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JOURNAL ARTICLE

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# Immunocytochemical localization of glutathione S-transferase Yo subunit in the rat testis and epididymis

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The glutathione S-transferases (GSTs) are a family of isozymes that catalyze the conjugation of glutathione with electrophiles. These proteins exist as homo- or heterodimers and are separated into five classes (alpha, mu, pi, theta, and sigma). In the present study, the distribution of the GST Yo subunit, a member of the mu family, was examined immunocytochemically in the adult rat testis and epididymis

using both light microscopy (LM) and electron microscopy (EM). In the testis, an intense immunoperoxidase reaction was observed over Leydig cells but not macrophages. Within the seminiferous epithelium, only weak reactivity was noted over Sertoli cells, spermatogonia, spermatocytes, and step 1-15 spermatids. There was, however, a progressive and dramatic increase in the intensity of staining in the cytoplasmic lobes of spermatids between steps 16 and 19. Residual bodies, representing the detached cytoplasmic lobes of the late step 19 spermatids, were also intensely stained. Initially seen near the lumen of the tubule, they eventually appeared at different levels of the tubule at stages IX-XI; none were present at stage XII. Cytoplasmic droplets of step 19 spermatids were also intensely reactive. After spermiation, the cytoplasmic droplets of spermatozoa within the proximal region of the epididymis remained intensely stained. A noticeable decrease in staining was observed in the cauda epididymidis in those droplets that were still there. Quantitation of the labeling density (number of gold particles representing anti-Yo antigenic sites/microns 2) paralleled the LM results; for example, between step 15 and 19 spermatids, a greater than sevenfold increase in labeling density was noted. In the epididymis, a progressive increase in immunoreactivity was observed over epithelial principal cells from the initial segment to the cauda region of this tissue. There was little reactivity over basal, halo, or clear cells. In all reactive cells, gold particles were distributed randomly throughout the cytoplasmic matrix and nucleus. The present work thus demonstrates that, at the end of spermiogenesis, the GST Yo subunit is expressed at high levels in late spermatids. Furthermore, the presence of this protein in late spermatids and cytoplasmic droplets of spermatozoa suggests that this conjugating enzyme may play a role in protecting these cells from electrophilic attack. Also interesting is the correlation between the loss of reactivity in cytoplasmic droplets of spermatozoa of the distal region of the epididymis and the concomitant increase of reactivity in principal cells of this region.

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