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MORPHOMETRIC AND BIOCHEMICAL PARAMETERS AS AGE INDICATORS IN THE NORWAY RAT (RATTUS NORVEGICUS BERK, 1769)

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Determination of both morphometric and biochemical parameters as a function of age in the Norway rat has demonstrated that dry tissue weight of the eye lens pairs enables estimation of age in animals from one to approximately 22 months of age. Tyrosine content in the insoluble eye lens fraction, body weight and ratio of body length and weight enable the estimation of age from one to about 10 or 5 months of age, respectively.

Body weight of animals at the time of early adulthood (at about 75 days of age) was about 190g for males and 165g for for females. The ratio of body length and weight averaged 0.96 for males and 1.09 for females, while dry tissue weight of the eye lens pairs was about 14 mg and tyrosine content in the insoluble fraction of the eye lens was around 1320 μg for animals of both sexes.

Key words: Morphometric parameters, rat, lens, tyrosine

INTRODUCTION

The Norway (brown) rat, similarly to other rodent species, grows during its whole lifetime considering this fact certain authors have used body weight and length, teeth wearing out rate, fur colour, cephalization index, testis diameter, condylobasal skull length and other morpho-and craniometric parameters as relatively good age indicators. For most small mammals, particularly small rodents, it has been shown that the weight of the eye lens is an important age indicator. However, there are indications that eye lens dry mass in Mus musculus, Lemmus lemmus and Microtus arvalis is not a reliable parameter for age determination (Martinet, 1966, Berry and Trusglove, 1968; Ostbye and Semb-Johansson, 1970). Observing the tyrosine content in the eye lens of Peromyscus polionotus, Otero and Dapson (1972) and Dapson and Irland (1972) found that accumulation of the soluble protein fraction is an indicator of growth, and that the change from soluble to insoluble protein is dependent on aging, which confirms the research and conclusions of Dishe et al. (1956). Birney et al. (1975) determined that tyrosine in the insoluble fraction is a better parameter for age determination in Sigmodon hispidus than eye lens weight.

The aim of this paper is to extend our previous research (Savić and Kataranovski, 1981; Kataranovski and Kataranovski, 1984; Kataranovski et al., 1989) on relatively reliable criteria for age determination in the Norway rat.

MATERIAL AND METHODS

Animals were captured from the natural populations in urban and suburban habitats and bred in simulated natural conditions on the small farm at the Institute for Biological Research, Belgrade. Animals of known age were sacrificed. Preparation of the eye lenses for measurement of dry tissue mass was done according to Yabe (1979). For determination of tyrosine content in the eye lens insoluble fraction, the method of Dapson et al. (1968) was employed. Also, body weight and body length-weight relationship were measured and correlated to age in Norway rats of both sexes.

RESULTS AND DISCUSSION

Body weight-age relationship. The results obtained during assessment of the body weightage relationship in the Norway rat are presented in Table 1. Comparison of mean values of weight for males and corresponding values for females aged up to 35-40 days did not indicate any significant differences, but these appeared with age greater than 45 days. The relationship of body weight increase and age during the period of 350 days showed a characteristic sigmoidal curve. Statistical analysis confirmed that body weight and age from the moment of birth until the age of 150 days are strongly correlated, with $r = 0.99$ ($Y = 7.641 + 2.433 X$ for males and $Y = 10.724 + 2.047$ for females).

The results obtained are in accordance with data presented by King, (1939) according to Leslie et al., 1952), Calhoun (1962), Olejar (1966) and Hirata and Nass (1974). Bishop and Hartley (1976) found that growth slows and body weight stabilizes when Norway rat specimens reached a weight slightly over 300 g, which agrees with our results. We obtained particularly high significance in the t-test ($p < 0.001$) when comparing body weight between males and females older than 3 months. Olejar (1966) and Hirata and Nass (1974) established that mean values for weight of males and females significantly differ at ages greater than 2 months.

Table 1. Changes in weight of the Norway rat dependent on age.

Age in days	Body weight in grams						
	OO			OO			
	x	SD	n	x	SD	n	
0	6.1		58	6.1		59	
5	11.1		55	10.9		58	
10	20.3		53	19.5		58	
15	28.7	2.4	52	28.5	2.9	58	
20	41.1	3.2	52	41.1	3.1	57	
30	68.7	7.7	51	66.2	8.3	56	
40	100.1	12.1	49	91.5	10.7	56	
50	130.7	15.4	48	118.6	14.3	56	
60	164.0	19.8	48	143.1	16.5	56	
70	192.7	21.6	48	172.5	18.6	55	
80	218.4	25.1	48	191.2	21.5	55	
90	247.6	30.2	47	208.3	22.6	55	
100	273.1	30.6	47	231.6	25.2	55	
120	297.4	33.1	47	258.4	30.2	55	
130	309.4	35.0	47	266.5	32.6	55	
140	322.1	38.2	47	274.3	32.0	55	
150	338.4	40.1	47	283.4	37.6	55	
160	340.2	40.0	47	286.3	38.4	55	
180	340.3	39.6	47	290.1	38.6	55	
190	338.7	40.1	47	286.5	39.2	55	
200	340.2	39.4	47	287.1	39.2	55	
220	341.2	42.6	47	283.4	40.1	55	
240	342.1	45.5	47	286.2	43.7	55	
260	348.8	44.6	47	291.3	44.2	55	
280	353.6	47.6	47	293.8	46.5	55	
290	356.2	48.3	47	294.2	46.7	55	
300	354.1	48.1	46	295.2	46.5	55	
330	354.1	47.8	46	291.3	46.6	53	
350	355.7	46.8	46	292.3	45.9	52	

The body length-weight relationship as a function of age. Observation of the length-weight relationship as a function of age in the Norway rat indicated that these parameters are strongly correlated with age from 25 to 150 days, with r= -0.99 for both sexes (Figure 1). During the age period from 180 to 255 and more days, statistically significant differences were not observed in the length-weight relationship values.

These results confirm data presented by Calhoun (1962). It is obvious that the length-weight relationship, like body weight, can be used as a relatively reliable morphometric characteristic for age determination in Norway rats of younger age classes. Besides the finding that rodent growth gradually slows with age, while certain morphometric parameters become significantly variable, it was established that some of those properties (e. g. body weight, testis

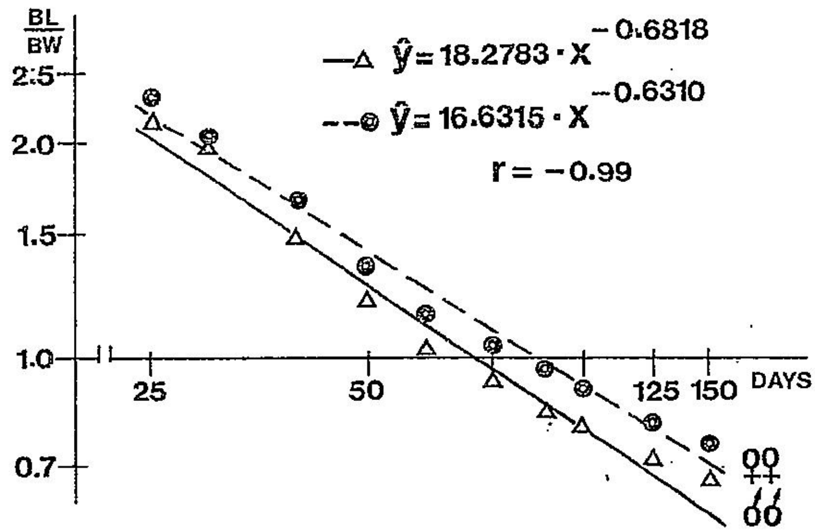

Figure 1. Body length-weight quotient BL/BW as a function of age (days) in the Norway rat.

diameter) change seasonally (Snyder et al., 1961; Skryja and Clark, 1970; Bishop and Hartley, 1976; Islam and Land, 1977).

*The dry eye lens tissue weight-age relationship.*The relationship of the dry mass of eye lens pairs with age was observed in Norway rat specimens (137 males and 124 females) from 1 to 25 months old on the day of sacrifice. We found that the dry mass of the eye lens pairs is correlated with age from 1 to 21.5 months, with the following regression curve values for 114 males:

$Y = 6.264 + 19.076 \log X$, $r = 0.89$, and for 98 females:

$Y = 6.552 + 18.723 \log X$, $r = 0.88$

(X = age in months, Y ≐ dry tissue mass of eye lens pairs in mg/.

Considering that sex differences do not exist regarding the weight of eye lens pairs-age relationship, an estimation of the regression curve was done jointly for both sexes. For this purpose mean values of eye lens dry tissue mass in specimens of known age were used (total of 23 age classes). The results are shown in Figure 2.

Analysis of the relationship between eye lens dry tissue mass and age has confirmed and extended our previous findings that this parameter can be used as an exceptionally good age indicator for specimens from 1 to approximately 22 months old. Taking into account that Donaldson and King (1937) (according to Yabe, 1979), and then Yabe (1979) studied eye lens dry tissue mass in Wistar type specimens and in offspring of wild and Wistar rats aged 1 to 12 months, Kataranovski and Kataranovski (1984), weighed the eye lenses of animals of the same type aged 1 to 19 months with the aim of comparing results. The obtained results (for 75 males: $Y = 6.732 + 18.471 \log X$, $r =$

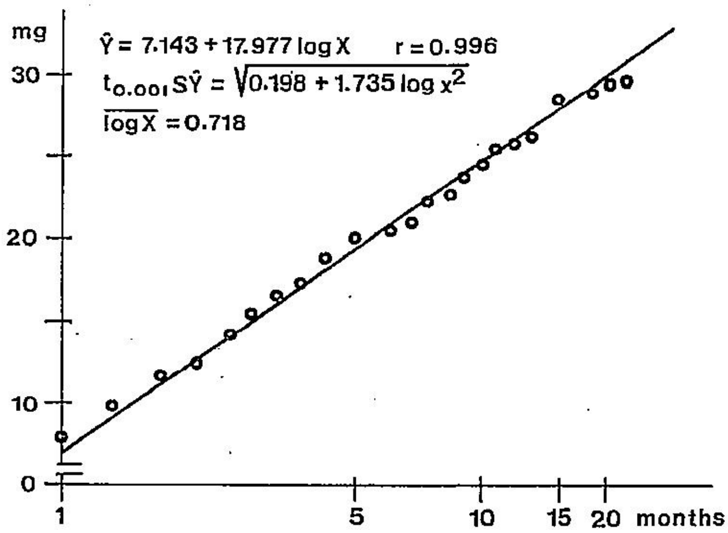

Figure 2. The dry tissue mass (mg) of eye lens pairs to age (months) relationship in Norway rats of both sexes.

0.89; for 78 females: Y = 7.010 + 18.207log X, r = 0.88) demonstrated that there are no significant differences between values for the weight of eye lens pairs in Norway and Wistar rats of the same age. The same authors also detected lower values for of the eye lens dry tissue mass in Black-hooded, Albino-Oxford and Dark-August specimens in comparison with corresponding values in Wistar and wild Norway rats of the same age.

Considering the results obtained by observing sexual maturity and reproductive ability in the Norway rat (Kataranovski, 1988), special care was dedicated to the weight of eye lenses in specimens aged 0.6 to 2.5 months. Based on the regression curve with:

$$Y = 7.180 + 17.262 \log X, r = 0.99,$$

it was determined that at the age of 2.5 months, when more than 75% of the specimens show sexual activity, the dry tissue mass of eye lens pairs equals 14.05 mg / Y ± t 0.001. SY = 13.5 to 14.6). These values do not differ from those shown in Figure 2 for animals aged 2.5 months (13.7 < 14.3 < 14.9 mg).

The (relationship) between tyrosine content in the insoluble fraction of the eye lens and age. Analysis of data obtained after the colorimetric estimation of tyrosine content in the insoluble fraction of the eye lens in rats of both sexes and known age from 1 to 16 months showed that tyrosine concentration and age of the animals were strongly correlated (r = 0.90) up to the age of approximately 10 months (Table 2). Comparison of values for the regression curve equation did not indicate any significant differences between sexes.

In contrast to our results, Dapson and Irland (1972) and Birney et al. (1975) detected a strong dependence between the observed parameters in

Peromiscus polionotus and *Sigmodon hispidus* up to the age of approximately 24 months. These authors also determined that tyrosine content in the insoluble fraction of the eye lens in specimens younger than 4 months did not represent a reliable age indicator.

Tyrosine content in the eye lens insoluble fraction was separatelly observed in specimens just reaching adulthood (13 males and 13 females) aged 2.2 to 2.7 months ($\bar{X}$ = 1.46 ± 0.15). We recorded that the tyrosine content was $\bar{x}$ = 1318.30 ± 68.60 μg (1160–1430) and that it is similar to the values shown in Table 2.

Table 2. Functional interdependence of tyrosine content in the crystalline lens insoluble fraction and age in the Norway rat.

Number of animals	Sex	Y = a + b log X		$+ t_{0.01} \cdot SY$	r	t_r
		a	b			
83	oo	365.124	2447.567	$68.093 \sqrt{5.159 + 83 (\log X - 0.602)^2}$	0.944	25.970
72	oo	396.023	2380.307	$77.771 \sqrt{4.609 + 72 (\log X - 0.647)^2}$	0.932	21.174

X in months; Y in μg

Besides the fact that tyrosine content in the insoluble eye lens fraction and dry or wet (see Rödel, 1987) tissue weight of the eye lens offer more accurate estimates than do the regular age determination methods, our results, together with data of other authors, support the opinion of Berry and Trusglove (1968) that these parameters must be checked for each animal species and cannot always be used.

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MORFOMETRIJSKI I BIOHEMIJSKI PARAMETRI KAO POKAZATELJI STAROSTI JEDINKI SIVOG PACOVA (RATTUS NORVEGICUS B E R K, 1769)

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SADRŽAJ

Praćenjem morfometrijskih i biohemijskih parametara u funkciji starosti jedinki sivog pacova ustanovljeno je da vrednosti suve mase parova očnog sočiva pružaju mogućnost određivanja uzrasta životinja do starosti od približno 22 meseca. Vrednosti sadržaja tirozina u nesolubilnoj frakciji očnog sočiva, telesne težine i količnika dužine i težine tela mogu biti primenjene u određivanju starosti jedinki do 10 (tirozin), odnosno 5 meseci) telesna težina i količnik dužine tela).

Članovi populacija sivog pacova sa tek stečenih adultnim statusom, pri starosti od približno 75 dana, imaju težinu tela oko 190 (mužjaci) i 165 g (ženke). Odnos između dužine tela ovih životinja iznosi približno 0,96 za mužjake i 1,09 za ženke, dok je vrednost suve mase parova očnog sočiva oko 14 mg a količine tirozina u nesolubilnoj frakciji očnog sočiva oko 1320 μg za oba pola.