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Action of Mercuric Chloride during One Cycle of Seminiferous Epithelium in the Rat

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Seminiferous tubule is surrounded by peritubular membrane (PTM) which serves as primary blood-testis barrier (Dym and Fawcett,1970). Hence, its structural and functional integrity are of greater significance. Analysis of testicular toxicity of various chemicals indicated that all the seminiferous tubules did not exhibit same or similar response to a chemical (Hoffer, 1983; Chapin et al., 1985; Shemi et al., 1988). Hoffer (1983) described that upon administration of gossypol, less than 50% tubules exhibit degeneration at different experimental durations.

The spermatogenic cycle in rat is divided into 14 different stages of seminiferous epithelium (SE). The Sertoli cells are already shown to possess a variability of characteristics at different stages of SE (Parvinen, 1982). It has been suspected that PTM, associated with the tubules at different stages of SE, might demonstrate differential responses to a toxicant. This study investigated the action of mercuric chloride (MC) on the peritubular membrane at different stages of SE in rats.

MATERIALS AND METHODS

30 male albino rats (Wistar, 250±10g), maintained in standard laboratory conditions were divided into two groups. Single i.p. dose of $HgCl_2$ (1mg/kg; 5% of LD_{50} value) was given to the animals of group II (n=18). Control rats group I (n=12) were given distilled water. Animals were sacrificed on 1,3,5,8 and 12 days post exposure. This duration was selected because one cycle of SE in rat takes around 12 days.

On each of the experimental duration , 2 rats from control group and 3 rats from treated group were sacrificed by cervical dislocation. Testes were cleanly dissected out and fixed in Bouin's fluid, 5 µm paraffin sections were stained with hematoxylin and eosin. The stages of SE and various spermatogenic cell types were identified. The status of seminiferous peritubular membrane (PTM), intactness of germinal epithelium and

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extent of tubular degeneration were analyzed at all the stages of SE. In each case, 5 tubules at each stage were analyzed. This data was compared with histopathological observations.

RESULTS AND DISCUSSION

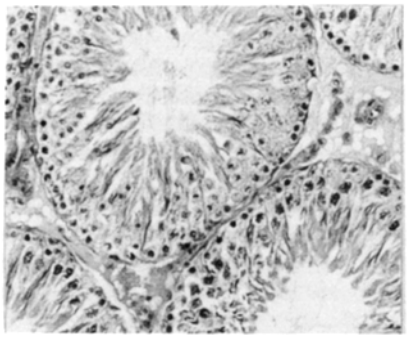
The status of PTM was observed in terms of loosening or detachment from the germinal epithelium(Table-1). On day 1, partial loosening of PTM was observed at stages VIII and IX while peritubular detachment was observed at stages X-XII. Around 50% of the PTM area was detached from germinal epithelium at stages IX-XII on day 3. The tubules at stages VII-VIII were less affected. On day 5, the PTM intactness was regained and only few tubules at stages IX-XII showed PTM detachment on days 8 and 12 (Table-1). Tubules at stages I-VI were normal.

Table 1. Number of tubules showing status of peritubular membrane.

Duration	PTM Detachment Area	STAGES OF SE			
		I-VI	VII-VIII	IX-XII	XIII-XIV
Day 1	< 50	-	-	15	-
	> 50	-	-	-	-
Day 3	< 50	-	8	12	10
	> 50	-	-	8	-
Day 5	< 50	-	5	7	2
	> 50	-	-	6	-
Day 8	< 50	-	-	5	-
	> 50	-	-	2	-
Day 12	< 50	-	-	3	-
	> 50	-	-	1	-

5 Tubules were analysed at each stage of SE.
Total Tubules : I-VI=30; VII-VIII=10; IX-XII=20; and XIII-XIV=10.

The control testis showed normal histoarchitecture, intact PTM and appropriate cellular associations (Fig.1). On day 1,although the PTM was detached, the tubules at stages IX-XIV exhibited normal germinal epithelium (Fig.2). On day 3, detachment of mature germ cells from the basal compartment was observed at stages VIII-XII. This was more prominent on day 5 where the round and elongated spermatids were detached from the germinal epithelium at stages VII and VIII (Fig.3). Analysis of germ layer detachment pattern showed that typical separation of adluminal from basal compartment was observed mainly on days 5 and 8 at stages IX-XII, while detachment of spermatids was observed at stages VII and VIII. Vacuolation of spermatids was also observed at stages VII-IX. Late elongating or elongated



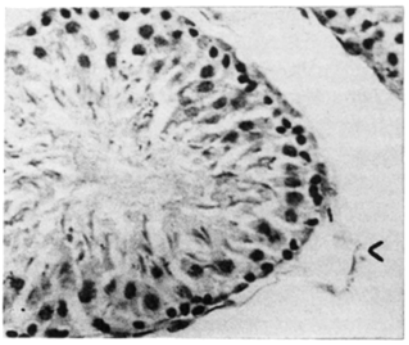


Figure 1. A tubule from control testis X 62.
Figure 2. PTM detachment (Λ) after MC treatment X 125.

spermatids were unaltered. Many tubules at IX-XIV regained normalcy over a period of 12 days.

Present observations showed that tubules at stages I-VI were not affected by MC. These tubules constitute around 40% of the total tubules (Amann, 1986). Among the tubules at stages VII-VIII and XIII-XIV some of the tubules appeared normal. As a whole, these tubules constitute around 40% of the total tubules. The tubules at stages IX-XII were affected the most. These constitute around 20% of the total tubules. In present studies a total of about 30% of the tubules were damaged. Earlier studies with MC showed that a particular spermatogenic cell type was affected differentially at various stages of SE (Vachhrajani,1989). Studies with various chemicals showed "focal degeneration" in the testis (Baccetti et al., 1986; Shemi et al., 1987). The plausible causes for such differences were not expressed. However, in present studies, we suspect that the differential susceptibility of PTM at different stages of SE may be the reason for variability of MC action.

Within the tubules, separation of germinal epithelium exhibited interesting patterns at different stages. Mercury can accumulate

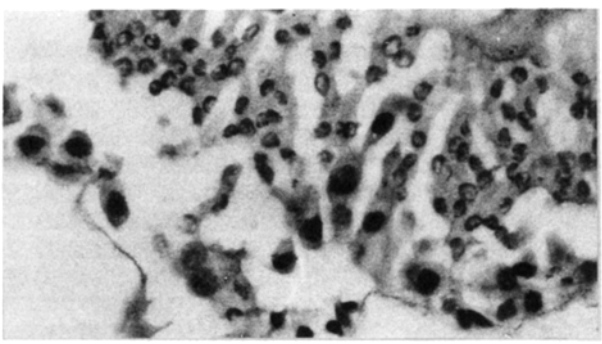


Figure 3. Separation of germ layers after MC treatment X 250.

in the adluminal compartment of the tubules. However, it could not dismantle the Sertoli-Sertoli-junctional complexes or spermatogenic cell-Sertoli complexes at stages I-VI. Hence the epithelium remained intact. While, deposited mercury could distort the crypt regions of Sertoli cells at stages VII-VIII. These are the sites of microtubules in the Sertoli cells beneath the elongated spermatids(Boekelheide et al., 1989), and mercury is a microtubule disrupting agent (Vogel et al., 1985). Thus, only spermatids were separated from Sertoli cells. It appears that either the deposition of mercury was less to affect the junctional complexes or these complexes were still resistant to deposited MC.

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