## The Localization of $^3$H-Corticosterone in Mast Cell Granules by Electron Microscopic Autoradiography

It has been described in a previous publication[1] that isotope-labelled corticosterone electively incorporates into the mast cells of the lymphatic organs of mice. At the beginning it can be found in the thymus in PAS positive cells, and later, after 6 h, also in the mast cells of lymph nodes. A more exact localization of corticosterone is made possible by the application of the autoradiographic method on ultra-thin slides, evaluated by the electron microscope. This allows a better approach to the interpretation of the mast cell effect of glycocorticoids[2-6]. In this study, therefore, investigation is aimed at the localization of $^3$H-corticosterone within the mast cells.

In the experiments, corticosterone-1, 2-$^3$H was given to BALB/C adult male mice. The specific activity of corticosterone was 945.9 mCi/m*M*. Labelled corticosterone was diluted with unlabelled, suspended with Tween 80 and injected i.p. Thus 1 mg/30 g animal corticosterone was given, $^1/_{10}$ of which was active. The given activity was 5 $\mu$Ci/g body weight. Six animals were treated in this manner. Material was taken from 2-2 animals 1, 3 and 6 h after treatment. The apex of the heart of the etherized animal was cut off and a cannula inserted into the aorta. Dextran solution at 37 °C, then buffered glutaraldehyde, were perfused through the cannula. The thymus, spleen and axillary lymph nodes of the animals were removed and fixed in glutaraldehyde for a further 2 h. Postfixation with $OSO_4$ was followed by the usual procedures applied in electron microscopic examinations. The slides were covered with Kodak or Ilford L4 emulsion, processed after 2–6 months' exposure, then contrasted and studied with the JEM 6C electron microscope.

In mice – as opposed to rats[7,8] – there are fewer mast cells in the thymus; they are, on the other hand, in great abundance in the spleen[9]. Thus the spleen proved to be most suitable for ultrastructural investigations. Figure 1 shows a mast cell in developmental form. The granules are small and the ergastoplasm satisfactorily developed. The precipitation of silver grains can be observed above the mast cell granules. Figure 2 shows 2 fully developed mast cells. Here the precipitation of the grains is clearly seen above the typical mast cell granules. No grains were found that precipitated as the effect of background radiation. Apart from mast cells, grains were found also above some lymphocytes; hence corticosterone is incorporated also in these cells.

These investigations support previous observations, according to which corticosterone incorporates into mast cells. At the same time they demonstrate that corticosterone becomes a component of the granule in the mast cell. This phenomenon was discussed in a previous publication[1].

Since this publication is of a preliminary character, we do not wish to deal at present with the role of corticosterone in the granules of the mast cell, but will discuss the subject in detail upon the termination of the experiments.

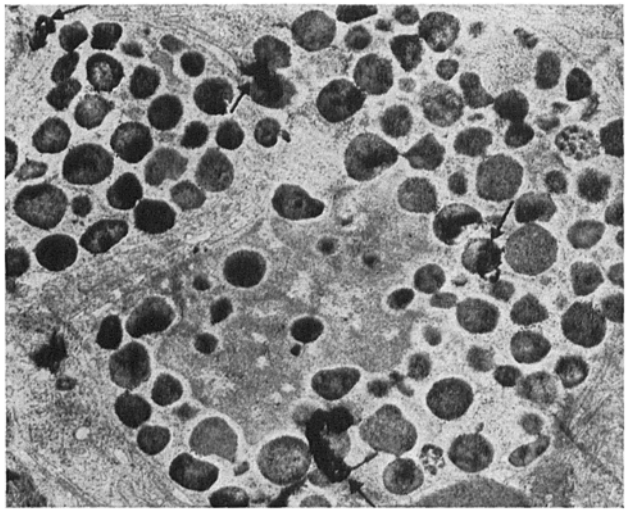

Fig. 1. Developing mast cell in mouse spleen. Grains indicated by arrows. × 9000.

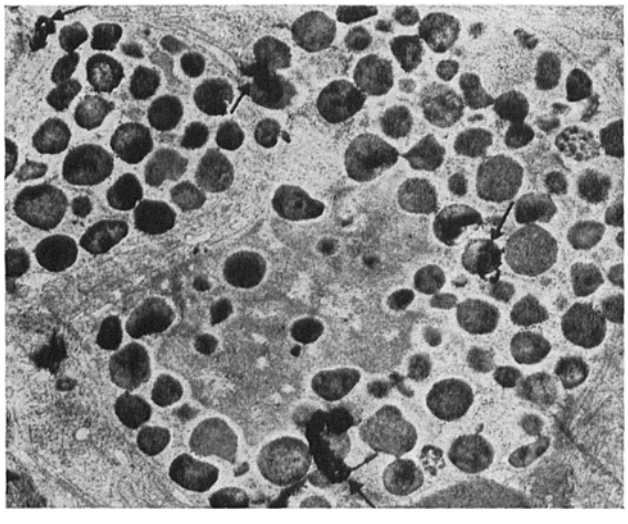

Fig. 2. Mature mast cell in mouse spleen. Grains indicated by arrows. × 10,200.

*Zusammenfassung.* Der Einbau von 1,2-$^3$H-Corticosteron in die Mastzellen der lymphatischen Organe wurde bei Mäusen untersucht. Bei gleichzeitiger Anwendung von Autoradiographie und Elektronenmikroskopie konnte das Corticosteron in den Mastzellen nachgewiesen werden.

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