RESEARCH ARTICLE

Effect of folic acid antagonist methotrexate on seminal vesicle of Indian palm Squirrel *Funambulus pennanti* (Wroughton)

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ABSTRACT

The toxic effect of Methotrexate on seminal vesicle have been studied by intramuscularly injecting low dose of 3 mg/ kgBW /day and 6 mg/kg BW/day for 15 days to adult male squirrel (*Funambulus pennanti*) during the breeding period January. For comparing the effects the saline treated vehicle was injected same amount of saline and was maintained for the same duration. Reduction in the size of secretory alveoli of seminal vesicle due to remarkable increase in the fibro-muscular coat, loss of secretory capacity all suggest toxic effect of MTX on seminal vesicle.

Key words: Methotrexate, toxic effects, Seminal vesicle.

INTRODUCTION

Methotrexate (Rheumatrex) is a medicine that is used to treat Rheumatoid arthritis (RA), psoriatic arthritis, Reiter's syndrome and other conditions. Aside from its antineoplastic activity, Methotrexate has also been used with benefit in the therapy of common skin disease psoriasis (Mcdonald, 1981). Additionally Methotrexate inhibits cell mediated immune reaction and is employed as an immunosuppressive agent, for example, in allogenic bone marrow and organ transplantation and for the treatment of dermatomyositis, rheumatoid arthritis, Wegener granulomatosis and Crohn's disease (Messmann and Allegra, 2001; Feagan *et al.,* 1995, Felig and Frohman 2001, Prasad *et al.,* 1996). Methotrexate was formerly known as amethopterin, is an antimetabolite drug used in treatment of cancer and autoimmune diseases. The present study embodies: Histopathological changes undergone by seminal vesicle.

MATERAILS AND METHOD

In all three sets of experiments using low and high-doses of Methotrexate (MTX) were performed for the present study for the duration of 15 days (Tables 1& 2). Animals were sacrificed using chloroform 24 hours after the last day of each experiment. Immediately the organs were excised seminal vesicle was used for histological studies.

Table 1: Experimental Design for Low Dose Methotrexate treatment

Number of animals and sex	Treatment	Dose mg/kg BW	Route	Duration
3 males (Experimental)	Methotrexate	3 mg daily	I.M.	15 days
3 males (Control)	Saline	E.V.	I.M.	15 days

Table 2: Experimental Design for High Dose Methotrexate treatment

Number of animals and sex	Treatment	Dose mg/kg BW	Route	Duration
3 males (Experimental)	Methotrexate	6 mg daily	I.M.	15 days
3 males (Control)	Saline	E.V.	I.M.	15 days

Abbreviations: E. V. = Equal volume, I. M. = Intra muscular, B W = Body weight

RESULT AND DISCUSSION

Histological Studies

Seminal vesicle fixed in Bouin's fluid for 24hrs and preserved in 70% alcohol. The tissues were dehydrated by passing through graded series of alcohol, cleared in xylol and after embedding in paraffin, blocks were prepared and serial sections were cut at various thicknesses between 5µ to 8µ. For routine histological study the sections were stained with Ehrlich's haematoxylin and counter-stained with eosin. Measurements when necessary were taken with the help of an occular micrometer calibrated to a stage micrometer. The photomicrographs were taken with the help of a Carl Zeiss camera attached to the microscope and enlarged to the required size.

Vehicle Treated Control

The seminal vesicle of the control squirrel was composed of a large number of acini embedded within the fibro muscular connective tissues. The acini were lined by tall columnar epithelial cells containing a prominent basal nucleus (fig. 1).

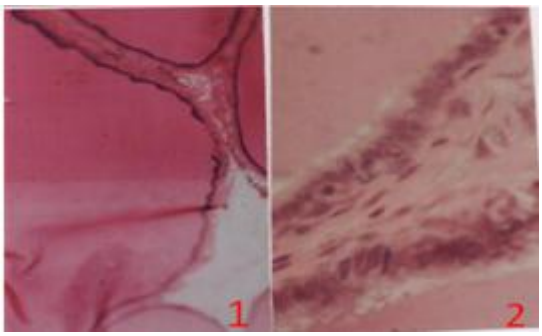

Fig. 1 and 2 Vehicle treated control

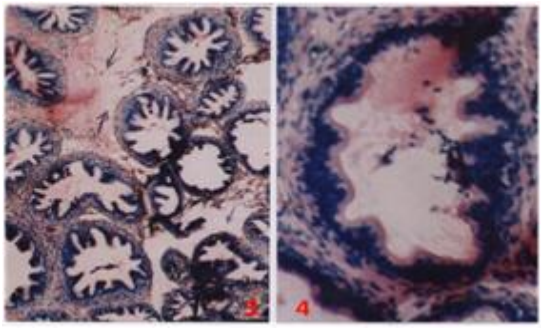

Fig. 3 and 4 Low dose 3 mg/KgBW

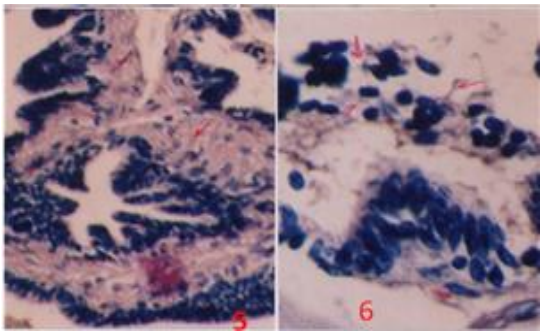

Fig. 5 and 6 High dose 6 mg/KgBW

Few basal cells, almost rounded in shape and basal in position were also observed between the columnar epithelial cells (fig. 1). Large numbers of dense secretory granules were visible in the apical cytoplasm. The lumen of acini was filled with the darkly stained secretory material. Lamina propria surrounding the epithelial cells was comprised of cellular connective tissues containing some smooth muscles rich in elastic fibers (fig. 2).

Low Dose Treatment (3mg/kg BW MTX for 15 days)

Histopathological study

An enormous increase in the fibro-muscular connective tissue in between the secretory tubules have resulted into remarkable reduction in there, inter tubular connective tissue was heavily lost at some places with large vacuole or streaks of cellular tissues. Each tubule was lined by secretory epithelium, which was either club-shaped or thrown into medium-sized, finger like projections, epithelium appeared partially damaged, due to the irregular placement of nuclei. The loss of secretory activity was evident by emptiness of most of the tubules (figs. 3 & 4).

High Dose Treatment (6mg/kg BW/day MTX for 15 days)

The nuclei of the connective tissue were reduced in number and randomly distributed. The secretory epithelium was thrown into crypt like structure and was highly vacuolated the nuclei were also randomly distributed due to extensive vacuolation of the cytoplasm. Almost all tubules showed loss of secretion (figs. 5 and 6).

There was significant reduction in the acinar size due to enormous increase in the intertubular connective tissues. Prevalence of vacuolation in the supranuclear region of regressed secretory epithelium directly suggested the depletion of secretory activity and hence scanty secretion in the lumen. The high dose 6 mg/kg BW/day for 15 days resulted into further increase in the intertubular connective tissue reducing further the size of secretory acini. The epithelial damage was more pronounced due to loss of secretory granules from the supra as well as infra-nuclear region and the total loss of secretion. The epithelial lining appeared disrupted and disorganized, the nuclei were scattered hapzardly with much spaces between them. These sloughed off nuclei were found to be intermingled with the dry flocculent secretion in the centre some tubules showed heavy deposits of such nuclei. The epithelium lining the tubule was manifested towards lumen, partially obliterating the lumen. Thus the enlargements of dense fibroblast cells following both the treatments were suggestive of alteration in the function of. Similar alterations in the sex accessory glands have been reported only by Takeda et al., 1985 that too on prostate, therefore this is the first kind of study which has evaluated the toxic effects of MTX on seminal vesicle which are dose, duration and androgen dependent.

REFERENCES

Takeda M, Hosaka M, Kitajima N, Noguchi K, Fujii H, Oshima H and Harada M (1985) Effects of anticancer agents on rat prostate. Evaluation of organ weight, histological finding and 5 alpha-reductose activities, *Hinyokika Kiyo*,31(1):71-80.

Feagan BG, Rochon, Fedorak RN, Irvin EJ, Wild G, Sutherland L, Steinhart AH, Greenberg AH, Gillies R, Hopkins M, Hanuer SB and McDonald JWD (1995) Methotrexate for the treatment of crohns disease. The North American Crohns study group investigators, *N Eng. J. Med*., 332:292-297.

Felig, P and Frohman LA (2001) The Testis In: Endocrinology and Metabolism, 4th edition Mc Graw- Hill, Inc. Medical Publishing Division.

Messmann, R and CJ Allegra (2001) Antifollates in cancer chemotherapy and Biotherapy: Principles and practice, 3rd ed. (chabner,BA and Longo, Dl. eds) Lippincott Williams and wilkins, Philadelphia in press.

Prasad, MRN, Seth GK, Shivashankar AK and Oberoi NK (1996) Biology of reproduction in the Indian palm squirrel *Funambulus pennnanti* (Wroughton), *Symp.Zool.Lond*.15:353