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# Punarna V

**TITLE: HORMONAL ASSAY OF VIRGARBHAKAR YOGA IN FEMALE ALBINO RATS W.S.R TO FERTILITY**

**MOHAR PAL MEENA, MUNESH, PROF.L.K.DWIVEDI**

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### HORMONAL ASSAY OF VIRGARBHAKAR YOGA IN FEMALE ALBINO RATS W.S.R TO FERTILITY

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#### ABSTRACT:

*Good health is considered as pre requisite to achieve the Purushartha Chatustaya (Dharma, Artha, Kamaand Moksha). Individual get these benefits when person have healthy life and will be able to give birth to healthy off spring. Ayurveda excellence and the Upanishad quotes that Putraishana is the primary desire of mankind. Infertility being major human problems shows its impact on emotional and social aspect. W.H.O states that 8-10% of couple experiences infertility problem worldwide. Organon Fertility Survey 15 million of Indian are suffering with infertility compared to U.S population. Survey of Infertility in India quotes that 1 out of 6 couple in India suffers from inability to procreate and about 7 % infertility is increased among Indian women. Infertility is the disorder which can be co-related with the Vandhyata. Many preparations are mentioned to manage the Vandhyata but among them Rasa Aushadhi's stands first. Present era being aware of toxicity of mineral drugs they won't prefer the medication prepared by these minerals. In this scenario Virgarbhakar Yoga which is specially indicated in infertility is studied with aim of standardizing the drug and to see the fertility and hormonal efficacy in female albino rats. Pharmaceutical, analytical and experimental study was the materials and methods and evidence of these claims that Virgarbhakar Yoga can be used safely to get fertility result.*

*Key words: Avurveda. Fertility. Vandhvata. VirgarbhakarYoga*

#### INTRODUCTION

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*Ayurveda* is the science of life which originated from the *Vedas*. It aims at achieving perfect health, through physical, mental and spiritual well-being. According to *Ayurveda* good

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health is considered as a prerequisite for achieving the goals of life i.e. *Dharma, Artha, Kama* and *Moksha*. A person gets these benefits by giving an offspring, which is the resort these qualities. Even in Upanishads *Putraishana* is mentioned as one of the primary desires of mankind. Infertility is one of the human problems having a social and emotional aspect. According to WHO estimates 8-10% of couple experience some form of infertility problem on world wide scale Given the hectic pace and stresses of modern day life , there is an increase in incidence of life style disorders, the incidence of hormonal imbalance is coming up as its consequences .*Vandhyata* is described by our *Acharyas* in a wide sense including *nidana* and *chikitsa*. Ayurvedic literature provides ample scope for effective and economic treatment.

Present study was undertaken to assess the effectiveness of *Virgarbhakar Yoga* on hormonal status of albino rats and to trace out mechanism by which drug is acting on reproductive system. *Virgarbhakar Yoga* is named after *Virgarbhakarikriya* mentioned in Ph.D thesis of *Dr.Lakshami Kant Dwivedi* entitled *Rudrayamala parade kalpa*

which had been quoted from the manuscript of *Rudrayamala Tantra* 84<sup>th</sup> *Patalajalasiddhikalpa*.The process is mentioned for the treatment of *vandhya*. Mineral content of the drug include the ingredients of *Chaturmukha Rasa* and herbs from *Charaka Samhita* and animal products from *Nayansukha*.

### Aims and objectives:

1. To standardize the herbomineral preparation: *Virgarbhakar Yoga*
2. To access the effect of the herbo-mineral formulation on infertility of female albino rats
3. To access the effect of *Virgarbhakar Yoga* on the hormonal status of albino rats
4. To trace out the mechanism by which drug is acting on the reproductive system.

### Materials and methods:

The herbomineral compound *Virgarbhakar Yoga* was prepared in the department of Rasa Shastra and Bhaishajya kalpana, N.I.A, Jaipur. Two samples of *Virgarbhakar Yoga* were prepared –*Virgarbhakar Yoga* 1 and *Virgarbhakar Yoga* 2 (eliminating mineral drugs). The ingredients of the

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compound along with the chemical analysis are given in the table 1 and 2.

**Table 1 Showing ingredients of *Virgarbhakar Yoga***

Sl.No.	Content	Sl.No.	Content
1	<i>Shuddha Parada</i>	10	<i>Ksheeravidari</i>
2	<i>Shuddha Gandhaka</i>	11	<i>Atibala</i>
3	<i>Abhraka Bhasma</i>	12	<i>Jivaka</i>
4	<i>Swarana Bhasma</i>	13	<i>Apamarga</i>
5	<i>Lauha Bhasma</i>	14	<i>Vata Shunga</i>
6	<i>Haritaki</i>	15	<i>Naga keshar</i>
7	<i>Mandukaparni</i>	16	<i>Ashwagandha</i>
8.	<i>Durva</i>	17	<i>Gorochana</i>
9	<i>Guduchi</i>		

**Table 2 Showing observations of AAS**

1	Au	1634ppm	6	Ca	5114ppm
2	Fe	3294 ppm	7	Al	306.5 ppm
3	Hg	3857 ppm	8	As	4.471 ppm
4	K	34070 ppm	9	Pb	9.029 ppm
5	Na	3195 ppm	10	Cd	.247 ppm

Study was conducted in three phases.

- Anti-fertility studies for assessing the anti-fertility effect of *Vidanga* extract.
- Fertility studies for assessing the effect of *Virgarbhakar Yoga*
- Study on immature rats to assess the effect of *Virgarbhakar Yoga* on reproductive maturity.

**Phase 1 study**

Albino rats of either sex 8-10 weeks weighing 70-80g were obtained

from animal house of Dept. of zoology Rajasthan University Jaipur. Total 8 male and 30 female albino rats were taken and kept in separate cages for 15 days to let them acclimatize with the environment. After fifteen days female rats were divided in to 2 Groups. GroupA n=9 (control) and Group B n=21(experimental).Group A was kept on normal diet and water. Group B were given alcoholic extract of *Vidanga* in a dose of 10mg/100g bow/rat/day in a single dose of 2ml having oil as vehicle for 30 days. After 30 days 3 animals each of groups were sacrificed.

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**Phase 2 study**

For second phase trial animals left in the first phase were taken. Group B animals were again divided in to 3 groups C, D, E. Group C was taken as infertile control group. Group D,E were given two different samples of *Virgarbhakar Yoga* 10mg/100g for 30 days for honey and water. After 30 days 3 animals each from A, D, E, C were sacrificed. Remaining animals of the four groups were left for mating with male rats. Female rats supposed to be pregnant after observing the vaginal mucous plug were kept in separate cages and observed regularly and autopsied on 21<sup>st</sup> day of gestational age when they did not deliver litters except group A females who delivered litters on 21<sup>st</sup>day.

**Phase 3 study**

For third phase of study 12 immature healthy female rats were divided in to 3 groups containing 4 rats each. The groups were named as F, G

and H. F and G were given samples of *Virgarbhakar Yoga* while H was kept as control. Biochemical parameters studied include Estrogen, Progesterone, FSH and LH. All animals were observed for morbidity and mortality twice daily. General clinical observations were made twice a day at the same time through out study Animals were sacrificed on 21<sup>st</sup> day and ovary and uterus were send for histopathological studies.

**Results**

The gross behavior of all animals remained normal throughout the study. There was increase in body weight in all groups which was statistically insignificant as compared to control group.

**Hormonal changes**

**Changes in phase 1:** The LH and Progesterone hormone in group B animals didn't show any significant changes while estrogen level was significantly increased.

**Table 3 Showing result of phase 1(antifertility) study in control group A and Vidanga extract treated group B**

Hormone Studies	Group A	Group B	t value	P value	Significance
	Mean ± S.E	Mean ± S.E			
LH	4.37±2.80	4.83±1.62	0.23	-	Insignificant
Estrogen	133.5±19.03	67.40±30.99	3.15	0.05	Significant
Progesterone	4.77±0.15	6.50± 4.07	0.74	-	Insignificant

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**Changes in phase 2:** On administration of *Virgarbhakar Yoga* 1 in group D the highly elevated Estrogen level come

down to the normal limits like that of control group, while level persist highly elevated in group C.

**Table 4 Showing result of phase 2(fertility) study**

Hormone Studies	Group A	Group D	Group E	Group C
	Mean ± S.E	Mean ± S.E	Mean ± S.E	Mean ± S.E
LH	0.1±0.0	0.63±0.61	15.57±24.64	0.40±0.44
Estrogen	86.73±11.72	73.73±11.88	83.38±14.22	13.4±19.4
Progesterone	19.33±3.11	16.10± 11.78	13.70±8.23	11.17±8.11

**Changes in phase 3:** The increase in LH hormone level was significant in F group treated with *Virgarbhakaryoga*

1.The group G showed increase in LH level which was also significant.

**Table 5 Showing result of phase 3 study**

Hormone Studies	GroupF Virgarbhakar 1	GroupG Virgarbhakar 2	GroupH ( Control )	t value F-H	t value G-H	p value F-H	P value F-H
	Mean ± S.E	Mean ± S.E	Mean ± S.E				
LH	5.12±1.78	4.52±2.02	0.95±1.57	3.51	2.79	0.02	0.05
Estrogen	31.35±4.81	29.23±20.6	33.95±11.5	0.42	0.40	-	-
Progesterone	1.25±0.59	2.88± 0.48	1.60±1.49	0.44	1.63	-	-

**Histopathological Changes:**

**Changes in phase 1**

The histopathological study showed a number of changes in ovary and uterus after *Vidanga* administration.

Ovarian changes.- The ovary of control group A showed normal structure having follicles of all sizes from nest of oocytes to large griffin follicles, theca external and internal, normal vascularity and normal corpus lutium cells. Ovary

of group B showed conspicuous follicular atresia, increased vascularity and necrosis of corpus lutium cells.

The atresia of oocyte is evident from following signs: viz contraction of chromatin material, wrinkling of nuclear membrane and presence of cells inside the oocyte were seen in large follicles, as atresia progressed further the collicles collapsed, leaving an irregular lumen or cavity filled with cells and fibers of connective tissue.

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The ovarian interstitial was also decreased in size.

Uterine changes- The uterus of Group A showed wide lumen lined up with columnar epithelial cells, 3 distinct layer of basal, myometrium and endometrium, normal endometrial glands with visible glandular epithelium having small lumen and outer periphery was smooth lined up with epithelial tissue. Group B show changes in uterine tissue as the vascularity was highly increased, the myometrium show noticeable shrinkage, the endometrial glands were undergoing hypotropic changes showing regression, the outer lining was disturbed and the lumen was narrow and slit like.

## Changes in phase 2

The histopathological study of ovary of group D and E showed that the changes were returning towards normal as graffian follicle were again seen in developing stages and corpus luteum having healthy cells as compared to group C in which changes of necrosis were persisting. The uterine

structure of group D and E showed decreased vascularity, normal endometrial glands with epithelial lining as compared to group C.

## Changes in phase 3

Study on immature rats showed that there were early changes in ovary and uterus of F and G as compared to control group H. Ovary showed graffian follicles in developing stage and presence of corpus leutium which were absent in group H. Groups C, D, E didn't delivered litters after mating so they were autopsied to see resorption on 22nd day. Absence of implantation indicated that none of them conceived. Study showed that though structural changes caused by *Vidanga* were returning to normal strategy after administering samples of *Virgarbhakar Yoga* but needed much longer time to let them become normal to conceive. Further study is needed to accomplish better results of prepared drugs.

## DISCUSSION

From time immemorial, fertility is an essential necessity and assumed overwhelming importance as evident from Vedic period describing

*Putraishana* as one of the three primary desires of mankind.

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In the AAS study potassium level was found higher than other which might have come from herbal parts. Gold, Iron and Mercury as the chief constituents showed their presence in AAS. Heavy metals were present in permissible limit.

**Table 6 Showing observations of  
AAS**

1	Hg	0.40%
2	Au	0.14%
3	Fe	0.42%
4	S	3.48%

In phase 1 study were alcohol. Extract of *Vidanga* were administered, the level of Estrogen hormone was significantly increased in *Vidanga* treated animals as compared to control the increase was further confirmed by changes seen in section of ovary and uterus. Changes in hormonal level of Progesterone and LH were insignificant. In phase 2 study, hormonal assay showed that elevated Estrogen level returned to normal. The ovary showed that changes were reversing as graffian follicles were

seen in developing stages. The changes were similar in *Virgarbhakar Yoga* 1 and 2 but were more advanced in *Virgarbhakar Yoga* 1 suggesting that it might be due to the presence of gold *Bhasma* and other minerals in *Virgarbhakar Yoga* 1. In phase three study there were significant increase in level of LH as compared to control group suggesting that the drug might be influencing the hypo pituitary axis for early release of gonado-tropins which further stimulate changes in gonads. Over all the study showed that the proposed drug might stimulate pituitary as well as gonads to secrete hormones essential for fertility in the normal range and thus enhancing fertility rates.

Proposed mechanism of action: The drug showed effect on hormonal level mainly on Estrogen. From Ayurvedic view we can say that the ingredients of the drug mainly possess *Madura* rasa and *vipaka*, *shitaveerya* and *vata pitta shamaka* properties and show *prajasthapaka* property.

### CONCLUSION

The study revealed that the *Virgarbhakar Yoga* can be safely used to enhance the activity of reproductive system in cases of unexplained

infertility. The trial drug is also beneficial in patients where Estrogen level is elevated as in cases of PCOD, endometriosis etc. However a



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large scale study is needed to accomplish better results.

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