

# A STUDY OF BIOLOGICALLY TREATED WASTEWATER IN IRRIGATION AND ITS EFFECT ON GROWTH AND (N, P, K) CONTENTS IN MAIZE

AHMED SLIIM HAIMED\*, ABD ALZAHRA TAHA THAHER  
AND SALWA JUAH FAKHIR

Soil Science and Water Resources, College of Agricultural, University of Basrah, Iraq  
[ASH, AATT, SJF].

[\*For Correspondence: E-mail: abdalzahrataha503@gmail.com]

## Article Information

### Editor(s):

(1) Dr. Tsygankova Victoria Anatolyivna, National Academy of Sciences, Ukraine.

### Reviewers:

(1) J. Amudha, ICAR-Central Institute for Cotton Research, India.

(2) Gabriela Medeiros, Brazil.

Received: 02 January 2021

Accepted: 09 March 2021

Published: 20 March 2021

Original Research Article

## ABSTRACT

The study aimed to use wastewater from Hamdan and Al-Jamaa stations after biological treatment by removing the two elements of lead by (*Staphylococcus aureus*) and cadmium with *Escherichia coli* bacteria isolated from this water. Shatt al-Arab water was used as a comparison. Biologically treated and untreated wastewater in addition to the Shatt al-Arab water was used to irrigate the growing *Zea mays* plant in pots containing (5) kg of soil taken from Qurna district and fertilized with chemical fertilizer (N, P and K) according to the fertilizer recommendation for *Zea mays* and after 60 days of growth. The results showed that the percentage of removal of cadmium from the liquid culture media pollutant with cadmium was 29.2% by *Escherichia coli* bacteria and that the percentage of removal of the element of lead from the culture liquid media pollutant with lead was 67.6% by bacteria *Staphylococcus aureus*. The highest dry weight of the maize crop was 21.64 g pot<sup>-1</sup> using the AL-jamaa wastewater treated with *E. coli* bacteria, and the highest nitrogen and phosphorous percentage in the maize plant was 4.38 and 0.4164% due to the effect of irrigation with the Hamdan sewage plant treated with *Staphylococcus aureus*, respectively, and potassium. By 4.183%, the effect of irrigation with water from Hamdan plant treated with *E. coli* bacteria.

**Keywords:** Waste water; bioremediation; *Staphylococcus aureus*; *Zea mays*.

## INTRODUCTION

The problem of water scarcity is one of the biggest problems facing humanity in the future around the world, and in this case [1], treatment and recycling of wastewater may be one of the most important solutions in developing water resources and

reducing the problem of water scarcity in agriculture [2] Biological water treatment is defined as a process in which microorganisms, green plants, or enzymes are used to treat polluted sites to restore their correct condition, as this technique is more appropriate than traditional techniques due to the low cost and economic

incomes and being environmentally friendly [3]. It is also the most effective and least expensive treatment for soils and water pollutants with heavy elements by using types of bacteria, fungi, algae, and yeasts isolated from industrial waste, soil pollutants with heavy metals, and wastewater to effectively remove heavy metals [4,5].

Microorganisms play an effective role in removing and reducing the concentrations of many heavy elements in water because they have a number of mechanisms that make them highly efficient, including the process of adsorption and formation of complexes on the microscopic cell wall [6,7]. The use of treated wastewater from pollution can save large quantities of river water for irrigation and reduce the increasing water needs for irrigation purposes in developing countries [8,9]. Moreover, it promotes the growth of soil microorganisms with essential nutrients [10,11]. Especially the nutrients for plants such as nitrogen, phosphorus, and potassium that sustain crop productivity and other growth parameters [12,13] showed that the use of treated wastewater increased the light of tomato plant by (35%) and 42% for the two agricultural seasons respectively compared to natural water sources. The research aims to isolate efficient bacteria in removing lead and cadmium elements from wastewater and using treated water to irrigate maize plants and their effect on dry weight and (K, P, N) concentrations in the plant.

## MATERIALS AND METHODS

### Collecting Water Samples

Sewage water samples were collected from Hamdan and al-Jamaa stations affiliated to Basra Governorate Sewage Directorate and from internal basins at a depth of 20 cm by sterile glass bottles with a capacity of (500 ml) and the samples were transferred to the laboratory and the chemical, physical and biological properties of the water were estimated according to [14].

### Isolation and Diagnosis of Lead and Cadmium Resistant Bacteria

The lead and cadmium resistant bacteria were isolated from the wastewater of Hamdan and

Jamea stations by dilution method and poured in plates ( $10^{-1}$ – $10^{-6}$ ) by transferring 0.1 ml of dilution to the nutrient medium Agar containing lead (1,2,3,4,5,6,7Mm) cadmium concentrations (1,2,3,4 Mm) and after incubation on  $37^{\circ}\text{C}$  for 48 hours. The large bacterial colonies growing in the highest concentrations of lead and cadmium were selected. These colonies were purified by re-cultivation on the same culture medium with the same concentrations of lead and cadmium and then the colonies were preserved in slant media after being covered with glycerol by freezing [15]. The bacterial isolates resistant to the highest concentration of lead (164) ppm were identified as (*Staphylococcus aureus*) and isolates resistant to the highest concentration of cadmium (11.24ppm) were identified as *Escherichia coli*, according to [16].

### Wastewater Bioremediation by Removing Lead and Cadmium and Preparing Treated Water for Irrigation

The wastewater was prepared for irrigation by placing 1000 ml of untreated sewage water for the Hamdan and Jamea plants in volumetric flasks.

This water was inoculated with (1) ml of bacterial inocula suspension containing  $10^8$  cfu of lead resistant (*Staphylococcus aureus*) and cadmium resistant (*Escherichai coli*) bacteria and the water was incubated at ( $37^{\circ}\text{C}$ ) for a period of (48) hours, then the treated water was filtered from 0.4Mm Millipore filter paper with using of a vacuum pump the treated water was collected for the Hamdan and al-Jamaa stations separately for use in the agricultural experiment.

### Agricultural Experiment

The wastewater was prepared for irrigation by placing 1000 ml of untreated sewage water for the Hamdan and Jamea plants in volumetric flasks.

The experiment was carried out in the plastic canopy - College of Agriculture - Basra University by taking soil samples from Qurna District / Basra Governorate, their properties were shown in Table 2 and after grinding and passing from a 4 mm sieve were placed in 5 kg pots-1 then fertilized the soil with urea, calcium superphosphate, and

potassium sulfate as recommended Fertilizers, of maize 150-90-150 kg ha<sup>-1</sup> N, P and K respectively. Pots were planted with (8) seeds pot<sup>-1</sup> and irrigated with wastewater of Hamdan and al-jamaa treatment plants with bacteria to limit field capacity with the use of Shatt al-Arab water as a comparison. The plants were thinned to 3 potted plants, the growth continued to 60 days, after which the plants were cut and dried at 65°C for 48

hours, and their dry weight was taken, then they were ground, 0.2 gm was taken, and they were digested with an acid mixture (96% concentrated sulfuric acid and 4% perchloric acid) according to [17], in which nitrogen was estimated by the Kaldahl apparatus and phosphorous by the optical absorption spectrophotometer and potassium in the Flame photometer apparatus according to [18].

**Table 1. The chemical and physical properties of water**

Adjectives	Unit	Sewage Hamdan	Whole wastewater	Shatt al-Arab water
pH	–	7.66	7.76	8.25
E.C	ds m <sup>-1</sup>	7.89	9.63	8.54
BOD		23.58	75.85	13.25
COD		211	564	172
Total hardness		280	254	554
Lead		3.7	0.23	2.84
Cadmium		1.2	0.4	1
Potassium		29	24	41
Phosphorous		0.872	0.711	0.766
Calcium		197	297.62	210
Sodium		189	914.23	338
TS	Mg L <sup>-1</sup>	3971	2890	5514
TSS		50	3235	92
TDS		4.48	4.02	5475
Turbidity		362	117	252
TPC	CFU	3.75 × 10 <sup>7</sup>	3.42 × 10 <sup>7</sup>	3 × 10 <sup>5</sup>

**Table 2. The primary physical and chemical characteristics of the soil used in the agricultural experiment**

Adjective	unit	The value	
Sand	gm Kg <sup>-1</sup>	279.5	
Silt		323.9	
Clay		395.95	
Tixture		Clay tixture	
Organic matter	gmkg <sup>-1</sup>	8.28	
Total carbonate		197.66	
PH	–	7.6	
E.C	ds m <sup>-1</sup>	6.5	
Ions dissolved	Ca <sup>++</sup>	7.78	
	Mg <sup>++</sup>	6.33	
	K <sup>+</sup>	0.375	
	Co <sub>3</sub> <sup>-2</sup>	0	
	Hco <sub>3</sub>	mmole L <sup>-1</sup>	2.73
	So <sub>4</sub>		6.17
	Cl		28.24
Heavy elements	Na	16.01	
	N	mgkg <sup>-1</sup>	17.15
	P	mgk <sup>-1</sup>	22.9
	Cd	mg kg <sup>-1</sup>	2
	Pb		20.081

## RESULTS AND DISCUSSION

Table 3 showed the biological removal of heavy elements from the contaminated agricultural medium with 164 parts per million of lead and 11.24 parts per million of cadmium, and it is noted that *Staphylococcus aureus* has removed (111.02) parts per million of lead with a removal rate of (69.6%). While the *Escherichia coli* bacteria removed 3.283 ppm of cadmium with a removal rate of (29.2%), *Staphylococcus aureus* also showed an ability to grow in solid and liquid media containing a concentration of 164 ppm of lead, while *Escherichia coli* bacteria were able to grow. In solid and liquid media containing 11.24 ppm of cadmium, this is due to the extent of the bacteria's ability to resist heavy elements through their accumulation in their cells, their adsorption, and the ridding of contaminated water from them [19].

It is noted from Table 4 indicated that irrigation with wastewater of Hamdan and Al-Jamaa stations and Shatt Al-Arab water had a significant effect on the dry weight of yellow corn. The highest rate was by al-jamaa wastewater 17.98 gm pot<sup>-1</sup>, while the lowest rate was 11.98 grams per 1 by the effect of irrigation Shatt al-Arab water, due to the fact that the wastewater is rich in organic matter and nitrogen that are important for plant growth. Also, the removal of cadmium from irrigation water by (*E. coli*) had a significant effect on the increase of dry weight of maize, as it gave the highest rate of 16.5gm of pot<sup>-1</sup>, which was not significantly different from the water treated with *S. aureus* bacteria removing lead, which had a dry weight of 15.80gm. pot<sup>-1</sup>, while the lowest rate was 11.64 gm pot<sup>-1</sup>, affected by non-use of bacteria. The interaction between the water used in irrigation and the bacterial used in its treatment was significant effect on the dry weight of maize, as the highest rate was 21.64 gm pot<sup>-1</sup> when irrigation with wastewater of the Al-Jamaa station and their treated with *E. coli* bacteria removing cadmium, while the lowest rate was 10.08 gm pot<sup>-1</sup> by the effect of irrigation with untreated Shatt al-Arab water, the reason for this was that the presence of cadmium in high concentrations in irrigation water negatively affects cell division and damage to the ends of the water-carrying roots [20]. Also, the high concentrations of heavy

elements inhibit the construction of chlorophyll due to its interference with the enzyme (proto chlorophyllide reductase), or due to the internal replacement of the magnesium atom in the center of the chlorophyll molecule with the heavy elements atoms, which leads to a decrease in the concentration of chlorophyll [21].

It is noted from Tables 5 and 6 indicated that the use of irrigation water with the Shatt al-Arab had a significant effect on increasing the concentration of nitrogen and phosphorous in the maize plant, as the highest rate reached 3.234% and 0.3302%, while the lowest rate were (3.126%) and (0.2992%) (When using Hamdan sewage water and AL-Jammaa sewage water respectively, the bacteria used in bioremediation had a significant effect in increasing the percentage of nitrogen and phosphorous, and the highest rate was 4.05% and 0.3626% due to the effect of lead-removing bacteria *Staphylococcus aureus*, and the lowest rate was 1.874% and 0.239% due by non-use bacteria respectively and the effect of the interaction between the type of water used for irrigation and the bacteria used in bioremediation were significant in the percentage of nitrogen and phosphorous. (4.383%) and (0.4167%) due to the effect of Hamdan irrigatim water with *S. aureus* bacteria removed the lead. This is due to the presence of lead in high concentrations in untreated sewage were decrease the plant height and leaf area due to its negative effect on the process of photosynthesis, formation of proteins and carbohydrates [22].

Table 7 show that the use of Hamdan treated water had a significant effect on increasing the concentration of potassium in the maize plant, and the highest rate was (3.767%), while the lowest rate was (3.392%) due to irrigation with the Shatt al-Arab water. The bacteria used through biological treatment had a significant effect on the potassium concentration in the plant. The highest rate was (3.792%) due to the effect of *E. coli* bacteria, while the lowest rate (3.203%) was found without using Bacteria. The interaction also had a significant effect on increasing the potassium concentration in maize plant, as the highest rate (4.183%) was due to the effect of using Hamdan sewage water biologically treated with *E. coli* bacteria. While the lowest rate (3.040%) was due

to the effect of using the Shatt al-Arab water that is not biologically treated with bacteria, due to the reduce and removal of the cadmium element from the sewage as a result of adsorption by *Escherichia coli* bacteria and the reduction of its effect on the transport tissues in the root system of the plant and the absorption of nutrients [10].

**Table 3. Bio-removal of heavy metals in the culture media contaminated with lead and cadmium**

Elements	Bacteria	Concentrate of element before treatment (ppm)	Concentrate of element after treatment(ppm)	Removal rate%
Lead	<i>Staphylococcus aureus</i>	164	52.98	67.6
Cadmium	<i>Escherichia coli</i>	11.24	7.957	29.2

**Table 4. The effect of irrigation with biologically treated and untreated water with lead and cadmium removal bacteria on the dry weight of maize (g/pot)**

Bacteria	Water quality			Average
	Hamdan sewage water	Al-Jammaa sewage water	Shatt al-Arab water	
S.aureus	17.45	17.73	12.21	15.80
E.coli	14.19	21.64	13.67	16.50
control	10.27	14.57	10.08	11.64
Average	13.97	17.98	11.98	L.S.D = Bacteria 1.869

Water quality L.S.D = 1.869  
L.S.D for interaction = 3.237

**Table 5. The effect of irrigation with biological treated and untreated water with lead and cadmium removal bacteria on nitrogen concentration in maize plant%**

Bacteria	Water quality			Average
	Hamdan sewage water	AL-Jamaa sewage water	Shatt al-Arab water	
S. aureus	4.353	4.093	3.675	4.050
E.coli	3.527	3.747	3.647	3.640
Control	1.467	1.773	2.383	1.874
Average	3.126	3.204	3.234	Bacteria L.S.D = 0.3586

Water quality L.S.D = 0.5586  
L.S.D for interaction = 0.6211

**Table 6. The effect of irrigation with biologically treated and untreated water with lead and cadmium removal bacteria on the phosphorus concentration of maize plant%**

Bacteria	Water quality			Average
	Hamdan sewage water	Al- jamaa sewage water	Shatt al-Arab water	
S.aureus	0.4167	0.3377	0.3333	0.3626
E.coli	0.3047	0.3200	0.3967	0.3404
control	0.2167	0.2400	0.2607	0.2391
Average	0.3127	0.2992	0.3302	Bacteria L.S.D = 0.02643

Water quality L.S.D = 0.02643  
L.S.D for interaction = 0.04577

**Table 7. The effect of irrigation with biologically treated and untreated water with lead and cadmium removal bacteria on potassium concentration of maize plant %**

Bacteria	Water quality			Average
	Hamdan sewage water	AL-Jamaa sewage water	Shatt al-Arab water	
<i>S. aureus</i>	4.030	3.920	3.387	3.779
<i>E. coli</i>	4.183	3.443	3.750	3.792
control	3.087	3.483	3.040	3.203
Average	3.767	3.616	3.392	bacteria.L.S.D = 0.2889

Water quality L.S.D = 0.2889  
L.S.D for interaction = 0.5004

## CONCLUSION

We conclude from this study the possibility of using the wastewater of Hamdan and the University plant to irrigate different plants after biologically treated with *Escherichia coli* and *Staphylococcus aureus* bacteria to remove heavy elements from them such as lead and cadmium to reduce the shortage of water suitable for irrigation.

## ACKNOWLEDGMENTS

The authors thank the staff of the Deep. of Soil Science and Water Resource, College of Agriculture, the University of Basra for supporting the study.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

## REFERENCES

- Al-Taey DKA, Imad JC, AL-Naely BH, Kshash. A study on effects of water quality, cultivars, organic and chemical fertilizers on potato (*Solanum tuberosum* L.) growth and yield to calculate the economic feasibility. *Bulgarian Journal of Agricultural Science*. 2019;25(6):1239-1245.
- Patel G. Effect of waste irrigation on heavy metal accumulation in soil and its removal by plants doctoral dissertation. JNKVV; 2013.
- Gaur N, Flora G, Yadav M, Tiwari A. A review with recent advancements on bioremediation-based abolition of heavy metals. *Environmental Science: Processes & Impacts*. 2014;16(2):180-193.
- Vishan I, Kalamdhad AS.. Heavy metal removal through bacterial biomass isolated from various contaminated sites. *International Journal of Environmental Sciences*. 2016;7(1):1-18.
- Slomy AK, Jasman AK, Kadhim FJ, AL-Taey DKA, Sahib MR. Study impact of some biofactors on the eggplant *Solanum melongena* L. Vegetative characteristics under glass houses conditions. *Int J Agriculture Stat Sci*. 2019;15(1):371-374.
- Balderian. Interactions of heavy metals with white-rot fungi. *Enzym Microb Technol*. 2009;32: 78–91
- Jatav MK, Kumar M, Trehan SP, Dua VK, Lal SS. Influence of microorganisms inoculation for nutrient economy in potato-radish crop sequence In North Western Himalayas. *Int J Agricult Stat Sci*. 2011; 7(1):309-316.
- Widaa AM, Saeed AB. Impact of using treated wastewater for irrigation on soil chemical properties, plant growth and forage yield. *University of Khartoum Journal of Agricultural Science*. 2008; 16(1):75-87
- AL-Taey DKA, Saadoon AH. Effect of treatment of salicylic acid and water salinity on the growth and nitrate accumulation with nitrate reductase activity in the leaves of spinach, *Spinacia oleracea* L. *Journal of Babylon University, Pure and Applied Sciences*. 2014;3(22):1188-1203.
- Al-Khafajy RA, AL-Taey DKA, AL-Mohammed MHS.. The impact of Water Quality, Bio fertilizers and Selenium Spraying on some Vegetative and

- Flowering Growth Parameters of *Calendula Officinalis* L. under Salinity Stress. *Int J Agricult Stat Sci.* 2020;16(1):1175-1180.
11. Hamza O.M. and D. K. A. AL-Taey. A study on the effect of Glutamic acid and benzyl adenine application upon growth and yield parameters and active components of two broccoli hybrids. *Int J Agricult Stat Sci.* 2020;16(1):1163-1167. Available:<https://connectjournals.com/03899.2020.16.1163>
  12. Abdoukader BA, Mohamed B, Nabil M, Alaoui-Sossé B, Eric C, Aleya L. Wastewater use in agriculture in Djibouti: Effectiveness of sand filtration treatments and impact of wastewater irrigation on growth and yield of *Panicum maximum*. *Ecological Engineering.* 2015;84:607-614.
  13. Akponikpe P, Wima K, Yakouba H, Mermoud A. Reuse of domestic wastewater treated in macrophyte ponds to irrigate tomato egg plants in semi-arid west-Africa: benefits and risks. *Agr water Manag.* 2011;98:834-840. DOI: 10.1016/j.agwat.2010.12.009
  14. Standard method for the examination of water and wastewater. American water public health assoc. American Water Works Assoc. New York. 2005;21<sup>st</sup> ed.
  15. Li W, Jiany Y, Xu L, Lin C. *Nocardia alba* sp Actinomycete strain isolated from soil in China. *Appl Microbiol.* 2004;27:308-312.
  16. Qureshi A, Hemant J, Purohit. Isolation of bacteria consortia for degradation of p-nitro phenol from agricultural soil, *Ann Appl Biol.* 2002;140: 159-162.
  17. Cresser MS, Parsons JW. Sulphuric—Perchloric acid digestion of plant material for the determination of nitrogen, phosphorus, potassium, calcium and magnesium. *Analytica Chimica Acta.* 1979; 109(2):431-436.
  18. Page AL, Miller RH, Keeney DR. Methods of soil analysis .Part 2.2<sup>nd</sup> Ed ASA. Inc Madison Wisconsin, U.S.A; 1982.
  19. Kalita D, Joshi SR. Study on bioremediation of Lead by exopolysaccharide producing metallophilic bacterium isolated from extreme habitat. *Biotechnology Reports.* 2017;16:48-57.
  20. Küpper H, Küpper F, Spiller M. Environmental relevance of heavy metal-substituted chlorophylls using the example of water plants. *Journal of Experimental Botany.* 1996;47(2):259-266.
  21. Athar R, Ahmad M. Heavy metal toxicity: effect on plant growth and metal uptake by wheat, and on free living *Azotobacter*. *Water Air and Soil Pollution.* 2002;138 (1-4):165-180.
  22. Wu FB, Chen F, Wei K, Zhang GP. Effect of cadmium on free amino acid, glutathione and ascorbic acid concentrations in two barley genotypes (*Hordeum vulgare* L.) differing in cadmium tolerance. *Chemosphere.* 2004;57(6):447-454.