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Normal Saline and Distilled Water for Isolation Parasites from Fresh Vegetables in Nassiriyah and Suq - AL- Shuyukh Cities in Thi-qar Province

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***Abstract*---**Normal saline is often preferred to isolate parasites from vegetables and other sources but distilled water is rarely used for this purpose. So in order to know the difference between ability of normal saline and distilled water to isolate the parasites, 64 samples of six types of fresh vegetables from Nassiriyah city and also the same number and types (64 samples of six types of fresh vegetables) from Suq - AL- Shuyukh city were examined by using sedimentation method with normal saline one time and distilled water in other times. Where these vegetables types are Garden Cress, Leek, Radish, Lettuce, Celery and Basil). In addition to additional samples of the most contamination vegetable were subjected to several ways of cleaning to find out what is the best way to get rid of these parasites. Four hundred forty one of patients of hospitals in Nassiriyah and Suq - AL-Shuyukh cities who infected with parasites were re-examined to know the most existence parasites. The results showed a convergence between the proportions of some parasites isolated by normal saline and parasites isolated by distilled water where percentage of Giardia lamblia was 70.3% using normal saline and 71.9% using distilled water. While some parasites were not isolated when using distilled water compared to using normal saline thus some parasitic species are affected by distilled water. Vinegar got rid of *Balantidium coli* and *Trichomonas hominis* that water alone could not get rid of them. Giardia lamblia was the most existence parasite in the hospitals that included in the study.

***Keywords*---**distilled water, normal saline, parasites, vinegar.

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Introduction

Fruits and vegetables have low calories, fat and sodium, and contain fibers, vitamins, minerals and other phytochemicals that promote health (Luke, 2017), so the essential materials that have important role in human resistance to some of diseases can be obtained by consuming fruits and vegetables (Su et al., 2012).

Although fresh vegetables are an important part of optimal health, the consumption of fresh raw vegetables without proper cooking or washing may make them the main route in the transmission of intestinal parasites to humans (Mahdi et al., 2013). Where Consumption of contaminated, undercooked and poorly washed vegetables can lead to infection of human with parasites such as *Entamoeba histolytica*, *Giardia intestinalis*, *Ascaris lumbricoides*, *Cryptosporidium* spp., *Enterobius vermicularis*, *Fasciola* spp., hookworm, *Hymenolepis* spp., *Taenia* spp., *Trichuris trichiura*, and *Taxocara* spp (Kozan et al., 2005).

Studies have varied about the preferred method for isolating parasites (Obaid, 2019), some of them preferred sedimentation and flotation methods (Al-Khamesi, 2014), that depend on solutions with low or high gravity used to precipitation or flotation of some parasites and examine them under a microscope (Arora & Arora, 2009), while other studies prefer other methods (Tefera et al.,2014). Washing vegetables properly is one of the prevention methods that reduce parasitic infection (Bekele et al., 2017), especially when solutions as vinegar or saturated salt solution are used where they have a decomposer effect on parasites (Agbalaka et al., 2018).

Materials and Methods

Samples collection

Sixty four of fresh vegetables samples (included six types) from Nassiriyah city and also 64 samples from Suq - AL- Shuyukh city in addition to 10 samples that were lift without treat with any liquid, 10 samples were treated with water, and 10 samples were treated with vinegar were collected from local markets of Nassiriyah and Suq - AL- Shuyukh cities.

Preparation of samples (Daryani et al., 2008)

- The parts of vegetables are not eaten were removed, and 100gm of each vegetable were cut to small parts and washed with normal saline (0.9%) on dish and soaked in it for 24 hours.
- The samples were raised from dish had rinsed by brush with small amount from special water then the vegetables were raised.
- Washing water after its filtration on metal sieve was left for one hour for sedimentation. The supernatant was discarded and sediment was examined.
- The sedimentation method (Al-Mozan & Al-Asady, 2015; Al – Mozan et al., 2014), with normal saline was done, where the sediment with

some normal saline that remained after removing the supernatant was centrifuged and examined.

The same mentioned steps were repeated with using distilled water to perform sedimentation methods with distilled water.

Soaking in vinegar

The parts of vegetables are not eaten were removed, and 100gm of each vegetable were cut to small parts and washed with water and soaked with vinegar for 15 minute and raised to soak in normal saline for 24 hours and the sediment was examined.

Examination of stool samples in the hospitals

Stool samples of patients in hospitals of Nassiriyah and Suq - AL- Shuyukh cities were examined using naked eye according to [14] and microscope according to (Paniker, 2007; Ansari et al., 2022).

Results and Discussions

Table 1
Comparison between the ability of normal saline and water to isolate parasites

Isolation solution / Parasite type	Normal saline Isolation percent	Distilled water Isolation percent
Giardia lamblia	45(70.3%)	46(71.9%)
Entamoeba histolytica	13(20.3%)	13(20.3%)
Balantidium coli	9(14.1%)	16(25%)
Trichomonas hominis	25(39.1%)	25(39.1%)
Entamoeba coli	5(7.8%)	1(1.6%)
Coccidia	3(4.7%)	2(3.1%)
Enterobius vermicularis	7(10.9%)	2(3.1%)
Heterophyes heterophyes	1(1.6%)	0(0.0%)
Hymenolepis nana	0(0.0%)	1(1.6%)
Strongyloides stercoralis	6(9.4%)	3(4.7%)
Taenia saginata	1(1.6%)	0(0.0%)
Diphyllobotherium latum	1(1.6%)	0(0.0%)
Ascaris lumbricoides	10(10.6%)	10(10.6%)
Ancylostoma duodenale	6(9.4%)	3(4.7%)
Trichuris trichiura	3(4.7%)	1(1.6%)
Schistosoma mansoni	1(1.6%)	0(0.0%)
Schistosoma japonicum	1(1.6%)	0(0.0%)
Schistosoma haematobium	1(1.6%)	0(0.0%)
Fasciola hepatica	1(1.6%)	0(0.0%)
Dipylidium caninum	1(1.6%)	0(0.0%)
Larva of nematoda	1(1.6%)	1(1.6%)

Percentages of *G. lamblia, E. histolytica, B. coli*, and *T. hominis* as examples of some parasites that were isolated using normal saline were 70.3%, 20.3%,14.1%,

and 39.1% respectively, also percentage of these parasites were 71.9%, 20.3%, 25%, and 39.1% respectively when were isolated using distilled water. On other hand, some parasites as *S. mansoni*, *S. haematobium, T. saginata,* and others were isolated with using normal saline but weren't isolated when distilled water was used Table (1).

This result is agreement with conclusion of (Zeibig, 1997; Olegovich Bokov et al., 2022), who indicated that the results between studies can be differed due to several factors as the techniques and handling method that are used.

Type of solution as one of materials that make up the method or technique has a role in identifying the diagnosed parasites, some parasites are not affected by the type of solution that used to isolate them while other parasites may be affected by the used solution, in other word, the distilled water may not affect the quantity but may have an effect on the type of parasite diagnosed (genus or diagnosed species).

Table 2
Parasites that were not affected by washing with water, but affected by vinegar

Parasites were eliminated by vinegar but weren't affected with water
Balantidium coli
Trichomonas hominis

Many parasites have been eliminated, whether using water or vinegar, and some have not been eliminated, neither with water nor with vinegar, while vinegar was able to get rid of *Balantidium coli* and *Trichomonas hominis*, which water could not get rid of them table (2). Where vinegar has a degrading effect on some parasites and this agrees somewhat with (Agbalaka et al., 2018; Huldani et al., 2022), who pointed out that vinegar and saturated salt solution can plasma lyse the parasites.

Table 3
Parasites that were found in patients of hospitals in areas from which the examined vegetables were taken

Parasites types in an infected patients of hospitals in Nassiriyah and Suq - AL-Shuyukh cities	Percentage of parasite
Giardia lamblia	397(90%)
Entamoeba histolytica	29(6.6%)
Entamoeba histolytica + *Giardia lamblia*	15(3.4%)
Total number of infected patients	441

The highest rate of infection in hospitals was 90% of *Giardia lamblia* table (3). This is evidence that vegetables are the main source of infection with *Giardia lamblia*.

The study recommends the need to use the recommended efficient solutions to isolate parasites where normal saline is the most efficient, but it is also preferable to use more than one type of solutions to increase certainty. Although vinegar was able to eliminate *Balantidium coli* and *Trichomonas hominis* that water could not get rid of them, it was not effective in eliminating parasites that were not eliminated by water, so it must be continuing to search for the material that has the efficiency of eliminating all parasites.

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