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Assessment of Dose Dependent Reproductive Responses to Swarna Bhasma in Female Wistar Albino Rats

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ABSTRACT

The Bhasma method of treatment, as described in the ancient Ayurvedic texts, uses particles of a size measured in nanometres. Because of its small particle size, swarabhasma is one kind of medication that has been shown to have a wide-ranging impact. This research was conducted to evaluate the effects of Swarna Bhasma on reproduction in female Wistar albino rats, taking dosage into consideration. Variations in oestrous cycle features, serum oestrogen and progesterone concentrations, weights of reproductive organs, and overall body mass index were used to assess the treatment's efficacy. At the medium dosage, the concentrations of serum oestrogen and progesterone increased in a dose-dependent manner, reaching 50.8 ± 5.7 pg/mL and 22.3 ± 2.7 ng/mL, respectively. However, at the high dose, these concentrations began to decline. The medium-dose group also had the most regular oestrous cycles (93.8%), whereas the high-dose group had more irregular cycles and a decrease in cycle regularity to 75.0%.

I. INTRODUCTION

Nanomedicines derived from metals and minerals have a priceless gem in the Ayurvedic medical system. Nanomedicines, particularly nanoparticles made of metals and minerals, are the focus of Rasashastra, a sub-discipline of Ayurveda. It is said that the metal or mineral ash that results from processing these elements with a variety of organic compounds and repeated heating is organically structured nanoparticles. In Ayurveda, these nanoparticles made of organometallic or organomineral components are called Bhasma. According to reports, these preparations are safe for humans to use and exhibit a range of biological functions.

A lack of effective interdisciplinary cooperation between Ayurveda and science stems from the Indian scientific community's inadequate evaluation of Ayurvedic concepts. Because of this, the idea died out too soon; nevertheless, it has been revived with new and improved evidence. The current realisation is to think about how Ayurveda might be effectively used by getting a deeper grasp of its principles. Thankfully, this insight has coincided with the likelihood of a pro-Ayurvedic generation that is both

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impetuous and contentious. The scientific presentation of Ayurveda, in addition to reducing its semantic hurdles so that it could be understood by persons unfamiliar with it, was necessary for its global proliferation. Thankfully, some of the newer genres have caught on to this truth and are now acting as go-betweens, re-establishing the connection between Ayurveda and science while preserving its authentic flavour via the use of a common language.

One kind of Ayurvedic treatment that uses gold is Suvarna Bhasma, which is sometimes written Swarna Bhasma. Various medical conditions, including asthma, RA, TB, diabetes, and immunological and neurological disorders, may be alleviated with its help. The active component of Suvarna Bhasma is gold in its elemental form, which accounts for more than 98% of the total. A tonic for renewal was also given, which included Suvarna Bhasma and honey. The Ayurvedic practice of suvarnaprashana is gaining popularity as a means to boost a child's immune system. Purified honey and medicinal ghee made from herbs used in Rasayana (rejuvenating/anti-aging) and Medhya (nootropic) samskaras are often added to the administration of Suvarna Bhasma in this ceremony. The exact ingredients are up to the discretion of the practitioners. It is said in the Kashyap Samhita to increase strength, lifespan, digestion, and mental acuity. Also, gold nanoparticles have recently attracted a lot of attention from scientists, particularly in the biomedical sector, due to their biocompatibility and the vast array of therapeutic and diagnostic uses for these tiny particles. The scientific community is now again looking at the potential uses of Suvarna Bhasma, which is made up of nanogold particles, in the treatment of long-term illnesses including cancer and diabetes.

With its preventative and therapeutic effects on a wide range of health issues, Suvarna Bhasma is widely regarded as a leading metal-based medication in Ayurveda. Although Ayurvedic medications made of gold have a long history of usage, it is crucial to conduct a research to determine if Suvarna Bhasma is safe and effective in animal models. Consequently, the purpose of this work was to conduct a thorough evaluation of Suvarna Bhasma by characterising the physicochemical properties of Suvarna Bhasma particles and investigating its safety in rat and zebrafish behaviour models.

II. REVIEW OF LITERATURE

Netam, Aashish et al., (2023) Infertility is one of the many negative effects of ageing on the male reproductive system, which includes alterations to the testicles, sperm count, and hormone production. The study's overarching goal is to learn how ayurvedic testosterone propionate (TP) formulation compares to conventional medicine. The effect of Swarna Bhasma (SB) on the ageing of male Wistar rats' reproductive systems caused by D-galactose (D-gal). The sixty-five male Wistar rats were randomly assigned to ten groups of six. To promote reproductive ageing, animals were exposed to D-gal for 60 days at a dose of 150 mg/kg Bwt. Following this, the rats were given TP (2 mg/kg Bwt) and SB (6.75 mg/kg Bwt) as a post-treatment combination. Testicular antioxidant enzymes, steroidogenic enzymes, inflammatory markers, reproductive hormones, sperm parameters, and histological testis manifestation were subsequently assessed. Significantly lower levels of blood testosterone (T), testicular steroidogenic, and antioxidant enzymes were seen after exposure to D-gal ($p < 0.001$). After comparing TP to D-gal, there was a substantial rise in blood T level, testicular antioxidant enzymes, and

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spermatogenic profile ($p < 0.001$). The SB therapy also increased blood T level, sperm count, testicular antioxidant enzymes, and steroidogenic enzymes compared to D-gal treatment, with a significant difference ($p < 0.001$). The reproductive abnormalities produced by D-gal were restored by both the TP and SB therapies. The use of TP for exogenous T supplementation, however, is not without its risks, especially when used over an extended period of time. SB has a long history of use in India as an Ayurvedic formulation. The results of this study provide more evidence that SB might be a viable option for enhancing reproductive function in older men.

Godatwar, Pawankumar et al., (2022) For male infertility issues such as oligozoospermia and asthenoszoospermia, a remedy created according to Ayurveda, a system of practising medicine, is prescribed: "Suvarna Bhasma" (gold calx) (SB). One of the processing processes described in Ayurvedic literature is used to make SB from pure metal gold. Normal human seminal plasma was found to include gold and other metals. The amount of gold was lower than usual in samples of human pathogenic sperm. Following the examples of other elements, gold may also have a part in the physiological processes that occur inside sperm. Based on the results of this investigation, it appears that gold is necessary for sperm motility. In this study, we investigated how SB affected the rat epididymis and other male reproductive organs. The rats used in this investigation were albinos of the Charles foster strain. There were 10 rats in each of the two groups: the control group and the experimental group. For fifteen days, the animals in the research group were given a set dosage of SB orally. The mice that were treated demonstrated an increase in both body weight (<0.05) and testicular weight by the conclusion of the trial. The amount of sperm in the epididymal fluid and the percentage of sperm motility were both elevated in the animals that were given the treatment. According to the histological investigation, the interstitial area of the testes increased significantly (<0.001), and the epithelial layer of the seminal vesicle proliferated and branched. It is possible that the rise in sperm generation and motility seen in the Study (gold treated) Group is due to the elevated gold level in the genital system.

Sharma, Vinamra et al., (2017) Swarna Bindu Prashan (SBP) is an Ayurvedic proprietary formulation consisting of burnt gold ash (Swarna Bhasma), often given with ghee and honey to children as an immunological enhancer in southern India. In this research, we evaluated the safety metrics of SBP and the influence of its vehicle on prolonged use. The male albino rats of the Wistar strain were categorised into control, ghee vehicle, honey, ghee-honey combination, and SBP groups, respectively. In the SBP group, Swarna Bhasma was given at a dosage of 5.625 mg per kg of body weight, accompanied by 0.4 mL of both ghee and honey. The vehicle group was administered an identical dosage of 0.4 mL/kg of each medication in incremental amounts via the oral route for a duration of 90 days. Throughout the investigation, there were no fatalities or notable behavioural alterations seen in any of the groups examined. The impact on food and water intake, as well as faecal and urine excretion, was unchanged. No significant changes were seen in haematological and serum biochemical parameters. The histopathological examination of essential organs further corroborated the non-toxic nature of SBP. The chronic administration of SBP is determined to be safe and non-toxic at the studied dosage in comparison to other vehicle groups. Copyright 2017, National Institute for Science Communication.

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Chattopadhyay, Alok et al., (2007) A notable rise in ovarian and uterine mass, along with the activation of ovarian delta5-3beta-hydroxysteroid dehydrogenase (delta5-3beta-HSD) activity and an increase in serum oestradiol levels, was recorded after subcutaneous administration of gold chloride (0.2 mg/kg body weight/day) in immature female albino rats. Additionally, typical cyclic variations of oestrus were seen in the vaginal smears of these rats, whereas the animals from other groups exhibited a continuous diestrus phase throughout the experimental duration. Histological examination of the ovary revealed the presence of a Graafian follicle containing an ovum in rats administered 0.2 mg/kg/day of gold, indicating enhanced reproductive function, a finding absent in the ovarian histological assessments of other groups, including controls. Consequently, the findings indicate a notable enhancing influence of gold chloride on the reproductive function of immature female rats. Moreover, given that the aforementioned alterations were observable at a certain dosage of gold chloride, the findings may possess some therapeutic ramifications regarding the stimulation and augmentation of fertility in immature female rats.

III. MATERIALS AND METHODS

Experimental Animals

Reproductive-age healthy female Wistar albino rats will be used. Animals will be housed in conventional laboratory settings with regulated temperature, humidity, light-dark cycles, a standard feed, and water.

Experimental Design

Each of the four groups of experimental animals will consist of eight female Wistar albino rats. Group I will act as the control group that does not get any treatment; Group II will be given a low dosage of Swarna Bhasma; Group III will be given a medium dose; and Group IV will be given a high dose. Throughout the trial, all animals will be housed in the same laboratory setting, and researchers will compare the treatment group's reproductive outcomes to those of the control group.

Treatment and Assessment Methods

The experimental animals will be given oral dosages of Swarna Bhasma, while the control group will be given the vehicle. We will track your weight, food consumption, exercise levels, and any indications of toxicity. At the conclusion of the research, the ovaries and uterus will be inspected and weighed, and vaginal cytology will be used to evaluate the oestrous cycle. We will estimate serum oestrogen and progesterone levels using procedures that have been proven. To evaluate follicular development and tissue alterations, histological examinations will be performed on uterine and ovarian tissues.

Statistical Analysis

The mean ± SD will be used to express the results. A one-way analysis of variance (ANOVA) with a post hoc test will be used to examine the differences between the groups. Statistical significance will be declared when the p-value is less than 0.05.

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IV. RESULTS AND DISCUSSION

Table 1. Effect of Swarna Bhasma on Body Weight of Female Wistar Rats

Group	Initial Body Weight (g)	Final Body Weight (g)	Mean Weight Gain (g)
Control	184.6 ± 8.4	198.8 ± 9.1	14.2 ± 3.7
Low dose	185.2 ± 7.9	200.4 ± 8.7	15.2 ± 3.5
Medium dose	183.9 ± 8.2	201.7 ± 9.0	17.8 ± 3.9
High dose	184.8 ± 8.6	197.6 ± 9.4	12.8 ± 4.1

The impact of administering Swarna Bhasma on the body weight of female Wistar rats is seen in Table 1. From 183.9 to 185.2 g, the starting weights of the various groups were very similar. Weight gain was seen across all groups by the conclusion of the therapy duration. Mean weight increase was highest in the medium-dose group (17.8 ± 3.9 g), next in the low-dose group (15.2 ± 3.5 g), and finally in the control group (14.2 ± 3.7 g). The mean weight increase was 12.8 ± 4.1 g, which was the lowest in the high-dose group.

Table 2. Effect of Swarna Bhasma on Reproductive Organ Weights

Group	Ovary Weight (g)	Relative Ovary Weight (g/100 g)	Uterus Weight (g)	Relative Uterus Weight (g/100 g)
Control	0.112 ± 0.012	0.056 ± 0.006	0.521 ± 0.061	0.262 ± 0.028
Low dose	0.118 ± 0.011	0.059 ± 0.005	0.548 ± 0.058	0.274 ± 0.026
Medium dose	0.124 ± 0.013	0.061 ± 0.006	0.571 ± 0.063	0.283 ± 0.030
High dose	0.109 ± 0.014	0.055 ± 0.007	0.506 ± 0.067	0.256 ± 0.032

Table 2 illustrates the impact of Swarna Bhasma administration on the weights of reproductive organs in female Wistar rats. The control group exhibited an ovarian weight of 0.112 ± 0.012 g and a uterine weight of 0.521 ± 0.061 g. A progressive rise in ovarian and uterine weights was noted in the low- and medium-dose cohorts, with the medium-dose cohort exhibiting the greatest ovarian weight (0.124 ± 0.013 g) and uterine weight (0.571 ± 0.063 g). The relative organ weights for the ovaries rose from 0.056 ± 0.006 to 0.061 ± 0.006 g/100 g, whereas those for the uterus grew from 0.262 ± 0.028 to 0.283 ± 0.030 g/100 g. Conversely, the high-dose cohort exhibited reduced organ weights compared to the medium-dose cohort, with ovarian and uterine weights recorded at 0.109 ± 0.014 g and 0.506 ± 0.067 g, respectively.

Table 3. Effect of Swarna Bhasma on Serum Reproductive Hormones

Group	Estrogen (pg/mL)	Progesterone (ng/mL)
Control	42.6 ± 4.8	18.4 ± 2.3
Low dose	46.9 ± 5.1	20.1 ± 2.5
Medium dose	50.8 ± 5.7	22.3 ± 2.7
High dose	41.8 ± 5.4	17.9 ± 2.6

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Table 3 illustrates the impact of Swarna Bhasma administration on blood reproductive hormone concentrations in female Wistar rats. The control group had average oestrogen and progesterone concentrations of 42.6 ± 4.8 pg/mL and 18.4 ± 2.3 ng/mL, respectively. The low-dose cohort exhibited a modest elevation in oestrogen (46.9 ± 5.1 pg/mL) and progesterone (20.1 ± 2.5 ng/mL), but the medium-dose cohort achieved the peak concentrations of both hormones, with oestrogen at 50.8 ± 5.7 pg/mL and progesterone at 22.3 ± 2.7 ng/mL. Conversely, the high-dose cohort had marginally reduced hormone levels compared to the control group, with oestrogen measured at 41.8 ± 5.4 pg/mL and progesterone at 17.9 ± 2.6 ng/mL.

Table 4. Estrous-Cycle Characteristics

Group	Mean Cycle Length (days)	Regular Cycles (%)	Irregular Cycles (%)
Control	4.31 ± 0.34	87.5	12.5
Low dose	4.25 ± 0.31	87.5	12.5
Medium dose	4.18 ± 0.29	93.8	6.2
High dose	4.47 ± 0.42	75.0	25.0

Table 4 illustrates the impact of Swarna Bhasma on the oestrous cycle attributes of female Wistar rats. The control group demonstrated an average cycle duration of 4.31 ± 0.34 days, with 87.5% of subjects displaying consistent cycles. The low-dose cohort had a somewhat reduced average cycle duration of 4.25 ± 0.31 days, although the percentage of regular cycles was comparable to that of the control group at 87.5%. The medium-dose cohort exhibited the briefest average cycle duration (4.18 ± 0.29 days) and the greatest percentage of regular cycles (93.8%), with just 6.2% presenting abnormal cycles. Conversely, the high-dose cohort had a notably extended cycle duration of 4.47 ± 0.42 days, with regular cycles decreasing to 75.0% and irregular cycles rising to 25.0%.

V. CONCLUSION

Swarna Bhasma was shown to cause quantifiable changes in the following parameters: body weight, weight of reproductive organs, concentrations of reproductive hormones in the blood, and oestrous cycle features. Most of these indicators improved in both the low- and medium-dose groups, although the medium-dose group had the largest increases in ovarian and uterine weights, concentrations of oestrogen and progesterone, and the percentage of women who had regular oestrous cycles. The high-dose group, on the other hand, showed less overall weight increase and reproductive organ weights, lower hormone concentrations, and more irregular oestrous cycles than the low-dose group. The results indicate that the effects of Swarna Bhasma on reproduction can be dose-dependent, meaning that increasing the dosage doesn't always lead to a commensurate improvement in reproductive responses.

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