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Detection of Parasitic Contamination of Cress Crop Irrigated with Two Types of Water in Jumka Village, Erbil – Iraq

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ABSTRACT

This study aims to evaluate the risk of using untreated wastewater for vegetable contamination. Water samples and vegetables cress and scientific name (Lepidium sativum) were collected from farms using wastewater and well water for irrigation for three seasons (Autumn, Winter, and Spring). Results show that untreated wastewater is characterized by an alkaline side of neutrality and has a high content of the Biochemical Oxygen Demand BOD₅ value of 124 mg. 1⁻¹. The Biochemical Oxygen Demand (BOD₅) values variations coincided with water temperature variations and no Dissolved Oxygen (DO) detection during the study periods. Cryptosporidium spp. and other intestinal parasites were observed in wastewater in nearly all studied seasons. Wastewater-contaminated vegetables irrigated by it. Well, water is characterized by the highest EC value of 1119 µS.cm⁻¹ during Winter and the highest pH value of 8.1 during the Spring season. Generally, well water is contaminated with parasitic organisms. Cryptosporidium spp. was detected in well water but no detection was found in cress irrigated by it.

Keywords: Wastewater, well water, *Cryptosporidium* spp., aquatic parasites, curly cress.

INTRODUCTION

In the past three decades, the demand for water in the city of Erbil has doubled and tripled, due to the population increase, urbanization, and the reception of immigrants from the neighboring and southern governorates in large numbers, in addition to the change in lifestyle and the inefficient management of water in the city, which has placed great burdens on the city's water resources. Also, the climatic changes and the lack of rain had a negative role on the amount of surface and groundwater in the governorate. In the agricultural sector, the farmers relied on rainfall in the wet seasons, but most of them now rely on wells; and at the southwest side of the city where the wastewater channel of Erbil and with its the villages that grow vegetables mainly depend on contaminated water for irrigation, eventually, polluted water discharged into the Greater Zab River (Shekha and Al-Abaychi, 2010b).

Urbanization increases wastewater production, and wastewater has emerged as a potential source of water for agricultural activities (Al-Samarrai, 2019). Generally, the use of potable water for irrigation is more expensive than the use of wastewater (Garrido *et al.*, 2006). Erbil untreated sewage water is considered a logical alternative for farmers to rainwater and groundwater, which have been greatly reduced, due to the advantages of this water in its abundance in their areas and its low cost, in addition to containing the nutrients necessary for the growth of their plants instead of using chemical fertilizers at a high cost. But what they do not know or ignore it is the risks of their use for public health, as it contains many heavy elements and pathogenic microorganisms, especially bacteria, viruses, and parasites (Aziz, 2020).

There are many studies that have been conducted on the Erbil wastewater channel since the beginning of the ninety last century for various assessments, including quantity and purification (Mustafa and Sabir, 2001), water quality (Shekha *et al.*, 2010, Aziz, 2020), reuse (Ganjo *et al.*, 2006a), types of microorganisms and their risks (Shekha and Al-Abaychi, 2010a). determination of heavy metals (Tariq, 2021).

Vegetables are one of the main ingredients in meals, eaten either fresh, as a salad, or cooked. Most of the markets in Erbil city are supplied with vegetables from farms irrigated with untreated wastewater. Vegetables are a high-risk source for the spread of microbial infections, particularly parasitic infections, with eaten of fresh vegetables playing a major role in the transmission of parasitic food-borne illnesses (Al-Mozan and Dakhil, 2019). One of the most important sources of parasites for Erbil wastewater channel comes from several sources, including animal slaughterhouse, in which all the contents of the intestines of animals, blood, and meat remains are thrown. In addition to the black water, where black water is transported from houses through tankers designated for this purpose and illegally and away from the eyes of the authorities it is thrown inside the waters of the Erbil wastewater channel, near the vegetable farms (Shekha, 2008). Various parasites that have been associated with vegetables include species of protozoans and helminths, protozoans such as Cryptosporidium spp., Giardia intestinalis, Ascaris lumbricoides, hookworms, Taenia spp., Fasciola spp. could infect humans if they consume contaminated, raw, or improperly washed vegetables (Yusuf et al., 2017). Based on (Mustafa and Sabir, 2001) report before two decades they estimated the vegetable farmlands were irrigated with sewage water by nearly 700 dunums, while these cultivated areas have expanded at present, but without any official estimation about them. There is also no statistical data from Erbil health sectors on the outbreak of infectious diseases or infection rates related to the consumption of vegetables contaminated with parasites.

This gap in information motivated researchers to study the effects of using untreated wastewater on contaminating cultivated vegetables with parasites and their negative impacts on both human and environmental health. The aim of this study is to evaluate the quality of untreated wastewater used for irrigation of vegetable farms in the study area and its parasite content.

MATERIALS AND METHODS

1. Samples collections

Water samples (Raw wastewater and well water) and vegetable samples (curly cress *Lepidium sativum*) from cultivated farmlands were collected with three replications for each season (Autumn 2021, Winter and Spring 2022). The studied area is near Jumka village which is located southwest of Erbil city about 19 Km far from the city center, and 9 km far from Turaq village Fig. (1). The selected vegetable is eaten freshly (non-cooked) and was collected from two farms one irrigated with raw wastewater and the other from farms that used well water, wells in this region usually do not exceed a depth of 60-70 meters. All samples were kept in a cold box and soon back to the laboratory for Physico-chemical and parasites examination (Apha, 2017).

2. Examination of parasites in water and vegetable:

Plastic bags were used to transport the vegetable samples to the laboratory. They were immediately immersed in distilled water for washing vegetables. 200g of cress samples were soaked in one of sterile physiological normal saline (0.95 percent NaCl), and each sample was mechanically shaken for 15 minutes. The top layer was removed, and the remaining wash solution was centrifuged at 2000 rpm for 15 minutes after being filtered through a sieve (micron pore size) to eliminate large debris. After decanting the supernatant, a few drops of the sediment were placed on glass slides, and smears stained with modified Ziehl-Neelsen were prepared to detect protozoan such as *Cryptosporidium* spp., *Isospora* spp., *Entamoeba* spp. and Trematods such as *Schistosoma haematobium*, *Fasciola* spp. (Monica, 2006).

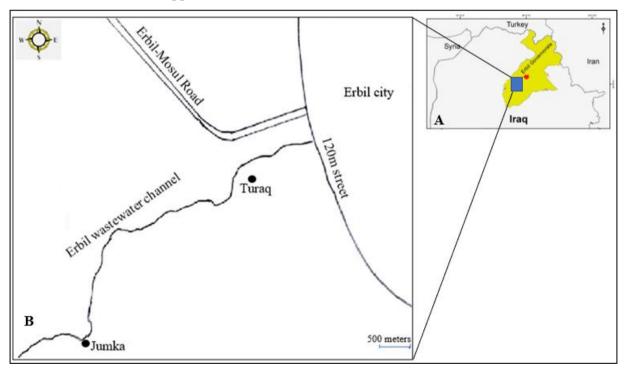


Fig. 1: Map Shows: A- Northern Iraq, Erbil governorate, studied area; B- studied site and Erbil wastewater channel.

3. Bailenger method

Bailenger method summarized as: a sample of wastewater is subjected to a settling and centrifugation process in order to collect specific sediment containing helminth eggs. After mixing and centrifuging, an acetic-acetate buffer and ethyl acetate are added to the sediment. An inferior layer containing sediment containing helminth eggs is collected. After concentration, the sample is

resuspended in zinc sulfate, and an aliquot is transferred to a McMaster chamber, where an egg count is performed after the flotation of the egg (Sharafi *et al.*, 2015).

4. Water analysis

For both water sources (wastewater and well-water) samples were collected. A mercury thermometer is used to measure water temperature; a pH meter (Philips 9420) is used to measure pH; and an EC meter is used to measure electrical conductivity (WTW D 8120). While chemical measurements were analyzed as follows (Apha, 2017). Dissolved oxygen, and BOD₅ using the azide modification method.

Table 1: Physiochemical parameters of Erbil wastewaters and well water from Jumka village-Erbil, Iraq.

Sites	Parameters	Autumn	Winter	Spring
	pН	7.37	7.43	8.10
Well water	EC (µS.cm ⁻¹)	834	1119	728
	Water temp.(°C)	27	20.5	10
	$BOD_5(mg. l^{-1})$	2	0.7	0.8
	pН	7.38	7.80	7.82
Wastewater	EC (µS.cm ⁻¹)	930	887	962
	Water temp.(°C)	26	11	20
	$BOD_5(mg. l^{-1})$	93.9	51.5	124.2

RESULTS AND DISCUSSION

Physio-chemical and biological results of both aquatic ecosystems are shown in (Table 1-3) Fig. (2 and 3). pH is one of the essential aquatic parameters that regulate the chemical reactions, activities and metabolic rates of organisms (Viessman and Hammer, 1993; Al-Hamdani and Kaplan, 2022). Generally, the value of well water ranged from (7.37-8), while waste water varied from (7.3 to 7.82). The highest pH value was recorded in the spring season, which may be related to detergents with alkaline nature, high photosynthesis rate of algae, and alkaline side of neutrality for catchment area in wastewater channel. Results came in accordance with previous studies in the same waste water channel (Al-Barzingy *et al.*, 2016a).

Electrical conductivity is the ability of water to pass on electrical current (Rice *et al.*, 2012), (Schwartz and Gruendling, 1985) observed a high EC value in the Steven Brook wetland, indicating a significant concentration of dissolved ions coming from sewage. (Ganjo *et al.*, 2006b) reported that the EC value of Erbil wastewater was more than 1000 μS.cm⁻¹. While the EC value for wastewater in the present study was 887 μS.cm⁻¹ in Winter and 962 μS.cm⁻¹ in Spring. Well water is characterized by the highest EC value (1119 μS.cm⁻¹) recorded in Winter. The value of the electrical conductivity depends on the concentration and equivalence of the dissolved ions present in the water and on the temperature of the water during the measurement (Nesaratnam, 2014).

Dissolved oxygen is crucial for the life of aquatic organisms and an indicator of aquatic health. During the study, the period DO for wastewater was non-detectable, the same results were reported by many authors (Ganjo *et al.*, 2006b) and (Shekha and Al-Abaychi, 2010b) in the same study area. High organic content various microorganisms' activities and biodegradable is the main reasons for DO consumption.(Bellinger and Sigee, 2010). Dissolved oxygen for well water was varied from highest level 5.1 mg. l⁻¹ during Autumn to less than 1 mg. l⁻¹ during Spring.

It was clear that these well water was not purified or chlorinated and it may contain many pollutants such as oocyst of *Cryptosporidium*, may be other microorganisms as it was observed at the same season (Table 2 and 3).

Table 2: Oocyst of Cryptosporidium spp. in Erbil untreated wastewater and well-water with

vegetable irrigated by them.

Type of Samples	Autumn		Winter		Spring	
	Samping1	Sampling2	Sampling1	Samping2	Sampling1	Sampling2
Wastewater	+	+	+	+	+	+
Vegetable irrigated by	+	+	-	-	+	+
wastewater						
Well-water	-	+	-	-	+	+
Vegetable irrigated by	-	-	-	-	-	-
well water						

Note: Sampling1, Sampling2; number of samplings repeating of each season.

Table 3: Parasitological contamination in Erbil untreated wastewaters and the vegetable

irrigated with it.

Taxa	Spring season			
	Sampling1	Sampling2		
Ascaris lumbricoides	+(egg)	-		
Schistosoma spp.	-	+(egg)		
Schistosoma haematobium	+(egg)	-		
Entamoeba spp.	+(cyste)	+(trophozoite)		
Isospora belli	+(Oocyst)	+(Oocyst)		
Giardia	+(cyst)	+(cyst)		
Fasciola spp.	-	+adult		

Note: Sampling1, Sampling2; number of samplings repeating of each season

Autumn had the highest BOD₅ value for well water (2 mg. 1⁻¹) as shown in Fig. (2), and as a result, it can be classified as good water (Aziz et al., 2014). Lowest BOD₅ was observed during Winter. The variations of water temperature were coincided with a change in the values of BOD₅. with an inverse relation between both parameters. Water temperature-controlled metabolic rate and activities of microorganisms, with increasing water temperature increase biodegradation of organic matter which decreased DO content and rise up BOD₅ values. While, highest BOD₅ value for wastewater was 124 mg. 1⁻¹ recorded during Autumn, and lowest value 51.5 mg. 1⁻¹ was observed during Winter coincided with lowest water temperature (Al-Barzingy et al., 2016b) and (Aziz, 2020) The proximity of water wells to the sewage channel and shallowness of wells, which usually do not exceed a depth of 60-70 meters, may be behind the contamination of these wells and their impacts by sewage water nearby.

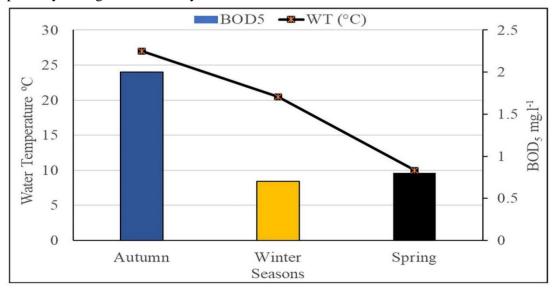


Fig. 2: Variations of water temperature (°C) and BOD₅ (mg. l⁻¹) for well water.

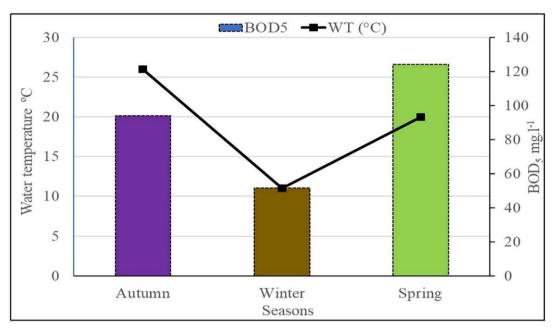


Fig. 3: Variations of water temperature (°C) and BOD₅ (mg. l⁻¹) for Erbil untreated wastewater.

Parasites in water and vegetable samples, developing countries are characterized by producing vegetables in unhealthy ways that created hazards for farmers and consumers by using untreated wastewater as a cheap source for irrigation (Abi Saab *et al.*, 2022). One of the major causes of human parasitic diseases is the consumption of raw vegetables without proper washing (Al-Shirifi and A Abdullah, 2005; Alemu *et al.*, 2019,).

During our investigations of vegetable farms in southwest Erbil city that used untreated wastewater for irrigation, many types of parasites were detected in water or vegetable samples. In both the Autumn and Spring seasons *Cryptosporidium* spp. and other intestinal parasites, liver and blood trematodes were observed in nearly all samples except vegetables irrigated by well water (Tables 2 and 3). Only vegetable irrigated with well water was clear from parasites despite the detection of *Cryptosporidium* spp. in well water. The Winter season was clear from any detection of parasites, it may be attributed to a decrease in water temperature and less availability of favorable conditions. The spring season was the most favorable period for the presence and activity of intestinal parasites in their various stages, which may be due to the increased activity of humans and animals that contaminated both water and vegetables, and in addition to the appropriate environmental conditions (Plates 1 and 2). The vegetables may have been contaminated during preharvesting, harvesting, and post-harvesting, according to their Studies (Ma *et al.*, 2014, Sleman Ali *et al.*, 2018). Because untreated wastewater is sometimes used to irrigate the vegetables, eating uncooked vegetables can result in parasitic infection if adequate washing is not performed.

CONCLUSIONS AND RECOMMENDATIONS

From the above results can conclude that untreated wastewater possesses various types of parasites with different stages which make risks for farmers and consumers. As well as, curly cress irrigated by polluted water. Well water is regarded as contaminated water through detection of *Cryptosporidium* spp. in two seasons with no detection of parasites in curly cress irrigated by it. From obtained results, it can be recommended to preventing the use of this polluted water for the purpose of irrigation and finding alternatives to polluted water and warning consumers against buying and eating vegetables irrigated with contaminated water or using these vegetables after using sterilizers when washing vegetables and washing them several times with clean water.

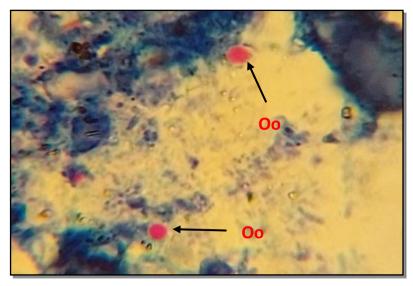


Plate 1: Direct modified acid-fast stain on untreated wastewater samples, showing acid fast positive *Cryptosporidium* spp. oocyst (Oo), 100X

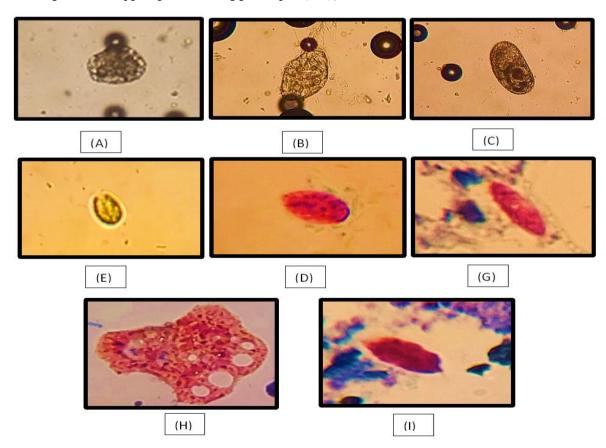


Plate 2: (A) Ascaris lumbricoides(egg), 10X (B) Schistosoma spp.(egg), 10X (C) Entamoeba spp.(cyste), 10X(D) Giardia spp.(cyste), 40X (E) Isospora belli (Oocyst), 100X (F) Schistosoma haematobium (egg), 100X (G) Entamoeba spp. (trophozoite), 100X (H) Fasciola spp. (adult), 40X

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كشف التلوث الطفيلي لمحصول حب الرشاد المروي بنوعين من المياه في قرية جومكا، أربيل - العراق

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الملخص

هدفت هذه الدراسة إلى تقييم مخاطر استخدام مياه الصرف الصحي غير المعالجة في تلويث الخضروات. تم جمع عينات من المياه و نبات الرشاد (Lepidium sativum) من المزارع المروية بمياه الصرف الصحي ومياه الآبار لأغراض الري لثلاثة مواسم (الخريف والشتاء والربيع). أظهرت النتائج أن مياه الصرف الصحي غير المعالجة تتميز بجانب قلوي من الحياد و تحتوي على نسبة عالية من الطلب على الأكسجين الكيموحيوي (BOD₅) بقيمة 124 ملغم لتر⁻¹، تزامنت التغيرات في قيم BOD₅ مع تغيرات درجة حرارة الماء، ولم يتم الكشف عن الأكسجين المذاب أثناء فترة الدراسة. و الأكسجين المذاب كان اقل من مستوى التحري عليه طوال فترة الدراسة. سجلت . Cryptosporidium spp وغيرها من الطفيليات المعوية في مياه الصرف الصحي في جميع المواسم المدروسة تقريبًا. كما تم تسجيلها في الخضروات المروية بمياه الصرف الصحي. تتميز مياه الآبار بأعلى قيمة للتوصيلية الكهربائية 119ميكروسيمنز .سم⁻¹ خلال فصل الشتاء وقيمة pH تبلغ 8.1. بشكل عام، تكون مياه الآبار ملوثة بالكائنات الطفيلية. خلال فصل الربيع تم الكشف عن الكشف عنها في مياه الآبار ولكن لم يتم الكشف عنها في نبات الرشاد المروى بها.

الكلمات الدالة: مياه الصرف الصحى، مياه الأبار، . Cryptosporidium spp، الطفيليات المائية، نبات الرشاد.