


Characterization of pregnancy rate and offspring counts in Wistar rats females after experimental treatment with propolis extract

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Abstract The current study was carried out on Twenty (20) female white rats aged 105-110 days; sourced from the Animal House/College of Veterinary Medicine/University of Al-Qadisiyah. The animals used in the study were equally divided into two groups; the treatment group (T) which made up of 10 female white rats, the animals of this group were weighed and dosed daily with 40 milligrams/kg of the alcoholic extract bee propolis for a period of 30 days and, the control group (C) which made up of 10 female white rats which were also weighed, but leaved without any treatment. Female rat's pregnancy rate and the number of newborns were investigated. The results recorded a superiority of the group treated with propolis in pregnancy rate of 80 % compared to the control group, which gave a pregnancy rate of only 60%. It was also demonstrated that the effect of alcoholic propolis extract is beneficial in increasing that of offspring compared to the number of offspring in the control group (55 vis 29) respectively.

Keywords: Female, pregnancy, Propolis, rats

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Introduction Traditionally propolis has been used for centuries in the treatment of many diseases, it is of great significance in defending honey bee cells from bacterial, fungal or other threats, it also play a role in protecting the body affective and prevent genetic disorders leading to tumor or cell death (1-6). The honey collection season and the geographical area for rearing also have a great influence on the physical and chemical properties, and this is reflected in nutritional and medicinal properties of propolis, these properties are also affected depending on the place where bees live and the food that bees eats, therefore, there is a wide difference in the composition of propolis; it contains over 180 types of important and useful compounds (7, 8). The most documented medicinal properties of propolis are antibacterial, antifungal, anti- inflammatory, antiviral, anaesthetic, antioxidant, antitumour, antiprotozoal and anticancer (3, 4, 9). It also has been used in the management of hypertension, anti-cancer and anti-hepatotoxicity as well as detoxacation of cells of the body (6). Moreover, owing to bioflavonoids, vitamin C, arginine, provitamin A, vitamin B-complex some minerals present in it, it also has the quality of healing the wound and thus helps to increase the healing of the injured and shortens the required time to heal the injury (10, 11).

In this study, there is attempted to uncover the effect of propolis on the rate of pregnancies, and clinically observed newborn.

Materials and method

Animal ethics and care

The current research was carried out according to the guidelines of the National Research Council for the care

and use of laboratory animal. The Ethical Council in the College of Veterinary Medicine, University of Al-Qadisiyah agreed to conduct this experiment.

Ethical Approval

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed. All procedures performed in studies involving animals were in accordance with the ethical standards of the institution or practice at which the studies were conducted. We were explaining the ethical state of animals in our study in details in the materials and method section and included the references that we were depending on.

Animals of the study

This study was carried out in laboratory animal house of College of Veterinary Medicine, Al-Qadisiyah University. Twenty female white rat were used for this experiment and which were 105 – 110 days old. The animals were housed in a plastic cages in laboratory environment, where light was around 12-14 hours and within a temperature ranged between 25-30 °C. The animal house was also having a controlled source of heating and cooling; water bottles of a peculiar type were used; the rats were fed with the special, balanced diet.

Preparation of alcoholic propolis extract

To obtain an alchoholic extract of propolis, 40 g of dry propolis powder was applied in to the thimble of the continuous extraction device (Soxhlet) (Korean company WISD). 500 ml of ethanol alcohol (70 %) was placed in the beaker of the device and the device was left to operate for 6- 8 h at a temperature of 70 °C, after that

the solution was filtered using Whatman No. 1 filter paper, then the filtrate was concentrated to dryness using rotary evaporator device at 60 °C. The remaining liquid was dried using an incubator at a temperature of 37°C for 48-72 hours to get the last and ultimate extract and placed the extract in the refrigerator at 4°C until it is used (12).

Design of study

The study animals, which 20 female white rats, were divided into two equal groups, where each group included 10 females as follows:

1-Control group (C): This groups had the animals weighed and leaved without any treatment; the pregnancy rate and total number of newborn were investigated after experiment ending.

2-Treatment group (T): In this group, the body weight was recorded, and the animals were treated with alcoholic extract propolis 40 mg/kg per day for a period of 30 days. After the completion of the study the pregnancy rate and number of newborns were documented. The mating of rats used on male for each two females (13).

Statistical analysis

The statistical program Statistical Package of Social Science (SPSS) was employed in the analysis of the results that were obtained in the study. The chi-square test was used to determine the significance of the differences in the percentage and the Independent T test to determine the significant differences for the rates at probability level of 0.01 (14).

Results and Discussion

Effect of propolis on the pregnancy female rate

As it is evidenced by Table (1) and Figure (1), the procedure of dosing propolis extract contributed to high increase in pregnancy rates in the treatment group in contrast to the control group of rats, which did not receive the treatment. In control group, pregnancy was 60%, while in the treatment group the pregnancy rate was 80%. This can account for the rising fertility rates due to the components of the propolis extract and this is in agreement with what was discovered (15, 16), who pointed out that there is a positive response to reproductive performance with influence on the rate of fertility by giving trace mineral elements. It is also in par with what was found by (17, 18), who stated that there was positive impacts of minerals on the rate of pregnancy and also used in treatment of cases of silent estrus, infertility and reproductive disorders and ; who noted that the advancement and regression of the ovarian follicles is influenced by the type of nutrition. The result of this study agreed with the studies done by (19) by histological examination of the ovaries where it has been confirmed that propolis had a positive influence on the number of mature ova.

Table 1: Alcoholic extract of propolis and its impact on pregnancy rate in female rats

groups	C	T
Pregnancy rates		
Pregnant females rats	60% ^b	80% ^a
Non Pregnant females rats	40% ^a	20% ^b

*Different letters mean significant ($P \leq 0.01$) in rows

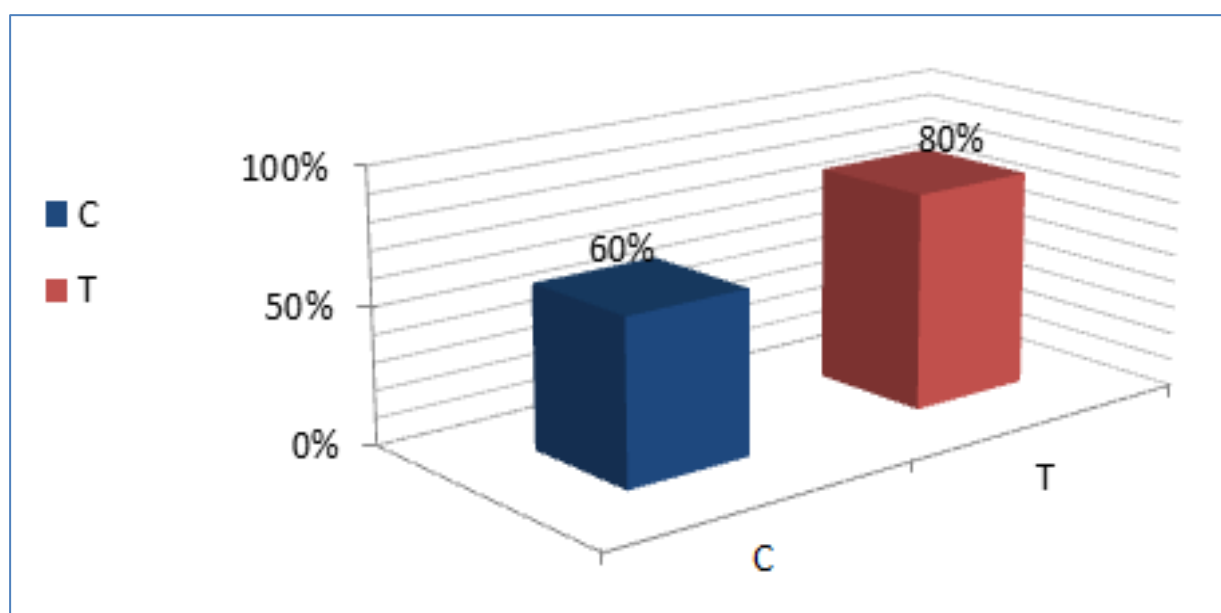


Figure 1: Alcoholic extract of propolis and its impact on pregnancy rate in female rat

Effect propolis on the number of newborn in female rats

According to the findings of the present study, there was increased delivery of newborn in rats in the female rats

treated with propolis extract (T) which were 55 newborn, while the female rats in the untreated group (C) delivered 29 newborn only (Table 2 and Figure 2). The difference in results can be explained by improving nutrition through the alcoholic extract of propolis, and this results are as they have been with what shown by (20), who found that when feeding animals with diet containing high levels of trace mineral elements, there was a high number of ovum observed. It is in par with what was found by (21) that organic-mineral elements contributes to the enhancement of reproductive performance via the promotion of ovarian functionality

Groups	C	T
Newborns		
Number of newborns	29 ^b	55 ^a
Ratio of newborns to mothers newborns	4.833	6.875
Ratio of newborns to all mothers	2.9	5.5

and healthy fetal outcome. This is in line with what (22) depicted regarding the relationship between the nutritional level and the development of the follicles as well as increase of the ovulation rate. In the same sentiment with (23) who also found out that significantly enhanced the number follicles and oocytes in the neonatal rat model exposed to stress.

Table 2: The positive impact of alcoholic extract of propolis on the number of newborns and ratio of newborns to mothers in the female rats.

*Different letters mean significant ($P \leq 0.01$) in rows.

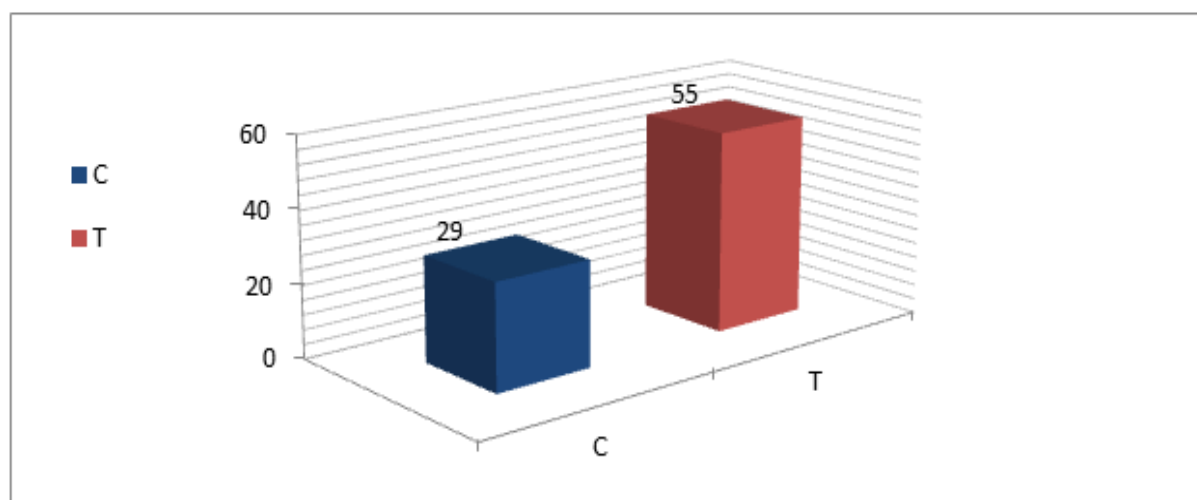


Figure 2: The impact of alcoholic extract of propolis on the number of newborn.

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Conflicts of Interest

The authors declare that there is no conflict of interest for the current study.

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References

1. Chan GC-F, Cheung K-W, Sze DM-Y. The immunomodulatory and anticancer properties of propolis. *Clin Rev Allergy Immunol*. 2013;44:262-73. <https://doi.org/10.1007/s12016-012-8322-2>
2. El Sohaimy S, Masry S. Phenolic content, antioxidant and antimicrobial activities of Egyptian and Chinese

propolis. *American-Eurasian Journal of Agricultural and Environmental Sciences*. 2014;14:1116-24.

3. Sforcin JM. Biological properties and therapeutic applications of propolis. *Phytother Res*. 2016;30(6):894-905. <https://doi.org/10.1002/ptr.5605>
4. Kuropatnicki AK, Szliszka E, Krol W. Historical aspects of propolis research in modern times. *Evidence-Based Complementary and Alternative Medicine*. 2013;2013(1):964149. <https://doi.org/10.1155/2013/964149>
5. Drescher N, Klein A-M, Neumann P, Yañez O, Leonhardt SD. Inside honeybee hives: Impact of natural propolis on the ectoparasitic mite *Varroa destructor* and viruses. *Insects*. 2017;8(1):15. <https://doi.org/10.3390/insects8010015>

6. Toreti VC, Sato HH, Pastore GM, Park YK. Recent progress of propolis for its biological and chemical compositions and its botanical origin. Evidence-Based Complementary and Alternative Medicine. 2013;2013(1):697390.
<https://doi.org/10.1155/2013/697390>
7. Afrouzan H, Zakeri S, Mehrizi AA, Molasalehi S, Tahghighi A, Shokrgozar MA, et al. Anti-plasmodial assessment of four different Iranian propolis extracts. Arch Iran Med. 2017;20(5):270-81.
8. Bueno-Silva B, Marsola A, Ikegaki M, Alencar SM, Rosalen PL. The effect of seasons on Brazilian red propolis and its botanical source: chemical composition and antibacterial activity. Nat Prod Res. 2017;31(11):1318-24.
<https://doi.org/10.1080/14786419.2016.1239088>
9. Abdulrhman M, Samir Elbarbary N, Ahmed Amin D, Saeid Ebrahim R. Honey and a mixture of honey, beeswax, and olive oil-propolis extract in treatment of chemotherapy-induced oral mucositis: a randomized controlled pilot study. Pediatric hematology and oncology. 2012;29(3):285-92.
<https://doi.org/10.3109/08880018.2012.669026>
10. Anjum SI, Ullah A, Khan KA, Attaullah M, Khan H, Ali H, et al. Composition and functional properties of propolis (bee glue): A review. Saudi J Biol Sci. 2019;26(7):1695-703.
<https://doi.org/10.1016/j.sjbs.2018.08.013>
11. Pasupuleti VR, Sammugam L, Ramesh N, Gan SH. Honey, propolis, and royal jelly: a comprehensive review of their biological actions and health benefits. Oxid Med Cell Longev. 2017;2017(1):1259510.
<https://doi.org/10.1155/2017/1259510>
12. Deshmukh S, Borle M. Studies on the insecticidal properties of indigenous plant products. 1975.
13. Mattaraia VGdM, Moura ASAMT. Productivity of Wistar rats in different mating systems. Ciência Rural. 2012;42:1490-6.
<https://doi.org/10.1590/S0103-84782012000800026>
14. Petrie A, Watson P. Statistics for veterinary and animal science: John Wiley & Sons; 2013.
15. Larson CK, editor Role of trace minerals in animal production. Nutrition Conference, Department of Animal Science, University of Tennessee <http://www.tennesseenutritionconference.org/Conference2005.htm>; 2005.
16. Ballantine H, Socha M, Tomlinson DAD, Johnson A, Fielding A, Shearer J, et al. Effects of feeding complexed zinc, manganese, copper, and cobalt to late gestation and lactating dairy cows on claw integrity, reproduction, and lactation performance. The professional animal scientist. 2002;18(3):211-8.
[https://doi.org/10.15232/S1080-7446\(15\)31524-2](https://doi.org/10.15232/S1080-7446(15)31524-2)
17. Subha Ganguly SG. Role of biochemical factors and mineral supplementation in livestock ration for maintenance of their fertility and healthy reproductive status: a review. 2013.
18. Islam N, Ray S, Batabyal S, Mukhopadhyay S, Ganguly S. Effect of certain micro minerals on fertility in mare. Indian Journal of Animal Nutrition. 2012;29(3):307-9.
19. Abbass AA, El-Asely AM, Kandiel MM. Effects of dietary propolis and pollen on growth performance, fecundity and some hematological parameters of *Oreochromis niloticus*. Turkish J Fish Aquat Sci. 2012;12(4):851-9.
20. Ansotegui R, Bailey J, Paterson J, Hatfield P, Swenson C, editors. Effects of supplemental trace mineral form on copper status, estrus, ovulation rate, and fertility in beef heifers. PROCEEDINGS-AMERICAN SOCIETY OF ANIMAL SCIENCE WESTERN SECTION; 1999: NEW MEXICO STATE UNIVERSITY.
21. Hackbart K, Ferreira R, Dietsche A, Socha M, Shaver R, Wiltbank M, et al. Effect of dietary organic zinc, manganese, copper, and cobalt supplementation on milk production, follicular growth, embryo quality, and tissue mineral concentrations in dairy cows. J Anim Sci. 2010;88(12):3856-70.
22. Viñoles Gil C. Effect of nutrition on follicle development and ovulation rate in the ewe. 2003.
23. Arabameri A, Sameni H, Bandegi A. The effects of propolis extract on ovarian tissue and oxidative stress in rats with maternal separation stress. International Journal of Reproductive BioMedicine. 2017;15(8):509.

