

Study of the Effect of Nano Iron Oxide on Biological Features of *Echinococcus Granulosus* and Comparing it with the Aqueous Extrat of the Colocynth Plant

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Abstract: Hydatidosis disease is a common disease between humans and animals and is caused by the larval stage of the tapeworm *echinococcus granulosus*, which is endemic in Iraq and neighboring countries. Therefore, the current study aimed to find out the effect of extracts of bitter melon and iron oxide nanoparticles on the protoscolex of *E. granulosus*. Isolated from the livers of infected sheep. The protoscolex isolated from the liver of sheep infected with aqueous sacs were incubated with extracts prepared in different concentrations at different intervals of 60, 30, 15, 10, 5 minutes. The average concentration of iron oxide nano, as the average concentration showed that there is a significant difference between the concentrations used to kill the vitality of the heads, and the concentration of 0.100 mg / ml achieved the highest killing rate, which amounted to 65.00%, which differed significantly from the concentrations of 0.050 and 0.025 mg / ml, which recorded a killing rate of 49.80% and 60.60% respectively. The alcoholic extract of the bitter melon plant was used from it concentrations of 1000, 500, 250 mg / ml The concentration of 1000 mg / ml differed significantly from the rest of the concentrations with a killing rate of 65.00%. Thus, the study indicates the effective role of iron oxide and alcoholic extract of the bitter melon plant in influencing the vitality of the initial heads of the beloved *echinococcus*.

Introduction

Hydatidosis is a serious health and epidemiological problem in most parts of the world, first described by Hippocrates as the bladder full of water (Aydin et al., 2022). Hydatid disease is a common human-animal disease Zoonosis, which is the larval stages of tapeworm. Tapeworm dating back to sex . (Zaffarano et al., 2023) *Echinococcus*

The *E. granulosus* parasite needs hosts to complete its life cycle: a definitive host, which is represented by carnivores from the canine family Canidae, and an intermediate host, represented by herbivores (Smyth, 1987). The cause of the disease in humans and intermediate hosts is their ingestion of food contaminated with tapeworm eggs (Tahir, 2022).). The seriousness of hydatid cyst disease and its health importance lies in the fact that the pathological symptoms do not appear in its initial stages and there is no evidence of infection with the disease until after a period that may reach several years due to the slow growth of the cyst as well as the increase in its numbers and large size, so the pathological symptoms of hydatid cysts appear due to the pressure of the cyst on the tissues adjacent to it (Pal et al., 2022) and the seriousness of the disease, as well as the slow appearance of symptoms, lies in the treatment being limited to the removal of water cysts by surgical intervention, which is The most effective treatment (Lightowers et al., 2021).

The importance of nanomaterial in its distinctive quantitative properties due to its small size and large surface. The ratio of the surface area of the nanomaterial to its mass is much greater than the same ratio in large molecules, which leads to a rapid chemical reaction (Khan et al., 2019).

However, the indiscriminate and excessive use of manufactured chemical pesticides caused pollution and damage in many environmental aspects, as well as the emergence of resistance against many pesticides and what requires the search for more toxic pesticides and more effective methods of control and this encouraged researchers to reduce reliance on chemical pesticides and search for new alternatives, so attention was directed to plants and the use of plant extracts because plants contain effective and toxic compounds against parasites of medical importance (Sohani, 2019).

The bitter melon plant is one of the plants of medical importance because it contains many effective and bioactive compounds that made this plant an important medicinal plant that enters into many pharmaceutical industries in addition to being a good nutrient as it contains glycosides, alkaloids, flavonoids, oils, fatty acids, resins, sapons, pectins, choline, proteins and carbohydrates. They are a mixture of glycosidic and alkaloid substances and an alcoholic called citrollol khatri et al., (2020).

Materials and methods

Collection of hydatid cyst samples and isolation of heads

Hydatid cyst samples were collected from the liver of naturally infected sheep. The infected livers were transported to the animal laboratory in Baghquba district / Diyala, by plastic bags in a container containing crushed ice, and were dealt with directly following the method (1980) Smyth and Barrett . The affected organs were washed with water for the purpose of getting rid of blood and suspended materials from the slaughter process and put the liver in a sterile dish and the outer surface of the bag was sterilized with 70% ethyl alcohol and then withdrew the largest amount of hydatid fluid Containing the protoscolex using a 3 ml medical syringe. Using sterile scissors, the bag was pierced and the bag was washed with a phosphate brackish buffer solution to remove the rest of the protoscolex deposited or stuck in the bag. After collecting the heads they were placed in test tubes for the purpose of deposition.

Collection of botanical models:

The fruits of the *Citrullus colocynthis* plant were collected from the central regions of Iraq (Mandali District - Diyala Governorate) during the autumn season 2022, as the fruits were washed with water well and then dried in shade after being placed in clean pots at laboratory temperature with continuous stirring to prevent rotting, then grinding well using an electric grinder, and kept in sterile and sealed containers, in moisture-free conditions until the plant extracts from it start working.

Preparation of alcoholic extract

75 g of the vegetable model was weighed and then placed in a glass flask containing 750 ml of ethanol, shake the flask in order to homogenize the mixture, leave the flask on the Shaker shaker for 24 hours, then filter the mixture using several layers of medical gauze to remove the large plant parts stuck in it, filtered again using Millipore filter papers with a diameter of 0.45µm to prevent the passage of impurities with the filtrate. Then put the mixture in an electric oven at a temperature of 40 ° C until the evaporation of ethanol The entire dry powder is deposited at the bottom, the dry extract is kept in clean sealed bottles in the refrigerator at a temperature of 4 ° C until use.

Results and discussion

Effect of Iron Oxide Nanoparticles on the Biology of Echinococcus granulosus Protoscolex Isolated from Sheep and its Relationship with Time

The results in Table (1) show the significant effect of the study factors (concentrations of iron oxide nanomaterial and time) and their interference in the vitality of the protoscolex of the granular echinococcus echinococcus granulosus, as the average concentration showed that there is a significant difference between the concentrations used to kill the vitality of the heads and the concentration of 0.100 mg / ml achieved the highest killing rate, which amounted to 65.00%, which differed significantly from the concentrations of 0.050, 0.025 mg/ml who recorded a killing rate of 49.80% and 60.60% respectively. From the same table, we find that there is a significant difference between the average time in killing the vitality of the heads, as the time recorded 60 minutes the

highest rate of killing, amounting to 97.66, morally superior to the rest of the times. As for the effect of interaction between the two factors, the treatment showed a concentration of 0.100 mg/ml with a time of 60 minutes and a treatment with a concentration of 0.050 mg/ml with a time of 60 Minute the highest killing rate was 98% and 100% respectively, which differed significantly from the rest of the transactions. While the treatment at a concentration of 0.025 mg/ml with a time of 5 minutes recorded the lowest killing rate, which amounted to 9%.

Table (1) Effect of Iron Oxide Nanoparticles on the Biology of Protoscolexof Echinococcus granulosus isolated from sheep and its relationship with time

Percentage of Killing by Concentration				
Average time	Concentration 0.1	Concentration 0.05	Concentration 0.025	Focus Time
13.33 E	17 h	14 Hi	9 i	5 minutes
35.66 D	41 ef	38 f	28 g	10 minutes
61.00 C	74 c	66 d	43 e	15 minutes
84.66 B	93 father	87 b	74 c	30 minutes
97.66 A	100 a	98 a	95 a	60 minutes
	65.00 a	60.60 A	49.80 b	Average concentration

Lowercase letters that are similar horizontally mean that there are no significant differences between them.

Vertically similar capital letters mean that there are no significant differences between them.

We find from the table that the best concentration of iron oxide nanoparticles in killing Echinococcus granulosus is 0.100 mg/ml at 60 minutes and therefore the relationship is direct with time and may attribute the cause of killing Nanoparticle complexes have a lethal effect against bacteria, fungi and parasites, making these complexes a large part in the proliferation of the drug (Huang et al., 2019). The results of the study were similar to the results of the study Do Carmo Neto et al. (2022) in the antiparasitic and anthelmintic activity of zinc oxide nanoparticles and proved the effective role of reactive oxygen catalysis that affect the balance of the pathogen. The results converged with the study of Shnawa et al. (2022) in the insecticidal properties of primers and biocompatibility of biosynthetic zinc oxide molecules from the leaves, where the concentration of 400 mg with a time of 60 minutes obtained the highest killing rate of 100%. The results of the study were similar to Albalawi et al. (2020) on the high efficacy of organic and inorganic nanoparticles for the treatment of echinococcosis cystic in vitro The results showed the wide effectiveness of organic and inorganic particles and the most commonly used in the treatment of hydatid cysts are metal nanoparticles. The results of the study were similar to the study of Abd-Algany (2022) on the effect of titanium dioxide nanoparticles against leishmania in the laboratory and different concentrations of 0.5, 1, 5, 10 mg/ml and particle size of 10, 100 nm and the results showed the effective effect of concentrations within 48 hours and size 10 was more effective in stabilizing parasite growth than size 100 nm.

Effect of Plant Alcoholic Extract Concentrations on the Vitality of Protoscolexof Echinococcus granulosus isolated from sheep and its relationship with time

The results of the statistical analysis of the average concentration in Table (2) on the effect of alcoholic extract of bitter melon plant on the vitality of protoscolex show that the concentration of 1000 mg / ml differed significantly from the rest of the concentrations with a killing rate of 65.00%.

As for the effect of the average time on the vitality of the heads, the time of 60 minutes achieved the highest average in killing of 94.33%, outperforming the rest of the times. As for the effect of factor interaction, the treatment showed a concentration of 1000 mg/ml with time 60 The highest killing rate was 100% minute, which differed significantly from all treatments, while the treatment at a concentration of 250 mg/ml was recorded with a time of 5 minutes, and the lowest killing rate was 4%.

From the table we find the best concentration for killing the protozoa of echinococcus granuloma E. Granulosus with alcoholic extract bitter melon 1000 mg at the time of exposure is 60 minutes, as the killing rate was 100%. The reason for the killing of primroses is due to the effect of the biological active substance of the active ingredients in the plant that have affected the physiology of the protozoa through the cessation of metabolic processes or their effect on enzymes, proteins and nucleic acids and the cessation of cell metabolic cycles that occur within the proprioritors.

The effects of water-soluble extracts and ethanol may have great potential in the development of new drugs for the treatment of Cystic echinococcosis (Yan, 2022).

Table (2) Effect of Alcoholic Extract Concentrations of Bitter Melon Plant on the Vitality of the Protoscolex of Echinococcus granulosus isolated from sheep and its relationship with time

Percentage of Killing by Concentration				
Average time	concentration 1000 mg	concentration 500 mg	concentration 250 mg	Focus Time
11.00 E	14 h	12 h	4 h	5 minutes
38.66 D	43 ef	39 f	34 g	10 minutes
58.00 C	77 c	47 This	50 d	15 minutes
82.33 B	91 b	83 c	73 c	30 minutes
94.33 A	100 a	93 b	90 b	60 minutes
	65.00 a	54.80 b	50.80 b	Average concentration

Lowercase letters that are similar horizontally mean that there are no significant . . differences between them.

Vertically similar capital letters mean that there are no significant differences between . them.

The results of the study were similar to those of Albani et al. (2022) Effect of Stevia multiristata on the vitability of primers of echinococcus granulomatosis The effective concentration was 50, 100 mg in vitro. The results converged with the laboratory study Abed and Ibrahim (2021) Ethanolic extracts of some medicinal plants against echinococcus granulomatosis as an environmentally friendly system, where the heads were exposed to different concentrations of 12.5, 25, 50, 75 mg for 10, 20, 30 minutes and after exposure for 20 minutes at a concentration of 75 Amalgam led to a significant decrease in the vitality of the heads. The results were also similar to the study of Hesari et al. (2020) in the laboratory study of pumpkin seed extracts Cucurbita moschata in the vitality of the protoscolex of granulocytes with different concentrations of 0.1, 1, 10, 100 mg and times of 5, 15, 30, 60 minutes and the highest killing rate was at a concentration of 100 mg with a time of 60 minutes.

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