

5. SUMMARY

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Because of many varigated adverse side effects, the synthetic preparations for controlling the fertility, in recent years, biologists all over the world taking interest in plant preparation to control human fertility since such plant preparations produce minimum side effects.

The present investigation was undertaken to study alteration in the male reproductive system of albino rats after administration of Picrorhiza kurroa extract, and to see its antispermatogenic potency. Many of the plant preparations were tried to explore their effects on only testes, but as these extracts may also exert some influence either directly or indirectly on the accessory male sex organs. Hence alterations in these organs, along with testis, have also been studied. This study was carried out with reference to alterations in body weight, organ weight, histoarchitecture, a lysosomal enzyme acid phosphatase, a non-lysosomal enzyme alkaline phosphatase in testes and accessory sex organs (excluding Cowper's gland, which did not show any detectable response to the extract treatment).

The petroleum ether extract of Picrorhiza kurroa was administered intraperitoneally to the albino rats of 90-100 days age weighing 210-230 g. for a period of 48th days. The control rats during this period received only vehicle (Tween 80 + saline). The histological changes were

studied by routine Haematoxyline Eosin technique, and enzymes were studied by employing recent and well known bioassay technique. All the studies were carried out at an interval of 8 day over a period of 48 days.

1. THE BODY WEIGHT :

Weights of the Picrorhiza treated rats decreased throughout the period of treatment.

2. TESTIS :

Weight of the testis decreased appreciably and the diameters of seminiferous tubules also showed reduction.

HISTOLOGY :

There are found many histological alteration in the testes. Except spermatogonia practically all testicular cell types were affected with Picrohiza. Spermatogenesis was arrested at primary spermatocytes. Giant cells (fused round spermatids) appeared and subsequently disappeared. Clear spaces in spermatocytic layers were formed. Tubular lumina contained cellular debris consisted of sloughed off various spermatogenic cells, the debris and its contents degenerated and lysed towards the end of the treatment. Some seminiferous tubules showed shrinkage. Basal membrane and tunica propria were thickened. Sertoli cells showed varying degree of vacuolization in cytoplasm. Leydig cells atrophied. All these alterations were noted in different

tubules at different time intervals, but there was no consistent sequential progress in damage. Severity of damage increased towards the end of the treatment.

ACID PHOSPHATASE AND ALKALINE PHOSPHATASE :

Testicular acid phosphatase and alkaline phosphatase activities were decreased towards the conclusion of the treatment. The alterations in the enzymes are discussed in relation to their control by androgen and also the role of lysosomes in degenerative atrophic changes occurring in testis during extract induced infertility.

3. EPIDIDYMISS :

Weight of caput and cauda were decreased significantly as an effect of extract treatment.

HISTOLOGY :

Epithelial cells showed slight decrease in height. Luminal contents, basal lamina and interstitium showed certain degenerating changes. Presence of cellular debris in the lumina of majority of tubules is a characteristic feature. The cellular debris contained immature germ cells like spermatids and spermatocytes, giant cells, degenerating spermatozoa. Most of these cellular elements derived from the damaged testes. In cauda identification of different cellular elements from luminal cellular debris, was not possible, probably due to mere severe necrotic changes. At

the termination of the treatment, majority of tubules were without spermatozoa.

The results, thus, indicated that in rats Picrorhiza extract caused the germ cells to develop upto primary spermatocyte stage, which then begin to exfoliate and appeared in lumina of seminiferous tubules and then were transferred to caput. The ultimate fate of the degenerating material in the lumina of caput could not be discerned in the present study, but possible phagocytic removal could not be ruled out. Antiandrogenic activity of extract might have been responsible for these changes.

ACID PHOSPHATASE AND ALKALINE PHOSPHATASE :

In both caput and cauda epididymis these two enzymes showed depletion in activity. The alterations in these enzymes are discussed in relation to their control by androgen and also the role of lysosomes in degenerative changes occurring in epididymis during the extract treatment.

4. SEMINAL VESICLES :

The weights of the seminal vesicles decreased.

HISTOLOGY :

The extract reduced insignificantly the height of epithelial cells but the mucosal folds which were highly arborized and reached upto the centre of the lumen in the controls, got reduced significantly in height and arborization also. Luminal secretion was reduced while in

some vesicular lumina almost no secretion is found except that present in the depths of the crypts. Depleted androgen level might have been resulted such changes in seminal vesicles.

ACID PHOSPHATASE AND ALKALINE PHOSPHATASE :

Picrorhiza extract decreased the acid and alkaline phosphatase activity in seminal vesicles, biochemically. Depleted enzyme activity may be due to depletion in androgen level.

5. PROSTATE GLAND :

Prostate gland showed reduction in weight after administration of Picrorhiza extract.

HISTOLOGY :

The extract reduced epithelial cell height insignificantly. Luminal secretion was affected and many acini were almost without secretion. It also resulted in slight thickening of basal lamina.

ACID PHOSPHATASE AND ALKALINE PHOSPHATASE :

The extract depleted the acid and alkaline phosphatase activities of the prostate gland. Such depletion may be due to deleted level of androgen.

FUTURE PLAN TO WORK :

The present investigation opens several avenues for further research on the Picrorhiza extract induced alterations in the male reproductive organs of rat. Some ideas for such further work are given here :

i) It seems from present investigation that Picrorhiza extract interfere in androgen production of testis in the treated rats. This conclusion is drawn from some indirect observations. Hence a direct investigation of bioassay of androgen in the treated animals is highly desired to confirm or modify the above conclusions.

ii) Picrorhiza extract caused atrophy of Leydig cells from such hypotrophy, the idea of possible depletion in androgenic level is projected. A histochemical study of steroid dehydrogenase in the Leydig cells, of the treated testes would have given a better information on their androgenic state.

iii) It will be very interesting to find out whether the extract induced alterations in sex organs can be reversed by administration of hormones like FSH, testosterone. Pituitary gland and adrenal glands are the sources of gonadotropins and sex hormones. Hence their studies in the treated animals will give some interesting information.

iv) To get a general picture of possible changes in the physiology of body as a whole, it is necessary to study the serum enzymes, proteins etc. in the serum of treated rats. This will also give information on the side effects of the Picrorhiza extract treatment.

v) Picrorhiza caused depletion in number of spermatozoa, decrease in secretion of epididymis, seminal vesicles and prostate. Optimum level of secretion of these accessory sex organs is necessary for sperm maturation and survival. Number of sperms should have also at optimum level, for fertilization. Extract treatment resulted oligospermia followed by azoospermia in many tubules of testes and epididymis, confirmation of which is possible only with epididymal sperm count, so it should be done.

vi) Protein, lipids and mucosubstances all play important role in physiology of reproduction. Optimum level of these metabolites is necessary for the normal process of reproduction and fertility. So alterations in these metabolites after administrations of Picrorhiza in rats is planned.

vii) In the present investigation many well known and recent available techniques have been used. But to get a thorough insight into the alterations application of better techniques is observed. It will include use of electronmicroscopy to study alterations especially in

various testicular cells, use of gel electrophoresis to study abnormalities in DNA of different cell types, appearance or disappearance of special proteins etc.

While conducting the present M.phil dissertation, the author would like to state the present work is by no means complete. He is fully aware of his shortcomings. Many aspects of male reproduction were remained to be explored, though maximum possible efforts were made to complete the dissertation. It is so due to the fact that the dissertation is a time bound and limitations of existing laboratory conditions etc.

The author feels gratified that he has made a detailed histological and biochemical study of male reproductive organs during administration of Picrorhiza extract and made original contributions which have yet not been reported. There is unlimited scope for further work on Picrorhiza induced alterations in male reproductive tract of rats. Some work on the problem outlined above is in progress, which it is hoped, might throw more light on the possible use of Picrorhiza extract as an antifertility agent having minimal side effects.