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Research Article

COMPARATIVE STUDY ON ANTI IMPLANTATION AND PREGNANCY INTERRUPTION ACTIVITY OF JAPAKUSUMA WITH HERBO MINERAL FORMULATIONS IN ALBINO RATS

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ABSTRACT

Increase in population has affected many socio-economic conditions of people by increasing crimes, illiteracy, destructive activities, diseases, improper food & shelter. Thus to control this population & limit the family size at a personal level and at a national level, modern contraceptive methods & 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. Oral contraceptives like 1) Pippali, (*Piper nigram*) + Vidanga (*Emblica ribes*) + Tankana (Borax). 2) Talisapatra (*Taxus baccata*) + Gairika (Hematite) with cold water 3) Kanji bhavita Japakusuma (*Hibiscus rosa sinensis*) are mentioned. An experimental study on above mentioned 1 & 2 formulations have proved its efficacy as temporary contraceptive medicine at BLDEA's AVS PGCRC, Ayurveda Mahavidyalaya vijayapur. An attempt was made to evaluate the permanent or long term temporary contraceptive effect of Japakusuma with Talisapatra Gairika. Study was conducted by Choudhry and Khanna method on 18 female, 36 male (for mating) albino rats. Japakusuma, Talisapatra, Shudha Gairika, Propylene glycol formed the materials. Single dose was administered on proestrous stage of rat estrous cycle & observed for anti implantation & pregnancy interruption activity. Test groups shown significant anti implantation & pregnancy interruption activity. Test drugs have shown significant anti implantation & pregnancy interruption of early pregnancy. Temporary contraceptive activity of Japakusuma (*Hibiscus rosa sinensis*). Talisapatra (*Taxus baccata*), shudha Gairik was observed.

Key words: Talisapatra, Shudha Gairika, Japakusuma, Anti implantation, Pregnancy interruption activity

INTRODUCTION

There is an increasing trend in the use of medicinal plants, botanicals or herbal preparations particularly in developing countries where these products are readily available.[1] It is a well known fact that the demand for the herbal drug treatment of various ailments is increasing and plant drugs from the Ayurvedic system are being explored more, not only in India but also globally. As a result, many research studies are being undertaken and there is a need for an update and to put them together. [2]

Rapid rise in population has caused serious problems in the economic growth and all round human development in developing countries like India. Family planning has been promoted through several methods of contraception, but due to serious adverse effects produced by synthetic steroidal contraceptives,[3-5] attention has now been focused on indigenous plants for possible contraceptive effect.

Hibiscus (Malvaceae) is a genus of herbs, shrubs and trees. Its 250 species are widely distributed in tropical and subtropical regions of the world and are reported to posses various medicinal properties viz; antitumor, antihypertensive, anticancer, antioxidant, anti ammoniac [6-10]. About 40 species are found in India. *Hibiscus rosa sinensis* Linn. is a native of China and is a potent medicinal plant. It is a common Indian garden perennial shrub [11] and often planted as a hedge or fence plant.

Some traditional practitioners used to dispense oral contraceptives. In Ayurvedic classical texts like Yogaratnakar, Bhavaprakash, Bhaishajya Ratnavali etc oral contraceptives like 1) kanji bhavita Japakusuma (*Hibiscus rosa sinensis*)[12-14] 2) Talisapatra+Gairika with cold water[12-14] 3) Pippali +Vidanga+Tankana are mentioned.[12-14] An experimental study on above mentioned 1 & 2 formulations have been proved as temporary contraceptive medicine at BLDEA's AVS, PGCRC, Ayurveda Mahavidyalaya, vijayapur. [15-18]
Not much research conducted to find a non surgical permanent contraceptive or long acting temporary contraceptive by which pregnancy can be prevented till date.

MATERIAL AND METHODS

Drugs: Talisapatra, Shodhita Gairika, Japakusuma (*Hibiscus rosa sinensis*) (Kanji bhavita Japakusuma)

Animals: Wister strain male & female albino rats 18 female & 36 male albino rats were taken from the animal house, BLDEA's AVS PGCRC Ayurveda mahavidyalaya vijayapur. Institutional ethical clearance no was AVS/PGCRC/IAEC/18/ 2007. All the experimental animals were maintained under standard laboratory conditions, fed with balanced food & water

as per the CFTRI formula prepared at Pranav food industries Sangali, Maharashtra. 12 hour light & darkness maintained in animal house with temperature of 18^0-25^0C. Different groups of animals placed separately in propylene rat cage.

Method of preparation of kanji bhavita japakusuma

Japakusuma pushpa was taken in clean & dry Khalwa yantra, pounded well & fine powder was prepared by vastra galana method. Thus prepared fine powder was mixed with kanji in a motor & pestle and subjected bhavana. After completion of bhavana dried under shade. Thus prepared fine powder stored in clean & air tight container. [12-14]

Gairika shodhana [19]

Raw gairika was subjected to bhavana with godugda. The process was repeated 7 times.
Talisa patra taken in clean & dry Khalwa yantra, pounded well & fine powder was prepared by vastra galana method. Thus prepared fine powder stored in clean & air tight container

Method of preparation of Medicine for administration

Group I: Fine powder of Talisapatra (*Taxus baccata*), shodhita Gairika & Japakusuma (*Hibiscus rosa sinensis*) (Kanji bhavita Japakusuma) were taken & mixed well into 2 ml of Propylene glycol, shacked vigorously in test tube & then the uniform suspension was fed to albino rats orally by a syringe.
Group II: Japakusuma (*Hibiscus rosa sinensis*) (Kanji bhavita Japakusuma) was taken & mixed well into 2 ml of Propelyne glycol, shacked vigorously in test tube & then the uniform suspension was fed to albino rats orally by a syringe.
Group III: Fine powder of Talisapatra (*Taxus baccata*) and shodhita Gairika were taken & mixed well into 2 ml of Propelyne glycol, shacked vigorously in test tube & then the uniform suspension was fed to albino rats orally by a syringe.

Method of experimental study [20- 25]
Method of selection of Animals

Inclusion criteria: Healthy fertile female albino rats of child bearing age & with normal oestrous cycle. Body weight between 150 to 200 grams. 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 200grams. Sterile male & female rats.

Anti implantation activity (by Choudary & Khanna Method)
It involves 6 stages.

- Collection of 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^{th} day of drug administration rats were subjected to laparotomy to observe for implantation

Sample size : n= 6 in each group

Drug schedule
Group I: Fine powder of Japakusuma 180 mg / 200 gm body weight of albino rats & Fine powder of shodhita Gairika 90 mg & Talisapatra (*Taxus baccata*) 90 mg total 180 mg / 200 gm body weight of albino rats with 2 ml of Propelyne glycol
Group II: Fine powder of Japakusuma 180 mg / 200 gm body weight of albino rats with 2 ml of Propelyne glycol
Group III: Fine powder of shodhita Gairika 90 mg & Talisapatra (*Taxus baccata*) 90 mg total 180 mg / 200 gm body weight of albino rats with 2 ml of Propelyne glycol

Outcome measures
Primary outcome

To compare the anti implantation activity of Talisapatra (*Taxus baccata*) + Gairika + Kanji bhavita Japakusuma (*Hibiscus rosa sinensis*), Kanji bhavita Japakusuma (*Hibiscus rosa sinensis*) Talisaapatra Gairika

Secondary outcome

- Mean birth weight of litters
- Survival of litters

OBSERVATION AND RESULTS

Table 1: Phytochemical constituents of aqueous extract of Talisapatra Gairika Japakusuma (*Hibiscus rosa sinensis*), Japakusuma & Talisapatra Gairika

Sl. No.	Organic Constituents	Talisapatra Gairika Japakusuma	Japakusuma	Talisapatra Gairika
1.	Alkaloids	-	-	-
2.	Carbohydrates	-	-	+
3.	Tannins	+	+	+
4.	Steroids	+	+	-
5.	Triterpenoids	-	-	+
6.	Saponins	+	+	+
7.	Flavonoids	+	+	+
8.	Carotenoids	-	-	-

Table 2: Anti implantation activity of all the Talisapatra Gairika Japakusuma, Japakusuma & Talisapatra Gairika (primary outcome measure): (n=06)

Group	Drugs	No. of Rats	Mean no. of implantations	% inhibition of implants
I	Talisapatra Gairika Japakusuma	6	0	100%
II	Japakusuma	6	0	100%
III	Talisapatra Gairika	6	0	100%

Table 3: Secondary outcome measures of Talisapatra Gairika Japakusuma, Japakusuma) & Gairika Talisapatra

Group	Drugs	% of rats delivered on full term	Mean no births	Mean weight of litters	Died within 2 days
I	Talisapatra Gairika Japakusuma	0%	0	0	0
II	Japakusuma	0%	0	0	0
III	Talisapatra Gairika	0%	0	0	0

DISCUSSION

In this anti implantation & Pregnancy interruption study, results of test sample were compared. Study was conducted in six stages. To assess contraceptive activity of test sample by the observation of anti implantation & pregnancy interruption activity in mature female albino rats. Anti implantation activity was conducted to assess contraceptive activity by following Choudary & Khanna method. Laparotomy was conducted on 10^{th} day after drug administration. Results of three groups were compared. In Group I, II& III implantations were not found, which indicates that all the test drugs have demonstrated anti implantation activity.

The anti implantation activity & pregnancy interruption activity might be postulated in the following ways based on the experimental & 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 estrogenic and anti progesteronic effect. Phytochemical analysis of Talisapatra (Taxus baccata) + Gairika & Japakusuma (Hibiscus rosa sinensis) (Kanji bhavita Japakusuma) has shown the presence of steroids, saponins, Flavonoids & tannins. Especially steroids & saponins are used as raw material for preparation of medically useful steroids & sex hormones like progesterone, oestradiol, & testosterone. Thus steroids, saponins might have contributed in the contraceptive activity of the drugs.

CONCLUSION

Significant anti implantation & pregnancy interruption activity was noted in all the three Groups indicating the contraceptive activity of the test drugs. Contraceptive activity may be due to the presence of Phytochemicals like Steroids, saponins, Flavonoids & Tannins.

Scope for further research

The promising results of this experimental study necessitate a well designed randomized clinical research before the test drug is recommended for clinical practice.

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