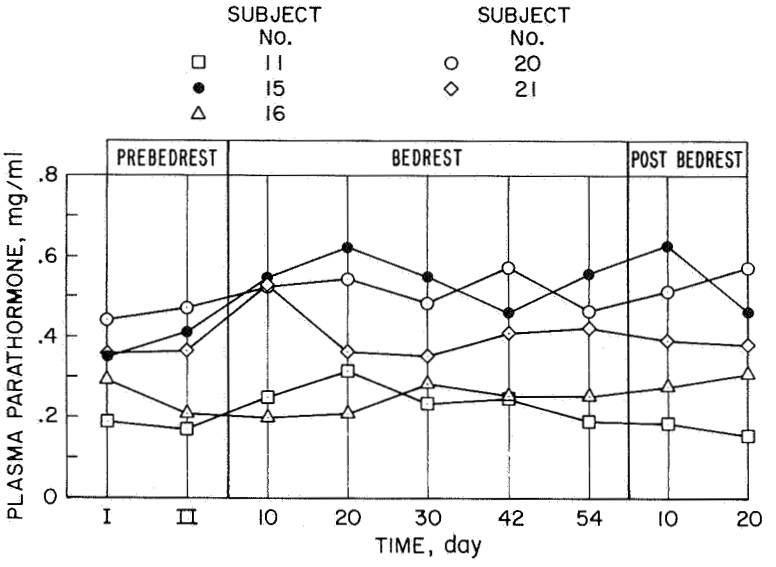


Figure 35.– Daily mean plasma parathormone in 56-day bedrest II.

Discussion

Conflicting reports on the presence or absence in man of circadian variations in thyroxine levels have appeared (refs. 51-54). In this study, the diurnal variation in the serum concentration of both tri-iodothyronine (T_3) and thyroxine (T_4) approximately paralleled that of adrenal activity, with the peak occurring about 0730 hours or before lights-on. Walfish *et al.* (ref. 54) reported a rise in blood thyroxine between 0200 and 0800 hours in a group of 11 euthyroid subjects. This also coincides with the peak in protein-bound iodine reported by others (ref. 50), while the peak in iodine-131 uptake of the thyroid gland occurs at about 1930 hours. The increase in plasma TSH levels would be expected to precede the increase in thyroid hormone secretion. A circadian rhythm in TSH concentration in man with peak values between 0200 and 0300 hours has indeed been reported (ref. 41). Bakke and Lawrence (ref. 55) report maximal levels of serum and pituitary TSH in the nocturnal hypothyroid (propylthiouracil-fed) rat between 8:00 a.m. and noon. Using more sensitive methods, Singh *et al.* (ref. 56) report peak plasma TSH levels at 1500 hours in euthyroid rats.

The low amplitude of the diurnal variation in thyroid activity in humans (as determined by measuring T_3 and T_4 in the blood) may be partly responsible for the confusion in the literature regarding the existence of such a rhythm. Furthermore, the relative instability of this rhythm may have led to erroneous results in past studies unless the environmental and activity conditions of the subjects were rigidly maintained. It is not unusual that subjects hospitalized for other reasons are used for studies of diurnal rhythmicity. For instance, Schatz and Volpe (ref. 52) used seven euthyroid patients, all in the hospital for nonthyroid illnesses, and found no marked fluctuations in protein-bound iodine in blood drawn at 0800, 1330, 1730, and 0300 hours. It is quite likely that these subjects were confined to bed and hence their results are not surprising since, in our subjects, bedrest promptly caused a disappearance or irregularity of both T_3 and T_4 rhythms which were not improved by exercise but were immediately restored when the subjects got out of bed.

Bedrest had little effect on the overall T_4 levels, except the sharp increase observed when the subjects got out of bed in study I. Also in study I, there was a prompt and sustained elevation of T_3 levels as the bedrest progressed.

Earlier experiments in humans subjected to bedrest for 6 days only showed decreased T_3 and normal T_4 levels during this early period (C. Leach, unpublished observations). However, a concomitant increase in thyroid binding globulin was also seen in these subjects, which may account for the apparent decrease in T_3 using the resin uptake procedure of Katz (ref. 57). In the 56-day bedrest studies reported here, thyroid binding globulin was not determined. Study II did not show the changes in mean daily circulating T_3 and T_4 observed in study I although bedrest again abolished the thyroid rhythm.

Nicoloff *et al.* (ref. 58) postulated a negative feedback action of circulating hydrocortisone in regulating the diurnal rhythm in thyroid function as measured by thyroidal iodine release and serum TSH values. Their hypothesis was based on the evidence that pharmacological doses of glucocorticoids suppress TSH secretion while a rebound in TSH release follows withdrawal of the steroid (ref. 59). Although the inverse relationship between adrenocortical and overall thyroid function has been demonstrated by several investigators, our data show a dissociation between T_3, T_4 and cortisol rhythmicity by bedrest which does not support the thesis that the diurnal rhythm in thyroid function is under corticosteroid regulation (ref. 58).

Although the thyroid hormone rhythms (T_3 and T_4) were of low amplitude and relatively unstable, their circadian rhythmicity was clearly demonstrable. This was not true for the circulating levels of parathyroid hormones. Although changes of as much as tenfold in the concentration of this hormone were observed in the ambulatory subjects, these did not appear to be of any particular frequency nor were they in any way associated with lights on or off or other obvious change in the environment. When the subjects were exposed to bedrest, an overall increase in the daily mean circulating parathormone concentration was evident in four of the five subjects. As with other hormones, this change was due to the greater amplitude in the sharp fluctuations of the hormone, the levels now showing as much as a 20-fold change within a given day. This showed a return to prebedrest values 20 days after the subjects had again become ambulatory.

7. SUMMARY AND CONCLUSIONS

Summary

Two bedrest studies of 56 days each, involving a total of 20 male subjects aged 20-26 (one subject aged 40), have been conducted to evaluate the effects of prolonged bedrest on circadian synchrony and endocrine and metabolic function. In addition, the contribution to the observed results of factors inevitably associated with bedrest (i.e., lack of activity, confinement, and the blood sampling schedule) was assessed. Around the clock blood sampling was designed (a) to determine the effects of bedrest on circadian synchrony as a measure of central nervous system function and (b) to determine if changes in hormone levels are sustained or intermittent. Measurements included the pituitary-adrenal, thyroid, parathyroid, insulin-glucose-growth hormones, catecholamine excretion, body temperature, and heart rate.

Significant Findings

1. Bedrest resulted in rhythm asynchrony (body temperature, heart rate, thyroid hormones, insulin, and other hormones) in spite of a well-regulated light/dark environment. The most drastic rephasing of heart-rate rhythms occurred suddenly on day 23 or 24 in all 14 bedrested subjects but not in ambulatory controls.

2. Mean daily body temperature decreased about 1°C in all subjects by the end of 56 days of bedrest.

3. Changes in hormone and glucose levels during bedrest were not sustained throughout the day, but reflected changes in the amplitude of the fluctuations.

4. Glucose homeostasis was maintained for the first 30 days of bedrest accompanied by a 2.5-fold increase in circulating insulin levels. Beyond that, insulin levels fell as did glucose (by day 54, the glucose level at 4:00 p.m. was 62.2 mg/100 ml).

5. Growth hormone increased initially with bedrest, but after 20 days it decreased to well below control levels. The pituitary did not respond to hypoglycemia by a rise in growth hormone secretion.

6. Plasma cortisol concentration doubled during the first 20 days of bedrest but decreased subsequently to levels below controls by day 54. Plasma ACTH remained relatively unchanged during the first 30 days but then showed a three-fold increase by day 54. Plasma cortisol was the only hormone whose circadian rhythm did not desynchronize with the photoperiod during bedrest.

7. Thyroxine and tri-iodothyronine levels in plasma showed minor increases or no change. However, their diurnal rhythmicity was abolished by bedrest.

8. Parathormone levels showed an overall increase and greater fluctuations during bedrest.

9. Sporadic urinary catecholamine measurements suggested increased excretion during bedrest.

Conclusions

1. A rigorous regimen of isotonic/isometric exercise did not prevent the endocrine and metabolic effects of prolonged bedrest.

2. Changes in circadian, endocrine, and metabolic functions in bedrest appear to be due to changes in hydrostatic pressure and lack of postural cues rather than to inactivity, confinement, or the bleeding schedule.

3. Changes in circulating metabolic and endocrine parameters are unreliable if measured once per day because their amplitude and time of peak of their diurnal fluctuations are altered during bedrest. Therefore, data should be expressed as units/24 hours.

4. Recovery periods up to 20 days are insufficient for full recovery from 56 days of bedrest.

5. Bedrest beyond 42 days results in periodic hypoglycemia, possibly in response to meals, which may warrant modification of meal composition.

6. Prolonged bedrest results in an apparent insensitivity of the glucose response to insulin, of cortisol secretion to ACTH, and of growth hormone secretion to hypoglycemia. This may be due to an effect of bedrest on the number or sensitivity of target organ receptors; it may reflect a change in radioimmunoassayable levels of the peptide hormones, or it may result from an alternation of the central nervous system's input/feedback integrating mechanisms.

7. Prolonged bedrest results in increased lability of all endocrine and metabolic parameters measured as indicated by larger daily fluctuations (except thyroid), and decreased circadian stability.

8. Prolonged bedrest, particularly beyond 24 days, results in rhythm desynchronization in spite of well-regulated light/dark cycles, temperature, humidity, activity, and meal times and meal composition.

9. The results suggest bedrest deconditioning interferes with central neuroregulatory mechanisms.

Ames Research Center
National Aeronautics and Space Administration
Moffett Field, Calif. 94035, January 1974

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