

MORPHOLOGICAL AND HISTOCHEMICAL CHANGES IN THE
CONNECTIVE TISSUE OF THE SKIN OF NEWBORN ALBINO RATS
FROM MOTHERS WITH PANCREATIC DYSFUNCTION

V. P. Zhuk

Department of Histology and Embryology (Head, Professor M. Ya. Subbotin),
Novosibirsk Medical Institute

(Presented by Active Member AMN SSSR, N. A. Kraevskii)

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The development and histogenesis of the fetus are closely associated with the state of the maternal hormonal systems and, in particular, with the state of the pancreas [2, 5-8]. Most research in this field, however, is concerned with the general problems of fetal development and not with the influence of the hormone of the maternal pancreas on the formation of the embryonic connective tissue.

We have studied the changes in the connective tissue of the skin of embryos and newborn albino rats from mothers with experimental pancreatic dysfunction. Connective tissue is known to react distinctively to changes in the carbohydrate metabolism.

EXPERIMENTAL METHOD

In the first series of experiment 15 rats received a daily subcutaneous injection of 0.04 ml insulin (40 units/ml) for 9-55 days. During the experiment some rats had three litters of young; altogether 90 young rats came under observation. In order to examine the action of insulin on the mother, the blood sugar of all the experimental rats was determined by the method of Hagedorn and Jensen. The mean blood sugar in the animals receiving insulin for 30 days or more was 57-60 mg%.

In the second series 10 rats received two subcutaneous injections, each of 1 ml of a 5% solution of alloxan, equivalent to 200 mg/kg body weight of alloxan (mean weight of experimental animals 250 g). Observations were made on 38 young rats. In order to discover whether the rats developed signs of diabetes, the sugar concentration in the urine was determined by means of an Althausen's saccharimeter. Its average value was 6-8%. All the rats had marked polyuria.

In the control series young animals from five litters of intact rats were investigated.

The material was fixed in 12% formalin and embedded in paraffin wax and celloidin. Sections were stained with hematoxylin and eosin, Heidenhain's iron-hematoxylin, azure-II-eosin, and by Mallory's method. Gomori's method of impregnation with silver nitrate was also used. We also used histochemical methods for detection of mucopolysaccharides—Hale's method, toluidine blue, the PAS reaction, and alcyan blue. Sections were treated with hyaluronidase for 18 h (lyophilized hyaluronidase manufactured by the firm Spofa of Prague), sulfated by the method of Moore and Schoenberg, and methylated (to block sulfate groups). Acetylation was carried out (to block 1,2-glycol groups by the method of McManus and Keyson) and aldehyde groups were blocked by Culling's method as controls of the PAS reaction.

EXPERIMENTAL RESULTS

The results of the experimental and control series of investigations corresponded to the morphological and histochemical characteristics of the connective-tissue stroma of the skin of newborn albino rats as described in the literature [1].

When we studied the dermis of the newborn rats from mothers receiving insulin for 30 days and more, we observed deviations from normal even when the ordinary histological methods of examination were used. The difference, however, was slight; after staining by Mallory's method the collagen and reticulin fibers of the dermis were

stained less intensively. Histochemical examination revealed a considerable decrease in the Hale-positive substance in the dermis of the experimental young rats compared with normal (Fig. 1): the staining was pale (Fig. 2). Staining with toluidine blue gave a strongly positive metachromasia of the ground substance in all the layers of the dermis, practically indistinguishable from normal. There were fewer mast cells than normally, and they contained fewer granules. In sections treated by the PAS method, the difference from normal was small.

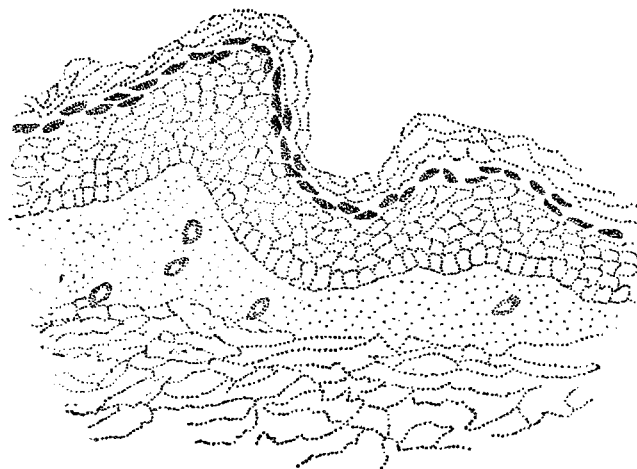


Fig. 1. Skin of a control newborn rat. Stained by Hale's method. Magnification: Objective 8X, eye-piece 10X.

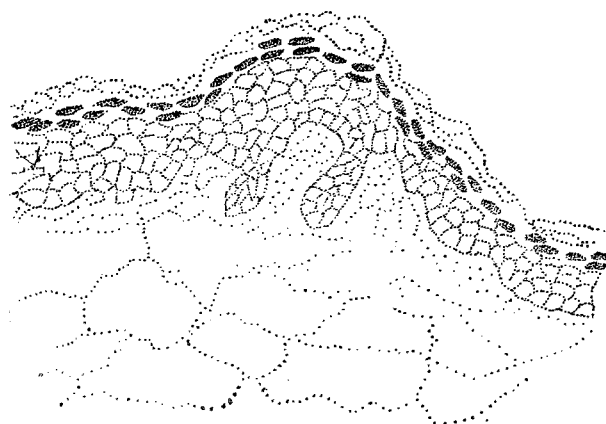


Fig. 2. Skin of a newborn rat from a mother receiving insulin for 38 days (hypoglycemia). Stained by Hale's method. Magnification: Objective 8X, eye-piece 10X.

A different picture was seen in the dermis of the newborn rats from mothers receiving two injections of alloxan during pregnancy. No abnormalities were found by the use of the ordinary histological methods of staining. The results of histochemical treatment of the sections were opposite to those obtained in the experiments after administration of insulin. The reaction with colloidal iron gave a somewhat lower intensity of staining of the ground substance (Fig. 3) by comparison with normal. Staining with toluidine blue showed an ill-defined metachromasia of the ground substance in this series of experiments. No difference from normal was seen in this respect. The mast cells were more numerous than normally, and their metachromatic granules were very conspicuous; sometimes extracellular granules were seen. In sections treated by the PAS reaction the ground substance was stained much more brightly than normal.

It follows from these findings that from a qualitative point of view the chemical structure of the ground substance of the connective tissue from the skin of the experimental young rats was unchanged. The changes which

were observed were quantitative, and were concerned with the mucopolysaccharide content. After administration of insulin to the experimental animals, the mucopolysaccharide content of the skin of the newborn rats and embryos was slightly lowered, and after administration of alloxan it was raised.

At first glance, these results appear paradoxical. They can be understood, however, if it is remembered that insulin does not pass through the placenta for a considerable period during pregnancy [3]. The placenta becomes permeable to insulin only after the 18th-19th day of pregnancy. As a result of the hypoglycemia developing in the

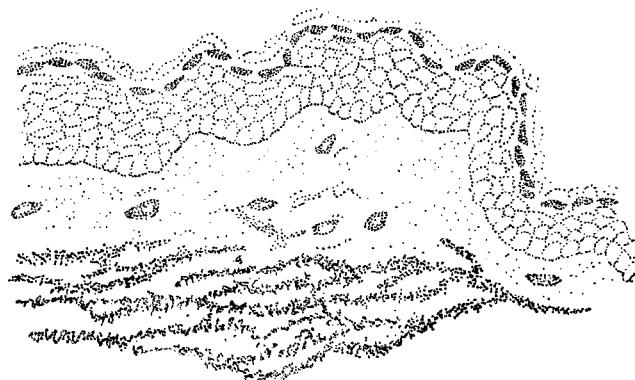


Fig. 3. Skin of a newborn rat from a mother with alloxan diabetes. Stained by Hale's method. Magnification: Objective 8X, eye-piece 10X.

mother, less sugar reaches the fetus than normally. This, in our opinion, accounts for the decrease in the mucopolysaccharide content in the skin of the newborn rats. At the same time, alloxan causes no changes in the cells of the islets of Langerhans in embryos [4]. Hence, the signs of alloxan diabetes in the mother are not accompanied by diabetes in the fetus. The increased content of mucopolysaccharides in the dermis of the young rats from mothers with diabetes is fully explained by the increased supply of sugar received by the fetus.

SUMMARY

There were changes of the quantitative content of mucopolysaccharides in the skin connective tissue of embryos and newborn rats from mother rats with pancreatic dysfunction. In experimentally induced hypoglycemia there is a reduction of the mucopolysaccharide content in the ground substance of the skin connective tissue; it rises in experimental alloxan diabetes. There were no other changes of the ground substance of the connective tissue of the skin in the experimental rats.

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