

## ULTRASTRUCTURAL STUDY OF THE NADPH-DIAPHORASE ACTIVITY IN MICE AGEING TESTES

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**INTRODUCTION:** A decline of testicular function occurs with ageing in mammals. Diminished sperm production and decreased testosterone secretion were in fact shown in humans and in laboratory rodents (1). A number of morphological alterations were also described in the Leydig cells and in the seminiferous tubules. The most frequent Leydig cell modifications consisted of poor development of the SER and of reduced both volume and number of the mitochondria. In addition, several Leydig cells exhibited lipofuscin granules in their cytoplasm. Recently, nitric oxide synthase (NOS), the enzyme responsible for the production of nitric oxide (NO), was recognized in the Leydig cells (2). Results so far achieved show NO to suppress the testosterone secretion (3). Modifications of NO production were described in several organs of ageing mammals. Therefore, the purpose of this study was to examine by TEM the distribution of the NOS enzymatic activity in the Leydig cells of ageing mice.

**MATERIAL AND METHODS:** Ten Swiss mice aged 12 months were anaesthetised, perfused by aldehydes and treated for NADPH-d histochemistry (4). In a previous study we demonstrated that NADPH-d reaction overlaps with NOS in the Leydig cells of mouse (5). The specimens were post-fixed in osmium tetroxide and embedded in Epon 812, following the usual procedures.

**RESULTS AND DISCUSSION:** In the ageing mice the majority of the Leydig cells were smaller in size than those of young adults, and their shape was elongated. These cells contained a small number of lipid droplets and poor developed both SER and mitochondria. In addition the NADPH-diaphorase reaction was strongly increased in respect to young animals. The reaction product was widely spreaded in the SER and several mitochondria were labeled. Leydig cells were also present filled with lipid vacuoles, showing a distribution of the enzymatic reaction like that observed in young animals. Some Leydig cells contained several lipofuscin granules. The present study indicates that most of Leydig cells of ageing mice undergo increase of NOS enzymatic activity, and suggests that NO could be involved in the age-associated decline of testosterone secretion.

### REFERENCES:

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