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# Evaluation of Fertility and Embryo Implantation in Rats After the Oral Administration of *Salvia officinalis* (Sage) Extract

## Evaluación de la Fertilidad y la Implantación de Embriones en Ratas Después de la Administración Oral de Extracto de Salvia officinalis (Salvia)

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**ABDRABOU, M. A.; ALHUMAIDI ALOTAIBI, M.; EL NAGAB, N. A. A. & AL-OTAIBIC, A. M.** Evaluation of fertility and embryo implantation in rats after the oral administration of *Salvia officinalis* (sage) extract. *Int. J. Morphol., 40(5)*:1404-1414, 2022.

**SUMMARY:** In Saudi Arabia, it is widely believed that women with reproductive problems can use the extract of the sage plant as a tea drink. This study was conducted to investigate the effects of this herb on the fertility of female rats and embryo implantation. Forty-eight Wistar virgin female rats were divided into four groups at random, with 12 rats in each group. The control group received distilled water orally. The three treatment groups received different concentrations of sage extract: 15, 60, or 100 mg/kg for 14 days before mating, then mated with a male and sacrificed on the 7th day of gestation, the uterine horns removed, and photographed. The total body weight of mothers, weight of uteri and ovaries and number of fetuses were determined. Ovarian and uteri tissues were cut into 5 µ sections and stained with hematoxylin and eosin. Serum FSH, LH were determined by the ELISA method. The present study showed that low dose of sage (15 mg/kg) have no effects on serum concentration levels of FSH and LH hormones, also has no effect on the number of growing follicles. The present study showed a significant differences (P≤0.05) in body weight, ovary and uterus weight in the groups treated with high doses of *Salvia officinalis* as compared to control group. Also a significant differences (P≤0.05) found in FSH, LH hormones. Histological study showed overall histomorphological structural configurations including growing and matured graafian follicular countable changes, besides a number of corpora lutea and regressed follicles in the treated groups with high doses of *Salvia officinalis* as compared to control group. The researchers concluded that the extract of the sage plant with high doses can stimulate the growth graafian follicles and improve fertility in female rats.

**KEY WORDS: *Salvia officinalis*; Follicle Stimulating Hormone; Luteinizing Hormone; Fertility Embryo implantation.**

## INTRODUCTION

Infertility is defined as a failure to get pregnant after a year of sexual contact. About 85 % of couples experience this issue. However, there are several known causes, ovulatory dysfunction, tubal disease (Carson & Kallen, 2021), abnormal uterine function (Mascarenhas *et al*., 2012; Lee *et al*., 2020).

According to the existing global data, roughly 25 % of couples may have primary or secondary infertility due to changes in hormone levels at some point throughout their reproductive lives (Burtis *et al*., 2008). Reproductive hormones have well-established roles in the reproductive process (Orieke *et al*., 2019).

In order to successfully carry out embryo implantation, it involves communicating between the receptive endometrium and the free-floating blastocyst (Mishra *et al*., 2021). The blastocyst anchors are then placed on the basal and stromal extracellular matrix. The invasion of the embryo into the uterus is the final phase of the procedure. Various factors, such as the presence of certain hormones and the cytokines, growth factors, adhesion molecules and receptive endometrium can affect the success of embryo implantation (Singh *et al*., 2012).

Because produced pharmaceuticals used to treat infertility are so expensive and have many side effects, many

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people turn to alternative approaches such as herbal therapy. *Salvia officinalis* is one of the herbs that has traditionally been used to treat infertility.

Medicinal plants have been in use for millenniums by the human race as a potential source of natural first aids against different diseases (Mehta, 2012). *Salvia officinalis* L.(sage) is a perennial round bush in the Labiatae/Lamiaceae family. Salvia has almost 900 species and is the largest genus in this family. The species of the sage plant that is common and native to the Mediterranean and the Middle East (Mossi *et al*., 2011). Therefore, this herb, called "Maramia" in Saudi Arabia, is widely sold and consumed as Maramia tea. Maramia tea is commonly consumed in Saudi Arabia.

It is regarded as the queen of herbs because it can be used in various cooking and medicinal preparations (Khan & Abourashed, 2011). Various studies have been conducted on the various biological effects of *Salvia officinalis*. These studies revealed its properties as anti-inflammatory, antimicrobials, and hypoglycemics. These studies revealed various drug-related activities, such as anti-inflammatory, antimicrobial, anticancer, antioxidant, hypoglycemic and hypolipidemic (Ghorbani & Esmaeilizadeh, 2017; Mendes *et al*., 2020). Various components of salvia are also studied such as fatty acids, carbohydrate, alkaloids, glycosidic products (flavonoid, saponins, cardiac glycosides), phenolic compound, polyacetylene, steroids, terpenes/ terpenoids (monoterpenoids, diterpenoids, triterpenoids) and waxes (Badiee *et al*., 2012).

*Salvia officinalis* consists of phytoestrogen steroids and isoflavonoids (Vitale *et al*., 2013). It has been known that the phytoestrogens in *Salvia officinalis* can help decrease the signs of pregnancy and recover infertility in women (Boroujeni & Gholami, 2017). Extract organized from this plant has been used for several therapeutic and pharmaceutical purposes (Russo *et al*., 2013; Pedro *et al*., 2016). In 2012, a study revealed that the clinical effects of *Salvia officinalis* were observed in postmenopausal women (Ciotta *et al*., 2012). In many ethnopharmacological reports, S. Officinalis has been described (Andrade *et al*., 2018) and suggested numerous gynecological sicknesses (Li *et al*., 2013; Alrezaki *et al*., 2021).

## MATERIAL AND METHOD

**Preparation of *Salvia officinalis* extract.** Dried *Salvia officinalis* was purchased from an herbal market in Sakaka city, Saudi Arabia. The water extract of this plant was obtained by boiling it for 5 min (4g/500 ml). It was then cooled down before being filtered. This extract was transmitted into a sterile container and kept in a refrigerator (Bisher *et al*., 2021).

**Experimental animals.** Forty-eight virgin female rats of the Albino species were used for this study, weighing 180 to 200 g. The female rats were kept in a climate-controlled environment and were exposed to natural light for 12 h a day. The animals were kept in suitable cages and were subjected to standard laboratory processes at temperature 25±2 ºC.

Their diet consisted of a normal pellet diet and water. They took one week to become comfortable with their new environment before the beginning of experimentation. Strictly adhering to international guidelines for the care and use of laboratory animals was done in this experiment. In addition, animals fasted for 12 h before the starting of the experiment. The current work was completed in the animal house and Pharmacognosy and medicinal plants laboratory, Faculty of Pharmacy, Cairo University, Egypt, from May to August 2021.

**Groups.** The female rats were separated into four groups (the control and 3 treated groups), each group contained 12 female rat. The control group (GIV) received saline water for 14 days daily. The three experimental groups (GI, GII, GIII) were given the S. officinalis extract at a doses of 15 mg/kg, 60 mg/kg, and 100 mg/kg respectively every day for fourteen day before mating, then mated with the fertile male and all groups dissected on the 7th day of pregnancy (embryo implantation). The extract was administered orally.

**Mating.** The females in the pro-estrous and estrous stages were housed together with productive male (2:1) under the controlled environmental condition of temperature (25 ± 2 ºC), humidity (60 ± 20 %). The following day, the females with positive vaginal smears or plug were considered pregnant. The day of presence of sperms in vaginal smears was considered the zero day of gestation (Abd Rabou & Alotaibi, 2021).

**Morphological Observations.** After the experiments were concluded, the pregnant rats were sacrificed and their uterine horns were removed and photographed.

**Quantitative Observations.** The number of embryos in the uteri were determined. The total body weight of pregnant rats, ovaries and uteri were also calculated using electronic balance.

**Hormone measurements.** Serum LH, FSH were measured by the ELISA technique using a kit purchased from Ray Biotech, Inc., USA (Zheng *et al*., 2018).

**Histopathological findings.** Rats of each group were weighed and sacrificed post fourteen day of the treatment and on the 7 day of pregnancy. The ovaries and uteri were fixed in neutral buffer formalin for twenty four hours. Five mm-thick sections of tissue were cut and stained with hematoxylin and eosin.

**Statistical analyses.** The data collected was analyzed using the statistical technique SPSS 22. Anova, a statistical test, was used to determine the statistical significance between groups of the results with a value of $p < 0.05$.

## RESULTS

**Morphological, quantities and hormonal studies.** On day 7 of pregnancy and post fourteen days of oral administration of *Salvia officinalis* extract, the uteri contained five and six embryos in the control (GIV) and GI group respectively, but contained ten and eleven embryos in the GII and GIII groups (Fig. 1).

Table I. Effect of different doses of *Salvia officinalis* extract on the number of fetuses on day 7 of pregnancy.

| Group | Number of fetuses (M ± SD) | P value |
|---|---|---|
| GI group (15 mg/kg) | 6.66 ±0.577 | 0.438 |
| GII group ( 60 mg/kg) | 8.66 ±1.52 | 0.011** |
| GIII group (100 mg/kg) | 10.33 ±0.57 | 0.001** |
| Control group (GIV) | 6.00 ± 1.00 | - |

M ± SD (mean ± standard deviation), ** refer to significant difference (P value ≤0.05) compared to the control group

Tables I and II showed the effect of different doses of *Salvia officinalis* on the number of embryos revealed that both high doses were significantly different compared to control set, but insignificantly different in the number of fetuses was detected in the group of the low dose. The total body weights of mothers and reproductive organs weight revealed a significant difference in total body weights, uteri weight of GII & GIII groups and ovary weight of GII group, but insignificant difference was detected in the total body weight and reproductive organs of GI group. The results of Table III showed that the GII&GIII groups had a significant difference in the levels of the hormones FSH and LH compared to the control group, while no any significant difference was detected in GI group.

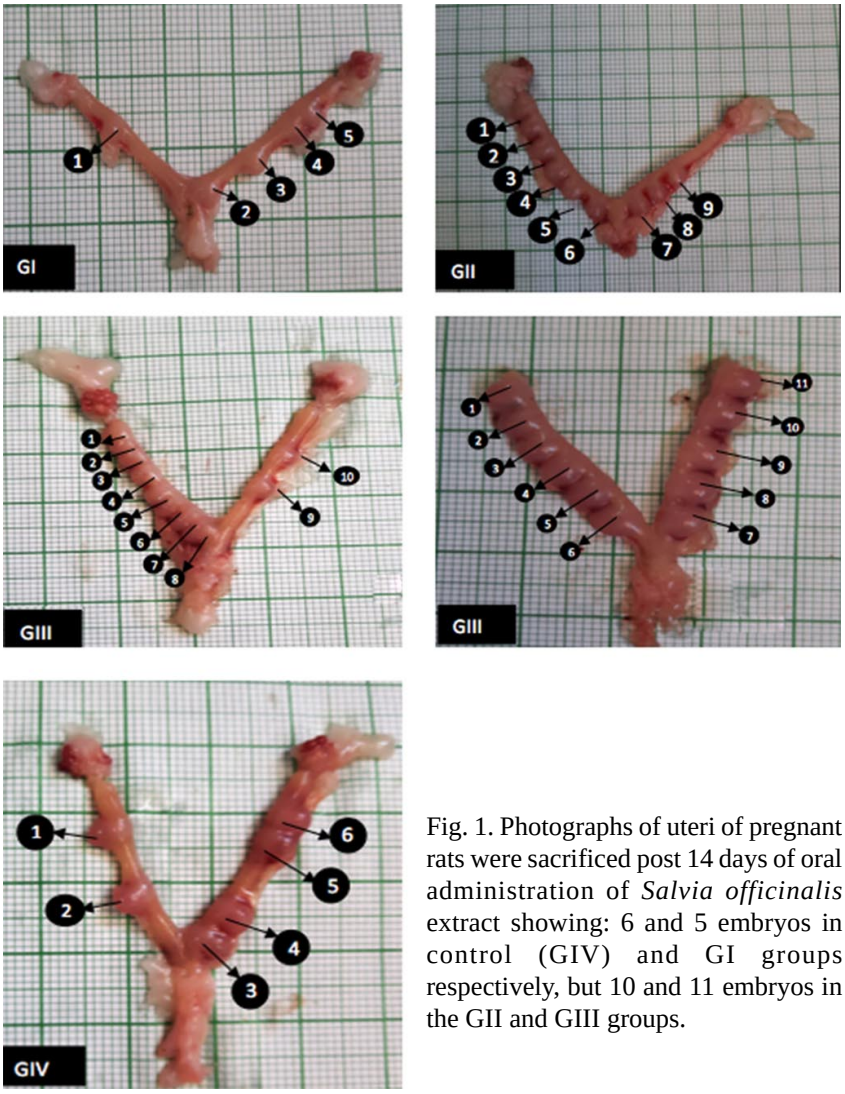


Fig. 1. Photographs of uteri of pregnant rats were sacrificed post 14 days of oral administration of *Salvia officinalis* extract showing: 6 and 5 embryos in control (GIV) and GI groups respectively, but 10 and 11 embryos in the GII and GIII groups.

**Histopathological Findings.** Investigation of serial sections from uterine implanted sites around the 7 th day of gestation revealed a ball of undifferentiated cells attached and polarly fixed to the maternal endometrium (The blastocyst) which specified into two layers. The outer portion of the trophoblast is composed of the trophectoderm which surrounds the inner cell mass (ICM), which consists of undifferentiated embryonic stem cells. Trophoblast is a type of cell that contributes only to the formation of the placenta. These cells are also responsible for producing hormones needed by the mother to recognize her pregnancy.

Table II. Effect of different doses of *Salvia officinalis* extract on total body weight and reproductive organs of pregnant rats.

| Group | Total body weight (g) (M ± SD) | P value | Ovary weight (g) (M ± SD) | P value | Uterus weight (g) (M ± SD) | P value |
|---|---|---|---|---|---|---|
| GI group (15 mg/kg) | 183.75±7.65 | 0.719 | 0.071±0.017 | 0.917 | 0.856±0.011 | 0.078 |
| GII group (60 mg/kg) | 193.83±9.68** | 0.013 | 0.145±0.037 | 0.382 | 1. 03±0.010 | 0.001** |
| GIII group (100 mg/kg) | 196.33±5. 49** | 0.002 | 0.285±0.324** | 0.028 | 1.137±0.015 | 0.000** |
| Control group (GIV) | 182.33±8.86 | - | 0.061±0.016 | - | 0.693± 0.127 | - |

M ± SD (mean ± standard deviation), ** refer to significant difference (P value ≤0.05) compared to the control group.

Table III. Effect of different doses of *Salvia officinalis* extract on reproductive hormones of pregnant rats post 14 day of administration.

| Group | Follicle Stimulating Hormone FSH (ng/ml) | P value | Luteinising Hormone LH (ng/ml) | P value |
|---|---|---|---|---|
| GI group (15 mg/kg) | 0.85±0.104 | 0.637 | 0.346±0.023 | 0.634 |
| GII group (60 mg/kg) | 1.13±0.12** | 0.015 | 0.853±0.135** | 0.000 |
| GIII group (100 mg/kg) | 1.29±0.205** | 0.002 | 0.993±0.015** | 0.000 |
| Control group (GIV) | 0.797±0.068 | - | 0.313±0.09 | - |

M ± SD (mean ± standard deviation), ** refer to significant difference (P value ≤0.05) compared to the control group.

Blastocysts from control from control and different treatment groups showed nearly the same morphogenesis and cytological and cytological properties of the primitive embryonal inner cell mass (ICM) and its derivatives including trophoblast cells that overlie the ICM (polar trophectoderm) which gives rise to fetal and syncyteotrophoblasts. ICM gives rise to all of the embryo proper as well as to mesodermal and endodermal components of the fetal placental structures and later on the fetal membranes and umbilical cord. Variability in the cytological properties of properties of the primitive embryonic structures at this stage of morphogenesis is very difficult among different among different experimental groups in concern with the aspects of stem cells proliferative capacities proliferative ,capacities, differentiability and physiologic apoptosis (Figs. 2 and 3).

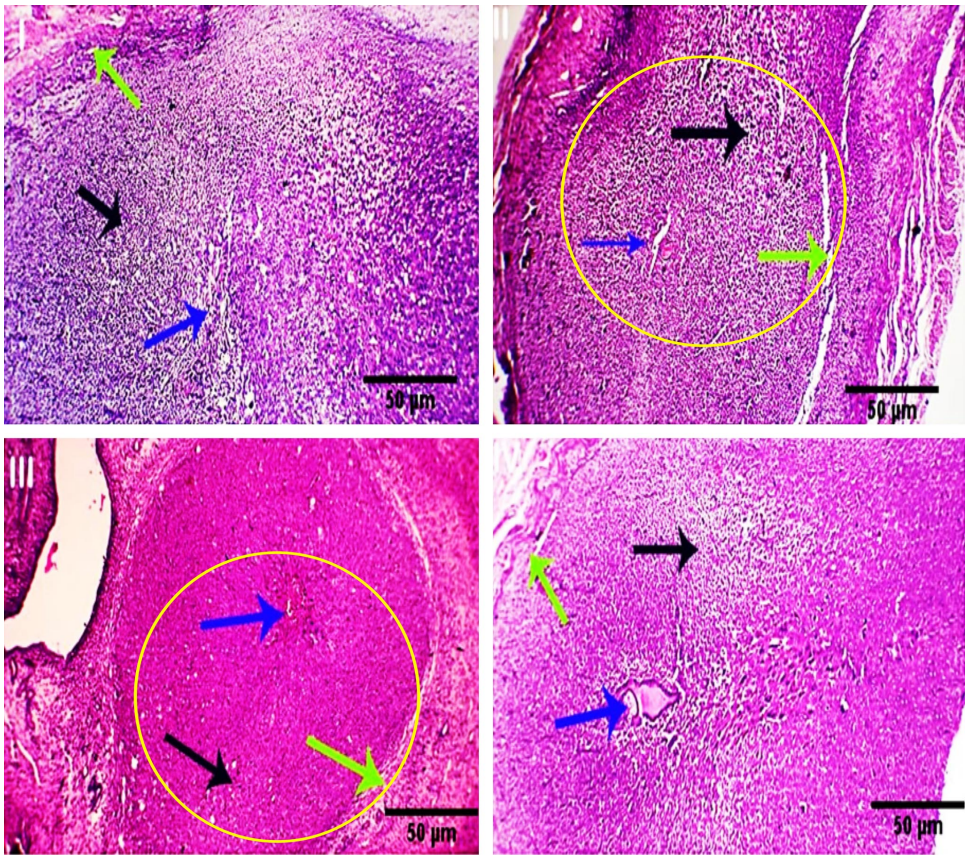


Fig. 2. Photomicrographs from Blastocysts of control rats (G IV) and different treatment groups ( G I G II and G III) showing normal process of primitive embryonal stem cell differentiation including the inner cell mass (ICM) (yellow circle), which consists of undifferentiated embryonic stem cells (the embryonic ectoderm). Trophoblast cells that overlie the ICM (polar trophectoderm) continue to proliferate, and give rise to fetal and syncyteotrophoblasts (black arrows). ICM give rise to all of the embryo properas well as to mesodermal and endodermal components of the placenta (green and blue arrows). A primitive endoderm is seen in the center of the blastocyst and appeared to be emerged from the ICM (Blue arrows). Scale bars 50 µm

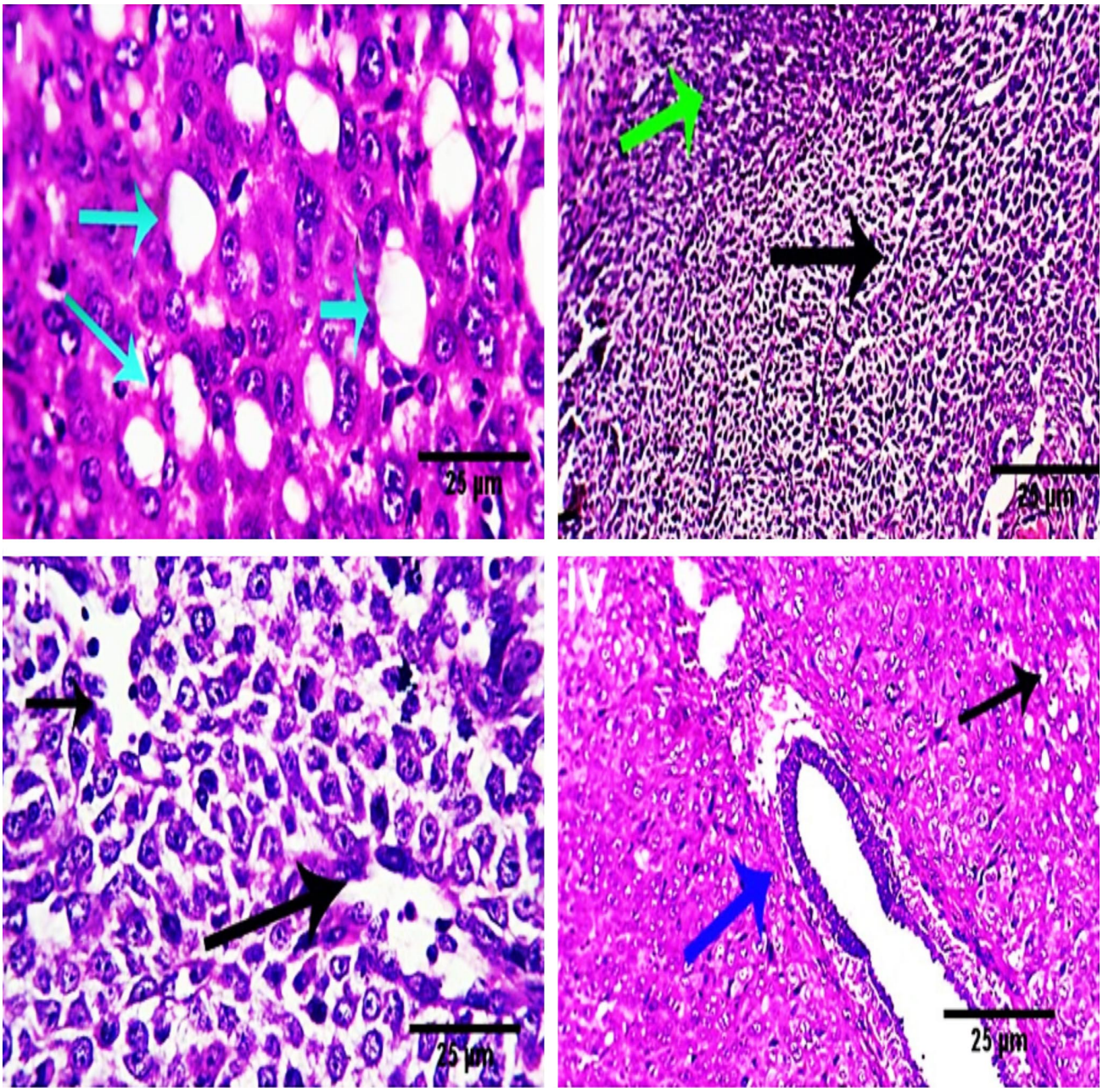


Fig. 3. Photomicrographs from Blastocysts of control rats (G IV) and different treatment groups (G I, G II and G III) showing normal process of primitive embryonal stem cell differentiation including the inner cell mass (ICM) (black arrows). Trophoblast cells that overlie the ICM (polar trophectoderm) continue to proliferate, and give rise to fetal and syncyteotrophoblasts (black arrows). A primitive mesodermal with new fetal capillary angiogenesis (green and light blue arrows) and endodermal (blue arrow) components of the placenta and fetal membranes appears to be emerged from the ICM. (Black arrows) .Scale bars 25 µm.

Sections from resorbed fetuses showed spontaneous resorption. The resorption was triggered by embryonic apoptosis, which is a process that occurs when a cell dies. The early resorptions were caused by motherly hemorrhage and a emphasis of motherly neutrophils (Fig. 4).

Examined sections from ovarian tissues of control pregnant rats and different treated groups demonstrated overall histomorphological structural configurations including growing matured Graafian follicular countable changes, changes, besides a number of corpora lutea and regressed

follicles. Histologically growing and matured Graafian follicles were represented by centrally or paracentrally located oocytes surrounded by granulosa cells which seen radially oriented (zona radiata) and peripherally arranged with formation of large antral space filled with eosinophilic materials saturated with estrogen sex hormone in fully matured follicles. The oocytes were either surrounded by solid sheet of granulosa cells (secondary and tertiary growing follicles or multiple small pre-antral spaces) in young growing follicles. Corpora Lutei were seen and represented by large number of fat-enriched progesterone secreting polygonal cells. Regressed follicles were also detected and showed as a group of nonfunctional

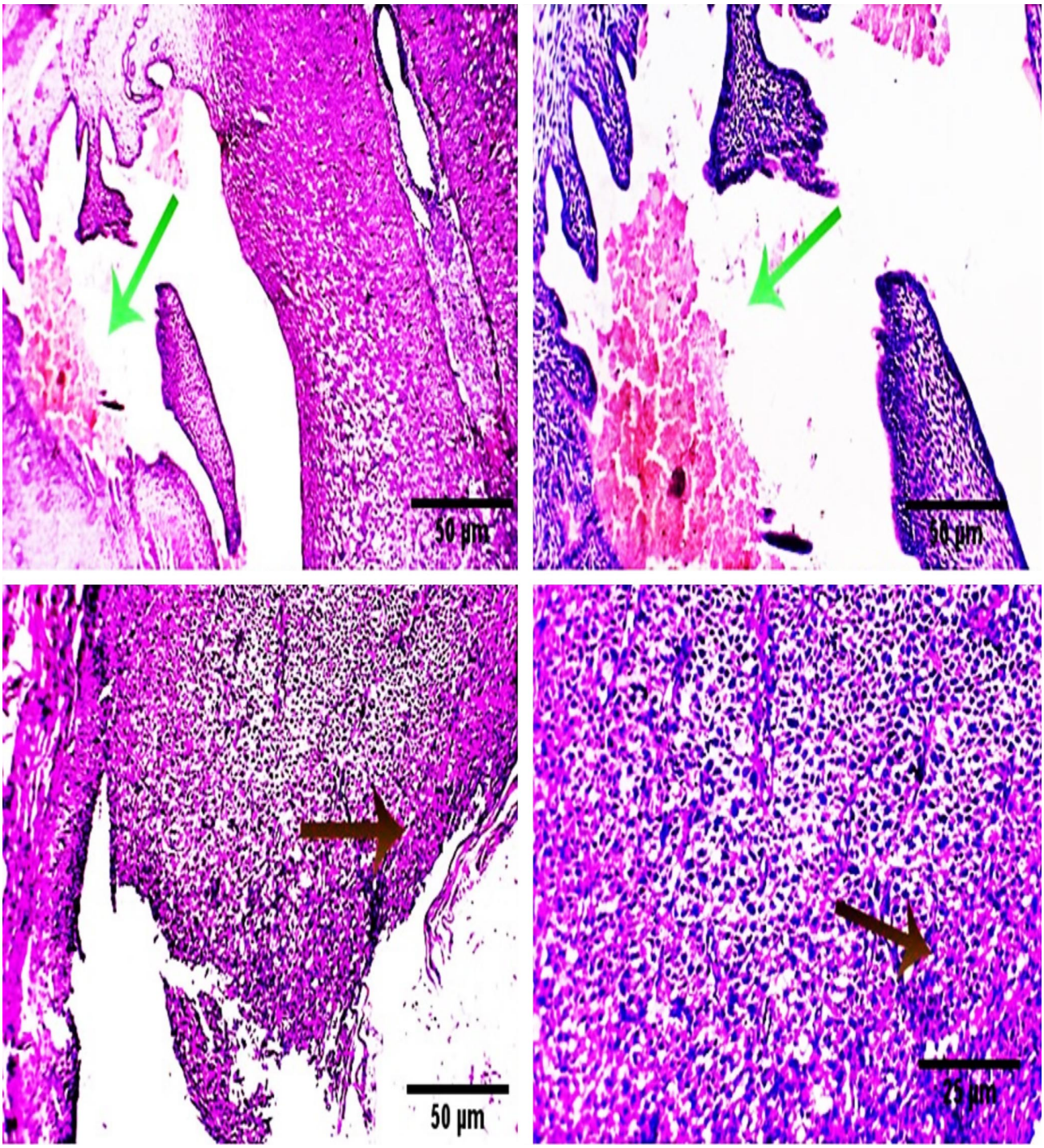


Fig. 4. Photomicrograph from Blastocyst of control rats showing the processes of fetal resorption which is represented by embryonic apoptosis (brown arrows). In the early resorptions detected at day 7, the embryo proper appears replaced by maternal hemorrhage and a suppurate focus of maternal neutrophils ( green arrows) .Scale bars 25-50 µm.

aggregates of granulosa cells with complete missing or degenerated oocytes. The numbers of young growing and matured follicles , corpora lutea and regressed follicles in different treated and control groups were calculated as follow : (7-9 , 1 and zero) , ( 13-15 , 1, 1 ) , ( 10-12 , 1, 1 ) , (8-10 , 1, 1) respectively (Figs. 5 and 6).

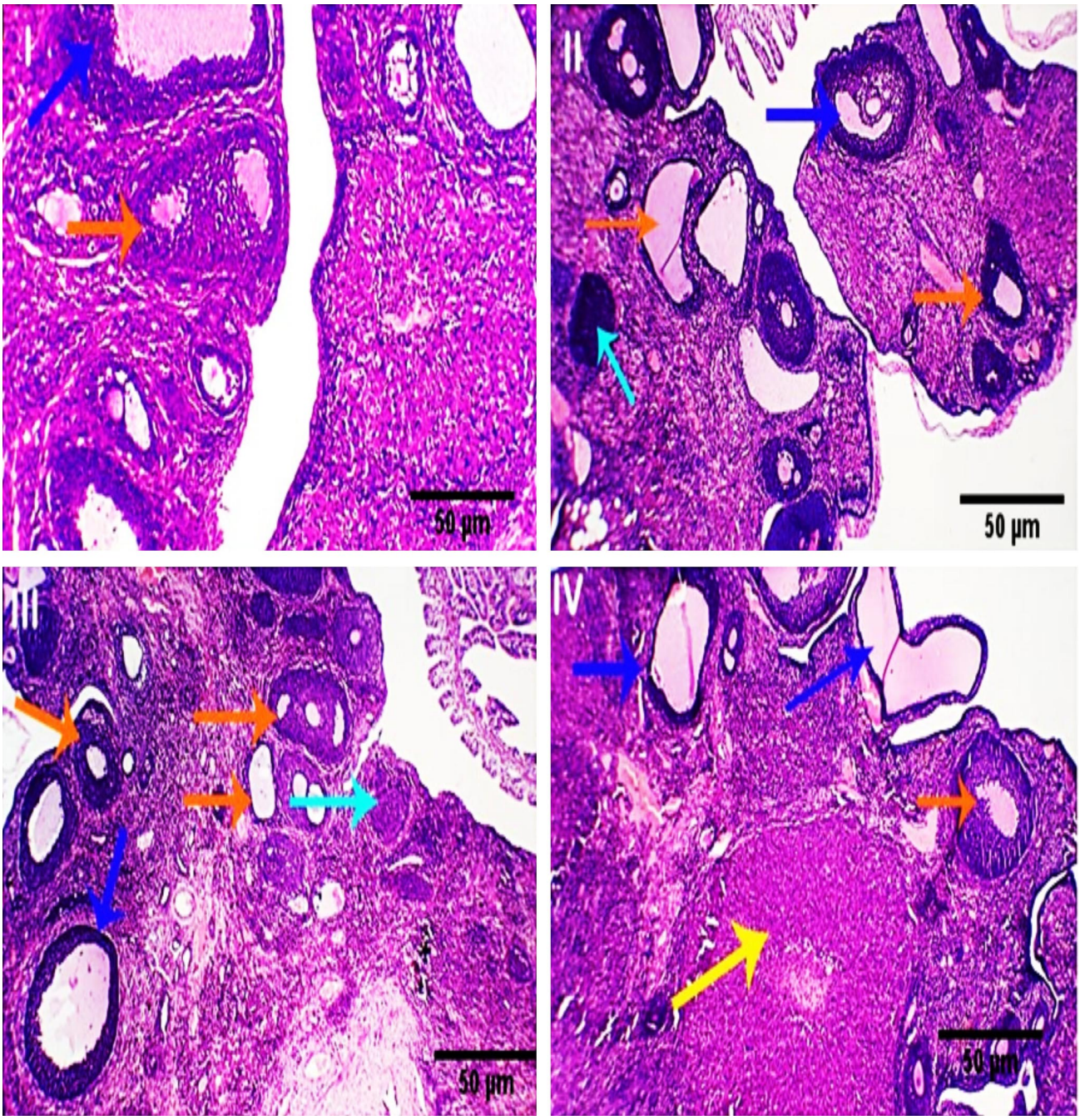


Fig. 5. Photomicrograph from ovaries of control pregnant rats ( G IV) and different treatment groups( G I, G II and G III) showing growing and matured Graafian follicles which are represented by centrally or paracentrally located oocytes surrounded by granulosa cells which seen radially oriented( zona radiata) and peripherally arranged with formation of large antral space filled with eosinophilic materials saturated with estrogen sex hormone in fully matured follicles. The oocytes were either surrounded by solid sheet of granulosa cells (secondary and tertiary growing follicles or multiple small pre-antral spaces) in young growing follicles (blue and orange arrows). Corpora Lutei are seen and represented by large number of fat-enriched progesterone secreting polygonal secreting polygonal cells (yellow arrows). Regressed follicles are demonstrating a group of nonfunctional aggregates of granulosa cells with complete missing or degenerated oocytes (light blue arrows). Scale bars 50 µm.

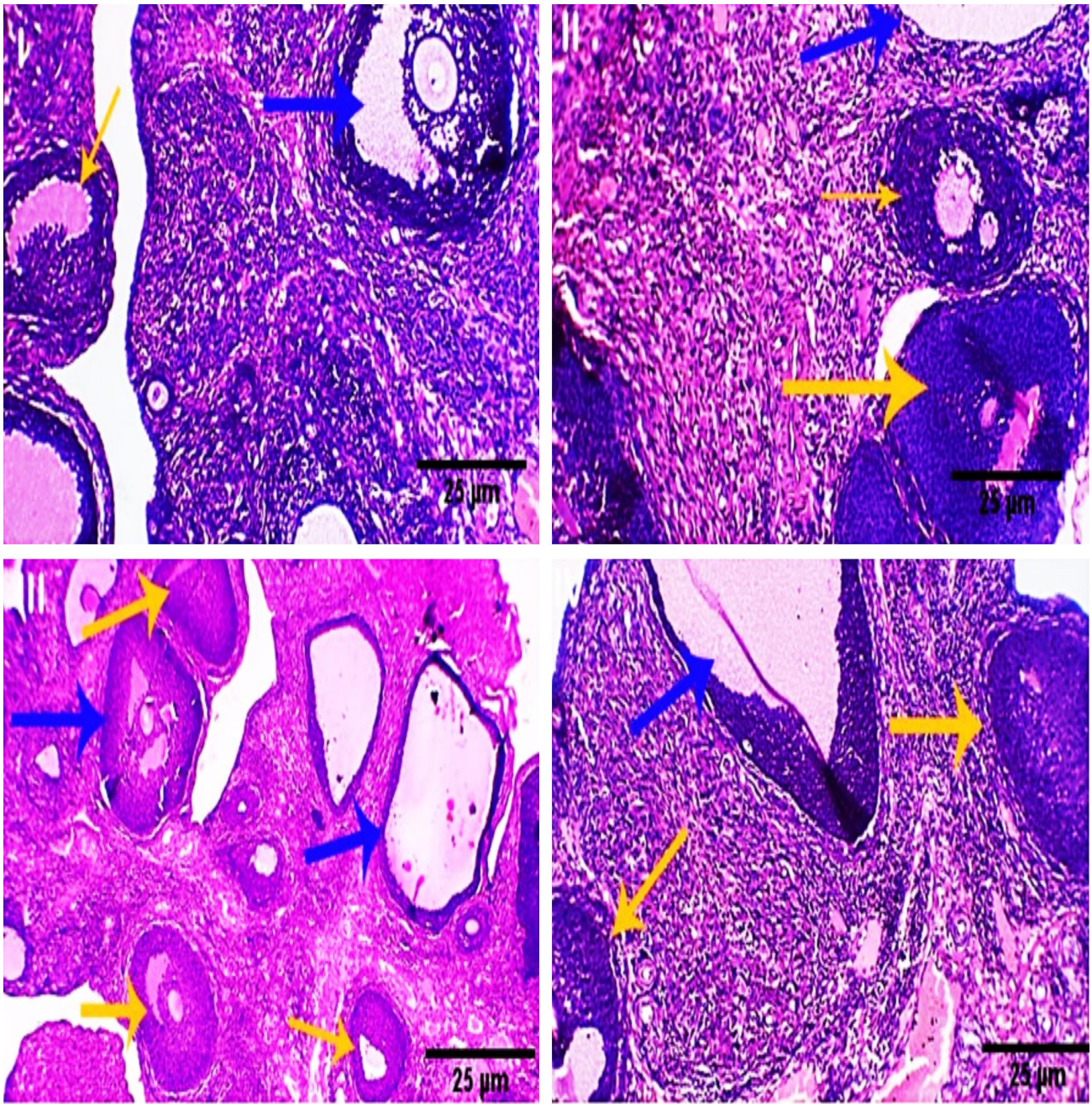


Fig. 6. Photomicrograph from ovaries of control pregnant rats ( G IV) and different treatment groups( G I, G II and G III) showing growing and matured Graafian follicles which are represented by centrally or paracentrally located oocytes surrounded by granulosa cells which seen radially oriented( zona radiata) and peripherally arranged with formation of large antral space filled with eosinophilic materials saturated with estrogen sex hormone in fully matured follicles. The oocytes were either surrounded by solid sheet of granulosa cells (secondary and tertiary growing follicles or multiple small pre-antral spaces) in young growing follicles (blue and orange arrows). Scale bars 25 µm.

## DISCUSSION

The results of a pregnant rat study revealed that the doses of sage herbs significantly increased the total body weight of the animal. This effect was attributed to the fact that these herbs can stimulate appetite and promote weight

gain (Koubaa-Ghorbel *et al*., 2021). The effects of the sage plant on the hypophysis (pituitary gland) can be attributed to its hypoglycemia effect, which can trigger the release of growth hormones (Fetrow & Avila, 2004).

These hormones are responsible for increasing muscle mass and regulating lipolysis. The results of the study revealed that the pregnant rats treated with different doses of sage herbs experienced a significant increase in their body weight. This effect was attributed to the fact that the herbs can stimulate appetite and promote weight gain (Al-Ani *et al*., 2020).

While the current work disagreement with the studies of Monsefi *et al*. (2015) and Alrezaki *et al*. (2021) which their results showed that the body weight of a mature female rat did not change after administration Sage extract, but the current study in covenant to the studies of Al-Syaad & Ibrahim (2014). Increasing in the weight of the reproductive organs of female rats post they were treated with extract of the plant with high doses. This rise due to the effect of Sage on the level of estrogen hormone and this can increase the epithelial cells and the endometrial glands diameter. This effect can lead to an increase in the blood flow and fluid content (AL-Taee, 2009). The present disagree with Koubaa-Ghorbel *et al*. (2021). Another study revealed that the presence of isoflavonoid in the plant which is considered as phytoestrogens which have direct influence on CVS which activate the division of cells and the development of the genital tract of the animals (Adaay *et al*., 2013). The current study supported the results of previous studies what found by (Adaay *et al*., 2013) that showed increasing in the weights of reproductive organs and body weight of the female mice post treatment with Sage extract.

The current work showed a significant rise in the follicle stimulated hormone (FSH) level and serum luteinize hormone (LH) level in groups GII and GIII, This increase can be attributed to the presence of a biological active compound found in these herbs that can stimulate the release of GnRH and increase the production of FSH and LH hormones in the hypophysis (Shi *et al*., 2019). A study conducted by Alrezaki *et al*. (2021) exposed that *Salvia officinalis* extract can increase the production of FSH and LH hormones in mice. Bisher *et al*. (2021) noted that the extract of *Salvia officinalis* can enhance the effects of these hormones in female rabbits. However, the present study disagreed with the results of Al-Ani *et al*. (2020), which showed a significant decrease in the production of both FSH and LH hormones in female mice post administration of Sage extract.

The present study revealed the various beneficial effects of *Salvia officinalis* on the development and maintenance of embryonic stem cells in rats. It also highlighted the properties of this plant in terms of its ovarian dams. This herb is native to the Mediterranean region and is commonly used in Europe and Iran. It is known for its various anti-inflammatory and phytoestrogenic properties due to the occurrence of various phenolic and flavonoids complexes (Barni *et al*., 2012). *Salvia officinalis* contain steroid and isoflavonoid which are documented as phytoestrogens. These complexes in this plant exert an estrogenic effect on CNC and stimulate the growth and division of female cells (Lu & Foo, 2000).

Studies have shown that sage can be used as an effective antioxidant, anti-inflammatory, anti-diabetic, and antimicrobial agent. It also can protect form cancer and cardiovascular diseases. The estrogenic activity of sage was demonstrated experimentally. It can be used as an ingredient for the treatment of various conditions such as pelvic congestion, irregular menses, premenstrual tension, fibroids and endometriosis (Croy *et al*., 2003). *Salvia officinalis* contain a rich collection of particles, its high concentration could improve the function of the ovaries through estrogen signaling pathways, ovarian steroidogenesis and estrogen receptors (Lienou *et al*., 2020). A study conducted on blastocysts revealed that the properties of the inner cell mass and its derivatives including trophoblast cells that cover ICM which give rise to fetal and syncyteotrophoblasts were similar in terms of their morphology and cellular functions. ICM give rise mesodermal and endodermal components of the fetal placental structures and later the membranes of fetuses and the umbilical cord. Variability in the cytological properties of the primitive embryonic structures at this stage of morphogenesis is very difficult among different experimental groups in concern with the aspects of stem cells proliferative capacities, differentiability and physiologic apoptosis.

A physio-anatomical phenomenon known as spontaneous embryo resorption was detected. This was triggered by the rapid removal of the embryo through maternal sterile inflammation. Resorptions were detected at day 7 due to maternal hemorrhage. The number of embryos that were reabsorbed was estimated to be higher in the untreated group than in the treated group. It has been noted that during the early stages of pregnancy, the natural killer cells are the most abundant in the uterus of rodents, monkeys, and humans (Croy *et al*., 2010). These cells are known to produce various growth factors and cytokines that help nourish and develop the fetus during the early stages of pregnancy (Lee *et al*., 2011). Increased uterine Natural Killer cells, lead to pregnancy loss through secretion of IFN-g (Walch & Huber, 2008). In addition, the presence of decidual angiogenesis, which is a crucial part of the primary stages of gestation, was observed in the treated rats. This process is known to maintain the viability of the fetus (Li *et al*., 2015). The dualization of these regions is

dependent on the presence of local factors and the hormones that regulate the growth of the fetus (Wang *et al*., 2016).

Pregnant rat's ovarian tissues of different treatments and control groups demonstrated overall histomorphological structural configurations including growing and matured Graafian follicular countable changes, besides a number of corpora lutea and regressed follicles, an ovarian stimulatory effect was noted in treated rats with compared to control group. The rise of the number of graafian follicles because of presence of estrogen in the herb, which lead to activate excretion of hormone LH level (Adaay *et al*., 2013). Raised estrogen synthesis post ethanolic extract of the *Salvia officinalis* because of phytoestrogenic effect of the extract lead in improvement the number of graffian follicles, corpus luteum and increase the level of LH hormone. Additional studies also discovered that S. officinalis promoted growth (Dadras *et al*., 2020). These may be due to presence of numerous compounds in this herb. GC–MS analysis showed that the dominant constituents (>1 %) were 1,8-cineole (47.40 %), 13-epimanool, (6.45 %), 4,4 dimethylandrost-5-ene (5.90 %), caryophyllene (5.11 %), camphene (4.88 %), a-pinene (4.40 %), camphor (4.32), b-pinene (2.94 %), and ledol (2.32 %). No previous studies have stated the influences of these molecules on function of ovaries (Alrezaki *et al*., 2021).

## CONCLUSION

We therefore conclude that *Salvia officinalis* decoction may promote fertility in females by stimulating FSH, LH and increase the number of graafian follicle, increase the number of implantation sites in pregnant rats with high doses, but the low doses don't promote the fertility in females.

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**ABDRABOU, M. A.; ALHUMAIDI ALOTAIBI, M.; EL NAGAB, N. A. A. & AL-OTAIBIC, A. M.** Evaluación de la fertilidad y la implantación de embriones en ratas después de la administración oral de extracto de *Salvia officinalis* (salvia). *Int. J. Morphol., 40(5*):1404-1414, 2022.

**RESUMEN:** En Arabia Saudita, se cree ampliamente que las mujeres con problemas reproductivos pueden usar el extracto de la planta de salvia como bebida de té. Este estudio se realizó para investigar los efectos de esta hierba sobre la fertilidad de las ratas hembra y la implantación del embrión. Se dividieron cuarenta y ocho ratas hembra vírgenes Wistar en cuatro grupos al azar, con 12 ratas en cada grupo. El grupo control recibió agua destilada por vía oral. Los tres grupos de tratamiento recibieron diferentes concentraciones de extracto de salvia: 15, 60 o 100 mg/kg durante 14 días antes del apareamiento, luego se aparearon con un macho y se sacrificaron el día 7 de gestación, se extrajeron los cuernos uterinos y se fotografiaron. Se determinó el peso corporal total de las madres, el peso del útero y los ovarios y el número de fetos. Los tejidos ováricos y uterinos se cortaron en secciones de 5 µ y se tiñeron con hematoxilina y eosina. FSH sérica, LH se determinaron por el método ELISA. El presente estudio mostró que dosis bajas de salvia (15 mg/kg) no tienen efectos sobre los niveles de concentración sérica de las hormonas FSH y LH, tampoco tienen efecto sobre el número de folículos en crecimiento. El presente estudio mostró diferencias significativas (P≤0,05) en el peso corporal, peso de ovario y útero en los grupos tratados con altas dosis de *Salvia officinalis* en comparación con el grupo control. También se encontraron diferencias significativas (P≤0,05) en las hormonas FSH, LH. El estudio histológico mostró configuraciones estructurales histomorfológicas generales que incluyen cambios contables en los folículos maduros (de Graaf) y en crecimiento, además de una cantidad de cuerpos lúteos y folículos en regresión en los grupos tratados con altas dosis de *Salvia officinalis* en comparación con el grupo de control. Los investigadores concluyeron que el extracto de la planta de salvia en altas dosis puede estimular el crecimiento de los folículos maduros y mejorar la fertilidad en ratas hembra.

**PALABRAS CLAVE: *Salvia officinalis*; Hormona estimuladora folicular; Hormona luteinizante; Fertilidad; Implantación de embriones.**

## REFERENCES

Abd Rabou, M. A. & Alotaibi, M. A. Toxicity of somadril compound on fetal ileum tissues of albino rats. Pak. J. Biol. Sci., 24(1):72-9, 2021.

Adaay, M. H.; Al-Dujaily, S. S. & Khazzal, F. K. Effect of aqueous extract of Medicago sativa and *Salvia officinalis* mixture on hormonal, ovarian and uterine parameters in mature female mice. *J. Mater. Environ. Sci., 4(4)*:424-33, 2013.

Al-Ani, B. T.; Al Saadi, R. R. & Reshan, R. G. Investigating effects of *Salvia officinalis* (Sage) on development of mice embryos kidney and some hormonal effect of treated mothers. *Indian J. Forensic Med. Toxicol., 14(1)*:649-54, 2020.

Al-Syaad, K. & Ibrahim, E. H. The protective effects of the aqueous extract of saliva against biochemical and histopathological changes in kidney and liver of male rats treated with the anticancer drug Doxorubicin. *Res. J. Pharm. Biol. Chem. Sci., 5(6)*:237-43, 2014.

AL-Taee, Z. K. *The Effect of Water Extract of Turmeric Plant in Reproductive Efficiency of Rat Females Exposed to Oxidative Stress Induced by Potassium Dichromate.* M.Sc. Degree. Mosul, College of Veterinary Medicine, University of Mosul, 2009.

Alrezaki, A.; Aldawood, N.; Alanazi, S.; Arafah, M.; Fabova, Z.; Badjah, Y.; Sirotkin, A. V.; Alwasel, A. & Harrath, A. H. Consumption of sage (*Salvia officinalis*) promotes ovarian function by stimulating estradiol hormone release and controlling folliculogenesis, steroidogenesis, and autophagy. *J. King Saud Univ. Sci., 33(2)*:101319, 2021.

Andrade, J. M.; Faustino, C.; Garcia, C.; Ladeiras, D.; Reis, C. P. & Rijo, P. Rosmarinus officinalis L.: an update review of its phytochemistry and biological activity. *Future Sci. OA, 4(4)*:FSO283, 2018.

Badiee, P.; Nasirzadeh, A. & Motaffaf, M. Comparison of *Salvia officinalis* L. essential oil and antifungal agents against candida species. *J. Pharm. Technol. Drug Res., (1)*:1-5, 2012.

Barni, M. V.; Carlini, M. J.; Cafferata, E. G.; Puricelli, L. &Moreno, S. Carnosic acid inhibits the proliferation and migration capacity of human colorectal cancer cells. *Oncol. Rep., 27(4)*:1041-8, 2012.

Bisher, M.; Ijioma, N. I.; Aghila, F. H. & Hamed, K. A. Effect of Saliva officinalis decoction on the hormone profile of normal and letrozole treated adult female rabbit. *Eur. J. Pharm. Med. Res., 8(3)*:132-6, 2021.

Boroujeni, N. & Gholami, M. Effect of the ethanolic extract of *Salvia officinalis* on ovarian angiogenesis in mice at preimplantation: a morphological and molecular analysis. *Jentashapir J. Health Res., 8(1)*:e44386, 2017.

Burtis, C. A.; Ashwood, E. R. & Bruns, D. E. *Tietz Fundamentals of Clinical Chemistry.* 6th ed. Philadelphia, W. B. Saunders, 2008. pp.797.

Carson, S. A. & Kallen, A. N. Diagnosis and management of infertility: a review. *JAMA, 326(1)*:65-76, 2021.

Ciotta, L.; Stracquadanio, M.; Pagano, I.; Andò, A.; Valenti, O. & Roccasalva, L. Clinical effects of treatment with phytoestroges in postmenopausal women. *Minerva Ginecol., 64(1)*:15-22, 2012.

Croy, B. A.; He, H.; Esadeg, S.; Wei, Q.; McCartney, D.; Zhang, J.; Borzychowski, A.; Ashkar, A. A.; Black, G. P.; Evans, S. S.; *et al*. Uterine natural killer cells: insights into their cellular and molecular biology from mouse modelling. *Reproduction, 126(2)*:149-60, 2003.

Croy, B. A.; Zhang, J.; Tayade, C.; Colucci, F.; Yadi, H. & Yamada, A. T. Analysis of uterine natural killer cells in mice. *Methods Mol. Biol., 612*:465-503, 2010.

Dadras, H.; Hayatbakhsh, M. R. & Golpour, A. Dietary administration of common sage (*Salvia officinalis*) and coneflower (Echinacea angustifolia) extracts affects growth, blood parameters and immune responses of Beluga, Huso huso. *Turk. J. Fish. Aquat. Sci., 20(5)*:367-74, 2020.

Fetrow, C. W. & Avila, J. R. *Professional's Handbook of Complementary and Alternative Medicines.* 3rd ed. Philadelphia, Lippincott Williams and Wilkins, 2004. pp.25-6, 728-9.

Ghorbani, A. & Esmaeilizadeh, M. Pharmacological properties of *Salvia officinalis* and its components. *J. Tradit Complement. Med., 7(4)*:433-40, 2017.

Khan, I. A. & Abourashed, E. A. *Leung's Encyclopedia of Common Natural Ingredients: Used in Food, Drugs and Cosmetics*. New Yor, John Wiley & Sons, 2011.

Koubaa-Ghorbel, F.; Chaâbane, M.; Jdidi, H.; Turki, M.; Makni-Ayadi, F. & El Feki, A. *Salvia officinalis* mitigates uterus and liver damages induced by an estrogen deficiency in ovariectomized rats. *J. Food Biochem., 45(5)*:e13542, 2021.

Lee, J. W.; Hyun, M. K. & Ang, L. Complementary therapy for female infertility: protocol for an overview of systematic reviews. *Integr. Med. Res., 9(1)*:62-4, 2020.

Lee, J. Y.; Lee, M. & Lee, S. K. Role of endometrial immune cells in implantation. *Clin. Exp. Reprod. Med., 38(3)*:119-25, 2011.

Li, M.; Li, Q.; Zhang, C.; Zhang, N.; Cui, Z.; Huang, L. & Xiao, P. An ethnopharmacological investigation of medicinal Salvia plants (Lamiaceae) in China. *Acta Pharm. Sin. B., 3(4)*:273-80, 2013.

Li, Y.; Gao, R.; Liu, X.; Chen, X.; Liao, X.; Geng, Y.; Ding, Y.; Wang, Y. & He, J. Folate deficiency could restrain decidual angiogenesis in pregnant mice. *Nutrients, 7(8)*:6425-45, 2015.

Lienou, L. L.; Telefo, P. B.; Rodrigues, G. Q.; Donfack, J. N.; Araújo, R. A.; Bruno, J. B.; Njimou, J. R.; Mbemya, T. G.; Santos, R. R.; Souza, J. F.; *et al*. Effect of different extracts and fractions of Senecio biafrae (Oliv. &Hiern) J. Moore on in vivo and in vitro parameters of folliculogenesis in experimental animals. *J. Ethnopharmacol., 251*:112571, 2020.

Lu, Y. & Foo, L. Y. Flavonoid and phenolic glycosides from *Salvia officinalis*. *Phytochemistry, 55(3)*:263-7, 2000.

Mascarenhas, M. N.; Flaxman, S. R.; Boerma, T.; Vanderpoel, S. & Stevens, G. A. National, regional, and global trends in infertility prevalence since 1990: a systematic analysis of 277 health surveys. *PLoS Med., 9(12)*:e1001356, 2012.

Mehta, M. *Salvia officinalis* Linn.: relevance to modern research drive. *Planta Act., 4*:203-7, 2012.

Mendes, F. S. F.; Garcia, L. M.; da Silva Moraes, T.; Casemiro, L. A.; de Alcantara, C. B.; Ambrósio, S. R.; Veneziani, R. C. S.; Miranda, M. L. D. & Martins, C. H. G. Antibacterial activity of *Salvia officinalis* L. against periodontopathogens: An *in vitro* study. *Anaerobe, 63*:102194, 2020.

Mishra, A.; Ashary, N.; Sharma, R. & Modi, D. Extracellular vesicles in embryo implantation and disorders of the endometrium. *Am. J. Reprod. Immunol., 85(2)*:e13360, 2021.

Monsefi, M.; Abedian, M.; Azarbahram, Z. & Ashraf, M. J. *Salvia officinalis* L. induces alveolar bud growing in adult female rat mammary glands. *Avicenna J. Phytomed., 5(6)*:560-7, 2015.

Mossi, A. J.; Cansian, R. L.; Paroul, N.; Toniazzo, G.; Oliveira J. V.; Pierozan, M. K.; Pauletti, G.; Rota, L.; Santos, A. C. & Serafini, L. A. Morphological characterisation and agronomical parameters of different species of Salvia sp. (Lamiaceae). *Braz. J. Biol., 71(1)*:121-9, 2011.

Orieke, D.; Ohaeri, O. C.; Ijeh, I. I. & Ijioma, S. N. Semen quality, hormone profile and histological changes in male albino rats treated with Corchorus olitorius leaf extract. *Avicenna J. Phytomed., 9(6)*:551-62, 2019.

Pedro, D. F. N.; Ramos, A. A.; Lima, C. F.; Baltazar, F. & Pereira-Wilson, C. Colon cancer chemoprevention by sage tea drinking: decreased DNA damage and cell proliferation. *Phytother. Res., 30(2)*:298-305, 2016.

Russo, A.; Formisano, C.; Rigano, D.; Senatore, F.; Delfine, S.; Cardile, V.; Rosselli, S. & Bruno, M. Chemical composition and anticancer activity of essential oils of Mediterranean sage (*Salvia officinalis* L.) grown in different environmental conditions. *Food Chem. Toxicol., 55*:42-7, 2013.

Shi, Y.; Li, L.; Zhou, J.; Sun, J.; Chen, L.; Zhao, J.; Wu, L.; Cui, Y.; Wu, L. & Wu, H. Efficacy of electroacupuncture in regulating the imbalance of AMH and FSH to improve follicle development and hyperandrogenism in PCOS rats. *Biomed. Pharmacother., 113*:108687, 2019.

Singh, M.; Chaudhry, P. & Asseli, E. Embryo implantation disorders. *Pregnancy Dis. Perinat. Outcomes, 2012*:3-19, 2012.

Vitale, D.C.; Piazza, C.; Melilli, B.; Drago, F. & Salomone, S. Isoflavones: estrogenic activity, biological effect and bioavailability. *Eur. J. Drug Metab. Pharmacokinet., 38(1)*:15-25, 2013.

Walch, K. T. & Huber, J. C. Progesterone for recurrent miscarriage: truth and deception. *Best. Pract. Res. Clin. Obstet. Gynaecol., 22(2)*:375-89, 2008.

Wang, X.; Tan, D.; Ma J, Liang, H.; Zhang, Q.; Tan, Y.; Wang, J. & Luo, W. Positive regulation of decidualization by l-type amino acid transporter 1 (lat1) in pregnant mice. *Nutrients, 8(11)*:704, 2016.

Zheng, R.; Zhao, Y.; Wu, J.; Wang, Y.; Liu J. L.; Zhou, Z. L.; Zhou, X. T.; Chen, D. N.; Liao, W. H. & Li, J. D. A novel PNPLA6 compound heterozygous mutation identified in a Chinese patient with Boucher-Neuhäuser syndrome. *Mol. Med. Rep., 18(1)*:261-7, 2018.

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