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Antisteroidogenic activity of floral extract of *Thespesia populnea* Corr. in mouse ovary

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Anti-steroidogenic activity of various extracts of *T.populnea* was screened in female albino mice. The weight of the uterus and ovaries were reduced significantly and the cholesterol and ascorbic acid content in ovaries were significantly elevated due to the treatment with extract of *T. populnea*. The significant inhibition of Δ5, 3β hydroxy steroid dehydrogenase and glucose-6-phosphate dehydrogenase, the two key enzymes involved in ovarian steroidogenesis were also observed in mouse ovaries after 15 days of treatment.

A large number of herbal drugs are used to control fertilisation with considerable success. Although the use of these drugs has a sound tradition, their place has yet to be validated in therapeutics using the current methodology. Scientific studies are therefore required to judge their actual efficacy, mode of action and other limitations to widen the scope of these drugs, if they are proved to be really effective[1]. The present study is designed to evaluate anti-steroidogenic activity of floral extracts of *Thespesia populnea* Corr.

Thespesia populnea[2] (Family-Malvaceae) can grow everywhere including saline soils, except in hilly areas, but prefers light and porous soils. The bark, leaves, flowers and fruits are reported to be useful in cutaneous affections such as scabies, psoriasis, ringworm, guineaworm and eczema[3]. A sample of petals yielded the following coloring principles, populnin, populnetin, herbacetin, populneol, guarcetin and gossypetin[4].

Flowers of *T. populnea* were collected in and around Tiruchirappalli and identified by the Department of Botany, St. Joseph's College, Tiruchirappalli. The shade dried flowers of *T.populnea* were powdered (500g) and were extracted using petroleum ether, ethyl acetate, chloroform and ethyl alcohol. The extracts were dried and the dried extracts were dissolved in arachis oil.

Animals—Female albino Swiss mice weighing 26-30g were procured from King Institute, Guindy, Chennai. The temperature of the animal room was maintained between 30° and 32°C. Twelve hours of lighting and twelve hours of darkness were provided in animal room for optimum growth. The mice showing three consecutive normal cycles were selected and divided into 6 groups, each consisting of 6 mice.

Design of experiment—Group 3,4,5 and 6 were treated (ip) with 100 mg/kg body wt of petroleum ether, ethyl acetate, chloroform and ethylalcohol extracts of *T.populnea* respectively. Groups 1 and 2 were treated with saline (0.9% NaCl w/v; 5ml/kg body wt) and arachis oil (0.2 ml/20g body wt) respectively (ip). All the treatments were carried out on every alternate days for 15 days and estrous cycle was observed everyday by microscopic examination of vaginal smear.

After 24 hr of the last treatment the mice were sacrificed, the ovary and uterus dissected out, weighed and kept on ice for further processing. The ascorbic acid[5], cholesterol content[6] glucose-6-phosphate dehydrogenase (G-6PD)[7] and Δ5, 3β-hydroxy steroid dehydrogenase (Δ5,3β HSD) activity[8] and the content of protein[9] in the ovaries were estimated. The results are presented in Tables 1-3.

Weight of the ovaries and uteri of drug treated mice was reduced significantly ($P<0.01$). The crude extracts significantly increased the level of cholesterol and ascorbic acid contents in mouse ovaries as compared to the saline control as well as vehicle control whereas, the activity of two ovarian enzymes Δ5, 3β-HSD and G-6-PD, essential for biosynthesis of estrogen and progesterone were reduced. They also produced arrest of estrous cycle at metestrous phase.

During proestrous and estrous phase of the estrous cycle, the secretion of progesterone and estrogen is maximum[10]. Cholesterol[11] and ascorbic acid[12] plays major role in steroidogenesis with the help of

Table 3—Effect of *T.populnea* on activity of two enzymes Δ5 - 3β hydroxy steroid dehydrogenase and glucose -6- phosphate dehydrogenase in mouse ovary
[Values are mean ± SE of 6 mice in each group]

Treatment	Glucose 6 phosphate dehydrogenase activity		Δ5 - 3β hydroxy steroid dehydrogenase activity	
	Unit activity/ .100mg tissue	Activity/mg tissue	Unit activity/ 100mg tissue	Activity/mg tissue
Saline control (0.2 ml / 20 g)	2.19 ± 0.6	0.44 ± 0.08	1.17 ± 0.7	0.37 ± 0.02
Vehicle control (0. 2 ml / 20 g)	2.08 ± 0.4	0.54 ± 0.06	1.50 ± 0.8	0.39 ± 0.03
Alcoholic extract (100 mg / kg)	0.57* ± 0.04	0.21 ± 0.07	1.10 ± 0.02	0.18 ± 0.02
Chloroform extract (100 mg / kg)	0.56* ± 0.08	0.30 ± 0.08	1.09 ± 0.2	0.20 ± 0.04
Ethyl acetate extract (100 mg / kg)	0.60* ± 0.07	0.27 ± 0.02	0.92 ± 0.04	0.36 ± 0.02
Petroleum ether extract (100 mg / kg)	0.60* ± 0.07	0.26 ± 0.04	0.96 ± 0.02	0.27 ± 0.04

* $P < 0.001$

Table 1—Effect of *T. populnea* on cholesterol and ascorbic acid content in mouse ovary
[Values, expressed as mg/100mg of ovary, are mean ± SE of 6 mice in each group]

Treatment	Cholesterol	Ascorbic acid
Saline control (0.2 ml / 20 g)	5.7 ± 0.8	0.83 ± 0.04
Vehicle control (0. 2 ml / 20 g)	6.0 ± 0.7	0.90 ± 0.06
Alcoholic extract (100 mg / kg)	8.7 ± 0.6*	4.2 ± 0.04*
Chloroform extract (100 mg / kg)	8.52 ± 0.6*	4.4 ± 0.04*
Ethyl acetate extract (100 mg / kg)	7.7 ± 0.7*	3.9 ± 0.06*
Petroleum ether extract (100 mg /kg)	7.9 ± 0.4*	4.8 ± 0.07*

* $P < 0.001$

Table 2— Effect of *T. populnea* on total body weight, weight of ovaries and uteri of mice
[Values are mean ± SE of 6 mice in each group]

Treatment	Difference in body wt (g) in 15 days	Weight of both ovaries (mg)	Weight of uteri (mg)
Saline control (0.2 ml / 20 g)	+ 4.0	1.5	2.90
Vehicle control (0. 2 ml / 20 g)	+ 4.0	1.45	2.80
Alcoholic extract (100 mg / kg)	+2.0	1.40	2.70
Chloroform extract (100 mg / kg)	+2.0	1.36	2.65*
Ethyl acetate extract (100 mg / kg)	+2.0	1.30*	2.5*
Petroleum ether extract (100 mg /kg)	+2.0	1.25*	2.65*

* $P < 0.001$

enzymes Δ5,3β HSD and G-6-PD.[13] The concentration of ascorbic acid increases in non-functional or hypo functional ovaries.[14]

In this present investigation, estrogen and progesterone synthesis is inhibited which is indicated as a reduction in the ovary and uterine weight and arrest of estrous cycle at metestrous phase.

The decreased activity of the enzymes Δ5,3β-HSD and G-6-PD indicated a reduction in steroidogenesis. It is further confirmed by the accumulation of cholesterol and ascorbic acid content. Therefore it can be concluded that the flowers extreact of *T.populnea* exhibit anti-steroidogenic activity in female mice.

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