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Bioaccumulation of Macro Elements in Spinach (*Spinacea oleraceac* L.) Irrigated with Wastewater from Quetta City

Zahoor Ahmad Bazai, Abdul Kabir Khan Achakzai and Shameem Gul
Department of Botany, University of Balochistan, Quetta, Pakistan

Abstract: This study was conducted to determine the concentration of macro elements (viz., Ca, Na and K) in spinach (*Spinacea oleraceac* L.), when irrigated with 5 different concentration (treatments) of waste water collected from three different localities (viz., Chiltan town, Chiltan Ghee Mill and Zarghoon town) of Quetta city. The spinach was taken as a test plant and grown in pots. Tap water was used as background water. Results showed that localities, treatments and their interactions exhibited highly significant difference ($p<0.01$). Statistically maximum concentration of Ca, Na and K (viz., 6906, 6008 and 1801 mg kg^{-1}) are obtained in Zarghoon town followed by a minimum of 4888, 3329 and 1448 mg kg^{-1} in Chiltan ghee mill effluents, respectively. Data also showed a significant linear increase in the amount of Na and K as the concentration of wastewater increases. While in case of Ca such increase was noted up to moderate level of effluents, beyond which reduction was noticed. Statistically maximum Ca, Na and K (viz., 6769, 6121 and 1798 mg kg^{-1}) were obtained in T_3 and T_5 levels of wastewater, respectively.

Key words: Wastewater, spinach, macro elements, calcium, sodium, potassium

INTRODUCTION

In the globally changing scenario, environment is the main focusing factor and a huge threat for the entire global community. The increased use of natural resources for human needs generated solid waste, toxic effluent and injurious gases emissions, which eventually adversely affects on human health and their environment[1] and major health risks are associated with poor quality/polluted water[2]. The practice of adding wastewater to agricultural land for raising vegetables yield is a common practice. It contains many plant nutrients. The kind and concentration of nutrients depend upon the source. Linked with environmental pollution, water pollution is a problem of worldwide concern and ground water is badly polluted due to unplanned disposal of untreated domestic sewage and industrial effluents into water courses.

Quetta is the capital of Balochistan province and one of the largest trade centers in Pakistan. It has a hardy, ugly and dull face, despite being the provincial metropolis nestled amidst the craggy mountains that form a ring around it, looks dusty and bone dry[3]. The population explosion, urbanization, persistent drought, the influx of Afghan refugees and industrialization are the factors showed the way to city a situation that the fresh water channels were replaced by the domestic sewage and industrial waste. But such changed situation could not influence the local farmers indulged in conventional practice of growing vegetables earlier by fresh water and now by wastewater. The direct use of untreated waste water is common in most cities which are due to the lack of alternative water sources and about 26% of the domestic vegetables are cultivated with waste water in Pakistan[4]. Quetta is one of the badly affected cities by different pollutions[5,6]. Like many other cities of the country, wastewater in Quetta is also directly used for irrigation to raise the net output of vegetables, which are considered as a major source of organic manure and plant nutrients as well[7,8]. But the use of city effluents may also result in accumulation of macro essential elements like Na, K, Mg and Ca[9,10], which may adversely affect the human metabolism. The elemental composition of both food grains and vegetables are important in human and animal nutrition. A great deal of work has been done and is known about the elemental composition of food grains, but much less is known about vegetables[11,12]. In view of the fact, number of sources of pollution and demand for food are increasing, the challenge of adequate knowledge about elements toxicities intensified. This study was therefore; mainly aimed to determine the concentration for accumulated macro elements by spinach irrigated with different sources (localities) and concentrations of wastewater.

Corresponding Author: Abdul Kabir Khan Achakzai, Department of Botany, University of Balochistan, Quetta, Pakistan

MATERIALS AND METHODS

The study presented deals to determine the concentration of macro elements in spinach (*Spinacea oleraceae* L.) when irrigated with five different concentrations (treatments) of wastewater collected from three different localities of Quetta city (viz., Chiltan town, Chiltan Ghee Mill and Zarghoon town). The treatments (T) were made by dissolving calculated amount of wastewater in Tap water i.e., control, 25, 50, 75 and 100%. These treatments were then designated by letters T_1, T_2, T_3, T_4 and T_5, respectively. Each treatment was replicated thrice for each locality

Spinach was grown in 45 pots contained well-mixed garden soil. They were then arranged in North-South direction at proper distance to avoid any possible external contamination along with the provision of possible uniform light condition. Ten seeds of *Spinacea oleraceae* (L.) were sown at one-centimeter depth at an equal distance with the help of clean policeman. Pots were arranged in a Completely Randomized Design (RCD) in an open field and were then equally irrigated with respective treatments (T) of wastewater. All agricultural practices were made during the course of germination, growth and development.

Before initiation of flowers, the plant samples were made by taking three leaves each from all treatments and replicates. Each treated plant sample washed separately with HCl (1%). They were then followed by 3-4 washings with deionized water to remove the foreign materials. These samples were then spread on blotting paper and air-dried, then oven dried at 70° C. After drying, leaf samples were crushed to powdered form in a grinder and then converted into liquid following wet digestion procedure i.e., one gram of dried leaf powder samples placed in a beaker, 10 mL of concentrated HNO_3 was added and allowed to stand overnight. It was then heated carefully on a hot plate until the production of red fumes ceased. The beakers were cooled and 2 mL of 70% $HClO_4$ added and heated it again and allowed to evaporate to a small volume. Filter the samples and transferred the filtrate to a 100 mL volumetric flask and made the volume up to 100 mL with the help of deionized water[13]. Macro elements (viz., Ca, Na and K) in plant samples were then analyzed by using flame photometer (Corning 400). Data obtained were statistically analyzed, following the procedure described by Steel and Torrie[14]. MSTAT-C computer software package was used for the purpose.

RESULTS AND DISCUSSION

Data presented in Table 1 deciphered that sampling localities (A), different levels of sewagewater (B) and their interactions (A x B) are statistically found highly significant ($p<0.01$) for all mentioned species.

Data pertaining to mean values showed that Ca contents of spinach are significantly different in water sampling localities. Statistically maximum amount of Ca i.e., 6906 mg kg^{-1} is obtained in Zarghoon town followed by minimum i.e., 4888 mg kg^{-1} in Chiltan ghee mill (Table 2). However, by comparing the treatments, Ca is significantly increased up to receiving moderate level of

Table 1: Analysis of variance (ANOVA) for few cations of spinach (*Spinacea oleracea* L.) subjected to various level of wastewater collected from different localities of Quetta

	Sum of square			Mean square			F-value of variables at an error of 28			
Variables (mg kg^{-1})	Localities (A)	Sewagewater levels (B)	A x B	Localities (A)	Sewagewater levels (B)	A x B	(A)	(B)	A x B	CV (%)
Calcium	34249889.54	6020416.36	27168617.69	17124944.27	1505104.09	3396077.2	81335.86**	7148.57**	16129.85**	0.24
Sodium	56130057.978	90882336.026	36612493.173	28065028.489	22720584.0	4576561.65	159502.6**	129128.4**	26010.07**	0.27
Potassium	933678.907	989796.174	857366.582	466839.453	247449.043	107170.823	6675.453**	3538.335**	1532.462**	0.51

** highly significant at 5% level of probability and CV = coefficient of variation

Table 2: Effect of five different level of wastewater of Quetta city on the calcium contents (Ca mg kg^{-1}) of spinach (*Spinacea oleracea* L.)

	Wastewater Treatments (T)					
Wastewater localities	T_1	T_2	T_3	T_4	T_5	Mean
Chiltan Ghee Mill	6101.16 I	5927.60 K	5698.34 L	3512.14 M	3202.60 N	4888.37 C
Chiltan Town	6221.14 G	6121.60 I	6775.18 E	6321.20 F	7099.16 D	6507.66 B
Zarghoon Town	6001.14 J	6190.87 H	7832.65 A	7301.21 B	7202.44 C	6905.66 A
Mean	6107.81 B	6080.02 C	6768.72 A	5711.52 E	5834.73 D	6100.562

LSD @ $p<0.05$ and $p<0.01$ both for localities and treatments are 24.27 and 32.74, respectively

Mean values followed by different letter(s) within right side column (localities) and bottom row (treatments) of table are not significantly differ with each other at 5% level of probability using LSD test. Similarly values followed by the same letter(s) within column and rows (localities x treatments) in the center of the given table are not significantly differ with each other

Table 3: Effect of five different level of wastewater of Quetta city on the sodium (Na mg kg^{-1}) of spinach (*Spinacea oleracea* L.)

Wastewater localities	Wastewater treatments (T) T_1	T_2	T_3	T_4	T_5	Mean
Chiltan ghee mill	2220.20 O	3404.76 K	4621.45 H	3777.17 J	2623.01 L	3329.32 C
Chiltan town	2440.20 M	4111.32 I	5812.31 F	6509.21 D	6877.35 C	5150.08 B
Zarghoon town	2322.30 N	5101.45 G	6121.32 E	7633.33 B	8861.23 A	6007.93 A
Mean	2327.57 E	4205.84 D	5518.36 C	5973.24 B	6120.53 A	4829.107

LSD @ $p<0.05$ and $p<0.01$ both for localities and treatments are 22.19 and 29.93, respectively

Mean values followed by different letter(s) within right side column (localities) and bottom row (treatments) of table are not significantly differ with each other at 5% level of probability using LSD test. Similarly values followed by the same letter(s) within column and rows (localities x treatments) in the center of the given table are not significantly differ with each other

Table 4: Effect of five different level of wastewater of Quetta city on the potassium (K mg kg^{-1}) contents of spinach (*Spinacea oleracea* L.)

Wastewater localities	Wastewater treatments (T) T_1	T_2	T_3	T_4	T_5	Mean
Chiltan ghee mill	1401.20 J	1272.56 L	1466.91 H	1799.17 D	1299.01 K	1447.77 C
Chiltan town	1416.20 I	1400.03 J	1699.63 F	1701.63 F	1904.74 B	1624.45 B
Zarghoon town	1503.10 G	1763.92 E	1701.73 F	1844.54 C	2189.72 A	1800.60 A
Mean	1440.17 E	1478.84 D	1622.76 C	1781.78 B	1797.82 A	1624.273

LSD @ $p<0.05$ and $p<0.01$ both for localities and treatments are 13.99 and 18.87, respectively

Mean values followed by different letter(s) within right side column (localities) and bottom row (treatments) of table are significantly differ with each other at 5% level of probability using LSD test. Similarly values followed by the same letter(s) within column and rows (localities x treatments) in the center of the given table are significantly differ with each other

effluents, beyond that significant reduction is observed. Statistically a maximum amount is obtained in T_3 (i.e., 6769 mg kg^{-1}). Data also showed a highly significant interaction between localities and treatments and their Ca concentration ranges from 3203-7833 mg kg^{-1}. These figures are therefore, far greater than those obtained by Shar *et al.*[11,12] in seven unpolished rice (i.e., 40.16-73.90 mg kg^{-1} and sixteen barley cultivars (i.e., 5.67-130.23 mg kg^{-1}).

Data concerned about Na (Table 3) exhibited that Na content in spinach leaf of all 3 localities are significantly differ with each other. Statistically maximum amount of Na i.e., 6008 mg kg^{-1} is received for Zarghoon town followed by minimum i.e., 3329 mg kg^{-1} in Chiltan ghee mill. However, by comparing the treatments, Na is significantly and linearly increased as the concentration of effluents increases and a maximum amount is obtained In T_5 (i.e., 6121 mg kg^{-1}). Data also exhibited a highly significant interaction between localities and treatments and their Na concentration ranges between 2220-8861 mg kg^{-1}. These values are also far greater than those obtained by Shar *et al.*[11,12] in seven unpolished rice (i.e., 436.12-616.01 mg kg^{-1}) and sixteen barley cultivars (i.e., 827-2409 mg kg^{-1}).

Data presented in Table 4 deciphered that in response to source of wastewater, K content in spinach significantly differs with each other. Statistically maximum amount i.e., 1800.60 mg kg^{-1} is found in Zarghoon town followed by minimum i.e., 1447.77 mg kg^{-1} in Chiltan ghee mill. However, by comparing the treatments, K is significantly and linearly increased as the level of effluent increases. Therefore, a maximum amount is obtained In T_5 (i.e., 1797.82 mg kg^{-1}). Data also deciphered a highly significant interaction between localities and treatments and their K level ranges between 1272.56-2189.72 mg kg^{-1}. These values are therefore, par lesser than those obtained by Shar *et al.*,[11,12] in seven unpolished rice (i.e., 2902-4097 mg kg^{-1}) and sixteen barley cultivars (i.e., 4353-10352 mg kg^{-1}). In present study K is also lower than the recommended adequate level i.e., 3800-5300 mg kg^{-1} for spinach[15].

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