



Histopathologic studies on the effect of repeated doses of doramectin on the urogenital organs of male guinea pigs

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ABSTRACT

In the present work, histopathologic studies were done in order to detect the adverse effects and tissue alterations in the urogenital organs of healthy male guinea pigs after repeated administration of variable dose levels of Dectomax (Doramectin). Total number of 72 adult male guinea pigs were used in a four treatments; non-treated control group, treated group with therapeutic dose of 0.2 mg/kg. b.wt, treated group with the double therapeutic doses (0.4 mg/kg. b.wt) and treated doses with the triple therapeutic dose (0.6 mg/kg. b.wt). The microscopic examination in case of repeated therapeutic doses revealed less or no changes, while the repeated double as well as triple therapeutic doses lead to numerous histopathologic changes of testicular degeneration and necrosis in addition to the interstitial edema and congestion in the prostate glands and seminal vesicles, especially after the 5th and 6th repeated triple doses. It was concluded that the repeated as well as duplication of the therapeutic dose levels lead to some adverse effects on the male genital organs and so on the reproductive performance.

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KEYWORDS

Doramectin;
Guinea pigs;
Urogenital organs;
Histopathologic;
Toxicity.

INTRODUCTION

Doramectin (Dectomax), a derivative of avermectin B which also include abamectin and ivermectin, is effective endoparasiticide agent in cattl^[7,12,15] and canines^[10] as well as in treatment of exoparasites in rabbits^[17]. It also is an orally effective microfilaricidal agent and considered as a drug of choice for treating patients infected with the nematode *Onchocerca volvulus* in human^[8]. Previous studies have shown the long-term persistence of unwanted residues of ivermectin and Doramectin in treated animal tissues and fluids^[4]. In addition, it was proved that ivermectin therapy did not affect the breeding performance of bulls^[11], rams^[13] and

Boars^[3]. Oral administrations of ivermectin were shown to induce neurotoxicity in brain tissue in mice particularly with P-glycoprotein deficiency^[9]. Furthermore, the administration with ivermectin and doramectin was found to result in some adverse effects on the reproductive performance and male sex organs of rats^[2,19,20] and ram^[1]. There is scarcity of information on the adverse effects of doramectin on reproductive performance of the guinea pigs. The present work aimed to study the adverse effects and tissue alterations in the kidneys and male sex organs (testes, epididymis, prostate gland and seminal vesicle) of male guinea pigs following repeated administration of variable dose levels of doramectin.

MATERIAL AND METHODS

Animals

A total number of 72 adult male guinea pigs, weighing 350-400 g., were used. Animals were grouped and housed in separate metal cages and fed on standard pelleted rabbit food. Both food and water were supplied *ad libitum*. The used injectable drug, Dectomax (Doramectin, Pfizer Co.), is a sterile, colorless to pale yellow solution. It contained 1 % (W/V) Doramectin (10 mg/ml).

Treatment

The experimental design is shown in TABLE 1

The animals were divided into 4 groups, each of 18 male guinea pigs. The first group (Gp-I) represent the non-treated control animals. In the second group (Gp-II) animals were weekly injected intramuscularly with a single therapeutic dose of 0.2 mg/kg body weight Dectomax for six weeks. The third group (Gp-III) were weekly injected with a single double therapeutic dose of 0.4 mg/kg for 6 weeks and the fourth group (Gp-IV) were injected with a triple dose of 0.6 mg/kg for 6 weeks. The animals of various groups were kept under observation. Three animals from each group were slaughtered and subjected for postmortem examination and specimen collection (from the kidneys, testes, Epididymis, seminal vesicles and prostate glands) for the histopathologic examination

Histopathologic studies

The collected specimens were fixed in 10 % neutral buffered formalin solution, washed in water and passed through the routine technique of the paraffin embedding and preparation of paraffin blocks. Paraffin sections of 3-5 microns thick were prepared on microscopic glass slides. The paraffin section were stained with hematoxylin and eosin according to the methods described by Drury and Wallington^[5] and then the prepared slides were subjected for light microscopy for microscopic examination.

RESULTS

Histological findings in organs of the control group

The microscopic examination of Kidneys, testes, epididymis, prostate gland and seminal vesicle of GpI

TABLE 1: Protocol of treatment of guinea pig with doramectin

Groups	No. of guinea pigs	Doses			Schedule of slaughtered animal / weeks					
		T	Dt	Tt	1 st W	2 nd W	3 rd W	4 th W	5 th W	6 th W
1 st	18	-	-	-	3	3	3	3	3	3
2 nd	18	+	-	-	3	3	3	3	3	3
3 rd	18	-	+	-	3	3	3	3	3	3
4 th	18	-	-	+	3	3	3	3	3	3
Total	72	-	-	-	12	12	12	12	12	3

T : Therapeutic dose, Dt : Double therapeutic dose, Tt : Triple therapeutic dose

appeared with normal histologic structures throughout the experiment period.

Administration with single therapeutic doses (group II)

Less or no microscopic changes could be seen in the examined organs of this group. The renal changes of the congested glomerular capillary tufts, cloudy swelling of the tubular epithelium as well as testicular changes of less active spermatogenesis, were the only detected changes after administration with the 5 and 6 weekly doses.

Administration with single double therapeutic doses (group III)

The kidneys after administration of 2 doses showed congestion of the various glomerular as well as the intertubular blood vessels. In the following weeks, some additional changes of periglomerular mononuclear cell infiltrations and aggregations were gradually seen after administration with the following doses. The male sex organs showed some mild changes only, after administration with the 5th and 6th doses interstitial edema and congested blood capillaries in the prostate glands in addition to changes of a less active spermatogenesis in the seminiferous tubules of the testes.

Administration with single triple therapeutic doses (group IV)

The vascular changes of congestion as well as granular degeneration of the renal tubular epithelium were seen early after administration of the 1st dose. Some large areas of intertubular hemorrhages appeared at administration of the 2nd doses (figure 1), while after the 3rd doses some hyaline cast formations appeared inside the lumina of renal tubules (figure 2). The testicular tissue appeared to be affected following administration of the 3rd doses. In these cases vacuolar degeneration

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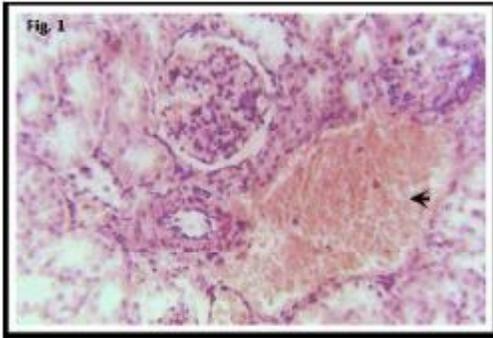


Figure 1: Kidney of male guinea pig received 2 triple therapeutic doses, showing tubular degeneration with large area of cortical hemorrhages (thick arrow). hematoxylin and eosin (H and E). X 250

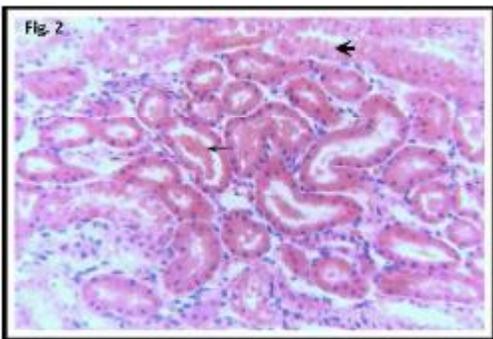


Figure 2: Kidney of male guinea pig received 3 triple therapeutic doses, showing epithelial cloudy swelling of some tubules (thick arrow) and luminal contents of hyaline cast (thin arrow) in other tubules. H and E. X 160

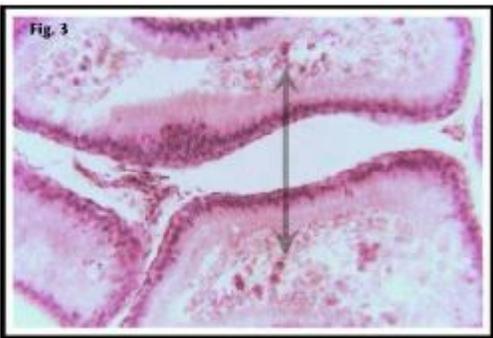


Figure 3: Head of epididymis of male guinea pig received 3 triple therapeutic doses, showing excess luminal contents of immature and necrotic spermatogonial cells (arrows). H and E. X 240

tion and necrosis of the spermatogenic cells with presence of some spermatocytic giant cells inside the lumina of some of the seminiferous tubules were seen. An excess of luminal contents of immature as well as necrotic spermatogonial cells were also seen inside the lumina of the epididymal tubules at the head region (figure 3).

After the administration for the 4th triple dose the

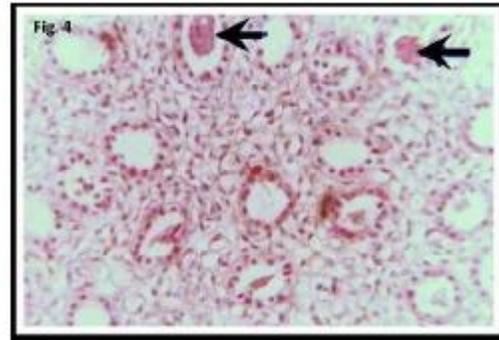


Figure 4: Kidney of male guinea pig received 4 triple therapeutic doses, showing dilated medullary tubules with excess of cast formations (arrows). H and E. X250

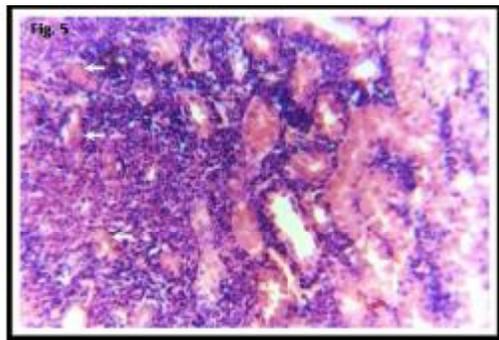


Figure 5: Kidney of male guinea pig received 5 triple therapeutic doses, showing tubular necrosis (white arrows) with replacement by an excess of mononuclear cell infiltrations and aggregations. H and E. X 250

kidneys were affected by degeneration and dilatation of the renal tubules in addition to excess formation of hyaline casts (figure 4). The testes of these cases were severely affected by vacuolar and hydropic degeneration in most of all spermatogenic cells with the appearance of luminal contents of liquefied necrotic materials inside the lumina of the severely affected seminiferous tubules. Similar liquefied contents were seen in the epididymis. The prostate glands of these cases were only affected by interstitial edema and congested blood capillaries.

The administration with the 5th triple doses was associated with progressive damage and necrosis of the renal tubules with replacement by an excess mononuclear cell infiltrations and aggregations (figure 5). The testes of these cases were also affected with degeneration and necrosis of the spermatogonial cells similar but more severe than those after the 4th triple doses. The seminiferous tubules in these severely affected testes appeared lined with only one cell layer of vacuolar and hydropic degenerated spermatogonial cells while the

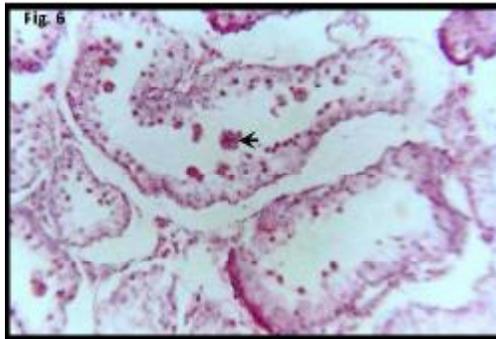


Figure 6: Testes of male guinea pig received 5 triple therapeutic doses, showing degenerated seminiferous tubules lined with one layer of vacuolar degenerated spermatogenic cells with presence of some spermatocytic giant cells (arrow). H and E. X 250

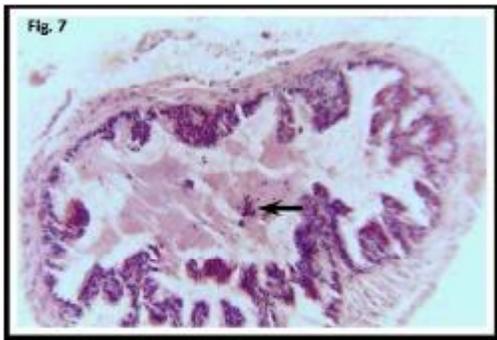


Figure 7: Prostate gland of male guinea pig received 5 triple therapeutic doses, showing interstitial edema with some luminal contents of degenerated and necrotic cellular debris (arrow). H and E. X 160.



Figure 8: Head of epididymis of male guinea pig received 6 triple therapeutic doses, showing excess luminal contents of the damaged and necrotic spermatogonial cells (arrow). H and E. X 250

lumina contained some spermatocytic giant cells (figure 6). The epididymis of these cases were also contained luminal contents some immature as well as necrotic spermatogonial cells. The prostate glands were also suffered degeneration, necrosis and desquamation, of its lining secretory epithelium into the lumina of the affected

acini (figure 7). The seminal vesicle appeared with cystic dilatation of its acini.

The detected microscopic changes after the administration with the 6th triple doses (end of the experiment) were somewhat progressed than previously described lesions. The kidneys appeared with excess of tubular necrosis and replacements with mononuclear cell aggregations. The testes appeared with several areas of damaged and necrotic seminiferous tubules accompanied with excess luminal contents of necrotic as well as spermatocytic giant cells. The epididymis contained also similar contents of the necrotic cellular debris and immature spermatogonial cells (figure 8). The prostate glands appeared with papillary hyperplasia, degenerated, necrotic and desquamated lining epithelium.

DISCUSSION

The intramuscular administration with the repeated therapeutic doses leads to less or no renal lesions and less active spermatogenesis, this is mainly correlated and attributed to the reported minute levels of Doramectin in the kidneys^[16]. The findings in case of the repeated double therapeutic doses revealed presence of more progressed lesions of nephrotoxicosis in form of sequential and variable degrees of glomerular as well as intertubular congestion with mononuclear cell infiltrations and aggregations. The testes were not severely affected, while the other sex organs (prostate and seminal vesicle) showed congestion with interstitial edema.

The findings in case of intramuscular administration with the repeated triple therapeutic doses were indicative for the more adverse effects in the kidneys, especially after the 2nd doses. These lesions of nephropathy included degenerative changes in the tubular epithelium (granular, vacuolar and hydropic degenerations and hyaline cast formation) followed by tubular necrosis, interstitial as well as periglomerular infiltration and aggregation of mononuclear cells, congestion and areas of hemorrhages. Some similar but mild lesions of nephrotoxicosis were reported in case of injection with single therapeutic doses of Ivermectin in guinea pig, rat and rabbits^[6,19] and in case of rat^[20]. The testes were also affected and showed spermatogonial degenerations and necrosis with presence of an excess of the necrotic debris and immature spermatozoal cells inside the lumina of the epididymal tubules. Edema and con-

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gestion were also seen in the prostate glands.

The obtained results for toxicoses in the kidneys and genital organs of the male guinea pigs, could be generally attributed to the chronic toxic effects and higher bioavailability of Doramectin at tissue level and its known long duration of action^[14,18]. Nevertheless, the loss of correlation between these results and other reports about the wide safety margins is probably due to either species difference in metabolites and excretion as well as dose dependent factors. It was concluded that the repeated single therapeutic doses of Doramectin have a wide safety margins, and lead to a minimal or no adverse effects on the urogenital organs of male guinea pigs; while the repeated higher doses lead to many adverse effects that may affects the reproductive performance of the male animals.

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