# Effect of Cobalt Oxide on the Vitality of the Protoscolex of Echinococcus granulosus Isolated from Middle Hosts

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#### Abstract

Hydatid cysts disease is a common disease between humans and animals and is caused by the larval phase of the tapeworm Echinococcus granulosus, which is endemic in Iraq and neighboring countries. Therefore, the current study aimed to find out the effect of cobalt oxide nanoparticles on the protoscolex of E. granulosus. Isolated from different hosts. The isolated protoscolex were incubated from the liver of sheep infected with aqueous bags with extracts prepared in different concentrations at different time intervals 60, 30, 15, 10, 5 minutes. As for the aqueous extract CO, it used concentrations of 0.100, 0.050, 0.025 mg / ml, where the treatment was shown in sheep livers with a concentration 0.100 mg/ ml with time 60 minutes and treatment with concentration 0.050 mg/ml with time 60 minutes Highest Kill Rate 96%, 100% In cattle livers, concentrations of 0.025, 0.050 mg/ml with a time of 60 minutes and concentration of 0.100 mg/ml with a time of 60 and 30 showed the highest homicide rates of 95%, 98%, 100% and 92%, respectively. In goat livers, the concentration showed 0.025 mg/ml with 60 minutes, concentration 0.050 mg/ml with 30 and 60 minutes, and concentration 0.100 mg/ml with 30 and 60 minutes, 100%, 100%.

#### INTRODUCTION

Hydatidosis is a serious economic, health and epidemiological problem in most parts of the world (Aydin et al., 2022). Hydatid cyst disease is a common disease between humans and animals Zoonosis It is a larval phase that belongs to the genus Echinococcus . (Zaffarano et al., 2023) Needs E granulosus hosts to complete its life cycle, a definitive the canine family, canidae host from carnivores, and an intermediate host. represented by herbivores . (Smyth, 1987). of the disease and its health importance lies in the fact that the symptoms do not appear in its initial stages and there is no evidence of infection with the disease until after a period, which reaches 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 etal., 2022) The seriousness of the disease, as well as the slow onset of symptoms, lies in the limited treatment of surgical removal of aqueous cysts, which is the most effective treatment (Lightowlers etal., 2021).).

The importance of the nanomaterial in its distinct quantum properties due to its small size and large surface. Also, 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 the speed of chemical reaction (Khan etal., 2019). Sensing Cobalt nanoparticles are commonly treated as magnetic nanoparticles, with little research

focused on verifying the usefulness of particles compared to other nanoparticles (Mei etal., 2019)..

#### Materials and methods

Samples of the Hydatid cysts were collected from the liver of naturally infected sheep. The affected livers were transported to the animal laboratory in Mansouriya / Diyala district, 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 placing the liver in a sterile dish and the outer surface of the bag was sterilized with ethyl alcohol 70% The largest amount of hydraid fluid containing the protoscolex was then withdrawn using a 3 ml medical syringe. Using sterile scissors, the bag was punctured and the bag was washed with a salt phosphate buffer solution to remove the rest of the protoscolex deposited or stuck in the bag. After collecting the protoscolex, they were placed in test tubes for the purpose of sedimentation.

### Co Chemicals used

Dissolve 0.9 g of cobalt aqueous chloride salt CoCl 2.6H2 O in 50 ml non-ionic water and put the solution on a magnetic motor for 30 minutes until the substance is completely dissolved, prepare a solution of sodium hydroxide NaOH by dissolving 1.6 g of it in a quantity of distilled water and then complete the volume to 200 ml non-ionic water, add 150 ml of this solution to the cobalt solution with continuous stirring using the engine and at a temperature of 80  $^{\circ}$  C, while monitoring that the pH does not rise more than 9 throughout the reaction period, dry the precipitate for 30 minutes at a temperature of 80 ° C and then burn at a temperature of 400 ° C for 2 hours and leave in the dryer for 24 hours at room temperature Figure (3. 3), and the appropriate

Stock Solution was obtained at a concentration of 0.1 mg / ml and from it the rest of the concentrations (0.025, 0.050) mg/ml were prepared using the law of V2 C2= V1 C1. (Barnawi,2020)

#### Structural Tests Results

#### - Diffraction No and Cobalt Diffraction Co

The crystal structure and phase purity of the cobalt oxide nanoparticles Co 3 O 4 prepared by X-ray diffraction as shown in Figure (1) The X-ray diffraction spectrum of the prepared cobalt oxide was matched with the standard spectrum of cobalt nanooxide Co 3 O 4 Cube by database (JCPDS 42-1467) The formation of cubic crystalline cobalt nanooxide was detected by diffraction peaks (37.40, 43.45, 62.95, 75.40 and 79.45), no other impurities were observed in the X-ray diffraction spectrum of cobalt nano oxide indicating that Co3O4 was obtained. Pure. The average crystal volume of cobalt nano oxide Co3O4 was calculated and was 22.70 nm using the Debye-Scherer formula))

- Diagnosis of cobalt oxide nanoparticles by (SEM)

The morphological and structural structures of Cobalt Oxide nanoparticles Co 3O4 were studied using scanning electron microscopy (SEM). Figure 2 shows that the nanoparticles were prepared in the nanometer range. SEM images indicated indicated that some of the nanoparticles were well separated from each other while most were present in a lumpy This agglomeration is due form. to electrostatic effects as well as the drying effect of the aqueous suspension. Similar behavior has also been reported for nanoparticle agglomeration

Figure 1 Scanning electron microscope image. Figure 2: X-ray diffraction model



**Results and discussion** 

The results in Table (1) show the effect of cobalt oxide nano concentrations on the vitality of the protoscolex of E. granulosus isolated from sheep The significant effect of the study factors (cobalt oxide nanomaterial concentrations + time) and their interference in the vitality of the protoscolex of the granular worm E. granulosus, as the average concentration showed that there is а difference significant between the concentrations used to kill the vitality of the protoscolex and the concentration achieved 0.100 mg / ml the highest killing rate, which reached 63.00%, which differed significantly from the concentrations of 0.050, 0.025 mg / ml, which recorded a killing rate of 50.00% and 56.20%. Respectively.

The reason for killing the protoscolex with cobalt oxide nano concentrations may be attributed decrease to а in surface agglomeration and providing more surface area to interfere with parasitic membranes, leading to greater toxicity. The results of the treatment of extracorporeal protoscolex with cobalt oxide nano extract were similar to the results obtained by Mohammed (2022) Effect of selenim nano the vitality of the protoscolex of E. granulosus in the laboratory, seven concentrations of 50, 100, 150, 200, 250, 350, 500 µg/ml were used for different exposure times of 10, 20, 30 and 60 respectively and the concentration of 350, 500 with 60 time obtained 100% kill. The results of the current study are consistent with those obtained by Shnawa et al. (2021) in the activity of zinc oxide nanoparticles biosynthesized from mint leaves against the protoscolex of E. granulosus, which obtained a 100% kill rate at a concentration of 400 ppm for the time of 150 minutes

Table (1) Effect of Cobalt OxideNanoconcentrations on the Vitality of theProtoscolex of Echinoccocus granulosusIsolated from Sheep

| Time      | Concent | Concent | Concent | Aver  |
|-----------|---------|---------|---------|-------|
| Concentr  | ration  | ration  | ration  | age   |
| ations    | 0.025   | 0.05    | 0.1     | time  |
| 5 minutes | 8       | 12      | 18      | 12.67 |
|           | Ι       | Ι       | h       | Ε     |
| 10        | 30      | 35      | 37      | 34.00 |
| minutes   | G       | fg      | f       | D     |
| 15 min    | 50      | 55      | 70      | 58.33 |
|           | E       | е       | d       | С     |
| 30        | 71      | 83      | 90      | 81.33 |
| minutes   | D       | С       | b       | В     |
| 60        | 91      | 96      | 100     | 95.67 |
| minutes   | В       | а       | а       | Α     |
| Medium    | 50.00   | 56.20   | 63.00   |       |
| concentr  | С       | b       | a       |       |
| ation     |         |         |         |       |

The results in Table (2) show the effect of cobalt oxide nano concentrations on the vitality of the protoscolex of E. granulosus isolated from cows The significant effect of

the study factors (cobalt oxide nanomaterial concentrations + time) and their interference in the vitality of the protoscolex of the granular worm E. granulosus), as the average concentration showed that there is a significant difference between the concentrations used to kill the vitality of the protoscolex and the concentration /achieved 0.100 mg / ml the highest killing rate, which amounted to 64.60%, which differed significantly from the concentrations of 0.050 and 0.025 mg / ml, which recorded a killing rate of 49.60% and 60.60%. respectively

Table (2) Effect of Cobalt Oxide Nano<br/>concentrations on the Vitality of<br/>Protoscolex of Echinoccocus granulosus<br/>Isolated from Cows

| Focus    | concentr | concentr | concentr          | Aver  |
|----------|----------|----------|-------------------|-------|
| Time     | etien    | etien    | concentr<br>a4ion |       |
| Ime      | ation    | ation    | auon              | age   |
|          | 0.025    | 0.05     | 0.1               | time  |
| 5        | 7        | 12       | 16                | 11.67 |
| minutes  | Ι        | hi       | h                 | Ε     |
| 10       | 26       | 39       | 40                | 35.00 |
| minutes  | G        | f        | ef                | D     |
| 15 min   | 45       | 65       | 75                | 61.67 |
|          | E        | d        | с                 | С     |
| 30       | 75       | 89       | 92                | 85.33 |
| minutes  | С        | b        | ab                | В     |
| 60       | 95       | 98       | 100               | 97.67 |
| minutes  | Α        | a        | a                 | Α     |
| Medium   | 49.60 b  | 60.60 a  | 64.60 a           |       |
| concentr |          |          |                   |       |
| ation    |          |          |                   |       |

The cause of the killing may attribute the nanoparticle complexes to have a lethal effect against bacteria, fungi and parasites, making those complexes a large part of the reproduction of the drug Huang etal.,2019). The results of the study were similar to the results of the do Carmo Neto study et al. (2022) in the anti parasitic and anthelmintic activity of zinc oxide nanoparticles and demonstrated the effective role of reactive oxygen dehydrophilation affecting pathological factor balance.

The results in Table (3) show the effect of cobalt oxide nano concentrations on the vitality of the protoscolex of the E. granulosus isolated from goats The significant effect of the study factors (cobalt oxide nanomaterial concentrations + time) and their interference in the vitality of the protoscolex of the E. granulosus worm as the average , concentration showed that there is а significant difference between the concentrations used to kill the vitality of the protoscolex and the concentration /achieved 0.100 mg / ml the highest killing rate, which amounted 70.40%, which to differed significantly from the concentrations of 0.050 and 0.025 mg / ml, which recorded a killing rate of 56.80% and 64.60%. Respectively. The killing cause is attributed to the effective effects of nanoparticles against the protoscolex of E. granulosus (Norouzi, 2020).

Table (3) Effect of Cobalt Oxide Nano<br/>concentrations on the Vitality of<br/>Protoscolexof Echinoccocus granulosus<br/>Isolated from Goat

| Focus    | concentr | concentr | concentr | Aver  |
|----------|----------|----------|----------|-------|
| Time     | ation    | ation    | ation    | age   |
|          | 0.025    | 0.05     | 0.10     | time  |
| 5        | 20       | 15       | 22       | 19.00 |
| minutes  | Hi       | i        | h        | Ε     |
| 10       | 35       | 43       | 45       | 41.00 |
| minutes  | G        | f        | f        | D     |
| 15 min   | 53       | 70       | 85       | 69.33 |
|          | Ε        | d        | bc       | С     |
| 30       | 80       | 95       | 100      | 91.67 |
| minutes  | С        | ab       | a        | В     |
| 60       | 96       | 100      | 100      | 98.70 |
| minutes  | Α        | Α        | a        | Α     |
| Medium   | 56.80 c  | 64.60 b  | 70.40 a  |       |
| concentr |          |          |          |       |
| ation    |          |          |          |       |

The results of the study were similar to the study of Kalpana et al. (2020) in strengthening the control of malaria with zinc nano oxide and its vectors Anopheles and its parasite Plasmodium falciparum, and the study concluded that zinc oxide prepared from L. siceraria peel A valuable green option in the **Reference** 

- Aydin, Y., Ulas, A. B., Ahmed, A. G., & Eroglu, A. (2022). Pulmonary Hydatid Cyst in Children and Adults: Diagnosis and Management. The Eurasian journal of medicine, 54(Suppl1), 133-140.
- Barnawi, N. A. M. (2020). Preparation, Characterization and Spectroscopic Studies of Metal Nanoparticles and Their Applications in The Environment.
- do Carmo Neto, J. R., Guerra, R. O., Machado, J. R., Silva, A. C., & da Silva, M. V. (2022). Antiprotozoal and anthelmintic activity of zinc oxide Nanoparticles. Current Medicinal Chemistry, 29(12), 2127-2141.
- Huang, T., Holden, J. A., Heath, D. E., O'Brien-Simpson, N. M., & O'Connor, A. J. (2019). Engineering highly effective antimicrobial selenium nanoparticles through control of particle size. Nanoscale, 11(31), 14937-14951.
- Kalpana, V. N., Alarjani, K. M., & Rajeswari,
  V. D. (2020). Enhancing malaria control using Lagenaria siceraria and its mediated zinc oxide nanoparticles against the vector Anopheles stephensi and its parasite Plasmodium falciparum. Scientific Reports, 10(1), 1-12.
- Kania-Dobrowolska, M., & Baraniak, J. (2022). Dandelion (Taraxacum officinale L.) as a source of biologically active compounds supporting the therapy of co-existing diseases in metabolic syndrome. Foods, 11(18), 2858.
- Lightowlers, M. W., Gasser, R. B., Hemphill, A., Romig, T., Tamarozzi, F., Deplazes, P., ... & Kern, P. (2021). Advances in the treatment, diagnosis, control and scientific understanding of taeniid cestode parasite infections over the past 50 years.

control of malaria vectors and parasites.

International journal for parasitology, 51(13-14), 1167-1192.

- Mohammed, S. A., & Ali, A. A. (2022). Effect of selenium nanoparticles against protoscoleces of Echinococcus granulosus in vitro and hydatid cysts in mice. Iraqi Journal of Veterinary Sciences, 36, 195-202.
- Mei, J., Liao, T., Ayoko, G. A., Bell, J., & Sun, Z. (2019). Cobalt oxide-based nanoarchitectures for electrochemical energy applications. Progress in Materials Science, 103, 596-677.
- Norouzi, R., Ataei, A., Hejazy, M., Noreddin, A., & El Zowalaty, M. E. (2020). Scolicidal effects of nanoparticles against hydatid cyst protoscolices in vitro. International journal of nanomedicine, 1095-1100.
- Pal, M., Alemu, H. H., Marami, L. M., Garedo, D. R., & Bodena, E. B. (2022). Cystic Echincoccoosis: A Comprehensive Review on Life Cycle, Epidemiology, Pathogenesis, Clinical Spectrum, Diagnosis, Public Health and Economic Implications, Treatment, and Control.
- Shnawa, B. H., Hamad, S. M., Barzinjy, A. A., Kareem, P. A., & Ahmed, M. H. (2021).
  Scolicidal activity of biosynthesized zinc oxide nanoparticles by Mentha longifolia L. leaves against Echinococcus granulosus protoscolices. Emergent Materials, 1-11.
- Smyth, J. D. (1987). Changing concepts in the microecology, macroecology and epidemiology of hydatid disease. Helminth Zoonoses, 1-11.
- Smyth, J. D., & Barrett, N. J. (1980). Procedures for testing the viability of human hydatid cysts following surgical removal, especially after chemotherapy.

Transactions of the Royal Society of Tropical Medicine and Hygiene, 74(5), 649-652.

- Tahir, K. Y. (2022). Hydatid cystic disease in man.salahaddin university-Erbil .
- Zaffarano, G. P., Miambo, R. D., Ussivane, É.
  E., Poglayen, G., Morandi, B., Mukaratirwa, S., & Afonso, S. S. (2023).
  Cystic echinococcosis in cattle (Bos taurus) from rural communities of Limpopo National Park, Gaza province, Mozambique: a One Health perspective.
  Rendiconti Lincei. Scienze Fisiche e Naturali, 1-10.