

# EFFECT OF DNA ON THE HYDROXYPROLINE CONTENT OF REGENERATING BONE

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The author has previously demonstrated the stimulant effect of homologous total RNA on the course of osteogenesis [1]. The hypothesis has been put forward that acceleration of regeneration in animal bone tissue is associated with increased synthesis of collagen in the zone of injury under the influence of RNA.

It has been shown that the introduction of various RNA fractions into a cell-free system [3, 4, 6, 8] or a tissue culture [5, 9] may stimulate protein synthesis. So far as bone tissue is concerned, there is a report in the literature [9] of the effect of heterologous RNA of yeast origin on osteoblastic differentiation in tissue culture.

The object of the present investigation was to study collagen biosynthesis (as hydroxyproline) at various stages of regeneration of bone in vivo during the administration of homologous total RNA.

## EXPERIMENTAL

Experiments were carried out on 42 rabbits in which a defect of the radius was formed at operation. Total RNA was obtained from the regenerating bone by Georgiev's method [2]. The RNA solution was injected intramuscularly at a point distant from the fracture, in a dose of 0.3-0.5 to 2 mg per injection, daily from the 2nd to the 30th day. The control animals received injections of 0.5 ml physiological saline. Hydroxyproline was determined quantitatively by the method described in the literature [7]. The numerical results obtained were analyzed by statistical methods.

## EXPERIMENTAL RESULTS

Analysis of the mature lamellar bone of a rabbit showed that the normal content of hydroxyproline in the cortical layer of the bone is  $17.06 \pm 0.18$  mg (results of 15 determinations). In regenerating bone, as the table shows, the hydroxyproline content on the 7th-10th day in the young regenerating bone in the zone of the artificial) defect of the experimental rabbits was 40-50% higher than in the controls.

On the 15th and 20th days a significant difference was also found between the hydroxyproline content in the experimental group of animals. By the 30th day the hydroxyproline content in the regenerating bone of both groups of animals was essentially the same. Comparison of the hydroxyproline content in the regenerating bone of the experimental rabbits with the normal values shows that in the early stages of callus formation the level in the experimental animals was 20-50% higher than normal ( $P < 0.05$  and  $0.01$ ), whereas in the control animals it varied within near normal limits. Starting on the 12th day the hydroxyproline content in the experimental group was essentially indistinguishable from normal ( $P > 0.05$ ). This period coincided in time with the beginning of well defined calcification of the zone of the defect. Meanwhile, in the control animals the hydroxyproline content was below normal until the 20th day of observation ( $P < 0.05$ ), and the processes of calcification of the organic bone matrix were retarded. Not until the 30th day was the normal hydroxyproline content found in the regenerating bone of these animals ( $P < 0.02$ ).

Total homologous RNA thus induces increased collagen synthesis in the fracture zone and significantly reduces the time required for bone regeneration. The results of the author's previous structural morphological investigations also confirm this conclusion concerning the inductive effect of RNA on osteoblastic differentiation and the more rapid formation of specific bone structures [1].

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Hydroxyproline Content  
of Regenerating Bone (in mg/g  
Fresh Substance)

Time (in days)	n	Expt.	Control	P
5	8	27,45±5,83	19,64±1,44	<0,05
7	8	21,14±2,18	14,71±1,57	<0,01
10	8	20,21±1,17	13,45±0,74	<0,01
12	6	16,27±3,29	14,48±0,45	>0,05
15	6	17,93±1,44	13,09±1,42	<0,05
20	6	17,71±1,11	13,43±0,68	<0,01
30	8	18,94±1,53	15,77±1,42	>0,05

This stimulant effect of total homologous RNA on the regeneration of bone tissue is possible associated with the messenger RNA.

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