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(ARTICLE REVIEW)

STUDY THE BIOLOGICAL EFFECT AND NANO PREPARATION OF OLEANDER PLANT *NERIUM OLEANDER* ON THE HAIRY GRAIN BEETLE *Trogoderma granarium* EVERTS (COLEOPTERA: DERMESTIDAE)

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ABSTRACT

Cereal crops, including wheat (*Triticum aestivum*), are among the most important vital and strategic crops in the world because they contain important food sources of vitamins, proteins, carbohydrates, fats, and other nutrients. Stored grains are affected by many insect pests, causing significant economic losses in grain weight that may reach 10% in one storage season. One of the most dangerous stored insects that infest grains is the hairy grain beetle (*Trogoderma granarium*).

The insect is known for its resistance to various liquid and gaseous chemical pesticides, as well as its ability to withstand long periods of drought and high temperatures. This adaptability has facilitated its spread across different countries and enabled it to conquer new geographical areas. Several methods have been employed to combat this insect, with chemical control being the most prominent. However, the use of chemical pesticides has resulted in negative environmental impacts and challenges in addressing the various roles insects play.

Due to the economic significance of the hairy grain beetle and the need to minimize reliance on harmful chemical pesticides, plant extracts and nano-preparations of iron from the leaves of the oleander plant (*Nerium oleander*) were utilized. These compounds have shown high effectiveness in controlling the Khabra insect in both laboratory and storage environments, while also being environmentally safe.

Keywords: *Nerium oleander,* proteins, pesticides, insects.

دراسة التاثير البيولوجي والمستحضر النانوي لنبات الدفلة على خنفساء الحبوب الشعرية ***Trogoderma granarium*** **Everts (Coleoptera : Dermestidae) .**

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الخلاصة

تعد محاصيل الحبوب ومنها محصول الحنطة (*Triticum aestivum*) من اهم المحاصيل الحيوية والاستراتيجية في العالم بسبب احتوائها على مصادر غذائية مهمة من فيتامينات وبروتينات وكربوهيدرات ودهون وعناصر غذائية أخرى. تتأثر الحبوب المخزونة بالعديد من الآفات الحشرية مسببة خسائر اقتصادية كبيرة في وزن الحبوب قد تصل الى 10% في موسم خزني واحد. ومن اخطر الحشرات المخزنية المصنفه عالميا التي تصيب الحبوب المخزونة حشرة خنفساء الحبوب الشعرية *Trogoderma granarium*.

وتعرف الحشرة بمقاومتها لمختلف المبيدات الكيماوية السائلة والغازية، فضلا عن قدرتها على تحمل فترات طويلة من الجفاف ودرجات الحرارة المرتفعة. وقد سهلت هذه القدرة على التكيف انتشاره عبر مختلف البلدان ومكنته من غزو مناطق جغرافية جديدة. وقد تم استخدام طرق متنوعة لمكافحة هذه الحشرة، وكان أبرزها المكافحة الكيميائية. إلا أن استخدام المبيدات الكيماوية أدى إلى آثار بيئية سلبية وتحديات في معالجة ادوار الحشرة المختلفة.

نظرًا للأهمية الاقتصادية لخنفساء الحبوب الشعرية ولتقليل الاعتماد على المبيدات الكيميائية الضارة، تم استخدام المستخلصات النباتية والمستحضرات النانوية للحديد المستخلص من أوراق نبات الدفلة (*Nerium oleander*) حيث أثبتت هذه االمركبات فعالية كبيرة في مكافحة حشرة الخابرا مختبريا ومخزنيا، كما أنها آمنة بيئيا.

الكلمات المفتاحية: الدفلة، بروتين، مبيدات، حشرات.

INTRODUCTION

Wheat (*T. aestivum*) is a strategic crop that is important for food security for the world's population, and its global demand is expected to reach 3.3 billion tons by 2050 **(Mustafa, 2017; UN report, 2022)**. Stored insects are among the most important pests that infect wheat seeds in stores, damaging them and reducing their quality, causing significant economic losses in the quality and weight of grains estimated at 6-73% **(Khalique *et al*., 2018; Rajendran, 2020).** The most important of these is the hairy grain beetle (Khabra) *T. granarium* (Everts), which is one of the dangerous and globally widespread insects and is characterized by its voracious feeding on completely stored grains in addition to its waste that is harmful to health **(Khan *et al*., 2019; Athanassiou *et al*., 2019).**

The widespread and frequent use of chemical pesticides to control stored insects, especially the grain beetle, has led to the emergence of resistance to pesticides, in addition to environmental pollution and toxicity to humans and animals. The most used materials are Phostoxin tablets and strips that produce phosphine gas **(Wakil *et al*., 2021; Yılmaz & Koçak, 2022)**. To reduce the risks of using pesticides, alternative methods have begun to be used for chemical pesticides that are safe for the environment and human health, such as the use of plant extracts that contain many biological compounds that are effective in controlling pests and protecting stored grains from insect infestation, which are environmentally friendly compounds due to their biodegradability and low toxicity to animals **(Alvi *et al*., 2018).**

The use of nano-preparations is one of the most modern and effective methods for combating stored pests. Plant extracts are used in the biosynthesis of nanoparticles and improving the control properties. It is an environmentally safe and economical method due to the speed and ease of preparation. It is expected that the global production of nano-preparations will reach ten thousand tons during the year 2026 **(Ovais *et al*., 2018).**

The economic importance of the grain beetle and its damages It belongs to the order of coleoptera, its scientific name is *T. granarium*, and it is considered one of the most important insect pests that attack stored grains and lead to their spoilage and the most dangerous in many countries of the world and in Iraq, causing a large economic loss in stored grains in terms of quantity and quality. In addition to the secretion of molting skins, dead insects, and insect waste, causing grain contamination, and as a result of the insect feeding in large numbers due to its rapid reproduction, it leads to an increase in the temperature of grain stores, which exposes them to infection with fungi and bacteria and makes them unfit for marketing, human consumption, and use as seeds **(Media, 2007; Ahmedani *et al*., 2009).**

The harmful stage of the insect is the larval stage, especially in advanced ages. The larvae feed on grains by gnawing them and destroying the embryo and endosperm, which makes them unsuitable for cultivation due to the low germination rate. The insect is characterized by its voracious feeding on different types of grains and dry food. One larva can consume 18 mg of grains per day, leaving behind waste at a rate of 14.8 mg. It is characterized by activity in a wide temperature range between 20- and 50°C, and the loss rate reaches 70% when suitable conditions are available **(Al-Iraqi, 2010).**

The importance of oleander plant extracts in pest controls. The ethanolic extract of ***N. oleander*** flowers has proven highly effective against the activity of plant pathogenic fungi, including *Fusarium oxysporum*, *Fusarium solani*, *Alternaria alternate*, and *Rhizoctonia solani* **(Hadizadeh *et al*., 2009)**. The bark, stem, leaves, flowers, and roots extract of oleander have also been used as an anti-feeding insecticide to control the larvae of the diamondback moth *Plutella xylostella* and Aedes aegypti (**Komalamisra *et al*., 2005**). The hexane and aqueous extracts of crude oleander flowers were highly effective in mortality of *Culex quinquefasciatus* larvae, with LC50 values reaching 102.54 and 61.11 ppm after 24 and 48 hours of treatment with the hexane extract **(Raveen *et al*., 2014).**

Al-Iraqi (2003) studied the efficiency of several plant powders against the hairy grain beetle (Khabra) *T. granarium*, including *N. oleander* L. powder, which was mixed with wheat grains at concentrations of 0.3, 0.6, and 0.9%. It influenced the generation duration, reaching 48.83, 48.96, and 54.09 days for the concentrations, respectively. It reached 40.18 days in the control treatment due to the anti-feeding effect of the oleander plant, as it reduced egg laying. **Suleiman (2005)** found that surface treatment of the adults of the rusty flour beetle *Tribolium castaneum* (Herbst.) with alcoholic and aqueous extracts of *N. oleander* L. leaves caused mortality of adults after 48 hours of treatment.

The cold aqueous extract of *N. oleander* leaves influenced the periodic death of eggs and larvae of the greater wax moth *Galleria mellonella* at several concentrations, as the mortality rate at the concentration of 4 mg/ml reached (99.4% and 96.6%) for eggs and first-stage larvae, respectively **(Najib, 2019)**. **Al-Mashhadani (2012)** also found that the powder of N. oleander leaves had a high repellent rate against larvae and adults of the hairy grain beetle *T. granarium* in the test he conducted to test the attraction and repellent of the powder. A study showed that the extract of *N. oleander* L. caused an inhibition in the severity of the disease early blight on tomato crop caused by the fungus *Alternaria solani* **(Nashwa & Abu-Elyousr, 2012)**.

Nanotechnology

The word nano refers to nanoparticles that are between n 1 an100) nanometers in size. Nanotechnology has spread rapidly in the world due to the features and properties of nanoparticles, their wide applications, their chemical composition, and their surface structure. Their small size increases the surface cohesion of nanoparticles **(Adhikari *et al*., 2013; Tawfeeq, 2014; Ahmida *et al*., 2017; Gulzar *et al*., 2020)**. Nanoparticles that have been converted into pesticides are characterized by green technology that selectively targets insects to preserve plants and the environment, as well as combating insects that consume stored grains **(Sahayaraj, 2017).**

The importance of nanoparticles in insect control:

The extract of the oleander plant was used to prepare silver nanoparticles by reducing silver nitrate and studied its effect in preventing *Culex pipiens mosquito* larvae from reaching the adult stage by using several concentrations of the plant extract and silver nanoparticles. The silver nanoparticles were more effective in preventing the emergence of adult insects from the extract by about 33.33 times, and both the plant extract and silver particles led to deformities in the treated larvae, which led to mortality of the insect and failure to complete its life cycle **(Al-Qurashi *et al*., 2015)**. **(Rouhani *et al*., 2008)** showed that treating the food of the southern cowpea beetle *Callosobruchus maculatus* with silver nanoparticles at a concentration of 2.5 g/kg grains led to their death by 75% after 14 days of treatment. Silver nanoparticles prepared using the aqueous extract of *Euphorbia prostste* leaves were used to control the adults of the rice weevil *Sitophilus oryzae* at a concentration of 250 mg/kg and a size of 25–80 nm, and the mortality rate reached 100% after 14 days of treatment **(Zahir *et al*., 2012)**.

(Chakravarthy *et al.* 2012) found that nanoparticles of titanium oxide 2TiO, silver AgNPS and cadmium sulphate Cds at a concentration of 2400 ppm used to control the cotton leafworm *Spodoptera lituralis* had a mortality rate of (73, 56 and 93%), respectively. **(Al-Shammari, 2015)** showed that silver nanoparticles prepared with aqueous extract of eucalyptus leaves at a concentration of 4250 ppm were used to control mealybugs, with a mortality rate of adults reaching 66.7% within 72 hours of treatment. **(Al-Shujairi, 2018)** prepared silver nanoparticles using alcoholic extract of myrtle leaves and used them against nymphs and adults of cotton A. gossypii, with a mortality rate of 99.2% after 72 hours of treatment.

(Najib, 2019) found that silver nanoparticles prepared from the cold aqueous extract of *N. oleander* and *Ricinus cmmunis* leaves had an effect in inhibiting the eggs and mortality the larvae of the greater wax moth *G. mellonella*, as the percentage of egg-stage inhibiting at a concentration of 4 mg/ml reached 98.56% and 100% for the two plants, respectively, and the percentage of mortality the first-stage larvae reached 92.3% and 100%, respectively, at the same concentration.

The importance of nanoparticles in controlling the grain beetle *T. granarium*:

Al-Hayali (2018) proved that treating wheat grains with nanoparticles EoEC-AAgNPs prepared from *Eucalyptus camaldulensis* oil and *Artemisia herba alba* at a concentration of 8000 ppm influenced mortality in the larvae of the grain beetle *T. granarium* with a mortality rate of 96.66% after 48 hours of treatment. The nanoparticles Agnps-Eoec prepared from Artemisia herba alba extract and *E. camaldulensis* oil, which were mixed with wheat grains at a concentration of 8000 ppm, moralized the larvae of the grain beetle *T. granarium* with a mortality rate of 96.66% within 48 hours **(Al-Hayali, 2018)**. **(Almadiy *et al*. 2018)** found that the use of silver nanoparticles prepared using the alcoholic extract of Peganmum harmala seeds was highly effective in killing larvae and adults of the hairy grain beetle *T. granarium*.

Uses of iron nanoparticles in insect control:

Iron nanoparticles are an environmentally friendly magnetic nanomaterial used as a pesticide against the larvae of the Egyptian mosquito *Aedes aegypti*. Its nanomaterial consists of two materials (SAMNs&chlorin), which are iron oxide nanoparticles γ-Fe_2O_3 and are called surface active nanoparticles SAMNs representing the core (Surface Active Maghemite Nanoparticles_SAMNs) and chlorin that forms the shell (chlorin-e6). It has a toxic effect on the larvae, and the material is characterized by its chemical effectiveness and stability and is a safe alternative to synthetic insecticides and has no harm to the environment **(Magro *et al*., 2019)**. **(Chen *et al*., 2014)** studied the toxic effect of iron nanoparticles on the fruit fly *Drosophila melanogaster*. **Reda *et al.* (2016)** conducted a study to evaluate the effectiveness of iron oxide nanoparticles (α-Fe_2O_3) against the larvae of the mosquito *C. pipiens*. The particles were prepared chemically using low temperature and had a lethal effect on larvae in the laboratory at concentrations of 0.025, 0.25, 2.5, and 25 ppm of the nanomaterial, as the larval mortality rates reached 6.67, 23.34, 35, and 68.34% for the four concentrations, respectively, after 48 hours of treatment.

Iron nanoparticles were used as an alternative to chemical pesticides to study their effectiveness against the larvae and pupae of the mosquito *Culex quinquefasciatus*, Iron nanoparticles Fe0 were prepared using the aqueous extract of *Ficus natalensis*, and iron oxide nanoparticles (Fe_2O_3) were prepared chemically. Both nanomaterials proved their efficiency in mortality the larvae and pupae of mosquitoes **(Murugan *et al*., 2018)**. **(Zhou *et al.* ,2012)** conducted a study to evaluate the toxicity of iron oxide nanoparticles against the discoid-cockroach Blaberus discoidalis by injecting iron nanoparticles into the central nervous system of the insect. The results showed a decrease in the activity of the treated insects, which proves that iron oxide nanoparticles caused neurotoxicity to the insects, which led to a decrease in their survival rate after 40 days of injection.

Iron nanoparticles (magnetite Fe3O4) showed high toxicity against fruit fly *D. melanogaster* by reducing their lifespan by 30% at low concentrations **(Vecchio *et al*., 2013)**. **(Vega-Alvarez *et al.* 2014)** confirmed that iron oxide nanoparticles caused the highest level of lethality in *Drosophila embryos* when compared with 8 other types of nanoparticles. A study was conducted on the effectiveness of nano-iron prepared with the alcoholic extract of *Dodonaea viscosa* L. leaves in influencing some aspects of the insect's life and its effect on the lethality of the stages of the greater wax moth *G. mellonella* L., using concentrations of 2, 5, and 10%. The direct treatment of the insect stages and the indirect treatment of the larval food proved the effectiveness of nano-iron particles compared to the alcoholic extract of Dodonaea leaves, as the percentage of inhibition of hatching of eggs treated superficially and at the age of 24 hours increased to 87.89, 91.13, and 93.34%, respectively, for the nano-preparation, while it reached 78.95, 85.86, and 87.04% for the concentrations, respectively, for the alcoholic extract, and it had a significant effect on some of the insect's life phenomena **(Al-Jubouri, 2020).**

CONCLUSION

Extracts and nano-preparations from Diphla plant sources can be used in the field of insect control in general and store insects in particular after recording a significant effect in lethality insects directly or indirectly (**Sahi, 2019**) and can be used in the field of integrated control.

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