ANCIENT SCIENCE OF LIFE, Vol. IX, No. 2, October 1989, Pages 95-98

‘STUDIES ON TESTICULAR REGENERTIVE POTENTIAL OF NAGA BHASMA’

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Received: 13 August 1985       Accepted: 6 September 1988

ABSTRACT: Naga Bhasma is used in various ailments and also as tonic in Ayurvedic practice. Mention has been made in Ayurvedic literature that this drug has regenerative action on testicular tissue. Therefore plan was made to study the regenerative capability on tests. It was observed that the test drug when given simultaneously with Cd cl2 showed marked prevention of toxic effects of Cd cl2 and when given alone after 36 hours of Cd cl2 administration, showed a notable regenerative potential on partially degenerated testes. It has showed specific regenerative effect on germinal epithelium of testes.

Introduction

Naga Bhasma, an Ayurvedic lead preparation is recommended for various ailments of genitor Urinary system. Of these vrisya and Balya properties are noteworthy. According to Ayurvedic concept ‘Vrisya’ is that which is good for ‘Vrisanas’ (Testes).

Materials & Methods:

i) Experimental animal: Thirty two healthy adult male albino rats weighing between 150-200 gms. Were taken and were kept on standard laboratory diet and water and. Libidium. They were marked and divided into following groups and kept in separate cages:

Group A (Control) – 4 rats.

Group B (Cd cl2) – 4 rats.

Group C (Simultaneous NB + Cd cl2) – 12 rats.

Sub Groups: I,II,III – each containing 4 rats

Group D (NB after 36 hours of adm. 12 rats. Of Cd cl2)

Sub Group: I,II,III – each containing 4 rats.

ii) Cadmium Chloride : Administered to produce partial testicular degeneration.

iii) NB (Test drug): Administered in different doses alone and simultaneously with Cd cl2 to see its regenerative potential on partially damaged testes and capability to prevent toxic effect of Cd Cl2.
Preparation of dosage form

i) Cadmium chloride Solution: 40.26 mg of Cd cl2 monohydrate (Sarabhai) was accurately weighed and dissolved in 100 ml. of triple distilled water. The concentration of Cd Cl2 solution is 0.4026 mg/ml or 0.2013 mg/0.5 ml.

ii) Suspension of NB (Test Drug): 110 gm. Of NB was weighed and about 3 gms. Of Gumacasia white powder was taken and dissolved in distilled water to make 15mls. Solution. NB was taken in a mortar added Gumacasia solution in small quantity and parted to make a paste, gradually poured all the solution and triturated well to make a uniform suspension. This suspension is of 6mg/1 ml. concentration. The test drug was to be administered in 3 different concentrations of different sub groups of animals belonging to group C & D. So as to administer the drug as follows:-

- 3mg/100 gm. Bd. wt. in (i) of group C&D.
- 6mg/100 gm.bd wt in (ii) of group C&D.
- 12 mg100 gm. bd. Wt. in (iii) of group C &D.

The above plan is devised to trace out the minimum effective dose for prevention of degeneration as well as regeneration of testes.

Administration of the Drugs:-

i) Group B (Cd Cl2) : Single S/c Inj. Of Cd cl2 n the dose o 0.2073 mg/100 gm. body weight was given to the animals of this group. Two animals each were sacrifie after 24 hours and 36 hours after the administration of Cd cl2 Animals sacrificed after 36 hours of Cd. Cl2 administration showed histoloically, the desired partial degeneration of testicular germinal epithelium.

ii) Group C (Simultaneously Cd Cl2 and NB: Aminals of this group were divided into three sub groups and NB suspension was admiistere orally 6 hours before the administration of Cd cl2 .2013 mg/100 gm. body wt S/c dose as follows:-
          C/1 = 3 mg/100 bd.wt., C/2 = 6 mg/100 gm. bd.wt. 
          C/3 =12 mg/100 gm. bd wt. continued the oral administration of NB in the same dose for a period of ten days.

iii) Group D (NB after 36 hours of Cd cl2 administration): Animals of this group received similar doses of Cd cl2 S/c inj. And NB orally as in group C. But the suspension of NB were administered after 36 hours of single inj of Cd cl2 administer. Continued the oral administration of drug for 10 days duration.

Group A (Control):-
Only 20% suspension of gumacasia was administered in the dose of 1 ml/100 gm body weight to the animals and was continued for ten days.

The animals of group A, C & D were sacrificed on 12th day of the starting the experiment. Testes of all the animals were dissected out and examined for any grass change. Tissues were then fixed in Bovin’s fixative and subjected for histopathological studies adopting standard laboratory techniques (Culli C.P., 1964).

Observation

Group A (Control): Testicles of control group revealed no significant change on gross and microscopic examination.

Group B (Cd cl2 treated): Testes of all the animals after 36 hours of Cd cl2 injection revealed diffuse fatty vacuolization and necrosis of the germinal epithelium with normal looking cells close to the basement membrane of seminiferous tubules. Few tubules were small and showed variation in shape. Additional findings include interstitial oedema and vascular congestion. In 2 animals Leydig cells were prominent.

Group C (Simlt. NB & Dc cl2)

a) At 3 mg/100gm bd wt, dose – Out of 4 animals, testes in two were normal looking. The other two animals revealed degenerative changes, mild in one and severe in other. Of these two in one animal single fatty vacuoles in 4-5 S.T (seminiferous tubules) along with mixed interstitial oedema and vascular congestion was observed while the other animal showed marked fatty vacuolization of germinal epithelium affecting most of the tubules reduction in thickness of germinal epithelium along with interstitial epithelium along with interstitial oedema and vascular congestion in one testicle while in the other testicles of the same animal, 1-4 fatty vacuoles in majority of the tubules associated with interstitial oedema, vascular congestion and prominent Leydig cell were noted.

b) At 6mg/100gm. body weight dose- Two out of 4 animals showed normal looking testes. The other animals revealed mild degenerative changes in the form of single fatty vacuoles affecting 2-4 seminiferous tubules. Mild interstitial oedema and vascular congestion was noted in all.

c) At 12 mg / 100gm. body weight dose – Single fatty vacuole in germinal epithelium of 3-4 seminiferous tubules was observed in only one animal. In the rest three seminiferous tubules were normal looking but mild interstitial oedema and vascular congestion was noted.

Group D – Testicular Regeneration:

a) At 3 mg/100 gm. body weight dose:-

At 3 mg/100 gm. body weight dose:- Fatty vacuole in germinal epithelium ranging from 1-2 in 2-4 seminiferous tubules along with mild interstitial oedema was noted in three animals. In the fourth animal majority of the seminiferous tubules showed fatty...
vacuoles in germinal epithelium interstitial oedema and vascular congestion.

A) At 6 mg/100 gm. bd, weight dose:-
Fatty vacuoles (1-2) in germinal epithelium of 2-4 seminiferous tubules alongwith mild interstitial oedema in all the animals of the group.

c) At 12 mg / 100 gm. bd wt. sose:-
There out of four animals of this group showed normal looking testes. Only in one animal 1-2 fatty vacuoles in germinal epithelium in 2-3 seminiferous tubules alongwith mild interstitial oedema were noted.

Animals of all the group showed no behavioural change during the period of experiment. Microscopically testes revealed no significant change. Basement membrane of the seminiferous tubules, Leydig cell, excretory testicular ducts, prostate etc. showed no appreciable pathologic change.

Discussion

NB is recommended in Prameha group of diseases and as Deepana, Pachana, Vrishya and Balya. Its vrishya and Balya properties are well known in Ayurvedic literature. The vrishya property of the drug indicate its beneficial effects on testes. However, such property of NB has not been worked out experimentally in animals. Considering the above facts it was planned to study the effects on NB on testes of albino rat.

As observed by other workers, out experimental study also revealed toxic effects of Cd cl2 on testes in the form of fatty vacuolization and necrosis of germinal epithelium of seminiferous tubules.

On simultaneous administration of Cd cl2 and the test drug it was observed that the degree of degeneration of germinal epithelium of seminiferous tubules and number of animals affected was in proportion to the dose of the NB administered higher the dose of test drug i.e lesser the (from 3 mg to 12 mg) toxic effects of Cd cl2 and the best result was noted at 12 mg dose where minimal degenerative changes were noted in one out of four animals.

NB when administered on partially damaged testes (by Cd Cl2) the drug showed its regenerative potential. The drug showed best regenerative capability at 12 mg dose. Where only one animals out of 4 revealed minimal degenerative change. At 6 mg dose level though all the four animals showed degenerative changes these were of very mild nature.

Summary and conclusion

The present study reveals that Cd Cl2 is toxic to testicular germinal epithelium and its effects can be minimize by NB. It has regenerative potential also. In higher doses the drug is very effective. Thus these findings are well corroborated with the Ayurvedic concept of vrishya property of NB. However to arrive at the valid conclusion further studies on larger groups of animals at different dose and duration are needed.

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Abbreviations used

NB – Naga bhasma.
Cd Cl2 – Cadmium chloride
