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Assessing Heavy Metal Contamination Using Biosensors and a Multi-Branch Integrated Catchment Model in the Awash River Basin, Ethiopia

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Abstract: Metal pollution in rivers from untreated industrial and domestic wastewater is a major issue in economically developing countries worldwide. The Awash River Basin in Ethiopia is one of those rivers that faces rising heavy metal concentrations due to poor wastewater management and loose law enforcement controlling effluent discharge into rivers. In this study, surface water and wastewater samples were collected within the Awash River Basin, with metals analysis using ICP-MS techniques. Acute toxicity of water was determined using new molecular biosensor technology based on engineered luminescent bacteria. A multi-branch Integrated Catchment Model (INCA) for metals, including Arsenic, Cadmium, Chromium, Copper, Lead, Manganese, and Zinc was applied to the Awash River Basin to simulate the impact of tannery discharge on the river water pollution levels and to evaluate a set of treatment scenarios for pollution control. Results show that all samples from tannery wastewater have high levels of metals, such as Chromium and Manganese with high levels of toxicities. River water samples from upper Awash near Addis Ababa showed elevated concentrations of heavy metals due to the untreated wastewater from the dense population and a large number of industries in that area. The modeling scenarios indicate that improved wastewater management will reduce the metal concentration significantly. With a 50% reduction in effluent concentrations, the mean concentrations of heavy metals (such as Chromium) over two years would be able to reach 20 to 50% reduction in river water samples.

Keywords: metal pollution; Ethiopia; tanneries; Awash River; biosensors; toxicity

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1. Introduction

Contamination of surface waters from heavy metals in industrial and domestic effluents is a common feature in many economically developing countries. Human activities, such as intensive agriculture and irrigation, urbanization, and industrialization create major issues for pollution control and, if not managed correctly, can cause major harm to river water quality and ecology and threaten public water supplies [1–7]. Consequently, in developing nations, water scarcity due to the water pollution of rivers and lakes represents a major challenge [8]. The UN Sustainable Development Goals (SDG 6.3) directly address this issue to close the gap on water scarcity, water quality, and pollution in order to protect the health of people and livestock, as well as to maintain and enhance livelihoods, especially of poor people, women, and children.

In Ethiopia, industrialization, urbanization, and human settlement are increasingly intense, especially around the major cities such as Addis Ababa. The impact on clean water resources from anthropogenic activities has been growing due to poor wastewater management, few monitoring systems, and weak enforcement of pollution control strategies [9–11]. In addition, there are many technical issues, for example, conventional treatment plants are not generally capable of removing emerging pollutants such as pesticides, pharmaceuticals, or heavy metal wastes. Heavy metal pollution in rivers is a major threat to stream ecology and human health due to metal persistence and toxicity [12–16]. Exposure to elevated concentrations of heavy metals, such as Chromium, Mercury, Lead, Arsenic, Cadmium, and other trace metals may cause cancer, retard human development, productivity and can cause severe health and environmental effects and also in extreme cases, death [17–21].

This paper focuses on the Awash River Basin in Ethiopia and, in particular, the upper reaches of the river where there are major discharges of partially treated or untreated industrial and domestic wastes. Tannery effluents are of serious concern and are typically characterized as pollution-intensive industrial complexes that generate widely varying high-strength wastewater [16,22,23]. Degradation of surface waters due to wastewater is becoming a serious problem as most of the industries in the basin are situated along the Awash or its tributaries including Modjo and Akaki Rivers. These then deliver polluting waters into the main river system, thereby degrading the river water of Awash.

Determination of heavy metals in waters has been a challenging task in Ethiopia, due to the lack of conventional instruments (ICP-MS, ICP-OES, GC-MS, UV, X-ray absorption spectroscopy), new technology (biosensors), and skilled professionals. This research provided the first comprehensive investigation of the Awash River Basin focusing on metals and water toxicity using ICP-MS and biosensors. In addition, a flow and water quality model INCA (Integrated Catchment) was applied to assess the metals and toxicity data and to simulate the Upper and Middle Awash River system. The model was used to investigate the effects of the tannery discharges, assess impacts on river water quality, and evaluate the effluent treatment levels required to meet international instream quality standards.

2. Study Area and Methods

2.1. Study Area

The Awash River, 1200 km long, is one of the twelve rivers in Ethiopia (Figure 1). It originates near Ginichi town, which is located to the west of the capital Addis Ababa, and flows through the Rift Valley until it reaches Lake Abe on the border shared by Ethiopia and Djibouti. The Awash River Basin is characterized by highlands escarpments and rift valley, with an elevation ranging from 3000 to 250 m a.s.l. The entire Awash River Basin covers a total area of 116,200 km^2, with the western catchment comprising 70,800 km^2 that drains into the main Awash River. In contrast, the eastern catchment (45,400 km^2) drains into a desert area and does not contribute flow to the main river course [24]. Thus, the eastern catchment is not considered in this study.

The recent development in the Awash River Basin such as industrialization, urbanization, and small and large-scale farming makes the Awash the most developed river basin in Ethiopia and the basin accounts for 25% of the agricultural production of the country. The upper catchment including the national capital, Addis Ababa, is densely populated. The basin comprises more than 12,500 industries and more than 65% of industries are located in the upper catchment, upstream of Koka Dam. Given the location of the industries, sampling collections were mainly focused on the upper catchment and middle valleys of the Awash River Basin (Figure 1). The modeling effort has been applied to the entire Awash River Basin so that the impact of the effluent could be assessed immediately after the discharge points as well as at the downstream sites.

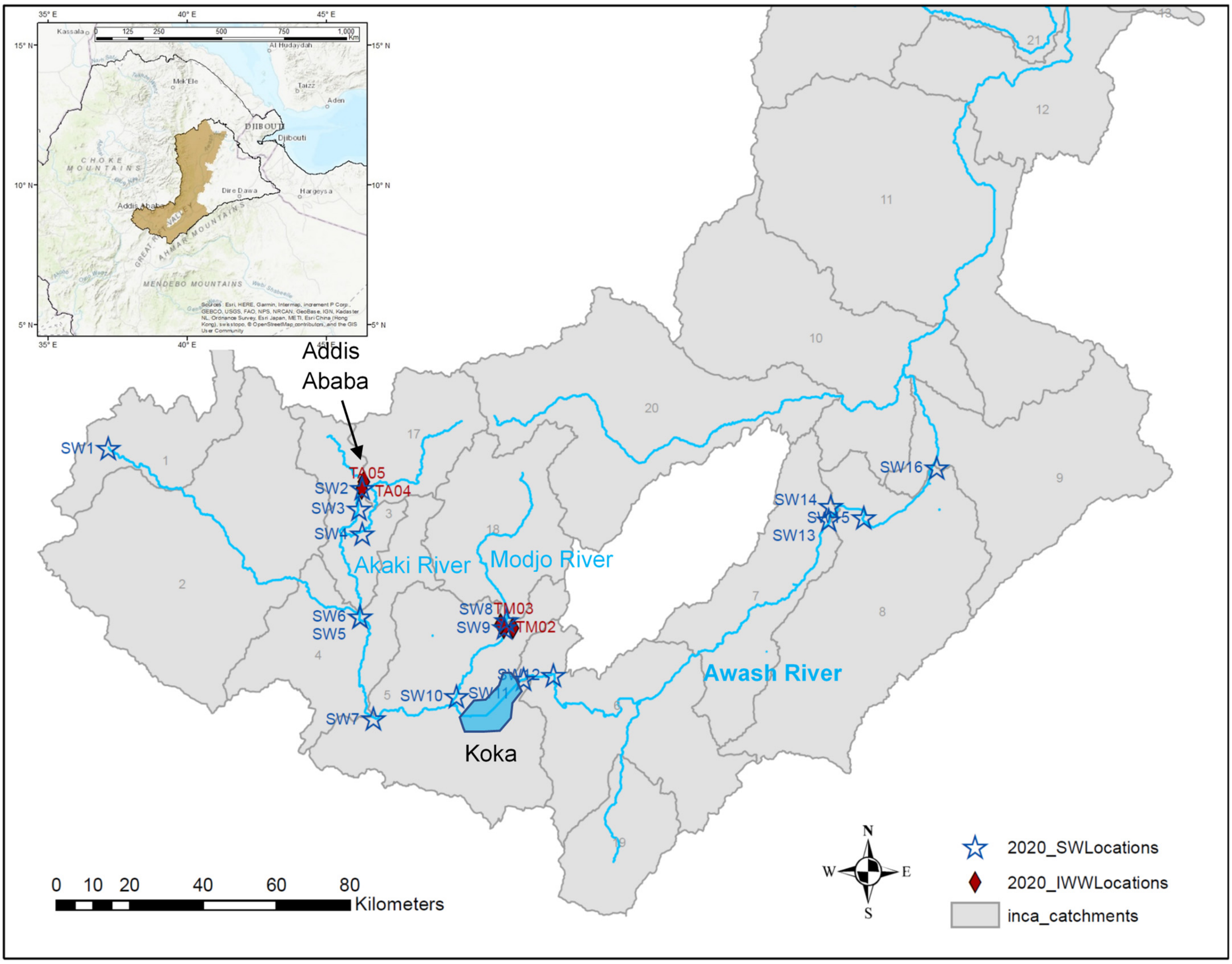


Figure 1. Locations of sixteen surface water samples (SW1-16) and five industrial wastewater samples (TA01-02 and TM01-03) in 2020. The numeric numbers in grey indicate INCA modeling catchments.

2.2. Sampling and Metals Analysis

Twenty-one samples were collected from sixteen surface water locations and five industrial wastewater effluents in 2020 (Figure 1).

After sample collection in the field, they were acidified before analysis, which allowed equilibrium overnight to permit the re-dissolution of any precipitates or materials adsorbed to the bottles. Over 30 heavy metals were analyzed by inductively coupled plasma mass spectrometry (ICP-MS) using a PerkinElmer Nexion 2000B instrument, which was coupled with an Elemental Scientific prepFAST M5 autosampler at the University of Oxford. The instrument was calibrated using the method of external calibration where the concentrations for the measured sample set were extrapolated from linear regressions generated from raw counts per second data from a series of standards. Specifically, for each element, a six-point calibration curve was created by measuring the ion count of six known concentrations and applying linear regression. Two commercial references were used to validate the calibration curves. Commercial references are commercially available solutions that are independent of the calibration solutions, with certified concentrations of a range of elements. The detection limits of each element are given in Table 1.

Table 1. Metal concentrations at sixteen sites in the river system and five tanneries. Concentrations are in µg/L (ppb). BDL indicates below the detection limit. The metal detection limits and the World Health Organization (WHO) drinking water standards were listed at the bottom.

Sample ID	Type	Fe	Al	Rb	Sr	Cr	Mn	Co	Ni	Cu	Mo	Zn	As	Li	Cd	Hg	Pb
SW1	River	155.1	121.4	2.1	293.4	0.3	108.1	0.8	1.9	2.0	1.7	4.9	0.5	1.3	BDL	BDL	0.1
SW2	River	3896.8	1980.4	35.3	442.4	214.2	2170.4	4.4	9.8	9.3	1.3	72.3	2.8	7.4	0.12	BDL	7.9
SW3	River	2695.0	1455.4	38.6	414.5	138.3	2240.2	5.5	12.1	15.4	1.8	75.7	2.6	7.5	0.10	BDL	9.0
SW4	River	1741.8	1065.0	19.7	314.4	5.4	1455.0	3.8	7.4	5.1	3.2	24.0	2.8	12.4	BDL	BDL	2.5
SW5	River	1657.6	1659.8	17.2	319.9	3.0	678.0	5.0	13.4	9.1	3.9	14.0	3.0	8.3	BDL	BDL	2.4
SW6	River	1082.7	1256.9	32.7	73.4	2.1	65.1	0.6	2.8	2.7	103.8	10.2	23.0	28.6	0.10	BDL	0.6
SW7	River	1431.5	1560.4	17.3	323.5	2.6	850.5	4.4	12.7	7.0	4.5	11.9	2.9	8.3	BDL	BDL	1.7
SW8	River	307.7	386.4	10.1	320.8	0.6	137.6	1.1	5.1	2.6	6.3	13.8	2.3	23.2	BDL	BDL	0.4
SW9	River	272.1	314.9	12.0	322.4	139.1	330.2	1.3	4.7	2.2	5.6	8.6	2.7	25.9	BDL	BDL	0.4
SW10	River	410.9	382.3	15.6	374.2	69.4	747.1	3.6	12.3	3.1	8.5	9.3	4.1	36.9	BDL	BDL	0.6
SW11	River	1003.4	1164.2	3.9	150.3	1.5	57.5	0.8	3.7	3.5	1.9	7.3	0.9	2.9	BDL	BDL	0.9
SW12	River	1162.3	1406.2	4.6	151.5	1.6	67.6	0.9	4.3	3.7	2.3	8.5	1.0	4.2	BDL	BDL	1.0
SW13	River	1833.0	1908.5	6.7	169.8	2.1	277.4	1.9	5.6	5.1	2.4	11.9	1.7	6.9	BDL	BDL	1.6
SW14	River	1014.8	1208.8	32.4	75.9	2.0	67.1	0.6	2.6	3.4	108.7	7.2	25.0	28.0	0.12	BDL	0.6
SW15	River	2158.1	2135.1	10.5	162.4	2.4	370.2	1.7	5.8	4.1	16.1	10.0	5.9	10.1	BDL	BDL	1.3
SW16	River	1732.5	1870.2	11.6	173.8	2.0	239.5	1.5	5.5	3.7	19.9	8.4	5.4	10.6	BDL	BDL	1.1
TM01	IWW	729.6	2239.6	77.5	469.0	18,038.1	168.3	0.8	17.5	3.2	BDL	31.9	BDL	67.9	BDL	BDL	0.6
TM02	IWW	37.2	67.9	85.7	235.1	1229.0	6.3	0.5	5.4	BDL	BDL	7.4	BDL	76.3	BDL	BDL	BDL
TM03	IWW	525.3	146.2	187.9	453.3	474.9	60.2	10.6	10.7	0.9	BDL	35.4	1.8	55.1	BDL	BDL	0.3
TA04	IWW	2278.8	3358.1	150.7	582.8	4329.5	3097.1	7.4	28.6	10.1	3.3	77.1	BDL	234.0	BDL	BDL	1.1
TA05	IWW	74.3	1881.0	46.2	438.8	34.1	472.6	3.5	3.5	0.8	1.1	11.1	BDL	23.4	BDL	1.81	0.1
Detection Limits		0.640	0.059	0.013	0.392	0.006	0.008	0.002	0.003	0.044	0.150	0.198	0.003	0.057	0.001	0.650	0.015
WHO standards		300	200	-	-	50	100	-	20	1300	70	5000	10	-	3	2	10

2.3. Biosensors

Molecular biosensors are bacterially derived sensors that determine the bioavailability of both specific chemicals and the overall toxic effect. These biosensors have been developed and provided by Oxford Molecular Biosensors, a spinout company from the University of Oxford (www.omb.co.uk, accessed on 1 June 2023). For this study, we employed a metabolic-based acute toxicity sensor, *Acinetobacter baylyi* ADP1 Tox2, which has previously successfully demonstrated the presence of cell damage or cytotoxicity in heavy metal-contaminated seawater [25] and river water [16]. The soil bacterium-based biosensor has been engineered, using a synthetic biology approach, to luminesce brightly in low-toxicity samples, and dimly in cytotoxic conditions, as illustrated in Figure 2. They sensitively respond at very low concentrations of toxic pollutants, with resolution over several orders of magnitude. Certain bacteria such as *Vibrio fischeri* naturally bioluminesce and have been used commercially for the detection of toxicity in freshwater toxicity detection for decades by companies such as Microtox [26,27], however, engineered whole-cell biosensors that are genetically designed for heightened sensitivity and detection of individually toxic components are new to the market. The OMB biosensor testing protocol utilizes positive and negative controls in every assay, allowing comparison between assays and a semi-quantitative result proportional to toxicity. This can then be calibrated against known concentrations of cytotoxicants for comparison, for example, Zinc.

The acute toxicity biosensor is important in determining water quality from an ecology standpoint and also to inform public supplies. Biosensor assays are rapid, taking less than one hour and so they are uniquely placed to act as an early warning of toxic pollution. The yellowstripe goby fish, *Mugilogobius chulae*, is often used as a model for toxicity. When the cytotoxicity biosensor was compared with the goby results, they were found to be comparable, demonstrating that the biosensor system can function as a proxy for cytotoxicity assessments in animal models [25]. The biosensor output encompasses the presence and bioavailability of toxicity, precluding the need to use multiple sensors and hence it can be used to determine whether further testing is required. It can test both fresh and saline water without additional pre-treatment, allowing more powerful comparisons between samples and flexibility of testing. The biosensor data in this study was collected and analyzed with reference to the metals data in order to ascertain the presence of any trends or patterns that might be important in the assessment of pollution around the Awash River Basin.

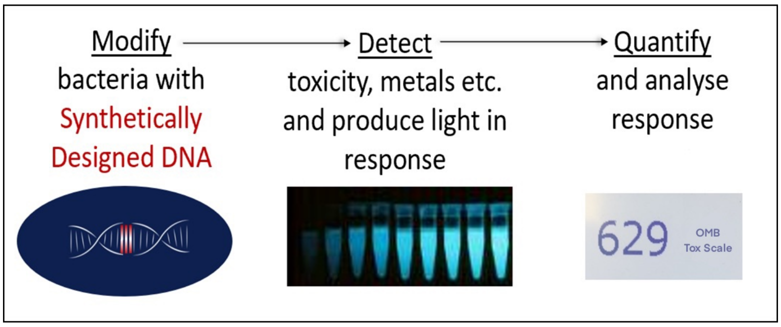


Figure 2. Luminescence from the bacterial biosensors reflecting acute toxicity.

Samples collected at the same 21 locations (Figure 1) were passed through a 0.2 uM filter before being challenged against the acute toxicity biosensor for 30 min. A positive control of 0.4 mM Zinc from Zinc Chloride is included as a known toxicant for reference. Results were then standardized to the negative control, 0.01 M sterile phosphate-buffered saline (PBS), and expressed as a percentage. Values above 110% (100% plus one standard deviation) were considered stimulatory; and values below 90% (100% minus one standard deviation) were considered toxic, with magnitude increasing as values decrease. When the tested value is significantly lower than the control, cell metabolism is inhibited, and toxicity is implied. When the tested value is significantly higher than the control, the result can be considered 'stimulatory'. This may be a result of hormesis, whereby the concentration of toxic compound(s) is below the threshold for toxicity, and instead has a beneficial or stimulatory effect on an organism [28]. The adage of 'the dose makes the poison' attributed to Swiss philosopher Paracelcus implies that toxicity is just a matter of dose. It is also possible that when perturbed, the cell mounts a response to the mixture by mobilizing energy reserves [29]. Therefore, stimulation of luminescence may suggest an early form of toxicity to a surmountable response, or a positive stimulation/beneficial effect.

2.4. *INCA-Metals Application to the Awash River Basin*

The Integrated Catchment Model (INCA) is a dynamic, daily time-step, catchment scale process-based model to calculate pollutant transfer from both diffuse sources and point sources to the catchment outlet. INCA model was originally created to simulate nitrogen [30]. Over the past two decades, new versions of INCA models were developed such as INCA-Metals [31]. The updated INCA-Metals model has a multibranch structure which allows it to simulate the in-stream metal concentrations in the dendric stream network [32].

INCA-Metals is a mass-balance dynamic model that estimates the daily fluxes and concentrations of flow, ammonia, cyanide, and eight metals (Cadmium, Lead, Zinc, Mercury, Arsenic, Copper, Chromium, and Manganese) based on 1 km^2 cell. The flow and metals are simulated by accounting for contributions from various inputs and transformations in the soil, groundwater, and rivers. The processes for INCA-Metals are based on those included in the Integrated Catchment model of Nitrogen but have been adapted to represent the metal adsorption to sediment, cyanide decay to ammonium, and cyanide volatilization [32]. The sources of pollutant inputs to the model include point sources such as abstractions, waste dumps, treated or untreated waste effluent as well as diffuse pollution from rural runoff. The hydrology component of the model is the same as other

INCA models where the river flow is generated by the soil flow, direct runoff flow, and groundwater flow [33]. Detailed equations for calculating flow and metal chemical fluxes can be found in Whitehead et al. [32]

The INCA models (Chloride, Nitrogen, and Phosphorus version) have already been applied to the Awash basin [34,35], however, this study is the first INCA application of the metals. The same INCA setup implemented in Bussi et al. [35] and Jin et al. [34] has been used here. To simulate flow and metal concentrations in the Awash River using INCA, the catchment was divided into 24 sub-catchments (Figure 1, shows the upper and middle Awash Basin), following the same setup as in Jin et al. [34]. The sub-catchment/reach boundaries were selected at confluences, effluent discharges, flow stations, and water quality monitoring stations, using the same principle as other INCA applications. Sub-catchments were delineated using a digital elevation model (DEM) in ArcGIS, which was obtained from the Shuttle Radar Topography Mission (SRTM) [34–36]. The land use information was from the GlobCover Portal and the most updated dataset from January to December 2009 was used in this study [34,35,37]. For the Awash River Basin application, six land use classes including arable, grassland/mixed, forest, water, bare, and urban were selected. The percentage of each land use class, sub-catchment area, and reach river length were calculated for each sub-catchment in ArcGIS which were used as INCA basic inputs that describe river network topology, reach characteristics, and sub-catchment characteristics.

Hydrological inputs to INCA include daily time series of precipitation, temperature, hydrologically effective rainfall (HER), and soil moisture deficit (SMD). HER and SMD are estimated using the Precipitation, Evapotranspiration, and Runoff Simulator for Solute Transport (PERSiST) model, which is a conceptual, daily time-step, semi-distributed model designed primarily for use with the INCA models [34,38]. In this study, daily rainfall data were acquired from the Climate Hazards Group Infrared Precipitation with Stations v2.0 (CHIRPS) [11,39]. Daily temperature data were obtained from a network of local weather stations. Both precipitation and temperature were acquired for the period between 1981 and 2021.

The Awash River Basin was divided into three climate zones: Upper Awash, Middle Awash, and Lower Awash to account for spatial variation of rainfall and temperature. To calculate average daily rainfall and temperature, the same procedure applied to Bussi et al. [35] and Jin et al. [34] were used in this study. A gridded dataset (10 $\times$ 10 km) of daily precipitation and temperature was created to compute the average of all the 10 $\times$ 10 km cells within each of the climate zones. The daily time series of HER and SMD between 1981 and 2021 were then generated by PERSiST for the Upper, Middle, and Lower catchments.

3. Results and Discussions

3.1. Metal Pollution in Awash River Basin

The metals results for sixteen locations along the river and five tanneries are shown in Table 1. The results indicate high concentrations of Chromium (Cr), Lithium (Li), and Cobalt (Co) in the tannery discharges, with these resulting in high concentrations in the downstream tributaries and river locations. For the river samples, at least one sample had concentrations of Iron (Fe), Aluminum (Al), Chromium (Cr), Manganese (Mn), Molybdenum (Mo), or Arsenic (As) that were higher than the WHO limits (Table 1). Specifically, for Mn, the majority of the samples (12 out of 16) exceeded the WHO limit. For As, two of out sixteen river samples had concentrations over the WHO limit. For Copper (Cu), Zinc (Zn), Cadmium (Cd), Mercury (Hg), and Lead (Pb), all samples were below the WHO limits.

Metal concentrations along the Awash River vary significantly (Figure 3). Concentrations of Cadmium, Chromium, Copper, Manganese, Lead, and Zinc were highest near the city of Addis Ababa reflecting the dense population and large number of industries in that area. Concentrations of Arsenic and Cadmium are high at the downstream locations in the middle Awash. This could be due to the mixing of the Beseka Lake water that is supported primarily by groundwater with high heavy metal concentrations [40].

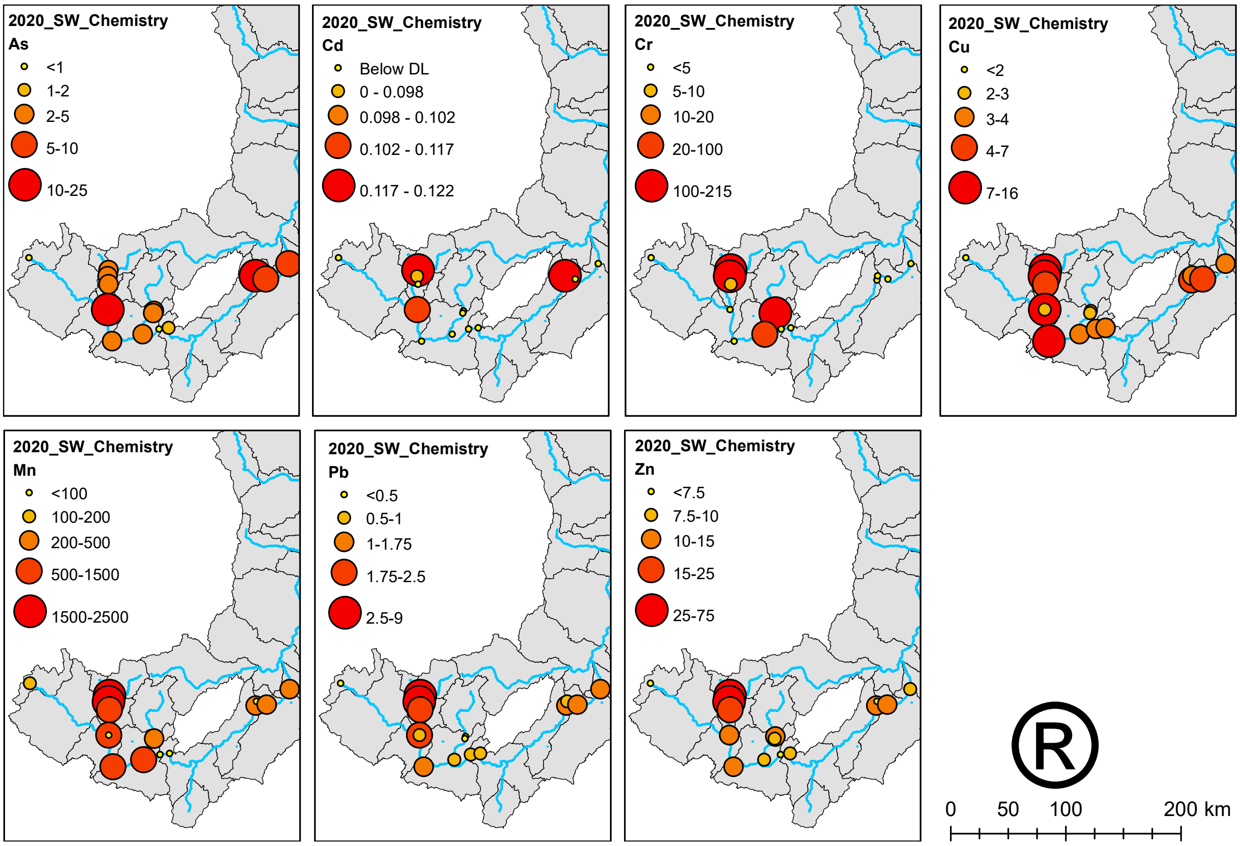


Figure 3. Spatial variations of selected metals in river water samples. Concentrations are in µg/L.

3.2. Biosensors Results and Toxicity

The results of the acute toxicity assay are displayed in Figure 4, with toxicity expressed as a percentage of luminescence relative to the PBS control.

The results from testing the instream samples using the molecular biosensors suggested that the river sample toxicity was never significantly greater than the control (less than 90% luminescence); however, there was significant stimulation in 13 out of 16 samples. The Awash after Akaki mix (SW6) and Lake Beseka samples (SW14) gave the highest toxicity readings of instream samples though they were not statistically different from the control ($p > 0.5$). The Akaki results may be lower than most samples tested due to the tannery discharges, and Lake Beseka may contribute high salinity levels and/or rock weathering (geogenic) effects due to subsurface flows of active rift and the presence of volcanic ash [34,40].

The stimulatory effect in the majority of samples may be a result of hormesis, whereby a low concentration of toxicant is stimulatory or beneficial to an organism. This may in turn indicate that the concentration is below the toxicity threshold; or may be due to the mobilization of energy reserves within the biosensor, in a protective response of a low-grade toxicant. With the tannery samples, three out of five samples were significantly toxic at the TM01, TM02, and TM03 locations. The sample from TA04 was not significantly different from the control, and the sample from TA05 was slightly stimulatory. The largest acute toxicity was observed at TM02 (2.9%) which was highly statistically significant.

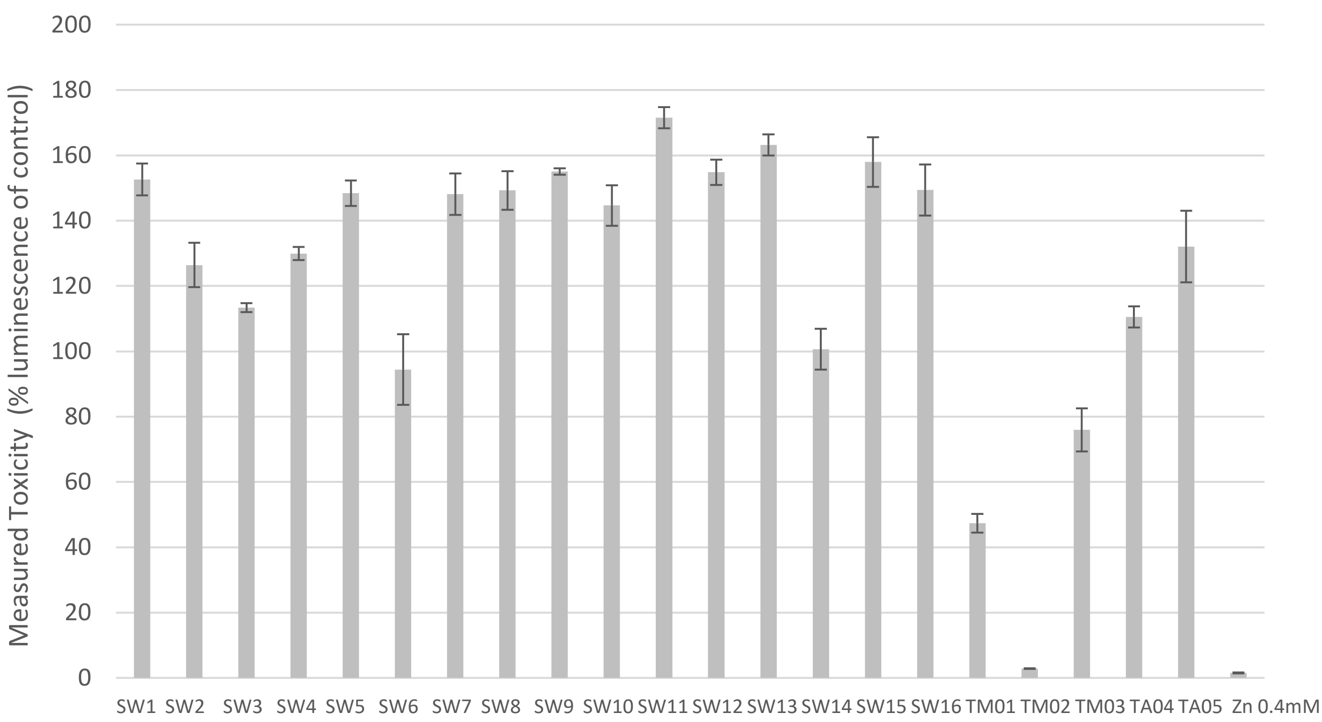


Figure 4. Acute toxicity results expressed as a percentage of luminescence relative to the phosphate-buffered saline (PBS) control taken at t = 30 min. All samples (n = 21) were tested in technical triplicate and error bars denote one standard deviation above and below the mean.

The output from the acute toxicity biosensors can be described as the net biological impact of the complex mixtures within a water sample. It is indicative of changes in metabolic activity within the cells. These biosensors have been engineered to constitutively express luminescence until metabolically perturbed. Luminescence-based biosensors report on the holistic whole-cell effects of pollutants on cellular metabolism, and therefore the output encompasses many toxic mechanisms such as genotoxicity, cell membrane disruption, or osmotic stress as just a few examples [26]. As they are affected by the individual components of a water sample as well as their combinatorial effects, they are not necessarily suitable for identifying specific individual drivers of toxicity; however, correlations between the concentrations of certain metals and the toxicity values indicated that some metals may have more of a pronounced toxic effect on the biosensors. In particular, a previous study identified Zinc, Cadmium, and Copper as biosensor toxicants [25].

Many metal species are known to have toxic effects through the generation of reactive oxygen species (ROS) which cause oxidative stress within the cell and can cause gene damage. As a holistic snapshot of water quality, biosensor technology has the potential to be used for rapid (30 min), in situ detection of toxic compounds in water. It can be calibrated to specific sample areas, making it a valuable tool for environmental monitoring and public health.

With the detailed analysis of metals available, toxicity/stimulation data of river samples (n = 16) was related to eight metals that the INCA-Metals model simulates. This exploratory exercise was to find statistically significant metals that relate to river water toxicity in the Awash River system. With an established relationship, metal simulation output from the INCA-Metals model could potentially be used to reasonably predict river toxicity values over time at any reach in the Awash River. To do this, backward regression analysis was applied to acute toxicity and eight metals. It was found that Lead, Zinc, Arsenic, Copper, and Manganese were highly linked to acute toxicity (Table 2). Multiple regression with the metals data yielded a predictive equation based on these metals with a high correlation of $r^2 = 0.94$ (Figure 5):

$$\text{Acute toxicity} = 17.364 \times [\text{Lead}] - 1.715 \times [\text{Zinc}] - 2.631 \times [\text{Arsenic}] - 3.361 \times [\text{Copper}] - 0.013 \times [\text{Manganese}] + 177.861 \quad \text{EQ.1}$$

Table 2. Statistical results of multiple regression analysis.

	Coefficients Estimate	Coefficients Standard Error	t	*p*
(Intercept)	177.861	6.106	29.127	<0.001
Lead	17.364	7.075	2.454	0.034
Zinc	−1.715	0.650	−2.638	0.025
Arsenic	−2.631	0.248	−10.605	<0.001
Copper	−3.361	1.779	−1.890	0.088
Manganese	−0.013	0.006	−2.058	0.067

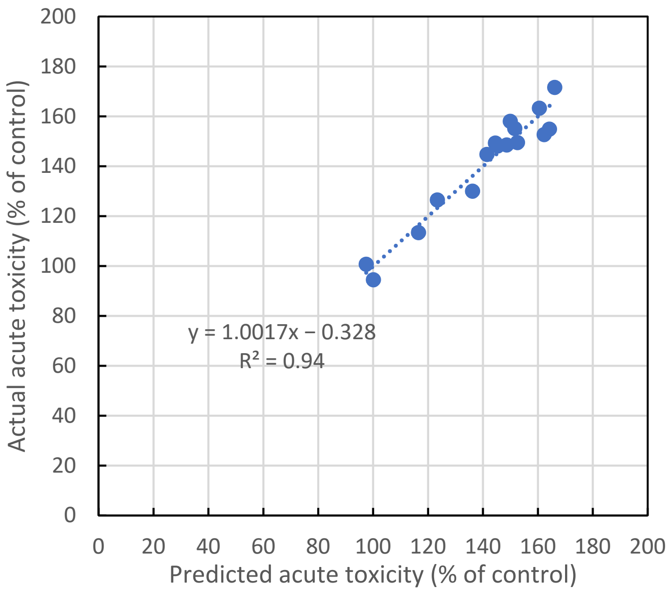


Figure 5. The statistical model predicted acute toxicity versus actual acute toxicity for the river samples.

3.3. Modelling Tannery Effluent Controls

To assess the impacts of the tanneries on the Awash River, the INCA-Metals model has been set up for the Awash River System, based on the previous applications of the model by Bussi et al. [35] and Jin et al. [34]. These applications were set up to address issues on nutrient management in the Awash and also the issues of salinity in the river and Lake Beseka. These two papers discuss in detail the INCA model setup and the definition of reach boundaries using topography (Figure 1) and other spatial data such as land use, animal numbers, and population, as well as the key driving data such as the daily rainfall, temperature, and soil moisture deficit.

Typical simulations for flow and salinity are shown in Figure 6 and demonstrate reasonable fits to the observed data with KGE statistics (Kling-Gupta efficiency) [41] of 0.7 for both flow and salinity [34]. The INCA-Metals model has been set up using the same information as the other INCA applications and, in this case, the INCA-Metals model has been set up with the 5 tanneries located at their reach locations (Figure 1). The model has then been run to assess the impacts of the metals on the downstream water quality. Figure 7 illustrates the simulation over 2 years showing the daily flows and the associated Chromium and Manganese water chemistry at a downstream river reach from tanneries (reach 5). The time series of Chromium show higher concentrations in the low flow period, due to the lack of dilution from rainwaters, and much lower concentration in the high flow periods. Manganese however has a different pattern. When the flow is higher, the Manganese concentration also goes higher which might reflect the contribution of high

Manganese from groundwater. Furthermore, the INCA model calculates the distributions of the simulated data. The spread of behaviors in Figure 8 indicates a non-gaussian type distribution with a mean Chromium of 0.0615 mg/L, which is above the WHO metals limit of 0.05 mg/L. Manganese has a mean of 0.112 mg/L, which is also above the WHO metals limit of 0.1 mg/L. The high mean concentrations of Chromium and Manganese in this reach are due to the discharge of tannery effluent into the Akaki River (Table 1). With untreated or partially treated effluents continuously discharging high levels of metals into the river system, the Awash River water will become increasingly impacted over time.

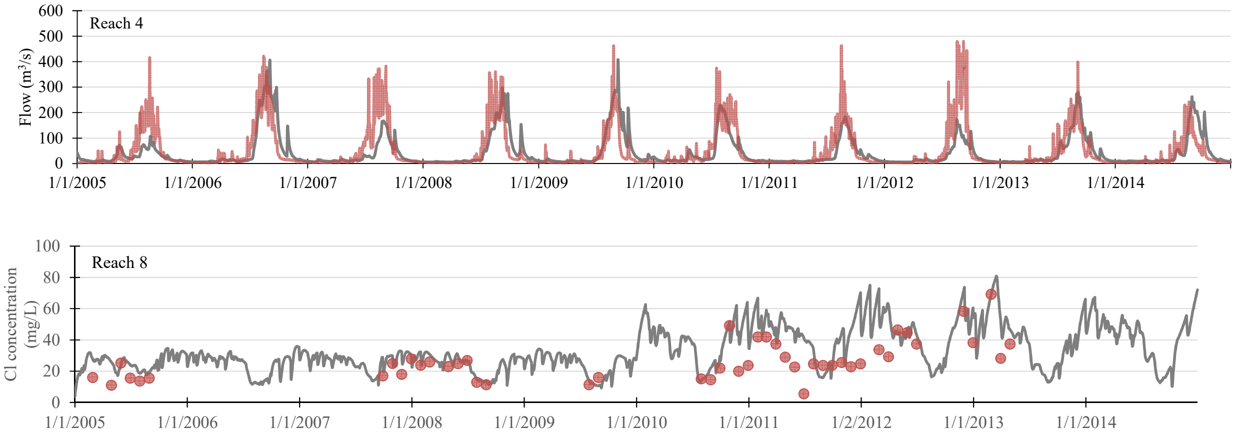


Figure 6. Flow and salinity model simulations (grey lines) compared to observed flow (red line) and salinity data (red circles) 2005–2014.

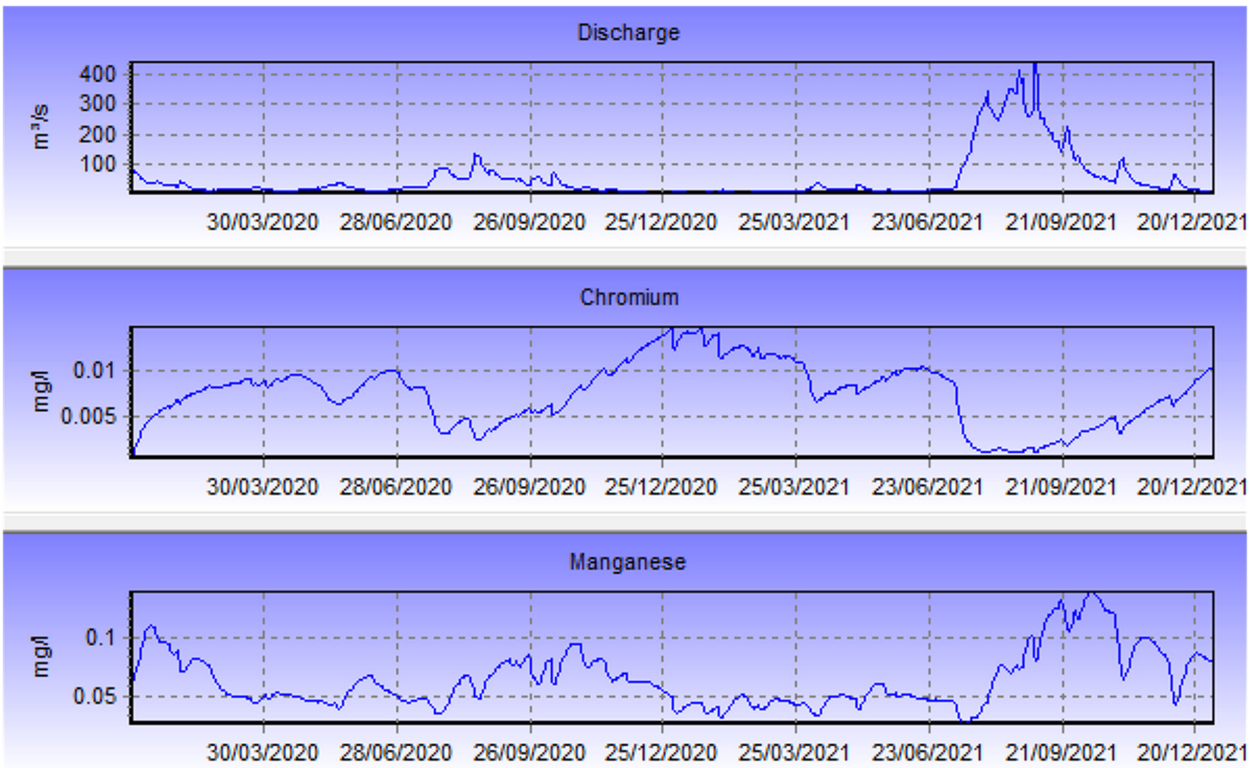
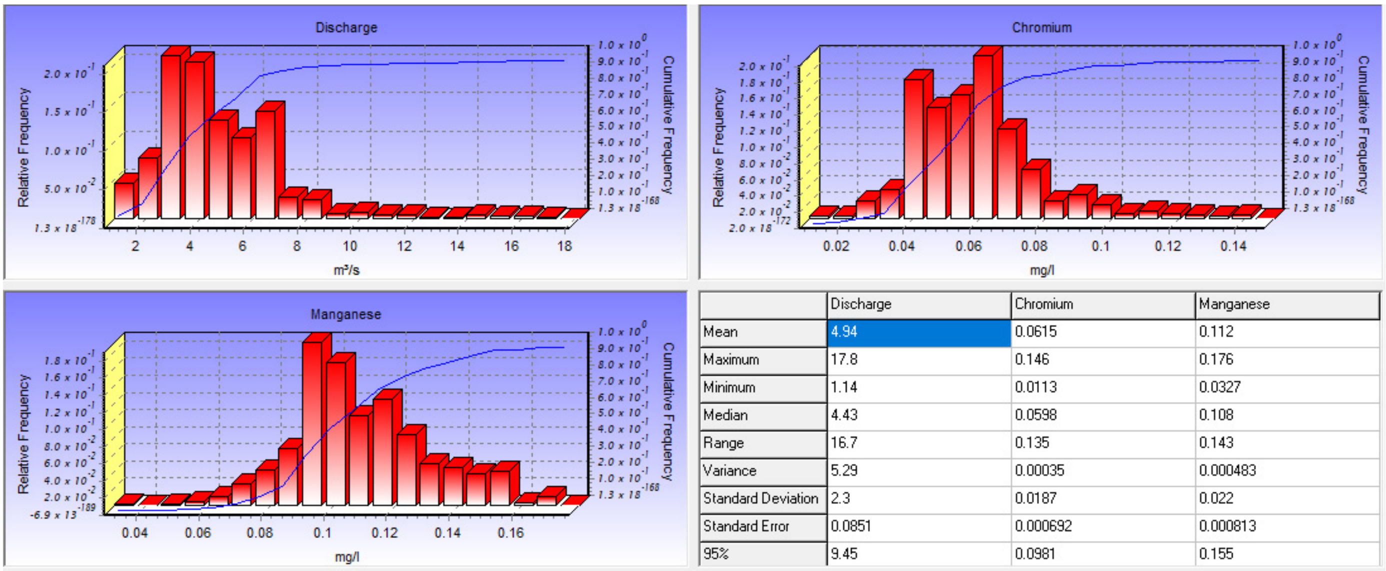


Figure 7. The simulated flow, Chromium, and Manganese concentrations in the main Awash River below the tanneries at reach 5, over 2 years 2020–2021.



	Discharge	Chromium	Manganese
Mean	4.94	0.0615	0.112
Maximum	17.8	0.146	0.176
Minimum	1.14	0.0113	0.0327
Median	4.43	0.0598	0.108
Range	16.7	0.135	0.143
Variance	5.29	0.00035	0.000483
Standard Deviation	2.3	0.0187	0.022
Standard Error	0.0851	0.000692	0.000813
95%	9.45	0.0981	0.155

Figure 8. Distributions of flow, Chromium, and Manganese in the river system downstream of the tanneries (reach 18).

To help assess the future condition, a scenario analysis has been undertaken as an example to simulate metal concentrations with future effluent treatment. Figure 9 shows the simulated Chromium concentrations with a 50% reduction in effluent concentrations compared to the original Chromium concentrations. The peak metal concentration at Reach 4 in the main Awash River profile reflects the tanneries discharge (Figure 9). With a 50% reduction in Chromium concentration in tannery effluent, the reduction of the mean Chromium concentration in river water is significant with a nearly 50% reduction at reach 4 which is immediately downstream from the tanneries. Moving down the river profile, the impact became less, and the reduction reached approximately 25% at reach 9 (Figure 9). The reduction in metals (e.g., 50% Chromium) can also be translated into toxicity, as shown in Figure 10, illustrating how the water toxicity will change based on the toxicity regression model (EQ.1). The values increasing suggest a more stimulatory nature, which could suggest an environment that might be more stimulatory or beneficial to an organism which may indicate a less toxic condition. The results suggest that serious consideration should be given to the treatment of the tannery effluents so that toxic conditions are reduced downstream of the tanneries.

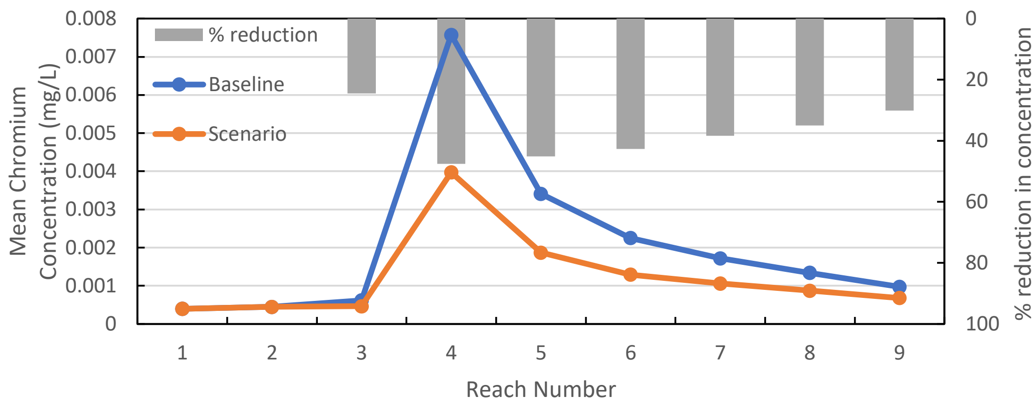


Figure 9. Profile down the main Awash River shows the reductions along the river using the mean Chromium concentrations from the model simulations.

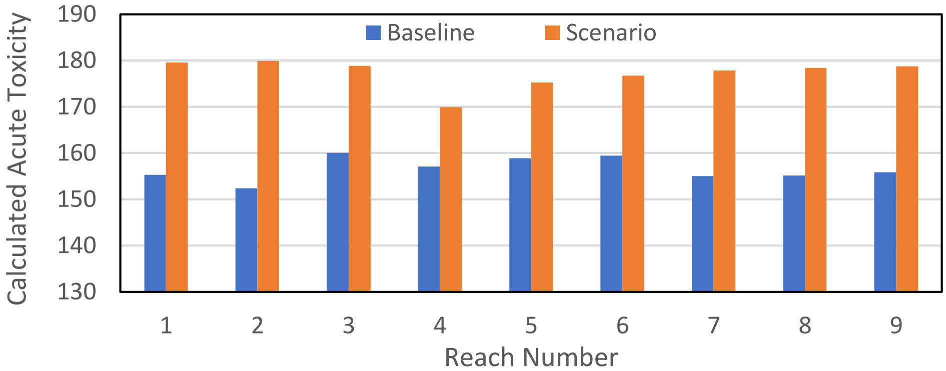


Figure 10. Calculated baseline and scenario river water toxicity along the main Awash River.

4. Conclusions

Water quality degradation due to rapid population growth, urbanization, and industrialization is a global environmental issue. The UN Sustainable Development Goal 6.3 aims to reduce pollution impacts globally and to encourage countries to close the gap between current poor water supplies and the ideal situation, where there are adequate water supplies for people, industry, and agriculture. Globally this gap is about 30% on average. Ethiopia has quite a long way to go to close this gap and, in many ways, the gap is expanding as industrial development continues and populations move towards the cities.

In this paper we consider elevated heavy metal concentrations such as Fe, Al, Cr, Mn, Mo, or As, resulting from untreated industrial effluent discharge into rivers. These are known to pose health concerns and risks to humans, especially where they drain into reservoirs or public water supplies. This is a serious concern for Ethiopia and requires action to put in place a suitable pollution control strategy, as well as ensure that the industry establishes adequate treatment facilities.

In countries like Ethiopia where conventional instruments and new technology are lacking, there have been few studies focusing on evaluating heavy metal levels in Ethiopian's river basins. This is the first comprehensive study that combines field sampling from surface waters and tanneries, chemical laboratory analysis, new biosensor technology, and mathematical modeling to assess the water chemistry and water toxicity in the Awash River. It was found that Lead, Zinc, Arsenic, Copper, and Manganese were highly linked to water toxicity in this river system. The study demonstrated the importance of metal-driven responses to toxicity and the value of a biosensor-based assessment of water quality. It must be noted that it was a small dataset in comparison to the number of metals tested; however, this work serves as a proof of concept for bacterial biosensor toxicity analysis in freshwater systems.

The multi-branch water quality model (INCA) was used to estimate the heavy metal concentrations and to simulate the impact of tannery discharge on the river water pollution levels. It is important to recognize that although most river water samples had heavy metal concentrations below the WHO drinking water standards, wastewater effluents constantly discharge polluted water into water bodies, significantly deteriorating the water quality. Several wastewater treatment plants that incorporate primary treatment with secondary or secondary with tertiary are currently under construction. The new wastewater treatment plants combined with strict implementation of wastewater discharge permit systems and enforcement of the law will improve future Awash River water quality.

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