THE INFLUENCE OF BACTERIAL POLYSACCHARIDES UPON EXPERIMENTAL TISSUE REGENERATION

A. I. Braude, G. E. Vaisberg, T. I. Afanas'eva and N. I. Givental'

Laboratory for New Antibiotics, Department of Microbiology (Director Corresponding Member Academy of Medical Sciences USSR, Professor Z. V. Ermol'eva) Central Postgraduate Medical School (Director M. D. Kovrigina), Moscow
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In recent years studies concerning the biologically active bacterial polysaccharides were met with considerable interest, as parenteral administration of these substances increases the resistance of laboratory animals to infection and to ionizing radiation to a considerable degree. The influence of bacterial polysaccharides upon tissue regeneration is of great practical importance and requires further experimental study.

We studied the influence of two polysaccharides – cyine, prepared from Azotobacter xylinum, and a polysaccharide prepared from E. coli strain K-12 (preparation K-12).

METHOD

One of the polysaccharides was administered to white mice at the times stated below before or after an injury was inflicted on the foot of the animals under deliberately non-aseptic conditions by cutting through along about one half of its length on the lateral edge of the foot the soft tissues and the fourth and fifth metatarsal bones. Cyine was administered in a single dose of 200 μg by intraperitoneal injection, preparation K-12 in a dose of 5 μg by intravenous injection. The control animals were given injections of normal saline. Altogether 135 mice (90 experimental and 45 control mice) were used which were killed consecutively in the course of 25 days.

RESULTS

In the control mice the injury is quickly followed within 20-30 min by massive edema of the foot, the tibiotarsal joint, the greater part of the shin, and sometimes the region of the knee joint and the adjacent part of the thigh. The marked edema accompanied by intensive hyperemia of the foot persisted for 4-5 days. In the majority of control mice the region of the wound was covered during the first seven-eight days by a purulent scab surrounded by a broad zone of hyperemia. Histological and microbiological investigation revealed during the first four-five days the presence of a great number of organisms in the wound.

On the sixth-seventh day fibroblasts and the first argyrophile fibers appeared in the region of the wound among extensive infiltrates consisting of histiocytes and lymphocytes. Along the line of the operative incision the cellular detritus persisted; no epidermization could be observed (Fig. 1, a). Resorption of the superficial layer of the facing ends of the transected bones could be observed. Along these surfaces poorly differentiated connective tissue cells could be seen. On the tenth day no typical scar tissue had yet formed in the wound region (Fig. 2, a). In some parts cartilage cells appeared along the surfaces of the transected bones. On the fifteenth day a well developed layer of hyaline cartilage could be seen on the ends of the transected bones in the majority of control animals; this layer was surrounded by rather thick bundles of collagenous fibers and fibroblasts. On the 20th-25th day the bone fragments became consolidated under formation of a callus; in the central part of the callus the hyaline cartilage persisted. Even on the 25th day considerable infiltrates consisting of histiocytes and lymphocytes could be found in the scar tissue.

In the experimental animals the edema quickly disappeared within 12-24 hours if cyine was injected 2-24 hours after the operation. If it was injected 24 hours before the operation a somewhat lesser edema developed in the injured foot than in the control animals. Injection of K-12 exerted no appreciable influence upon the develop-

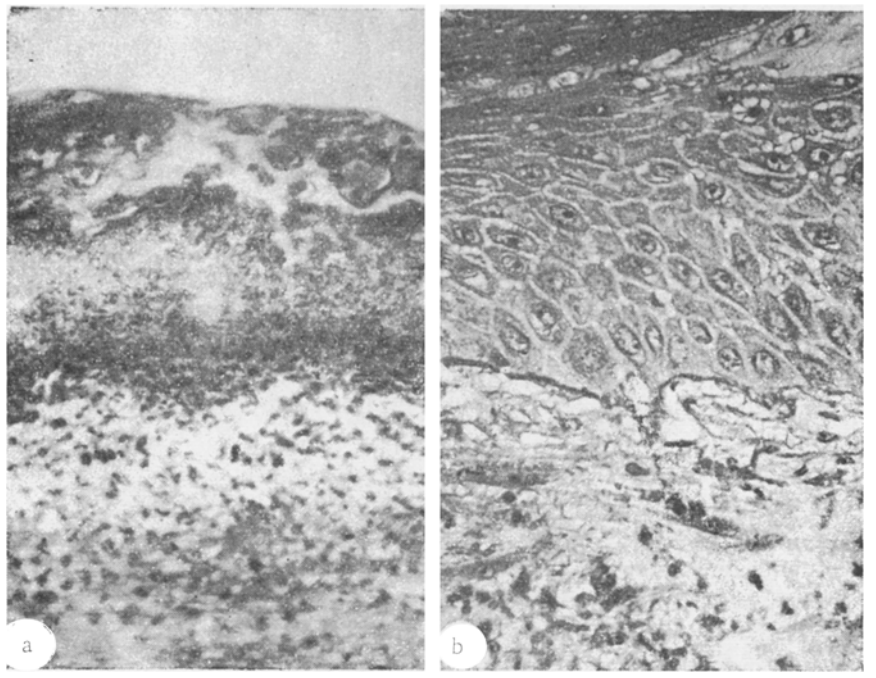

Fig. 1. State of the site of incision in a control mouse (a) and an experimental mouse (b) on the seventh day of the experiment. In the control animal a mass of cellular detritus can be seen; in the experimental animal epithelization of the cicatrizing wound can be seen. Staining with iron-hematoxylin and aniline blue. Magnification × 300.

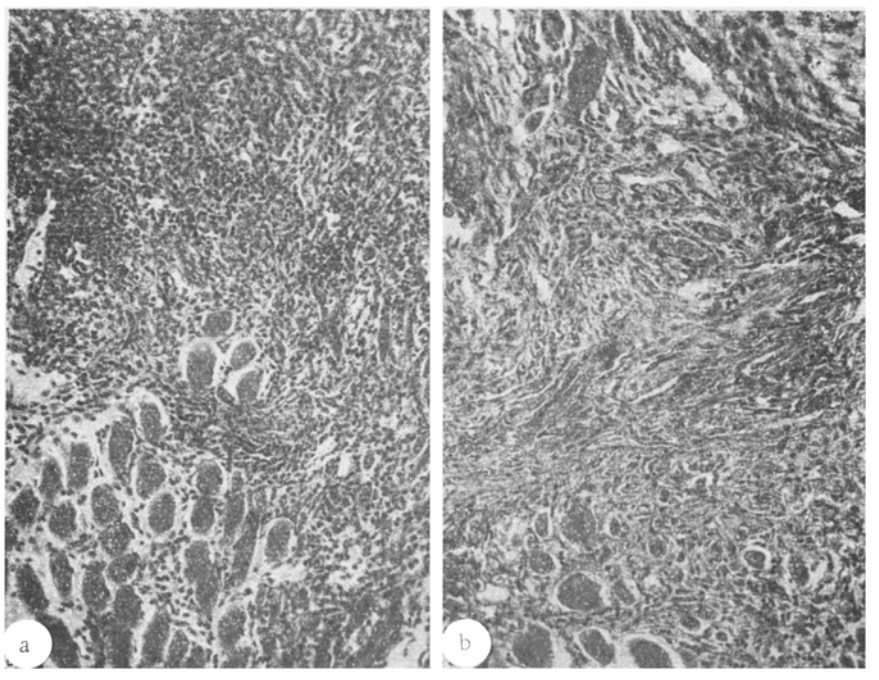

Fig. 2. The state of the wound in a control mouse (a) and an experimental mouse (b) on the tenth day of the experiment. In the control animal an intensive inflammatory reaction, in the experimental animal, the presence of scar tissue can be seen. Van Gieson stain, magnification × 160.

ment of edema. Microscopical investigation of preparations and selective cultures revealed that the wounds of animals treated with either cyine or preparation K-12 before the operation or within the first day after the operation were practically free of microorganisms. If one of the two polysaccharides was administered before the operation or in the first days after the injury the inflammatory infiltrate was less extensive.

On the seventh day of the experiment a considerable number of newly formed collagenous fibers could be seen in seven out of ten mice which had been given injections of cyine and four out of nine mice which had been given injections of the preparation K-12. The greatest part of the incision was covered by regenerating epidermis; a basal membrane appeared along its internal border (Fig. 1, b). Ten days after the operation almost no residue of necrotic muscle fibers could be seen in the region of the wound in five out of 11 mice treated with cyine and three out of nine mice treated with the preparation K-12; the formation of scar tissue could be observed (Fig. 2, b) and along the surface of the transected bones a broad zone of hyaline cartilage could be seen. On the 20th day bone tissue developed on the ends of the transected bones in five out of eight mice treated with cyine and three out of seven mice treated with the preparation K-12; the clefts in the bone were filled with elements of the red bone marrow, a phenomenon which in the control animals could be observed only on the 25th day. In mice killed on the tenth day of the experiment the scars were as a rule much narrower than in the control mice.

Administration of cyine or preparation K-12 was never followed by a deterioration in the state of the wound or delayed regeneration. Consequently the unfavorable influence upon the regeneration and the state of the tissues reported by some authors [3,4,5] must not be regarded as characteristic for all polysaccharides, in particular not for those which stimulate resistance to infection. Above we remarked that following the administration of cyine and the preparation K-12 the wound regeneration took place in a considerable part of the experimental animals more rapidly than in the control animals. At the same time we had shown earlier that a single cyine injection has no influence upon the speed of encapsulation of foreign bodies. In consequence the more rapid regeneration found on the model of traumatic inflammation can hardly be regarded as the consequence of the direct influence of polysaccharides upon those elements which secure the proliferative processes. It seems that in the case of traumatic inflammation the more rapid regeneration is enhanced (among the factors studied so far) by the very rapid liquidation of the wound infection and the more rapid lysis of the necrotic tissues due to the activation of the phagocytic elements [1], and improved blood supply in the wound region due to the absence of edema [2], which latter causes compression of small veins.

SUMMARY

There was no unfavorable effect noted from parenteral administration of polysaccharides obtained from Az. xylinum (cyine) or from E. Coli K-12 (preparation K-12) on the tissue regeneration in foot trauma inflicted on mice. In a number of cases tissue regeneration in animals which received cyine or K-12 preparation was even quicker than in control animals. Evidently, this results from the complex of favorable shifts, such as a more rapid elimination of the infective agents, activation of macrophagic elements and removal of the factor of venous compression by edema fluid.

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
