

Study of anti-fertility activity of alcoholic extract of bark of *Moringa oleifera*

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Unlike animals, man can control the reproduction at will and plan a family. Research for the evolution of a safe acceptable cheap effective and reversible antifertility agents of plant origin is progressing world wide. The present study is an effort towards finding the antifertility activity of *Moringa Oleifera* commonly known as 'Drum Stick'. The results of this study indicate that the alcoholic extract of bark of *Moringa Oleifera* could prolong the di-oestrous phase and shorten the pro-oestrous and oestrous phases in proven fertile female albino rats. Anti implantation activity was observed in 71% and post implantation resorption in 29%.

Keywords: Fertility, implantation, oestrous cycle.

Man's great reproductive potential and greatly increased survival rate in recent years have posed a major problem of population growth with far reaching consequences. Unlike animals he alone can control the reproduction at his will and plan his family. Scientific works are presently engaged in the search for a safe, acceptable effective, easily administrable, cheap, reversible antifertility agent from the extracts of herbs and plants commonly grown in India and elsewhere. Such herbs and plants have been used since ages for fertility control purposes. The present study is an effort towards finding the antifertility activity of *Moringa oleifera* commonly known as "Drum Stick", a member of family 'Moringaceae'. An alcoholic extract of root and bark of the plant has been reported to cause foetal resorption in the later day of pregnancy.

Materials and methods

Collection and preparation of plant extract: The bark of *Moringa Oleifera* was collected and dried

in shade and was coarsely powdered and sieved to get 1.5 kg of fine powder. This was exhaustively extracted with ethyl alcohol in a soxhlet apparatus which yielded orange coloured extract. It was concentrated on steam bath to yield a solid mass, which was dissolved in 60% ethyl alcohol to get a concentration of 200 mg of extract per 1 ml of solution.

Animals: Healthy adult female albino rats of wistar strain weighing between 150-175 gms were selected for the study. All the animals were maintained under uniform husbandry, light and temperature and were fed with rat pellets and water.

Acute toxicity studies: Acute toxicities were conducted in 8 groups of albino rats of wistar strain

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weighing about 150-175 gms to determine the effects of alcoholic extracts of bark of *Moringa oleifera*. After overnight fasting the 8 groups of rats were administered intraperitoneally a calculated dose of 0.05-0.5 gms/kg body weight.

The animals were observed for every hour for 4 hrs, on the first day and once daily for the remaining 6 days. The group of rats which received the test extract upto 100 mg/kg/body weight showed no mortality. The group of rats which received the test extract of 150 mg, 200 mg and 400 mg/kg/body weight and onwards showed a mortality of 25%, 50%, 75% and 100% respectively.

Experimental methods

Action on oestrous cycle of female albino rats: Proven fertile uniform conditions were selected for the study. The rats were checked daily for different stages of oestrous cycle by studying the vaginal smears. The smears were prepared by putting a few drops of saline into the vagina of the animal taking care that the cervix was not touched. The fluid was sucked out by the dropper and the same was transferred on to a clean slide and examined under microscope. Only those rats that showed normal cyclic changes for three consecutive cycles were selected and divided into 2 groups, each group containing 6 rats.

One group was treated with the alcoholic extract of bark of *Moringa oleifera* for 12 days at a dose of 100 mg/kg/day. The other group was given 1.5 ml of 1% gum accacia solution daily for 12 days. Vaginal smears were examined every day in both the groups to find out whether the oestrous cycle was altered or not. The changes in the pattern of oestrous cycles were studied for 6 cycles, statistically analysed and tabulated.

Action on implantation: Proven fertile female albino rats of wistar strain weighing between 150-175 gm were taken up for the study. The rats were checked daily for normal oestrous cycle by studying their vaginal smears. The rats which showed normal oestrous cycle were selected for the study. Those rats which were in pro-oestrous or oestrous phase were left overnight with males of proven fertility in the ratio of 1:3. Next morning the vaginal smears were checked for the presence of spermatozoa. The day the sperms were detected in the vaginal smears was considered as Day 1 of pregnancy. 12 such pregnant animals were selected and they were divided into 2 groups of 6 rats each, one group was fed orally with 1.5 ml of 1% gum accacia solution. The test group was injected with the alcoholic extract of bark of *Moringa oleifera* at a dose of 100 mg/kg/rat/day for 5 days starting from the first day of pregnancy.

Laparotomy was performed under semi-sterile conditions and under light ether anaesthesia on the 10th day of pregnancy. Uterine horns were observed for the size of implantation sites.

Action on post-implantation activity: Young female rats of proven fertility weighing between 150-175 gms. were taken up for the normal oestrous cycle by studying their vaginal smears.

The rats which showed normal oestrous cycle were selected for the study. Those rats which were in pro-oestrous or oestrous phase were left overnight with males of proven fertility in the ratio 1:3. Next morning the vaginal smears were checked for the presence of spermatozoa. The day the sperms were detected in the vaginal smears was considered as day 1 of pregnancy. They were divided into 2 groups of 6 each, one group of six animals was taken as control and fed with 1.5 ml. of 1% gum accacia solution. The second group was taken as the test group and injected with the alcoholic extract of bark of *Moringa oleifera* at a dose of 100 mg/kg/rat/day.

starting from the 7th to 11th day of pregnancy. Laparotomy was performed under light ether anaesthesia under semi-sterile conditions. The uterine horns were observed for implantation and resorption sites.

Results

Anti-ovulatory activity:

a) Action of the test extract on oestrous cycle in albino rats: In the test group of animals fed with the alcoholic test extract the di-oestrous phase was prolonged in many of the six cycle observed and the pro-oestrous and oestrous phase was shortened.

In the test group, 5 out of 6 animals showed a prolongation of the di-oestrous phase in the last three cycles which may indicate delayed ovulation. The average number of di-oestrous days in 6 cycles is 19.6% days as compared to the control group of 6 animals in which the average number of di-oestrous days are shortened by 61% and the di-oestrous days are prolonged by 80% in the extract fed group of animals.

The results when statistically analysed showed a mean value of 19.6 days, and 't' value of 4.9 ($P < 0.001$) comprising of di-oestrous days which is statistically significant. However with respect to oestrous days the mean value was 6, 't' value is 3.24 ($P < 0.02$) which is not significant.

b) Action on implantation: Laparotomy conducted on the 10th day of pregnancy in the test extract fed group of animals showed a complete absence of implantation in the uteri of four out of six experimental animals and a reduced number of implantation sites in the remaining two experimental animals. The results when statistically analysed showed

a 't' value of 8.33 ($P > 0.01$) with respect to the implantation sites.

c) Post implantation activity: The test extract fed group of animals when observed throughout the entire period of pregnancy did not show any evidence of vaginal bleeding. Laparotomy conducted on the 16th day of pregnancy showed a reduced number of implantation sites and a few resorption sites when compared with the control group.

The results when statistically analysed showed that the 't' value is 4.37 ($P < 0.01$) with respect to the implantation sites.

Discussion

The anti fertility effect of *Moringa oleifera* has been reported by various laboratories [3,6,7]. However controversy exists with regard to doses used, methodology and the process of extraction from crude material.

The WHO, while, preparing a computer search of the literature for fertility regulation plants, has rated this plant with a high index priority for further studies.

The present findings indicate that, the alcoholic extract of bark of *Moringa oleifera* showed significant effect on the oestrous cycle of the albino rats (80%), anti implantation activity (71%) and a moderate post implantation activity (29%).

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