Mining induced soil and water contamination: exposure assessment and phytoremediation options for a gold mine in Burkina Faso.

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Summary

The gold rush in Burkina Faso has many facets, the most expected one being the economic contribution for the country and local development. Since 2012, gold has become the main exportation product of Burkina Faso, reaching 90% of the total export budget in 2018. On the other hand, mining has many disadvantages such as contamination of land and water, water depletion and health risks linked to the consumption of contaminated food. It is therefore imperative that solutions to environmental problems be found for the "greening" of mining.

The focal point of this thesis is a gold mine in the southern part of Burkina Faso in the Boulgou province, and close to the border with Ghana Republic. The site is in the catchment of the Nakambé River. This study has three main objectives: (1) to assess the contamination of soil and water by mining, and this in terms of potentially toxic elements; (2) to assess the exposure to potentially toxic elements linked to the consumption of contaminated foodstuff cultivated on and nearby the mining site; and (3) to investigate phytoremediation as a possible way to control potentially toxic elements in soil and water.

Regarding the first objective, soil and water samples were taken for analysis. Total digestion and sequential extraction were performed on soil samples taken within the mining site and at nearby, non-affected control sites. The study investigated the concentration of potentially toxic elements, namely arsenic, cadmium, cobalt, chromium, copper, nickel, lead and zinc. Results of analysis of soil samples were interpreted using enrichment factor and geoaccumulation index. Water samples were taken from the pit lakes and the nearby Nakambé river as a control, and the same potentially toxic elements were analyzed. The investigation revealed a high level of As on the site with hotspots around the processing plant and the tailings storage facility. As, Cd, Co, Cr, Ni and Pb were exceeding the agriculture threshold and were therefore of concern. Cu, Zn and Mn were below threshold for agriculture and therefore not a concern. The subsequent calculation of enrichment factors and geoaccumulation indices expressed strong to extreme contamination for As, Co, Cr, Cu, Ni, Pb and Zn. Water analysis revealed that the pit lakes did not meet World Health Organization criteria for drinking water and reflects the impact of the surrounding mining activities. This study highlighted that mining does have an impact on land and water and that the impact is of anthropogenic origin.

For the second objective, four cereal types were sampled in four sites, i.e. the mining site itself and three nearby villages (Songo, Youga and Sighnoguin), and those were analyzed for potentially toxic elements. Also, fish were collected from two pit lakes and from the Nakambé river and analyzed for potentially toxic elements. This was complemented by a dietary intake

survey of the local populations living near the gold mine. The survey revealed a cereals consumption of 0.43 ± 0.19 kg/person/day and a fish consumption of 5.34± 2.60 g/person/day. The Hazard Index (HI) related to consumption of cereals exhibited a value higher than unity in all locations considered, expressing a health concern by the combined toxicity of the elements investigated. The Target Hazard Quotient (THQ) of fish consumption was below unity, as well as the HI. The results revealed that Target Cancer Risk (TR) results for As, Ni and Pb from combined cereals and fish consumption were higher than acceptable, expressing a concern of risk of cancer due to the long-term consumption of cereals and fish.

Finally, for the third objective, soil phytoremediation was considered in a lab experiment using *Leucaena leucocephala* which is a (sub)tropical shrub endogenous to Burkina Faso, and treatment of seepage from the tailing's storage pond was investigated by means of a full-scale constructed wetland populated with *Chrisopogon zizanioides* (vetiver) and *Typha domingensis* (southern cattail). With As being the main element of concern, the capacity of *L. leucocephala* to absorb arsenic has been studied under different conditions: with EDTA supplementation and/or with a growth enhancer containing *Mycorrhiza sp*. and *Bacillus sp*. The results showed an effective uptake of arsenic by *Leucaena leucocephala* while EDTA and the growth enhancer did not cause any additional As uptake by the plant. On the contrary, the treatment with EDTA or growth enhancer reduced the plants' total biomass production. Accumulation of As in the below ground part makes *Leucaena* more suitable for phytosequestration than phytoextraction and allows its subsequent use as forage for livestock. In the constructed wetland, spontaneously grown *T. domingensis* thrived and presented better adaptability than *C. zizanioides*. Due to its high biomass production, standing stocks (amount of potentially toxic elements accumulated in aboveground biomass per area unit) of As, Co, Cr, Cu, Mn, Ni, Pb and Zn were 3, 7, 4, 7, 14, 7, 5, times higher than in *C. zizaniodes*. The outcomes on seepage treatment and the contribution of plant uptake towards treatment were not conclusive, as there was an additional and significant backload from the sediments in the constructed wetland. Nevertheless, the use of *T. domingensis* for gold mine tailing storage seepage treatment element uptake is recommended in combination with uprooting in order to achieve a high level of standing stock removal.

Samenvatting

De goudkoorts in Burkina Faso heeft vele facetten: de meest verwachte is de economische bijdrage voor de natie en de lokale ontwikkeling. Sinds 2012 is goud het belangrijkste exportproduct van Burkina Faso geworden; in 2018 maakte het zelfs 90% uit van het totale exportbudget. Aan de andere kant heeft mijnbouw vele nadelen, zoals verontreiniging van land en water, wateruitputting en gezondheidsrisico's door de consumptie van gecontamineerd voedsel. Het is daarom uitermate belangrijk dat er oplossingen gevonden worden voor deze milieuproblemen om zo de mijnbouw te "vergroenen".

De focus van dit proefschrift ligt op een goudmijn in het zuidelijke deel van Burkina Faso in de provincie Boulgou, en dicht bij de grens met Ghana. De site ligt in het stroomgebied van de Nakambé rivier. Deze studie heeft drie doelstellingen : (1) de verontreiniging van bodem en water door mijnbouw beoordelen op het gebied van potentieel toxische elementen; (2) evalueren van de blootstelling aan potentieel toxische elementen door de consumptie van gecontamineerde levensmiddelen geteeld op en nabij de mijnsite; en (3) fytoremediatie bestuderen als een mogelijkheid om de potentieel toxische elementen in bodem en water te beheersen.

In het kader van de eerste doelstelling werden grond- en watermonsters genomen voor analyse. Een ontsluiting ter bepaling van totaalgehaltes, en sequentiële extractie werden uitgevoerd op bodemmonsters van de mijnsite en van nabijgelegen, niet-aangetaste controleplaatsen. De studie onderzocht de concentratie van potentieel toxische elementen, namelijk arseen, cadmium, kobalt, chroom, koper, nikkel, lood en zink. De resultaten werden verder geïnterpreteerd met behulp van de verrijkingsfactor en de geoaccumulatie-index. Watermonsters werden genomen uit de verlaten mijnputten en de nabijgelegen Nakambé-rivier als controle, en dezelfde potentieel toxische elementen werden geanalyseerd. Uit het onderzoek bleken hoge concentraties aan As in de bodem van de site met hotspots rond de verwerkingsfabriek en de opslagplaats voor residuen. As, Cd, Co, Cr, Ni en Pb overschreden de drempel voor landbouwgebruik en waren daarom zorgwekkend. Cu, Zn en Mn lagen daarentegen onder de drempel voor landbouwgebruik en vormen daarom geen probleem. De daaropvolgende berekening van verrijkingsfactoren en geoaccumulatie-indices bracht sterke tot extreme verontreiniging aan het licht voor As, Co, Cr, Cu, Ni, Pb en Zn. Uit de wateranalyses bleek dat water uit de verlaten mijnputten niet voldeed aan de criteria van de Wereldgezondheidsorganisatie voor drinkwater en de impact van de omliggende mijnbouwactiviteiten weerspiegelde. Deze studie toonde dus aan dat mijnbouw een impact heeft op land en water en dat de impact van antropogene oorsprong is.

Voor de tweede doelstelling werden vier graansoorten bemonsterd op vier locaties, namelijk de mijnsite zelf en drie nabijgelegen dorpen (Songo, Youga en Sighnoguin), en deze werden vervolgens ook geanalyseerd op potentieel toxische elementen. Ook werden vissen verzameld uit twee verlaten mijnputten en uit de Nakambé-rivier en geanalyseerd op potentieel toxische elementen. Dit werd aangevuld met een voedingsonderzoek bij de lokale bevolking die in de buurt van de goudmijn woont. Uit de enquête bleek een consumptie van van 0,43 ± 0,19 kg granen / persoon / dag en een visconsumptie van 5,34 ± 2,60 g / persoon / dag. De *Hazard Index* met betrekking tot de consumptie van granen vertoonde op alle beschouwde locaties een hogere waarde dan één, wat een gezondheidsrisico uitdrukt door de gecombineerde toxiciteit van de onderzochte elementen. De *Target Hazard Quotient* door visconsumptie was lager dan één, evenals de *Hazard Index*. De resultaten toonden aan dat de *Target Cancer Risk* -resultaten voor As, Ni en Pb van gecombineerde granen- en visconsumptie hoger waren dan acceptabel, wat de bezorgdheid over het risico op kanker als gevolg van de langdurige consumptie van granen en vis tot uiting bracht.

Ten slotte werd voor de derde doelstelling fytoremediatie bekeken in een laboratoriumexperiment met *Leucaena leucocephala*, een (sub)tropische struik die inheems is voor Burkina Faso; ook werd de behandeling van afvalwater uit de residubekkens onderzocht door middel van een helofytenfilter begroeid met *Chrisopogon zizanioides* (vetiver) en *Typha domingensis* (zuidelijke lisdodde). Met As als belangrijkste aandachtspunt werd het vermogen van *L. leucocephala* om arseen te absorberen onderzocht, en dit onder verschillende omstandigheden: met EDTA-suppletie en / of met een groeibevorderaar met *Mycorrhiza* sp. en *Bacillus* sp. De resultaten toonden een effectieve opname van arseen door *Leucaena leucocephala*, terwijl EDTA en de groeibevorderaar geen extra As-opname door de plant veroorzaakten. Integendeel, de behandeling met EDTA of groeibevorderaar verminderde de totale biomassaproductie van de planten. Opstapeling van As in het ondergrondse deel maakt *Leucaena* daarom geschikter voor fytosequestratie dan fytoextractie en maakt een eventueel later gebruik als veevoer mogelijk. In de helofytenfilter groeide *T. domingensis* zeer goed en vertoonde een beter aanpassingsvermogen dan *C. zizanioides*. Vanwege de hoge biomassaproductie waren de hoeveelheden potentieel toxische elementen in de bovengrondse biomassa per oppervlakte-eenheid bij *T. domingensis* 3, 7, 4, 7, 14, 7, 5 keer hoger dan bij *C. zizaniodes* voor respectievelijk As, Co, Cr, Cu, Mn, Ni, Pb en Zn. De uitkomsten op het gebied van afvalwaterbehandeling en de bijdrage van plantopname aan de behandeling waren niet overtuigend, aangezien er een bijkomende en significante nalevering was vanuit de sedimenten. Desalniettemin wordt het gebruik van *T. domingensis* voor de verwijdering van potentieel toxische elementen uit afvalwater afkomstig van de residubekkens mits regelmatig maaien en afvoeren van de bovengrondse biomassa.

Résumé

La ruée vers l'or au Burkina Faso a de multiples facettes, la plus attendue étant la contribution économique du pays et le développement local. Depuis 2012, l'or est devenu le principal produit d'exportation du Burkina Faso, atteignant 90% du budget d'exportation total en 2018. En revanche, l'exploitation minière présente de nombreux inconvénients tels que la contamination des terres et de l'eau, l'appauvrissement en eau et les risques sanitaires liés à la consommation d'aliments contaminés. Il est donc impératif de trouver des solutions aux problèmes environnementaux pour le "verdissement" de l'exploitation minière.

Le point focal de cette thèse est une mine d'or dans la partie sud du Burkina Faso dans la province de Boulgou, et près de la frontière avec la République du Ghana. Le site se trouve dans le bassin versant de la rivière Nakambé. Cette étude a trois objectifs principaux: (1) évaluer la contamination du sol et de l'eau par l'exploitation minière, et ce en termes d'éléments potentiellement toxiques; (2) évaluer l'exposition à des éléments potentiellement toxiques liés à la consommation d'aliments contaminés cultivés sur et à proximité du site minier; et (3) d'étudier la phytoremédiation comme moyen possible de contrôler les éléments potentiellement toxiques dans le sol et l'eau.

Concernant le premier objectif, des échantillons de sol et d'eau ont été prélevés pour analyse. La digestion totale et l'extraction séquentielle ont été effectuées sur des échantillons de sol prélevés sur le site minier et sur des sites témoins non affectés à proximité. L'étude a porté sur la concentration d'éléments potentiellement toxiques, à savoir l'arsenic, le cadmium, le cobalt, le chrome, le cuivre, le nickel, le plomb et le zinc. Les résultats de l'analyse des échantillons de sol ont été interprétés à l'aide du facteur d'enrichissement et de l'indice de géoaccumulation. Des échantillons d'eau ont été prélevés dans les fosses transformées en retenue d'eau et à la rivière Nakambé à proximité comme contrôle, et les mêmes éléments potentiellement toxiques ont été analysés. L'enquête a révélé un niveau élevé d'As sur le site avec des points à forte concentration autour de l'usine de traitement et du parc à résidus. L'As, le Cd, le Co, le Cr, le Ni et le Pb dépassaient le seuil agricole et étaient donc préoccupants. Le Cu, le Zn et le Mn étaient inférieurs au seuil pour l'agriculture et n'étaient donc pas préoccupants. Le calcul ultérieur des facteurs d'enrichissement et des indices de géoaccumulation a exprimé une contamination forte à extrême pour As, Co, Cr, Cu, Ni, Pb et Zn. L'analyse de l'eau a révélé que les fosses transformées en reservoir d'eau ne répondaient pas aux critères de l'Organisation mondiale de la santé pour l'eau potable et reflètent l'impact des activités minières environnantes. Cette étude a souligné que l'exploitation minière a un impact sur la terre et l'eau et que l'impact est d'origine anthropique.

Pour le deuxième objectif, quatre types de céréales ont été échantillonnés sur quatre sites, à savoir le site minier lui-même et trois villages voisins (Songo, Youga et Sighnoguin), et ceux-ci ont été analysés pour les éléments potentiellement toxiques. De plus, des poissons ont été recueillis dans deux fosses transformées en retenue d'eau et dans la rivière Nakambé et analysés pour détecter des éléments potentiellement toxiques. Cela a été complété par une enquête sur l'apport alimentaire des populations locales vivant près de la mine d'or. L'enquête a révélé une consommation de céréales de 0,43 ± 0,19 kg/personne/jour et une consommation de poisson de 5,34 ± 2,60 g / personne / jour. L'indice de danger (HI) lié à la consommation de céréales a montré une valeur supérieure à l'unité dans tous les sites considérés, exprimant un problème de santé par la toxicité combinée des éléments étudiés. Le quotient de danger cible (THQ) de la consommation de poisson était inférieur à l'unité, ainsi que le HI. Les résultats ont révélé que les résultats du risque de cancer (TR) pour As, Ni et Pb provenant de la consommation combinée de céréales et de poisson étaient plus élevés qu'acceptables, exprimant une préoccupation quant au risque de cancer en raison de la consommation à long terme de céréales et de poisson.

Enfin, pour le troisième objectif, la phytoremédiation du sol a été envisagée dans une expérience de laboratoire utilisant le *Leucaena leucocephala* qui est un arbuste (sub) tropical endogène au Burkina Faso, et le traitement des infiltrations du parc à résidus a été étudié au moyen d'une construction à grande échelle d'une zone humide peuplée de *Chrisopogon zizanioides* (vétiver) et de *Typha domingensis* (quenouille du Sud). L'arsenic étant le principal sujet de préoccupation, la capacité du *L. leucocephala* à absorber l'arsenic a été étudiée dans différentes conditions: avec supplémentation en EDTA et/ou avec un activateur de croissance contenant *Mycorrhiza* sp. et *Bacillus* sp. Les résultats ont montré une absorption efficace d'arsenic par le *Leucaena leucocephala* tandis que l'EDTA et le stimulateur de croissance n'ont pas provoqué une absorption supplémentaire d'As par la plante. Au contraire, le traitement avec de l'EDTA ou un activateur de croissance a réduit la production totale de biomasse des plantes. L'accumulation de As dans la partie inferieure rend le *Leucaena* plus adaptée à la phytosequestration qu'à la phytoextraction et permet son utilisation ultérieure comme fourrage pour le bétail. Dans la zone humide construite, le *T. domingensis* qui s'est spontannement developpé, a prospéré et a présenté une meilleure adaptabilité que le *C. zizanioides*. En raison de sa production élevée de biomasse, les stocks sur pied (quantité d'éléments potentiellement toxiques accumulés dans la biomasse aérienne par unité de surface) d'As, Co, Cr, Cu, Mn, Ni, Pb et Zn étaient de 3, 7, 4, 7, 14, 7, 5 fois plus élevé que chez le *C. zizaniodes*. Les résultats sur le traitement des infiltrations et la contribution de l'assimilation des plantes au traitement n'étaient pas concluants, car il y avait une charge supplémentaire et importante des sédiments dans la zone humide construite. Néanmoins, l'utilisation de *T. domingensis* pour l'absorption des éléments de traitement de l'infiltration du parc à résidus miniers d'or est recommandée en combinaison avec le déracinement afin d'atteindre un niveau élevé d'enlèvement de matière sur pied.

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List of Acronyms

ADI:	Acceptable daily intake
AHC:	Agglomerative hierarchical clustering
AMDC :	African Minerals Development Centre
ANOVA:	Analysis of variance
ANZFS:	Australia New Zealand Food Standards
BAC:	Biological accumulation coefficient
BCF:	Biological concentration factor
BCR:	Community Bureau of Reference
CEC:	Cation exchange capacity
CFA:	Communauté Financière africaine
CIL:	Carbon-in-leach
CSQG:	Canadian water quality guidelines
DO:	Dissolved oxygen
EDI:	Estimated daily intake
EDTA:	Ethylenediaminetetraacetic acid
EF:	Enrichment factor
EFSA:	European Food Safety Authority
ELAW:	Environmental Law Alliance Worldwide
FAO:	Food and Agriculture Organization of the United Nations
GE:	Growth enhancer
HI:	Hazard Index
HRT:	Hydraulic residence time
IFC:	International Finance Corporation
Igeo:	Geoaccumulation Index
INSD:	Institut National de la statistique et demographie
ICP-OES :	Inductively Coupled Plasma - Optical Emission Spectrometry
ISO:	International Standard Organisation
MDL:	Method detection limit
MME:	Ministère des mines et de l’energie
PTE:	Potentially toxic element
NF:	Norme Francaise/French standard
NGOs:	Non-governmental organization
NR:	Nakambé River
ORP:	Oxidation-reduction potential
PCA:	Principal component analysis

PNDES:	National Economic and Social Development Plan
PWTI:	Provisional weekly tolerable intake
RfD:	reference dose
Rompad:	Run of mining pad
SAWQG:	South African water quality guidelines
SCADD:	Strategy for Accelerated Growth and Sustainable Development program
TDS:	Total dissolved solids
TF:	Transfer factor
THQ:	Target hazard quotient
TOM:	Total Organic matter
TSF:	Tailing storage facilities
Tukey HSD:	Tukey Honest Significant Difference
USEPA:	United states Environmental Protection Agency
UTM:	Universal Transverse Mercator
WGS 84:	World Geodetic System 1984
WHO:	World Health Organisation

Section 1

Chapter 1: Introduction, Objectives and Outline

1.1. Introduction

Africa is considered to be the ‘granary of the mineral world’, with numerous commodities produced (Africa Union, 2009; Dialga, 2018). With a huge reserve of 40% of total gold deposits and an increase in the rate of extraction of world production, the continent is keen to continue providing minerals to the world for the foreseeable future. The economic impact of extraction is interesting. On the one hand, it provides financial assets to power the continent’s development needs (Pokorny et al., 2019). On the other hand, criticism on the impact of mineral extraction in Africa has been on the rise, starting with NGOs, and international and regional institutions (Africa Union, 2009; AMDC, 2014).

At the country level, the movement against mineral extraction is also on the rise.

To deal with these issues, the continent, in the form of the African Union, unified its vision for mining. In February 2009, the Union issued a document entitled “Africa Mining Vision,” with the clear objective of achieving a “transparent, equitable and optimal exploitation of mineral resources to underpin broad-based sustainable growth and socio-economic development,” and ensuring “a sustainable and well-governed mining sector that effectively garners and deploys resource rents, and that is safe, healthy, gender and ethnically inclusive, environmentally friendly, socially responsible and appreciated by surrounding communities” (Africa Union, 2009). Africa’s mineral potential, if used appropriately, will benefit the continent and power its development (Mainguy, 2011). Thus, strategies to accentuate the positive aspects and techniques to reduce any negative aspects are to be put in place, such as increasing the income for the local population and government and reducing the environmental impact through environmentally friendly activities and original land reclamation (Andriamasinoro & Angel, 2012; Luning, 2014).

Burkina Faso, a landlocked country located in the center of West Africa with 18 million inhabitants (INSD, 2015), joined the exclusive club of mine producers a few years ago. Burkina Faso is experiencing a boom in the mining industry, with a suitable economic contribution towards the African Union’s ambitious plan which should advance the country’s development. In 2003, Burkina Faso issued an incentive mining code to attract investors. The process of getting approval for exploitation was easy (Ouoba, 2017). These regulations favored a company’s asset management, offered free tax for mining equipment, and eased in the labor and mining code. This all contributed to making the mining boom a reality, with industrial mining settling in all corners of the country; from south to north, and east to west.

The ease of the process accelerated the entry of financial resources but, unfortunately, led to shortcuts being taken in some essential aspects. The country is classified as the third highest gold producer in West Africa, and the fourth in Africa behind South Africa, Ghana and Mali (Chambres des mines, 2018). According to mineral exploration specialists, Burkina Faso is committed to be a leading world mineral provider if explorations continue (Metelka et al., 2011; Béziat et al., 2008). The whole landscape of Burkina Faso is changing as a result of both gold mining and the mining of other minerals (Table 1-1). Mining is spread all over the country with eight gold mines in the exploitation stage in 2017, ten in exploitation stage in 2019 and many more in advanced exploration and construction stages (Béziat et al., 2008; Tshibubudze & Hein, 2016; Augustin et al., 2016; Hein, 2016b; Hein & Tshibubudze, 2016; Ouiya et al., 2016; McCuaig et al., 2016; Ilboudo et al., 2018; Woodman, et al., 2016).

Table 1-1 Extractive industries of Burkina Faso, source: Chambre des mines du Burkina, accessed December 27th 2019

Group	**Exploitation Company**	**Ore**	**Observations**
Amara Mining	Kalsaka Mining SA	Au	Closure stage
Amara Mining	Seguenega Mining SA	Au	Closure stage
Orezone Inc	Orezone Bombore SA	Au	Approved
B2Gold	Kiaka Gold SA	Au	Approved
West African Ressources	Société des mines de sabrado	Au	Approved
Endeavour Mining	Bouere douhoun SA	Au	Approved
NordGold	NordGold Yeou SA	Au	Approved
CENTAMIN	Konkera SA	Au	Approved
Pan African Burkina	Pan African Tambao SA	Mn	Interrupted
Metal Mass	Burkina Manganèse SARL	Mn	Exploitation
NordGold	Sociétés des Mines de Taparko (SOMITA - SA)	Au	Exploitation
Avesoro	Burkina Mining Company (BMC) SA	Au	Exploitation
Avocet Mining	Société des mines de bélahouro (SMB) SA	Au	Interrupted
SEMAFO	Mana SEMAFO SA	Au	Exploitation
IAMGOLD	Iamgold Essakane SA	Au	Exploitation
NordGold	Bissa Gold SA	Au	Exploitation
Glencore International	Nantou Mining	Zn	Exploitation
KOMET Resources	KOMET Resources	Au	Exploitation
Endeavour	Hounde Gold Operations SA	Au	Exploitation
Teranga Gold	Société Minière Gryphon	Au	Construction
SEMAFO	Boungou SEMAFO SA	Au	Construction

The production of gold in Burkina Faso rose from 12.2 tonnes in 2009, to 24 tonnes in 2010 and 32.6 tonnes in 2011, 30.2 tonnes in 2012, and 36.6 tonnes in 2014 to 53 tonnes in 2018 (Figure 1-1). Since 2009, gold has become the main provider of funds for the country, along with cotton and livestock (Werthmann, 2009; Ouoba, 2017).

The financial contribution of the sector to the country's financial assets is very appreciable. The mining sector became the main income provider (Figure 1-2), more than cotton and agricultural products, and is expected to contribute to lifting the country from developing to emerging status, according to the Strategy for Accelerated Growth and Sustainable Development program (SCADD, 2010) and the National Economic and Social Development Plan (PNDES, 2015) set by the Government. Gold exportation in 2015 and 2016 was accounted for 908 and 1023 Billions Francs CFA, respectively (Chambre des Mines du Burkina, 2019).

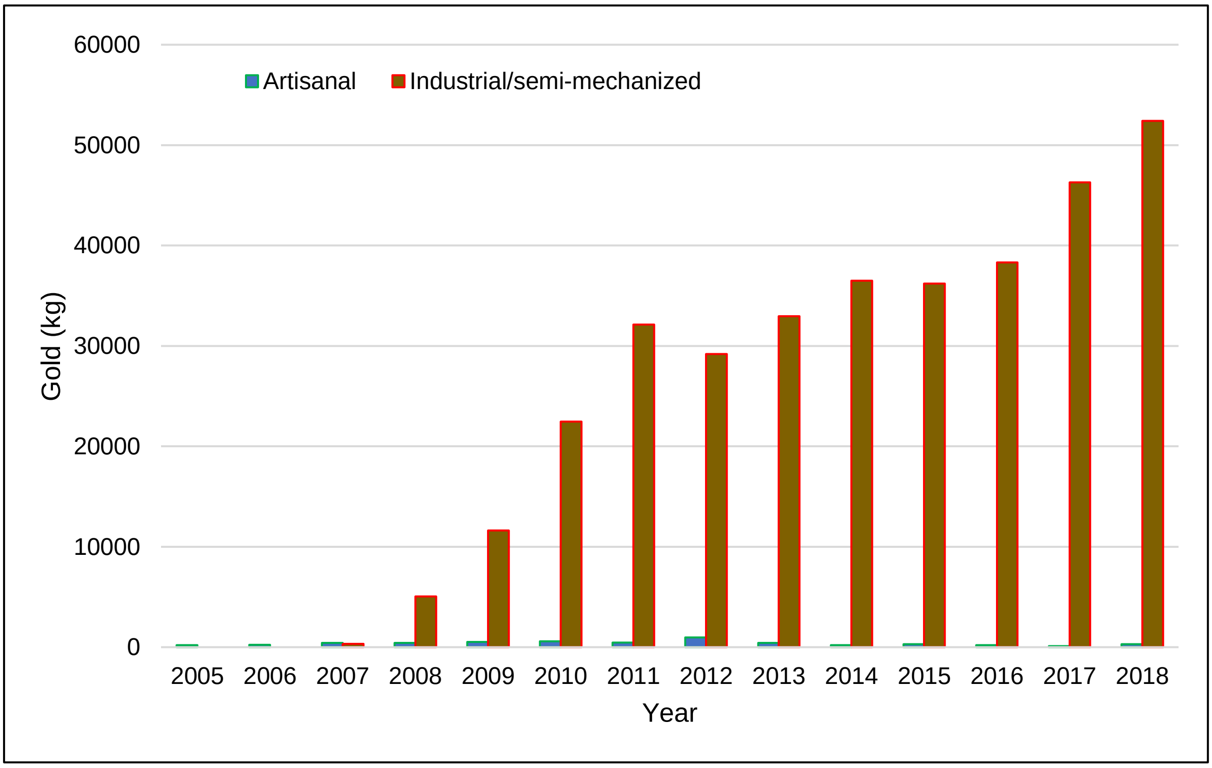

Figure 1-1 Evolution of artisanal and industrial gold production in Burkina Faso, source: INSD, 2018

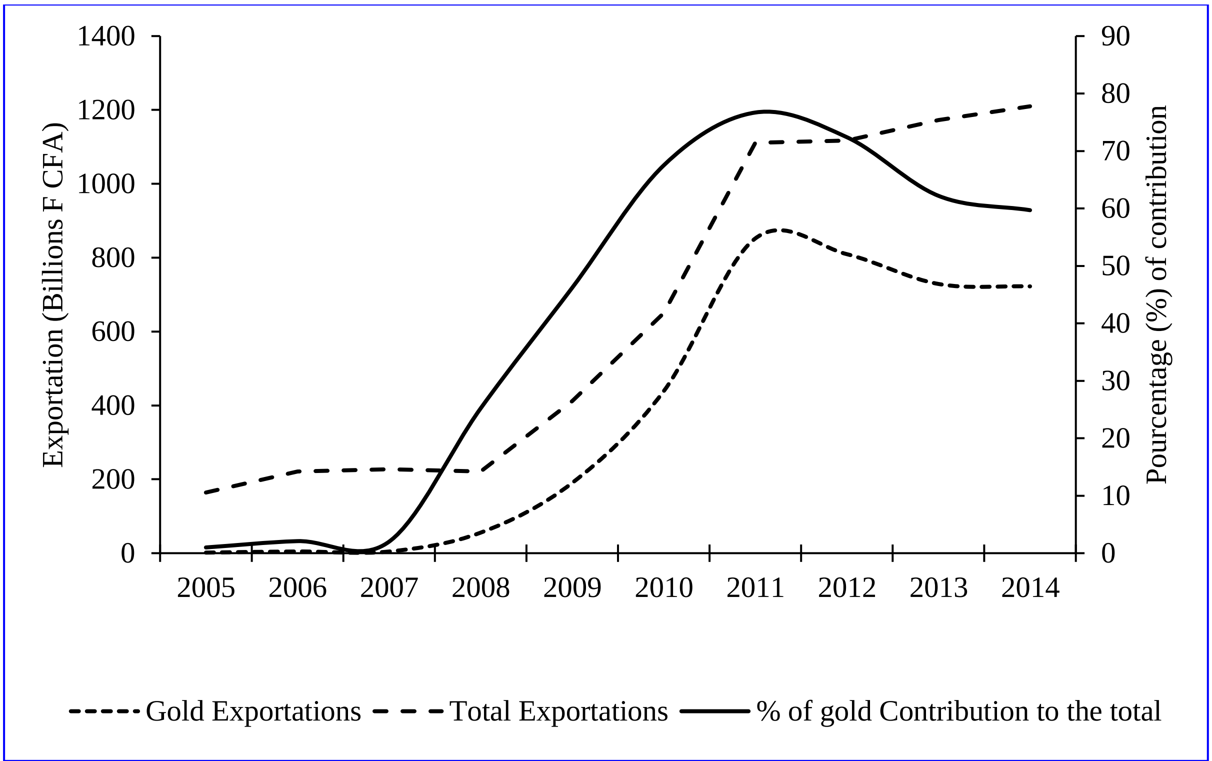

Figure 1-2 Gold contribution to the exportation (Chambre des mines, 2016)

The opening of a mine is a very important event for all stakeholders. From the local community to the national government, the mine is a huge opportunity and, during the opening stage, mining companies showed a strong commitment to fulfill local communities' requests and regulation requirements. However, the closure stage is entirely the opposite (Kitula, 2006; Bainton & Holcombe, 2018). Broadly interpreted, ‘‘closure’’ requires leaving viable ecosystems on mining lands compatible with a healthy environment and human activities, that are of low hazard and which encompass measures to prevent ongoing pollution from the site in the long-term (Peck & Sinding, 2009).

Burkina Faso, through its Mining Sectorial Policy 2013-2025 (MME, 2013) is willing to use its mineral potential to boost the country's economy and move towards sustainable development by reducing the drawbacks of mining and ensuring a better, balanced development for all citizens. To achieve such objectives, a proper environmental management policy should be enforced in the mining sector, in order to reduce the sector's impact during all stages of mining, and particularly in the reclamation of the land after the mine's closure.

1.2. Problem Statement

It is clear that mining has positive financial impacts for a developing country (AMDC, 2014; African Union, 2009; Kumah, 2006; Ouoba, 2017) in the context of economic crises, but also has negative impacts (Andriamasinoro & Angel, 2012; Brewer et al., 2012; Dialga, 2018). During all stages of mining, the chemicals used, polymers for drilling, blasting agents, gold lixiviation chemicals (cyanide), and laboratory analysis reagents, could potentially reach the soil or water bodies and threaten local communities through the food chain by uptake through crop growth near contaminated sites (Liu et al., 2005; Wu et al., 2010; Kinimo et al., 2018).

Use of open-cut mine pits as water reservoirs has been considered (de Lange et al., 2018; Sondergaard et al., 2018; Mollema et al., 2015); however, pit lakes used by the local population, instead of being a help, could jeopardize the lives of local people when contaminated, especially when they use these pit lakes for agricultural purpose or for fulfilling daily basic needs. The blasting, excavating and moving of ore and waste reveal deeply buried contaminant-containing rocks, which could be mobilized by wind and water to water bodies, soil, and the surrounding environment. Handing over anthropogenic-impacted environments to the local population for agricultural purpose without proper prior environmental remediation inevitably leads to the growing of a contaminated crop (Zhou et al., 2018) and an increase in short-, middle- and long-term health issues in the local population.

Risk assessment to identify the hazards of a mining environment must be carried out. Solutions to the hazards should be identified and sound corrective actions implemented (de Lange et al., 2018) to fix the issues, taking into account local parameters such as climate conditions, local population expectations, availability of remediation materials, affordability, ease of maintenance, and other uses of the environment.

In 2001, Burkina Faso experienced the sudden closure of the Poura mine, which led to a significant upheaval in the community and local government, and for all other stakeholders. The after-closure environment management has been marked by many deficiencies. Unfortunately, the excitement that surrounds the opening of a new mine is never present when it finally closes (Bainton & Holcombe, 2018).

Mining site reclamation now forms part of the environmental impact study prior to getting government go-ahead for any mine exploitation in Burkina Faso (Mining Code, 2015). Mining reclamation has been undertaken in many countries worldwide, with less or acceptable success. Reclamation is highly encouraged, and could prove to be beneficial for another country which, like Burkina Faso, has just entered in the exclusive club of mining countries like Mali (Traore, 2016), Gambia (Niane et al., 2014) and Niger (Dan-Badjo et al., 2019). Pioneering studies are needed.

1.3. Research Questions and Thesis Structure

This research study investigates contamination of a mining site in Burkina Faso and provides an exposure assessment of foodstuffs impacted by the mining to evaluate local community exposure. A solution through exploring the potential of naturally growing plant phytoremediation on gold mining impacted site remediation was investigated. The focus in this thesis is on As, Cd, Co, Cr, Cu, Fe, Ni, Mn, Mg, Pb and Zn designated as Potentially Toxic Elements (PTE), a term recommended by Pourret & Bollinger, 2018; Pourret & Hursthouse, 2019, after the term "Heavy metals" was described by Duffus, 2002 as a meaningless term.

The site under scrutiny is a mid size gold mining site located in the southeastern side of Burkina Faso. The gold production started in 2008 after a year and a half of construction. The Youga deposits are characterized by two distinct styles of mineralization: moderately to weakly silicified host rock with quartz stockwork veining and pyrite as the predominant sulfide; and the intensely silicified arkose, with abundant quartz veins and more diverse sulfides. The site is located between the watershed of Nakamber and Nazino with annual rainfall ranging from 900 mm to 1200 mm. Local communities are farmers with livestock breeding.

The study objectives were (Figure 1-3):

- To assess the impact of the gold mining on the soil potentially toxic element contaminations and to identify hotspots. This involved both total potentially toxic element contaminations and sequential extraction to identify repartition of contaminant in the different fractions in order to predict the mobility and bioavailability.
- To evaluate the water quality of open-pit mining transformed to pit lakes and to assess the potential use of those pit lakes by the local community. Potentially toxic elements and field physicochemical parameters were measured and benchmarked against selected national guidelines and the nearest river water quality parameters.
- To measure the quality of the crops (crop potentially toxic element content) grown in the mining site and in the vicinity to check the impact of gold mining on crop quality and intake risk for the local population.
- To estimate the quality of common locally available fish from the pit lakes and estimate the intake risks for the local population. Potentially toxic elements were analyzed and benchmarked against fish from the Nakambé river and international guidelines.

Further objectives concerned remediation options for the contaminated land.

- *Leucaena leucocephala* was used in laboratory scale experiments with mining soil, spiked with different concentrations of arsenic, and with supplementation of EDTA and growth enhancer to evaluate its capability to uptake arsenic from soil.
- A field-scale constructed wetland populated by spontaneously grown *Typha* and introduced *Chrisopogon* were tested to assess their uptake capacity for potentially toxic elements from gold mine tailing storage facilities seepage.

The present study is structured in three sections and eight chapters. The first section contains two chapters: the introduction, and the literature review. The second section contains five chapters reporting experimental results. The third section contains one chapter; focused on the interpretation of the study findings and giving some future perspectives as well.

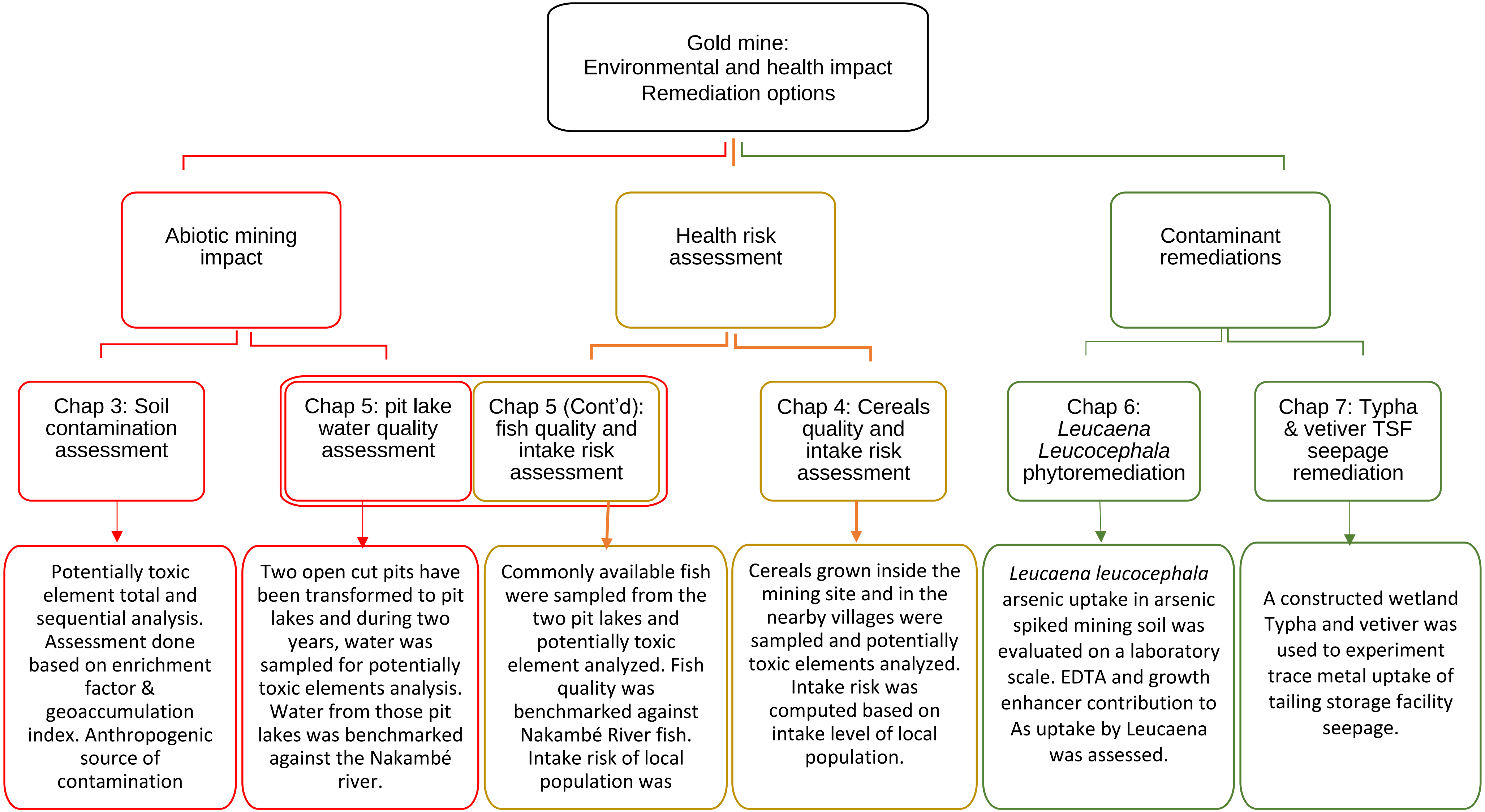

Figure 1-3: Study objectives diagramme and chapters organization

The chapters in the second section are as follows:

Chapter 3: Mining activities impact on soil. Mining impact can be localized in hotspots within the perimeters, or contaminants can be mobilized in the vicinity of the perimeters. This chapter characterizes mining-induced potentially toxic elements contamination by comparing recent analysis results to background values. Enrichment factor (EF) and Geoaccumulation Index (Igeo) were both used to evaluate the extent of contamination. Further comparison with selected national standards was used to check the potential use of the soil.

Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (2019a). Gold mine impact on soil quality, Youga, southern Burkina Faso, West Africa. *Water, air and soil pollution*, 230:207. http://dx.doi.org/10.1007/s11270-019-4257-z.

Chapter 4: Cereals grown in soil impacted by mining activities can accumulate potentially toxic elements and jeopardize local community health. Commonly-grown cereals were sampled and potentially toxic elements were analyzed. Based on the results, referential doses and a survey on intake rates, risk assessments were carried out to measure exposure.

Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (2019b). Trace element content in cereals from a gold mining site in Burkina Faso and intake risk assessment. *Journal of Environmental Management*, 248, 109292. http://dx.doi.org/10.1016/j.jenvman.2019.109292.

Chapter 5: Open-pit mining ends with pits which can be used for many purposes. One purpose is filling the pit with water, creating a pit lake, and using it for livestock watering, recreational use, a fishing pool, swimming pool, market gardening, and domestic use. This chapter measured the water quality on a regular basis over a period of two years. Fish from the pit lakes were sampled and analyzed for potentially toxic elements. In respect of water quality, the fish sampling results were compared to the results of a nearby water body, the Nakambé River. Exposure of the local population was estimated based on the intake rate obtained through surveys and referential doses from regulatory institutes.

Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (2019c). Metals and metalloid in gold mine pit lakes and fish intake risk assessment, Burkina Faso. *Environmental Geochemistry and Health*, pp 1-15, http://dx.doi.org/10.1007/s10653-019-00390-8.

Chapter 6: This chapter describes the evaluation of arsenic (As) uptake by *L. leucocephala* in a 45-day laboratory experiment with gold mine soil from Burkina Faso. A soil spiked with 25 and 50 mg As/kg, obtained from arsenic trioxide (As_2O_3), stabilized for 45 days and mixed with compost and supplemented with growth enhancer (GE) or ethylenediaminetetraacetic acid (EDTA) or both, was used. The objectives were to evaluate the contribution of EDTA and growth enhancer containing mycorrhiza to *Leucaena leucocephala* As uptake capability.

Chapter 7: This chapter reports an experiment analyzing the capability of two plant species to take up potentially toxic elements from gold mine tailing storage facility seepage in a constructed wetland with horizontal subsurface flow. A naturally populating *Typha domingensis* and introduced *Chrisopogon zizaniodes* were sampled on a regular basis in two experimentation cycles: *Typha* only in the first cycle, and *Typha* plus *Chrisopogon* in the second. Corresponding water samples were taken from the inflow and outflow of the system. Potentially toxic elements were analyzed in both sets of samples.

Chapter 2: Literature review

2.1. Mining and its Impact

Generally, the gold mining industry, despite providing substantial economic benefits (Gajigo et al., 2012), has a negative image because it is potentially high-polluting, plus the fact that it is not renewable and its costs are often externalized on the local communities that host its operations (Kumah, 2006). Mining has several common stages or activities: exploration, construction, exploitation, and closure; each of which has potentially adverse impacts on the natural environment, society, and cultural heritage, on the health and safety of mine workers, and on local communities. Local community impact includes the displacement of local people from ancestral lands, marginalization, an increase in living standards beyond local community capability, and the oppression of people belonging to lower economic classes (Kitula, 2006).

2.1.1. Socioeconomic Impact

Direct social challenges are primary, such as displacement of communities and disruption of livelihoods, social conflict (Engels, 2018), post-mine closure issues, community and worker health, safety, and pay. Other major challenges in the sector include relations between mining companies, the government, and mining communities (AMDC, 2014; Bainton & Holcombe, 2018; Luning, 2012; Hilson et al., 2019). Mine-located zones witness an increase in the poverty gap, according to a study undertaken by Zabsonré et al. (2018). The same study also revealed an increase in child labor in zones hosting mining industries, compared to zones without mining activities. Migration can also be included in mining socioeconomic impact. Additionally, mining is influencing gender: firstly by migration of men to the mining site leaving women behind in their village; secondly by an increase in payment difference as mining is seen as male activity which leads to an increase in wealth unbalance and inequality in economic opportunities between male and female (Kotsadam & Tolonen, 2016). Female workforce in mining sector worldwide, according to the International Finance Corporation, represented 14% of the total workforce (WBG, 2018).

The contribution of mining to a country's economy has been reported as a positive impact, despite Ouoba's (2017) study revealing that 40% of mining added-value represented natural capital depreciation and highlighting the government's inefficiency in spending mining revenue. On the other hand, improvement in basic and social infrastructure and services built as part of mining companies' social responsibility actions have been witnessed in zones hosting mining activities (Garvin et al., 2009). Employment by mining companies is also important (Figure 2-1). Pokorny et al. (2019) assessed the impact of mining on rural livelihoods in Northern Burkina Faso, and through surveys across six districts representing different mining schemes (industrial, artisanal, no mining), established that artisanal gold mining can actually generate job opportunities and cash income for local households, whereas industrial gold mining widely fails to do so. In 2014, 640,800 of Burkinabè (Burkina Faso citizens), representing nearly 4% of the total population of the country, were directly involved in artisanal mining activities. Multiplying each artisanal miner by five dependents, makes artisanal mining central to the livelihood of 3,200,000 of Burkinabè, corresponding to 18% of the country's population (Bazillier & Girard, 2018). However, a drawback of artisanal mining is the presence of clandestine activities, which takes a very high priority (Andriamasinoro & Angel, 2012; Hein & Funyufunyu, 2014).

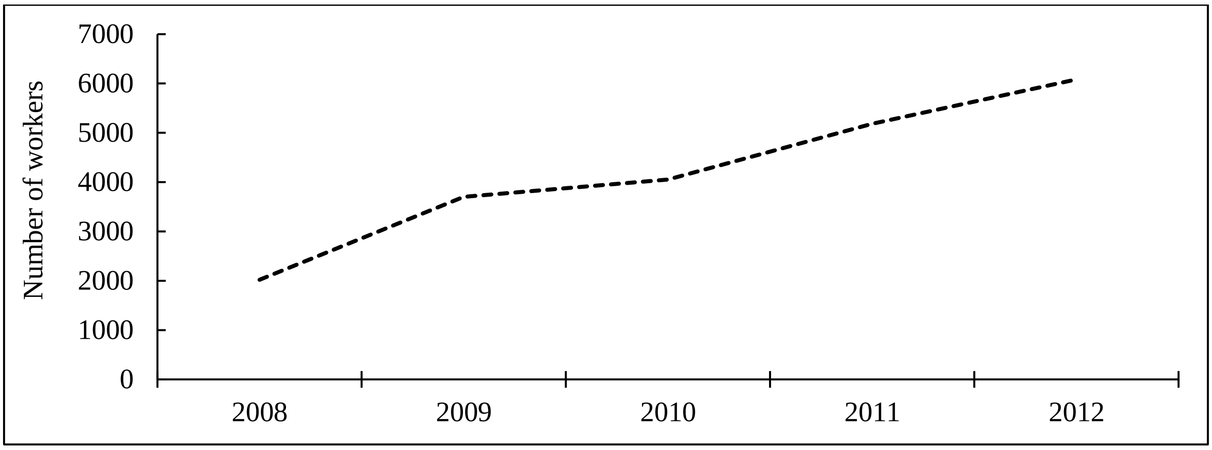

Figure 2-1: Employment in industrial mining sector (Chambre des mines du Burkina, 2016)

2.1.2. Environmental Impact

There are different phases in a mining project, beginning with mineral ore exploration and ending with the post-closure period. Each mining phase is associated with different sets of environmental impacts (ELAW, 2010) (Table 2-1). Mining causes environmental impacts from visual intrusion, dust, noise, blasting, traffic, and pollution of water bodies and groundwater by both industrial and artisanal mining (Andriamasinoro & Angel, 2012; von der Goltz & Barnwal, 2019). Mining causes environmental impacts from visual intrusion, dust, noise, blasting, traffic, and pollution of water bodies and groundwater by both industrial and artisanal mining (Andriamasinoro & Angel, 2012; von der Goltz & Barnwal, 2019). This results among others in cyanide pollution (Griffiths et al., 2009), land degradation, and potentially toxic element contamination (Sousa et al., 2011) (Table 2-1). In addition, Owusu et al. (2018), investigating the secondary impact of mining on primates and other medium to large mammals in forest reserves in southwestern Ghana, revealed that mammal diversity in the mining area had declined due to noise from mining operations and hunting.

Table 2-1 Mining activities with a potential environment incidence, adapted from Sousa et al. (2011)

Soil physical alteration	Flora alteration
Soil chemical alteration	Fauna alteration
Soil microbiological alteration	Depletion of renewable resources
Water contamination (chemical/biological)	Noise emission
Water siltation	Human health alteration
Water, soil and fauna alteration	Greenhouse effect
Air quality alteration	Abandoned pit

Deforestation is one of the main visible impacts of mining activities on the environment. The stripping of land for pits, waste dumps, camps, buildings, processing plants, fuel storage tanks, workshops, and roads mostly in pristine lands, is very common and will impact the landscape for a very long time unless adequate and acceptable reclamation procedures are put in place.

In Burkina Faso, around 1244.43 km^2 of land (based on permits issued by the government) was used for industrial mining in the exploitation stage in 2016 (Figure 2-2), and a further 330 km^2 in the following two years. These areas should all be reclaimed by mining industries before they are handed back to their owners. It is anticipated that 0.54% (around 1500 km^2) of the country's land will need to be rehabilitated in the coming years.

Perhaps the most significant impact of a mining project is its effect on water and soil quality, and on availability of water resources within the project area. Key questions are whether surface and groundwater supplies, and soils, will remain fit for human consumption and use, and whether water and soil quality in the project area will remain adequate to support native aquatic life and terrestrial wildlife (ELAW, 2010), and whether water availability will be granted to the local community.

Land is considered contaminated when it contains hazardous materials above background or naturally occurring levels (IFC, 2007). Contaminated land may involve surficial soils or subsurface soils that, through leaching and transport, may affect groundwater, surface water, and adjacent sites. Contamination comes from chemicals used for blasting and mineral processing, as well as compounds contained in the dust, fuel spillages, washing of processing pads, etc. According to the ore being processed, the contaminants are potentially toxic elements represented such as arsenic, cadmium, lead, or zinc. Water pollutants of concern include potentially toxic elements, which occur in low concentrations in natural aquatic ecosystems (Brewer et al., 2012). They occur in the earth's crust but are enriched in soil mainly by anthropogenic actions such as mining, wastewater discharges, or coal burning.

One of mining's environmental challenges with an immediately visible impact is certainly waste. Mining produces a huge quantity of waste which, without an adequate management system, could significantly impact the environment. Waste is produced in many ways in mining, and toxicity is therefore linked to the waste's origin, and its processing pathway. In general, mining produces solid waste as burden rock stored in waste dumps. There is also waste from camps and laboratory slags. Liquid waste consists mainly of the tailings of processing, water pumped from pits, laboratory liquid waste, and toilet wastewater from camps and plant facilities. Air pollutants comes from moving-equipment engine exhaust systems and power generators.

The major environmental impact of waste disposal at mine sites can be divided into two categories: loss of productive land following its conversion to waste storage area, and the introduction of sediment, acidity, and other contaminants into the surrounding surface and groundwater, from water running over exposed problematic or chemically reactive wastes. Mining processing typically uses sodium cyanide, calcium hydroxide, sodium hydroxide and hydrochloric acid.

Potentially toxic element contamination continues after mining ceases, through the leaching of waste rocks, and tailings storage (Doe et al., 2017) if appropriate actions are not put in place to mitigate contamination. These potentially toxic elements are reaching the food chain and threatening human health (Álvarez-Ayuso et al., 2016; Mileusnić et al., 2014). The most toxic forms of potentially toxic elements in their ionic species are the most stable oxidation states, e.g., Cd^{2+}, Pb^{2+}, Hg^{2+}, in which they react with the body's biomolecules to form extremely stable biotoxic compounds which are difficult to dissociate (Hashim et al., 2011). The most toxic arsenic species are inorganic ones: Arsine, followed by As (III), then As (V) and organic As. Arsine, arsenate and arsenite are inorganic compounds (Hashim et al., 2011).

The processes of mineral extraction, processing, smelting and refining can never approximate to become environmentally neutral; however, the areas of impact can be ameliorated, sometimes to a major degree, by long-term monitoring; from the initiation of a project to the phases of a restored or remediated mine by taking appropriate action (Kwolek, 1999). To respond to these increasing environmental concerns, mining and mineral companies should adopt good environmental management practices.

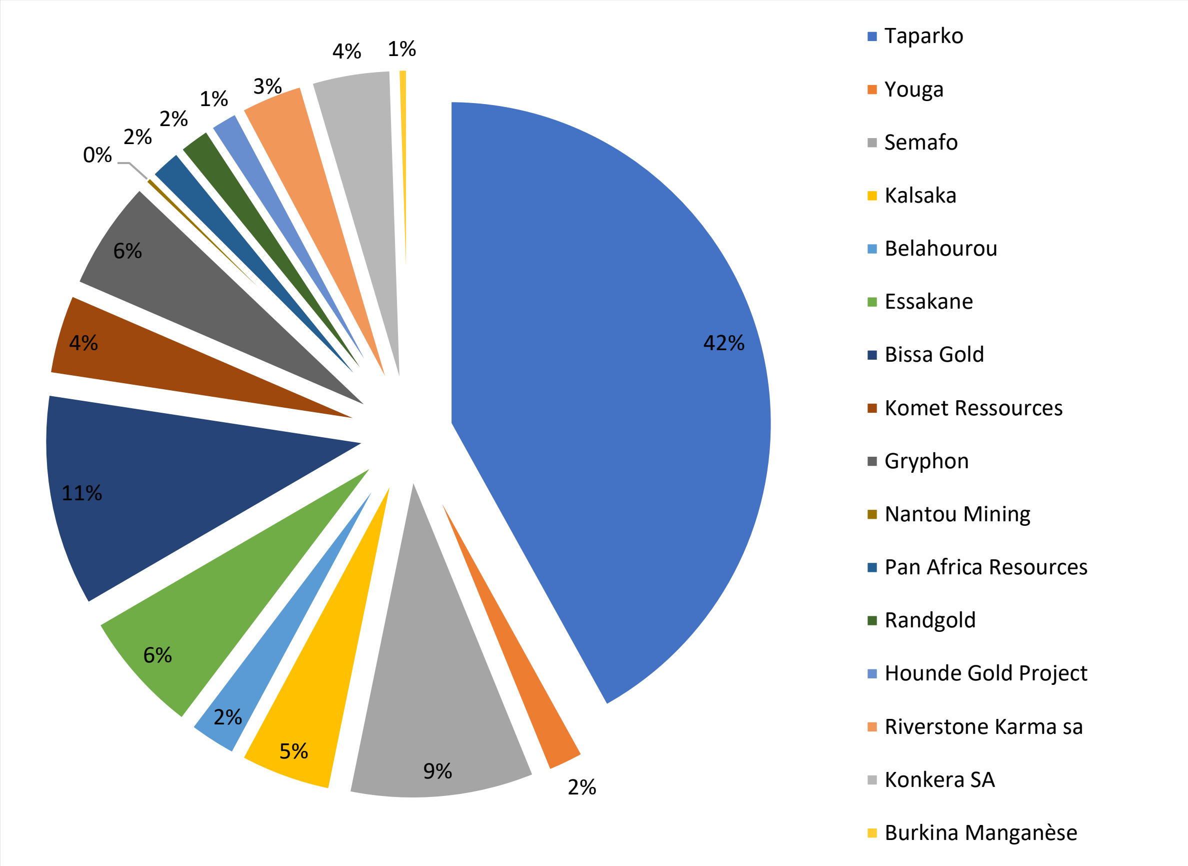

Figure 2-2: Permit area per mining site (data collected from the permits).

2.2. Mining Sites, Pit use and Health Risk

2.2.1. Exposure Assessment

Consumption of foodstuffs is a basic need for human survival, and these foodstuffs should normally provide all nutrients the body needs for its daily functioning. However, some toxic substances could be taken in through food which, at a certain level (Figure 2-3 and Table 2-2), could instead of being a benefit for the body, threaten consumers' health in the short- or long-term.

Despite the mining activities, the cultivation of crops helps to secure the basic necessities of daily life in much of the mining area in Burkina Faso where rural livelihoods remain precarious. In addition to high social pressure for land demand, it is certain that mining sites will be used in the after-closure stage for crop cultivation. Use of mining-impacted sites without any soil characterization, land reclamation for crop cultivation and market gardening using pit lakes, could lead to contamination of these foodstuffs and, indirectly, affect the local population.

Consuming contaminated food in a particular quantity is a risk to humans. To avoid adverse health effects when consuming contaminants, international institutes and organizations have set thresholds for Acceptable Daily Intake (ADI) or Provisional Weekly Tolerable Intake (PWTI) (EFSA, 2015; EFSA, 2009; EFSA, 2010; WHO, 1996; USEPA, 1997b). In mining areas, foodborne contaminant exposure to local communities should be assessed and action taken accordingly to mitigate any exposure beyond regulated thresholds.

Potentially toxic elements can be seen as essential (macro and micro) and non-essential (toxic) elements. Some essential elements include Cr, Co, Cu, Fe, Mn, Ni, Zn, while some non-essential elements include Pb, As and Cd.

Direct toxicological effects of potentially toxic elements on living organisms can lead to inhibition of membrane function, enzyme activities, respiration and protein synthesis, as well as genetic disruptions, oxidative damage (Clemente et al., 2015). Inorganic As exposure causes skin, lungs and bladder cancer and also associated with many non-cancerous outcomes like cardiovascular dysfunction, it could lead to immune dysfunction by hindering cellular and humoral immune response (Sattar et al., 2016). Exposure to Cd above the guidelines is of great concern and could lead to malfunctioning of renal tubular and results in the 'itai-itai' disease (Inaba et al., 2005). Inside organism, Pb blocks the N-methyl-D-aspartate receptor leading to impaired cognitive functions and neuronal synaptic plasticity with a disruption of memory function. Pb long term exposure is a major health issue especially for children for their growth and mental development (Tinggi and Schoendorfer, 2018). Cu, Cr, Co, Fe, Mn and Zn did not show EDI above Referential dose and could at this level of intake contribute to the good functioning of organs. Zn and Cu are essential to the normal functioning of various components of the immune system (Teklić et al., 2013). Zn deficiency results in depressed lymphocyte response to mitogens and abnormal skin hypersensitivity responses. Cu deficiency could lead to failure of the immune system to react properly to infection (Rai et al., 2019).

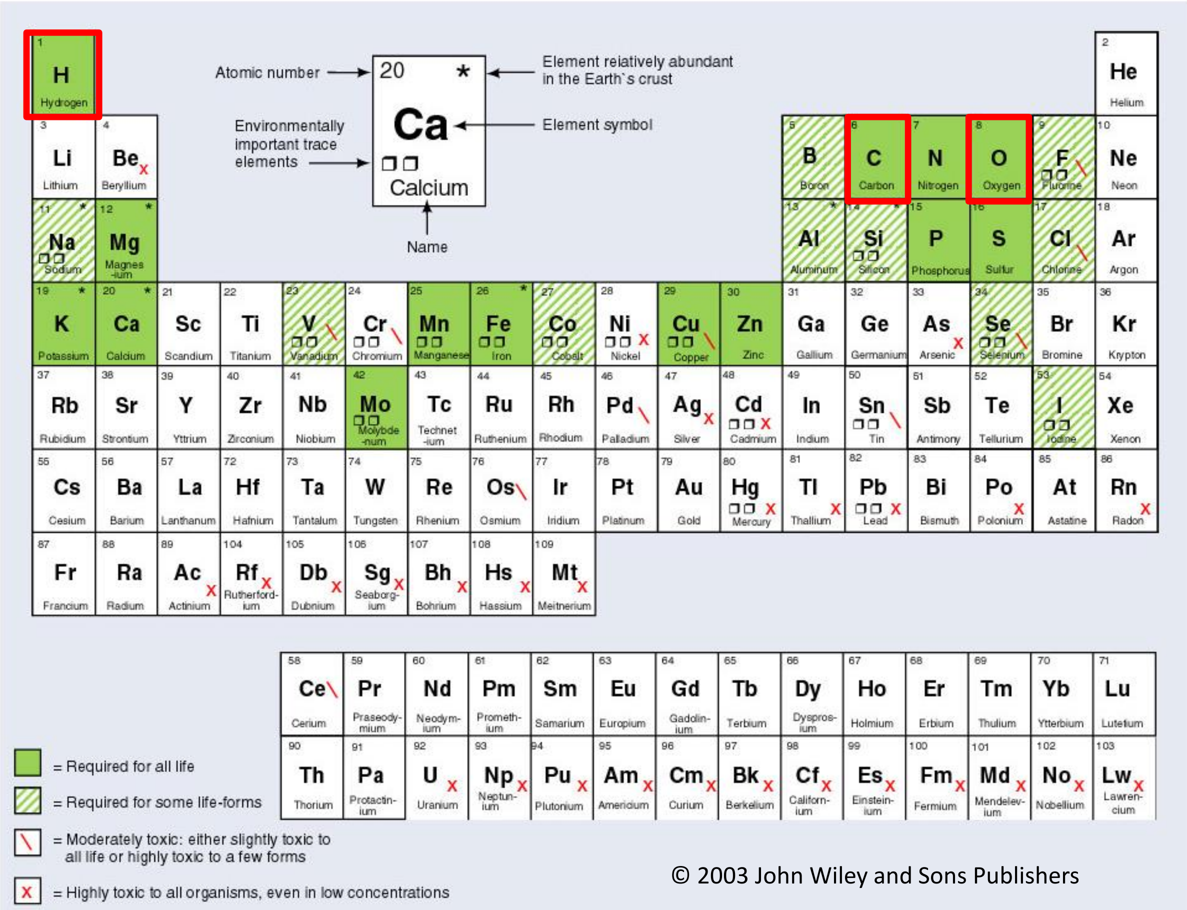

Figure 2-3 : Figure representing the periodic table with compound characteristic,

Table 2-2 Reference doses of contaminants, where needed converted to the same unit of µg kgbw^{-1} day^{-1} by assuming a bodyweight (bw) of 70 kg per adult.

Element	Reference doses (µg kgbw^{-1} day^{-1})	Reference
As	2.14	(FAO/WHO 2011)
Cd	0.36	(ESFA 2009)
Co	1.4	(RIVM 2001)
Cr	300.00	(EFSA 2014)
Cu	71.43	(ESFA 2009)
Fe	642.86	(Institute of Medicine 2000)
Mn	160.00	(ATSDR 2012)
Ni	11.00	(WHO 2007)
Pb	3.60	(RIVM 2001,)
Zn	357.14	(ESFA 2009)

2.2.2. Pit use and Exposure Assessment

Fish consumption is encouraged because it balances the typcal diet of local population, particularly in the context of Burkina Faso where malnutrition spreads over the nation as dictated by the more and more scarce rainy seasons.

Mining pits are often used by local populations for recreational use, irrigation, livestock breeding and fishing; however, mining pits are concomitant with contaminant-contained rocks (Woodman et al., 2016). Such a situation could lead to contaminant exposure for the local population through direct use of the water, or through intake of pit lake-based food or irrigated products. In gold mining, sulfide-based minerals particularly often contain As and more potentially toxic elements, and remobilization of these contaminants leads to environmental problems (Smedley et al., 2007; Bretzler et al., 2017). Studies on anthropogenic or naturally-occurring contaminants in water have been reported in Burkina Faso (Ouédraogo & Amyot 2013; Smedley et al., 2007), and even contamination according to rock type has been reported by Bretzler et al. (2017). Potentially toxic elements content in fish and cereals were reported in many studies worldwide (Table 2-3).

Fish quality and intake risk assessments should be carried out prior to a decision on pit closure options of either refilling the pit or using it as a water reservoir and handing it back to the local population. Quality control and risk assessments could be used to edit a pit lake use procedure for the nearby population, plus sensitizing sessions.

Table 2-3 Literature data about fish and cereals, mg/kg dry weight for cereals, mg/kg wet weight for fish and mg/L for water,,

Literature values for *O. niloticus*									
Element	As	Cd	Co	Cr	Cu	Mn	Ni	Pb	Zn
Pit lakes, Malaysia(i)		0.016±0.003			0.03± 0.01	nd		0.05± 0.05	0.43± 0.11
Nil River, Egypt(ii)		nd				3.57 ± 0.20		1.62± 0.05	7.92 ± 0.59
Nakivubo, Uganda(iii)				0.56 ± 0.20	0.57 ± 0.02	0.16 ± 0.07			0.58 ± 0.27
Drainage water, Egypt(iv)		5.11±0.10						4.59± 0.05	
Drainage canal, Egypt(v)		3.344±0.067	0.16 ± 0.06					1.96± 0.12	0.32± 0.02
Ogulagha, Nigeria (vi)	0.01 ± 0.00	0.68 ± 0.01	1.54 ± 0.12	2.69 ± 0.06				4.77± 0.02	5.44 0.01
Literature values for cereals									
Multiple*		<0.01		<0.04	3.18±0.39	9.56±10.11	<0.01	<0.08	19.75±6.23
Maize**	0.64±0.17	0.08±0.02		1.8±0.33	2.3±0.39		1.3±0.44	0.17±0.08	
Maize***		0.80±0.07		1.4±0.7	2.75±1.10			2.56±0.33	12.9±5.71
Burkina Faso									
Water reservoirs[2]	0.0005±0.0001								
Boreholes[1]	0.0005 to 1.630								
Fish[2]	0.04 to 0.42								

(Baharom and Ishak 2015), (ii) (Talab et al. 2016), (iii) (Birungi et al. 2007), (iv) (Authman, et al. 2012), (v) (Younis et al. 2015), (vi) (Oyibo et al. 2017). * (Akinyele and Shokunbi 2015) Wet method results were considered, results were average of content for three cereals (rice, maize and wheat); ** (Islam et al. 2015); *** (Wang et al. 2017). [1] smedley et al. (2007). [2] Ouedraogo et al. (2013).

2.2.3. Mining Site Use and Exposure Assessment

Cereals are the main dietary component of the population in Burkina Faso and are consumed at least twice per day. Thus, contaminated cereals would be a public health issue. Different types of cereals are grown in Burkina Faso, particularly in the study site. Basically, corn and sorghum are grown as main crops. Cereal crops are grown near the mine and inside the mining perimeter, and quality assessment of crops is a good indicator of soil quality, and, above all, could be used to evaluate local population exposure to cereals from impacted soil. Site use in the after-closure stage could be deduced from cereal quality and intake risk assessments of cereals.

2.3. Mining Impacted Land Reclamation

2.3.1. Reclamation Importance

When active mining ceases, mine facilities and the site are reclaimed and closed. The goal of mine site reclamation and closure should always be to return the site to a condition that most resembles its pre-mining condition (ELAW, 2010; Nehring & Cheng, 2016). The most serious and far-reaching environmental consequences of mining projects occur during the closure period, after mining ceases. Waste rock piles, open pits, tailings impoundments, and leach piles left behind and unattended by the mining company, can begin generating and releasing toxic wastewaters that cause huge damage to soil, water resources and aquatic life, and also to human beings (Doe et al., 2017; Cao, 2007). Reclaimed sites have a wide range of potential functions such as pasture, haylands, recreational areas, wildlife habitat, wetlands, fishing ponds, and swimming pools.

Remediation of industrial wasteland depends on many factors; for example, country regulation and enforcement bodies, localization in either a developing country or in a developed country, types of industry, the final use of the land, and contamination type and degree. Remediation will depend on parameters like the incipient compound, the setting, the cost, the after-land use request, the regulation of the country, and availability of techniques at the time. It will also depend on the image that the company wants to show to the world, and their goodwill.

Mining land reclamation has been practiced worldwide for many years, both in developing and developed countries. The mining land reclamation ratio in the US, Germany, Canada, Brazil and Spain is between 50% and 70%. In China, it is 12% (Cao, 2007). However, in Burkina Faso, as a young mining country, reclamation is not common.

2.3.2. Reclamation Strategies

Reclamation of industrial wasteland, especially mining land, should be done continuously from exploration, construction, exploitation, dismantlement, and closure (Nikolaou & Evangelinos, 2010). Subsequent reclamations are mainly of poor quality and do not respond to stakeholder expectations. In lifelong mining reclamation strategies, mining attracts interest for using existing fleet and reducing costs; for example, by filling the old pit with waste materials from the new pit. However, with falling mineral grade quality, and in order to achieve cost reductions, reclamation is sometimes done as a legal constraint rather than a commitment to environmentally friendly policy of mining companies.

Closure and post-closure plans should include appropriate aftercare, and continued monitoring of the site, pollutant emissions, and related potential impacts. The duration of post-closure monitoring should be defined on a risk basis; however, site conditions typically require a minimum monitoring period of five years after closure, or longer (World Bank, 2008).

2.3.3. Reclamation Methods

Remediation could be carried out using a wide range of techniques that can be classified as chemical, biological, and physical (Thapliyal & Malik, 2006; Hashim et al., 2011). Industrial mining wasteland reclamation using physical methods has been widely investigated and has been applied with interesting results despite some side-effects. Physical technologies include removal of the contaminated land and construction of a barrier, which could be inert or with chemical properties to interact with the targeted compound. Chemical methods are used to decrease the toxicity or mobility of potentially toxic element contaminants by converting them to inactive states. Chemical technologies are used for the treatment of land contamination and water; either surface water or ground water. In soil reclamation, these methods are applicable where the loss of soil fertility can be replenished by the addition of inorganic or organic amendments; for example, acidic soils can be neutralized by the addition of lime. In many cases, chemical technologies have been combined with physical or biological methods to reach acceptable results swiftly.

Biological methods are the most accepted and investigated methods today due to the use of natural properties of living elements to reclaim contaminated soil or water body (Mohanty & Patra, 2020; Patra et al., 2020; Jaskulak et al., 2020). In recent decades, intensive research and development efforts have been directed towards finding cost-effective and eco-compatible solutions to deal with potentially toxic element contaminants in an industrial wasteland (Ashraf et al., 2019; Odoh et al., 2019). There has been increased interest in the possibility of using vegetation to remediate potentially toxic element-contaminated mining sites (Robinson et al., 1997; Fosso-Kankeu & Mulaba-Bafubiandi, 2014; Abdullahi, 2015). It is unacceptable, even for reclamation land, to contribute to using methods that do not consider environmental aspects during their implementation (Hashim et al., 2011).

Biological methods imply the use of plants or microorganisms for reclamation of the wasteland. Examples of in-situ and ex-situ potentially toxic element bioremediation include land-farming, composting, use of bioreactors, bioventing by oxygen, using biofilters, bioaugmentation by microbial cultures, and bio-stimulation by providing nutrients. Some of the other processes include bioaccumulation, bioleaching, and phytoremediation (Table 2-4).

Table 2-4 Techniques and their application for As reported (Gonzalez-Martinez et al., 2019)

Techniques	Articles	Reviews
As		
Adsorption	48	4
Electrocoagulation	2	
Precipitation	47	2
Bioleaching	3	
Others	110	9

2.4. Phytoremediation

2.4.1. Phytoremediation Principles

Phytoremediation uses plants to extract, degrade, contain or immobilize contaminants in soil, groundwater, and other contaminated media. Plants are solar-powered systems, which are the center of primary production in all ecosystems. There are several advantages to reclaiming wasteland through plants, such as; natural regeneration ability, multidimensional improvement in soil properties (due to leaf-litter and beneficial rhizosphere microbiota), and multipurpose utilization of the produced biomass (as fuel, fodder, food, etc.). Further, revegetation of wastelands also improves the aesthetic beauty and supports valuable wildlife. However, this again is not a simple task, as not every plant can grow on the harsh conditions prevailing on the wastelands. Nevertheless, there are different plant species suitable for growth on different types of problematic soils such as those arising due to acidity, alkalinity, and accumulation of potentially toxic elements (Thapliyal & Malik, 2006). The success of phytoremediation as an environmental cleanup technology depends on several factors, including the bioavailability of potentially toxic elements in soils and the ability of the plants to uptake, translocate and accumulate potentially toxic elements in shoots and plant-microbe interactions.

Mining as the main provider of raw materials for the modern industries is spread all over the world due to the huge demand. Mainly as a non-renewable resource, mining use to close. The main reason seems to be depletion of the ore stock, but other factors could lead to stoppage and closure. Mining for mineral, induced contamination by this mineral and by the other element associated to this mineral. Reclamation in this study refer to mining wasteland remediation.

2.4.2. Types of Phytoremediation

There are six basic phytoremediation mechanisms that can clean up mine sites: 1) phytosequestration, 2) rhizodegradation, 3) phytohydraulics, 4) phytoextraction, 5) phytodegradation, and 6) phytovolatilization (Table 2-5). Phytoremediation is best used to treat large areas of shallow contamination. Because high levels of contaminants may be toxic to plants and inhibit their growth, phytoremediation is best applied to low and moderate levels of contamination used in conjunction with other treatment methods or used as a final polishing step in site remediation. The various mechanisms of phytoremediation can treat a wide range of contaminants, including potentially toxic elements; although not all mechanisms are applicable to all contaminants.

Long-term maintenance is minimal once the vegetation is established. Phytoremediation is a sustainable and green technology that does not require supplemental energy. It generates minimal air emissions, water discharge, and secondary wastes, and improves air quality and sequesters greenhouse gases. Biomass from phytoremediation process was incinerated and ash environmentally friendly disposed. Phytoremediation may take longer than other technologies to treat a site, but it has the potential to be less expensive than excavating and treating large volumes of soil ex-situ (USEPA, 2006).

Phytoremediation is an attractive option for alleviation of potentially toxic elements from contaminated sites but it also has some challenges like (i) Several years needed for remediation of contaminated sites. (ii) Most of the hyperaccumulators species have low biomass, slow growth and are not easy to cultivate. (iii) Bioaccumulation ability of plants may be decreased due to pests (iv) Introduction of new species in contaminated sites may affect the local biodiversity. (v) Disposal of contaminated biomass is a matter of concern. (vi) Sustainable remediation of potentially toxic elements by using plants depends mainly on climatic conditions (vii) Phytoremediation technology can be applicable for sites where the potentially toxic element concentrations have low to moderate levels. (viii) Potentially toxic elements can be transferred to food chain in case of mismanagement of contaminated biomass. (ix) Phytoremediation is limited to shallow ground water, soils and sediments. (x)

Soil phytoremediation is applicable to surface soils only. (xi) Phytoremediation is not capable of reducing to 100% (Patra et al., 2020).

Table 2-5: Common phytoremediation mechanisms used in mining remediation

Mechanism (Cleanup Goal)	Description
Phytosequestration (Containment)	The ability of plants to sequester certain contaminants in the rhizosphere through exudation of phytochemicals and on the root through transport proteins and cellular processes.
Phytohydraulics (Containment by controlling hydrology)	The ability of plants to capture and evaporate water and take up and transpire water.
Phytoextraction (Remediation by removal of plants)	The ability of plants to take up contaminants in the transpiration stream.
Phytodegradation (Remediation by destruction)	The ability of plants to take up PTE and break down organic contaminants in the transpiration stream through internal enzymatic activity and photosynthetic oxidation/reduction.
Phytovolatilization (Remediation by removal through plants)	The ability of plants to take up, translocate, and subsequently transpire volatile contaminants in the transpiration stream.

2.4.3. Species for Phytoremediation

Several plant species can be used to phytoremediate mining and processing tailings and for revegetation of mining sites (Mohanty et al., 2010). It has been known since the late 1800s that a special category of plants, the so-called hyperaccumulators, can accumulate extraordinary levels of potentially toxic elements (Hooda, 2007). A hyperaccumulator is defined as a plant with the ability to yield 0.1% Cr, Co, Cu, Ni or 1% Zn, Mn in the above-ground shoots on a dry weight basis. At present, at least 45 plant families are known to contain potentially toxic element-accumulating species. More than 775 plants exist with capabilities to accumulate or hyperaccumulate one or several key metallic elements.

Some plants accumulate larger quantities of potentially toxic elements in their tissues than do others. A key success factor when trying to establish an effective phytoremediating plant community is to find native plant species that grow well in the area to be remediated; but also, to choose plants that are effective absorbers of targeted toxic elements from the soil. Land reclamation commonly fails when carried out as a ‘single-phased’ attempt, even when indigenous species are used for plantations (Thapliyal & Malik, 2006). Use of native plants avoids the introduction of non-native and potentially invasive new species that could threaten regional plant diversity. Few field trials have yet tried to take advantage of native plant diversity; not doing so has often resulted in poor plant colonization at waste sites (Mohanty et al., 2010). Simply said, not using native plants in field trials leads to failure in remediation.

Phytotechnologies provide restoration and land reclamation benefits during and after cleanup, as well as habitats for plants and wildlife. Identifying appropriate plant species and soil amendments is essential to treatment success. Native site species were selected for the remediation experimentations. These species consisted of *Chrisopogon zizanioides,* which is native to the area and widely used in Burkina Faso for erosion control. *Typha domingensis*, spontaneously grown in trenches for tailing seepage control, was considered alongside the objective to control contaminated water runoff such as the tailing in the after-closure stage, or the dewatering of pits with subsequent load in potentially toxic element. Thirdly, *Leucaena leucocephala*, a common species in Burkina Faso, was used for remediation experimentation in unflooded soil.

Chrisopogon zizanioides, or vetiver, is an evergreen perennial tropical grass which can reach 2 to 5 meters in height and is characterized by an extensive root system which can penetrate 4 meters deep. Among other characteristics, vetiver is used for slope control, erosion mitigation, and its essential oil is used in perfume and drinks. Vetiver is renowned for its capabilities to survive in harsh environmental conditions, and, above all, for its capability to uptake some potentially toxic elements. The root and above-ground parts of vetiver are used in some regions for making baskets, fans and mats. In addition, Vetiver is used in paper pulp processing. Studies around the world underpinned the use of vetiver for potentially toxic element mitigation, and extensive results were reported in mining site reclamations and potentially toxic element control. In Burkina Faso, Vetiver is a common weed, and the study of its use in contaminant control is reported by Ondo Zue Abaga, (2012). Vetiver use for mining site remediation was experimented in both laboratory scale and field scale.

Typha domingensis or cattail is a perennial, monoecious, unbranched tropical and subtropical plant widespread in Tropical Africa, which can reach 5 meters. As with vetiver, *Typha* is grown and used for the production of basic daily items such as baskets, fans, mats, rods and fences. In some regions in Africa, the stems and rhizome are eaten, and in Argentina, the pollen is eaten. Medical properties have been found for *Typha*. *Typha* use for land reclamation and mining site potentially toxic element mitigation was reported in many studies, and reliable results have been highlighted (Mukhtar & Abdullahi, 2017). *Typha* has emerged as the species most frequently used for phytoremediation across the world (Eid et al., 2012a). The ability of plants in the *Typha* family to grow and accumulate potentially toxic elements has been investigated, and dependable results have been reported by Bonanno & Cirelli (2017) and

Fernandes & Henriques (1990). *Typha domingensis'* tolerance to wastewater was widely studied (Di Luca et al. 2019), as was its capacity to accumulate Cd, Pb, Cr, Ni, Zn, and Cu (Demirezen & Aksoy, 2004; Eid et al., 2012b). Oliveira et al. (2018) confirmed that *T. domingensis* can sustain up to 50 µM of Cd concentration without subsequent damage to its morphophysiology.

Leucaena leucocephala is a widely studied tropical and subtropical shrub (Normaniza et al., 2008) from Australia (Bray, 1986), India (Neelam et al., 1993; Bhatnagar et al., 1993; Tewari et al., 2004), South and North America (Lugo et al., 1990; Felker et al., 1988; Glumac et al., 1987; Lins et al., 2006), Thailand (Chotchutima et al., 2016), Malaysia (Normaniza et al., 2008), Philippines (Goudie & Moore, 1987), Tanzania (Lulandala & Hall, 1987), Kenya (Heineman et al., 1997), because of its multiple benefits as degraded land restoration (Normaniza et al., 2008), forest regeneration, biomass production and energy (Lugo et al., 1990; Narayanaswami et al., 1986; Tewari et al., 2004; Heineman et al., 1997) animal feed (Glumac et al., 1987), cosmetic and pharmaceutical preparations (Nehdi et al., 2014), phytoremediation properties (Schneider et al., 2013; Ssenku et al., 2017), and particularly for its robustness to growth in difficult conditions (Felker et al., 1988; Glumac et al., 1987). *Leucaena leucocephala* is a high-branched tree, growing between 6 to 20 meters high depending on type (Hawaiian, Peru and Salvador), and flowering at around 8 weeks to six months (Bagyaraj et al., 1989). *Leucaena leucocephala* is a tree common in Burkina Faso. Popular in the south and southeast of the country, it is a tree appreciated by animals.

Phytoremediation in mining soil was widely investigated using different plants or weeds/grass. In China, giant reed (*Arundo donax* L.) was used for remediation of potentially toxic element contaminated soil in non-ferrous mining (Liu et al., 2019), *Jatropha curcus L.* was experimented in highly contaminated soil in conditions simulating South of Spain spring climate (Alvarez-Mateos et al., 2019). Use of phytoremediation in mining focused in mining soil (Lu et al, 2019; Midhat et al., 2019), in waste dump contaminant control and in processing tailings deposits.

2.5. Wasteland Reclamation Experience in Burkina Faso

2.5.1. Land Reclamation Experience

Despite three decades of experience in industrial mining, and long experience in artisanal mining (Hein & Funyufunyu, 2014), Burkina Faso has little experience in industrial wasteland remediation. This could be due to a lack of assessment of likely contaminated sites, lack of enforcement and regulatory institutes, opening up to investors (not willing to put pressure on existing ones), and particularly due to corruption (Kumah, 2006; World Bank, 2008; Gajigo et al., 2012; MME, 2013; Ouoba, 2017; Dialga, 2018). Studies on degraded land reclamation for crop yield growth in Burkina Faso have been successfully carried out (Zougmore et al., 1999). These studies were specific to soil with too-few nutrients for crops. Solutions for the impact of cotton cultivation have also been investigated (Dipama, 2009). The use of vetiver for containing potentially toxic element dispersion in cotton-cultivated soils has been researched, and promising results have been highlighted (Ondo Zue Abaga, 2012). A traditional strategy for reclamation of wastelands and degraded lands is based on topsoiling methods followed by the intensive use of fertilizers and planting of various grass mixtures.

But those strategies did not consider industrial wasteland especially mining industries. Wasteland of industrial mining as well as the artisanal mining using chemicals for gold processing were left without any remediation, forming hotspots from which contaminants spread over. Mining industries impact needs to be investigated, and tailored affordable, aesthetic and easy to implement techniques proposed.

2.5.2. Mining Impacted Land Reclamation Experience in Burkina

When a company relinquishes a mining title, whether for an exploration or mining site, it is responsible for carrying out the rehabilitation of that site prior to departure. National jurisdictions now require some form of closure plan or rehabilitation program to be submitted to the regulatory authority prior to commencement of any work on the site (Environment Code, 2013; Mining Code, 2015). It is an increasingly common requirement for the closure plan to contain details of the estimated cost of rehabilitation, and for a financial surety to be established at the same time (World Bank, 2008; Mining Code, 2015).

Burkina Faso has made a comparatively late start in mining-impacted land reclamation, as they have only recently become a mineral producer, and currently most mining is still in the exploitation phase. However, according to national regulations, mining wastelands must be rehabilitated prior to handing over land to the local community. A deposit secured in the UEMOA central bank or any national bank is a prerequisite during exploitation and provisioning the account as per the environmental impact study and recommendations, is compulsory (Environment Code, 2013; Mining Code, 2015).

As for 2019, several industrial mines closed their operations. The first was Poura, located 180 km west from the capital city and operated by “Société de recherches et d’exploitation minières du Burkina (SOREMIB)”. The exploitation began in 1985 (inaugurated on October 18, 1984 by President Thomas Sankara) until 1999 and produced approximately 15 tons of gold

(Ouedraogo et al., 1987). However, the area of 250 km² was not properly rehabilitated. The second industrial mine was Kalsaka gold mine, located approximately 150 km northwest of Ouagadougou, the capital city. The Kalsaka gold mine, which began operation in 2008, operated by Amara Mining Plc, ceased its activities in the first quarter of 2015. The site is awaiting complete rehabilitation.

2.6. Mining Environmental Regulations in Burkina Faso

2.6.1. Mining Codes

Burkina Faso mining sector is driven by many Code, law and decree which specify how mining should be perform in the country (Table 2-6). One of them is the Mining Code.

The Mining Code is first drafted in 2003 with a clear intention to boost the mining sector and attract and promote investment, enhance and stimulate research and exploration, as an objective to set an economic and social development of the country (Mining Code, 2003). This Mining Code went through slightly modification by the “Loi de Finance” of 2009. The first Mining Code was set after the mining policy statement of 1997 (Declaration de politique manière). In 2015, the transitional government set after departure of the long-lasting president changed the Mining Code, now with the objectives for the mining to benefit the local communities, the regions and the country widely (Mining code, 2015). This Mining Code was enacted by the decree n°2015-885/PRES-TRANS of the same year. Environmental aspects were described in the chapter 5 “Articles 139 to 142” which mainly highlighted need for rehabilitation and found to be deposited in BCEAO (West African states Central bank) or commercial banks in Burkina for the purpose of the rehabilitation of the mine.

2.6.2. Environment Codes

The actual Environmental Code was enacted in 2013 (Environment Code, 2013), and was updated based on the previous Environment Code of 1997 (Environment Code, 1997). The Environment Code stipulates that necessary actions should be taken to protect the environment and defined the principle of “Pollueur-Payeur” (who is responsible for a pollution must be accountable for the remediation). Broadly, the mining code regulated use of chemical substances, waste management, environmental evaluations for permitting, specifies industries functioning mode regarding environment, issues air, water and soil pollution control and inspections and management of environmental risks and catastrophes.

2.6.3. Other Codes in Relation with Mining Activities

Many other law and code interfere in mining (Table 2-6). The Code of forestry was issued in 2011 to aims in particular to set a symbiosis between the need to protect natural forest, wildlife and fishery resources and meet the economic, cultural and social needs of the population (Code of Forestry, 1997 and 2011). There is also the Public health Act (Public Health Code, 1994) which give guidelines on the management of health in Burkina Faso, management of waste, water and air pollution and pubic hygiene.

Table 2-6 Codes and laws driving mining activities in Burkina Faso

Type	Number	Date	Title	Reference
Loi	n°001-2003/AN	08 Mai 2003	Code minier	Mining Code, 2003
Loi	n°005/97/ADP	30 Janvier 1997	Code de l'environnement	Environment Code, 1997
Loi	n°006/2013/AN	02 Avril 2013	Code de l'environnement au Burkina Faso	Environment Code, 2013
Loi	n°14/96/ADP	23 Mai 1996	Réorganisation Agraire et Foncière	Land Management Code, 2001
Loi	n°034-2012/AN	02 Juillet 2012	Réorganisation Agraire et Foncière au Burkina Faso	Land Management Code, 2012
Loi	n°006/97/ADP	31 Janvier 1997	Code forestier au Burkina Faso	Code of Forestry, 1997
Loi	n°003-2011/AN	05 Avril 2011	Code Forestier au Burkina Faso	Code of Forestry, 2011
Loi	n°23/94/ADP	19 Mai 1994	Code de la Santé Publique	Public Health Code, 1994
Loi	n° 022-2005/AN	24 Mai 2005	Code de l'hygiene publique au Burkina Faso	Public Health Code, 2005
Loi	n°002-2001/AN	6 Février 2001	Relative à la gestion de l'eau	Water Management Code, 2001
Loi	n° 008-2014/AN	08 Avril 2014	Orientation sur le développement durable	Sustainable development Code, 2014

2.7. Canadian and South African Standards

Canadian and south African soil and water quality guidelines were used for benchmarking of the soil and water investigated in this study (Table 2-7 and Table 2-8).

Table 2-7 Canadian Soil Quality Guidelines for the Protection of Environmental and Human Health, (CCME, 2007), Concentration (mg/kg dry weight),

N°	Chemical name	Agricultural*	Residential/ parkland**	Commercial***	Industrial****
1	As	12	12	12	12
2	Cd	1.4	10	22	22
3	Cr	64	64	87	87
4	Co	40	50	300	300
5	Cu	63	63	91	91
6	Pb	70	140	260	600
8	Ni	45	45	89	89
9	Zn	250	250	410	410

*Agricultural: where the primary land use is growing crops or tending livestock. This also includes agricultural lands that provide habitat for resident and transitory wildlife and native flora.

**Residential/Parkland: where the primary activity is residential or recreational activity; parkland is defined as a buffer between areas of residency, and includes campground areas, but excludes wildlands such as national or provincial parks.

***Commercial: where the primary activity is commercial (e.g., shopping mall) and not residential or manufacturing. This does not include zones where food is grown.

****Industrial: where the primary activity involves the production, manufacture, or construction of goods.

Table 2-8: South African water quality guidelines, field guide, 1st Edition, 1996. Values in mg/L, exept for Cd in µg/L, DO for dissolved oxygen, TDS for total dissolved solids,

	Human consumption	Recreation*	Industry*	Agriculture		
				Livestock	Irrigation	Aquaculture
pH	6-9	6.5-8.5	7.0-8.0	NA	6.5-8.4	6.5-9.0
NH_3	0.1					2.0
DO						5-8
F^-	1			2	2	
NO_2	6			10		
NO_3	0			100		0.05
SO_4^{2-}	200		30	1000		
TDS	450		100	2000		
As	0.01	NA	NA	1	0.1	0.05
Cd	5	NA	NA	10	10	0.2
Ca	32			1000		
Cl	100		20	1500	1	600
Cr	0.05			1	0.1	0.002
Co				1	0.05	
Cu	1			1	0.2	0.005
CN						0.05
Fe	0.1		0.1	10	5	0.01
Pb	0.01			0.1	0.2	0.01
Mg	30			500		
Mn	0.05		0.05	10	0.02	0.1
Ni				1	0.20	
Zn	3			20	1	0.03

*For recreation use, “full contact” guideline values were shown. For Industry, “Category 1” data was shown.

2.8. Study Site Specificities

2.8.1. Location

The site under investigation is in the southern part of Burkina Faso in the Boulgou province, 180 km from Ouagadougou, the city town, and close to the border with Ghana Republic (Figure 2-4 and Table 2-9). The site is in the catchment area of the Nakambé River (formerly White Volta). The Zéra secondary basin passes inside the site area from west to east. Along its course, several small streams flow into this waterway, including the Gossé. The Zéra River joins the Nakambé a few kilometers after crossing the border with Ghana.

The mine poured its first gold in 2008, and is still in the operation phase; and at December 31, 2014 had produced 537621 oz (15.24 tons) of gold (Woodman et al., 2015). The site is delimited by the coordinates detailed in Table 2-9:

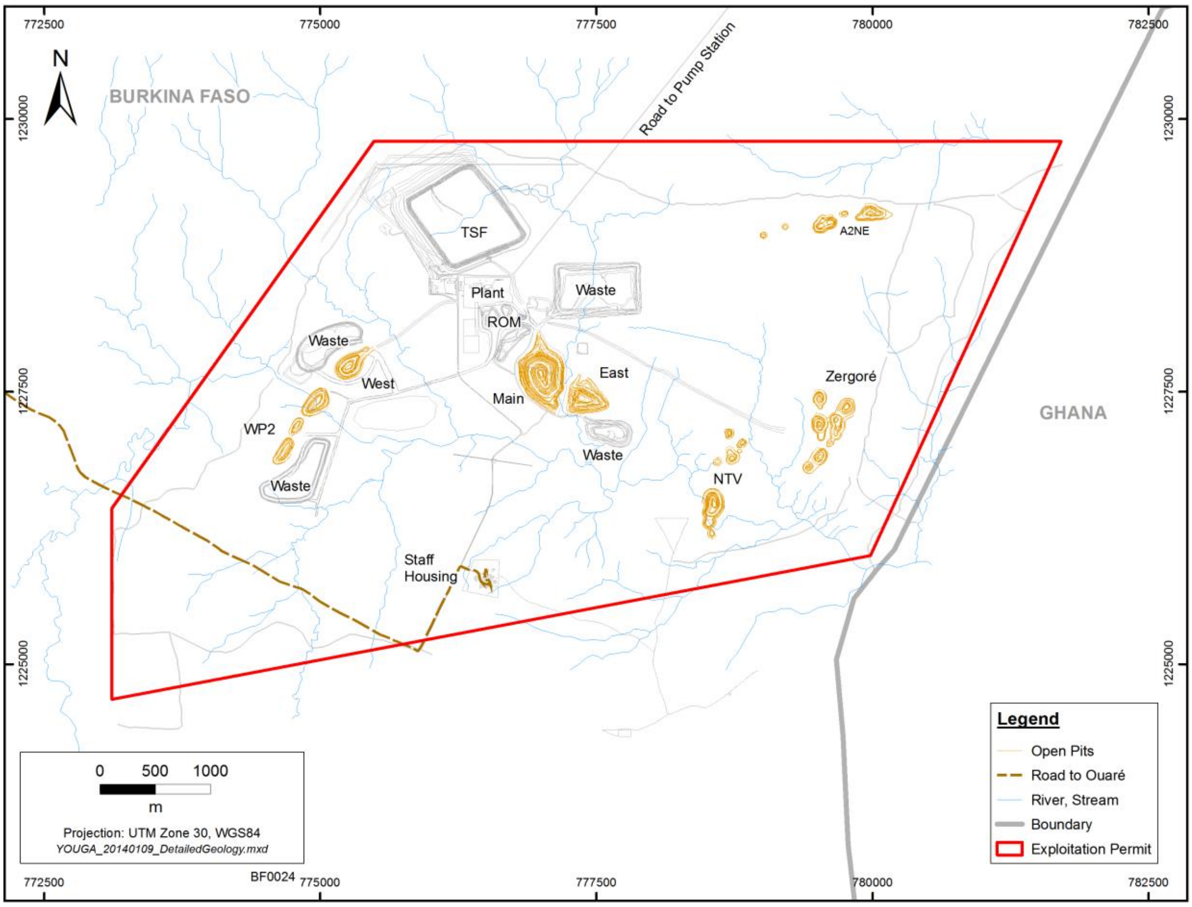

Figure 2-4 Youga gold Mine site Perimeter, source: Burkina mining company,

Table 2-9: The Youga gold mine site limit coordinates, UTM 30, WGS 84, source: Burkina mining company,

Limit	Longitudes (X)	Latitudes (Y)	
A	780001	1225783	Ghana border
B	773134	1224464	
C	773134	1226217	
D	775505	1229580	
E	781729	1229580	Ghana border

The site comprises a processing plant, a tailing storage facility (TSF) containing the processing residue, mainly a mixture of pulps and chemicals such as cyanide, lime and carbon, a run-of-mine (ROM) pad, staff housing and multiple pits with their associated waste dumps.

2.8.2. Gold Mining and Processing

For mining, the site under our scrutiny utilizes a conventional open pit, selective mining exploitation method. The ore extracted from the mine is conveyed by trucks to the mill. The processing plant uses the conventional gravity/carbon-in-leach (CIL) gold recovery process and consists of three stage crushing, ball milling, gravity concentration and cyanidation by carbon in leach (Figure 2-5). Pressure Zadra elution is utilized for recovery of gold from loaded carbon.

The discharge of the grinding circuit advances to feed the leaching circuit. Slurry of crushed gold bearing ore, and cyanide solution enters the first tank of the CIL circuit.

The first tank is used to adjust the pH and to condition the slurry with oxygen. The discharge of the first tank is gravity fed into the second tank where the cyanide is added for a target concentration of 150 ppm of free cyanide in the solution. Gold that dissolves from the ore is continuously adsorbed onto the coconut-shell activated carbon. The discharge of the last tank flows to the tailing's ponds (Blanchette et al., 2011).

As the slurry passes through the circuit, gold continuously leaches in the cyanide solution. Carbon particles are suspended in the tanks to simultaneously adsorb gold from cyanide solution. Fresh carbon from the elution circuit enters the last CIL tank. Counter-current transfer of carbon particles takes place using slurry pumps at regular intervals. The carbon in the first tank has highest gold loading and is sent to the elution circuit for gold removal (Wadnerkar et al., 2015).

The striping circuit (pressure Zadra) consists of an acid leaching column, followed by an elution column operated in closed circuit. The stripped carbon is regenerated in a rotary kiln prior to being returned to the CIL circuit. The process consists of leaching the gold-laden carbon with a caustic sodium sulfide solution. The operation is conducted in close circuit with simultaneous leaching and electrolysis (Zadra, 1950).

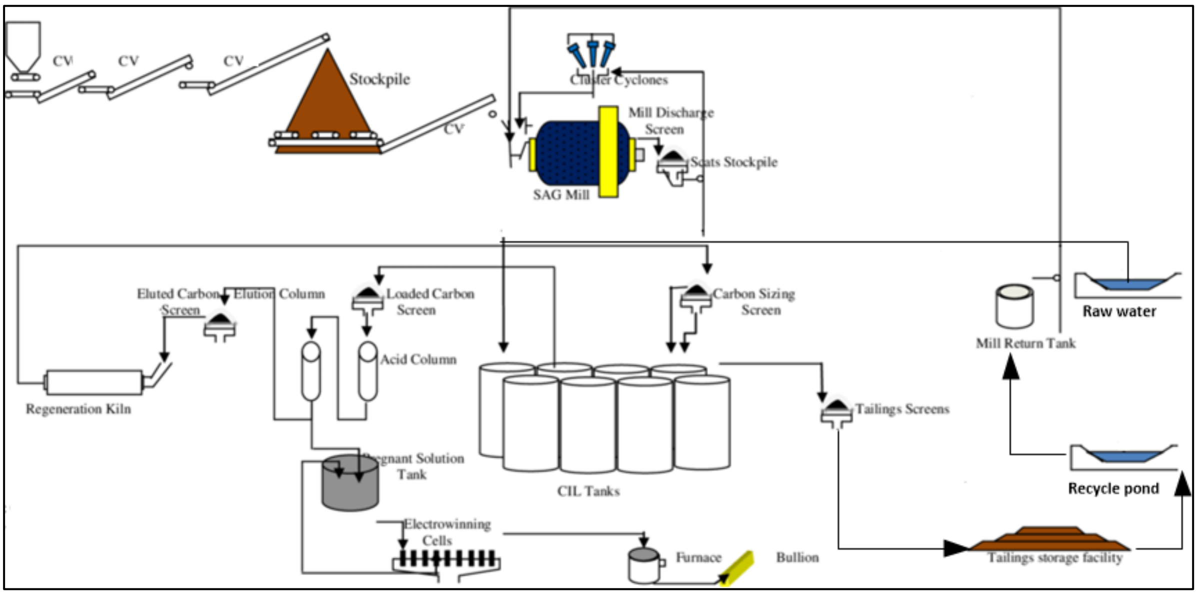

Figure 2-5 Carbon in leach (CIL) process plant flow diagram, updated from Faanu, (2011),

2.8.3. Geological Aspects

The study area is defined by two types of rock that cover the zone (Figure 2-6): volcanic rocks in the south, and sedimentary rocks in the north. Two kinds of aquifer can be found in the locality: a shallow aquifer in which water can reach trough well, all year round, at less than 10 meters depth; and a fractured aquifer (Yeates et al., 2005).

The Youga deposits are characterized by two distinct styles of mineralization: the moderately to weakly silicified host rock with quartz stockwork veining and pyrite as the predominant sulfide; and the intensely silicified arkose, with abundant quartz veins and more diverse sulfides (pyrite, arsenopyrite, chalcopyrite, pyrrhotite, and galena). The alteration paragenesis associated with the mineralized vein stockwork is characterized by quartz, ankerite, albite, chlorite and pyrite (Woodman et al., 2016). The presence of sulfide predisposes the site to contain some arsenic (Smedley et al., 2007).

Six soil types falling into four distinct classes were identified within the perimeter during the environmental impact assessment of the project (Yeates et al., 2005): (i) the class of mineral soils with lithosol subtype, mainly sandy silt texture; (ii) the class of poorly developed soils from soil erosions, developed from the decomposition of original rocks (schist, granite, quartz) and presenting a sandy silt in texture on the surface; (iii) the class of brown soils developed from alkaline rocks with montmorillonite as the dominant type of clay, represented by three subtypes: the first subtype is characterized by a brown color and sandy silty clay at the surface and developed biological activities; the second, a ferric tropical eutrophic brown soil characterized by a silty clay to sandy clay at the surface and developed biological activities; The third subtype is a hydromorphous tropical eutrophic brown soil with a silty clay texture on the surface, with calcareous nodules whose percentage increases in depth, and (iv) the last class of subtype is the hydromorphous soil with a silty clay texture on the surface, found only in certain low lying areas, especially on the surface of flooded areas as well as areas prone to flood. Its coarse matter consists of approximately 5% ferruginous concretion and ferro-manganese concretion (Yeates et al., 2005).

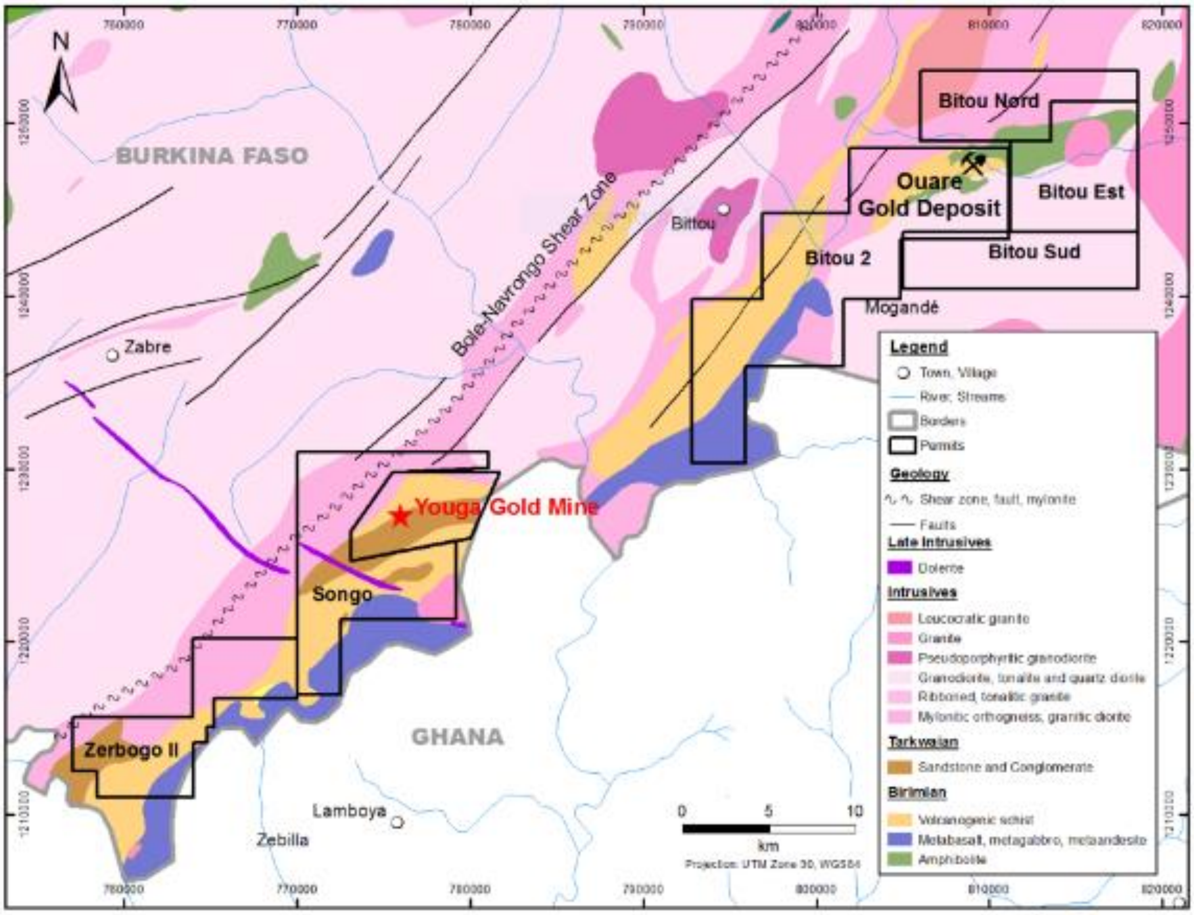

Figure 2-6 Youga Belt and geology (from Woodman et al., 2015).

2.8.4. Climate, Vegetation and Land Use

The zone under investigation has a Guinea Savannah climate condition. The vegetal cover is also rich, as annual rainfall reaches 900-1200 mm, and is mainly colonized by diversified shrubs and trees. Native plants dominate. There are even some exotic plants, planted by the mining company. Around the processing plant and the pits, the trenches for containment or to divert rainwater are colonized by hydrophytes such as *Typha domingensis* and *Chrisopogon zizanioides*. In waste dumps and naked rocks, *Calotropis procera* and *Laptadenia hastata* can now be found creeping through the rocks.

The highest temperatures are experienced at the end of the dry season, with mean monthly maxima exceeding 39°C. Annual evaporation is high; 2870 mm in the region. The wind is dominated by the Harmattan (Figure 2-7).

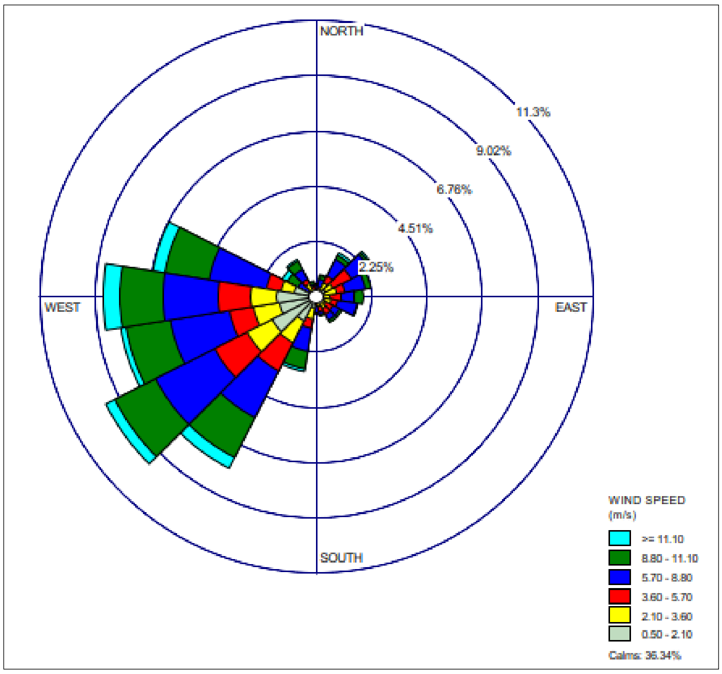

Figure 2-7 Windrose of harmattan blowing from January to April 2017, plotted using data measured on Youga gold mine site.

Primary land use in the locality is for agricultural purpose, growing cereals, crops and rice in the lowlands. Traditional animal breeding systems are still ongoing in the locality, with animals gathered during the rainy season and left to themselves during the dry season, which puts pressure on the shrub vegetation. The nomadic population settled around the perimeter of the mine, to benefit from facilities such as the potable water (WHO, 2011), and also the river water pumped from the river which they use to water their herds.

Section 2

Chapter 3: Gold mine impact on soil quality

This chapter is based on the publication:
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ABSTRACT: The present study aims to assess the impact of a gold mine located in the south-eastern part of Burkina Faso on local soil quality. This information is needed in order to determine any health hazards and potential remediation strategies as the mining site is expected to be turned over to the local community after the closure of the mine. For the purpose total potentially toxic elements analysis as well as a sequential extraction were performed and results were interpreted using different methodologies: enrichment factor (EF), geoaccumulation index (Igeo) computed using two separate background samples and comparison to selected national standard. The soil analysis revealed a moderate to significant soil EF and Igeo with hotspots located closer to the ore processing plant and on the east side of the site. Cobalt, chromium, copper, manganese, nickel, lead and zinc expressed moderate to significant contamination context. Sequential extraction revealed, however, that less than 2% of the arsenic is found in the exchangeable part. Cobalt and zinc are more distributed in the different fractions than arsenic, chromium, copper, nickel and lead. Anthropogenic contributions were revealed by the study.

3.1. Introduction

In Burkina Faso, an attractive legal framework combined with an investor incentive system led to an increase in the number of large-scale mining projects. Currently, eight gold mines and one zinc mine are in operation. In addition, large numbers of small-scale artisanal mines exist (Chambre des Mines du Burkina, 2018). Clearly, mining is a great contributor to the country's economy but, on the other hand, the industry gives rise to concerns about environmental pollution.

Studies identifying mining as a source of contamination in the environment are emerging worldwide (Khaska et al., 2015; Cott et al., 2016; Cuvier et al., 2016). In Africa in particular, studies from Tunisia (Wali et al., 2004), Ghana (Abdul-Wadood & Ashraf, 2015), Kenya (Ngure et al., 2014), South Africa (Hansen, 2015) and Namibia (Mileusnić et al., 2014) have highlighted contamination of the environment as well as the threat to the continents' ecosystems and way of life (Hilson, 2002; Hilson, 2016).

One of the main environmental issues with mining is the impact it has on water quality and availability and on associated soil characteristics. The contribution of mining to pollution could be both anthropogenic (associated with man-made actions like chemical use, waste generation, and traffic) and geogenic (associated with parent rocks) (Barbieri et al., 2018). The quality of the soil will determine whether human life and terrestrial wildlife can be supported and whether safety will be guaranteed for local communities. Pollution with potentially toxic elements may persist for many years, threatening local ecosystems and jeopardizing local community health (Křibek et al., 2014). Mining site rehabilitation may thus be required before handing over the site to the local population and local government bodies.

This study, therefore, aims to assess the impact of a gold mine on local soil quality. PTE namely arsenic, cadmium, cobalt, chromium, cupper, manganese, nickel, lead and zinc were investiageted based on the mining site geology and soil background (Etruscan, 2005). This information can be used to determine the level of contamination of the site, assess health hazards affecting the local population and, if necessary, decide on a remediation strategy.

3.2. Materials and Methods

3.2.1. Soil Sampling

3.2.1.1. Surface Soil Sampling

Thirty-four samples were collected in 2016 inside the mine perimeter (Study site has been described in 2.6), labeled S01-S34 in Figure 3-1, and two reference samples were taken outside the perimeter for comparison purposes. The mining site was divided into five zones and samples were taken in each zone to cover the entire mine perimeter. Reference samples were selected in zones where it is supposed to not have spatial geochemical variation.

The mining site was divided into five subgroups, namely Zones 1 to 5. Zone 1 consisted of an area covering the processing plant and the tailings storage facility (TSF) with a grouping of six samples. Zone 2 consisted of seven samples covering the west pits and their waste dumps. Zone 3 covered the senior camps and the access between the camp and the mine, with seven samples. Zone 4 covered the main and east pits and their waste dumps and accounted for five samples. Zone 5 covered the south part of the site and consisted of Nanga tail village backfilled pits, Zergoré pits and waste dump, and A2 north-east pit and waste dumps; nine samples were taken in this area.

Approximately 3 kg of soil per location was sampled from the layer between 5 and 10 cm deep (to not include the surface litter, twigs, rocks, or crop residue) (USEPA, 2000) using a plastic rod and collected by hand using plastic gloves. The soil was stored in a plastic bag and transported to the on-site laboratory for drying at 105 °C until it reached a constant weight. Coarse rock material, leaves and wood were removed, and the remaining fraction was mechanically ground. The laboratory sample was obtained by passing the sample into an autosubsampler, and 300 g was collected in a paper bag and brought to the laboratory for analysis.

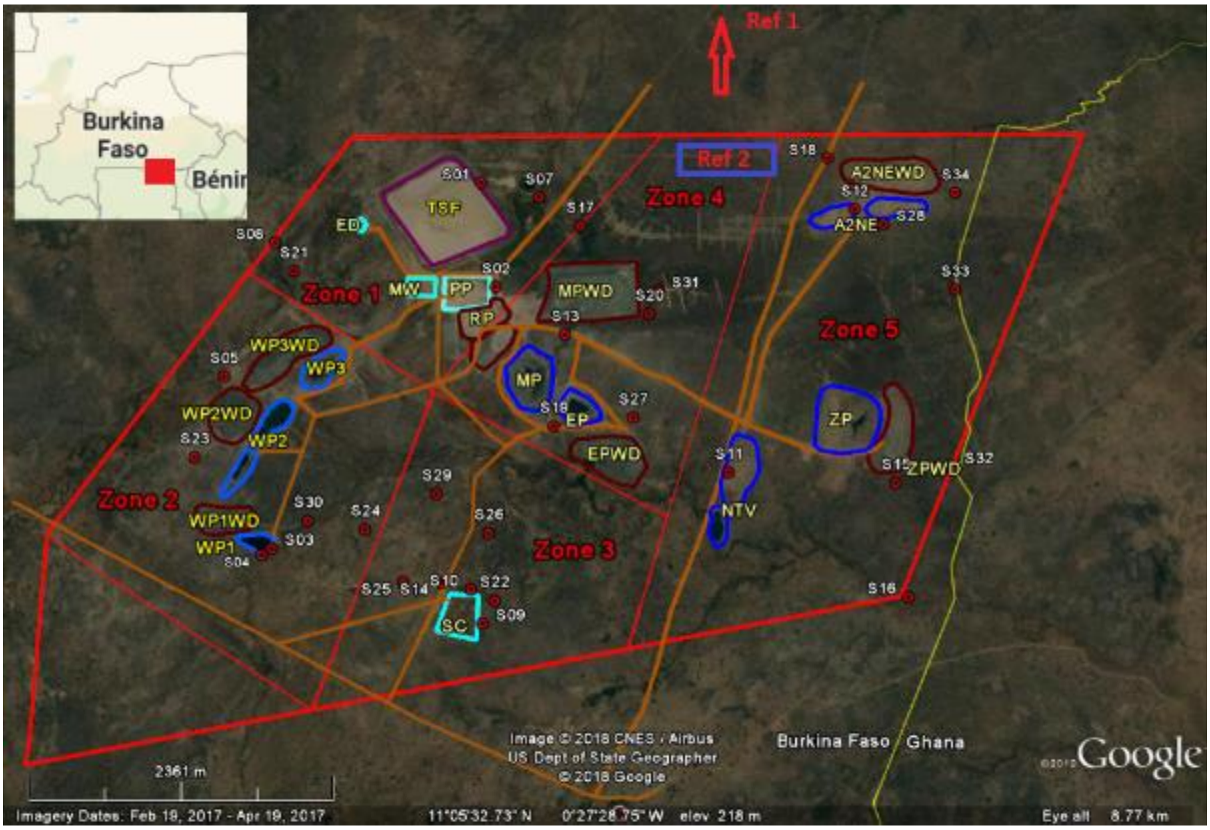


Figure 3-1 Site overview and zoning. TSF: tailing storage facilities, MW: maintenance workshop, PP: process plant, ED: explosives store, RP: ROM-pad, MP: main pit, EP: east pit, MPWD: main pit waste dump, EPWD: east pit waste dump, A2NE: A2 north-east pit, A2NEWD: A2 north-east pit waste dump, ZP: Zergoré pits, ZPWD: Zergoré waste dump, WP1: west pit 1, WP2: west pit 2, WP3: west pit 3, WP1WD: west pit 1 waste dump, WP2WD: west pit 2 waste dumps, WP3WD: west pit 3 waste dumps, SC: senior camp. Ref 1 for the direction where reference surface soils were sampled (At 5 km away). Ref 2 where core samples were taken.

3.2.1.2. Subsoil Sampling

Diamond drilling cores were sampled for PTE analysis in 2016, when a total of six samples taken in the northeast part of the site were considered. At that time, the Northeast part of the site was not impacted by mining activities like trenching, blasting and intensive drilling. Around 3 kg of each core from 3.5 to 4.5 m depth were sent to ALS Geochemistry Burkina Faso for analysis. The results of the core samples were used for the calculation of the geoaccumulation index.

3.2.2. Soil Analysis

3.2.2.1. Soil pH, Total Organic Matter and Cation Exchange Capacity

The soil pH was measured using a daily calibrated multiparameter probe HI9829 (Hanna Instruments Inc, Woonsocket, USA) in a mixture of soil and bi-distilled water at a ratio of 1:5 that was shaken overnight. Total organic matter (TOM) was analyzed gravimetrically using the loss on ignition method at 550 °C (Heiri et al., 2001). Cation exchange capacity (CEC) was analyzed using Chapman's method with ammonium acetate at pH 7, as described by Ross & Ketterings (2011). The resulting ammonium concentration was measured using LCK 303 test kits and a DR 2800 spectrophotometer (Hach Lange, Germany). Soil texture data was obtained from the Environmental impact assessment study done in 2005 (Etruscan, 2005).

3.2.2.2. Total Potentially Toxic Element Analysis

The method used for the analysis of the total potentially toxic element (PTE) was that of Bettinelli et al. (2000) and Melaku et al. (2005), with prior microwave digestion (an ETHOS Touch Control Advanced Microwave Labstation from Milestone Inc® Monroe, CT, USA) using a mixture of nitric acid, hydrochloric acid, hydrofluoric acid and boric acid in a ratio of 2:6:2:2. In short, 0.5 grams of sample was weighed directly in a Teflon digestion tube with the first three acids and heated in the microwave according to the first three steps given in Table 3-1. After cooling the vessels, 2 mL of saturated boric acid was added, and the sample was heated and cooled again (Steps 4 and 5, Table 3-1). The digestate was then filtered with a Whatman filter n°41 into a 50-mL flask and made to volume. Acid matching standard solutions were prepared based on a Certipur® ICP (inductively coupled plasma) multi-element standard IV of 23 elements at 1000 mg/L from Merck. This standard was also used for spiking samples for recovery calculation. An additional 1000 mg/L arsenic standard was prepared from arsenic trioxide (AsO_3). The equipment used for the analysis of PTE was a Vista-MPX, CCD simultaneous ICP-OES, from Varian. The PTE considered were arsenic (As), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Copper (Cu), Manganese (Mn), Nickel (Ni), Lead (Pb) and Zinc (Zn). Core samples were analyzed by ICP-OES in the ALS laboratory.

Table 3-1 Microwave assisted digestion program

Step	1	2	3	Cooling	4	5
Power (W)	250	400	600	0	300	0
Hold time (min)	8	4	6	10	3	2

3.2.2.3. Sequential Extraction

Sequential extraction was carried out using the modified Community Bureau of Reference (BCR) three-step method (Rauret et al., 2000) for PTE speciations of soil samples, with subsequent analysis on ICP-OES as previously described: (i) an acid extractable/exchangeable fraction was obtained after treatment with a 0.11M acetic acid solution; (ii) then, the easily reducible fraction was obtained after reaction with a 0.5M hydroxylamine hydrochloride solution; and, (iii) the oxidizable fraction was obtained after reaction with hydrogen peroxide. The residual fraction was calculated as the difference between the total potentially toxic element concentration and the sum of the above three fractions (Fernández-Ondoño et al., 2017). Note that this is an idealized representation as by definition this closes the mass balance to 100%, whereas in reality this is often not the case when the residual fraction is also experimentally determined; e.g. Malaj et al (2012) found recovery rates between 70-136% and Wen et al. (2016) found recovery rates between 86-101% when comparing the sum of the fractions to the total concentration.

3.2.3. Quality Control

All reagents used were analytical grade at least, and bi-distilled water was used during analysis. A calibration blank consisting of reagent water with the same acid mix was used for the calibration of the ICP instrument. In each sequence of digestion, a procedural blank was prepared and used during analysis. Spiked soil samples in which a known quantity of the target element had been added were processed in the same way as normal samples and used to evaluate the recovery. All soil values are expressed in dry matter. The MDL was determined by running the lowest standard of 0.1 ppm seven times and calculating the MDL based on three times the standard deviation of the seven runs (EPA Document 40 CFR 136). Two independent replicates were performed in parallel for each sample.

3.2.4. Pollution Assessment

The mining impact on soil was evaluated using enrichment factors (EF) compared with a reference soil sampled at the same depth (5 to 10 cm) from a location 5 km from the site and further using geoaccumulation index (Igeo), calculated based on core samples from the site taken at a depth of between 3.5 and 4.5 m. In addition, a comparison against international standards was carried out.

3.2.4.1. Enrichment Factor Calculation of Soil Contaminants

The EF is defined as the concentration ratio of a given element to a reference element in a sample, divided by the same ratio in a reference material (Cuvier et al., 2016).

$$EF = \frac{\left[\frac{Element}{Reference\ Element}\right]Sample}{\left[\frac{Element}{Reference\ Element}\right]Reference\ Material} \qquad \text{(Equation 1)}$$

Soil from outside the site was considered to be the reference sample and was taken at two locations with the same soil texture as the mining site that were far enough away not to be impacted by the mining operation. The reference element considered in this study is manganese (Mn) due to its particular stability in soils, characterized by the absence of vertical mobility and/or degradation phenomena (Isaac & Ololade, 2014; Zhang et al., 2014a). EF values ranging from 0.5 to 2 are considered to be due to natural variability, whereas ratios above 2 indicate enrichment corresponding to anthropogenic inputs. Five categories were considered in the interpretation of the results (Cuvier et al., 2016; Barbieri et al., 2015):

- EF < 2 = deficient to minimal enrichment (DE)
- 2 ≤ EF < 5 = moderate enrichment (ME)
- 5 ≤ EF < 20 = significant enrichment (SE)
- 20 ≤ EF < 40 = high enrichment (HE)
- EF ≥ 40 = extremely high enrichment (EE)

3.2.4.2. Geoaccumulation Index

Geoaccumulation was computed based on background values of core samples taken between 3.5 and 4.5 meters (Barbieri et al., 2018).

$\mathrm{Igeo} = \ln\left(Cn/1.5\ Bn\right)$ (equation 2) (Barbieri et al., 2018).

When Cn is the measured concentration of the element in surface soil and Bn is the subsoil value of the element, the natural contribution correction factor of 1.5 enables the analysis of natural fluctuations in the content of a given substance in the environment; simply put, 1.5 represents the attenuation factor which accounts for lithogenic variations in background concentrations at the site. Interpretation of Igeo was carried out based on seven classes ranging from below 0 (unpolluted) to above 5 (extremely polluted) (Nowrouzi & Pourkhabbaz, 2014):

- <0 Uncontaminated
- 0-1 Uncontaminated to moderately contaminated
- 1-2 Moderately contaminated
- 2-3 Moderately to strongly contaminated
- 3-4 Strongly contaminated
- 4-5 Strongly to extremely strong contaminated
- >5 Extremely contaminated

3.2.4.3. Soil Quality Assessment

Soil quality was benchmarked against international standards as no specific standards exist in Burkina Faso. The Canadian Soil Quality Guidelines for the Protection of Environmental and Human Health (CCME, 2007) (Table 2-8) were used to compare soil values due to Canada gold mining experience and dominance in Burkina Faso's mining sector.

3.2.5. Statistical Analysis of the Results and Mapping

The Shapiro–Wilk's test was used to determine the normal distribution of the dataset while the Levene's test was undertaken to verify the homoscedasticity of values. If a dataset passed these two conditions of validity, one-way analysis of variance (ANOVA) was used to evaluate the existence of significant differences between the PTE concentrations from different zones at a level of significance of 0.05. In the case of the absence of homoscedasticity, Welch's test was applied. Otherwise, if the Normal set data was rejected by the Shapiro–Wilk's test, the Mann–Whitney test was used. Excel (Microsoft 2016), SPSS 24 (IBM statistic software, USA) and XLSTAT-Base (Addinsoft, V19.7, 2018) were used for the treatment of the results. Spatial distribution was plotted using MapInfo 11.0 (Pitney Bowes Software Inc, USA). A handheld Garmin eTrex 10 (Garmin, USA) was used for coordinates reading.

3.3. Results and Discussion

3.3.1. Total Organic Matter Content, Cation Exchange Capacities and pH

Results of TOM, CEC and pH analyses are represented in Figure 3-2. TOM values ranged from 2.58 ± 1.40 to 3.71 ± 0.76%. CEC results ranged from 4.55 ± 1.24 to 7.80 ± 4.12 meq/100g. The pH level ranged from 6.96 ± 0.39 to 7.77 ± 1.11, expressing the slightly alkaline nature of the soils which, in turn, may cause low mobility of PTE; PTE uptake by plants is inversely correlated with a decrease in pH (Zhou et al., 2018). pH, CEC and TOM dictate the distribution of potentially toxic element species in soils and therefore can be used to forecast PTE toxicity to plants, microbes and human beings (Liu et al., 2018). Carbon occurring in the

TOM is the primary source of electrons, and so TOM and CEC should be correlated (Khaledian et al., 2017); this was confirmed, with a significant correlation between CEC and TOM (F=0.554, p<0.001). Reference soils sampled gave TOM, CEC and pH values of 2.35 ± 1.11%, 4.27 ± 1.90 meq/100g and 6.51 ± 0.16 respectively. In comparison, Liu et al. (2005) found in a lead/zinc mine a pH of 5.32, a CEC of 7.80 ranging from 5.7 to 10 meq/100 g and a TOM of 3.62 ranging from 3.21 to 3.97%. In a gold mine in Cote d'Ivoire, Sako et al. (2018) found soil sample TOM results ranging from 0.2 to 1.6 %.

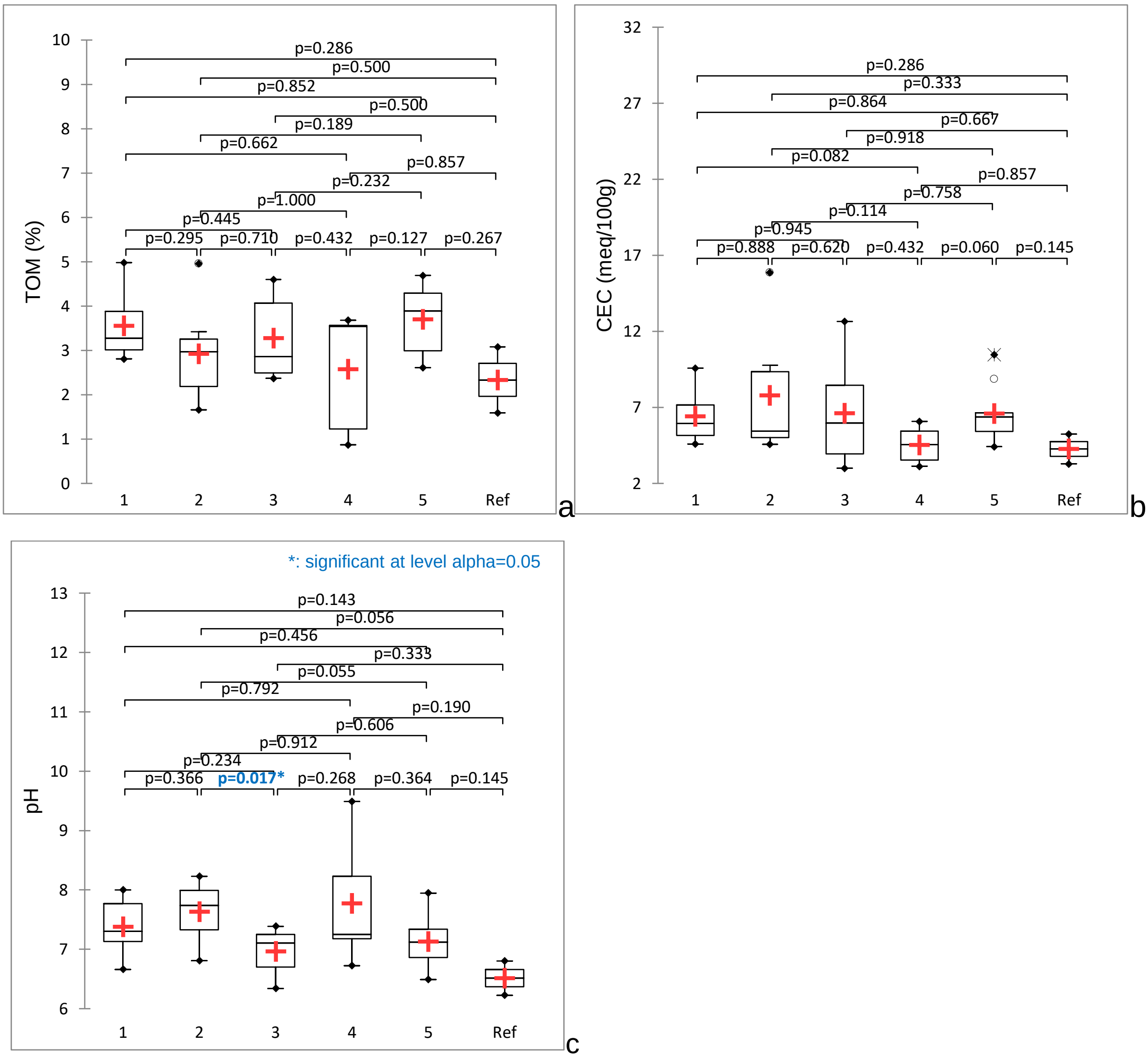
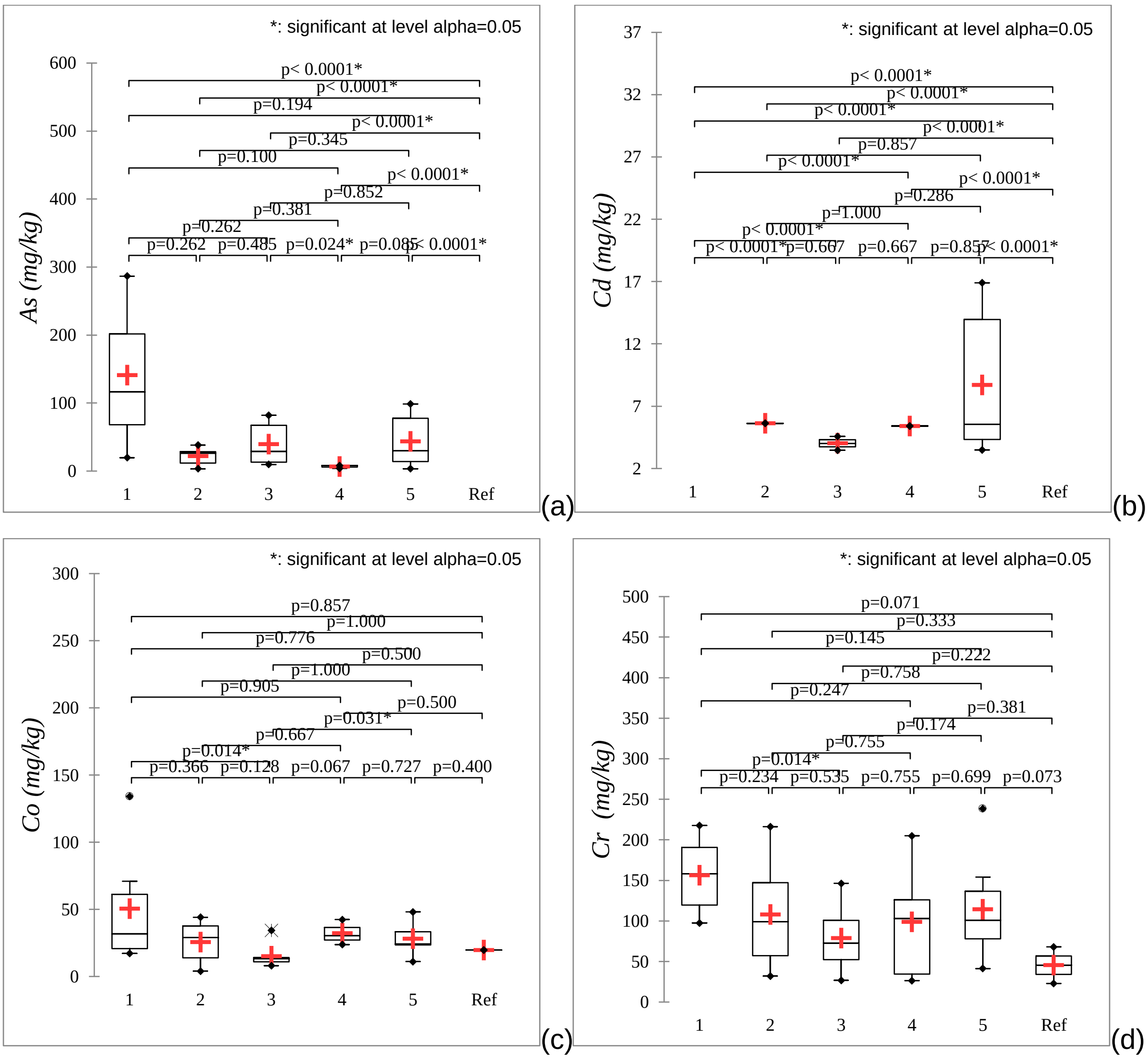


Figure 3-2 TOM (a), CEC (b) and pH (c) boxplot representations per zones (1, 2, 3, 4, and 5), and Mann–Whitney test, "Ref" for reference soil.

3.3.2. Total Potentially Toxic Element Contents

PTE analysis of soil samples is a key aspect of investigating soil contamination. PTE analysis of the 34 soil samples showed an uneven distribution of contaminants from one location to another (Figure 3-3). A higher standard deviation was observed for some potentially toxic elements. Such high deviation can be due to the lack of uniformity of the distribution of potentially toxic elements across the site as found by Islam et al. (2017) in different soils. In this context, the difference could be linked to the difference in soil types met within the mining site (sandy silt texture type, clay type, silty clay type) (Etruscan, 2005) and to the influence of mining activities, excavations, dewatering activities, emissions from fuels combustion.



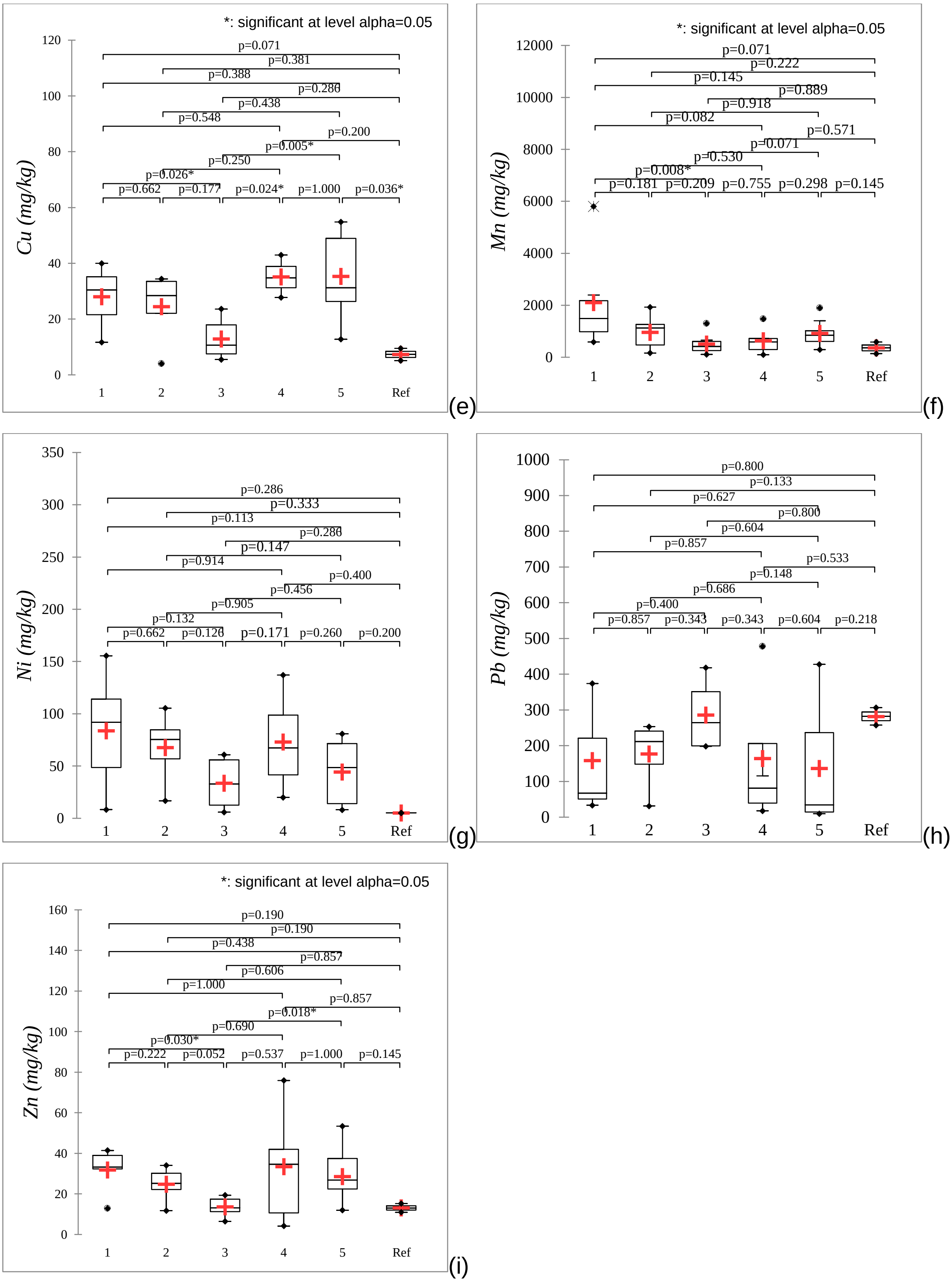


Figure 3-3 Potentially toxic elements distribution per zones and Mann-Whitney test, “Ref” for reference soil. As (a), Cd (b), Co (c), Cr (d), Cu (e), Mn (f), Ni (g), Pb (h) and Zn (i). Cd and As below MDL in reference soil. Cd in Zone 1 below MDL.

The overall site As average (44.7±59.3 mg/kg) exceeded the CSQG guideline value for agricultural use of 12 mg/kg. Sako et al. (2018) found in gold mining soil samples As up to 278.6 mg/kg, and Kinimo et al. (2018) found in industrial mining 294.4 ± 195.4 mg/kg of As at Afema and 7.38 ± 4.39 mg/kg As at Bonikro in Cote d'Ivoire.

29.4% of sites samples showing Cd higher than the MDL were clustered in Zone 5 and were higher than the CSQG limit of 1.4 mg/kg for agricultural use. The site average was 7.12±5.16 mg/kg. The surface reference soil Cd was below the MDL. All zones were below the residential use value of 10 mg/kg.

Co was found in 94.1% of samples with site average of 29.3±23.8 mg/kg. The highest Co value was identified in Zone 1, and the lowest value was found in Zone 3. Except for Zone 3, averages in all zones were higher than the surface reference soil. The level in Zone 1 exceeded the CSQG agricultural guideline of 40 mg/kg.

Cr was detected in all soil samples and site average was 111± 9.5 mg/kg . The highest Cr content was found in Zone 1 (157± 47.5 mg/kg), and the lowest Cr concentrations were found in Zone 3 (78.8 ± 41.9 mg/kg). Concentrations in all zones were above the CSQG guideline for soil intended for agricultural and residential/parkland use (64 mg/kg) and were also above the reference soil value.

Cu in all the zones exhibited values higher than the surface reference soil but were lower than the CSQG agricultural guidelines values of 63 mg/kg.

Mn concentrations in zones followed this trend Zone 1 > Zone 5 > Zone 2 > Zone 4 > Zone 3. Except for Zone 1, all zones exhibited average values below surface reference soil.

Zone 1 showed the highest Ni average and Zone 3 had the lowest value. All zone average values were high compared to the surface reference soil value of 5.24 ± 0.00 mg/kg. Zone 3 and 5 Ni levels were below the CSQG guidelines values of 45 mg/kg for agricultural use.

The mine perimeter exhibited Pb levels that exceeded the CSQG agricultural guideline of 70 mg/kg. Other than the southern side of the site, which exhibited a value of 137±153.2 mg/kg, all the other zones showed Pb concentrations above the residential value of 140 mg/kg. Pb values in Zone 3 even exceeded the guideline for commercial land use of 260 mg/kg. Furthermore, Pb could have been underestimated due to volatilization during drying at 105°C.

Zn concentrations in zones follow this trend: Zone 4 > Zone 1 > Zone 5 > Zone 2 > Zone 3. All zone Zn values were below the CSQG agricultural guideline of 250 mg/kg.

Mann-Whitney's test revealed significant differences in Cd concentrations between firstly Zone 1 and 2, secondly between Zone 1 and 3 and thirdly between Zone 1 and 4. Significant differences in Co, Cr, Cu, Mn, and Zn were found between Zone 1 and Zone 3. Significant differences in As and Cu concentrations were found between Zone 3 and Zone 4. Significant differences were found in Co, Cu and Zn concentrations between Zone 4 and Zone 5.

All in all, the investigation revealed that As level is high and therefore is of concern. Cd is also of concern with value higher than the safe threshold for agriculture. Co is a concern in zone 1 only. Cr and Pb were exceeding the agriculture threshold. Investigated Cu, Zn and Mn were below threshold for agriculture and therefore not a concern. Ni was a concern in zone 3 and 5.

3.3.3. Multivariate Potentially Toxic Element Analysis

Principal component analysis (PCA) is shown in Figure 3-4 to illustrate the inter-relationship between PTE, soil physical parameters, and samples. If significant correlation exists among elements, then they may be controlled by a single factor (Lin et al., 2019). PCA showed the tendency of Cr, Mn, Co and Cu to be more or less related to TOM, CEC. Generally, soils with high TOM and CEC sequester more Cr (III) through (i) strong adsorption onto the negatively charged soil surfaces; (ii) formation of complexes with organic matter; and, (iii) formation of insoluble oxides and hydroxides in soil (Stewart et al., 2003). The analysis also expresses the inter-relation between Zn, Ni, As and Cd with samples from zone 1 (S02, S17, S07), zone 2 (S04, S05, S06), zone 4 (S20) and zone 5 (S11, S15, S12). Additionally, a grouping of As, Zn and Ni confirms the assumption that they have the same pyrite and arsenopyrite source. A third group constituted by samples from zone 2 (S23, S03, S23), zones 3 (S10, S09), Zone 4 (S14, S27, S13) and zone 5 (S18) did not show affinity to any element investigated. Sample S22 from zone 3 showed a good affinity to Pb as, to a lesser degree, did samples from zone 1 (S21), zone 2 (S30), zone 3 (S25, S29, S26) and zone 5 (S32, S28, S33).

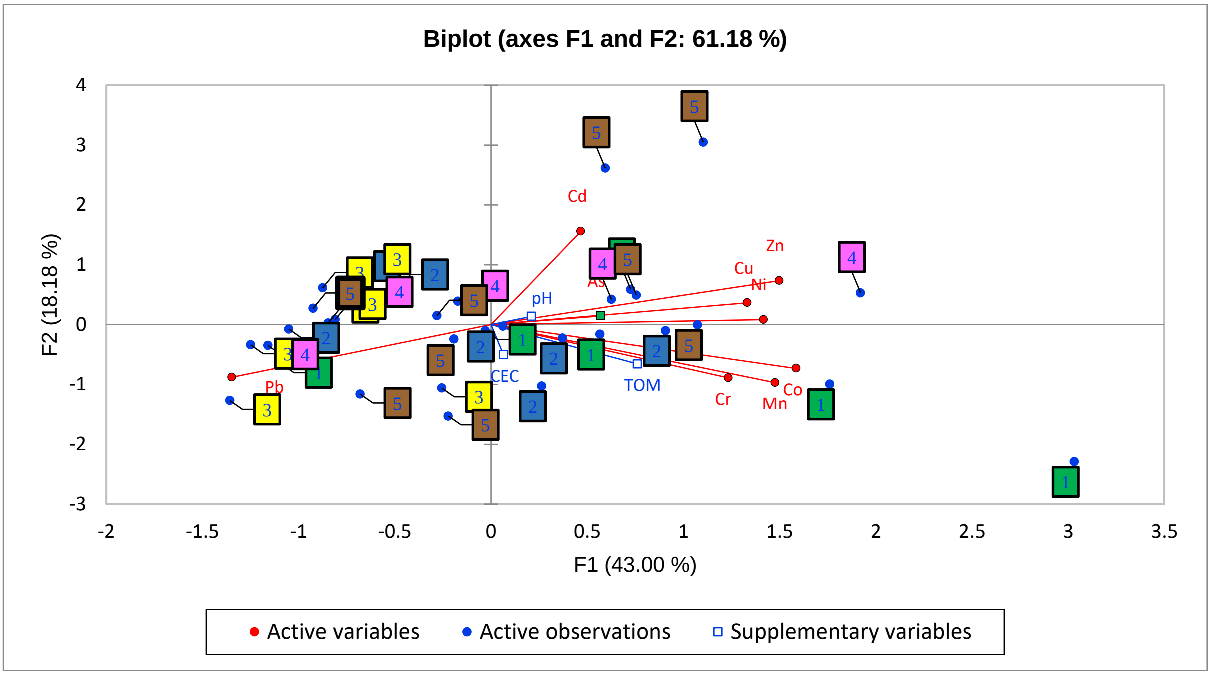


Figure 3-4 Principal component analysis of PTE of soil samples in the mine site. Missing data estimated on the base of mean or mode. Pearson Correlation. Number stand for zone number. Done using XLSTAT-Base (Addinsoft, V19.7, 2018),

PTE contamination of mining sites is common and reported in the literature (Arenas-Lago et al., 2014; Wu et al., 2010; Liu et al., 2005). The presence of elements is influenced by the parent rocks in the area, the dominant activities and by mobilization/transportation mechanisms by wind or water from one location to another. The Zone 1 impact could be explained by plant cleaning water runoff, and dust from the crushing unit, the TSF, and the maintenance workshops. Zone 2, Zone 4 and Zone 5 contamination was mainly dependent on ore geochemistry. The Zone 3 PTE influence came from the human settlement in the senior camp. An interaction between zones was observed. Zones 2 and 3 were located in the dominant wind impact of Zone 1. In particular, TSF wind impacts Zone 2 and crushing in Zone 2 and truck circulation dust impact Zone 3. The presence of As can be directly linked to the presence of arsenopyrite (FeAsS) in the Youga ore deposit. Co, Zn and Ni could come from their natural occurrence with pyrite (FeS_2) and arsenopyrite. Galena (PbS) was likely to be the main source of Pb in the site. Cu could be a derivative of chalcopyrite ($CuFeS_2$) (Woodman et al., 2016; Sako et al., 2018). Furthermore, element presence could be explained by artisanal miners in the vicinity using chemicals for gold complexation. Contamination from fuel and grease rainfall runoff from improperly managed workshops is another possibility, lead-fuel could have contributed as well as mining fleet tire abrasion, vehicular exhaust, and brake pad degradation (Wang et al., 2017). In addition, processing strategies, residue management techniques and overall plant management all influence mine site contamination.

3.3.4. Enrichment Factors and Geoaccumulation Indexes

Based on the monitoring results of the soil quality of the mining site and on the results of surface and subsurface referential soil, quantitative and qualitative assessment of the site contamination was carried out. The enrichment factor was computed for qualitative assessment of the site based on reference soil samples located 5 km north-east of the site. Further Igeo was calculated and comparisons with standards were made. Geoaccumulation indexes were computed based on the averages of the core sample results used as background values. The background soil potentially toxic element contents were 5.57±3.69, 21.4±9.00, 65.9±47.9, 46.1±20.4, 671±208, 50.3±24.0, 4.57±3.41, 67.4±21.1 mg/kg As, Co, Cr, Cu, Mn, Ni, Pb and Zn, respectively. The EF and Igeo are shown in Table 3-2.

Table 3-2: Enrichment factor and geoaccumulation index per zone and site average

	As		Co		Cr		Cu		Ni		Pb		Zn	
Zone	EF	Igeo	EF	Igeo	EF	Igeo	EF	Igeo	EF	Igeo	EF	Igeo	EF	Igeo
Zone 1	10.4	6.26	0.43	6.58	0.95	8.84	0.90	6.76	3.80	7.94	0.12	6.18	0.60	7.26
Zone 2	2.06	4.39	0.54	5.91	1.09	8.46	0.66	6.62	4.26	7.73	0.37	6.29	0.39	7.01
Zone 3	17.1	4.99	0.70	5.37	1.54	8.15	1.59	5.98	9.37	7.03	0.43	6.77	1.19	6.41
Zone 4	0.92	3.22	0.40	6.13	1.40	8.38	1.24	6.99	5.90	7.8	1.34	6.22	1.38	7.32
Zone 5	5.39	5.09	0.64	5.99	1.23	8.52	2.38	6.99	3.80	7.31	0.26	6.03	1.10	7.16
Average	7.33	5.11	0.56	6.04	1.24	8.49	1.43	6.73	5.35	7.57	0.45	6.28	0.92	7.07

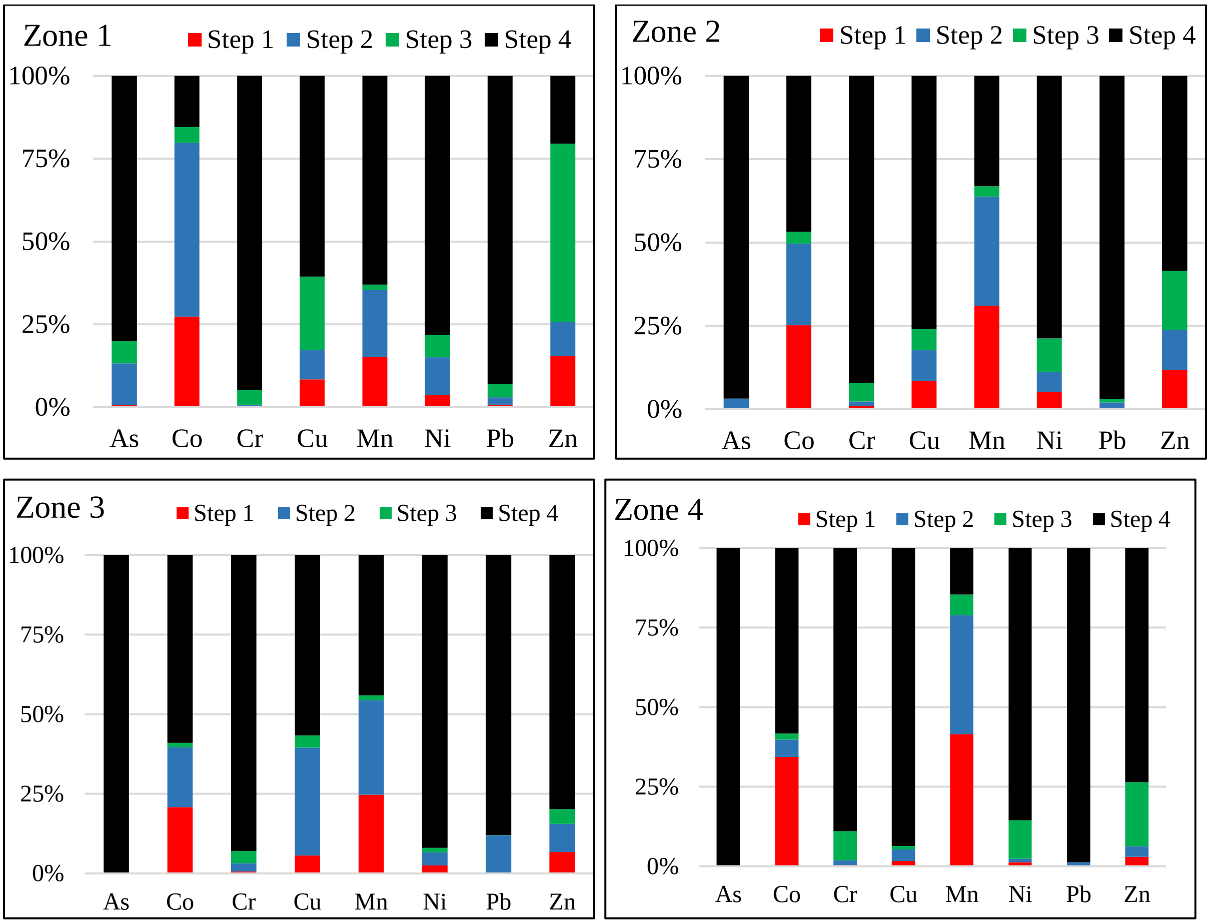
The average EF of As was 7, indicating significant enrichment and an acute anthropogenic action, whilst the Igeo average was 5 expressing strong to extremely strong contamination. The EF for As followed this trend: Zone 3 > Zone 1 > Zone 5 > Zone 2 > Zone > 4. Igeo revealed this trend for As: Zone 1 > Zone 5 > Zone 3 > Zone 2 > Zone > 4. Co showed an average EF of 0.6, which indicates a deficiency to minimal enrichment. The highest Zn EF values were found in Zone 4, whilst the highest Co and Ni EF was found in Zone 3. A deficient to minimal enrichment was noticed for Cr in all zones. A minimal EF of Pb was noticed for all zones, with the highest value for Zone 4 at 1.34. Zn contamination was minimal, with an EF average of 1. On the other hand, however, Co, Cr, Cu, Mn, Ni, Pb and Zn from all zones expressed an extreme contamination context according to Igeo. Noticeable enrichment highlighted by both EF and Igeo in Zone 1 could be linked to the washing of plant, water runoff, and TSF impact and mining activities like crushing, lixiviation, storage of residue, mechanical and chemical moving of elements. Zone 2 was influenced by mining activities and waste dump element mobilizations. The results for Zone 3 could be explained by the dominant wind direction which transports dust from the ore crushing unit and spreads it over Zone 3.

Enrichment is dependent on the location and the element but, all-in-all, EF and Igeo results infer soil contamination and indicate an anthropogenic contribution.

A comparison of zone PTE levels with the background values of core samples taken at a depth of 3.5-4.5 m revealed different results. Higher concentrations were found in all five zones than in the background for As, Cr and Pb, in four zones for Co and Mn, and in three zones for Ni. In contrast, background values for Cu and Zn were higher than each of the zone values.

3.3.5. Sequential Extraction Results

Sequential extraction was undertaken to evaluate the individual fraction of each element in the soil to forecast accurately the potential and actual hazards, given that total concentration of PTEs in soil does not always represent the bioavailable fraction (Fernández-Ondoño et al., 2017). Results of the sequential extraction are represented in Figure 3-5. Anthropogenic potentially toxic elements, in comparison to potentially toxic elements of natural origin, are more loosely associated with the soil and, therefore, can be released (Wali et al., 2004).



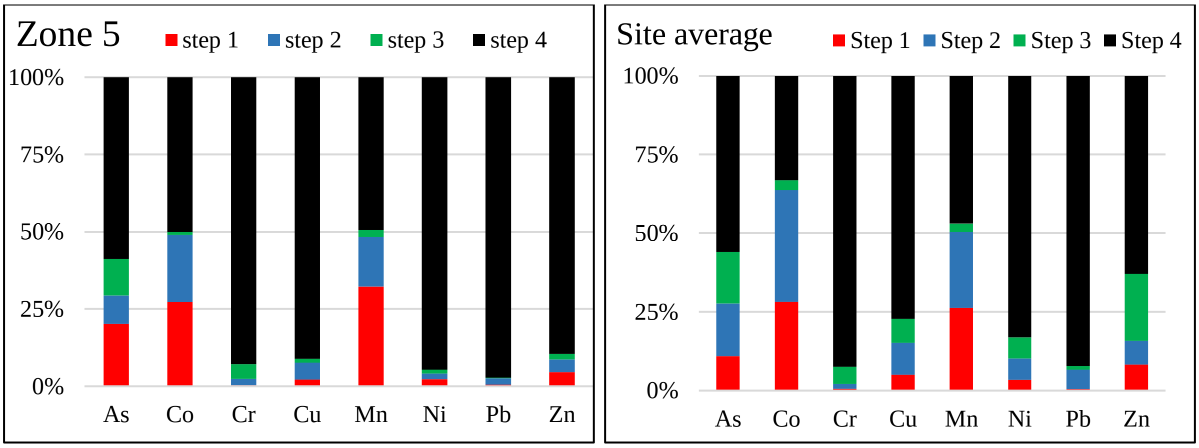


Figure 3-5 Fraction distribution per zone and site average, step 1 stands for the first fraction extracted using acetic acid solution, step 2 is the second fraction extracted using hydroxylamine hydrochloride solution, and step 3 is the third fraction obtained after treatment with hydrogen peroxide and step 4 is the residual fraction, calculated as the difference between the total potentially toxic element concentration and the sum of the above three fractions. Cd was below MDL for steps 1, 2 and 3, and therefore not added in the graph.

In general, the average values of elements in the exchangeable fraction varied in this decreasing order: Mn > Co > As > Zn > Cu > Ni > Pb > Cr, whereas the average percentage of this fraction to the total digestion values follows this pattern: Mn > Co > Zn > Cu > Ni > As > Cr and Pb. Zone 5 exhibited the highest share for As follow by Zone 1. Zones 2, 3 and 4 presented a value below the MDL. For Co, this trend was: Zone 1 > Zone 4 > Zone 5 > Zone 2 > Zone 3. In comparison, the highest values were found in Zone 1 for Cu, Mn, Zn and Pb.

PTE in the reducible fraction (F2), those bound to Fe and Mn oxides, follow this pattern based on site average: Mn > Pb > Co > As > Ni > Cu > Zn > Cr. The average percentage compared to the total digestion values followed this sequence: Mn > Co > Cu > Zn > Ni > Pb < Cr > As.

Elements associated with the oxidizable phase (F3) are assumed to stay in the soil for longer periods and are considered not to be bioavailable because of association with stable high molecular weight humic substances (Fernandez et al., 2004; Bogusz & Oleszczuk, 2018). The average share of each element in this fraction follows this pattern: As < Pb < Co < Mn < Ni < Cr < Cu < Zn compared to the total PTE content.

The greater part of PTE was associated with the residual fraction, which is also called the mineral fraction (F4). The average shares of those elements were 96.3 %, 70.1 %, 94.1 %, 84.1 %, 58.3 %, 88.7 %, 97.5 % and 81.1 % for As, Co, Cr, Cu, Mn, Ni, Pb and Zn.

Fraction F1 is the more mobile one and is considered to be more hazardous as it is used by organisms. The fraction F2 is the fraction which could be mobilized under reducing conditions which occur in water-saturated environments caused by flooding or by high groundwater levels (Dorau, 2017). Consequently, lowering the groundwater level due to dewatering of pits could delay the mobilization of this fraction and the refilling of pits with water could, in turn, lead to a rise in the groundwater level and the potential mobilization of this fraction. The particular presence of elements at high percentages in fraction F3 could be explained by the affinity of the organic matter behaving as a scavenger for PTE (Wali et al., 2014). The residual fraction (F4) is mostly associated with the Earth's crust and hence lithogenic in origin (Hasan et al., 2018). This analysis revealed that the highest shares of elements were in the residual fraction. Nevertheless, a change in soil physicochemical parameters could lead to mobilization of such amounts (non-residual fraction) that health risks would result.

3.3.6. Soil Characteristics and Sequential Extraction Fractions

The influence of soil characteristics on sequential extraction fractions was investigated using Pearson's correlation statistics. The computation excluded As from the first, second and third fraction of the sequential extraction due to values being below the detection limit for some zones.

In the second fraction, Cr exhibited a significant positive correlation (0.582). In the third fraction, Ni again exhibited a significant negative correlation (-0.545) with TOM. When considering the total digestion analysis results, more elements exhibited a significant correlation with TOM: Co (0.393), Cr (0.757), Cu (0.483), Mn (0.386). Elements which did not show a relationship with TOM are associated with the inorganic fraction of the soil (Klubi et al., 2018).

No significant correlations were found between CEC and sequential extraction for the first three fractions. In the total digestion analysis results, only Cr presented a significant correlation with CEC (0.344).

Correlations with pH were found with elements from the first fraction of the sequential extraction; Cu (0.632), Ni (0.660) and Zn (0.581). In the second fraction, pH correlations were with Cr (-0.514), Mn (0.555), and Zn (0.711). The pH correlations for the third fraction were with Mn (0.554), Ni (0.788) and Zn (0.563). Soil pH plays a crucial role in the mobility and bioavailability of soil potentially toxic elements and affects the solution and surface complexation reactions of cations, ion exchange, and other metal-binding processes. An increase in the pH results in the generation of more negatively charged sorption sites on the soil colloid and organic matter surfaces and, consequently, a decrease in positive potentially toxic element availability (Xu et al., 2017). Potentially toxic elements occurring as anionic species may behave differently. Studies have demonstrated that the availability of potentially toxic elements was influenced by competitive adsorption-desorption processes which, in turn, are determined by soil properties including pH, redox potential, organic matter content, electric conductivity (EC), quantity and type of clay minerals, and hydrous oxides of Fe, Al and Mn (Islam et al., 2017; Cheng et al., 2018).

Differences in soil PTE can also be explained by the diversity of soil types encountered within the site. The clayish styles of soil could explain the content of elements in the soil, as higher clay and higher pH sequesters more elements (Stewart et al., 2003). Clay minerals are able to increase soil pH and decrease the chemical-extractable fractions and bioavailability of potentially toxic elements in soils (Xu et al., 2017), acting as a natural scavenger of pollutants by taking up cations and anions either through ion exchange or adsorption. Silt and the silty type of soil encountered within the perimeter, with their small pore predominance, moderate to low permeability and medium water holding capacity, influence sequential extraction fractions levels (Ouadjenia-Marouf et al., 2013).

3.3.7. Implications for Mining Environmental Management

This study contributed to the highlighting of gold mining site contamination and used multiple methods to investigate the contamination (total analysis and sequential analysis of trace and minor elements). It also used two different pollution assessment methods (enrichment factor and geo-accumulation index) for contamination evaluation. In addition, the use of multiple methods for potentially toxic element contamination level evaluation and distribution could be adopted by other researcher in evaluating potential site contamination. Further comparison to selected standards dictated the potential use of the soil. These findings could be used as decision-making criteria for the further use of the site and the remediation level needed (Ashraf et al., 2019; Varedaa et al., 2019). Mining site remediation strategies and the financial assets required could be derived from this study. It could also be used by local government technicians and the local population in the rehabilitation of the mining site. Selection of plants capable of growing in this specific environment is a prerequisite if phytoremediation strategies are to be employed.

3.4. Conclusions

Soil screening by analyzing total potentially toxic element (namely As, Cd, Co, Cr, Cu, Ni, Mn, Pb, Zn) content interpreted using the enrichment factor and geoaccumulation index, as well as sequential extraction, revealed soil contamination. The study showed that potentially toxic element concentrations in the mining soils vary with different mining activities. Higher degrees of contamination mostly occur around the processing plant and the TSF in Zone 1 and in Zone 5 covering the old artisanal mining locations. The main causes of contamination were anthropogenic (associated with chemical use for gold lixiviation, waste management, vehicle, and fuel use). Actions should be taken for the remediation of the mining site prior to handing it over to local population.

Chapter 4. Potentially Toxic Element Content in Cereals and Exposure Risk Assessment

This chapter is based on the publication:
Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (2019b). Metals and metalloid content in cereals from a gold mining site in Burkina Faso and intake risk assessment. Journal of Environmental Management, 248, 109292. http://dx.doi.org/10.1016/j.jenvman.2019.109292

ABSTRACT: Cereals grown close to mining sites could contain high levels of potentially toxic elements which could jeopardize local population health through intake of those crops. This study investigated for the first time the concentration of potentially toxic elements, namely arsenic, cadmium, cobalt, chromium, copper, iron, manganese, nickel, lead and zinc in four types of cereals (two of maize and two of sorghum) grown within the perimeter of a gold mine and at three surrounding villages in Burkina Faso. A total of 47 samples were collected. Cereal consumption surveys in those villages were undertaken to evaluate the intake hazard. Average arsenic content trend was Site (0.31±0.56 mg kg^{-1}, dry weight (dw)) > Songo (0.18±0.17 mg $kgdw^{-1}$) > Sighnoguin (0.15±0.10 mg $kgdw^{-1}$) > Youga (0.10±0.00 mg $kgdw^{-1}$); subsequently, the average estimated daily intake of arsenic followed this pattern: Site > Songo > Sighnoguin > Youga with 1.93, 1.08, 0.89 and 0.63 µg $kgbw^{-1}$ day^{-1} respectively which all fall below a target hazard quotient of 1. Non-parametric Kruskal-Wallis tests confirmed significant difference of Co, Cu, Fe, Mn and Ni between locations whilst no significant differences were found for As, Cd, Pb and Zn. Considering cereals types, yellow corn from the mine site exhibited As value higher than the referential dose (2.14 µg $kgbw^{-1}$ day^{-1}) and consequently a target hazard quotient of 1.97. This finding indicates that there is an intake risk to the local population from dietary intake. Contamination by As could be linked to mining activities on parent rocks that contain As with spread by wind to Songo and Youga. Sighnoguin village is more subjected to contamination by agricultural practices. Decontamination of the site and selection of cereals with low uptake capability and some changes to agricultural practices could reduce the hazards.

4.1. Introduction

Potentially toxic elements are inherent in the terrestrial crust and are present naturally in the topsoil in some locations or are distributed by anthropogenic means (Liu et al., 2018). The presence of these elements in soil has a negative impact on human health through direct intake or consumption of products which accumulate the elements (Sharma et al., 2018). Mining activities have been identified by many researchers as the principal contributing factor in the spread of contaminants (Eijsackers et al., 2017; Tepanosyan et al., 2018; Zhou et al., 2018; González-Fernández et al., 2018). The target minerals for mining are complexed with toxic elements which need to be removed by chemical treatment, creating significant levels of waste that requires disposal. Contaminants end up in nearby water bodies and on farms with the inherent consequences (Soyol-Erdene et al., 2018). Mobilization of potentially toxic elements by water follows the water runoff, while mobilization by wind follows wind direction, with influencing characteristics including wind speed, particle size and obstacles. Potentially toxic elements accumulation in agricultural soils around mining sites, resulting in uptake by cereals, is concerning because of the potential health risk to the local population. Many researchers have identified cereals from contaminated soils as a source of contaminant intake by populations (Zhuang et al., 2009a; Islam et al., 2015; Kwon et al., 2017). And the studies by Xu et al. (2013) and Cheyns et al. (2014) clearly revealed mining impact on foodstuffs.

Cereals are the main component of the diet of the population of Burkina Faso (Vom Brocke et al., 2010), and so contamination of cereals will be a public health concern. To avoid health effects when consuming contaminants, international institutions and organizations set levels for the acceptable daily intake (ADI) or provisional weekly tolerable intake (PWTI) (Cuadrado et al., 2000; EFSA, 2009; EFSA, 2010; FAO/WHO, 2011; EFSA, 2015; Filippini et al., 2018). In West Africa, sorghum is particularly appreciated because of its good adaptation to environmental conditions and in rural areas, it is a major source of nutrients (energy, protein, vitamins and minerals) in human food (Guindo et al., 2019).

Ten years mining operations in addition to several years of exploration campaigns impacted the site. In addition, depletion of the ore deposit is expected in the near future. Loss of employment of local population and migrants could lead to a return to agriculture in the mining site or in the close vicinity. Cereals grown in the mining site or in the vicinities could contain high levels of potentially toxic elements which could jeopardize local population health through intake of those crops. Yet, no potentially toxic element related health issues were reported so far by local health centers but concerns on possible uptake of contaminants by cereals arise and need investigation. This study is part of research focused on assessing the impact of mining on biotic and abiotic environments and on the population, looking at solutions based on phytoremediation. The chapter 3 and 5 investigated soil quality in the mining perimeter and water from the pit lakes revealed an anthropogenic contribution reflecting the parent rock type mined in the zone and mechanical (wind) and chemical (water) mobilization of potentially toxic elements.

The objectives of the present study are to (1) investigate potentially toxic elements content in cereals grown within the gold mine site and compare them to those from the vicinity, (2) assess the potential exposure of the local population, (3) determine the potential use of the mine site perimeter in the after-closure stage and (4) specify the need for remediation prior to handing over the site to the local population.”

4.2. Materials and Methods

4.2.1. Site Under Investigation

The site under investigation is described in section 2.6. The climate is a tropical savannah type with an annual rainfall average of 1100 mm. The 2017 rainfall was 1200 mm with 55 days of rains. The average temperature of the region is 37°C, ranging from 18 to 40°C. The dominant wind is the dry dust-laden Harmattan wind (Ribolzi et al., 2006) that blows from the north-east during the dry season and from the south-west during the wet season (Figure 4-1). There are six villages within a radius of 11 km from the processing plant: Youga, Gonsé, Wilgo, Songo, Youguego and Sighnoguin. Only the three closest villages to the mine were considered for the cereals sampling and analyzing, i.e. Sighnoguin (8 – 14 km); Songo (3 – 8.7 km) and Youga (5 – 9 km).

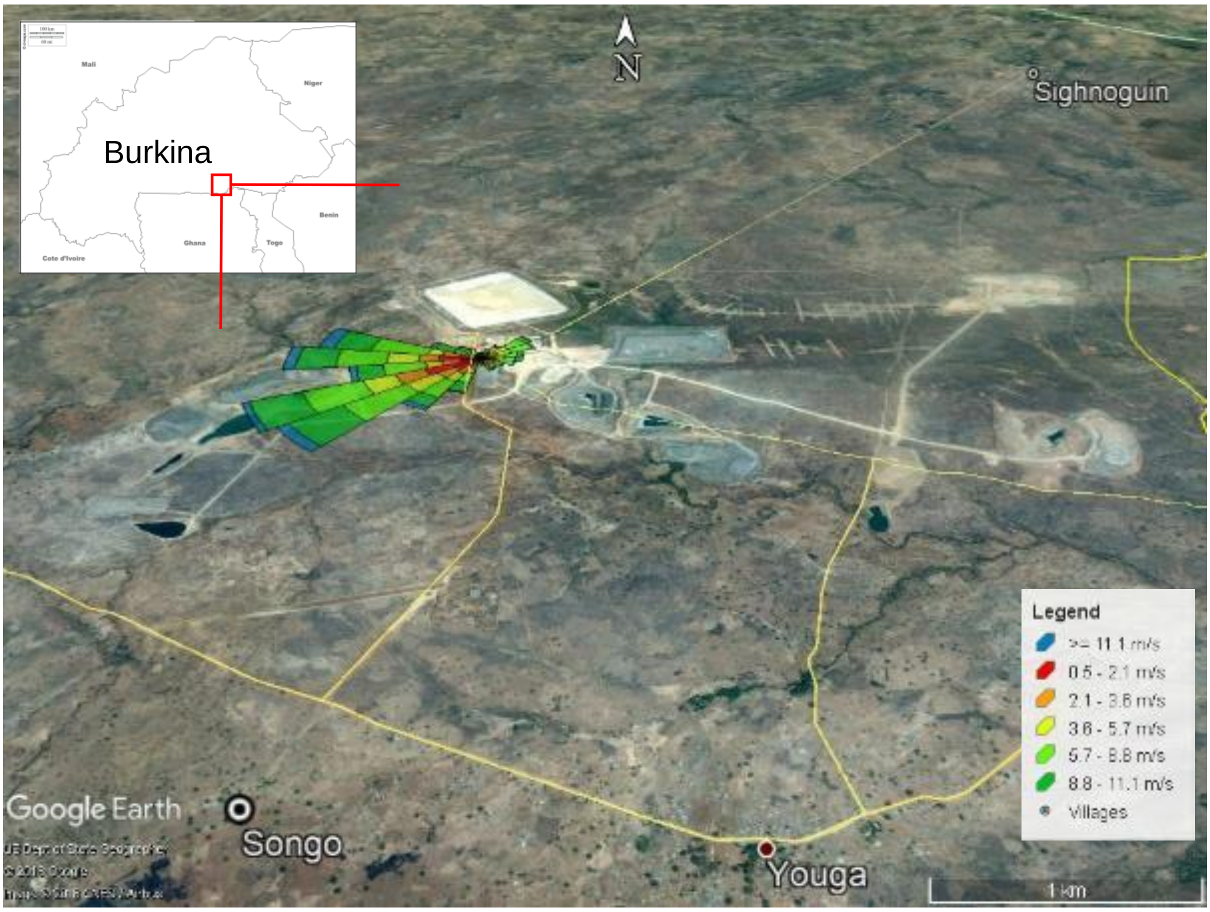


Figure 4-1 The three closest villages from each side of the mining site and Harmattan wind rose representation

4.2.2. Cereal Sampling

Cereals cultivated in the rainy season were sampled from farms inside the mining perimeter and from the nearby villages. The two common cereals species, sorghum (Sorghum bicolor) and maize (Zea mays) were sampled for potentially toxic elements analysis. In Sighnoguin village, 11 samples were taken representing 4 red sorghum, 4 white corn and 3 yellow corn. In Songo village, a total of 9 samples were taken, 5, 1 and 3 for white corn, white sorghum and yellow corn respectively. In Youga village, a total of 12 samples were taken, 4 for each type of cereals (white corn, white sorghum and yellow corn). Within the mine perimeter, 15 cereal samples were taken directly from the farms, 2 white corn, 2 yellow corn and 11 white sorghum. Fifteen samples represented all sorghum and corn farms in the perimeter.

From each farm, one sample was considered by taking a number of cobs and mixing them together to make a representative sample (Huang et al., 2008). These samples, taken directly from the farm, were sun-dried, stored in plastic bags and sent to laboratory for analysis. For farms inside the perimeter, coordinates were taken at the sampling point. Outside the mine perimeter, cobs or grains samples were obtained from farmers in the three considered villages (Sighnoguin, Songo and Youga) of the total six villages and went for the same treatment as samples from the mining perimeter. Harmattan winds were plotted based on wind direction and wind speed and transposed onto the Google map of the site (Figure 4-1).

4.2.3. Local Community Cereal Intake Survey

A survey was carried out to estimate cereal consumption rates. Ten per cent (10%) of the households were randomly selected for the survey, equivalent to 73 households on a total of 714 households in the six villages surrounding the mining site. The survey consisted of going to the selected households and completing a questionnaire of seven questions all about cereals and fish consumption: frequency, provenance, quantity consumed, preparation. Questionnaires were anonymously filled by an experienced interviewer (Table 4-1).

The characteristics of the questionnaires and the techniques of survey were based on recommendation made by Conforti et al. (2017), especially their “Types 3” approach for obtaining accurate data on the measurement of food consumption. Using this approach, the survey reported on food actually consumed, and whether that same food was purchased or own-produced (Friedman et al., 2017). Local units were used for the quantification and later on converted to international units. The survey took place during the dry season of 2017.

Table 4-1 Questionnaire for cereals and fish intake survey in local community

1	Localité	Date	Nombre/consession	Sexe repondant
				M ☐ F ☐
1	Consommez vous des cereals ?	Oui ☐	Non ☐	
1.1	Combien de fois en manger vous ?	1 ☐	2 ☐	3 ☐
1.2	D'où provient ils ?	Champ ☐	Marché ☐	Autre ☐
1.3	Quelle quantité en consommez vous ?		Peser du recipient :	
2	Consommez vous des poissons ?	Oui ☐	Non ☐	
2.1	Combien de fois en manger vous ?	1 ☐	2 ☐	3 ☐
2.2	D'où provient t-ils ?	Peche direct ☐	Marché☐	Fosses☐
2.3	Comment vous les manger ?	Frais☐	Sec☐	Autres
2.4	Quelle quantité en consommez vous ?	Par jour ( )	Par semaine ( )	
Remarque :				

4.2.4. Cereals Potentially Toxic Elements analysis

The method described by Jha et al. (2017) was used for the analysis of total PTE in cereals. The samples were digested using a Milestone Microwave Laboratory System (Milestones Inc, Monroe, CT, USA). In short, 0.5 g of cereals was weighted in a Teflon vessel for the microwave digestion process; 4 mL of nitric acid and 2 mL of hydrogen peroxide were added and placed in the microwave for a digestion program of 5 min at 450 W followed by 600 W for 10 min. After cooling, the digests were diluted to 50 mL. These samples were analyzed immediately or stored in a refrigerator at 4°C until they were analyzed.

Calibration standards were prepared as described in 3.2.2.2. Metal-based recipients were avoided during the sequence of sampling, handling, processing and analyzing. Cereals samples were analyzed as taken from farms without processing, although a study revealed that food processing practices may have positive impact on PTE availability and toxicity (Hajeb et al., 2014). Prior to analysis, the cereal samples were thoroughly washed with tap water in the lab, then with nitric acid 10% and three times with bi-distilled water in order to remove any dust that might have settled on the samples in the field or during sun-drying.

Potentially toxic elements in cereals were analyzed using an ICP 7200 equipped with Qtegra Software (Thermo Scientific™ iCAP™ 7000 Plus Series ICP-OES, Thermo Fisher Scientific Brand, USA) and a CETAC AXP 560 autosampler (Teledyne Technology, USA).

The soil to cereal contaminant transfer factor was assessed. The transfer factor was computed using the formula below in which C_{soil} = the average contaminant concentration in the soil from the site and $C_{cereals}$ = the concentration of the respective contaminant in cereals from the site.

$T_F = {}^{C_{cereals}}/_{C_{soil}}$ C_{soil} and $C_{cereals}$ are in the same unit. T_F = transfer factor (Equation 3)

4.2.5. Potentially Toxic Element Exposure Assessment

Estimated Daily Intake (EDI) and Target Hazard Quotient (THQ) were used to estimate the health risks related to consumption of cereals (Zhuang et al., 2009a; Islam et al., 2015). For the purpose of risk assessment, samples with values beneath the limit of detection were replaced by values as half the limit of detection. Referential doses used for the calculation was recorded in Table 2-3. Interpretation of the data was done by comparison with New Zealand, Netherlands and FAO standards, as there are no national nor regional guidance values available. Those standards were selected to cover all PTE analyzed.

The **EDI** was computed based on the concentration of potentially toxic elements in cereals, the daily cereal consumption rate and the average body weight of an individual consumer. Average body weight was taken to be 70 kg.

$\mathrm{EDI} = \ (C\_Food \times Food\ daily\ intake)/W$, (Equation 4)

C_{Food} = Concentration of chemical contaminant in the cereals (mg kg^{-1} or ppm)

$Cereals\ daily\ intake$ = mean daily consumption rate of cereals (kg day^{-1}),

W = Average body weight of an individual consumer (70 kg),

The **THQ** was estimated based on the formula below which considers the food intake rate by the population, the average body weight and Referential dose of the respective potentially toxic elements.

$\mathrm{THQ} = EDI/RfD$, (Equation 5)

EDI = $Estimated\ daily\ intake$

RfD= Tolerable daily intake value for each element (g kg^{-1} body weight day^{-1}).

Hazard index (HI) values were computed in order to evaluate the overall potential for non-carcinogenic effects of combined potentially toxic elements. HI was computed as the sum of THQ of individual element (Huang et al., 2008).

4.2.6. Quality Control and Statistical Analysis

The method used for the analysis of the cereal samples was validated. The validation consisted of verification of the applicability and fit for the purpose of the method by selecting a proved one. The validation also included the verification of the specificity and selectivity of the method and calibration investigation which involved range, sensitivity and detection limits. The accuracy and performance were checked through the recovery rate and use of referential sample. Reagent blank and spiked samples were prepared for method precision and recovery checks. Reagent blanks were measured prior to sample analysis and values were subtracted from sample values. Two recovery tests were included and the average used. Water acidified with nitric acid at 1% was used as the calibration blank. The method detection limit (MDL) was determined as in 3.2.3. and statistical analysis was done as per the description in 3.2.5. Additionally, a non-parametric (Kruskal-Wallis) test was used. Principal component analysis (PCA) was used to obtain the detailed information of the dataset and gain insights into the distribution of potentially toxic elements by detecting similarities or differences between samples. Agglomerative hierarchical clustering (AHC) with Spearman correlation were used to look for cereals samples patterns according to location, cereals types and potentially toxic elements investigated. Weather data were acquired by a stand-alone weather station (ultra-precision weather station, WRM300A, Oregon scientific, USA) and was used with Lakes Environmental software for the wind rose plots of meteorological data (WRPLOT View™, Version 8.0.2) and projected onto Google Earth. Results of potentially toxic elements are expressed in mg kg^{-1} dry weight or as otherwise stated. Values were expressed as the mean ± standard deviation. XLSTAT-Base (Addinsoft, V19.7, 2018) were used for PCA and AHC.

4.3. Results

4.3.1. Characteristics of the population

The survey covered seventy-three households from the six villages in the vicinity of the mine. Men (43.8% (32)) and women (56.2% (41)) responded to the survey. The number of persons per household ranged from 1 to 47 persons with the range of 6 to 10 being the most common. The order between villages – based on average numbers per household – was as follows: Sighnoguin (16±13) > Gonsé (14±6) > Songo (12±7) > Wilgo (10±8) > Youga (8±5) > Youguego (7±3). The number of persons per household was significantly different from one village to another $F(5, 139.471) = 2.702$ ($p<0.05$). Bernhard & Hoffmann (1999) reported similar population characteristics for the rural area of Burkina Faso: 26.2% of households with 2-10 persons, 40.8% of households with 11-20 persons, 24.6% of households with 21-30 persons and 8.5% of households with more than 30 persons.

4.3.2. Local Population Cereal Consumption Rate

Cereal consumption rates were estimated; the results are shown in Table 4-2. Cereal consumption rates were numerically in this order: Gonsé) < Youga < Sighnoguin < Youguego < Wilgo < Songo (Table 4-2). The cereal intake average of the six villages was 0.43±0.19 kg day^{-1} person^{-1}. An ANOVA test revealed that there was no significant difference between the cereal intake averages from the villages: $F(5, 66) = 2.138$ $p = 0.072$. National cereals consumption is 0.52 kg day^{-1} person^{-1} for the year 2016/2017 (INSD, 2017). Ceareals intake is expressed in dry weight.

Table 4-2 Population data, survey sample numbers and cereal intake rates (kg person^{-1} day^{-1}) given as mean ±standard deviation (sd) and range.

Villages	Population data and survey sample numbers			Cereal intake rates	
	Total population in village	Number of households participating in survey	Number of persons participating in survey	Mean±sd	Range
Gonsé	290	10	133	0.33±0.15	0.13-0.60
Youga	1153	12	95	0.34±0.16	0.17-0.80
Wilgo	976	11	107	0.48±0.21	0.27-0.86
Sighnoguin	610	13	202	0.43±0.18	0.22-0.76
Youguego	499	14	101	0.44±0.21	0.22-1.00
Songo	1526	13	149	0.53±0.15	0.30-0.83
Total	5054	73	787	0.43±0.19	0.13-1.00

The food intake survey revealed that six types of cereals or leguminous crops were consumed by local people: millet, white and red sorghum, rice, maize, and beans with sorghum and maize the most consumed cereals which will be sampled for analysis in this study. The survey showed that only in a minority of cases (8% in Youga, 8% in Songo, zero in the other villages), households don't do farming and purchase 100% of their food; in all other cases households relied to some degree on cultivation for food production (Table 4-3). For this latter category, the bottom two lines in (Table 4-3) shows the ratio between the amount of cereals purchased versus the amount of cereals obtained by farming.

Contrary to nationwide cereals production figures, in this region (Centre Est) of 14 710 km² and 1 515 518 inhabitants (2016), enough rainfall (up to 1100 mm) spread over a long period of time is favorable for good cereals production. Cereals production in the region for the season 2016/2017 was 422 223 tons with corn and sorghum the most produced, with 121 824 and 160 907 tons respectively, covering 113.2% of cereals demand (INSD, 2017).

Table 4-3 Cereals supply conditions, expressed in %

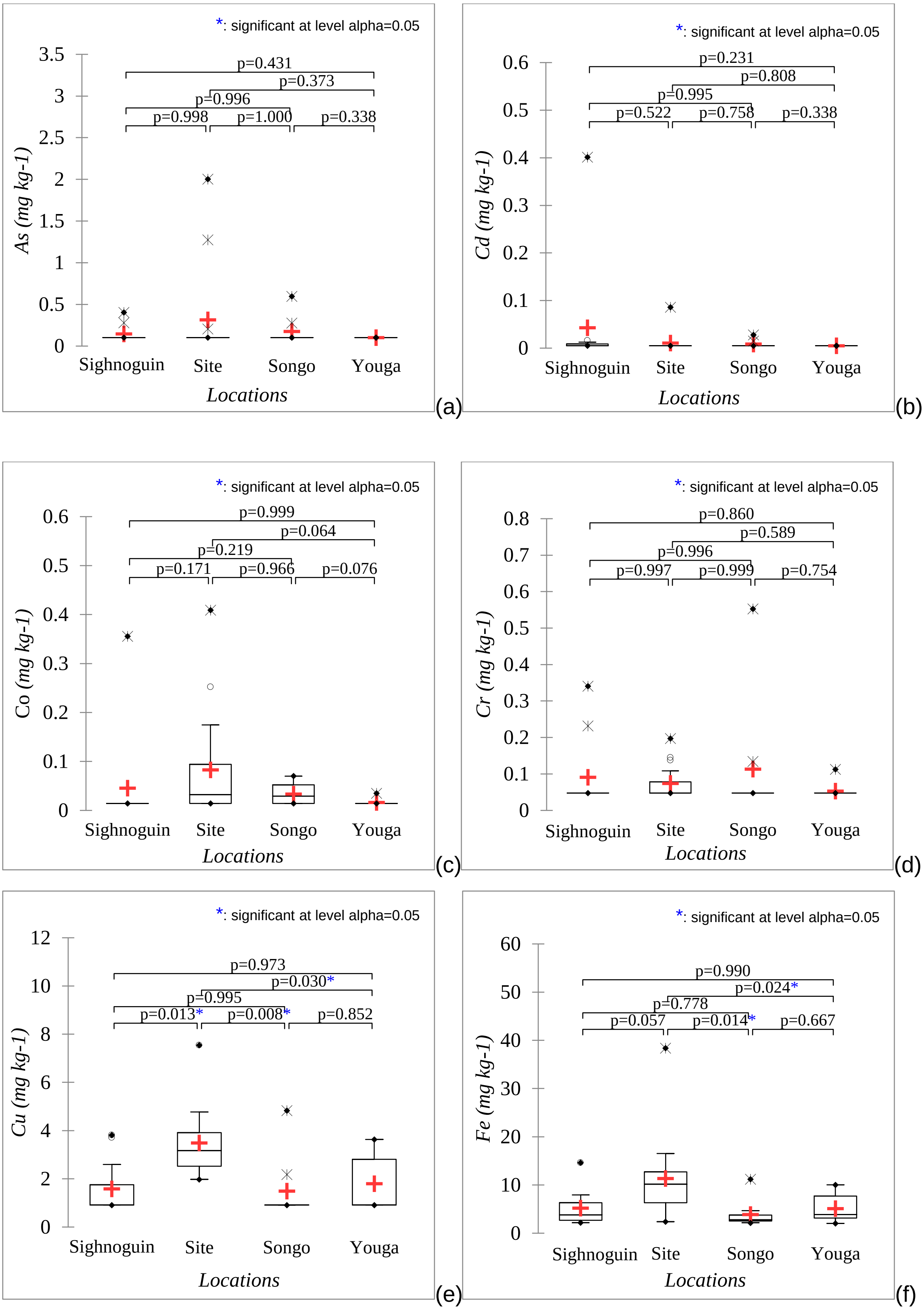
Supply conditions	Gonsé	Youga	Wilgo	Sighnoguin	Youguego	Songo
All purchased	0	8	0	0	0	8
Cultivated	100	92	100	100	100	92
Self-sufficient farming	30	0	36	46	7	15
Additional purchase	70	92	64	54	93	85

Cereals samples from the three closest villages were further considered for analysis.

4.3.3. Potentially Toxic Element Analysis in Cereals Samples

Potentially toxic elements in cereals samples from the three villages (Sighnoguin, Songo and Youga) and from the mining perimeter analysis results were displayed in Figure 4-2. The average of potentially toxic elements by location shows that cereals from the mining site have the highest values for six out of the ten elements investigated (As (0.31±0.56 mg/kg), Co (0.08±0.11 mg/kg), Cu (3.49±1.40 mg/kg), Fe (11.36±8.57 mg/kg), Mn (9.74±3.56 mg/kg) and Ni (0.89±0.57 mg/kg)) while Songo exhibited the highest values for Cr (0.11±0.17 mg/kg) and Zn (26.40 ± 10.84 mg/kg) and Sighnoguin for Cd (0.04±0.12 mg/kg) and Pb (4.16±9.80 mg/kg). Average potentially toxic elements values by location exhibited this pattern: Mine > Songo > Youga and Sighnoguin. Non-parametric Kruskal-Wallis tests confirmed significant difference of Co, Cu, Fe, Mn and Ni between locations. The content of As in all cereals samples did not exceed 1 mg kg^{-1} as set by the New Zealand regulation (ANZFS, 2011). Cd did not exceed the 0.1 mg kg^{-1} set by Codex (1995). Zn levels in cereals samples ranged between 19.54±4.36

mg kg^{-1} and 26.40±10.84 mg kg^{-1} (Table 4-4). The highest value for Pb content in cereals investigated exceed the limit value of 0.2 mg kg^{-1} for cereals (Codex, 1995).



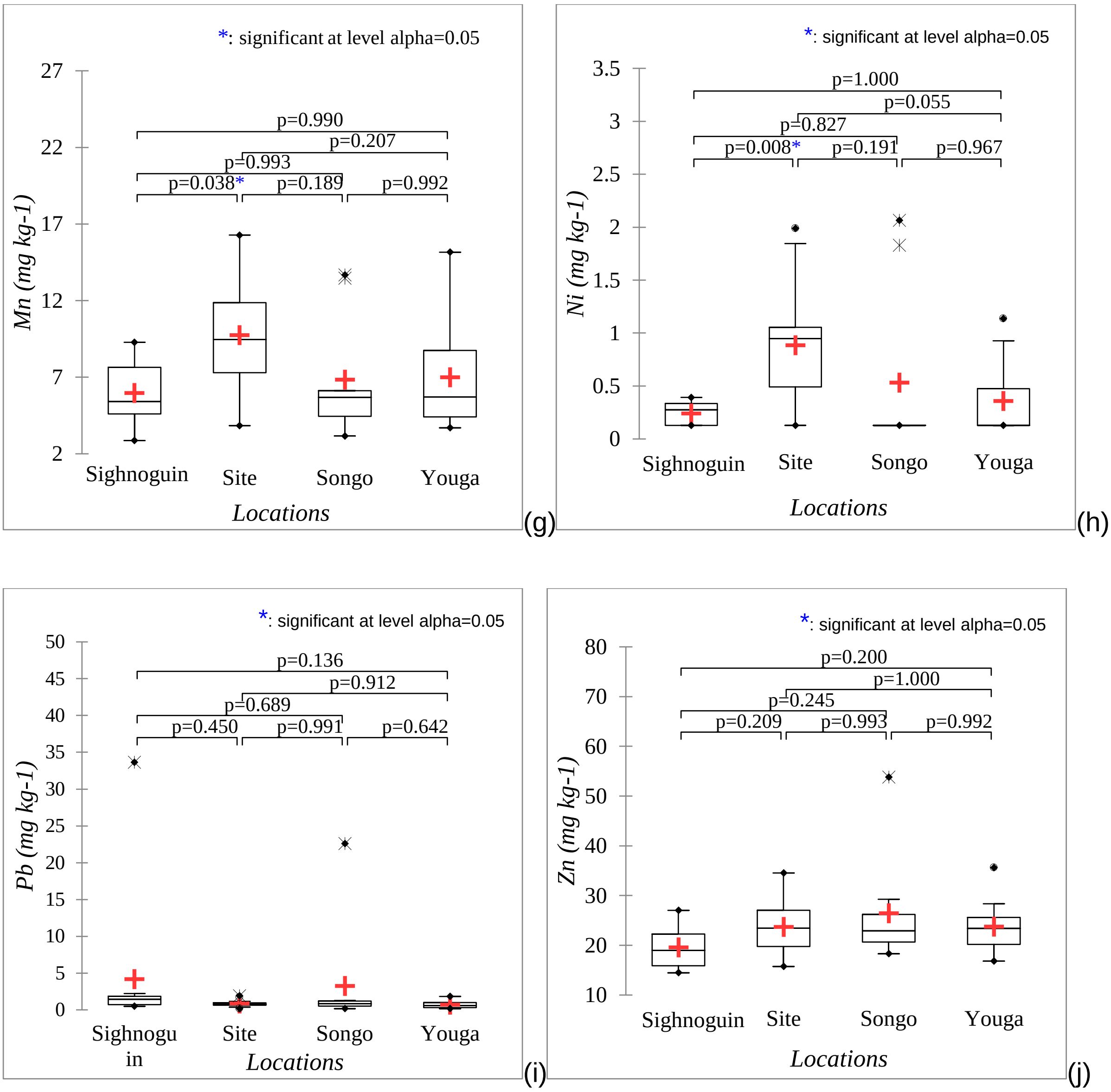


Figure 4-2 Boxplot of potentially toxic element contents (expressed in mg kg^{-1}, dry weight) according to the four locations considered (Site, Sighnoguin, Songo and Youga). As (a), Cd (b), Co (c), Cr (d), Cu (e), Fe (f), Mn (g), Ni (h), Pb (i), Zn (j). p values and *significance (< 0.05) level included, + for mean, ⁎ for outliers.

Table 4-4 Potentially toxic elements in cereals samples, values are in mg kg^{-1} dry weight. RS means red sorghum, WC means white sorghum, YC means yellow corn, WC means white corn. SD for standard deviation.

		Sighnoguin (8 – 14 km)				Site < 1 km				Songo (3 – 8.7 km)				Youga (5 – 9 km)				Overall mean
		RS	WC	YC	**Mean**	WC	WS	YC	**Mean**	WC	WS	YC	**Mean**	WC	WS	YC	**Mean**	
As	Mean	0.10	0.10	0.26	**0.15**	0.10	0.28	0.69	**0.31**	0.14	0.10	0.27	**0.18**	0.10	0.10	0.10	**0.10**	**0.19**
	SD	0.00	0.00	0.15	**0.10**	0.00	0.57	0.83	**0.56**	0.08	0.00	0.28	**0.17**	0.00	0.00	0.00	**0.00**	**0.33**
Cd	Mean	0.01	0.01	0.14	**0.04**	0.01	0.01	0.05	**0.01**	0.01	0.01	0.01	**0.01**	0.01	0.01	0.01	**0.01**	**0.02**
	SD	0.00	0.01	0.23	**0.12**	0.00	0.00	0.06	**0.02**	0.01	0.00	0.01	**0.01**	0.00	0.00	0.00	**0.00**	**0.06**
Co	Mean	0.01	0.01	0.13	**0.05**	0.13	0.04	0.29	**0.08**	0.03	0.01	0.05	**0.03**	0.01	0.01	0.02	**0.02**	**0.05**
	SD	0.00	0.00	0.20	**0.10**	0.17	0.04	0.17	**0.11**	0.02	0.00	0.02	**0.02**	0.00	0.00	0.01	**0.01**	**0.08**
Cr	Mean	0.09	0.05	0.15	**0.09**	0.12	0.07	0.05	**0.07**	0.05	0.13	0.22	**0.11**	0.05	0.06	0.05	**0.05**	**0.08**
	SD	0.09	0.00	0.17	**0.10**	0.11	0.04	0.00	**0.05**	0.00	0.00	0.29	**0.17**	0.00	0.03	0.00	**0.02**	**0.09**
Cu	Mean	2.34	1.33	0.91	**1.59**	2.19	3.88	2.66	**3.49**	0.91	2.18	2.22	**1.49**	1.59	2.47	1.35	**1.81**	**2.23**
	SD	1.65	0.84	0.00	**1.19**	0.30	1.45	0.21	**1.40**	0.00	0.00	2.26	**1.32**	1.36	1.20	0.88	**1.17**	**1.52**
Fe	Mean	8.80	3.16	3.13	**5.20**	3.61	14.05	4.34	**11.36**	2.60	11.22	3.63	**3.90**	2.75	8.72	3.88	**5.12**	**6.90**
	SD	3.98	1.55	0.59	**3.70**	1.72	8.53	0.12	**8.57**	0.29	0.00	1.13	**2.85**	0.78	1.10	0.53	**2.81**	**6.20**
Mn	Mean	8.04	4.30	5.43	**5.97**	7.78	10.66	6.60	**9.74**	4.85	13.68	7.86	**6.84**	4.58	11.18	5.20	**6.99**	**7.60**
	SD	1.85	1.14	1.17	**2.15**	4.36	3.26	3.90	**3.56**	1.20	0.00	4.90	**3.93**	0.60	2.91	1.32	**3.54**	**3.59**
Ni	Mean	0.22	0.25	0.26	**0.24**	0.34	1.08	0.35	**0.89**	0.13	1.83	0.77	**0.53**	0.13	0.78	0.17	**0.36**	**0.53**
	SD	0.10	0.14	0.12	**0.11**	0.09	0.54	0.32	**0.57**	0.00	0.00	1.12	**0.80**	0.00	0.32	0.08	**0.35**	**0.56**
Pb	Mean	1.83	0.64	11.97	**4.16**	0.63	0.95	0.54	**0.85**	5.17	1.17	0.63	**3.21**	0.55	1.15	0.40	**0.70**	**2.04**
	SD	0.33	0.13	18.77	**9.80**	0.27	0.50	0.23	**0.46**	9.75	0.00	0.53	**7.28**	0.44	0.48	0.35	**0.51**	**5.70**
Zn	Mean	15.28	21.49	22.61	**19.54**	20.90	23.56	27.05	**23.67**	21.36	29.25	33.85	**26.40**	25.53	20.98	24.68	**23.73**	**23.24**
	SD	0.86	3.78	3.78	**4.36**	3.57	5.61	1.67	**5.13**	3.19	0.00	17.29	**10.84**	7.72	2.02	3.36	**4.97**	**6.62**

Principal component analysis (PCA) was computed. PCA of mine site samples (Figure 4-3) revealed that WS was relatively correlated to Pb, Ni, Fe, Cu Mn, and Zn which highlights its tolerance to those elements as stated by Zhuang et al. (2009b) and Jia et al. (2017). YC is correlated with Cd, As, and Co which reflects a tendency of this cereal to contain these elements. WC is more subject to harm by Cr and is less correlated with the other elements investigated. PCA of all locations' cereal samples (Figure 4-4) revealed that WS had an affinity with elements such as Mn, Cu, Ni, Fe and Zn. YC had an affinity with Cd, Co, Cr and As. WC is correlated to Pb.

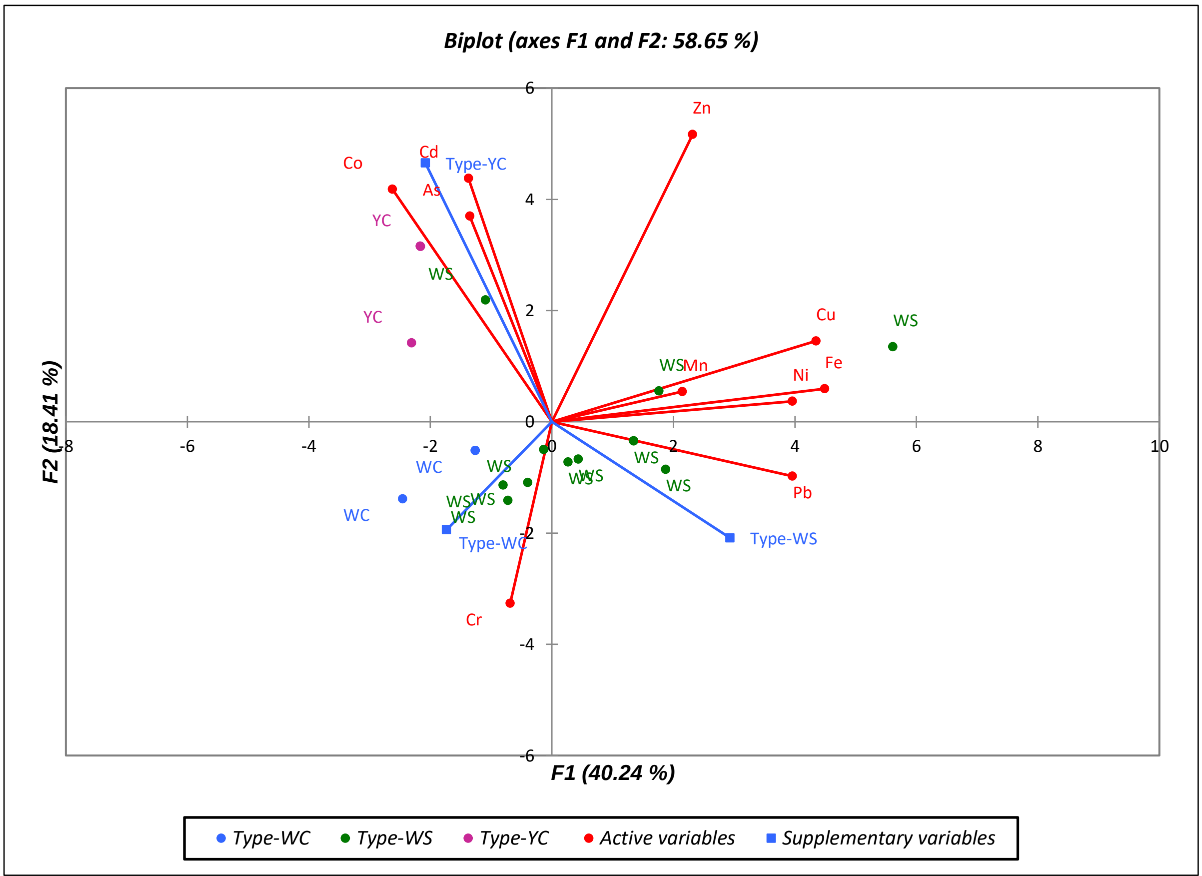


Figure 4-3 Principal component analysis (PCA) of samples from the mine site, WC for white corn, WS for white sorghum, YC for yellow corn. Color coding of samples per types (white corn (Type-WC), white sorghum (Type-WS), yellow corn (Type-YC)).

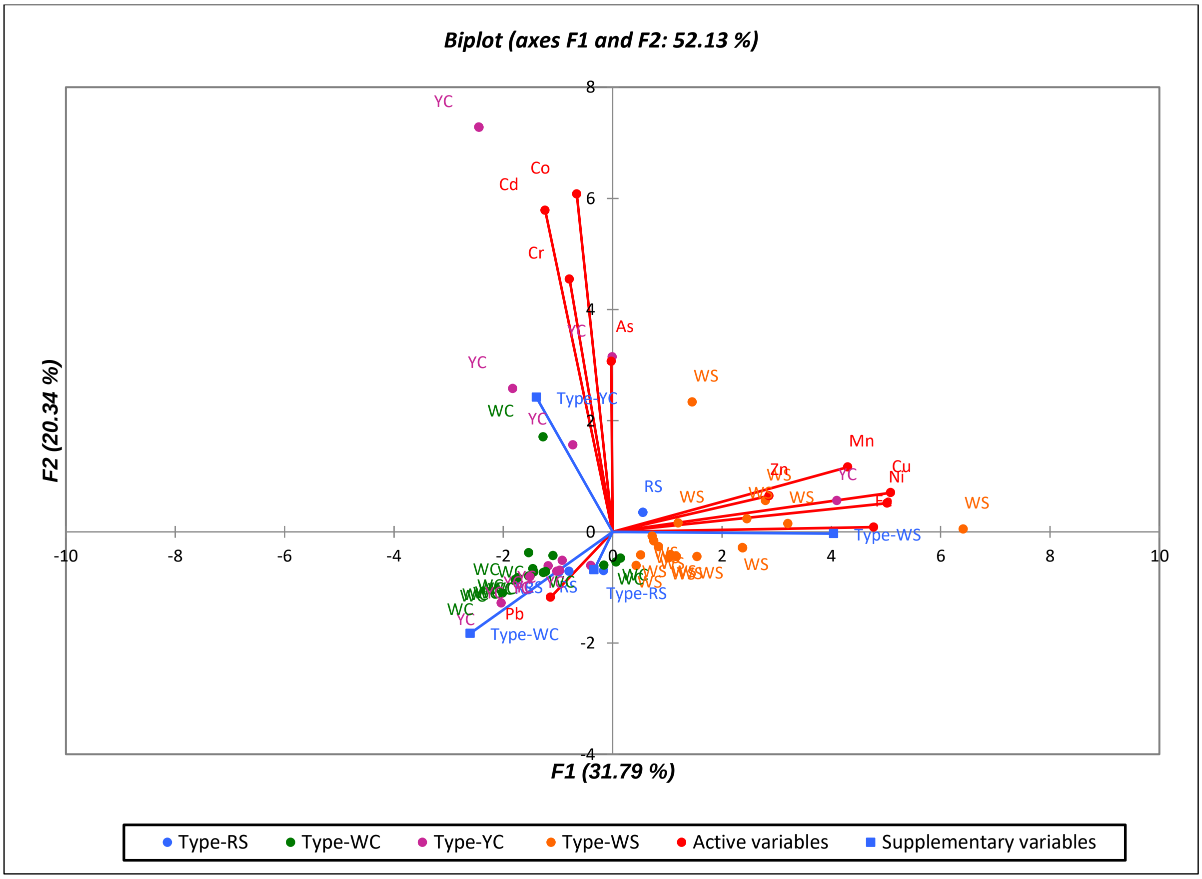


Figure 4-4 Principal component analysis (PCA) of all samples from all locations, WC for white corn, WS for white sorghum, YC for yellow corn, RS for red Sorghum. Color coding of samples per types (white corn (Type-WC), white sorghum (Type-WS), yellow corn (Type-YC), red sorghum (Type-RS)).

4.3.4. Bio-accumulation from Soils to Cereals from the Mine Site

Transfer factors from soil to cereal for the mine site were calculated based on soil average potentially toxic elements content and cereal average contaminant content by species; the results are displayed in Table 4-5. TF for As and Zn were, in increasing order: YC > WS > WC. The maximum TF was detected for Cu, Mn, Ni and Pb in WS and the maximum for Cr in WC.

Table 4-5 Transfer factors (TF, 10^{-3}) of cereals from the site, expressing the ratio of cereal to soil potentially toxic element concentrations (dimensionless).

Element	As	Cd	Co	Cr	Cu	Mn	Ni	Pb	Zn
White corn	2.29	0.71	4.55	1.10	80.4	7.72	5.96	3.57	796.4
White sorghum	6.38	0.71	1.22	0.63	142.5	10.59	18.69	5.40	897.4
Yellow corn	15.42	6.39	9.95	0.43	97.5	6.55	6.10	3.05	1030.6

These results show that some potentially toxic elements are more difficult than others to transfer, or crops have some preferential uptake. In support of these findings, a study by Memoli et al. (2017) to assess potentially toxic element compartmentalization in sorghum found transfer factors of 0.29, 0.37, 0.00, 0.00 for respectively Cd, Cu, Ni, and Pb. Arsenic content in soils and in cereals should be monitored closely as the potential for contamination exists given that mined ore contains pyrite and arsenopyrite. The presence of Galena (Pb contained rocks) in ore (Woodman et al., 2016) could also result in the Pb exposure of soil and crops in the vicinity. Lead deposits on cereals from dust could have contributed to Pb content on cereals (Ma et al., 2019), even when washing of cereals could have solved this issue. Ma et al. (2019) found that atmospheric deposition was the main source of Pb in wheat tissues. They then advised that taking measures to reduce the absorption of Pb from atmospheric deposition can effectively ensure food safety. Lead presence in the atmosphere was mainly due to mining fleet exhaust.

4.4. Discussion

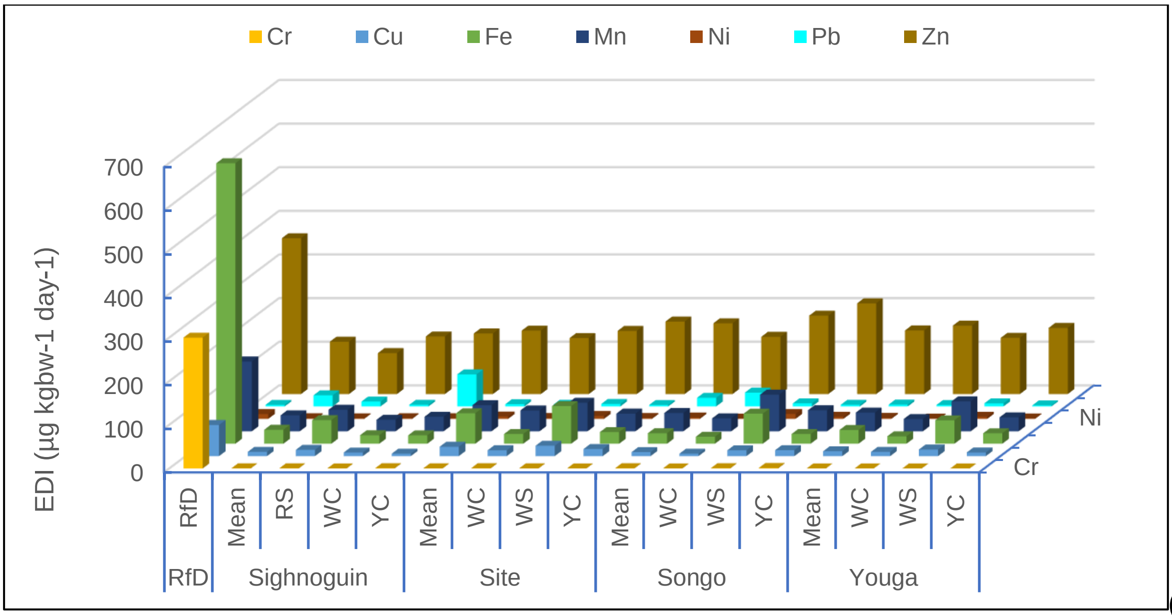
4.4.1. Cereals Contaminant Uptake

The presence and variation of potentially toxic elements in samples from the different locations could be explained by activities at the mine site, with wind mobilization to farms in the vicinity, variation in absorption and accumulation capabilities, the bioavailability of contaminants, and farming techniques such as use of fertilizers. Low-level As content in RS, WC, WS could be explained by the absence of bioavailable As from soil in those locations and also the physiological barrier to potentially toxic element transfer acting during seed maturation (Murillo et al., 1999). It is generally accepted that edible plants grown on uncontaminated or unmineralized soils contain 0.01–1.5 mg kg^{-1} As (Kwon et al., 2017). The level of Cd in Sighnoguin could be explained by the intensive use of phosphate fertilizers (Sharma et al., 2018) for market gardening along the river, which are major sources of Cd in soil. Sighnoguin is not impacted by mining activity contaminant mobilization channels. The presence of Cd at the mining site could be partly explained by years of poor management of Ni-Cd batteries (El-Kady & Abdel-Wahhab, 2018) and gold amalgamation by artisanal miners using illegal impure chemicals. The low level of Ni in sorghum is explained by a poor transfer factor of Ni from roots to aerial parts (Kaplan et al., 2005). The difference in Zn content could be explained by mining fleet tire abrasion, lubricating oil, vehicular exhaust, and brake pad degradation (Wang et al., 2017). An additional factor may be the affinity to Zn of crops like sorghum or corn used for remediation of Zn contaminated soil (Soudek et al., 2014). Pb presence in mining site samples could be justified by presence of Pb in parent rock mined (Woodman et al., 2016) and in samples from villages by ore processing by artisanal miners at their homes and poor Pb-contained solar system battery management and leaded gasoline.

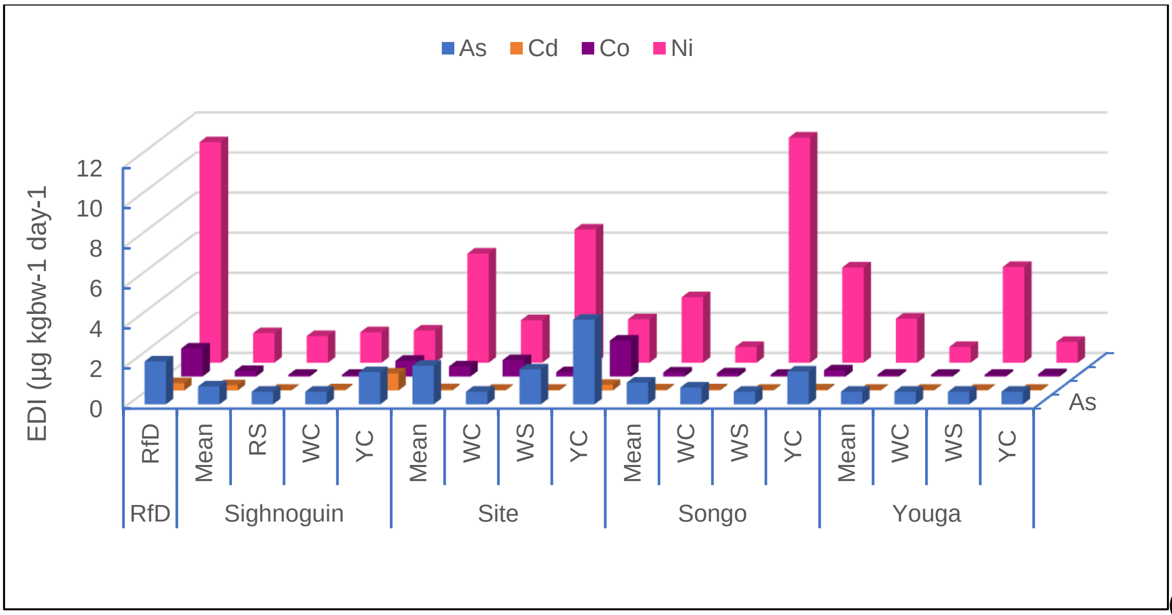
4.4.2. Intake Risk Potential to the Population

Consuming contaminated cereals can lead to health hazards. Intake risks to nearby populations were estimated based on the EDI of contaminants and individual element intake THQ. Two scenarios were used for the calculation of the EDI: the average of potentially toxic elements and a pessimistic scenario using the maximum value of potentially toxic elements content in cereals. The pattern of average EDI of potentially toxic elements was: Mine > Songo > Youga > Sighnoguin.

EDI of single potentially toxic elements according to locations are shown in Figure 4-5 and THQ in Figure 4-6. Average EDI of As followed this pattern: Mine > Songo > Sighnoguin > Youga with 1.93, 1.08, 0.89 and 0.63 µg kgbw^{-1} day^{-1} respectively which all fall below a THQ of 1. Considering cereals types and computing with the maximum values, WC and YC from Songo village, WS and YC from the site and YC from Sighnoguin show EDI of As approximatively above the referential dose. Average EDI of Cd was in this pattern: Sighnoguin > Mine > Songo > Youga and all locations had a THQ below 1. However, computing with the maximum values of contaminant level in cereals showed YC within the mining perimeter with a THQ of 1.5. Pb EDI by location followed this pattern: Sighnoguin > Songo > Mine > Youga, with TQH for all locations above 1. EDI of Ni followed this pattern: Mine > Songo > Youga > Sighnoguin. EDI of Ni consumption of WS from Songo, exceeded the referential dose. When evaluating using the maximum data, YC from Songo showed a THQ above 1.

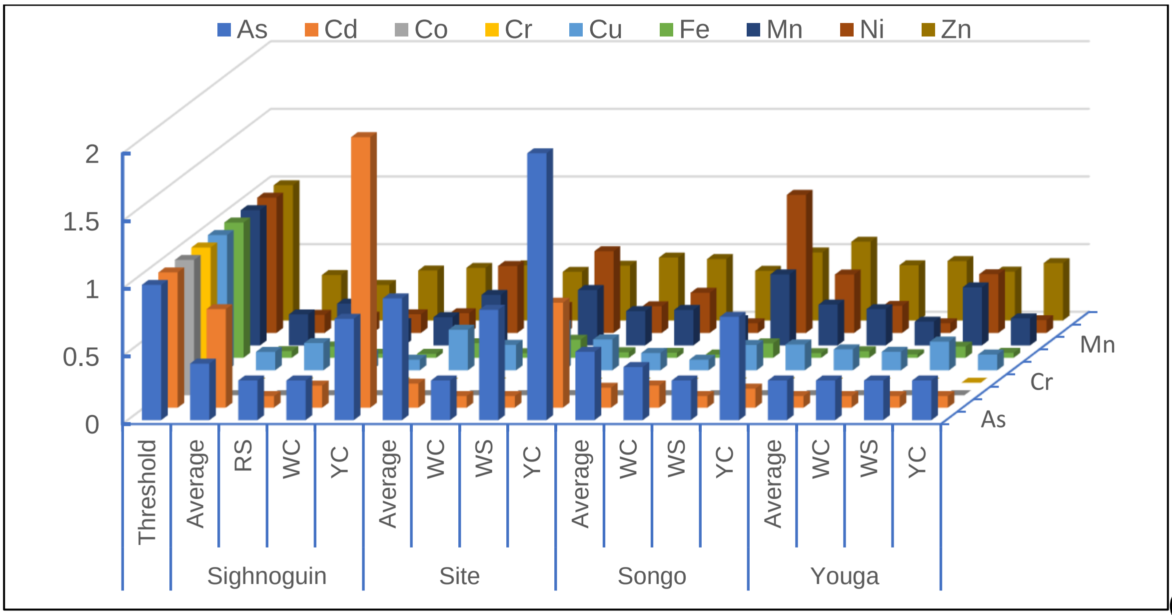


(a)

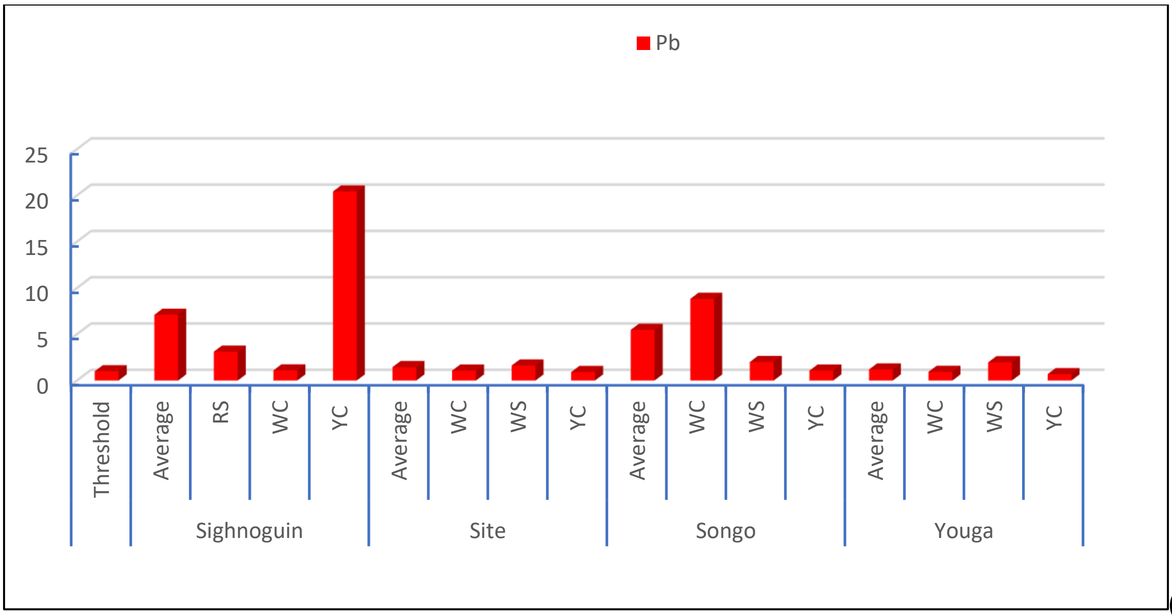


(b)

Figure 4-5 Estimated daily intake (EDI, µg kgbw^{-1} day^{-1}) of potentially toxic elements per cereals type and the average for the location, WC for white corn, WS for white sorghum, YC for yellow corn, RS for red Sorghum. Figure (a) for Cr, Cu, Fe, Mn, Ni, Pb and Zn; figure (b) for As, Cd, Co and Ni. RFD for referential dose.



(a)



(b)

Figure 4-6 Target hazard quotients (THQ) per cereals type and the average of the location, WC for white corn, WS for white sorghum, YC for yellow corn, RS for red Sorghum. (a) for As, Cd, Co, Cu, Fe, Mn, Ni and Zn. (b) for Pb.

The Hazard Index (HI) was computed to estimate the health risk when consuming the cereals. When the HI value is higher than unity, then potential health effects could raise. All locations exhibited HI more than the unity, expressing a health concern for combined toxicity. The trend Youga < Mine site < Songo < Sighnoguin was observed with 2.70, 4.30, 7.30 and 8.90, respectively. An average of 5.80 was observed. Pb contribution to the HI was 80 %, 75 %, 44 % and 35 % for Sighnoguin, Songo, Youga and the mine site respectively. In comparison, Huang et al. (2008) found for wheat growth in developed industry city in China, HI of rural adult and children of 1.09 and 1.20, respectively. Furthermore, other sources of potentially toxic elements such as water and fish from pit lakes at the mining site, could increase potentially toxic elements intake (Huang et al., 2008).

Direct toxicological effects of potentially toxic elements on living organisms can lead to inhibition of membrane function, enzyme activities, respiration and protein synthesis, as well as genetic disruptions, oxidative damage (Clemente et al., 2015). Inorganic As exposure causes skin, lungs and bladder cancer and is also associated with many non-cancerous outcomes like cardiovascular dysfunction, it could lead to immune dysfunction by hindering cellular and humoral immune response (Sattar et al., 2016). Exposure to Cd above the guidelines is of great concern and could lead to malfunctioning of renal tubular and results in the ‘itai-itai’ disease (Inaba et al., 2005). Inside organism, Pb blocks the N-methyl-D-aspartate receptor leading to impaired cognitive functions and neuronal synaptic plasticity with a disruption of memory function. Pb long term exposure is a major health issue especially for children for their growth and mental development (Tinggi and Schoendorfer, 2018). Cu, Cr, Co, Fe, Mn and Zn did not show EDI above Referential dose and could at this level of intake contribute to the good functioning of organs. Zn and Cu are essential to the normal functioning of various components of the immune system (Teklić et al., 2013). Zn deficiency results in depressed lymphocyte response to mitogens and abnormal skin hypersensitivity responses. Cu deficiency could lead to failure of the immune system to react properly to infection (Rai et al., 2019).

Contamination could be found in other agricultural products as peanuts (*Arachis hypogea* (L.)), rice (*Oriza sativa* L.), and cowpea (*Vigna unguiculata* (L.)) grown within the site or in the mining impacted areas. Use of peanut paste in sauce and increasing consumption of rice could increase health risks.

4.4.3. Managing Cereals Contaminations

Mining impact to cereals grown within and nearby the perimeter was revealed for cereals investigated and according to potentially toxic elements considered. EDI above referential dose thus TQH more than 1 and HI also above unity call upon proactive solutions to reduce cereal contaminants content and post-harvest solutions to mitigate effect on human beings. Removal of potentially toxic elements in soil should be done (Sharma et al., 2018) by the mining company, preferably using phytoremediation strategies, all along their exploitation and closure stages until substantial results have been obtained (Yao et al., 2012; El Rasafi., et al 2017). Avoidance of using impacted sites should be considered when pollution could lead to health effects on the local population. Farming strategies which could reduce uptake by cereals should be disclosed to farmers, such as the techniques described by Marchiol et al. (2007) and Del Coco et al. (2019) who discovered that the provision of organic amendment in a contaminated soil lowers Cd concentration in sorghum roots. Silica was reported by Greger et al. (2016) to reduce Cd uptake in wheat. Selection of crops with low potentially toxic elements uptake capability (Rosas-Castor et al., 2014) should be done and shared with the local population. Local environmental and agricultural services and local responsibles should play an important role in following local farmers. Close monitoring of local population health should be included in recommendations of the environmental impact assessment study and enforced closely by local and central government.

Management of industrial sites and probable impacted areas should involve industries, central and local government, local responsible, local technical services and health specialists. Mining impact monitoring including land, water, fish and cereals contamination should be enforced in mining impact assessment studies in developing world as local government and population can not afford these analyses. Furthermore, impact study findings should be used for local population awareness raising (Sana et al., 2017). Actions and technologies to minimize mining impact by preventing pollution and its dissemination must be encouraged by all stakeholders.

4.5. Conclusions

This study consisted of a dietary intake survey of local populations living near a gold mine in south-eastern Burkina Faso, cereals potentially toxic elements analysis (As, Cd, Cr, Cu, Co, Fe, Ni, Mn, Zn, Pb) and subsequently exposure assessment. The study investigated four types of cereals (red and white sorghum and white and yellow corn) grown in four locations (Mine site itself and three surrounding villages Songo, Youga and Sighnoguin). For individual elements and cereals, certain values exceeded the Rfd. For instance yellow corn from the site revealed As EDI above the RfD, and yellow corn from Sighnoguin revealed Cd EDI above RfD. White sorghum from Songo consumption lead to EDI above the RfD. All cereals from all the location except for white sorghum from Youga and yellow corn from both Site and Youga, exhibited EDI of Pb above the Referential dose and consequently a THQ above 1. Average EDI of elements followed this pattern Mine > Songo > Youga > Sighnoguin. Combined toxicity expressed by HI were all above 1 highlighting a potential health concern.

This situation means that actors should address the need for contaminant removal before agricultural practices can be allowed in mining sites and impacted zones. Decontamination of sites and selection of cereals with a low uptake capability and specific agricultural practices could reduce these hazards. Proper management of lead-batteries in rural areas and leaded gasoline should be implemented. Health centers in the vicinity of the mining perimeter should consider these hazards during diagnosis of diseases. This study focused on cereals, but full dietary intake (cereals, meat, fruits, water) survey should be carried out to estimate potentially toxic elements intake and to take actions accordingly.

Chapter 5: Potentially Toxic Elements in Pit Lakes and Fishes and Fish Intake Exposure Assessment

This chapter is based on the publication:
Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (2019c). Metals and metalloid in gold mine pit lakes and fish intake risk assessment, Burkina Faso. Environmental Geochemistry and Health, pp 1-15, http://dx.doi.org/10.1007/s10653-019-00390-8.

ABSTRACT: This study aimed to determine the levels of potentially toxic elements (As, Cd, Co, Cu, Cr, Mn, Ni, Pb and Zn) in pit lakes from a gold mining site and in their fishes and assess their potential health effect on the local human population, in order to evaluate whether pit lakes can be safely used for aquaculture. Water quality data were collected from two pit lakes namely West pit 1 (WP1) and West pit 2 (WP2), and the Nakambé river (NR) in Burkina Faso. Fish consumption rates in different villages were assessed through a survey. Commonly available fish were sampled from the pit lakes and the NR. Fish from the pit lakes contained higher amounts of potentially toxic elements than fish from the river (WP1 ~ WP2 > NR). Of the four species of fish considered, *Oreochromis niloticus* and *Hydrocynus forskahlii* had the highest potentially toxic elements content and *Bagrus bajad* and *Clarias anguillaris* had the lowest. The results indicated that the consumption of the whole fish results in higher potentially toxic elements intake than consumption of the fleshy part only. Due to the low fish intake of 5.34±2.60 g/day/adult deduced from the nutritional survey, exposure to potentially toxic elements was below referential doses. The highest arsenic intake comes from eating entire *Oreochromis niloticus* (0.058 mg/day/adult) from WP1. Eating *Oreochromis niloticus* and *Clarias anguillaris* exposes people to an arsenic intake of 0.01 mg/day/adult. The arsenic contents of *Hydrocynus forskahlii* and *Bagrus bajad* were below the method detection limit.

5.1. Introduction

It is very common for mining to have an impact on soil and water bodies, with mining sites behaving as hotspots for spreading contaminants through leaching, wind, and water runoff (Camizuli et al., 2014). Potentially toxic elements levels change due to gold processing and the presence of tailing storage facilities, waste dumps, fuel storage and excavated contaminant-bearing rocks. These potentially toxic elements could threaten the ecosystem and negatively affect human health through the intake of contaminated food from these areas (Bortey-Sam et al., 2015; Squadrone, 2016).

When open pit mining ends, pit lakes could potentially be used for irrigation, fish farming and providing water for the rearing of livestock (Fowe et al., 2015; De Lange et al., 2018). Fish farming in pit lakes could be an opportunity to improve local community food resilience, improve diet quality and provide nutrients to meet daily requirements, as well as acting as a source of income for the local population. However, those options are only viable if the quality of pit lake water is suitable for such a use and fish from the pit lakes are safe and not a danger to human health. The quality of water in pit lakes is influenced by geochemical characteristics and proximity to waste dumps and tailing storage facilities, and the change in water quality could affect the life of aquatic organisms. Fish have the ability to accumulate contaminants from their environment (Talab et al., 2016) and are used as bioindicators of contaminants in water bodies (Birungi et al., 2007; Linde-Arias et al., 2008). Essentially, fish assimilate essential (cobalt, chromium, copper, manganese, nickel and zinc) and non-essential (Arsenic, cadmium and lead) potentially toxic elements, major elements (magnesium) by ingestion of particulate material suspended in water, by ingestion of food, by ion-exchange of dissolved potentially toxic elements across lipophilic membranes, and by adsorption through tissue and membrane surfaces (Shah et al., 2009). Contaminants in fish originate predominantly from their diet, and levels of bioaccumulative contaminants are greater in fish that are higher in the food chain (EFSA, 2005). To avoid an adverse effect on health from consuming contaminants, international organizations set acceptable daily intake (ADI) or provisional weekly tolerable intake (PWTI) (WHO, 1996; USEPA, 2011; EFSA, 2009; EFSA, 2010; EFSA, 2015).

To be sure that pit lakes can be used in the stage after mine closure for fish breeding, experiments were carried out in pit lakes that had been created by diverting a nearby small river. Levels of potentially toxic elements from pit lakes were found to be above the standards for drinking water (WHO, 2011), and this could affect the health of the local population if used as a source of drinking water. Against South African and Canadian water quality guidelines, water from the pit lakes was found to be suitable for aquaculture. However, species specificity should be considered as the ability to take up potentially toxic elements differs (Birungi et al., 2007) and, therefore, so does the contribution of different species to contaminant uptake by the local population (Bortey-Sam et al., 2015).

The study objectives were to: (i) assess the water quality of two pit lakes and benchmark with a nearby river, (ii) assess the quality of the fish from the pit lakes and benchmark against fish from the nearest local river, (iii) evaluate the local common fish bioaccumulation capability and the transfer factor, and (iv) estimate hazards from intake of fish by the local community to decide on potential use of the pit lakes. The fish consumption rate was estimated through a survey submitted to the six villages surrounding the mining site.

5.2. Materials and Methods

5.2.1. Study Area

The mining site under investigation is described in 2.6. Two half-a-million cubic meter pits were used. West Pit 1 (WP1) was mined from October 2009 for six months and then West Pit 2 (WP2) was mined for the same period. The approximate surface area is 0.02 km^2 for each pit with a depth of 40 meters. WP1 was filled in 2011 by diverting the Zéra River tributary passing close to the pit. WP2 was filled during the rainy season (May to October) in 2016 through a channel dug to divert the Zéra River and passing between WP2 and West Pit 3 waste dumps.

5.2.2. Water Sampling

WP1, WP2 and NR were sampled 14 times each from January 2015 to April 2017 on a bimonthly basis for potentially toxic element analysis. NR located 11 km away from the site was considered as reference. Water samples were taken from close to the banks, between 5 and 10 cm depth to avoid floating litter. For practical reasons, stratifications and thus a potential heterogeneity of water quality over depth, has been ignored. Glass bottles of half a liter capacity were used to sample and stored in an ice container. The containers were then sent to the laboratory for analysis in the same day. During water sampling, in-field parameters were measured. A handheld multiparameter probe HI9829 (Hanna Instruments Inc, USA) equipped with a pH/ORP sensor (HI 7609829-1) which is a combined pH/ORP sensor features a glass sensitive bulb for pH readings, a platinum sensor for redox measurements and a silver/silver chloride double junction reference with gelled electrolyte, dissolved oxygen sensor (HI 7609829-2) and EC sencor (HI 7609829-3), was used for the measurement of pH, dissolved oxygen (DO, mg/L), oxidation-reduction potential (ORP, mV), total dissolved solids (TDS, mg/L) and conductivity (µS/cm). ORP data were corrected to standard hydrogen electrode (SHE) readings by adding 205 mV to the readings, according to the manufacturer handbook.

5.2.3. Fish Sampling

Fish were sampled from WP1 and WP2 and from the NR 11 km from the mining site. Fish from WP1 come from the diverted small river, supplemented by fish bought from the Bagré dam area for the experiment. Fish from WP2 come from the diverted small river. Local fishermen were instructed to catch the fish at the two different pit lakes using both active and passive fishing systems; fish nets and trammel nets were used. After the fishing process, the length of fish was measured and fresh weight was also recorded. Each fish was put in a labeled clear plastic bag, temporarily stored in an ice cooler, brought to the site, thoroughly cleaned with distilled water and stored in a deep freezer prior to further processing. Fish from the NR were purchased from local fisherman and underwent the same treatment as previously described. Further processing consisted of drying at 60 °C until a consistent dry weight was reached. After the drying process, the fish samples were labelled and stored in transparent plastic bags at ambient conditions and sent to Ghent University Campus Kortrijk for analysis.

Common local fish were included in this study. The selection was based on the fish available from the NR and frequently consumed by locals, as described by Ouédraogo & Amyot (2013). The selected fish families and species (Froese & Pauly, 2017) are shown in Table 5-1. *Bagrus Bajad* (Forsskäll 1775) (BB) lives and feeds on or near the bottom. The adults are piscivorous. *Clarias anguillaris* (Linné 1758) (CA) is an omnivorous fish commonly found in inundated areas (Lévêque et al., 1992). *Hydrocynus forskahlii* (HF) (Cuvier 1819) is an open-water predator often found near the water surface feeding on fish. *Oreochromis niloticus* (ON) (Linnaeus 1758) is mainly diurnal and feeds on phytoplankton or benthic algae. They are oviparous and omnivorous.

Sampling was carried out in two rounds. 30 fish samples were sampled during the first round on November 2016 and entire fishes were analyzed. 39 fish samples were sampled during the second round in March 2017 and entire fish as well as the fleshy part was also analyzed. NR fish samples were used for benchmarking.

Table 5-1 Selected fish samples and descriptions

Description			Fish sampled						
Classification			First round			Second round			Total
Order	Family	Species	WP1	WP2	NR	WP1	WP2	NR	
Characiformes	Alestidae	*Hydrocynus forskalii*	4	1	0	0	7	9	21
Perciformes	Cichlidae	*Oreochromis niloticus*	10	3	5	11	2	6	37
Siluriformes	Bagridae	*Bagrus bajad*	0	4	0	0	0	2	6
Siluriformes	Clariidae	*Clarias anguillaris*	1	0	2	0	0	2	5

5.2.4. Local Community Food Intake Survey

A survey was carried out to estimate the fish consumption rate as the method described in 4.2.3. Fishes were typically consumed dried and grinded and used in sauce. In this condition, entire fishes were consumed instead of the fleshy part if the fishes are small.

5.2.5. Major Water Quality Variables Analysis

Major anions (F^-, Cl^-, NO_2^-, NO_3^-, SO_4^{2-}) and Na, K, Ca, Mg, Fe, As, Zn, Mn were analyzed in a commercial laboratory in Ouagadougou in Burkina Faso. The methods used for the analysis of the major water variables were NF ISO 9297: 2000 (chloride), NF EN ISO 7980 (Ca and Mg), Hach method 8051 (sulfates), Hach method 8507 (nitrites), Hach method 8171 (nitrates), Hach method 8029 (fluorides), and NF T 90-019 (Na and K). In addition, atomic absorption spectrometry was used for analysis of iron (Fe), As, Zn and Mn. Detection limit were As (0.5 µg/L), Fe (0.005 mg/L), Mn (0.002 mg/L), Zn (0.001 mg/L), Chloride (0.5 mg/L), sulfate (2.5 mg/L), fluoride (0.005 mg/L).

Water quality was benchmarked against international standards as no specific standards exist in Burkina Faso; Canadian water quality guidelines (CSQG) (CCME, 2008) and South African water quality guidelines (SAWQG) (Department of Water Affairs and Forestry, 1996).

5.2.6. Potentially Toxic Element Analysis of Fish

The method used for the analysis of total potentially toxic elements is described in USEPA 3052 and slightly modified: microwave-assisted acid digestion of siliceous and organically based matrices (USEPA, 1996). Briefly, for each fish sample, 0.5 g of the fleshy part was taken from the whole dried fish. The remainder of the whole fish was ground using a mortar and mixed well; 0.5 g was taken from the entire fish sample for analysis. 0.5 g was weighted into the Teflon vessel for the microwave digestion process (Milestone Microwave Laboratory System, Milestones Inc, Monroe, CT, USA); 8 mL of concentrated nitric acid (65%) was added and placed in the microwave at 330 Watts for 6 minutes. After a cooling down stage, 2 mL of pure hydrogen peroxide was added, and the mixture heated for 4 minutes at 450 Watts. After another cooling stage, a second 2 mL of hydrogen peroxide was added and heated for 4 minutes at 600 Watts. The vessels were cooled down and then filtered with a VWR filter N° 413 and diluted to 25 mL with bi-distilled water. The samples were analyzed immediately or stored in a refrigerator at 4°C until analysis. The measurements were made using an ICP-OES equipped with Qtegra Software (Thermo Scientific™ iCAP™ 7000 Plus Series ICP-OES, Thermo Fisher Scientific Brand, USA). Method detection limits (MDL) of 1.78, 0.04, 0.44, 0.37, 0.35, 1.63, 0.40, 0.45, 0.41 and 0.23 mg/kg for As, Cd, Co, Cr, Cu, Mg, Mn, Ni, Pb and Zn respectively were measured. Calibration solutions were prepared as indicated in section 3.2.2.2. The water used was bi-distilled water and the chemicals were all analytical grades.

The water-fish transfer factor (TF) was assessed. The TF is the ratio between the contaminant concentration in the pit lakes or the NR water in mg/L (C_{Water}) and the concentration of the respective contaminant in the fish in mg/kg dry weight (C_{Fish}).

$T_F = {}^{C_{Fish}}/_{C_{Water}}$ (Equation 6)

5.2.7. Health Risk Evaluation

Estimated daily intake (EDI) and target hazard quotient (THQ) were used to estimate the intake risks related to consumption of the fish (Birungi et al., 2007; Oyibo et al., 2017). Allowable daily consumption (ADC) and the allowable daily number of fish (ADN) which could be safely eaten were calculated (Ouédraogo & Amyot, 2013; Taweel et al., 2013).

The EDI (mg/adult/day) was computed based on the concentration of potentially toxic elements in fish, the daily fish consumption rate and the average body weight of an individual consumer, which was considered equal to 70 kg (Equation 4).

The THQ was estimated based on the formula expressed in Equation 5 which considers the fish intake rate by the population, the average body weight W (70 kg) and the referential doses of respective potentially toxic elements (Table 4-1).

The ADC was computed for each fish species which could be eaten while remaining in the safe range, based on the daily food intake of the local population and the contaminant level in the fish. The safe amount of fish and the number of fish per species which could be eaten by the local community were calculated using the formula of Ouédraogo & Amyot (2013).

$ADC = W \times (TDI/C_{Fish})$ (Equation 7),

Where ADC = Allowable daily consumption, amount of fish which could be consumed with respect to reference values (grams),

W = average body weight (70 kg for an adult man),

TDI= Tolerable daily intake value for each element (mg/kg body weight/day).

C_{Fish} = Concentration of chemical contaminant in the fish (mg/kg),

5.2.8. Quality Control and Statistical Analysis

The method used for the analysis of the fish samples was validated through a set of parameters like the recovery and analysis of certified reference material. Quality control of the analysis was carried out using reference material ERM-BB422, fish muscle (potentially toxic elements) N° 354 from the Institute of Reference Materials and Measurements of the Joint Research Centre of the European Commission and the results are found to be within the range of 85 to 115%. The MDL was determined as described in 3.2.3 and statistical analysis as per the description in 3.2.5. Values were expressed as the means ± standard deviation.

5.3. Results and Discussion

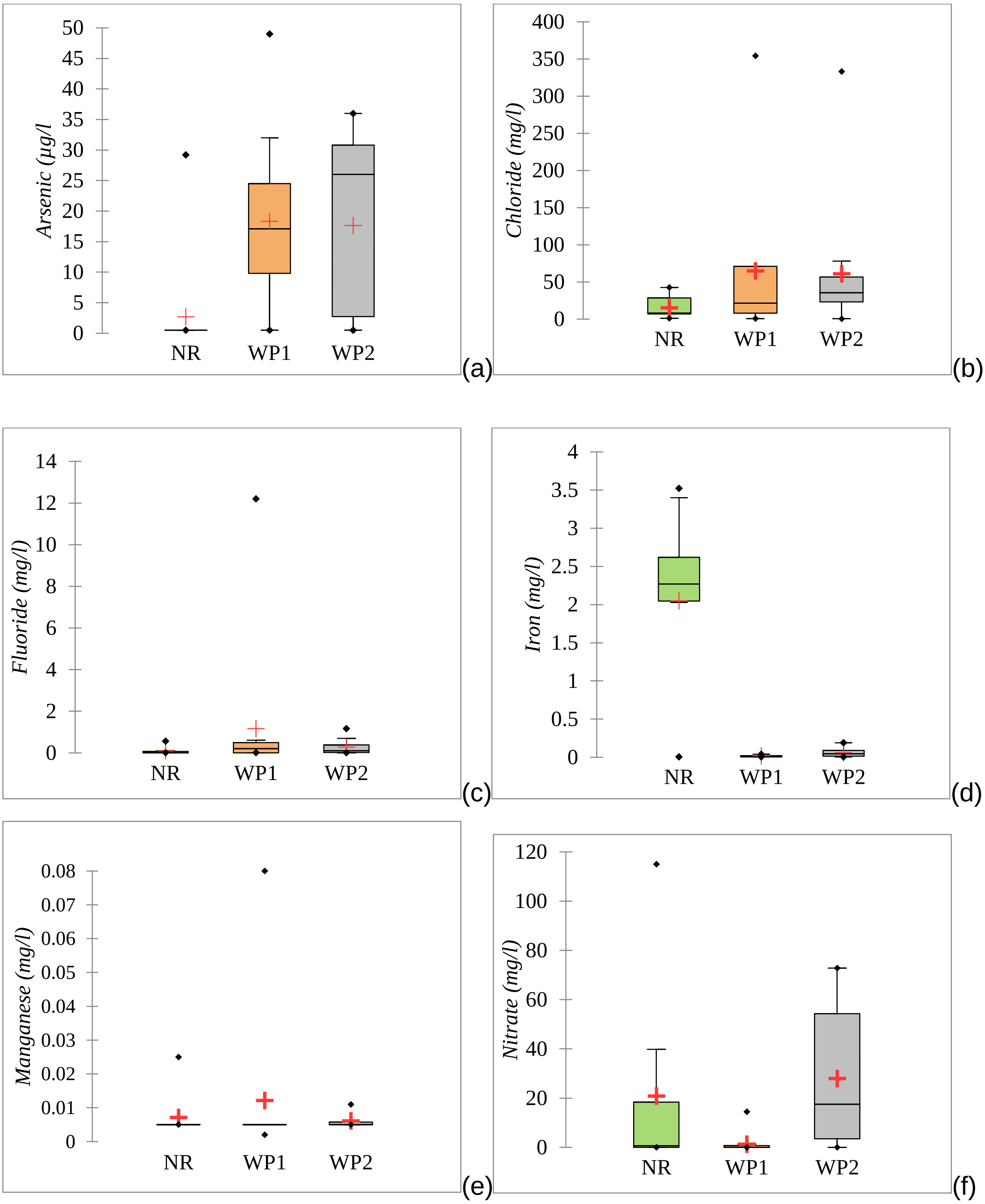
5.3.1. Water Quality

5.3.1.1. In-field Parameters

Measurements were carried out from January 2016 to July 2017, except for DO and ORP which were measured only in 2017. The temperature annual averages were 28.1±2.5°C, 25.7±5.6, and 27.5±2.5°C for WP1, WP2 and the NR respectively. The values for pH, ORP, TDS and conductivity were higher in the pit lakes compared to the NR, whilst DO was lower. High conductivity reveals the presence of salt in the water (Sako et al., 2018), and mineralization was higher in the pit lakes than in NR. The pH of the pit lakes, as well as NR, was alkaline, with annual averages of 8.33±0.48, 8.32±0.35 and 7.95±0.24 for WP1, WP2 and NR respectively. ORP in pit lakes were 269.53±38.93 mV for WP1, 212.27±7.35 mV for WP2 and 246.83±22.06 mV for the NR. The redox potential value gives a general description of many redox reactions and is viewed as a measure of a system's overall reducing or oxidizing capacity (Søndergaard, 2009). Redox potentials of less than -100 mV indicate anaerobic environments, while values greater than 100 mV indicate aerobic environments (Delaune & Reddy, 2005; Scholz, 2019). When the ORP value is high, there is lots of oxygen present in the water. DO of water bodies was consistently above 80% saturation. The 2017 annual averages were 88.23±14.93 % and 87.77±16.16 % for WP1 and WP2 and 89.90 ± 16.99 % for the NR. In-field parameters are dictating physicochemical interactions, potentially toxic elements availability, speciation and uptake by fishes. DO characterizes the water reservoir's capability to contain living animals. ORP is important in terms of element speciation. The effect of oxidation-reduction potential on potentially toxic element mobility revealed the transportability of chromium with increasing redox potential while arsenic was barely mobile with increasing oxidation level (Kartal, 2003). Also, zinc mobility diminished strongly with increasing oxidation-reduction potential.

5.3.1.2. Other Water Quality Parameters

Grab samples were collected from the pit lakes and the NR during 2016 and 2017. Results are represented in (Figure 5-1). Pb, Hg, free Cyanide, Ni, Cu, Co, Cr, WAD Cyanide, Cd, Se, were analyzed and found to be below method detection limit (0.005 mg/L).



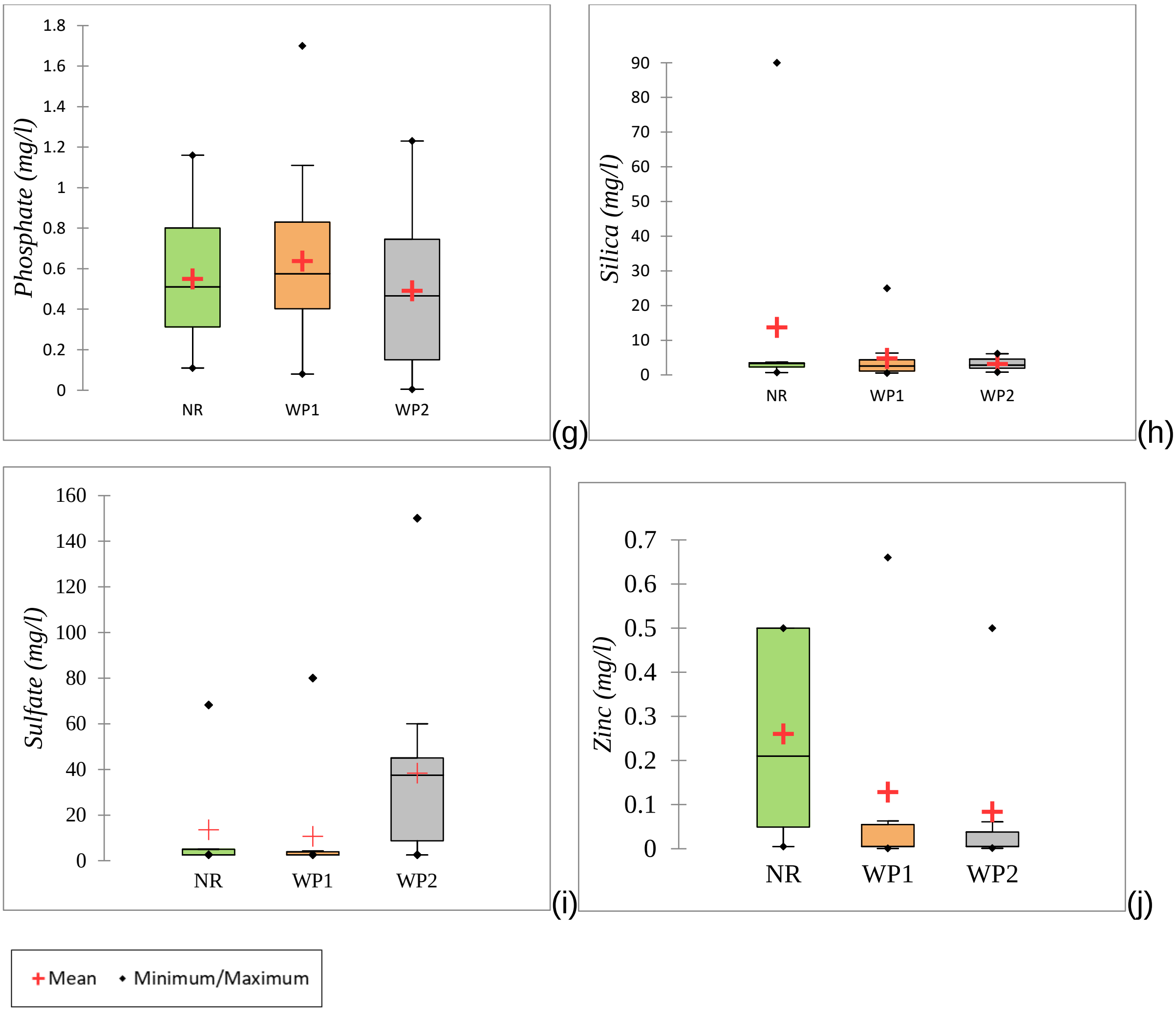


Figure 5-1 Boxplot of selected water parameters from the three locations,

NO_2^-, NO_3^-, Cl^-, F^- and Zn did not show a significant difference between the three-year means of the three water bodies involved in this study according to the ANOVA analysis. Similar content of those elements in the three water bodies reflected the same sources and did not demonstrate a mining contribution to their content in West pit water. The source of nitrate could be fertilizers contained in water runoff from nearby farms to the site and the NR. On the other hand, ANOVA revealed significant difference between the three-year averages of the three water bodies for Na, K, Ca, Mg, Fe and As. The pits' As content averages were not statistically different from each other. Hierarchical cluster analysis confirmed three distinct water bodies with a greater similarity between WP1 and WP2 than between WP1, WP2 and NR. Selected major anions and cations analyzed did not allow ion balance computation.

5.3.1.3. *Benchmarking and potential use of pit lakes*

The local community can safely use pit lake water if water quality parameters meet standard requirements. Alongside the benchmarking of pit lake water chemistry with the nearest river, water quality was benchmarked against international standards and selected national regulations as per Table 5-2 for potentially toxic elements. Two national regulations have been chosen for the purpose: SAWQG and CWQG. In-field parameters of all water bodies comply to South Africa Water Quality Guidelines for agricultural use.

Table 5-2 Water quality parameters compared to Canada and South Africa water quality guidelines, In mg/L except for arsenic in µg/L, F^- for fluoride, Cl^- for chloride. Bold data express values exceeding a limit which could be for one standard or for both Canadian and South African Water Quality Guidelines. Empty cells indicate that no standards are included in those guidelines.

			As	Ca	Fe	Mg	Na	K	Zn	NO_3^-	NO_2^-	SO_4^{2-}	Cl^-	F^-
Canada Water Quality Guidelines	Aquatic Life	Freshwater	5		0.3				0.03	13	0.06			
	Protection of Agriculture	Irrigation	100		5									
		Livestock	25	1000					50		10	1000		
South Africa Water Quality Guidelines	Aquatic Life	Freshwater	10						0.002					0.75
	Domestic	Use	10	32	0.1	30	100	50	3			200	100	1.0
	Agricultural Use	Livestock	1000	1000	10	500	2000		20	100	10	1000	3000	6
		Irrigation	100		5		70		1				1.00	2
		Aquaculture	50		0.01				0.03	0.05			600	
Site water bodies and NR parameters	WP1	Mean	**18.32**	18.05	**0.02**	7.14	9.48	1.78	**0.13**	**1.28**	**0.03**	10.68	64.83	1.60
		SD	12.89	9.38	0.01	5.02	11.65	0.66	0.23	3.83	0.03	21.30	101.2	3.33
		Min	0.5	0.46		0.48	0.075	0.106	0.001	0.005	0.005	2.5	0.7	0.005
		Max	49	36.8	0.04	19.76	47.1	2.8	0.66	14.5	0.10	80	354.5	12.2
	WP2	Mean	**17.65**	**36.93**	**0.06**	11.64	18.69	2.71	**0.08**	**27.98**	**0.12**	38.29	60.97	0.27
		SD	15.00	14.09	0.06	10.08	10.42	1.14	0.17	29.02	0.12	40.11	89.22	0.35
		Min	0.5	16	0.005	0.48	5.6	0.6	0.001	0.005	0.005	2.5	0.5	0.005
		Max	36	67.33	0.19	36.78	37.9	3.8	0.5	72.80	0.33	150	333.23	1.16
	NR	Mean	2.69	9.50	**2.03**	4.97	2.25	2.75	**0.26**	**20.81**	**1.44**	13.46	15.17	0.10
		SD	7.64	6.38	1.15	3.47	1.40	1.34	0.23	38.95	5.06	24.50	14.29	0.19
		Min	0.5	0.2	0.005	0.48	0.3	0.22	0.005	0.005	0.005	2.5	1	0.005
		Max	29.2	23.2	3.52	12.1	5.95	4.0	0.5	115	19	68.2	42.54	0.56

The pit lakes' arsenic content exceeds both SAWQG and CWQG for aquatic life but falls within the requirements of both regulations for agricultural use. Consequently, water from the pit lakes can be deemed fit for agricultural use. In the case of Fe, pit lake water shows levels below the requirement for the protection of aquatic life of 0.3 mg/kg set by the CWQG and SAWQG but is above the requirement of 0-0.1 mg/L for aquaculture.

The arsenic content in WP1 and WP2 was above the limit of 10 µg/L set by the World Health Organization guideline (WHO, 2011) for drinking water but beneath the 50 µg/L guideline for SAWQG for aquaculture. Seasonal water runoff to the pit lakes will continue to influence pit lake water chemistry depending on the water runoff source, the runoff bed soil type and sediment quality. Long-term stabilization could be predicted (Kalin et al., 2001) as wall rock leachate will diminish and show an overall improvement of water quality as found in the Getchell (Getchell Gold Corporation, Nevada) pit lake as reported by Shevenell et al. (1999).

5.3.2. Local Community Fish Consumption Rate

The main consumption mode in the study region is dry entire fish which is ground to a powder and mixed in sauce for taste. The fish consumption survey showed that consumption rates of dry fish varied between 4.77±2.01 g per day per person in the village of Songo to 6.93±3.94 g per day per person in the village of Youga (Table 5-3). An ANOVA test revealed that these fish consumption rates were not significantly different between the six villages. The overall average consumption rate of fish was 5.34±2.6 g/day/person. These fish consumption rates are in line with national data which reported a fish consumption rate of 3.29 g/day/person in 2001 and 4.93 g/day/person in 2010. Again, Bernhard & Hoffmann (1999) found similar fish consumption patterns for Burkina Faso, with average fish consumption rates of 2.57 kg/year/person equivalent to 7.04 g/day/person in rural areas, and 4.37 kg/year/person (11.97 g/day/person) for urban populations. A negative significant correlation found between the number of persons in a household and the fish consumption rate ($r = -0.443$; $p < 0.001$) expressed that larger households have a lower fish consumption rate.

Table 5-3 Fish consumption rate (dry weight(g)/person/day), obtained through the survey as described in 4.2.3.

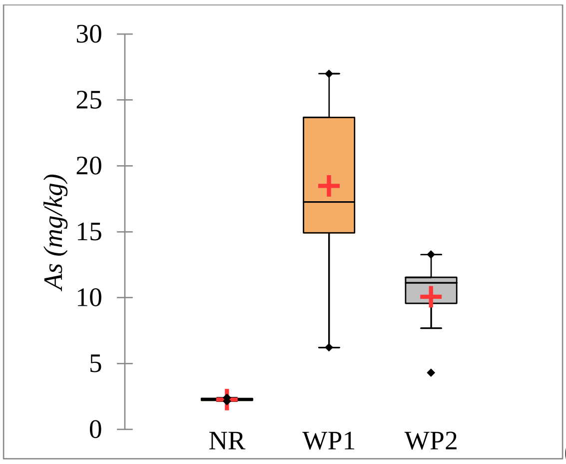
	Gonsé	Youga	Wilgo	Sighnoguin	Youguego	Songo	**Total**
Mean	4.94±3.03	6.93±3.95	5.34±2.16	4.82±1.96	5.29±1.99	4.77±2.01	**5.34±2.60**
Range	1.74-12.00	1.00-15.00	0.56-7.50	2.22-7.50	2.00-8.57	2.50-10.00	**0.56-15.00**

5.3.3. Sampled Fish Characteristics

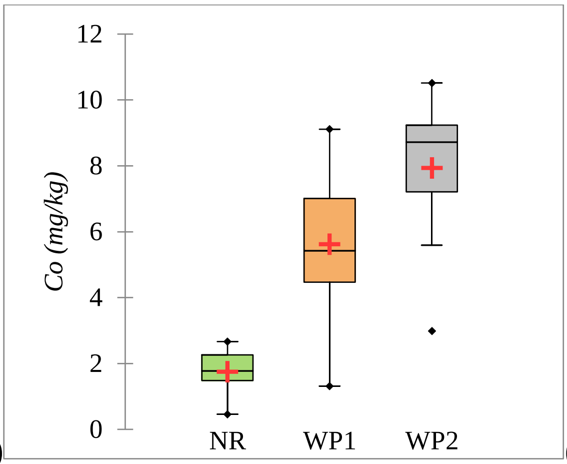
The fish length, fresh weight and dry weight were measured. The average fresh weight of the different species was in this sequence from heaviest to lightest: *C. anguillaris* > *H. forskahlii* > *O. niloticus* > *B. bajad* with their corresponding fresh weights being 145.8±81.46 g, 82.64±24.20 g, 28.21±5.97 g and 27.9±5.37 g, respectively. Average dry matter contents were in the range of 23-25%, comparable to the study of Talab et al. (2016) on *O. niloticus*. The average length of *C. anguillaris* was 27.5±6.36 cm, *H. forskahlii* 20.55±2.21 cm, *B. bajad* 15.55±0.78 cm, and *O. niloticus* 11.81±0.98 cm. No significant differences were found between fish species according to the locations.

5.3.4. Potentially Toxic Element Concentrations in Fish Samples

Entire fish and flesh part of fish were analyzed to assess the potentially toxic elements and intake risk related to fish consumption. In general, potentially toxic elements content per location were as follows: WP2 > WP1 > NR for Co, Cr, Ni and Pb, NR ~ WP1 < WP2 for Mg, WP1 ~ WP2 > NR for Cu, WP1 > WP2 > NR for Mn and As and WP2 ~ NR < WP1 for Zn (Figure 5-2). Fleshy parts of fish from Pit lakes analysis results were displayed in Figure 5-3. Comparison between entire fish analysis and fleshy part of fish analysis was represented in Figure 5-4.



(a)



(b)

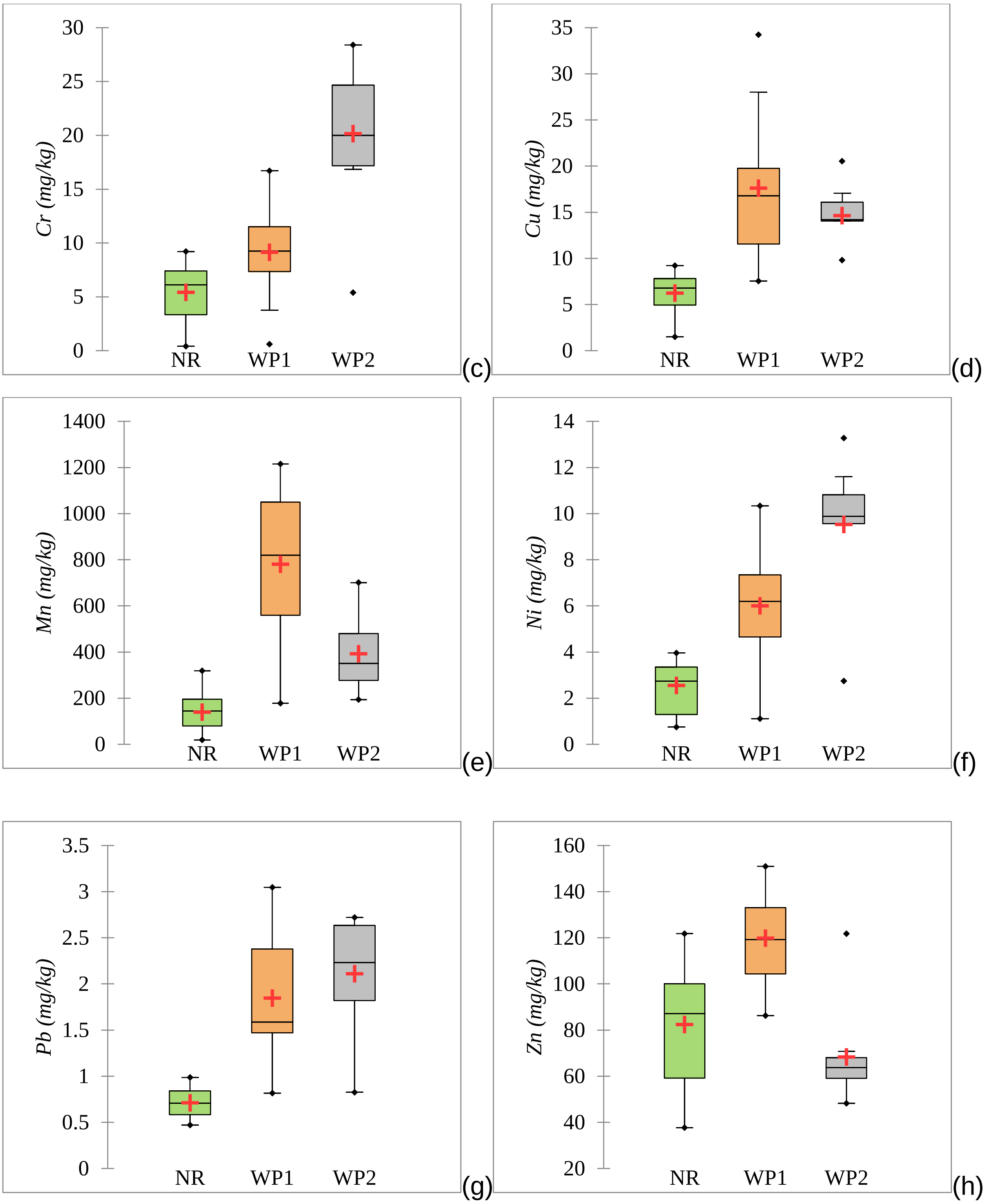
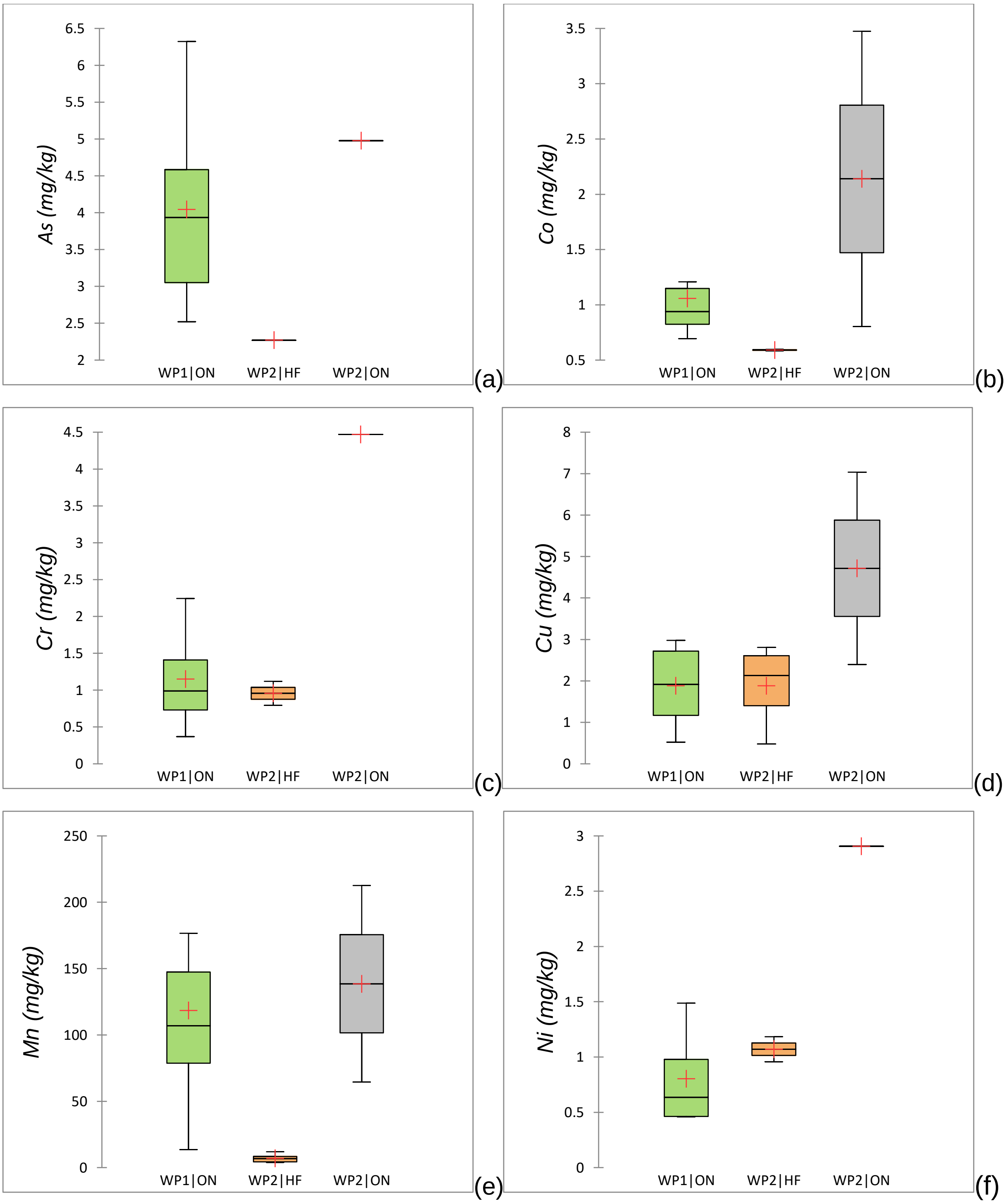


Figure 5-2 Potentially toxic elements distribution in entire fish according to locations, As (a), Co (b), Cr (c), Cu (d), Mn (e), Ni (f), Pb (g) and Zn (h). NR for Nakambé river, WP1 for West Pit 1 and WP2 for West Pit 2.


As (mg/kg)
6.5
6
5.5
5
4.5
4
3.5
3
2.5
2
WP1|ON
WP2|HF
WP2|ON
(a)
Co (mg/kg)
3.5
3
2.5
2
1.5
1
0.5
WP1|ON
WP2|HF
WP2|ON
(b)
Cr (mg/kg)
4.5
4
3.5
3
2.5
2
1.5
1
0.5
0
WP1|ON
WP2|HF
WP2|ON
(c)
Cu (mg/kg)
8
7
6
5
4
3
2
1
0
WP1|ON
WP2|HF
WP2|ON
(d)
Mn (mg/kg)
250
200
150
100
50
0
WP1|ON
WP2|HF
WP2|ON
(e)
Ni (mg/kg)
3
2.5
2
1.5
1
0.5
0
WP1|ON
WP2|HF
WP2|ON
(f)

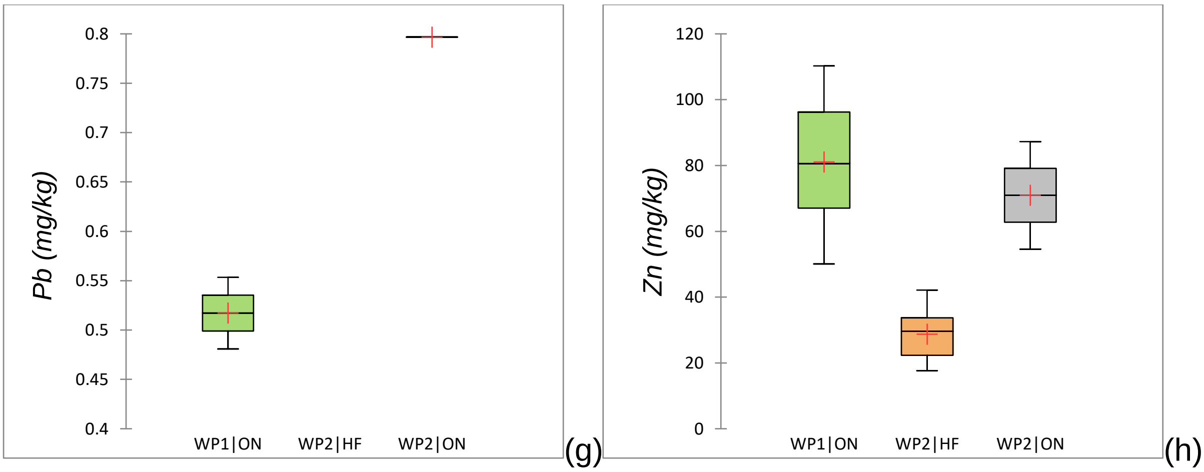


Figure 5-3 Potentially toxic element distribution in fleshy fish from west pit lakes and fish species, As (a), Co (b), Cr (c), Cu (d), Mn (e), Ni (f), Pb (g) and Zn (h). WP1 for West Pit 1 and WP2 for West Pit 2,

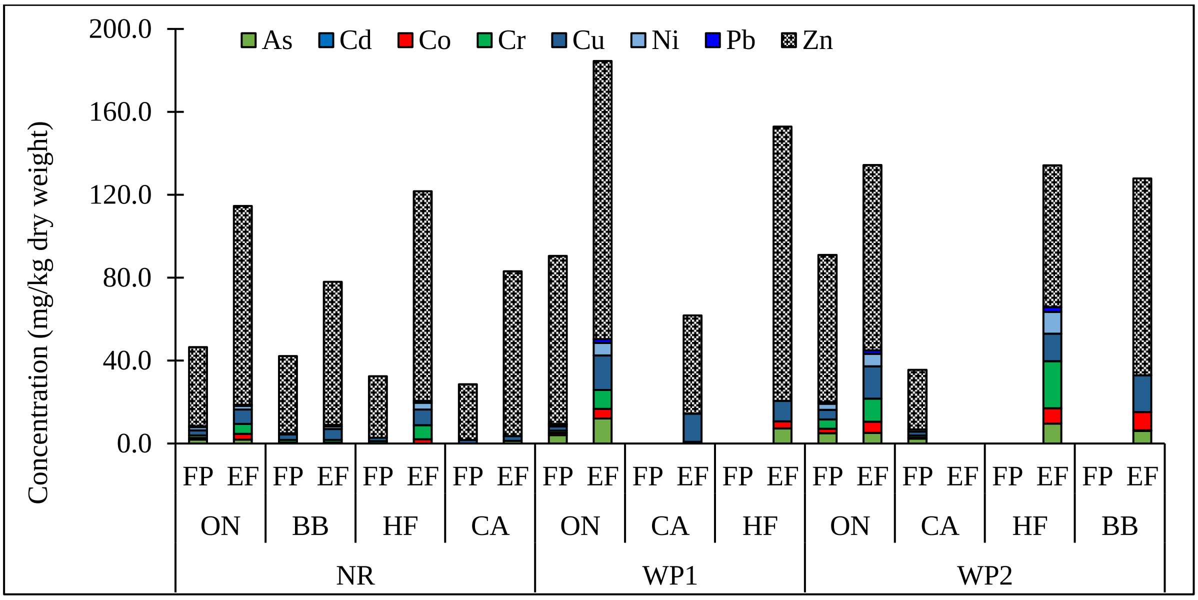


Figure 5-4 Comparison of PTE distribution in fish according to species, locations and parts analyzed, FP for flesh part and EF for entire fish analysis results,

Agglomerative hierarchical clustering (AHC) with similarity based on Pearson correlation highlighted three distinct classes, which from the percentage of dominance, could be attributed to each of the three water bodies (Figure 5-5). Further, the dendrogram representation showed the split of WP1 samples into the two of the three major classes inferring similarity of WP1 fish samples to NR and WP2. WP1 *O. niloticus* intrusions in the first class dominated by NR samples was observed and interpreted as similarity between *O. niloticus* from those two locations. Three locations with different water qualities results in different potentially toxic element contents in fish.

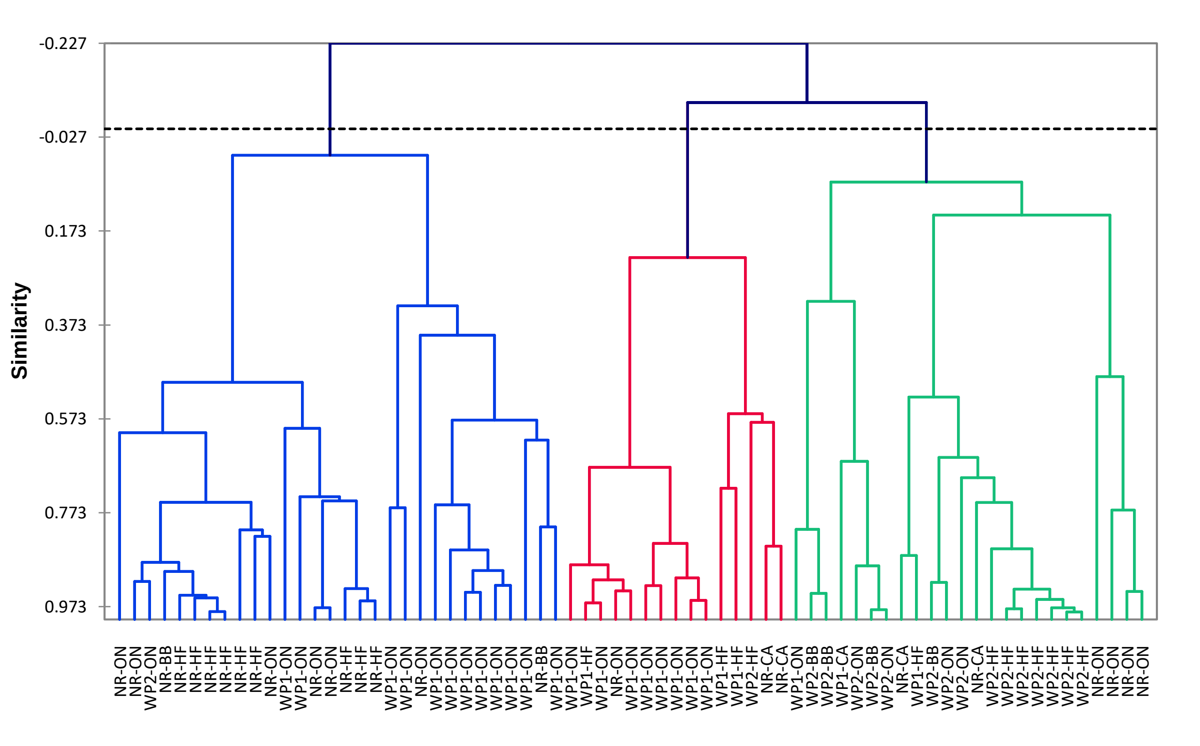


Figure 5-5 Agglomerative hierarchical clustering with similarity based on Pearson correlation.

The arsenic contents of *O. niloticus* from NR and WP1 were significantly different. A significant difference was also found in the As content of *O. niloticus* between WP1 and WP2, while no significant difference was found between WP2 and NR. Other fish species' As content was below MDL.

The cobalt contents of *B. bajab* from NR and WP2 were significantly different, with WP2 samples presenting the highest levels. *H. forskahlii* Co contents were significantly different between locations.

The copper contents of *O. niloticus* from NR and WP1 were significantly different. No significant differences were found in O. *niloticus* between WP1 and WP2 or between WP2 and NR or in *H. forskahlii* between WP2 and NR. All fish Cu content were below limit set by FAO/WHO, 1989 which was 30 mg/kg wet weight.

The Cr content in *H. forskahlii* from WP2 was significantly different from the Cr content from NR and also significantly different between WP1 and WP2.

C. anguillaris samples from NR and WP1 show significant differences in Mg content. *H. forskahlii* Mg content in fish was significantly different according to location. A significant difference was found in *O. nolitic*us Mg contents (WP1 ~ NR < WP2).

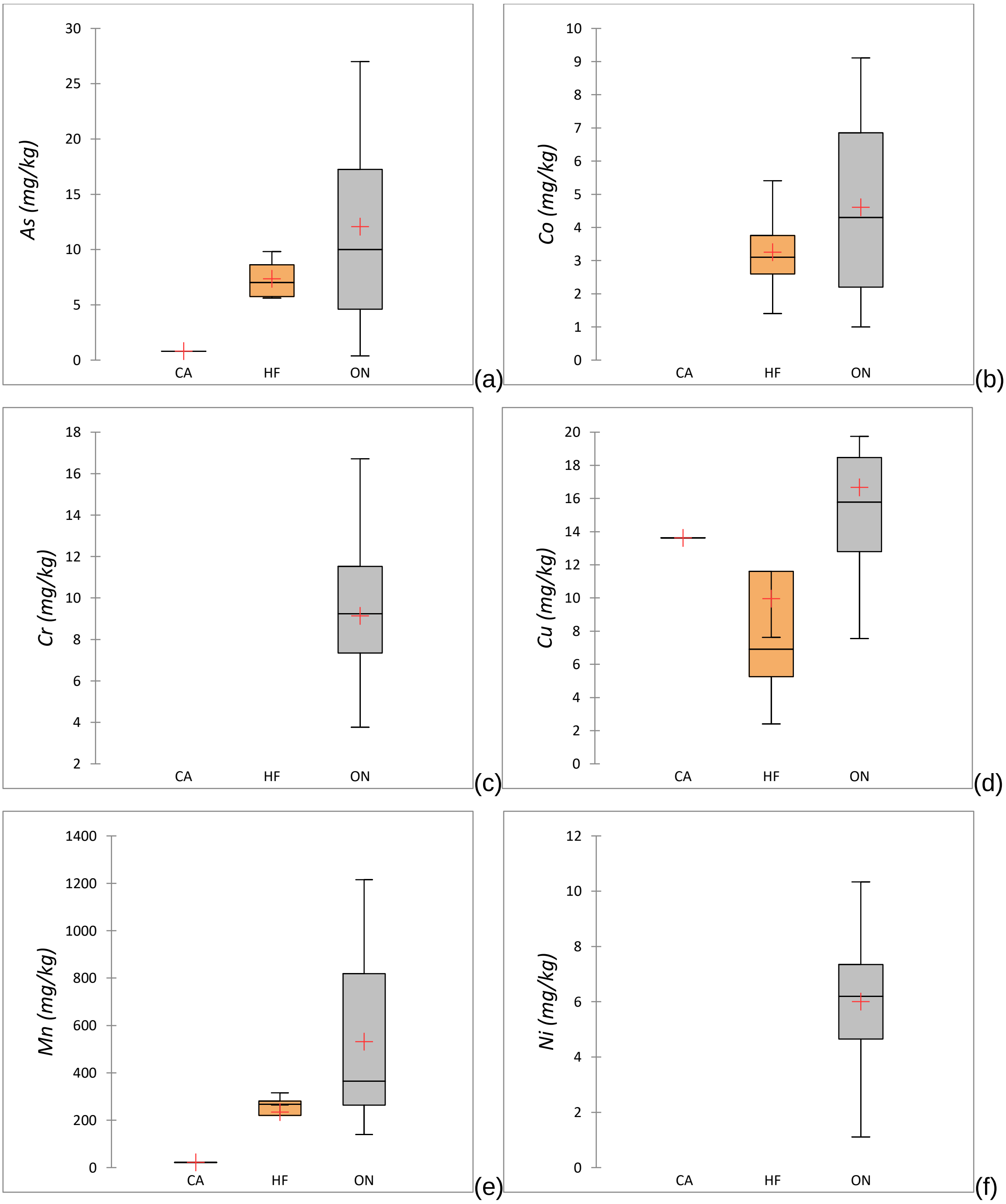
The nickel contents of *H. forskahlii* from WP2 and NR were significantly different. Significant differences were also found in the Mn content in *O. niloticus* between WP1 and NR.

Significant differences were found in *H. forskahlii* Pb content between WP2 and NR and also in *O. niloticus* between WP1 and NR. No significant differences were found in *O. niloticus* Pb content between WP1 and WP2, or between WP2 and NR. Except for WP2 fishes, NR and WP1 fishes exhibited Pb content below FAO/WHO limit of 0.5 mg/kg wet weight.

The zinc contents of *O. niloticus* from WP1 and WP2 were significantly different from Zn contained in the same species from NR. No significant difference was found between WP1 and WP2 for *O. niloticus*. The Zn contents of *H. forskahlii* in NR and WP2 were significantly different. All fishes' samples exhibited Zn content below limit set by FAO/WHO, 1989 which was 40 mg/kg wet weight. Differences in entire fish analysis results could be explained by potentially toxic elements content in the water body and the fish feeding style. The higher Zn content in *O. niloticus* could be related to its feeding on benthic worms and crustaceans (Baharom and Ishak, 2015).

A comparison was made between the fleshy part results and the entire fish results of the second round of sampling. This comparison revealed lower levels of potentially toxic elements in flesh than in the entire fish. Higher potentially toxic element contents in entire fish compared with the fleshy part could be attributed to the presence of gills, which are known to accumulate high amounts of potentially toxic elements (Birungi et al., 2007); the intestine, liver, kidneys also accumulate potentially toxic elements (Atobatele and Olutona, 2015; Pouil et al., 2017), and to low transfer of dietary elements like Cd into fish muscle (2 – 6 %) (EFSA, 2005). Consuming entire fish will expose consumers to higher potentially toxic elements intake rate than would eating the fleshy part.

When comparing PTE content according to fish species from WP1, *O. niloticus* had the highest content follow by *H. forskhalli* and by *C. angularis* (Figure 5-5) following the weight sequence from heaviest to lightest: *C. anguillaris* (omnivorous) > *H. forskahlii* (piscivorous) > *O. niloticus* (omnivorous) > *B. bajad* (piscivorous) with their corresponding fresh weights being 145.8±81.46 g, 82.64±24.20 g, 28.21±5.97 g and 27.9±5.37 g respectively (Figure 5-6). In WP2, the PTE content of fish sampled followed the sequence: HF > BB > ON for As, Cr, Ni, Pb; this sequence BB > HF > ON for Co, Mn, and this sequence BB > ON > HF for Cu, Zn. Lastly Mg followed this sequence HF > ON > BB (Figure 5-7).



As (mg/kg)
0
5
10
15
20
25
30
CA
HF
ON
(a)
Co (mg/kg)
0
1
2
3
4
5
6
7
8
9
10
CA
HF
ON
(b)
Cr (mg/kg)
2
4
6
8
10
12
14
16
18
CA
HF
ON
(c)
Cu (mg/kg)
0
2
4
6
8
10
12
14
16
18
20
CA
HF
ON
(d)
Mn (mg/kg)
0
200
400
600
800
1000
1200
1400
CA
HF
ON
(e)
Ni (mg/kg)
0
2
4
6
8
10
12
CA
HF
ON
(f)

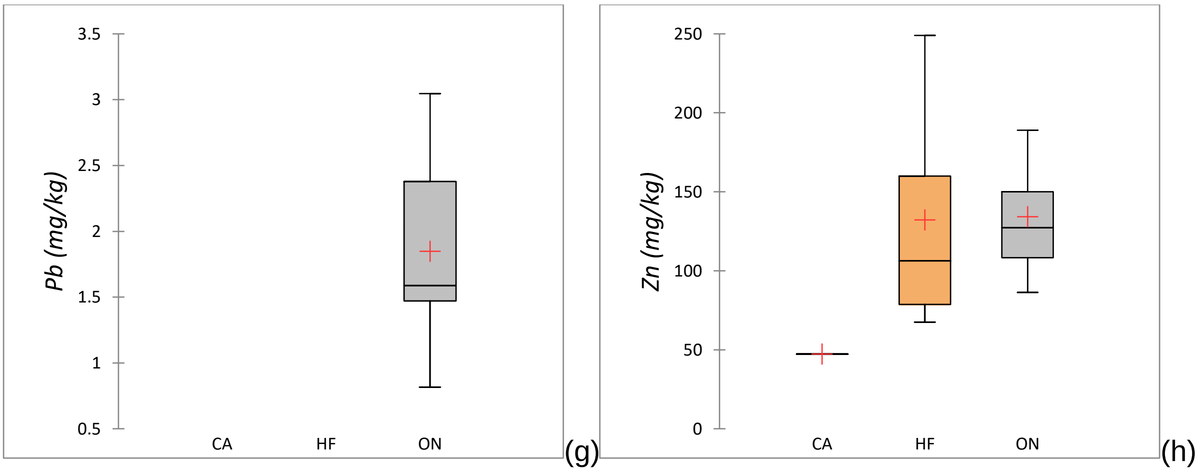
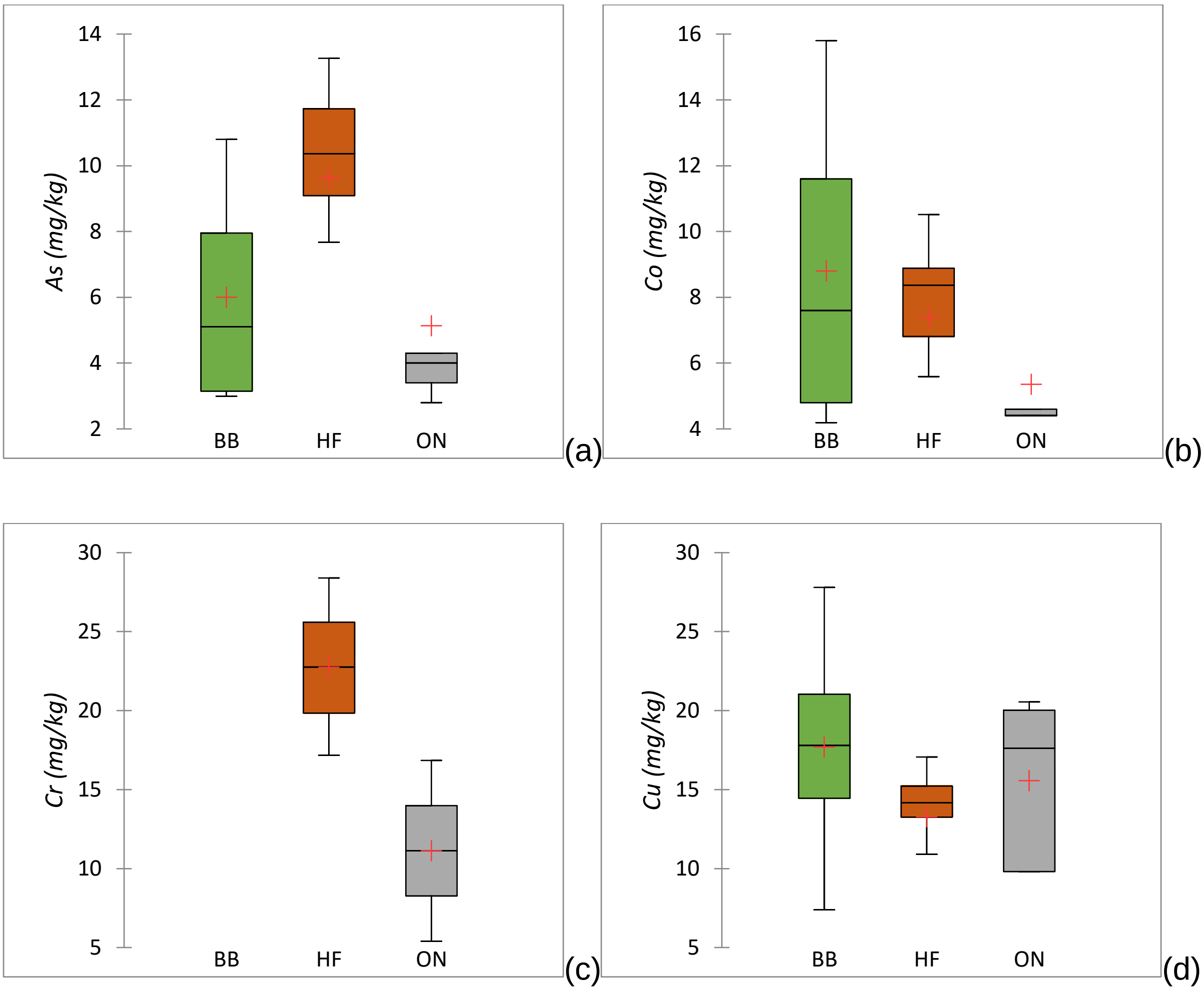


Figure 5-6 Potentially toxic element contents in fish species in West pit lake 1 (WP1), As (a), Co (b), Cr (c), Cu (d), Mn (e), Ni (f), Pb (g) and Zn (h). Values expressed in mg/kg dry weight, values below MDL were not plotted.



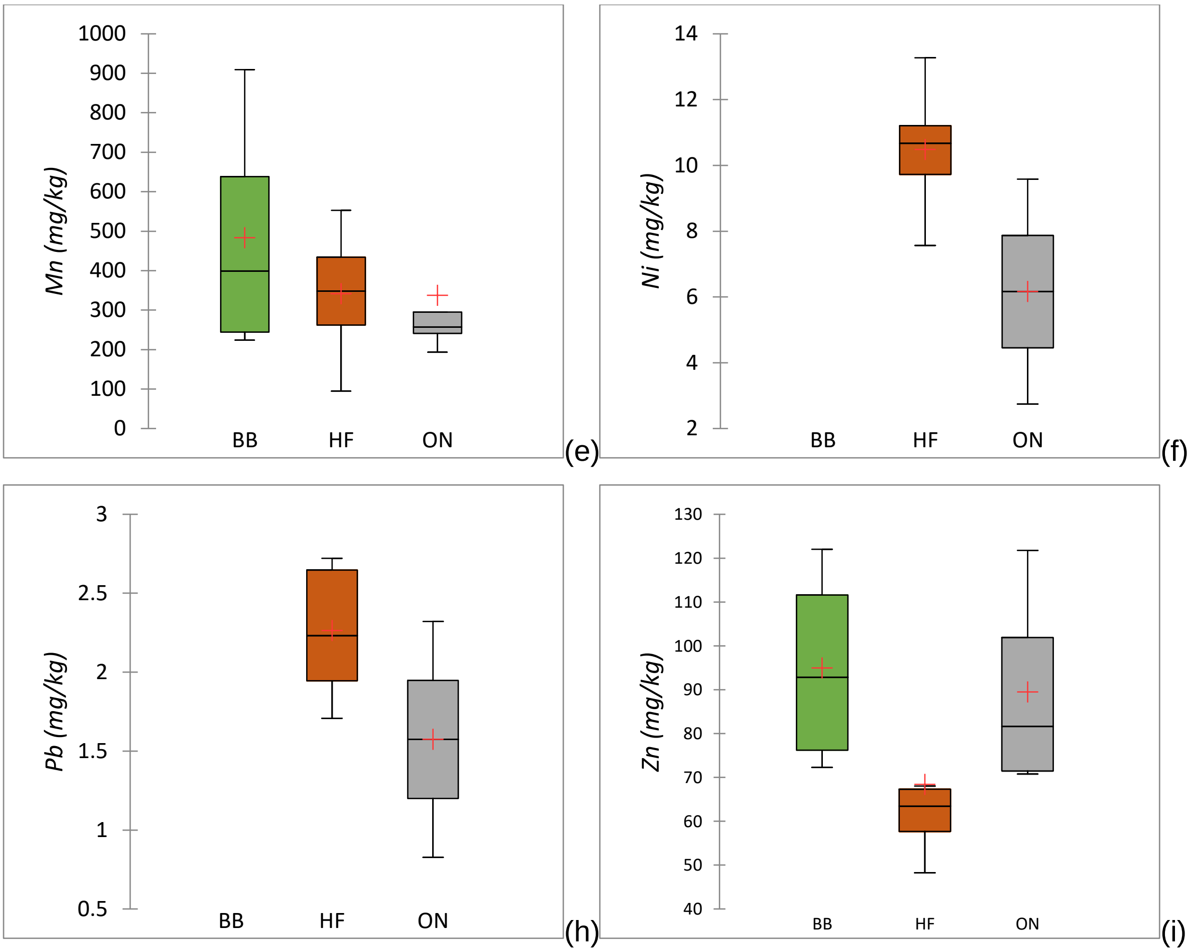


Figure 5-7 Potentially toxic element contents in fish species in West pit lake 2 (WP2), As (a), Co (b), Cr (c), Cu (d), Mn (e), Ni (f), Pb (g) and Zn (h). Values expressed in mg/kg dry weight, values below MDL were not plotted.

All in all, difference in water qualities resulted in difference in potentially toxic element contents in fish. Fish are affected by water quality.

5.3.5. Water to Fish Contaminant Transfer Factors

Transfer factors of potentially toxic elements from pit lake water to the fish were computed and benchmarked with those from NR (Table 5-4). Computing was carried out using water parameters. Based on TF, all fish considered in this study accumulated the potentially toxic elements investigated. High TF from water to fish could be explained by the bioavailability of potentially toxic elements due to specific physicochemical parameters of the pit lakes and NR, including concentration, solubility, pH, dissolved oxygen, temperature, salinity and hardness (Cogun and Kargın, 2004), as well as the biological factors of species such as feeding mechanism and age. Potentially toxic elements uptake follow several pathways in an aquatic environment: diffusion, absorption, ingestion of water and feeding on contaminated algae and bottom trophic levels (Magellan et al., 2014).

Cu levels in fish could have been influenced by the alkaline pH, which is above 7 in all the water bodies, with precipitation of Cu as carbonates reducing its biological availability (Birungi et al., 2007). The presence of Ca (18.05±9.38 mg/kg and 36.93 ± 14.09 mg/kg for WP1 and WP2 respectively) due to mobilization of soften calcareous soil, reduces Cu toxicity and enhances fish growth (Abdel-Tawwab et al., 2007). Furthermore, the presence of Ca^{2+} ions in the water bodies could lead to competition with the Mn^{2+} ion and reduce intake by fish. Zn as a key essential element which acts as a growth promoter in fish (Kumar et al., 2017) is found in all the fish investigated. In addition, uptake and accumulation of potentially toxic elements in fish could have been influenced by competition among potentially toxic elements for high affinity biotic ligand sites (Santore et al., 2001 and 2002; Balistrieri and Mebane, 2014).

Differences in transfer factors were found when results from either the entire fish or the fleshy part were used. Assessment of potentially toxic elements in fish, and particularly in *O. niloticus*, could be of interest in monitoring the possible impact of water contaminants on humans, as well as in anticipating their toxicity effect to the local population (Linde-Arias et al., 2008; Birungi et al., 2003).

Table 5-4 Estimated daily intake of contaminant through consumption of entire fish expressed in mg/adult/day and transfer factor (TF) of potentially toxic elements from water to fish. RfD = reference dose for EDI benchmarking (mg/Adult (70kg)/day except for Co in µg/adult (70kg)/day). Missing results were those not computed due to values below method limit of detection of elements, and/or no fishes sampled.

Elements (RfD)		As (0.15)		Cd (0.07)		Co (98)		Cr (21)		Cu (5)		Mg (250)		Mn (11)		Ni (0.20)		Pb (0.25)		Zn (25)	
Factors		EDI	TF	EDI	TF	EDI	TF	EDI	TF	EDI	TF	EDI	TF	EDI	TF	EDI	TF	EDI	TF	EDI	TF
ON	WP1	0.06	537	0.00		0.02		0.05		0.09		12.98	474	2.77	1899	0.03		0.01		0.72	332
	WP2	0.03	254	0.00		0.03		0.06		0.08	15495	16.6	423	1.9	1576	0.03		0.01		0.48	1270
	NR	0.01	1222	0.00		0.02		0.03		0.04		13.3	469	1.77	2139	0.01		0.00		0.57	4
HF	WP1	0.04	363	0.00		0.02		0.00		0.05		10.94	399	1.25	855	0.00		0.00		0.71	326
	WP2	0.03	290	0.00		0.03		0.12		0.05	9640	15.94	407	1.26	1045	0.06		0.01		0.49	1294
	NR	0.00		0.00		0.01		0.04		0.04		14.42	509	1.05	1268	0.02		0.00		0.54	4
BB	WP1																				
	WP2	0.03	274	0.00		0.05		0.00		0.09	17690	13.18	336	2.58	2140	0.00		0.00		0.51	1331
	NR	0.00				0.00		0.01		0.03		12.15	429	0.31	373	0.00		0.00		0.37	3
CA	WP1	0.00	39	0.00		0.00		0.00		0.07		7.02	256	0.12	79	0.00		0.00		0.25	117
	WP2																				
	NR	0.01				0.00		0.00		0.01		10.85	290	0.15	30			0.00		0.42	1

5.3.6. Fish Intake Risk Assessment

5.3.6.1. Estimated Daily Intake of Potentially Toxic Elements from Fishes

The population estimated daily food consumption rates were used to calculate the EDI of potentially toxic elements through consumption of fish. The assumptions on EDI of potentially toxic elements through fish have been made based on two suppositions: the first was that consumed fish all come from the pit lakes, and the second was that one kind of fish was eaten daily. The worst-case scenario results, computed based on entire fish analysis, is shown in Table 5-4.

Due to low consumption rates of fish in the nearby villages, potentially toxic elements intakes were below the referential doses. The EDI of potentially toxic elements varies between the fish. Entire fish consumption exposes an individual to higher intake of potentially toxic elements than consumption of only the fleshy part of fish. For As, the highest intake was consumption of entire *O. niloticus* from WP1 with a value of 0.406 mg/adult/week (0.06 mg/adult/day) which is 2.6 times less than the provisional tolerable weekly intake (PTWI) of 1.05 mg/adult/week (0.15 mg/adult/day). The Cd intake rate of the population regardless of the consumption mode (entire or fleshy part) was below the PTWI of 7 µg/kgbw/week. Despite the high content of Pb in entire fish, low consumption rates help to limit its intake to a maximum of 0.012 mg/adult/day. Consumption of entire fish exposes people to greater potentially toxic elements intake than consumption of the fleshy part.

5.3.6.2. Target hazard quotient

Target hazard quotient (THQ) was computed to estimate the potential health risks associated with the intake of fish. Fish from pit lakes showed higher values than fish from NR. *H. forskahlii* and *O. niloticus* had the highest quotient among the four species. Values computed were below one for both entire fish and the fleshy part. The THQ value when eating entire fish was higher than when consuming the fleshy part.

5.3.6.3. Allowable daily consumption

Allowable daily intake of fish depends on the potentially toxic elements content of the fish, the daily intake and the referential permissible intake values. As and Mn were the most common limiting factors from the intake of fish, followed by Ni and Zn. All allowable daily intake amounts were more than the intake rate of fish from all villages. Family members from the smaller households were exposed to more potentially toxic elements due to the higher intake of fish than families with a large number of persons in which consumption rates were lower. The tendency of families to have fewer members could in the future increase intake of fish, hence an increase in consumption of potentially toxic elements is anticipated. However, a margin exists, and the consumption rate could be increased by 264% when considering eating the fleshy part of fish instead of entire fish.

The most vulnerable age group are teenagers who could use the pit lakes for recreational fishing during holidays and class-free days. The body weight of the age group of 7 to 16 year-olds is estimated at 44.8 kg (USEPA, 2011) and was used to compute the allowable daily consumption of this age group; no excessive exposure was observed.

5.4. Conclusions

Firstly, the study investigated gold mine open pit lake water chemistry and revealed higher potentially toxic elements contents compared to a nearby river, attributable to leaching, washing and mobilization of excavated contaminant-bearing rocks. The potential use of pit lakes was decided after comparing the water chemistry to both CWQG and SAWQG. This comparison revealed that the water quality did not meet the requirement for the protection of aquatic life but was fully compliant with requirements for irrigation, animal watering and recreational use. Except for Fe, Zn and NO_3^-, it was also compliant with SAWQG aquaculture requirements.

Secondly, the study investigated potentially toxic elements contents in fish from the two pit lakes and found a similarity in fish between the two pit lakes and differences with the nearby river. Differences were linked to fish species, elements, fish parts analyzed (flesh or entire fish) and location. Higher potentially toxic elements contents were found in fish samples from the pit lakes than from the NR. In addition, analysis of entire fish showed potentially toxic elements content values higher than in the fleshy part. Pit lake water chemistry influences the elemental contents of fish. The transfer factors of all the fish species investigated were high, and values were even higher when considering entire fish compared to the fleshy part.

Thirdly, the fish intake survey revealed low fish consumption in the local villages, which significantly reduced exposure to potentially toxic elements to below referential doses for either consuming the fleshy part or the entire fish. The exposure risk assessment, given the low fish consumption, showed that pit lake fish were safe for human consumption; therefore, the pit lakes could be used for fish breeding to meet the nutritional needs of the local population.

The decision to use mining pit lakes as water reservoirs for the local population should be considered after assessing water quality, benchmarked to a nearby water body and to existing standards. Further exposure risk assessment for the local population is also a key factor in the decision-making process for the use of the pit lakes. It is then advised to consume fleshy part of the fish. Rehabilitation of open pit to pit lake should be done after long term analysis of water and fish, including intake survey for estimation of daily intake of PTE by local communities. National guidelines on PTE content in fish as well as threshold in PTE intake are also awaited.

Section 3

Chapter 6: EDTA and Growth Enhancer Influence on Arsenic Uptake by *Leucaena leucocephala* grown on Gold Mining Soil

This chapter is based on the paper:

Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (in preparation). EDTA and Growth Enhancer Influence on Arsenic Uptake by *Leucaena leucocephala* grown on Gold Mining Soil.

ABSTRACT: Arsenic (As) uptake by *L. leucocephala* was evaluated in a 45-day laboratory experiment with gold mine soil from Burkina Faso. A soil spiked with 25 and 50 mg As/kg, stabilized for 45 days, mixed with compost and supplemented with growth enhancer (GE) containing mycorrhiza or ethylenediaminetetraacetic acid (EDTA) or both was used. Parts of *L. leucocephala* plant grown in soil treated with As at 25 mg/kg with GE supplementation did not exhibit a significant difference compared with parts from plants grown in the same condition without GE. In 50 mg/kg As spiked soil, GE supplementation or EDTA supplementation or both did not reveal a significant difference in As uptake by roots and stems compared to each other when leaves did. The combined action of EDTA and GE significantly decreased As uptake in leaves compared to the same soil without EDTA supplementation. Multi-element spiked soil supplemented with both EDTA and GE revealed a significant increase in As uptake in leaves compared to the same soil spiked with As whilst no significant differences were found between roots and stems. EDTA and growth enhancer did not increase As uptake by *L. leucocephala*. It can also be deduced that treatment with EDTA or GE reduced the plants' total biomass production. Accumulation of As in the below ground part makes this plant more suitable for phytosequestration than phytoextraction and allows its use as forage for livestock in the after mine closure stage.

6.1. Introduction

The economic benefit of mining tends to override concerns about the environmental impact at the time of opening a mine. However, when the mine is in the exploitation stage, and especially when closure is approaching, environmental concerns become more prominent and land reclamation problems are highlighted. The environmental impacts of gold mines are considerable and need scientifically driven technology to mitigate the effects. The highest risk aspect of mining in terms of environmental impact is the contamination of land, water and air by potentially toxic elements which dictate land use after mine closure; land that is safe to use is characterized by the absence of these elements at dangerous levels. In cases where these elements are present at toxic levels, strategies to remove them should be put in place. One of the methods used to tackle this issue is phytoremediation which is an easy to implement, easy to operate and maintain, and low-cost technique (Wan et al., 2016). Phytoremediation is defined as a plant-based environmental-friendly technology for the remediation of contaminated sites, using plants and microbes to clean up contaminated air, soil and water (Singh et al., 2015; Fayiga & Saha, 2016). There are six basic phytoremediation mechanisms that can clean up mine sites: phytostabilization, rhizodegradation, phytoextraction, phytohydraulic, phytodegradation and phytovolatilization (Table 2-5); phytostabilization and phytoextraction are the ones relevant for this study as the concerned pollutants cannot be degraded nor volatilized. Phytostabilization also known as phytosequestration, is the ability of plants to sequester certain contaminants in the rhizosphere through exudation of phytochemicals and on the root through transport proteins and cellular processes. Phytoextraction also known as phytoaccumulation is the ability of plants to take up contaminants in the transpiration stream, stem or leaves tissues. Also other mechanisms were involved, i.e. roots prevent erosion, leaves prevent erosion from falling rain drops, evapotranspiration reduces infiltration of water.

Among many plant species that can be used for phytoremediation, *Leucaena leucocephala* seems to have several attractive properties (see 2.4.2). The plant offers multiple benefits in degraded land restoration (Normaniza et al., 2008), forest regeneration, biomass and energy production (Narayanaswami et al., 1986; Heineman et al., 1997), animal feed, cosmetic and pharmaceutical preparations (Nehdi et al., 2014) and phytoremediation (Schneider et al., 2013; Ssenku et al., 2017).

The ability of plants to grow in contaminated soils can be influenced by the addition of chelating agents and mycorrhizal fungi. When added to the soil, a chelating agent such as ethylenediaminetetraacetic acid (EDTA), can influence potentially toxic element bioavailability and tolerance by plants (Rahman et al., 2008; Yan et al., 2012; Abbas & Abdelhafez, 2013; Vigliotta et al., 2016; Luo et al., 2017; Han et al., 2018).

These chelating agents remove PTE with less impact on soil properties than other decontamination systems. In comparison with other chelating agents, EDTA presents the following advantages: a low degree of biodegradability in soil and moreover a high level of complexing capacity. EDTA not only can form soluble complexes with metals but may also influence the distribution of metals in the fractions by moving metals from less water-soluble fractions to more soluble fractions (Xia et al., 2009).

Application of EDTA in soil increased Mn uptake in tissue of *Polygonum pubescens* (Yu et al., 2019), alleviated the Pb-induced toxicity of *Juncus effusus* L. by improving its growth, biomass and antioxidative enzyme activities (Najeeb et al., 2017). The role of EDTA in arsenic mobilization and its uptake by maize grown on an As-polluted soil was investigated by Abbas & Abdelhafez (2013) and they found that EDTA contributed to As uptake by the plant. Further, Rahman et al. (2008) explained that EDTA phosphate interaction could influence As uptake in plant as it is well documented that As has the same pathway as phosphate. EDTA may not directly act toward As, but protects the added micro-nutrients from precipitations and fixation reactions, facilitating then As uptake by plant roots. EDTA also interacts with iron plaque to solubilize the binding of iron plaque containing As and can contribute to As mobilization to the environment. Arbuscular mycorrhizal fungi (AMF) can alleviate PTE-induced toxicity in plants by increasing plant tolerance. Pavithra & Yapa (2018) demonstrated that mycorrhizal fungi could help improve health and growth in harsh condition as drought. EDTA effect on Pb and Zn bioavailability and their influence on As uptake was investigated by Barbafieri et al. (2017) and found that EDTA impact on Pb and Zn bioavalibility influence As uptake. Gua et al. (2014) confirmed that EDTA combined to plant growth-promoting rhizobacterium and to CO_2, promoted ryegrass growth and increased As, Cd, Pb and Zn uptake.

The study reported in chapter 3 expressed soil contamination by potentially toxic elements with hotspots found in processing and tailings storage areas. Mining soil arsenic contents was found to be varying between 3.20 to 283.40 mg/kg with first quartile at 9.80 mg/kg, median at 26.30 mg/kg and an average of 44.56±59.30 mg/kg. The mining site therefore needs that contaminants be removed and a suitable way in this context is phytoremediation. *L. leucocephala* as an endogenous tree to the country is used for the purpose. This species has the ability to survive in degraded environments. Mining waste dumps lixiviation, tailing storage facility spillage and seepage contain very high arsenic content reported to around 0.4 mg/L and in acid mining drainage condition reach 1.5 mg/L (Migaszewski et al., 2019) which may arise in the after mine closure stage (Tabelin et al., 2020) to up to 1440 mg/L of As in shallow groundwater (Jelenová et al., 2018; Teixeira et al., 2020). Progressive oxidative decomposition of arsenopyrite in mining is an important source of As in surface environment (Wang et al., 2018) which could be largely accelerated by EDTA. Anticipating very high arsenic drainage to trees used in rehabilitation of mine appears as a wise strategy to achieve sound phytoremediation in ore bearing arsenopyrite rocks. The innovation comes from the fact that this study uses mining soil spiked with EDTA and mycorrhiza fungi inoculum to measure *L. leucocephala* capability to uptake contaminants. This study will then fill the gap of lack of data on use of *Leucaena leucocephala* in spiked soil with As and supplemented with a growth enhancer; soil being taken from the mining area.

The objectives of this study were to evaluate the capability of *L. leucocephala* to uptake As in an As contaminated soil watered with As contaminated water. The laboratory experiment was a simulation of a contaminated mining site drained by runoff from As contained mining waste dumps. Further, the influence of chelating agent EDTA and growth enhancer (GE) containing mycorrhiza sp. on the uptake capability was assessed. In addition, As translocation was computed to check if *L. leucocephala* has phytoextraction potential in these specific conditions.

6.2. Materials and methods

6.2.1. Soil Preparation

A hydromorphous soil with a silty clay texture (Etruscan, 2005) was obtained from a gold mine in Burkina Faso (11°10’N, 00°18’W), sun dried, and then transported to Ghent University Campus Kortrijk. Subsequently, coarse rocks and plant roots were removed from the soil by hand picking. Then the soil was spiked by means of an arsenic (prepared by dissolving solid As_2O_3) solution equivalent to a level of 25 mg/kg or 50 mg/kg As and kept for 45 days to stabilize, allowing As to interact with substances affecting its bioavailability in the soil prior to seeding as recommended by Schneider et al. (2017). A moisture level of 6% (w/w) was maintained during this incubation period. Choice of spiking levels was based on previous soil screening (Compaoré et al., 2019a).

After the 45-day stabilization period, 25 and 50 mg/kg spiked soils and non-spiked control soils were mixed with compost (Vlaco vzw, Mechelen, Belgium) at a level of 50% (w/w) to provide nutrients for plant growth. They are designated in this chapter as “spike_25”, “spike_50” and “spike_0” respectively. One kg of the respective mixes was then put in a polyvinyl chloride conical pot (15 cm depth) for further experimentation.

6.2.2. *Leucaena leucocephala* Seedlings

Seeds of *L. leucocephala* were purchased from Zadengigant, The Netherlands, and scarified first to accelerate germination (Bray, 1986). Subsequently, the seeds were soaked for two days in tap water to facilitate the germination, before being transferred to a pot containing a commercial compost substrate, and left to germinate and grow for 45 days in a growth chamber. Conditions were set to a temperature of 21 ± 2°C and a light/dark regime of 12/12 hours. After 45 days, seedlings were transplanted to the previously prepared PVC pots with soil-compost mixes, at one seedling per pot. Four seedlings, randomly selected, were kept aside and used to determine their initial content of As, Pb and Zn (see 2.5). Their initial dry weight was also recorded.

6.2.3. Pot Experimental Set Up

The experiment was designed with one control and nine treatments, each set up in eight replicates except for treatment 9 for which only 4 replicates were available (Table 6-1). The control (coded 00NINE) consisted of spike_0 medium without addition of growth enhancer (GE) as inoculum of mycorrhiza nor EDTA, and no metals in the irrigation water. Treatments 1-4 evaluated the effect of GE and/or EDTA addition to spike_25 medium and received irrigation water with a concentration of 25 mg/L As; treatments 5-8 evaluated the effect of GE and/or EDTA addition to spike_50 medium, and received irrigation water with a concentration of 50 mg/L As; treatment 9 repeated the conditions of treatment 4, except for the irrigation water which consisted in this case of a multi-element solution of As, Pb and Zn at 25 mg/L each to evaluate the effect of metal ion competition on potentially toxic element uptake. EDTA was added weekly as a 25 mL 5 mM solution based on the positive results obtained by Najeeb et al. (2017).

The GE consisted of a commercial mixture of *Mycorrhiza sp., Bacillus sp.*, yeast and enzymes and was added only once at the start of the experiment at a dose of 1 kg/m^3, as recommended by the supplier (Bio Nova B.V., Elzenweg, Netherlands). Irrigation water was supplied every other day, in 40 mL aliquots and this during 20 repeated occasions. Distilled water was used for the control, for treatments 1-8 an arsenic solution prepared from As_2O_3 was used, and for treatment 9 a solution of $PbCl_2$, $ZnC_4H_6O_4$ and arsenic solution was used. During the experiment, the following climatic conditions were set: a temperature of 21 ±2°C and a light/dark regime of 12/12 hours.

Table 6-1 Overview of the experimental set up. (N)I = (not) inoculated with growth enhancer (GE); (N)E = (no) EDTA added; ME = multi-element.

		Code	Spike level (As)	EDTA (weekly 25 mL)	GE (at start only)	Irrigation water (40 mL every other day)	n
Control		00NINE	00 (mg/kg)	-	-	00 mg/L As	8
Treatments	1	25NINE	25 (mg/kg)	-	-	25 mg/L As	8
	2	25NIE	25 (mg/kg)	5 mM	-	25 mg/L As	8
	3	25INE	25 (mg/kg)	-	1 kg/m^3	25 mg/L As	8
	4	25IE	25 (mg/kg)	5 mM	1 kg/m^3	25 mg/L As	8
	5	50NINE	50 (mg/kg)	-	-	50 mg/L As	8
	6	50NIE	50 (mg/kg)	5 mM	-	50 mg/L As	8
	7	50INE	50 (mg/kg)	-	1 kg/m^3	50 mg/L As	8
	8	50IE	50 (mg/kg)	5 mM	1 kg/m^3	50 mg/L As	8
	9	25ME	25 (mg/kg)	5 mM	1 kg/m^3	25 mg/L As/Pb/Zn	4

6.2.4. Soil Parameter Measurement

All soil samples were oven dried at 40 °C overnight prior to further analysis. As, Pb and Zn concentrations in the original soil and in the soil-compost mixes were determined via total digestion analysis. The method used was that of Bettinelli et al. (2000) and Melaku et al. (2005), with prior microwave digestion (ETHOS Touch Control Advanced Microwave Labstation from Milestone Inc® Monroe, CT, USA) using a mixture of nitric acid, hydrochloric acid, hydrofluoric acid and boric acid in a volumetric ratio of 2:6:2:2. In short, 0.5 grams of sample was weighed directly in a Teflon digestion tube with the first three acids and heated in the microwave at 250 W for 8 minutes, followed by 400 W for 4 minutes and 600 W for 6 minutes and then the vessels were cooled down for 10 minutes. After cooling the vessels, 2 mL of saturated boric acid was added, and the sample was heated again at 300 W for 3 minutes and cooled down again. The digestate was then filtered with a Whatman filter n°41 into a 50-mL flask and made to volume with bi-distilled water. Potentially toxic elements were then analyzed by ICP 7000 (Thermo Scientific™ iCAP™ 7000 Plus Series ICP-OES, Thermo Fisher Scientific Brand, USA) equipped with Qtegra Software and CETAC ASX-560 autosampler (Teledyne CETAC Technology, USA).

Total organic matter (TOM) was analyzed gravimetrically using the loss on ignition method at 550 °C (Heiri et al., 2001). Cation exchange capacity (CEC) was analyzed using Chapman's method with ammonium acetate at pH 7, as described by Ross & Ketterings (2011). The resulting ammonium concentration was measured using LCK 303 test kits and a DR 2800 spectrophotometer (Hach Lange, Germany). The Olsen phosphorus method (Sims 2000) was used to estimate the relative bioavailability of ortho-phosphate (PO_4-P) in soils by extraction using alkaline sodium bicarbonate (pH 8.5) solution and colorimetric determination using refractometry (UV-VIS spectrophotometer, UV-1280, Shimadzu, Shimadzu Europa Gmbh, Germany). Finally, pH was measured in a ratio of 1:5 soil in bi-distilled water.

6.2.5. Potentially Toxic Elements Determination in *Leucaena leucocephala*

After 45 days of growth, all plants were carefully uprooted, and above-ground leaves and stems were separated, as were roots. Roots, stems and leaves were separately washed with tap water then with 10% nitric acid followed by distilled water. All parts were then oven dried at 40°C overnight. Weights were recorded.

Subsequently, potentially toxic element (As, Pb and Zn) concentrations were analyzed. Approximately 0.2 g of dried biomass was taken and microwave-digested in Teflon tubes with a mixture of commercial 65% concentrated nitric acid and oxygen peroxide at a ratio of 4:2

mL using a digestion program of 400 W for four minutes followed by 600 W for ten minutes. After cooling the Teflon tubes, digestate was filtrated with a Whatman filter N° 413 and made up to 25 mL with bi-distilled water and analyzed on the same ICP-OES as described before. Acid matching standard solutions were prepared based on a Certipur® ICP (inductively coupled plasma) multi-element standard XIII from Merck.

6.2.6. Contaminant Uptake and Factors Calculation

L. leucocephala As uptake was evaluated based on biological concentration factor (BCF), biological accumulation coefficient (BAC) and translocation factor (TF).

The BCF describes the ability of a plant to transfer potentially toxic elements from soil to root and was calculated as the ratio of the concentration of targeted potentially toxic elements in the *L. leucocephala* roots to the concentration of potentially toxic elements in soil (Hosseini et al., 2018).

The BAC describes the ability of the plant to translocate potentially toxic elements from soil to the above-ground parts (stem and leaves) of the plant and was calculated as the ratio of the concentration of potentially toxic elements in plant above-ground parts to the concentration of target elements in soil (Fitz & Wenzel, 2002).

Note that for both the BCF and BAC, the calculation was done based on soil concentrations after spiking and mixing with compost and including the additions by watering (Table 6-2).

The TF measures the capacity of the plant to translocate potentially toxic elements from roots to the above-ground parts of the plant (Han et al., 2018). It is calculated as the ratio of the concentration of potentially toxic elements in plant above-ground parts to the concentration in below-ground parts.

6.2.7. Quality Assurance and Statistics

Results obtained from the analysis of plants and soils were submitted to an ANOVA and post hoc Tukey HSD (honest significant difference) test. The method detection limit (MDL) was determined as described in 3.2.3 and statistical analysis was done as in 3.2.5. Potentially toxic elements are expressed in mg/kg dry weight or as otherwise stated. Values were expressed as the mean ± standard deviation.

6.3. Results

6.3.1. Physicochemical Properties of Soil

The pH of the control-mix (00NINE) was 7.51 ± 1.2, Olsen P was 125.90 ± 28.95 mg/kg, TOM was 20 ± 3 % and the CEC 6.13 ± 0.16 meq/100g. These parameters are key as they influence element uptake by the plant by directly affecting plant behavior or by making elements bioavailable for uptake by the plant. Table 6-2 gives an overview of potentially toxic elements in the different set. Analysis of As, Pb and Zn was done after mixing with compost in order to know the value including compost contribution.

Table 6-2 Total digestion analysis of potentially toxic elements (mg/kg) in the original soil and in the spiked soil-compost mixes, and input via watering (mg). (n/a = not applicable),

	Soil mixture content			Input via watering		
Label	As	Pb	Zn	As	Pb	Zn
Original soil	6.19±1.85	65.94±6.17	13.10±2.57	n/a	n/a	n/a
Spike_0 (00NINE)	6.01±0.00	26.02±8.01	24.40±4.45	n/a	n/a	n/a
Spike_25 (25NINE)	15.03±2.32	26.02±8.01	24.40±4.45	20	0	0
Spike_50 (50NINE)	32.66±4.69	26.02±8.01	24.40±4.45	40	0	0
Multi-element (25ME)	15.03± 2.32	26.02±8.01	24.40±4.45	20	20	20

6.3.2. Plant Biomass Production

Plants grown in the control (00NINE) showed higher biomass production than those grown in soil spiked and watered with As solution (25NINE and 50NINE), and the effect is more pronounced at higher As concentrations (Figure 6-1). On the other hand, plants grown in a medium supplemented with EDTA revealed slightly lower biomass production than those without ETDA, regardless of GE supplementation. Moreover, biomass values were slightly higher for plants with GE supplementation than plants without GE supplementation. In addition, plants grown in medium supplemented with both GE and EDTA exhibited biomass production lower than those with no supplemented medium and also those grown with a single supplement (EDTA or GE). It can be deduced that treatment with EDTA or GE reduced the plants' total biomass production. The decrease in biomass can been explained by the fact that EDTA could affect the mineral balance, then disturb the metabolism of cells and impact cell membranes. The effect could also be due to the enhanced dissolution of potentially toxic elements. Statistically significant differences were not found between experimentation sets regarding biomass production.

Variations were noticed when considering plant parts. From the lowest to the highest, the leaves biomasses were 0.04 g to 0.06 g per plant following this trend 50NIE < 50IE < 25NIE < 50NINE < 25NINE < 25INE < ONINE. The root biomass ranged from 0.02 to 0.03 g per plant in this trend 25NIE < 25IE < 50IE < 50INE < ONINE < 50NINE < 25NINE < 25INE < 50NIE respectively. The biomass of the stem ranged from 0.01 to 0.3 g per plant and in this trend 25NIE < 25MEIE < 25IE < 50INE < 50IE < 25NINE < 0NINE < 50NINE < 50NIE < 25INE. Experimenting with supplementation of chelating agents like EDTA, Vigliotta et al. (2016) noticed significant decreases in leaf and stem biomass of *Zea mays* L., compared to medium without EDTA, and no substantial change in root biomass. Furthermore, the fluctuation of biomass of roots in this study when soil was supplemented with GE As, was in line with the study of Schneider et al. (2013) who reported a reduction in root dry matter with an increase in As supplementation and with *G. clarum* inoculation; they also concluded that AMF had an effect ranging from reduction to increased levels of As depending on the species.

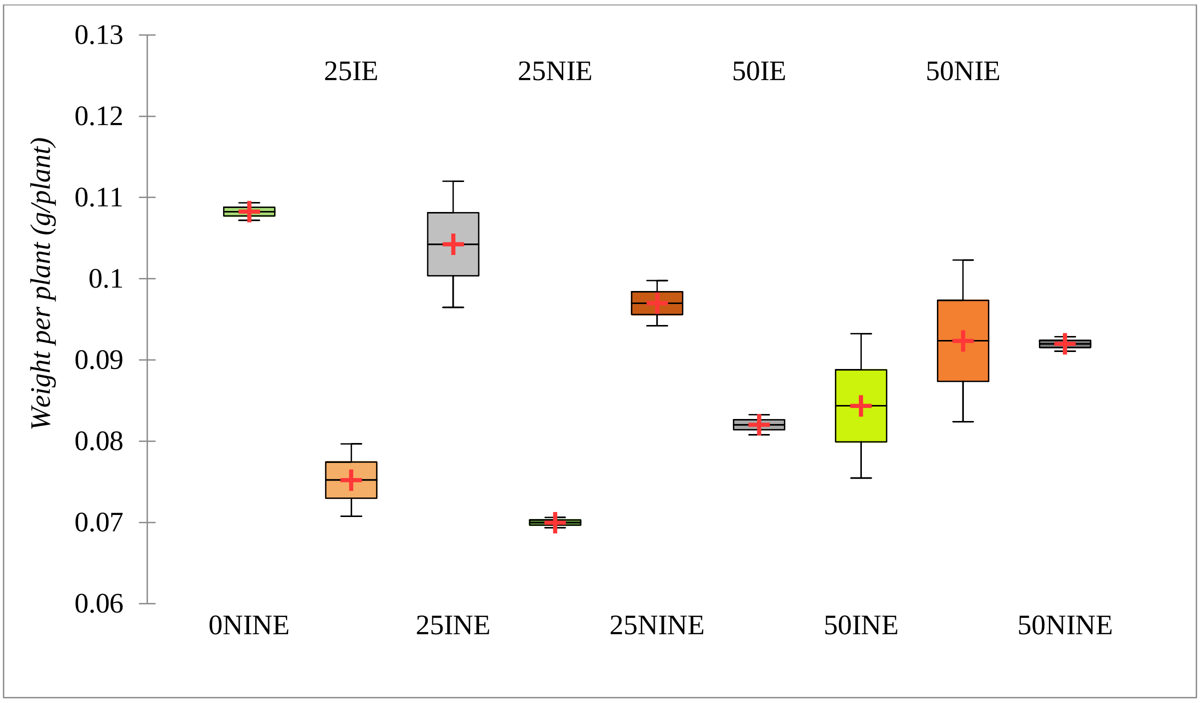


Figure 6-1: Plant biomass dry weight after 45 days according to supplementation growth factors. 25 means spike_25 and watered with 25 mg/kg As solution, 50 means spike_50 and watered with 50 mg/kg As solution, (N)I means (No) GE supplementation, (N)E means (No) EDTA supplementation.

6.3.3. Arsenic Uptake by *Leucaena leucocephala* in As Spiked Soil

The potentially toxic element levels (As, Pb and Zn) of *L. leucocephala* seedlings analyzed before transferring of seedlings from the compost medium to the pot for the experiment were found to be below the 0.36 mg/kg, 1.10 mg/kg and 0.50 mg/kg method detection limits (MDL) for As, Pb and Zn, respectively.

L. leucocephala grown in soil treated with different levels of As and without EDTA and GE (25NINE and 50NINE) supplementation was compared to the control soil plants (00NINE) (Figure 6-2). In general, *L. leucocephala* As uptake followed this pattern: roots > stems > leaves. The HSD test revealed that differences existed between *L. leucocephala* parts grown in spiked soils and parts of *L. leucocephala* grown in the unspiked control soil, especially in leaves and roots. However, no significant differences in As uptake were found between spike_25 and spike_50.

L. leucocephala grown in spike_25 supplemented with GE exhibited As content levels for both roots and stem two-times less than plants grown in the same soil without supplementation with GE (22.86 ±13.17 mg/kg to 47.55 ± 19.32 mg/kg and 7.36 ± 0.39 mg/kg to 13.34 ± 1.49 mg/kg for roots and stems respectively). Moreover, the pattern roots > stems > leaves were maintained (Figure 6-2). In leaves, the same trend was observed, with a significant decrease in As content compared to samples from the same soil without GE supplementation. In stems, no significant difference was observed. In high As spiked soil (spike_50), unlike moderate (spike_25) As contaminated soil, roots in soil supplemented with GE did not exhibit a significant decrease in As content compared to roots grown in the same soil without GE supplementation. Stems in spike_50 samples did not show a significant decrease in As content compared to stems in spike_50 without GE supplementation. On the other hand, leaves from spike_50 exhibited significantly higher As levels (36% high) when supplemented with GE than when not supplemented with GE. When considering stems, no significant differences were found between settings.

L. leucocephala growth in control and spiked soils supplemented with EDTA followed this trend: leaves < stems < roots. Differences were found between parts of *L. leucocephala* grown in spiked soil versus the control soil (Figure 2). Tukey HSD pairwise comparison revealed significant differences in stem, roots and to some extent in leaves of *L. leucocephala* grown in spike_25 and spike_50 compared to the unspiked control soil. These differences included a 14-fold increase in root As level from samples grown in the unspiked control soil compared to the level found in spike_25 and spike_50 supplemented with EDTA. Also, an 18-fold increase was found between stems of *L. leucocephala* in unspiked control soil and both spike_25 and spike_50 with EDTA supplemented. No significant differences were found in roots from *L. leucocepaha* grown in spike_25 and spike_50 supplemented with or without EDTA and roots from spike_25 and spike_50 without any supplements. Significant differences were found in leaves of *L. leucocephala* grown in spike_50 supplemented with EDTA and control soil. In spike_50 supplemented with EDTA, As concentration in leaves increased by 350 % compared to unspiked control sample leaves. Other scenarios did not reveal significant differences. Based on these findings, it can be seen that As was translocated to leaves when As was in high concentration in the medium. EDTA supplementation in soil for *L. leucocephala* did not reveal any significant difference for As content in *L. leucocephala* above-ground parts (leaves in spike_25 and stems in spike_50) compared to *L. leucocephala* grown in a medium without an EDTA supplement.

In spiked soils treated with both GE and EDTA, As accumulations in *L. leucocephala* parts follow the trend: roots > stems > leaves (Figure 6-2). Tukey HSD test showed that stems of *L. leucocephala* grown in spike_25 and spike_50 did not exhibit a significant difference to unspiked control soil. In the same trend, no significant differences were found between stems from *L. leucocephala* grown in spike_25 or spike_50 supplemented with EDTA and GE and stems from *L. leucocephala* grown in spike_25 and spike_50 supplemented with EDTA and also stems from spike_25 and spike_50 supplemented with GE. Leaves from *L. leucocephala* grown in spike_50 and supplemented with both EDTA and GE revealed a significant difference with the unspiked control soil, whilst As from leaves of *L. leucocephala* grown from spike_25 supplemented with both EDTA and GE did not show a significant difference compared to unspiked control soil samples. *L. leucocephala* leaves As content from spike_25 supplemented with EDTA and GE was significantly different from As in leaves grown in spike_25 without EDTA and GE. In spike_50 supplemented with EDTA and GE, *L. leucocephala* leaves did not reveal a significant difference with spike_50 without EDTA and GE supplementation. In addition, spike_25 with both EDTA and GE supplementations did not reveal a significant difference in As content in leaves with those grown in the same spike_25 with only EDTA or with only GE. A significant difference was found between spike_50 supplemented with both EDTA and GE and unspiked control soil, when there was no significant difference between spike_50 and EDTA and GE samples and spike_50 control samples. In addition, spike_50 supplemented with only EDTA or only GE did not show a significant difference with spike_50 supplemented with both EDTA and GE. However, compared with the same supplement condition, not supplementing with GE exhibited a significant difference at spike_50 but not at spike_25. Also, there was no significant difference when not supplementing with EDTA.

A set consisting of 25 mg/kg As spiked soil and supplemented with both EDTA and GE watered with a solution containing As, Pb and Zn has been considered and the results compared with single As spiked soil (Figure 6-3). In general, As content in leaves grown in soil watered with multi-elements revealed a significant difference (increase in As level) compared with leaves of plants grown in spike_25 and in spike_50, whether supplemented or not with EDTA or GE or both EDTA and GE. However, there was an exception in that there was no significant difference compared with spike_50 supplemented with only GE. In roots, supplementation with both EDTA and GE and watering with multi-element solution showed a significant difference (decrease in As level) with firstly spike_25 control soil (25NINE), secondly with spike_50 control soil (50NINE). Other scenarios did not show significant differences (Table 6.3).

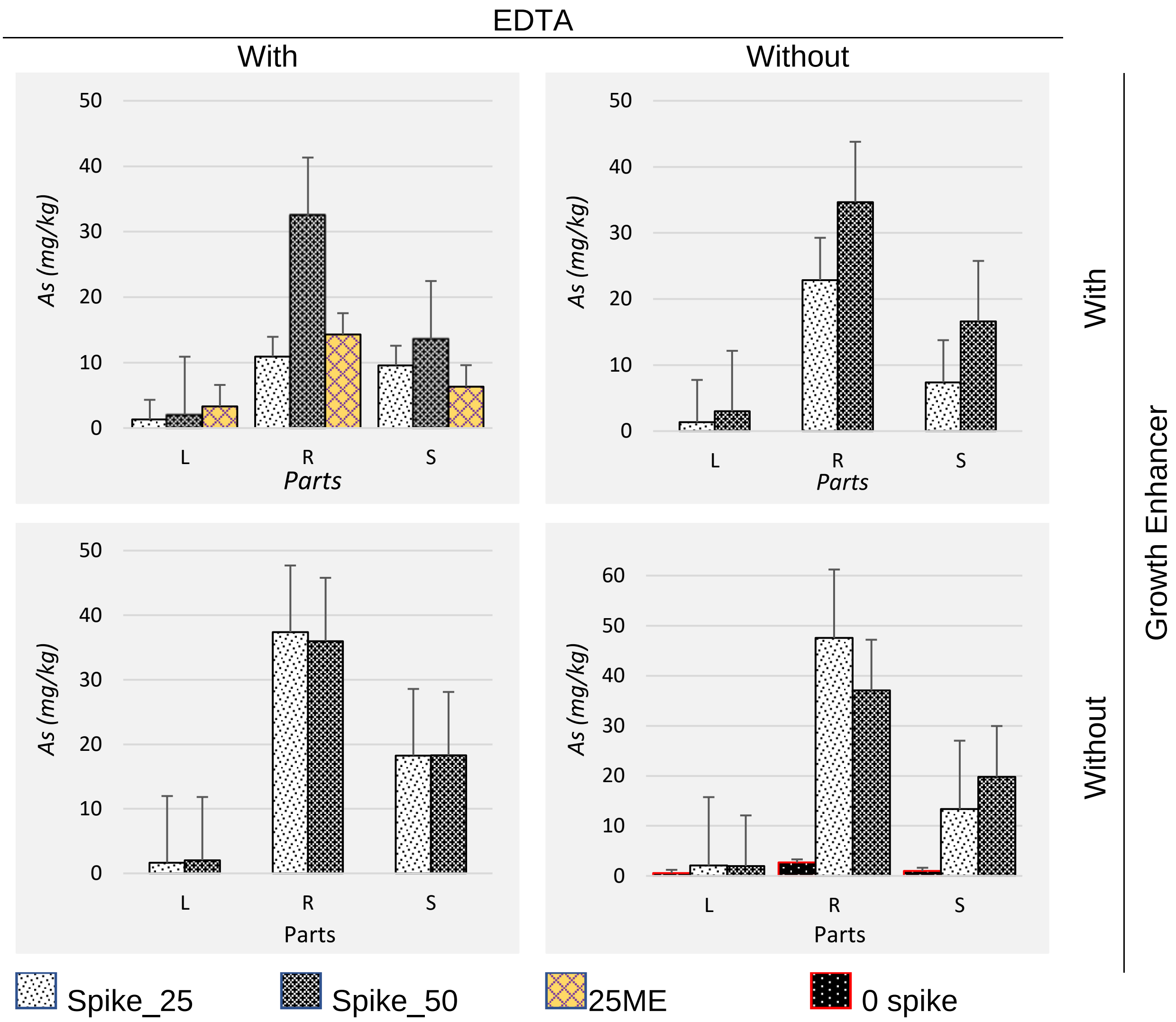


Figure 6-2 As uptake by *L. leucocephala* growth in medium supplemented with different factors, spike_25 and watered with 25 mg/L As solution; spike_50 and watered with 50 mg/L As solution, 25ME means 25 mg/L multi-elements treatment. L is for leaves, R for roots and S for stems. Positive error bars shown only.

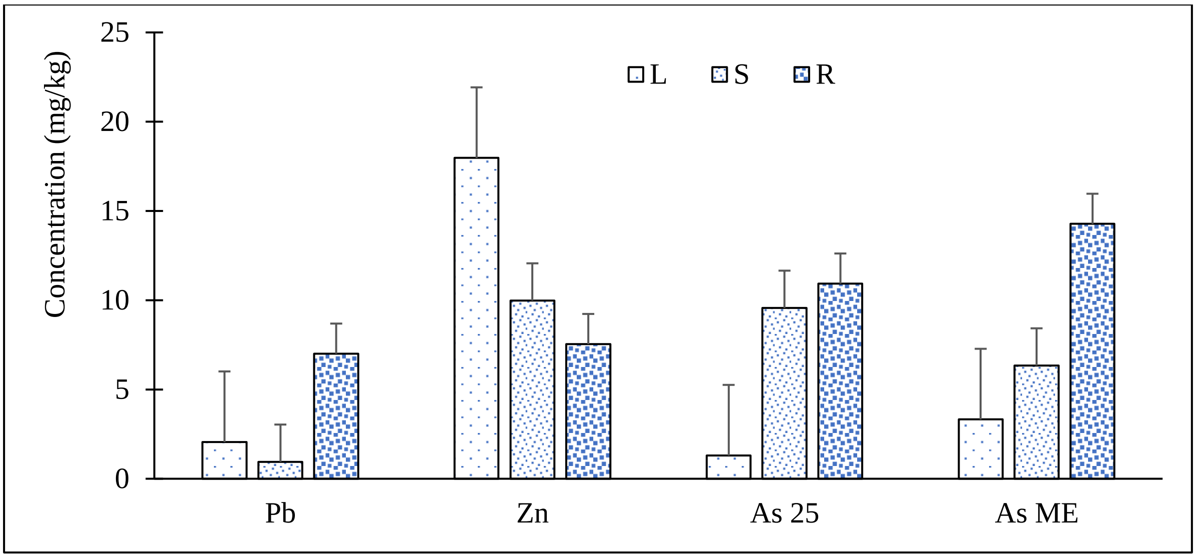


Figure 6-3 As uptake in 25 mg/kg single element spiked (As 25) and in 25 mg/kg multi-element spiked growth medium (As ME) with the element spiked uptake, Zn and Pb, L for leaves, R for roots and S for stems.

Table 6-3 As (mg/kg, dry weight) uptake by *L. leucocephala* according to growth condition

Growth condition			L	S	R
As Spike	GE	EDTA	Mean	Mean	Mean
00	NI	NE	0.58±0.23^{d}	0.99±0.28^{bc}	2.67±0.41^{d}
25	I	E	1.30±0.12^{d}	9.57±3.08^{abc}	10.93±0.99^{cd}
		NE	1.35±0.21^{cd}	7.36±0.39^{bc}	22.86±13.17^{bc}
	NI	E	1.66±0.26^{bcd}	18.25±2.96^{ab}	37.38±12.20^{ab}
		NE	2.06±0.19^{b}	13.34±1.49^{abc}	47.55±19.32^{a}
50	I	E	2.03±0.26^{b}	13.59±5.28^{abc}	32.46±6.04^{ab}
		NE	2.99±0.22^{a}	16.60±2.36^{abc}	34.63±10.16^{ab}
	NI	E	2.03±0.24^{b}	18.30±11.36^{ab}	35.98±1.56^{ab}
		NE	1.95±0.13b^{c}	19.81±5.76^{a}	37.06±2.52^{ab}
25ME	I	E	3.32±0.06^{a}	6.34±0.99^{c}	14.28±0.27^{bcd}

Two combinations in the same column with no letter in common are significantly different

6.3.4. Biological Accumulation Coefficient and Biological Concentration and Translocation Factors

The BCF, BAC and TF are shown in Figure 6-4. The BAC for all scenarios was below one. With regards to As BAC, *L. leucocephala* in the condition of this experimentation, spiked soil with continuous watering with arsenic solution, regardless of the supplementation, did not exhibit phytoextraction potential.

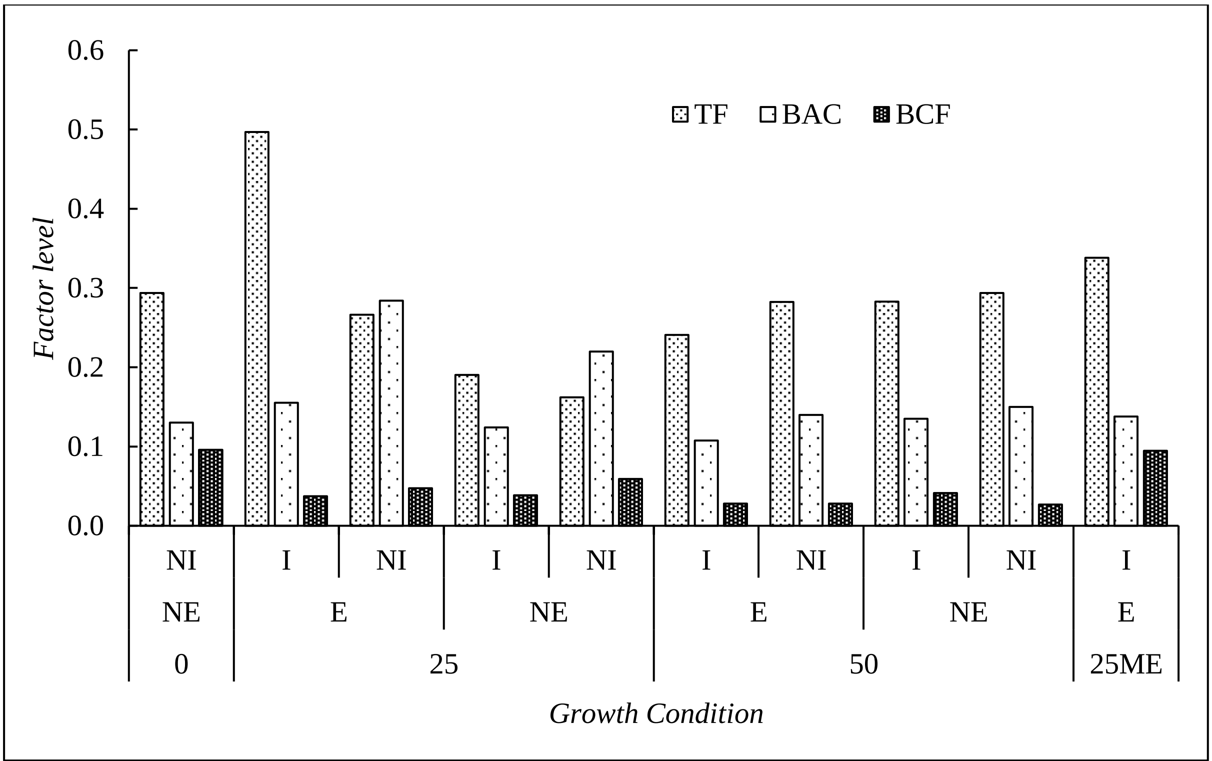


Figure 6-4 *L. leucocephala* As uptake factors

The BCF values ranged from 0.03 to 0.10. The BCF in plants grown in soil treated with 25 mg/kg of As was higher than the BCF of those grown in soil contaminated with 50 mg/kg As. This difference could represent a limit in *L. leucocephala* uptake capability from soil and the absence of a linear root to soil ratio. The highest BCF value (0.10) was from plants grown in control soil watered with tap water. In general, plants grown in spike_25 with the presence of GE showed increased BCF values. In spike 2, the BCF values were between 0.03 and 0.04. In addition, the multi-element spiking condition showed the a BCF value of 0.09 revealing that the presence of other elements could increase As transfer from soil to roots of *L. leucocephala*.

The TF of *L. leucocephala* grown in the control soil, not spiked and not supplemented with GE or EDTA, exhibited a value of 0.29. The TF of As in *L. leucocephala* increased in the presence of EDTA with supplementation with GE in spike 1. On the other hand, in spike 2, the TF decreased when GE supplement was present with or without EDTA. The TF of *L. leucocephala* grown in spike_25 and watered with multi-element solution (As, Pb and Zn) and supplemented with both EDTA and GE showed a value above the TF of spike 2 *L. leucocephala* regardless of the supplementation. The TF of *L. leucocephala* were below 1, making *L. leucocephala* better suited for phytostabilization than phytoextraction in these conditions and non-hyper-accumulator (Fayiga and Saha, 2016).

Translocations from soil to the above-ground parts (stem and leaves) were higher in spike_25 than in spike 2. In both spike_25 and spike_50, absence of supplementation with GE increased BAC value.

6.4. Discussion

6.4.1. *Leucaena leucocephala* As Uptake in Soil without any Supplementations

Firstly, As uptake of *L. leucocephala* parts is correlated with the content of As in the soil (25 mg/kg spiked soil and 50 mg/kg spiked soil). Differences were found between *L. leucocephala* parts grown in spiked soil and parts of *L. leucocephala* grown in unspiked soil. Significant increases in As content in leaves, stem and roots were observed between *L. leucocephala* grown in control soil and those grown in spiked soils except between stem *of L. leucocephala* from spike_25 and control sample. Aboveground parts (stem and leaves) As uptake in control soil did not exceed 1 mg/kg which increased by 4, 13 and 18 times for leaves, stems and roots respectively compared to As content in parts of L. l*eucocephala* grown in spike_25 and to 3, 20 and 14 times for leaves, stems and roots respectively compared to same part of *L. leucocephala* frown in spike_50. As uptake is related to As content in soil. The relationship Predictive relationship between As uptake by *L. leucocephala* and As content in soil was estimated (Figure 6-5). The linear regression of results of control, spike_25 without supplementation nor EDTA addition showed R squared values of 0.53, 0.77 and 0.42 for respectively leaves, stems and roots.

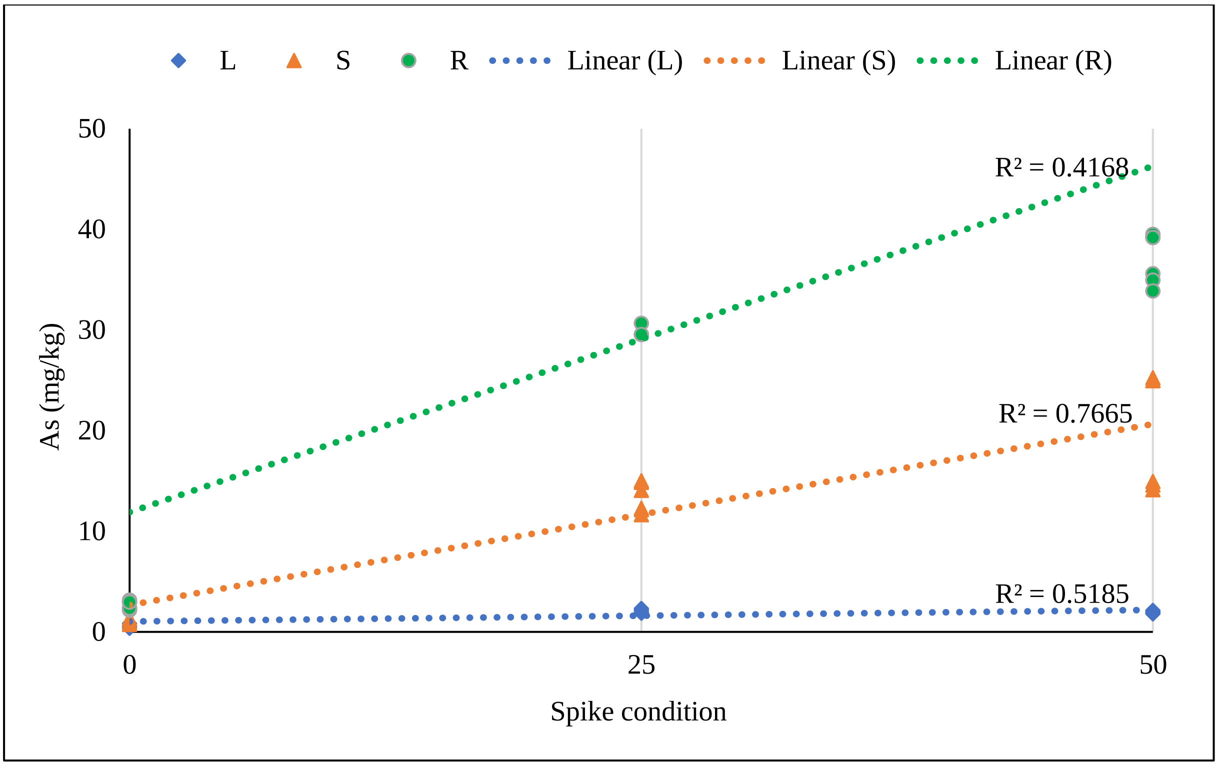


Figure 6-5 As uptake by *L. leucocephala* grown in control soil and spike_25 and spike_50, without EDTA and GE supplementation, L for leaves, S for stems and R for roots.

The As content in stems and roots of *L leucocephala* grown on spike_25 was not significantly higher than in stems and roots from *L. leucocephala* grown in spike_50. Lack of significant difference between spike_25 and spike_50 in stems and root could be due to the presence of a threshold in As uptake and tolerance by *L. leucocephala*. Arsenic accumulation is factor-dependent, and the factors are mainly the plant species, and the level of As in the soil (Figure 6-6), as well as the soil physicochemical properties, including pH, redox potential, and electrical conductivity. Uptake of As by plants uses the same transporter pathway and carrier as phosphate due to their similarity (chemical analogue: $H_2PO_4^-$ and $H_2AsO_4^-$) (Fitz & Wenzel, 2002; Zhao et al., 2009; Rahman & Hasegawa, 2011). Fitz & Wenzel (2002) found that nitrogen nutrition responsible for the cation/anion uptake ratio affects pH; therefore, fertilization increases pH and then As accumulation in plant tissue. Compost addition could then have contributed to As uptake by *Leucaena leucocephala*. Organic matter provided mainly by the compost could have induced adsorption/immobilization mechanisms which could have interacted with cations, rendering anions more bioavailable in the milieu for uptake.

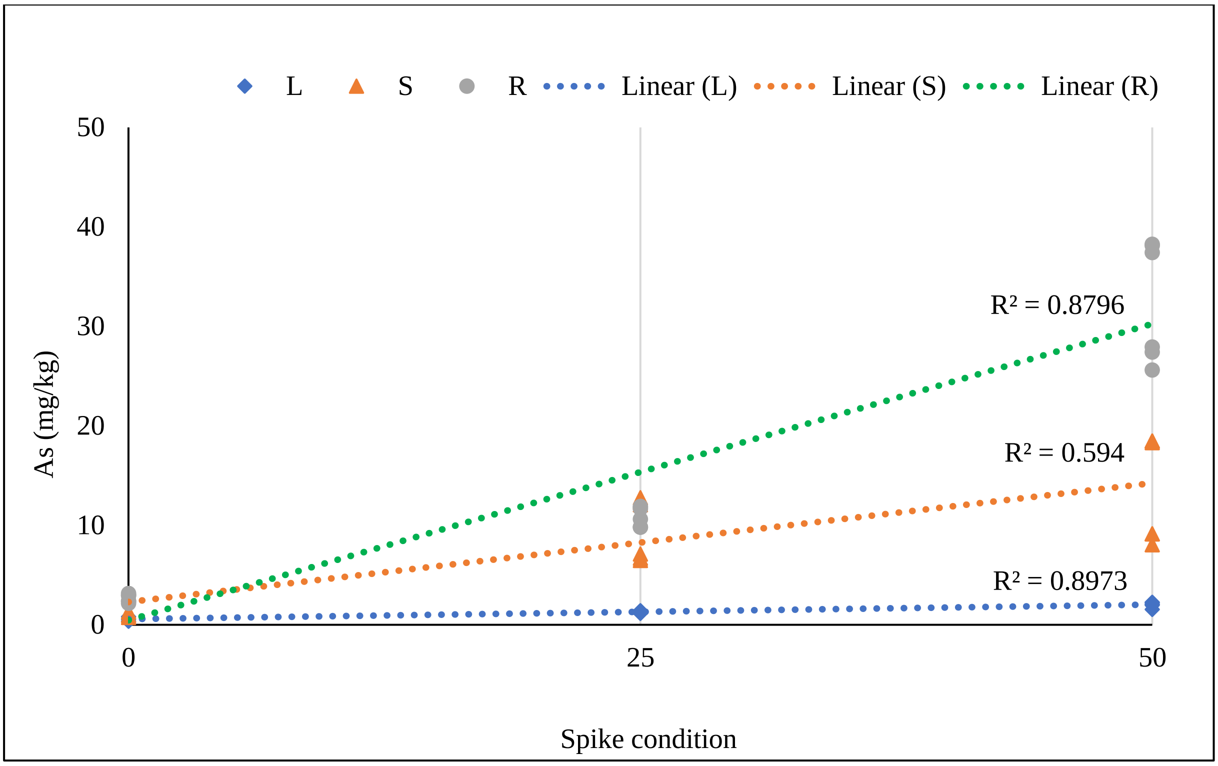


Figure 6-6 As uptake by *L. leucocephala* grown in control soil and spike_25 and spike_50, with EDTA and GE supplementation, L for leaves, S for stems and R for roots.

The comparatively high As content in roots compared with other parts of plants could be explained by lower translocation factors, the presence of iron and sulfur in the medium, As speciation in the soil, the phosphate uptake system (similarity of arsenate to phosphate and arsenite uptake through glycerol channel) (Shoji et al., 2008) and accumulation in vacuole cells. Arsenic uptake by plants is dictated by As bioavailability and plant physiology; therefore, action to increase As bioavailability or improve plant capability could speed up the remediation process.

6.4.2. Influence of Growth Enhancer on As Uptake by *L. leucocephala*

The role of a GE in plant growth in As-contaminated soil has been widely investigated and multiple mechanisms could explain the decrease in As uptake by *L. leucocephala* treated with GE composed of M*ycorrhiza sp., Bacillus sp*., yeast and enzymes. First, mycorrhizal fungi increase the resistance of the host plant by suppressing the high-affinity phosphate uptake system and therefore reducing arsenate uptake. A second explanation comes from the fact that mycorrhizal fungi may enhance As resistance in plants by effluxing As to the external medium. Zhao et al. (2009) reported that mycorrhizal fungi might reduce As translocation from roots to stems and Meharg & Hartley-Whitaker (2002) found that the presence of bacteria and yeast in GE could increase As resistance through enhancement of As efflux and reduction of the arsenate assimilation mechanism. Schneider et al. (2013) used three AMF isolates from uncontaminated soil (*Acaulospora morrowiae, Glomus clarum*, and *Gigaspora albida*) singly or in combination and three AMF isolates from As-contaminated areas (*Acaulospora morrowiae, Glomus clarum* and *Paraglomus occultum*) for *L. leucocephala* phytoremediation of As-contaminated soil and found a disparity in isolates' influence on uptake and growth.

When considering stems, no significant differences were found between settings, which confirmed the fact that *L. leucocephala* does not accumulate As in its stems. For leaves, GE influenced As uptake: in spike_50, As content in leaves was significantly higher than As content in leaves of *L. leucocephala* grown in spike_25 supplemented with GE, whilst no significant differences were found between spike_25 leaves and leaves of the control set.

6.4.3. Influence of EDTA on As Uptake

The EDTA influence on As uptake by *L. leucocephala* in mining soil from Burkina Faso was examined. Tukey HSD pairwise comparison revealed significant differences in stems, roots and to some extent in leaves of *L. leucocephala* grown in spike_25 and spike_50 compared to the unspiked control soil. In spike_50 supplemented with EDTA, As concentration in leaves increased by 350 % compared to unspiked control sample leaves. Other scenarios did not reveal significant differences. Based on these findings, it can be seen that As was translocated to leaves when As was in high concentration in the medium. Abbas & Abdelhafez (2013) concluded after investigating maize As uptake in EDTA supplemented As contaminated soil that in the presence of relatively lower concentrations of available As in soil, sequestering of As in roots took place to minimize its translocation from roots to shoots. EDTA supplementation in soil spike_25 did not significantly increase As uptake by *L. leucocephala* leaves and stem comparatively to the control soil. EDTA supplementation in spike_50 did not induce significant increase in leaves, stem and roots of *L. leucocephala* compared to same parts of *L. leucocephala* grown in EDTA supplemented spike_25 soil.

EDTA influence on As uptake by *Leucaena* could be induced by the capability of the chelating agent to solubilize iron-bound arsenic. In fact, arsenic has a high binding affinity with iron hydroxide precipitation. This affinity could be explained by the co-precipitation of iron (Fe(III)) with arsenic (As(III) and As(V)) (Rahman et al., 2008). Rahman et al. (2008) concluded that the addition of EDTA to the culture media increased the uptake of As(V) and As(III) into the plant tissue and about 4-6% of the inorganic arsenic species were desorbed or mobilized from iron plaque by EDTA. EDTA used in arsenopyrite bearing rocks can accelerate oxidative dissolution of the arsenopyrite leading to increase in availability of As in the environment (Wang et al., 2018) which could have increased readily available As and its toxicity due to the high concentration in the milieu.

Low uptake of As by *L. leucocephala* could have been induced by combined phytotoxicity of As and EDTA (2 mM) (Barbafieri et al., 2017) leading to some damage to transporters and cells. Additional use of NaOH, dithionite and H_3PO_4 in EDTA solution would have increased As availability for removal as reported by Wang et al. (2017). Kartal (2003) reached to the same conclusion when reporting use of EDTA (1%) for leaching of arsenic in chromated copper arsenate (CCA-C) treated wood.

6.4.4. Effect of Combined EDTA and Growth Enhancer on As Accumulation

The addition of EDTA and growth enhancer (GE) to spike_25 revealed a significant increase in As uptake by *Leucaena leucocephala* leaves and roots compared to spike_25 without either EDTA or GE. The same trend was observed with control soil. The combined action of EDTA and GE tended to reduce As uptake by roots and translocation to leaves in moderately contaminated soil. In highly contaminated soil (spike_50), the combined action of EDTA and GE significantly increased As translocation to leaves, while there was no significant decrease in As uptake by roots or translocation to the stem in spike_50 without EDTA or GE. Briefly, the combined action of EDTA and GE decreased As uptake by roots and translocation to the stem and leaves, except for leaves in spike_50.

Low As uptake and translocation could be explained by the probable increasing of resistance through enhancement of As efflux and reduction of the arsenate assimilation mechanism (Meharg & Hartley-Whitaker, 2002).

6.4.5. Growth Enhancer and EDTA Effect in Multi-elements Contaminated Soil

As content in leaves grown in soil watered with multi-elements revealed a significant increase in As level compared with leaves of plants grown in spike_25 and in spike_50, whether supplemented or not with EDTA or GE or both EDTA and GE. Low As uptake by plants roots grown in multi-element contaminated soil could be explained by plant physiology due to potential element toxicity in the plant and by preference in the uptake of elements. However, the As content of leaves was different except in the case of spike_50 supplemented with GE only.

6.4.6. *Leucaena leucocephala* As Uptake Factors

The BAC for all scenarios was below one, which implies that *L. leucocephala* in those conditions remains a plant that is tolerant to As and is not a hyper-accumulator taking up As and translocating it to the above-ground parts (Fitz and Wenzel, 2002). This result is in line with the finding of Schneider et al. (2013) who noticed poor translocation from roots to shoot in *L. leucocephala*. However, a ratio higher than 0.6 is considered acceptable for As phytoextraction (Barbafieri et al., 2017).

For BCF, in general, plants grown in spike_25 with the presence of GE showed increased BCF values. In spike_50, the BCF values were between 0.03 and 0.04. In addition, the multi-element spiking condition showed the a BCF value of 0.09 revealing that the presence of other elements could increase As transfer from soil to roots of *L. leucocephala*. The TF of *L. leucocephala* were below 1, making *L. leucocephala* better suited for phytostabilization than phytoextraction in these conditions and non-hyper-accumulator (Fayiga and Saha, 2016).

In high arsenic contained water, *L. leucocephala* was still showing competencies to uptake As.

6.4.7. Considerations for Field Test

Soil was spiked and kept for 45 days for balancing in order to let reactions take place between As and other compounds in the soil; during the experiment, the soil was watered with As prepared from arsenic trioxide. However, 45 days balancing time and use of an arsenic trioxide-based solution could deliver different As behavior than in normal soil gradually receiving As over a decade from mining activities. Fitz and Wenzel (2002) reported that As got involved in binding reactions with soil components over time. Actions to get As in a more available state for uptake by plants in the phytoremediation process are keys to the success of the operation. As revealed by the study undertaken by Kim et al., (2016), the addition of reducing agents, which includes sodium oxalate ($Na_2C_2O_4$), ascorbic acid ($C_6H_8O_6$) and sodium dithionite ($Na_2S_2O_4$), greatly enhanced the EDTA extraction of both As and potentially toxic elements from the contaminated soils due to the increased mobility of the potentially toxic elements under the reduced conditions. According to results found in this study, EDTA supplementation to soil, is not increasing significantly As uptake by *L. leucocephala* and an alternative should be searched like phosphate (K_2HPO_4) or ethylenediamine-N,N'-disuccinic acid (EDDS). Also, a GE isolated from contaminated soil could be more efficient (Jankong et al., 2007, Schneider et al., 2013). *Leucaena leucocephala* biomass use in methanisation process was investigated by Narayanaswami et Al. (1986) and was found to be an sustainable option.

6.5. Conclusion

This study revealed that the As uptake of *L. leucocephala* parts is correlated with the content of As in the soil (25 mg/kg spiked soil and 50 mg/kg spiked soil), with a significant difference between the control soil without supplementation and spiked soil without supplementation. Supplementation of EDTA does not significantly influence As uptake in the condition of this experiment. Also, supplementing with GE containing *Mycorrhiza sp*., *Bacillus sp*., does not significantly increase As uptake. The combined action of EDTA and GE in 50 mg/kg spiked soil does not increase uptake as expected. In 25 mg/kg spiked soil, no significant effect was noticed by the combined action in roots and leaves compared to the same level spiked control soil.

This study was based on a worse case scenario when contaminant concentration is high in soil and runoff water from tailings storage facilities and from waste dump. Anticipating in waste dumps and tailing storage facilities remediation, by for example avoiding oxidation of the arsenopyrite rocks which could lead to release of arsenic in the environment, could help in reducing the overall site contaminants. Arsenic average in water runoff will therefore still be in the acceptable range for *L. leucocephala* uptake.

In the condition of this study, with poor As translocation to leaves, *L. leucocephala* leaves could be used as cattle fodder. Transforming the biomass to charcoal could be considered as an extra option with an additional appropriate management of ash.

Chapter 7:

Phytoremediation of seepage from a gold mine tailing storage facility by means of a constructed wetland populated with spontaneously grown *Typha domingensis* and introduced *Chrysopogon zizanioides*

This chapter is based on the paper:
Compaore, W.F., Dumoulin, A., Rousseau, D.P.L. (in preparation). Phytoremediation of seepage from a gold mine tailing storage facility by means of a constructed wetland populated with spontaneously grown Typha domingensis and introduced Chrysopogon zizanioides.

ABSTRACT: This study analyzed the capability of two species to take up potentially toxic elements from gold mine tailing storage facility seepage in a constructed wetland with horizontal subsurface flow. Naturally populated *Typha domingensis* (cattail) and introduced *Chrisopogon zizanioides* (vetiver) were sampled on a regular basis in two experimentation cycles; *T. domingensis* only in the first cycle and *T. domingensis* plus *Chrisopogon* in the second. Corresponding water samples were taken from the inflow and outflow of the system. Potentially toxic elements were analyzed in both sets of samples. After 75 days' growth, *T. domingensis* exhibited an above-ground biomass production of 12.30 to 14.18 g per plant higher than *C. zizaniodes* which exhibited a biomass production of 6.65 g per plant. In the experimentation with the wetland populated by *T. domingensis* only, As concentration increased by 6% between the inlet and the outlet against a decrease of 37% in the experimentation populated by *T. domingensis* and *C. zizaniodes*. *T. domingensis* revealed an average bioaccumulation factor of 7, 5, 293, 1997, 413, 225 and 583 for As, Co, Cu, Mn, Ni, Pb and Zn, respectively, whilst *C. zizaniodes* exhibited bioaccumulation factors of 6, 2, 278, 503, 228 and 1184 for As, Co, Cu, Mn, Pb and Zn respectively. However, the translocation factor (TF), as the ratio of the concentration of an element in the aerial part divided by the concentration of the same element in the root, found in *C. zizaniodes* was higher compared to *T. domingensis*. *T. domingensis* translocation of Mn, Pb and Zn was 1 and 0, 58 and 0.8 Vetiver translocation of Zn is > 2. Due to the high biomass production, standing stock (amount of potentially toxic elements accumulated in aboveground biomass per area unit) of As, Co, Cr, Cu, Mn, Ni, Pb and Zn were 3, 7, 4, 7, 14, 7, 5, times higher in *T. domingensis* than in *C. zizaniodes*. Therefore, *T. domingensis* could be considered as a promising alternative for gold mine tailing seepage phytoremediation when in an adequate wetland design, whilst *C. zizaniodes* revealed very low growth rates and showed adaptation difficulties. Use of *T. domingensis* for gold mine tailing storage seepage element uptake is possible with uprooting giving high level of standing stock removal.

7.1. Introduction

Mining is a source of anthropogenic potentially toxic element contribution to environmental contamination in today's era of industrialization. Mine tailing storage facilities (TSF) are the main hotspots from which contaminants spread all around the mine area, threatening local soil and site water, as well as surrounding surface and underground water bodies. Attempts to use mining pit lakes as water reservoirs have been hindered by their possible contamination by nearby TSF. The TSF of a gold mine receives process residues, which are basically the ore mixed with lixiviation, washing, elution and melting chemicals. The TSF contains all the potentially toxic elements which go with the ore, including arsenic, iron, copper and nickel.

Site contamination from TSF comes from various sources. Transportation by wind occurs when the TSF is dry and the contaminant follows the wind pattern. Transportation by water occurs when the contaminant follows the water runoff and ends up in nearby surface or underground water bodies. Contaminant spread through water to the environment or water bodies can be stopped using methods such as physical, chemical or biological barriers. A phytoremediation method could be interesting in many ways, as appropriate plants are faster-growing and more cost-effective compared with other methods (Chandra & Yadav, 2010); they are also easy to implement and easy to maintain (Zhang et al., 2014b), they do not require high-tech equipment (Liu et al., 2008), and they are a particularly good fit for developing countries and a poor rural context. The contribution of wetlands to pollution control is presently a subject of interest, and wetland ecosystems are being studied worldwide with a focus on plants with bioaccumulation capability (Marchand et al., 2010; Soda et al., 2012; Vymazal & Brezinová, 2016).

Regarding the gold mine that is the subject of this study, a trench surrounding the TSF built for seepage control became spontaneously populated with monotype *Typha domingensis* species. In addition to *T. domingensis, C. zizanioides* was introduced for the purpose of this study. *T. domingensis* is described in section 2.4.3. Vetiver grass (*Chrisopogon zizanioides*) is used for all kinds of erosion control (Cordon, 1993), and is suitable under almost all conditions in almost all soil types and almost all climatic regions, except those with freezing conditions for long periods each year (Pratt et al., 1984). In addition, according to Pandey & Singh (2015), vetiver is one of the four most important aromatic grasses, with a capability to take up As and an economic value through the essential oil that can be extracted. Its use in constructed wetland for remediation of potentially toxic elements has been detailed in literature. Klomjek & Nitisoravut (2005) proved its tolerance to high salinity conditions. Vetiver's ability to treat acid mine drainage was reported by Kiiskila et al. (2019) in hydroponic conditions and also by Banerjee et al. (2019) for the phytoremediation of iron ore mine spoil dumps.

Studies of the two species' ability to remove potentially toxic elements have been widely reported. However, studies of their use to mitigate gold ore TSF seepage have not yet been carried out. The aim of this study was to assess the ability of *T. domingensis* and *C. zizanioides* to mitigate potentially toxic elements before seepage reaches the environment after the mining closure stage in which the TSF should be rehabilitated and erosion controlled. First, the ability of these two species to grow in this specific condition was assessed, based on their growth rate. Second, their potentially toxic element uptake capability was measured, using bioaccumulation factor, translocation factor and standing stock. A comparison was made between the two species and, finally, a sustainable harvesting period was estimated.

7.2. Materials and methods

7.2.1. Study site

The 22-hectare TSF was constructed to collect the tailings of carbon-in-leach gold processing. Annual evapotranspiration in the region has been estimated to 2827 mm (Etruscan, 2005). Average annual rainfall is 1,100 mm, with the rainy season running from May to October.

The experiments were carried out during the dry season due to practical reasons. Weather conditions during the experimentation were monitored with an Oregon ultra-precision weather station (Oregon, USA). The average air temperature during the experiment was 31.5 °C, the relative humidity was 52%, the average wind speed was 6 m/s, and the dominant direction was south-west.

7.2.2. Full-scale constructed wetland design

The experimental set-up consisted of a lined trench (HDPE 1.5 mm thick membrane) built around the TSF to contain the seepage and runoff. Only a portion of the lined trench was considered for the study, i.e. a section 50 meters long and 1.5 meters wide, containing 0.15 meters of soil with a light inclination allowing gravity flow of the seepage from the inlet to the outlet (Figure 7-1 and Figure 7-2). The system could be classified as horizontal flow constructed wetland as no water was visible at the surface.

The seepage flow rate was estimated daily during the first month of each experiment cycle and every three days thereafter. A volumetric flask and chronometer were used for the measurement. Three measurements were taken every time, and the average calculated. The seepage flow rate was controlled by installing a bypass pipe for the excess. This system maintained the flow rate at 2.98 ± 0.04 liters per minute, which equates to a hydraulic loading rate (HLR) of 5.72 cm/day.

The water balance describing the inflow and outflow was estimated based on the formula below, ignoring precipitation as the experiment was carried out during the dry season:

$\mathrm{SD} = \mathrm{PD} - ET * \mathrm{A}$ (Equation 8)

PD: the pipe discharge from the TSF (L/day); SD: discharge from the system (L/day); ET: evapotranspiration (mm/day = L/m²/day); A = wetland surface area (m²).

With the measured inflow rate of 2.98 L/min and an average daily ET of 7.74 mm/day, this yields an estimated outflow of 2.58 L/min. Using the average of SD and PD, and assuming a soil porosity of 0.35, the nominal hydraulic residence time (HRT) is estimated to be 1 day.

Figure 7-1 view of the lined-trench and the TSF wall, before setting up the experiementation,

Figure 7-2 View of the experimental plot and the inlet pipe, first experimentation setup with monotype species

7.2.3. Population of *Typha domingensis* and *Chrisopogon zizanioides*

Two cycles of experimentation were carried out, both during the dry season; from December 2017 to March 2018 and from October 2018 to January 2019. In the first experimentation cycle, *Typha* which spontaneously grew inside the 50 m trench as monotype species was considered. In the early stage of this experiment, the existing population of *Typha* was cut at the bottom edge of the shoot, allowing the plant to regenerate, and the population was adjusted to 30 stems per m². In the second experimentation cycle, *Typha* was removed from the final 25-meter length of the trench and replaced with *Chrisopogon* collected in the vicinity of the site, cut at the edge of the shoot and washed with running water.

7.2.4. Water Sampling and In-field Parameter Monitoring

Water was sampled on a two-weekly basis using a plastic bottle and conserved in a refrigerator at 4°C until preparation and analysis. Water was collected at the inlet and at the outlet during the first experimentation cycle, and at the inlet, the middle and the outlet during the second cycle of experimentation. Analysis of potentially toxic elements was carried out at Ghent University Campus Kortrijk. In-field water parameters were measured weekly using a hand-held Hanna instrument multiparameter HI9829 (Hanna Instrument Inc, Woonsocket, USA) properly calibrated with a set of solutions (Hanna instrument SRL, Romania) for the measurement of pH, dissolved oxygen (DO, mg/L), oxidation-reduction potential (ORP, mV), total dissolved solids (TDS, mg/L), conductivity (µS/cm), and resistivity (Ω*cm). The turbidity of the water was analyzed using a Hanna HI 93414 meter calibrated with a set of turbidity standards according to the manufacturer's instructions. The device used allowed measurement of ORP relative to an Ag/AgCl reference electrode. After readings, values of ORP were corrected to standard hydrogen electrode (SHE) according to the manufacturer advises (HI 9829 Instruction Manuel, Hanna Instrument):

SHE readings (corrected value) = value from the device (Ag/AgCl) + 205 mV (equation 9).

7.2.5. *Typha domingensis* and *Chrisopogon zizanioides* Sampling

Every two weeks, five samples of each species were collected. For each sample, ten random shoots were cut all over the wetland at the same level as they were the first time for the regeneration. For the first sample, the ten shoots were uprooted, and the roots were analyzed. Afterwards, only shoots were sampled for analysis as the objective was to remove potentially toxic elements through the upper part of the plants. After sampling, length and weight were measured, and the samples were washed with tap water followed by distilled water and then

10% nitric acid. Washed samples were dried at 105 °C until they reached a constant weight and recorded. Parts were labeled and kept in plastic bags in ambient conditions until analysis for potentially toxic elements. For comparison purposes, *T. domingensis* shoot samples were collected from an unimpacted location near the Bagré dam spillway (11°28'18.17"N, 0°32'27.00"W). These samples followed the same treatment as samples from the experiment.

7.2.6. Relative Growth Rate

Relative growth rate (RGR) was estimated based on the method described by Chandra and Yadav (2010). For *T. domingensis*, the dry mass at the two harvests was divided by the time span between the harvests. The start weight was considered as zero, as the stems had been cut before. For *C. zizanioides*, due to low growth, RGR was estimated by considering a time span of two months. The RGR is an indicator of the capacity of the species to survive in this seepage condition and thus of the suitability of the species for phytoremediation.

7.2.7. Water and plant Potentially Toxic Elements Analytical Methods

Potentially toxic elements dissolved in water were analyzed using USEPA method 200.7. Rev 2001 (USEPA, 2001). Briefly, the method consisted of filtration through a 0.45 µm filter, acidification to a pH below 2 (1.8±1) with commercial concentrated nitric acid (65%, 63.01 g/mol) and analysis using ICP-OES of Thermo Scientific. Plant samples were ground and thoroughly mixed, and 0.5 g was weighed in Teflon tubes for microwave-assisted acid digestion with a mix of hydrochloric acid and hydrogen peroxide in a (v/v) ratio of 4:2. The microwave was heated at 300 W for 5 minutes, followed by 600 W for 8 minutes and cooled down using the built-in venting system for 15 minutes. The digestates were filled to 50 mL with bidistilled water and analyzed using the ICP OES, iCAP 7200. The same parameters of operation of the ICP were maintained throughout. A Certipur® ICP standard XIII was used to calibrate the equipment. This standard was also used for spiking the samples for recovery calculations. The potentially toxic elements considered were As, Cd, Co, Cr, Cu, Mn, Ni, Pb, and Zn.

7.2.8. Soil Sample Potentially Toxic Element Analysis

Soil samples from the wetland were taken for potentially toxic element analysis at the end of the second cycle of experimentation. Approximately 3 kg of wet soil was collected in a plastic bag and sent to an external certified laboratory for analysis by ICP-MS (Method ME-MS61).

7.2.9. Bioaccumulation and Translocation Factors

Bioaccumulation factor (BF), which expresses the amount of element uptake by the plant, was calculated as the ratio of the element concentration in the plant divided by the element content in the water (Equation 5). The content of PTE considered in the calculation were that from the inlet. Within wetland samples were not available. The translocation factor (TF) was computed as the ratio of the concentration of the element in the aerial part divided by the concentration of the same element in the root (Equation 5).

7.2.10. Standing Stock

Standing stock was defined as the total accumulation of an element in a particular part of the plant (Brezinová & Vymazal, 2015) and was computed based on the biomass production per unit area and the concentration of the element. Results were expressed in mass per unit area g/m^2. The standing stock growth trend was used to estimate the optimum time to harvest for subsequent removal of potentially toxic elements.

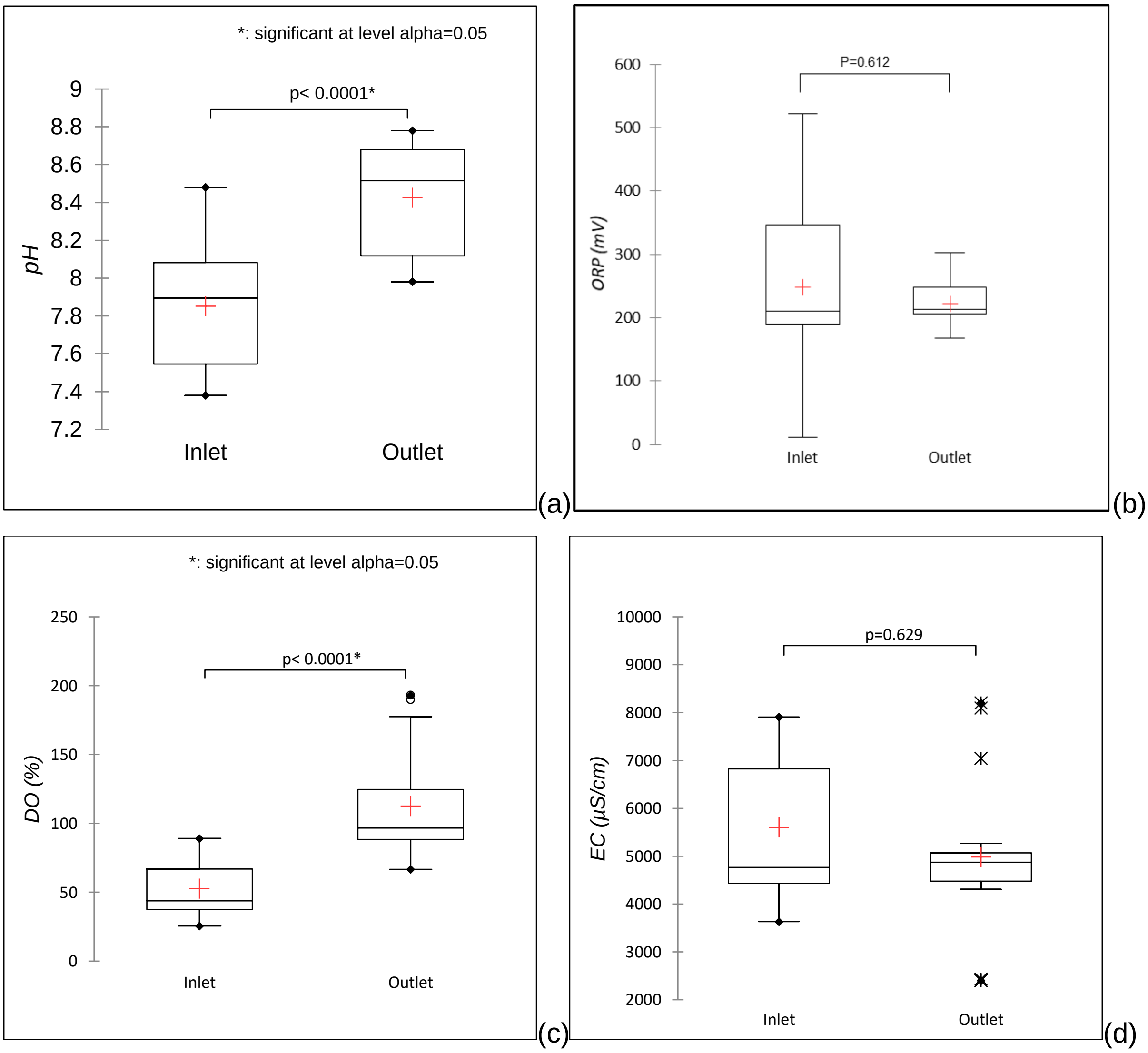
7.2.11. Quality Control and Statistical Processing

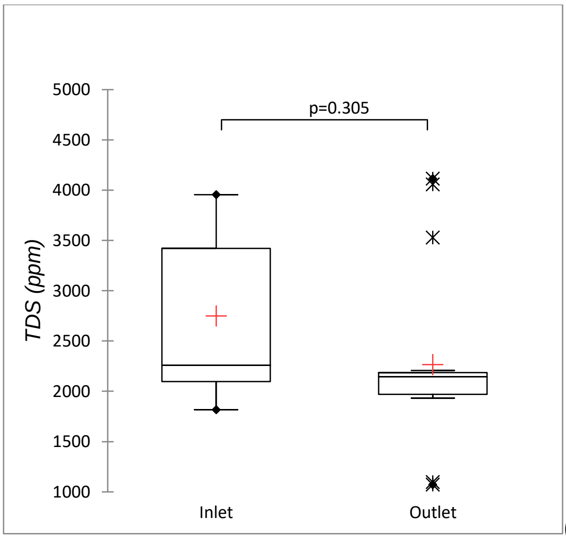
During potentially toxic elements analysis, a number of steps were taken to guarantee the reliability of the measurement. A field blank, procedural blank, and spiked samples were used. Within each digestion batch, a procedural blank was included consisting of all the reagents except the samples. During laboratory analysis, metal-based materials were avoided. A field reagent blank consisting of distilled water was used in each sampling campaign and went through all the steps in-field, from container rinsing to transmission to the laboratory for analysis. The method detection limit (MDL) was determined as in 3.2.3 and statistical analysis as in 3.2.5.

7.3. Results

7.3.1. Field Parameters

The results of the field monitoring of water quality are shown in Figure 7-3. Significant differences between inflow and outflow were observed for pH, DO and T°; indicating some form of activity within the wetland and evapotranspiration. The ORP didn't significantly decrease from the inlet to the outflow as did not the conductivity (EC). The same trend was observed with TDS. Dissolved oxygen increased significantly when comparing inflow and outflow. The temperature of the seepage at the inflow was significantly higher than at the outflow. Values of turbidity were 1.57±0.69 NTU for the inlet and 2.64±0.55 NTU for the outlet.





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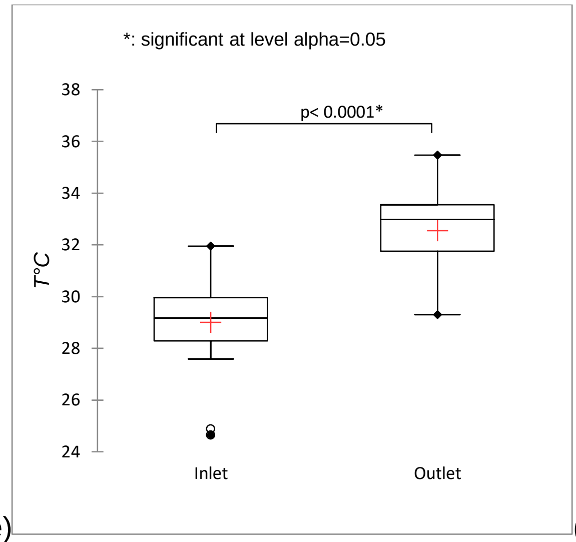

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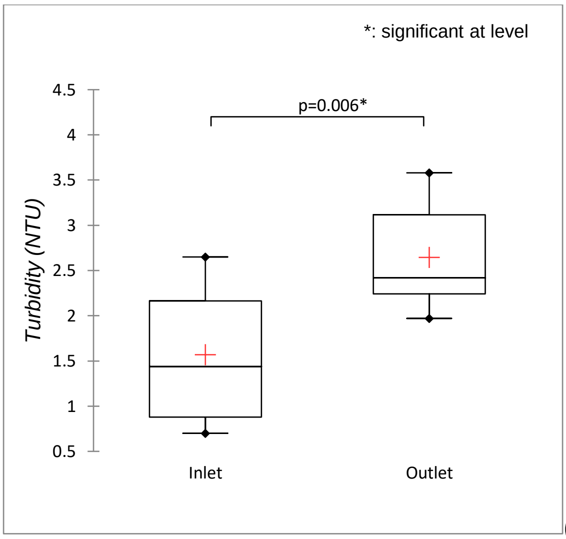


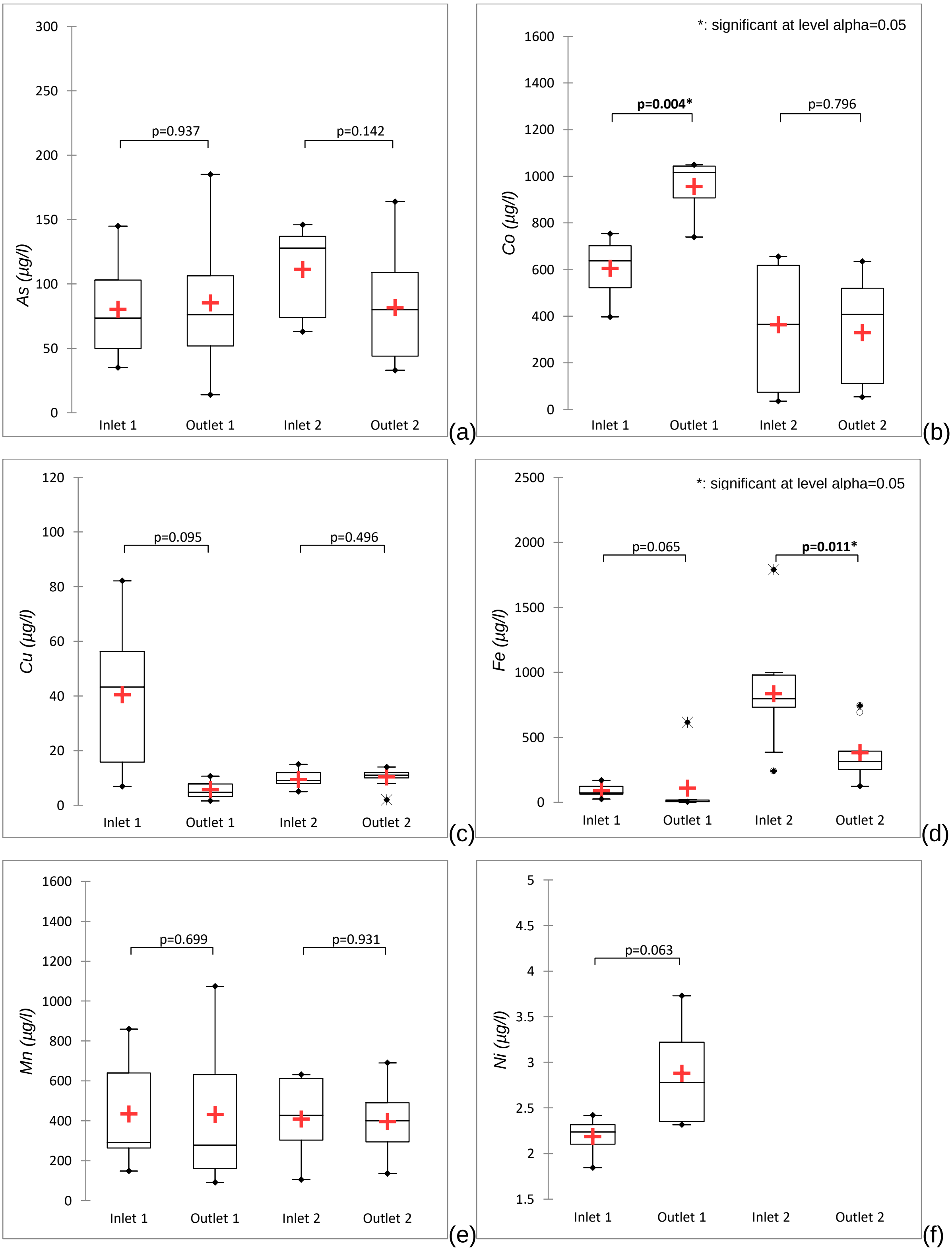
(g)

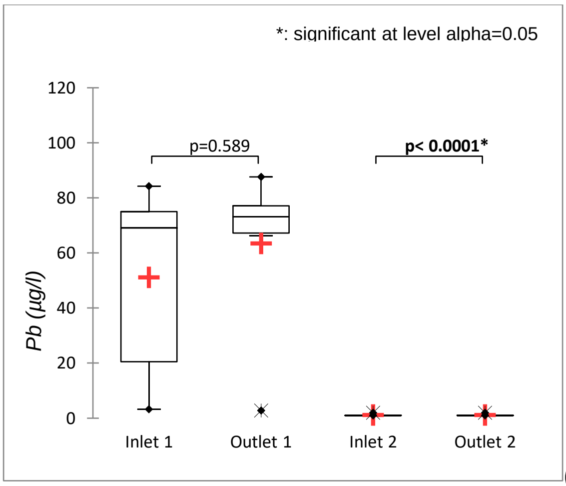
Figure 7-3 In-field parameters of the seepage. Dissolved oxygen (DO, %), oxidation-reduction potential (ORP, mV), total dissolved solids (TDS, mg/L) and conductivity (EC, µS/cm), Temperature (T, °C), Turbidity (NTU, NTU) and Mann-Whitney's statistics.

7.3.2. Potentially Toxic Elements in Seepage

Analysis of potentially toxic elements revealed that the seepage from the tailing storage facility had a high content of potentially toxic elements. An overview of the results is given in Figure 7-4.

Differences were noticed between the two experimentations inlet values which in turn influence outlet values. Inlet Fe significantly increased, while Co, Pb and Zn significantly decreased from the first experimentation with only Typha to the second experimentation with Typha and vetiver. In the experimentation with only *T. domingensis*, inlet and outlet water potentially toxic elements analysis did not reveal significant differences for As, Cu, Fe, Mn, Ni and Pb, though there was a significant increase for Co and a significant decrease for Zn. When setting up the experimentation with both *T. domingensis and C. zizaniodes*, Pb and Fe contents decreased significantly between the inlet and outlet. As, Co, Cu, Mn, and Zn analysis did not reveal any significant differences.





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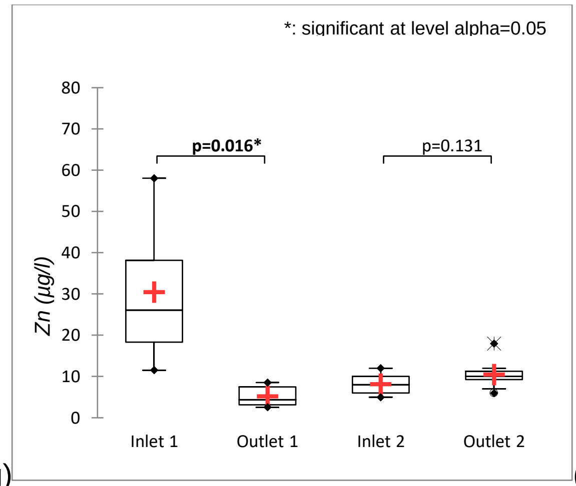


(h)

Figure 7-4 Potentially toxic elements in the inflow and outflow of the constructed wetland during the first experimentation populated with only Typha (n=6) and second experimentation populated with Typha and vetiver (n=9). Cd and Cr were below the MDL. Ni in the second cycle was below the MDL. Statistics based on Mann-Whitney tests.

Average potentially toxic element contents in seepage were high due to characteristics of the tailings which were defined by the type of rock being processed.

Tailings from the process plant were not authorized to be discharged into the environment nor the seepage (Etruscan, 2005). In the rehabilitation stage, when mining will not run to recycle the tailing water, precaution should be taken to avoid runoff to the environment.

7.3.3. Biomass Production of *Typha domingensis* and *Chrisopogon zizanioides*

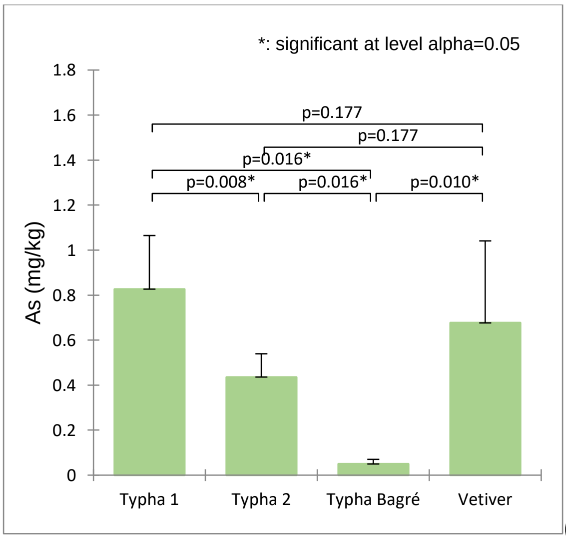
Biomass production could be an indicator of the capacity of the species to survive in the TSF seepage milieu and could indicate the suitability of the species for phytoremediation. Biomass production of *T. domingensis* during the first experiment was estimated to be 14.18 g dry weight per stem in 75 days' growth, reaching a height of 1.61 ± 0.22 m, with 9.30 ± 1.95 leaves per stem and a fresh weight of 145.34 ± 62.42 g. During the second experiment, the biomass production of *T. domingensis* was estimated to 12.30 g dry weight per stem over a growth period of 75 days with a fresh weight of 122.50±11.43 g.

C. zizanioides in two months' growing time (75 days) reached 0.65 ± 0.11 m height, a biomass of 6.65 ± 3.99 g (fresh weight) and 2.50 ± 1.39 g (dry weight) per stem.

7.3.4. Potentially Toxic Elements in *Typha domingensis* and *Chrisopogon zizanioides*

Plant potentially toxic element contents in the above-ground parts are represented in Figure 7-5. Results show that above-ground parts of both *T. domingensis* and *C. zizanioides* accumulate potentially toxic elements.

Potentially toxic elements in the above-ground parts of *T. domingensis* from the first experimentation which was populated by *T. domingensis* followed this trend: Mn > Zn > Pb > Cu > Co > Ni > As > Cr. Potentially toxic elements in roots from this first experimentation showed this trend: Mn > Co > As > Cr > Zn > Pb > Ni > Cu. *T. domingensis* above-ground parts from the second experimentation where the wetland was populated with *T. domingensis* and *C. zizaniodes* showed potentially toxic elements results with this trend: Mn > Zn > Cu > Co > As > Cr > Ni > Pb. Cu and Zn concentrations in the above-ground parts of *T. domingensis* during the first experimentation were significantly different from Cu and Zn concentration in the above-ground parts in the second experimentation, whilst Cu content in the inflow was not significantly different between the first and second experimentation. Comparatively, Bagré dam aboveground part results were lower compare to *Typha* from the first and the second cycles.



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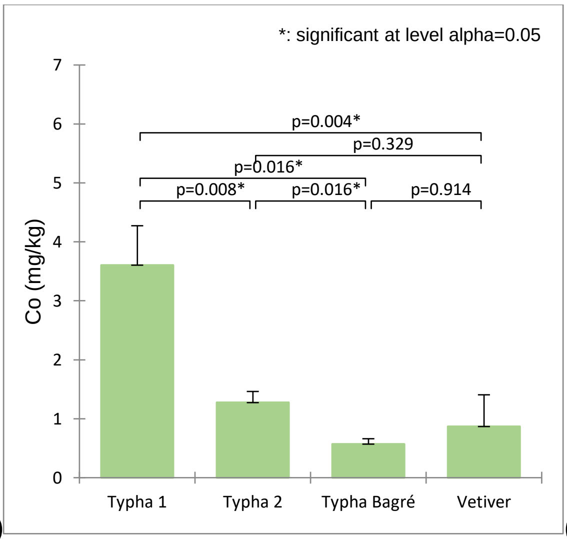

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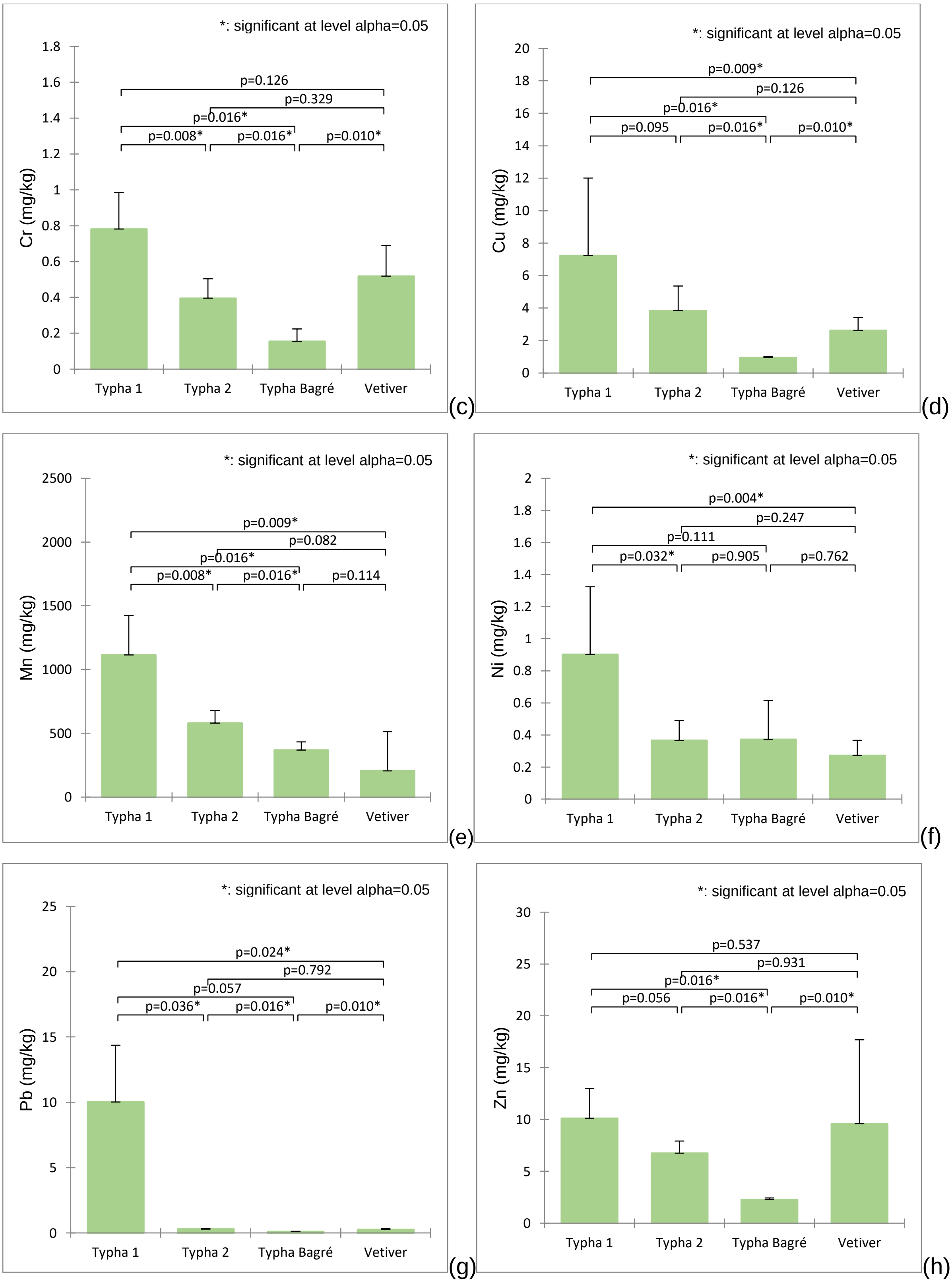


Figure 7-5 Aboveground potentially toxic elements content (mg/kg) in *Typha* and *Chrisopogon* from both cycles and *Typha* from Bagré. Statistic based on Kruskal test.

As, Cd, Co, Cr, Mn, Ni, Pb and Zn content in *Typha* roots were 69.47±19.10, 0.31±0.08, 123.0±30.5, 16.20±3.96, 793.92±282.53, 6.74±2.36, 8.94±3.35 and 11.02±6.70 mg/kg, respectively.

7.3.5. Bioaccumulation and Translocation Factors

The bioaccumulation factor of *T. domingensis* and *C. zizanioides* parts were computed against the seepage inlet potentially toxic elements content and are shown in Figure 7-6. When comparing the two cycles of experimentation, As BF decreased from 10 to 4, Co went from 6 to 3 and Mn dropped from 2,574 to 1,420 while Pb increased from 196 to 257 and Zn from 333 to 834. C. zizanioides exhibited a BF of 6; 2; 278; 503; 228 and 1,184 for As, Co, Cu, Mn, Pb, and Zn respectively. Compared to the second cycle data of *T. domingensis* in the same condition, As, Cr and Zn BF were higher. Co, Cu, Mn, Pb exhibited BF values lower than *T. domingensis*. The BF computed, based on above-ground parts, revealed that BF was dependent on the growth period. The highest BF was found for As at the end of the second month of growth, as it was for Ni. The highest BF for Pb was detected in the early stage of growth, as it was for K, Cr and Co. Zn, Mn and Cu accumulation factors reached their highest values after 30 days of growth.

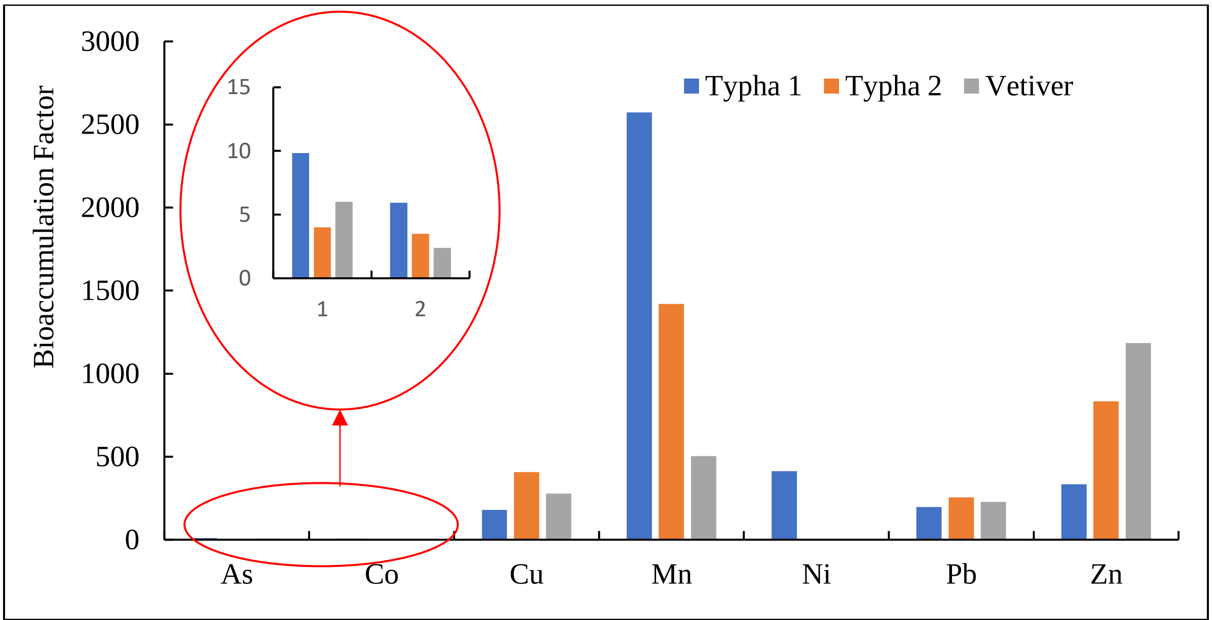


Figure 7-6 Bioaccumulation factors of *T. domingensis* in the first and second experimentation cycle and *C. zizanioides*.

The TF of *T. domingensis* and *C. zizanioides* was calculated based on elements in the above-ground parts and roots and revealed that most elements remained in the roots, although this is dependent on the element (Figure 7-7). Arsenic TF was very low, meaning that the greater part of As uptake by the plant remains in the roots. In the first cycle of the experiment, roots of *T. domingensis* accumulated 84 times the As content of the above-ground parts, 34 times for cobalt, 21 times for Cr, 7 times for nickel and as much as the above-ground parts for Mn, Pb, and Zn. In the second cycle, the TF increased except for Mn. Root As concentration was 158 times the above-ground parts, 97 times for Co, 41 times for Cr, 18 times for Ni, 31 for Pb and 2 for Zn.

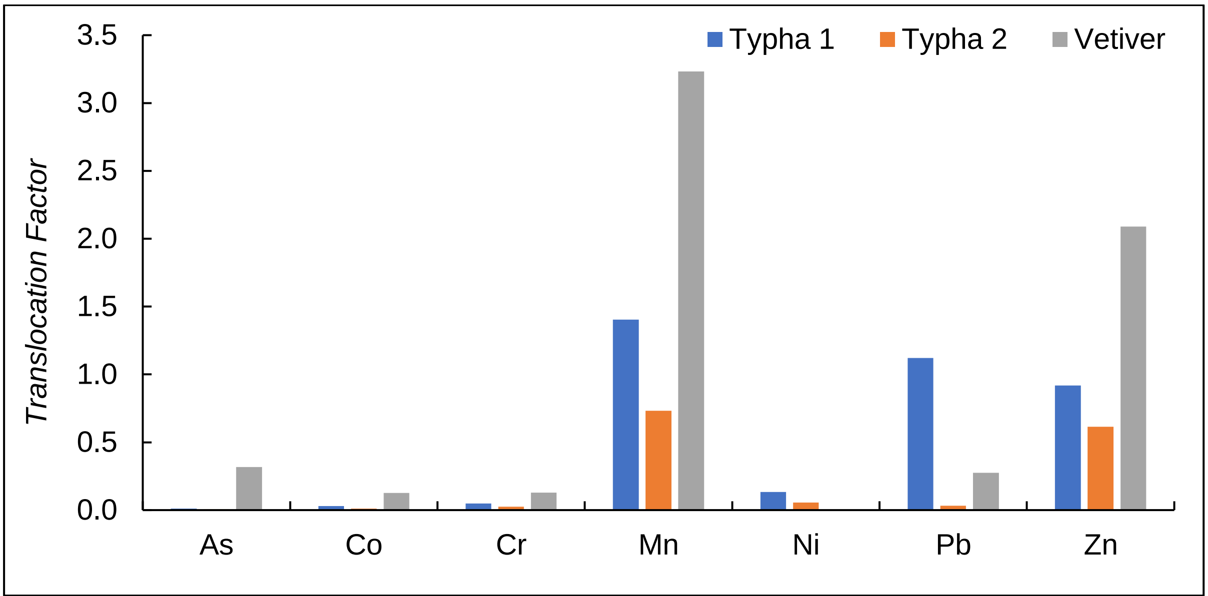


Figure 7-7 Translocation factor representation of *T. domingensis* in the first and second experimentation cycle and *C. zizanioides*.

7.3.6. Soil potentially Toxic Element Content

Results on the potentially toxic element content of wetland soils are recorded in Table 7-1.

Table 7-1 Wetland soil samples, results from total digestion analysis results in mg/kg dry soil, expressed per dry weight of soil.

Element	As	Ca	Cd	Co	Cr	Cu	Fe	K
MDL	0.2	0.1	0.02	0.1	1	0.2	0.1	0.1
Soil sample	52.4	2.1	0.02	55.3	350	31.3	84.4	6.9
Element	**Mg**	**Mn**	**Na**	**Ni**	**P**	**Pb**	**S**	**Zn**
MDL	0.1	5	0.1	0.2	10	0.5	0.1	2
Soil sample	2.6	1440	4.2	94.7	270	24.2	0.8	32

7.3.7. Standing Stock of *Typha domingensis* and *Chrisopogon zizanioides*

Standing stocks of the aerial parts of *T. domingensis* at each stage are shown in Table 7-2. The As level in standing stock of *T. domingensis* during the first cycle of the experiment was 353 mg/m^2, Co was 1,531, Cr was 332, Cu was 3,080, Mn was 474,398, Ni was 383, Pb was 4,263 and Zn was 4,309. In the second cycle, the values were 2, 3, 2, 2, 2, 3, 40 and 2 times less for As, Co, Cr, Cu, Mn, Ni, Pb, and Zn, respectively. *C. zizanioides* stand stock were 3, 7, 4, 7, 14, 7, 5, and 3 times lower compared with *T. domingensis* grown in the same experimentation set-up.

Table 7-2 Standing Stock of above-ground part of *T. domingensis* and *C. zizanioides* (mg/m^2) after a 10 weeks growth period,

	As	Cd	Co	Cr	Cu	Mn	Ni	Pb	Zn
T. domingensis 1	353	30	1531	332	3080	474	383	4263	4309
T. domingensis 2	162		469	148	1417	214	137	107	2494
C. zizanioides	51		65	39	197	15	20	20	720

7.4. Discussion

7.4.1. Inflow and Outflow Physicochemical Field Parameters

The study expressed an increase in DO between the inlet and the outlet. Significant increase of DO from the inlet to the outlet is due to the thickness of the experimentation system of only 0.15 m allowing diffusion of air. The height of the tailing in the storage facility is increasing leading to an increase of hydrostatic pressure and the saturation of gas and therefore reducing the DO at the inlet. It is thus expected that DO will fluctuation over time. The measured ORP expressed an moderately oxydizing context with ORP value at the inlet similar to the ORP value at the outlet (248±128 mV and 222±40 mV). When considering each of the two experimentation setups, a significant decrease in ORP was noticed (from 352±70 to 212±55 mV). During the second experiment using the same milieu and vegetated with polytype species, the ORP increased from a moderate reducing context (144±75 mV) to moderate oxidizing context (228±27 mV) (Aigberua et al., 2018). ORP fluctuation over time could also be linked to ore type being processed at the process plant, and decrease in the second experimentation could be linked to change in the milieu especially porosity. ORP change will dictate As and Cr speciation and their uptake profile by both species. In general, the seepage showed a slightly alkaline condition which could be induced by the use of lime to control the pH of the ore lixiviation to avoid formation of cyanide gas during processing. The pH of the outflow of both cycles exceeded the optimal range for *Typha*, which is estimated to be from 4.0 to 8.1 (Pratt et al., 1984) and also vetiver optimal pH ranging from 4 to 7.5 (USDA, 2009). The conductivity of the seepage coming from the tailing storage facility was very high (> 4600 µS/Cm), indicating a high content of ions. Fluctuation of EC could be due to the evapotranspiration and solubilization of elements induced by other physicochemical parameters. Low growth rate of *C. zizanioides* could have lead to variation of evapotranspiration factors as low leaf surface and absence of shadow in the *C. zizanioides* part. TDS of the inflow was significantly lower than that of the outflow in the first cycle, whereas in the second cycle it was significantly higher. The same trend was observed for conductivity. For TDS, adsorption by soil could explain this decrease in the first cycle. In the second cycle, saturation of the soil medium could explain this increase. The temperature was above 27°C during the experiments and was thus conducive for biological processes. Variation of temperature between the first experimentation vegetated with monoculture and the second experimentation vegetated with polyculture was not significantly different. The turbidity of the inlet was significantly low compare to the outlet. The seepage got charged between the inlet and the outlet.

7.4.2. Potentially Toxic Elements of Inflow and Outflow

As discussed before, variations in ore composition, tailing quantities and pouring location, were affecting the influent contents in terms of potentially toxic elements. Inlet and outlet water analysis revealed significant changes in most of the PTE. Briefly, some potentially toxic elements (Fe and Zn from the first cycle) significantly decreased from the inlet to the outlet while some potentially toxic elements (Co and Pb from the first cycle) increased (Table 7-3). Pb solubility was found to increase with decreasing redox potential, confirmed with high Pb uptake by plant.

Different variation in potentially toxic elements content between the inlet and the outlet could be linked to the concentration effect of evapotranspiration expressed by EC variability, which decreased by 29% between the inlet and outlet in the first experimentation cycle and increased by 12% between the inlet and outlet in the second experimentation cycle. Further, decrease in water potentially toxic elements content could be due to phytoremediation by the species populating the wetland and to some extend to adsorption effect. Equally, a higher decreasing ratio in the first than in the second cycle could be explained by the overall physicochemical state of the seepage (DO, TDS, T°), low pH or oxidative state, which influence water potentially toxic elements uptake by *Typha Chrisopogon* and potential adsorption and settling mechanism occurring in the wetland (Carling et al. 2011).

Table 7-3 : Inlet and outlet variation according to experimentation set up, expressed in µg/L,

	As	Co	Cu	Fe	Mn	Ni	Pb	Zn
Experimentation with only T. domingensis								
Inlet	80.3±41.5	606±137	40.4±29.5	90.2±54.1	433±295	2.18±0.25	51.2±36.7	30.4±20.16
Outlet	85.3±59.4	957±124	5.69±4.60	109±248	432±390	2.88±0.60	63.4±30.6	5.22±2.66
Variation	6%	37%	-611%	17%	0%	24%	19%	-483%
Experimentation with only T. domingensis and C. zizaniodes								
Inlet	111.4±33.0	364±270	9.44±3.40	835±437	409±189	<MDL	1.14±0.38	8.11±2.42
Outlet	81.6±44.0	330±230	10.3±3.57	381±207	396±180	<MDL	1.13±0.35	10.5±3.63
Variation	-37%	-10%	9%	-119%	-3%		-2%	23%
Average from the two experimentations								
Inlet	99.0±38.5	461±252	21.9±23.8	537±503	419±227	2.18±0.25	24.2±35.2	15.0±14.9
Outlet	83.0±48.7	581±370	9.17±4.19	273±256	410±271	2.88±0.60	27.8±37.2	8.47±4.14
Variation	16%	-26%	58%	49%	2%	-32%	-15%	43%

However, fluctuation in outlet water potentially toxic elements values over time was explained by the fluctuation in the inlet values, the physicochemical parameters driving water potentially toxic elements uptake and uptake capabilities (UN-HABITAT, 2008) and influence of *C. zizanioides* absent at the first cycle of experiment. Adsorption of investigated potentially toxic element is influenced by the overall state of the wetland and the physicochemical parameters driving this mechanism in the wetland. It involves several physico-chemical mechanisms as absorption, ion exchange, surface complexation and precipitation (Fawzy et al., 2016), flocculation, sedimentation, reduction and oxidation (Batool & Saleh, 2020) plant uptake and microbial oxidation and reduction. Removal of potentially toxic elements in a constructed wetland includes plant uptake, abiotic and biotic reactions such as microorganism actions, adsorption and precipitations. Variation of ORP can lead to variation of the As species and the solubility in combination with other elements . As is lightly increasing in the first cycle when the ORP is decreasing from inlet to outlet. Arsenic uptake in the first experimentation vegetated with a monotype species was high compared to the second experimentation vegetated with a polytype species, this decrease in As uptake by *T. domingensis* present in both experimentation could have been induced by precipitation of As due to high pH compare to the first set up with lower pH. This could also be explained by the lowering of ORP conditions which indicate a decrease in the oxidizing state of the wetland, which could have led to change in As speciation and therefore to it uptake profile (Akter et al., 2005). The overall moderately oxydizing context of the wetland (248±128 mV inlet and 222±40 mV outlet) and the pH of 8 were a condition for As (V) predominance. However, when considering experimentation cycle, the wetland expressed an oxidizing context in the first which As (V) would predominante. At the second set up, with an ORP at the inlet of 143±74 mV and a pH of 8, As (III) could have prevailed (Yazdi & Darban, 2010). Use of calcium hydroxide, in the gold processing for carbon in leach pH control, could have influenced As in the wetland because both As (V) and As (III) behave as chelates and precipitates with Calcium given the most stable As(V) species in oxidized alkaline context. Ferric hydroxide [$Fe(OH)_3$] plays an important role in controlling the As concentration in soils. Both As(V) and As (III) are adsorbed onto the surface of $Fe(OH)_3$, but the adsorption of As (V) is much higher than As (III) (Akter et al., 2005). The presence of iron, aluminium, and calcium compounds is the most important factor in controlling the fixation in soil. ORP fluctuation could have dictated Cr speciation and it uptake profile. As and Cr are directly sensitive to redox changes (Aigberua et al., 2018).

7.4.3. Potentially Toxic Element in *Typha domingensis* and *Chrisopogon zizanioides*

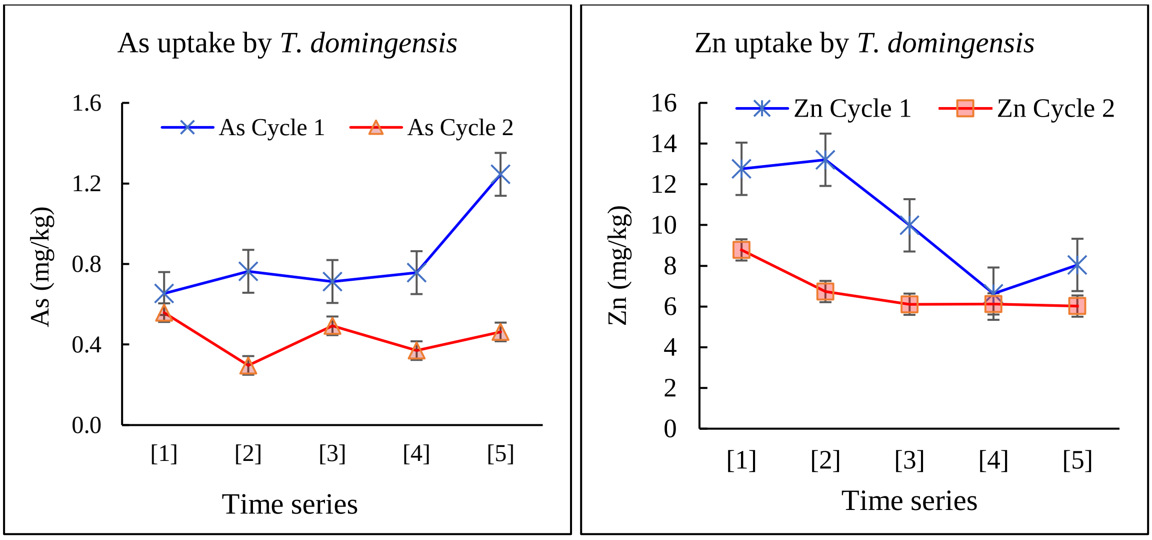
The study revealed that in the same condition of experimentation, the two species were performing similarly. The results for introduced *C. zizanioides* clearly confirmed its ability to take up some potentially toxic elements from mining tailings (Chiu et al., 2006). *C. zizanioides* potentially toxic element concentrations followed this trend: Mn > Zn > Cu > Co > As> Cr > Ni > Pb. Compared to the results for *T. domingensis* from the second cycle, *C. zizanioides* potentially toxic elements concentration according to the Mann-Whitney test was not significantly different to *T. domingensis* for all potentially toxic elements considered except for Mn. For instance, vetiver As uptake was 0.68±0.36 mg/kg against 0.44±0.10 mg/kg for Typha. This performance could be explained by wetland conditions favoring the uptake like pH, ORP, EC and DO affecting speciation of element and particularly the plant physiological state. ORP condition of the wetland, influencing As speciation, dictated arsenic uptake by both species.

Analysis of *T. domingensis* above-ground potentially toxic elements according to growth period revealed fluctuations of potentially toxic elements content (Figure 7-8), which is consistent with many studies (Kossoff et al., 2014; Schoenberger, 2016; Ceniceros-Gómez et al., 2018). For instance, in the first experimentation cycle, As concentration in *T. domingensis* aboveground increased over the first weeks to double by the end of the eighth week. In the second cycle, concentration of As in aboveground part of *T. domingensis* was almost constant. Sensitivity of As to ORP, could explain the As uptake variation in Typha in the first and second cycle. With high ORP in the first cycle, predominance of arsenate could have been easily uptaken through the phosphate uptake pathway than in the second cycle where the ORP was low. Also difference in toxicity between As (III) and As (V) could explain difference in uptake due to toxicity to plant (Aigberua et al., 2018). The high concentrations of Mn in the two cycles were not beneficial to arsenic uptake. In addition, the presence of manganese dioxide in the binary oxides can influence As(III) sorption in two aspects. On the one hand, manganese dioxide may effectively oxidize As(III) to As(V). In addition, during As(III) oxidation, the reductive dissolution of MnO_2 could result in the production of fresh adsorption sites at the solid surface, which will favor the adsorption of formed As(V) (Zhang et al., 2012).

Fawzy et al. (2016) investigated Ni(II) uptake by *T. domingensis* in laboratory scale and found that the removal rate of Ni(II) declined when the pH levels were higher than 6.0 as it is in this seepage. Eid et al. (2012) investigated Ni, Co and Ag concentration by *T. domingenis* and found no significant difference in concentration over a period of eight months.

Cr concentration by *T. domingensis* in both experimentation cycles did not exceed 1 mg/kg, which is considered as the threshold of toxicity for *T. domingensis* species. In contrast, Bonanno & Girelli (2017) found for *Typha* species concentration values higher than 1 mg/kg with BF varying between 0.15 and 0.19 and TF varying between 0.21 and 0.29 according to season (spring or autumn).

All in all, fluctuation of potentially toxic element concentrations over time in *T. domingensis* and *C. zizanioides* could be linked to the changing physiological state of the species and the variation of inlet potentially toxic element concentrations, as well as by the influence of fluctuating field parameters (DO, EC, pH, salinity etc.) and media behavior, and the effect of weather conditions (sun, temperature) on wetland efficiency (Zhang et al., 2014b). Differences in results between the first and the second cycle of Typha could also be explained by the reaction of *Typha* to harvesting and to media change (mainly saturation effect and porosity filling).



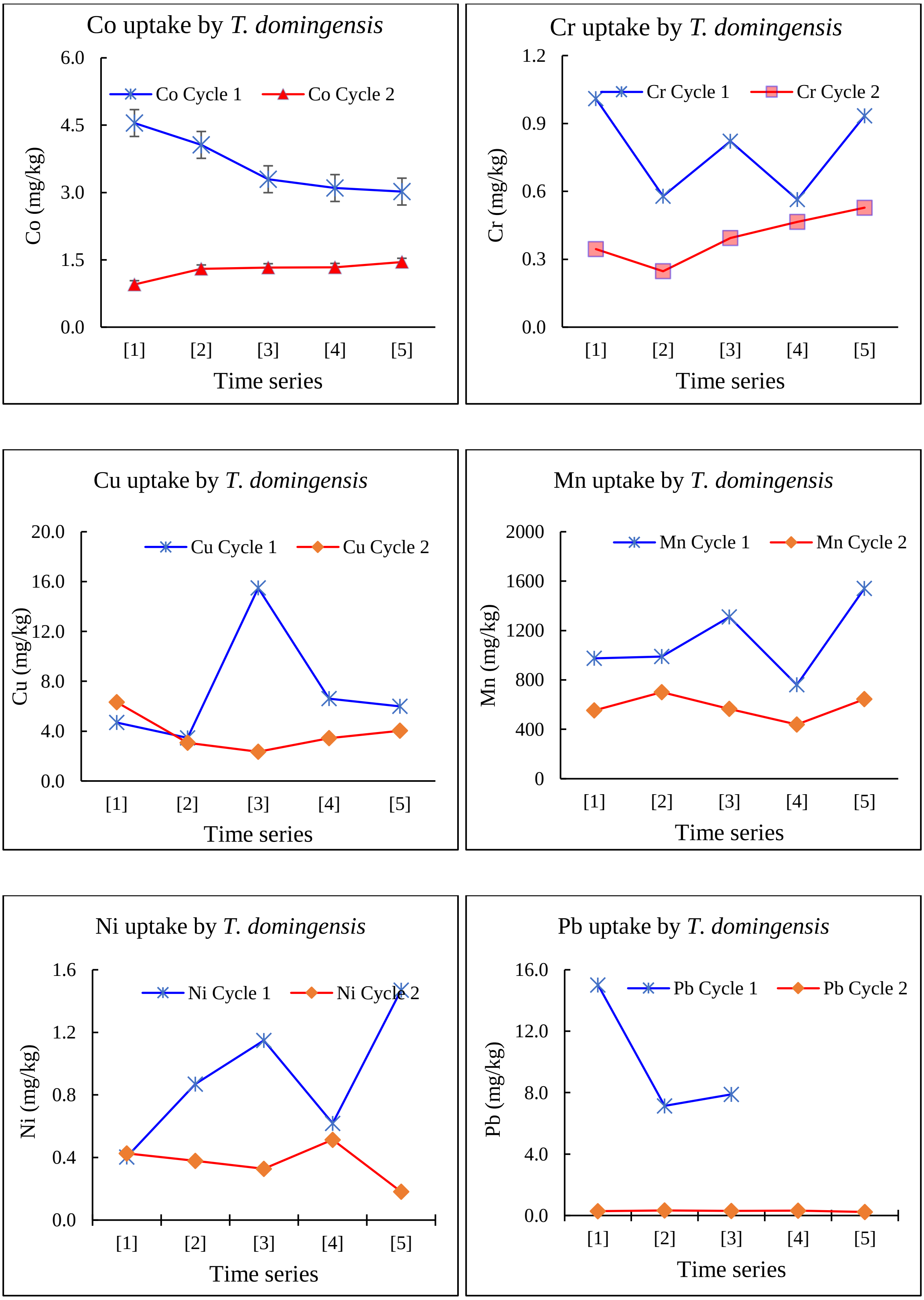


Figure 7-8 Potentially toxic elements concentration profile over time during the first and second cycle of experimentation. Times series expressing 14 days intervals, Standard error (vertical bars).

7.4.4. Bioaccumulation and Translocation Factors

Bioaccumulation of potentially toxic elements in macrophytes depends on plant uptake capability as well as the transportation mechanism within the plant and also includes the chemical state of the target element. Plant detoxification capacity also plays a key role in plant accumulation of potentially toxic elements (Vymazal & Brezinová, 2016).

In both natural and constructed wetlands, research reveals complex interactions leading to the removal of pollutants (Vymazal, 2013). Badejo et al. (2017) reported that the wetland processes that contribute to the removal or reduction of pollutants in constructed wetlands are: microbial-mediated processes (removal as a result of the activity of bacteria or other microorganisms); chemical networks (creating products that are themselves contaminants of interest); sorption (either linear, Freundlich or Langmuir); sedimentation (mostly involving suspended solids); plant uptake (in which plants take up nutrients and trace chemicals to sustain their metabolism and for storage); transpiration flux; vertical diffusion in soils and sediments; seasonal cycle, and accretions (creation of new soils and sediments). The wetland media analysis after the two experimentation cycles revealed an accumulation in the media.

The lower accumulation of potentially toxic elements in above-ground plant tissues protects the aerial parts and especially the photosynthetic apparatus from the phytotoxic effects of some potentially toxic elements (Salem et al., 2014). In industrial effluent in Nigeria, Mukhtar & Abdullahi (2017) found for *T. domingensis* a BF of 21.7, 13.3, 8.78, 16.73 and 33.85 for Pb, Cu, Zn, Cr and Fe respectively and a TF of 0.96, 2.00, 1.15, 2.01 and 1.67. The TF of Co and Ni for *T. domingensis* growth in Burulus Lake in Egypt were below one (Eid et al., 2012). These values could be linked to the effluent difference between the Burulus Lake and the gold mine tailing storage facility seepage. *C. zizanioides* is known as a hyperaccumulator of Pb, which explains a TF close to 1 (Pidatala et al., 2018).

T. domingensis and *C. zizanioides* exhibited a preference in the uptake of element and locations. As had a BF of less than 1 and a TF of 0.01 reflecting the preference of the plant to keep As in the root system rather than in the aerial parts. Rahman et al. (2011) revealed that in a constructed wetland, 44 to 49% of the total inflowing As was recovered or concentrated within the plant roots (*Juncus Effuses*). The same trend was observed in this study for Cr, Co, Fe and Ni. On the other hand, Pb exhibits a BF and a translocation factor higher than one. K, Mg, Mn and Zn followed the same trend as Pb, which leads to the same conclusion as Chiu et al. (2006) who found Zn uptake by *C. zizanioides* and translocation within the plant. All BF values were greater than one, showing that the plant accumulates potentially toxic elements in the organism. Therefore, based on the mitigation of the targeted element and according to its translocation factors, aerial-only use or total uprooting of the plant should be considered.

The findings of this study revealed that *T. domingensis* has a poor translocation capability (<1) except for Mn, expressing a weak translocation of the element from roots to the above-ground parts. Therefore *T. domingensis* is more suited for phytostabilization (phytosequestration) than phytoextraction, as confirmed by Bonanno & Cirelli (2017) for Al, As, Cd, Cr, Cu, Hg, Mn, Ni, Pb and Zn. The basic mechanism underlying phytostabilization is the complexation of potentially toxic element ions with the root exudates/mucilage or with the cell walls and also binding with potentially toxic element-binding molecules like phytochelatins and metallothioneins and finally sequestering them to the root vacuole (Shackira & Puthur, 2019).

7.4.5. Biomass Production

The decline in biomass production of *T. domingensis* noticed in the second cycle could be linked to the fact that *T. domingensis* does not sustain repeated harvesting, as shown in congener species like *T. augustifolia* by Jinadasa et al. (2008) in which biomass production declined significantly after four consecutive harvests. Biomass reduction from cycles could also be linked to changes in inlet water quality such as inflow ORP dropping from 352.87 ± 70.47 mV to 143.54 ± 74.59 mV. Pezeshki et al. (1996) revealed that total biomass of *T. domingensis* reduced in response to low ORP conditions.

C. zizanioides biomass production is comparatively low, which could be explained by the adaptability of this species to conditions that could be described as abiotic stresses (George et al., 2017). High salinity (EC and TDS) could have hindered development and reduced metabolism. Edelstein et al. (2009) found that irrigation with water with EC of 6 dS/m decreased the growth rate of *C. zizanioides* significantly and concluded that the salinity threshold value for *C. zizanioides* is between 3 and 6 dS/m. Klomjek & Nitisoravuk (2005) testing species in different salinity levels confirmed that *C. zizanioides* is not tolerant to saline conditions (14-16 mS/cm). Also, cyanide content was not measured, but it was used in the mining process and it could induce some toxicity in the species and impede development. In addition, disturbance of microorganisms in the specific conditions (high salinity, poor nutriment) has been found to contribute to the decrease in the development of vetiver (Leaungvutiviroj et al., 2010). These biomass production results were lower than the results found by Elhaak et al. (2015), where plant mass reached 24.94 g in the summer time and 41.22 g (dry weight) in the winter time when they investigated *T. domingenis* population on Burullus Lake at the Mediterranean coast of Egypt. Oliveira et al. (2018) found in a Cd-spiked experiment with *T. domingensis* a biomass of 33.04±14.70 to 48.38±13.90 g (dry weight per plant). This comparatively low biomass production for both *T. domingensis* and *C. zizaniodes* could be explained by the pH range of the wetland (Pratt et al., 1984; USDA, 2009).

7.4.6. Standing Stock and Harvesting Period

As for the growth rate, standing stocks (Figure 7-9) fluctuated over time. For As, the standing stock increased at the start of the experiment and reached the highest value at the end of the experiment. The standing stock growth stagnated until the middle of the second month and then rapidly increased. In order to remove an adequate amount of potentially toxic elements from the seepage water stream, the standing stock increase should be considered. When targeting As, the end of the second month is likely to see the highest plant stock capability.

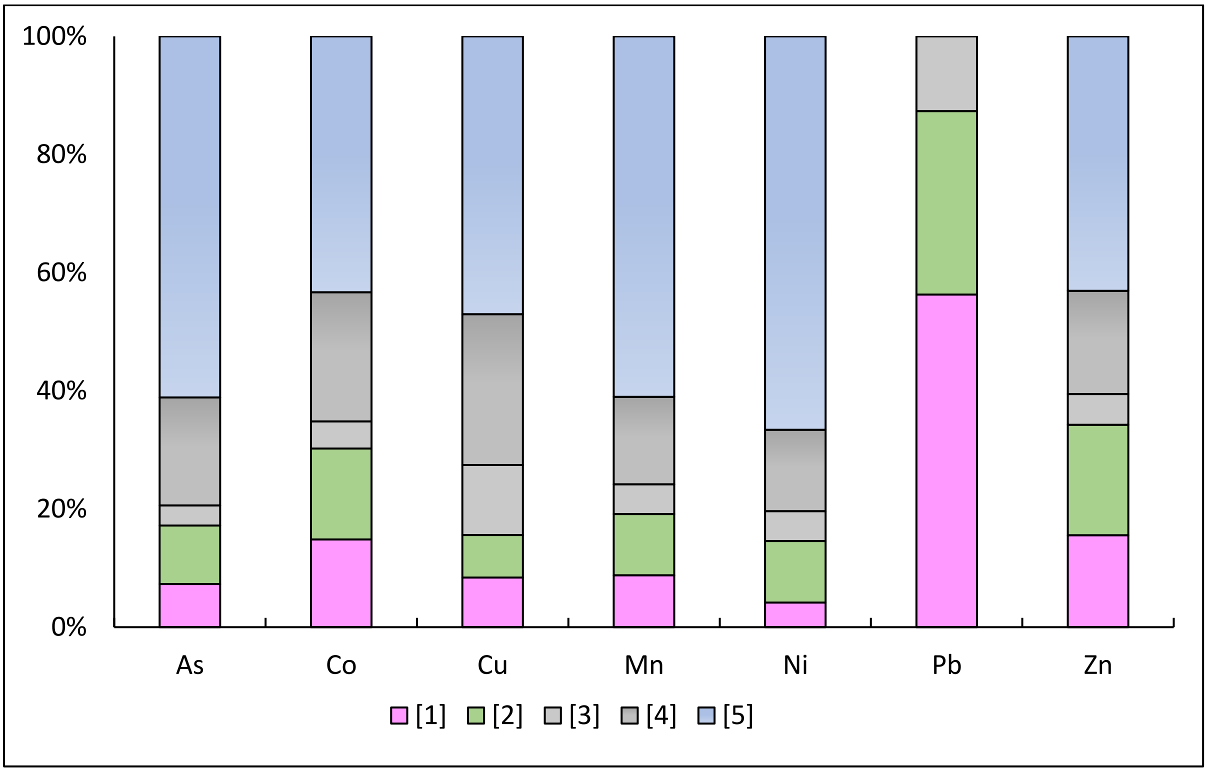


Figure 7-9 Share of potentially toxic elements in the five intervals of sampling, each interval equal to two weeks,

This growth rate was obtained because the roots systems of the plant were maintained. Uprooting the species leads to important removal of contaminants but also extends the next growth cycle. With cellulose, hemicellulose and lignin content of 26.33%, 38.67% and 4.67% respectively (dry weight), *T. domingensis* has a high potential for use as biofuel, as a source of ligno-cellulosic biomass containing exceptionally high cellulose (26%), hemi-celluloses (38%) and lignin (4%) (Abideen et al., 2011). Use of *T. domingensis* biomass for bioethanol was investigated and found to be an advantage compared to other species. *T. domingensis* may challenge any second generation species for biofuel production, for instance water-hyacinth (*Eichhornia crassipes*) which has 35-50% of hemi-cellulose but with high (12%) tannin which is undesirable for fermentation. Currently, several options for using Typha plant material are being considered including: compost for local horticulture farms and energy use via charcoal production or incineration with further management of the ash. Charcoal production appears to be the most logical and pragmatic route, taking into consideration the large volume of material that has to be dealt with (Henning, 2002). In Mauritania, use of Typha for coal was implemented in large scale with sound results involving seven small-scale Typha coal units, 1 pilot industrial unit which produced 25 tons of coal (www.gret.org). Typha coal project could provide a complementary income for rural household and could be of benefit for local communities around the mine during the exploitation stage and in the after-closure stage.

Further use of biomass could include anaerobic digestion and aerobic degradation. The recycling options of the plant biomass produced during the phytoremediation of contaminated soils include the production of renewable energy (such as biogas or thermal energy) and also of useful products such as compost (Bernal et al., 2019). *T. domingensis* polysaccharides content could also be valorized (Sorourian et al., 2020).

7.4.7. Mass Balance in the Wetland

Mass balance in the wetland was calculated to estimate the contribution of the wetland to the uptake or retention of PTE (Table 7-4).

In the first experimentation populated with only *T. domingensis*, As uptake from the whole plant biomass was 26.5 g. The seepage contribution was 24.1 g and what was going out was 22.1 g. Dry tailings transportation by wind into the wetland could explain the variation. At the second experimentation populated with *T. domingensis* and *C. zizaniodes*, the inlet As contribution was 33.5 g, 21.2 was going out and the total biomass uptake was estimated to 8 g. At the second experimentation soil contribution was not noticed. Around 5 g were retained in the wetland by adsorption and other retention mechanisms. Thus, at first, soil contribution was noticed whilst at the second experimentation, the wetland was retaining PTE.

Potentially toxic elements bioavailability is dictated by the pH condition of a wetland. Fluctuation in pH could have influenced the wetland adsorption, precipitation or release of PTE (Qureshi et al., 2004), driving the mass balance in the wetland.

Table 7-4 Potentially toxic element mass balance in the entire wetland, values are in g,

Point	As	Co	Cu	Mn	Ni	Pb	Zn
Mass Inlet	28.805	145.63	7.495	126.49	0.33	7.855	5.79
Mass Outlet	21.665	167.155	2.08	107.49	0.375	8.38	2.045
Plant uptake	17.24	67.415	145.75	22.075	17.315	162.265	221.84

7.5. Conclusions

Seepage water from the TSF had a high content of potentially toxic elements, characterized by large fluctuations depending on the ore type being processed at a particular time. Spontaneously grown *T. domingensis* showed an ability to grow in a constructed wetland watered by seepage and demonstrated a fast-growing rate of 0.18 g (dry weight) per day on average, reaching 1.61 meters high after 75 days' growth and presenting better adaptability than *C. zizanioides* which showed a growth rate of 0.03 g (dry weight) per day, reaching only 0.65 ± 0.11 m height. *T. domingensis* also achieved a higher yield of potentially toxic elements than *C. zizanioides*. *C. zizanioides* standing stocks were, depending on the potentially toxic element, 3-14 times lower compared to *T. domingensis* grown in the same set-up at the end of the tenth week.

In the context of gold mine tailing seepage phytoremediation where inflow water is characterized by high salinity, fluctuating ORP, and alkaline pH, *T. domingensis* with an acceptable biomass production is more indicated than *C. zizanioides* which has low biomass production. The optimal harvesting time is the end of the tenth week for *T. domingensis*. C. zizanioides biomass production is very low in the context of this study.

Chapter 8: Study synopsis, Conclusions and Perspectives

8.1. Introduction

The economic impact of extractive industries is interesting. On the one hand, it provides financial assets to power a country's development needs. On the other hand, criticism of the impact of mineral extraction has arisen across the board, starting with local populations, NGOs, and international and regional institutions.

Burkina Faso, a landlocked country located in the center of West Africa with 18 million inhabitants, joined the exclusive club of mine producers a few years ago. The whole landscape of Burkina Faso is changing as a result of gold and other minerals mining activities.

The production of gold in Burkina Faso rose from 12.2 tons in 2009, to 24 tons in 2010 and 32.6 tons in 2011, 30.2 tons in 2012, and 36.6 tons in 2014. Since 2009, gold has become the main provider of funds for the country, along with cotton and livestock. And it is expected to contribute to lifting the country from developing to emerging status, according to the two five-year plans set by the Government: Strategy for Accelerated Growth and Sustainable Development program (SCADD) edited in 2010 and the National Economic and Social Development Plan (PNDES) edited in 2015.

This research study sets out the background in terms of the characterization of a mining site in Burkina Faso and provides an exposure assessment of foodstuffs impacted by the mining to evaluate local community exposure. A solution through exploring the potential of plant phytoremediation on gold mining impacted site remediation was investigated.

The study objectives were to assess the contribution of the gold mine to the soil contamination as well as to the crops potentially toxic element content grown in those soils and the exposure of the local population through food intake. Pit lake use by local population in the after-mining closure stage for resilience was part of the objectives with exposure through consumption of fish evaluated. Further objectives were remediations of the contaminated land *using Leucaena leucocephala* and the TSF seepage using spontaneously grown *Typha* and introduced *Vetiver*.

The site under our scrutiny is a mid-term gold mining site located in the southeastern side of Burkina Faso which started gold production in 2008 after year and half of construction. The Youga deposits are characterized by two distinct styles of mineralization: moderately to weakly silicified host rock with quartz stockwork veining and pyrite as the predominant sulfide; and the intensely silicified arkose, with abundant quartz veins and more diverse sulfides. The site is located between the watershed of Nakamber and Nazino with annual rainfall ranging from 900 mm to 1200 mm. Local community are farmers with livestock's breeding.

8.2. Study Synopsis

8.2.1. Research Question 1: Mining Impact on Environment Quality

Mining impact on soil and water was investigated. Soil and water sampled within the site were analyzed for potentially toxic elements. Soil sample potentially toxic elements were analyzed by total digestion and also by sequential extraction analysis. Soil impact was interpreted using the enrichment factor (EF) and the geoaccumulation index (Igeo), which were computed using two different referential soils: surface soil and subsurface soil. Water of pit lakes and the nearby river was sampled on a regular basis during two years for analysis.

The mining site was divided into five areas, namely Zones 1 to 5. Zone 1 consisted of an area covering the processing plant and the TSF with a grouping of six samples. Zone 2 consisted of seven samples covering the west pits and their waste dumps. The pit lakes considered for the water sampling and analysis were located in this zone. Zone 3 covered the senior camps and the access between the camp and the mine, with seven samples. Zone 4 covered the main and east pits and their waste dumps and accounted for five samples. Zone 5 covered the south part of the site and consisted of NTV backfilled pits, Zergoré pits and waste dump, and A2 northeast pit and waste dumps; nine samples were taken in this area. Higher degrees of contamination mostly occur around the processing plant and the TSF in Zone 1 and Zone 5 covering the old artisanal mining locations.

The investigation into potentially toxic element content in mining site soil revealed anthropogenic contributions to contamination. Contaminants in the mining site exhibited huge variation due to excavation work, mining extraction and processing. Also, the tailings storage facility (TSF) created some hotspots from where contaminants were spread by wind or water. In benchmarking against Canadian Soil Quality Guidelines (CSQG) value for agricultural use, the site potentially toxic element (As, Cd, Co, Cr, Mn, Ni and Pb) contents were found to be higher. Sequential extraction revealed that the lowest share was located in the first fraction, which is readily available for biota uptake. However, the share from the second and third fractions could be mobilized in some conditions, increasing the availability of potentially toxic elements to biota.

Mining activities are well known for their huge water demand, which contributes to surface water competition with the local population, underground water depletion due to dewatering of pits for mining, and contamination due both TSF contaminant mobilization and toxic elements from ore-bearing rocks.

In this specific context, in addition against the background of national water scarcity, the use of pits as water reservoirs to compensate for mining water demand was considered. However, contamination from the site has contributed to pit lake water quality, as has wall rock leachate during filling. Water from the two pit lakes which were analyzed revealed differences with the nearby river, which was used as a benchmark.

Pit lakes water chemistry was influenced by both the mining soil washed by rainfall into the pit and the pit wall rocks. Correlation between soil parameters and water parameters was carried out. West Pit 1 As content was negatively and strongly correlated with As measured in surface soil on the site. WP1 Zn was strongly positively correlated to the soil CEC. WP1 Mn was strongly positively correlated with Co and Mn from soil. For WP2, only Zn showed strongly positive correlation with As from soil and strongly negatively correlation with both Cu and Zn from soil. NR water parameters did not show any strong nor moderate correlation with mine site soil parameters.

TSF seepage water chemistry was computed to check the correlation with pit lake water and soil. Except for Mn from the TSF seepage which showed a moderate positive correlation (0.567) with WP1 Mn, all other potentially toxic elements exhibited weak correlation. Correlation between soil total digestion potentially toxic elements concentrations and TSF seepage potentially toxic element content was carried out. Soil As was strongly correlated to WP1 As (-0.657) and to WP2 Zn (0.797). Soil Co and Mn were strongly positively correlated to WP1 Mn. Soil Zn was negatively correlated to WP2 Zn. No site soil potentially toxic elements were correlated to the NR water parameters.

Water chemistry showed that pit lake water could not be used for drinking purposes, as some elements were found at levels higher than WHO standards (As equal to 18 µg/L higher than WHO standard of 10 µg/L). The possibility of using the pit lakes for fishing was assessed against South African and Canadian guidelines.

It appears that mining sites need remediation to avoid contamination of pit lakes. Measures to prevent contaminants from spreading and to remediate soil should be taken. There are different risks arising from mining site contamination. The first risk is the spread to nearby locations, widening the impacted area. The second risk is the transfer to crops, which can lead to human health hazards. There is a high probability of use of the site for agriculture after the mining stage, and this gives rise to concern about the local population growing contaminated crops, with the associated health risks.

8.2.2. Research Question 2: Mining Potential Impact to Human Health

Mining potential impact to human health was investigated based on potentially toxic element intake through foodstuff, particularly cereals grown within or in the vicinity of the mining site. This investigation was also aimed at determining the potential for the use of the pit lakes and the fish intake risk by the local community. An exposure risk assessment and assessment of the health hazards linked to consumption of these fish by local population were undertaken.

For the intake risk assessment, a food consumption survey was undertaken in the nearby five villages. The survey consisted of cereals and fish consumption and also considered the family size. A dietary intake survey of the local populations living near the gold mine revealed cereals consumption of 0.43 ± 0.19 kg/person/day; the nationwide intake is 0.52 kg/person/day, as reported by the INDS (2017). The survey revealed that overall fish consumption was low, 5.34 ± 2.6 g/day/person, in line with nationwide figures, and confirmed that fish were mainly consumed in the form of powder mixed with sauce.

The impact of mining on cereals grown in the mining perimeter and in the vicinity was assessed by analyzing cereal samples for potentially toxic elements. Cereal PTE analysis revealed that, when considering the average by locations, EDI is below RfD. However, when considering EDI of PTE by cereal type, As, Cd, Ni and Zn exhibited EDI above the RfD and consequently a THQ above 1. These findings indicate that, with monotype cereal consumption, there is a health risk to the local population. Average EDI of elements followed this pattern: mine > Songo > Youga > Sighnoguin. This study highlights variability in PTE content in cereals and finds that some elements are higher in crops grown in some villages than those grown in the mining site. Some cereals exhibited EDI above the RfD with a THQ above 1, indicating a health risk to consumers. Combined toxicity expressed by HI were all above 1, highlighting a potential health concern. The trend Youga < Mine site < Songo < Sighnoguin was observed with an HI of 2.7, 4.3, 7.3 and 8.9 respectively. An average HI of 5.8 was observed. Pb contribution to the HI was 80 %, 75 %, 44 % and 35 % for Sighnoguin, Songo, Youga and the mine site respectively.

With the actual fish consumption, intake of potentially toxic elements by the local population does not expose them to intake higher than the guidelines. However, a reduction in the number of family members per household increases fish consumption and also increases intake of potentially toxic elements, creating health hazards. While pit lake potentially toxic elements tend to stabilize over time due to wall rock chemistry changes, runoff from nearby waste dumps and TSF can keep the contaminant level high in pit lake and increase by the same way bioaccumulation potential of fishes. Target hazard quotient (THQ) from consumption of fish was computed to estimate the potential health risks associated with the intake of fish. Values computed were below one for both entire fish and the fleshy part.

Combined exposure to cereals and fish is significantly increasing potentially toxic element intake hazards. Water from pit lakes however is not used for drinking water purposes.

Further to THQ assessment used to express the risk of non-carcinogenic effects, Target cancer risk (TR) was computed. Cancer risk is estimated by the incremental probability of an individual developing cancer, over a lifetime (60 years for Burkina Faso average life expectancy), as a result of exposure to a carcinogen. Target cancer risk (TR) for Ni, Pb and As were calculated according to following equation (Sadeghi et al., 2019):

$$TR = \frac{M_C \times IR \times 10^{-3} \times CSF \times EF \times ED}{BW \times ATc} \quad \text{(Equation 10)}$$

where M_C is the potentially toxic element concentration in fish or cereals (mg/kg), IR is the cereals or fish ingestion rate (g/person/day), CSF is the oral carcinogenic slope factor from the Integrated Risk Information System (for Ni: 1.7; Pb: 8.5 x 10^{-3}; As: 1.5 mg/kg/day), EF is exposure frequency (365 days per year), ED is the exposure duration (60 years), BW is the body weight (70 kg), ATc is the averaged exposure time to the carcinogen (365 days x 60 years) and 10^{-3} is the unit conversion factor. TR is dimensionless.

Combined THQ values were below unity except for Pb which exhibited a value of 3.85. HI of the consumption of fish and cereals by local community was 6.59 and reveals a potential adverse health effect. Target cancer risk was computed, and values are shown in Table 8-1.

Table 8-1 THQ, HI and TR of cereals and entire fish consumption

	THQ										HI	TR (10^{-3})		
	As	Cd	Co	Cr	Cu	Fe	Mn	Ni	Pb	Zn		As	Ni	Pb
Cereals	0.53	0.29	0.00	0.00	0.18	0.06	0.28	0.28	3.81	0.40	5.83	1.79	5.56	0.11
Fish	0.31		0.00	0.00	0.01	0.05	0.16	0.16	0.04	0.02	0.76	0.99	0.77	0.00
Combined	0.84	0.29	0.00	0.00	0.19	0.11	0.44	0.44	3.85	0.42	6.59	**2.78**	**6.33**	**0.11**

According to Sadeghi et al. (2019), TR values are ranged as: negligible (less than 10^{-6}), acceptable (between 10^{-4} and 10^{-6}) and unacceptable (higher than 10^{-4}). The results revealed that combined TR for As, Ni and Pb were higher than acceptable value range. The risk of cancer due to the long-term consumption of cereals and fish could be a concern.

Pb alone represent 65.4% of the HI from cereals. The other nine potentially toxic elements investigated represent 34.6% of Cereals HI.

This situation means that actors should address the need for contaminant removal before agricultural practices can be allowed. Decontamination of sites and selection of cereals with a low uptake capability and some agricultural practices could reduce these hazards. To avoid any consequence from agricultural activities in the after-closure stage, the site should be reclaimed, and contaminants removed or sequestered, or their availability reduced. The best approach is to use a low-cost, low-technology strategy and an easy to maintain method like phytoremediation.

Environmental regulation enforcement bodies should consider requesting foodstuff contamination investigation, local population exposure survey and risk assessment as part of environmental impact assessment during mining exploitation stage and in the after-closure stage.

The first two research questions impose a question about solution of remediation. These studies highlighted the need for site contamination containment and site land rehabilitation, which will reduce potentially toxic elements below the levels given in land use guidelines and also reduce availability to biota.

8.2.3. Research Question 3: Phytoremediation Option for Mining Site Remediation,

This research question was answered in two parts: the first was about investigating the capability of *Leucaena leucocephala* capability to uptake contaminants from mining soil and the second one was about tailings storage facilities seepage remediation through constructed wetland.

The use of *L. leucocephala* for mining soil phytoremediation was investigated. The objectives of this study were to evaluate the capability of *L. leucocephala* to uptake As in an As-contaminated soil watered with As-contaminated water. The laboratory experiment simulated a contaminated mining site drained by runoff from As-containing mining waste dumps. Further, the influence on uptake capability of the chelating agent ethylenediaminetetraacetic acid (EDTA) and growth enhancer containing mycorrhiza sp. was assessed. In addition, As translocation was computed to check whether *L. leucocephala* has phytoextraction potential in these specific conditions. A soil spiked with 25 and 50 mg As/kg, stabilized for 45 days, mixed with compost and supplemented with growth enhancer (GE) or EDTA or both was used.

This study revealed that the As uptake by *L. leucocephala* is correlated with the content of As in the soil, with a significant difference between the control soil and spiked soil, both without supplementation. However, no significant difference was found between 25 mg/kg spiked soil and 50 mg/kg spiked soil samples, which could be explained by toxicity which hindered As uptake by the plant. Supplementation of EDTA does not significantly influence As uptake in the conditions of this experiment. Also, supplementing with GE containing *Mycorrhiza* sp., *Bacillus* sp., does not significantly increase As uptake. Similarly, the combined action of EDTA and GE in 50 mg/kg spiked soil does not affect uptake. In 25 mg/kg spiked soil, the combined action does not significantly influence uptake in root and leaves compared to the same level spiked control soil. Accumulation of As in the below-ground parts make this plant more suitable for phytosequestration than phytoextraction and for use as forage for livestock in the after-mine closure stage.

Tailings storage facility seepage was controlled using constructed wetland. This study analyzed the capability of two species to take up potentially toxic elements from gold mine TSF seepage in a constructed wetland with horizontal subsurface flow. A naturally populated *Typha domingensis* and introduced *Chrisopogon zizanioides* were sampled on a regular basis in two experimental cycles; *Typha* only in the first cycle and *Typha* plus *Chrisopogon* in the second.

The aim of this study was to assess the ability of *T. domingensis* and *C. zizanioides* to mitigate potentially toxic elements before seepage reaches the environment after the mining closure stage in which the TSF should be rehabilitated and erosion controlled. First, the ability of these weeds to grow in this specific condition was assessed, based on their growth rate. Second, their potentially toxic element uptake capability was measured, using bioaccumulation factor, translocation factor and standing stock. Finally, a sustainable harvesting period, a period where harvesting will guarantee high removal of PTE, was estimated.

Seepage water from the TSF contained a high content of potentially toxic elements, which is influenced by the processing residue which, in turn, depends on the ore type being processed. The pouring location inside the TSF affects seepage turbidity; closer to the wall, water turbidity was higher. Spontaneously grown *T. domingensis* showed an ability to grow inside the bottom-lined trench watered by seepage and demonstrated a fast-growing rate of 0.44 g (dry weight) per day on average, reaching 1.61 meters high after 75 days' growth and presenting better adaptability than *C. zizanioides*. Typha also achieved a higher yield than vetiver. At the end of the tenth week, vetiver stand stocks were 3, 7, 4, 7, 14, 7, 5, and 3 times lower than typha grown in the same set-up for As, Co, Cr, Cu, Mn, Ni, Pb and Zn respectively.

In the context of gold mine tailing seepage phytoremediation, when inflow water is characterized by high salinity, fluctuating ORP, and alkaline pH, *T. domingensis* with an acceptable biomass production is more indicated than *C. zizanioides* which has low biomass production.

In field implementation of soil remediation through use of *L. leucocephala* must consider the soil physicochemical conditions influencing the targeted element speciation, the organic matter, CEC and humidity. Selection of chelating agent which could enhance uptake of the targeted element is strongly suggested. Native microorganisms for example fungi could influence positively PTE uptake by plant. Plantation of *Leucaena leucocephala* should be done according to recommendation of Prasad et al. (2011) study which found that 1-meter distance between *Leucaena* plants guarantees an optimum biomass yield. Use of manure which is locally available could also help.

Regarding the wetland populated with vetiver and Typha for remediation and PTE spreading control, in field full scale experimentation must consider particularly the fluctuation of the contaminants. During mining exploitation stage, continuous pouring of process tailings guarantees almost continuous seepage and feeding of the wetlands. However, when processing ceases, fluctuations will be huge, varying between periods with zero feeding of the wetland and periods of high feeding (during rainy season). Full scale remediation should be designated to consider those parameters: selecting plants which could sustain the dry season, dimension which could handle peak period or alternatively go for mix flow wetland (surface and subsurface), adjust flow to the capability of the plant (standing stock) for efficient removal of PTE. The high variability of As concentrations in the site should be considered during the design of the real remediation process. The wetland should be designed based on the mass balance to achieve a optimal uptake by species for an outlet suitable to discharge guidelines.

Several options for using Typha plant material are being considered including: compost for local horticulture farms and energy use via charcoal production. In Mauritania, use of Typha for coal was implemented in large coal with sound results involving seven small-scale Typha coal units, 1 pilot industrial unit which produced 25 tons of coal (Gret, 2016). Typha coal project increased a complementary income for rural household and could be of benefit for local communities around the mine during the exploitation stage and in the after-closure stage.

Further use of biomass could include anaerobic degradation and aerobic degradation (Bernal et al., 2019). *T. domingensis* polysaccharides content could also be valorized (Sorourian et al., 2020).

8.3. General Conclusion

An investigation into mining soil potentially toxic elements revealed mining site soil contamination, which could be attributable to anthropogenic action (chemicals used for gold extraction, fuel emission, tire abrasion, etc.).

Results of both analyses clearly revealed anthropogenic contribution to pollution. Zoning of the site highlighted that the process plant, the old artisanal mining and the TSF were the hotspots and fear of overspread of contaminants to water bodies as well as uptake by cereals grown on those land was constant. The findings from this study are valuable for providing effective management options for risk prevention and control of the persistent PTE accumulation.

Cereals grown inside the perimeter and at local villages were sampled and analyzed to investigate the contribution of mining to cereals potentially toxic element content. Subsequently, intake risk assessment was carried out to evaluate the intake risk of local population. The cereals intake survey done, and calculation made give a cereals consumption of 0.43 kg/person/day.

Site soil contamination leads to cereals grown in the site having high levels of potentially toxic elements and to pit lake water contamination. Cereals grown within the site were found to have some intake risk, and caution was required for consumption of pit lake fish.

In addition, an exposure assessment was carried out to evaluate the intake hazard when consuming fish from those pits. Comparison was again made with fishes from the local river (Nakambé). Mining impact on water resource is established and this study tried to check the potential of balancing this by creating water reservoir using the open cut pit of mining activities. Two pits filled with water by diverting nearby river were considered and the quality analysis for both potentially toxic element and infield parameters. The pit lakes water quality was benchmarked with a nearby river (Nakambé) and to selected national standard.

Site soil contaminant remediation was examined with *Leucaena leucocephala* in vitro. Solution to uptake arsenic and potentially toxic element was investigated using *Leucaena leucocephala*. In laboratory investigation, supplementation of the growth media with EDTA and growth enhancer was verified. Further, tailings storage facility contaminant spread was studied in a wetland populated with *Typha domingensis* and *Chrisopogon zizanioides*. While *Typha* and *Chrisopogon* both showed similar As uptake, *Typha* was found to have a higher biomass production than *Chrisopogon*.

It comes out through this study that Burkina Faso:

- should consider environmental impact assessment including mining induced potentially toxic element exposure assessment of local population through food intake,
- enforce continuous assessment of mining impact on nearby agriculture, fishery and breeding,
- should set clear objectives on soil and water remediation and the guideline values to achieve; also mining companies must integrate in their remediation process the recommendations about land and water quantity and quality management,
- should accompany institutes to enforce mining regulations and texts,
- should sensitize on good agricultural practices, livestock management and fishery for safe food for local population around mining companies,
- Promote cost effective, easy to handle remediation techniques like phytoremediation based on indigenous species,

Those findings could be of benefit for Burkina Faso mining and could benefit neighboring countries like Côte d'Ivoire, Mali and Ghana which are close to the mining site under scrutiny. The benefit could come from the methodology used for the investigation of contamination, the exposure assessment and investigation of species for phytoremediation.

8.4. Contribution to the State of the Science

When focusing on mining site characterization and remediation, this doctoral study used preexisting methods and techniques to assess mining site soil impact by considering two soil sample analysis methods (total digestion and sequential extraction analysis) and interpreting data using two different factors: the enrichment factor and the geoaccumulation index. Pit lakes formed from open-cut pits were influenced by local soil geochemistry and contaminant mobilization pathways from hotspots like the processing plant and TSF. Pit lake water quality does not meet World Health Organization standards for drinking water due the geogenic contribution of As in ore. The uptake of contaminants by cereals grown in mining sites varies according to cereal species, with wind mobilization of contaminants giving a pattern to the contamination nearby. In general, mining site cereal contaminant levels are higher than in the nearby villages, but levels of some elements are higher in cereals from the villages. This particularly the case with lead (Pb) due to intensive use of lead-based batteries in the nearby villages, mostly for artisanal mining.

Contaminant containment of the mining site under investigation was envisaged during this doctoral research. Mining site soil remediation was investigated using *L. leucocephala*. The experiment was carried out in a greenhouse condition with EDTA and growth enhancer supplementation revealing that supplementation does not significantly influence *L. leucocephala* As uptake. TSF seepage remediation was investigated using two different species: *T. domingensis* spontaneously grown in a lined trench and *C. zizanioides* intentionally added. This investigation revealed that both species take up As in a similar way, but *T. domingensis* has a higher biomass production.

8.5. Future Research

Gold mining, and mining in general, have environmental issues, which should be understood as a prerequisite for developing adequate remediation strategies. Mining soil contamination must be investigated by mining actors and government inspections bodies. For the purpose, mining site characterization methodology must be edited including, sampling methods, transportation, analysis methods and interpretation of results. Increasing existing institute capacities in terms of both human capital and analysis equipment for analysis and expertise is welcome. Tailored procedures considering the country specificities all along the process of investigating, sampling, analyzing and interpreting with a set of background values for comparison is highly needed.

Contaminated soil may lead to contaminated crops and uptake by local population. Thorough risks assessments based on full diet intake survey must be undertaken to accurately estimate PTE uptake by local population and to assess the risk linked to this intake. Further, sensitizing local health officer to those risks in the consultation could help in anticipating chronic exposure or diseases being ignored. Institutional survey of food intake for a larger population will lead to more accurate values. In addition, interference of food processing to bioavailability of PTE must be considered as processing could enhance or diminish PTE toxicity. Local population food processing style may induce some changes and must be known and integrated in a full risk assessment. For full exposure assessment all PTE uptake pathways should be investigated and considered like airborne PTE due to mining induced dust, child playing on the soil, skin transfer when swimming in contaminated pit lakes. Human exposure to potentially toxic elements through consumption of livestock-based products (meat and milk) and cereals should be investigated as well as the exposure through consumption of garden market products.

It came out that mining wasteland reclamation must be done to reduce the impact and to respond to regulation requirements. Investigation was done on locally available species for the remediation. The most important way to deepen understanding is to further examine the use of local plants for the remediation of contaminated sites. As EDTA contribution to As uptake by *L. leucocephala* was not satisfying, investigation using other chelating agents (EDDS) is recommended considering the toxicity, the availability and impact to PTE mobilization to underground or nearby water bodies. Thus, it is advisable to monitor the movement of potentially toxic elements in soil through an appropriate control of water movements, in order to prevent a downward displacement and/or the contamination of groundwater resources with soluble potentially toxic element species (Barbafieri et al., 2017). Typha presented an interesting standing stock, biomass could be used for coal for local communities uses and income generating activities. Plants with accumulated potentially toxic elements could be used as firewood and ash could be collected for further treatment or used as construction material. Typha or vetiver could be transformed to coal blocks for used in rural stoves for food processing.

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Curriculum Vitae

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Baccalauréat Série D (Mathématiques et Sciences de la nature)

2002, Diplôme de Bachelier de l'enseignement du second degré,
Université de Ouagadougou,

Key trainings

- ⇒ *Loss Prevention and safety promotion in chemical process Industries*. Organization of the Prohibition of Chemical Weapons (OPCW), the Bergische University of Wuppertal (BUW), the Federal Republic of Germany; Wuppertal, Germany, from 03 – 07 April 2017.
- ⇒ *Energy Management and Cleaner Production in small and medium scale industries*. CSTM Twente Centres for studies in Technology and sustainable development, 2nd November-4th December 2009. University of Twente, Enscheda, Netherland,
- ⇒ *Environmental and social impact assessment in Burkina Faso,* ISTAPEM, Ouagadougou, Burkina, 07 – O8 Dec 2018,
- ⇒ *Training on Monitoring of air pollution,* International Agency for Atomic Energy (IAEA) and National nuclear safety institute (ARSN). Burkina Faso, 13 – 15 Nov 2017,
- ⇒ Summer school on *Wastewater Treatment Plants and Management*, November 04–11, 2012. Excellence Center for Development Cooperation (EXCEED), UFSCar, Federal University of São Carlos, USP-SC, São Paulo University, São Carlos, Brazil.
- ⇒ Summer school on *Global Warming and Sustainable Water Management*. 16th to 22th November 2011. Technische Universität Braunschweig, Braunschweig, Germany,
- ⇒ Conference on *Innovative water technologies* 11-13 may, 2015, Ouagadougou, Burkina Faso, USAID West Africa Water Supply, Sanitation and Hygiene Program, Global water for sustainability (GWS), Florida International university (FIU),
- ⇒ International workshop of Young professionals on the *Management of waste in West African middle cities*. Organised by Réseau Projection at the " International institute for water and environmental engineering (2IE)", 5th-7th July 2010, Ouagadougou, Burkina,
- ⇒ 3rd international biofuels Conference. "*Biofuel prospects and opportunities for Africa*" 14 to 16 November 2011. 2IE (international institute for water and environmental engineering), MMCE (Ministry of Mine), CIRAD. Ouagadougou, Burkina Faso,
- ⇒ Formation pour les *Personnes compétentes en radioprotection*, Autorité Nationale de la Radioprotection et de la sureté nucléaire (ARSN), Ouagadougou, 27 au 31 Juillet 2015,
- ⇒ Intensive Training on *Mycotoxin Analysis*, University of Ghent, Department of Bioanalyse – Laboratory of Food Analysis, Belgium, 28 August to 10 September 2014,
- ⇒ 3rd Africa-wide Women & Young Professionals in Science competitions, "*Scientific writing, communication and policy advocacy*", Workshop, 24-28 Sep 2012. Entebbé, Uganda.
- ⇒ Western Africa Region workshop on the *Application of the renewable energy to rural electrification*. 2nd Regional Workshop Agenda, 7th – 11th November 2011. European Union Energie Initiative (EUEI), Institute for Energy and Transport Renewable Energy (ISPRA). EC training initiative for experts in energy. Ouagadougou, Burkina Faso,
- ⇒ *Mycotoxin inspection for food safety*. Trainers Training Program. Training and Dialogue Programs of the Japan International Cooperation Agency (JICA), From Jan. 31, 2011 to Apr. 23, 2011. Hyogo, Japan,

Scholarship/Award/Gratitude

⇒ Merit scholarship for technology for PhD study, Islamic development bank, 2014

⇒ Charlotte conservation fellowship, Africa Wildlife Foundation, (AWF) Capacity Building Program Award,

⇒ Excellent Leadership Awards, Islamic development bank, November 2014, Promising contribution to science and community,

⇒ Semi-finalist of the 3rd competition on youth professional and science in "feed one billion African in a changing context" 2012. Organized by RUFORUM, ACP-EU (CTA), IFS, the Forum for Agricultural Research in Africa (FARA), ANAFE, NEPAD AGRA,

Conférence participations

⇒ COMPAORE, W.F., DUMOULIN, A., ROUSSEAU, D.P.L. *A constructed wetland populated with Typha and vetiver for gold mine tailing storage facilities seepage treatment*. 8th International Symposium on Wetlands Pollutant Dynamics and Control, WETPOL 2019. 17 – 21 June 2019, Aarhus University, Denmark.

⇒ COMPAORE, W.F., DUMOULIN, A., ROUSSEAU, D.P.L. *Gold mine pit lakes use for aqualculture potential health risk assessment: case study from new booming country, Burkina Faso*. Water Security and climate change conference, 03-05 December 2018, Nairobi, Kenya.

⇒ COMPAORE, W.F., DUMOULIN, A., ROUSSEAU, D.P.L. *Gold mine pit lakes as water reservoir for local population in a water resourceless country: putting into practice*. Youth Global actions map for a safe water future - nature and community based experiences for water solutions, 8th World water forum, Brazil, March 2018.

⇒ COMPAORE, W.F., DUMOULIN, A., ROUSSEAU, D.P.L. *Pit lakes as water reservoir for local population: opportunities and threats*. IWA young Water Professionals, 5th Benelux, young Water Professional Regional Conference, 5 – 7 July 2017, Ghent.

⇒ COMPAORÉ, W.F., *Life cycle assessment of rural main activities source of revenue as tool for water management in rural field*. In solution platform, World Water Forum, Marseille, France. 12-17 March 2012, co-organized by the French Government, the city of Marseille and the World Water Council.

⇒ W.F. COMPAORÉ, KABORE, S., POUAN, O., KOUDOUGOU, K. Inventory and statement of wastewater management in Burkina Faso. (Poster). Summer school on *Wastewater Treatment Plants and Management*, November 04 – 11, 2012, Excellence Center for Development Cooperation (EXCEED), UFSCar, Federal University of São Carlos, USP-SC, São Paulo University, São Carlos, Brazil.

Scientific publications/Researches

⇒ COMPAORE, W.F., DUMOULIN, A., ROUSSEAU, D.P.L. (2019). Trace elements content in cereals from a gold mining site in Burkina Faso and intake risk assessment. *Journal of Environmental Management*, 248, 109292. https://doi.org/10.1016/j.jenvman.2019.109292

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