tract of incubation mixture, the identification of individual metabolites was carried out on a broader scale on the basis of their chromatographic properties and recrystallization to a constant specific radioactivity, respectively.

The amounts of metabolites formed by 100 mg of each tissue are shown in the Table. It is evident that 7α-hydroxylase of DHA is present in both testis and epididymis and that its activity is practically identical in both cases. However, the total metabolic activity is higher in the testis, which results from higher decrease of ^{14}C-DHA. There is a very active 17α-hydroxysteroid:dehydrogenase in the testis, as demonstrated by formation of a high yield of 5-androstene-3β, 17β-diol and testosterone. Due to the activity of this enzyme, 5-androstene-3β, 7α, 17β-triol prevailed among the 7-hydroxylated metabolites of DHA in the samples incubated with the testis. In epididymis, the dehydrogenases of 3β- and 17β-hydroxysteroids are weakly active, therefore predominantly 7α-hydoxydehydroepiandrosterone was there found. The formation of epimeric 7β-hydroxy-DHA in samples incubated with both testis and minced epididymis suggests either the presence of DHA: 7β-hydroxylase or 7α- and 7β-hydroxysteroid:dehydrogenase or 7α/7β epimerase in these tissues. However, the yield of 7β-epimer was essentially lower than that of 7α-hydroxy DHA.

The total amount of 7-hydroxylated metabolites of DHA in incubation mixtures with testis is relatively high, i.e. approx. 70% of that of testosterone formed from DHA under the same conditions. This fact agrees with the results reported by INANO et al.[3]. In their experiments, 7α-hydroxyandrostenedione was often obtained in higher yield from androstenedione than testosterone itself by microsomal fraction of mature rat testes supplemented with NADPH. They postulated a physiological role of the hydroxylated metabolite in relation to testicular endocrine function and/or to regulation of androgen production and its secretion. However, a definite comparison of our results with those of INANO et al. is difficult with respect to a different substrate, origin of the tissue and incubation conditions (cofactors). In our experiments, when we incubated both [4-^{14}C]androstenedione and [4-^{14}C]testosterone with minced human testis under the same conditions as we did with ^{14}C-DHA, 1 compound in the fraction of polar metabolites of testosterone and 2 compounds among polar metabolites of androstenedione, respectively, were found, the chromatographic mobilities of which corresponded to 7α-hydroxytestosterone and 7α-hydroxyandrostenedione. Owing to their very small yield, they could not be identified further. Unlike the rat testis, in the human testis dehydroepiandrosterone is 7-hydroxylated preferentially to androstenedione and testosterone. The finding of 7-hydroxylase activity in both testis and epididymis confirms, however, the non-specifity of this enzymatic system already known before[2,5]. The problem of physiological importance of 7-hydroxylation process in human testes remains still unexplained.

Formation of [4-^{14}C] dehydroepiandrosterone metabolites in healthy human testis and epididymis in vitro

Metabolite	Testis (dpm)	Epididymis (dpm)
DHA recovered	118,535	371,345
5-Androstene-3β, 17β-diol	108,768	984
Testosterone	15,506	315
4-Androstene-3, 17-dione	1,393	28
5-Androstene-3β, 7α, 17β-triol	9,672	469
7α-Hydroxy-DHA	826	10,747
7β-Hydroxy-DHA	238	1,223
Total of 7-hydroxylated metabolites	10,736	12,439

In each experiment 100 mg of tissue was incubated with 0.2 μCi of [4-^{14}C] dehydroepiandrosterone. The values are averages of 2 parallel experiments. The values are corrected in respect to the blank, but not to methodological losses.

Zusammenfassung. Zerhackte menschliche Hoden und Epididymis wurden mit Dehydroepiandrosteron ohne Kofaktoren bebrütet und es wurden unter anderem 7α- und 7β-Hydroxydehydroepiandrosteron und 5-Androsten-3β, 7α, 17β-triol charakterisiert. Die Ausbeute an 7-hydroxylierten Metaboliten war fast gleich in Hoden und Epididymis.

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[5] J. ŠULCOVÁ and L. STÁRKA, Experientia *19*, 632 (1963).

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Degenerating Pituicytes in the Neural Lobe of Osmotically Stressed Rats

It is well known that the pituicyte is a specialized glia-like interstitial cell interposed among the neurosecretory endings of the hypophysial neural lobe. The importance of this cell type is due to the fact that to pituicytes has been attributed, beyond a supporting role common to other glial cell types, also an intermediary role in transport and release of neurosecretory hormones[1]. In this context, the present note deals with some degenerative aspects of pituicytes in the neural lobe of dehydrated rats.

Materials and methods. 20 male adult rats of a Wistar strain were deprived of water during 5, 10, 15 days. Some rats were fed with standard diet and water ad libitum and used as controls. The samples were prefixed with 2% glutaraldehyde in phosphate or in cacodylate buffer, postfixed in osmium tetroxide, dehydrated in ethanol and embedded in Epon.

Results and discussion. Our results concern the latest stages of the dehydrating experiment, i.e. the fine pituicytic modifications appearing 10–15 days after the beginning of the experiment, being the early stages already described[2].

The lipid droplets undergo a progressive vacuolization and reduction in number. The cisternae of the smooth endoplasmic reticulum swell progressively up to assume a vacuolized and variably-shaped appearance. It is also possible to observe swollen mitochondria, disorganized Golgi complexes and some scattered lipofuscin bodies (Figure 1). Many glycogen-like particles permeate especially the peripheral areas of the pituicytic cytoplasm

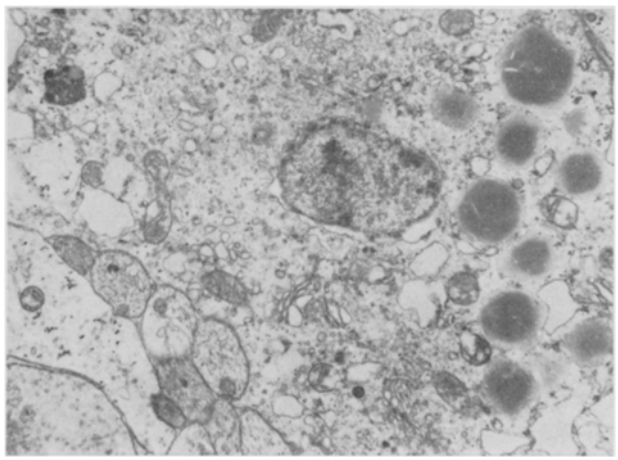

Fig. 1. Pituicyte of neural lobe. The micrograph shows the typical cytoplasmic disorganization with swollen cisternae of the endoplasmic reticulum and mitochondria, disordered Golgi complexes, a lipofuscin body and the picnotic nucleus. The lipid droplets are reduced in number and density. ×16,000.

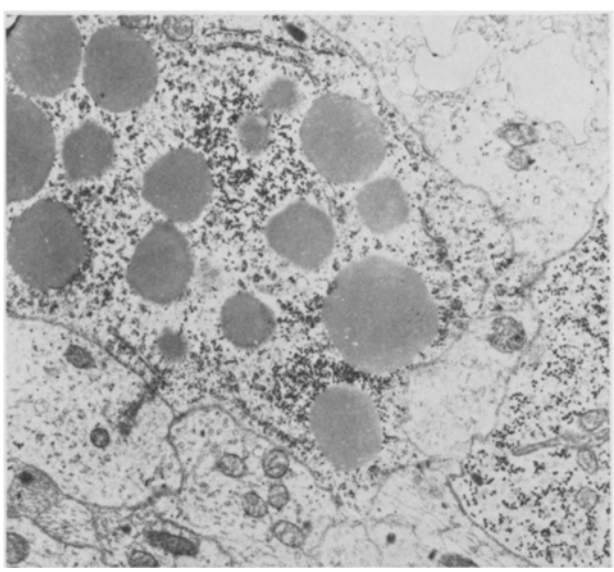

Fig. 2. Pituicyte of neural lobe. Many glycogen-like particles permeate a peripheral cytoplasmic process which is still relatively rich in lipid droplets. Note a synaptoid contact in the left and low part of the figure. ×16,000.

(Figure 2). The nucleus becomes picnotic, variably altered in shape, and is surrounded by a swollen perinuclear cisterna (Figure 1). In the final degenerative stages the organelles-empty cytoplasm contains only a finely fuzzy material and some scattered profiles due to membrane residua (Figure 3). Finally it must be emphasized that such degenerative pattern is statistically quite frequent in the pituicytes of such experimentally-treated rats.

It has been proposed that pituicytes may be involved in transport and release of neurosecretory hormones: a morphological aspect of such relationship between the neurosecretory neuron and the pituicyte itself is given by the 'synaptoid' contacts interposed between the pituicytes and the neurosecretory endings[3,4]. Furthermore, it has been shown that in dehydrated rats – parallel with the neurohormonal release – there is an apparent increase in

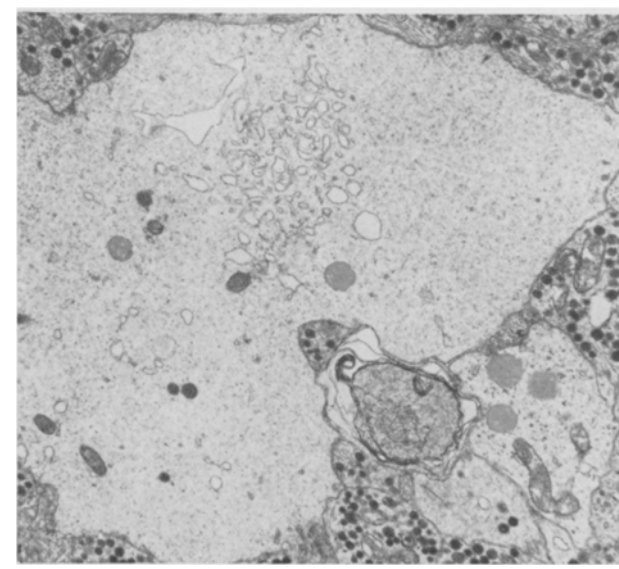

Fig. 3. Pituicyte of neural lobe. A degenerated pituicyte is shown containing a finely fuzzy material, some small lipid droplets and mitohondria and a clumping of membrane profiles. ×16,000.

the size and in the mitotic activity of pituicytes[5]. Finally, fine cytoplasmic modifications of pituicytes under the same experimental conditions have been described[2]. Our experiments show that after prolonged dehydration such modifications assume a clear-cut degenerative character which leads progressively to the total destruction of the cell. Furthermore, the high number of degenerating pituicytes under such conditions proves that it is a generalized process.

It can be suggested that in the early stages of dehydration the cytoplasmic modifications of the pituicyte may be due to its metabolic hyperactivity in connection with an increased neurohormonal release. On the contrary, in the latest stages hyperosmotic stimulation results in a progressive exhaustion of the pituicyte so that it undergoes an irreversible degeneration. This finding may be a further element supporting the hypothesis that the pituicyte can be involved in the final stages of the neurosecretory process.

Riassunto. Nella presente nota sono descritti gli aspetti degenerativi di pituiciti appartenenti al lobo neurale ipofisario di ratti sottoposti a disidratazione sperimentale.

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[1] F. M. Rodriguez and J. La Pointe, Z. Zellforsch. *95*, 37 (1969).
[2] J. Krsuslovic and G. Brückner, Z. Zellforsch. *99*, 210 (1969).
[3] F. Knowles and L. Vollrath, Nature, Lond. *206*, 1168 (1965).
[4] Y. Nakai, Z. Zellforsch. *110*, 27 (1970).
[5] T. F. Leveque and M. Small, Endocrinology *6*, 909 (1969).
[6] Acknowledgment. The technical help of V. Panetta is greatly appreciated.