

CIRCADIAN RHYTHMS DURING AND AFTER THREE MONTHS IN SOLITUDE UNDERGROUND

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The existence and persistence of the habitual circadian (Halberg, Halberg, Barnum & Bittner, 1959) rhythms of renal function have been studied in Eskimos and other Arctic dwellers who are not subjected to the usual alternation of night and day, as well as in inhabitants of temperate latitudes spending the summer in the Arctic and living on abnormal time schedules (Lobban, 1960). When it was learnt that Mr G. Workman intended to spend 100 days in solitude in a pot-hole, the opportunity was taken to find out how his renal rhythm would respond to this complete isolation from any environmental rhythm, and almost complete isolation from contact with other human beings.

METHODS

Workman entered Stump Cross Cavern, near Pateley Bridge, on 16 June, 1963 and remained there until 29 September. His intention was to follow normal habits of sleep and waking, meals, activity and so on. He used a watch reading British Summer Time (B.S.T.), and had a field telephone with which he spoke to the owner of the cave, at the surface, once a day; apart from this, he had no contact with human beings. The preparation of meals, and other necessary chores involved in living alone, occupied a fair proportion of his time, and he was also actively engaged in further exploration of the cave system and clearing a blocked passage. No special effort was made to adhere to a uniform diet, but since all his food was taken down with him, his diet was somewhat monotonous; as assessed from 24 hr electrolyte excretions, it supplied potassium, 22–54 m-equiv, mean 37, and chloride, 80–250 m-equiv, mean 158; sodium was similar to chloride. The low intake of potassium resulted in an unusually high Na:K ratio in the urine. The temperature was constant, 7° C, and the atmosphere nearly saturated with water vapour. Light was provided mainly by candles, or by a miner's forehead lamp when he was exploring, but he used a Tilley lamp for short periods. Once a week, while he was underground, all the urine produced during two sleep periods and the intervening 'day' was collected, by spontaneous voiding at times convenient to the subject. When sleep was broken by the need to pass urine, the two samples were collected and analysed separately. On these days of urine collection he was usually, but not always, physically idle. The urine volume was read to the nearest 10, or occasionally 5 ml., and the time to the nearest 5 min, and a portion of each sample was preserved; these portions were left at a pre-arranged place for collection and despatch to Manchester, where they were analysed for chloride (Sanderson, 1952), sodium and potassium (flame photometer), phosphate (Fiske & Subbarow, 1925) and creatinine (Bonsnes & Taussky, 1945). When samples were received reasonably fresh, pH was measured by a glass electrode, and if

this exceeded 7.0, bicarbonate was also measured (Van Slyke & Neill, 1924); if decomposition was suspected, ammonia was estimated by aeration-titration, and if pH and concentration of bicarbonate and ammonium were all high, it was presumed that urea was being hydrolysed. Similar collections and analysis of a day and two nights' urine was made on the day before he descended, and on 2 days some months after he emerged. Four blood samples were collected for steroid estimation (Mattingly, 1962) during a 24 hr period before descent.

When he emerged, at 13.30 hr on 29 September, after 105 days underground, he spent some time being interviewed and photographed by press and TV reporters, and then bathed, shaved, was psychiatrically examined, had a meal, spent a few hours with his wife and children, and was taken to the Metabolic Ward at Manchester Royal Infirmary, where he arrived about 19.30 hr and was put to bed. There he remained for 3½ days, with no visitors except nursing and medical staff, and received identical meals every 6 hr; judging by renal excretion, these supplied about 30 m-equiv of potassium, and 50–60 m-equiv of sodium and chloride, per 24 hr. Blood samples were collected 6-hourly for steroid estimation, and all urine voided was collected and analysed, time and volume being now recorded to the nearest minute and millilitre. Unfortunately, no blind nor dark curtains were available, so he was subjected, for the first time for 3 months, to the customary alternation of light and darkness, though this was minimized by leaving an electric light continuously alight. He was instructed to sleep when he felt inclined, and was not supplied with a watch.

RESULTS

Control days

Some of the findings on the day before descent, 15 to 16 June, and on the 2 further control days in December, are summarized in Table 1, and illustrated in Fig. 2. It will be seen that the electrolyte excretory peaks occurred rather later than in other reported series (Stanbury & Thomson,

TABLE 1. Electrolyte excretion on 3 controls days

	Potassium	Chloride	Sodium
Mean excretion and range (μ -equiv/min)	38 (18–64) 32 (18–73) 27 (19–33)	129 (59–287) 87 (42–232) 54 (29–95)	124 (43–248) 93 (31–288) 58 (23–108)
Time of maximal excretion and s.e. of estimate, hr	15.59 \pm 0.34 18.28 \pm 0.45 18.40 \pm 0.46	14.15 \pm 0.38 17.34 \pm 0.68 16.26 \pm 0.48	14.37 \pm 0.38 17.38 \pm 0.69 17.00 \pm 0.46
Time of maximal excretion, hr and min after waking	8.09 8.58 9.40	6.29 8.04 7.26	6.47 8.08 8.00

1951; Mills, Thomas & Yates, 1954; Lewis & Lobban, 1957*a*; Imrie, Mills & Williamson, 1963*a*), and not at a very constant time each day. The data were, however, rather more consistent if the peak times are calculated by reference to the time of waking. The amplitude of the rhythm of potassium, sodium and chloride excretion was rather variable, whether in absolute terms, or expressed as a proportion of the mean excretion. Phosphate excretion fell sharply on waking, and rose again 4–8 hr later, but thereafter followed no regular course. Creatinine excretion

followed no regular pattern during the day, but was always somewhat lower at night, the mean (geometric) day:night ratio being 1.13:1.

Plasma was collected for hydroxycorticosteroid determination on 14 to 15 June at 16.15, 22.00, 08.00 and 11.15 hr, and the concentrations found were 12, 5, 21.5 and 5 $\mu\text{g}/100$ ml. respectively. The subject was much distressed by the venepuncture at 08.00 hr, and nearly fainted; this may account for the high steroid concentration, which was not, however, associated with any irregularity in his electrolyte excretion.

Sleeping habits

Workman's intention to go to sleep underground at his normal time, around 23.00 hr, was frustrated by a progressive tendency to lie awake to a later hour every night, and then to sleep beyond his usual waking time of 08.00 hr. After thus attempting to adhere to normal habits for the first 3 weeks, he changed his plan and, from 5 July onwards, went to bed when he felt tired, which became later every night (Fig. 1). Thus, by 13 July he was falling asleep around 09.00 hr and waking again at 17.00 hr. By 30 July he was round to normal time again, and by the time he emerged he had 'lost' 2½ days.

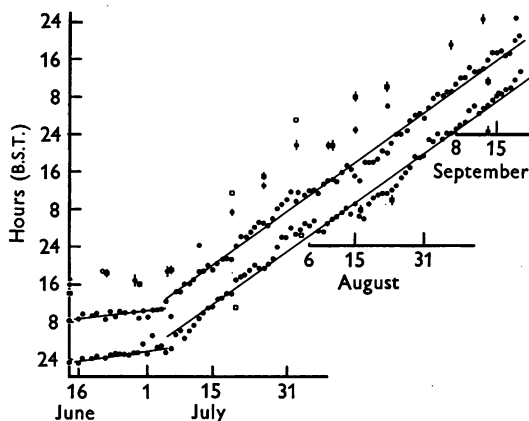


Fig. 1. ● Times of going to sleep and waking, with regression lines. ● Time of maximum K^+ excretion; ■ time of maximum Cl^- excretion, determined from sine curves fitted to excretory data, with s.e. where this is large enough to be shown. ○, □, Actual times of maximal K^+ and Cl^- excretion when there was a clear maximum but a sine curve could not be fitted. A few Cl^- points are shifted slightly to the right for clarity. The points on the ordinate represent the last day above-ground. Note that as ordinate extends over more than 24 hr, some points are represented twice.

The best estimate of the mean length of his 'day' is the slope of the regression line, of time of falling asleep and of waking, upon sequential

order. Such regression lines were fitted for the two periods underground: 16 June to 5 July, when he was attempting to sleep at the customary time, and the shift was slow, and 5 July to 22 September. These regression lines are shown in Fig. 1. The mean duration of the 'day' thus calculated from time of going to sleep was 24.17 ± 0.03 hr (s.e. of regression coefficient) from 16 June to 5 July, and 24.71 ± 0.01 hr from 5 July to 22 September. Similar calculations from times of waking give 24.12 ± 0.03 and 24.72 ± 0.01 hr, respectively.

Potassium, sodium, chloride, pH

The mean and range of excretion rates of chloride, sodium and potassium are shown in Table 2. These have been calculated over a 24 hr period from approximately the middle of one 'night' to the middle of the next. The full findings for the excretion of potassium and chloride on a day before descent, and on 16-17 August, are shown in Fig. 2. These show a reasonable approximation to a circadian rhythm. Lewis & Lobban (1956) point out that such data can best be described by a sine curve, thereby reducing to a minimum the number of parameters necessary to define this rhythm and ignoring random fluctuations, and providing an objective estimate of the time of maximum excretion. The method of Whittaker & Robinson (1926) has therefore been used approximately as described by Lewis & Lobban and the fitted curves are inserted in Fig. 2. Since the urine collection periods were not of 2 hr, the mean excretion rates over consecutive 2 hr collection periods had first to be calculated and this will result in some

TABLE 2. Range and mean of excretion rate (μ -equiv/min) during 24 hr collection periods underground

Date	Chloride		Sodium		Potassium	
	Range	Mean	Range	Mean	Range	Mean
22 June	53-370	142	44-374	138	14- 42	22
29 June	36-294	118	18-269	101	18- 69	35
6 July	55-328	154	49-437	168	17- 90	38
13 July	83-219	175	71-222	148	23- 82	42
20 July	126-418	199	105-548	198	25-112	54
27 July	63-196	111	61-211	115	21- 52	30
3 Aug.	106-338	170	104-302	168	17- 42	28
10 Aug.	67-184	101	61-137	97	16- 34	24
17 Aug.	32-229	82	19-272	85	9- 37	24
24 Aug.	162-333	210	114-336	150	21- 69	24
31 Aug.	101-240	145	72-223	135	36- 60	46
7 Sept.	168-415	248	112-432	231	19- 92	42
14 Sept.	139-260	194	126-257	200	23- 70	41
21 Sept.	131-336	190	91-335	185	17- 69	45
28 Sept.	82-341	148	79-344	148	25-152	47

further flattening of the computed curve beyond that which results from treating the mean excretion over 2 hr as if it were the instantaneous excretion rate at the mid-point of this period.

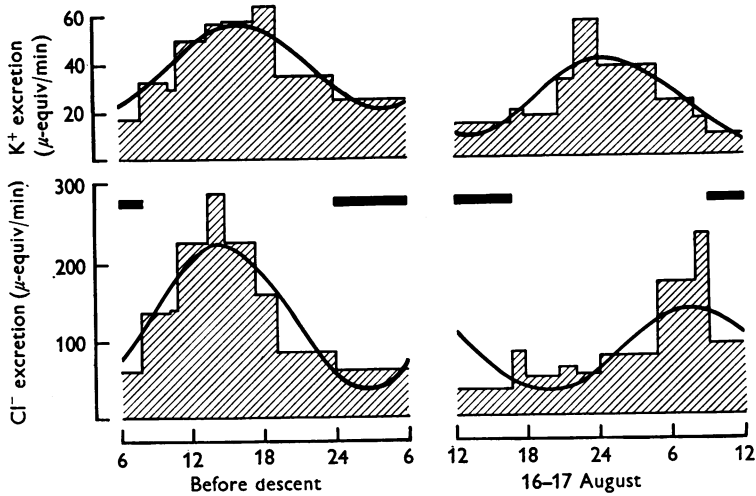


Fig. 2. Excretion of K^+ and Cl^- on 15-16 June, the day before descent, and on 16-17 August, with fitted sine curves. In this and subsequent figures solid rectangles indicate times of sleep.

The fit of a sine curve was very good for both potassium and chloride excretion on all 3 control days, as judged by an F test on the residual variance (P always below 0.01, and usually below 0.001). During the period underground, behaviour was not quite so regular, particularly for chloride, but when the F test gave a P value below 0.05 the time of maximal excretion derived from the sine curve is indicated in Fig. 1, with its s.e. On three occasions, two for potassium and one for chloride, when no sine curve could be fitted, excretion was high for a short period and low and uniform for most of the 24 hr so that a time of peak excretion could be assessed visually; these times are separately indicated in Fig. 1. On one occasion potassium excretion showed no circadian rhythm, and the same was true of chloride on five occasions.

The times of maximal potassium excretion became progressively later each week, as indicated in Fig. 1, in much the same way as did the sleeping habits. Electrolyte excretion was only measured weekly but if it be presumed that the drift was continuous throughout the week, the rhythmic excretion of potassium had a cycle length somewhat greater than 24 hr. Strictly speaking, therefore, the fitted sine curves should have a period slightly over 24 hr; there is, however, little difference between a fitted curve of 24 or 25 hr period, so no attempt has been made at such further precision.

For the first eight weeks underground, until 10 August, the timing of the curves of chloride and of potassium excretion were similar (Fig. 1), but

on 17 and 24 August (Figs. 2 and 3) the chloride excretory cycle was completely out of phase with that of potassium, with maximal excretion 7 and $4\frac{1}{2}$ hr later; thereafter, behaviour of the two electrolytes was completely dissociated, and chloride excretion only twice conformed to a sinusoidal pattern.

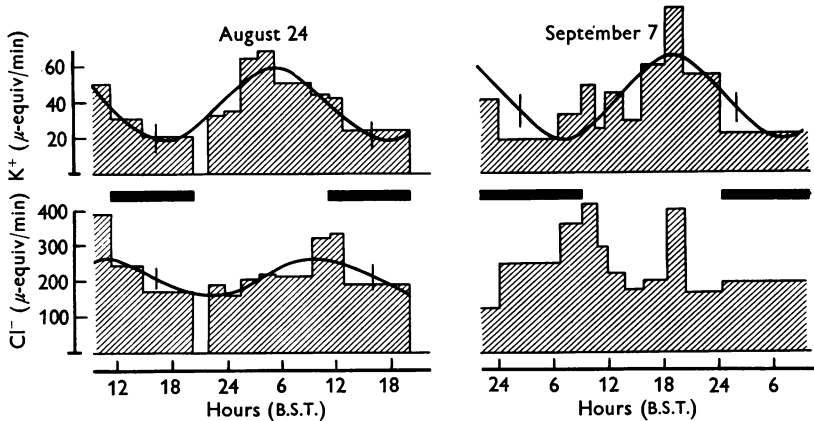


Fig. 3. Excretion of K^+ and Cl^- on 24 August and 7 September, with fitted sine curves. The vertical lines on the sine curve delimit the 24 hr period to which the curve was fitted.

Sodium and chloride excretory patterns were always closely similar to one another, and, though sine curves have been fitted to only some of the sodium excretory data, it is believed that the values derived for chloride apply to sodium also.

When electrolyte excretion followed a sinusoidal course in two successive collection periods, the mean length of the excretory cycle can be calculated by dividing the total shift in a week by the number of cycles presumed to have elapsed, usually seven but occasionally six, with results shown in Table 3.

TABLE 3. Mean length (hr) of the physiological 'day', as defined by the cycle length of the potassium and chloride excretory rhythms

Period	Potassium	Chloride
16-22 June	—	24.66 ± 0.15
22-29 June	—	23.66 ± 0.13
29 June-6 July	24.26 ± 0.23	24.44 ± 0.12
6-13 July	24.79 ± 0.17	—
13-20 July	25.18 ± 0.13	—
20-27 July	24.82 ± 0.12	—
27 July-Aug. 3	25.23 ± 0.18	—
3-10 Aug.	24.00 ± 0.19	—
10-17 Aug.	24.45 ± 0.11	25.47 ± 0.23
17-24 Aug.	24.69 ± 0.11	24.31 ± 0.20
7-14 Sept.	24.80 ± 0.19	—
14-21 Sept.	23.99 ± 0.15	—

Note. The dates are only approximate, since the periods of observation were not always centred around a calendar day.

Urinary pH was roughly correlated with rate of potassium excretion, with high values of each occurring in the same urine samples; sometimes, however, the highest pH was observed before the peak of potassium excretion. When excretion of sodium and potassium were grossly out of phase, pH was in phase with potassium rather than with sodium.

Urine flow

The rate of urine flow has been considered by others, e.g. Lewis, Lobban & Shaw (1956), as an independent component of diurnal excretory rhythms. The present experiments were not designed to study this aspect, since water intake was uncontrolled, but high rates of flow were usually associated with high excretion rates of sodium and chloride, and so were presumably osmotically determined.

During eight 'nights' when urine was collected, the subject was awakened prematurely by the need to pass urine. This was always due to a large flow associated with high rate of sodium and chloride excretion, and was liable to occur when sodium and chloride rhythm was out of phase with sleeping habits. The various techniques used by Lewis *et al.* (1956) to demonstrate inherent rhythms in urine flow have been applied to the present data, with wholly unconvincing results except that the incidence of periods with a flow in excess of 1.6 ml./min was lowest around midnight. Beyond an association between high flow and high chloride excretion, this is of doubtful significance.

Phosphate excretion

Excretion of phosphate was always low immediately after waking, irrespective of B.S.T. The mean drop from the rate during the previous period of sleep was 5.6 ± 1.1 μ -mole/min (s.e. of mean of fourteen observations, $P < 0.01$). This fall was probably proceeding during the later hours of sleep; for on these eight nights when the subject had to pass urine during the night, the phosphate excretion in the second sample was below that in the first by a mean of 4.8 ± 1.2 μ -mole/min (s.e. of mean, $P < 0.01$). This behaviour is also shown by plotting the incidence of low excretion rates, below 10 μ -mole/min (Fig. 4); these were only observed during the 6 hr after waking, except on one occasion when excretion was low continuously for 15 hr. By contrast, if the incidence of periods of low phosphate excretion is plotted against B.S.T., they are uniformly scattered throughout the 24 hr.

Excretion usually rose again steadily, reaching a peak 6–12 hr after the minimum (Fig. 5); this variable interval between minimum and maximum excretion precluded the fitting of sine curves of standard period. Phosphate

excretion sometimes behaved irregularly during the second half of the 'day', with a second minimum and maximum. The behaviour, considered in relation to sleeping habits, was the same underground as on the 3 control days.

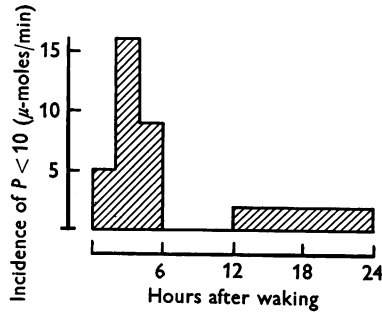


Fig. 4. Incidence, by 2-hourly periods, of hours in which mean excretion of phosphate was below 10 μ-mole/min.

During the final 3 days in hospital, phosphate excretion was unusually steady, almost always between 10 and 20 μ-mole/min, and did not show the usual relation to periods of waking and sleeping.

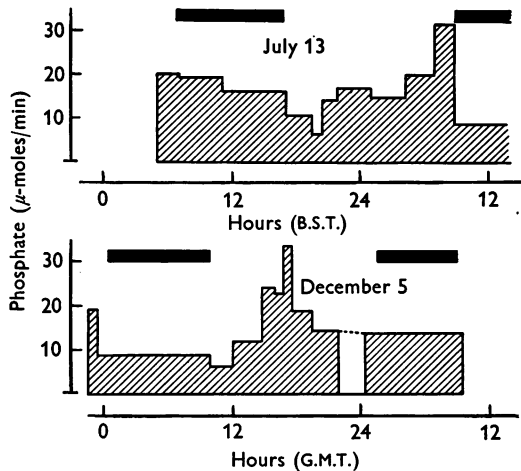


Fig. 5. Phosphate excretion on 13 July and 5 December.

Creatinine

Creatinine excretion varied somewhat irregularly, with most of the values (82%) between 0.8 and 1.2 mg/min. It was low during sleep periods (mean 0.83 mg/min) being on every occasion below the mean value of the preceding or following 'day' (mean 1.01 mg/min), with a (geometric) mean

day:night ratio of 1.22:1. This sometimes resembled the behaviour of sodium and chloride excretion. During waking hours there was, however, no close parallelism, and excretion of sodium and of creatinine often changed in opposite directions. Further evidence that any parallelism between sodium and creatinine excretion was fortuitous is that on three days (20 July, 17 and 24 August, Figs. 3 and 6) sodium and chloride rhythms were out of phase with sleep habits, showing a late peak, although creatinine excretion rose immediately on waking. On 2 more days (7 and 14 September, Fig. 3) sodium and chloride excretion was high during sleep while creatinine excretion was low.

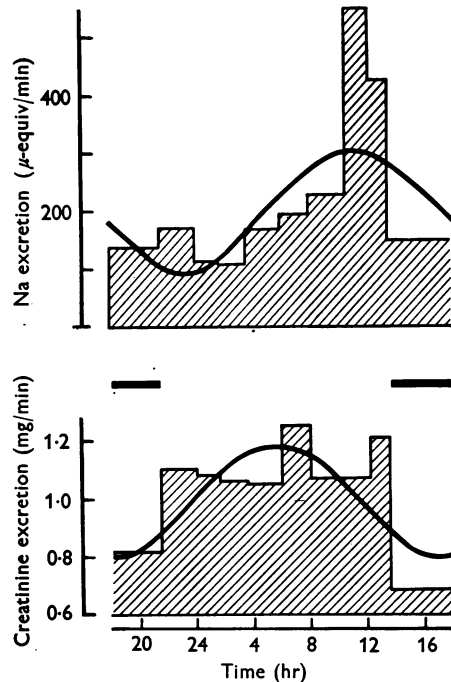


Fig. 6. Excretion of Na^+ (Cl^- behaved similarly) and of creatinine on 20 July, with fitted sine curves.

Recovery period

When the subject entered hospital, excretion of potassium, sodium and chloride were high, as was the plasma concentration of hydroxycorticosteroids, and the urine was alkaline with a pH of 7.3. A variety of stresses may well have contributed to these values. During the next 24 hr potassium excretion (Fig. 7) followed a sine curve with a maximum at $00.47 \text{ hr} \pm 1.24 \text{ hr}$, almost the exact reverse of the normal pattern. Sodium and chloride excretion were irregular, and no sine curve could be fitted. During

the next 36 hr the excretion of all three ions rose slowly and rather irregularly, but without any sign of a circadian rhythmic component. During the last 24 hr excretion again followed a sine curve (Figs. 7 and 8), but

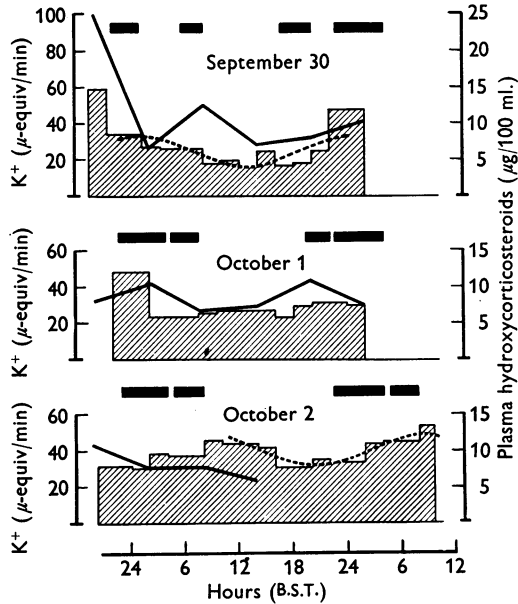


Fig. 7. Broken line: K^+ excretion, with fitted sine curve; full line: plasma concentration of 11-hydroxysteroids, and times of sleep, during $3\frac{1}{2}$ days in hospital after 3 months underground.

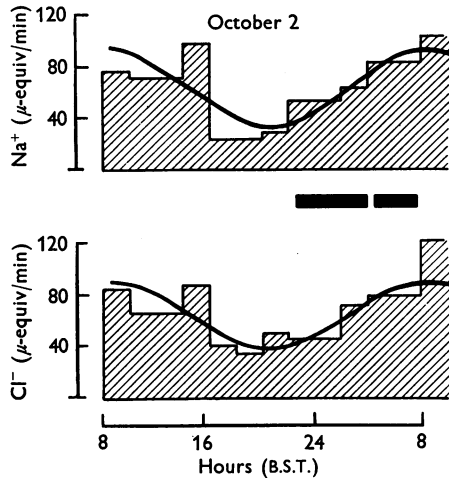


Fig. 8. Na^+ and Cl^- excretion during last 24 hr of $3\frac{1}{2}$ days in hospital, after 3 months underground, with fitted sine curves.

the phase had now reverted to nearly normal, with maximal excretions of potassium, sodium and chloride at 08.23 ± 0.54 , 08.32 ± 0.44 , and 08.42 ± 1.00 hr, respectively. pH followed the changes of potassium excretion during the first and last days. Excretion of creatinine and phosphate showed only irregular variations throughout the period in hospital, with no indication of either circadian rhythm or any relation to the somewhat irregular hours of sleep. Plasma concentration of hydrocorticosteroids (Fig. 7) fell rapidly from the initial high value and thereafter remained low; but unfortunately the last blood sample was collected before the excretory rhythm had resumed its normal pattern.

DISCUSSION

Rhythmically oscillating biological variables may be 'endogenous', independent of any rhythm in habit or environment, 'free-running' in the sense of Halberg *et al.* (1959); they may be 'exogenous', or immediately dependent upon an external rhythm; or they may result from an interaction between the two. Thus Halberg *et al.* (1959) found that the 24 hr temperature rhythm in blinded mice was disturbed in that the cycle length departed slightly but significantly from 24 hr, and the temperature was thus sometimes completely out of phase with solar time. Since no external rhythms are known with the same period, slightly above or below 24 hr, these authors assume the existence of an endogenous, or free-running, rhythm, whose phase in the intact animal is repeatedly checked and slightly adjusted by the rhythmic stimulus of external illumination. Light is thus referred to as the 'synchronizer', which is probably synonymous with Aschoff's (1955) 'Zeitgeber'. When Lewis & Lobban (1957*a, b*) observed a persistent rhythm of 24 hr cycle length in subjects living on a 21, 22 or 27 hr 'day', it is highly probable that they were observing an endogenous rhythm, though it is hard to exclude rigidly such trifling residual clues as the passage of the sun around the horizon (Aschoff, Discussion in Lobban, 1960). When Sharp (1960*a*) found that phase reversal of habits could reverse the phase of these usual rhythms within six days, and that further phase shift was achieved by remaining awake but in darkness (1960*b*), it is evidence that the 'synchronizer' can more readily adjust phase angle than cycle length, and that light is a major component of this synchronizer. Lobban's (1958) observations on Arctic dwellers do not enable one to discriminate between light, and social habits induced by the regular alternation of light and darkness, as major synchronizers in man, though Halberg *et al.* (1959) favour the latter in urbanized communities.

The observations most nearly comparable to the present are those of Siffre (1963), and of Aschoff & Wever (1962). Siffre spent 63 days at a

greater depth, and at subzero temperature; he had no time-piece, but telephoned to the surface his times of going to bed and waking, where observers recorded them. His time of waking behaved very much like Workman's, becoming slightly later every day, with a mean interval of 24.52 hr. His own attempts to estimate the passage of time were grossly erroneous, and in the diary in which he recorded his estimate of time many of his 'nights' of sleep appear as after-lunch naps. Aschoff & Wever (1962) confined their subjects for 8–19 days in solitude in a deep bunker without a time-piece, and instructed them to adopt regular habits of sleeping, eating, etc., whose times were recorded. Eight of nine subjects adopted a 'day' whose mean length, assessed from time of waking, exceeded 24 hr, with a range of 24.7–25.8 hr. Body temperature, and urinary volume and excretion of electrolytes and corticoids, also varied rhythmically with a cycle slightly above 24 hr; but the published data do not permit any assessment of the association or dissociation of the different components of these circadian rhythms. It is remarkable that the sleep cycle of Workman, who was well aware of the time, of the subjects of Aschoff & Wever (1962), and of Siffre (1963), who was unaware of the time and made such erroneous estimates, were of similar duration. All clearly displayed a free-running rhythm.

The connexion between the different components of the rhythm now needs consideration. When an endogenous and an exogenous rhythm appear to be competing, two different components—such as potassium and chloride excretion in the experiments of Lewis & Lobban (1957*b*)—may become dissociated, one being more heavily affected by the exogenous, the other by the endogenous rhythm. When several components maintain their normal synchronization, as was the case with potassium excretion and sleeping habits throughout much of the period underground here reported, a common cause is probable.

The possibility must, however, be considered that the abnormal renal rhythm was a simple consequence of the abnormal sleeping habits, even though sleep does not of itself diminish potassium excretion. If these habits were the cause of the renal behaviour, the circumstances would be formally similar to those of Lewis & Lobban (1957*a*) in which subjects were becoming artificially 'adapted' to a cycle of unusual length, except that the 24½ hr day adopted by Workman departed less from the solar cycle than did the 21, 22 and 27 hr days of Lewis & Lobban, and it therefore took several weeks instead of days to become out of phase with solar time. The techniques of presentation employed by these workers have been applied to the present findings, with entirely different results. Figure 6 of Lewis & Lobban (1956) and Figs. 6 and 7 of Lewis & Lobban (1957*a*) are similar presentations to the electrolyte data in Fig. 1 of the present paper;

but whereas all the subjects of Lewis & Lobban showed some influence of 24 hr time, this was not apparent in the present subject. Figure 7 of Lewis & Lobban (1956) and Fig. 8 of Lewis & Lobban (1957*a*) show that the relative amplitude of the sine curve fitted to the excretion of sodium and of potassium was less when experimental time was out of phase with B.S.T., suggesting an interaction between a persistent cyclical influence with a 24 hr period, and the abnormal period of the subjects' activities. Nothing of the sort was seen in the present investigation.

Workman is himself certain that his unorthodox sleeping habits were not due to any discomfort. He states that he was very comfortable; that he sleeps soundly under much less comfortable conditions; and that his only difficulty was that he did not feel sleepy. Moreover, numerous observations, published (e.g. Kleitman, 1963; Mills, 1951) and unpublished, indicate that an inherent rhythm of sleepiness and wakefulness is more important in determining the hours of sleep than are such factors as fatigue or discomfort. Moreover, high and low potassium excretion do not cause wakefulness and sleepiness. Thus, the approximate synchrony between the sleep cycle and potassium excretion, which was usually maximal 6–9 hr after waking, suggests a common cause for both rhythms: that some intrinsically rhythmic process, a biological 'clock', governed alike the rhythmicity in sleep and in potassium excretion, and that this 'clock' was not keeping perfect 24 hr time. The close association, over many weeks, of chloride and potassium excretion suggests that the 'clock' governed chloride excretion also; but the subsequent drift of the chloride rhythm suggests that it was determined eventually by some other physiological process. The coincidence of the two chloride excretory peaks on 7 September (Fig. 3) with a minor and the major potassium peaks suggests that the 'clock' affecting potassium excretion has some effect also on chloride, and vice versa. These two peaks occurred at times of fairly constant creatinine excretion, so are unlikely to be artifacts due to errors in recording time or volume of urine samples.

Through what intermediation the 'clock' or 'clocks' affect the kidney is still largely unknown. Halberg *et al.* (1959) suggest the secretion of cortisol, or corticosterone, as an effector mechanism controlling a large number of such processes with a circadian rhythm. Imrie, Mills & Williamson (1963*a, b*) have produced evidence that adrenal secretion is of great but not exclusive importance, and the latter conclusion is borne out by the present results. The obvious irregularity of adrenal activity shown on 15 June did not prevent a simple sinusoidal rhythm in electrolyte excretion; Bayliss (1955) mentions a similar persistence of normal rhythm in electrolyte excretion, despite a rise of plasma steroid concentration owing to apprehension. Conversely, a sinusoidal rhythm—though in a

different phase—was seen on the first day after emerging, despite a virtually constant plasma steroid concentration. No rhythmic influence on the kidney which can adequately account for the rhythmic renal behaviour has been demonstrated.

Phosphate excretion maintained the normal relation (Kleitman, 1925) to waking and sleeping, but this is hardly remarkable since it adjusts itself so readily (Mills & Thomas, 1957) to altered habits of living.

SUMMARY

1. A record of times of sleep of a man spending 105 days in solitude underground showed that he fell asleep and woke a little later day by day, following a sleep-waking cycle of roughly $24\frac{1}{2}$ hr.

2. Collection and analysis each week of 36 hr urine in a series of samples showed that potassium excretion followed a similar rhythm.

3. For eight weeks, excretion of sodium and chloride followed a rhythm similar to that of potassium, but thereafter they became dissociated from it, and increasingly irregular.

4. Creatinine excretion was always low during sleep.

5. Phosphate excretion always fell about the time of waking.

6. It is maintained that sleep-wakefulness, potassium excretion, and for a time chloride excretion, followed a free-running rhythm with cycle length slightly over 24 hr.

7. On emerging, the subject spent $3\frac{1}{2}$ days in bed in hospital; during the first 24 hr his potassium excretion followed a circadian rhythm with maximum about 01.00 hr, whereas during the last 24 hr excretion of potassium, sodium and chloride had returned approximately to normal phase relations.

My special thanks are due to Mr Geoffrey Workman for permitting me to study him and for submitting to further inconvenience to this end. I also wish to thank Mr G. Gill for sending me the urine samples, Dr S. W. Stanbury for arranging the admission to Manchester Royal Infirmary, Mr M. Bell for plasma steroid analyses, Dr H. Kidd for collecting blood samples before descent, and E. Hesford and J. Collins for technical assistance.

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