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EFFECT OF WATER STRESS AND CONTAMINATED WATER ON SEED GERMINATION TRAITS AND EARLY GROWTH IN MAIZE (*Zea mays*)

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Abstract

Irrigation is an increasingly important practice for sustainable agriculture. Water reuse as refined or non-refined wastewater can help agroecosystem productivity. Contaminated water may influence seed germination. Two experiments as laboratory and pot were carried out to assess impact of different levels of laundry detergent and water stress on seed germination and early growth in maize. At the laboratory experiment, seeds were treated with solutions of 0, -0.01, -0.09, -0.6 MPa polyethylene glycol 6000 and iso-osmotic concentration of laundry detergent. The pot experiment included different doses of laundry detergent (20, 2, 0.2 and 0 grams per litre) and two irrigation intervals (1 and 2-day irrigation interval). Results showed that -0.6 MPa osmotic potential reduced maize seed germination percentage, caulicle length, radicle length, seedling weight and seed vigor. 20 and 2 grams per litre of laundry detergent reduced plant height, leaf number per plant, leaf area, stem weight and leaf weight. 20 grams per litre of detergent increased specific leaf weight and reduced total biomass. The results show that irrigating maize by contaminated water with laundry detergent at high dose is not recommended.

Keywords: biomass, growth parameters, laundry detergent, maize, seed vigor

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1. Introduction

As population grows, demand for food products increases. It is needed to increase plant yield to supply food for people. Drought reduces agricultural products in the world. Due to water reduction in many countries, farmers tend to use wastewater. Reuse of water helps agroecosystems. This is reduction in pollution of ground water due to wastewater and increase in arable land (Anderson, 2003). Sodium, chlorine and nutrients are ingredients of refined water. These ingredients can be absorbed by plants through plant irrigation (EPA, 1992). Wastewater constituents such as detergent and pesticide can affect seed germination. Shakir et al. (2016) reported that pesticides are harmful for tomato seed germination. High levels of zinc, lead and cadmium did not reduce survival of *Coronilla*

varia (Bae et al., 2016). Oil crops irrigated with treated domestic wastewater produced higher oil than fresh water. This kind of oil has industrial uses (Hussein et al., 2004). Irrigation by wastewater produced lower growth and grain yield than irrigation by fresh water in canola (Ahmad et al., 2011). Sinha and Paul (2013) reported that *Cicer arietinum* had the highest seed vigor index at 20% sewage concentration over control and seed germination percentage was reduced from concentration of 30% onwards. Sarwar et al. (2011) reported that among different industrial wastewater treatments, the wastewater of textile and dyeing industries resulted in the lowest seed germination in red amaranth (*Amaranthus tricolor* L). Millioli et al. (2009) reported that the addition of rhamnolipid biosurfactant in a petroleum-bearing soil had a negative impact on microbial activity and lettuce

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(*Lactuca sativa*) seed germination. Triticale seed germination percentage and growth were reduced as osmotic potentials decreased. Seed treated by polyethyleneglycol had lower germination characteristics compared to seed treated by sodium chloride (Kaydan and Yagmur, 2008). Seed germination, emergence, seedling weight and root length in bitter vetch (*Vicia ervilia* L.) were significantly reduced by NaCl (Cocu and Uzun, 2011). In *Chenopodium glaucum* L., salinity caused by $MgCl_2$, Na_2SO_4 , Na_2CO_3 , NaCl and $MgSO_4$ and water stress caused by PEG reduced seed germination. NaCl had more adverse effect on seed germination percentage than that of PEG at osmotic potential less than -0.5 MPa under iso-osmotic solutions (Duan et al., 2004). Chlorination in water is a way to reduce disease. When this wastewater is used for irrigating crop plants, crop performance can be affected (Akanke et al., 2010).

One of the components of sewage is heavy metal. The lowest seed germination was reported in copper concentration of 20 to 200 mg per litre for both fescue and ryegrass (Taghizadeh and Solgi, 2017). Heavy metals had toxic effect on seed germination of *Lepidium sativum*. This toxicity was depended on metal type and concentration (Pavel et al., 2013). Zinc and copper had higher adverse impact on plant growth than germination in *Arabidopsis thaliana* (Burada et al., 2017; Li et al., 2005).

Maize plant irrigated by 20 g/L of cloth detergent had the lowest biomass (Heidari, 2012). Janmohammadi et al. (2008) reported that water stress reduces radical and caulicle length in maize (*Zea mays*). Afzali et al. (2006) found that *Matricaria chamomilla* biomass and seed germination were reduced as water stress level increased. Millet leaf area was reduced by drought (Heidari et al., 2011). Maize is a C_4 plant and widely cultivated in most part of Iran. Many researchers studied effect of wastewater on plant growth but little information is available about effect of laundry detergent on seed germination. Detergent may increase plant growth due to some nutrient elements and may inhibit plant growth due to toxicity of salts and increasing water osmotic potential. In the research, impacts of water deficit stress and laundry detergent on growth and germination of maize were assessed.

2. Material and methods

2.1. Laboratory experiment

At the laboratory experiment, seeds were treated with solutions of 0, -0.01, -0.09, -0.6 MPa polyethylene glycol 6000 (Michel and Kaufmann, 1973) and iso-osmotic concentration of laundry detergent (0, 0.2, 2, 20 g/L, respectively). Detergent powder was dissolved in distilled water. Control was distilled water. Detergent osmotic potential was measured using osmometer. Chemical ingredients of

used cloth washing powder were reported by Berger (2002). The layout of laboratory experiment was completely randomized design with three replicates. The experiment was conducted at Razi University, Iran in 2012. Seeds of maize (*Zea mays*, Cv. single cross 704) were gathered from one year maternal plants under non-stress condition to have high germination.

Firstly, maize seeds were sterilized. Then 20 seeds were allocated to each Petri dish. Each Petri dish received 8 mL of prepared solution. Petri dishes were kept at $26 \pm 1^\circ C$. After a week, seed germination traits were counted. Seedlings were weighed after drying by oven. Abdul-Baki and Anderson (1973) formula was applied to compute seed vigor.

2.2. Pot experiment

The layout of pot experiment was factorial arranged in RCBD. Pot experiment had three replicates and was conducted at Razi University in 2012. One factor was laundry detergent levels and the other factor was irrigation interval. Laundry detergent concentrations included 20, 2, 0.2 and 0 grams per litre. Detergent was dissolved in tap water. Irrigation interval included 1 and 2-day.

Five maize plants Cv. single cross 704 were grown per pot. Firstly plants irrigated daily. Irrigation treatments were imposed eight days after sowing. To wet soil well, pot soil was irrigated enough as excess water was drained. Plants nitrogen requirement was supplied by urea fertilizer.

23 days after sowing, all plants per pot were cut and used for sampling. Dry matter parameters (stem and leaf biomass) were measured after drying plant samples in oven. After measuring leaf length and width by ruler, leaf area (LA) was computed. Specific leaf weight (SLW) was calculated by dividing leaf dry weight by LA.

Data were tested for normality by Minitab software. Then normal data were analyzed. Mean comparisons were done by test of Duncan ($P < 0.05$) and the SAS software. The SPSS software was used for calculating correlation coefficients.

3. Results and discussion

3.1. Laboratory experiment

3.1.1. Results

Seed germination percentage was reduced under osmotic potential of -0.09 MPa laundry detergent, but seed germination percentage was not reduced under osmotic potential of -0.09 MPa polyethylene glycol. Germination percentage was higher under osmotic potential of -0.6 MPa polyethylene glycol compared to laundry detergent (Fig. 1). Correlation between all traits and seed germination percentage was positive and significant (Table 1). Caulicle length was reduced as osmotic potential increased. Caulicle length was reduced

under osmotic potential of -0.01 MPa detergent, but no significant reduction was observed under osmotic potential of -0.01 MPa polyethylene glycol (Fig. 1). Correlation between caulicle length and all traits was positive and significant (Table 1).

Radical length was severely reduced under osmotic potential of -0.6 MPa compared to control (Fig. 1). Polyethylene glycol had higher radical length than laundry detergent under osmotic potential of -0.09 MPa. Correlation between radicle length and all traits was positive and significant (Table 1). The lowest seedling weight was observed under osmotic potential of -0.6 MPa (Fig. 1). Osmotic potential of -0.6 MPa detergent produced lower seedling weight than osmotic potential of -0.6 MPa polyethylene glycol. Correlation between seedling weight and all traits was positive and significant (Table 1). The lowest seed vigor was observed under osmotic potential of -0.6 MPa (Fig. 1). Osmotic potential of -0.09 MPa detergent had lower seed vigor than

osmotic potential of -0.09 MPa polyethylene glycol. Correlation between seed vigor and all traits was positive and significant (Table 1).

3.1.2. Discussion

A -0.6 MPa osmotic potential reduced all maize seed germination traits. Some parts of these results are in agreement with results of other research (Shakir et al., 2016). Seed germination percentage and seed vigor in sunflower were reduced under high concentration of laundry detergent (Heidari, 2013) that is in agreement with our finding. At the first stage of seed germination, seed absorbs water and seed imbibition occurs. By increasing water potential, seed germination decreases.

At the second stage of seed germination, seed needs oxygen for respiration. If oxygen reduces fewer than 20%, seed germination reduces (Gardner et al., 2010). Both detergent and PEG can reduce water oxygen diffusion and capacity.

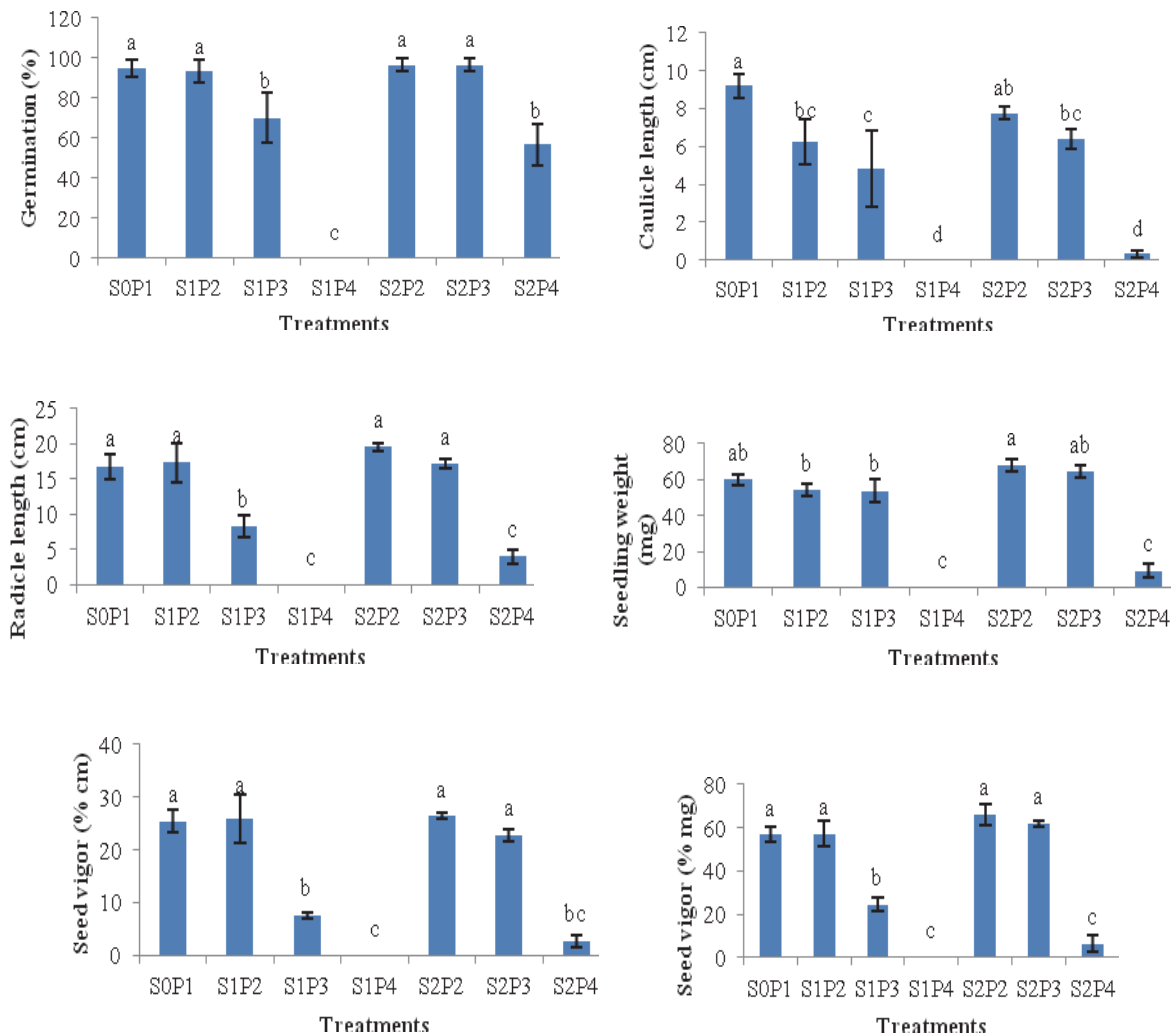


Fig. 1. Effect of different osmotic potentials of laundry detergent and polyethylene glycol on maize seed traits. S0, S1 and S2 are distilled water, laundry detergent and polyethylene glycol, respectively. P1, P2, P3 and P4 are osmotic potential of 0, -0.01, -0.09, -0.6 MPa. Within each seed germination trait, means with the same letter have no significant difference as determined by test of Duncan ($P < 0.05$). Values are mean (three replicates) $\pm$ standard deviation

Table 1. Coefficients of correlation among maize seed traits as affected by different osmotic potentials of laundry detergent and polyethylene glycol

	<i>Germination percent</i>	<i>Caulicle length</i>	<i>Radicle length</i>	<i>Seedling weight</i>	<i>Seed vigor (based on weight)</i>	<i>Seed vigor (based on length)</i>
Germination percent	1	0.855**	0.926**	0.912**	0.902**	0.880**
Caulicle length	0.855**	1	0.878**	0.921**	0.911**	0.911**
Radicle length	0.926**	0.878**	1	0.920**	0.985**	0.974**
Seedling weight	0.912**	0.921**	0.920**	1	0.927**	0.872**
Seed vigor (based on weight)	0.902**	0.911**	0.985**	0.927**	1	0.984**
Seed vigor (based on length)	0.880**	0.911**	0.974**	0.872**	0.984**	1

*, **: significant at the 0.05 and 0.01 level, respectively

Low osmotic potential reduces water absorption by seed. Dehydration inhibits cell division. Plants reply to drought by production of reactive oxygen species known as ROS. One of reasons for cellular destruction is ROS (Kar, 2011).

It was observed that laundry detergent had more adverse effect on seed germination traits, especially under lower osmotic potential, than iso-osmotic potential of polyethylene glycol. It is probably due to the uptake of ions such as Na⁺ by the seed, destruction of seed coat and ion toxicity. Janmohammadi et al. (2008) reported that seed treatment in maize by NaCl produced higher seed germination than that of PEG. It is probably due to that Na⁺ and Cl⁻ ions were uptaken by the seed. So, water uptake by seed was retained during seed germination process. Heavy metals immobilize the seed starch and restrict nutrient sources. Heavy metals also reduce proteolytic enzyme activity (Seneviratne et al., 2017).

At the first stage of germination, seed starts to absorb ions including heavy metals. These metals can be accumulated into the root or exported to the shoot by transpiration. Reduction in seed germination characteristics as a result of high level of detergent may be due to heavy metal (Srivastava et al., 2017). Osmotic potential had more adverse effect on caulicle length than radicle length. Using root to shoot ratio as an index for drought was declared, however interpretation of this index is not easy (Silva et al., 2012). Root length was used as an indicator of tolerance to heavy metal because this organ is in direct contact with metals compared to other organ. Low dose of lead had no impact on seed viability in mung beans, but the high dose of the metal delayed germination. Many mechanisms like chelating have a potential to reduce toxicity of heavy metals (Coscione et al., 2009). When sewage water containing heavy metals was used for irrigating plants, there is high chance for the metals to be absorbed by plant root and the chance of chelating is low compared to soil polluted by heavy metals that their metals may be bound. Plants can absorb and accumulate heavy metals in their tissues. Then heavy metals enter into food chains and contaminate them. Ultimately the human health is threatened. Positive correlation among seed germination traits indicates

that all seed germination traits can be affected by drought and detergent, simultaneously and reduction in seed vigor can be related to reduction in seed germination, weight and length of seedling.

3.2. Pot experiment

3.2.1. Results

20 and 2 grams per litre of detergent severely reduced plant height (Fig. 2). Under each contaminated concentration, difference between irrigation intervals was not significant. Leaf number per plant was severely reduced by 20 grams per litre of detergent (Fig. 2). Correlation of plant height with many traits was positively significant (Table 2). Correlation of plant height and leaf number with specific leaf weight was negatively significant (Table 2). As contamination increased, leaf area was reduced (Fig. 2). Under detergent dose of 0.2 g/L, irrigation interval of 2-day produced higher leaf area than irrigation interval of 1-day. Leaf weight was reduced as contaminated water dose increased (Fig. 2). Under each pollution level, irrigation interval difference was not significant in terms of leaf weight. Correlation of leaf area and leaf weight with most traits was positively significant (Table 2). Correlation between leaf area and SLW was negatively significant (Table 2).

Stem weight was severely reduced by high doses of detergent (Fig. 2). Total dry matter production was reduced by only detergent dose of 20 grams per litre (Fig. 2). Under each contamination concentration, difference between irrigation intervals was not significant in terms of total biomass and stem weight. Correlation coefficient of stem weight and total biomass with most traits was positively significant (Table 2).

Under detergent concentration of 2 grams per litre, plants irrigated by interval of 1-day produced higher leaf to stem ratio compared to plants watered by tap water (Fig. 2). Specific leaf weight was severely increased by 20 grams per litre of detergent (Fig. 2). Under 20 grams per litre of detergent, irrigation interval of 1-day produced higher SLW compared to irrigation interval of 2-day. Correlation between SLW with leaf area, leaf number per plant and plant height was negatively significant (Table 2).

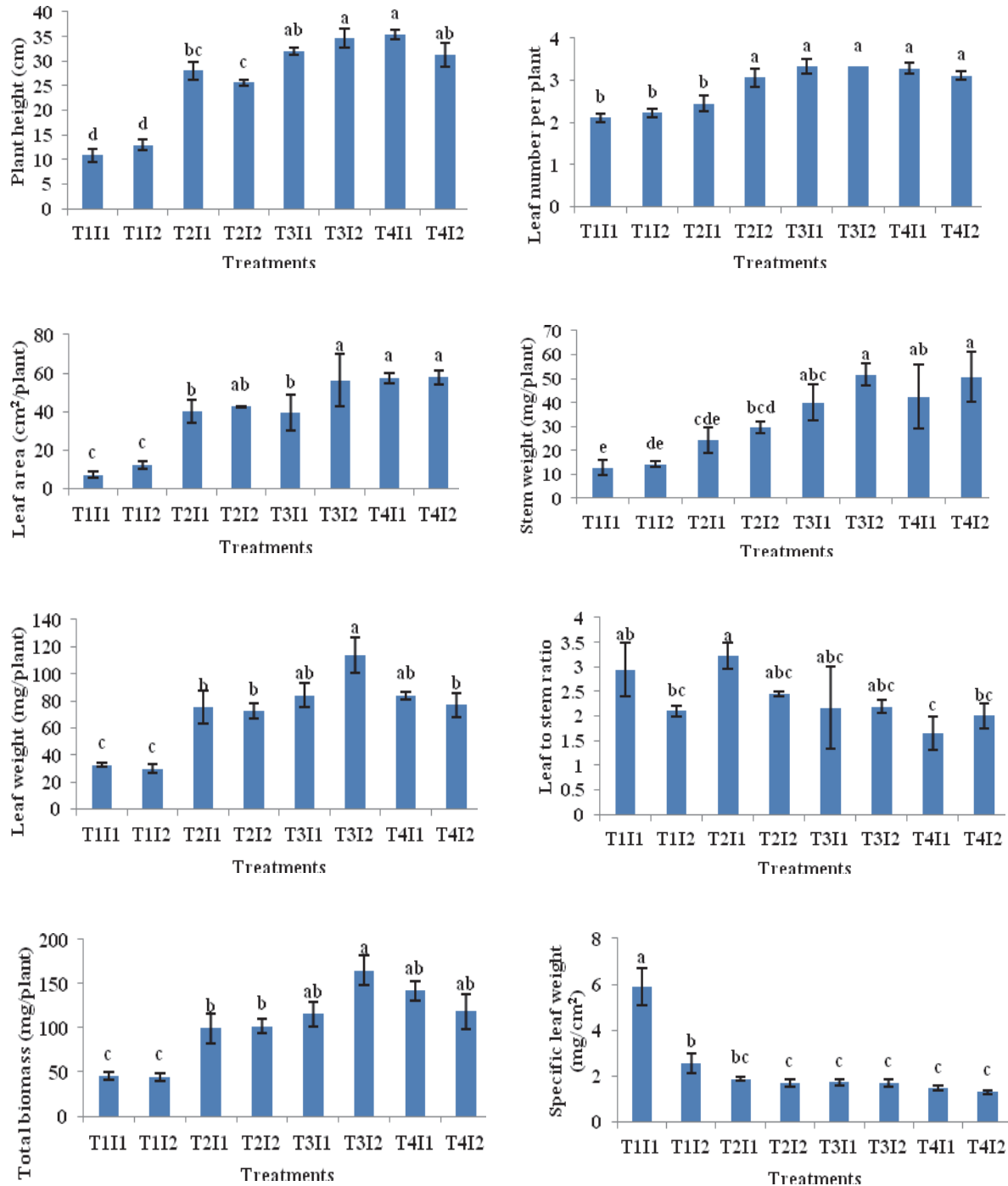


Fig. 2. Maize vegetative growth characteristics as affected by detergent and water stress.

T1 to T4 indicate laundry detergent levels of 20, 2, 0.2 and 0 grams per liter, respectively. I1 and I2 indicate irrigation interval of 1 and 2 days. Within each plant growth characteristic, means with the same letter have no significant difference as determined by test of Duncan ($P < 0.05$). Values are mean (three replicates) $\pm$ standard deviation

3.2.2. Discussion

High concentration of laundry detergent (20 and 2 grams per litre) reduced leaf weight, stem weight, leaf area, leaf number per plant and plant height. Laundry detergent concentration of 20 grams per litre reduced total biomass and increased specific leaf weight. Environmental stresses can reduce plant height. Under drought condition, ROS was generated.

This process is mediated by ABA, ethylene and other hormones. ROS can damage cells (Kar, 2011). Accumulation of abscisic acid leads to plant height reduction. Plants reduce their shoot intelligently to reduce transpiration (Mishra and Pradhan, 1972). Increasing SLW under high concentration of detergent may be as a result of increasing solute concentration and decreasing cell size (Heidari et al., 2011).

Table 2. Coefficients of correlation among maize plant growth traits as affected by different concentration of laundry detergent and water stress

	<i>Plant height</i>	<i>Leaf number per plant</i>	<i>Leaf area</i>	<i>Stem weight</i>	<i>Leaf weight</i>	<i>Leaf to stem ratio</i>	<i>Total biomass</i>	<i>Specific leaf weight</i>
Plant height	1	0.904**	0.957**	0.903**	0.937**	-0.434	0.956**	-0.801*
Leaf number per plant	0.904**	1	0.870**	0.912**	0.867**	-0.626	0.899**	-0.728*
Leaf area	0.957**	0.870**	1	0.922**	0.892**	-0.465	0.936**	-0.809*
Stem weight	0.903**	0.912**	0.922**	1	0.880**	-0.582	0.921**	-0.696
Leaf weight	0.937**	0.867**	0.892**	0.880**	1	-0.289	0.977**	-0.688
Leaf to stem ratio	-0.434	-0.626	-0.465	-0.582	-0.289	1	-0.440	0.507
Total biomass	0.956**	0.899**	0.936**	0.921**	0.977**	-0.440	1	-0.699
Specific leaf weight	-0.801*	-0.728*	-0.809*	-0.696	-0.688	0.507	-0.699	1

*, **: significant at the 0.05 and 0.01 level, respectively

Heidari (2012) declared similar findings. In sunflower, high dose of cloth washing detergent reduced plant biomass and growth parameters (Heidari, 2013) that are in agreement with our finding. Amirmoradi et al. (2012) showed that high dose of lead and cadmium reduced leaf area and dry matter production in peppermint. Some elements, in contaminated water like sodium, act as soil disturbing agent. This element also has adverse impact on plant growth.

Salinity effect on plant growth should be considered at cellular level. Ions can pass into the cell plasma membrane. Plant responds to salinity at two stages. At the first stage, plant reduces its leaf area. It is because of accumulated solutes in plant root. At the second phase, plant succumbs and leaf production rate reduces so plant gradually dies (Volkmar et al., 1998).

Contaminated water with laundry detergent had high electrical conductivity, high sodium concentration and high sodium adsorption ratio, so prolonged application of the contaminated water for irrigation may result in many physical and chemical changes in soil properties such as pH, availability of nutrient etc. Some heavy metals such as zinc (II), nickel (II), copper (II) and chromium (VI) are micronutrient and promote plant growth, (Cocarta et al., 2017). Some plant species are used in phytoremediation process. Phytoremediation process is a process in which plants were applied to immobilize, remove or detoxify contaminants in environment. For example *Eruca* had high tolerance to high levels of nickel (Zhi et al., 2015). In the study, maize stand out at low to moderate concentration of laundry detergent, so it can be recommended for possible phytoremediation.

Clover leaf number was not changed by moderate drought, but it was reduced by extreme drought (Belaygue et al., 1996). Shoot reduction due to stress can increase plant tolerance to drought. Salinity and drought cause similar effect on plant growth. Overall, laundry detergent can reduce plant growth parameters by multiple stresses. Positive correlation among most plant traits shows that water stress and contaminated water affect plant traits simultaneously and reduction in total biomass relates

to reduction of plant growth parameters. Negative correlation between SLW with leaf area, shows that under water and contamination stress plant decrease its leaf area and increase its density to reduce transpiration rate and absorbed radiation from sunlight (Heidari et al., 2012).

4. Conclusions

An osmotic potential of -0.6 MPa reduced most seed germination traits in maize. Under iso-osmotic potential, more adverse effect of laundry detergent on seed germination traits was observed compared to polyethylene glycol. Heavy metals and salts existing in detergent can damage seed and disturb seed germination processes. Detergent concentration of 20 and 2 grams per litre reduced most maize traits at early plant growth stage.

Multiple stresses are reasons for this reduction. It is concluded that maize farm should not be irrigated by sewage water containing high dose of laundry detergent at germination and seedling stages. Laundry detergent effect on weed seed germination can be studied at the next researches. Positive impact of detergent on weed germination is possible, because detergent can remove some germination inhibitors in seed coat or it may affect their dormancy.

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