Concentration of Heavy Metals in *Teff* Straw, Water and Milk in Some Selected Areas of Central Ethiopia

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Abstract: The concentration of cadmium (Cd), lead (Pb), arsenic (As) and chromium (Cr) in *teff* straw, water and milk were studied in East and West Shoa, Ethiopia. The concentration of heavy metals in feed, water and milk were investigated using Graphite Furnace Atomic Absorption Spectrophotometer. Heavy metals in *teff* straw in West and East Shoa were 1543.54 ± 318.70 µg/kg and 1486.92 ± 279.73 µg/kg, respectively and were found in the order of Cr > As > Pb > Cd. The concentration of heavy metals in water in East Shoa (28.08 ± 7.02 µg/L) was significantly higher ($p<0.05$) than in West Shoa (1.96 ± 0.28 µg/L) and were in the order of Cr > As > Pb > Cd. Water samples obtained from Mojo Tannery had the highest level of heavy metals (86.89 µg/L) followed by Mojo Lake (67.89 µg/L) and a river near Mojo town (43.64 µg/L). Except for pH of water from Mojo Lake (10.4) and Gelan dye factory (8.9), the rest of water samples from East Shoa were found within the recommended pH limit of 6.5-8.5 for livestock drinking. Water samples from West Shoa had an average pH of 7.0 ranging 6.4–7.7 and were within the permitted limit. Water from Mojo had the highest concentration of Cr and the contamination level of heavy metals in water from East Shoa was in the order Mojo > Awash River > Gelan > Akaki > Bishoftu. The concentration of heavy metals in water from East Shoa was more than ten times higher than West Shoa. The concentration of heavy metals in livestock water in the study areas did not pass the standard limit. The overall concentrations of heavy metals in milk were in the order Cr > Cd > Pb > As. Compared to WHO/JECFA 1989 standards, the concentrations of Cd and As in milk were within the permitted limits, whereas the concentrations of Cr and Pb were found beyond the standard limit. It is concluded that the level of heavy metals near industrial areas were higher than those sites in non-industrial areas. It is recommended that further study is required on heavy metals content of soils, fodder feeds and livestock products in these study areas.

Keywords: *Arsenic, Cadmium, Chromium, East Shoa, Lead, Milk, Teff straw, West Shoa*

Introduction

Environmental pollution is a major global problem that pose risks to human beings and animals. The development of modern technology and industrialization are among the major phenomena that bring environmental pollution (Ramchander *et al*., 2015). Heavy metals are one of the pollutants that enter the environment naturally from weathering of rocks (Kaplan *et al*., 2011) and through human activity such as discharge of industrial effluents, sewage sludge and application of agrochemicals (Baloach and Farid, 2012). Heavy metals are extremely persistent, non-biodegradable and non-thermodegradable and they remain accumulated in the environment (Akan *et al*., 2009). Cd, Pb, As and Cr are some of the most toxic heavy metals that are accumulated in soil and water. They are rated with varying toxicity degrees: Cd (highly toxic), Pb (toxic), As (moderately toxic) and Cr (slightly toxic) (FAO, 1999).

Heavy metals are becoming a challenge to agricultural production because they contaminate soil, water, grains and feeds. Water is a major reservoir of chemicals, as it is polluted with effluents from industries and dissolved gasses (Ramchander *et al.,* 2015), then channels the soil system and contaminates feeds and crops which consequently affect the quality of milk and milk products and finally enter to human beings through food chain (Kaplan *et al*., 2011; Kodric *et al*., 2011). Heavy metals have the capability of interfering with the normal process of milk synthesis and affect milk yields in dairy cattle (Khan *et al*., 2012). Feeds containing excess Cd, Pb and As appear in toxic levels and their toxicity interferes with copper and zinc absorption, resulting in decreased milk production (Khan *et al*., 2012). Heavy metals are also capable of displacing other essential minerals from where they do vital biochemical and enzymatic reactions in the metabolism process (Dawd, 2010). Heavy metals also affect the health status of livestock species (NRC, 2001). Consumption of Pb at higher concentration in acute cases resulted in death of cattle within 24 hours (Mukesh *et al*., 2008). Heavy metals have the capacity of

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damaging vital organs like the kidneys and liver in chronic situations (Bala *et al*., 2014) and can also lead to death in acute cases when consumed at higher concentrations (Rajaganapathy *et al*., 2011).

Scanty information (Dawd, 2010; Muluken, 2014; Tassew *et al*., 2014) was reported on the heavy metal content of water and milk and no information is available on the heavy metals content of livestock feeds. Moreover, these reports did not compare the pollution status of industrialized *vis,* non-industrialized places of the country. Hence, this study was designed to fill this gap by determining the concentration of toxic heavy metals specifically Cd, Pb, Cr and As in livestock feed (*teff* straw), water and cow milk in industrialized and non-industrialized areas of East and West Shoa Zones of Ethiopia respectively.

Materials and Methods

Study Areas

The study was carried out in West Shoa (Menagesha to Holetta) and East Showa (Akaki to Mojo) Zones of Ethiopia and the detailed description of the study areas are presented on the maps indicated in Figures 1 and 2. West Shoa, the direction to Holetta is located 45 km west of Addis Ababa at an altitude of 2400 m above sea level with an annual rainfall of 1066 mm and average minimum and maximum temperatures of 6 and 22°C, respectively. The soil type was reported to be predominantly Eutric Nitosols, pellic and chromic vertisols (Getachew *et al*., 2014). The characteristics of the soils vary from sandy to clay soils and the pH of the soils is mainly acidic, 5.24 (Gemechu, 2007). This location is a potential area for agricultural production where both crop and livestock farming has been undertaken. This place has been hosting the prominent research center (Holetta Agricultural Research Center), many improved agricultural technologies of crop varieties, pesticides and fertilizers have been applied for verification and demonstration purposes for the past 50 years. In addition, this location has been considered as a prominent place for growing and cultivation of rose (flower) since 2004.

East Shoa (the direction to Bishoftu) is located at 45 km Addis Ababa at an altitude of 1950 m above sea level with a mean minimum and maximum temperature of 10.6 and 25.0 ℃, respectively (Samuel *et al*., 2009). The soil type towards Akaki area is predominantly vertisol and fluvisol in which the fluvisol is more contaminated with polluted wastewater than the vertisol (Fisseha *et al*., 2003).

Akaki area is one of the industrialized areas in Ethiopia where it hosts over half of the industries in Addis Ababa. Large and medium industrial plants like foods, beverages, furniture, detergent, dye, leather, paper, metallic products and textiles industries are running in this area. The polluted Akaki River also flows through this area after passing Addis Ababa (Ellen *et al*., 2015). Gelan (the direction to Bishoftu) also accommodates different factories from which effluents are released and commonly meet in Gerbecha River (the river that drains Gelan area into the Awash River). Since neighboring farmers were experiencing water shortages for livestock drinking, they use this River as an alternative water source for livestock drinking.

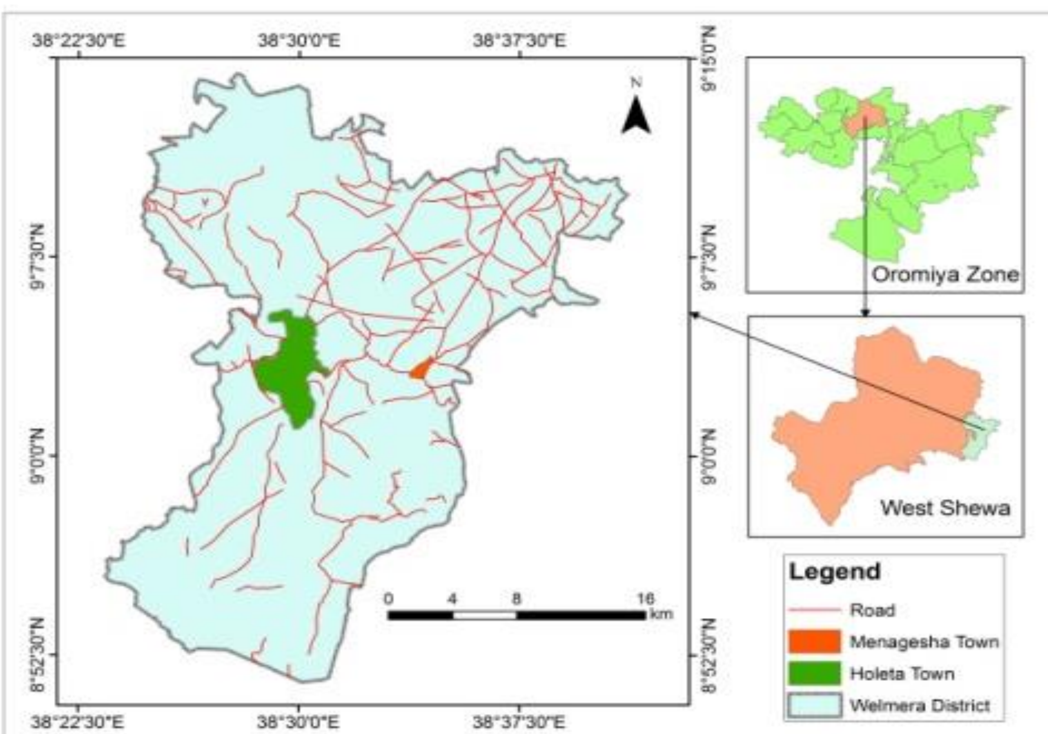

Figure 1. Map of West Shoa.

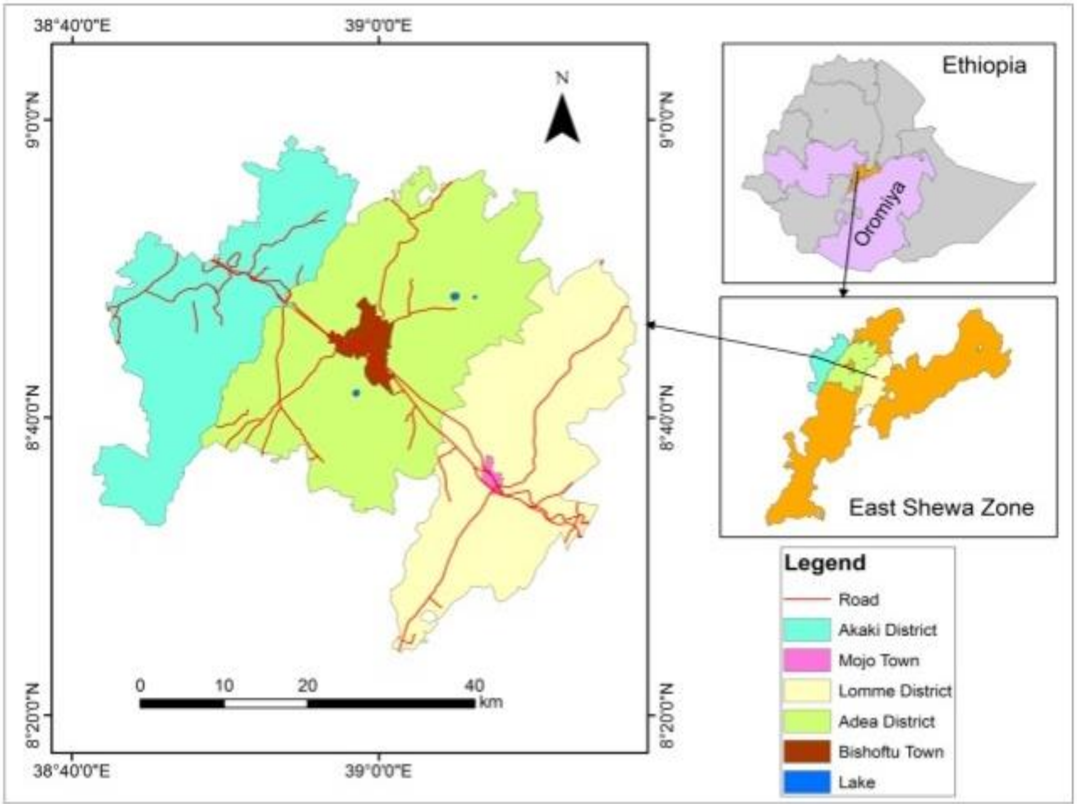

Figure 2. Map of East Shoa.

Data Collection and Preparation of Samples

Fifteen samples of *teff* straw were collected from the study sites of Holetta and Bishoftu. The samples were taken from different parts of the hips and maintained in labeled plastic bags. The expensive nature of laboratory analysis of heavy metals in this study forced us to use the minimum number of samples (15 per study location). Before grinding, samples of *teff* straw were chopped into smaller sizes using a pair of scissors, and ground on a feed miller; first with a 2 mm, and then with a 1 mm size mesh. The milled samples were preserved in labeled paper bags and sent for investigation of heavy metals to the laboratory of the Environmental Protection Authority in Addis Ababa, Ethiopia.

Fifteen representative samples of water were collected at a village level (field) from West Shoa (Menagesha to Holetta) and East Shoa (Akaki to Mojo). Criteria for the selection of sampling sites for water collection were of two: the first is water sources that livestock species are commonly used for drinking and the second is the closeness of water sources to different manufacturing industries. A capacity of 0.5 L labeled plastic containers were used for water collection from surface waters mainly from streams, surface runoff and rivers. To avoid contamination with unnecessary elements, the containers were properly washed with clean water and rinsed with de-ionized water and finally with the actual water sample. Water samples from the water bodies were taken using plastic cups and then poured into the labeled plastic water containers. The containers were filled to the top, capped tightly, and were delivered to Holetta Agricultural Research Center for pH measurement using pH/ion-meter, WTW, Inolab (Germany).

Concerning milk samples, fifteen fresh whole milk samples were collected from farmers in West Shoa (Welmera) and East Shoa having local milking cows. The feeding management of the cows was based on grazing on the field (outdoors). The roughage feed provided to these cows in both study locations was *teff* straw. Cattle drink water from available water bodies in their respective vicinities.

Labeled plastic sample bottles were carefully washed with clean water, soaked in 20% HNO_3 for 24 hours, and rinsed with de-ionized water to avoid contaminations. A hundred mL of fresh milk samples were collected from the morning milking of each farm and placed in an icebox and was transported to the laboratory of Holetta Agricultural Research Center and preserved at -20 °C in a deep freezer. The samples were then sent to the laboratory of the Environmental Protection Authority at Addis Ababa for laboratory analysis of heavy metals.

Laboratory Analysis of Feed, Water and Milk Samples

Mineralization of feed samples: Mineralization of feed samples was carried out following dry ashing. One gram of ground feed samples was transferred into a porcelain crucible and placed in a muffle furnace at 550^{0}c for 2 hours until complete mineralization took place. The crucibles were removed from the muffle furnace using tongs and placed directly on a hot plate (AOAC, 1990).

Digestion process of feed samples: Following ashing, the residue of each sample was subjected to the wet digestion process. Using a glass rod, the ash residue was transferred into an Erlenmeyer flask. Then the glass rod and crucibles were rinsed with 1% HNO_3. Five mL of 6 M HNO_3 was added to the Erlenmeyer flask in a hot plate and digested by gentle boiling until it remained to the level of 1 mL. Then 5 mL of 3 M HNO_3 was added to the extract and reheated for 30 minutes. The warm solution was filtered into a 100 mL volumetric flask using a Whatman 42 filter paper. The filtrate was allowed to cool and diluted to 100 mL with de-ionized water and the flask was sealed with a stopper. Blanks consisting of the reagents and de-ionized water were subjected to similar sample preparation and analytical procedures and the extracts were subjected for measurement of heavy metals (AOAC, 1990).

Mineralization and digestion processes were not undertaken on water samples; rather, they were directly subjected to the assay process. Since mineralization is used to burn the organic matter obtain the non-organic matter (metals), water is not expected to contain organic matter. Likewise, the digestion process aims to facilitate dilution. water by itself is diluent and water can easily pass the nebulizer without any blocking and no need for doing digestion process in water samples.

Regarding milk analysis, wet digestion, using nitric acid (HNO_3), perchloric acid ($HCLO_4$), hydrogen peroxide (H_2O_2) and de-ionized water was undertaken by placing in a hot plate. A mixture of 9 mL of 65% HNO_3 and 1 mL of 72% $HCLO_4$ was added into a beaker containing 3 ml milk samples. Then 3 mL of 30% H_2O_2 was added, mixed, and placed in an electric hot plate at 90 ℃ for digestion until a clear solution was left. To avoid blocking of the nebulizer, the samples were filtered through Whatman 42 filter paper. Then the samples were transferred to 10 mL volumetric flasks and filled up to the mark with de-ionized water. Then 10 mL of milk samples was injected and nebulized into the tube of GFAAS and the concentration of each metal was read from the computer that was interfaced with the GFAAS. Blanks consisting of the reagents and de-ionized water were subjected to similar sample preparation and analytical procedure and the metals were determined by the same instrument (GFAAS).

Preparation of standard solution and calibration curve: Before determination of the concentration of each metal, the GFAAS instrument was calibrated with a standard solution according to the procedures indicated in (AOAC, 1990).

Study Design and Statistical Analysis

The study was undertaken based on Completely Randomized Design (CRD) and the statistical model was Yi= μ + Li + ei Where,

Yi = level of heavy metals
μ = the overall mean
Li = effect of location
ei = the error term

The data was subjected to analysis of variance using Statistical Analysis System (SAS, 2002); GLM and t-test were used to analyze the data. The comparison was made in two ways: one is between two different locations of industrialized and non-industrialized areas. The other comparison was made among sampling sites within a location.

Results and Discussion

pH Values of Livestock Water in East and West Shoa of Different Sampling Sites

The pH of livestock water collected from different sampling sites of East and West Shoa is indicated in Table 1. Except for pH of water from Mojo Lake (10.4) and Gelan dye factory (8.9), the rest of samples taken from East Shoa were within the recommended pH limit of 6.5-8.5 for livestock drinking. Water samples from Holetta had an average pH of 7.0 ranging 6.4–7.7 and were within the permitted limit. The highest pH measurement (9.55) obtained in the Mojo area (10.4) is comparable with the findings of Dejene (2011). The observed high pH could be related to the use of lime as raw materials for the removal of hair from hides and skins in the production process.

pH value in water is a measure of quality (Beed, 2006) and has implications on the health and productivity of livestock species. Highly acidic pH in livestock water, especially, pH lower than 5.5 leads to acidosis and reduced feed intake, while highly alkaline water (pH greater than 9.0) causes chronic or mild alkalosis (Adams and Sharpe, 1995), digestive upsets, diarrhea, reduced water intake, lower feed intake and lower feed conversion efficiency (Jane, 2009).

Concentration of Heavy Metals in Livestock Water in East Shoa of Different Sampling Sites

Among the sampling sites (Table 2), water samples from the effluent of Mojo tannery had the highest concentrations of Pb (24.66 μg/L) and chromium (214.05 μg/L). The highest level of chromium in water samples collected from Mojo area is due to the utilization of chrome for the tanning process in tanneries and textile industries (Solomon *et al.,* 2015). Among the heavy metals, the lowest mean value (0.80 μg/l) was recorded for cadmium (Table 2).

The lead contents of water samples significantly varied from a negligible quantity of 0.0001 μg/L in Bishoftu to a relatively highest value of 24.66 μg/L in Mojo Lake. The arsenic content of water also varied from the lowest level of 4.45 μg/L in the Glean area to the relatively higher level of 19.04 μg/L in Awash. The data showed that the mean concentration of all heavy metals in water from Mojo (66.14 ± 7.58) significantly ($p<0.05$) differed from Awash (29.92 ± 13.14), Gelan (23.94 ± 5.36), Akaki (20.59 ± 6.57) and Bishoftu areas (16.87 ± 13.14).

Except for chromium, the concentrations of other heavy metals in water samples collected from East Shoa were lower than the levels reported from Hyderabad, India where the values of lead, chromium, and cadmium were 37.5 μg/L, 62.25 μg/L and 3.75 μg/L, respectively (Ramchander *et al.,* 2015). The mean concentration of chromium (92.77 ± 20.33) in livestock water in East Shoa significantly ($P<0.05$) differed from arsenic (9.61 ± 1.50), lead (9.14 ± 3.54) and cadmium (0.80 ± 0.18) and it was in the order of Cr > As > Pb > Cd which was in similar order with heavy metal levels of industrial wastewater released from Hawassa textile factory (Solomon *et al.,* 2015).

According to this study, majority of the water samples were highly contaminated with heavy metals due to heavy pollution from urban and industrial activities around the sampling areas. Overall, the contamination levels with heavy metals in livestock water in the sampling sites of East Shoa, Ethiopia, were in the order Mojo > Awash > Gelan > Akaki > Ada (Bishoftu). Studies on the heavy metal content of livestock water in East Shoa were very scarce, and among the scanty studies, Prabu (2009) showed that samples of wastewater from the polluted Akaki River had higher chromium and cadmium levels as compared to the level of heavy metals found in unpolluted water. Studies on heavy metals in soils and on vegetables in these areas particularly around the polluted area of Akaki River (Fisseha *et al*., 2003; Prabu, 2009) and Zeway (Amare, 2007) showed that soils and vegetables might have been irrigated with industrial wastewater contaminated with heavy metals.

Table 1. pH values of livestock water in East and West Shoa of different sampling sites

East Shoa (Akaki to Mojo)	**pH**	**West Shoa (Menagesha to Holetta)**	**pH**
Gerbecha River (Gelan)	7.7	Welmera (Sademo karsa)	7.1
Gelan surface water	7.5	Welmera (Burka/Finland School)	7.2
Gelan dye & ceramic factory	7.5	Welmera (Met)	6.4
Mojo factory of tannery	7.1	Welmera (Qorejilla 2)	7.2
Gelan steel factory	7.4	Welmera (Qorejilla 1)	7.3
Gelan garment factory	7.0	Holetta River	7.7
Gelan dye factory	8.9	Welmera (Kui River)	6.9
Lome River	7.6	Welmera (Ureni River)	7.2
Akaki Dellolo River	6.5	Welmera (Kateba)	7.0
Debrezeit surface water	6.6	Welmera (Sademo/Medrok flower farm gate)	7.0
Surface water near UNISA University	7.5	Welmera (Ama flower farm gate)	6.8
Akaki *Kebele* 06 surface water	7.2	Welmera (Sademo/Kresher sefer)	6.8
Mojo Lake	10.4	Menagesha (Chefekto)	6.6
Akaki Beseka	7.6	Welmera (Sademo near steel industry)	6.9
Awash River	7.5	Welmera (Melkato)	7.4
Average	**7.6**		**7.0**

Note: Holetta= Town; Welmera= District; Sademo=Sub-district.

Table 2. Concentration of heavy metals in water in East Shoa of different sampling sites

Sampling sites	Concentration of heavy metals (μg/L)				
	Pb	Cr	Cd	As	Mean ± SE
Gelan	5.21	67.45	0.75	4.45	23.94 ± 5.36^{b}
Akaki	3.91	56.73	0.99	12.69	20.59 ± 6.57^{c}
Bishoftu	0.0001	34.26	0.54	15.82	16.87 ± 13.14^{e}
Mojo	24.66	214.05	0.67	10.64	66.14 ± 7.58^{d}
Awash	16.18	83.52	0.92	19.04	29.92 ± 13.14^{a}
Overall mean	**9.14 ± 3.54^{b}**	**92.77±20.33^{a}**	**0.80±0.18^{c}**	**9.61±1.50^{b}**	**31.41± 13.14**

LS-means with different superscripts among rows holding mean values were significantly different (last column).

Concentration of Heavy Metals in Livestock Water by Location

The levels of heavy metals in livestock water in East Shoa and West Shoa are shown in Table 3. With the exception of cadmium, the levels of heavy metals in livestock water were significantly different ($P<0.05$) between study sites in East Shoa and West Shoa. Accordingly, higher concentrations of heavy metals were observed in livestock water in study sites of East Shoa (28.08 ± 7.02μg/kg) than in West Shoa (1.96 ± 0.28 μg/kg), associated with the different factories existing in the study locations and effluents released from these factories to nearby water bodies. As a result, water in these areas has become the primary reservoir for heavy metals.

Except for lead and arsenic, the elemental differences between the heavy metals in study sites in West and East Shoa was significant (P<0.05) and the levels of heavy metals in livestock water in the study locations were in the order of Cr > As > Pb > Cd (Table 3). The average concentrations of Pb (4.99 µg/L) and Cd (0.76 µg/L) observed in this study were also lower than the Pb (270 µg/L) and Cd (70 µg/L) content of water collected from shallow wells in Egypt (EI-Bassiony, *et al.*, 2016). This may be due to the reason that water samples were collected from the shallow and deep parts of underground water which is the usual water source of Egyptians that might be protected from exposure to different pollutants.

Table 3. Concentrations of heavy metals (µg/L) in livestock water by location

Element	N	West Shoa (Menagesha to Holetta)	East Shoa (Akaki to Mojo)	Average
Cadmium	30	0.72 ± 0.09^{a}	0.80 ± 0.18^{a}	0.76 ± 0.1^{d}
Lead	30	0.84 ± 2.84^{b}	9.14 ± 5.80^{a}	4.99 ± 1.93^{c}
Arsenic	30	4.03 ± 0.41^{b}	9.61 ± 1.5^{a}	6.82 ± 0.92^{bc}
Chromium	30	2.26 ± 0.25^{b}	92.77 ± 20.33^{a}	47.52 ± 13.05^{a}
Overall mean	120	1.96 ± 0.28^{b}	28.08 ± 7.02^{a}	15.0 ± 12.34
pH (mean and range)	120	7 (6.4-7.7)	7.6 (6.51-10.37)	

Ls-means with different superscripts between columns were significantly different (P<0.05) (column 3 and 4); Ls-means with different superscripts between rows were significantly different (P<0.05) (column 5); N= Number of samples.

Concentration of Heavy Metals (µg/L) in Water Close Factory Gates and Far from Factories in East Shoa (Akaki to Mojo)

There was a significant difference (p<0.05) in the concentration of heavy metals between water samples taken from factory gates and those ones collected far from the existing factories. Moreover, the concentration of Cr (123.23 ± 29.13) from the factory gates was significantly (p<0.05) higher than the concentration of heavy metals in water samples collected from sites far from the existing factories (Table 4).

Table 4. Concentration of heavy metals (µg/L) in water from factory gates and far from factories in East Shoa (Akaki to Mojo)

Water source	**As**	**Cd**	**Cr**	**Pb**
Near to factories	7.37 ± 1.57^{b}	0.95 ± 0.28^{c}	123.23 ± 29.13^{a}	10.0 ± 5.25^{d}
Far from factories	11.43 ± 2.17^{f}	0.89 ± 0.42^{h}	50.79 ± 15.14^{e}	7.07 ± 4.35^{g}

Concentration of Heavy Metals in Livestock Water in the Study Areas as Compared to NRC (2001) Upper Limit Guideline for Cattle

Results of this study showed that the concentrations of heavy metals in livestock water in both study locations were lower than the maximum limit indicated in NRC (2001) upper limit guideline for cattle (Table 5). Industrial activities in East Shoa contributed to the existence of higher heavy metals in livestock water in these locations.

Table 5. Concentration of heavy metals in livestock water in the study areas as compared to NRC (2001) upper limit guideline for cattle

Element	**Concentration (µg/L)**		**NRC (2001)**
	East Shoa (Akaki to Mojo)	West Shoa (Menagesha to Holetta)	(µg/L or ppb)
Cadmium	0.80 ± 0.18	0.72 ± 0.09	5
Lead	9.14 ± 3.54	0.84 ± 2.84	15
Chromium	92.77±20.33	2.26 ± 0.25	100
Arsenic	9.61±1.50	4.03 ± 0.41	50

MAC= Maximum allowable concentration.

Concentration of Heavy Metals (Mg/Kg) in Feeds (Teff Straw) in East and West Shoa

The concentrations of heavy metals in *teff* straw in East and West Shoa are shown in Table 6. Higher levels of As (2205.3µg/kg) and Cr (3204 µg/kg) were observed in *teff* straw when compared with the relatively lower concentrations of Cd (125.6µg/kg) and Pb (526.1µg/kg) in this study, indicating that there were significant (p<0.05) variations among individual elements and the concentrations of heavy metals in *teff* straw were in the order of Cr > As > Pb > Cd (Table 6).

As indicated in table 6, the cadmium, chromium and lead concentration of maize fodder collected from polluted areas of Pakistan (Farid and Baloch, 2012) was 750µg/kg, 29340µg/kg and 24280µg/kg, respectively, which is much higher than the cadmium (125.61µg/kg), chromium (3204.00 µg/kg) and lead (526.1 µg/kg) concentration in *teff* straw in this study.

Except for cadmium in (Holetta 168.81 ± 18.12 μg/kg compared to Bishoftu 82.41 ± 10.79 μg/kg), the levels of the other heavy metals in *teff* straw did not show significant ($p > 0.05$) variation between West Shoa (1543.54 ± 318.70 μg/kg) and East Shoa (1486.92 ± 279.73 μg/kg) Zones.

The cadmium and lead content of *teff* straw in the present study were also lower than the cadmium (155μg/kg) and lead (563μg/kg) content studied in wheat fodder and the cadmium (305 μg/kg) and lead (844 μg/kg) content studied in maize fodder in Pakistan (Iftikhar *et al.*, 2014).

Similarly, the cadmium (125.6 μg/kg) and lead (526.1μg/kg) content of *teff* straw found in this study were lower than 160 μg/kg and 2090 μg/kg of the same elements in maize fodder (livestock feed) grown in unpolluted areas of Pakistan and these levels were also lower than the cadmium (873.3 μg/kg), lead (24317 μg/kg) and chromium (29250 μg/kg) levels in fodder feeds grown in a polluted area of Pakistan (Farid and Baloch, 2012). *Teff* straw is categorized in Gramineae (grasses) and in cereals which is similar to wheat and maize fodder which is expected to have a similar degree of exposure to pollution of heavy metals. The fact that cadmium, lead and chromium levels in *teff* straw in this study were lower than the results reported in Pakistan by Farid and Baloch (2012) is an indication that *teff* growing areas in Ethiopia were not relatively highly polluted.

Table 6. Concentration of heavy metals (μg/kg) in feeds (*teff* straw) in East and West Shoa

Element	N	East Shoa (Akaki to Mojo)	West Shoa (Menagesha to Holetta)	Average
Cadmium	30	168.81 ± 18.13[a]	82.41 ± 10.80[b]	125.61 ± 13.11[d]
Lead	30	608.34 ± 103.16[a]	443.86 ± 68.76[a]	526.1 ± 62.80[c]
Arsenic	30	1979.51 ± 986.35[b]	2431.00 ± 797.95[a]	2205.3 ± 624.73[b]
Chromium	30	3417.50 ± 516.16[a]	2990.40 ± 483.48[b]	3204.00 ± 349.72[a]
Overall	**120**	**1543.54 ± 318.70[a]**	**1486.92 ± 279.73[a]**	**1515.23 ± 295**

Ls-means with different superscripts between columns were significantly different (P<0.05) (column 3 and 4); Ls-means with different superscripts between rows were significantly different (P<0.05) (column 5); N= Number of samples.

The chromium content of *teff* straw in the present study (3204 μg/kg) was 15-fold higher than the chromium contents of forage grasses (208 μg/kg) grown on the roadside of Botswana (Moreki *et al.*, 2013) and 2-fold higher than the chromium content of maize fodder studied in Pakistan which was 1340 μg/kg (Farid and Baloch, 2012).

Concentration of Heavy Metals (Mg/L) in Cow Milk in East and West Shoa

The levels of heavy metals in milk samples from the different study sites are presented in Table 7. Accordingly, the concentration of chromium (95.35 ± 14.16 μg/L) was significantly ($p < 0.05$) higher than the concentrations of cadmium 34.77 ± 4.41 μg/L, lead 33.01 ± 2.63 μg/L and arsenic 6.70 ± 1.15 μg/L. Except for chromium, the concentrations of cadmium, lead and arsenic in the milk samples did not show significant ($P>0.05$) differences between the study sites in East and West Shoa.

The overall levels of heavy metals in cow milk samples in the study locations were in the order of Cr > Cd > Pb > As. It is interesting to note that cadmium (34.24 ± 4.95) and lead contents (33.01 ± 2.63) of milk in this study were lower (3 and 30 times) than the cadmium (100.25 μg/L) and lead (998.25 μg/L) contents of milk collected from high yielding crossbred cows that were fed grass hay and agro-industrial by-products in Addis Abba (Dawd, 2010), respectively.

Table 7. Concentration of heavy metals (μg/L) in cow milk in East and West Shoa

Element	N	West Shoa (Menagesha to Holetta)	East Shoa (Akaki to Mojo)	Average
Cadmium	30	37.69 ± 3.49[a]	30.78 ± 9.36[a]	34.24 ± 4.95[b]
Lead	30	34.77 ± 4.41[a]	31.24 ± 2.95[a]	33.01 ± 2.63[bc]
Arsenic	30	7.03 ± 2.24[a]	6.36 ± 0.68[a]	6.70 ± 1.15[d]
Chromium	30	139.73 ± 23.00[a]	50.97 ± 4.56[b]	95.35 ± 14.16[a]
Overall mean	**120**	**54.81 ± 8.76[a]**	**29.84 ± 3.35[b]**	**42.32 ± 35.79**

Ls-means with different superscripts between columns were significantly different (P<0.05) (column 3 and 4); Ls-means with different superscripts between rows were significantly different (P<0.05) (column 5); N= Number of samples.

The lead content of milk in the present study was 3 times lower than the lead level of milk (91μg/L) collected from cows reared within 2 km of the industrial zone of India (Roy *et al.*, 2009), and that of Pb level (110μg/L) of milk samples studied in Pakistan (Farid and Baloch, 2012). Similarly, the cadmium (34.24 μg/L) and chromium (95.35 μg/L) levels of milk samples in this study were 2 and 3 times lower than the cadmium (60 μg/L) and chromium (290 μg/L) level of milk studied in Pakistan, respectively (Farid and Baloch, 2012). The lead, cadmium, and chromium content of milk of cows grazed around an industrial estate in Nigeria were 550 μg/l, 163 μg/l and 1756 μg/l, respectively (Ogabiela *et al.*, 2011) which was higher than the lead (33.01 µg), cadmium (34.24 µg) and chromium (95.35 µg) contents of milk observed in this study.

Studies on heavy metals chromium, arsenic and lead content of buffaloes milk from the contaminated area of Ludhiana district Punjab, India indicated that chromium, arsenic and lead contained 37000 µg/L, 33000 and 5000 µg/L, respectively which is much higher than cow milk collected from the polluted area of Bishoftu which was 50.97 µg/L, 6.36 µg/L, 31.24 µg/L, respectively (Yeotikar *et al.*, 2018).

The level of chromium in milk in this study was higher (95.35 g/L), which could be attributed to the high concentration of chromium in *teff* straw collected from the study locations implying that the level of chromium is higher in agricultural soil in the study areas, especially in Holetta. Attah and Melkamu (2013) in their study on the heavy metal content of the soil, river water and effluent in Holetta around the floriculture area reported that the levels of chromium were higher in the sample of soils (129000 µg/kg) than in effluent (470 µg/kg) and river water (370 µg/kg) in which the chromium content in soil was beyond the recommended limit in agricultural soils (100000 µg/kg). Based on another study, the heavy metal content of waste water in Eastern Industry Zone of central Ethiopia was reported to be 0.69 mg/L, which was above the recommended limit of WHO, which is 0.1 mg/L (Dagne, 2020). A higher level of Cr (20) above the recommended level (2.3) was also found in vegetables grown on polluted soils of Akaki area that was irrigated with waste water of the little Akaki River (Agajie, 2007).

Concentration of Heavy Metals in Milk in This Study as Compared to The Standard Set By WHO/JECFA

The levels of heavy metals in milk were compared with the standard set by WHO/JECFA (Table 8). Accordingly, cadmium and arsenic levels of milk in both study locations were within the WHO/JECFA standards. In addition, 27% and 40% of the milk samples collected from Bishoftu and Holetta respectively were within the WHO/JECFA standard set for lead in milk. Whereas, the rest milk samples were above the WHO/JECFA standard. Results having a similar trend were also reported in Iran in which the concentration of lead in 11% of cow milk samples was higher than the maximum level reported in codex which is 0.02 µg/L (Derakhshesh and Rahimi, 2012). In addition, the concentrations of cadmium, chromium and lead in milk samples collected from cows fed on effluent irrigated fodder in Pakistan were above the permissible limit for human consumption (Farid and Baloach, 2012). It was reported that in humans, exposure to lead toxicity at lower levels results in loss of cognitive power (NRC, 2001). Like other heavy metals, consumption of lead and chromium beyond the permissible limit could result in damaging vital organs like the kidneys and liver in chronic situations (Bala *et al.*, 2014) and can also lead to death in acute cases when consumed at higher concentrations (Rajaganapathy *et al.*, 2011). Moreover, none of the chromium content of milk samples in both study locations was within the WHO/JECFA standards.

Table 8. Concentration of heavy metals in milk in this study as compared to WHO/JECFA (1989) standards

Site	Heavy metal	Mean heavy metal (µg/L)	Permissible standard (µg/L)	Within standard (%)	Beyond standard (%)
Bishoftu	Cd	30.78	71	100	0
	Pb	31.24	25	27	73
	As	6.36	156	100	0
	Cr	50.97	17	0	100
Holetta	Cd	37.69	71	100	0
	Pb	34.77	25	40	60
	As	7.53	156	100	0
	Cr	139.74	17	0	100

Conclusion

Results of this study showed that the average pH values of water collected from both study locations were within the normal pH range for livestock water. The concentration of heavy meatal in both study areas were found less than the standard limit of livestock water. The concentration of chromium in Mojo town was recorded the highest level. The concentration of heavy metals in water samples collected from East Shoa was more than ten times higher than that of West Shoa. The concentrations of chromium and lead in majority of the milk samples examined in this study were beyond the permissible limit. To come up with strong recommendation, there is a need of further study on heavy metals level of soils, other fodder feeds and livestock products in these study areas.

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Conflict of Interests

The authors declare that they have no competing interests.

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