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The Effect of Various Photoperiods on Daily Oscillations of Serum Corticosterone and Insulin in Rats

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Summary

The effect of various photoperiods on circadian rhythms of chosen parameters was investigated in laboratory rats. SPF male Wistar rats were adapted for six weeks to artificial light-dark cycles (LD 8 : 16, 12 : 12, 16 : 8). The light was switched on at 07.00 h in all regimens. The rats were killed at 3-hour intervals within 24 h, the serum concentration of corticosterone, insulin, glucose, food and water intake was determined. The external and computative acrophases of corticosterone varied in every photoperiod being dependent on the duration of light, the mesor values decreased in LD 16 : 8 in comparison with other photoperiods. The external acrophase of insulin was located 4 h after light onset in LD 8 : 16 and 12 : 12, in LD 16 : 8 one hour before light onset. The mesor values were approximately equal in all photoperiods. The circadian rhythms of glucose were similar in all regimens. Circadian variation of food and water consumption culminated at the same time in all regimens, the amount of food consumed in light increased with the light duration. Various photoperiods remarkably influenced circadian oscillations of corticosterone and in part food and water intake which could be considered as photoperiodic traits.

Key words

Serum corticosterone – Insulin and food intake – Circadian oscillations – Photoperiods – Rats

Introduction

The circadian oscillations of serum corticosterone and insulin concentrations in laboratory rats were found to be influenced by the seasons (Ahlers *et al.* 1980, Ahlersová *et al.* 1984, 1989). The present experiment was designed to examine the effects of various photoperiods simulating light conditions in the course of the year using artificial light on circadian rhythms of the given hormones.

Material and Methods

Adult male SPF Wistar rats aged three months were adapted simultaneously to three arteficial light - dark (LD 8 : 16,12 : 12, 16 : 8) regimens for six weeks under standard conditions (temperature 22±2 °C, relative humidity 60-70 %). Cool light (fluorescent lamps Tesla, 40 W) of 150 lux intensity in each cage was automatically switched on at 07.00 h in all regimens, and switched off at 15.00 h (LD 8 : 16), 19.00 h (LD 12 : 12) and 23.00 h (LD 16 : 8). The animals had free access to food (LD pellets, Velaz, Praha) and water.After adaptation to these conditions the rats were quickly decapitated at 3 h intervals within 24 h (dimmed red light of less than 1 lux intensity was employed in the dark) in December. The corticosterone, insulin and glucose concentrations were estimated in mixed blood serum, stored at -20 °C until used : corticosterone was determined fluorimetrically according to Guillemin *et al.* (1958), insulin by radioimmunoassay using commercial kits (Radioisotope Centre, Swierk, Poland), glucose was determined enzymatically (Hugget and Nixon, 1957). Each group consisted of 8 rats. The results were evaluated by the cosinor test (Halberg *et al.* 1967), the mesor values were compared by the t-test. One week before the rats were killed, food and water consumption was measured at 3 h intervals throughout the 24 h.

Results

To study the effects of various photoperiods on circadian oscillations of the chosen parameters we used two acrophase patterns : the computative acrophase given as relation of the peak to 0.00 h of local time (the values are given in the table of cosinor

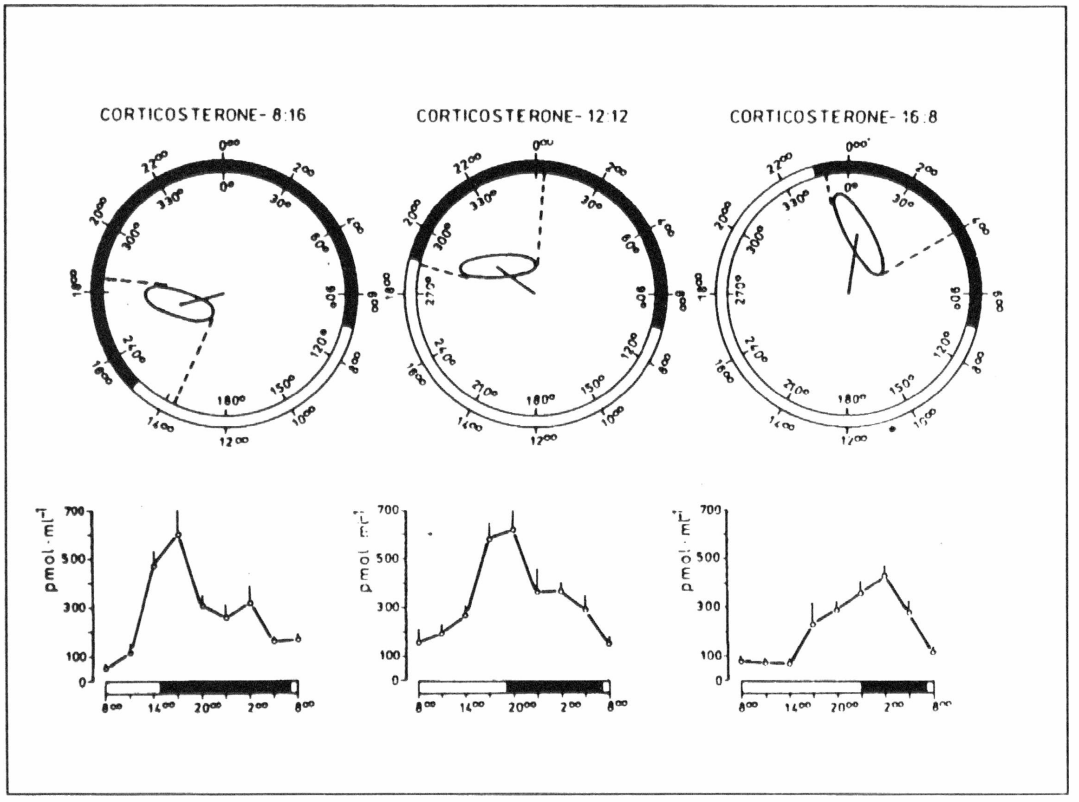

Fig. 1

Circadian oscillations (below) and cosinor diagram (above) of the serum corticosterone concentration in the rats from various photoperiods: light-dark (LD) 8:16, 12:12, 16:8. The values are given as the means ± S.E.M. In the cosinor diagram the vectors originating from the centre of the circular system of coordinates represent amplitudes of oscillations; their position denotes the acrophase. The rhythm is present if the ellipse of the errors does not overlap the origin of the coordinates. The tangents to the ellipses represent the 95 % confidence interval for the acrophase. Light part of time data - light, dark part - darkness.

analysis) and the external acrophase which is related to a definite point on the external synchronizing cycle, in our experiment to the switching on of light.

The circadian oscillations of the corticosterone concentration culminated in all photoperiods depending on the duration of light: the computative acrophases were located at 17.00 h in LD 8 : 16, at 22.00 h in LD 12 : 12 and 00.32 h in LD 16 : 8. A similar phase shift was recorded in external acrophases: 10 h and 13 h after switching on (LD 8 : 16, LD 12 : 12), and 6 h before switching on (LD 16 : 8). The mesor value in LD 16 : 8 was lower than that of shorter photoperiods namely in comparison with LD 12 : 12 (Figs. 1, 4 and Tab. 1).

The computative acrophases of insulin circadian oscillations were between 10.00 h and 11.00 h in LD 8 : 16 and 12 : 12 and 05.44 h in LD 16 : 8. The external acrophases were located 4 h after light onset (LD 8 : 16, LD 12 : 12) and 1 h before light onset (LD 16 : 8). The mesor values were approximately the same in all photoperiods (Figs. 2, 4 and Tab. 1).

The circadian variation of the serum glucose concentration had two peaks, the external acrophases were located 1-2 h after light onset (between 08.00–09.00 h – computative acrophases) in all light regimens,the mesor values were similar (Figs. 3, 4 and Tab. 1).

The circadian oscillations of serum corticosterone, insulin and glucose were rhythmic (the 24 h period of rhythm was chosen) in each photoperiod.

The oscillations of food and water intake culminated in all regimens at 23.00 h with the second lower peak at 05.00 h. The amount of food consumed in light part of the day had been increased with the light length from 10 % in LD 8 : 16, to 22 % in LD 12 : 12 and to 50 % in LD 16 : 8. The mean daily value of consumed food and water was the highest in LD 8 : 16, the lowest in LD 12 : 12 (Figs. 5, 6 and Tab. 1).

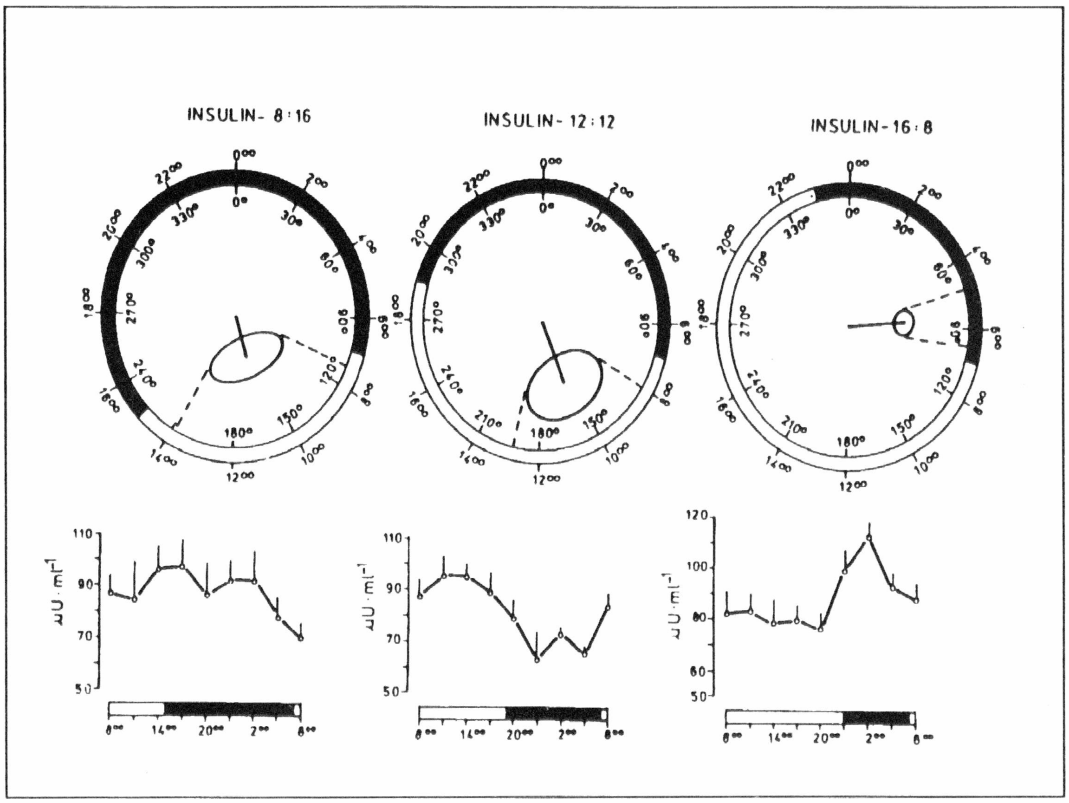

Fig. 2
Circadian oscillations and cosinor diagram of the serum insulin concentration in the rats from various photoperiods. Other details as in Fig. 1.

Discussion

Various photoperiods did not substantially modify the circadian oscillations of serum glucose in the present work. However, the long days (LD 16 : 8) shifted both acrophases of insulin daily oscillations, the external and the computative ones, in comparison with other photoperiods.The relative stability of the circadian oscillations of blood glucose in various photoperiods was recorded in laboratory rats (Pauly and Scheving 1967, Ahlersová *et al.* 1982). De Souza and Meier (1987) demonstrated the stability of daily rhythms of serum glucose, insulin and corticosterone in scotoresistant Syrian hamsters (*Mesocricetus auratus*) with the change of the long photoperiod (LD 14 : 10) to the shorter one (LD 10 : 14).Inversion of the light regimen from LD to DL in laboratory mice did not influence the acrophase localisation of plasma insulin oscillations but reduced the mesor values in comparison with the LD regimen (Weinert *et al.*1986).

Circadian oscillation of serum insulin concentration in rats exposed to a short photoperiod of artificial (LD 8 : 16) and natural (LD approx. 8 : 16) light (Ahlersová *et al.* in press) in December have the similar acrophase but different amplitudes and mesors.

The circadian properties of serum corticosterone (CS) oscillations namely the acrophases and mesors were markedly influenced by photoperiods in the present experiment. Scheving and Pauly (1966) have studied the dependence of serum CS oscillations in Sprague-Dawley rats on various light regimens. Obvious pattern of CS oscillations in LD 12 : 12 was changed in continuous light (LL) or darkness (DD) to a free running rhythm with lower amplitudes, high mean daily values in LL and low in DD. The response of the hypothalamic-hypophyseal-adrenal axis to different light regimens in Sprague-Dawley rats was analysed by Fischman *et al.* (1988). Synchronous oscillations of corticotropin-releasing factor-like immunoreactivity (CRF-LI) in hypothalamus and of adreno-corticotrophin (ACTH) and corticosterone in the plasma were noted in LD 12 : 12. Continuous darkness produced the changes in CRF-LI and ACTH concentrations and shifted the peaks of all three parameters. The desynchronization between rhythms of plasma ACTH and CS was found in continuous light without changes in synchronization in CRF-LI and ACTH rhythms.

Comparing the circadian oscillations of serum CS concentration in rats exposed to a short

Table 1
Characteristics of cosinor test: the mesor (mean value of the fitted curve) and amplitude are given in the employed units; the acrophase is given in an angular and a time interpretation
CI – confidence interval; its limits in the presence of rhzthm are given in brackets

Serum	Photo-periods (LD = light:dark)	Rhythm detec-tion (95% level)	Mesor ± S.E.M.	Amplitude ± CI (95%)	Acrophase ± CI (95%)	
					Degrees/°	Hours/minutes
Cortico-sterone pmol/ml	LD 8:16	+	269.50 ± 31.14	120.25(56.97;183.53)	255(205;218)	$17^{00}(13^{40};18^{32})$
	12:12	+	300.97 ± 30.83	127.89(91.39;164.39)	304(284;5)	$20^{16}(18^{56};0^{20})$
	16:8	+	197.86 ± 37.83	154.05(110.92;197.18)	8(349;60)	$0^{32}(23^{16};4^{00})$
Insulin μU/ml	LD 8:16	+	83.23 ± 3.79	16.02(8.52;23.52)	164(110;20)	$10^{56}(7^{20};13^{56})$
	12:12	+	83.52 ± 5.79	24.54(14.14;34.94)	158(120;193)	$10^{32}(8^{00};12^{52})$
	16:8	+	84.99 ± 5.81	22.66(18.89;26.43)	86(73;100)	$5^{44}(4^{52};6^{40})$
Glucose mmol/l	LD 8:16	+	7.91 ± 0.37	1.55(1.39;1.71)	140(98;18)	$9^{20}(6^{32};12^{00})$
	12:12	+	8.07 ± 0.51	2.11(1.95;2.27)	120(83;140)	$8^{00}(5^{32};9^{40})$
	16:8	+	8.02 ± 0.43	1.77(1.5;2.09)	117(95;136)	$7^{48}(6^{20};9^{04})$

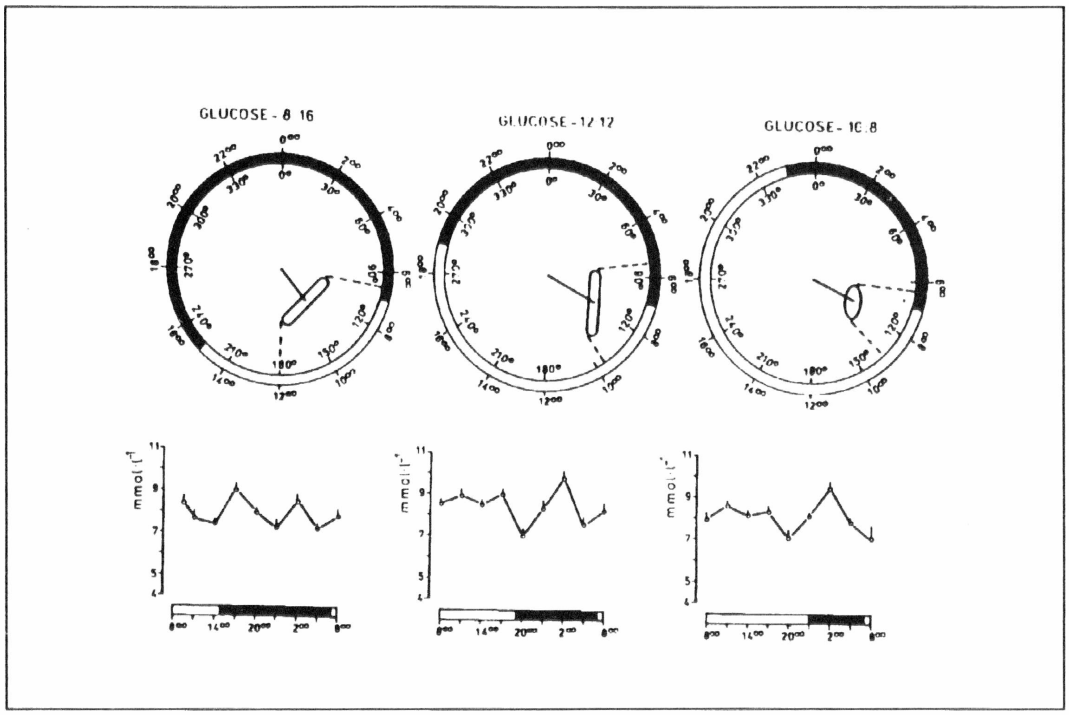

Fig. 3
Circadian oscillations and cosinor diagram of the serum glucose concentration in the rats from various photoperiodics. Other details as in Fig. 1.

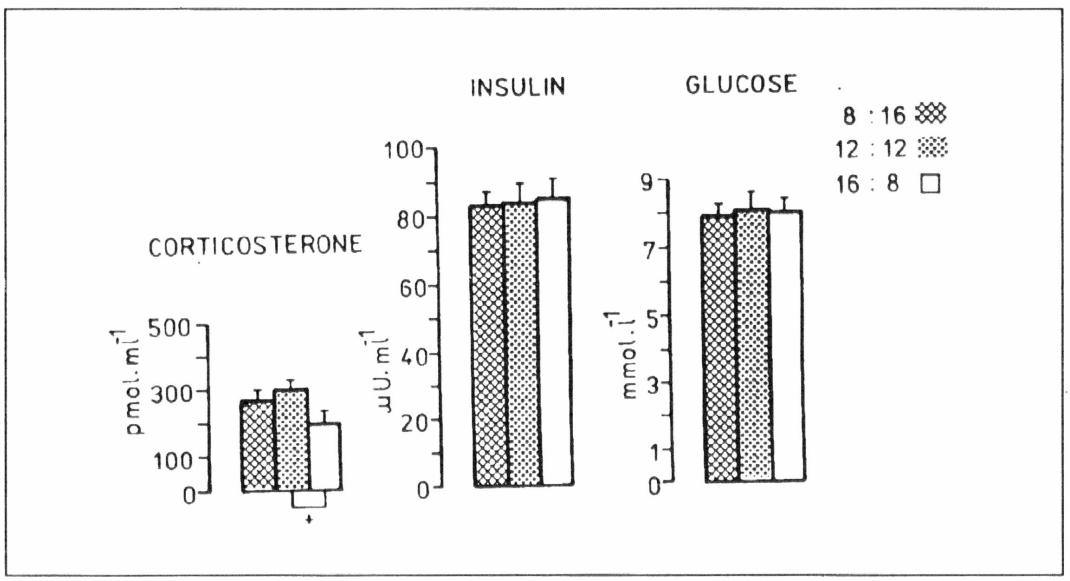

Fig. 4
The mesors – mean daily values of fitted curves of the serum corticosterone, insulin and glucose concentration (mean ± S.E.M.) in the various photoperiods. Significance of mesor value differences: + for $P < 0.05$.

photoperiod of artificial (LD 8 : 16) and natural (LD approx. 8 : 16) light (Ahlersová *et al.* in press) in December, the same acrophase, but different shapes of curves with different amplitudes and mesors were

detected. In both the CS and insulin, this may result from different light intensity (150 lux vs below 150 lux

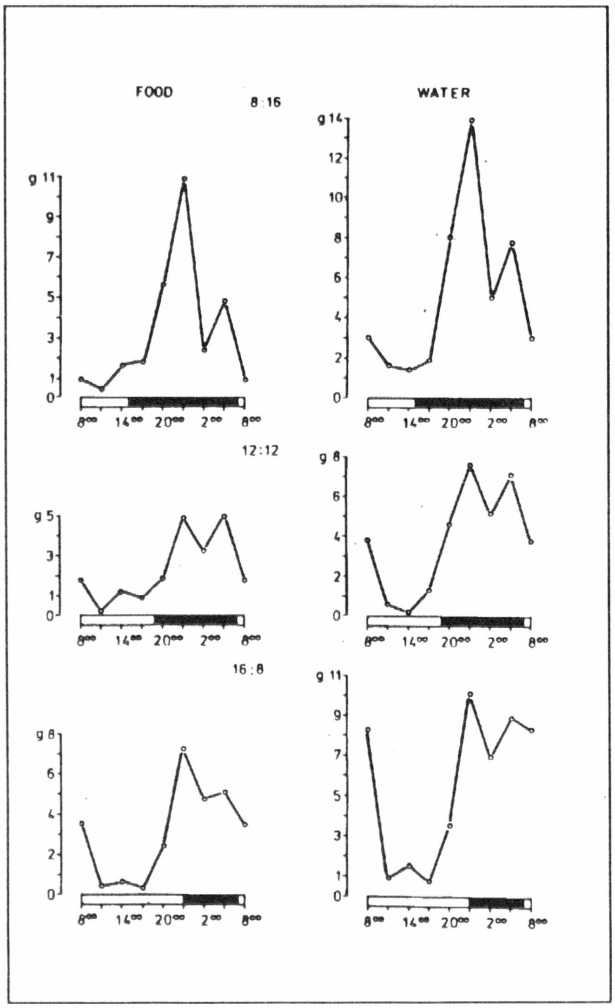

Fig. 5
Circadian oscillations of food and water intake in the rats from various photoperiods. Other details as in Fig. 1.

in artificial vs natural light) or from the rate of light and dark onset (switching on, off vs dawn, dusk) and different spectral composition of both kinds of the light. The shifts of external acrophases (greater than of computative one) were recorded in the circadian oscillations of serum CS, insulin and glucose in the rats kept in the conditions of natural light during the year. The mesors of all parameters were higher in summer compared to the other seasons (Ahlersová *et al.* 1989).

The light as a dominant synchronizer of biological rhythms plays an important role in the formation of circadian rhythm in food intake of laboratory animals. Exposure of rats kept on LD 12 : 12 to constant light or darkness led after some time to the loss of circadian rhythms in food and water intake. In rats sixty days after blinding the circadian rhythms of food and water intake disappeared. Inversion from LD to DL (12 : 12) caused inversion of food and water intake rhythm (Zucker 1971). The circadian rhythm of food and water intake in our work was dependent in some way on used light regimen: the magnitude of food consumed in the daylight increased by lengtheninng of the photoperiod from 10 % in LD 8 : 16 to 22 % in LD 12 : 12 and to 50 % in LD 16 : 8. In the short day there was an increase in mean food and water intake compared with other light regimens. These data agree with the results of Ten Hoor *et al.* (1980).These authors have described the onset of food intake activity in rats at the beginning of darkness in the short photoperiod; by lengthening of the photoperiod the onset was shifted to the end of the light and in long photoperiods the fundamental part of food intake activity was situated in the light.

According to our opinion the clear modification of circadian rhythms in serum corticosterone and partially in food and water intake by various photoperiods in the laboratory rat belongs to the photoperiodic traits. Persisting of these traits in laboratory conditions documents the genetic fixation of evolutionary adaptative mechanisms to the changes of environmental factor, such as light and food availability.

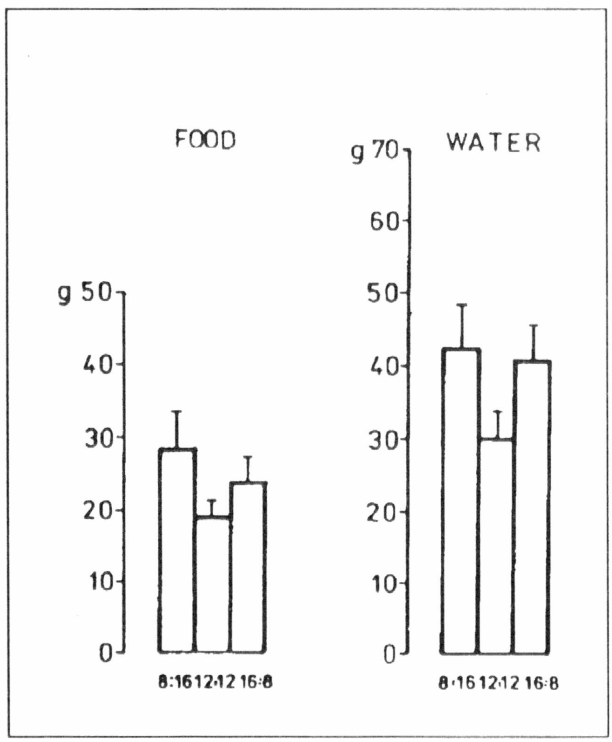

Fig. 6
The mean daily values of food and water consumption (mean ± S.E.M.) in the rats from various photoperiods.

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