

# Research Article

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# ANTI IMPLANTATION AND PREGNANCY INTERRUPTION ACTIVITY OF JAPAKUSUMA (HIBISCUS ROSA SINENSIS) IN ALBINO RATS

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#### ARSTRACT

Increase in population has affected many socio-economic conditions of people by increasing crimes, illiteracy, destructive activities, diseases, improper food and shelter. Thus to control this population and limit the family size at a personal level and at a national level, modern contraceptive methods and medicines were introduced long back. There are many new contraceptives available now, but they have various side effects. Some traditional practitioners used to dispense oral contraceptives mentioned in Ayurvedic classics. Such as 1) Pippali (Piper longum), Vidanga (Emblica ribes) and Tankana (Sodii Biboras). 2) Talisapatra (Taxus baccata) and Gairika (Hematite) with cold water and 3) Kanji bhavita Japakusuma (Hibiscus rosa sinensis). An experimental study on above mentioned 1 and 2 formulations has proved its efficacy as temporary contraceptive medicine. To evaluate the permanent or long term temporary contraceptive effect of Japakusuma, an attempt was made in this study. Study was conducted by Choudary and Khanna method on 18 female, 36 male (for mating) albino rats. Japakusuma, Propylene glycol, Ovral L formed the materials. Single dose was administered on proestrous stage of rat oestrous cycle and observed for anti-implantation and pregnancy interruption activity. Test drug showed significant anti implantation and pregnancy interruption activity. Thus showing the temporary contraceptive activity of Japakusuma (Hibiscus rosa sinensis).

Key words: Anti-implantation, Pregnancy interruption activity, Japakusuma. Ovral L

#### INTRODUCTION

Reproduction is a unique property and vital process of the living being. This can produce young ones like themselves after having attained maturity. Thus continuity of life can be maintained. In ancient times it was believed that without having a child, one cannot get moksha. Man having numerous progeny is like one having numerous forms, faces, troop's action, eyes, knowledge and selves. He was praised as auspicious, commendable, worthy, potent and numerously branched. Such belief made the man to get more number of children, as well as man's great reproductive potential and the greatly increased survival rate. In recent years we have posed a major problem of population growth with far reaching consequences<sup>2</sup>.

This kind of beliefs became foundation to population explosion. According to UN projection 2001, world population is expected to be 7973 million at 2025 and 9322 million at 2050. Censes reports of India show that, India has reached one billion on 12<sup>th</sup> May 2000<sup>3,5</sup>. There will be no proper check on crimes, diseases, illiteracy, other destructive activities, food and shelter.

Above results created need to limit family size at a personal level and for the control of population at a national level. With the increase in population it is obvious that serious problems loom ahead, unless the number of our progeny is controlled<sup>4</sup>.

In 1960 various types of contraceptive method were tried on women such as IUCD's, Tubal ligations, paraenteral (injectables) and oral contraceptives (OC). Unfortunately none of them have proved to be absolutely safe method though almost 100% contraception was seen in the case of combined OC's and Tubectomy.<sup>5</sup>

PID (Pelvic Inflammatory Disease), Pain during insertion, spontaneous expulsion and possibilities of change in position made the women to stay far from adopting the IUCDs. Ligation needs a minor surgery. Injectable contraceptives contain formulated depot medroxy progesterone acetate (Depo provera) or Norethisterone enanthate or noristerat which may induce weight gain, nausea, headache, etc.<sup>5</sup>.

Presently available Oral contraceptives like Mala-D, Mala-N, Ovral, Novelan etc. may induce weight gain, nausea, headache, Cancer of Cervix, CA (Carcinoma) of breast, certain neurologic and neuro-opthalmologic syndrome, candida vaginitis, trichomans vaginitis.<sup>5</sup> Because of all these hazards actually women are in need of a safe, acceptable, economic contraceptive medicine.

The concept of an ideal contraception is not newer to Indian women. The first documented method of birth control in India was available 2000 year ago. The first authoritative book was Kamasutra. It is also explained in Ratrirahasy or Kokla shastra <sup>5</sup>

Some traditional practitioners used to dispense oral contraceptives. In Ayurvedic classical texts like Yogaratnakar, Bhavaprakash, Bhaishajya Ratnavali etc oral contraceptives like

- 1) Pippali (*Piper longum*), Vidanga (*Emblica ribes*) and Tankana (*Sodii Biboras*). <sup>6,7,8</sup>
- 2) Talisapatra (*Taxus baccata*) and Gairika (*Hematite*) with cold water<sup>6,7,8</sup>
- 3) Kanji bhavita Japakusuma (*Hibiscus rosa sinensis*) <sup>6,7,8</sup> are mentioned. An experimental study on above

mentioned 1 and 2 formulations have proved its efficacy as temporary contraceptive medicine <sup>9,10,11</sup>.

But till today, there is no research conducted to find out a non-surgical permanent contraceptive or long acting temporary contraceptive by which pregnancy can be prevented.

In order to evaluate the contraceptive effect and to provide safe economic contraceptive formulation, an attempt was made in this study.

# MATERIAL AND METHODS

# Materials

#### Drugs

Japakusuma (*Hibiscus rosa Sinensis*) (Kanji bhavita Japakusuma) (Oral) (Test Drug)

Ovral L (0.15mg levonorgestrel + 0.03mg ethinyl estradiol) (Oral) (Standard Drug)

Propylene glycol (to prepare suspension for test and standard drug) (Oral) (Control Drug)

Animals: Wistar strain male and female albino rats: 18 female albino rats, 36 male were taken from the animal house, BLDEA's AVS **PGCRC** Ayurveda mahavidyalaya. Bijapur, India with Ethical Committee clearance No: AVS/PGCRC/IAEC/18/2007. All the experimental animals were maintained under standard laboratory conditions, fed with balanced food and water as per the CFTRI formula prepared at Pranav food industries Sangali, Maharashtra, India. 12 hour light and darkness maintained in animal house with temperature of 18°-25°C. Different groups of animals placed separately in propylene rat cage.

# Methods

# Method of preparation of Kanji Bhavita Japakusuma

Japakusuma pushpa was taken in clean and dry Khalwa yantra, pounded well and fine powder was prepared by vastra galana method. Thus prepared fine powder was mixed with kanji in a Khalwa yantra and subjected to bhavana. <sup>6,7,8</sup>

# Method of preparation of Medicine for administration

Group I: Fine powder of *Hibiscus rosa sinensis* was taken and mixed well into 2 ml of Propelyne glycol, shaked vigorously in test tube and then the uniform suspension was fed to albino rats orally by a syringe.

Group II: Ovral L was mixed well into 2 ml of Propelyne glycol and shaked vigorously in test tube and was fed to albino rats orally by syringe.

Group III: 2 ml of Propelyne glycol was fed to albino rats orally by syringe.

#### Method of selection of Animals

**Inclusion criteria:** Healthy fertile female albino rats of child bearing age and with normal oestrous cycle. Body

weight between 150 to 200 gm. Fertile male rats were taken for mating.

**Exclusion criteria:** Unhealthy albino rats, female albino rats of body weight less than 150 grams and more than 200 gm. Sterile male and female rats.

#### Anti implantation activity

Anti implantation activity was conducted by following Choudary and Khanna method <sup>12-14</sup> It involves 6 steps.

- · Taking vaginal smear.
- Examination of smear to know the phase of oestrous cycle.
- Allowing animals for mating 1 : 2 (female : male) ratio.
- Observation for sperm clumps to confirm mating.
- Drug administration
- On 10<sup>th</sup> day of drug administration rats were subjected to laparotomy to observe for implantation

Sample size: n= 6 in each group

## Drug schedule

Group I: 4.5 mg / 200 mg body weight of albino rats test drug *Hibiscus rosa sinensis* with 2 ml of Propelyne glycol Group II: Overal L with 2 ml of Propelyne glycol Group III: 2 ml of Propelyne glycol.

# **Outcome Measures**

#### **Primary outcome**

To compare the anti implantation activity of *Hibiscus* rosa sinensis to that of standard drug i.e. Ovral L and Control Propelyne glycol

# Secondary outcome

- Mean birth weight of litters
- Survival of litters

#### **OBSERVATION AND RESULTS**

Table 1: Phytochemical constituents of aqueous extract of Japakusuma (Hibiscus rosa sinensis)

Organic Constituents	Japakusuma (Hibiscus rosa sinensis)		
Alkaloids	-		
Carbohydrates	-		
Tannins	+		
Steroids	+		
Triterpenoids	-		
Saponins	+		
Flavanoids	+		
Carotenoids	-		

Aqueous extract of Japakusuma showed presence of phytochemicals like tannins, saponins, steroids and flavonoids.

Table 2: Anti implantation activity of Japakusuma (Hibiscus rosa sinensis) (primary outcome measure): (n=06)

Group	Drugs	No. of Rats	Mean no. of implantations	% inhibition of implants
I	Japakusuma (Hibiscus rosa sinensis)	6	0	100%
II	Overal L	6	0	100%
III	Propylene Glycol	6	9 <u>+</u> 0.53	0%

All the animals in Test group and Standard group have shown 0% mean number of implants and 100% inhibition of implants. Control group shown  $9\pm0.53$  % mean number of implants and 0% inhibition of implants.

Table 3: Secondary outcome measures

Group	Drugs	% of rats delivered on full term	Mean no births	Mean weight of litters	Died within 2 days
I	Japakusuma (Hibiscus rosa sinensis)	0%	0	0	0
II	Overal L	0%	0	0	0
III	Propylene Glycol	100%	7.7 <u>+</u> 0.48	6.3 <u>+</u> 0.048	2 <u>+</u> 0.44

In Japakusuma (test drug) and Overal L (Standard drug) 0% of rat delivered on full term, mean number of births were 0 (zero), 0 (zero) mean weight of litters and 0 (zero) litters died within two days.

In Control group 100 % rats were delivered after full term, 7.7 + 0.48 mean number of births, 6.3 + 0.048 gm and  $2 \pm 0.44$  litters died within two days of birth.

#### DISCUSSION

In this anti-implantation and pregnancy interruption study, results of test sample were compared with standard and control groups. Study was conducted in six stages. To assess contraceptive activity of test sample by the observation of anti implantation and pregnancy interruption activity in mature female albino rats.

In Ist stage, anti implantation activity was conducted to assess contraceptive activity by following Choudary and Khanna method. Laparotomy was conducted on 10<sup>th</sup> day after drug administration. Results of three groups were compared. In Group I and II implantations were not found, which indicates that both the test and standard drugs have demonstrated anti-implantation activity.

The anti implantation activity and pregnancy interruption activity might be postulated in the following ways based on the experimental and phytochemical studies. Estrogen and progesterone both hormones are essential for maintenance of regular menstruation cycle, production of ovum, maintenance of pregnancy in all stages. Hence anti-implantation activity seen in this study may be due to anti-oestrogenic and anti-progestrogenic effect.

Phytochemical analysis of Japakusuma (Hibiscus rosa sinensis) has shown the presence of steroids, saponins, flavanoids and tannins. Especially steroids and saponins are used as raw material for preparation of medically useful steroids and sex hormones like progesterone, oestradiol, and testosterone. Thus steroids, saponins might have contributed in the contraceptive activity of the drugs.

### CONCLUSION

Significant anti implantation and pregnancy interruption activity was noted in the Group I indicating the contraceptive activity of the test drug. The presence of steroids, saponins, flavanoids and tannins in japakusuma might have contributed in contraceptive activity.

# Scope for further research

This is an animal based experimental study but the promising results of this study necessitate a well designed

randomized clinical research before the test drug is recommended for clinical practice.

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