Physical growth and brain development of captive-bred male and female squirrel monkeys, *Saimiri sciureus*

S.L. Manocha

Yerkes Primate Research Center, Emory University, Atlanta (Georgia 30322, USA), 5 June 1978

Summary. The physical growth and brain development of the captive bred male and female squirrel monkeys have shown that the male grows at a significantly faster rate with respect to these parameters in the postnatal life, starting from preweaning stage as compared to the female infant. During the prenatal life the male and the female fetuses grow at similar rates and show comparable brain development as indicated by its weight.

Because of the growing restrictions on the international shipments of large numbers of wild caught primates by the governments of India and several Latin American countries, it is imperative for the major consumers, such as the United States, to breed the primates at home for use in biomedical research. The use of squirrel monkeys - a South American primate, is next only to the rhesus as a laboratory animal[2-4]. Although small in size, which makes their housing, feeding and maintenance a relatively easy job, the squirrel monkey is endowed with a large sized brain and is quite suitable for projects which involve complex behavioral tasks or physiological functioning of a high order.

Recent studies on the breeding of this primate have given a clearer picture of its reproductive physiology[5-7]. With proper management, Manocha and Long[8] achieved a pregnancy rate of over 85% in their squirrel monkey colony. It is important to have baseline data on the physical development of the growing animals in pre- and postnatal life so that these monkeys can be used as appropriate scientific models with a full understanding of their age, growth patterns, nutritional requirements and general health. Beveridge[9] described the advantages of laboratory bred monkeys over wild caught animals. Some baseline data on the body weight and head and body length of squirrel monkeys during the postnatal period has been collected by Long and Cooper[10]. The present report deals with the physical growth and brain development of the captive bred male and female squirrel monkeys from 115 days of gestation to 36 months of postnatal life.

A breeding colony composed of 65 squirrel monkeys has been used over the last 6 years to collect data for this report. Details of breeding have been published elsewhere[8]. Before conception, the female squirrel monkeys weighed 620–710 g and ranged from 4 to 8 years of age. The day of conception was determined by physical examination of the uterus, the study of vaginal smears along with criteria

Table 1. Relationship of body, brain and cerebellum weights in male squirrel monkeys in the pre- and post-natal periods

Age sacrificed	Specimen	Body weight	Brain weight	Cerebellum
115-day gestation	EFS 3	55.85	10.32	0.649
115-day gestation	EFS 5	57.36	10.51	0.679
	$\bar{X}$	56.605	10.415	0.664
145-day gestation	EMO	66.18	13.75	0.777
145-day gestation	BFS 34	64.222	13.413	0.803
	$\bar{X}$	65.201	13.582	0.79
Neonate	BFS 11	105.0	13.132	–
Neonate	BFS 51	96.2	13.047	0.802
Neonate	BFS 12	118.08	13.841	0.928
Neonate	BFS 52	102.0	13.578	0.945
Neonate	BFS 41	124.0	14.56	–
Neonate	BFS 50	124.5	13.762	1.102
Neonate	BFS 54	119.5	13.99	1.105
Neonate	BFS 55	120.0	14.105	1.112
Neonate	BFS 56	117.5	14.217	1.117
Neonate	BFS 57	110.0	13.991	1.109
	$\bar{X}$	113.678	13.822	1.028
15 days	BFS 76	167.0	21.343	1.187
15 days	BFS 78	193.0	19.996	1.215
	$\bar{X}$	180.0	20.670	1.201
30 days	BFS 60	207.0	24.131	1.375
30 days	BFS 61	214.5	25.16	1.192
30 days	BFS 62	207.0	24.02	1.385
30 days	BFS 63	198.0	21.206	1.312
30 days	L 13–75	231.0	25.415	1.405
	$\bar{X}$	211.5	23.986	1.334
60 days	L 4–75	290.0	29.863	1.983
60 days	L 10–75	222.5	27.416	1.609
60 days	L 8–75	262.5	26.85	1.784
	$\bar{X}$	258.333	28.043	1.792
6 months	L 12–75	339.0	28.0	2.11
6 months	L 9–75	409.0	29.1	2.02
	$\bar{X}$	374.0	28.55	2.065
1 year	L 1–75	612	28.306	2.31
3 years	ADH 8	890	29.47	2.578
3 years	ADH 7	910	28.54	2.412
	$\bar{X}$	900.0	29.005	2.495

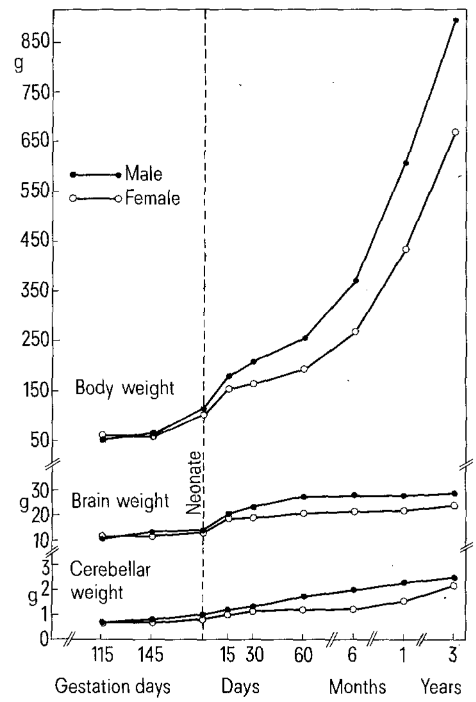
established by Nathan et al.[11], Goss et al.[12], and Hutchinson[13]. Fetuses at 115 and 145 days of gestation were removed by c-sections and the fetuses were sacrificed immediately thereafter. Beginning at 35 days of conception, the pregnant animals were removed from the outdoor breeding colony and housed singly in cages where their daily food and water intake was monitored and weight gain recorded at weekly intervals. The pregnant mothers were given a well-balanced diet containing 25% protein, 35% fat and 40% carbohydrate calories with adequate amounts of vitamins and minerals[9,14]. A similar diet was given to male and female infants after their weaning from the mothers at the age of 60 days. Several infants were sacrificed within hours after birth and animals sacrificed at the ages of 15, 30, and 60 days and 6 months, 1 year and 3 years in the postnatal life were also studied.

A study of tables 1 and 2, as well as the figure, clearly shows the differences in size and rates of development of male and female offspring. While at the age of 3 years, the female has acquired the full adult size, which does not alter significantly in terms of b.wt, the male continues to add extra weight and most of them cross the 1 kg mark during their 4th year of life. Up to 145 days of gestation, the difference between the growth and development of the male and the female fetus is negligible. The average b.wt of female fetuses at 115 days of gestation is 60.15 g compared to 56.605 g in the male. The brain weight at this stage of development averages 10.855 g in the female and 10.415 g in the male fetus. It is only in the neonatal period that the differences in b.wt between the sexes become significant; the average birth weight of a male neonate being 113.678 compared to 105.247 in the case of a female birth. However, there is no significant difference between the brain weights of the 2 sexes at the time of birth. Both male and female infants appear similar to each other in their abilities to cling to the mother, nursing and early exploration of their environments. During the postnatal period, the male infant appears to have a significant advantage over the female infant in terms of rate of development of the brain as well as accretion of weight. Such a difference is manifested even before weaning when the infant depends solely on the mother for breast feeding. It would be interesting to find out whether the mother of a male infant produces larger quantities of milk as compared to when she bears a female infant. The differences between physical growth and brain development of the male and the female infants become more pronounced after 15 days of postnatal life and remain distinct throughout life. By 3 years of age, the average b.wt, brain wt, and cerebellum wt is 900 g, 29.005 g, and 2.495 g, respectively, in the male and 675.44 g, 24.116 g, and 2.194 g, respectively, in the female.

Table 2. Relationship of body, brain and cerebellum weights in female squirrel monkeys in the pre- and post-natal periods

Age sacrified	Specimen	Body weight	Brain weight	Cerebellum
115-day gestation	BFS 27	60.78	10.99	0.688
115-day gestation	BFS 70	59.52	10.724	-
	$\bar{X}$	60.15	10.855	-
145-day gestation	EFS 2	63.101	12.322	0.779
145-day gestation	EM 2	62.43	11.231	0.755
	$\bar{X}$	62.766	11.777	0.767
Neonate	BFS 53	91.843	13.951	1.001
Neonate	BFS 13	114.53	14.005	0.910
Neonate	BFS 58	97.0	12.004	0.731
Neonate	BFS 17	117.16	14.321	0.974
Neonate	BFS 19	112.54	13.981	-
Neonate	L 6–75	118.5	16.002	0.799
Neonate	F 3–77	91.59	14.96	0.714
Neonate	BFS 36	96.344	12.796	0.800
Neonate	PN-2	107.72	14.417	0.787
	$\bar{X}$	105.247	14.049	0.840
15 days	BFS 75	150.5	19.165	1.117
15 days	BFS 77	160.5	18.521	1.054
	$\bar{X}$	155.5	18.843	1.086
30 days	BFS 21	169.0	18.235	1.190
30 days	PN-3	163.5	18.905	1.175
30 days	BFS 59	162.0	19.134	1.185
	$\bar{X}$	164.833	18.758	1.183
60 days	L 7–75	192.0	21.35	1.212
60 days	PN-4	195.5	21.760	1.230
	$\bar{X}$	193.75	21.555	1.221
6 months	PN-5	270.5	21.97	1.205
1 year	L 11–75	454	22.25	1.625
1 year	PN-6	425	22.32	1.572
	$\bar{X}$	439.5	22.285	1.599
3 years	L-76	625.0	22.88	1.746
3 years	M-29	650.0	22.74	2.458
3 years	L-9	727.0	22.696	1.875
3 years	M-9	640.0	25.11	2.478
3 years	M-10	702.0	25.13	2.480
3 years	M-12	689.0	24.871	2.399
3 years	M-13	680.0	24.98	2.314
3 years	M-18	676.0	23.87	1.987
3 years	M-28	690.0	24.765	2.007
	$\bar{X}$	675.444	24.116	2.194

Males not only have no advantage over the females in terms of physical growth and brain development during prenatal life, but there is another interesting aspect. In our colony, maintained at about 65 animals during the breeding seasons from 1971 to the present, an average of 4 animals are stillborn and 16 are aborted each year. Out of a total number of 24 prematurely extruded fetuses, including stillbirths, 15 were males and 9 females. (Some aborted fetuses and infants born dead were cannibalized by the mothers[15].) This clearly indicates that in their prenatal life, although more males are conceived, they suffer higher rates of mortality as compared to the females. It is only in postnatal life, that they gain an edge over the females by showing accelerated rates of physical and brain growth.



1 Acknowledgments. This work was supported by USPHS Grants RR-00165 and HD-06087 from National Institutes of Health. The technical assistance of Mrs Judie Wells is greatly appreciated.
2 T.B. Clarkston, B.C. Bullock, N.D.M. Lehner and M.A. Feldner, in: Animal Models for Biomedical Research, p.64. National Academy of Science, Washington 1969.
3 Institute of Laboratory Animal Resources, ILAR News *15*, 1 (1971).
4 Institute of Laboratory Animal Resources, ILAR News *16*, 1 (1972).
5 D.W. Ploog, in: Recent Advances in Biological Psychology, vol.8, p.199. Plenum Press, New York 1966.
6 R.W. Cooper, in: The Squirrel Monkey. Academic Press, New York 1968.
7 E.W. Hupp, in: Internationaler Kongress für tierische Fortpflanzung, p.2313. München 1972.
8 S.L. Manocha and J. Long, Primates *18*, 923 (1977).
9 W.I.B. Beveridge, WHO Symposium, p.490 (1971).
10 J.O. Long and R.W. Cooper, in: The Squirrel Monkey, p.193. Academic Press, New York 1968.
11 T.S. Nathan, L.A. Rosenblum, G. Limson and J.H. Nelson, Jr, Anat. Rec. *155*, 531 (1966).
12 C.M. Goss, L.T. Popejoy II, J.L. Fusiler and T.M. Smith, in: The Squirrel Monkey, p.171. Academic Press, New York 1968.
13 T.C. Hutchinson, Folia primat. *12*, 212 (1970).
14 R.S. Harris, in: Feeding and Nutrition of Non-human Primates, p.159. Academic Press, New York 1970.
15 S.L. Manocha, Lab. Animal Sci. *26*, 650 (1976).

Multinucleated cells in the retinal pigment epithelium: A scanning electron microscopic study

D. Puzzolo and Italia de Simone

Institute of Histology and General Embryology, Faculty of Medicine, University of Messina, Messina (Italy), 22 May 1978

Summary. Multinucleated cells in the retinal pigment epithelium of the albino rat are shown with the scanning electron microscope beside normal mononucleated cells. We suggest that such elements are formed during foetal life, owing to the absence of any mitotic activity in adult rats.

Recent studies of the ultrastructure of the retinal pigment epithelium have added much to our knowledge of its morphological features[1], its relationships with photoreceptors[2-4], and its role during the renewal of the rods[5] and, more recently, of the cones outer segments[6]. In spite of the many studies on the ultrastructural features, little attention has been paid to a characteristic detail of pigment cells, that is the observation of multinucleated elements; they were described for the first time by Ts'o and Friedman[7] with the light microscope. None of the above-mentioned authors were able to confirm such results with the transmission electron microscope; Hansson[8], with the scanning electron microscope (SEM), showed non-detailed photographs, obtained by the 'mostly possible' observation of some nuclear contours. The aim of the present study was, therefore, to show the first detailed images, obtained with the SEM, of multinucleated cells in the retinal pigment epithelium of the rat.

Materials and methods. Sprague-Dawley albino rats (200–250 g) were used and their activity rhythm (sleep-wake regimen) was synchronized by light-darkness 12:12 h, at 24±1 °C in a sound dampened room. The animals were killed by decapitation, the eyeballs rapidly excised and cut into 2 parts; the retina was picked with a pair of tweezers and then detached with a single sudden movement. The pigment epithelium was fixed in OsO_4 1% in phosphate-sucrose buffer 0.2 M at pH 7.4, dehydrated in ethanol, and then transferred to Freon 12, and finally to Freon 13 in a critical point apparatus[9]. The specimens were coated with gold and examined in a ETEC Autoscan SEM at 20 kV.

Results and discussion. The retinal pigment epithelium of the albino rat is formed by cells varying in their shape and size (figure 1). As to the shape, pentagonal (a_1), hexagonal (a_2) (both mononucleated) and heptagonal [bi- (b) or trinucleated (c)] cells are seen in a very close proximity; the hexagonal mononucleated cells have larger (5.19 µm) nuclei than the pentagonal ones (4.34 µm). As to size, next to mononucleated cells of about 12.68 µm diameter, binucleated cells of about 17.59 µm and trinucleated cells (figure 2) of about 17.82 µm diameter are seen. The