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Influence of maternal thymectomy on development of the offspring in the rat

By

G. Csaba, A. Hajpál, I. Juvancz

Institute of Biology, Semmelweis University Medical School, and Biometric Unit Hungarian Academy of Sciences, Budapest

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Average body weight of the offspring of tyhmectomized mother rats is significantly less than normal at the age of 17—20 days. At other ages the difference is less pronounced. Though the mortality rate of such offspring seemed to be doubled, the difference was not significant. As compared to the controls, the sham operation itself decreased the thymus weight.

The thymus is constructed of specific epithelial reticulum and lymphoid elements [11]; its role in immunological processes is assumed to depend on this structure [9, 10]. Though the attempts at isolating its hormone have failed, there are some proofs of its existence [8]. The fate of adult animals is not or is only slightly influenced by thymectomy [3, 4, 10], while in the newborn rat the operation induces wasting disease, a condition that is often fatal [9, 10]. In the survivors, resistance to infections and injuries is considerably impaired [5, 6]. There are findings to suggest that the life-span of thymectomized animals was shortened even without signs of wasting [1].

Recent studies [7, 8] have shown that the thymus exerted an influence on immunological processes apart from the direct cell-to-cell contact by humoral means. Thus, this factor contacting the fetus in utero might influence its later resistance. This has made us to investigate the behaviour and fate of offsprings of thymectomized mother rats. It seemed even more such, as for human the state of the thymus displays great variety even within the some age-groups: it may be an epithelial-lymphoid unit of complete value or may be an adipose thymus [2].

Material and Methods

Forty-five Wistar CB closed bred adult female rats of 200 g weight were used. They were divided in three groups of 15 animals each. In the treated group the sternum was split and the thymus was removed together with its capsule. In the sham operated group only the sternum was split. In the control group there was no intervention. All the females were mated to the same sire. In the treated group all the 15 females, in the sham group 11, and in the control group 13 became pregnant.

Prior to delivery the mothers were transferred to separate cages. After delivery the litter sizes were registered and the individual body weights of the newborns were registered. Weighting was repeated every 3—4 days till the 30th day, when all the animals were killed. The thymus of all the offsprings and of the mothers from the sham and control groups was weighed and fixed in Carnoy's solution for light microscopic study.

Results

Three indices were applied for estimating the effect of maternal thymectomy on the progeny, *viz.* mortality, gain in body weight, and weight of the thymus. Mortality and weight gain being dependent on litter-size, a preliminary analysis of this supposed influence was performed.

Influence of litter-size on mortality

The litter numbered from 2 to 13. Consequently, their individual shares were too low to yield reliable information. Nor could matching the litters of the same size be utilized as in several cases the matching size was missing for one of the groups. The best founded partition seemed to be whether the offsprings outnumbered eight, the number of mamillae. To compare the treated and sham groups, four subgroups were formed of the small and large litters for the treated and similar two for the sham operated animals.

Analysis of the mortality data by angle transformation, the results proved that the litter-size had little influence on mortality, *viz.* 15—15% for the treated and 7—10% for the sham groups. These results thus justified to pool the data for group mortality.

Influence of maternal thymectomy on the mortality of the offspring

The pooled frequency of the treated group was 20, or 15%; and for the sham group 10, or 8%. Even this mortality rate failed to prove significant though it was near to it.

As ancillary statistics, the velocity of the process leading to death was analyzed, i.e. the reciprocals of the age at death. As the best estimate of the age at death, the arithmetic means of the last "still alive" and the first "already dead" was considered. The harmonic means of ages at death was for the treated group 5.2 days, and for the sham group, 4.6 days. This would suggest the velocity being equal or perhaps somewhat higher for the sham group. This was, however, difficult to explain as in the treated group the mortality seemed higher and the onset mostly occurred in the first week. This contradiction seems to be due to the death on the first day of 4 progenies in a litter of 10 in the sham group. Consequently, repetition of the experiment on a major scale might show a higher mortality rate and velocity in the treated group.

Influence of litter-size on individual weight

Weight were analysed at the same ages. Age-groups of ranges not exceeding 4 days were formed. First

it was tested which litter-size had to be taken in regard; the one synchronous with the weight or the one of 10 days prior to testing. The regressions of weights on litter-size proved to be practically the same for both possibilities, so the analysis was performed with the synchronous litter-size. For no group was the deviation from linearity unacceptable.

Influence of maternal thymectomy on weight of the offspring

The effect seemed to be overt for all the age groups though less pronounced in some. In the 3—6 days group it was rather vague (Fig. 1); in the 13—16 one it seemed more convincing (Fig. 2); in the 17—20 one it was marked (Fig. 3). Detailed analysis of this age group proved the regression lines for the two treatment groups running practically parallel with the sham group being 4 g above the treated group. The difference proved highly significant statistically ($P < 0.01$). On the 30th day, the difference was much less distinct (Fig. 4).

Influence of maternal thymectomy and sham operation on thymus weight in the offspring

Thymus weight in the three (thymectomy, sham, control) groups was compared by analysis of variance. The differences between the means, 369, 388, 402 mg, respectively, did not prove significant. Though the data suggested an increasing tendency, samples multiple in size would be needed to prove its significance.

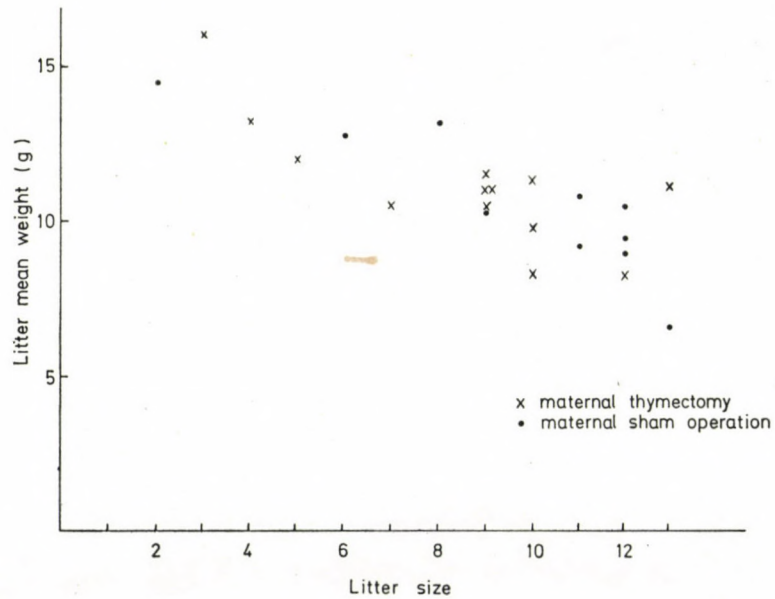

FIG. 1.

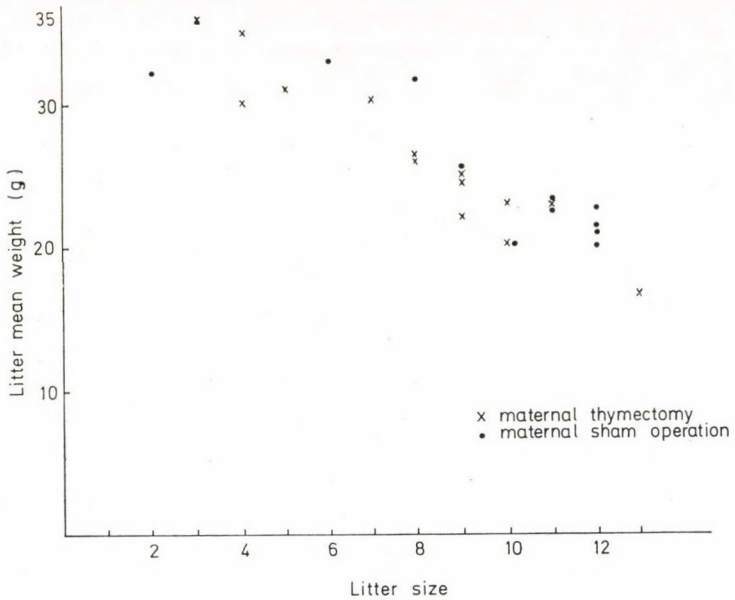
35
30
20
10
Litter mean weight (g)
x maternal thymectomy
• maternal sham operation
2
4
6
8
10
12
Litter size

FIG. 2.

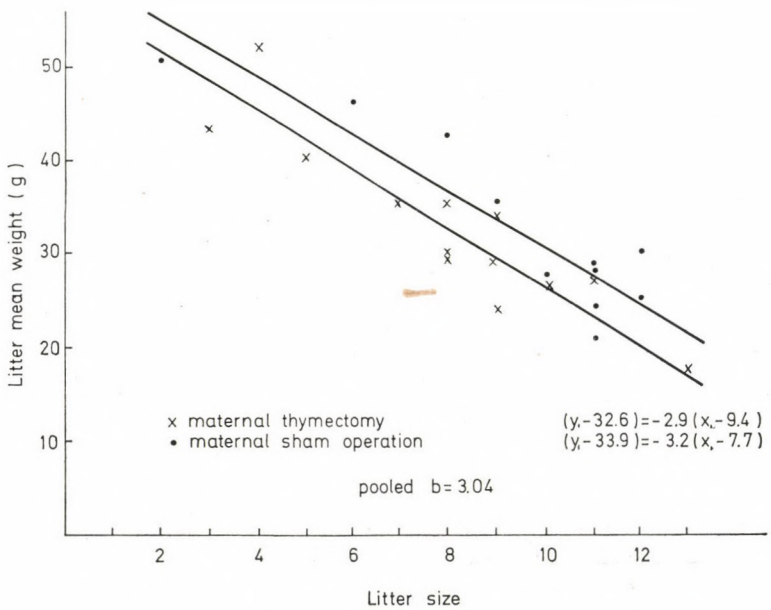
50
40
30
20
10
Litter mean weight (g)
x maternal thymectomy
• maternal sham operation
(y,-32.6)=-2.9(x,-9.4)
(y,-33.9)=-3.2(x,-7.7)
pooled b=3.04
2
4
6
8
10
12
Litter size

FIG. 3.

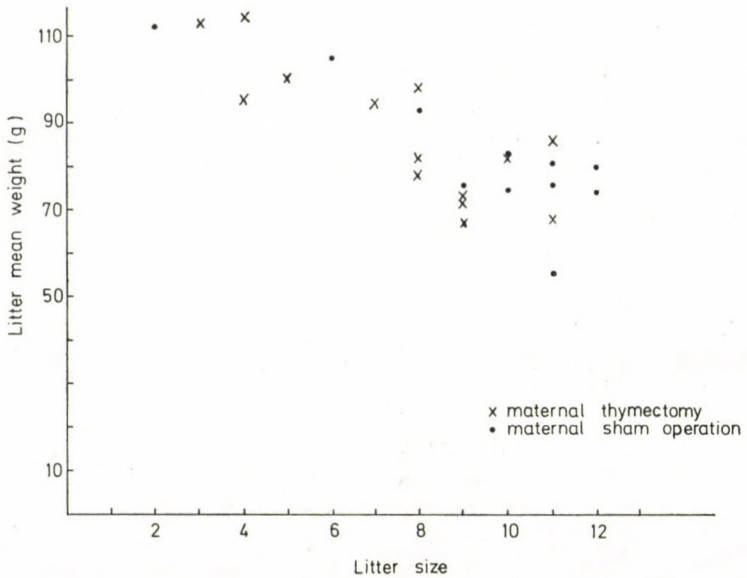

FIG. 4.

FIGS 1—4. Influence of maternal thymectomy on body weight of the offspring. 1) 3—6 day. 2) 13—16 day. 3) 17—20 day. 4) 28—30 day age groups

Influence of sham operation on thymus weight

For the sham group the mean was 183 mg, for the control 273 mg; the *t*-test yielded a significant result.

Histological studies

The thymus of the offsprings showed no differences in the three groups.

DISCUSSION

The results suggest that maternal thymectomy exerted some kind of harmful influence on the offsprings. Taking in regard the samples sizes, not only the significant differences but also the tendencies proved informative. This holds first of all for body weight, that displayed a pronounced and significant difference for the 17—20 days age groups and non-significant ones for the other ages. The age coincided with the retardation registered in wasting disease [3, 4, 9, 10]. The results seem to suggest that the influence of maternal thymectomy was similar to neonatal thymectomy though of much lesser grade. As no sign of retardation could be observed in the newborns and the effect developed gradually, the thymus factor might reach the sucklings by the mother's milk and its lack in the case of maternal thymectomy would be harmful. The histological similarity of the thymus in the three groups of progenies suggests that the maternal thymus has no direct effect on the

gland of the offspring. In this case the maternal thymus factor would directly influence the thymus-dependent organs of the suckling. The suckling's thymus alone would not be sufficient to stimulate these organs. The 30th day data suggested that the young rat's thymus attains its full capacity by that age, enabling it to overtake the regulation.

The significant difference in post-lactational thymus weight in the sham and control group showed that the sham operation in itself was hindering thymic functions. The data of the treated group were compared to those of the sham one. It seems justified to suppose that the differences of the treated versus the control not reduced by the effect of sham operation, would be greater than the difference between treated and sham values.

No direct inferences from animal to man are ever permitted. However, the above observations have some importance in neonatology as, in contrast to rats, in man the functioning thymus substance displays wide varieties in dependence on age [2].

Professor G. Csaba
Tűzoltó u. 58
H-1450 Budapest, Hungary

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