THE EFFECTS ON RATS OF CHRONIC EXPOSURE TO 34°C

IV. REPRODUCTION

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Summary

Vaginal opening occurred later in rats reared at 34°C than in those reared at lower temperatures. The vaginal plate broke down earlier in animals at all temperatures when they were given a high protein diet.

Oestrous cycles were longer in all animals kept at constant temperatures (21, 27, and 34°C) than they were in animals kept at room temperature. There was no significant difference between the results for the animals at the three constant temperatures.

The usual length of gestation amongst animals at room temperature on stock diets was 22 days. Gestation was prolonged in animals kept at 34°C on the same diets. There was no significant difference between the gestation periods for animals kept at 27 and 34°C on purified diets.

The rate of mating was less at 34°C than at lower temperatures, but the number of pregnancies following service was unaffected by the environmental temperature used. Female rats reared at 34°C mated as readily as animals reared at room temperature when they were paired at the latter temperature.

Ovulations were significantly depressed in females reared at 34°C; losses of embryos prior to implantation and losses of foetuses at birth were slightly higher at 34°C than at lower temperatures; losses during lactation were greatly increased by exposure to 34°C. Losses at birth appeared to be associated with an increase in the duration of gestation, losses during lactation to the effects of heat on the mother prior to parturition and to depression of lactation.

The similarities between these responses of the animals exposed to heat and those of hypophysectomized animals are discussed.

I. INTRODUCTION

Exposure to high environmental temperatures has been observed to affect the reproductive potential of rodents by influencing the age at which sexual maturity is reached, the length of the oestrous cycle, male fertility, female fertility, and the survivals of embryos, foetuses, and nursing young. Unfortunately, the responses have not always been in the same direction.

Vaginal opening has been observed (Sundstroem 1930) to occur earlier in rats exposed to 32°C than in controls. In mice Ogle (1934) and Mills (1945) found that this process was slightly retarded by heat. The discrepancy between these findings could be due to species differences or to differences in the diets used. The vitamin contents of the diets used were not stated, however.

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Przibram (1919, quoted by Sundstroem 1927) and Sundstroem (1930) found that the duration of the oestrous cycle was the same in experimental and control animals, but the experiments of Sundstroem (1930) showed that lengthened cycles appeared if the temperature of the warm environment was increased from 32 to 34°C.

Young first-generation male rats kept at 32°C were not as willing to mate as control animals but nearly all services led to pregnancy (Sundstroem 1930). As they became older these males had periods of sterility though they later became fertile again. Towards the end of the first year of life they showed signs of permanent sterility. The same pattern of behaviour appeared in several control animals so it is difficult to know whether there was any significant difference between the two groups. This sterility of older males was not observed in animals of later generations reared at high temperatures. Ogle (1934) mated female mice reared at 88–92°F (31·1–33·3°C) with males reared at the same temperature and with males reared at 70–80°F (21·1–26·7°C). The percentage of pregnancies was lower for the females paired with the heated males than for females mated with males kept at the lower temperature. Litter size was also reduced in the former group. Reductions in the percentage of pregnancies and in litter size were apparently due to infertility of the male. Mills (1945) stated that mice mated at the same rate at both high and control temperatures but that conceptions were fewer at high temperatures. Conceptions were probably measured by observing the numbers of litters produced. Since this depends on intra-uterine losses as well as on male fertility it is not possible to use this observation as a method of assessing male fertility.

There is some evidence that female fertility is reduced by exposure to high environmental temperatures. Sundstroem (1930) found that when males were placed with a group of female rats exposed to 32°C, some of the females failed to mate. Several authors have observed a reduction in the size of the litters born to rats (Steinach and Kammerer 1920; Sundstroem 1930) and mice (Ogle 1934; Mills 1945; Biggers *et al.* 1958), exposed to high temperatures. However, Sundstroem (1922) failed to observe any difference in the litter sizes of mice at 18–24°C and at 29–34°C. The reason for this is not clear; possibly the mouse strain used was not as susceptible to heat as those used in other experiments.

Sundstroem (1930) attributed prenatal losses to resorption and abortion while Ogle (1934) noted that losses were due in part to a number of the pups being born dead. Macfarlane, Pennycuik, and Thrift (1957) found that reduction in the number of pups born following acclimatization to heat for 2–11 weeks prior to mating was due to a reduction in the number of ovulations; losses due to resorption were no higher than those of controls and abortions were not observed. The discrepancy between the results of Sundstroem (1930) and of Macfarlane, Pennycuik, and Thrift (1957) may have been due to dietary deficiencies in Sundstroem's animals, since resorptions are known to occur in cases of vitamin deficiency, and heated animals are known to require more of certain vitamins than control animals (Pennycuik 1964*a*). Sundstroem (1930) gave no vitamin analyses of his diets so it is impossible to check this point.

Postnatal losses have also been found to be increased in heated rodents. Mills (1945) found that when mice were exposed to high environmental temperatures,

losses at birth and losses during the first week of life were higher than those for controls over the same period. Biggers *et al.* (1958) also observed that losses during this period were higher in heated mice than in controls.

These observations were made on several strains of animals at different temperatures and fed various (and possibly inadequate) diets. It was therefore decided to re-examine the effects of high environmental temperatures (34°C) on the reproductive potential of animals fed diets with adequate vitamin supplies.

II. Materials and Methods

(*a*) *Temperature Control*

The incubators used were the same as those described in Part I of this series (Pennycuik 1964*a*). Animals were kept at room temperatures, and at constant temperatures of 21, 27, and 34°C.

(*b*) *Diets*

All animals at room temperature and some of those kept at 34°C were given Barastoc dog cubes supplemented with Pentavite and Vetemul–vitamin E mixture. Animals kept at 21 and 27°C and the remainder of those kept at 34°C were given a purified diet supplemented with wheat and liver. Both these diets are described in Part I (Pennycuik 1964*a*).

(*c*) *Procedures*

The time of vaginal opening was determined by examining each rat each day from weaning until breakdown of the vaginal plate occurred. These measurements were made for pups born to mothers kept at the temperatures investigated.

The length of the oestrous cycle was estimated from records of vaginal smears made when the rats were 18 weeks old or older. These were taken over three or four consecutive cycles for each rat, smears being made once a day between 9 and 10 a.m.

Mating was carried out both at room temperature and in the incubators. The mating ability of the female rats was measured in terms of the number of oestrous cycles which occurred after the introduction of a male into the cage before service was observed. The ratio of males to females was 1 : 6. Service was detected by smearing; the presence of sperm or of vaginal plugs was taken as an indication that mating had occurred. This method was not entirely reliable, since in 2 out of 72 females kept at room temperature, pregnancy followed exposure to the male although sperm were not observed in the smear. This indicates the probable accuracy of the method.

The mating ability of the male was measured by offering each animal one female in oestrus each night for 12 consecutive nights. Females in oestrus were detected by examing vaginal smears for epithelial cells. Service was detected by looking for cornified cells and sperm in the smear on the day following exposure to the male.

The procedure followed for handling the animals during pregnancy was similar to that described in Part I (Pennycuik 1964*a*). Following parturition, animals at

34°C were kept at room temperature for about 4 days before they were returned to the incubator. This was done because early experiments had shown that losses of both mothers and young were high if they were kept at 34°C during parturition and early lactation. Nursing boxes were examined for new-born litters in the mornings and evenings. The young were counted at this time and at regular intervals throughout lactation, i.e. at 1, 2, 4, 7, 10, 18, and 21 days of age.

III. Results

(*a*) *Age at which Vaginal Opening Occurred*

It is clear from Table 1 that diet had a more pronounced effect on the time of vaginal opening than had environmental temperature. However, temperature did exert a significant effect, for rats on both diets matured later when kept at high environmental temperatures. Ogle (1934) also found that animals at high environmental temperatures (88–92°F, i.e. 31·1–33·3°C) matured later than those at 70–80°F (21·1–26·7°C).

The ages at which vaginal opening occurred in the groups at 21, 27, and 34°C are comparable with those of Ogle (1934) but they are below those observed by Asdell and Crowell (1935) and Murray (1941). Asdell and Crowell (1935) found that the vagina opened in their rats at an age of $53 \cdot 5 \pm 0 \cdot 19$ days when the animals weighed $136 \cdot 7 \pm 2 \cdot 1$ g, and Murray (1941) estimated that opening occurred at ages between 42·7 and 47·2 days. The latter author found that opening occurred earlier in animals which grew quickly. This may perhaps explain why opening occurred earlier in the animals on purified diets than in those on stock diets for the former grew more rapidly than the latter.

(*b*) *Length of Oestrous Cycle*

The findings for rats kept at room temperature, and at 21, 27, and 34°C are illustrated in Figure 1. Rats at room temperature showed a predominance of 4-day cycles, those at constant temperatures showed a predominance of 5-day cycles. This did not appear to be due to differences in diet, since oestrous cycles in rats kept at 34°C on stock diets were the same as those in rats at 34°C on purified diets. Lamond and Braden (1959) found that there was a diurnal variation in the response of mice to gonadotrophin injection. So far there is no evidence that this is related to temperature fluctuations, but it is conceivable that these may affect the pattern of response. Sundstroem (1930) considered that there was no increase in the cycle length of heat-adapted animals, though disturbed cycles occurred commonly in those exposed suddenly to a slight increase in external temperature. The values he quoted, however, show that a high percentage of rats at 32°C had 5- or 6-day cycles.

(*c*) *Duration of Gestation*

At room temperature, 75% of the rats on stock diets had a gestation period of 22 days (Fig. 2). The gestation periods of the majority of rats kept at 34°C on the same diet were 23 or 24 days. For rats reared on purified diets at 27 and 34°C

the differences in gestation time were not great but the proportion of 23-day pregnancies was much higher than it was for rats at room temperature. It is possible that this prolongation of the gestation period was due to the constancy of the environment.

TABLE 1

REPRODUCTIVE PERFORMANCES OF RATS REARED AND MATED AT ROOM TEMPERATURE AND AT 21, 27, AND 34°C

Numbers in parentheses indicate the number of rats in each group; the rats used for estimating survivals were the same as those used for estimating the duration of gestation

	Room Temperature	21°C	27°C	34°C
Males				
Services (%)	81·9 (6)	87·5 (2)	81·9 (5)	50·0 (7)
Pregnancies following service (%)	78·8 (6)	90·0 (2)	90·4 (5)	82·7 (7)
Females				
Time of vaginal opening (days)				
Stock diet	38·8±0·62 (41)			41·0±0·81 (24)
Purified diet		27·3±1·02 (15)	32·4±0·49 (65)	34·8±1·06 (14)
Duration of gestation (days)	22·2±0·07 (54)	22·9±0·21 (8)	22·6±0·12 (45)	22·9±0·10 (86)
No. of corpora lutea	9·26±0·20	8·50±0·52	8·78±0·22	7·72±0·16
Survivals				
Implants	8·24±0·21	7·13±0·54	7·42±0·23	6·14±0·17
Viable 16-day foetuses	7·65±0·25	6·13±0·65	6·64±0·27	5·35±0·20
Viable pups at day of birth	7·37±0·30	6·00±0·78	6·20±0·33	3·91±0·24
Viable pups at weaning (21 days)	7·00±0·35	4·38±0·90	4·45±0·38	0·81*
Mean mammary gland weight (g)	7·04±0·56 (8)	7·16±0·79 (5)	6·88±0·71 (6)	4·27±1·12 (3)

* Values for this group showed a skew distribution due to the fact that a number of mothers lost entire litters. Standard errors have therefore not been quoted.

(*d*) *Rates of Mating*

The rates at which males kept at room temperature and at 21, 27, and 34°C mated with females in oestrus are shown in Table 1. Males at the lower temperatures mated with 80% of the females offered, males at 34°C with only 50%. However, the fertility of the males, as measured by the number of pregnancies following service, does not appear to have been lessened by rearing the animals at 34°C.

Table 2 illustrates the number of females served during each oestrous cycle after the introduction of the male. Approximately 60% of the matings of rats reared and mated at room temperature and at 27°C took place at the first cycle.

Amongst rats reared and mated at 34°C only 40% mated at the first cycle. The mating abilities of the females were not apparently impaired by exposure to 34°C since those paired at room temperature mated as readily as rats reared at room temperature or 27°C. Whether the lower rate of mating is the result of male or

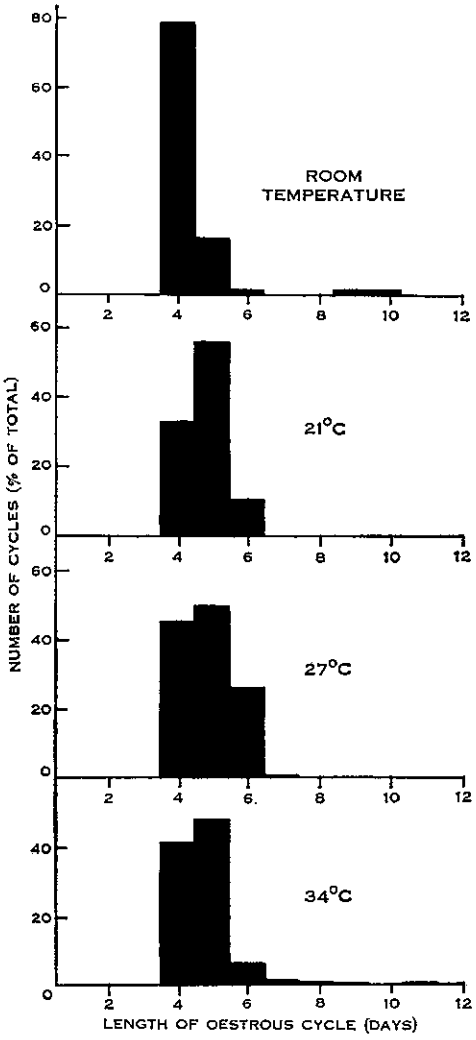

Fig. 1

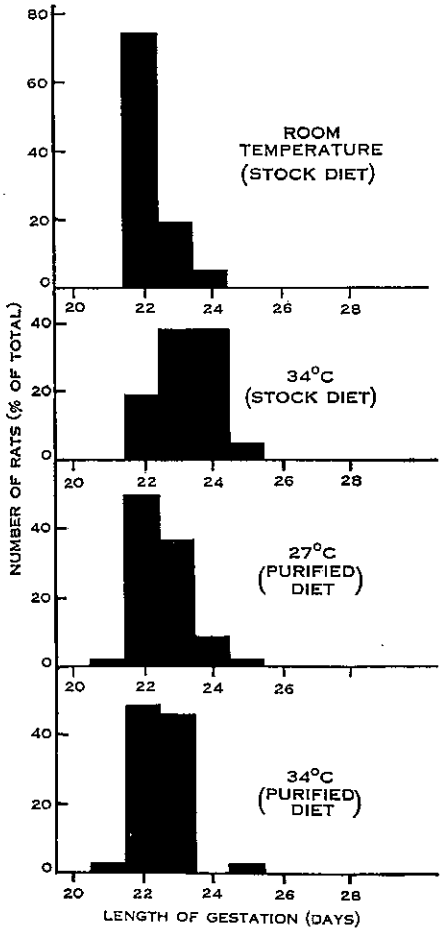

Fig. 2

Fig. 1.—Lengths of oestrous cycles of rats reared at room temperature (22 rats), 21°C (16 rats), 27°C (38 rats), and 34°C (61 rats).

Fig. 2.—Duration of gestation period of rats living at room temperature (71 rats) and at 34°C (21 rats) on stock diets and of rats living at 27°C (46 rats) and at 34°C (37 rats) on purified diets.

female depression, or both, cannot be determined from these experiments. It may possibly be related to the lowered activity of the rats at higher temperatures (Howard *et al.* 1959).

The findings in the experiment on the mating ability of the male are at variance with those of Mills (1945) but in agreement with those of Sundstroem (1930) for his young first-generation rats and also for his second-generation rats. They are also in agreement with Ogle's (1934) observations for mice. The high percentage of pregnancies following service confirms Sundstroem's (1930) results for rats born and reared at 32°C. The sizes of litters born to heat-reared mothers and sired by heat-reared males were no smaller than those of litters born to heat-reared mothers but sired by control males. This is at variance with the results of Ogle (1934) and Mills (1945).

TABLE 2

PERCENTAGE OF FEMALE RATS SERVED AT EACH OESTROUS CYCLE AFTER THE INTRODUCTION OF THE MALE

Female		Male	Temperature of Mating	1st Cycle	2nd Cycle	3rd Cycle	4th Cycle	5th Cycle
Temperature of Rearing	No. of Rats	Temperature of Rearing						
Room	128	Room	Room	57·0	34·4	6·3	2·3	
27°C	35	27°C	27°C	68·6	22·8	8·6		
34°C	50	34°C	34°C	42·0	38·0	10·0	6·0	4·0
34°C	12	Room	Room	66·7	33·3			

(e) *Corpus Luteum Counts and the Viability of the Young*

Corpus luteum counts and the number of young surviving at different ages are shown in Table 1 and Figure 3. The corpus luteum count for rats at 34°C was significantly different from those of rats at room temperature and 27°C, but the difference between animals at these two more moderate temperatures was not significant. In addition to the reduction in the number of ovulations the weights of the ovaries were significantly reduced in rats reared at 34°C (at room temperature, $25{\cdot}39 \pm 1{\cdot}20$ mg/100 g body weight; at 34°C, $19{\cdot}29 \pm 1{\cdot}25$ mg/100 g body weight). This reduction is in line with Sundstroem's (1930) observation that the weight of the testes was reduced in animals reared at higher temperatures. Body weight of the mother is known to affect the number of corpora lutea formed (Brambell and Rowlands 1936; Eckstein and McKeown 1955). When regression lines relating these two parameters were calculated for rats at room temperature and at 34°C the following equations were obtained:

$$y = 5{\cdot}75x + 109, \quad \text{(room temperature, 117 rats, S.E. } 20{\cdot}78)$$
$$y = 4{\cdot}90x + 99, \quad \text{(34°C, 86 rats, S.E. } 12{\cdot}22)$$

where y is the body weight and x is the number of corpora lutea formed. There is a 10-g difference between the values for the two groups suggesting that body weight

is more severely affected by heat exposure than the rate of ovulation. It is clear, therefore, that the lowered ovulation rate at 34°C is not solely a consequence of reduced body size.

The survival rate of pups during pregnancy and lactation is illustrated in Table I and Figure 3. Loss prior to implantation was higher for the animals reared at 34°C than for those reared at lower temperatures. This is in agreement with Ryle's (1961) findings for ewes kept at high environmental temperatures; namely that the animals tended to have fewer multiple ovulations, that fewer of these eggs developed into foetuses and, of these, fewer were alive 25 days after copulation.

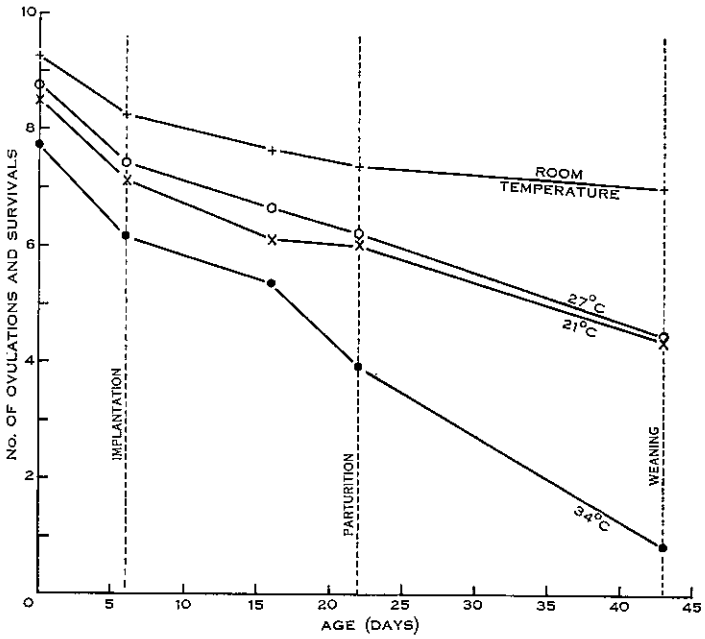

Fig. 3.—Ovulations and survivals of pups of mothers gestating and lactating at room temperature, and at 21, 27, and 34°C.

Resorption losses were the same for all four groups. This is at variance with the results for animals exposed suddenly to high temperatures where resorption loss was high due to corticoid release (Macfarlane, Pennycuik, and Thrift 1957). Losses at parturition were higher for animals at 34°C than for those at lower temperatures and losses during lactation were enormously increased by exposure to heat.

Loss between the sixteenth day of pregnancy and the day of birth was due to several causes: (i) loss by resorption between 16 days pregnancy and birth; (ii) loss by death of foetuses just prior to birth (which appeared to be a common cause of loss); (iii) loss due to death of pups just after birth but before pups were counted; (iv) loss due to the mothers killing and eating pups which were born alive.

The relationship between gestation length and the number of pups alive at birth for rats reared at 34°C is illustrated in Table 3. It was found that as litter size

decreased, pregnancy duration increased and the survival rate of whole litters and of pups fell. Hammond (1934) observed a similar relationship in rabbits. He attributed prolongation of pregnancy to the persistence of luteal tissue, and death of foetuses in the longer pregnancies to a failure of synchronization of the various physiological activities connected with the act of birth. Slonaker (1925) observed that longer pregnancies occurred in rats carrying small litters.

Losses of pups and of litters during lactation are illustrated in Figure 4. Most deaths occurred in all groups during the first day of extra-uterine life; these were fewest for the group reared at room temperature, more for the groups reared at 21 and 27°C, and most for the group reared at 34°C. Comparison with curves for litter survivals shows that loss at this time was largely due to the death of whole litters (Fig. 4). Losses for the period from day 1 to day 4 were smaller for all groups than

TABLE 3

NUMBER OF LITTERS AND OF PUPS SURVIVING AT BIRTH AFTER GESTATION PERIODS OF DIFFERENT LENGTHS IN RATS REARED AT 34°C

Period of Gestation (days)	Total of Mothers Pregnant	No. of Litters Surviving (in whole or in part)	No. of Young Viable on 16th Day of Gestation	Average No. in Litter on 16th Day of Gestation	No. of Young Alive on Day of Birth	No. of Survivals (expressed as % of 16-day foetuses)
22	31	29 (= 93·5%)	186	6·00	165	88·7
23	33	29 (= 87·9%)	169	5·1	125	74·0
24	15	12 (= 80%)	79	5·3	37	46·8
25	4	2 (= 50%)	15	3·8	3	20·0

those for the first day of lactation. They were again associated with deaths of entire litters. For the period from day 5 to day 10, losses for rats reared at room temperature, 21, and 27°C were negligible but loss for those reared at 34°C was still high. For the period from day 11 to day 18, losses for all groups were at a minimum. There was a further increase in pup mortality during the 18–20-day period for rats reared at the higher temperature, which did not occur for the other three groups.

Losses appeared to be due to a variety of causes operating at different stages of lactation. For the first day deaths of pups reared at 21 and 27°C are difficult to explain but those for the rats reared at 34°C were probably due to the effects of high temperature on mother or foetuses during the pregnancy period, for parturition took place at room temperature. The tendency of the mother to eat her offspring suggests that the losses were due, in part at least, to the effect of high temperature on the behaviour of the mother.

Losses over the 2–4-day period remained high for the rats reared at 34°C. As these were still living at room temperature, deaths must have been the result of heat exposure during pregnancy. Figure 5, in which weight changes of pups are graphed against time, shows that the pups which died during this period lost weight steadily

from birth to the time they died. Starvation due to failure of lactation seems to be the most likely cause, though it is possible that pups failed to learn how to suck.

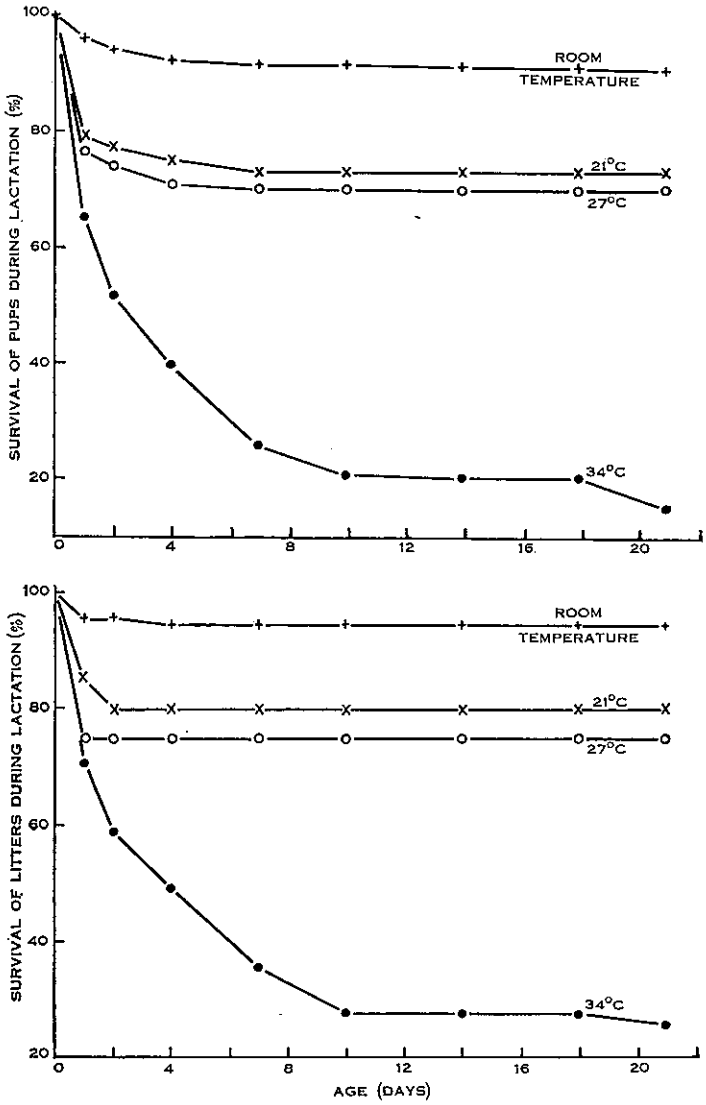

Fig. 4.—Survivals of individual pups and of litters during lactation at room temperature, and at 21, 27, and 34°C. In each case the numbers alive at birth have been taken as 100% and survivors at different stages during lactation have been expressed as percentages of this.

Rats reared at 34°C were the only ones showing significant losses over the 5–10-day period. By this time mothers and litters had been returned to 34°C. The weight of pups dying in this period (Fig. 5) increased over the first few days of extra-uterine life then fell, suggesting a failure of lactation when the mother was returned

to the high environmental temperature. Loss could not have been due to failure to suckle in this case. The pups which survived to weaning showed a break in the growth curve over this same period. This period is also the one characterized by rapid maternal weight loss and depressed food intakes (Pennycuik 1964*b*, 1964*c*). These changes preceded the loss of weight by the pups, suggesting that appetite is first affected by heat exposure and that reduced lactation and pup survival occur as consequences, but this is almost certainly an oversimplification of the situation.

Losses over the 18–20-day period occurred only in one high temperature group (2 rats) allowed to suckle to 21 days (in most of the other groups pups were weaned

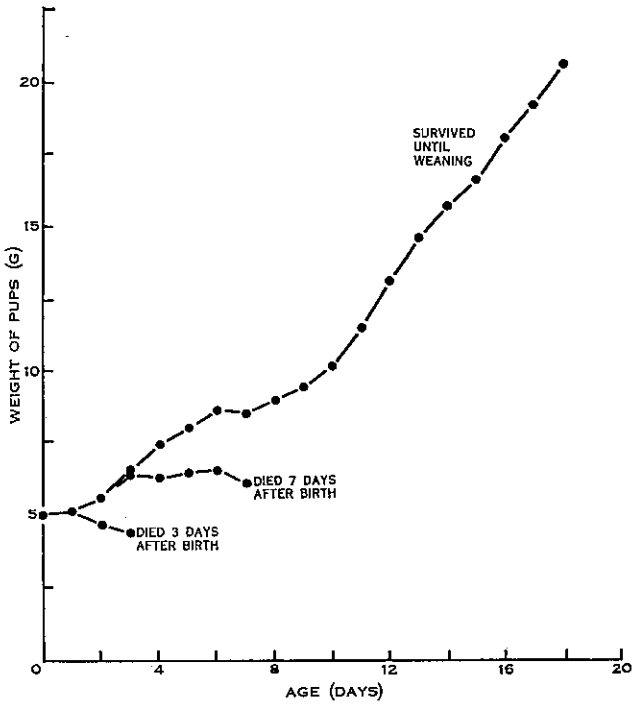

Fig. 5.—Weight changes of pups reared at 34°C and which survived 3 and 7 days after birth and until weaning.

at 18 days). The mothers lost a good deal of weight during lactation and at 19 or 20 days they killed their offspring by nipping through their spinal cords just below the skull base.

Evidence for changes in behaviour at 34°C was also seen in the earlier stages of lactation. Mothers kept at lower temperatures tended to retrieve young pups which strayed from the nest, and to remain still while the pups suckled. Many of the females kept at 34°C showed no interest in keeping their offspring about them and were restless when pups attempted to suckle.

In addition to reduced pup growth at 34°C there is further evidence for reduced lactation at this temperature. Table 1 shows the absolute weight of mammary tissue present at 21 days in females lactating at room temperature, 21, 27, and 34°C. The

amount of tissue present was approximately the same for the animals at the lower temperatures but was reduced in those at 34°C. Figure 6 shows that the amount of mammary tissue developed for each pup was less for the rats kept at 34°C than for those at the other temperatures used. This is in agreement with the findings of Ragsdale *et al.* (1948) and with Morris (personal communication 1961) that milk production of cattle and ewes is lower in animals gestating at high temperatures.

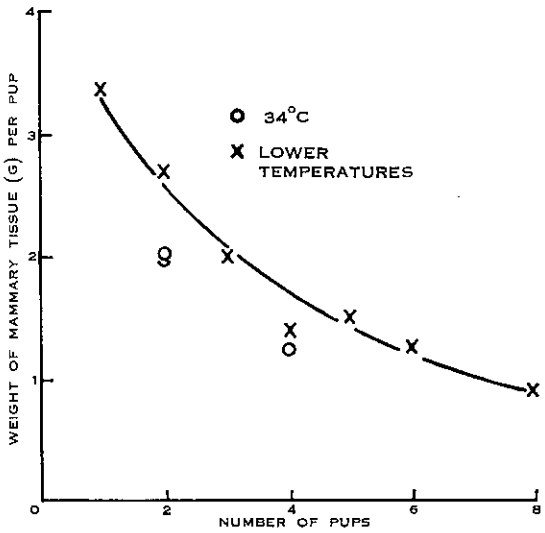

Fig. 6.—Relationship between the amount of mammary tissue and the number of pups of mothers reared at 34°C (3 rats) and at lower (room, 21, and 27°C) temperatures (19 rats).

The reduction in the number of ovulations observed at 34°C is in agreement with results of earlier experiments carried out in this Laboratory (Macfarlane, Pennycuik, and Thrift 1957). The reduction in implantation rate does not appear to have been observed previously in rats, but it has been observed in sheep (Ulberg 1958; Ryle 1961). Sundstroem (1930) attributed the reduction in the number of survivors at birth to resorption and abortion. The elimination of this source of loss in the present experiments can possibly be attributed to the use of more satisfactory diets. The losses at parturition are comparable with those of Ogle (1934) and the losses during lactation with those observed by Mills (1945) and Biggers *et al.* (1958).

IV. Discussion

High environmental temperature affected many phases in reproduction and in all cases the changes produced tended to reduce the number of rats in succeeding generations. The main causes of this reduction are the later maturation of the female, the lower rate of mating, a reduction in the number of eggs shed, reduced implantation, and increased losses of pups at parturition and during lactation. Probably there are several reasons for this wide array of changes, though all may be ultimately related to a depression of ovarian function.

The ovary is known to be depressed by a number of factors. Rinoldini (1950) showed that inanition caused a loss of weight of the gonads. Brambell and Rowlands (1936) and Eckstein and McKeown (1955) demonstrated that there was a correlation between body size and the number of corpora lutea produced. Increasing dosages of gonadotrophin injected into immature mice (Green 1955) caused a proportional increase in ovarian weight, and superovulation following gonadotrophin injection is a well-known phenomenon (Evans and Simpson 1940; Austin 1950) so it would be expected that lowered gonadotrophin production would cause a reduction in the number of follicles ripening and the number of corpora lutea formed. A reduction in the number of corpora lutea in adrenalectomized rats was reported by Mandl (1954). This was related to lowered sensitivity of the ovary to follicle-stimulating hormone in the absence of corticoids. Leatham (1951) found that ovarian weight was reduced in rats given thiouracil over a long period.

Reduced food intake is a possible cause of lowered ovarian activity in the heated animals. However, they did not display the usual signs of starvation observed by Asdell and Crowell (1935), and their fat stores were only 20% below those of control animals (Pennycuik 1964c).

Reduction of body weight is also a possible explanation, but the regression equations relating body weight and numbers of corpora lutea in animals at room temperature and 34°C illustrate clearly that these parameters were affected differently by heat exposure.

The remaining explanations, namely reduction of gonadotrophins, corticoids, and thryoxine, share the common feature of hypofunction of the appropriate endocrine gland. Since both thyroid and adrenal are finally dependent on the anterior pituitary, hypofunction of this organ could be involved in all cases. Bonsma (quoted by Hammond 1949) has already speculated that this may be the explanation of lowered fertility in cattle exposed to tropical conditions.

Many characteristics of the heated rat resemble those of the hypophysectomized animal though they are less extreme in degree. Pituitary removal at 6 days of age is known to reduce the growth rate of rats to 50% of that of normal animals (Walker *et al.* 1950). Removal of the gland from older animals inhibits growth entirely (Lee and Ayres 1936; Walker *et al.* 1950). A similar depression in growth rate has also been observed under hot conditions by Sundstroem (1930) and in the present series of experiments. Smith (1930) showed that there was a reduction in the relative weight of the liver, spleen, and kidneys following hypophysectomy; similar changes have also been observed after heat treatment (Sundstroem 1930). Crafts and Walker (1947) observed a reduction in gastric acidity in the hypophysectomized rat; the acidity of the gastric juice of the heated animal has also been observed to be reduced (Ichiyoshi 1954). Smith (1930) found that there was an immediate disappearance of libido following pituitary removal; males exposed to 34°C were definitely less inclined to mate than their brothers at room temperature. Pencharz and Long (1933) found that pituitary ablation between the eleventh and twentieth day caused an increase in length of pregnancy by as much as 4 days, difficult birth, and a failure of milk secretion; these also are very similar to the results of the present experiments though the effects were not so severe. Knobil and Canton (1953) weighed pups of

control and hypophysectomized mothers on the twenty-first day of pregnancy and found that those of hypophysectomized mothers were significantly lighter, while in the present series of experiments new-born pups of mothers reared at 34°C were found to be lighter than those born of mothers reared at lower temperatures (Pennycuik 1964*b*). Food intake was found to decrease in the hypophysectomized rats studied by Rinoldini (1950); this is also a characteristic of animals reared at higher temperatures.

In view of these resemblances the hypothesis that hypofunction of the anterior pituitary is responsible for some of the changes observed in the heat-exposed rat seems a reasonable one to investigate. Should this hypothesis prove tenable, it will probably be found that some of the changes produced by heat are not due to the direct action of pituitary hormones but rather to the effect of pituitary hypofunction on other endocrines. Corticoids have been shown to influence the production of corpora lutea (Mandl 1954), body weight (Heroux and Hart 1954), and food intake (Kochakian and Robertson 1951). Thyroid depression has been shown to cause an increase in length of the oestrous cycle, a decrease in the percentage of fertile matings, and a decrease in the percentage of young born alive (Peterson *et al.* 1952); it also causes a depression in adrenal and ovarian weight (Leatham 1951) and a slight prolongation of pregnancy (Krohn and White 1949).

Pituitary hypofunction has been postulated as a cause of hibernation (Foster, Foster, and Meyer 1939). Unexpectedly, heat-treated animals show resemblance to those adapted to this other extreme of temperature toleration. These adaptations include lowered activity, lowered food intake, reduced sizes of endocrine glands (Ryle and Morris 1961), lowered blood sugar levels, and a tendency towards ketosis. If these changes are due to pituitary hypofunction, it would be interesting to know how function of this gland varies over the intervening range and how this fall-off is brought about.

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VI. References

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