cervical luxation, under ether anesthesia. Samples of the ascending colon were removed, weighed, fixed in Clarke's solution to remove unincorporated thymidine and the samples counted employing standard liquid scintillation techniques. The resultant data was expressed as disintegrations per min per mg of tissue (dpm/mg). This technique is based on the observation that over 95% of the cells which incorporate ^{3}H-TdR in detectable amounts are the S-phase proliferative cells of the intestinal mucosa[14]. The dpm/mg parameter thus provides a reliable estimate of S-phase cellularity. Individual serum calcium levels were determined from samples taken via the jugular vein immediately prior to sacrifice, employing standard atomic absorption procedures.

Results and discussion. The colonic response to fasting and refeeding in rats begins within 12 h after refeeding, reaching a maximum between 24 and 48 h and regaining control levels by 72 h. This contrasts with the response in the mouse[3], where maximal response is observed between 12 and 24 h with a duration of 36 h. Additionally, the rat exhibits no significant depression in thymidine incorporation per mg tissue after fasting, as is the case with mice[3].

Total serum calcium content in sham-operated and parathyroidectomized (PTX) rats during dietary manipulation (mg%) ± 1 SE.

	Sham	PTX
Control	10.09 ± 0.18%	4.06 ± 0.23%
120 h fast	9.37 ± 0.46%	4.23 ± 0.25%
24 h refeed	9.75 ± 0.05%	4.17 ± 0.07%
48 h refeed	10.35 ± 0.09%	4.44 ± 0.22%
72 h refeed	9.80 ± 0.10%	4.14 ± 0.18%

The parathyroidectomy procedure employed here significantly reduced the serum calcium levels (table), while the sham-operated animals showed no difference from control. Animal survival in PTX-animals exceeded 80% following fasting. Parathyroidectomy did not in itself alter colonic cell proliferation when compared to sham-operated control mice.

Parathyroidectomized rats exhibited a refeeding response equivalent in both duration and magnitude to that seen in sham-operated and control rats. The serum calcium levels attained via parathyroidectomy in the present study were equivalent to those which restricted bone marrow, thymus and liver proliferation in vivo[7,9,11]. This suggests that the colonic response is not mediated through serum calcium. The exact nature of the dietary mineral requirement in the colonic refeeding response is currently under investigation.

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Oxidative activity during the sexual cycle of the central nervous system, adrenal glands and ovaries in the hamster (*Mesocricetus auratus*)

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Summary. The results indicate significant increases of the oxidative metabolism in the oestrus of the ovaries, hypothalamus and the posterior cortex, while in the amygdala this increase occurs in the phase of diestrus.

In the rat it is known that the hypothalamus regulates the secretion of the anterohypophysis and that there exists a close relationship between the oxidative metabolism of the hypothalamus and the secretion of gonadotrophines[1,2]. We know, the participation of the limbic system in the axis hypothalamus-hypophysis-gonad[3–5] and that the gonads show cyclical variations in their oxidative metabolism[6]. Recently[7], it has been shown that in the rat there seems to exist a possible participation of the posterior cortex (latero-occipital) in the control of the sexual cycle. There are numerous works[8,9] which note that the neuroendocrinal processes of the hamster seem to be different from those of the rat and other species of vertebrates. This information moved us to verify if the previously mentioned structures, which in the rat experience cyclic changes in their consumption of O_2, in the hamster experience the same changes as an index of their participation in the regulation of the sexual processes.

Material and methods. 30 female hamsters, whose weight varied from 130 g to 146 g, were used. They were fed 'ad libitum' the standard diet of the Interfacultative Department of Physiology of the University of Oviedo, with free access to drinking water. The light (12 h light, 12 h dark), temperature (23 ± 3 °C) and absolute humidity were controlled. The selection of animals was made following the study of the vaginal cytology, and only those which had complete cylces of 4 days were used. They were decapited and the following materials were dissected in accordance with Hoffman and Robinson[10]: hypophysis, hypothalamus, amygdala, posterior cortex (latero-occipital) and the septal area. Additionally, the ovaries and adrenal glands were extracted and weighed. The glucose level in the blood was determined by glucose oxidase method.

One oxidative metabolism (consumption O_2) was determined by Warburg's Manometric Method[11]. This method was used because of the abundant evidence which shows the close relationship between the oxidative metabolism of the areas of the CNS and the ovaries with the secretion of gonadotrophines[1,4]. The statistical treatment of the results was done in accordance with the test 't' of Fisher and Yates[12].

Results. Table 1 shows the results of the oxidative metabolism of the different structures studied. As we can see, the ovary, the hypothalamus and the posterior cortex suffer a significant increase in the phase of estrus, while amygdala experience it in the phase of diestrus; however the hypophysis, septal area and the adrenal glands did not experience changes through the sexual cycle. Table 2 reflects the values of the weight of the ovaries and adrenal glands (mg) as well as the glucemias in the phases of estrus and diestrus. Taking into account that there is no significant difference in the total weights of

Table 1. Oxidative metabolism of different organs in female hamster during the sexual cycle

Tissues	QO2: µl O_2/mg wet tissue/h Estrus	Diestrus	't'	p value
Ovary	0.92 ± 0.04* (12)	0.72 ± 0.04* (14)	3.17	0.01
Adrenal glands	0.49 ± 0.04 (12)	0.61 ± 0.06 (15)	1.55	NS
Hypophysis	0.82 ± 0.04 (6)	1.03 ± 0.11 (7)	1.61	NS
Hypothalamus	1.29 ± 0.04 (12)	1.04 ± 0.10 (11)	2.29	0.05
Amygdala	1.14 ± 0.06 (10)	1.41 ± 0.06 (10)	3.03	0.01
Posterior cortex	1.16 ± 0.08 (9)	0.90 ± 0.05 (9)	2.60	0.05
Septal area	0.93 ± 0.09 (7)	0.91 ± 0.13 (8)	0.12	NS

*Mean ± SE. Figures in parentheses = number of determinations. NS = Statistically nonsignificant differences.

Table 2.

Tissues	Estrus	Diestrus	't'	p value
Ovary	34.50 ± 1.36 mg* (12)	27.77 ± 1.41 mg* (18)	3.27	0.01
Adrenal glands	34.50 ± 2.05 mg (12)	23.23 ± 1.94 mg (17)	3.90	0.001
Glycemia (mg/100 ml)	65.30 ± 4.14* (12)	66.80 ± 2.42* (17)	0.34	NS

*Mean ± SE. Figures in parentheses = number of determinations. NS = Statistically nonsignificant differences.

the animals (t = 0.90; df = 27; p = NS) we have to detach the highest weight of the ovary and adrenal glands in the phase of estrus from that of diestrus, which throws up elevated significant differences. On the other hand, the glucemia does not show significant differences.

Discussion. The results of this work (table 1) indicate that in the structures of the CNS which were studied, the hypothalamus, the amygdala and posterior cortex (latero-occipital) show cyclic changes in their oxidative activity. In relation to the hypothalamus of the rat, it has been seen for some years now[13] that the sexual activity changes the oxidative metabolism of the hypothalamus and that castration of male rats causes a decrease in the consumption of O_2 of the anterior and posterior hypothalamus, which together with other findings points to the participation of the hypothalamus in the control of the secretion of gonadotrophines in the rat.

Our data, which coincide with those found in the bibliography, seem to indicate that, in the hamster, the hypothalamus experiences important cyclic changes in its consumption of O_2 which would seem to indicate its participation in the control of the sexual cycle. In relation to the posterior cortex (latero-occipital) cyclic variations in consumption of O_2 have been recently encountered in the rat, which would seem to indicate its action in the sexual regulation. Our results with the female hamster corroborate those obtained in the rat.

In the amygdala, cyclic changes occur as well, but while in the rat fhe oxygen consumption reaches its maximum in the phase of estrus and its minimum in the phase of diestrus, in the hamster the opposite occurs (table 1) behaving in a similar manner to that of the hyppocampus in the female rat. This distinct behaviour of the amygdala does not seem to support the theory of Payne and Swanson[8,9] that the neuroendocrinal activity of the hamster is not identical to that of the rat. The structures of the limbic system, which was studied, i.e. the septal area, showed no cyclic variations, the same as in the rat[5], although its relevant role in sexual behaviour has been proved; also that its injury produces the opposite effects to that of the amygdalines in relation to the sexual conduct[14,15]. Nevertheless, it has to be noted that the pattern of liberation of gonadotrophines is affected by the septal lesions[14].

In the hypophysis (table 1), cyclic variations have not been detected, although it is known that in the rat the secretion of gonadotrophines takes place in a cyclic form[16]. It is necessary to point out that the cyclic variations in the oxygen consumption have been detected in the adenohypophysis[5] and in our study we studied the complete hypophysis, which could distort the results. The study of the ovaries in the distinct phases of the sexual cycle has demonstrated cyclic variations in the consumption of O_2 like those encountered in this structure in the rat[6,17] which points out the cyclic action of this

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structure throughout the estrual cycles. This is shown also by the differences in its weight (table 2) which shows a statiscally larger weight in the phase of estrus than in that of diestrus.

The action of the adrenal glands in the sexual cycle of the hamster does not show any statistically significant differences from the point of view of its oxidative metabolism. It has to be pointed out that it is possible that the effects of the adrenal cortex remain hidden because of the medula, because if we take into account the total weight of these glands (table 2) in the phase of estrus, they reach a very high value compared to that reached in the phase of diestrus, which indicates a higher activity in this gland at that moment. Finally, in table 2, the values of glucemia are shown. As can be seen, statistically significant differences do not exist, which seems to indicate that the level of glucose in the blood does not experience changes during the sexual cycle.

Rhythmicity of aminotransferase in the cockroach, *Periplaneta americana*

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Summary. Aspartate (AAT) and alanine aminotransferase (AlAT) activities in nervous system and coxal leg muscle of the cockroach showed circadian variations with maximal activity around midnight, alternating with minimal activity at 12.00 noon of the solar day. The enhanced activity levels of the enzymes observed during night dark hours may be related to higher energy requirements during increased locomotor activity of the animals.

Studies on circadian rhythmicity in enzymatic activities have been gaining momentum in the recent years[2–5]. Diurnal variations in aminotransferases in vertebrates have been reported[6–8]. But such reports are lacking in invertebrates. Aminotransferases play an important role in transamination of amino acids to their respective keto acids and constitute a junction between the metabolism of protein and that of carbohydrates and lipids. The present study demonstrates the existence of cyclic variations in aspartate (AAT) and alanine aminotransferase (AlAT) activities in the nervous system and coxal leg muscle of the cockroach, *Periplaneta americana*.

Methods. Adult male cockroaches collected in Tirupati were acclimatized for 1 month to the laboratory conditions (27 ± 3 °C; 75 ± 5% RH). The animals were fed daily with bread pieces. 6-time-periods, viz., 8.00, 12.00, 16.00, 20.00, 0.00 and 4.00 h, were selected for experimentation to cover the 24 h period of the day. Nervous system, including all the ganglia and coxal leg mucsle, were isolated and pooled each time from a minimum of 3 animals. The tissues were preserved in ice-cold glass tubes till experimentation. Each experiment was repeated 5 times.

AAT and AlAT activities were estimated by the method of Reitman and Frankel[9] as given by Bergmeyer[10]. The incubation mixture contained 100 μmoles of phosphate buffer (pH 7.2), 2.5 μmoles of α-ketoglutarate, 50 μmoles of L-aspartic acid (AAT), 50 μmoles of DL-alanine (AlAT) and 0.2 ml of clear supernatant fraction of 1% tissue homogenates prepared in 0.25 M ice-cold sucrose solution. The contents were thoroughly mixed and incubated for 1 h for AAT and 30 min for AlAT at 37 °C, as they represent initial velocities. The reaction was stopped by the addition of 1.0 ml of 2,4-dinitrophenyl hydrazine (ketone reagent) in 0.1 N HCl. 10.0 ml of 0.4 N sodium hydroxide solution were added and the colour developed was read at 546 nm in Bausch and Lomb Spectronic 20. The enzyme activity was expressed as μmoles of pyruvate formed/mg protein/h.

Results and discussion. AAT and AlAT activities in nerve and muscle tissue showed cyclic variations with maximal activity at 0.00 h for AAT and 20.00 h for AlAT, alternating with minimal activity at 12.00 noon for both the enzymes (table 1). During the 24 h period, the levels of the enzymes were higher during dark h (20.00 to 4.00 h) than during light h (8.00 to 16.00 h) (table 1). From De Ritis quotient, it is observed that the tissues were pyruvate preponderant (table 2).

Table 1. Rhythmicity in aminotransferases in *Periplaneta americana*

	Time of day in h 8.00	12.00	16.00	20.00	0.00	4.00	MEL	A	B
Aspartate aminotransferase									
NS	3.43 ± 0.21	2.25 ± 0.36	2.66 ± 0.27	4.35 ± 0.57	4.81 ± 0.33	3.9 ± 0.42	3.57	2.78	4.36
MS	4.26 ± 0.35	2.5 ± 0.47	3.06 ± 0.32	5.21 ± 0.64	5.65 ± 0.49	4.62 ± 0.22	4.22	3.27	5.16
Alanine aminotransferase									
NS	7.58 ± 0.29	4.41 ± 0.66	5.51 ± 0.44	11.23 ± 0.37	9.55 ± 0.5	7.82 ± 0.6	7.67	5.83	9.53
MS	9.55 ± 0.74	6.27 ± 0.79	6.94 ± 0.66	12.22 ± 0.46	11.19 ± 0.84	9.97 ± 0.98	9.35	7.58	11.12

Enzyme activity is expressed a μmoles of pyruvate formed/mg protein/h.
NS, Nervous system; MS, coxal leg muscle; ± indicates SD; MEL, mean enzyme level of 6 periods; A, average enzyme level during 8.00–16.00 h; B, average enzyme level during 20.00–4.00 h.

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