REPEATED POSTTRAUMATIC REGENERATION OF BONE TISSUE IN THYMECTOMIZED RATS

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Incorporation of Ca^{45} into bone tissue during primary and secondary (after 50 days) regeneration is depressed in thymectomized rats. This depression is particularly marked during secondary regeneration of bone after thymectomy and is observed in both injured and uninjured parts of the skeleton.

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Regeneration after repeated experimental trauma is more marked in degree and more rapid in its course than after identical primary trauma [2, 4, 6, 7, 9]. However, the mechanism of this "anamnestic reaction" has received inadequate study.

The object of the present investigation was to study the role of the thymus in activation of secondary regeneration, in view of reports indicating its importance in the regulation of mineral metabolism and the course of regeneration in calcified tissues [1, 3, 8].

EXPERIMENTAL METHOD

Experiments were carried out on 128 Wistar rats weighing 80-120 g, distributed among 7 groups: 1) control, 2) primary trauma, 3) repeated trauma, 4) thymectomy, 5) thymectomy plus primary trauma, 6) thymectomy plus repeated trauma, 7) repeated trauma plus thymectomy plus primary trauma. Measured trauma [6] was inflicted on the femoral diaphysis. An identical second trauma was inflicted to the same part of the bone 50 days after primary trauma. Thymectomy was carried out as described elsewhere [10]. The animals were sacrificed 3 and 15 days after the beginning of the experiment (groups 1, 2, 4, and 5) or after repeated trauma (groups 3, 6, 7). $Ca^{45}Cl_2$ was injected subcutaneously into the rats 24 h before sacrifice in a dose of 1000 pulses/min/g (specific activity 0.116 mCi/mmole). Radioactivity was determined in ash samples (10 mg) from bone tissue on a type PP-16 apparatus with a T-25-BFL end-type counter (thickness of mica 1.1 mg/cm^2). The error of determination of radioactivity did not exceed 2%. The results were expressed in pulses/min/mg ash and subjected to statistical analysis using Student's criterion. Differences were regarded as significant when $P \leq 0.05$.

EXPERIMENTAL RESULTS

Incorporation of Ca^{45} into bone tissue of thymectomized animals (group 4) was reduced on the 3rd day to 85.5% compared with the control (group 1), after which it increased to 134% on the 15th day (Table 1). The difference is statistically significant ($P < 0.001$). After primary and secondary trauma (groups 2 and 3), incorporation of the isotope into regenerating bone was increased, this increase being observed by the 7th day of secondary regeneration, i.e., sooner than during primary regeneration. The difference is significant ($P < 0.05$). These changes in calcium metabolism in the calcified tissues are generally in agreement with those discovered previously [1, 5, 6].

The course of primary posttraumatic regeneration in thymectomized animals (group 5) was marked by absence of the early phase of activation of metabolism, and it was evidently determined only by the thymectomy itself; the differences between groups 5 and 4 are not significant. Changes in calcium metabolism developed in a similar manner during secondary regeneration in thymectomized animals (group 6). On the 3rd day of the experiment, for instance, Ca^{45} incorporation into the injured femur fell to 84.6% of the level in intact control animals ($P = 0.05$), and then increased by the 15th day to 136% ($P < 0.001$). Changes in metabolism in the other bones of the animals of this group were similar in direction. The dynamics of incorporation of Ca^{45} was the same as in thymectomized rats (group 4).

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TABLE 1. Incorporation of Ca^{45} (pulses/min/mg ash) into Femur in Normal Animals, During Regeneration, and After Thymectomy ($M \pm m$)

Group	Days of experiment		
	3-rd	7-th	15-th
1	934±50 (5)	966±74 (5)	800±24 (6)
2	1 046±142 (4)	732±82 (4)	1120±107 (5)
3	1 020±37 (5)	1 044±70 (5)	1165±63 (7)
4	799±62 (8)		1077±27 (10)
5	806±44 (9)		1087±45 (9)
6	790±47 (8)		1016±44 (10)
7	895±14 (10)	1 133±43 (9)	940±64 (9)

Note. Number of rats in parentheses.

Repeated regeneration was disturbed if thymectomy was carried out in addition to primary trauma (group 7). In these animals the metabolic level in the injured femur was lowered on the 3rd and 15th days of the experiment to 87.7 and 80.6% respectively of its level in animals with secondary trauma (group 3). The difference is statistically significant ($P < 0.01$; $P < 0.05$). On the 7th day of the experiment in animals with secondary regeneration following thymectomy, activation of regeneration was observed: 155% compared with primary trauma (group 2), $P < 0.002$. This is similar to the result observed in normal secondary regeneration.

It can be concluded from these results (Table 1) that after thymectomy in rats depression of primary and secondary posttraumatic regeneration of bone tissue is observed, expressed as a decrease in the rate of Ca^{45} incorporation. This reaction is generalized in character and is exhibited both in the injured and uninjured parts of the skeleton. Suppression of the "anamnestic reaction" to preceding trauma is independent of whether the thymectomy is carried out at the time of primary or secondary trauma.

There are several possible explanations of the pathogenetic mechanisms of this phenomenon. After thymectomy, for instance, the "memory" of the primary regeneration process is disturbed, as a result of which the principal phenomenon of secondary regeneration—activation of mineral metabolism—does not take place.

On the other hand, it can be postulated that the thymus plays no part in the formation of the "anamnestic reaction" to preceding trauma, and activation of calcium metabolism does not take place during secondary regeneration through the direct or indirect inhibitory effect of thymectomy on mineral metabolism in calcified tissues. The possibility cannot be ruled out that mechanisms of regulation of mineral metabolism in bone are restored to normal in the case of regeneration combined with thymectomy at later periods than when thymectomy and regeneration occur separately. A further study of these matters is required.

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