PHA auftraten, fanden sich in den Ausstrichpräparaten auch nicht.

Die Mesothelzellveränderungen, die qualitativ denen nach der Injektion von nativem PHA entsprechen, waren jedoch sehr viel geringer ausgeprägt. Sowohl Zytoplasmavakuolen als auch Siegelringzellen kamen nur selten vor. Dagegen führte auch die Applikation von denaturiertem PHA zu einer Proliferation von Mesothelzellen.

Durch die Hitzedenaturierung verliert das PHA die Fähigkeit, eine messbare Hyaluronsäureproduktion zu stimulieren. Die morphologischen Veränderungen der Mesothelzellen sind nur gering. Da durch die histochemischen Untersuchungen eine Hyaluronsäuresynthese in den multivakuolären Mesothelzellen und Siegelringzellen wahrscheinlich gemacht werden konnte (Mohr et al.[2d]), ist zu vermuten, dass es nur zu einer geringen Hyaluronsäureproduktion unter dem Einfluss des denaturierten PHA gekommen ist, die mit der angewandten Methode, bei der die unterste Grenze der noch messbaren Hyaluronsäurekonzentration bei 5 mg/100 ml liegt, nicht ermittelt werden konnte.

Summary. After the i.p. injection of heat denatured PHA, mesothelial cell changes are similar to those found after the application of natural PHA. While vacuolization of mesothelial cells and signet ring cell formation is strongly marked after the injection of natural PHA, this reaction is minor if denatured PHA is used. Furthermore, the peritoneal fluid does not contain hyaluronic acid in detectable amounts.

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Mitochondrial Changes in the Guinea-Pig Muscle after Envenomation with *Vespa orientalis* Venom

Most wasps sting and kill their prey by paralysis[1]. The prey of wasps consists mainly of insects. Reports concerning wasp stings in humans indicate that hymenoptera stings result in an allergic response manifested by urticaria, angiodema, syncope, vascular collapse and respiratory obstruction[2,3]. Neurological symptoms due to wasp stings are rare but, when they do occur, they complicate anaphylactic shock[2-5]. The site of action of the paralysing venom is as yet unidentified: neuromuscular junction, peripheral nerves and the central nervous system have been incriminated[6].

In the present study, the ultrastructure of muscle and neuromuscular junction following envenomation with *Vespa orientalis* venom has been investigated.

The venom was milked from ether-anesthetised wasps by pressure on the upper abdomen and then by collecting the venom into a capillary. The venom was dried in a lyophyliser. 100 µg of the venom were dissolved in 1 ml of saline, and 0.1 ml was injected into the lower lip of a guinea-pig. A wedge dissection of the injected area was cut 3 h after the injection, and the specimen was divided into 2 halves. One half was taken for optical microscopic study, and the other for electron microscopic study.

Microscopic examination revealed mild edema of the sub-epidermal tissue, with an inflammatory exudate composed mainly of polymorphonuclear leucocytes. Under the electron microscope the inflammatory reaction was seen in the sub-epidermal tissue. The mitochondria of fibroblasts and endothelial cells were normal. Significant changes were observed in the mitochondria of the striated muscle and the neuromuscular junction. The cristae gave the impression of having collapsed, agglutinating in the middle or adherring to the inner membrane of the mitochondrion. Clumped cristae dissolved into an amorphous granular material in which remnants of the mitochondrial

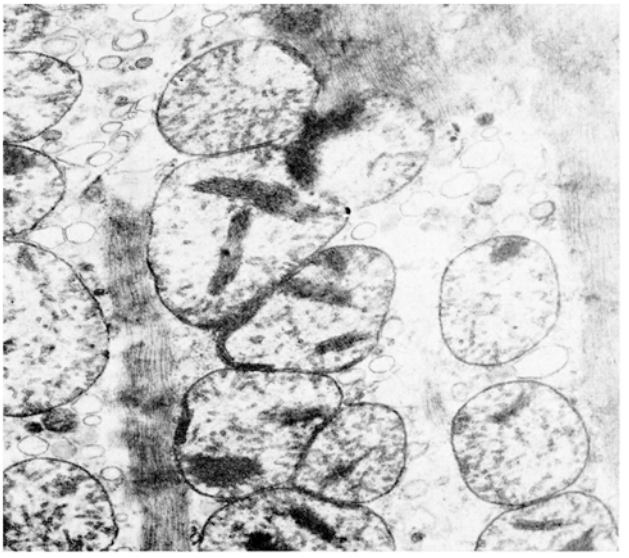

Fig. 1. Group of mitochondria near neuromuscular junction with severe alteration of inner structure.

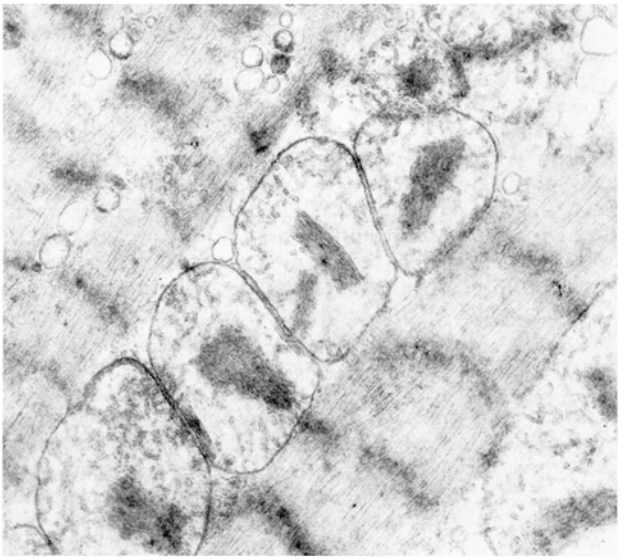

Fig. 2. Higher magnification of mitochondria with clumping and dissolution of mitochondria.

cristae were seen (Figures 1 and 2). In animals injected with saline only, no mitochondrial changes were observed. A preliminary study with rat liver mitochondria examined with a oximeter after addition of wasp venom registered an immediate arrest of oxidative phosphorilation, also indicating a functional lesion. The effect of hymenopterae venom on the function of mitochondria has recently been reviewed by HABERMAN[6]. As numerous lipolitic enzymes are present in the venom, functional and morphological mitochondrial changes are expected.

In numerous conditions, alterations in the internal structure of muscle mitochondria have been reported. These were observed in human muscle diseases[7], in human rhabdomyoma[8-10] under environmental conditions influenced by acid pH[11], under the influence of estrogenic hormones[12], and in vitamin E deficiency[13,14]. Lately changes in mitochondrial ultrastructure in Nickel-Sulfide-induced rhabdomyosarcoma was reported[15].

It seems, therefore, that muscle mitochondria exposed to many different noxious agents manifest morphological changes. The observation that wasp venom cause both morphological and functional changes in muscle mitochondria may explain the paralysing effect of the venom.

Résumé. Du venin de *Vespa orientalis*, injecté dans la lèvre inférieure du cobaye, a donné une reaction inflammatoire non spécifique mais aiguë. Dans les mitochondries du muscle contigu strié de graves changement apparurent. Les cristae se sont agglutinées à mi-hauteur et les mitochondries agglutinées ont été disloquées et prirent l'aspect d'une matière granulaire amorphe. Après l'arrêt immediat de phosphorylation oxydative on constate que la lésion mitochondriale pourrait expliquer l'effet névrotoxique du rénin de la guêpe.

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Ultrastructural Response of the Neural Plate Cells of Chick Embryos to Dithiodiglycol

It has been reported[1,2] that in amphibians the morphogenetic movement leading to the closure of the neural tube was linked with the reversible denaturation of a fibrous protein. It seems that as –SH groups in the protein are being oxidized into –SS– groups the fibrous molecule is transformed into a globular one and changes in cell shape[3] thus follow. This valuable hypothesis led to a series of studies of the effect on morphogenesis, in a variety of biological species, of a number of sulphydryl reagents including mercaptoethanol, a strongly reducing sylphydryl reagent[3-6], and dithiodiglycol, its oxydation product[3,7].

Dithiodiglycol was shown to be an efficient inhibitor of neurulation in amphibians[8] and in chick embryos[7]. This oxidizer of –SH groups was also reported[9] to favor the isolation of the mitotic apparatus of sea urchins through its stabilizing effect on the apparatus, and to inhibit cell division in cleaving amphibian eggs[10] by opposing the regression of the achromatic figure.

The purpose of this note is to give an account of the ultrastructural response of the neural plate cells of chick embryos when neurulation is inhibited with dithiodiglycol.

Abbreviated materials and methods. Chick embryos were explanted at stage 7 + or 8 — of Hamilton's table and cultivated for 5.5 h at 38 °C on Spratt's medium, but with $10^{-3}M$ dithiodiglycol added. Control embryos, growing on standard media, were cultivated, fixed and embedded at the same time as the treated ones.

All embryos were fixed first in phosphate buffered glutaraldehyde and then in osmium tetroxyde. Blocks were embedded in epon or araldite and sections double stained with uranyl acetate and lead.

Principal observations. Control embryos were comparable to those previously described in detail[11].

In experimental animals, thin sections cut transversely in front of the first pair of somites showed a wide open nervous system still appearing in the form of a plate. The cellular contours were identical to those observed in controls. After a 5.5 h treatment all cellular organelles, save microtubules, appeared entirely comparable to those seen in control embryos.

Microtubules were apparently the only structures affected by the treatment. In control embryos microtubules follow a straight line or occasionally show a slightly wavy and smooth undulatory path (Figure 2). Under dithiodiglycol treatment similar outlines were

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