and genetic transfer. Among others, '*Aspercillium*' or '*Penigillus*' strains, the results of *Aspergillus-Penicillium* protoplast fusion, would be especially interesting from both theoretical and practical points of view. Results of attempts at interspecific complementation will be published elsewhere.

Zusammenfassung. Es gelang, die intraspezifische Protoplastfusion von *Aspergillus flavus*, *A. nidulans*, *A. niger*, *Penicillium frequentans* und *P. ramigena* mit grosser Häufigkeit zu verwirklichen. Die Fusionshäufigkeit wurde durch Komplementation von Mangelmutanten festgestellt.

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Electron-Microscopic Investigation on Peroxisomes in the Epithelia of Mice Gall-Bladder

The term 'peroxisome' or 'microbody' is understood as a special type of cytoplasmic organelle characterized biochemically by the association of one or more oxidases producing hydrogen peroxide with catalase, which destroys the hydrogen peroxide[1–3]. The name 'microbody' was first proposed by Rhodin in 1954[4] to describe roughly spherical singl-membrane limited particles in the proximal convoluted tubular cells of the mouse kidney 0.3–0.5 nm in diameter containing a fine-grained moderately electron dense matrix. Subsequently, analogous organelles were detected by Rouiller et al.[5,6] in hepatic parenchymal cells of the rat. The microbodies of the liver cells have an inner electron density or nucleoid. These initial studies have been confirmed and extended. Further studies indicated that microbodies or peroxysomes were constituent organelles of animal cell[7–14]. In the course of electron-cytochemical investigation, peroxisomes were found in the epithelial cells of mice gall-bladder.

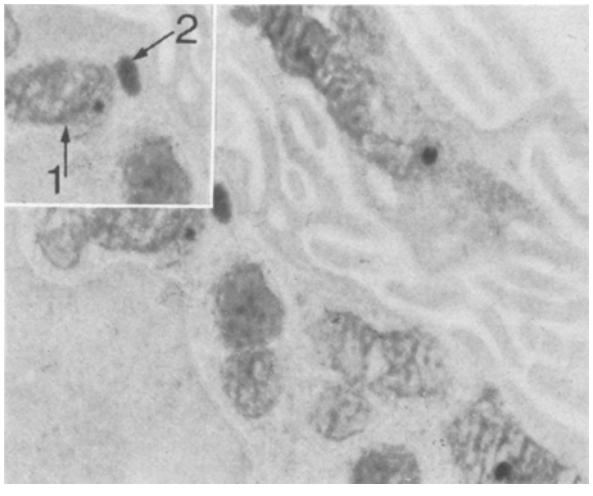

Fig. 1. Fragment of epithelial cell of mice gall-bladder. Mitochondria (1). Peroxisome (2). ×23,000.

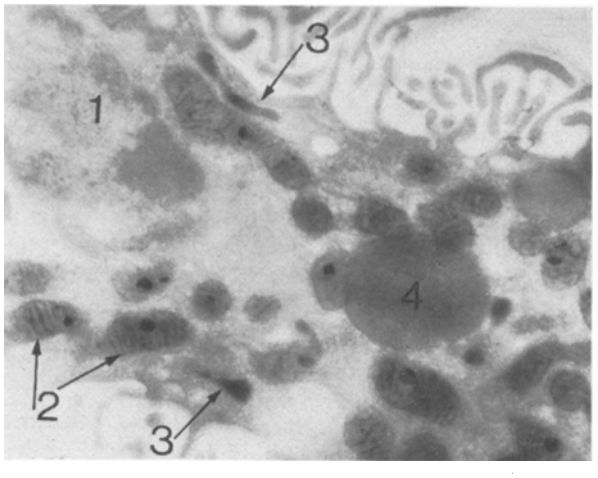

Fig. 2. Epithelial cell of mice gall-bladder. Nucleus (1). Mitochondria (2). Peroxisomes (3). Lipid drop (4). ×12,000.

Materials and methods. Normal male albino mice (18–20 g) were anesthetized with ether and small pieces of the gall-bladder were removed, fixed in ice-cold 4% glutaraldehyde in 0.1 *M* phosphate buffer, pH 7.2 for 6 h at 0 °C. The specimens were preincubated for 5 min in *Tris*-HCl, pH 9.5 containing 3.5% sucrose (v/v) and incubated for cytochemical demonstration of the peroxidatic activity of catalase in a modified DAB-medium[15] pH 9.5 at 37 °C for 3 h[16,10]. The final medium contained 20 mg DAB (3,3′-diaminobenzidine tetrahydrochloride, Chemapol, Praha), 0.2 ml 1% H_2O_2, 9.8 ml *Tris*-HCl, pH 9.5 and 3.5% sucrose.

The following control experiments were carried out: a) omission DAB, b) omission H_2O_2, c) heating at 100 °C for 10 min. After the incubation, the specimens were washed in 7% sucrose solution and postfixed in unbuffered 4% O_sO_4 for 15 h at 0 °C. They were dehydrated through graded concentrations of acetone and embedded in Epon

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812. Thin sections were cut on a LKB II ultramicrotome. Unstained and stained[17] sections were examined in a Hitachi HU IIB electron microscope.

Results. Our morphological observations are in agreement with the published results on the ultrastructure of epithelial cells of the gall-bladder[18, 19]. Intensively stained structures are found in the cytoplasm of epithelial cells of mice gall-bladder (Figure 1). These structures are single-membrane bound and have a round, oval or rod-like shape. The diameters of round organelles measure from 0.2 to 0.3 nm. The reaction product within the organelles shows a granular texture. No crystalloid core is found in the matrices. These organelles are found throughout cytoplasm of the epithelial cells. It is not possible to demonstrate a continuity of the limiting membranes of our organelles with the smooth surfaced endoplasmic reticulum. The membranes and crystes of mitochondria are stained. This is most probably due to peroxidatic activity of cytochrome oxidase[20] or cytochrome c[21]. The staining of some lysosomes of epithelial cells is probably due to peroxidatic activity of lipid peroxides[22] or hemes and metals[23].

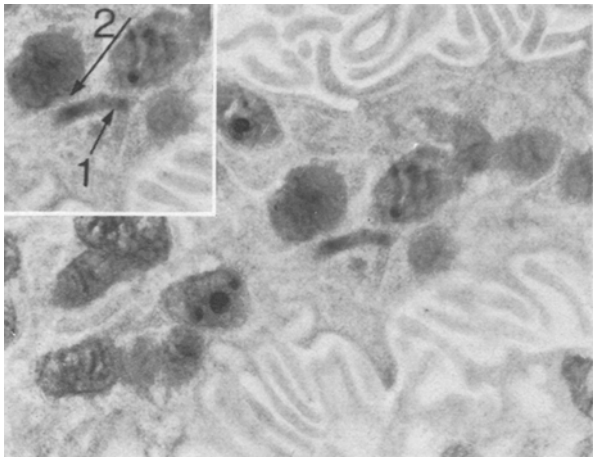

Fig. 3. Fragment of epithelial cell of mice gall-bladder. Peroxisome (1). Mitochondria (2). ×21,000.

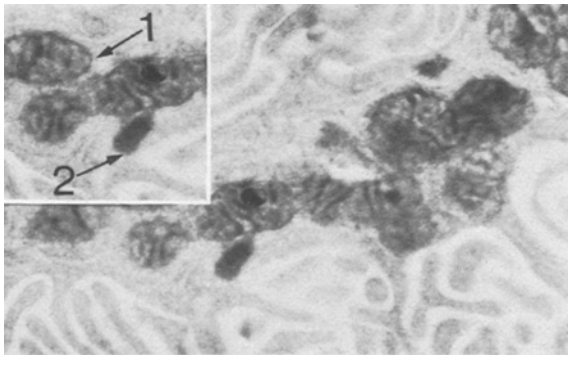

Fig. 4. Fragment of epithelial cell of mice gall-bladder. Mitochondria (1). Peroxisome (2). ×17,000.

No product reaction was observed in our organelles if the tissue was incubated in the medium without DAB or after heating. A very weak reaction remained if the tissue was incubated in DAB solutions without H_2O_2.

Discussion. The organelles described in the epithelial cells of mice gall-bladder are similar to peroxisomes found in many animal cells[7]. We think that our results point to the fact that the organelles described may be peroxisomes. The function of peroxisomes in epithelial cells of gall-bladder remains to be elucidated. The role of the organelles in animal cells remains obscure. But it is possible, as we suggested previously[24, 25], that one of their functions is antimicrobial. Probably, peroxisomes of gall-bladder epithelia are antimicrobial organelles.

Summary. Electron-cytochemically peroxysomes were found in the epithelial cells of mice gall-bladder. The possibility is discussed that these are antimicrobial organelles.

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Ultrastructural Cytochemistry of Peroxidase and Acid Phosphatase in Mice Maturing Eosinophils

Materials and methods. Observations were made on eosinophils from mice bone marrow. Small pieces were fixed in 4% glutaraldehyde in 0.1 *M* cacodylate buffer, pH 7.4 for 5 h at 0–4 °C. After a wash in 0.1 *M* cacodylate buffer, pH 7.4 containing 7% sucrose for 18–20 h in cold, the specimens were incubated for peroxidase in the modified Graham-Karnovsky medium[1] for 10 min at 37 °C. The incubation medium composed of 0.01% H_2O_2, 7% sucrose and 20 mg 3, 3'-diaminobenzidine tetrachloride (DAB) in 10 ml 0.1 *M Tris*-buffer, pH 7.6. After the incubation, the specimens were washed in 7% sucrose for 5–7 min and postfixed in 2% OsO_4 for 20 h, dehydrated through graded concentrations of acetone and embedded in Durcupan.

Controls for activity of peroxidase included: 1. omitting DAB from the incubation medium, 2. omitting H_2O_2 from the incubation medium (in this case the specimens were incubated for 10 min and 60 min), 3. preincubation with absolute acetone for 40 min – 24 h following the incubation in the complete medium and the medium without H_2O_2, 4. preincubation with the following inhibitors:

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