

# Female Anti-Fertility Screening of Plants *Mucuna prurita*, *Mesua Ferrea* and *Punica granatum* on Rats

Sonu Sharma\* and Ashish K. Sharma

NIMS institute of Pharmacy, NIMS University, Shobha Nagar, Jaipur, India.

## ABSTRACT

India has a rich culture of medicinal herbs and spices, including Ayurvedic, Unani, Siddha and other traditional medicines but only very few have been studied chemically and pharmacologically for their potential medicinal value. According to the World Health Organization, most populations still rely on traditional medicines for their psychological and physical health requirements. Herbal medicines are in great demand in both developed and developing countries as a source of primary health care owing to their attributes having wide biological and medicinal activities, high safety margins and lesser costs. As far as population growth is concerned India will be the leading country within few years of time span. Current pandemic population explosion demands an immediate betterment of new potential contraception. Thus, there is growing need to look for aphrodisiac more from natural plant or herbal origin as opposed to synthetic compounds which are known to cause severe unwanted side effects. In this regard, we have taken the plants *Mucuna prurita*, *Mesua ferrea*, *punica granatum* that has been used traditionally as antifertility agent in women and aphrodisiac, so that the couples without issue may be benefited for better acceptance in society and they may have better psychological health. In these plants so many secondary metabolites are present i.e carbohydrates, glycosides, alkaloids, tannins, phytosterol etc. All these three plants have pharmacological activity like Astringent, anthelmintic, nervine tonic, aphrodisiac, diuretic, vermifuge and stimulant, anodyne, antidotal, psychedelic, leucorrhoea, spermatorrhea, facial paralysis and powerfully aphrodisiac, leprosy, scabies, skin diseases, pruritus, haemorrhoids, ulcers, depresso, impotency, leucorrhoea, haemoptysis, cephalalgia, fever and cardiac debility.

**Keywords:** Pruritus, Haemorrhoids, Ulcers, Depressio, Impotency, Leucorrhoea, Haemoptysis, Cephalalgia, Fever, Cardiac Debility.

## INTRODUCTION

As far as population growth is concerned India will be the leading country within few years of time span. Current pandemic population explosion demands an immediate betterment of new potential contraception.<sup>1</sup> Family planning has been promoted through several methods of contraception, including oral contraceptives. Therefore, there is a need of drug which is effective but with lesser side effects.<sup>2</sup> Global search on anti-fertility agents is going on, to tackle the problem of population explosion. Many hormonal drugs are available for the purpose but they are not free from side effects. Hence, the search for a suitable product from indigenous medicinal plants is proposed which could be effectively used in the place of oral Pills.<sup>3</sup>

India has a rich culture of medicinal herbs and spices, including Ayurvedic, Unani, Siddha and other traditional medicines but only very few have been studied chemically and pharmacologically for their potential medicinal value. According to the World Health Organization, most populations still rely on traditional medicines for their psychological and physical health requirements. Herbal medicines are in great demand in both developed and developing countries as a source of primary health care owing to their attributes having wide biological and medicinal activities, high safety margins and lesser costs.<sup>4</sup> The practice of traditional medicine for the control of fertility in most parts of Africa is based on the use of plant medicines for many years. The fact that the herbal medicines have been employed for such a long time there are no reports on both ethno botanical and pharmacological profile of these plants.<sup>5</sup> In fertility regulation; the ancient literature has mentioned the use of a number of plants/preparations as abortifaciant and local contraceptives. WHO and ICMR provide systemic guidelines for the evaluation of anti-fertility plants to generate reproducible results, i.e., proper authentication and systemic screening

methods. Many plant preparations are reported to possess antifertility properties in ancient Indian literature. Many plants have been tested for their anti-fertility activity in laboratory animals. Only one plant through vascicine from *Adhatoda vasika* have been claimed to have abortifaciant properties but it could not be used clinically; the centchroman, non hormonal oral contraceptives also has not been proved very successful in phase IV clinical trials. Hence, the search needs to be continued.<sup>6</sup> this may reduce the population burden of our country.

Aphrodisiacs are the substances which are used to increase sexual activity and help in fertility. Sexual feelings are an inevitable part of life. The basic and fundamental purpose of sex and sexuality is the "continuation of progeny" and the survival of human race.<sup>7</sup> the sex is the most intimate, indispensable and an integral part of every individual and can be a source of pleasure and fulfillment. The sexual myths and misconception also leads to sexual dysfunctions.<sup>8</sup>

Infertility is a worldwide medical and social problem. It affects above 10-15% of married couples. WHO estimates that there is 60-80 million infertile couples worldwide.<sup>9</sup> Infertility in itself may not only threaten physical health but it can certainly have a serious impact on the mental and social wellbeing of infertile couple, especially in our country. In many countries the stigma of infertility often leads to marital disharmony, divorce or ostracism.<sup>10</sup> Research during the past two decades has an unfolded focus on impotence (erectile failure), premature ejaculation and male infertility. There are a number of prescription drugs which may act as sex stimulant and enhancing the sexual desire and activity in both men and women, although their use have shown significant improvement in treating sexual disorders, but at the same time they are not devoid of large number of side effects. These include arrhythmia, suicidal tendencies, mental disorders, tremors etc. The use of synthetic aphrodisiacs results in the dilatation of blood vessels in other parts of the body also, causing headache and even fainting. Other side effects include facial flushing, blurred vision and sensitivity to light which usually occur at higher doses.<sup>11</sup>

Thus, there is growing need to look for aphrodisiac more from natural plant or herbal origin as opposed to synthetic compounds which are known to cause severe unwanted side effects. In this regard, we have taken the plants *Mucuna prurita*, *Mesua ferrea*, *punica granatum* that has been used traditionally as antifertility agent in women and aphrodisiac, so that the couples without issue may be benefited for better acceptance in society and they may have better psychological health.

## PLANT PROFILE

### 1. *Mucuna prurita*

**Family:**

Leguminosae

**Common name:**

Kaunch Beej

**Habitat:**

It is found almost all over the country

**Parts used:**

Seeds, roots and legumes

**Pharmacological action**

Astringent, anthelmentic, nervine tonic, aphrodisiac, diuretic, vermifuge and stimulant, anodyne, antidotal, psychedelic, leucorrhoea, spermatorrhea, dyspepsia, colic, hemiplegic, and facial paralysis and powerfully aphrodisiac.<sup>12</sup>

**Chemical Constituents**

L-dopa, sulphur, manganese, 4-dihydroxy phenylalanine, glutathayon, lacithine, galic acid and glucoside. These seeds contain saturated fatty acid stearic and palmitic acid, oliac acid and linolinic acid.<sup>13</sup>

**Therapeutic classification index**

**Digestive system:** it is used in intestinal worms and colic

**Central nervous system:** it is used in paralysis, hemiplegia and other nervine disorders and spasms associated with Parkinson's or Bell's palsy.

**Genito- urinary system:** it is used in leucorrhoea and profuse menstruation

**Reproductive system:** it is used as an aphrodisiac and used in seminal weakness, spermatorrhea.<sup>14</sup>

### 2. *Mesua ferrea*

**Common name :**

Nagkeshar

**Family :**

Guttiferae

**Habitat :**

Eastern Himalayas, Assam, West Bengal, Western Ghats, Travancore, and Andaman

**Parts used:**

Flower (stamen), oil, bark, leaf, bud, seed.

**Medicinal uses :**

It has astringent, digestant, and ant-poisonous, antimicrobial, anti-inflammatory, antipyretic and anthelmintic. It is used in fever, itching, nausea, leprosy, skin disorders, erysipelas, bleeding piles, metrorrhagea, menorrhagea, excessive thirst, and sweating

**Uses as home remedy:**

1. In bleeding piles, it should be used in the dose of 3 gm along with mishri.
2. Its oil should be used externally to cure skin disorders.
3. Its paste or dusting of its powder is very effective to get relief from excessive sweating.

**Chemical composition:**

Mesulol, mammeisin, mamiflegin, mesulol sitosterol, octadecatriefloic, hexadecanolic. 1, 5-dihydroxyxanthone (ii), euxanthone 7-methyl ether (iv) and  $\beta$ -sitosterol, mesuanic acid,  $\alpha$ - and  $\beta$ - amyrrin,  $\beta$ - sitosterol, mesulol, mesuaferrol, Leucoanthocyanidin, mesuone, mammeigin.

**Medicinal properties:**

Digestive, carminative, constipating, anthelmintic, diuretic, expectorant, stomachic, haemostatic, aphrodisiac, febrifuge and cardiotoxic. They are useful in asthma, cough, hiccup, leprosy, scabies, skin diseases, pruritus, pharyngodynia, vomiting, dysentery, haemorrhoids, ulcers, dyspepsia, impotency, leucorrhoea, haemoptysis, cephalalgia, fever and cardiac debility. The seed oil is used in skin diseases. Pericarp of fruit is astringent and stomachic.<sup>15</sup>

**Circulatory system-**

Cardiac tonic and haemostatic, it is used in cardiac debility, rakta pitta and blood disorders.

**Central nervous system-**

Brain tonic it is useful in brain debility and hysteria.

**Respiratory system-**

It is used in cough induced by kapha, dyspnoea and hiccups as it alleviates kapha.

**Reproductive system-**

It is used as an aphrodisiac and as a haemostatic in menorrhagia.

**Digestive system-**

It is appetizer, mainly digestive, antidiabetic, antiemetic, antihaemorrhoid, astringent and vermicide; hence it is used in anorexia, distaste, dyspepsia, emesis, worms, bleeding piles, dysentery. It acts as a haemostatic in bleeding piles.

**Urinary system-**

Diuretic, hence useful in retention of urine.<sup>16</sup>

**3. Punica Granatum****Common Name :**

Annar

**Family Name :**

Lythraceae

**Part Used :**

Seed, Roots, Leaves

**Habitat :**

Southern Europe, Northern Africa, tropical Africa, Central Asia, India, America, California

**Constituents:**

Vitamin C, vitamin K, polyphenols, such as ellagitannins and flavonoids, Pomegranate seed oil contains punicalic acid, palmitic acid, stearic acid, oleic acid and linoleic acid. Juice, seeds and peel apparently contain steroid hormones, including estrone.<sup>17, 18</sup>

**Uses :** Diarrhea, dysentery and intestinal parasites. The seeds and juice are considered a tonic for the heart and throat, especially when sweet, pomegranate fruit is nourishing for (pitta or fire) systems and is

known as a blood builder. The astringent qualities of the flower juice, rind and tree bark are considered valuable for a variety of purposes, such as stopping nose bleeds and gum bleeds, toning skin, firming-up sagging breasts, and treating hemorrhoids. Pomegranate juice (of specific fruit strains) is also used as an eye drop, as it is believed to slow the development of cataracts. Pomegranate has been used as a contraceptive and abortifaciant by means of consuming the seeds, or rind, as well as by using the rind as a vaginal suppository. This practice is recorded in ancient Indian literature, in medieval sources, and in modern folk medicin.<sup>19</sup>

## 1. MATERIALS AND METHODS<sup>20,21,22</sup>

### 1.1. Animals

Female Swiss albino mice (18–22 g), Wistar albino rats 150–200 g, and immature female Wistar albino rats of 21–23 days old (40–60 g) were used in this study. They were procured from animal house, College of Pharmacy, NIMS University, Jaipur. The animals were acclimatized for ten days under laboratory conditions. They were housed in polypropylene cages and maintained at 27°C ± 2°C, relative humidity 65 ± 10% under a 12-hour light/dark cycle. The animals were fed with rodent pellet diet and water ad libitum. Animal ethical clearance for performing the experiments on animals was obtained from the Institutional Animal Ethical Committee (IAEC).

Each experimental group had a separate set of animals and care was taken to ensure that animals used for one response were not employed elsewhere. Animals were habituated to laboratory conditions for 48 h prior to experimental protocol which minimizes any nonspecific stress.

### 1.2 Acute Toxicity Study

The acute toxicity for ethanol extracts of *Mucuna prurita*, *Mesua ferrea*, *Punica granatum* and their combinations i.e. **Combination A** (*Mucuna prurita*+*Mesua ferrea*), **Combination B** (*Mesua ferrea* + *Punica granatum*), **Combination C** (*Mucuna prurita* + *Punica granatum*) and **Combination D** (*Mucuna prurita* + *Mesua ferrea* + *Punica granatum*) was determined in albino mice, maintained under standard conditions. The animals fasted overnight prior to the experiment, and fixed dose method was adopted as per OECD Guideline no. 420—fixed dose method.

### 1.3 Antifertility Activity

#### 1.3.1. Estrogenic Activity in Immature Female Rats

Immature female rats of Wistar strain 21–23 days old and weighing 40–60 g were used. They were divided into nine groups of six animals each. The various groups were treated as follows:

- **Group I:**  
Control (saline solution) p.o.,
- **Group II:**  
Reference standard (Ethinyl Estradiol 0.02 mg kg<sup>-1</sup>, p.o.),
- **Group III:**  
Ethanol extract of *Mucuna prurita* (200 mg kg<sup>-1</sup>, p.o.),
- **Group IV:**  
Ethanol extract of *Mesua ferrea* (200 mg kg<sup>-1</sup>, p.o.),
- **Group V:**  
Ethanol extract of *Punica granatum* (200 mg kg<sup>-1</sup>, p.o.),
- **Group VI:**  
Ethanol extract of **Combination A** (*Mucuna prurita*+*Mesua ferrea*) (200 mg kg<sup>-1</sup>, p.o.),
- **Group VII:**  
Ethanol extract of **Combination B** (*Mesua ferrea* + *Punica granatum*) (200 mg kg<sup>-1</sup>, p.o.),
- **Group VIII:**  
Ethanol extract of **Combination C** (*Mucuna prurita* + *Punica granatum*) (200 mg kg<sup>-1</sup>, p.o.),
- **Group IX:**  
Ethanol extract of **Combination D** (*Mucuna prurita* + *Mesua ferrea* + *Punica granatum*) (200 mg kg<sup>-1</sup>, p.o.),

The treatment was given for six days, 24 h after the last treatment; all the animals were sacrificed by decapitation, and uteri were dissected out, cleared off the adhesive tissue, blotted on filter paper, and

weighed quickly on a sensitive balance. The tissues were fixed in Bouin's fixative for 24 h, dehydrated in alcohol, and embedded in paraffin. The paraffin blocks were sectioned at 6  $\mu$  and stained with hematoxylineosin solution (H and E Stain) for histological observations.

### 1.3.2 Anti-Implantation Activity

Female rats of proestrus phase were kept with male rats of proven fertility in the ratio of 2 : 1. The female rats were examined in the following morning for evidence of copulation. The animals which showed thick clumps of spermatozoa in vaginal smear was separated from the male partner. Only the rats with normal estrous cycles were selected for the experiment. The animals divided into nine groups of six animals each. The various groups were treated as follows:

- **Group I:**  
Control (saline solution) p.o.,
- **Group II:**  
Reference standard (Ethinyl Estradiol 0.02 mg kg<sup>-1</sup>, p.o.),
- **Group III:**  
Ethanol extract of *Mucuna prurita* (200 mg kg<sup>-1</sup>, p.o.),
- **Group IV:**  
Ethanol extract of *Mesua ferrea* (200 mg kg<sup>-1</sup>, p.o.),
- **Group V:**  
Ethanol extract of *Punica granatum* (200 mg kg<sup>-1</sup>, p.o.),
- **Group VI:**  
Ethanol extract of **Combination A**(*Mucuna prurita*+*Mesua ferrea*) (200 mg kg<sup>-1</sup>, p.o.),
- **Group VII:**  
Ethanol extract of **Combination B**(*Mesua ferrea* + *Punica granatum*) (200 mg kg<sup>-1</sup>, p.o.),
- **Group VIII:**  
Ethanol extract of **Combination C**(*Mucuna prurita* + *Punica granatum*) (200 mg kg<sup>-1</sup>, p.o.),
- **Group IX:**  
Ethanol extract of **Combination D**(*Mucuna prurita* + *Mesua ferrea* + *Punica granatum*) (200 mg kg<sup>-1</sup>, p.o.),

The extracts were administered orally from day 1 to day 7 of gestation. On the 10th day, laparotomy was carried out under light ether anesthesia in sterile conditions. The uteri were examined to determine the number of implantation sites; the numbers of corpora lutea in ovaries were recorded. The abdomen was sutured, and the animals were left in cages. The drugs were administered orally again for 3 days (days 14–16). On the 18th day, laparotomy was carried out again for evaluating the early abortifacient activity. The percentages of anti-implantation activity were calculated using the formula given in

$$\% \text{ Abortifacient activity} = \frac{\text{Number of resorptions}}{\text{Number of corpus luteum}} \times 100$$

$$\% \text{ Anti-implantation activity} = \frac{\text{Number of implantations}}{\text{Number of corpus luteum}} \times 100$$

### 1.4 Statistical Analysis

Values were expressed as  $\bar{x} \pm s$  from 6 animals. Statistical difference in the mean analyzed using one-way ANOVA followed by Turkey's multiple comparison tests  $P < 0.05$  was considered as statistically significant.

## 1.5 RESULTS

### 1.5.1 . Acute Toxicity Study

No morbidity and mortality were detected till 2000 mg kg<sup>-1</sup>, p.o. for all ethanol extracts; hence, ethanol extracts and their combinations were considered to be safe till 2000 mg kg<sup>-1</sup>, p.o.

## 1.5.2 Antifertility Activity

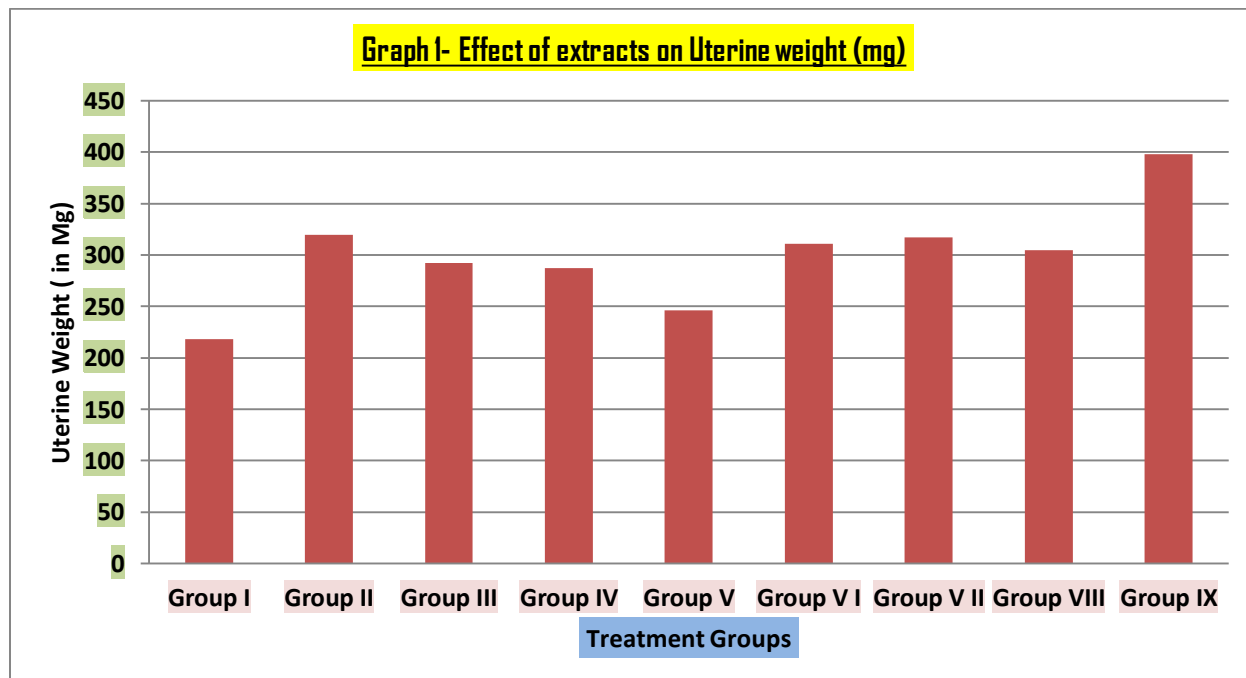
### 1.5.2.1 Estrogenic Activity on Immature Female Rats

Treatment with ethanolic extracts (200 mg kg<sup>-1</sup>, p.o.) and their combinations (200 mg kg<sup>-1</sup>, p.o.) had showed significant increase in uterine weight in a dose-dependent manner compared to vehicle control. The estrogenic effect of Combination D extract at 200 mg kg<sup>-1</sup> p.o. was comparable with reference standard Ethinyl estradiol (0.02 mg kg<sup>-1</sup>, p.o.). Furthermore, the ethanolic Combination D extract at 200 mg kg<sup>-1</sup> offered more potent estrogenic activity than the reference standard ethinyl estradiol. The extract significantly increased the weights of uteri (Table 1), and results obtained were also correlated and supported by the histopathological findings, where the combination of ethanolic extracts A, B and C (200 mg kg<sup>-1</sup>, p.o.) showed synergistic effect showed significant increase in the height of luminal epithelium and loose and edematous stroma with stimulated uterine glands, while the individual ethanolic extract (200 mg kg<sup>-1</sup>, p.o.) showed moderate increase in the height of luminal epithelium with stimulated uterine glands (Figures 1, 2, 3, 4, 5, and 6)

**TABLE 1: EFFECT OF SELECTED PLANT DRUG EXTRACTS AND THEIR SYNERGISTIC (COMBINATION) EXTRACTS ON UTERINE WEIGHT OF IMMATURE FEMALE RATS**

Group	Extracts/drug	Dose (mg kg <sup>-1</sup> )	Uterine weight (mg)
Group I	Control (saline solution)	-	218.27±16.79
Group II	Ethinyl Estradiol ( Control Standard)	0.02 mg kg <sup>-1</sup> , p.o	319.57±19.24**
Group III	Ethanolic extract of <i>Mucuna prurita</i>	200 mg kg <sup>-1</sup> , p.o	292.34±16.27*
Group IV	Ethanolic extract of <i>Mesua ferrea</i>	200 mg kg <sup>-1</sup> , p.o	287.45±57.24*
Group V	Ethanolic extract of <i>Punica granatum</i>	200 mg kg <sup>-1</sup> , p.o	246.27±64
Group V I	Ethanolic extract of <b>Combination A</b> ( <i>Mucuna prurita</i> + <i>Mesua ferrea</i> )	200 mg kg <sup>-1</sup> , p.o	311.24±12.47**
Group V II	Ethanolic extract of <b>Combination B</b> ( <i>Mesua ferrea</i> + <i>Punica granatum</i> )	200 mg kg <sup>-1</sup> , p.o	317.29±15.98**
Group VIII	Ethanolic extract of <b>Combination C</b> ( <i>Mucuna prurita</i> + <i>Punica granatum</i> )	200 mg kg <sup>-1</sup> , p.o	304.87±29.32**
Group IX	Ethanolic extract of <b>Combination D</b> ( <i>Mucuna prurita</i> + <i>Mesua ferrea</i> + <i>Punica granatum</i> )	200 mg kg <sup>-1</sup> , p.o	398.27±54.27***

Values are mean ± SEM (n=06), \*P<0.05, \*\*P<0.01, and \*\*\*P<0.001 as compared to control group.



Histopathological studies are supported by following figures

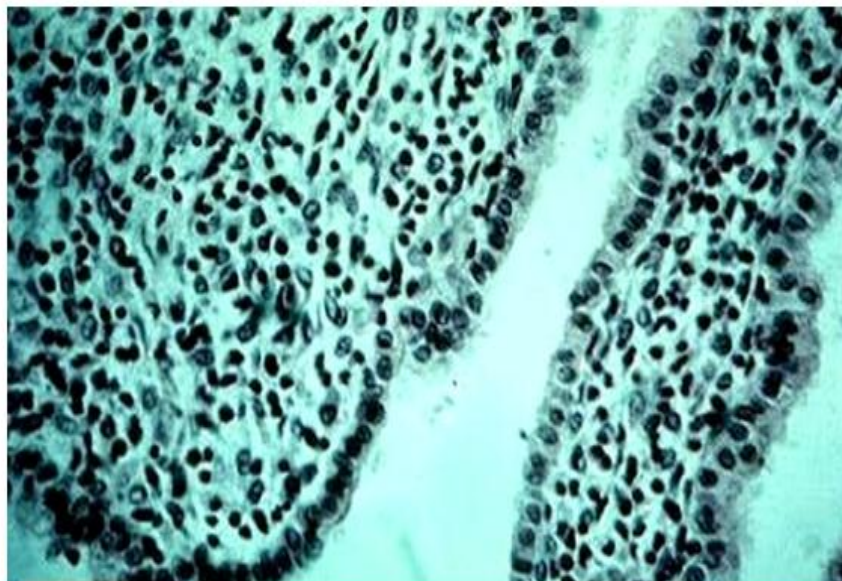


Fig. 1: Photomicrograph showing section of uterus indicating surface epithelium with no secretory activity (control group) HE 300x

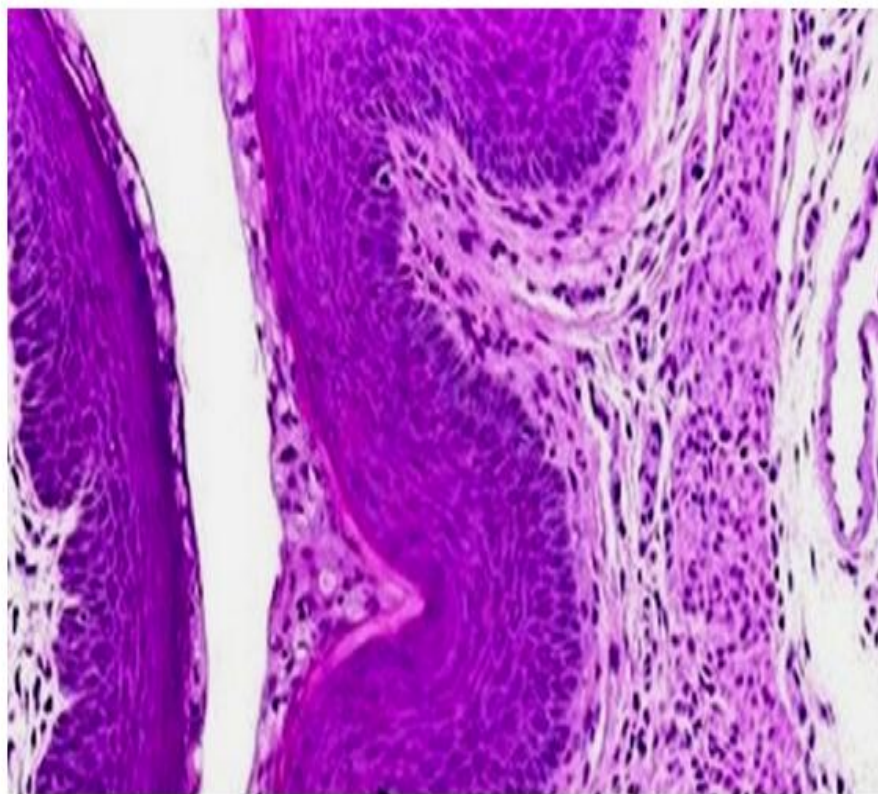
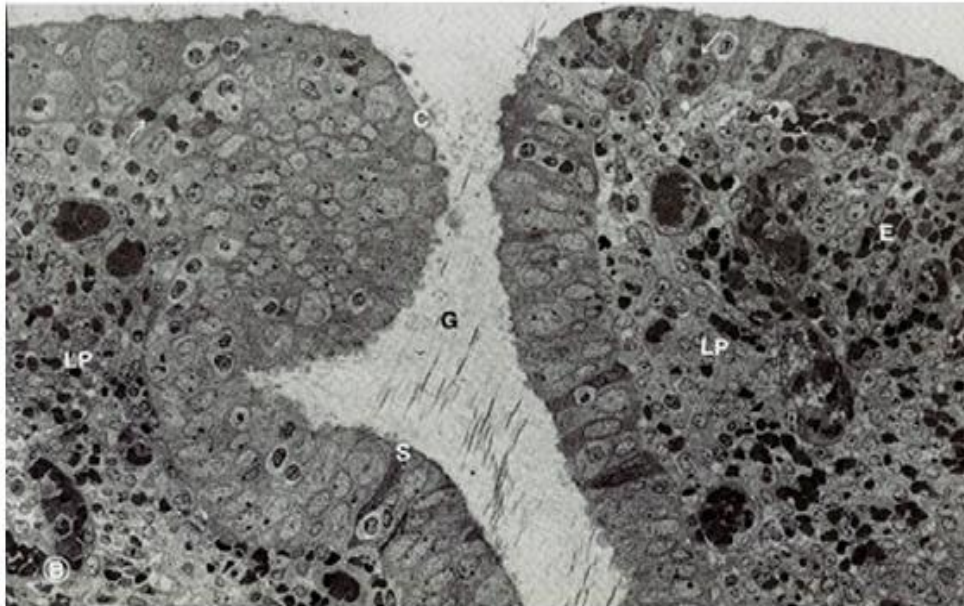
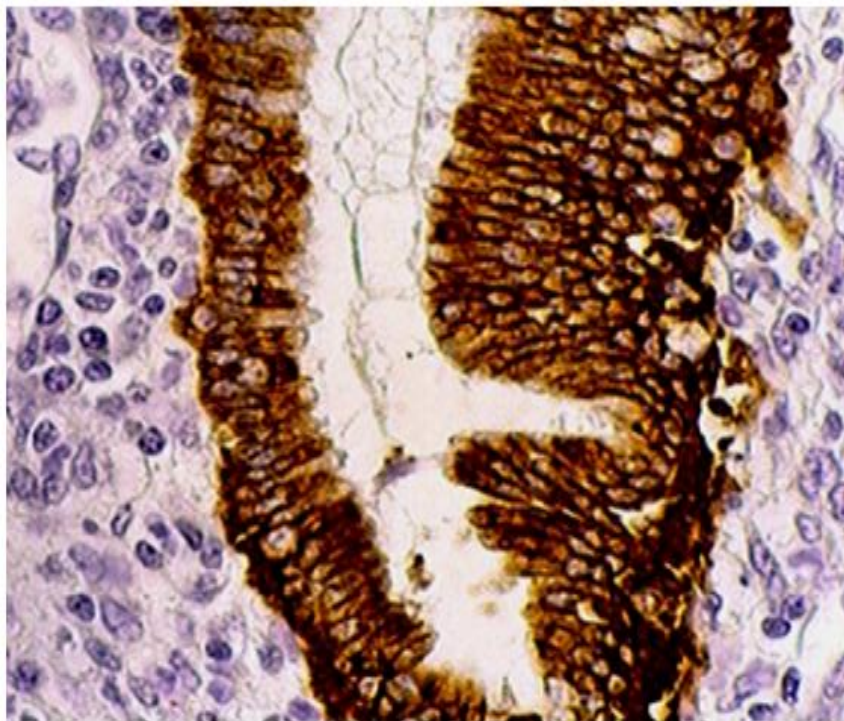


Fig. 2: Photomicrograph showing section of uterus indicating increasing height of luminal epithelium (Ethinyl estradiol) HE 300x

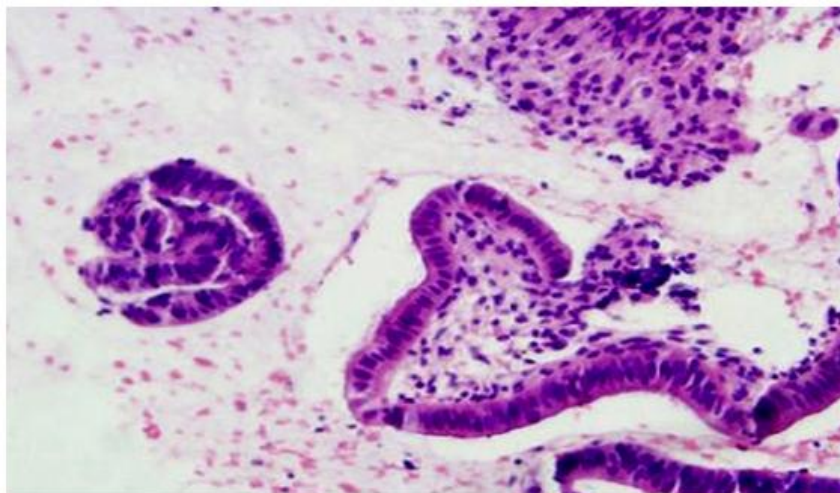


**Fig. 3: Photomicrograph showing section of uterus indicating increase in height of luminal epithelium (Combination C extract: 200 mg kg<sup>-1</sup>) HE 300x**

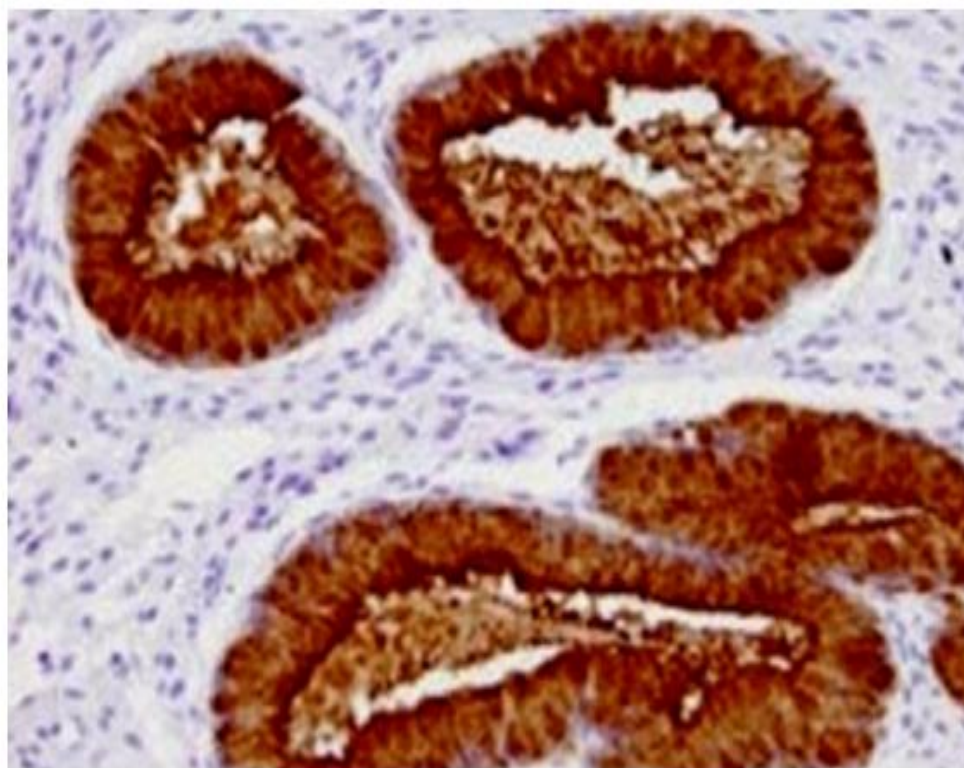


**Fig. 4: Photomicrograph showing section of uterus indicating increase in height of luminal epithelium and loose and edematous stroma with stimulated uterine glands (Combination D extract: 200 mg kg<sup>-1</sup>) HE 300x**





**Fig. 5:** Photomicrograph shows section of uterus indicating moderate increase in height of luminal epithelium with moderate stimulation of uterine weight (Combination A extract: 200 mg kg<sup>-1</sup>) HE 300x



**Fig. 6:** Photomicrograph showing section of uterus indicating moderate increase in height of luminal epithelium with stimulated uterine glands (Combination B extract: 200 mg kg<sup>-1</sup>) HE 300x

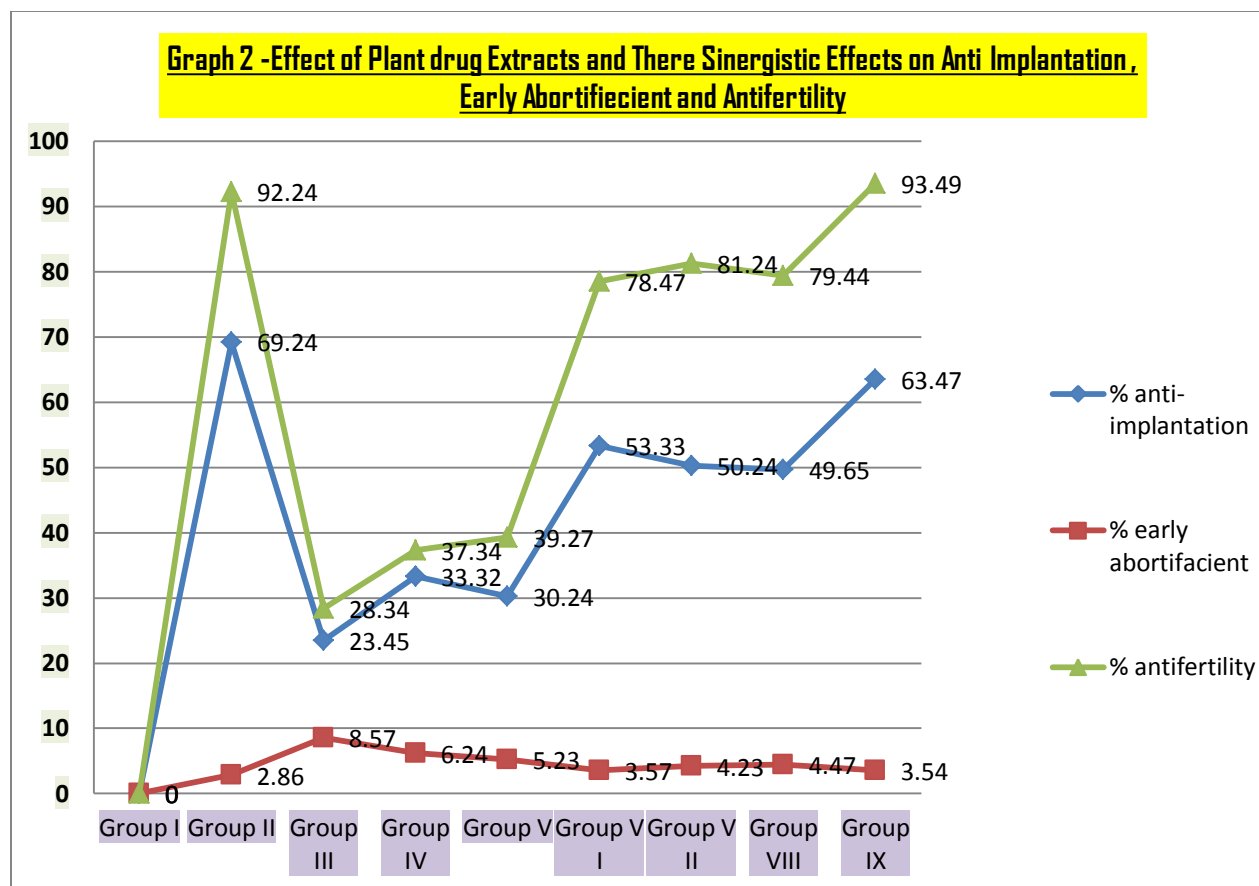
### 1.5.3 Anti-Implantation Activity

The anti-implantation activity is expressed as the percentage decrease in the number of implantations in the uteri on day 10 of pregnancy, and the number of resorbed implants from the existing number of implants will be recorded on day 18 for evaluating the early abortifacient activity. The ethanolic extracts and there combinations have offered significant and dependent anti-implantation and early abortifacient activity by decreasing the number of implantation sites and showed significant resorption of the existing implants compared to vehicle control. The Combination D ethanolic extract at 200 mg kg<sup>-1</sup> p.o. showed 84.27% antifertility activity and it was found to be more potent than other extracts and combinations ; at 200 mg kg<sup>-1</sup> p.o..The results are shown in Table 2.

**TABLE 2: EFFECT OF SELECTED PLANT DRUG EXTRACTS AND THERE SYNERGISTIC (COMBINATION) EXTRACTS ON ANTI-IMPLANTATION AND EARLY ABORTIFACIENT ACTIVITY IN RATS ( $\bar{x} \pm s$ ; n=6 ).**

Group	Extracts/drug	Dose (mg kg <sup>-1</sup> )	% anti-implantation activity	% early abortifacient activity	% antifertility activity
Group I	Control (saline solution)	-	0	0	0
Group II	Ethinyl Estradiol ( Control Standard)	0.02 mg kg <sup>-1</sup> , p.o	69.24±23.24**	2.86±0.54***	92.24±24.27***
Group III	Ethanolic extract of <i>Mucuna prurita</i>	200 mg kg <sup>-1</sup> , p.o	23.45±0.47*	8.57±1.24*	28.34±5.27*
Group IV	Ethanolic extract of <i>Mesua ferrea</i>	200 mg kg <sup>-1</sup> , p.o	33.32±1.89*	6.24±0.57*	37.34±2.41*
Group V	Ethanolic extract of <i>Punica granatum</i>	200 mg kg <sup>-1</sup> , p.o	30.24±1.48*	5.23±0.82*	39.27±3.23*
Group V I	Ethanolic extract of <b>Combination A</b> ( <i>Mucuna prurita</i> + <i>Mesua ferrea</i> )	200 mg kg <sup>-1</sup> , p.o	53.33±0.49**	3.57±1.24*	78.47±3.17**
Group V II	Ethanolic extract of <b>Combination B</b> ( <i>Mesua ferrea</i> + <i>Punica granatum</i> )	200 mg kg <sup>-1</sup> , p.o	50.24±1.28*	4.23±0.21*	81.24±3.15**
Group VIII	Ethanolic extract of <b>Combination C</b> ( <i>Mucuna prurita</i> + <i>Punica granatum</i> )	200 mg kg <sup>-1</sup> , p.o	49.65±0.38*	4.47±1.12*	79.44±3.62**
Group IX	Ethanolic extract of <b>Combination D</b> ( <i>Mucuna prurita</i> + <i>Mesua ferrea</i> + <i>Punica granatum</i> )	200 mg kg <sup>-1</sup> , p.o	63.47±2.49*	3.54±0.49*	93.49±3.74***

Values are mean ± SEM (n=06), \*P<0.05, \*\*P<0.01, and \*\*\*P<0.001 as compared to control group.



## CONCLUSION

Treatment with ethanolic extracts (200 mg kg<sup>-1</sup>, p.o.) and their combinations (200 mg kg<sup>-1</sup>, p.o.) had showed significant increase in uterine weight in a dose-dependent manner compared to vehicle control. The estrogenic effect of Combination D extract at 200 mg kg<sup>-1</sup> p.o. was comparable with reference standard Ethinyl estradiol (0.02 mg kg<sup>-1</sup>, p.o.). Furthermore, the ethanolic Combination D extract at 200 mg kg<sup>-1</sup> offered more potent estrogenic activity than the reference standard ethinyl estradiol. The extract significantly increased the weights of uteri (Table 1), and results obtained were also correlated and supported by the histopathological findings, where the combination of ethanolic extracts A, B and C (200 mg kg<sup>-1</sup>, p.o.) showed synergistic effect showed significant increase in the height of luminal epithelium and loose and edematous stroma with stimulated uterine glands, while the individual ethanolic extract (200 mg kg<sup>-1</sup>, p.o.) showed moderate increase in the height of luminal epithelium with stimulated uterine glands.

The anti-implantation activity is expressed as the percentage decrease in the number of implantations in the uteri on day 10 of pregnancy, and the number of resorbed implants from the existing number of implants will be recorded on day 18 for evaluating the early abortifacient activity. The ethanolic extracts and their combinations have offered significant and dependent anti-implantation and early abortifacient activity by decreasing the number of implantation sites and showed significant resorption of the existing implants compared to vehicle control. The Combination D ethanolic extract at 200 mg kg<sup>-1</sup> p.o. showed 84.27% antifertility activity and it was found to be more potent than other extracts and combinations; at 200 mg kg<sup>-1</sup> p.o.

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