Reproductive and Lactational Performance of Rats Bred at Different Ages[1] (34680)

DONALD E. PRITCHARD AND H. ALLEN TUCKER

Animal Reproduction Laboratory, Department of Dairy, Michigan State University, East Lansing, Michigan 48823

For years management of laboratory rats has included the practice of not breeding females until they are about 60 to 80 days of age. This practice stems from the idea that an adequate body size must be attained before the stress of pregnancy and lactation is imposed upon the female (1, 2). This view was challenged by Cole and Hart (3, 4) who demonstrated that 26- to 31-day-old female rats, induced to precocious puberty with a single injection of pregnant mare serum gonadotropin, successfully maintained pregnancies which resulted in the birth of live normal pups.

Several workers (2–7) reported that smaller litters, both in number and individual pup weight, are born to younger, smaller females. But, in one study no difference in individual pup weaning weights was found between pups born to females which were 87 or 134 days old at first parturition (8). Although studies have been conducted to measure the reproductive and lactational performance of rats bred at various ages, the studies have not used postpuberal rats which have been younger than 50 days at the time of conception, nor has the lactational performance of these rats been thoroughly studied. This paper reports the reproductive and lactational performances of rats bred between 40 and 90 days of age. The usual age of the female rats at first breeding in our laboratory is 60 to 70 days.

Materials and Methods. Seventy-two female Sprague-Dawley rats were housed 6 to a cage from day 21 of age until they were transferred to individual lactation cages prior to parturition. They were kept under conditions of controlled temperature (25°) and lighting (14 hr daily), and received our regular rat feed free choice. The feed contains 21% crude protein, 3.9% fat, 2.5% crude fiber, antibiotics, and all the vitamins, minerals, and other nutrients recommended for rats by the National Research Council of the National Academy of Sciences. At day 21 of age they were randomly assigned to be cohabited with an adult male at the ratio of 6 females to 1 male for a 10-day period beginning at either 40, 60, or 80 days of age. Within each age group 12 females were assigned to be killed on the day of parturition and 12 on day 16 of lactation. Vaginal smears were examined daily during the breeding period to determine when mating occurred. At parturition body weight of the female and litter size and weight were recorded. Litter size was adjusted to 8 pups on the third day postpartum. Litter weights were recorded daily from day 3 to 16 as a measure of lactational performance. Lactating rats were killed by decapitation, and the 6 abdominal-inguinal mammary glands were removed, weighed, and stored in 0.25 *M* sucrose at −20° until analyzed for DNA and RNA as previously described (9).

Results and Discussion. Within each group of 12 rats, 8 to 10 of the females maintained normal pregnancies and delivered live young (Table I). Rats bred at an average age of 44 days weighed less at breeding, parturition, and autopsy ($p<0.01$) than those bred at an average age of 63 or 82 days (Table I). The compensatory growth that often occurs during lactation in smaller females of some species was not evident among the rats in our

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TABLE I. Body Weight and Litter Size of Rats Bred at Various Ages.

Day of lactation when killed	No. of rats with litters	Av age when bred (days)[a]	Wt when cohabited with male[a] (g)	Wt at parturition (g)	Wt at autopsy (g)	Litter size[c]	Litter wt at parturition[c] (g)
0	10	44	141	219[b]		8.6	56.6
	8	63	196	252		8.8	56.9
	9	82	223	256		10.2	64.8
16	9	44	144	227[a]	239[a]	9.1	61.6
	9	63	197	248	261	10.0	67.2
	9	82	216	269	277	10.6	69.3

[a] Values within time of kill are significantly different ($p < 0.01$).
[b] Significantly smaller than the other two values at that time of kill ($p < 0.01$).
[c] No significant difference between any of the values ($p > 0.05$).

study. Litter sizes and weights at birth were not significantly affected by age or size of the dam. These findings fail to agree with earlier reports (2–7) that younger females produced smaller litter sizes and weights. Perhaps improvements in dietary formulations and genetic selection for laboratory rats since these earlier studies have resulted in animals which are capable of producing larger litters (both in size and weight) at an earlier age.

The 6 abdominal-inguinal mammary glands of the rats killed at parturition were smaller ($p < 0.025$) for rats bred between 40 and 50 days of age than for rats bred during the other two breeding periods (Table II). However, by day 16 of lactation no differences in mammary weight ($p > 0.05$) were detected among the three age groups. Total mammary gland DNA was greater ($p < 0.01$) at day 16 of lactation than at parturition, indicating that mammary growth occurred during lactation in rats of all three ages. At parturition the oldest rats contained significantly more ($p < 0.01$) mammary DNA than the other two groups. By day 16 of lactation this difference was not apparent, indicating that more mammary gland growth occurred during lactation in the rats bred between 40–50 and 60–70 days than in those bred between 80–90 days of age.

Total mammary RNA also increased ($p < 0.01$) during lactation in all three age groups (Table II). This indicates that the hormonal stimulus of lactation causes an increase in protein synthetic activity (RNA) of

TABLE II. Mammary Weight and Nucleic Acid Content, and Litter Weight Gains of Rats Bred at Various Ages.

Day of lactation when killed	Av age when bred (days)	6 Abdominal-inguinal mammary glands				Day 3–16 litter wt gain (g)
		Wt (g)	Total DNA (mg)[c]	Total RNA (mg)[c]	RNA/DNA ratio[c]	
0	44	5.97[a]	13.87	46.84[b]	3.31	—
	63	7.51	15.59	58.61	3.80	—
	82	7.09	17.78[a]	45.94	2.59[a]	—
16	44	7.51[b]	22.76[b]	99.64[b]	4.43[b]	127.2[b]
	63	7.04	24.06	97.83	4.06	124.7
	82	7.53	24.08	106.21	4.47	152.5

[a] Significantly different from the other two values at that time of kill ($p < 0.025$).
[b] No significant difference among values within that time of kill ($p > 0.05$).
[c] Values between two times of kill significantly different ($p < 0.01$).

the mammary glands. There was no difference ($p>0.05$) in RNA among the three age groups at parturition or at day 16.

The average RNA/DNA ratio for the tion than the comparable averages of the other two groups. But, there was no signifioldest rats was smaller ($p<0.01$) at parturicant difference ($p<0.05$) between the three groups at day 16 of lactation. This indicates that the mammary glands of the oldest group began lactation with a lower protein synthetic activity per cell, but by day 16 protein synthetic activity per cell was equal to that of the other two age groups.

Litter weight gains from days 3 to 16 are shown in Table II. Orthogonal contrasts revealed no significant difference ($p>0.05$) in litter weight gains among the three age groups. This finding might be expected since there were no significant differences in the other mammary gland parameters measured. The correlation coefficients between days 3–16 litter weight gain and total mammary gland RNA were 0.84, 0.85, and 0.67 for rats bred between 40–50, 60–70, and 80–90 days, respectively, and these coefficients were significant ($p<0.05$). This supports similar findings by Tucker (9) who suggested that total mammary RNA at day 16 can be used as an indicator of the lactational performance of rats. All other coefficients between litter weight gain and mammary gland parameters were not significant.

Summary. Seventy-two female rats were randomly allotted to three groups and mated at average ages of 44, 63, and 82 days. One half of each age group was killed at parturition and the other one half at day 16 of lactation. Although rats in the youngest age group weighed less than those in the other two groups at each weighing, these age and weight differences did not significantly affect litter weight or number of pups per litter at birth. The oldest rats had more total mammary DNA at parturition, but no significant difference existed by the 16th day of lactation. Total RNA doubled during lactation for all three age groups, but there was no significant difference among age groups at parturition or at day 16. Average litter weight gains were not significantly affected by age at mating, indicating that rats bred as early as 40 days of age lactated and raised litters as well as rats bred after 60 days of age.

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