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Response Some Types of Parasites to the Influence of Some Heavy Metals Ions under Laboratory Conditions.

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ABSRACT

Used parasites *Naegleria fowleri, Acanthamoeba sp. , Balantidium coli* in the current study to determine the effect of ions of some heavy metals (copper, aluminum, Zinc, manganese, iron) on growth under laboratory conditions in culture media. The addition of copper in culture media to kill or inhibition of parasite growth *Naegleria fowleri, Acanthamoeba sp. , Balantidium coli* ,both separately. While the addition of both aluminum , zinc, manganese and iron to encourage growth and increase the number of parasite. Show that the use of ions concentration (40 mg / L) led to the killing of parasites while was observed increasing the numbers in the case of addition of ions concentration (20 mg / L) compared to the control group.

Keywords: parasites, heavy metals, laboratory.

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INTRODUCTION

Subtract heavy elements into the environment from many sources: including industrial waste and agricultural waste and household and other affect those elements positive effects and other negative growth of organisms in general and microbiology in particular, such as parasites and effect of different elements depending on the heavy element and focus on the environment and the period of exposure object to him [1].

Protozoa are still important a worldwide public health problem. *Naegleria* is an ameba commonly found in warm freshwater and soil. Only one species of *Naegleria* infects people, *Naegleria fowleri*. It causes a very rare but severe brain infection [2].

Infection with *Naegleria* causes the disease primary amebic meningoencephalitis (PAM), a brain infection that leads to the destruction of brain tissue. In its early stages, [3]

Acanthamoeba species are commonly found in air, soil, and water. Their ubiquity is illustrated by the fact that 80% of the human population carries antibodies against *Acanthamoebae* infection.[4]

Acanthamoeba spp. are also named as a causative agent of granular amoebic encephalitis (GAE), an opportunistic, chronic infection of the nervous system. The first cases of GAE were diagnosed in 1972 by Jager and Stamm. [5]

Balantidium is the only ciliated protozoan known to infect humans. Balantidiasis is a zoonotic disease and is acquired by humans via the feco-oral route from the normal host, the pig, where it is asymptomatic. Contaminated water is the most common mechanism of transmission[6].

There are several elements affect the growth of parasites, variously including elements (copper, iron, manganese and zinc) and those elements are toxic and cause problems when subtract to the environment[7]

As found[8] that *Naegleria fowleri , Entamoeba histolytica* ability to growth the presence of iron and moderate density as iron works to increase the density of growth in culture media while the copper negative effect works on the inhibition growth *Naegleria fowleri* in the environment

Amoeba is one of the most important protozoa the aim of this study was to determined response of three type of protozoa *Naegleria fowleri* , *Acanthamoeba* sp. ،*Balantidium coli* against (copper, aluminum, Zinc, manganese, iron)

MATERIALS AND METHODS

Washed a large group of cultures of *Acanthamoeba sp*. *Balantidium coli* and *Naegleria fowleri* containing phase (cyst and trophozoite) with sterile water and collected in sterile tubes both separately and conducted a process centrifuges by at 4000 cycle / min for

five minutes, washing the sediment with sterile water and returned to him the process of centrifugal 2-3 times, collect sediment and add his 2 ml of sterile water and after good shaker , calculated number of the parasite in suspension by Hemacytometer using a compound microscope[9]

Prepared Soil extract medium according to (Himedia company , India)

17.750 g of clay soil and add (1000 ml) of distilled water and good shaker, then leaved a period to allow the deposition of Minutes and suspended impurities to the bottom, taking of Leaky soil and dissolved 15 g of Agar Sterility media in the Autocleave temperature 121 ° C for 15 minutes

Prepared Page solution based on[10] **of the following materials dissolved in a liter of distilled water**:

NaCl	0.12g
MgSO4.7H2O	0.004 g
CaC12 • 2H2O	0.004 g
Na2HPO4	0.142 g
KH2PO4	0.136 g

With the addition of 200 units / ml of each of the Penicillin and Streptomycin cultivated all media with 1 ml of suspension parasites both separately containing 800 phaes / Mm^3 and added(2 ml)of Page solution, and then add ions of heavy metals (Zn, Cu, Al, Fe, Mn) are salts($znso_4$, $cuso_4$, Al_2o_3 , $Feso_4$, $Mnso_4$) dissolved in distilled water free of ions after sterilization culture media user. The added concentrations (20 mg / L) , (40 mg / L) for each element respectively, while taking control of the sample to not contain any ions and by 3 replications for each group., And after 24 hours calculated numbers parasite daily for four days.

RESULTS

Test the direct effect of ions of heavy metals on the growth rates of the parasite *Naegleria fowleri* in vitro

Proved the results of the current study, the direct effect of heavy metals on the growth of the parasite *Naegleria fowleri* The effects of ions (aluminum, zinc ,manganese ,iron) big as it led added to obtain a significant increase in the numbers of the parasite while the effects of ions copper negative as it has led to a decrease in the numbers of the parasite compared to Control group (Table 1)

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Table (1) the effect of ions of heavy metals (copper,, aluminum, Zinc, Manganese iron) on the growth rates of the parasite *Naegleria fowleri* in vitro

S	Elements	Rates of parasite numbers Mm^3			
		24 hours	48 hours	72 hours	96 hours
1	Control	487	737	1037	1456
2	**Cu1**	300	387	406	700
3	**Cu2**	243	300	368	531
4	**Al1**	2012	2062	2131	3206
5	**Al2**	1762	2043	2031	2568
6	**Zn1**	1537	1768	2250	2443
7	**Zn2**	1393	1618	2062	2237
8	**Mn 1**	1062	1456	1662	2018
9	**Mn 2**	1031	1275	1600	1875
10	**Fe1**	718	1012	1350	1806
11	**Fe2**	581	781	1156	1406

1= Concentration of 20 mg / l
2 = concentration of 40 mg / l

Test the direct effect of ions of heavy metals on the growth rates of the parasite *Acanthamoeba* sp in vitro

Proved the results of the current study, the direct effect of heavy metals on the growth of the parasite *Acanthamoeba sp* was observed when adding copper to the culture media -killing parasite few of numbers compared to control group while the effects of ions (aluminum, zinc ,manganese ,iron) large as led added to obtain a significant increase in the numbers of the parasite (Table 2)

Table (2) the effect of ions of heavy metals (copper,, aluminum, Zinc, Manganese iron) on the growth rates of the parasite *Acanthamoeba* sp **in vitro**

s	Elements	Rates of parasite numbers Mm^3			
		24 hours	48 hours	72 hours	96 hours
1	Control	725	1000	1218	1437
2	**Cu1**	268	437	481	618
3	**Cu2**	206	387	425	512
4	**Al1**	1875	2050	2312	2525
5	**Al2**	1743	1887	2156	2343
6	**Zn1**	1493	1775	1987	2281
7	**Zn2**	1412	1650	1775	2112
8	**Mn 1**	1162	1475	1762	2068
9	**Mn 2**	1031	1287	1625	1762
10	**Fe1**	793	1200	1531	1881
11	**Fe2**	737	1100	1375	1625

1= Concentration of 20 mg / l
2 = concentration of 40 mg / l

Test the direct effect of ions of heavy metals on the growth rates of the parasite *Balantidium coli* **in vitro**

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Proved the results of the current study, the direct effect of heavy metals on the growth of the parasite *Balantidium coli* was found when adding ions (aluminum, zinc ,manganese ,iron) to the culture media led to increase the number of parasite compared to control group while the effects of ion copper few where led added to the inhibition of the growth of the parasite few numbers (Table 3)

Table (3) the effect of ions of heavy metals (copper,, aluminum, Zinc, Manganese iron) on the growth rates of the parasite *Balantidium coli* in vitro

s	Elements	Rates of parasite numbers Mm^3			
		24 hours	48 hours	72 hours	96 hours
1	Control	937	1175	1387	1662
2	**Cu1**	737	837	987	1275
3	**Cu2**	562	725	825	1025
4	**Al1**	2012	2275	2525	2875
5	**Al2**	1775	2150	2275	2687
6	**Zn1**	1662	2025	2212	2343
7	**Zn2**	1525	1850	2012	2200
8	**Mn 1**	1337	1562	1912	2025
9	**Mn 2**	1212	1387	1762	1875
10	**Fe1**	1187	1350	1525	1812
11	**Fe2**	987	1200	1425	1687

1= Concentration of 20 mg / l
2 = concentration of 40 mg / l

DISCUSSION

Showed current study metabolize experiment protozoa were conduct to examine the effect of supplemental (copper,Aluminum, manganese,zinc,iron) to culture media and test effect on growth of three type of parasite *Naegleria fowleri, Acanthamoeba* sp.
With two concentration for all element (20mg/ l , 40mg/ l) *coli Balantidium*

Proved the results of the current study that the increase of the concentration of elements toward all tested isolates led to her death, and this is consistent with the study[11] to indicated that some of the ions of heavy elements are essential for the growth of microorganisms when added low concentrations take place within the limits of carrying microorganism, but when increase the concentrations of these ions on the limits of the microorganism, it carries a negative affect on the growth and reproduction.

He also noted[8] to a positive relationship between the effect of trace elements and influence positive or negative, where the greater the absorption component increased toxicity and killing the parasite, as in copper, the less absorption deficiency toxicity and increased growth and reproduction, and because of the variation of the absorption of aluminum, zinc, manganese, iron, copper led to a variation in numbers of the parasite

Copper inhibition its protozoa growth while iron in the culture media positive effect because the protozoa can uptake copper 60 times comparaed with iron inside cell , so the copper is more toxic than iron and it leads to death[13]

Aluminum in the environment utilized efficiently interact with hydrogen cell parasite and this gives the fuel cell leads to increased division invitro[14]

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