ANTI FERTILITY AND ANTI IMPLANTATION EFFECT OF GREEN TEA AQUEOUS EXTRACT ON FEMALE WISTAR RATS
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ABSTRACT
Green Tea is obtained from the leaves of Camellia Sinensis plant. The green Tea comprises of caffeine as a major constituent. A lot has been listed out on the health benefits of drinking Green Tea. The plant belongs to family Theaceae. The aqueous extract (200mg/kg) was orally given to female wistar rats for fifteen days in treated group and saline solution to control group. We maintained five groups each of control and treated so as to get a clear picture and cumulative result of our research. The male rats were kept in each cage in ratio of 2:1 (Male: Female) from day 2-4 and then withdrawn. When the implantation and fertility studies were carried out a decrease in both the activities (Implantation and Fertility) was reported in case of treated group. The parameters of weight was also studied which reflected a increase in case of control group as compared to treated group. When the results were calculated and observed a decrease in implantation and fertility was shown by treated female rats which were on aqueous green tea dosing. The previous studies have been conducted on male wistar rats which proved a decrease in size of gonads thus helping us to aim at the female wistar rats in our research. The control groups showed a total of 16 implants as compared to the treated groups which collectively showed only 9 implants. This made our observation clear that both the fertility and implantation is affected under regular dosing of Green Tea aqueous extract.

KEYWORDS: Estrous cycle, Fertility, implantation, Gestation period, GTE

INTRODUCTION
Tea is a popular beverage which is commonly used all over world. Tea is commonly used in Asian countries like China and Japan which contribute for about 20% of the consumption of tea worldwide.1 Commercial cultivation of green tea takes place in Asia, Africa and South America”. Green tea is derived from Camellia Sinensis a plant of Theaceae family. Some researchers have shown a hint of beneficiary uses of green tea extract in treatment of cancer, although the evidences are yet to be shown.3,5 Green Tea is a treasure from nature to the making as is consumed majorly by the population.6

Many effects of green tea have been reported , consumption of green tea reduces risk of cardiovascular diseases, cancer, increases neuroprotective activity, lowers cholesterol levels, increases antibacterial activity, anti viral activity, lowers blood glucose levels, anti oxidant activity. However some side effects have been reported when green tea is consumed regularly and at regular intervals8 The researches have proven that black tea consumption reduces heart diseases and stroke risks when consumed three or more cups regularly.8-12

As the cultivation of tea requires humid climate provinces like southern part of China mainland, Northern India slopes, Sri Lanka, Tibet are ideal growing area of tea.

Objective
The objective of this research is to determine anti fertility and anti implantation effect of green tea on female albino wistar rat using aqueous infusion of green tea dosing orally.

MATERIAL AND METHODS
Investigated Samples: Investigated samples were aqueous (infusion) extracts of green tea (Camellia Sinensis ). The source of drug was from the company GAIA green tea bags. The aqueous extract was prepared after taking 2 grams of green tea from tea bag.

Rats: Ten female albino rats, each weighing 35-100 g. were taken from animal house of PSIT, Kanpur approved by the CPCSEA. The CPCSEA number is 1273/ ac/ 09/CPCSEA. Each rat was kept in a separate cage and monitored for a complete duration of fifteen days.

Preparation of Green tea extract (GTE)
Green tea was collected from market .Composition of green tea was epicatechin (EC) 1.55%, epigallo catechin gallate (EGCG) 9.00% as specified by the company GAIA on their product description. Then we weighed 2 gm of green tea packed it in tea bag, 100 ml water taken in a beaker and subjected to heating plate to make it lukewarm . Infused tea bag in lukewarm water was kept for 10 minutes and then the mixture was shaken occasionally .After 10 minutes tea bags are removed and pressed for complete extraction and the green tea aqueous extract was prepared by infusion method daily for dosing . A dose of 200mg/kg of green tea was given orally to treated group rats. According to OECD guidelines a drug of 1.2 ml was administered daily to an animal of 120 gm and respectively all the groups were treated the same way.

Methodology
Two groups were maintained of adult female albino wistar rats weighing around 100 gm. Then all rats were kept in separate cages. Now we examined estrous cycle stage by taking vaginal smear at noon for five consecutive days. Then 5 rats for control
The second dose of test drug was given on the third day. The dosing was repeated on the third day. Same procure was repeated for the control group in which saline was orally administered regularly. After taking the vaginal smear the male rat are removed on the 2 day. From 4th and 7th day test drug is given in the morning. Control rats are given the vehicle (saline) only. The test drug (green tea aqueous extract) was given daily for duration of fifteen days to test group. Now the number of implants was counted in both groups when the rats were sacrificed on fifteenth day.

**RESULT**

The weight of wistar rats was monitored daily in both the control and treated groups and it was found that the weight in treated group increased moderately as compared to the control group where there was tremendous increase in weight.
Figure 5: Gradual increase in weight in case of Treated group and rapid increase of Weight in case of Control group

Figure 6: Moderate increase in weight in Treated group animals

Figure 7: Rapid increase in weight in Control group animals
We have a large number of research papers which say that a low body weight or loss of weight is not only a parameter of reproductive capacity of reproduction. It is well established that the implantation index directly correlates to the number of corpora lutea and indicates blastocyst implantation in the endometrium as well as the normal capacity of reproduction.

When the data of animal weight was analyzed a fast increase in weight was observed in case of control group animals. The treated groups were showing a comparatively little increase in weight. But as the weight is not only a parameter of judgment so we studied the number of implants in the female rat. The average numbers of the implantation sites were found to be reduced in treated animals as compared to the one in control groups. The number of pregnant rats out of total 5 control animals was found to be 4. The percentage of fertility was 80% while 3 rats were found pregnant out of 5 in treated group. The percentage fertility was 60%. As per the research conducted 20% decrease in fertility was found. When the control group was given saline drug and treated group given aqueous saline extract 43.75% decrease in implantation was found in the research. Oral aqueous extract of green tea to female rats resulted in a significant decrease (20%) in the numbers of implantation sites and live births when extract was daily administered to wistar female rats. The screening was carried out using anti fertility methodology. The observations reflected that green tea has induced anti fertility and anti implantation effects. Previous researches carried out on male wistar rats have shown a decrease in size of male gonads with reference to the research we carried the research further. The female rats have also shown a decrease in fertility and implantation.

We have a large number of research papers which say that Green tea causes a decrease in weight of humans as well as mice which attracts most women but the extract causing decreased Implantation and fertility might capture the eyes of people therefore breaking continuous consumption of Green Tea.

**DISCUSSION**

A low body weight or less gain in body weight suggests a few or nil implantations. It is well established that the implantation index directly correlates to the number of corpora lutea and indicates blastocyst implantation in the endometrium as well as the normal capacity of reproduction.

The implantation rate was reduced by 43.75% in case of treated animals when compared to the control group. Thus green tea extract treated animals were showing a rapid decrease in implantation rate as compared to the saline dosed animals.

The fertility rate is calculated as:

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\text{Fertility rate} = \frac{\text{Number of implants in control group} - \text{Number of implants in treated group}}{\text{Number of implants in control group}} \times 100
\]

CONCLUSION

The research carried out proves that there is a decrease in fertility and implantation rates in female albino rats when green tea extract was administered continuously for duration of fifteen days. Our research further points out that decrease in implantation and fertility further promotes research work which involves the determination of doses which is causing anti fertility and anti implantation activities. Thus we can say that regular consumption of green tea can decrease fertility rates in rats which hint that humans might also be affected due to regular consumption of green tea.

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