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*Full Length Research Paper*

# Fertigation response of *Abelmoschus esculentus* L. (Okra) with sugar mill effluent in two different seasons

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**The fertigation response of sugar mill effluent doses namely: 5%, 10%, 25%, 50%, 75% and 100% on *Abelmoschus esculentus* (var. IHR 31) in two different seasons, that is, rainy (*Kharif*) and summer (*Zaid*) seasons was investigated. The study revealed that fertigant had significant (P<0.01) effect on moisture content, EC, pH, $Cl^-$, OC, $HCO_3^-$, $CO_3^{-2}$, $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, $Fe^{2+}$, TKN, $NO_3^{2-}$, $PO_4^{3-}$, $SO_4^{2-}$, Cd, Cu, Pb, Mn and Zn and insignificant (P>0.05) effect on WHC and bulk density of the soil in both seasons. Fertigation with 100% sugar mill effluent concentration decreased moisture content (18.84-22.69%), WHC (13.26-15.61%), BD (1.40%) and pH (16.66-17.28%), and increased EC (84.13-86.47%), OC (3811.62-3961.90%), $Na^+$ (273.00-302.59%), $K^+$ (31.59-33.14%), $Ca^{2+}$ (729.76-788.31%), $Mg^{2+}$ (740.47-805.12%), TKN (1390.63-1445.54%), $PO_4^{3-}$ (337.79-346.52%), $SO_4^{2-}$ (77.78-81.07%), $Fe^{2+}$ (301.90-345.26%), Cd (437.73-565.00%), Cu (417.57-419.13%), Pb (1487.50-1706.66%), Mn (1365.38-1382.52%) and Zn (333.33-348.32%) in the soil used for the cultivation of *A. esculentus* in both seasons. The agronomical performance of *A. esculentus* was recorded to be in increasing order from 5 to 25% in both the *Kharif* and *Zaid* season when compared to the control. The accumulation of heavy metals was increased in the soil as *A. esculentus* increased from 5 to 100% concentrations in both cultivated seasons. The contamination factor (Cf) of various heavy metals was in the order of Mn>Cd>Cu>Zn>Pb for soil and Cu>Mn>Zn>Cd>Pb for *A. esculentus* in both the *Kharif* and *Zaid* season after fertigation with sugar mill effluent.**

**Key words:** Sugar mill effluent, *Abelmoschus esculentus*, fertigation, heavy metals, *Kharif* and *Zaid* seasons, contamination factor.

**ABBREVIATIONS: ANOVA** - Analysis of variance; **BD** - Bulk density; **BIS** - Bureau of Indian standards; **BWW** - Bore-well water; **CD** - Critical difference; **Cf** - Contamination factor; **HI** - Harvest index; **LAI** - Leaf area index; **MPN** - Most probable numbers; **RT** - Relative toxicity; **SPC** - Standard plate count; **WHC** - Water holding capacity.

## INTRODUCTION

The application of industrial wastewater in the cultivation of agricultural crops is becoming common practice (Ayyasamy et al., 2008; Ezhilvannan et al., 2011; Kumar and Chopra, 2012, 2014a, b). The increasing application of wastewater in the agricultural fields may serve as a viable method of disposing the wastewater and sustaining agriculture in non-irrigated areas having shortage of fresh water for irrigation. In the agriculture, irrigation water quality is believed to have effects on the soils characteristics and agricultural crops growth (Kisku et al., 2000; Maliwal et al., 2004; Kumar and Chopra, 2013a, c). Irrigation with wastewater is a common practice in urban and suburban areas. Effluent generated from various sources like municipal, household, small and big industries are the important sources of wastewater generation in urban and sub-urban areas (Kisku et al., 2000; Kaushik et al., 2004; Kannan and Upreti, 2008). Application of wastewater in agriculture is most prevalent in these areas, in that it reduces the fertilizer and irrigation water cost as it is available without paying any cost, and is rich in various plant nutrients as well. Besides this, effluent contains heavy metals which accumulate in plant and vegetable parts and cause various health effects (Ramakrishnan et al., 2001; Orhue et al., 2005; Kannan and Upreti, 2008; Sridhara et al., 2008). Long term irrigation with effluents increases heavy metals accumulation in soil and increases the chances of their entrance into food chain, thus, ultimately causes significant geoaccumulation, bioaccumulation and biomagnifications (Ferguson, 1990; Hati et al., 2007; Kumar and Chopra, 2013d, e, f). The amount of heavy metals mobilized in soil environment is a function of pH, clay content, organic matter, cation exchange capacity and other soil properties making each soil unique in terms of pollution management (Kisku et al., 2000; Tandi et al., 2004; Kumar and Chopra, 2013b, g).

*Abelmoschus esculentus* (Okra) is cultivated two times in two different seasons (rainy and summer) in a year in India, one sown at the end of February for summer (*Zaid*) crop and at the end of August for rainy (*Kharif*) crop (Kumar and Chopra, 2013b). During the last decade, cultivation of *A. esculentus* has become a centre of magnetism of Indian vegetable growers due to its short growing period, biannual growing habit and high market significance (Kumar and Chopra, 2013b). It is extensively used as green vegetable and also has medicinal values. The green pods are mildly diuretic and contain a substance that reduces the blood sugar level. The dried mature pod is used in the treatment of diabetes and in the treatment of ulcers. The seeds are diuretic, hypoglycemic, anti-diabetic, anti-viral and hypotensive and also used in the treatment of blood cancer (Kumar and Chopra, 2013b). Most crops give higher potential yields with wastewater irrigation and reduce the need for chemical fertilizers, resulting in net cost savings to farmers (Osaigbovo et al., 2006; Bharagava et al., 2008; Ezhilvannan et al., 2011). So it is important to understand the specificity of crop-effluent liaison for their appropriate application in irrigation practices (Krishna, 2002; Osaigbovo et al., 2006; Vijayaragavan et al., 2011; Kumar and Chopra, 2013g). In recent past, various studies have been made on the characteristics of effluent of industries, agronomical properties of various crop plants (Tiwari et al., 2000; Swamy et al., 2001; Yadav et al., 2002; Sharma et al., 2007). But much attention has not been paid so far on the use of industrial effluents on the cultivation of agricultural crops like *A. esculentus* (Rathore et al., 2000; Ramana et al., 2002; Rattan et al., 2005). Keeping in view the reuse of wastewater as fertigant and the economic importance of *A. esculentus*, the present investigation was undertaken to study the response of *Abelmoschus esculentus* L. (Okra) after fertigation with sugar mill effluent in two different seasons.

## MATERIALS AND METHODS

### Experimental design

A field study was conducted in the Experimental Garden of the Department of Zoology and Environmental Sciences, Faculty of Life Sciences, Gurukula Kangri University, Haridwar (29°55'10.81'' N and 78°07'08.12'' E), for studying the fertigation effect of sugar mill effluent on *A. esculentus.* The crop was cultivated in two seasons namely *Zaid* and *Kharif* season in the year 2012 and 2013, thus it was grown four times (that is, two times in *Zaid* and two times in *Kharif* seasons). Poly bags (dia-30 cm) were used for growing the *A. esculentus* plant. The experiment was replicated by six times. The number of poly bags (42) having soil were used for the cultivation of *A. esculentus*. Proper distance was maintained between each replicate (30 cm) and between all treatments (60 cm) for the maximum growth performance of the crop. Each poly bag was made porous for aeration and it was labeled for the various treatments namely: 5%, 10%, 25%, 50%, 75% and 100%.

### Effluent collection and analysis

Uttam Sugar Mills Ltd. Libberheri, Haridwar (29°44'38''N 77°51'14''E) was selected for collection of effluent sample. The effluents were collected from an outlet of the effluent treatment plant using plastic container and brought to the laboratory and was analyzed for various physico-chemical and microbiological parameters namely: total dissolved solids (TDS), pH, electrical conductivity (EC), dissolved oxygen (DO), biochemical oxygen demand (BOD), chemical oxygen demand (COD), chlorides ($Cl^-$), bicarbonates ($HCO_3^-$), carbonates ($CO_3^{2-}$), sodium ($Na^+$), potassium ($K^+$), calcium ($Ca^{2+}$), magnesium ($Mg^{2+}$), total Kjeldahl nitrogen (TKN), nitrate ($NO_3^{2-}$), phosphate ($PO_4^{3-}$) and sulphate ($SO_4^{2-}$) and iron (Fe), cadmium (Cd), copper (Cu), lead (Pb), manganese (Mn), zinc (Zn), standard plate count (SPC) and most probable number (MPN) following standard methods (APHA, 2005; Chaturvedi and Sankar, 2006) and further used as fertigant in different concentrations namely: 5%, 10%, 25%, 50%, 75% and 100% for cultivation of *A. esculentus*.

### Soil preparation, filling of poly bags, sampling and analysis

The soil used was collected from a depth of 0 - 15 cm. Each poly bag (30×30 cm) was filled with 5 Kg well prepared soil, earlier air-dried and sieved to remove debris and mixed with equal quantity of farmyard manure. About 5 Kg of soil in each of the forty-two poly bags were fertigated twice in a week with 500 ml of sugar mill effluent in six concentrations 5%, 10%, 25%, 50%, 75% and 100% along with bore well water (control). The soil was analyzed before crop sowing and after crop harvesting as per effluent concentration for various physico-chemical parameters namely: moisture content, water holding capacity (WHC), bulk density (BD), EC, pH, OC, $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, TKN, $PO_4^{3-}$, $SO_4^{2-}$, $Fe^{2+}$, Cd, Cu, Pb, Mn and Zn following standard methods (Chaturvedi and Sankar, 2006).

**Sowing of seeds, irrigation pattern and collection of crop parameters data**

The seeds of *A. esculentus* were sown at the end of February 2012 and 2013 for summer (*Zaid*) crop and at the end of August 2012 and 2013 for rainy (*Kharif*) crop. The seeds of *A. esculentus* (var. IHR 31) were procured from Indian Council of Agriculture Research (ICAR), Pusa, New Delhi and sterilized with 0.01 mercuric chloride and was soaked for 12 h. The sugar mill effluent was applied with its concentrations namely: 0%, 5%, 10%, 25%, 50%, 75% and 100% concentration per 5 Kg soil and then left for 2 weeks to allow for mineralization and further irrigation of the crop plant. Seven seeds were initially sown in each poly bag at equal distance between plant to plant (7.5 cm). Five plants were maintained in each bag by thinning out of the seven and each set was replicated six times as thirty plants were grown for each treatment group including the control group. The crop plants received the effluent with concentrations of 5%, 10%, 25%, 50%, 75% and 100% of effluent as fertigant doses (500 ml) twice in a week/as per the crop requirement and no drainage was allowed. The various agronomical parameters namely: seed germination, relative toxicity, shoot length, root length, dry weight, chlorophyll content, leaf area index (LAI), number of flowers, number of pods and crop yield and harvest index (HI) of *A. esculentus* at different stages (0-90 days) were determined following standard methods (Kumar and Chopra, 2012):

Relative toxicity:

$$\text{Relative toxicity (RT)} = \frac{\text{Germination percentage with control}}{\text{Germination percentage with effluent}} \times 100$$

Leaf area index: $$\text{Leaf area index (LAI)} = \frac{\text{Leaf area}}{\text{Land area}}$$

Harvest index:

$$\text{Harvest index (HI)} = \frac{\text{Grain weight (g.)}}{\text{Total plant weight (g.)}} \times 100$$

**Heavy metals analysis**

For heavy metal analysis, 5-10 ml sample of effluent, 0.5-1.0 g sample of air dried soil/plant was taken in digestion tube and added to 3 ml conc. $HNO_3$ digested on electrically heated block for 1 h at 145°C. Then 4 ml of $HClO_4$ was added to it and heated to 240°C for an additional hour. It was allowed to cool down and filtered through Whatman # 42 filter paper and makeup volume up to 50 ml and used for analysis. Metals were analyzed using an atomic absorption spectrophotometer (PerkinElmer, Analyst 800 AAS, GenTech Scientific Inc., Arcade, NY) following the methods of APHA (2005) and Chaturvedi and Sankar (2006). The contamination factor (Cf) for heavy metals accumulated in sugar mill effluent irrigated soil and *A. esculentus* was calculated by the following procedure (Ferguson, 1990; Kumar and Chopra, 2013a):

$$\text{Contamination factor (Cf)} = \frac{\text{Mean content of metal in the sample}}{\text{Background metal content of the substance}}$$

**Statistical analysis**

Data were analyzed for one way analysis of variance (ANOVA) to determine the difference between soil and crop parameters before and after sugar mill effluent irrigation, as per effluent concentration. Standard deviation, coefficient of correlation for soil and crop parameter as per effluent concentration was also calculated with the help of MS Excel, SPSS12.0 and Sigma plot, 2000.

**RESULTS AND DISCUSSION**

**Characteristics of effluent**

The mean ± SD values of physico-chemical and microbiological parameters TDS, EC, pH, DO, BOD, COD, $Cl^-$, $HCO_3^-$, $CO_3^{2-}$, $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, TKN, $NO_3^{2-}$, $PO_4^{3-}$, $SO_4^{2-}$, $Fe^{2+}$, Cd, Cu, Pb, Mn, Zn, SPC and MPN of sugar mill effluent are given in Table 1.

The results revealed that the effluent was acidic in nature pH (6.11). Among various parameters, BOD (3480.20 mg $L^{-1}$), COD (8560.00 mg $L^{-1}$), $Cl^-$ (1680.50 mg $L^{-1}$), $Ca^{2+}$ (1820.40 mg $L^{-1}$), $NO_3^{2-}$ (1430.50 mg $L^{-1}$), $Fe^{2+}$ (28.90 mg $L^{-1}$), TKN (572.50 mg $L^{-1}$), $SO_4^{2-}$ (1280.00 mg $L^{-1}$), MPN ($4.58 \times 10^6$ 100 $ml^{-1}$) and SPC ($3.64 \times 10^{10}$ $ml^{-1}$) were found beyond the prescribed limit of Indian Irrigation Standards (BIS, 1991). The higher values of TDS (4868), BOD (1664.56) and COD (2285.80) indicated the higher inorganic and organic load in sugar

**Table 1.** Physico-chemical and microbiological characteristics of control (Bore-well water) and Uttam Sugar Mill effluent.

| Parameter | Effluent concentration (%) | | | | | | | BIS for drinking water | BIS for irrigation water |
|---|---|---|---|---|---|---|---|---|---|
| | 0 (BWW) | 5 | 10 | 25 | 50 | 75 | 100 | | |
| TDS (mg $L^{-1}$) | 221.50±10.75 | 478.50±7.55 | 860.00±4.76 | 1964.80±9.57 | 2413.00±6.22 | 4396.80±8.41 | 7680.00±4.43 | 500 | 1900 |
| EC (dS $m^{-1}$) | 0.34±0.12 | 0.74±0.11 | 1.34±0.19 | 3.07±0.21 | 3.77±1.03 | 6.87±1.42 | 12.00±1.83 | - | - |
| pH | 7.52±0.24 | 7.49±0.25 | 7.41±0.26 | 7.02±18 | 6.65±0.33 | 6.27±0.31 | 6.11±0.50 | 6.5-8.5 | 5.5-9.0 |
| DO (mg $L^{-1}$) | 8.24±2.65 | 6.36±2.74 | 4.68±2.86 | 4.87±2.89 | 2.42±0.23 | 1.18±0.14 | NIL | 6-8 | - |
| BOD (mg $L^{-1}$) | 3.83±0.59 | 164.00±7.48 | 334.25±10.28 | 850.50±15.00 | 1635.50±5.97 | 2462.50±11.82 | 3480.20±8.54 | 4.0 | 100 |
| COD (mg $L^{-1}$) | 5.88±1.37 | 448.00±30.59 | 897.75±63.05 | 2231.00±11.94 | 4352.50±10.25 | 6495.50±11.82 | 8560.00±9.87 | 150-200 | 250 |
| $Cl^-$ (mg $L^{-1}$) | 15.68±2.50 | 105.25±3.40 | 185.50±7.72 | 460.50±17.31 | 859.00±14.09 | 1248.00±10.95 | 1680.50±8.81 | 250 | 500 |
| $HCO_3^-$ (mg $L^{-1}$) | 282.00±13.95 | 585.00±7.75 | 650.50±13.99 | 834.50±9.15 | 1389.50±5.97 | 1884.50±8.39 | 2254.50±7.55 | - | - |
| $CO_3^{2-}$ (mg $L^{-1}$) | 105.75±5.91 | 165.50±8.70 | 188.00±7.83 | 227.50±9.57 | 245.50±9.29 | 285.25±9.57 | 356.50±9.98 | - | - |
| $Na^+$ (mg $L^{-1}$) | 9.65±1.25 | 14.00±1.63 | 27.50±3.42 | 69.00±8.41 | 138.00±16.81 | 217.00±5.29 | 277.00±9.31 | - | - |
| $K^+$ ( mg $L^{-1}$) | 5.54±2.25 | 35.00±5.29 | 61.50±3.42 | 154.50±7.00 | 278.00±7.12 | 408.75±7.72 | 536.50±9.29 | - | - |
| $Ca^{2+}$ (mg $L^{-1}$) | 23.46±4.16 | 135.00±6.22 | 213.25±4.43 | 480.00±12.11 | 953.00±11.14 | 1425.00±10.00 | 1820.40±6.83 | 75 | 200 |
| $Mg^{2+}$(mg $L^{-1}$) | 12.15±1.50 | 37.50±4.43 | 46.00±5.89 | 96.50±8.39 | 159.50±03.42 | 231.00±5.29 | 284.00±11.78 | - | - |
| TKN (mg $L^{-1}$) | 24.27±5.08 | 60.59±3.07 | 75.56±6.49 | 136.00±8.60 | 320.50±5.17 | 456.00±10.77 | 572.50±8.29 | | 100 |
| $NO_3^{2-}$ (mg $L^{-1}$) | 25.17±4.16 | 117.50±5.00 | 173.25±7.80 | 430.50±5.26 | 760.00±6.73 | 1139.50±7.72 | 1430.50±8.14 | 45 | 100 |
| $PO_4^{3-}$ (mg $L^{-1}$) | 0.04±0.01 | 32.00±4.32 | 66.50±5.97 | 168.00±7.12 | 323.00±8.08 | 476.50±9.71 | 637.50±9.15 | - | - |
| $SO_4^{2-}$ (mg $L^{-1}$) | 17.64±2.57 | 96.50±9.57 | 155.25±4.99 | 298.50±18.41 | 633.00±7.75 | 954.50±5.97 | 1280.00±10.58 | 200 | 1000 |
| $Fe^{2+}$ (mg $L^{-1}$) | 0.28±0.04 | 1.53±0.30 | 3.05±0.60 | 7.75±1.71 | 15.25±2.99 | 22.75±4.27 | 28.90±5.97 | 0.30 | 1.0 |
| Zn (ppm) | 0.06±0.02 | 0.31±0.29 | 0.62±0.57 | 3.08±0.80 | 6.16±1.61 | 9.24±2.41 | 12.96±3.22 | 5.00 | 15 |
| Cd (ppm) | 0.1±0.01 | 0.13±0.05 | 0.16±0.02 | 0.34±0.06 | 0.67±0.09 | 0.99±0.07 | 1.33±0.08 | 0.1 | 2.00 |
| Cu (ppm) | 0.04±0.01 | 0.09±0.01 | 0.17±0.03 | 0.43±0.06 | 0.86±0.03 | 1.29±0.09 | 1.72±0.05 | 0.05 | 3.00 |
| Pb (ppm) | 0.02±0.01 | 0.09±0.01 | 0.18±0.04 | 0.45±0.03 | 0.91±0.06 | 1.36±0.09 | 1.81±0.06 | 0.05 | 1.00 |
| Mn (ppm) | 0.04±0.02 | 0.5±0.01 | 0.07±0.01 | 18±0.04 | 0.36±0.07 | 0.54±0.03 | 0.72±0.04 | 0.05 | 2.00 |
| SPC (SPC $ml^{-1}$) | 63±6.20 | $3.84\times10^4$±172 | $5.26\times10^5$±211 | $7.42\times10^6$±245 | $4.56\times10^7$±231 | $2.36\times10^8$±236 | $3.64\times10^{10}$±245 | - | 10000 |
| MPN (MPN100 $ml^{-1}$) | $2.56\times10^1$±15.25 | $4.86\times10^3$±236 | $6.75\times10^3$±342 | $8.36\times10^3$±423 | $4.56\times10^4$±652 | $6.62\times10^5$±864 | $4.58\times10^6$±1000 | 50 | 5000 |

Mean ± of six values; BWW - Borewell water; BIS - Bureau of Indian standard.

mill effluent of R.B.N.S. Sugar Mill, Laksar, Haridwar (Uttarakhand), India as also reported by Kumar and Chopra (2013e).

### Effect of sugar mill effluent on characteristics of soil

The mean ± SD values of various physico-chemical characteristics and heavy metals namely: soil texture, moisture content, WHC, BD, EC, pH, OC, $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, TKN, $PO_4^{3-}$, $SO_4^{2-}$, $Fe^{2+}$, Cd, Cu, Pb, Mn and Zn of the soil before and after irrigation of *A. esculentus* with different concentrations of sugar mill effluent namely 0% (BWW), 5%, 10%, 25%, 50%, 75% and 100% in two different seasons, that is, in the *Kharif* and *Zaid* season are given in Table 2. In the recent studies, Kaushik et al. (2004) reported that the distillery effluent irrigation increase the EC, pH, total organic carbon (TOC), total Kjeldahl nitrogen (TKN), available phosphorus, exchangeable Na, K, Ca, Mg, microbial population

**Table 2.** Physico-chemical characteristics of soil before and after irrigation of *A. esculentus* with sugar mill effluent, that is, after crop harvesting of 90 days in rainy (*Kharif*) and summer (*Zaid*) seasons.

| Parameter | Season | Before effluent irrigation | After effluent irrigation | | | | | | | r-value | F-calculated | Critical difference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Effluent concentration (%) | | | | | | | | | |
| | | | 0 (BWW) | 5 | 10 | 25 | 50 | 75 | 100 | | | |
| Soil texture | - | Loamy | Loamy | Loamy | Loamy | Loamy | Loamy | Loamy | Loamy | - | - | - |
| Soil moisture (%) | KS | 61.16±2.90 | 61.08±6.21 | 60.82±2.50 (-0.42) | 56.61±2.26 (-7.32) | 53.83±2.90 (-11.86) | 51.87a±3.97 (-15.07) | 50.27a±2.96 (-17.70) | 49.57a±2.13 (-18.84) | -0.95 | 6.26*** | 8.19 |
| | ZS | 61.16±2.90 | 61.08±6.21 | 58.64±2.50 (-3.99) | 54.84±2.26 (-10.21) | 51.23a±2.90 (-16.12) | 49.56a±3.97 (-18.86) | 48.24a±2.96 (-21.02) | 47.22a±2.13 (-22.69) | -0.95 | 7.65*** | 7.1 |
| WHC (%) | KS | 46.54±1.77 | 45.98±3.03 | 45.31±4.44 (-1.46) | 44.15±4.14 (-3.98) | 42.79±3.37 (-6.94) | 41.82±3.11 (-9.05) | 40.85±3.17 (-11.16) | 39.88±3.65 (-13.26) | -0.96 | 1.16NS | 0.2 |
| | ZS | 46.54±1.77 | 45.98±3.03 | 44.22±4.44 (-3.82) | 43.35±4.14 (-5.71) | 41.56±3.37 (-9.61) | 40.84±3.11 (-11.17) | 39.65±3.17 (-13.76) | 38.80±3.65 (-15.61) | -0.96 | 2.59NS | 10.64 |
| BD (gm $cm^{-3}$) | KS | 1.42±0.12 | 1.42±0.08 | 1.42±0.07 (0.00) | 1.42±0.07 (0.00) | 1.41±0.07 (-0.70) | 1.41±0.01 (-0.70) | 1.40±0.02 (-1.40) | 1.40±0.05 (-1.40) | -0.97 | 0.52NS | 8.96 |
| | ZS | 1.42±0.12 | 1.42±0.08 | 1.42±0.07 (0.00) | 1.42±0.07 (0.00) | 1.41±0.07 (-0.70) | 1.41±0.01 (-0.70) | 1.40±0.02 (-1.40) | 1.40±0.05 (-1.40) | -0.98 | 0.37NS | 0.2 |
| EC (dS $m^{-1}$) | KS | 2.18±0.73 | 2.08±0.08 | 2.42a±0.32 (+16.34) | 2.52a±14 (+21.15) | 2.80a±13 (+34.61) | 3.13a±0.20 (+50.48) | 3.40a±13 (+63.46) | 3.83a±0.33 (+84.13) | +0.98 | 33.68*** | 0.31 |
| | ZS | 2.18±0.73 | 2.07±0.88 | 2.45a±0.37 (+18.35) | 2.54a±16 (+22.70) | 2.82a±16 (+36.23) | 3.15a±0.22 (+52.17) | 3.42a±0.09 (+65.21) | 3.86a±0.37 (+86.47) | +0.98 | 27.4*** | 0.35 |
| pH | KS | 7.99±18 | 7.50±0.26 | 7.47±0.25 (-0.40) | 7.40±0.26 (-1.33) | 7.02±18 (-6.40) | 6.65a±0.33 (-11.33) | 6.27a±0.31 (-16.40) | 6.25a±0.85 (-16.66) | -0.97 | 7.39*** | 0.6 |
| | ZS | 7.99±18 | 7.52±0.26 | 7.42±17 (-1.32) | 7.37±0.22 (-1.99) | 7.00±0.23 (-6.91) | 6.63a±0.37 (-11.83) | 6.24a±0.35 (-17.02) | 6.22a±0.81 (-17.28) | -0.98 | 7.63*** | 0.59 |
| OC (mg $Kg^{-1}$) | KS | 0.51±10 | 0.43±10 | 0.99±10 (+130.23) | 1.95±19 (+353.48) | 5.16a±0.38 (+1100.00) | 9.81a±0.63 (+2181.39) | 14.79a±1.43 (+3339.53) | 16.82a±1.93 (+3811.62) | +0.99 | 199.83*** | 1.4 |
| | ZS | 0.51±10 | 0.42±10 | 1.00±0.07 (+138.09) | 1.97a±17 (+369.04) | 5.17a±0.36 (+1130.95) | 9.82a±0.61 (+2238.09) | 14.80a±1.41 (+3423.80) | 17.06a±1.86 (+3961.90) | +0.99 | 216.53*** | 1.36 |

**Table 2.** Cont'd.

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $Na^+$ (mg Kg $^{-1}$) | KS | 21.32±2.50 | 17.56±2.51 | 22.81±1.71 (+29.89) | 24.22±2.95 (+37.92) | 27.10a±4.57 (+54.32) | 33.28a±3.48 (+89.52) | 49.84a±6.09 (+183.82) | 65.50a±8.70 (+273.00) | +0.98 | 50.82*** | 7.11 |
| | ZS | 21.32±2.50 | 16.58±2.51 | 24.06a±2.83 (+45.11) | 25.47a±4.01 (+53.61) | 28.35a±3.31 (+70.98) | 34.53a±3.95 (+108.26) | 49.84a±6.09 (+200.60) | 66.75a±7.54 (+302.59) | +0.98 | 55.25*** | 6.83 |
| $K^+$ (mg Kg $^{-1}$) | KS | 169.01±3.06 | 154.09±6.70 | 160.63±3.74 (+4.24) | 171.84±5.48 (+11.51) | 224.55a±8.21 (+45.72) | 219.97a±9.85 (+42.75) | 210.06a±4.30 (+36.33) | 202.77a±6.03 (+31.59) | +0.64 | 77.37*** | 9.76 |
| | ZS | 169.01±3.06 | 152.29±6.70 | 160.63±3.74 (+45.53) | 171.84a±5.48 (+12.83) | 224.55a±8.21 (+47.44) | 219.97a±9.85 (+44.44) | 210.06a±4.30 (+37.93) | 202.77a±6.03 (+33.14) | +0.64 | 77.37*** | 9.76 |
| $Ca^{2+}$ (mg Kg $^{-1}$) | KS | 17.73±1.94 | 14.11±2.69 | 19.15±3.24 (+35.71) | 23.56a±2.42 (+66.97) | 132.54a±7.36 (+839.33) | 129.92a±4.02 (+820.76) | 122.03a±4.59 (+764.84) | 117.08a±4.76 (+729.96) | +0.75 | 498.99*** | 7.53 |
| | ZS | 17.73±1.94 | 13.18±2.69 | 19.15±3.24 (+45.29) | 23.56a±2.42 (+78.75) | 132.54a±7.36 (+905.61) | 129.92a±4.02 (+885.70) | 122.03a±4.59 (+825.87) | 117.08a±4.76 (+788.31) | +0.75 | 498.99*** | 7.53 |

**Table 2.** Contd.

| Parameter | Season | Before effluent irrigation | After effluent irrigation | | | | | | | r-value | F-calculated | Critical difference |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Effluent concentration (%) | | | | | | | | | |
| | | | 0 (BWW) | 5 | 10 | 25 | 50 | 75 | 100 | | | |
| $Mg^{2+}$ (mg Kg $^{-1}$) | KS | 1.72±10 | 1.68±0.59 | 3.43±0.77 (+104.16) | 4.43±0.65 (+163.69) | 21.04±2.38 (+1152.38) | 18.94±5.62 (+1027.38) | 16.75a±4.05 (+897.02) | 14.12a±3.04 (+740.47) | +0.62 | 28.49*** | 4.46 |
| | ZS | 1.72±10 | 1.56±0.59 | 3.43±0.77 (+119.87) | 4.43±0.65 (+183.97) | 21.04±2.38 (+1248.70) | 18.94a±5.62 (+1114.10) | 16.75a±4.05 (+973.71) | 14.12a±3.04 (+805.12) | +0.62 | 28.49*** | 4.46 |
| TKN (mg Kg $^{-1}$) | KS | 33.41±3.94 | 30.96±4.09 | 58.63a±3.11 (+89.37) | 74.50a±8.85 (+140.63) | 144.08a±3.27 (+365.37) | 285.18a±8.48 (+821.12) | 405.50a±5.97 (+1209.75) | 461.50a±5.00 (+1390.63) | +0.99 | 3478.5*** | 8.75 |
| | ZS | 33.41±3.94 | 29.86±4.09 | 58.63a±3.11 (+96.34) | 74.50a±8.85 (+149.49) | 144.08a±3.27 (+382.51) | 285.18a±8.48 (+855.05) | 405.50a±5.97 (+1258.00) | 461.50a±5.00 (+1445.54) | +0.99 | 3478.5*** | 8.75 |
| $PO_4^{3-}$ (mg Kg $^{-1}$) | KS | 55.54±5.72 | 51.75±4.79 | 57.39±6.65 (+10.89) | 66.78a±8.34 (+29.04) | 70.56a±7.24 (+36.34) | 121.06a±5.23 (+133.93) | 173.27a±4.55 (+234.82) | 226.56a±6.63 (+337.79) | +0.99 | 453.43*** | 9.32 |
| | ZS | 55.54±5.72 | 50.85±4.79 | 57.89±5.81 (+13.84) | 67.28a±9.16 (+32.31) | 71.06a±6.52 (+39.74) | 121.56a±4.77 (+139.05) | 173.77a±4.30 (+241.73) | 227.06a±5.79 (+346.52) | +0.99 | 494.9*** | 8.93 |

**Table 2.** Cont'd.

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $SO_4^{2-}$ (mg $Kg^{-1}$) | KS | 75.64±5.13 | 73.12±7.37 | 79.09±3.33 (+8.16) | 89.72a±6.86 (+22.70) | 102.39a±8.43 (+40.03) | 110.95a±5.19 (+51.73) | 121.18a±8.92 (+65.72) | 130.00a±9.52 (+77.78) | +0.96 | 33.52*** | 10.84 |
| | ZS | 75.64±5.13 | 72.62±7.37 | 80.34±4.61 (+10.63) | 90.97a±6.08 (+25.26) | 103.64a±7.05 (+42.71) | 112.20a±4.23 (+54.50) | 122.43a±7.26 (+68.58) | 131.50a±7.72 (+81.07) | +0.96 | 44.98*** | 9.51 |
| $Fe^{2+}$ (mg $Kg^{-1}$) | KS | 3.13±0.44 | 2.63±0.85 | 3.14±0.38 (+19.39) | 3.63±0.40 (+38.02) | 4.76a±0.37 (+80.78) | 5.71a±0.44 (+117.11) | 8.71a±0.54 (+231.17) | 10.57a±2.21 (+301.90) | +0.99 | 28.72*** | 1.64 |
| | ZS | 3.13±0.44 | 2.43±0.85 | 3.39±0.33 (+39.50) | 3.88±0.45 (+59.67) | 5.01a±0.54 (+106.17) | 5.96a±0.61 (+145.26) | 8.96a±0.75 (+268.72) | 10.82a±2.10 (+345.26) | +0.99 | 29.05*** | 1.66 |
| Zn (ppm) | KS | 1.096±17 | 0.765±16 | 1.068a±12 (+39.60) | 1.405a±12 (+83.66) | 2.481a±0.27 (+224.31) | 2.745a±0.29 (+258.82) | 3.275a±0.35 (+328.10) | 3.315a±0.64 (+333.33) | +0.93 | 42.28*** | 0.48 |
| | ZS | 1.096±17 | 0.745±16 | 1.093±11 (+46.71) | 1.430a±10 (+91.94) | 2.506a±0.24 (+236.37) | 2.770a±0.29 (+271.81) | 3.300a±0.39 (+342.95) | 3.340a±0.63 (+348.32) | +0.92 | 43.37*** | 0.48 |
| Cd (ppm) | KS | 0.080±0.07 | 0.040±0.06 | 0.096a±0.06 (+140.00) | 0.101a±0.1 (+152.50) | 0.160a±0.02 (+300.00) | 0.166a±0.02 (+315.00) | 0.213a±0.03 (+432.50) | 0.266a±0.03 (+565.00) | +0.96 | 17.65*** | 0.05 |
| | ZS | 0.080±0.07 | 0.053±0.06 | 0.098±0.06 (+84.90) | 0.103±0.02 (+94.33) | 0.163a±0.02 (+207.54) | 0.168a±0.03 (+216.98) | 0.216a±0.03 (+307.54) | 0.285a±0.04 (+437.73) | +0.97 | 16.06*** | 0.06 |
| Cu (ppm) | KS | 2.137±0.35 | 2.003±0.33 | 2.199±0.32 (+9.78) | 2.305±0.33 (+15.07) | 5.049a±0.20 (+152.07) | 6.667a±1.07 (+232.85) | 9.420a±0.91 (+370.29) | 10.367a±0.80 (+417.57) | +0.99 | 116.55*** | 0.96 |
| | ZS | 2.137±0.35 | 2.002±0.33 | 2.224±0.29 (+11.08) | 2.330±0.30 (+16.38) | 5.074a±0.23 (+153.44) | 6.692a±1.02 (+234.26) | 9.445a±0.92 (+371.77) | 10.393a±0.77 (+419.13) | +0.99 | 123.09*** | 0.93 |
| Pb (ppm) | KS | 0.17±0.1 | 0.16±0.1 | 0.041±0.03 (+156.25) | 0.046±0.02 (+187.50) | 0.096±0.1 (+500.00) | 0.155±0.1 (+868.75) | 0.236a±0.02 (+1375.00) | 0.254a±0.09 (+1487.50) | +0.99 | 27.29*** | 0.05 |
| | ZS | 0.17±0.1 | 0.15±0.1 | 0.043±0.00 (+186.66) | 0.048±0.02 (+220.00) | 0.098±0.1 (+553.33) | 0.158±0.1 (+953.33) | 0.238a±0.1 (+1486.66) | 0.271a±11 (+1706.66) | +0.99 | 22.18*** | 0.06 |
| Mn (ppm) | KS | 0.117±0.06 | 0.104±0.06 | 0.248±0.1 (+138.46) | 0.269±0.1 (+158.65) | 0.572a±0.04 (+450.00) | 0.883a±0.09 (+749.03) | 1.381a±16 (+1227.88) | 1.524a±0.38 (+1365.38) | +0.99 | 49.37*** | 0.24 |
| | ZS | 0.117±0.06 | 0.103±0.06 | 0.250±0.1 (+142.71) | 0.272±0.1 (+164.07) | 0.575a±5 (+462.74) | 0.885a±0.09 (+759.22) | 1.383a±16 (+1242.71) | 1.527a±0.38 (+1382.52) | +0.99 | 50.2*** | 0.24 |

Mean ± of six values; Significant F - ***P > 0.1%; r - Coefficient of correlation; % Increase or decrease in comparison to control given in parenthesis; a - significantly different to the control; NS - Not Significant; BWW - Borewell water; KS - *Kharif* season; ZS - *Zaid* season.

and soil enzyme activities. Baskaran et al. (2009) found that distillery effluent (spent wash) discharged as wastewater contains various nutrients and toxic chemicals that can contaminate water and soil and may affect the common crops if used for agricultural irrigation. The pH is an important parameter as many nutrients are available only at a particular range of pH for plant uptake. A range of pH 6.0-8.2 provides predominating bacterial activity and is favorable for maximum yield of crops. A shift in pH outside that range renders the nutrients less available, even though they remain in the soil. Under acidic conditions, elements such as iron, aluminium, manganese and the heavy metals (zinc, copper, and chromium) become highly soluble and may create problems for vegetation (Roy et al., 2007; Samuel and Muthukkaruppan, 2011; Kumar and Chopra, 2014).

The higher concentration of Na in the soil after effluent irrigation is associated with presence of higher concentration of carbonate, bicarbonate in the effluent (Rattan et al., 2005; Hati et al., 2007; Kumar and Chopra, 2013g). Long term application of distillery effluent significantly increased the OC, TKN, K, P and soil enzymatic activities in the soil. Short terms application of 50% distillery effluent proved to be the most useful in improving the properties of sodic soil reported by Kaushik et al. (2004). Effluent irrigation generally adds significant quantity of salts to the soil environment, such as sulfates, phosphates, bicarbonates, chlorides of the cations sodium, calcium, potassium and magnesium; they stimulate the growth at lower concentration but inhibit at higher concentration (Maliwal et al., 2004; Kumar and Chopra, 2013e, f). Kumar and Chopra (2012) reported that the wastewater irrigation significantly increased the concentration of various heavy metals like Cu, Zn, Cd, Ni, Cr and Pb in the soil and vegetables. These heavy metals accumulate in vegetables and pose various health problems in humans and animals after ingestion.

During the present study, the soil characteristics have been found to change on irrigation with sugar mill effluent. It was observed that after 90 days of *A. esculentus* crop-harvesting, the soil particle size depicted that the soil was loamy and no drastic change in soil texture occurred with the application of all the concentrations of sugar mill effluent throughout the period of the experiment. The soil characteristics changed on irrigation with different concentrations (5 to 100%) of the effluent. Among different concentrations of sugar mill effluent, irrigation with 100% effluent concentration showed the maximum decrease in moisture content, WHC and BD, and maximum increase in EC, OC, $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, $Fe^{2+}$, TKN, $PO_4^{3-}$, $SO_4^{2-}$, Cd, Cu, Pb, Mn and Zn in both cultivated seasons (Table 2).

The ANOVA showed that different concentrations of sugar mill effluent affected differently on various soil characteristics. The sugar mill effluent concentrations 50%, 75% and 100% showed significant ($P<0.001$) effect on moisture content in both cultivated seasons, that is, *Kharif* and *Zaid* seasons. It was quite interesting to note that the moisture content was also found to be significantly ($P<0.001$) affected with 25% effluent concentrations in *Zaid* season. WHC and BD were found to be insignificantly ($P>0.05$) affected with different concentrations of sugar mill effluent in both cultivated seasons.

Among effluent concentrations, 10% to 100% of sugar mill effluent showed significant ($P<0.001$) effect on EC, OC, TKN, $K^+$, $Ca^{2+}$, $PO_4^{3-}$, $SO_4^{2-}$, Zn and Cd in the *A. esculentus* cultivated soil in both seasons. The concentration of 5% of sugar mill effluent was also shown to have significant ($P<0.001$) effect on EC and TKN in both cultivated seasons. The sugar mill effluent concentrations of 25% to 100% significantly ($P<0.001$) affected the $Na^+$, $Mg^{2+}$, $Fe^{2+}$, Cu and Mn of the soil in both cultivated seasons. The pH of the treated soil was affected significantly ($P<0.001$) with 50%, 75% and 100% concentration of sugar mill effluent, while that of Pb was affected with 75% and 100% concentration of sugar mill effluent. The $Na^+$ of the soil were also affected significantly ($P<0.001$) with 5% sugar mill effluent in the *Zaid* season (Table 2).

The r value of the soil parameters: EC (r = +0.98), OC (r = +0.99), $Na^+$ (r = +0.98), $K^+$ (r = +0.64), $Ca^{2+}$ (r = +0.75), $Mg^{2+}$(r = +0.62), $Fe^{2+}$ (r = +0.99), TKN (r = +0.99), $PO_4^{3-}$ (r = +0.99), $SO_4^{2-}$ (r = +0.96), Cd (r = +0.96 to +0.97), Cu (r = +0.99), Pb (r = +0.99), Mn (r = +0.99) and Zn (r = +0.92 to +0.93) were shown to have positive correlation with different concentration of sugar mill effluent in both the *Kharif* and *Zaid* seasons (Table 2).

The content of heavy metals, Cd, Cu, Pb, Mn and Zn in the soil was increased as the effluent concentration increased (Table 2). Among heavy metals, the concentration of Mn was found to be the maximum, while that of Pb was found to be the minimum after sugar mill effluent irrigation in both cultivated seasons. The contamination factor (Cf) indicated the contamination rate of heavy metals in the soil after sugar mill effluent irrigation.

The contamination factor of various heavy metals, Mn (14.65 and 14.82), showed maximum contamination in both cultivated seasons, while that of Pb (1.58 and 1.80) showed minimum contamination in both the *Kharif* and *Zaid* seasons with 100% concentration of sugar mill effluent irrigated soil. The contamination factor (Cf) of various heavy metals was recorded in order of Mn>Cd>Cu>Zn>Pb after irrigation with sugar mill effluent in both seasons (Figure 1). Kumar and Chopra (2012) reported that distillery effluent irrigation increased the EC, $Cl^-$, TOC, $HCO_3^-$, $CO_3^{2-}$, $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, TKN, $NO_3^{2-}$, $PO_4^{3-}$ and $SO_4^{2-}$, Fe, Zn, Cd, Cu, Pb and Cr of the soil. Among the heavy metals, the maximum enrichment factor (Ef) was shown by Cd (31.33), while the minimum was shown by Fe (4.59) and it was in order of Cd>Cr>Pb>Zn>Cu>Fe after irrigation with distillery effluent.

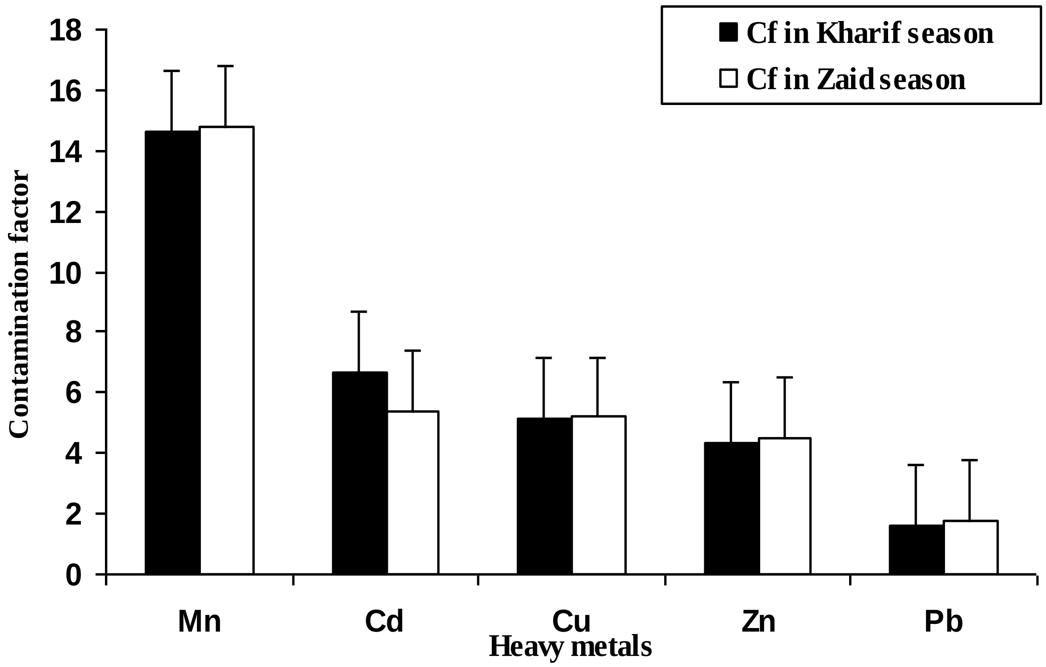


**Figure 1.** Contamination factor of heavy metals in soil after irrigation with sugar mill effluent.

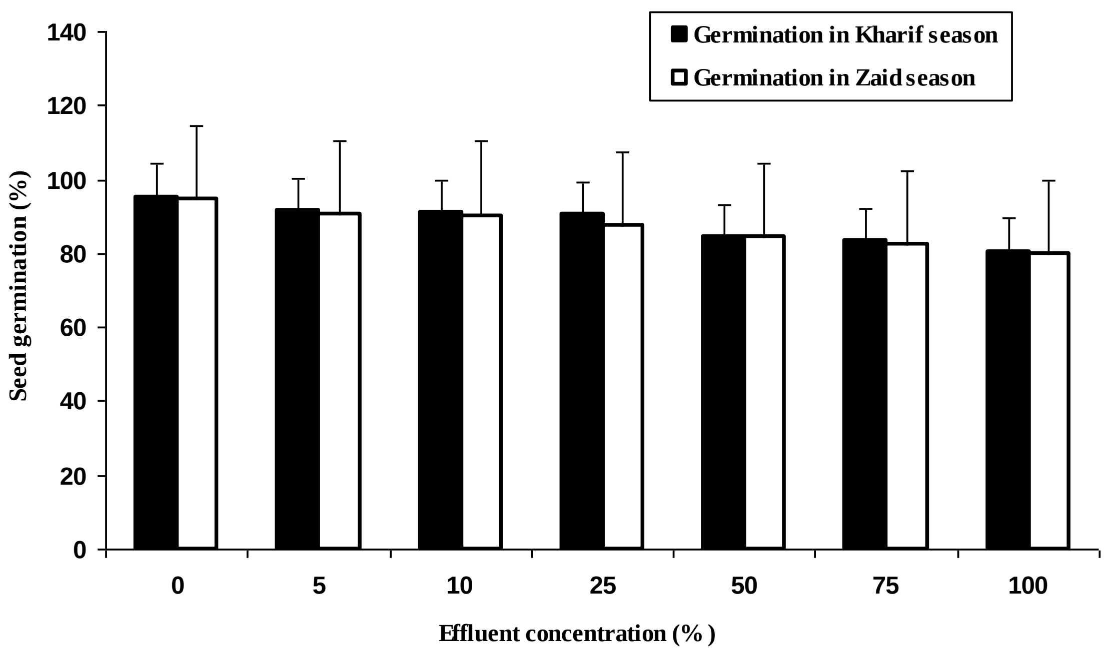


**Figure 2.** Seed germination of *A. esculentus* after irrigation with sugar mill effluent.

## Agronomical characteristics of *A. esculentus*

### *Effect of A. esculentus on germination*

The seed emergence period, seed germination and relative toxicity (RT) against seed germination of *A. esculentus* at different concentrations, namely: 5%, 10%, 25%, 50%, 75% and 100% of sugar mill effluent along with control (BWW) in the *Kharif* and *Zaid* seasons are shown in Figures 2 and 3.

In the recent studies, it has been observed that the availability, uptake and leaching of nutrients is greatly

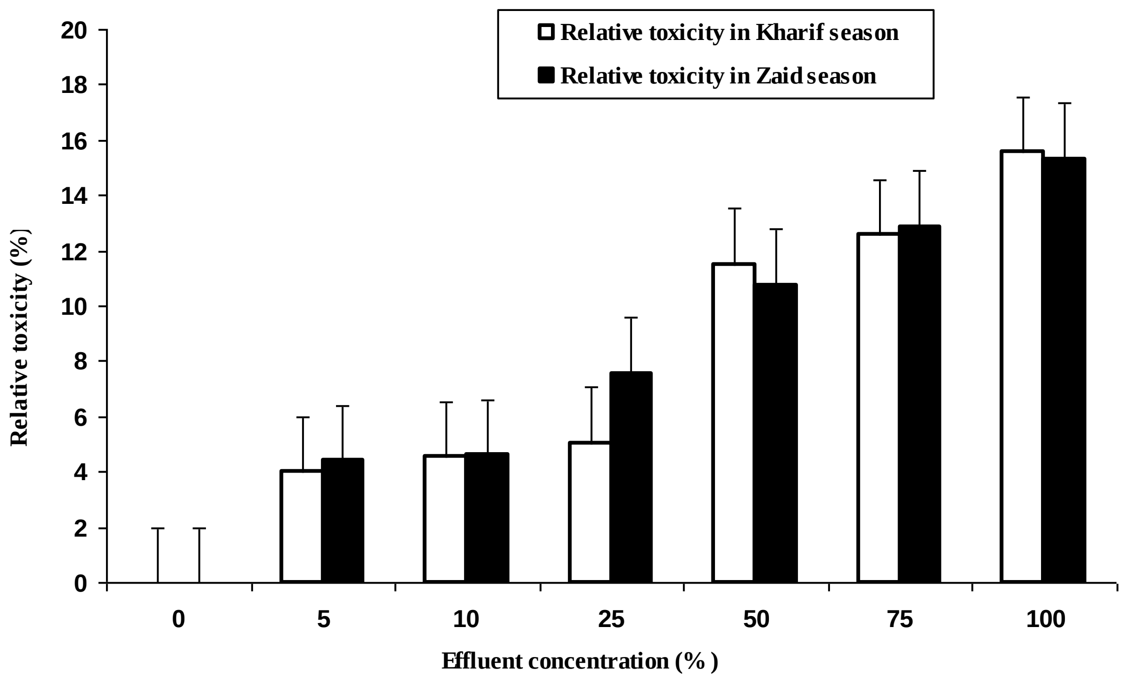


**Figure 3.** Relative toxicity of sugar mill effluent against seed germination of *A. esculentus*.

influenced by a number of physico-chemical factors. Among various physico-chemical factors, pH plays a significant role in the soil. In the acidic soil environment, the availability of the basic cations like $Ca^{2+}$, $Mg^{2+}$, $K^{+}$ and $Na^{+}$ becomes lower due to leaching. Thus, the availability of these nutrients decreases as the acidic character of the soil increases; however it directly affects the vegetative growth of the crop plants as it gradually decreased as the effluent concentration increased (Osaigbovo et al., 2006; Ezhilvannan et al., 2011; Vijayaragavan et al., 2011). Salts are usually most damaging to young plants but not necessarily at the time of germination, although high salt concentration can slow seed germination by several days or completely inhibit it. Because soluble salts move readily with water, evaporation moves salts to the soil surface where they accumulate and make the soil surface harden as a result of the delay in germination (Krishna, 2002; Kannan and Upreti, 2008).

The present study showed that at germination stage 0-15 days, the maximum seed germination (94.50% and 95.33%) of *A. esculentus* was recorded with control (BWW) while that of minimum germination (80.00% and 80.50%) was recorded with 100% concentration of sugar mill effluent. It was found to be negatively correlated (r = -0.95 and -0.96) and significantly different with sugar mill effluent concentrations in both the *Kharif* and *Zaid* seasons. The ANOVA showed that the concentrations of 50%, 75% and 100% of sugar mill effluent showed significant (P<0.001) effect on seed germination of *A. esculentus* in both cultivated seasons. The maximum relative toxicity (15.34 and 15.55%) of the sugar mill effluent against the seed germination of *A. esculentus* was recorded with 100% concentration of sugar mill effluent and it was positively correlated (r = +0.96) with different concentrations of sugar mill effluent in both cultivated seasons (Figures 2 and 3).

This type of germination pattern of *A. esculentus* is likely due to the presence of toxicants in the higher concentration of effluent which may inhibit the germination at higher concentrations as observed earlier for the crop, *T. foenum-graecum* (Kumar and Chopra, 2012). Ramana et al. (2002) observed that the effect of the distillery effluent is crop-specific. The distillery effluent did not show any inhibitory effect on seed germination at low concentration except in tomato, but in onion the germination was significantly higher (84%) at 10% concentration in comparison to 63% in the control.

### *Effect of A. esculentus on vegetative growth stage*

The various parameters of vegetative growth stage, that is, (at 45 days) such as shoot length, root length, dry weight, chlorophyll content and LAI/plant of *A. esculentus* at different concentrations namely: 5%, 10%, 25%, 50%, 75% and 100% of sugar mill effluent along with control (BWW) in both the *Kharif* and *Zaid* seasons are given in Table 3.

In this study, the maximum shoot length, root length, dry weight, chlorophyll content and LAI/plant of *A. esculentus* were recorded with 25% concentration of sugar mill effluent in both the *Kharif* and *Zaid* seasons

**Table 3.** Agronomical characteristics of *A. esculentus* after irrigation with sugar mill effluent at vegetative (45 days) and flowering stage (60 days) in rainy (*Kharif*) and summer (*Zaid*) seasons.

| Effluent Concentration | Agronomical parameters | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Vegetative growth stage (45 days) | | | | | | | | | | Flowering stage (60 days) | |
| | Shoot length (cm) | | Root length (cm) | | Dry Wt. (g) | | Chlo. content (mg./g.f.wt) | | LAI | | No. of flowers/plant | |
| | KS | ZS | KS | ZS | KS | ZS | | KS | ZS | KS | KS | ZS |
| 0 | 122.45±4.98 | 124.29±3.82 | 12.28±2.78 | 12.78±3.13 | 17.25±2.84 | 17.61±3.12 | 2.99±0.87 | 3.01±0.88 | 2.31±0.68 | 2.33±0.72 | 32.33±3.44 | 32.50±3.21 |
| 5 | 124.60±4.44 | 126.60±4.93 | 12.61±2.89 | 13.36±2.97 | 17.86±2.35 | 19.03±4.91 | 3.34±0.96 | 3.38±0.90 | 3.08±0.80 | 3.12±0.74 | 35.50±3.21 | 35.67±3.08 |
| 10 | 125.79±4.88 | 127.79±4.25 | 13.42±3.77 | 13.75±3.39 | 18.40±2.84 | 19.56±4.68 | 3.48±1.00 | 3.51±0.95 | 3.52±0.95 | 3.55±0.97 | 36.33±5.13 | 36.50±5.13 |
| 25 | 137.24a±5.12 | 139.34a±3.44 | 18.00a±3.82 | 18.30a±3.23 | 22.66a±1.35 | 24.05a±1.75 | 5.71a±1.91 | 5.74a±1.89 | 5.08a±1.02 | 5.15a±0.96 | 37.00±7.87 | 44.00±3.41 |
| 50 | 133.76a±3.46 | 135.77a±4.18 | 16.62±3.50 | 15.70±3.93 | 19.99±1.97 | 20.86a±2.53 | 4.24±1.56 | 4.28±1.55 | 4.72a±1.54 | 4.89a±1.45 | 43.83±3.54 | 41.27±5.78 |
| 75 | 133.68a±2.76 | 135.68a±2.05 | 14.44±2.75 | 14.17±2.36 | 19.15±2.58 | 21.75±3.47 | 4.05±1.09 | 4.08±1.09 | 4.36a±1.24 | 4.75a±1.53 | 40.00±5.76 | 38.00±7.82 |
| 100 | 133.06a±3.16 | 135.06a±2.57 | 13.03±3.35 | 12.24±2.61 | 18.96±2.43 | 20.35±1.31 | 3.97±0.89 | 4.00±0.89 | 3.81±1.09 | 3.84±1.08 | 37.83±7.78 | 37.17±7.94 |
| r-Value | +0.66 | +0.65 | +13 | -14 | +0.23 | +0.38 | +0.27 | +0.27 | +0.47 | +0.50 | +0.59 | +0.27 |
| CD | 6.04 | 5.34 | 4.73 | 4.5 | 3.69 | 4.82 | 1.78 | 1.75 | 1.55 | 2.12 | 7.99 | 7.96 |
| F-calculated | 10.90*** | 14.24*** | 2.63* | 2.62* | 2.95* | 3.17* | 3.05* | 3.17* | 4.82** | 3.14* | 2.54* | 2.56* |

Mean ± SD of six values; Significant F - ***P - 0.1%, **P - 1% level, *P - 5% level, r - Coefficient of correlation; BWW - Borewell water; CD - Critical difference; KS - *Kharif* season; ZS - *Zaid* season.

(Table 3). The ANOVA indicated that the concentrations of sugar mill effluent had significant (P<0.05) effect on shoot length, root length, dry weight, chlorophyll content and LAI/plant of *A. esculentus* in both the *Kharif* and *Zaid* seasons. Shoot length and chlorophyll content of *A. esculentus* was also found more significantly (P<0.01) different in the *Zaid* season with different concentrations of sugar mill effluent. The 25% concentration of sugar mill effluent showed significant (P<0.05) effect on shoot length, root length, dry weight, chlorophyll content and LAI/plant of *A. esculentus* in both the *Kharif* and *Zaid* seasons. Shoot length of *A. esculentus* was also found significantly (P<0.01) different with 50% to 100% concentration of sugar mill effluent in both cultivated seasons. The sugar mill effluent concentrations of 50% and 75% also showed significant effect on LAI in both cultivated seasons (Table 3).

Bharagava et al. (2008) reported that post methanated distillery effluent irrigation increased the chlorophyll and protein contents in Indian mustard plants (*Brassica nigra* L.) at the lower concentrations (25% and 50%) of distillery effluent followed by a decrease at higher concentrations (75% and 100%) of distillery effluent as compared to their respective controls. In *Zea mays*, the maximum chlorophyll content was found with 25% concentration of brewery effluent while that of plant height, biomass and leaf number were maximum at 100% concentration as reported by Orhue et al. (2005). Osaigbovo et al. (2006) reported maximum plant height and number of leaves and chlorophyll content at 25% of pharmaceutical effluent treatment on maize plant.

**Table 4.** Agronomical characteristics of *A. esculentus* after irrigation with sugar mill effluent at maturity stage (90 days) in rainy (*Kharif*) and summer (*Zaid*) seasons.

| Effluent Concentration | Agronomical parameters - Maturity stage (90 days) | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | No. of pods (I-H) | | No. of pods (II-H) | | No. of pods (III-H) | | Crop yield (g) | | HI (%) | |
| | KS | ZS | KS | ZS | KS | ZS | KS | ZS | KS | ZS |
| 0 | 11.00±3.10 | 11.33±3.33 | 12.00±3.79 | 12.33±4.27 | 8.17±2.32 | 8.33±2.42 | 221.78±33.53 | 222.62±30.90 | 553.37±8.36 | 555.19±3.66 |
| 5 | 13.50±2.26 | 13.67±2.50 | 13.50±2.35 | 13.67±2.58 | 9.67±3.08 | 9.83±3.06 | 234.29±38.85 | 245.80±26.79 | 594.02a±8.57 | 618.60a±4.56 |
| 10 | 14.50±2.59 | 14.67±2.73 | 14.83±3.60 | 15.00±3.69 | 9.83±3.31 | 10.00±3.52 | 242.76±15.79 | 251.08±9.24 | 596.07a±6.85 | 6312a±7.52 |
| 25 | 17.33a±2.94 | 17.50a±2.88 | 19.17a±3.31 | 19.33a±3.14 | 15.17a±2.14 | 15.33a±1.86 | 288.06a±39.69 | 301.16a±40.39 | 620.93a±7.57 | 639.00a±8.32 |
| 50 | 15.17a±2.71 | 15.33±2.80 | 16.00±2.83 | 16.17±2.86 | 12.83±3.76 | 13.00±3.95 | 249.54±29.76 | 257.88±18.34 | 611.41a±8.24 | 615.07a±4.25 |
| 75 | 15.17a±2.32 | 15.33±2.42 | 15.50±1.97 | 15.67±2.07 | 12.67±3.72 | 12.83±3.92 | 234.35±31.22 | 242.69±27.28 | 602.62a±5.99 | 606.89a±4.78 |
| 100 | 14.83±2.79 | 15.00±2.90 | 15.33±4.18 | 15.50±4.23 | 10.00±5.18 | 11.74±5.42 | 230.86±30.07 | 239.19±26.98 | 600.33a±6.33 | 598.55a±6.58 |
| r-Value | +0.58 | +0.41 | +0.48 | +0.32 | +0.27 | +0.27 | +10 | -0.06 | +0.47 | +0.1 |
| CD | 3.86 | 4.03 | 4.64 | 4.81 | 5.01 | 5.19 | 46.11 | 39.12 | 9.64 | 5.68 |
| F-calculated | 3.08* | 2.68* | 2.84* | 2.55* | 2.91* | 2.71* | 2.73* | 4.85*** | 2.54* | 3.22* |

Mean ± SD of six values; Significant F - ***P - 0.1%, **P - 1% level, *P - 5% level, r - Coefficient of correlation; BWW - Borewell water; CD - Critical difference; KS - *Kharif* season; ZS - *Zaid* season.

Total chlorophyll content has been found to decrease gradually with the increase in distillery/sugar mill effluent concentration in *V. faba* as reported by Kumar and Chopra (2013c, e). Therefore, the 25% sugar mill effluent concentration contains optimum contents of nutrients required for maximum vegetative growth of *A. esculentus.*

***Effect of A. esculentus on flowering stage***

The parameters of flowering stage, that is, (at 60 days)
such as number of flowers/plant of *A. esculentus* at different concentrations namely: 5%, 10%, 25%, 50%, 75% and 100% of sugar mill effluent along with control (BWW) in both the *Kharif* and *Zaid* seasons are given in Table 3. In the present study, at flowering stage, that is, at 60 days, the maximum flowers and pods/plant of *A. esculentus* were recorded with 25% concentration of sugar mill effluent in both the *Kharif* and *Zaid* seasons. The 25% concentration of sugar mill effluent had significant (P<0.05) effect on number of flowers/plant of *A. esculentus* in both cultivated seasons (Table 3). Thus, the 25% sugar mill effluent concentration contains optimum contents of nutrients required for maximum flowering of *A. esculentus.* Kumar and Chopra (2012) also recorded the same trend of flowering of *T. foenum-graecum* after irrigation with distillery effluent.

***Effect of A. esculentus on maturity stage***

The various parameters of maturity stage (at 90 days) such as number of pods/plant, crop yield/plant and harvest index (HI) of *A. esculentus* at different concentrations namely: 5%, 10%, 25%, 50%, 75% and 100% of sugar mill effluent along with control (BWW) in both the *Kharif* and *Zaid* seasons are given in Table 4.

The present study showed that the maximum number of pods/plant at I, II and III harvest, yield/plant and HI of *A. esculentus* was recorded with 25% concentration of sugar mill effluent in both the *Kharif* and *Zaid* seasons. The 25% concentration of sugar mill effluent showed significant (P<0.05) effect on number of pods/plant at I, II and III harvest, yield/plant and HI of *A. esculentus* in both cultivated seasons (Table 4). Number of pods at I harvest of *A. esculentus* was also found to be significantly (P<0.05) different in the *Kharif* season with 50% and 75% concentration of sugar mill effluent. The yield of *A. esculentus* was also found to be significantly (P<0.05) different with 50% concentration of sugar mill effluent and it was also recorded to be significantly (P<0.001) affected with 25% concentration of sugar mill effluent. The harvest index of *A. esculentus* was significantly (P<0.05) affected with 5% to 100% concentrations of sugar mill effluent in the *Kharif* season, and 10% to

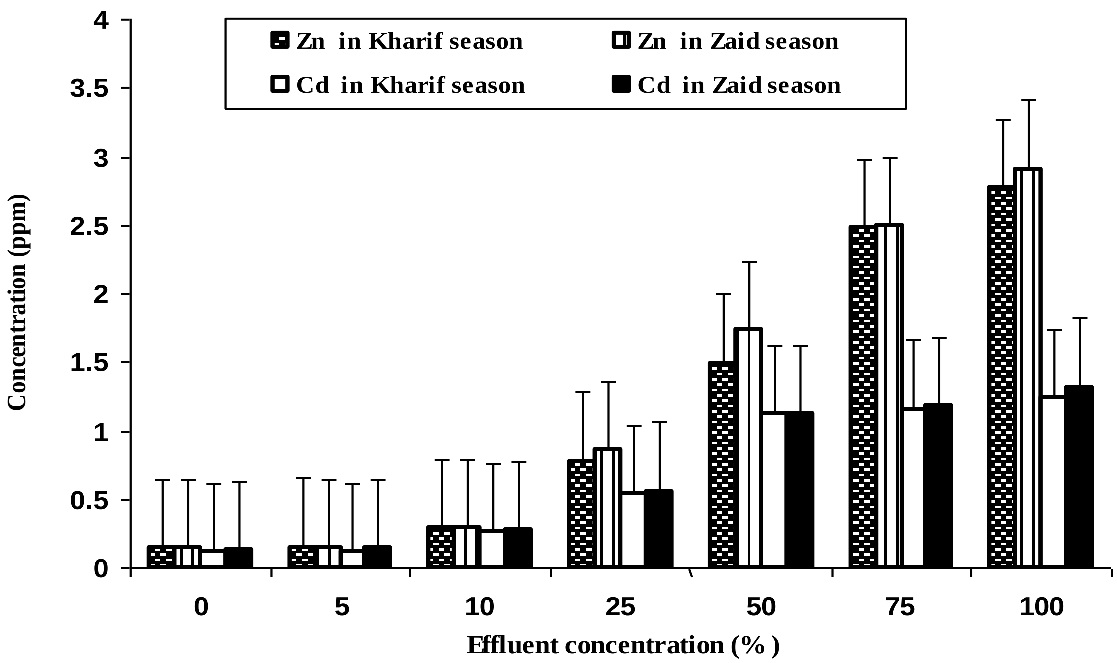


**Figure 4.** Zn and Cd concentration in *A. esculentus* after irrigation with sugar mill effluent.

100% concentrations of sugar mill effluent in the *Zaid* season (Table 4). The findings were in accordance with those of Kumar and Chopra (2013b) who reported that the number of pods, crop yield and harvest index of *A. esculentus* decreased when the concentration of paper mill effluent increased. The 25% sugar mill effluent concentration was observed to be favourable for maturity of *A. esculentus.* Therefore, 25% sugar mill effluent concentration can be used for the maximum growth and yield of *A. esculentus.*

## Micronutrients in *A. esculentus*

The content of various heavy metals namely: Cd, Cu, Pb, Mn and Zn, of *A. esculentus* and their contamination factor (Cf) after irrigation with different concentrations of 5%, 10%, 25%, 50%, 75% and 100% of sugar mill effluent along with control (BWW) in both the *Kharif* and *Zaid* seasons are given in Figures 4, 5, 6 and 7.

During the present study, ANOVA showed that the concentrations of 25%, 50%, 75% and 100% of sugar mill effluent had significant ($P<0.001$) effect on the content of Cd, Cu, Pb, Mn and Zn of *A. esculentus.* It is likely to occur due to the presence of significant quantity of these metals in the sugar mill effluent and irrigated soil due to more irrigation frequency with increase in the irrigant concentrations. The content of Cd, Cu, Pb, Mn and Zn in *A. esculentus* was recorded maximum with 100% concentration of sugar mill effluent (Figures 4, 5, 6). The r value of the heavy metals, Cd ($r = +0.98$ to $+0.99$), Cu ($r = +0.97$ and $+0.98$), Pb ($r = +0.98$ to $+0.99$) and Mn ($r = +0.99$) and Zn ($r = +0.98$), in *A. esculentus* was shown to have positive correlation with different concentrations of sugar mill effluent in both the *Kharif* and *Zaid* seasons (Figures 4, 5, 6).

The contamination factor (Cf) indicated the contamination rate of heavy metals in *A. esculentus* after sugar mill effluent irrigation. The Cf of various heavy metals was in order of Cu>Mn>Zn>Cd>Pb in *A. esculentus* after irrigation with sugar mill effluent in both cultivated seasons (Figure 7). The maximum contamination factor was found for Cu (25.55 and 29.19) while the minimum was found for Pb (1.79 and 1.88) in *A. esculentus* with 100% concentration of sugar mill effluent in both cultivated seasons. Though, contents of heavy metals in *A. esculentus* were slightly higher in the effluent irrigated plants as compared to the control, these were under the permissible limits of FAO/WHO-Codex Alimentarius Commission (2011). Kumar and Chopra (2012) reported the maximum accumulation of heavy metals of Fe, Zn, Cd, Cu, Pb and Cr in soil and *T. foenum-graecum* with 100% concentration of distillery effluent. The maximum enrichment factor of Pb (24.00) and the minimum of Fe (4.21) in *T. foenum-graecum* were recorded with 100% concentration of distillery effluent irrigation and it was in the order of Pb>Cr>Cd>Cu>Zn>Fe after irrigation with distillery effluent.

## Conclusion

The present study concluded that the effluent of the

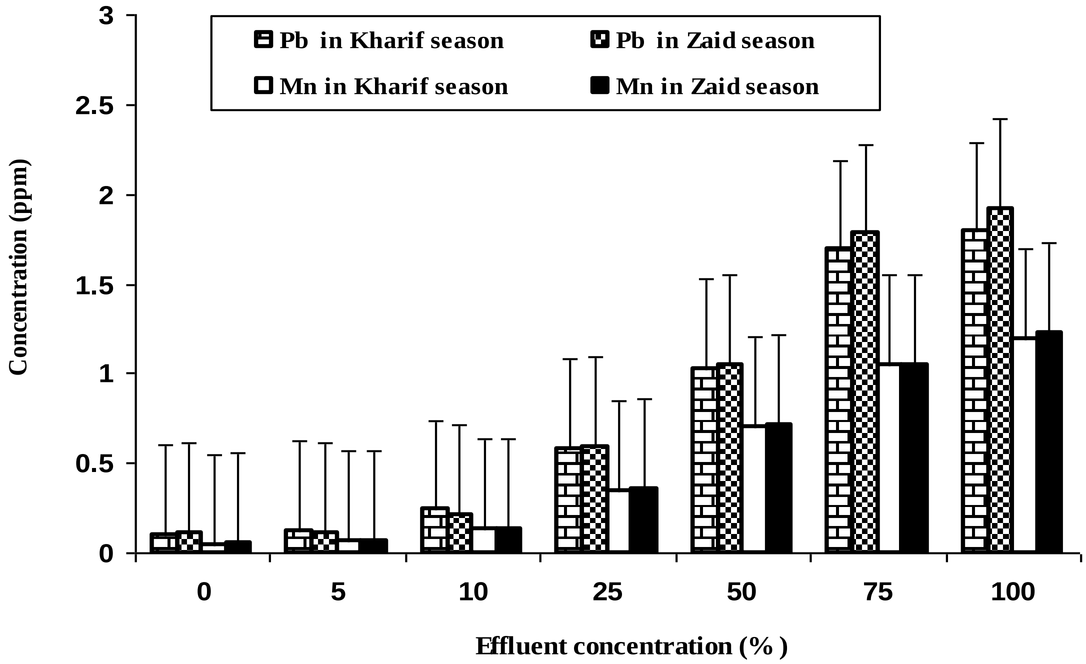


**Figure 5.** Pb and Mn concentration in *A. esculentus* after irrigation with sugar mill effluent.

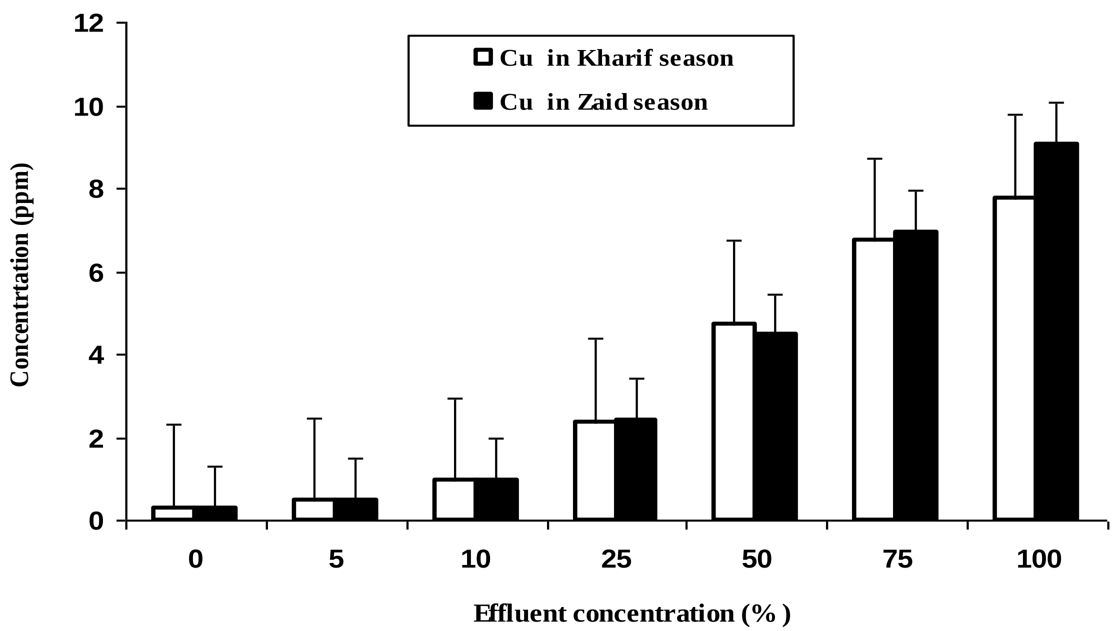


**Figure 6.** Cu concentration in *A. esculentus* after irrigation with sugar mill effluent.

Uttam Sugar Mill decreased the moisture content, WHC, bulk density and pH, and increased the EC, OC, $Na^{+}$, $K^{+}$, $Ca^{2+}$, $Mg^{2+}$, $Fe^{2+}$, TKN, $PO_4^{3-}$, $SO_4^{2-}$, Cd, Cu, Pb, Mn and Zn of the *A. esculentus* cultivated soil in both the *Kharif* and *Zaid* seasons. Thus, fertigation improved the soil nutrient status in both seasons. The agronomical performance of *A. esculentus* was recorded in gradually increasing order at lower effluent concentrations, that is, from 5% to 25% while in decreasing order at higher effluent concentrations from 50% to 100% as compared to the control. The maximum performance of *A. esculentus* was observed at 25% effluent concentration in

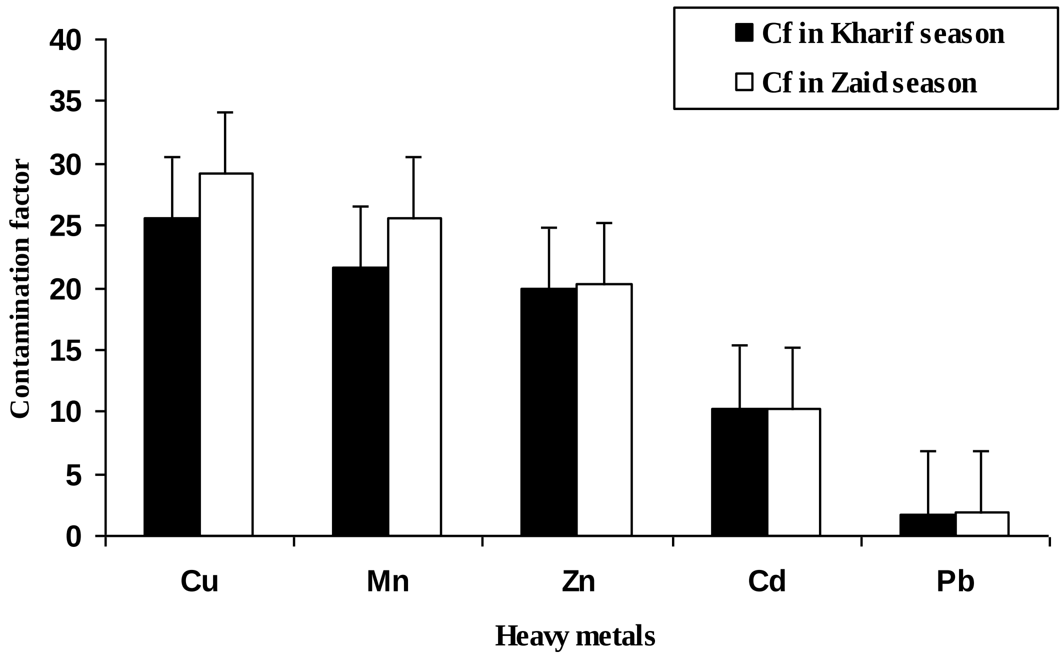


**Figure 7.** Contamination factor of heavy metals in *A. esculentus* after irrigation with sugar mill effluent.

both *Kharif* and *Zaid* seasons. It may be due to the supportable nutrients accumulation in the soil at this 25% effluent concentration that might have stimulated the growth performance. However, more irrigation increased the accumulation of nutrients at higher effluent concentrations, that is, 50%, 75% and 100%, thus it inhibited the overall performance of the *A. esculentus* crop plants. The contamination of heavy metals in the soil and *A. esculentus* was increased with the increase in effluent concentrations. Among heavy metals, the maximum contamination factor was found in Mn (14.65 and 14.82) for soil and Cu (25.55 and 29.19) for *A. esculentus* with 100% concentration of sugar mill effluent in both cultivated seasons. The contamination factor (Cf) indicated the order of contamination of various heavy metals in the soil and *A. esculentus* after sugar mill effluent irrigation. The Cf of various heavy metals was noted in the order of Mn>Cd>Cu>Zn>Pb for soil and Cu>Mn>Zn>Cd>Pb for *A. esculentus* after irrigation with sugar mill effluent in both the *Kharif* and *Zaid* seasons. Therefore, sugar mill effluent can be used as biofertigant after its appropriate dilution (25%) for irrigation of *A. esculentus* to mitigate the irrigation water scarcity and manage the problem of effluent disposal in sugar industries.

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