DESYNCHRONIZATION OF HUMAN CIRCADIAN RHYTHMS

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In man as well as in animals, diurnal rhythms are based on endogenous periodic processes which can be considered self-sustained oscillations in a technical sense[2]. Evidence for this is given by the observation that, under constant conditions, the rhythm continues a) undamped and b) with a frequency which deviates from that of the earth's rotation[6]. It is this deviation of the free-running 'circadian'[11] period from 24 hours which excludes external cues as possible causes of the rhythm. In many aspects, the whole organism behaves like a single oscillator which obeys the law of oscillation theory. Some findings, however, suggest that the circadian system consists of a multiplicity of oscillators which normally are synchronized with each other but which can become desynchronized under special circumstances[13,14]. This hypothesis seems to be supported by recent results from experiments with human subjects which will be discussed below.

METHODS

Single subjects are kept in isolation and without watches in an underground sound-proof bunker which is continuously illuminated. Next to the bed-sitting-room is a toilet and a small kitchen. The subject prepares his own meals. His rectal temperature is measured continuously by means of an electric thermometer and recorded outside the experimental chamber. The subject is asked to sample his urine in intervals of his own choice. The samples are stored in an ice box between the two lock-gates of the entrance from where they are removed for analysis. The subject can not perceive the time of removal. The records of several system of electric contacts, activated by the subject either consciously or unconsciously, serve to survey the subjects general activity. Communication between experimenter and subject is restricted to letters. Other details of the arrangements are given in two earlier publications[4,8].

RESULTS

A subject who is kept in isolation and without a time-telling device con-

·tinues to alternate regularily between states of wakefulness and of sleep. He also shows regular rhythms of body temperature and of urine excretion. An example is given in FIG. 1. As shown on the upper margin, the subject ·arises and goes to bed each day at a later time; the sleep times (black bars) ·drift steadily towards the right (midnight indicated by vertical lines). Con- ·current with each activity-time (wakefulness=white bars), there is a maximum ·of body temperature and a maximum of urine excretion. The average length ·of the circadian period for the activity cycle as well as for the vegetative functions is 24.9 hours. In other words: The exclusion of external Zeitgebers which normally synchronize the circadian oscillation to 24 hours[1], results in ·one free-running period of the whole circadian system. As another conse- ·quence, the normal phase-relationship between the vegetative functions and the activity cycle is slightly changed (see below). In general, however, the internal 'phase-map' as it has been called[3], is kept in order, even in the absence of the phase-setting effects of Zeitgebers.

Results like that presented in FIG. 1 seem to be in accordance with the assumption that the rhythms of body temperature and of urine excretion are mere consequences of the rhythm of wakefulness and sleep. However, such a causal relationship is contradicted by the results of experiments which show a dissociation between the activity cycle and the vegetative rhythms. As can be seen in FIG. 2, it may happen that a subject is awake for about 29.0 hours in each cycle and sleeps for about 21.0 hours. In contrast to this unusual
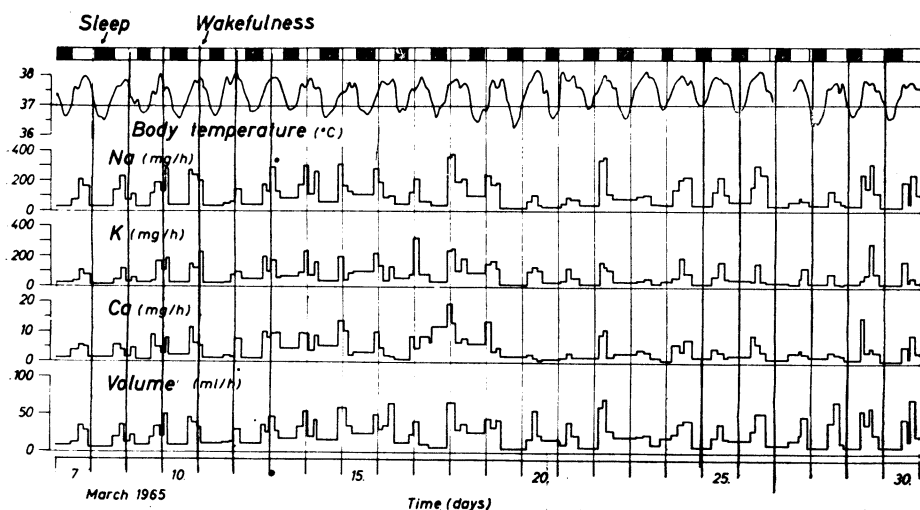
FIG. 1. Free-running circadian rhythms in a human subject, enclosed in an under- ground bunker without time cues for 24 days. The four curves from below give the urine excretion for water, calcium, potassium and sodium respectively. Vertical lines drawn at midnight.
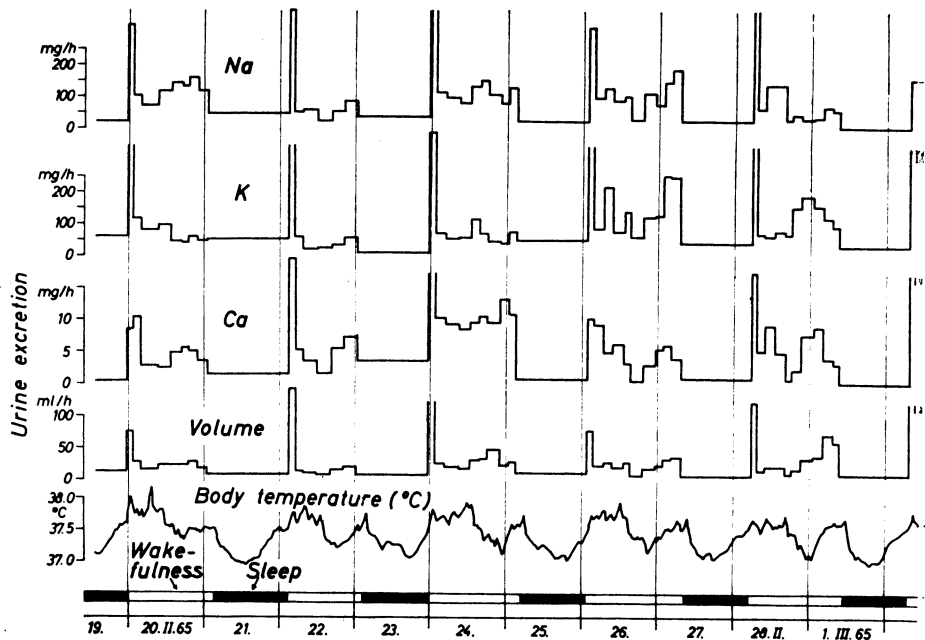
FIG. 2. Circadian rhythms for urine excretion (sodium, potassium, calcium and' water), rectal temperature and activity in a human subject, enclosed in isolation with-- out time cues.

activity cycle of 50 hours, the rhythms of body temperature and of urine ex-- cretion have 'normal' circadian periods of about 25 hours. There are two- peaks in the vegetative functions for each period of wakefulness and sleep. The two peaks differ in their extreme values, the higher one coinciding with.
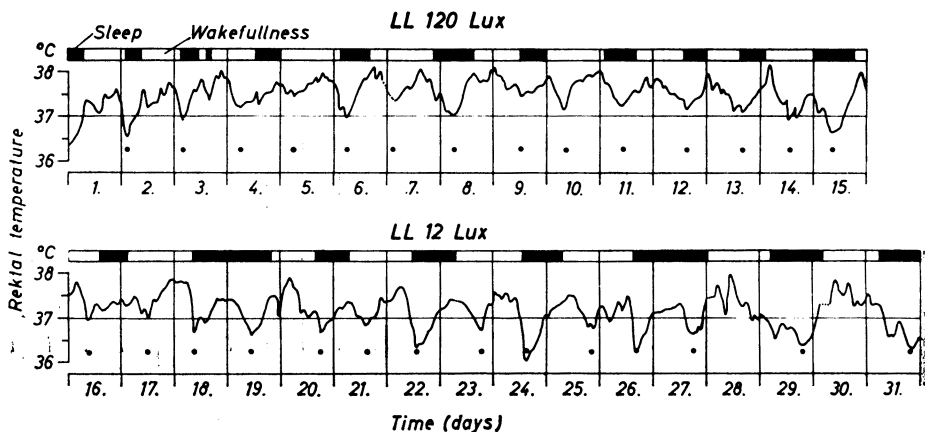
FIG. 3. Circadian rhythms of activity and of rectal temperature in subject B., enclosed in isolation at two different intensities of constant illumination. Minima. of body temperature as used for computations indicated by closed circles.

wakefulness, the lower one with sleep. This can easily be explained as a consequence of the interaction between the cycles of vegetative functions and the activity cycle which, in terms of a multi-oscillator system, are entrained to each other in a 2 : 1-ratio. The phenomenon can be considered as an example for 'synchronization by demultiplication', well known from experiments with animals entrained by Zeitgebers of a rather high frequency[9].

Of even greater interest are the results of two other experiments which demonstrate that the rhythms in activity and in body temperature can have
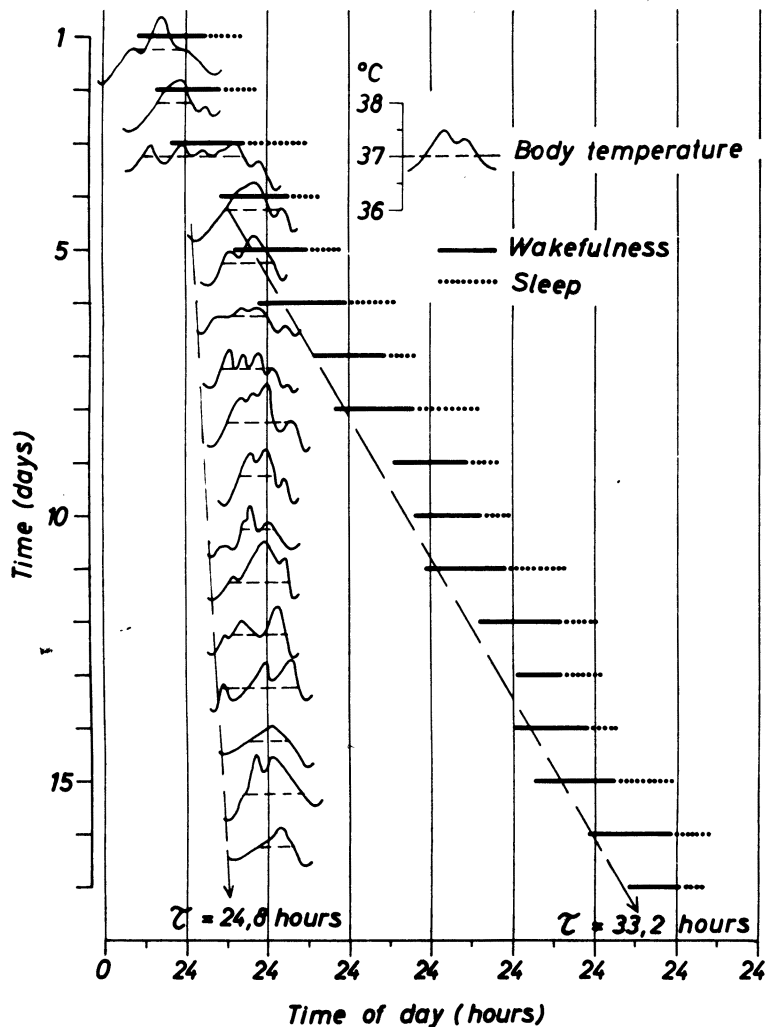
FIG. 4. Circadian rhythms of rectal temperature and of activity of subject F., enclosed in isolation without time cues. Consecutive periods are drawn underneath each other.

different, non-integral circadian periods. In the case of FIG. 3, the subject
shows 18 activity cycles only, as against 28 cycles of body temperature. Dur-
ing the first 15 days of confinement (with an intensity of illumination of 120
lux), the average circadian period for body temperature is 24.8 hours, the
period for wakefulness and sleep 34.4 hours. During the following 16 days
(at an illumination of 12 lux), the average period of body temperature (with-
out the last two cycles) is 24.7 hours and that of the activity cycle is 49.4
hours. In all but the two last cycles, body temperature and activity are free-
running with obviously different circadian frequencies. Results of a second
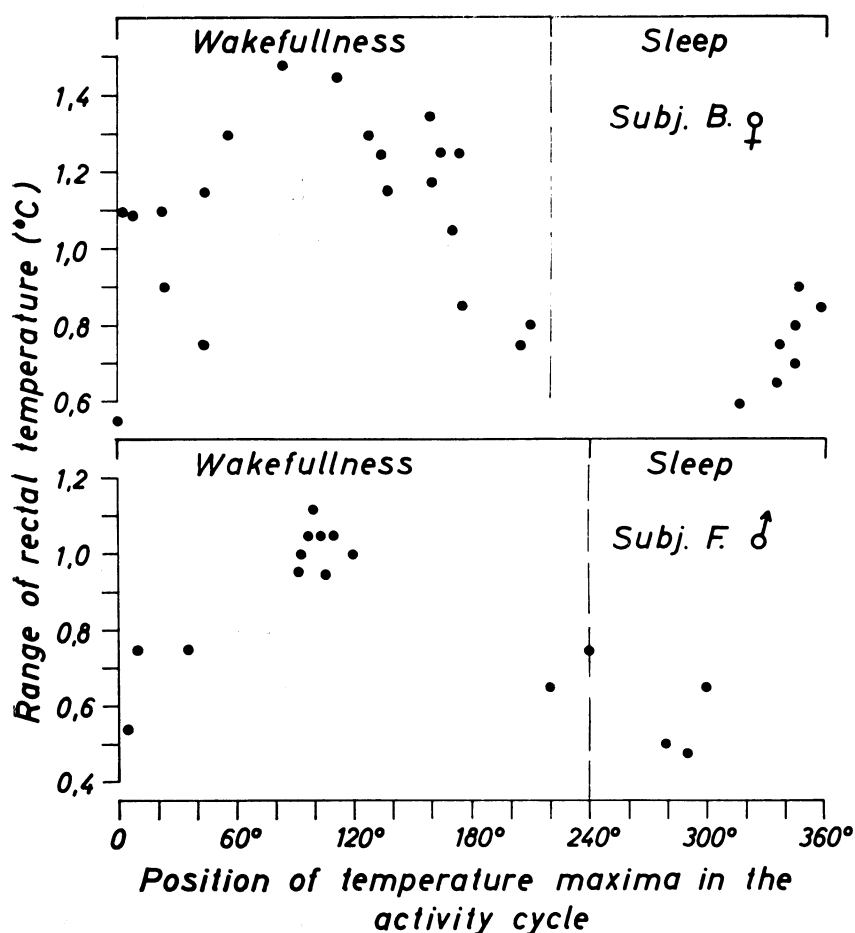
FIG. 5. Range of rectal temperature (measured from minimum to maximum in each
circadian period) as a function of the position of the temperature maximum in the
activity cycle. Computations from the data of the two experiments graphed in FIG. 3
and 4. The average circadian period of the activity cycle (given on the abscissa as
360°) was 42.5 hours for subject B. and 33.2 hours for subject F.

experiment are graphed in FIG. 4 in another way. The horizontal solid lines represent the times when the subject is out of bed; the dotted lines represent the following sleep times. For each cycle, the curve of body temperature is also shown. Both rhythms are in phase during the first two days of confinement and again on the fourth day. Thereafter, the rhythm of activity and rest starts to drift away with an average period of 33.2 hours while the rhythm of body temperature shows a more circadian-like period of 24.8 hours.

If the two rhythms of activity and of body temperature can each be considered as representing a separate oscillation, and if there is interaction between the two oscillators—as one has to assume (c. f. FIG. 2)—, one should expect a resonance-phenomenon. Since the two rhythms are free-running with different frequencies, they are crossing each other, sometimes reaching their normal phase-relationship, mostly being out of phase. In case of two coupled oscillators, such changes in the phase-relationship are reflected in changes of the range ('amplitude'=distance from minimum to maximum in a period) of at least one of the two oscillators. Applying this general law to the experiments shown in FIG. 3 and 4, one should expect a large range of the rhythm of body temperature when it has it's 'normal' phase-relationship to the activity cycle, and a small range when it is 180° out of phase. Computations from both sets of data are in agreement with this prediction (FIG. 5). In both experiments, the range of the rhythm of body temperature is largest when the maximum of temperature coincides roughly with subjective 'noon'; the range is smallest, when the maximum of temperature coincides with sleep.

From 50 subjects studied so far, two showed internal synchronized in a 1 : 2 ratio from the beginning, three others during the last experimental days. Two subjects showed desynchronization for most of the time of confinement. Seven more subjects were first internally synchronized and started desynchronization after 9 to 23 days of confinement. The remaining 36 subjects were internally synchronized for the whole experiment.

DISCUSSION

From these results, at least two conclusions can be drawn : 1) The rhythms of body temperature and of urine excretion are not mere consequences of the rhythms of wakefulness and sleep. They represent one or several separate self-sustained circadian oscillators. 2) Interaction between the rhythms of activity and the rhythms of vegetative functions results either in mutual synchronization (in a 1 : 1- or in a 1 : 2-ratio), or in relative coordination between rhythms which are free-running with different frequencies, as described by v. HOLST[12] for rhythms of fin movements. Relative coordination is characterized by two phenomena : a) The two rhythms cross each other with regular changing speeds[5,10]. That means: The two rhythms show a

tendency to adhere to some preferred phase-relationship. Therefore, in two graphs in FIG. 5 all possible phase-relationships are not equally represented (points missing from 230° to 310° and from 120° to 200° respectively). b) The range of the oscillation depends on the phase-relationship, reaching maximal values at the point of resonance between the two rhythms. It is of interest to note, that, in the two examples given in FIG. 5, resonance occurs at about 28° before subjective noon. This disagrees with the phase-relationship between body temperature and activity as it is observed under natural conditions. From measurements of many workers it is well known that, in the entrained organism, body temperature reaches its maximum towards the end of activity-time (late afternoon). However, this 'normal' phase-relationship is changed when the circadian rhythms are free-running. As can be seen in FIG. 1, the internally synchronized, free-running human circadian system is characterized by a maximum of body temperature which coincides with subjective 'noon' rather than with late afternoon, and by a minimum which coincides with the beginning of sleep rather than with its end. A detailed analysis of these differences between the internal phase-relationship of entrained and of free-running human circadian rhythms has been given by ASCHOFF and coworkers[7]. From this analysis it is clear that the point of resonance, as shown in the graphs of FIG. 5, is in full agreement with the typical phase-relationship between body temperature and activity in internally synchronized, but free-running circadian rhythms.

The two main findings—synchronization between the rhythms of activity and of body temperature in a 1:2-ratio and relative coordination between the two rhythms when they are free-running with different frequencies—are consistent with the hypothesis that both rhythms represent separate self-sustained circadian oscillations. There is no reason to doubt this interpretation in the case of body temperature. However, the sometimes unusually long periods of activity and rest are out of the range of circadian periods that have been observed so far, and that can be expected from a theoretical point of view. The question therefore arises whether this rhythm can be considered a circadian oscillator in the strict sense. It depends on the answer to this question to what an extent the results reported here truely support the hypothesis of a multi-oscillatory circadian system. These uncertainties do not weaken the strength and relevance of the conclusion that the rhythm of body temperature (like that of other vegetative functions) represents a circadian oscillator in itself which might or might not be synchronized with the activity cycle.

SUMMARY

Fifty human subjects have been kept in an underground bunker in conditions of continuous illumination, each in complete isolation and without any

time-telling device. Rectal temperature was recorded continuously by means of an electric thermometer, and urine samples, collected in intervals of the subjects own choice, were analyzed for excretion of water, sodium, calcium and potassium. Several systems of electric contacts served to survey the subjects general activities. All subjects showed free-running circadian rhythms, the average periods of wakefulness and sleep ranging from 23.9 to 50.0 hours. 36 subjects remained internally synchronized during the whole experiment. In 5 cases, the rhythm of activity and the rhythms of vegetative functions were synchronized in a 1:2-ratio for parts of the experiment. 9 subjects showed different circadian frequencies in activity and in body temperature; in two of these subjects, desynchronization started immediately after being enclosed in the bunker, in the remaining 7 subjects after 9 to 23 days of confinement.

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