

REVERSIBLE OESTROUS CYCLE DISTURBING ACTIVITY OF THE EXTRACT OF BUPLEURUM MARGINATUM

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ABSTRACT: *Effect of ethanolic extract of Buplerum marginatum has been studied on the periodicity of oestrous cycle in adult rats. It is found that the extract at a dose of 50 mg/kg/day when given for 10 days caused significant increase in the duration of metaestrus phase of the oestrous cycle. Other longer durations of treatment also caused similar changes. Percent increase in the duration of metaestrus phase at 10,20 and 30 days has found to be identical. No significant effect on body weight was recorded during and after the treatment with extract. Irregularity in the oestrous cycle of treatment with extract. Irregularity in the oestrous cycle of treated rats resumed to normal after discontinuation of treatment, however, the period of recovery depended on the dose administrated.*

INTRODUCTION

Medicinal plants being the source of natural gifts have been considered as man's utmost need especially in the field of medicine. Even in ancient times these herbs were being used in birth control, to induce abortion or to reconstitute uterus after birth. These herbs are now under scientific investigations for their efficacy and mechanism(s). Thousands of plants have been screened in laboratory animals for potent antifertility activity (Dhawan *et al.*, 1977; Prakash *et al.*, 1978; Aswal *et al.*, 1993) but majority of them have failed due to number of reasons, of which, potent estrogenicity is the major one (Qureshi and Dixit, 1980; Saxena *et al.*, 1985; Patak and Prakash, 1989). Synchronization in the events at ovarian and uterine level is essential for normal reproductive functions and this phenomena are easily judged with a simple technique of vaginal cytology without manipulating the reproductive organs. Disturbance or irregularity in the oestrous cycle in rats is considered as an end

point to assess reproductive disorders. Antifertility agents of hormonal virtues usually alter the reproductive cyclicity. (Khokute and Udupa, 1976; Pakrashi and Pakrashi, 1977; Prakash *et al.*, 1985) but ideology is that of effect should not be persisted for longer duration after discontinuation of the treatment. These qualities depend on many factors including the presence of chemical constituents in extract and their interactions with reproductive tissues. Many of herbs are known to induce sterility in wild animals as the herbs disturbed the reproductive cyclicity of animals which never faced estrus.

Ethanolic extract of *Buplerum marginatum* Wall (sipil) has been reported earlier to possess significant anti – implantation and abortifacient activity (Prakash *et al.*, 1993). Present paper highlights the events occurring in the vaginal epithelium of adult

cyclic rats in relation to the administration of extract of *B.marginatum*.

MATERIALS AND METHODS

Healthy adult female rats (160 ± 10 g) of the Sparague – Dawley strain were collected from the departmental colony and were kept in separate cages individually. These were maintained under uniform husbandry conditions of light and temperature. Their vaginal smear was examined daily at 10.00 hrs to identify the stage of oestrous cycle. Animals showing 2 – 3 regular oestrous cycles were selected for the study and divided into different groups of control and experimental. Ethanolic extract of *B.marginatum* prepared as described earlier (Prakash *et al.*, 1993) was macerated with gum acacia suspended in distilled water and a dose of 50 mg/kg body weight was prepared. This dose is effected to inhibit implantation in rats (Prakash *et al.*, 1993). It was administered orally with the help of intragastric catheter to three different groups

of animals for 10, 20 and 30 days separately. Parallel controls were run with each group and these animals received vehicle only. Throughout the experimentation the vaginal smear of each rat was examined daily at 10.00 hrs. After the last administration of extract the treatment was discontinued, however, vaginal smear of each rat was continuously examined daily till the animals showed regular cycle. At the end of the experiment the record of different stages of oestrous cycle of each rat was analysed.

RESULTS

Table – I shows the effect of ethanolic extract of *B.marginatum* on the duration of various stages of the oestrous cycle. Control animals showed regular and normal oestrous cycle. Administration of extract for 10 days significantly lengthened the duration of metastrus phase in 7 rats out of 10. Similarly other longer durations of treatment caused significant increase in the duration of metastrus phase in majority of animals.

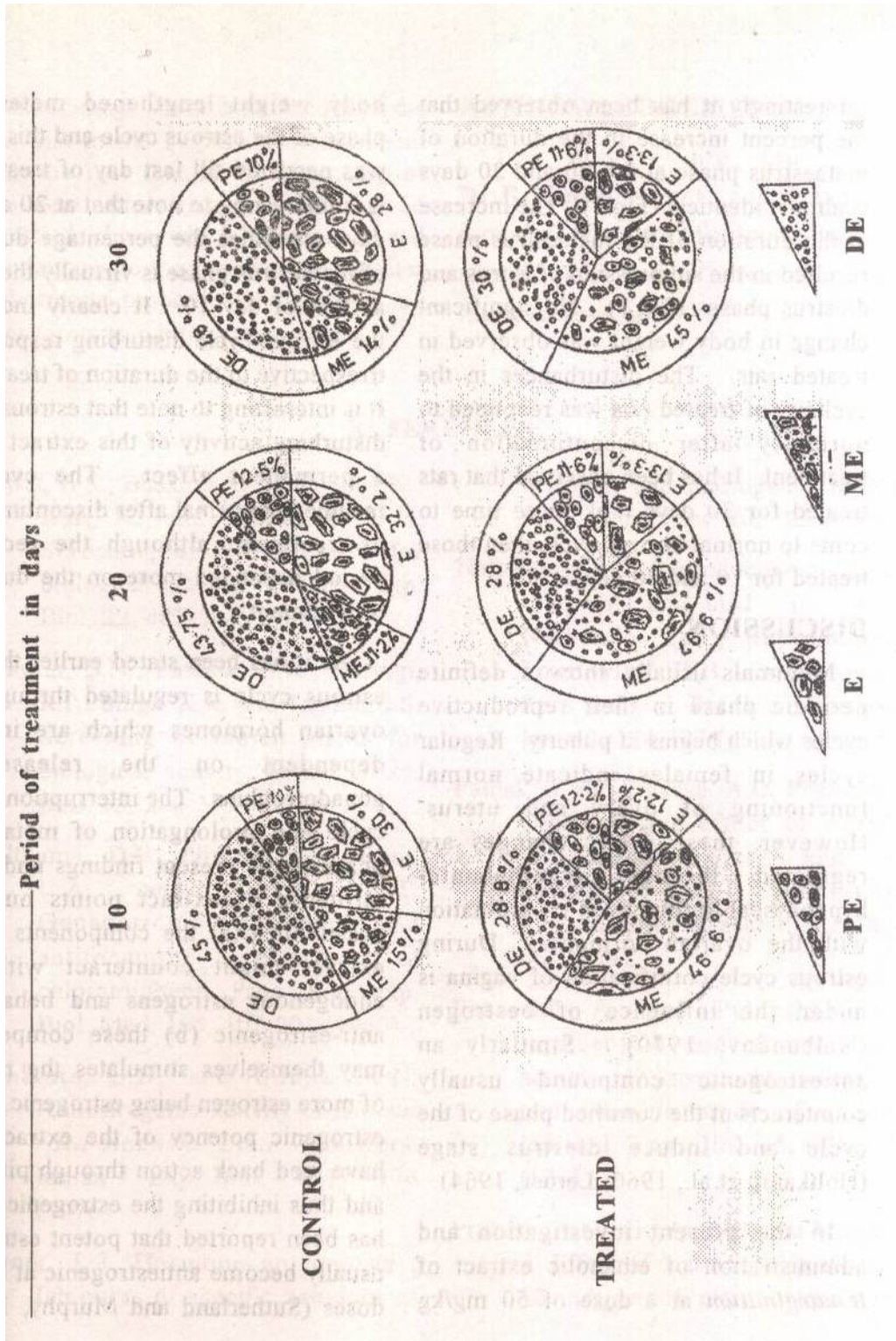
TABLE – I Effect of ethanolic extract of *B.marginatum* on the oestrous cycle of rats. (Ten rats were used in each experiments)

Dose mg/kg	Period of treatment days	Number of rats depicted change in the stages of oestrous cycle				Cyclicity Regular	Irregular**	Time Taken for restoration of normalcy in cycle (days)*
		Prolonged Diestrus	Prolonged Proestrus	Prolonged Estrus	Prolonged Metaestrus			
Control (Vehicle only)	-	0	0	0	0	7	3	Nil
50	10	0	0	3	7	3	7	6.2 ± 0.2
	20	1	0	2	7	3	7	10.1 ± 0.5
	30	1	0	2	7	5	5	15.4 ± 0.6

* Minimum period in days when animals showed regular cycle after

** Animals showing zigzag stages.

Fig. I – Effect of Ethanolic extract of *B. marginatum* wall on the periodicity of oestrous cycle in rats



Interestingly it has been observed that the present increase in the duration of metaestrus phase at 10,20 and 30 days is almost identical (Fig.I). The increase in the duration of the metaestrus phase resulted in the suppression of estrus and diestrus phases (Fig.I). No significant change in body weight was observed in treated rats. The disturbances in the cyclicity of treated rats was recouped to normalcy after discontinuation of treatment. It has been observed that rats treated for 30 days took more time to come to normal oestrus cycle than those treated for 10 and 20 days.

DISCUSSION

Mammals usually shows a definite periodic phase in their reproductive cycles which begins at puberty. Regular cycles in females indicate normal functioning of ovary and uterus. However, these cyclic changes are regulated through hypothalamic hypophyseal hormones in co-ordination with the ovarian hormones. During estrous cycle cornification of vagina is under the influence of oestrogen (Balbandov, 1970). Similarly an antiestrogenic compound usually counteracts at the cornified phase of the cycle and induce diestrus stage (Holtkamp et al., 1960; Lerner, 1964).

In the present investigation and administration of ethanolic extract of *B.marginatum* at a dose of 50 mg/kg body weight lengthened metaestrus phase of the estrous cycle and this phase was persisted till last day of treatment. It is interesting to note that at 20 and 30 days schedules the percentage duration of metaestrus phase is virtually the same as that of day 10. It clearly indicates the estrous cycle disturbing response is irrespective to the duration of

treatment. It is interesting to note that estrous cycle disturbing activity of this extract is not a permanent effect. The cyclicity resumed to normal after discontinuation of treatment, although the recovery period depended more on the duration of treatment.

As it has been stated earlier that the estrous cycle is regulated through the ovarian hormones which are in turn dependent on the release of gonadotrophins. The interruption in the cycle and prolongation of metaestrus phase in the present findings under the influence of extract points out two possibilities (a) the components in the extract might counteract with the endogenous estrogens and behave as anti-estrogenic (b) these components may themselves stimulates the release of more estrogen being estrogenic. High estrogenic potency of the extract may have feed back action through pituitary and thus inhibiting the estrogenicity. It has been reported the potent estrogens usually become antiestrogenic at higher doses (Sutherland and Murphy, 1982).

These findings are strengthened with the fact that in the beginning of treatment with the fact that in the beginning of treatment with extract some rats showed cornified phase which later on turned into leucocytic stage. However, studies on hormonal properties of the extract are in progress to confirm these facts.

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