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Effects of Certain Herbal and Synthetic Drugs on the Gonado-Somatic Index and Utero-Somatic Index of Adult Female Albino Rats (*Rattus Albicans*)

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Abstract:

Many plant products as well as some synthetic drugs, often used for the treatment of different reproductive ailments may cause certain disturbance to the reproductive as well as to the other organs of an individual. In the present investigation effects of bark extract of the plant *Saraca indica* and Levonorgestrel, a synthetic progestogen, on gonado-somatic index (GSI) and utero-somatic index (USI) were evaluated, in adult female albino rats. While *Saraca indica* is used as tonic to reproductive system, Levonorgestrel is used as a synthetic contraceptive in birth control. *Saraca indica* at a dose of 500mg/kg body weight/day was orally administered for 12 consecutive days and Levonorgestrel was administered subcutaneously for 3 consecutive days at a dose of 1.5mg/kg body weight/day. Increased GSI and USI were observed in the plant extract treated female albino rats when compared to that of control group. Increase in GSI and USI after the administration of the plant product may be due to its estrogenic effect and can be taken as a positive indicator of increased fertility rate. Statistically significant ($p<0.05$) decrease in GSI and USI were observed in the synthetic Levonorgestrel (LNG) treated female rats which may be indicative of negative effect of the product on the reproductive physiology of the organism.

Key words: Anti-estrogenecity, Estrogenecity, Fertility, GSI, USI..

Introduction:

Certain plants have gained popularity in the world because people are interested on the medication that contain components of natural origin (Holanda *et al.,* 2009). *Saraca indica* is one such plant, the different parts of which viz. bark, leaf, flower etc. are used traditionally to cure different human diseases. *Saraca indica*, locally known as Asoka belongs to the family Caesalpiniaceae (Mishra, 2013) found mostly in Assam, Garo hills, Lushai hills and Himalayan foot hills and Western Ghats of India.

The bark of *Saraca indica* is smooth and dark grayish-brown in colour (Yadav *et al.,* 2013). The bark portion has a stimulatory effect on the endometrial and ovarian tissues (Shilpakala, *et al*., 2009). *Saraca indica* is especially useful in curing menorrhagia due to uterine fibroids (Dharmananda, 2004). The Indian Materia Medica (1500A.D) said that this plant can be used as a uterine tonic because of its effectiveness in regularizing menstrual disorders. The bark of this plant is also use to treat leucorrhoea, respiratory complications, and kidney stone. Seeds of *Saraca indica* are used in treating bone fractures whereas the dried flowers are used to treat diabetes (Shyam, 2014). Ashokarishta, a common herbal tonic for female reproductive system contains 26.62g *Saraca indica* bark powder per100ml of the drug.

Blockage in the development and maturation of ovarian follicles, prevention of passage of ova through the oviduct, interference in fertilisation of ovulated oocytes, inhibition of implantation, gestation and lactation are the main targets during fertility control in females (Balser, 1964; Farnsworth, *et al.,* 1975; Chambers *et al.,* 1999).Synthetic steroids are one such group of chemicals which are used for fertility control in females. However, it has been reported that all progestogens or synthetic progestins, when administered alone at contraceptive dose, cause some

disruption of menstrual bleeding pattern (Benagiano, 2000). Such hormonal contraceptives suppress ovulation and at the same time affect the endometrium though the net effect of these steroids into the endometrium cannot be predicted (Rawal, 2016). Progestogens were found to have a range of actions which are dose dependent (Larsson-Cohn *et al.,* 1970). Levonorgestrel (LNG), a synthetic progestin, is one of the active components used in different types of birth control measures like OCPs, IUDs etc. It works either by preventing ovulation or by thickening the cervical mucus. However, a number of menstrual disturbances are also reported in users of synthetic progestin which include lengthened cycles, spotting and breakthrough bleeding (Hatcher *et al.,* 1988; Speroff *et al.,* 1992). Continuous menstrual disturbance in the reproductive cycle due to the use of contraceptives may result in infertility. Therefore the present investigation aimed to search the effectiveness of both the plant product and synthetic drug on the gonado- somatic index (GSI) and utero-somatic index (USI) of adult female albino rats as GSI and USI help in evaluating the reproductive potency of concerning individual.

Materials & Methods

Experimental animal:

Female albino rats weighing 130 g to 150 g were used for the present work. Rats were supplied sufficient food and water and housed in standard poly cages, bedded with paddy husk under natural light/dark cycle at constant room temperature. Experiments were done in accordance with the institutional ethical guidelines. Twenty numbers of adult female albino rats were divided into four groups, five in each. Group- I (normal control) received water and normal food, while group-II was treated with *Saraca indic*a bark extract at a dose of 500mg/kg body weight/day for 12 days, group-III received Levonorgestrel at a dose of 1.5mg/kg body weight/day for 03 days and group-IV received co treatment with both Levonorgestrel (for 3 days) and *Saraca indic*a bark extract (for 12 days). The ovaries and the uterus of all animals were collected immediately after the end of treatment and weighed in an electronic balance (model no. DJ602A) and then fixed in 10% formalin histology.

Preparation and administration of the plant extract:

The bark of *Saraca indica* was collected and authenticated by the Department of Botany, Gauhati University, Assam. The barks were cleaned, air dried and then grounded to fine powder. The bark powder was dissolved in pure methanol at room temperature for 72-96 hours. The solution was then filtered using glass funnel and Whatman No.1 filter paper and concentrated by evaporation at room temperature until a semi solid mass appeared. The concentrated extract was weighted and reconstituted in distilled water to get the desired concentration for all experiments. The semi solid extract can be stored at 4ºC until use. The bark extract was administered orally for 12 consecutive days at an interval of 24 hours at the dose of 500mg/kg body weight/day.

Administration of Levonorgestrel (LNG):

Levonorgestrel or l-norgestrel or D-norgestrel is a synthetic progestogen. It was taken for the present study as it is used as an active ingredient in some of the birth control measures. A dose of 1.5mg/kg body weight/day was prepared according to the body weight of the animal. The required dose was prepared using the formula $C_1V_1=C_2V_2$. The required stock solution was mixed with sesame oil to make 1ml volume and administered by sub cutaneous injection for 3 consecutive days at 24 hours time interval.

Calculation of GSI and USI:

Mean body weight of each rat group and weight of gonads and uteri were recorded. The GSI and USI were calculated considering the body weight, weight of the ovary and uterus by the following formulae:

$$GSI = \frac{\text{Wet weight of the ovary}}{\text{Body weight of the rat}} \times 100$$

$$USI = \frac{\text{Wet weight of the uterus}}{\text{Body weight of the rat}} \times 100$$

Data analysis:

The mean values and standard error of means were calculated for both the body weight and genital organ weight. Student's t test was applied to establish any significant difference at 95% confidence interval. P-values less than 0.05 were considered significant.

Results

Effect of *Saraca indica* bark extract and LNG on the body weight and genital organ weight of female albino rats were presented as mean ± standard error of mean (S.E.M) in Table 1. GSI and USI of all the rat groups were also presented in Table 1. After 12 days of bark extract administration, an increase in the body weight of rats from 142g to 150g was observed in group-II. When compared to the body weight of group-I (control), a gradual decrease in the body weight was observed (from 140g to 128g) in the rats of group-III which were administered LNG subcutaneously for 03 consecutive days. But almost similar results to that of group-I was seen in the group-IV (from 140g to 142g) where the rats were treated with both LNG and *S.indica.*

Table 1: Table showing Mean ± S.E.M (* p< 0.05) of Body weight and Genital organ weight with GSI and USI in different experimental groups:

Experimental groups	Average weight of body and Genital organs (mean ± S.E.M)				GSI	USI
	Initial body weight (in g)	Final body weight (in g)	Ovaries (in g)	Uterus (in g)		
Group-I (Control)	142 ± 3.742	148 ± 3.742	0.056 ± 0.004	0.480 ± 0.027	0.038	0.324
Group-II (*S.indica*)	142 ± 3.742	150 ± 3.536	0.060 ± 0.004	0.518 ± 0.017	0.040	0.345
Group-III (LNG)	140 ± 3.742	128 ± 3.674*	0.042 ± 0.002*	0.350 ± 0.030*	0.032	0.273
Group-IV (*S.indica* + LNG)	140 ± 3.742	142 ± 3.391	0.055 ± 0.002	0.460 ± 0.011	0.038	0.323

Increase in gonado-somatic index of the rats treated with the bark extract (0.042) when compared with group-I rats (0.039g) was observed. A decrease ($P < 0.05$) in the GSI of the rats treated with LNG (group-III) i.e., 0.032g was observed when compared with the control (group-I). These observations were similar for both the ovaries combined. But group-IV, treated with both bark extract and synthetic LNG exhibited almost similar results (i.e., 0.038g) to that of group-I (0.039g). Similar results were obtained for the uterus of treated and control groups.

Differences were also observed in the utero-somatic index among different treatment groups. Increase in uterine index of the group-II (0.345g) when compared with group-I (0.338g) was found. A decrease ($P < 0.05$) in the uterine index of the rats treated with LNG (group-III) i.e., 0.273g was observed when compared with the control (group-I). But group-IV, treated with both bark extract and synthetic LNG exhibited almost similar results (i.e., 0.334g) to that of group-I. The increase and decrease in GSI and USI were presented in the form of histogram in Fig1.

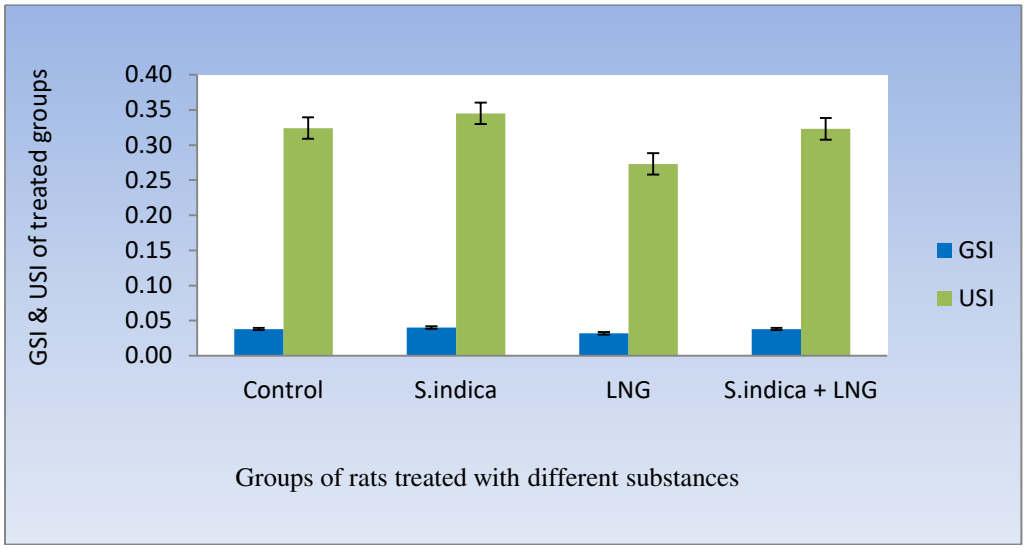

Fig. 1: Effect of *Saraca indica* bark extract and Levonorgestrel on the GSI and USI of female albino rats

Discussion

Main objective of the present investigation is to study the effect of a plant product and a synthetic drug on the gonado-somatic index (GSI) and utero-somatic index (USI) of adult female albino rats. GSI helps in evaluating the reproductive potency of concerning individual. While increase in GSI indicates hyperactivity of the gonads, decreased GSI indicates poor development, disfunctioning or inactiveness of gonads (Gupta, 2000). Decreased GSI is also indicative of reduction in the rate of folliculogenesis or an increased rate of follicular atresia (Oluwatoyin, *et al.,* 2012).

In the present investigation, a little increase in GSI was observed in the *Saraca indica* bark extract treated rats (group-II) which can be correlated to the work of Gaind (1971) who reported that the increased GSI in albino rats was due to the estrogenic effect of some intermediate Azasteroid. On the other hand, decreased GSI of group-III, administered with LNG can be correlated to the results of Arora *et al.,*(1990) after *Embelia ribes* extract treatment. Similar results were obtained by Choudhury *et.al.,*(1991) after treatment with *Piper betle* leaf stalk**,** pandey *et al.,*(1992) after *Hibiscus rosa sinensis* flower extract treatment, and Srinivasulu (1992) after CDRI 85/287 compound treatment where they clearly reported that reduction in the GSI was due to the anti- estrogenic activity of the treated plant extract or chemical. The decreased GSI of the ovaries observed in the present work may be due to the initiation of follicular atresia in the rat ovaries by Gonadotropin releasing hormone (GnRH) (Parborell *et al.,* 2005). Because sexual maturation and reproductive functions in mammals are generally regulated by the GnRH which is released from the hypothalamus in a pulsatile manner, GnRH triggers the synthesis and secretion of gonadotropins, (Singh and Krishna, 2010). The hypothalamo-pituitary-gonadal axis controls the female reproductive processes in mammal and the estrogen and progesterone from the ovaries are known to play important role in the maintenance of the uterine function (Guyton, 2006).

Increased USI of rats of group II, treated with the bark extract may be due to the estrogenic activity of the *Saraca indica* bark extract and this can also be correlated to the work of Gaind (1971), who stated that the increased uterine weight in albino rats was due to the estrogenic effect of some intermediate Azasteroid which was indicated by the gradual increase in ovarian weight. But the present results are contradictory to the results of Arora *et al.,*(1990) in *Embelia ribes* extract treatment, Choudhury et.al.,(1991) in piper betle leaf stalk treatment**,** pandey *et al.,*(1992) in *Hibiscus rosa sinensis* flower extract treatment and Srinivasulu, (1992) in CDRI 85/287 compound treatment where they clearly stated that reduction in the uterine weight is due to the anti- estrogenic activity of the treated plant

extract or chemical. However, this can be correlated to the result of the present work where a decrease in utero-somatic index was found in rats treated with LNG (group-III).

Conclusion

It can be concluded that plant product with estrogenic activity are helpful in increasing the fertility rate as it increases the rate of folliculogenesis whereas synthetic drugs with anti-estrogenic activity always tend to reduce the rate of folliculogenesis and hence the rate of fertility.

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