

ALTERATION OF EPIDIDYMAL FUNCTION AFTER CONVULVUS MICROPHYLLUS (WHOLE PLANT) ETOH ADMINISTRATION IN MALE RATS

***Dr. Sulochana Sharma**

Abstract

The epididymis performs an important role in the maturation of spermatozoa including their acquisition of progressive motility and fertilizing ability. This study is to find a reversible male contraceptive which would be effective at a post testicular site. The inhibitory effect of alcoholic extract of Convolvulus microphyllus on sperm motility and density of epididymal sperms was noticed in dose dependent manner. The drug may also interfere with the production of glucose from other sources by glycolytic pathway as evident from the significant fall in testicular glycogen. There was a significant decrease in level of sialic acid and protein of testes and epididymis.

INTRODUCTION

The mammalian epididymis is one of the longest single duct in the body. It allows post meiotic gamete development that result in immature testicular sperms to acquire the potential to fertilize eggs. Sperm maturation takes place principally within the caput and corpus regions of the epididymal duct whereas the caudal region serves more as a storage area where mature sperm are maintained.

The highly ordered and precise process that occurs during the genesis and maturation of sperm should allow numerous approaches to their regulation and thus, to male contraception.

A review of literature shows that a number of contraceptive devices, drugs and methods are used to regulate and to control fertility. Methods employed either have direct effects on hormonal milieu or disrupt spermatogenesis or interfere with sperm maturation i.e. epididymal function or inhibit sperm transport, ejaculation and deposition etc.

Efforts to evolve new compounds as male contraceptive agents being effective, safe, reversible and relatively more rapid in its antifertility action.

There is always a hope that search among medicinal plants may provide certain plant products as reliable contraceptive agents. In the past decade a bulk of literature has been accumulated on the efficacy of plant extracts as fertility regulating agents. Indigenous medical system represent the end product of many years of experimentation and observation.

MATERIALS AND METHODS

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Plant extract:-

Convolvulus microphyllus (Sieb) Convolvulaceae known as Shweta Shankhpushpi. It has a great value in the ayurvedic system of medicine. It occurs as an ingredient in Brahma-rasayang, Aindrarasayana, Aagastyaharitki, Medhyarasayana, Manasamitram and in number of other composite drugs. The plant is shade dried and powdered. The alcoholic extract was prepared by soxhalation.

Animal model:-

Mature male albino rats weighing about 200gms maintained on standard diet (Hindustan Lever Ltd.) and water ad libitum were distributed in groups (5 in each). One is for vehicle treated (Distilled water) controls and remaining three for ethanol extract dissolved in water at the dose level of 100, 200 & 300 mg/day/rat.

Fertility test:-

The animals fertility test was carried out with females on day 55 of experiment in the ratio of 1:3. The animals were sacrificed on last day of experiment. The epididymis was taken out, cleaned the altering tissues and cauda separated for sperm motility and density.

Sperm motility:-

Sperm motility was assayed by the method of(1). The epididymis was removed immediately after anaesthesia and known weight of cauda epididymis was gently teased in a specific volume of physiological saline (.9% NaCl) to release spermatozoa from the tubules. The sperm suspension was examined within five min. after their isolation from epididymis. The results were determined by counting both motile and immotile sperms in at least ten separate and randomly selected fields. The results were finally expressed as percent fertility.

Sperm density:-

Sperm density was assayed(1). Briefly total number of sperms were counted using haemocytometer after further diluted sperms suspension from cauda epididymis and testes. The sperm density was calculated in million/ml as per the dilution.

Biochemistry :-

The total protein(2), sialic acid(3) and glycogen(4) were assayed in testes and epididymis.

Statistical analysis:-

All the values were expressed in terms of mean value \pm standard error. The different groups were compared among each other using student's t test.

RESULT AND DISCUSSION

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Post drug administration of *Convolvulus microphyllus* resulted in strong impairment of sperm motility. Two important enzyme systems are associated with sperm activity-

(1) acrosomal enzymes and (2) enzymes involved in glycolysis, citric acid cycle and oxidative phosphorylation (5). According to (6) inhibition of any of these enzyme systems can be utilized for the development of new contraceptives.

(7) have further suggested that reduction of sperm motility after gossypol treatment may be due to inhibition of ATPase activity in the spermatozoa. (8) has described that the motility of spermatozoa is regulated by the activity of mid piece enzyme. Inhibition of their activity will therefore lead to immobilization (9).

It has been suggested that spermatids being the target tissue, are more susceptible to plant extract during their maturation in the seminiferous tubules as previously observed with gossypol treatment (10), (11) and (12).

The sperm density decreased after *Convolvulus* feeding concurrently with an increase in the percentage of non motile and immature spermatozoa. Sperm count is considered to be one of the important parameters that affect fertility. Sperms have two principle attributes, namely motility and fertilizing ability (13). Motility is an important prerequisite for fertilization. Any negative impact on motility would seriously affect fertilizing ability(14). Decrease in sperm count and motility and an increased number of abnormal sperms is associated with infertility (15), (16). The inhibition of rat epididymal sperm was also observed by (17), (18), (19) and (20).

Testicular sperms lack motility and fertility but they acquire these abilities by the time they reach the tail of epididymis (21). The acquisition of these abilities by sperm depends upon their passage through a specific environment, which in turn, is regulated by the absorptive and secretory activities of the epithelium lining the excurrent ducts (22).

A fall in glycogen level may be due to the interference in glycogenolysis. Since glycogen is an energy source for general metabolism and constant supply of glucose is essential for proper functioning of epididymis. The decrease in testicular glycogen is in accordance with the view of (23). Who speculated that protein synthesis in spermatogenic cells is dependent on glucose. A marked decrease in glycogen content could affect protein synthesis and thus subsequently inhibit spermatogenesis.

Sialic acid might play a physiological role in the process of fertilization. Its level in spermatozoa and luminal plasma may influence maturation of spermatozoa in the epididymis and is involved in the maintenance of ionic balance, in the antigenic interaction between epididymis and spermatozoa and in the stabilization of the acrosome and at the structural integrity of spermatozoa. (24) suggested that the lower content of sialic acid in seminal plasma have a deteriorating effect on structural integrity of sperm. This further supports our results for lowered sialic acid concentrations in epididymides.

Convolvulus treatment in rats resulted in significant reduction in protein contents of testes and

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epididymis. (25) suggested that nuclear changes and disorganization of the granular reticulum of epididymis seriously interfere with the cell's capacity for protein synthesis. The nucleolar changes result in disturbance of ribosome production while the progressive loss of polyribosomes from the reticulum is probably an indication of a dissociation of ribosomes from m-RNA and cessation of protein synthesis (26). (27) suggested the reason for reduction in testicular protein as the absence of later stages of spermatogenesis. The decrease or increase in protein concentration depends upon the absence or presence of spermatozoa and luminal fluid.

CONCLUSION

In the present study dose dependent treatment of *Convolvulus microphyllus* (whole plant) EtOH extract caused marked alteration in the epididymal functions. Significant loss of sperm motility and density in testicular and epididymal sperms of treated male rats. Biochemical analysis of protein, sialic acid and glycogen further supports the antifertility like activity of *Convolvulus microphyllus* at post testicular site.

OBSERVATION:-

Table No. 1: Sperm Dynamics And Tissue Biochemistry Of Control And *Convolvulus Microphyllus* Treated Intact Male Rate (Mean \pm SEM of 5 Animals)

GRO-UPS	TREATMENT	SPERM MOTILITY (%)	STERM DENSITY (million/m ³)		FERTI-LITY TEST	GLY-COGEN (mg/gm)	SIALIC ACID (mg/gm)			PROTEIN (mg/gm)		
			Testes	Cauda epididy-mides			Testes	Caput Epidi-dymides	Cauda Epidi-dymides	Testes	Caput Epidi-dymides	Cauda Epidi-dymides
I.	Control	69.16 ± 1.26	3.66 ± 0.24	39.79 ± 1.01	100(+)	2.06 ± 0.01	4.30 ± 0.06	5.90 ± 0.002	6.91 ± 0.01	166.52 ± 3.18	216.2 ± 5.31	248.86 ± 1.32
II.	100mg/day for 60 days	56.94** ± 1.74	2.47* ± 0.13	30.61** ± 0.36	83(-)	1.64** ± 0.06	2.82** ± 0.06	3.69** ± 0.12	4.26** ± 0.09	133.32** ± 2.09	177.76** ± 2.09	199.98** ± 2.09
III.	200mg/day for 60 days	47.78** ± 5.36	0.98** ± 0.16	15.9** ± 0.87	100(-)	1.16** ± 0.06	2.52** ± 0.01	3.32** ± 0.02	3.51** ± 0.01	119.98** ± 2.09	164.43** ± 2.09	186.45** ± 2.09
IV.	300mg/day for 60 days	28.13** ± 1.23	0.65** ± 0.07	12.65** ± 4.85	100(-)	0.68** ± 0.06	2.32** ± 0.04	3.06** ± 0.02	3.29** ± 0.02	106.65** ± 2.09	151.09** ± 2.09	173.32** ± 2.09

ns = nonsignificant * = P 0.01 compared with control ** = P 0.001 compared with control

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