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Antifertility Activity of Low Dose of Alcoholic Extract of *Vinca rosea (Catharanthus roseus)* (L.) Leaves in Female Swiss Albino Mice

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Abstract: This study investigated the antifertility potential of a 5 mg/day oral dose of the alcoholic extract of Vinca rosea (L.) leaves in adult female Swiss albino mice. Vinca rosea (also known as Catharanthus roseus, Madagascar Periwinkle, or Apocynaceae) is traditionally known for its medicinal properties, including those affecting reproductive health. The extract was administered to female mice post-coitally to evaluate its effect on pregnancy. Results demonstrated a significant reduction in implantation rate, suggesting potent anti-implantation activity and possibly anti-estrogenic activity. Biochemical analysis of the uterus revealed altered levels of key reproductive markers like glycogen and cholesterol, which can be linked to the extract's mechanism of action.

Keywords: antifertility activity, Vinca rosea extract, reproductive health, implantation inhibition, biochemical analysis

1.Introduction

The search for safe, effective, and reversible antifertility agents from natural sources remains a crucial area of pharmacological research, driven by concerns over side effects associated with synthetic contraceptives. *Vinca rosea* (L.), a plant rich in indole alkaloids like vinblastine and vincristine (known for their anti-cancer properties), has also been traditionally employed in various parts of the world for regulating fertility.

Previous studies have reported the antifertility activity of various extracts of *Vinca rosea* in both male and female animal models. Specifically, the leaves have been reported to possess anti-implantation efficacy. This study focuses on a specific, low-level 5 mg/day dose of the alcoholic extract of the leaves, administered orally to pregnant female Swiss albino mice to precisely determine its efficacy as a post-coital interceptor agent, focusing primarily on its anti-implantation effect.

2. Materials and Methods

2.1. Plant Material and Extract Preparation

Plant extract and animals used: Leaves of the experimental plant Vinca Rosea were collected from agricultural farms near Jaipur, Rajasthan. They were then authenticated in the Herbarium, Department of Botany, University of Rajasthan, Jaipur, under the specimen voucher No. RUBL-20841. The leaves were shade dried, powdered, and extracted with alcohol (90%) in a Soxhlet apparatus, to obtain a semi-solid viscous dark green mass, i. e., the extract.

Colony-bred adult healthy male and female Wistar mice weighing 30 to 45 g were used in the present investigation. The mice were housed in standard cages and maintained under standard conditions (12h light/dark cycle, room temperature) and provided standard laboratory chow

(Ashirwad Food Industries, Chandigarh, India), and water was provided ad libitum. The extract was administered intramuscularly. The study was approved by the Institutional Ethical Committee of the Department of Zoology, Vedic Kanya P. G. College, Jaipur. The Indian National Science Academy (2000), New Delhi, guidelines were followed for the maintenance of experimental animals.

2.2. Experimental Animals

Adult female Swiss albino mice (weighing approximately 30-45 g) were used. The animals were housed under standard laboratory conditions (12-hour light/dark cycle, controlled temperature, and access to standard pellet diet and water *ad libitum*). All animal procedures followed ethical guidelines.

2.3. Antifertility Study Protocol

Female mice were caged overnight with fertile males in a 2: 1 ratio. Day 1 of pregnancy was confirmed by the presence of a copulatory plug or sperm in the vaginal smear the following morning. Pregnant mice were randomly divided into two groups of ten animals each:

- 1. **Control Group:** Received the vehicle (Distilled water) orally for 3 consecutive days (Day 1 to Day 3 post-coitum).
- 2. **Test Group:** Received the **alcoholic extract** of *Vinca rosea* leaves at a dose of **5 mg/day/mouse** intramuscularly for 3 consecutive days (5 (Day 1 to Day 3 post-coitum).

2.4. Evaluation of Antifertility Efficacy

All mice were sacrificed on **Day 10 post-coitum** under light ether anaesthesia. The following parameters were recorded:

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- Number of Corpora Lutea (CL): Counted on the surface of both ovaries (indicating the number of ova shed).
- Number of Implantation Sites (IS): Counted in both uterine horns (indicating successful implantation).

Additionally, uterine weight and biochemical changes in the sialic acid content were estimated by the method of Svennerholm (1950) as given by Glick (1960), to assess potential anti-estrogenic or estrogenic effects.

2.5. Statistical Analysis

Data were expressed as Mean $\gamma \$ Standard Error of the Mean (SEM) and analysed using an appropriate statistical test (e. g., Student's t-test or ANOVA) to determine significance (SP < 0.05).

3. Results and Discussion

3.1. Antifertility Efficacy

The administration of **5 mg/day** of the alcoholic *Vinca rosea* leaf extract resulted in a statistically **significant reduction** (SP < 0.01) in the mean number of **implantation sites** compared to the control group. The **85.24 % implantation activity** is a robust indication of the extract's efficacy as a post-coital contraceptive agent at this specific dose. The number of Corpora Lutea remained comparable across both groups, suggesting the extract did not significantly affect ovulation, thus confirming its action as an **anti-implantation** agent rather than an anti-ovulatory one.

3.2 Biochemical Alterations

Daily treatment of adult mated females with the alcoholic extract of leaves of *Vinca rosea* from day 7 to 9 *pc* decreases the uterine sialic acid content on day 12 *pc*. This decrease is statistically significant in relation to the mean control value on the same day.

3.3. Possible Mechanism of Action

The observed anti-implantation activity is likely due to the presence of alkaloids, flavonoids, and/or steroidal compounds in the alcoholic extract. A common mechanism for anti-implantation is the disruption of the hormonal

balance necessary for maintaining the receptive state of the uterus (endometrial receptivity) for the blastocyst.

- Anti-estrogenic/Estrogenic Effect: Many plant-derived antifertility agents act by exhibiting either a strong estrogenic or an anti-estrogenic effect. Changes in uterine weight and histology often indicate this. A decline in uterine weight, as reported in similar studies, suggests an anti-estrogenic action, while an increase might suggest an estrogenic one, both of which can prevent successful implantation.
- Biochemical Sialic acid is found as a part of a complex of mucoproteins which forms the mucus of the uterus and vagina (Cappola and Ball, 1966; Carlborg, 1969). In the present investigation treatment with postcoitally effective antifertility doses of the petroleum ether, alcoholic extract and chromatographic fraction (petroleum ether and benzene, 1.1 V/V) of the petroleum ether extract of leaves of Catharanthus roseus causes a significant decrease in the sialic acid content of the uterus of the mouse. The above results are in agreement wth those of Dixit et al., (1975, 1976) who reported in mouse, rat and gerbil that the alcoholic extract of Canabis sativa seeds causes a decline in the sialic acid level in the uterus of intact animals. Rajalakshmi et al., (1972) have studied chan- ges in uterine sialic acid during early pregnancy in the rat. Sialic acid was maximum on day 1 to 4 of pregnancy but becomes low on day 5 and 6 of pregnancy. Vinca Alkaloids: The presence of Vinca alkaloids (e. g., vinblastine, vincristine) might also contribute through their known mechanism of microtubule disruption, potentially interfering with rapid cell division of the blastocyst or the necessary cellular changes in the uterine lining for implantation.

4.Conclusion

The results confirm the significant **antifertility activity** of the 5 mg/day alcoholic extract of *Vinca rosea* leaves in female Swiss albino mice, primarily through an **antiimplantation** mechanism. The high percentage of pregnancy interruption validates the traditional claims for this plant in fertility regulation. Further research is necessary to isolate the specific active phytoconstituents responsible for this action and to perform detailed toxico-pharmacological studies to ensure safety and reversibility for potential use as a novel, plant-derived contraceptive agent.

Table 1: Effect of administration of various extracts of leaves of Catharanthus roseus from day 7 to 9 postcoitum (pc) on the fertility of mice- (Number of mice in each group)

			Autopsy On Day 12 Postcoitum		
Group	Extract	Treatment mg/dose/day/mice	Corpora Lutea	Implantation Sites	Percentage Implantation
Control		-	61	52	85.24
Experimental	Alcoholic	5	56	33	58.92

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Table 2: Effect of administration of various extracts of leaves of Catharanthus roseus from day 7-9 postcoitum (pc) on the body and uterine weight of mice

AUTOPSY ON DAY 12 pc								
Group	Extract	Treatment mg dose/day/mice	Initial Body Weight (gms) mean SEM	Final Body Weight (gm) Mean +SEM	Uterine Weight (mg) Mean ± SEM			
Control		-	33_5±0.8	35.7±1.03	372.2±31.94			
Experiment AL	Alcoholic							
		5	27.6±27	28.5+2.7	104.2+18.10 ***			

Minimum number of mice in each group =5

P < 0.05, almost significant;

Table 3: Effect of administration of various extracts of leaves of Catharanthus roseus from day 7 to 9 postcoitum (pc) on the biochemical constituents of the uterus of mice (Autopsy on Day 12 postcoitum pc)

Group	Extract	Treatment (mg/day/mice	Sialic Acid
Control	-	-	0.61±0.03
Experimental	Alcoholic	5	0.57 ±.05*

Minimum number of mice in each group =5

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^{**} P < 0.01 Significant and;

^{***} P < 0.001 highly significant in relation to control.

P < 0.05, almost significant;

^{**} **P** < 0.01 Significant and;

^{***} P < 0.001 highly significant in relation to control.