



### ANTIFERTILITY ACTIVITY OF LEAVES OF *TABERNAEMONTANA DIVARICATA* (LINN) R.Br IN FEMALE RATS

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#### ABSTRACT

The petroleum ether, ethanolic and aqueous extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br. were found to possess significant estrogenic activity at dose of 400mg/kg as indicated by significant increase in uterine weight compared with the control in a dose dependent manner. However petroleum ether extract at dose of 200 mg/kg body weight and 400 mg/kg body weight. and ethanolic extract at dose of 400 mg/kg body weight when compared with standard (Ethinyl estradiol) found to possess greater effect than that of standard but not statistically significant. All the three extracts (petroleum ether, ethanolic and aqueous) have shown significant Anti-implantation and early abortifacient activity at dose of 400 mg/kg body weight. However the activity shown by aqueous extract was found to be less significant than other two extracts.

**Key Words:** *Tabernaemontana divaricata* (Linn) R.Br. leaves, Estrogenic activity, Antiimplantation and abortifacient, Ethinyl estradiol.

#### INTRODUCTION

The population problem has thrown challenge world over to change the outlook, attitude and behaviour of individuals, families and nations towards life. In history of man never before his very existence has been threaten so danger. And it is threatened by the very force of numbers which he had all along thought as an insurance against the deadly punish of wars, epidemics, natural disasters and pestilence. But since the advent of modern scientific achievements, death rate has been considerably checked and yet birth rate continues to be high. The pace of population growth is faster than that of increase in production of commodities.

Population explosion may lead to economic and health impact on family in particular and society in general especially in developing countries like India where the population growth is very high. The wealth of nations lies in its healthy population not in its prosperities. Thus the control of human fertility in the sense of its limitation, is

the most important and urgent of all biosocial and medical problems confronting mankind today.

The development of new fertility regulating drug from medicinal plants is an attractive preposition. Man since time immemorial has been using herbs or plant products as medicine for abortifacient and antifertility activities. Although in recent times, synthetic drugs are used extensively in modern medicine systems. However many modern medicines are developed through the clues obtained from phytochemicals. More over the phytochemicals are even today are important resources for medicinal uses. The plant products are becoming more popular than the synthetic drugs due to their low toxicity and long standing experience of exposure of these drugs in ethnic medicine system like Ayurveda. Family planning has been promoted through several methods of contraception, but due to serious adverse effects produced by synthetic steroidal contraceptives attention has now been focused on indigenous plants for possible contraceptive effect. Hence there is need for searching suitable product from indigenous plants that could be effectively used in the place of pills.

Already several scientific papers have been published related to fertility control from medicinal plants

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but still number of plants are yet to be screened for their efficacy. *Tabernaemontana divaricata* (Linn) R.Br. is one such plant used traditionally in folk medicine as thermogenic, anodyne, astringent, vermifuge, anti-inflammatory [1-2], anthelmintic, emmenagogue, aphrodisiac, tonic to the brain, liver, spleen [3] and also advocated as a traditional medicine for family planning<sup>3</sup>. *Tabernaemontana divaricata* (Linn) R.Br. reported to contain phytochemical constituents such as, Flavonoids, Steroids, Alkaloids, Tannins and others [4].

Keeping this in view after extensive literature survey available from all scientific sources revealed no information about the pharmacological validation of the antifertility activity of leaves of *Tabernaemontana divaricata* (Linn) R.Br. Thus the present study deals with the screening of antifertility efficacy by using different experimental models.

## MATERIALS AND METHODS

### Plant Material

Leaves of *Tabernaemontana divaricata* (Linn) R.Br. were collected in the month of March and April from Harapanahalli, Karnataka. The authentication was done by Prof. K. Prabhu, Department of Pharmacognosy, S.C.S. College of Pharmacy, Harapanahalli. A voucher specimen has been deposited at the museum of our college.

### Preparation of Extracts

The leaves of *Tabernaemontana divaricata* (Linn) R.Br. were collected and shade dried. The dried leaves were coarse powdered and the powder was packed in to soxhlet column and extracted successively with petroleum ether (60 – 80°C), ethanol (60°C) and distilled water. The extracts were concentrated by using rotary flash evaporator under reduced pressure. The dried extracts were stored in airtight container in refrigerator below 10°C. The percentage yield of the each extract shown in Table No. 1. The solution of ethanolic and aqueous extracts were prepared using distilled water and petroleum ether extract was suspended in 2% gum acacia were subjected to

#### I. Preliminary phytochemical screening.

Based on results of the preliminary phytochemical screening, petroleum ether, ethanolic and aqueous extracts have been selected for the following studies.

#### II. Determination of acute toxicity (LD<sub>50</sub>).

#### III. Antifertility activity.

1. Estrogenic activity
2. Antiimplantation and early abortifacient activity.

### Preliminary Phytochemical Screening

The preliminary phytochemical screening was carried out on petroleum ether, ethanol and aqueous extracts of *Tabernaemontana divaricata* (Linn) R.Br. leaves for the detection of various phytochemicals. Tests for common phytochemicals were carried out by standard

methods described in practical pharmacognosy by C.K Kokate [5] and K.R. Khandelwal [6].

### Animals used

Female and male albino rats (wistar strain) weighing 150-200gms, immature female rats of 21-23 days old (wistar strain) weighing 40-60gms and albino mice weighing 20-25gms of either sex were used in this study. They were procured from Nijalingappa Medical College, Bagalkot and Venkateshwara Animal Suppliers, Bangalore. 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 12 hours light / dark cycle. The animals were fed with rodent pellet diet (Gold Mohur Lipton India Ltd.) and water ad libitum. Animal ethical clearance for performing the experiments on animals was obtained from the Institutional Animal Ethical Committee (IAEC).

### Determination of acute toxicity (LD<sub>50</sub>)

The acute toxicity for ethanolic and aqueous extracts of *Tabernaemontana divaricata* (Linn) R.Br. leaves were determined in albino mice, maintained under standard conditions. The animals were fasted overnight prior to the experiment, fixed dose method was adopted as per OECD Guideline No. 420; (Annexure-2d) of CPCSEA [7].

### Evaluation of Antifertility Activity

#### 1) Estrogenic activity on immature female rats

Immature female rats of wistar strain 21-23 days old weighing 40-60 gms were used. They were divided in to eight groups of six animals each. The various groups were treated as follows ;

Group I - Control : (Vehicle) p.o.

Group II - Standard : Ethinyl estradiol 0.02mg/kg s.c.

Group III - Petroleum ether extract of leaves of *Tabernaemontana divaricata* (Linn) R.Br. 200mg/kg p.o.

Group IV - Petroleum ether extract of leaves of *Tabernaemontana divaricata* (Linn) R.Br. 400mg/kg p.o.

Group V - Ethanolic extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 200mg/kg p.o.

Group VI - Ethanolic extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 400mg/kg p.o.

Group VII - Aqueous extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 200mg/kg p.o.

Group VIII - Aqueous extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 400mg/kg p.o.

The treatment was given for three days, 24 hours after the last treatment, all the animals were sacrificed by decapitation and uterus 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 hours, dehydrated in alcohol and embedded in paraffin. The paraffin blocks were sectioned at 6 $\mu$  and stained with haemotoxylene-eosin solution (H & E Stain) for histological observations [8-9].

#### Antiimplantation and early abortifacient activity in rats

The method of Khanna and Chowdhary [10] was adopted with the modification for the anti-implantation and early abortifacient activities of petroleum ether, ethanolic and aqueous extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br.

Female albino rats (Wistar strain) weighing 150-200gms were used to assess anti-implantation and early abortifacient activity. All the animals were maintained under controlled standard animal house condition with access to food and water ad libitum. Vaginal smears from each rat was monitored daily. Only the rats with normal oestrous cycles were selected for the experiment.

Female rats of proestrus phase were kept with male rats of proven fertility for mating in a ratio of 2:1. The females were examined, the following morning for evidence of copulation. The animals exhibiting thick clumps of spermatozoa in vaginal smears were separated from male partner. That day when spermatozoa were detected in the vaginal smear was considered as day one of gestation.

The separated pregnant rats were divided into seven groups of six rats each.

Group I - Control : (Vehicle) p.o.

Group II- Petroleum ether extract of leaves of *Tabernaemontana divaricata* (Linn) R.Br. 200mg/kg p.o.

Group III - Petroleum ether extract of leaves of *Tabernaemontana divaricata* (Linn) R.Br. 400mg/kg p.o.

Group IV - Ethanolic extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 200mg/kg p.o.

Group V- Ethanolic extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 400mg/kg p.o.

Group VI - Aqueous extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 200mg/kg p.o.

Group VII - Aqueous extract of leaves of *Tabernaemontana divaricata* (Linn)

R.Br. 400mg/kg p.o.

The extracts were administered orally from first day to day seven of gestation. The control animals received only vehicle. On the tenth day laparotomy was carried out under light ether anesthesia and semisterile condition. The uteri were examined to determine the number of implantation sites. The number of corpora lutea in ovaries were also recorded. The abdomen was sutured

and animals left in cages. The drugs were administered orally again for 3 days (day 14- 16). On the eighteenth day laprotomy was carried out once again under light ether anesthesia for the abortifacient study.

The percentages of anti-implantation and early abortifacient activities were calculated by using following formulas.

$$\% \text{ of anti-implantation activity} = 100 - \frac{\text{No. of implantation}}{\text{No. of Corpora lutea}} \times 100$$

$$\% \text{ of abortifacient activity} = \frac{\text{No. of resorptions}}{\text{No. of Corpora lutea}} \times 100$$

## Results

### Preliminary phytochemical screening

Preliminary phytochemical screening was carried out for the presence of carbohydrate, proteins, amino acids, steroids, saponins, flavonoids, alkaloids, tannins and glycosides for petroleum ether, ethanolic and aqueous extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br. Results are shown in Table 2.

### Determination of acute toxicity (LD<sub>50</sub>)

The acute toxicity studies of petroleum ether, ethanolic and aqueous extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br. were found to be safe and no mortality was found at dose of 2000 mg/kg body weight. Hence 2000 mg/kg was LD<sub>50</sub> cutoff value for petroleum ether, ethanolic and aqueous extracts.

So that the doses selected for all extracts as per OECD Guideline No. 420 (Annexure 2d) fixed dose method are mentioned below :

Petroleum ether extract - 200 mg/kg (<sup>1</sup>/<sub>10</sub> of 2000 mg/kg)

Petroleum ether extract - 400 mg/kg (<sup>1</sup>/<sub>5</sub> of 2000 mg/kg)

Ethanolic extract - 200 mg/kg (<sup>1</sup>/<sub>10</sub> of 2000 mg/kg)

Ethanolic extract - 400 mg/kg (<sup>1</sup>/<sub>5</sub> of 2000 mg/kg)

Aqueous extract - 200 mg/kg (<sup>1</sup>/<sub>10</sub> of 2000 mg/kg)

Aqueous extract - 400 mg/kg (<sup>1</sup>/<sub>5</sub> of 2000 mg/kg)

### Evaluation of Antifertility Activity

#### Estrogenic Activity on Immature Female Rats

The effect of petroleum ether, ethanolic and aqueous extracts on immature female rat uterus is shown in Table No- 3 & Fig. 2. Oral administration of test extracts caused a significant increase in uterine weight in dose dependent manner when compared to those of control rats. The petroleum ether and ethanolic extracts at dose of 200 mg/kg b.w. and 400 mg/kg b.w. and aqueous extract a dose of 400 mg/kg b.w. were found to possess significant (p < 0.001) estrogenic activity as indicated by increase in the uterine weight of immature female rats when compared to control. However the petroleum ether at dose of 200 mg/kg b.w. and 400 mg/kg b.w. and ethanolic extract at dose of 400 mg/kg b.w. when compared with standard found to

posses greater effect than that of standard but not significantly greater ( $p > 0.05$ ). Where as the aqueous extract at dose of 400 mg/kg b.w. and ethanolic extract at dose of 200 mg/kg b.w. when compared with standard found to possess lesser effect than that of standard. The aqueous extract at dose of 200 mg/kg b.w. was less significantly ( $p < 0.01$ ) increased the uterine weight of immature female rats when compare to that of control.

The above results obtained with the test extracts on uterine weight of immature female rats also supported by histological architecture shown in Fig. 3-8. Statistical analysis was carried out using analysis of variance (ANOVA) test. The results were judged significant if ( $p < 0.05$ ).

### Anti-implantation and Early Abortifacient Activity

A dose dependent anti-implantation and early

abortifacient activity of the petroleum ether, ethanolic and aqueous extracts was evident by significant decrease in number of implantation sites and increase in number of resorptions when compared with the control group.

The percentage of antifertility activity of the petroleum ether extract at dose of 400 mg/kg b.w. and 200 mg/kg b.w. were found to be (93.18%) and (74%) respectively in a dose dependent manner when compare to control. Where as percentage of antifertility activity of ethanolic extract at dose of 400 mg/kg b.w. and 200 mg/kg b.w. were found to be (83.32%) and (67.73%) respectively in a dose dependent manner when compare to control. However percentage antifertility activity of aqueous extract at dose of 400 mg/kg b.w. and 200 mg b.w. were found to be (38.09%) and (29.89%) respectively in a dose dependent manner when compared to control. The results are shown in the Table 4.

**Table 1. Percentage yield of crude extracts of *Tabernaemontana divaricata* (Linn) R.Br. leaves**

S. No.	Solvent	Colour and Consistency	Percentage yield
1	Petroleum Ether (60-80°C)	Blackish and sticky	6.74%
2	Ethanol	Blackish green and pasty	22.09%
3	Aqueous	Brownish and pasty	17.44%

**Table 2. Preliminary phytochemical screening of *Tabernaemontana divaricata* (Linn) R.Br. leaves**

Phytochemical constituents	Ethanolic Extract	Aqueous Extract	Petroleum ether extract
Carbohydrates	+	+	--
Proteins	--	--	--
Amino acids	+	+	--
Steroids	+	+	+
Glycosides	+	+	--
Flavonoids	+	+	--
Alkaloids	+	+	--
Tannins	+	+	--

-- absent + present

**Table 3. Effect of *Tabernaemontana divaricata* (Linn) R.Br. leaves on uterine weight of immature female rats**

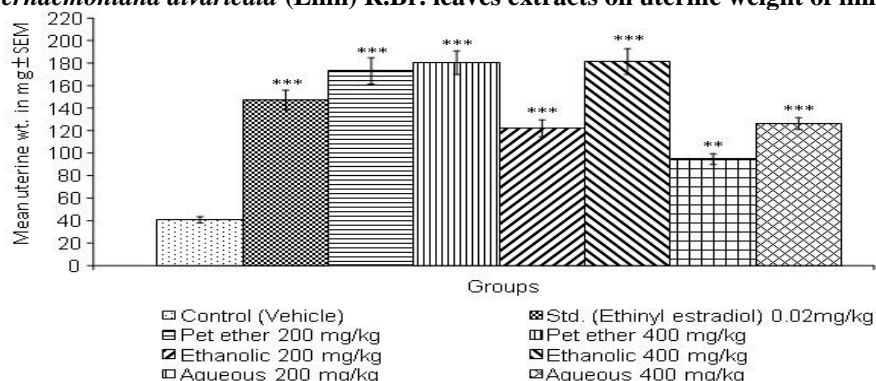
Group No.	Extracts / Drug	Dose mg/kg	Uterine Weight in mg Mean $\pm$ SEM
I	Control (vehicle)	--	40.618 $\pm$ 2.793
II	Ethinyl estradiol	0.02	147.13 $\pm$ 8.644***
III	Petroleum ether	200	173.08 $\pm$ 11.730***
IV	Petroleum ether	400	180.42 $\pm$ 10.457***
V	Ethanolic Extract	200	121.94 $\pm$ 7.533***
VI	Ethanolic Extract	400	181.39 $\pm$ 11.535***
VII	Aqueous Extract	200	94.667 $\pm$ 4.807**
VIII	Aqueous Extract	400	126.22 $\pm$ 5.288***

\*\*\*P < 0.001 (n=6) when compared with control.

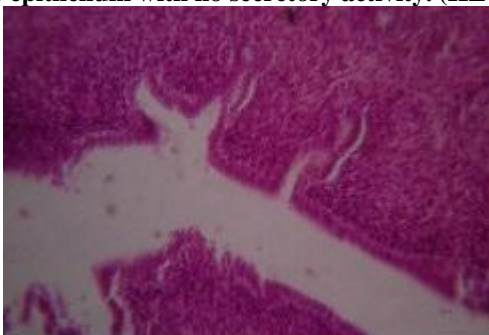
\*\*p < 0.01 (n=6) when compared with control.

**Table 4. The percentage of antifertility activity**

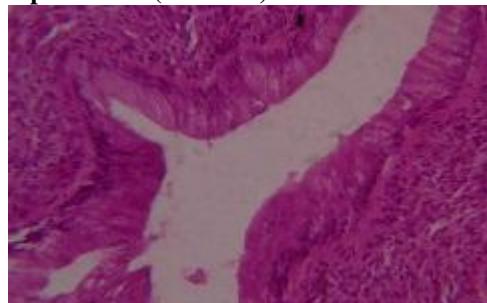
Group and Treatment	Animals Used	Anti-implantation		Early Abortifacient	% of Antiimplantation activity	% of Early Abortifacient activity	% of Antifertility activity
		No. of Implan-tation	No. of Corpora-lutea	No. of Resorbed Implantation			
Control (vehicle)	6	11	13	--	10.38	0.00	10.38
		11	13	--			
		12	13	--			
		11	12	--			
		12	13	--			
Petroleum ether extract (200mg/kg)	6	4	9	1	68	6	74
		3	8	--			
		0	7	--			
		0	9	--			
		4	8	1			
Petroleum ether extract (400mg/kg)	6	0	7	--	84.09	9.09	93.18
		0	7	--			
		3	8	2			
		0	8	--			
		4	7	2			
Ethanollic extract (200 mg/kg)	6	0	10	--	62.90	4.83	67.73
		6	10	1			
		6	11	1			
		6	11	--			
		0	10	--			
Ethanollic extract (400 mg/kg)	6	5	10	2	74.07	9.25	83.32
		0	10	--			
		0	8	--			
		5	9	2			
		4	9	1			
Aqueous extract (200 mg/kg)	6	8	11	--	26.86	2.98	29.84
		8	11	--			
		8	12	--			
		9	12	1			
		8	10	--			
VII. Aqueous extract (400 mg/kg)	6	8	11	1	34.92	3.17	38.09
		7	10	--			
		7	11	1			
		7	10	--			
		6	11	--			
		8	11	1			
		6	10	--			
		8	11	1			
		6	10	--			
		6	10	--			

**Fig 1. Effect of *Tabernaemontana divaricata* (Linn) R.Br. leaves extracts on uterine weight of immature female rats**

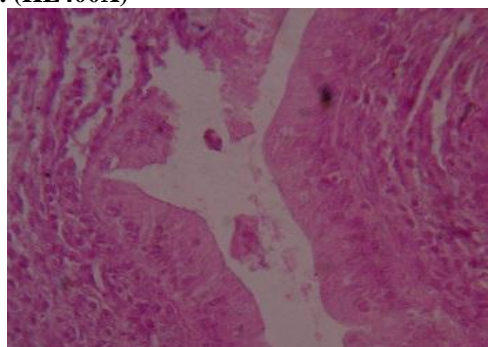
**Fig 2. Photomicrograph of control rat uterus showing surface epithelium with no secretory activity. (HE400X)**



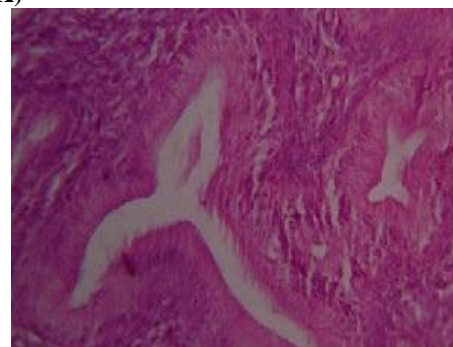
**Fig 3. Photomicrograph of rat uterus treated with Standard drug showing remarkable stimulation in the luminal epithelium. (HE400X)**



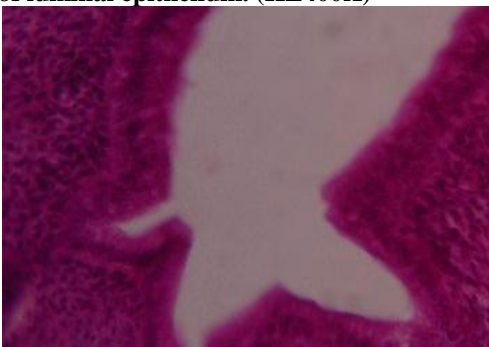
**Fig 4. Photomicrograph of rats uterus treated with petroleum ether extract 200mg/kg dose showing breakdown in the endometrial epithelium and loose stroma. (HE400X)**



**Fig 5. Photomicrograph of rat uterus treated with petroleum ether 400mg/kg dose showing disturbances in stroma and uterine gland with obliterated luminal. (HE400X)**



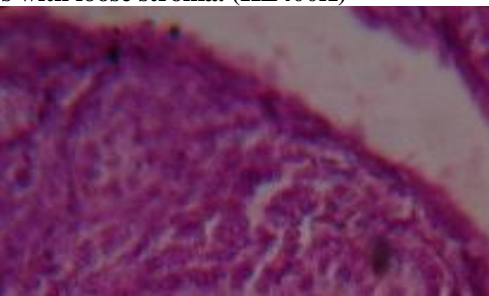
**Fig 6. Photomicrograph of rat uterus treated with ethnolic extract 200mg/kg dose showing increase in height of luminal epithelium. (HE400X)**



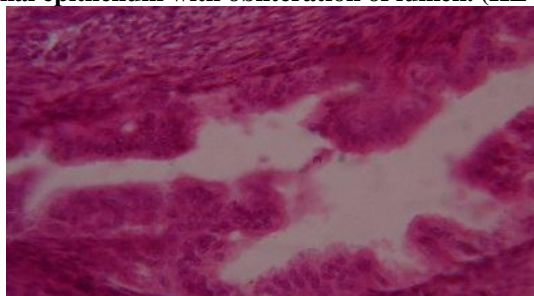
**Fig 7. Photomicrograph of rat uterus treated with ethnolic extract 400mg/kg dose showing increase in height of luminal epithelium with loose stroma and stimulated uterine gland. (HE400X)**



**Fig 8. Photomicrograph of rat uterus treated with aqueous extract 200mg/kg dose showing stimulated features with loose stroma. (HE400X)**



**Fig 8. Photomicrograph of rat uterus treated with aqueous extract 400mg/kg dose showing breakdown in luminal epithelium with obliteration of lumen. (HE400X)**



## DISCUSSION

A large number of indigenous plants having contraceptive activities are recorded in ancient Indian literature. Research on antifertility activity of native plants has been exhaustively reviewed by Chowdhary *et al* [11-12] and Kamboj and Dhawan [13-14]. Whole plant or different parts of various plants have been tested for antifertility activity. Whole plant of *Biophytum sensitivum* [15], roots of *Achyranthes aspera* [16], Seeds of *Trigonella foenum graecum* [17], Fruits of *Balanites roxburghii* [18], Flowers of *Woodfordia fruticosa* [19] and leaves of *Inula viscosa* [20] have been reported for their antioviulatory and anti-implantation/antifertility activity.

*Tabernaemontana divaricata* (Linn) R.Br. was a traditional plant used for family planning [21]. It was found rich in phytochemical constituents which may have variety of pharmacological actions. In the literature survey the phytochemicals investigation made on this plant have reported the presence of phytochemicals like flavonoids, alkaloids, phytosterols, tannins and amino acids etc [22-23].

The present preliminary phytochemical investigation on *Tabernaemontana divaricata* (Linn) R.Br. leaves reveals the presence of carbohydrates, aminoacids, steroids, glycosides, flavonoids, alkaloids and tannins in ethanolic and aqueous extracts, but only steroids was found to be present in petroleum ether extract. Hence petroleum ether, ethanolic and aqueous extracts of *Tabernaemontana divaricata* (Linn) R.Br. leaves were taken for evaluation of the antifertility activity.

The data obtained in the present study indicates that petroleum ether and ethanolic extracts of *Tabernaemontana divaricata* (Linn) R.Br. leaves exhibited more significant and aqueous extract exhibited less significant antifertility activity against estrogenic, anti-implantation and abortifacient activity in dose dependent manner.

The petroleum ether and ethanolic extracts at dose of 200 mg/kg body weight and 400 mg/kg body weight and aqueous extract at dose of 400 mg/kg body weight found to possess significant estrogenic activity as indicated by increase in uterine weight of immature female rats when compare to control. However petroleum ether extract at dose of 200 mg/kg body weight and 400 mg/kg body weight and ethanolic extract at dose of 400 mg/kg body weight when compared with standard found to possess greater effect than that of standard but not significantly greater. Where as the aqueous extract at dose of 400 mg/kg body weight and ethanolic extract at dose of 200 mg/kg body weight when compared with standard found to possess lesser effect than that of standard.

In the present study petroleum ether, ethanolic extracts at dose of 200 mg/kg body weight and 400 mg/kg body weight tested for anti-implantation and abortifacient activity exhibited a significant decrease in number of implantation sites and increase in number of resorptions in

a dose dependent manner. Where as the aqueous extract exhibited less significant anti-implantation and abortifacient activity when compare to other extracts in a dose dependent manner. The number of rats that had shown no implantation and the reduction in the number of implants suggests the possible antifertility activity of the extracts.

Estrogenic activity is shared by many steroidal and non-steroidal compounds. The three principal native forms of known endogenous estrogens are 17  $\beta$ - estradiol, estrone and estriol. The most potent biologic form is 17  $\beta$ - estradiol which is used as a component of oral contraceptives for inhibiting gonadotropin secretion. The non-steroidal compounds with estrogenic activity including flavonoids (flavones, flavonones hydroxychalcones and isoflavonoids) coumestrans with isoflavones, lignans, alkaloids, phenolics occur in variety of plants are well documented as producing infertility [24-25].

It is well known fact that estrogenic substances inhibit pregnancy by suppressing the level of both follicular stimulating hormone (FSH) and luteinizing hormone (LH) which in turn prevent the implantation. Estrogen and progesterone are the hormones responsible for histology and functional modifications of female genital tract. The exogenous administration of physiological doses of estrogen, in sexual immature rats stimulated histoarchitecture of uterus [26]. According to Laurence [27] compounds possessing estrogenic activity may exhibit antifertility activity, they act by suppressing gonadotropin secretion, with consequent inhibition of ovulation. In the present study the petroleum ether, ethanolic and aqueous extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br. exhibited definite estrogenic activity in immature female rats in dose dependent manner. Hence the anti-implantation activity of these extracts may be due to an imbalance in endogenous estrogen and progesterone levels. The loss of implantation caused by extracts may be due to their antizygotic, blastocytotoxic or anti-implantation activity as described by Hafez [28]. In this study, the histological evidences of uterus treated with petroleum ether, ethanolic and aqueous extracts clearly supports an unfavorable uterine milieu, showing obliterated lumen with loose stroma, increased height of luminal epithelium and stimulated uterine glands in respective extracts.

The literature survey on the plant *Tabernaemontana divaricata* (Linn) R.Br. reveals the presence of flavonoids, steroids, alkaloids, tannins, and other constituents in the leaves. The phytochemical constituent coronaridine reported to be predominant alkaloid in this plant and at single 30 mg/kg p.o. dose prevented pregnancy in rats when given on day 1, 2, 3 or 4 after coitus. When given on day 5, 6, 7 or 8 of pregnancy, the results were only partially successful. The substance showed estrogenic activity, which appeared to be responsible for the antifertility action. The flowers of



*Tabernaemontana divaricata* (Linn) R.Br have reported to posses antifertility [29] activity and leaves shown to posses antifertility effect on chronic administration in male rats [30].

In the present investigation also, flavonoids and alkaloids present in ethanolic and aqueous extracts and steroids present in the petroleum ether extract (preliminary phytochemical screening) may be responsible for significant antifertility activity against the selected experimental models.

Thus the data obtained from phytochemical and pharmacological evaluations of leaves extracts of *Tabernaemontana divaricata* (Linn) R.Br. tend to suggest that petroleum ether and ethanolic extract possess significant and aqueous extract possess less significant estrogenic, anti-implantation and early abortifacient activity in a dose dependent manner.

It is interesting to note that administration of these extracts not caused permanent sterility in rats, since discontinuation of the treatment allowed a prompt return to normal fertility.

## CONCLUSION

It is thus concluded that petroleum ether and ethanolic extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br. found to possess significant and aqueous extract found possess less significant antifertility activity in estrogenic, anti-implantation and early abortifacient study in a dose dependent manner.

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The estrogenic activity of petroleum ether and ethanolic extracts at dose of 200 mg/kg b.w. and 400 mg/kg b.w. and aqueous extract at dose of 400 mg/kg b.w. were found to be significant as indicated by increase in uterine weight of immature female rats when compared to control. However petroleum ether extract at dose of 200 mg/kg b.w. and 400 mg/kg b.w. and ethanolic extract at dose of 400 mg/kg b.w. when compared with standard drug ethinyl estradiol (0.02 mg/kg) found to possess greater effect than that of standard but not significantly greater ( $p > 0.05$ ). Where as the aqueous extract at dose of 400 mg/kg b.w. and ethanolic extract at dose of 200 mg/kg b.w. when compared with standard found to posses lesser effect than that of standard.

The anti-implantation and early abortifacient activity of petroleum ether and ethanolic extracts of leaves of *Tabernaemontana divaricata* (Linn) R.Br. at dose of 400 mg/kg b.w. were found to be more significant when compared to the other tested doses of the extracts. Where as the aqueous extract showed less significant anti-implantation and early abortifacient activity when compared to other two extracts in a dose dependent manner.

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Nil

## CONFLICT OF INTREST

No interest.



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