CHAPTER ONE
INTRODUCTION

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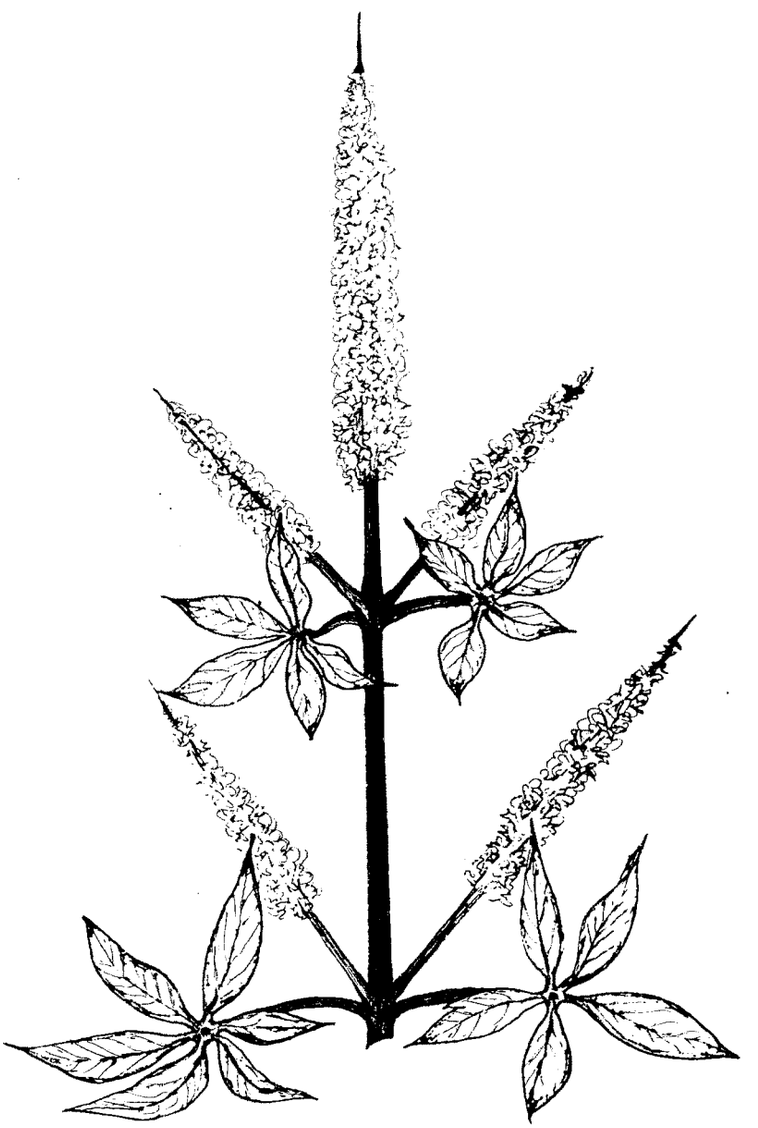
PLATE NO. 1



Vitex - Agnus castus (Linn.)

CHAPTER ONE

INTRODUCTION

1.1. General introduction :

Survival of human race as a biological species - is a problem, we are facing due to overpopulation. This tremendous pressure of population, creating so many problems all over the world. It affects almost all fields of human interest. Malnutrition, ill - health, poverty and shortage of basic needs are the few consequences of the population explosion. This has alarmed the scientist and thinkers of the world to find out solution and search for various birth controlling methods.

There are various methods of contraception such as surgical procedures, bioactive devices, mechanical devices and drugs for the control of fertility. But still newer, easier safer and most effective methods are being searched.

Female reproductive system has been studied for a long time for a control of fertility; the area male physiology of reproduction has bee neglected in this aspect. But now many Biologists are studying the alterations induced by various chemicals and plant preparation in the male reproductive system leading to aspermatogenesis. It is observed that chemical produce many toxic effects in body. Therefore number

of research workers, now actively engaged in exploring the effects of various plant preparations, having minimal side effects, on the male reproductive system, inducing sterility.

1.2. Review of plants having antifertility activity :

Following is a brief review of the plants affecting fertility of male animals.

Extract of roots of Lithospermum impaired development of the gonads and accessory sex organs of male rats (Plunket and Noble, 1951). Punica granatum produced infertile matings in rats and guinea pigs (Gujaral et al., 1960). Joshi et al., (1965) reported antispermatogenic property in the extract of the bark of Hippophae salicifolia. Antifertility effects of Butea frondosa were demonstrated in mouse and rats (Razdhan et al., 1969) and in house sparrow (Shrivastava, 1982). ROC - 101, a herbal preparation composed of a mixture of three different plants, impaired fertility and caused sterility in male mice (Munshi and Rao, 1972). Dixit et al., (1974) observed that an extract of Cannabis species caused testicular disfunction in frog. Oscimum sanctum induces reversible aspermatogenesis in male mice (Kashinathan et al., 1972), Opium seed extract was shown to decrease number and size of spermatocytes and decrease size of seminiferous tubules without

any alterations in Leydig cells in pigeon (Vyas and Singh, 1976). Aristolochia indica extract induces disfunction of seminiferous epithelium (Pakrashi and Pakrashi, 1977). Malvaviscus conzanttii extract induced changes in histoarchitecture of testis in bats (Dixit, 1977). Fresh aquous extract of Momordica charantia when administered to fertile male albino rats, altered differential count of germ cells in the testis (Biswas et al., 1977). Saxena et al., (1977) demonstrated that the saponine obtained from Blighia sapida had 100% spermicidal activity. Hot alcoholic extract of seads of Butea monosperma had antifertility activity. (Dreishbach, 1963; Khanna and Chaudhary, 1968). Singh et al., (1978) studied ten plants for their antifertility activity. The plants were Anona squamosa, Buddleja asiatica, Celastrus paniculata, Cyperus rotundus, Hibiscus rosa sinensis, Luffa acutangula, Mentha arvensis, Sapindus trifdiatus, Vitex negundo and Withania somifera. Out of these only Vitex negundo was found to be effective in male animals. Testicular degeneration was shown aqueous ethanolic extracts of Vinca rosea and Embelia ribes (Chauhan et al., 1979). Artabotrys odoratissimus caused temporary sterility (Prakash, 1979). Garg (1979) demonstrated certain testicular necrotic changes with the extract of Calotropis procera. Hibiscus rosa sinensis

extract caused damage to testicular germinal epithelium (Kholkute, 1977). In sparrow the extract of Hibiscus rosa sineus flowers decreased the weight and volume of gonads in both the sexes (Singh et al., 1980). Plumbagin, an active principle isolated from Plumbago zeylenica roots has profund influence on male reproductive system in rats (Shantakumari et al., 1981) and in dogs (Bhargava, 1984). Shrivastava (1981) reported antifertility activity with Butea monosperma flower extract in house sparrow. Seth et al., (1981) observed that Oscimum extract reduced sperm count and sperm motility in male rats. Dixit and Joshi (1982) reported that garlic powder reduces body weight and testicular weight significantly. Hoffer (1983) studied effects of Gossypol and observed the presence of severaly damaged and entirely normal seminiferous tubules adjacent to one another in the same section. Toro (1984) with Vinca alkaloids from Vinca rosea leaves, Shah (1985) with Daucus carota seeds, Sohani (1985) with Vitex negundo leaves and Awati (1985) with Butea monosperma leaves extracts reported various changes on the male reproductive organs in albino rats, leading to aspermatogenesis. Oscimum sanctum leaves decrease sperm count, spermmotility and weight of male reproductive organs in albino rats. Solanum xantho-carpum brought about depopulation of spermatogenic elements

in seminiferous tubules (Rao, 1988). Chinoy et al., (1988) reported alterations in the histoarchitecture of the testis in albino rats with Vinca rosea leaves extract. Gossypol known to inhibit spermatogenesis and result into sterility (Bhiwagade et al., 1988; Nair et al., 1988; Bhiwgade and Nair, 1989; Nair and Bhiwgade, 1989). Celastrus paniculatus administration reported to exhibit antifertility and antispermatogenic effects on male rats (Wango, 1988). Piper betle leaves extract decreased number and motility of sperms in albino rats (Hiremath, 1988; Adhikary et al., 1989). Oleanolic acid, a triterpene isolated from Eugenia jambolana flowers decreases fertilizing capacity of the male rats (Rajsekaran et al., 1988). Bhargava (1988) studied five plant products and found all of them effective in females as well as in males. These plant products and plants were Aristolic acid (Aristolochia indica), butin (Butea monosperma), embelin (Embelia ribes), plumbagin (Plumbago zeylenica) and solasodine (Solanum xanthocarpum). Definite impairment in the spermatogenesis with Malvadin chloride isolated from Malvaviscus couzanttii was reported by Bhargava (1988, b) in the langur monkeys. Crude extract combinations of Azadirachta indica, piper longum and Emblica officinalis exhibited conspicuous histological changes in testes in rats (Reddy et al., 1989). Alkaloid of Tylophora

asthamatica caused marked changes in Testis of rats (Dikshith et al., 1990). Antifertility effects of Andrographis paniculata on the male reproductive system were reported by Akbarsha et al., 1988 a; 1988 b; 1988 c; 1990. Jadhav (1988) showed reducticn in the fertilizing capacity of male rats after the administration of plumbagin. Syzygium cumuni seed extract shown to decrease body weight and reproductive organ weights of male rats with alterations in histoarchitecture of the organs (Ambaldage, 1990). Patne (1990) reported many changes in testes and accessory reproductive organs with Picrorhiza kurroa in albino rats. Aqueous extract of Vinca rosea leaves caused regressive changes in testes of mice (Murugavel and Akbarsha, 1991).

1.3. Reasons that lead to undertaking of the present investigation :

A critical evaluation of the earlier review of the plant extract induced aspermatogenesis reveals that :

i. Intention of most of these studies is the 'destruction' caused in the seminiferous tubules by the 'active principle' in the plant extract. Not much is known about the mechanism of such destruction.

ii. Some workers have focussed their attention on histological alterations in testis only, some have only studied

epididymis, others have selected either seminal vesicle or prostate gland only.

It is hence felt desirable that in a study of plant extract induced aspermatogenesis all the reproductive organs should be studied simulteneously.

With these views, it was decided to study the effects of Agnus castus on the male reproductive organs of the albino rats. To make the study consolidated it was decided to study changes in body weight and organ weights and histological changes in testes and the accessory reproductive organs of the laboratory albino rat.

1.4. Choice of the plant :

1.4.1. Botanical consideration :

Agnus castus is commonly called as "Chaste tree" or "Monk's pepper tree" or "Hemp tree". It is a strongly aromatic Wooly - tomentose shrub or a small tree, found from the Mediterranean region through south west Asian countries upto Baluchistan. It is occasionally cultivated in Indian gardens.

Leaves are long petiolate digitately five or rarely seven foliate and velvety, The leaflets lanceolate. Flowers are pale purple or violet and are compact, subsessile, forming long

spiked verticilasters. Fruits are drupe, spherical, obtuse.

1.4.2. Classification of Agnus castus :

Systematic position of the plant in the plant kingdom is as follows :

Division	-	Spermatophyta
Sub division	-	Angiospermae
Class	-	Dicotyledons
Sub class	-	Gamopetalae
Order	-	Polemoniales
Family	-	Verbenaceae
Genus	-	Agnus
Species	-	castus

1.4.3. Biological properties of Agnus castus :

Berries are used as a stimulant, diuretic, abortificient, alexiteric, given in enlargement of liver and spleen and in dropsy. The fruits have antiaphrodisiac action.

Leaves are used for pains due to chills. The plant also cure eye diseases and stomach - ache.

The seeds are prescribed in colic and in veternary surgery for the same purpose to horses. (Indian materia medica,

1976, Kirtikar and Basu, 1980; Wealth of India, 1982).

1.4.4. Chemical composition of Agnus castus :

The leaves contain an essential oil (Yield 0.75 % on dry basis). It has following constants = d 20°, 0.910, (α)D - 12.5°, n 1.477, acid val. 11.5 and ester val. 42.0. The constituents present in the oil are eucalyptol, piene, sabiene and quinone.

The leaves also contains agnuside ($C_{22}H_{26}O_{11}$, m.p. 146°, yield 0.3 %), aucubin, luteolin - 7 - glucoside, α - D - glucoside of tetrahydroxymonomethoxyflavone ($C_{22}H_{24}O_{12}$, m.p. 245°), casticin and homoorientin (isoorientin).

Gaschromatographical analysis of leaf oil show 31 compounds of which major components are 1, 8 - cineole (50.9 %), sabinene (10.8 %), α - pinene (9 %), terpinen - 4 - 01 (4.8 %), p - cymene (4.2 %), limonene (2.5 %) and α - terpenoid (2.3 %), eight sesquiterpene hydrocarbons (2.8 %) and four sesquiterpene alcohols (2.4 %) (Ekundayo et al., 1990).

Berries contain a bitter principle called castine, a violet aerid substance, a free acid and oil (Indian materia medica, 1976).

The seeds yield 10 % of dark oil (dry basis) which shows progesterone like effect on mature female rats.

1.5. Reasons for selecting Agnus castus for present investigation :

1. Agnus castus which is used in the present investigation is a mother tincture, derived from the berries of Vitex agnus castus.
2. Indian Vitex species (Vitex negundo) has antifertility properties in the male animals (Singh et al., 1978; Bhargava and Dixit, 1982; Sohani, 1985).
3. Agnus castus is antiaphrodisiac and known to affect the sexual sphere causing depression of function, lowering of sexual vitality (Clarke, 1983).

Thus Agnus castus causes antifertility, sterilization and reduction in libido. The available literature showed that no such work is carried out on the male albino rats. Hence it was thought desirable to study the details of the histological alterations to confirm the above findings. To get a clear insight into the mechanism of aspermatogenesis induced by the drug Agnus castus, not only testes but accessory reproductive organs were also decided to be studied.

1.6. Choice of Parameter :

As mentioned earlier it was decided to study Agnus castus induced alterations in adult male albino rats. The following parameters were selected for studies.

1. Body weight.
2. Organ weight (Gonadosomatic index).
3. Histology.
4. Fertility.

Alterations in the organ weights may be an indication of alterations in the structural or functional state.

The functional significance of any organ or system, normal or affected will be known only when the structural alterations are studied and this is possible only with histological studies. Therefore a thorought knowledge of histological alterations in the testis and accessory reproductive organs caused due to the administration of Agnus castus is essential for the understanding of the exact locus of the action during such changes.

1.7. Presentation of the thesis :

It was selected to divide the present thesis into eight chapters. The first chapter being an introduction giving a

review of the literature on agents causing aspermatogenesis in vertebrates, reasons that stinulated the present investigation, details of the plant Agnus castus and reasons for the selection of the parameters for the present investigation. The second chapter devoted to the detailed description of the materials and methodology and techniquies employed in the present investigation.

Third, fourth, fifth, sixth and seventh chapters deal with the alterations caused by administration of Agnus castus in the weights and histoarchitecture of testes, epididymis, vasa deferentia, seminal vesicles and prostate glands of albino rats, respectively. These five chapters contain a brief review of earlier work, detailed report on observations and discussion; the observations have been duely compared with the available literature on effects of various antifertility agents and certains conclusions have been arrived at. The observations are well supported by tabular data, graphical representation and by photomicrographs of histological observations.

In the final chapter, eighth, of the thesis, summary with concluding remarks is given. Bibliography of exhaustive literature is cited at the end.