

## Effect of indigenous medicinal plants on onset of puberty in immature female rats

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### ABSTRACT

Mature fresh green leaves of medicinal plants *Murraya koenigii* and young shoots of *Urtica dioica* were collected, botanically identified, shade dried, powdered and extracted with 50% ethanol under reflux. The effect of 50% ethanolic extracts of *Myrraya* and *Urtica* on onset of puberty was examined at three dose levels in immature rats i.e. 100, 300 and 1000 mg kg<sup>-1</sup> based on LD50 studies. Administration was performed orally from day 25-34 of age and induction of vaginal opening was used as an index of puberty. It was found that onset of puberty was substantially advanced along with body weight gain in *Murraya* 1000 mg kg<sup>-1</sup> dose group.

Key words : Medicinal plants, puberty, rats

Considerable attention has been focussed on reproductive endocrinology as a mean to identify specific problems and to adopt therapeutic measures by using exogenous hormones for augmenting fertility in farm animals (Hukeri, 1995) but hormones do not always give consistent results which may be due to a number of variables including age, weight, diet and management. Besides this other major constraints in the use of various hormonal preparations viz. high cost, adverse effects on reproductive performance, residual effects in food animals and their products, lack of easy availability and quick assay facilities etc. seriously demands minimization of our dependence on hormones for therapeutic/fertility augmentation purposes, by evolving suitable alternatives.

The advantages like affordable cost, efficacious, readily available, acceptable, with minimal side effects, gentle in action etc. make herbal remedies, as a potent alternative to hormones. Moreover, presence of minerals and salts are also beneficial in treatment (Atal and Kapoor, 1982). So far, meagre attention has been paid for identifying plants for animal fertility augmentation in contrast to fertility suppression in human beings. There is an immense need of hour for reproductive

biologists to get involved in the medicinal plant research in order to evolve suitable phytotherapies for fertility augmentation in livestock. Therefore, the present study was undertaken with two medicinal plants viz. *Murraya koenigii* (Curry leaf plant) and *Urtica dioica* (Bichchu grass), mentioned in traditional practice, in order to examine their effects on onset of puberty in immature female rats.

### MATERIALS AND METHODS

**Preparation of plant material :** Mature fresh green leaves of *Murraya koenigii* (Mur) and young shoots of *Urtica dioica* (Urt) were collected from Bareilly and Nainital Districts of U.P. and Uttaranchal respectively followed by botanical identification of plant material at National Botanical Research Institute (NBRI), Lucknow as *Murraya koenigii* (Accession No. 90068) of family Rutaceae and *Urtica dioica* (Accession No. 90067) of family Urticaceae. The collected materials were shade dried, powdered and exhaustively extracted with 50% ethanol under reflux. Ethanolic extracts were concentrated under reduced pressure to semisolid sticky mass and were made free of solvent. The plant extract were suspended in vehicle polysorbate-80 or Tween-80 (Polyoxyethylene sorbitan Mono-oleate : Himedia), which was added @0.1 ml/ml of distilled water to 1 g of extract to make a stock solution of 1g/ml concentration, separately. This stock solution was stored at 0°C and required dose levels were prepared fresh from this just before administration. Equal volume of

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polysorbate-80 was taken with distilled water to serve as vehicle for control groups.

**Acute toxicity studies and determination of LD<sub>50</sub>:** Thirty six adult, male mice were divided into nine groups of four animals each. Eight groups of mice received, Mur and Urt in logarithmic doses of 3.0, 4.5, 6.75 and 10.125 extract/kg body weight P.O. respectively, using tuberculin syringe fitted with a 22 gauge tilted blunt needle. The ninth group served as control. The mice were observed for gross behavioural changes, toxic signs or mortality. The mortality, if any was recorded upto a time period of 24 h and the LD<sub>50</sub> was determined according to the method of Weil (1952). The further experimental doses for both the extracts in rats were chosen as 100, 300 and 1000 mg/kg-1, respectively, based on LD<sub>50</sub> values.

**Effect of 50% ethanolic extract of Mur and Urt on onset of puberty in immature rats:** A total number of 42 immature female rats of 22 days age were taken for the study and they were divided into seven groups consisting of 6 animals in each group. Group I served as control where as group II to VII received Mur and Urt extract @100, 300 and 1000 mg/kg<sup>-1</sup> respectively, using same mode of administration as of mice. The animals were dosed once daily for ten days (from day 25 to 34 of age) and observed for onset of puberty. Onset of puberty was adjudged by vaginal opening (Bennet and Vickery, 1970) from day 26 onwards till all the control animals attained puberty. At the appearance of vaginal opening animal's body weight and age were recorded and the data was analysed statistically.

#### RESULTS AND DISCUSSION

**Acute toxicity studies and determination of LD<sub>50</sub>:** The extracts did not show any sign of toxicity or mortality over the

observation period of 24 h upto the dose level of 10.125 g/kg<sup>-1</sup>. Thus, it was assumed that the extracts were devoid of any acute toxic effects proving their wide margin of safety.

**Effect of 50% ethanolic extract of Mur and Urt on onset of puberty and body weight in immature rats:** The pertinent data is depicted in Table 1. Analysis of variance revealed that mean age at onset of puberty and body weight did not differ significantly between groups. However, earlier onset of puberty was observed in group IV animals as compared to control values (37.33±0.98 vs 46.5±3.09 days) with simultaneously higher body weight (41.16±2.30 vs 35.16±1.51 g). It was evident that there was a greater spread of age at onset of puberty than body weight indicating that puberty is more closely related to body weight than age, which is in agreement with Kannedy and Mitra (1963b). Parker and Mahesh (1976) have suggested that estrogen serve as primary trigger for setting up the sequence of events leading to puberty. Vaginal opening is an estrogen dependent event (Ojeda and Urbanski, 1994). Pubertal advancement under present study might be either through steroid (estrogen) stimulating action of Mur or by stimulation of steroidogenic activity of ovary under the hypothalamic effect. The gonads and the pituitary gland of prepubertal animals are not insensitive to the stimulus of tropic hormones. However, the stimulus from the hypothalamus is missing before puberty (Donovan and Harris, 1966).

Though statistical significance could not be obtained in this study but the findings has its economic importance. Since the endocrine profiles were not studied, further studies are warranted to ascertain the mechanism of action of plant extract in pubertal advancement and growth promoting effects.

Table 1 : Effect of *Murraya* and *Urtica* on onset of puberty and body weight in immature female rats (Mean±SE)

Experimental group (n=6)	Onset of puberty (days)	Body weight at puberty (g)
I (Control)	46.5±3.09	35.16±1.51
II (Mur 100)	43.16±2.83	37.83±3.48
III (Mur 300)	44.83±2.94	38.83±3.99
IV (Mur 1000)	37.33±0.98	41.16±2.30
V (Urt 100)	45.33±1.82	38.5±3.11
VI (Urt 300)	48.16±2.73	39.83±1.72
VII (Urt 1000)	46.0±2.75	37.16±2.53

Means do not differ significantly between groups

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