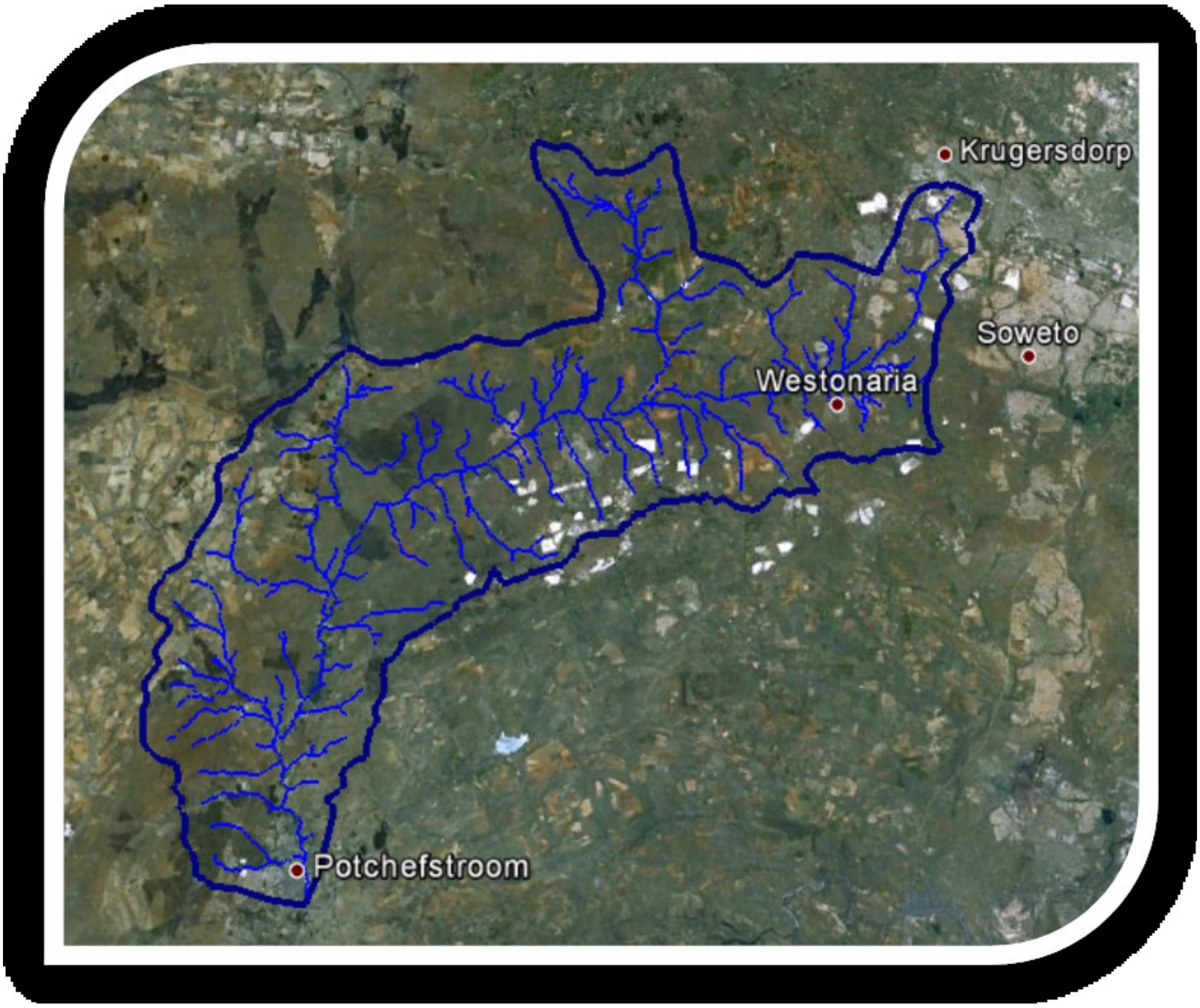
A holistic view on the impact of gold and uranium mining on the Wonderfonteinspruit



David Hamman

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Supervisor: Prof. Leon van Rensburg
Co-Supervisor: Prof. Ingrid Dennis
Assistant Supervisor: Dr. Rainier Dennis

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“Most of the fundamental ideas of science are essentially simple, and may, as a rule, be expressed in a language comprehensible to everyone.” Albert Einstein

“To raise new questions, new possibilities, to regard old problems from a new angle, require creative imagination and marks real advance in science.” Albert Einstein

“If we knew what it was we were doing, it would not be called research, would it?” Albert Einstein

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Summary

The Wonderfonteinspruit (WFS) flows through the richest gold mining region in the world and has subsequently been exposed to the related pollution for more than a century. In order to determine the extent of mining related pollution in the WFS, sediment, water, soil, grass and cattle tissue samples were collected, analysed and compared from an experimental group and a control group.

This study identified cobalt, nickel, zinc, selenium, cadmium, gold, lead and uranium as elements of interest by comparing sediment samples from the WFS and the Mooi River (MR) (which served as a control or background site). The cobalt concentration was found to be 16.37 times higher, the nickel concentration was 30.4 times higher, the copper concentration was 3.59 times higher, the zinc concentration was 103.49 times higher, the selenium concentration was 7.14 times higher, the cadmium concentration was 17.88 times higher, the gold concentration was 4.78 times higher, the lead concentration was 1.32 times higher and the uranium concentration was 375.78 times higher in the initial comparison with sediments from the MR. These results were all found to be significant.

All these elements are by products of non-ferrous mining activities as was described in the literature review. The elevated concentrations of these elements, which were found in the streambed sediment of a site in the Lower-Wonderfonteinspruit, suggest that they could have resulted due to upstream gold mining activities. These gold mining activities were initiated more than a century ago and continue to this day.

Analysis of the different particle size fractions (sand, silt and clay fractions) revealed that the highest elemental concentrations were found in the clay sized fractions. The clay sized fraction usually contains secondary soil minerals which have the ability to adsorb dissolved cations onto their surface areas. Further analysis revealed that the sand fraction of the WFS sediment contained a substantial concentration of cobalt, nickel, copper, zinc, lead and uranium which, upon initial inspection could not be explained.

X-Ray Diffraction (XRD) analysis revealed that more than 90 % of the WFS sand, silt and clay fractions consisted of quartz, which was much higher than that of the MR. Due to the particle size of quartz, it generally dominates the sand and silt fractions, and finding it at levels above 90 % in the clay sized

fraction is thought to be highly irregular. This could be explained by the extraction and processing of gold reefs from the goldfields in the catchment. The gold reefs consisted of quartz veins that were milled to a fine dust and pumped onto slime and sand dumps after the gold was extracted. The most abundant ore minerals found within these dumps were uraninite(UO2), brannerite (UO3Ti2O4), arsenopyrite (FeAsS), cobaltite (CoAsS), galena (PbS), pyrrhotite (FeS), gersdofite (NiAsS) and chromite (FeCr2O4), which contain some of the elements of interest. These dumps are either located in close proximity to the WFS or connected to the WFs via canals or pipelines. Erosion of these dumps would then introduce this finely milled quartz into the stream system. Therefore, the elements found in the sediment of the WFS were not only introduced to the system in the dissolved form, but also in the particulate form.

The water samples that were collected from the experimental site (WFS) were found to exceed the cobalt, nickel, copper, zinc, selenium and cadmium concentrations ranges which are normally found in natural waters. In addition to this, the cadmium, lead and nickel concentration in the WFS water samples were found to occasionally exceed the target water quality ranges for livestock water as set by DWAF (1996). Water samples that were collected from the control group were found to exceed only the selenium concentration found in natural water sources as found by Crittenden *et al*., (2005). Cattle in the experimental group drink directly from the WFS and may stir up the sediment and thereby increasing the elemental concentrations within the water prior to ingestion. The target water quality ranges (TWQR) for livestock watering, as set by DWAF 1996, were exceeded by the average nickel and lead concentrations found in the disturbed WFS water samples. Although the elemental concentrations in the respective water samples were fairly low there was a definite practical significant difference between the WFS water and the MR water samples, as well as the disturbed WFS water and the MR water samples. The WFS water quality seemed to have a very large standard deviation which could serve as an indication that the elemental concentrations are highly variable over time.

The elemental concentrations that were found in soil samples from the respective sites were compared to elemental concentrations found in normal agricultural soil as presented by Bergman (1992), which revealed the following results. The cobalt concentrations in the soil samples from the soil along WFS site, soil along MR site and irrigation MR site exceeded the agricultural threshold value. The nickel concentrations in the soil samples from the soil along WFS site, soil along MR site, wetland WFS and irrigation MR site exceeded the agricultural threshold value. The zinc concentrations in the soil samples from the soil along WFS site exceeded the agricultural threshold value. Copper, selenium, cadmium and lead concentrations did not exceed the agricultural threshold values in any of the respective sites. The agricultural threshold value for uranium concentrations was exceeded in the soil samples from the soil along the WFS site and the wetland WFS site.

The comparison between the elemental concentrations that were found in the soil samples from the irrigated soil WFS site and the irrigated soil MR site revealed a practically significant difference for the copper, zinc and uranium concentrations. The comparison between the elemental concentrations found in soil samples from the soil along the WFS site and the soil along the MR site revealed a practically significant difference for all elements of interest. The analysis of the elemental concentration in the different particle size fractions of soil samples from all the sites (excluding the irrigated pastures) displayed highest elemental concentrations in the clay sized fraction. The elemental concentrations that were found in this fraction are generally considered to be available for plant uptake, as most of them are usually bound to the surface of secondary soil minerals. The sites with the highest concentration of plant available elements were found to be the soil along WFS site and the wetland WFS site.

The elemental concentrations found in the grass samples from the respective sampling sites were compared to elemental concentrations that are normally found in grass pastures (Underwood & Suttle, 2001). The cobalt, nickel, copper and concentrations that were found in the grass samples from most of the sites in both the control and experimental groups were all found to exceed the concentration ranges found in natural pastures. The cadmium and zinc concentrations in the grass samples from the soil along WFS site were found to exceed the respective concentration ranges found in natural pastures.

The normal uranium concentration found in irrigated or natural grasses could not be found in an extensive search. Dreesen *et al*. (1982) reported 0.16 mg/kg uranium in grasses and 1.8 mg/kg uranium in shrubs that grew on soil-covered tailings material. All the sites in the experimental group, including the control WFS site, drastically exceeded these concentrations, which may suggest that the grasses in the experimental sites have been exposed to elevated uranium concentrations.

The grass samples with the highest average elemental concentrations were found in the soil along WFS site and irrigated soil WFS site. Lead was to be the only element of interest to have the highest concentration in grass samples from the irrigated soil WFS site. The irrigated soil WFS site portrayed significant transfer factors for nickel, copper, zinc, lead and uranium. This could serve as an indication that the grasses under irrigation in the WFS site absorb and accumulate the highest concentration of elements in respect to the soil concentrations found in the various sites. Therefore, the irrigation from the WFS has a profound effect on the nickel, copper, zinc, lead and uranium concentration in the grass samples under irrigation.

The results obtained from the comparative analysis of the elemental concentration in grass samples from the irrigation WFS and irrigation MR sites revealed that all elemental concentrations except for that of zinc had a difference that was practically significant, with the uranium concentration having the largest effect size.

The results obtained from the comparative analysis of the elemental concentration in grass samples from the soil along WFS and soil along MR sites revealed that all elemental concentrations had a difference that was practically significant uranium, nickel and zinc concentrations having the largest effect sizes. Considering that a large effect size is achieved at a value equal to or greater than 0.8, the uranium concentration therefore had a massive difference in both comparisons.

The results obtained from the comparative analysis of the elemental concentration in grass samples from the wetland WFS and control WFS sites revealed that only the cobalt, nickel and uranium concentrations had differences that were practically significant, with the cobalt concentration having the largest effect size.

The results obtained from the comparative analysis of the elemental concentration in the grass samples from the soil along WFS and control WFS sites revealed that all the elemental concentrations except for the lead concentration had a difference that was practically significant. The cobalt, nickel and zinc had the largest effect sizes.

The elemental concentrations that were found in cattle liver, kidney and muscle tissue samples from both the experimental and control groups were compared to elemental concentrations normally found in cattle samples as found in Pulse (1994), ATSDR (2004), and ATSDR (2011). This comparison revealed the following results:

- The nickel, cadmium and lead concentration that were found in the cattle liver, kidney and muscle tissue samples from both the experimental and control groups were found to be within the ranges normally found in cattle.

- Cobalt concentrations found in the liver and muscle tissue samples of cattle from both the experimental and control groups exceeded the normal ranges, and the cobalt concentrations found in the kidney samples from the experimental group exceeded the normal range.

- The copper concentration found in the kidney samples from the cattle in the experimental group exceeded that of the normal concentration range.

- The zinc concentration found in the liver and kidney samples in the cattle from the experimental group, and the kidney samples from the cattle in the control group exceeded the normal range.

- The selenium concentration found in the liver, kidney and muscle tissue samples in the cattle from the experimental group, and the kidney samples from the cattle in the control group exceeded the normal range.

- The uranium concentration found in the liver, kidney and muscle tissue samples in the cattle from the experimental group exceeded the normal range.

The comparison between cattle tissue samples from the experimental and control group revealed that nickel, zinc, selenium, lead and uranium concentrations all reveal a practically significant difference. Uranium, nickel and lead portrayed the largest differences between the two groups. The uranium concentration in the cattle samples from the experimental group was 126.75 times higher in the liver, 4350 times higher in the kidney, 47.75 times higher in the spleen, 31.6 times higher in the muscle tissue, 60 times higher in the bone and 129 times higher in the hair than that of the cattle samples from the control group. In addition to this, the uranium did not only accumulate in the predicted tissue samples (bone, liver and kidney), but also in the muscle tissue samples. The nickel concentrations in the cattle samples were all found to be higher in the experimental group, with liver 1.4 times higher, kidney 387.5 times higher, spleen 2.1 times higher, muscle tissue 2.8 times higher, bone 167.5 times higher and hair 76.5 times higher than that of the cattle samples from the control group. The lead concentrations found in the cattle samples from the experimental group were found to be 3.8 times higher in the liver, 17.3 times higher in the kidney, 3.3 times higher in the spleen, 3.2 times higher in the muscle tissue, 9 times higher in the bone and 12.2 times higher in the hair than the cattle samples from the control group.

Furthermore, the study revealed that the major route of ingestion for all the elements of interest, excluding nickel and cobalt was *via* the ingestion of grass. The major route for nickel and cobalt ingestion was *via* soil ingestion. The elemental concentrations from water ingestion were found to be a less significant.

It was shown that a predictive cattle consumption model was developed and calibrated from data gathered from a control and experimental group. Animal matter analysed for both groups were related to the cattle age of six years. Although good correlation between observed and simulated values was achieved, the exiting model fit is non-unique. To obtain a more precise model fit a similar dataset is required for both groups, but at a different age.

The predictive model also showed that if only grass were to be used as input, there were no significant changes in the correlation between observed and simulated values. This has a huge advantage in terms of costs associated with laboratory analyses as the analysis of grass will be sufficient for using the model.

A human health risk assessment was performed based on the results of the cattle consumption model. It was shown that no toxic risk exits for both the control and experimental groups if an intake rate of 0.13 kg of meat per day was assumed. Furthermore, Figure 6-11 clearly indicates that an intake rate of up to 0.38 kg of meat per day also has no toxic risk for both groups, which strongly suggests that there is no risk to the human food chain.

The cattle grazing in the WFS appear to be in a good physical condition and according to the farmer; the reproduction rate is at desirable levels. Good farming practices would have also played a significant role to achieve this.

***Key words**: Heavy metals, trace elements, acid mine drainage, sediment, water, grass, soil, cattle, risk assessment, Wonderfonteinspruit, Mooi River, ICP-MS, XRD, cattle consumption model*

List of Abbreviations

AMD	Acid Mine Drainage
CPF	Cancer Potency Factor
DWAF	Department of Water Affairs and Forestry
i.e.	id est
ICPMS	Inductive Plasma Coupled Spectroscopy
EPA	Environmental Protection Agency
MR	Mooi River
NWRS	National Water Resource Strategy
ND	Non Detect
RfD	Reference Dose
TWQR	Target Water Quality Range
WFS	Wonderfonteinspruit
XRD	X-Ray powder Diffraction

Units of Measurements

°C	Degrees Celsius
cm	centimetre
d	day
g	gram
g/cm^3	gram per cubic centimetre
ha	hectare
km	kilometre
km^2	square kilometre
L	litre
m	meter
mm	millimetre
mg/kg	milligrams per kilogram
mg/L	milligrams per litre
mL	millilitre

Chemical Parameters

Al	Aluminium
As	Arsenic
Ba	Barium
B	Boron
Ca	Calcium
Cd	Cadmium
Cl	Chlorine
Co	Cobalt
Cr	Chromium
Cu	Copper
Fe	Iron
Hg	Mercury
I	Iodine
K	Potassium
Li	Lithium
Mg	Magnesium
Mn	Manganese
Mo	Molybdenum
Na	Sodium
Ni	Nickel
P	Phosphorus
Pb	Lead
S	Sulphur
Se	Selenium
Si	Silicon
Sn	Tin
V	Vanadium
Zn	Zinc
U	Uranium

Table of Contents

List of Figures

List of Tables

List of Pictures

1 Introduction

1.1 Historical Background

The Wonderfonteinspruit (WFS) has its origin at the surface water divide immediately to the south of Krugersdorp in the Gauteng Province and flows into the Mooi River, close to Potchefstroom in the Northwest Province of South Africa (Coetzee *et al*., 2004).

It forms part off the Mooi River (Kromdraai) Catchment, which constitutes an important component of the Vaal River System. The name WFS means "Wonderful-fountain-stream", a name it derived from the large volumes of dolomitic groundwater that once fed the stream via karst springs. It drains a catchment area of approximately 1600 km² and flows for approximately 90 km through an area known to have the richest gold deposits in the world (Winde, 2010). The WFS has been divided into the Upper and Lower Wonderfonteinspruit areas (Coetzee *et al*., 2004). A map of the catchment can be seen in Figure 1-1.

1.1.1 The Upper-Wonderfonteinspruit

The Upper WFS originates at Tudor Dam, south of Krugersdorp and ends in Donaldson Dam near Westonaria. The West Rand goldfield, which has produced more than 1900 tons of gold, was first mined in 1887, only a year after the discovery of gold in the Witwatersrand (Handley quoted in McCarthy 2006). Most of the mines in the Upper WFS have been closed or abandoned and the area is dominated by unrehabilitated slimes dams, rock dumps and sand dumps (Coetzee *et al*., 2004).

1.1.2 The Lower-Wonderfonteinspruit

The Lower WFS starts below Donaldson Dam, at the beginning of the 1 m pipeline which was constructed in 1977 to transport water from various gold mines over three dewatered dolomitic compartments (Oberholzer, Venterspos, and Bank). The pipeline was constructed to prevent recirculation of the water that was pumped to the surface from underground mine workings (Coetzee, 2004). The 1-m pipeline stretches for approximately 32 km and prevents recirculation of pumped water; it ends immediately to the north of Carletonville. The Lower WFS area comes to an end at its confluence with the Mooi River above Boskop Dam (Coetzee *et al*., 2004). The Mooi River originates at the Bovenste Eye in the Mathopestad area (near Ventersdorp) and flows into Klerkskraal Dam. From Klerkskraal Dam it flows into Boskop Dam, then Potchefstroom Dam and finally, the Vaal River (DWAF, 2002)

The goldfield in this area is often referred to as the West Wits Line and has supported 10 major mines that produced more than 7300 tons of gold (McCarthy 2006). Mining only commenced after the dolomitic compartments were dewatered in the 1930's. Several of the gold reefs in this area also contained elevated concentrations of uranium, which lead to large-scale uranium production in the early 1950's. The mining activities gradually transformed the Lower WFS into a barren streambed littered with several sinkholes and four dried up springs (Swart *et al*., 2002).

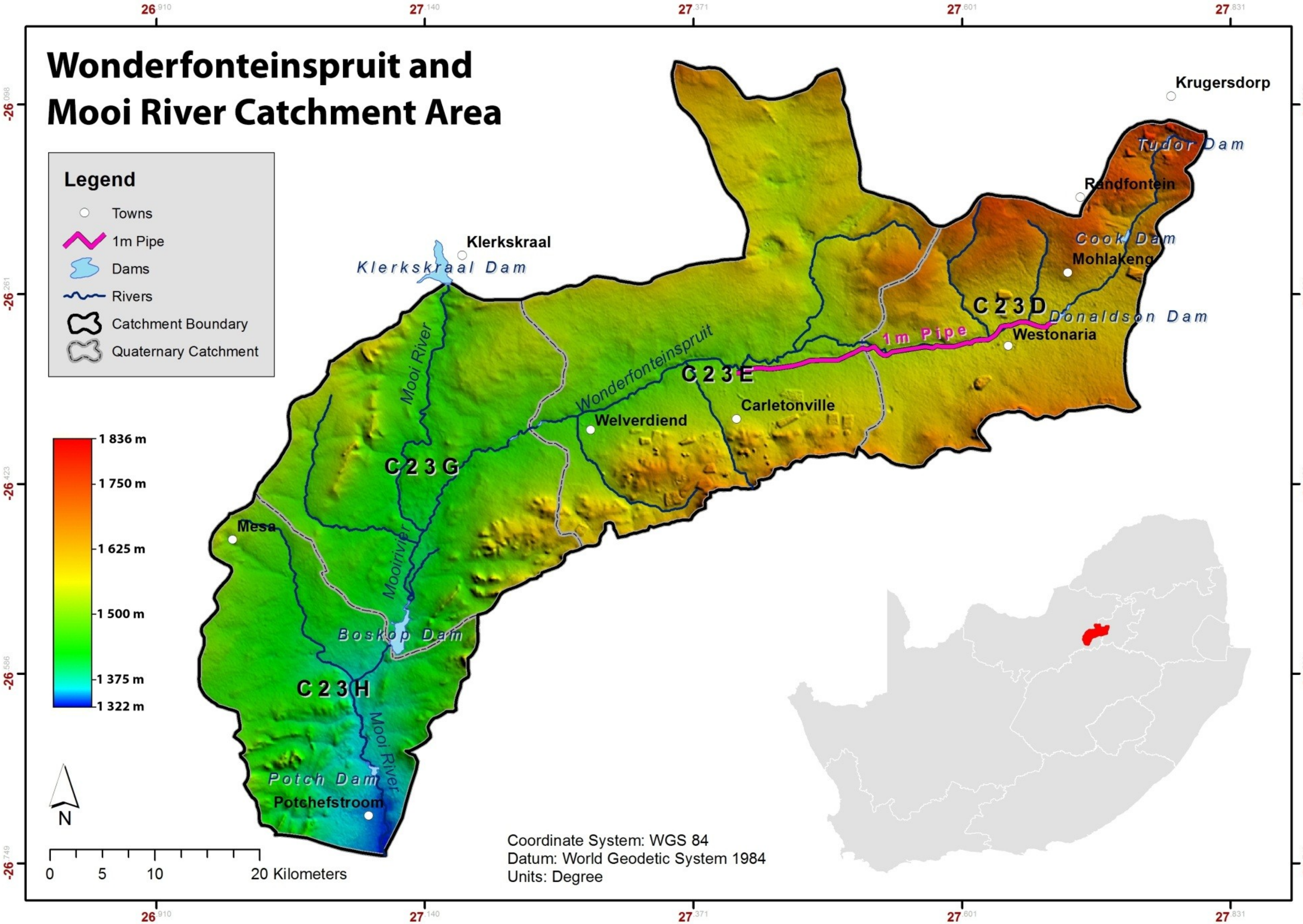


Figure 1-1: Map of the Wonderfonteinspruit, surrounding areas and the Mooi River Catchment

1.2 Pollution of the Wonderfonteinspruit

Mining, which involves the extraction and processing of ores, generally affects relatively small areas. It is the tailings and waste rock deposits close to the mining site, which is the source of metal contamination of water resources (Salomons, 1995).

Surface water and groundwater pollution is a major concern throughout the world (Sparks, 2003). According to NWRS (2004), as quoted by (Lin & Harichund, 2011) there is an even greater concern for all South African water users as water is a scarce resource being placed under pressure by ongoing mining pollution. According to Stoch 2008 as quoted by (Winde, 2010) the issue of radioactive contamination of the WFS was raised for the first time in 1967.

In 1991 the Council for Geoscience conducted a high – resolution airborne radiometric survey over the Witwatersrand area. The survey indicated that portions of the WFS (mostly wetlands) have been polluted with radioactive material as a result of upstream mining activities (Coetzee *et al*., 2004).

The pollution in the WFS has already reached headlines in local and international media (Winde, 2010). It has also been the subject of numerous studies and reports by the National Nuclear Regulator, Department of Water Affairs and the Council of Geoscience (Coetzee *et al*., 2004).

Mining and processing of uraniferous gold ores are mainly responsible for radioactive and heavy metal pollution that the WFS has been associated with (Coetzee *et al*., 2004). The main mechanism for release of metals from mine wastes are through leaching into surface and groundwater, fugitive dust emissions and from tailing solutions (Spitz & Trudinger, 2009).

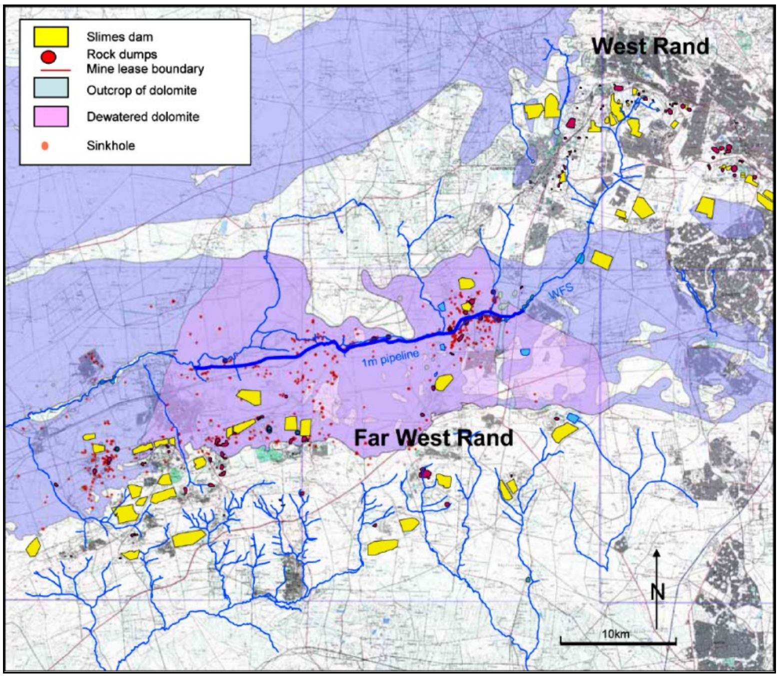


Figure 1-2: Distribution of slimes dams, rock dumps and sinkholes in relation to the WFS and sinkholes in the West Rand and Far West Rand goldfields (Winde, 2010)

The metals that are found in rocks generally occur in the form of insoluble minerals such as silicates and sulphides. Once these minerals are exposed to harsh atmospheric processes they decompose slowly and the metals are liberated. The liberated metals are dissolved in surface and groundwater and become bio-available (Spitz & Trudinger, 2009). The uraniferous gold ores in the WFS catchment also contains significant levels of pyrite, which is oxidised in the presence of oxygen and water to produce sulphuric acid or Acid Mine Drainage (AMD) as it is more commonly known. AMD solubilises heavy metals from the waste rock and these leached metals enter the surface and ground water systems (Rios *et al*., 2007).

The leaching of metals depends strongly on metal solubility which is largely pH-depended. An increase in acidity results in an increase in free metal ion concentration in solution. Therefore, for most metals, the solubility increases with a decrease in pH value (Spitz & Trudinger, 2009). However, there are exceptions to this such as uranium. Different uranium species show different solubility at various pH ranges as seen in Figure 1-3. In aqueous solutions, uranium exists as uranyl ions and readily forms complexes with carbonates and hydroxides. In the pH range of most natural water sources, uranyl ions primarily form complexes with carbonates. At pH values between 5 and 6.5, the primary species is $UO_2CO_3^0$. At pH values between 6.5 and 7.5, the primary species is $UO_2(CO)_2^{2-}$. Small amounts of uranyl hydroxide complexes such as, UO_2OH^+ and $(UO_2)_3(OH)_5^+$ are respectively formed at pH values between 4 - 6 and 6.5 - 9 (Crittenden *et al*., 2005).

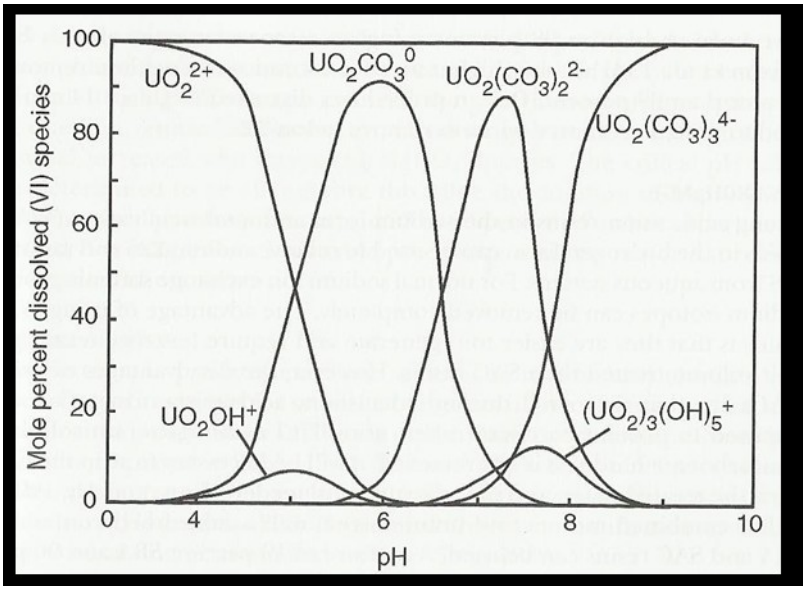


Figure 1-3: Distribution of uranium species in water (Crittenden *et al*., 2005)

Although metal solubility is mostly pH –dependent, it can also be influenced by redox potential, occurrence of organic and inorganic complexing agents, the presence of electron donors and acceptors etc. Most metals show optimum solubility in water typified with a low pH values. Solubility characteristics vary widely due to the extensive variety of physical and chemical states that metals exist in (Spitz & Trudinger, 2009).

The WFS catchment is not solely affected by mining activities as it has 21 discharge points and numerous non-point discharges from mines (gold, uranium and peat), sewage works, settlements (formal and informal), industry and agriculture (Coetzee *et al*., 2004).

Pollutants that arise from a specific site and can be traced to a particular source such as a waste water treatment plant or industrial site are defined as point source pollutants (Sparks, 2003). Groundwater is most often polluted by point source pollution (Crittenden *et al*., 2005). Non-point source pollutants enter the aquatic environment over a broad area and not from any one source and include both human and natural activities such as agriculture, mining activities, forestry, construction and atmospheric deposition (Sparks, 2003). Surface water is usually subjected to pollution arising from point and non point sources (Crittenden *et al*., 2005).

Once heavy metals enter the WFS they can be adsorbed onto the streambed sediment which acts as a sink for these metals (Stackelberg 1997; Munn & Gruber, 1997). The sorption of a metal is dependent on pH, sorptive concentration, surface coverage and the sorbent type, with pH paying a major role once again. Maximum sorption for most metals occurs as soon as the water pH increases and reaches alkaline conditions as can be seen in Figure 1-4. Metal sorption generally occurs on the surface of secondary soil minerals such as clay minerals (Phyllosillicates), oxides, hydroxides and oxyhydroxides, due to high surface areas, surface functional groups and a constant surface charge (Sparks, 2003).

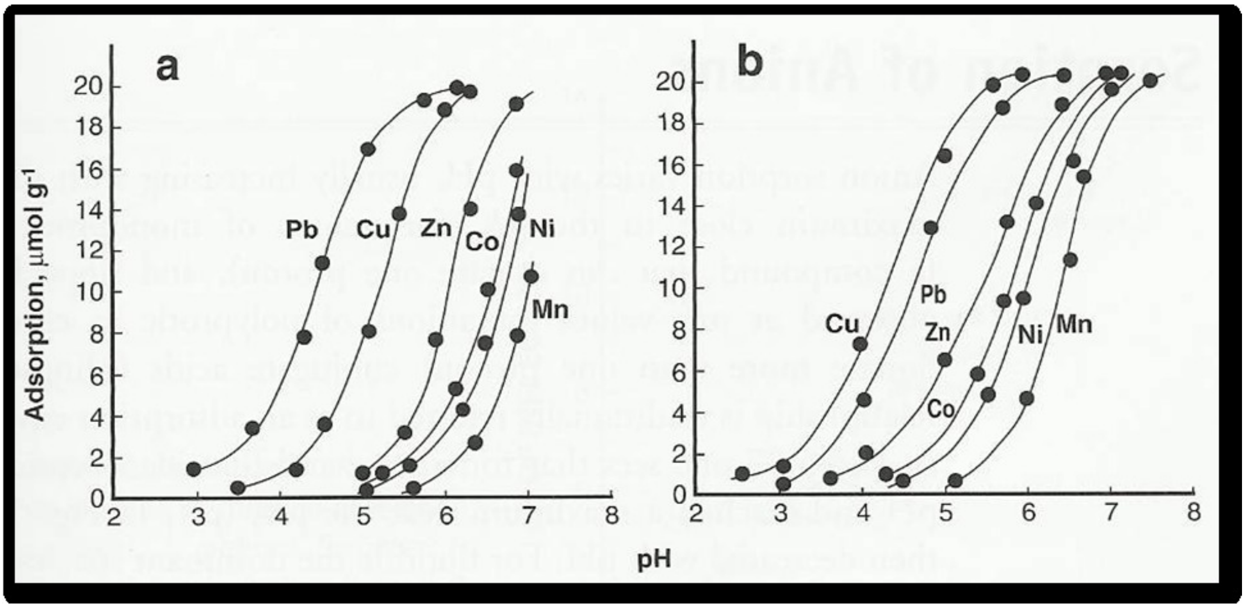


Figure 1-4: Sorption of a range of metals on (a) hematite and (b) goethite when they were added at a rate of 20 µmol g-1 of adsorbate (Sparks, 2003).

Hematite and goethite are both iron oxides and both form part of the inorganic surface functional groups in soils and sediment. Surface functional groups play a significant role in the adsorption process and for this reason hematite and goethite are excellent examples to demonstrate adsorption of metals (Sparks, 2003). The surface water in the WFS is mostly alkaline due to the buffering capacity of the dolomite it flows over and possible lime treatment done by mining companies in the area. Dolomite is a variety of limestone that consists of carbonates such as magnesium carbonate and calcium carbonate (Swart, 2002). These carbonates have the ability to buffer AMD from mine workings and acid seepage from slimes dams that will reach the water systems (Coetzee *et al.*, 2004).This creates ideal conditions for metal sorption onto secondary soil minerals in the WFS streambed sediments (Sparks, 2003).

2 Perspective and Outline of Thesis

2.1 Problem Statement

Although the WFS has been the subject of various reports, projects and articles, the focus was mostly placed on the contamination of water and sediments within the stream and the potentially associated risk posed to downstream users, mostly humans. Most of these investigations are not published and have limited circulation.

Municipalities within the catchment have erected warning sigh along the WFS, as can be seen in Picture 2-1.

WARNING/WAARSKUWING/ISILUMKISO/KITSISO

The water in the Wonderfonteinspruit is unsafe for human consumption.
Please do not drink the water or use it for cooking purposes.
Thank you!

Die water van die Wonderfonteinspruit is nie veilig vir menslike gebruik nie.
Moet dit asseblief nie drink of vir kookdoeleindes gebruik nie.
Dankie!

Amanzi ase Wonderfontein awalunganga ukuselwa anobungozi angasetyenziswa ngabantu angaselwa okanye kungaphekwa ngawo.
siya bonga!

Metsi a molapo wa Wonderfonteinspruit ga a lokelwa go nwewa go a dirisa go apea gonne a nale ditwatsi.
Re a leboga!

Picture 2-1: Warning signs that have been erected at various sites along the Wonderfonteinspruit

The notice clearly states that the water in the WFS is not to be used for human consumption, but there is no mention of utilizing the water for livestock watering or irrigation purposes. There are very few farms in the catchment that still irrigate from the WFS, but almost all farmers with property along the WFS use it for livestock watering purposes. There are also some individuals from informal settlements along the WFS that use the water to irrigate their vegetables and to water their livestock.

This is a cause of growing concern as not much research has been done on this topic. It is therefore imperative that research has to be done on the transfer and accumulation of heavy metals from the WFS into the adjacent and surrounding environment. In order to establish whether or not the heavy metals are isolated to the WFS streambed or if they are transported beyond the streambed boundary and possibly pose a threat to animals and the human food chain. This subject could be the topic of numerous studies and therefore it is important to define the aim and objectives of such a study.

2.2 Aim and Objectives

Hypothesis: It is expected that toxic and trace elements do transfer and accumulate from the WFS into the surrounding environment and into the cattle grazing in the area.

The aim of this study was to detect and quantify toxic and trace elements that may transfer and accumulate from the WFS into the surrounding environment, such as soil, grass and cattle grazing in the area. Specific objectives of the investigation include:

- Identifying elements of interest
- Detecting and quantifying the accumulation of the identified elements of interest in water, sediment, soil, grass and cattle tissue samples
- Assess the water quality of the WFS for livestock watering and purposes.
- Identifying the major route of elemental ingestion by cattle
- Develop a tool to determine possible impacts of pollutants in the WFS and associated human health risks
- Quantification of impacts and making recommendations to reduce these impacts

2.3 Literature Review

Nature has dispersed all trace elements in rather equally within the earth’s crust albeit at different concentrations. Therefore, metals are naturally occurring elements and life has evolved in their presence (Spitz & Trudinger, 2009). The actual concentration of a particular element that is found in any soil sample depends primarily on the nature of the parent rock from which it was formed (Reilly, 1991). Human exploitation of metals and industrial activities have liberated, transformed and accumulated metals in some geographic areas and for some metals, these changes can be detected on a global scale (Spitz & Trudinger, 2009).

Trace elements include trace metals, heavy metals, metalloids, micronutrients, and trace inorganics are elements that are present at a level <0.1% in the lithosphere (Sparks, 2003). There are about eighty elements of the Periodic table that can be classified as metals (Reilly, 1991). Heavy metals are amongst these metals and are environmentally of most concern (Spitz & Trudinger, 2009).

The term “heavy metals” is sometimes used incorrectly to describe some potentially harmful elements that are metals or metalloids (Spitz & Trudinger, 2009). A better description is to define heavy metals as elements with densities greater than 5.0 g/cm^3, which include elements such as cadmium, chromium, cobalt, copper, lead, nickel, mercury and uranium (Sparks, 2003).

According to Duffs as quoted by (Newman, 2010) this classification of elements is unhelpful to toxicologists and chemists. Nieboer and Richardson consider this type of classification of metals to have substantial shortcomings (Newman, 2010). A more relevant approach would be to group elements according to their essentiality to the health and well-being of living organisms.

2.3.1 Essentially Beneficial Elements

Essentially beneficial elements are those compounds that need to be present in an organism’s diet to maintain normal physiological functions. The concentrations of essential elements must usually be maintained within quite narrow limits if the functional and structural integrity of the tissues are to be protected. Severe disabilities arise when dietary concentrations are low and active transport mechanisms are then invoked to ameliorate the deficiency. This includes calcium (Ca), phosphorus (P), magnesium (Mg), sodium (Na), chlorine (Cl),potassium (K), sulphur (S), cobalt (Co), copper (Cu), iodine (I), iron (Fe), manganese (Mn), selenium (Se) and zinc (Zn) (Underwood & Suttle, 2001). The following elements are of interest to this study and will be discussed briefly:

Cobalt

Cobalt is a hard, magnetic, silver-gray metal that is also a by-product of non-ferrous metal mining. Cobalt levels that are above desirable levels may adversely affect heart and lung function. Cobalt is present in vitamin B12 and plays a biological role in N2-fixation (Spitz & Trudinger, 2009).

Copper

Copper was one of the first metals known to mankind. This versatile and durable metal appears everywhere in our everyday lives. It can be found in cytochrome and hemocyanin and in cellular molecules that are involved in respiration. Large doses may induce vomiting, nausea, diarrhoea, cramps or hepatic damage. It is toxic to fish and aquatic life at low levels (Spitz & Trudinger, 2009).

Selenium

Selenium is a naturally occurring, solid element that is widely distributed in the earth's crust and it’s commonly found in rocks and soil (ATSDR, 2003). Selenium is an essential element in the animal body for effective metabolism and health reasons. It can also have toxic effects if ingested in amounts exceeding the body’s metabolic requirements for extended periods (Underwood and Suttle, 2001). It can behave similarly to arsenic and can be produces as a by-product of gold, copper and nickel mining (Newman, 2010).

Zinc

Zinc, a blue-gray metal is the fourth most commonly used metal. It is found in sulphide ores in combination with copper, silver and lead. It is essential in several enzymes that catalyze the metabolism of proteins and nucleic acids. Zinc may affect water taste at high levels. It can cause irritation of the digestive system and is toxic to some plants and fish (Spitz & Trudinger, 2009).

2.3.2 Occasionally Beneficial Elements

Occasionally beneficial elements are argued to also be essential, albeit in ‘ultra trace’ concentrations and include boron (B), chromium (Cr), lithium (Li), molybdenum (Mo), nickel (Ni), silicon (Si), tin (Sn) and vanadium (V) (Underwood & Suttle, 2001). The following element is of interest to this study and will be discussed briefly:

Nickel

Nickel is a hard grey, magnetic metal that is extremely resistant to corrosion. It is an essential element but some nickel compounds can be toxic and carcinogenic. It occurs in sulphide ores and in laterites in association with iron and magnesium (Spitz & Trudinger, 2009).

2.3.3 Essentially Toxic Elements

Essentially toxic elements are those elements which are renowned for their toxicity and include aluminium (Al), arsenic (As), cadmium (Cd), lead (Pb), mercury (Hg) and uranium (U) (Underwood & Suttle, 2001). The following elements are of interest to this study and will be discussed briefly:

Cadmium

Cadmium is emitted to soil, water, and air by non-ferrous metal mining and refining e.g. gold, zinc, lead and copper mining, and is not usually found in water at concentrations greater than 1µg/L (ATSDR, 2008). It is recognized to produce toxic effects in humans and may be linked to renal arterial hypertension and violent nausea (Spitz & Trudinger, 2009). Most of the cadmium that enters the body enters the kidneys and liver and can remain there for many years. A small portion of the ingested cadmium is excreted in the urine and faeces (ATSDR, 2008).

Lead

Lead is a bluish-grey metal and has no taste or smell. Lead in the environment is known to be toxic to microorganisms, plants and animals. It is readily formed from the reduction of Galena. Lead is a cumulative body toxin and has been shown to affect virtually every organ and body system in humans and animals (Spitz & Trudinger, 2009). It may cause anaemia and neurological dysfunction with chronic exposures (Newman, 2010).

Uranium

Uranium is radioactive and the heaviest naturally occurring element that is found in varying but small amounts in soil, rocks, water, plants and animals (Barillet *et al*., 2006). This silver-gray metal is chemically reactive and weakly radioactive, and is therefore its toxicity is of more concern than its radioactivity. There are three isotopes: U-234, U-235: and U-238. U-238 is the most common isotope, accounting for more than 99% of all natural occurring uranium. It is chemically reactive and oxidizes readily (Spitz & Trudinger, 2009). Less than 0.1–6 % of the uranium is absorbed by the digestive tract, depending on the solubility of the uranium compound. The highest levels of uranium are found in the bone, liver, and kidney. It takes 11 days for half of the uranium to leave the bones, and 2-6 days for the kidneys. Most of the absorbed uranium is excreted via the urine (ATSDR, 2011).

2.3.4 Trace Elements in Organisms

Organisms have developed numerous mechanisms for the uptake, and excretion, regulation and detoxification of both essential and non-essential elements (Spitz & Trudinger, 2009). Therefore, all vegetation species and animal tissues contain inorganic trace elements in widely varying concentrations. Most of the elements that plants require for normal growth are drawn from the soil, (Reilly, 1991). Plants have a limited capacity for the selective uptake of essentially beneficial elements. They are also able to take up and accumulate, sometimes to very high levels, certain elements which are not necessary for growth and which may even be toxic (Marschner, 1997). Due to the great genetic variability that is found among populations of plants, there will be individuals that have the ability to survive and even prosper in highly contaminated soils. These plants may not be as vigorous as if they were growing in normal uncontaminated soils, but they will survive and develop, as they don't have to compete for space and nutrients with other plants (Reilly, 1991). Climate, season, genetics, concentration and availability in soil and stage of maturity are all factors that influence metal uptake by plants.

Trace elements exist in the animal body's cells and tissues in a variety of functional and chemical combinations (Underwood & Suttle, 2001). Soil, water and vegetation are the primary sources of all elements found in animal tissues. Livestock usually derive a high proportion of their trace elements from the feed and vegetation that they consume (Underwood & Suttle, 2001). The typical dry matter intake for beef cattle ranges between 2.5 -3% of body mass, depending on the quality of the pasture. A mature beef cow weighing 450 kg, will therefore consume 11.25 – 13.5 kg of grass per day and has an estimated daily requirement of 36-41 litres of water (Dickinson *et al*., 2007). Drinking-water is not normally a major source of minerals to livestock due to the relatively low concentrations (Underwood & Suttle, 2001).

Soil ingestion can be accidental or a result of mineral seeking behaviour by the cattle called geophagia. The contamination of vegetation by soil and dust will increase with high grazing intensities or low pasture availability (Underwood & Suttle, 2001). Studies indicate that cattle ingest 0.5 – 0.9 kg of soil per day (Mayland *et al*., 1997) and (Mayland *et al*., 1975).

Other sources of trace elements include atmospheric inputs, fertilizers used on the planted pastures and mineral supplements that are provided to cattle in the winter months.

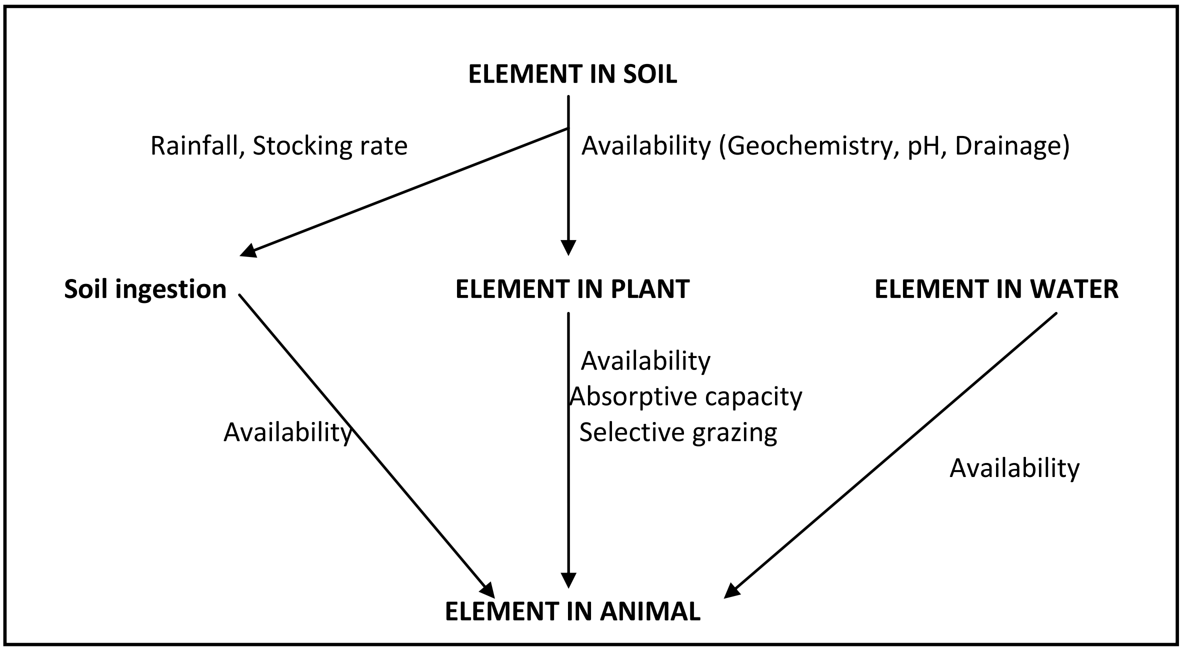


Figure 2-1: Summary of the many and varied factors which can influence the flow of an element from the soil, water and vegetation to the grazing animal.

Figure 2-1 is a modified version of a figure available in (Underwood & Suttle, 2001). It illustrates the different pathways and factors that influence the flow of elements from sources into cattle.

Normal animal health and performance is established by maintaining the concentration of functional forms of trace elements within narrow ranges. The animal body has developed strategies to deal with excess and toxic elements by means of adsorption, excretion and tissue deposition of these elements (Miller, 1979).

2.4 Limitations related to Field Work and Assessment

The scope of this study is restricted to analysis of inorganic metals in various sampled mediums. It will therefore not consider any radiological (except U-238), bacteriological or organic pollutants.

When referring to uranium, only the U-238 isotope is considered as it is the only isotope that the ICP-Ms instrument is able to detect. Only the U -238 isotope will be referred to as uranium in this study. The U-238 isotope is the most prevalent isotope, Making up about 99% of natural uranium.

Although mercury (Hg) pollution is often associated with gold mining activities, it will not be part of this study due to the inability of the ICP-Ms instrument to accurately quantify the mercury concentrations.

This study only considers the total element concentration and does not examine the biological availability of the elements. Furthermore, the study does not examine the effects of the elements on biota, but only quantifies the accumulation of various elements in cattle, grass, soil, sediments and water samples.

For the purpose of this study, the sources of elements will be restricted to those cattle can ingest orally.

2.5 Outline of Thesis

This thesis is divided into seven chapters including:

- Chapter 1 is the introduction to this study, which includes historical background of the Wonderfonteinspruit.
- Chapter 2 includes the Problem statement, aim and objectives, literature review and limitations related to field work and assessment.
- Chapter 3 discusses the materials and methods that were used in this study.
- Chapter 4 provides a summary of the study area, summary of the receiving environment and site descriptions.
- Chapter 5 is a general discussion of the sediment, water, soil, grass and cattle results.
- Chapter 6 includes data interpretation to reveal elemental transfer factors for each respective site, the major route of elemental ingestion and the cattle consumption model.
- Chapter 7 presents the conclusion and recommendations.

3 Materials and Methods

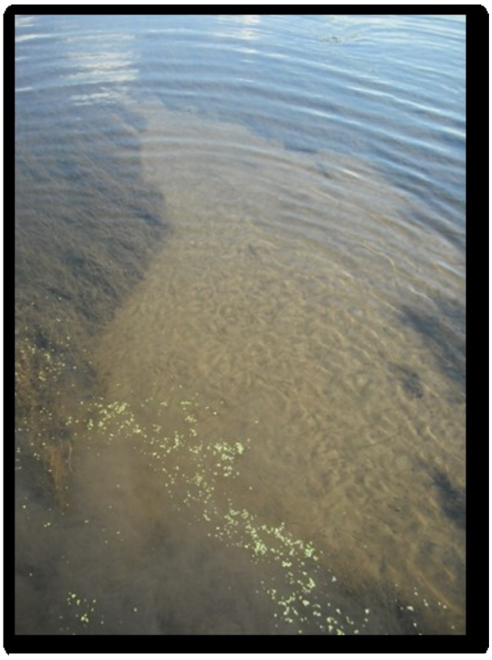
3.1 Sampling Procedure

Water, sediment, soil, grass and cattle tissue samples were collected on two farms, the one farm was located in the Lower-Wonderfonteinspruit area and will serve as the experimental group, the other farm was located along the Mooi River, between Klerkskraal Dam and Boskop Dam and will serve as the control group as it has not been impacted my mining activities. All samples were collected by the author to prevent contamination, mislabelling and to ensure that the correct samples were collected which proved to be quite a challenge at the abattoir.

3.1.1 Sediment Sampling

A grab-sampler and a shovel were used to collect approximately 500 g samples of sediment at an approximate depth of 10 - 20 cm. In the experimental group samples were collected in a dam with a water depth ranging from 30 - 100 cm. During sampling it was evident that a distinction could be made between light coloured sand banks and dark coloured compacted fine grained material. The samples in the control group were very homogenous and collected in areas with slow moving water at a depth ranging from 120 – 160 cm.

Picture 3-2: Sediment sample from the Wonderfonteinspruit (dark coloured sediment)



Picture 3-1: A sand bank in the Wonderfonteinspruit (light coloured sediment)

In order to identify elements of interest sediment samples from the experimental and control groups were collected, dried, analyses and compared. Eight light coloured sediment samples and eight dark coloured sediment samples were collected from the experimental group and five sediment samples from the control group were collected. After the elements of interest were identified, it was deemed necessary to establish the elemental concentrations and mineralogical composition within the different particle size fractions (sand, silt and clay fractions) of the sediment samples. For this reason, four sediment samples were collected from the experimental group and one sample was collected from the control group.

3.1.2 Water Sampling

Approximately 250 mL water was sampled at four sites along a 1 km stretch of both the Wonderfonteinspruit and Mooi River. The water samples were collected in the middle of the streams, as shown in Figure 3-1 at a minimum depth of 20 cm. Thirty five water samples were collected in the experimental group and fifteen water samples were collected in the control group.

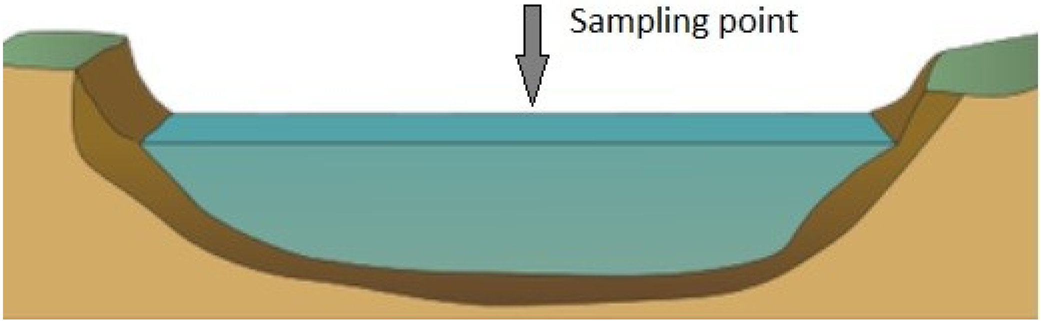


Figure 3-1: Cross section of the streambed

Disturbed water

During the water sampling process on the experimental farm, it was observed that cattle actually wander into the stream and dam to drink. This stirs up the sediment and the water becomes turbid due to the presence of suspended particles. In order to establish the true elemental ingestion by cattle from the water in the Wonderfonteinspruit, this “disturbed water” had to be sampled and analysed. This was done by wading into the Wonderfonteinspruit with gumboots, and collecting five 500 mL water samples at different site.

3.1.3 Soil Sampling

A ground auger was used to collect five 500 g soil samples at each of the designated sites within the control and experimental groups. These soil samples were collected at an approximate depth of 20 cm and cleared from organic matter.

3.1.4 Vegetation Sampling

The dominant grass species at each of the respective sites were collected for analysis. Sampling took place during the growing season and five samples weighing approximately 100 g was collected at each of the sites. The grass samples were then dried at 45 °C for two day before being digested and analyzed by means of ICP-MS to determine the elemental concentrations of each sample.

3.1.5 Cattle Tissue Sampling

Kidneys, liver, spleen, muscle tissue (meat), bone and hair samples were collected from non-pregnant, non-lactating cows that were of the same breed and age. Ten cattle from the experimental group and five cattle from the control group were slaughtered at a local abattoir and approximately 100 g samples were collected from each tissue type. The collected samples were cut up into fine pieces, frozen at -75°C for three days before being dried in a freeze dried for four days. The dry samples were then digested and analyzed by means of ICP-MS to determine the elemental concentrations of each sample. The cattle from both groups were Brahman crossbred cows that were all older than 5 years.

3.1.6 Mineral Supplements and Fertilizer Sampling

The fertilizer that was used on the irrigated grass on the experimental farm was collected, dried in an oven set to 45 °C for two days digested and analyzed by means of ICP-MS to determine the elemental concentrations. The mineral supplement provided to cattle was also collected, dried digested and analyzed in a similar fashion.

3.2 Sample Preparation

3.2.1 Chemical Analysis

The pH value of all the water samples was lowered to 3, in order to dissolve the suspended solids, before being analysed for element concentrations by means of inductive plasma coupled spectroscopy (ICP-MS).

The rest of the sampled items – the grass, cattle tissue, sediments and soil samples – were prepped separately, using the EPA 3050B method, as required to enable analysis with an ICP-MS (Agilent 7500, Chemetrix, 2003).

U.S. EPA method 3050B - Total Sorbed Metals (U.S. EPA, 2012)

1. Two grams of each sample was weighed of into a 150 mL beaker

2. Fifteen millilitres of concentrated HNO_3 was added and the beaker was covered with watch glass. The beaker was then placed on a hot plate and brought to 95 °C for least one hour. As soon as the fuming started to decrease and the samples appeared to be completely digested the watch glass was removed in order to permit greater evaporation.

3. Heating was continued until the sample volume was reduced to 5 mL, where after the sample was removed and allowed to cool down to room temperature.

4. Three millilitres of 30 % H_2O_2 was added after the sample reached room temperature.

5. Ten millilitres 3N HCL was then added and the beaker was once again covered with the watch glass.

6. The beaker was once again placed on the hotplate and brought to 95 °C for least one hour.

7. After the hour passed the beaker was removed from the hotplate and allowed to cool down to room temperature.

8. The dissolved sample was then filtered through a Whatman 40 paper into 50 mL volumetric flask and the volume of the sample was brought to 50 mL with deionised water.

3.3 Particle Size Distribution of Sediment and Soil Samples

3.3.1 The Hydrometer and Calgon Method (Laker & Du Preez, 1982) and (Standards Association of Australia, 1976)

Soil consists of an assembly of ultimate soil particles (discrete particles) of various shapes and sizes. The object of a particle size analysis is to group these particles into separate ranges of sizes and to determine the relative proportion by weight of each size range. The method employs sieving and sedimentation of a soil/water/dispersant suspension to separate the particles. The sedimentation technique is based on an application of Stokes' law to a soil/water suspension and periodic measurement of the density of the suspension.

3.3.1.1 Equipment

- Soil hydrometer (ASTM 152H hydrometer).
- Sedimentation cylinders with internal depth of 340 ± 20 mm and capacity of1 L.
- 0.20, 2.00 and 4.75 mm sieves.
- Interval timer.
- End-over-end shaker, rotating at 15 rpm.

3.3.1.2 Reagents

- Calgon (25% sodium hexametaphosphate).
- Two hundred and fifty grams of Calgon was dissolved into 900 mL of warm deionised water. After the solution cooled down to room temperature, sodium carbonate was added to bring to pH value to 8.
- Deionised water was then added to dilute the solution until a volume of 1L was reached.

3.3.1.3 Sample Preparation

The samples were weighed and passed through the 4.75 mm and 2 mm sieves making sure that no aggregates are retained on the sieves. The amounts of particles >4.75 mm and 2.0 to 4.75 mm were weighed and recorded. After this, 50 g of the air dried soil (<2 mm) was placed into a shaking bottle and 200 mL of deionised water and 20 mL of the Caolgon mixture was added. The shaking bottle was then placed on an end-over-end shaker and shook for 16 hours (overnight) at 15 rpm.

The hydrometer

For all readings, the hydrometer was placed in the suspensions for 20 seconds before each reading. The value of the effective depth (L) for each hydrometer and the sedimentation cylinder, in which it was used in, was calculated as follows:

$$L = L1 + 0.5(L2 - Vb/A)$$

Where:

L = Effective depth (mm)

L1 = Distance along the stem of the hydrometer from the top of the bulb to the mark for a hydrometer reading (mm)

L2 = Overall length of the hydrometer bulb (mm)

Vb = Volume of hydrometer bulb (cm^3)

A = Cross-sectional area of sedimentation cylinder (cm^2)

The volume of the hydrometer bulb (Vb) was measured by the rise in level of water in a 250 mL measuring cylinder, which was filled to the 150 mL mark. An effective depth (L) was calculated for each of the major calibration marks from 60 to –5 g/L on each hydrometer.

3.3.1.4 Sedimentation

1. Once the shaking was completed, the samples was transferred from the shaking battle into a 1 L measuring cylinders, which was then filled with deionised water to the 1 L mark.

2. The sample was then stirred with a plunger for 20–30 seconds to ensure that all material at the bottom was brought into suspension. At the end of stirring, the plunger was removed and the interval timer was started.

3. After 4 minutes of sedimentation, the hydrometer was immerged to a depth slightly below its floating position and allowed to float freely. A reading at the top of the meniscus was recorded after 5 minutes.

4. The hydrometer was then removed, rinse cleaned and placed in a sedimentation cylinder filled with deionised water and 20 mL of Calgon (blank solution), which was kept at the same temperature as that of the soil suspension. A hydrometer and temperature reading was recorded in the blank solution at the same time as the recording of the soil suspension.

5. The hydrometer was then re-inserted in the soil suspension to record readings at periods of 30, 93 and 420 minutes, taken in the same manner as above.

6. The hydrometer was left in the blank solution between readings.

3.3.1.5 Sand measurement

1. The soil suspension was then passed through the 0.20 mm sieve and thoroughly washed free of all fine particles. Those particles retained on the sieve were the coarse sand fraction.

2. The coarse sand fraction was then transferred into a weighing tin and dried in an oven between 105 °C and 110 °C until it was dried. After the sample was dried it was left to cool before weighing it.

3.3.1.6 Calculations

The summation percentage (P) for each hydrometer reading is calculated as follows:

$$P(\%) = \frac{(H - B)\text{x}\,100}{W}$$

Where:

H = Hydrometer reading in soil suspension (g/L)

B = Hydrometer reading in blank solution (g/L)

For the coarse sand fraction:

$$Coarse\ sand(\%) = \frac{J\ \text{x}\ 100}{W}$$

Where:

J = Oven-dry (OD) weight of sand from sieve (g)

Where on a whole soil basis:

$$W = \frac{ODW\ x\ (G + ODWF)}{ODW}$$

Where:

ODW = OD weight of fine earth (<2.0 mm) used in hydrometer analysis (g)

J = OD weight of gravel (>2.0 mm) from sieve (g)

ODWF = OD weight of total fine earth (<2.0 mm) (g)

G = Weight of gravel (>2.0 mm) from sieve (g)

For the gravel fraction:

$$Gravel(\%) = \frac{G\ \text{x}\ 100}{ODWF}$$

Where:

ODWF = OD weight of total fine earth (<2.0 mm) (g)

G = Weight of gravel (>2.0 mm) from sieve (g)

Calculate the particle size (D) at different times from the relationship:

$$D\ (mm) = 0.315x\ K\ x\ \sqrt{L}/T$$

Where:

K = Sedimentation constant, which varies with temperature and particle density

L = Tabulated effective length for the hydrometer reading (mm)

T = Time (minutes)

If the particle density of the soil was known, that value could have been used in the K table. As this was not the case, a given value of 2.65 Mg/m^3 was used instead.

The cumulative percentage (P) versus particle size diameter (D) was plotted on a semi-logarithmic, 4 cycle graph paper. The curve was used to interpolate the percentages for:

- Clay = <0.002 mm
- Silt = 0.002–0.02 mm
- Fine sand = 0.02–0.20 mm (See Note 4.)
- Coarse sand = 0.20–2.0 mm
- Gravel = >2.0 mm

3.3.2 Manual Method (ASTM Committee E-29, 1985) and (Chepil, 1961)

After the initial particle size distribution analysis was completed; similar soil and sediment samples were collected and separated into sand, silt and clay samples by drying, grinding and sieving without the addition of any chemicals. This was done in order to determine the elemental concentrations within each particle size fraction of every sample. Although the accuracy of the particle size distribution may have been slightly compromised, it was essential to ensure the accuracy of the elemental concentrations of each fraction.

3.3.3 Soil Texture Classes (Soil Classification Working Group, 1991)

The soil texture classes were determined after the particle size distribution of the sand, silt and clay fractions was completed. This was done by using a soil texture chart as seen in Figure 3-2.

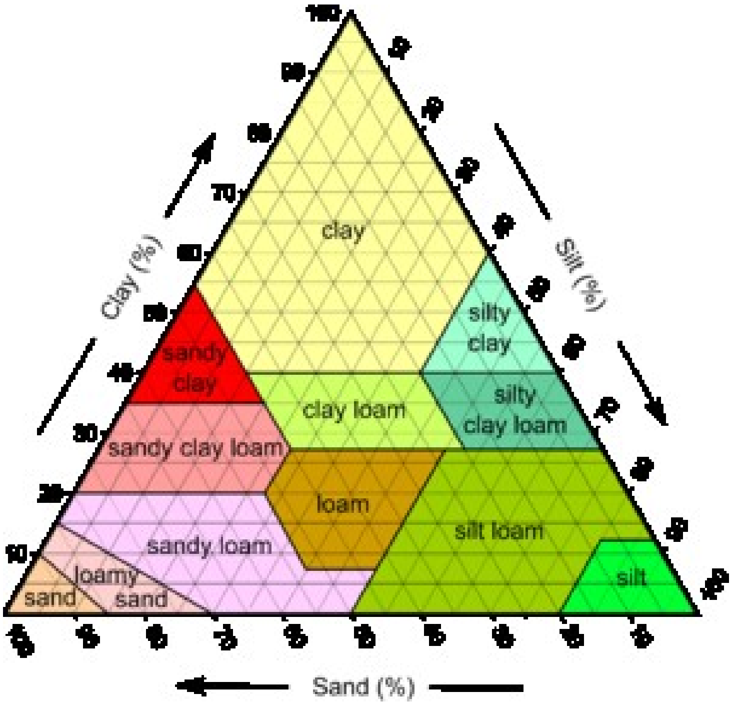


Figure 3-2: Soil texture chart

3.4 Soil Classification (Soil Classification Working Group, 1991) and (The Non-Affiliated Soil Analysis Work Committee, 1990)

Soil classification was conducted by making use of the South African Standard for Soil Testing Methods and a book -Soil Classification – a Taxonomic System for South Africa. The 73 soil forms which are found in the South African classification can be placed into 14 groups which are identified by using an eliminative key based on the presence of defined diagnostic horizons or materials.

3.5 Sediment XRD Analysis

General Description

In this study, XRD analysis was done on sediment samples to determine the structural characteristics of the materials**.** X-Ray powder Diffraction (XRD) analysis is a powerful method by which X-Rays of a known wavelength are passed through a sample to be identified in order to identify the crystal structure. The wave nature of the X-Rays occurs due to diffraction by the lattice of the crystal to give a unique pattern of peaks of 'reflections' at differing angles and of different intensity, just as light can be diffracted by a grating of suitably spaced lines. The diffracted beams from atoms in successive planes cancel unless they are in phase.

XRD investigates crystalline material structure, including atomic arrangement, crystallite size, and imperfections. In this study, XRD analysis was done on sediment samples to determine the structural characteristics of the materials.

Samples were sent to the NWU's XRD laboratory which uses the following equipment:

Equipment
XRD machine name: Röntgen difraktometer system
Model: PW3040/60 X'Pert Pro
Complies with safety standards: EN61010/IEC1010/CSA C22.2 No.1010
Safety requirements for electrical equipment for measurement, control and laboratory use.
Radiation level: less than 1µSv/hour at 10 cm from the outside surface.

XRD Ceramic tube used:
A Cu-tube (PW – 3373/10) used at 40kV and 45mA.

Computer software used:

- PDF – 4 + 2007 Relational Database. International centre for Diffraction Data, 12 Campus Boulevard, Newtown Square, Pennsylvania 19073-3273, USA.
- X'Pert High Score Plus PW3212, Second Edition, January 2004. PANalytical, The Netherlands.
- X'Pert Data Collector, Sixth Edition, January 2006. PANalytical, The Netherlands.

3.6 Vegetation Identification (Oudsthoorn, F. 2006)

The dominant grass species that were found in each of the respective sites were identified by inspecting the leaves, stems and inflorescences (seed heads) of each sample and comparing the finding to species found in the following book: Guide to Grasses of South Africa 2006.

3.7 Soil-to-plant Transfer Factors

This ratio describes the amount of elements expected to enter a plant from its substrate, under equilibrium conditions (Sheppard and Sheppard, 1985).

The Transfer Factor (*TF*) was expressed as:

$$TF = \frac{Element\ concentration\ of\ plant\ (mg/kg\ dry\ weight)}{Element\ concentration\ of\ soil\ (mg/kg\ dry\ weight)}$$

Factors such as soil characteristics, climatic conditions, type of plants, part of the plant concerned, physico–chemical form of the elements and the effect of the competitive species can influence the *TF* values (Bettencourt *et al*., 1988).

3.8 Statistical Analysis

All statistical analyses were done with Microsoft Excel 2007, and Matlab version 7.1. EXCEL was used to determine the variation, deviations, mean values and averages of the elemental concentrations in the specific areas.

As the aim of this study was to establish if the elemental concentrations found in samples from the experimental site differs from the control site.

There are two methods to establish if the difference would be significant or not:

Statistical significance

This method is used to determine whether or not the differences between two samples (a control sample and en experimental sample) are statistically significant. This is done by calculating the p-value and determining if the hypothesis that the respective mean values of the samples are equal was found to be true. If the p-value is smaller than some specified significance level, the hypothesis is rejected and the difference between the mean values of the respective samples is then considered to be statistically significant (Steyn & Swanepoel, 2007).

The problem with this type of test is that it only proves that the means of the two groups under consideration differ, it does not indicate the size of the difference. Another problem is that the test is more likely to detect differences with a larger number of samples (Steyn & Swanepoel, 2007).

Practical significance

The effect size is used to detect the presence of a practically significant difference between the mean of two samples (Steyn & Swanepoel, 2007). Effect size is calculated as:

$$Effect\ size = \frac{|\text{Experimental Average} - \text{Control Average}|}{\text{Control Standard Deviation}}$$

After calculating the effect size, the result has to be interpreted in light with the project data. In this study the interpretation as documented in Table 3-1 was used.

Table 3-1: Interpretation of effect size

Effect size:	Interpretation
0 to 0.2	Small effect
0.2 to 0.8	Medium effect
larger than 0.8	Large effect

The difference between the two sample means was found to be practically significant when the effect size was interpreted as large (Steyn & Swanepoel, 2007).

3.9 Graphics

The Map of the Wonderfonteinspruit (Figure 1-1) and the Geology map (Figure 4-1) were created by using Arcgis 10. The other maps were obtained from Google earth.

4 Study Area

4.1 Site Description

Two areas were selected for this project – that represent an experimental and control group respectively. Both groups were located in the Wonderfonteinspruit Catchment and fall within the same quaternary catchment (C23G). The two groups share similar geology, which is Dolomite, subordinate chert, minor carbonaceous shale, limestone and quartzite as illustrated in Figure 4-1 . The two groups also share similar soil types as will be described later in this section.

The Wonderfonteinspruit flows directly through the experimental group which is considered to be a source of contamination to the environment.

4.1.1 The Experimental Group

The farm Blaauwbank is located in the Gauteng, South-Africa. The farm is situated approximately 45 km Northeast of Potchefstroom towards Carletonville. Reasons for selecting the site include:

- It is downstream of all the mining activities in the Wonderfonteinspruit Catchment. It therefore receives water from all mining activities in the catchment via canals or pipelines.
- The farmer follows good management practices and keeps records of reproductive rates, deaths and abnormalities.
- A good vaccination program is followed.
- No chicken litter is fed to the cattle. Chicken litter could be a source of metals such as Zn, Mn and Cu (Han *et al*., 2000).
- Cattle drink directly from the Wonderfonteinspruit.
- Cattle graze on natural pastures adjacent to the Wonderfonteinspruit and on planted pastures that are irrigated from the Wonderfonteinspruit.

4.1.2 The Control Group

The farms Muiskraal and Rooidraai would serve as the control group. They are located along the Mooi River, below Klerkskraal Dam and above the confluence with the Wonderfonteinspruit. There is no upstream gold or uranium mining activities, therefore all contaminants will result from the small town, agricultural activities and small scale diamond mining (DWAF, 2002).

4.2 Summary of the Receiving Environment

4.2.1 Rainfall and Temperature

Carletonville normally receives about 500 mm of rain per year as seen in Table 4-1, with most rainfall occurring mainly during mid-summer. It receives the lowest rainfall (0 mm) in June and the highest (111 mm) in January. The average midday temperatures for Carletonville range from 17°C in June to 28°C in January. The region is the coldest during June and July when the mercury drops to 2°C on average during the night.

Table 4-1: The average temperature and rainfall per month for Carletonville, which is close to both sites

Month	January	February	March	April	May	June
Average Max Temperature	28 °C	27 °C	25 °C	23 °C	20 °C	17 °C
Average Min Temperature	15 °C	15 °C	13 °C	9 °C	5 °C	2 °C
Average Rainfall	111 mm	42 mm	66 mm	30 mm	6 mm	0 mm
Month	**July**	**August**	**September**	**October**	**November**	**December**
Average Max Temperature	18 °C	20 °C	22 °C	23 °C	26 °C	25 °C
Average Min Temperature	2 °C	4 °C	8 °C	12 °C	13 °C	14 °C
Average Rainfall	6 mm	12 mm	15 mm	57 mm	63 mm	72 mm

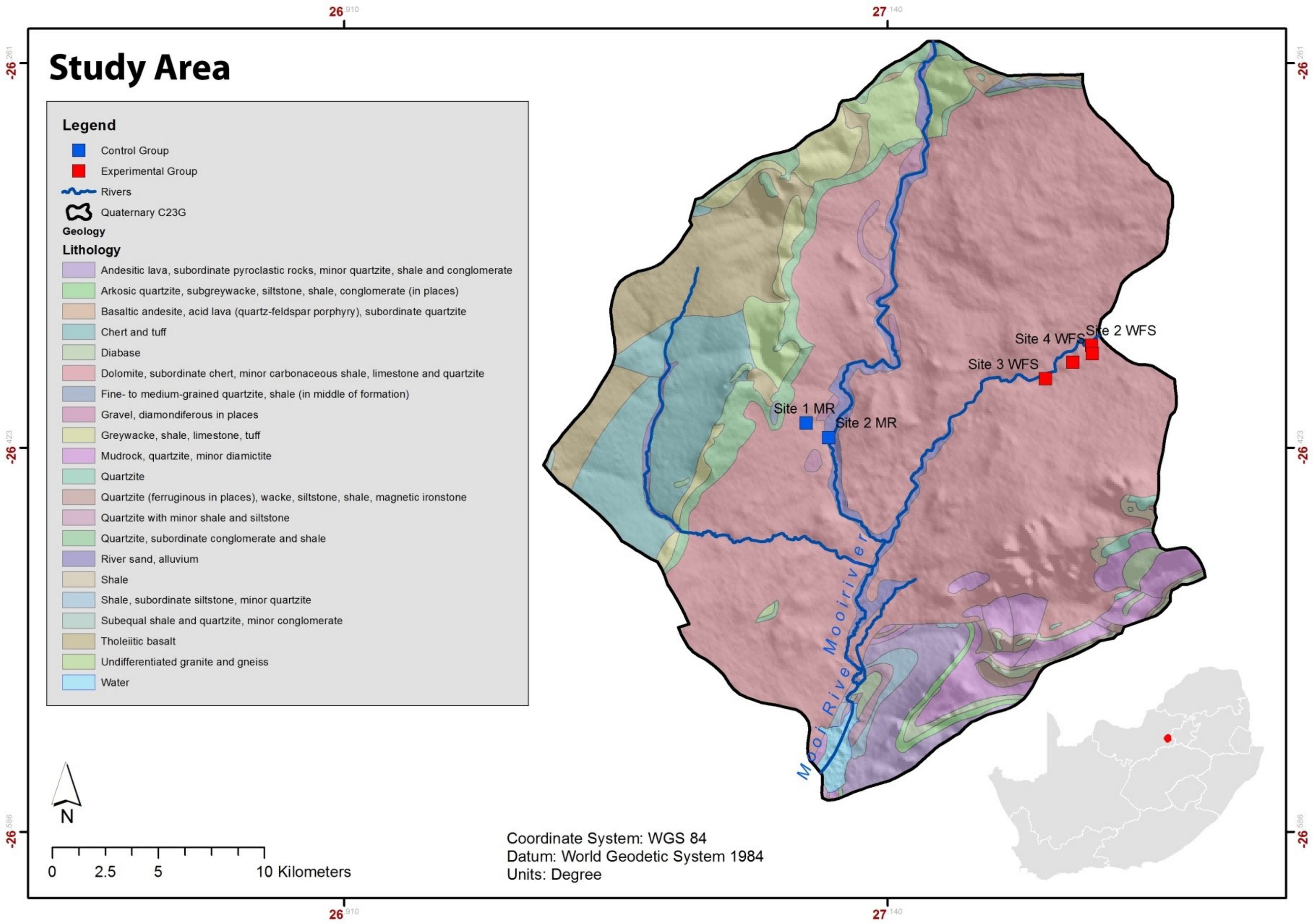


Figure 4-1: Geological map indicating the position of the experimental and control sites

4.2.2 Natural Vegetation

The experimental and control groups were characterized by Bankenveld Western Variation veld type and consist of tall tufted sour grassveld component with shrubs and trees found on rocky outcrops (Acocks, 1998). Furthermore both groups fall within the grassland biome, the fire climax grassland of potential savanna areas. This grassland generally occurs in areas with a lower or unreliable rainfall pattern. These areas have been greatly affected by past misuse and it is unlikely that the original grassland will ever replace these communities which are dominated by thatch grass (*Hyparrhenia hirta*). Growth is confined to September – April. Summer yields are limited due to drought and high evaporation and transpiration rates (Tainton, 1999).

4.3 Experimental Group Sites

Samples were collected at four different sites in the Experimental Group. The sites are displayed in Figure 4-2. Thirty five water samples (250 mL) were collected along a 1 km stretch of the Wonderfonteinspruit.

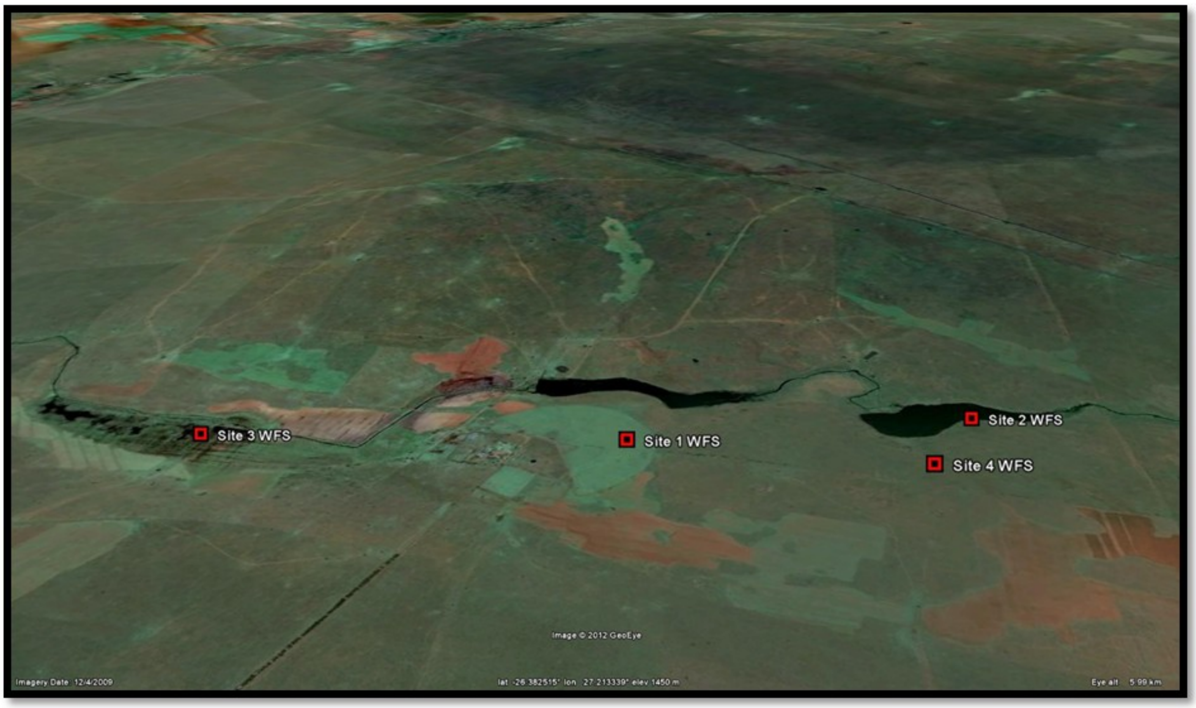


Figure 4-2: Google earth aerial map of the Experimental Group Imagery date: 12/10/2009

4.3.1 Site 1 WFS

Site 1 WFS (Picture 4-1) consisted of approximately 18 ha of planted Cocksfoot Grass (*Dactylus glomerata*) that is irrigated directly from the Wonderfonteinspruit. It is located approximately 150 m to the south of the Wonderfonteinspruit. Five soil (500 g) and grass (100 g) samples were collected at this site. Cocksfoot grass is a robust, tufted, perennial grass which grows best in spring, summer and autumn. The soils are classified in Table 4-2. Cold winter months are marked with slow growth (Dickinson *et al*., 2007). The pasture is fertilized annually with an application of nitrogen. Cattle do not graze this pasture continuously as they are rotated onto natural vegetation as well.

Picture 4-1: Irrigation of planted pastures in the Experimental Group

Table 4-2: Site 1 WFS soil classification, texture class and location

Soil Classification	
Soil form	HUTTON
Soil group:	Oxidic, red apedal
Soil Texture Class	Silt loam
Location	
Latitude	-26.387249°
Longitude	27.218897°
Altitude	1448 m

4.3.2 Site 2 WFS

Site 2 WFS (Pictures 4-2 and 4-3) consisted of the area in and around the dam on this site. Twenty one sediment samples (500 g) were collected from this dam. Fifteen grass (100 g) samples and sixteen soil samples (500 g) were collected along the southern edge, inlet and outlet of the dam. The soils are classified in Table 4-3.

The dam is quite shallow (depth of 1 meter) and cattle drink directly from the dam, often walking a couple of meters in to take a drink. This movement into the dam disturbs the sediment and the clear water becomes turbid with suspended sediments. The main grasses found along the dam, the inlet and outlet were Couch Grass (*Cynodon dactylon*) and Kikuyu (*Pennisetum clandestinum*). These grasses are actively grazed by the cattle throughout the year. Couch Grass is a creeping, perennial species with rhizomes and stolons. Kikuyu is a robust, creeping perennial grass which develops thick rhizomes (Dickinson *et al*., 2007).

Picture 4-3: Grass growing along the dam in the Experimental Group

Picture 4-2: Grass growing along the outlet of the dam in the Experimental group

Table 4-3: Site 2 WFS sediment texture class, soil classification, soil texture class and location

Sediment Texture Class	
Light coloured sediment	Sand
Dark coloured sediment	Sandy loam, Loamy sand, Loam
Soil Classification	
Soil form	CLOVELY
Soil group:	Oxidic, yellow-brown apedal
Soil Texture Class	Silt loam
Location	

Latitude	-26.380039°
Longitude	27.226827°
Altitude	1451 m

4.3.3 Site 3 WFS

Site 3 WFS (Picture 4-4) consisted of approximately 30 ha of wetlands. Five grass samples (100 g) and six soil samples (500 g) were collected at this site. The soils are classified in Table 4-4. Cattle graze this area throughout the year especially during winter as the grass in this area remains green and growing in winter months (Dickinson *et al.*, 2007).

Picture 4-4: The wetland in the Experimental Group

The main grasses found in the wetland site were Couch Grass (*Cynodon dactylon*) and Kikuyu (*Pennisetum clandestinum*) (Dickinson *et al.*, 2007).

Table 4-4: Site 3 WFS soil classification, texture class and location

Soil Classification	
Soil form	SHORTLANDS
Soil group:	Oxidic, red structured B
Soil Texture Class	Silt clay
Location	
Latitude	-26.394258°
Longitude	27.207332°
Altitude	1439 m

4.3.4 Site 4 WFS

Site 4 WFS was located approximately 250 m to the south of the Wonderfonteinspruit and well beyond the floodplain. This site was selected to serve as a control site within the experimental group as it cannot be directly affected by the Wonderfonteinspruit. Although the sight could not be directly affected by the Wonderfonteinspruit it is still subjected to contaminants arising from windblown dust as there are a number of tailings facilities within 10 km. The Wonderfonteinspruit is actually visible in the centre of Picture 2-5. This site is a typical representation of the natural vegetation that is not adjacent to the Wonderfonteinspruit. Six soil samples (500 g) and five grass samples (100 g) were collected at this site. The soils are classified in Table 4-5.

Picture 4-5: The control site in the Experimental Group

The dominant grass species in this site is Red Grass (*Themeda triandra*) and cattle are able to graze it throughout the year. Red Grass is perennial, tufted and a very important grazing grass (Dickinson *et al*., 2007).

Table 4-5: Site 4 WFS soil classification, texture class and location

Soil Classification	
Soil form	HUTTON
Soil group:	Oxidic, red apedal
Soil Texture Class	Sandy loam
Location	
Latitude	-26.383542°
Longitude	27.227160°
Altitude	1459 m

4.4 Control Group Sites

Samples were collected at two different sites in the Control Group. The sites are displayed in Figure 4-3. Fifteen water samples (250 mL) were collected along a 1.5 km stretch of the Wonderfonteinspruit, still remaining within the Muiskraal farm.

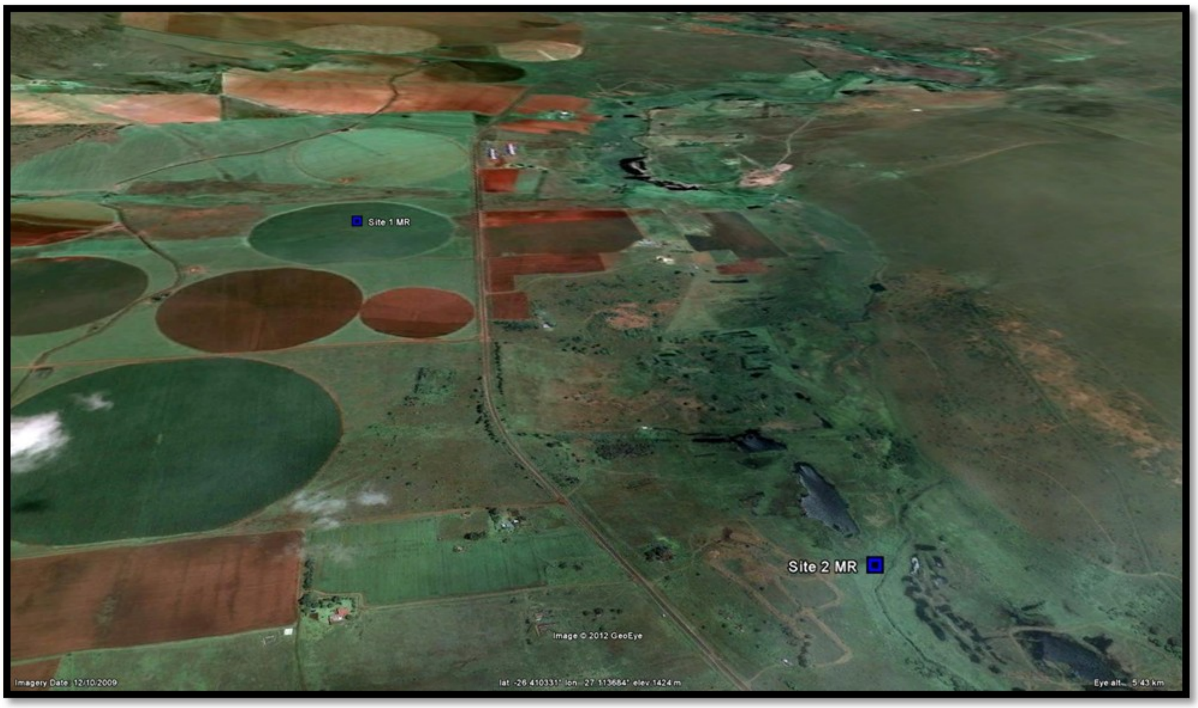


Figure 4-3: Google earth aerial map of the Control Group. Imagery date: 12/10/2009

4.4.1 Site 1 MR

Site 1 MR consists of approximately 25 ha of planted Annual Ryegrass (*Lolium multiflorum*) on the Roodraai farm. This pasture is irrigated directly from a canal that is fed by the Mooi River (Picture 4-6).

Picture 4-6: Irrigation of planted pastures in the Control Group

Six soil and five grass samples were collected at this site. The soils are classified in Table 4-6. Annual Ryegrass is an evergreen, tuft forming annual species (Dickinson *et al*., 2007). The site is not used for pastures exclusively as maize is planted in the summers and the Annual Ryegrass is established every year after the maize has been harvested.

Table 4-6: Site 1 MR sediment texture class, soil classification, soil texture class and location

Sediment Texture Class	
Loam, Clay loam	
Soil Classification	
Soil form	HUTTON
Soil group:	Oxidic, red apedal
Soil Texture Class	Silt loam
Location	
Latitude	-26.402464°
Longitude	27.110471°
Altitude	1437 m

4.4.2 Site 2 MR

Site 2 MR (Picture 4-7) consist of a 1 km stretch along the Mooi River on the Muiskraal farm. Five grass samples and six soil samples were collected along the Mooi River, in areas that were accessible to grazing. The soils are classified in Table 4-7. Six sediment samples were collected from the Mooi River in areas with slow moving water.

Picture 4-7: Grass growing along the Mooi River in the Control Group

The main grasses found in the wetland site were Couch Grass (*Cynodon dactylon*) and Kikuyu (*Pennisetum clandestinum*) (Dickinson *et al*., 2007).

Table 4-7: Site 2 MR soil classification, texture class and location

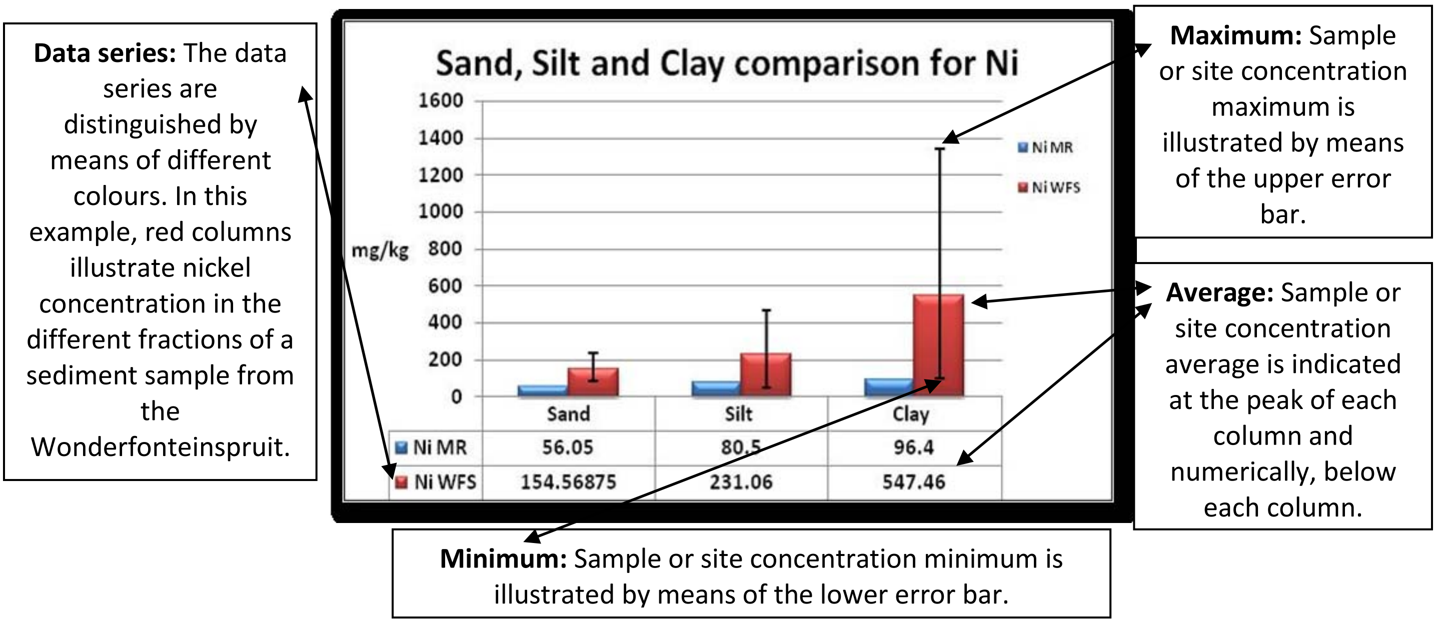
Soil Classification	
Soil form	CLOVELY
Soil group:	Oxidic, yellow-brown apedal
Soil Texture Class	Silt loam
Location	
Latitude	26.419100°
Longitude	27.115401°
Altitude	1421 m

5 Data Evaluation

5.1 Presentation of results

Large volumes of data were produced during this study, predominantly from ICP-MS scans. The raw data was processes and presented in the form of figures and tables. Due to sheer volume, most of the data that will be presented in this chapter will focus on the essentially toxic elements, cadmium, lead and uranium. These elements which are renowned for their toxicity and lack of benefits to living organisms will be discussed in more detail. This does however not suggest that these elements are of greater concern or that the other elements are less important. As the renowned Paracelsus once said: “All things are poison and nothing is without poison, only the dose permits something not to be poisonous" (Stawicki, 2007). This implies that even the essential elements could be toxic if they are ingested at a dose that permits them to be. The actual data tables and graphs from all elements of interest tables can be found in the respected appendixes.

An example of the results that are illustrated by means of figures is shown in Figure 5-1.



Data series: The data series are distinguished by means of different colours. In this example, red columns illustrate nickel concentration in the different fractions of a sediment sample from the Wonderfonteinspruit.

Maximum: Sample or site concentration maximum is illustrated by means of the upper error bar.

Average: Sample or site concentration average is indicated at the peak of each column and numerically, below each column.

Minimum: Sample or site concentration minimum is illustrated by means of the lower error bar.

Figure 5-1: Nickel concentration in the different fractions (sand, silt and clay) that are found in sediment samples from the Wonderfonteinspruit (Red) and the Mooi River (Blue)

Many of the data sets that were used exhibited large standard deviations relative to their means. To accommodate this characteristic, the customary error bars were not calculated as the mean plus/minus one standard deviation, as the lower error bar would have a negative value in many of the cases (giving the mistaken impression that negative concentrations were measured). In order to circumvent this problem the error bars illustrate the minimum and the maximum values obtained in the sample under discussion. The standard deviation of each sampled site is however used to determine the practically significant difference between experimental and control samples.

In some instances abbreviations were used and the most common are WFS (Wonderfonteinspruit), indicating that the site or sample is from the experimental group, and MR (Mooi River), indicating that the site or samples is from the control group. All other abbreviations were presented in the list of abbreviations.

The practically significant difference between sites or samples from the experimental and control group will be illustrated in the form of tables.

5.2 Sediment

5.2.1 Identification of Elements of Interest

As mentioned earlier, streambed sediments can act like a sink for elements entering the water body. (Stackelberg 1997; Munn & Gruber, 1997). The comparison of the elemental concentrations within the sediments of the WFS with background levels, can serve as an indication of mining pollution. The sediment samples from the MR will serve as the back ground levels (normal concentrations).

In order to identify the elements of interest, eight light coloured sediment samples from the WFS, eight dark coloured sediment samples from the WFS and five sediment samples from the MR were collected, analyzed and compared.

This comparison resulted in the identification and prioritization of the following elements: cobalt, nickel, copper, zinc, selenium, cadmium, lead, gold and uranium. These elemental concentrations were all elevated above background levels and will be discussed in more detail.

Other elements such as vanadium, chromium, manganese, iron, arsenic and barium were also part of the investigation but were found to have higher concentration in the sediment samples from the

MR when compared to the sediment samples from the WFS. Although the average arsenic concentration in the dark coloured sediment samples was found to be in close proximity to that of the sediment samples from the MR, the MR sediment samples exhibited a greater maximum value and for this reason arsenic was excluded from the elements of interest. The actual data tables for the three sediment groups can be found in Appendix A, in Table 9-1 and Table 9-3.

The complete particle size distribution of each sample is presented in Appendix A. The eight light coloured sediment samples from the WFS all had a predominantly sandy texture (Table 9-4). The dark coloured sediment samples consisted of 4 sandy loam samples, 2 loamy sand samples, 1 silt loam sample and 1 loam sample (Table 9-5). The MR sediments consisted of 4 loam samples and 1 clay loam sample (Table 9-6).

The elements of interest were discussed by means of graphs.

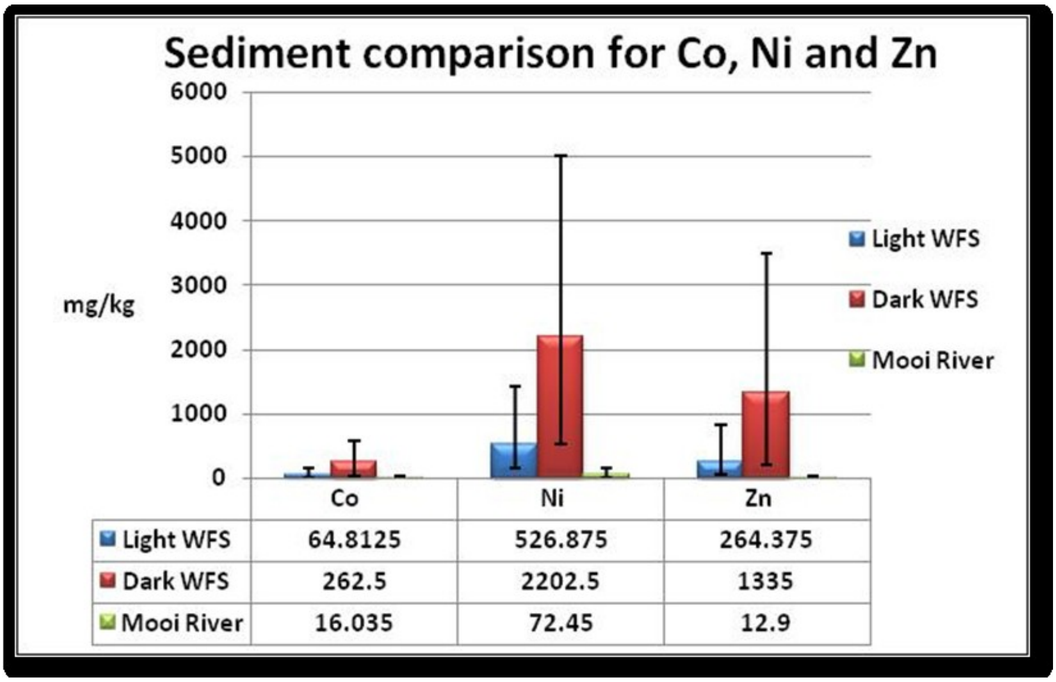
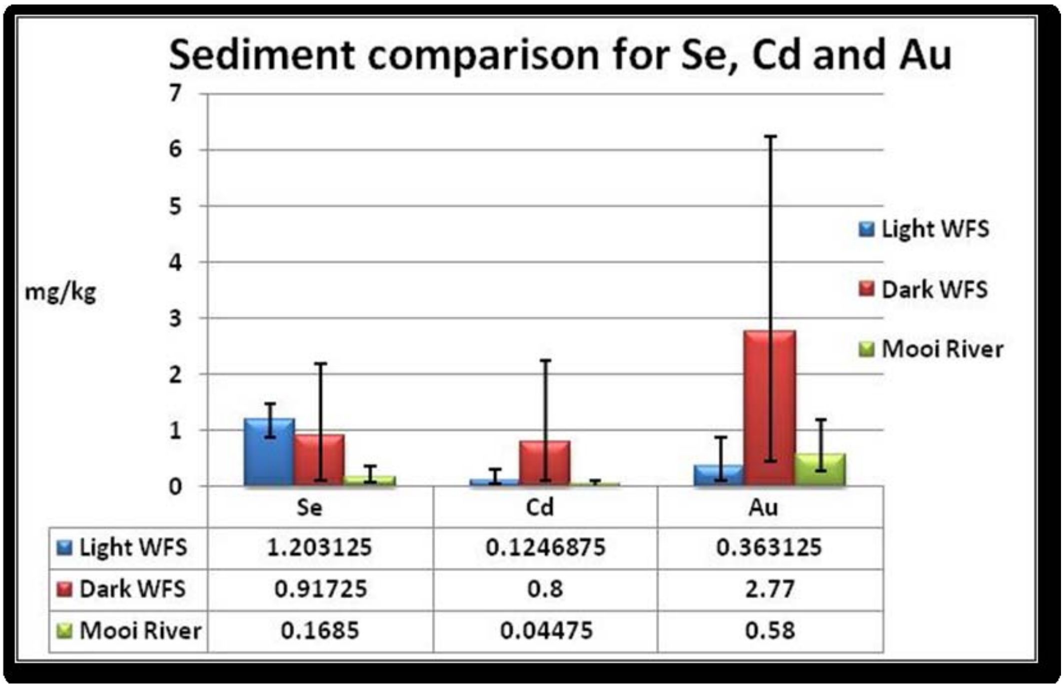


Figure 5-2: A comparison of sediments from the WFS and MR for cobalt, nickel and zinc concentrations. Light coloured sediments from the WFS in blue, dark coloured sediment samples from the WFS in red and the background sediment samples from the MR in green

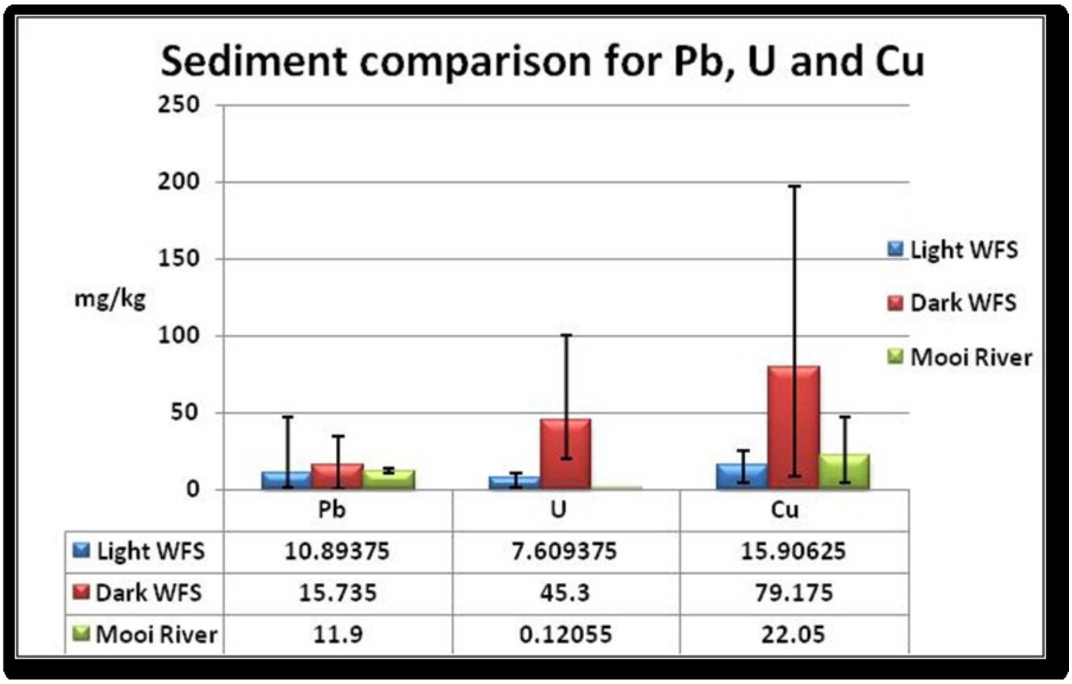
The cobalt, nickel, and zinc concentrations of the three different samples types are illustrated in Figure 5-2. Both WFS groups, i.e. light and dark coloured samples, had higher elemental concentrations than that of the MR samples. The comparison illustrated that the dark coloured sediments consisted of higher elemental concentrations when compared to the light coloured WFS sediments and MR sediments. The cobalt concentration was 16.37 times higher than back ground, the nickel concentration was 30.4 times higher and the zinc concentration 103.49 times higher than back ground levels.



	Se	Cd	Au
🟦 Light WFS	1.203125	0.1246875	0.363125
🟥 Dark WFS	0.91725	0.8	2.77
🟩 Mooi River	0.1685	0.04475	0.58

Figure 5-3: A comparison of sediments from the WFS and MR for selenium, cadmium and gold concentrations. Light coloured sediments from the WFS in blue, dark coloured sediment samples from the WFS in red and the background sediment samples from the MR in green

The selenium, cadmium, and gold concentrations of the three different sample types are illustrated in Figure 5-3. Selenium concentrations were found to be higher in the light coloured sediment samples (7.14 times higher than back ground levels). The dark coloured sediment samples were found to have higher concentrations of cadmium (17.88 times higher than back ground levels) and gold (4.78 times higher than back ground levels). As gold does not pose any hazard to living organisms (mainly due to its low concentration) it will only be discussed in the sediment section of this study. The gold concentration could however serve as motivation for the removal and processing of the sediments in order to extract the gold. According to Naicker, *et al*., (2002), tailings dumps with a gold concentration around 0.5 g/ton are being re-mined. The dark coloured sediments from the WFS contain a gold concentration of 2.77 g/ton, which is five times higher than the tailings dumps, but these sediments are some distance away from any mining facilities and might pose a greater challenge to rework than the tailing dumps.



	Pb	U	Cu
Light WFS	10.89375	7.609375	15.90625
Dark WFS	15.735	45.3	79.175
Mooi River	11.9	0.12055	22.05

Figure 5-4: A comparison of sediments from the WFS and MR for lead, uranium and copper concentrations. Light coloured sediments from the WFS in blue, dark coloured sediment samples from the WFS in red and the background sediment samples from the MR in green

The lead, uranium and copper concentrations of the three different sample types were illustrated in Figure 5-4. Uranium concentrations were found to be higher than the background level in both light and dark coloured sediment samples, with the concentration in the dark coloured sediment samples being 375.78 times higher than the back ground levels. The lead and copper concentrations were found to be higher than background values only in the dark coloured WFS samples, with the lead concentration being 1.32 times higher than background levels and the copper concentration being 3.59 times higher than background levels.

Table 5-1 illustrates the practically significant differences between the sediments from the WFS and the MR. The comparison between the elemental concentrations of the light coloured sediments from the WFS and the MR sediments indicated that the cobalt, nickel, zinc, selenium, cadmium and uranium concentrations had a large effect size, and therefore a practically significant difference. Copper, gold and lead only displayed a medium effect size, therefore not having a difference that is practically significant. The comparison between the elemental concentrations of the dark coloured sediment samples from the WFS and the sediments from the MR indicated that all the elements of interest had a large effect size, and therefore also had a difference that was found to be practically significant.

Table 5-1: The practically significant differences between elemental concentrations of the Light coloured WFS sediment and the Mooi River and; The dark coloured WFS sediment and The Mooi River sediment.

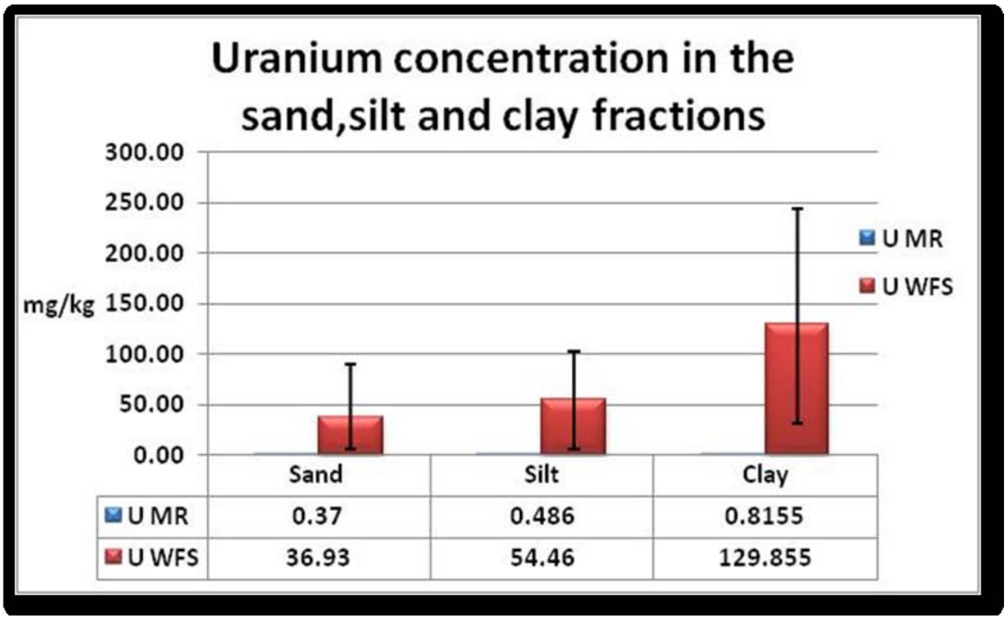
Light coloured sediment WFS vs. Sediment from the MR			Dark coloured sediment WFS vs. Sediment from the MR		
Element	**Effect size**	**Practical significance**	**Element**	**Effect size**	**Practical significance**
Cobalt	16.55	Large	**Cobalt**	3.28	Large
Nickel	7.65	Large	**Nickel**	35.84	Large
Copper	0.39	Medium	**Copper**	3.60	Large
Zinc	24.54	Large	**Zinc**	128.99	Large
Selenium	8.73	Large	**Selenium**	6.32	Large
Cadmium	1.53	Large	**Cadmium**	14.42	Large
Gold	0.61	Medium	**Gold**	6.12	Large
Lead	0.74	Medium	**Lead**	2.81	Large
Uranium	57.41	Large	**Uranium**	346.34	Large

Although the light and dark coloured sediment samples were collected from the same site they differ significantly in terms of in texture and elemental concentrations. This difference in texture, in all probability is caused by different particle size distribution of the sediment fractions. As mentioned previously the light coloured sediment samples were found to consist predominantly of sand (93.5 – 96.4% sand, 0.9 – 1.4% silt, 2.8 – 5.4% clay) as can be seen in Appendix A (Table 9-4). The dark coloured sediment samples were also dominated by a sand fraction, but the silt and clay fractions are much higher, (87.6 – 34.4 % sand, 6.2 – 52.9% silt, 6.1 – 21.8 clay) than that of the lighter sediment samples from the WFS, as can be seen in Appendix A, Table 9-5. The higher elemental concentrations contained in the dark sediment samples from the WFS could have been caused by the higher silt and clay fractions that generally consist of secondary soil minerals which have the ability to adsorb dissolved element cations onto their surface areas (Sparks, 2003). It was therefore deemed necessary to establish the elemental concentrations within the different particle size fractions (sand, silt and clay) in order to gain a better understanding of the sediment contamination.

5.2.2 Comparison of the Elemental Concentrations within the different particle size fractions of Sediments Samples

Samples from the WFS and MR were collected and sieved to form sand, silt and clay fractions based on their particle size distribution. These samples were sieved without the use of other chemicals in order to portray the most accurate chemical composition possible. This method does however allow a small margin of error, where particles with small sizes stay bound to each other due to their surface charges and end up in a larger particle size fraction (Sparks, 2003). The WFS sediments displayed great heterogeneity in the previous section that was thought to have been caused by the large variations in elemental concentrations and particle size distribution. For this reason five samples of sediment from the WFS and one sample from the MR was collected and sieved into sand, silt and clay fractions. The previous sediment samples from the MR displayed much lower elemental concentrations with a greater homogeneity in particle size distribution and elemental concentrations compared to that of the WFS sediment samples.

Figure 5-5 illustrates the different uranium concentrations of the sediment samples from the WFS (in red) and MR (in blue). It is evident that in both groups, the uranium concentration is significantly higher in the clay fraction.

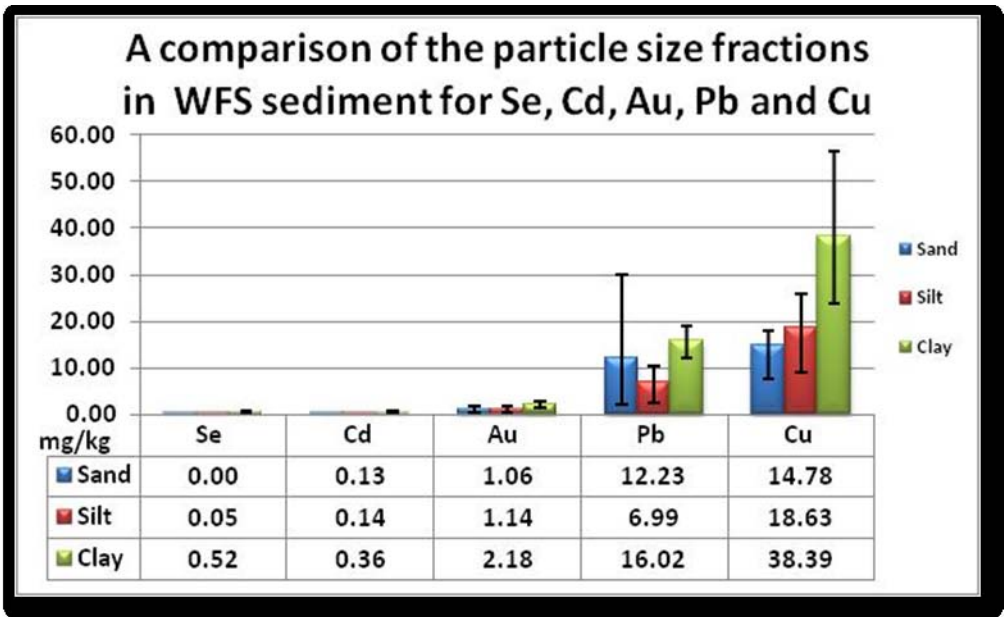


	Sand	Silt	Clay
U MR	0.37	0.486	0.8155
U WFS	36.93	54.46	129.855

Figure 5-5: A comparison of the uranium concentration in the different fractions (sand, silt and clay) between sediment samples from the WFS (in red) and MR (in blue)

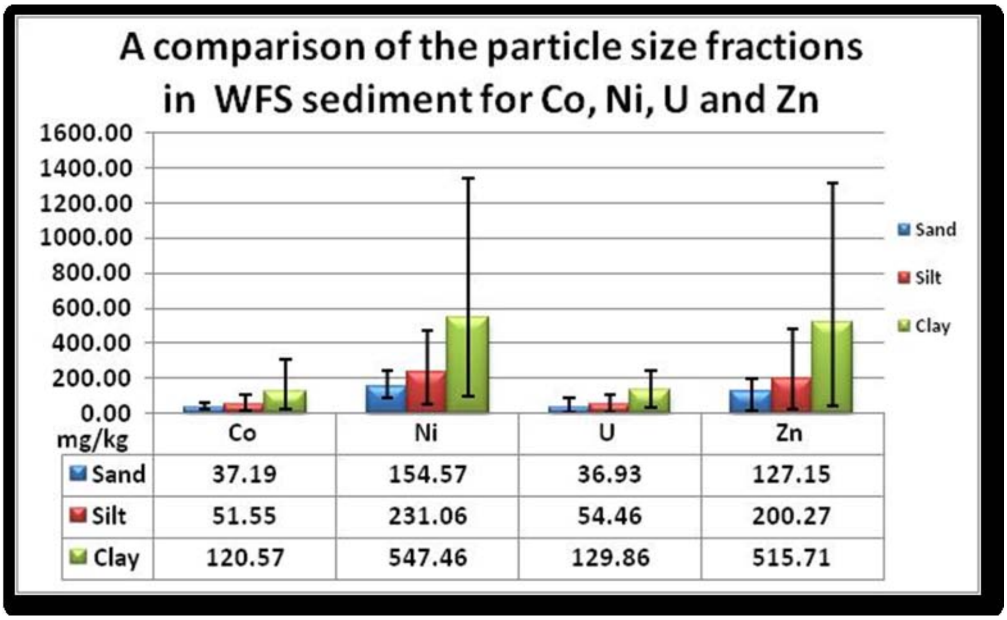
The uranium concentration in the WFS sediment was found to be 100.42 times higher in the sand fraction, 112.01 times higher in the silt fraction and 159.23 times higher in the clay fraction, than that of the MR sediment particle size fractions.

The elemental concentrations of the different particle size fractions from the WFS sediments were presented in Figure 5-6 and Figure 5-7.



mg/kg	Se	Cd	Au	Pb	Cu
🟦 Sand	0.00	0.13	1.06	12.23	14.78
🟥 Silt	0.05	0.14	1.14	6.99	18.63
🟩 Clay	0.52	0.36	2.18	16.02	38.39

Figure 5-6: A comparison of the selenium, cadmium, gold, lead and copper concentrations in the different particle size fractions from the WFS sediment samples

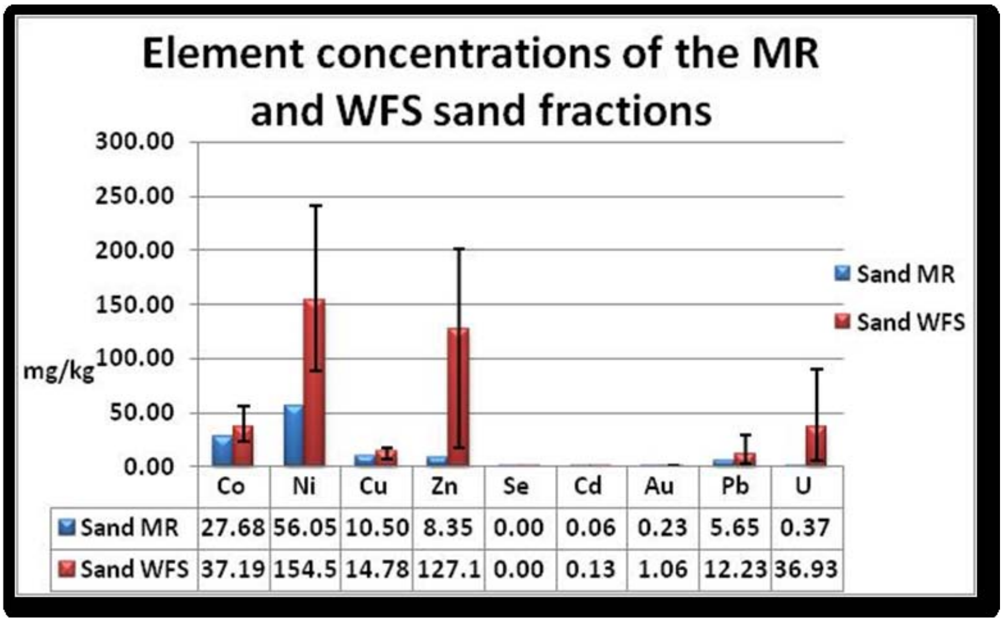


mg/kg	Co	Ni	U	Zn
🟦 Sand	37.19	154.57	36.93	127.15
🟥 Silt	51.55	231.06	54.46	200.27
🟩 Clay	120.57	547.46	129.86	515.71

Figure 5-7: A comparison of the cobalt, nickel, uranium and zinc concentrations in the different particle size fractions from the WFS sediment samples

Most of the other elements illustrated the same paradigm of elemental concentrations within the different particle size fractions as portrayed by that of uranium in Figure 5-5, except for the lead concentrations. The elemental concentrations of the different particle size fractions from the MR sediments were presented in Appendix A, Figure 9-6 and Figure 9-7.

Sand fractions generally consist of primary soil minerals, such as feldspars and quartz (Brady & Weil, 2008). These primary soil minerals are large particles with small specific surface areas and therefore have a very low capacity to adsorb dissolved elemental cations onto their surfaces (Brady & Weil, 2008). The fact that the average lead concentration was higher in the sand fraction than that of the silt fraction, and that the maximum lead concentration was found in the sand fraction of the WFS sediment samples, proved to be an anomaly that would require further investigation. Furthermore, the other elemental concentrations in the sand fraction of the WFS sediment samples were found to be much higher than that of the sand fraction in the MR sediments, as can be seen in Figure 5-8.



	Co	Ni	Cu	Zn	Se	Cd	Au	Pb	U
Sand MR	27.68	56.05	10.50	8.35	0.00	0.06	0.23	5.65	0.37
Sand WFS	37.19	154.5	14.78	127.1	0.00	0.13	1.06	12.23	36.93

Figure 5-8: A comparison of the different elemental concentrations in the sand fraction of sediments from the WFS (in red) and MR (in blue)

The preliminary investigation seemed to illustrate that the sediment samples from the WFS did not abide to the principles of soil chemistry and further investigation by means of an X-ray diffraction analysis was required to achieve a better understanding of the aforementioned anomalies.

5.2.3 XRD Comparison

X-ray diffraction reveals information about the crystal structure and physical properties of materials. In this study it was used to identify the mineral composition of the different particle size fractions (sand, silt and clay) of sediment samples from the WFS and MR. The results are portrayed in Table 5-2 and Table 5-3.

Table 5-2: X-Ray Diffraction results of the different particle size fractions (sand, silt and clay) from WFS sediments

Wonderfonteinspruit sediment			
Minerals Identified	Sand fraction	Silt fraction	Clay fraction
Quartz	94	90	91
Goethite	3	4	2
Nacrite	3	0	0
Koalite	0	5	6
Jacobsite	0	1	0
Hematite	0	0	1

Table 5-3:-Ray Diffraction results of the different particle size fractions (sand, silt and clay) from MR sediments

Mooi River sediment			
Minerals Identified	Sand fraction	Silt fraction	Clay fraction
Quartz	63	68	52
Goethite	4	5	8
Montmorillonite	1	1	1
Mica	28	20	24
Nacrite	4	7	15

The minerals that were identified in the WFS sediment fractions were quartz, goethite nacrite, koalite, jacobsite and hematite and the minerals that were identified in the MR sediment fractions were quartz, goethite, montmorillonite, mica and nacrite.

Quartz was the only primary soil mineral in both groups; all the other minerals were identified secondary soil minerals (Sparks, 2003). Table 5-4 displays the primary and secondary soil composition of the different particle size fractions from both sets of sediment samples.

Table 5-4: The primary and secondary soil composition of the different particle size fractions (sand, silt and clay) from WFS and MR sediment samples

Wonderfonteinspruit sediment			
	Sand fraction	**Silt fraction**	**Clay fraction**
Primary soil mineral composition	**94**	**90**	**91**
Secondary soil mineral composition	**6**	**10**	**9**
Mooi River sediment			
	Sand fraction	**Silt fraction**	**Clay fraction**
Primary soil mineral composition	**63**	**68**	**52**
Secondary soil mineral composition	**27**	**22**	**48**

The WFS sediment was found to contain more primary soil minerals than the MR sediment; this high primary soil mineral content significantly reduces the WFS sediment’s capacity to adsorb dissolved elemental cations and begs the question, where did the high elemental concentrations in the WFS sediment arise from. The answer, in all probability lied in the composition of the gold reefs that were mined in and around the WFS catchment.

The two gold fields, the Westrand Goldfield and the West Wits Line contained gold reefs that consisted of primary gold quartz veins (McCarthy 2006). These quartz veins consisted of quartz pebbles (1- 3 cm in diameter) set in a quartz sand matrix, and contained some 70 different ore minerals according to Feather and Koen (1975), as quoted by (Naicker *et al.*, 2002). The most abundant ore minerals were pyrite, uraninite(UO_2), brannerite ($UO_3Ti_2O_4$), arsenopyrite ($FeAsS$), cobaltite ($CoAsS$), galena (PbS), pyrrhotite (FeS), gersdofite ($NiAsS$) and chromite ($FeCr_2O_4$) (Naicker *et al.*, 2002).

These reefs were located at depths that exceeded the reach of manual extraction methods and therefore required sophisticated techniques and equipment (McCarthy 2006). The ore was brought to the surface and milled to fine sand during which it was exposed to a mercury film which was spread on copper plates. These plates were subsequently removed in order to recover the gold amalgamation. The remaining sand was transported to dumps, forming the so-called “sand dumps”. These sand dumps were then milled even finer particle size in order to recover more gold via the MacArthur-Forrest process of gold extraction, which used cyanide to dissolve the gold. After the gold had been removed, the finely milled ore was pumped to large dumps, known as "slime dumps" (Naicker *et al*., 2002).

Most of these sand and slimes dumps are located along drainage lines and canals that are able to flow into the WFS. Erosion of these dumps and the direct discharge of tailings into the WFS could therefore have resulted in the introduction of elements in particulate form. According to Miller, 1997, it is not uncommon for 90 – 99 % of the total elemental load in rivers to be transported in the particulate phase if tailings material is dumped in close proximity to the stream (Miller, 1997).

This could explain why the compositions of the sediment samples from the WFS were dominated by the presence of quartz, and why the quartz was found in the all the different particle size fractions of these WFS sediments. The elements found in the sediments could have been incorporated within the quartz minerals or, could have formed a coating onto the quartz. Aluminium and iron silicate clays have the ability to form a surface coating on the surface of primary soil minerals such as quartz. These clay coatings can then adsorb elemental cations onto their surface areas and then transport these adsorbed elements through the environment (Harrison *et al*., 2005) and (Coston *et al*., 1995). Most of the elements of interest, as identified in this study are by-products from gold mining activities and gold mining techniques. The quartz in the MR sediment samples was likely to have formed by means of natural weathering processes as it contains relatively low elemental concentrations when compared to that of the WFS.

5.2.4 Case study: Radioactivity study on Sediment in a Dam in the Wonderfonteinspruit Catchment

In 2002 Coetzee *et al*. produced a report for the Department of Water Affairs and Forestry on the radioactivity of sediments from the WFS (hereafter referred to as the Coetzee report). The aim of the Coetzee report was to establish the potential release of radioactivity and other heavy metals from a dam in the WFS by analysis sediment and water samples.

Sediment and water samples were collected form a dam on the Blaauwbank farm and compared to samples from Klerkskraal Dam, which served as a local background value. The data collected from both sites identified uranium and nickel to be elements of high priority, cobalt, copper and chrome to be elements of concern and arsenic and zinc to be elements of possible concern. Furthermore the report suggests that radionuclides and heavy metals were being adsorbed and concentrated in the sediments from the WFS, causing the dam to be hazardous in its current state and remedial action was deemed to be necessary.

The Coetzee report is based on work conducted on a dam in the WFS which is approximately 1 km upstream from the experimental group monitored in the study presented in this document. There are many similarities and few distinctions between the Coetzee report and the findings of the experimental group presented in this document.

The experimental group results revealed that cadmium, selenium and lead concentrations were elevated above background values and that uranium, zinc, nickel and cadmium concentrations had the largest practically significant differences of the elements of interest, identifying these elements to be high priority based on the elevated elemental concentrations compared to the background values.

The inclusion of chrome and arsenic as high priority elements in the Coetzee report contrast the experimental group results as these elements were found to have higher concentrations in the sediment samples from the control group (MR) than that of the WFS as indicated in Table 9-1, Table 11-2 and Table 9-3, which can be found in Appendix A . The inclusion of chromium and arsenic and exclusion cadmium and lead was therefore found to be key dissimilarities between the two studies.

Furthermore the Coetzee report does not specify in which particle size fraction the highest elemental concentrations were found and did not fully investigate the mineralogy of the sediment samples. The fact that sediment samples were dominated by sand sized particles and the secondary soil mineral, quartz, which contrary to popular understanding contained high elemental concentrations, serves as an indication that an investigation into the elemental concentrations of the particle size fractions (sand, silt and clay) and the mineralogical composition of the sediment samples was an essential component of the investigation. The Coetzee report states that heavy metals are adsorbed and concentrated onto the surface of the WFS sediments, but the WFS's capacity to adsorb and concentrate these heavy metals may have been over estimated due to the dominance of secondary soil minerals in the WFS sediments which have no or little ability to adsorb elemental cations.

The two studies do however reach a similar conclusion in that this particular study area in the WFS and the immediate vicinity could be hazardous and would require remedial actions.

5.3 Water

From this point on, only the essentially toxic elements (cadmium, lead and uranium) will be illustrated in this chapter. The rest of the elements of interest (cobalt, nickel, copper, zinc and selenium) will be discussed and their graphs can be found in Appendix B. The focus of the study expands to encompass the transfer and distribution of elements from the WFS into the surrounding environment.

5.3.1 Comparison with Natural Waters

Water samples from the experimental group (WFS) and the control group (MR) were collected, analysed and their elemental concentrations were compared with each other and with concentrations that are found in natural waters. The elemental concentrations of natural waters were provided by NAS (1997), Livingstone (1963), Turekian (1971) and Hem (1971) as found in Crittenden *et al*., (2005).

Table 5-5 portrays the comparison of the elemental concentrations found in natural waters, water samples from the MR and water samples from the WFS.

Table 5-5: A comparison of elemental concentrations found in natural waters, MR water samples and WFS water samples

Element	Concentrations in natural waters	MR water samples		WFS water samples	
		Average	Max	Average	Max
Co (mg/ℓ)	ND – 0.001	0.00004	0.00006	0.02417	0.20000
Ni (mg/ℓ)	0.01	0.00192	0.00200	0.12746	0.64000
Cu (mg/ℓ)	0.01	0.00382	0.00390	0.10267	0.28604
Zn (mg/ℓ)	0.01	0.00280	0.00390	0.10707	0.44299
Se (mg/ℓ)	0.0002	0.00688	0.00750	0.06253	0.10733
Cd (mg/ℓ)	ND – 0.01	ND	ND	0.00184	0.02438
Pb (mg/ℓ)	0.001 – 0.01	0.00054	0.00057	0.00509	0.02440
U (mg/ℓ)	0.00002 – 0.2	0.00078	0.00080	0.05704	0.23358

The water samples from the MR had cobalt, nickel, copper, zinc and lead concentrations that were below the concentrations found in natural waters. The MR samples exhibited a higher selenium concentration and similar cadmium and uranium concentrations when compared to the respective concentrations that were found in the natural waters. The water samples from the WFS were found to contain higher elemental concentrations than that of the natural waters for cobalt, nickel, copper, zinc, selenium and cadmium. Although the average uranium and lead concentrations of the WFS water samples were within the same range as the natural waters, the maximum values for both uranium and lead exceeded the average concentrations found in the natural waters.

5.3.2 Livestock Watering

The WFS water is no longer used as a source for domestic water, therefore there will be no comparison to domestic use standards. The WFS water was still used for agricultural purposes, with livestock water being one of the main methods of utilization. Cattle in the experimental group drink directly from the WFS and often disturbed the underlying sediments which could increase the elemental concentrations of the water that is ingested by the cattle. It was therefore necessary to recreate this disturbance of the sediment and collect and analyze this disturbed water (murky water) in order to establish whether or not the elemental concentrations increase.

The water samples that were collected from the experimental and control groups, MR water, WFS water and the disturbed water from the WFS will be compared with each other. Additionally the livestock Target Water Quality Range (TWQR) for each element, as given by the South African Water Quality Guidelines (DWAF, 1996) will be indicated on the graphs of each element. Table 5-6 illustrates the TWQR's for each element (DWAF, 1996).

Table 5-6: Target Water Quality Ranges for the elements as found in DWAF, 1996

Element	Target Water Quality Range
Cobalt	0.5 – 1 mg/ℓ
Nickel	0 – 1 mg/ℓ
Copper	0 – 1 mg/ℓ
Zinc	0 – 20 mg/ℓ
Selenium	0 – 50 mg/ℓ
Cadmium	0 – 0.1 mg/ℓ
Lead	0 – 0.1 mg/ℓ

The results obtained from the comparative analysis of the cadmium concentration in the respective water samples were illustrated in Figure 5-9. The disturbed water samples contained higher cadmium concentration that that of the WFS and MR water samples and exceeded the TWQR. Although the average cadmium concentration in the WFS sample was below the TWQR, the maximum value did exceed it. This could be an indication that the water quality in the WFS varies over time. Cadmium could not be detected in the water samples from the MR.

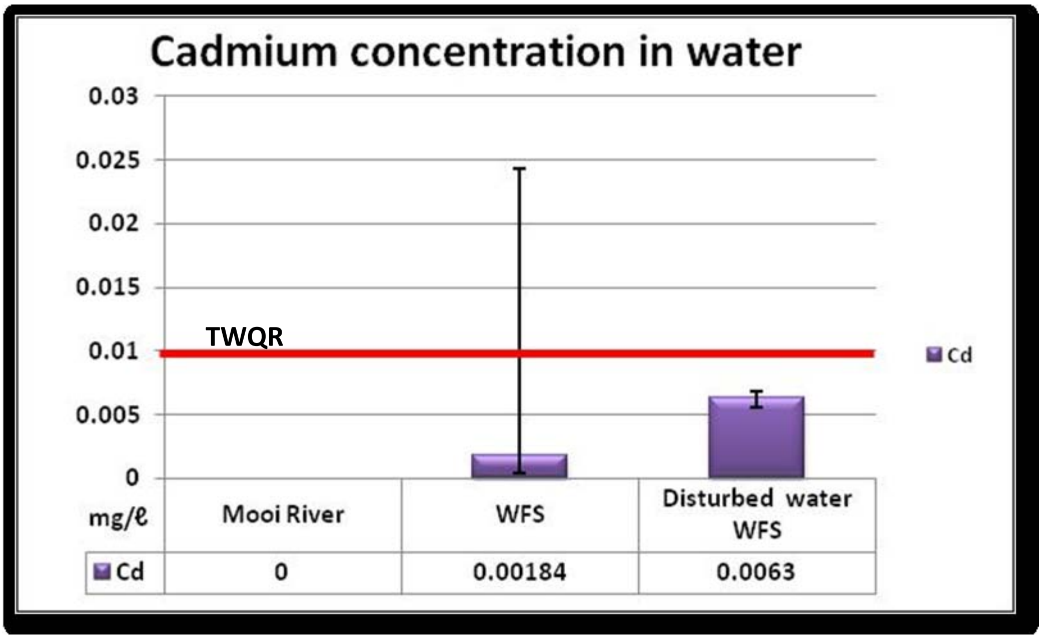


Figure 5-9: A comparison of the cadmium concentration in the respective water samples

The results obtained from the comparative analysis of the lead concentration in the respective water samples were illustrated in Figure 5-10. Its results were very similar to that of cadmium. The disturbed water samples had the highest lead concentration and exceeded the TWQR. Although the average lead concentration in the WFS sample was below the TWQR, the maximum value did exceed it.

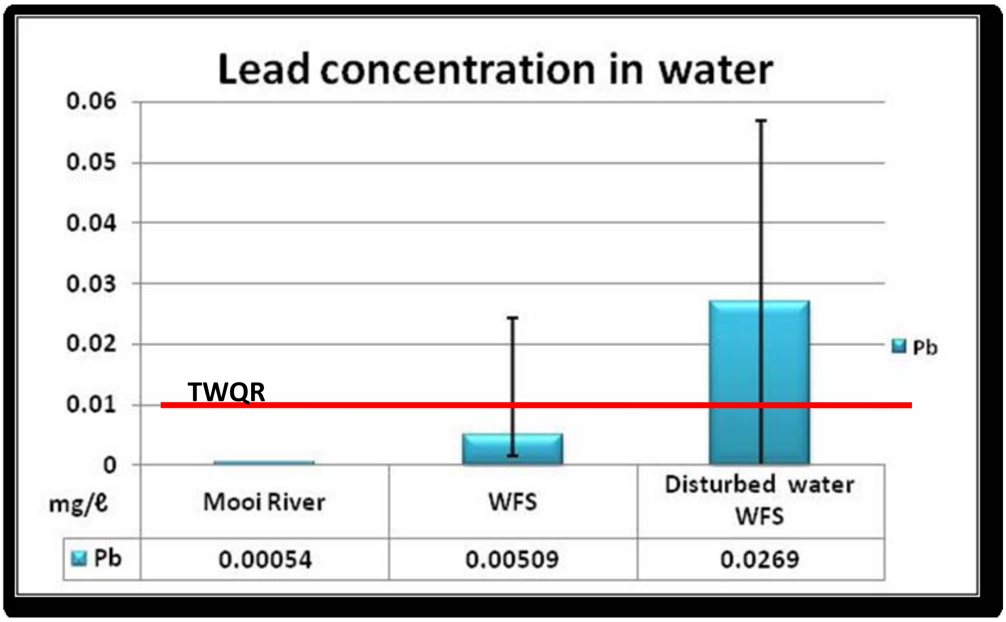


Figure 5-10: A comparison of the lead concentration in the respective water samples

The results obtained from the comparative analysis of the uranium concentration in the respective water samples were illustrated in Figure 5-11. There is no uranium TWQR for livestock in the South African Water Quality Guidelines. The disturbed water samples once again contained the highest average when compared to the WFS and MR samples.

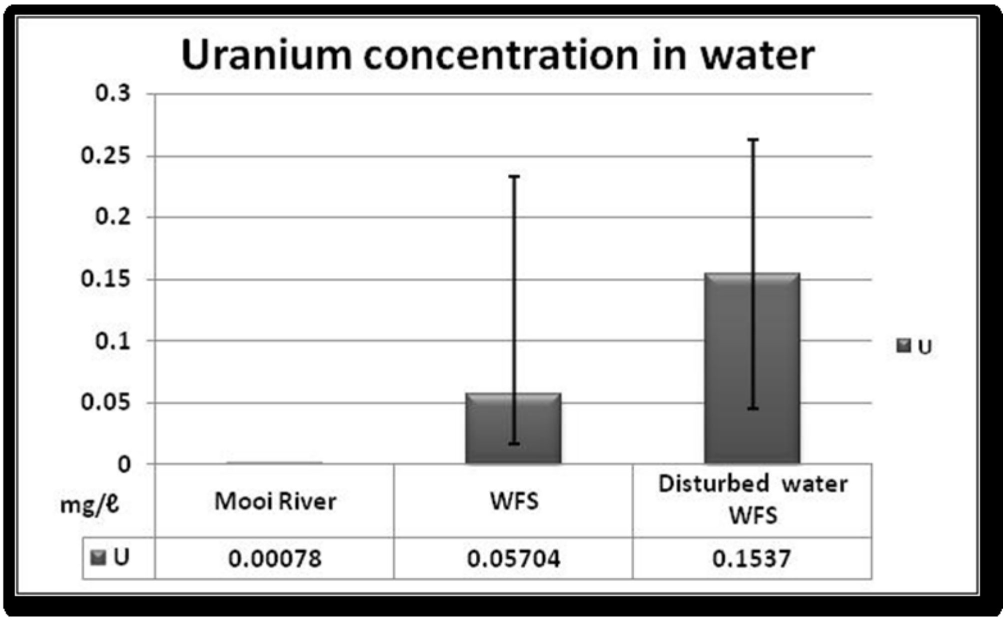


Figure 5-11: A comparison of the uranium concentration in the respective water samples

The cobalt (Figure 10-1), zinc (Figure 10-4) and nickel (Figure 10-2) concentrations were fairly similar to the cadmium, lead and uranium concentrations, with low concentrations in the MR and WFS samples and high concentrations in the disturbed water samples. The nickel concentration in the disturbed water was observed as exceeding the TWQR. The selenium (Figure 10-5) and copper (Figure 10-3) concentrations were the highest in the samples from the WFS (undisturbed), and did not exceed the TWQR. These graphs can all be found in Appendix B.

Table 5-7 illustrates the practically significant differences between the elemental concentrations in the WFS and MR water samples; and the elemental concentrations in the disturbed water samples from the WFS and the MR.

Table 5-7: The practically significant differences between the elemental concentrations in water samples from the WFS and the MR and; The disturbed water samples from the WFS and the MR water samples

WFS vs. Mooi River			Disturbed water vs. Mooi river		
Element	**Effect size**	**Practical significance**	**Element**	**Effect size**	**Practical significance**
Cobalt	1256.35	Large	**Cobalt**	20191.19	Large
Nickel	2807.15	Large	**Nickel**	53939.33	Large
Copper	902.39	Large	**Copper**	875.23	Large
Zinc	130.34	Large	**Zinc**	5249.00	Large
Selenium	84.18	Large	**Selenium**	8.62	Large
Cadmium	10000.00	Large	**Cadmium**	1000.00	Large
Lead	171.83	Large	**Lead**	995.58	Large
Uranium	3977.90	Large	**Uranium**	10810.25	Large

The comparison between the water samples from the WFS and the MR indicated that all the elements of interest have a large effect size, and therefore have a difference which is practically significant.

5.3.3 Case study: Radioactivity Study on Sediment in a Dam in the Wonderfonteinspruit Catchment

The Coetzee report found that the water quality results exceeded the domestic use TWQR for nickel, mercury and uranium. The study presented in this document does not consider domestic use, but rather livestock watering and found that cadmium, lead and nickel concentration could possibly exceed the TWQR. There is no uranium TWQR for livestock watering and the method of analysis did not allow for the quantification of mercury concentrations.

The Coetzee report furthermore predicts that heavy metals would be released from the sediments if the dam was allowed to run dry, followed by a pulse of water. This prediction is similar to the results obtained from the disturbed water samples from the WFS in the study presented in this document.

5.4 Soil

Soil samples were collected on four sites in the experimental group, and two sites in the control group. The two sites from the control group, irrigation soil MR and soil along MR, and one site from the experimental group, control site WFS were selected to serve as background values in the comparison with the other sites, irrigation soil WFS, soil along WFS and wetland soil WFS.

5.4.1 Elemental Concentrations commonly found in Agricultural Soils

Agricultural soils generally contain elements from a number of sources, predominantly the rocks from which the soil was formed, waste material and fertilizers. The soil in the experimental group has additionally been exposed to elements from mining activities via the WFS. Table 5-8 displays the elemental concentrations that are commonly found in agricultural soils.

Table 5-8: Elemental concentration found in natural soil, according to Kloke 1980a, as found in Bergman (1992)

Element	Common agricultural soil	Agricultural threshold values
Cobalt (mg/kg)	1 – 10	50
Nickel (mg/kg)	2 – 50	50
Copper (mg/kg)	1 – 20	100
Zinc (mg/kg)	3 – 50	300
Selenium (mg/kg)	0.1 – 5	10
Cadmium (mg/kg)	0.1 – 1	3
Lead (mg/kg)	0.1 – 20	100
Uranium (mg/kg)	<0.1 – 1	5

The elemental concentrations that were observed in the respective sampling sites in both the experimental and control groups can be found in Table 11-1 in Appendix C. The cobalt concentrations in the soil samples from the soil along WFS site, soil along MR site and irrigation MR site exceeded the agricultural threshold value. The nickel concentrations in the soil samples from the soil along WFS site, soil along MR site, wetland WFS and irrigation MR site exceeded the agricultural threshold value. The zinc concentrations in the soil samples from the soil along WFS site exceeded the agricultural threshold value. Copper, selenium, cadmium and lead concentrations did not exceed the agricultural threshold values in any of the respective sites. . The agricultural threshold value for uranium concentrations was exceeded in the soil samples from the soil along the WFS site and the wetland WFS site.

5.4.2 Site Specific comparison of Soil Samples

In this section, the elemental concentrations in soil samples from the respective sites were illustrated, compared and discussed. The results obtained from the comparative analysis of the cadmium concentration in soil samples from the respective sites were illustrated in Figure 5-12. The highest concentration was found in the soil along the WFS. A result that was contrary to results expected by the hypothesis was that the cadmium concentration was found to be higher in the MR irrigation site than the WFS irrigation site.

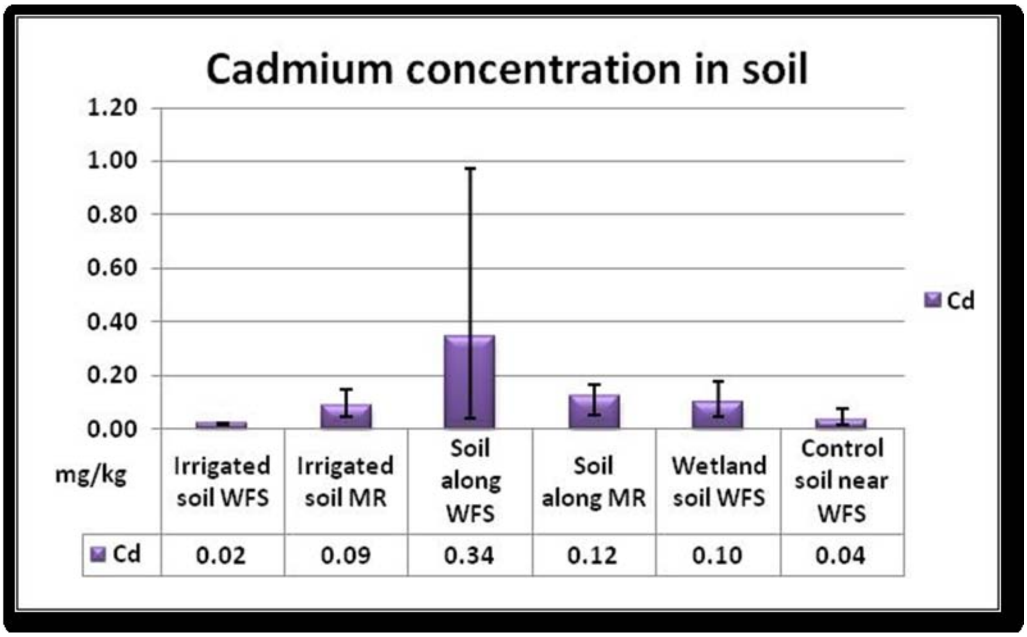


Figure 5-12: Cadmium concentration in the soil samples from the respective sampling sites

The results obtained from the comparative analysis of the lead concentration in soil samples from the respective sites were illustrated in Figure 5-13. The highest concentration was found in the soil from the irrigation site MR, which was also contrary to results expected by the hypothesis. This unexpected result will be discussed in more detail during the comparison of the elemental concentrations from the different particle size fractions of the soil samples that were collected from the respective sites. The second highest lead concentration was found in the soil along the WFS site.

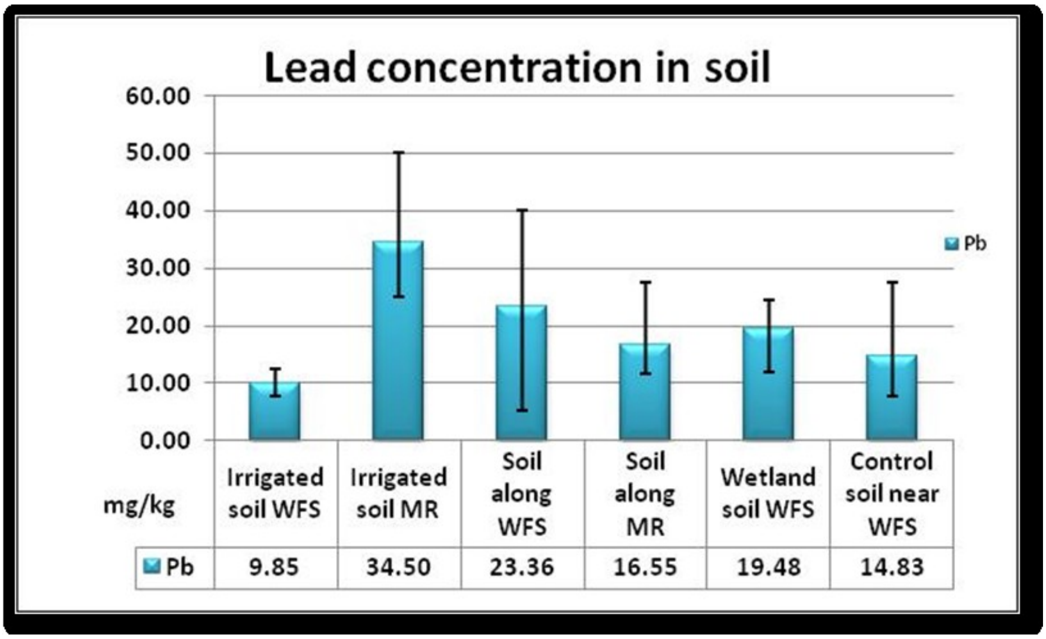


Figure 5-13: Lead concentration in the soil samples from the respective sampling sites

The results obtained from the comparative analysis of the uranium concentration in soil samples from the respective sites were illustrated in Figure 5-14. The highest concentrations were found in the soil along the WFS site and the wetland WFS site, which were both orders of magnitude higher than the rest of the sites.

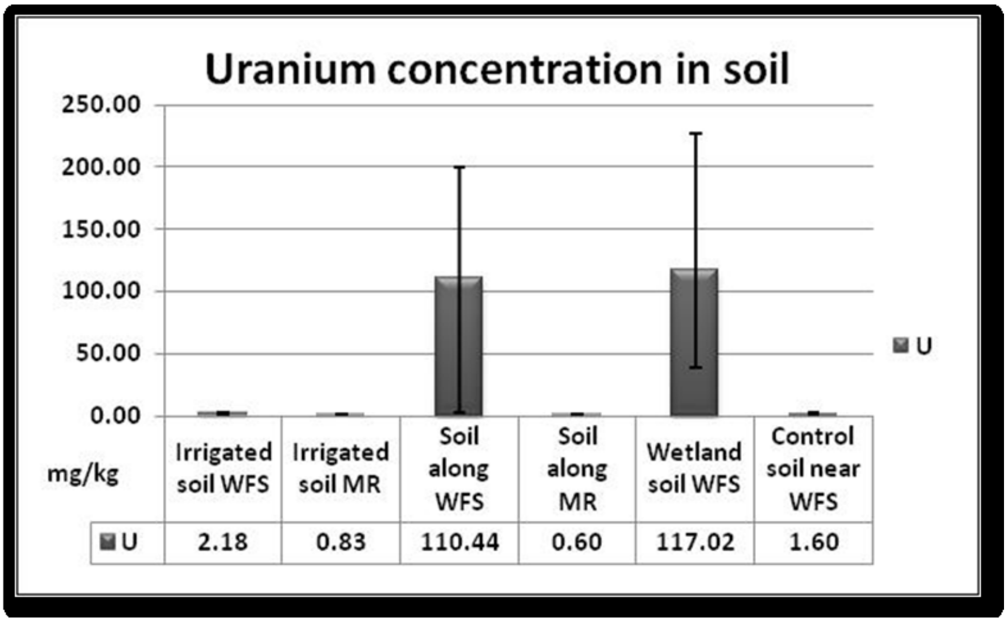


Figure 5-14: Uranium concentration in the soil samples from the respective sampling sites

The other elements, which can be found in Appendix C, were found to have the highest concentrations in the soil along the WFS site. All the elemental concentrations, except uranium, were found to be higher in the irrigated soil MR site than that of the irrigated WFS site. This was truly unexpected and contrary to results expected by the hypothesis as it would be very unlikely that these elemental concentrations were a result of irrigation from the MR, as the water samples from the MR were found to contain exceptionally low elemental concentrations. The higher elemental concentrations had to arise from source other than the water, such as the parent rock or other anthropogenic activities. A comparison of the elemental concentrations that were found in the irrigation MR site and soil along MR site revealed fairly similar concentrations, which creates the impression that these concentrations are typical of the area or have been produced by the same source. Although the experimental and control groups are both located on geological formations, there is a possibility that the soils from the respective groups were formed form different parental rock.

The practically significant differences between the elemental concentrations in the soil samples from the irrigated WFS site and irrigated MR site; and the elemental concentrations in the soil samples from the soil along WFS site and the soil along MR sites were illustrated Table 5-9.

Table 5-9: The practically significant differences between the elemental concentrations in soil samples from the irrigated pastures in the WFS and MR; and the elemental concentrations in the soil samples along the WFS and along the MR

Irrigated soil WFS vs. Irrigated soil MR			Soil along WFS vs. Soil along MR		
Element	**Effect size**	**Practical significance**	**Element**	**Effect size**	**Practical significance**
Cobalt	NA	NA	**Cobalt**	9.20	Large
Nickel	NA	NA	**Nickel**	28.95	Large
Copper	NA	NA	**Copper**	2.89	Large
Zinc	NA	NA	**Zinc**	108.83	Large
Selenium	NA	NA	**Selenium**	5.61	Large
Cadmium	NA	NA	**Cadmium**	5.50	Large
Lead	NA	NA	**Lead**	1.07	Large
Uranium	3.21	Large	**Uranium**	915.33	Large

The results obtained from the comparative analysis of the elemental concentrations in soil samples from the irrigated WFS site and irrigated MR site revealed that the uranium was the only element of interest to have a higher concentration in the irrigated WFS site. The other elemental concentrations were higher in the irrigation MR site, which was contrary to the results expected by the hypothesis and therefore were denoted with *ND* to indicate that these difference between the two sites are not practically significant. The uranium concentration did however produce a difference that was found to be practically significant.

The results obtained from the comparative analysis of the elemental concentrations in soil samples from the soil along WFS site and the soil along MR site revealed a difference that was practically significant for all elements of interest, with uranium, zinc and nickel concentrations portraying the largest effect sizes.

The practically significant differences between the elemental concentrations in the soil samples from the wetland WFS and control WFS sites; and the elemental concentrations in the soil samples from the soil along WFS and control WFS sites were illustrated in Table 5-10.

Table 5-10: The practically significant differences between the elemental concentrations in soil samples from the wetland WFS site and the control WFS site; and the elemental concentrations in the soil along the WFS site and the control WFS site

Wetland WFS vs. Control WFS			Soil along WFS vs. Control WFS		
Element	**Effect size**	**Practical significance**	**Element**	**Effect size**	**Practical significance**
Cobalt	0.24	Medium	**Cobalt**	68.16	Large
Nickel	NA	NA	**Nickel**	52.01	Large
Copper	0.45	Medium	**Copper**	7.73	Large
Zinc	15.70	Large	**Zinc**	464.64	Large
Selenium	1.38	Large	**Selenium**	12.50	Large
Cadmium	3.00	Large	**Cadmium**	15.00	Large
Lead	0.63	Medium	**Lead**	1.16	Large
Uranium	183.21	Large	**Uranium**	172.76	Large

The results obtained from the comparative analysis of the elemental concentrations in soil samples from the wetland WFS site and control WFS site revealed that the zinc, selenium, cadmium and uranium concentrations had differences that were practically significant, with the uranium concentration having the largest effect size.

The results obtained from the comparative analysis of the elemental concentrations in soil samples from the soil along WFS site and the control WFS site revealed that all the elemental concentrations had differences that were practically significant, with zinc, uranium, cobalt and nickel concentrations having the largest effect sizes.

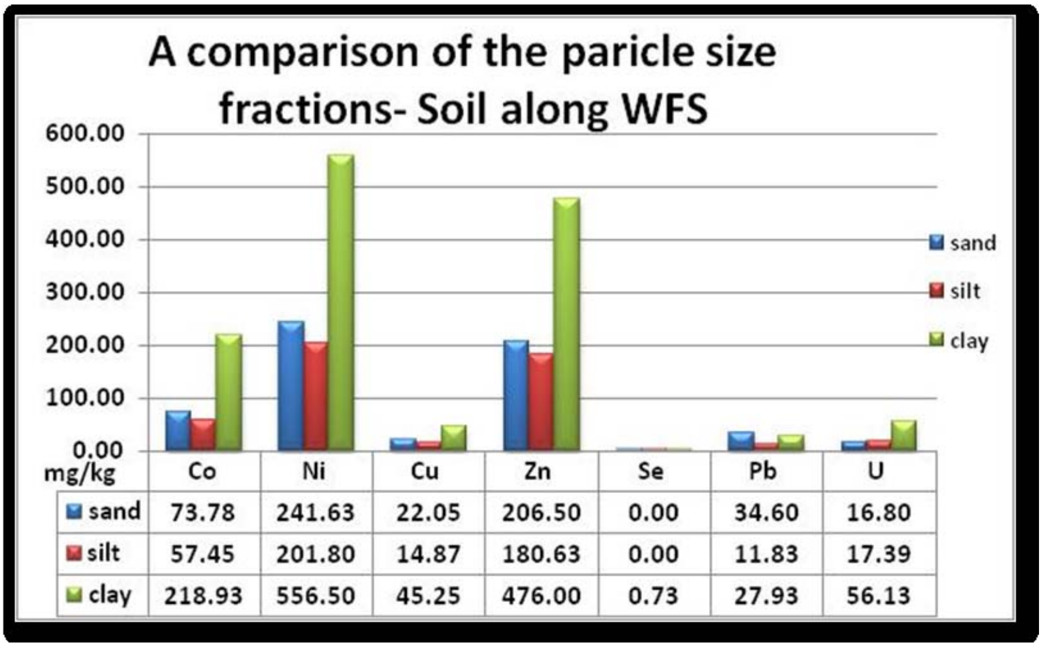
5.4.3 Comparison of the Elemental Concentration within the different particle size fractions of Soil Samples from the Respective Sites

As mentioned earlier, soil is known to be the primary source of all elements that are found in plants, and plants have a limited ability for the selective uptake of elements. This means that plants will take up any elements that are available to them from the soil. Most of the elements that are available to plants are found in the mineral crystalline structure of secondary soil minerals, which are found in the clay fraction. It is therefore necessary to establish the elemental composition and concentrations within the different soil fractions as secondary soil minerals are generally found in the clay sized fraction of soil (Brady & Weil, 2008).

Clay is just a general term for inorganic material that is smaller than 0.002 mm in size; this is not to be confused with clay minerals (synonym for secondary soil minerals) that occur in this clay sized fraction. Secondary soil minerals not only contain elements in their structure, but they also have the ability to bind charged cations (ions or elements with a positive charge) on their surface (Sparks, 2003). The method for establishing the elemental concentrations which are readily available to plants is a 1:2 water extraction method (1 part soil, 2 parts water). This method will however, not include both the elemental cations within the structure of secondary soil minerals, and the elemental cations that are bound onto their surfaces.

In order to quantify the total concentration of elemental cations that are available to plants, a total acid digestion of the clay fraction has to be done. One soil sample from each of the six sites was collected and separated into the different fractions by drying, grinding and sieving without the addition of any chemicals. The sand, silt and clay fractions were digested with an acid mixture and analysed separately. This method may produce results with a slight over estimation of the available elemental cations, as all soil minerals (not only the secondary soil minerals but also primary soil minerals) in the clay sized fraction will be included. Primary soil minerals generally dominate the sand and silt fractions, but small quantities are also found in the clay fraction. The primary soil minerals do not have the ability to bind cations onto their surface and the elements that are contained within their structure are not available for uptake by plants (Brady & Weil, 2008) and (Sparks, 2003). By analysing the different fractions separately, a good understanding of the available elemental concentrations could be obtained.

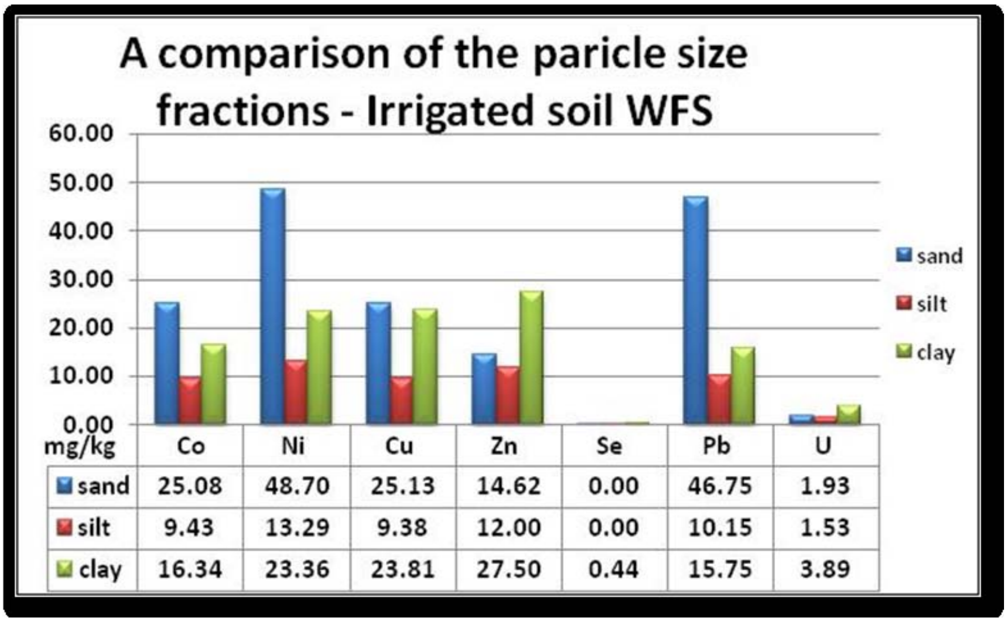
Due to the similarity between certain sites only a few of the sites will be displayed in this section, the rest of the graphs can be found in Appendix C. Figure 5-15 displays the elemental concentrations in the sand, silt and clay fractions of the soil sample from the soil along WFS site. All the elements, excluding lead, displayed the highest concentration in the clay fraction. Soil samples from the soil along MR site (Figure 11-10) and the wetland WFS site (Figure 11-13) showed similar results and can be found in Appendix C.



mg/kg	Co	Ni	Cu	Zn	Se	Pb	U
🟦 sand	73.78	241.63	22.05	206.50	0.00	34.60	16.80
🟥 silt	57.45	201.80	14.87	180.63	0.00	11.83	17.39
🟩 clay	218.93	556.50	45.25	476.00	0.73	27.93	56.13

Figure 5-15: A comparison of the elemental concentrations in the different particle size fractions (sand, silt and clay) in a soil samples from the soil along the WFS

The elemental concentrations in the sand, silt and clay fractions of the soil samples from the irrigated MR site are displayed in Figure 5-16. Cobalt, nickel, copper, cadmium and lead concentrations were all found to be highest in the sand fraction. The soil sample from the control WFS site (Figure 11-14), and irrigation WFS site (Figure 11-11) showed similar results, which could mean that this is natural back ground values or a result of agricultural practises such as the addition of lime and fertilizers. These graphs can also be found in Appendix C.



mg/kg	Co	Ni	Cu	Zn	Se	Pb	U
sand	25.08	48.70	25.13	14.62	0.00	46.75	1.93
silt	9.43	13.29	9.38	12.00	0.00	10.15	1.53
clay	16.34	23.36	23.81	27.50	0.44	15.75	3.89

Figure 5-16: A comparison of the elemental concentrations in the different fractions (sand, silt and clay) in a soil samples from the soil under irrigation in the MR (control site)

Table 5-11 illustrates the different particle size composition of the soil samples from the six respective sites. All of the sites consisted mostly of silt sized soil except for the control WFS site, which was dominated by sand sized soil.

Table 5-11: The different particle size composition of the soil samples of the respective sites

Site	Sand	Silt	Clay
Soil along WFS	30.42	57.02	12.56
Soil along MR	19.60	55.13	25.28
Irrigation WFS	16.15	68.63	15.22
Irrigation MR	23.24	63.30	13.46
Wetland WFS	6.29	50.70	43.01
Control WFS	74.30	19.80	5.90

An estimation of the plant available elemental concentrations can be made by multiplying the elemental concentrations in the clay fraction with the percentage of clay in each soil sample. The results for each site and each element are displayed in Table 5-12. The soil samples from the soil along WFS site were typified by the highest cobalt, nickel, zinc and cadmium concentrations and the soil samples from the wetland WFS was typified by the highest copper, selenium and lead concentrations.

Table 5-12: An estimation of the concentration of elements that are available to plants

Element	Irrigation WFS	Irrigation MR	Soil along WFS	Soil along MR	Wetland WFS	Control WFS
Co (mg/kg)	2.49	2.38	27.50	9.65	3.27	0.48
Ni (mg/kg)	3.55	2.83	69.90	23.78	36.59	1.20
Cu (mg/kg)	3.62	2.83	5.68	11.81	14.11	0.91
Zn (mg/kg)	4.19	4.72	59.79	4.36	23.91	0.66
Se (mg/kg)	0.07	0.03	0.09	0.04	0.28	0.06
Cd (mg/kg)	0.01	0.01	0.04	0.02	0.02	0.00
Pb (mg/kg)	2.40	3.23	3.51	3.08	9.19	0.45
U (mg/kg)	0.59	0.19	7.05	0.22	86.39	0.06

The results only serve as an indication as it does not include other factors which will influence elemental availability, such as pH, oxidation state, cation exchange capacity, organic matter content, plant species, plant growth stage etc. (Brady & Weil, 2008).

5.5 Vegetation

Plants have a limited capacity for the selective uptake of essentially beneficial elements. They are also able to take up and accumulate, sometimes to very high levels, certain elements which are not necessary for growth and which may even be toxic (Marschner, 1997). Due to the great genetic variability that is found among populations of plants, there will be individuals that have the ability to survive and even prosper in highly contaminated soils.

5.5.1 Elemental Concentrations commonly found in Natural Pastures

The elemental concentrations of natural pastures in the UK were provided by Minson (1990), Kabata *et al*. (1992), Whelan *et al*., (1994), Archer and Hodgson (1987) and Bacon *et al*. (1992) as found in Underwood & Suttle, (2001) and displayed in Table 5-13. The elemental concentrations that were found in grass samples from the respective sampling sites were compared to the concentration ranges found in natural pastures and can be found in Table 12-1 in Appendix D.

Table 5-13: Elemental concentrations of natural pastures

Element	Normal pastures
Cobalt	0.02 – 0.22 mg/kg
Nickel	0.13 – 1.1 mg/kg
Copper	4.7 – 7.8 mg/kg
Zinc	7 – 100 mg/kg
Selenium	<0.05 mg/kg
Cadmium	<0.1 mg/kg
Lead	rarely exceeds 5 mg/kg

The cobalt and nickel concentrations that were found in the grass samples from the respective sites were all found to exceed the concentration ranges found in natural pastures. Bergmann (1992) revealed a wider cobalt concentration range (0.01 – 0.4 mg/kg) which was still exceeded by all sites except the control WFS site. It seems that grasses in both the experimental and control groups have the ability to accumulate cobalt at rates far beyond expected. Evans *et al*. (1979) found 6.3 mg/kg nickel in grasses that were irrigated with municipal sewage water, and although this concentration is much higher (almost 6 times higher) than that of the nickel concentration found in natural pastures, the nickel concentration in grass samples from the soil along WFS, irrigation WFS and wetland WFS sites were found at even higher concentrations. The grass samples from the soil along WFS site contained 65.12 mg/kg cobalt which is orders of magnitudes higher than the results found in natural pastures and grasses that were irrigated with municipal sewage water.

All the sites except for the wetland WFS site contained grass samples with copper and selenium concentrations that exceeded those found in natural pastures. According to Kluge & Podlesale (1984) as quoted by Bergmann (1992) it is not unusual for plants to contain 20 – 30 mg/kg copper, a range in which all the respective sampling site fall within. Underwood & Suttle (2001) also reported that most pastures generally contain less than 0.05 mg/kg selenium, which is significantly lower than that the selenium concentration that was found in grass samples from the respective sites. The cadmium and zinc concentrations in the grass samples from the soil along WFS site were found to exceed the respective concentration ranges found in natural pastures. Evans *et al*., (1979) found that grasses that were irrigated with municipal sewage water contained 1.1 mg/kg cadmium, which is higher than the cadmium concentration in the soil along WFS site and 171 mg/kg zinc, which is lower than the zinc concentration found in the soil along WFS site. This could serve as an indication that the grass samples from the soil along WFS have accumulated a very large zinc concentration.

The lead concentrations in grass samples from all the respective sites were found to be within the range found in natural pastures.

The normal uranium concentration found in irrigated or natural grasses could not be found in an extensive search. Dreesen *et al*. (1982) reported 0.16 mg/kg uranium in grasses and 1.8 mg/kg uranium in shrubs that grew on soil-covered tailings material. All the sites in the experimental group, including the control WFS site, drastically exceeded these concentrations, which may suggest that the grasses in the experimental sites have been exposed to elevated uranium concentrations.

5.5.2 Elemental Concentrations in Grass Samples from the Respective Sites

In this section, the elemental concentrations in grass samples from the respective sites will be illustrated, compared and discussed. The cadmium concentrations in the grass samples were illustrated in Figure 5-17. The highest concentration was found in the grass samples from the soil along WFS site. It was also interesting to note that the second highest cadmium concentration was found in the control site WFS.

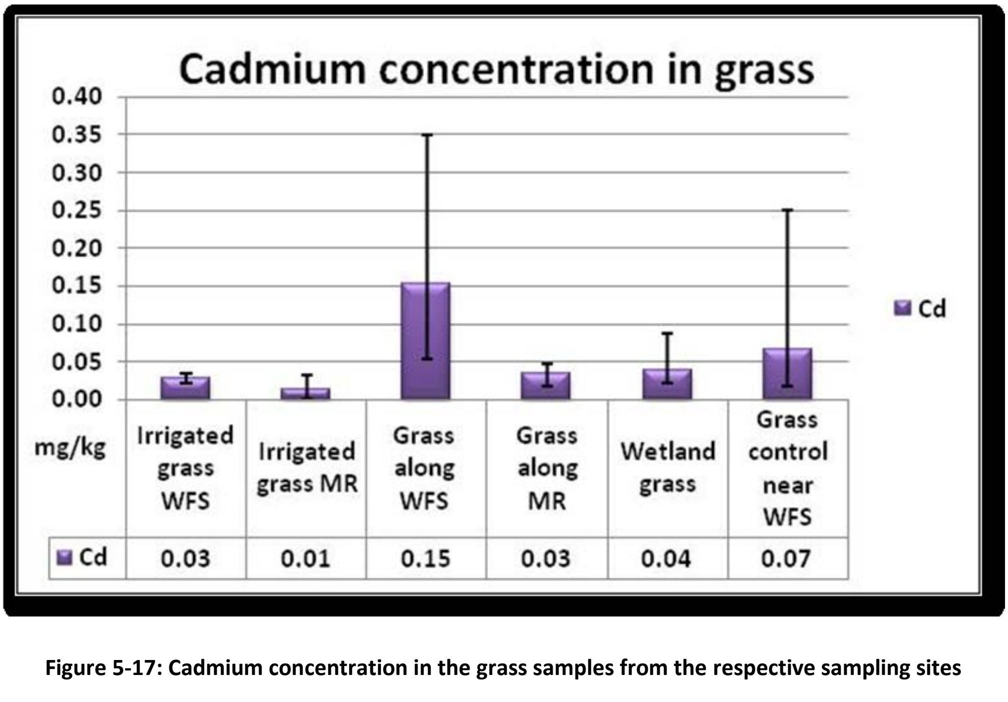


Figure 5-17: Cadmium concentration in the grass samples from the respective sampling sites

The lead concentrations in the grass samples were illustrated in Figure 5-18, with the highest concentration found in the grass samples from the irrigation WFS site. This site was typified with a very high maximum value, and a minimum value that is near or above the average concentrations in the other sites. The lead concentration in the grass form the irrigation WFS site is 5.9 timed higher than that in the irrigation MR site, which is interesting as the highest lead concentration in soil was found in the soil under irrigation in the MR.

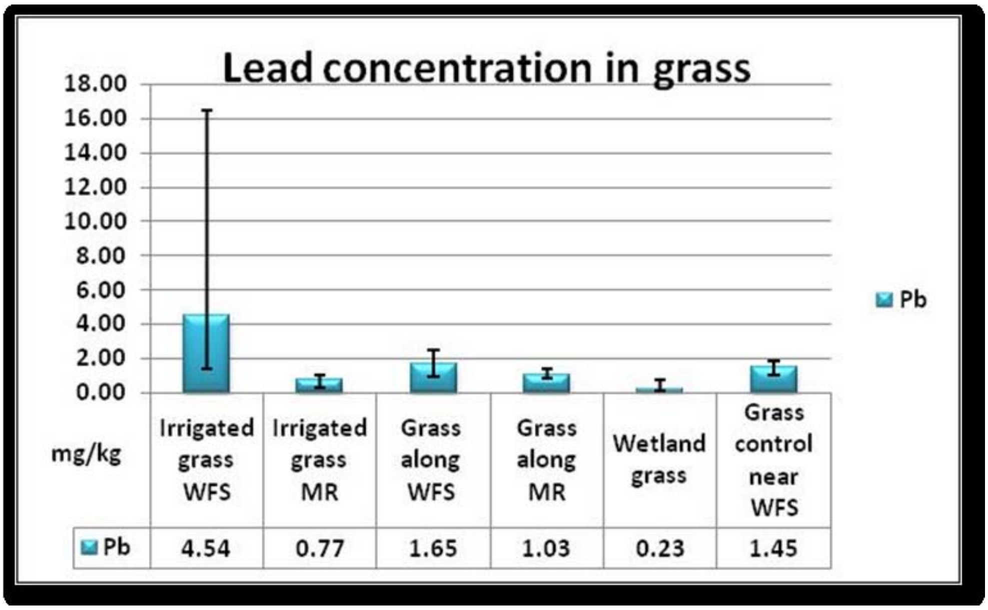


Figure 5-18: Lead concentration in the grass samples from the different sampled sites

Although the concentration of lead in soil was found to be highest in the irrigated MR site (Figure 5-13), the lead concentration in the grass was second to lowest in this site. The particle size distribution of this soil sample indicated that the highest lead concentration was found in the sand sized fraction (Figure 5-16). This example could serve as proof to the point that the majority of plant available elements are primarily found in the clay sized fraction of soil sample due to the dominance of secondary soil minerals.

The uranium concentrations in the grass samples were illustrated in Figure 5-19. The highest concentration was found in the grass samples from the soil along WFS site, with a maximum value of 350 mg/kg. This site was typified with a uranium concentration that was 1002.5 times higher than that of the grass samples from the soil along MR site. The grass samples from the irrigation WFS site also contained a high uranium concentration, being 396.25 times higher than that of the grass samples from the irrigation MR site.

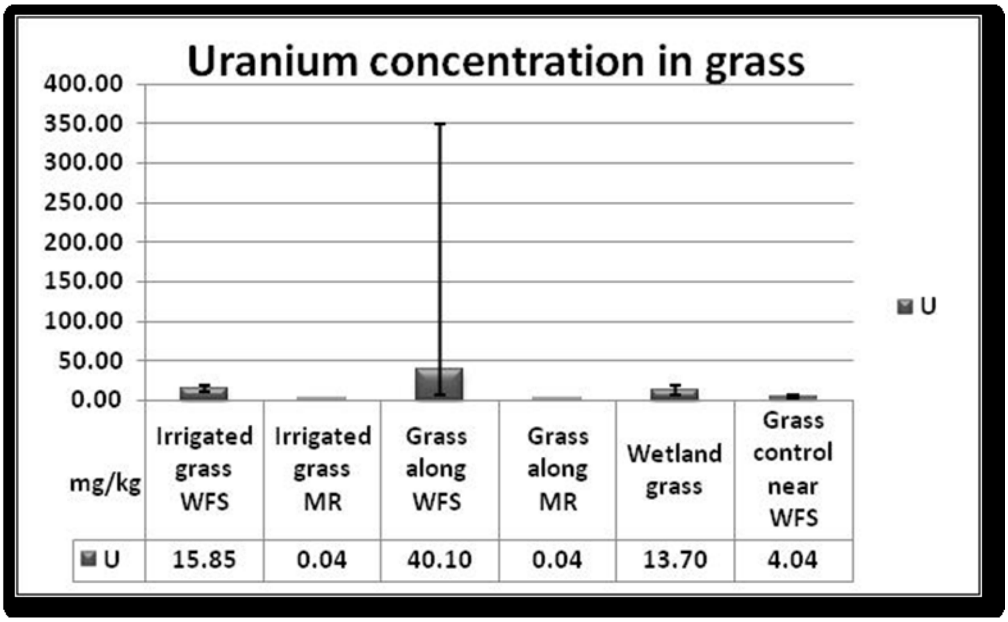


Figure 5-19: Uranium concentration in the grass samples from the different sampled sites

Most of the other elements of interest displayed similar results with highest elemental concentrations found in grass samples from the soil along WFS site. The graphs of the other elements of interest can be found in Appendix D.

The cobalt concentration was found to be highest in the grass samples from the soil along WFS site (Figure 12-1), being 5.98 times higher than that of the grass samples from the soil along MR site.

The nickel concentration was found to be highest in the grass samples from the soil along WFS site (Figure 12-2); this concentration was 11.04 times higher than that of the grass samples from the soil along MR site. The nickel concentration in the grass samples from the irrigation WFS site was 2.93 times higher than that of the grass samples from the irrigation MR site.

The copper concentration was found to be highest in the grass samples from the soil along WFS site (Figure 12-3); this concentration was 2.57 times higher than that of the grass samples from the soil along MR site. The other sites did not exhibit significant variations.

The zinc concentration was found to be highest in the grass samples from the soil along WFS site (Figure 12-4); this concentration was 11.17 times higher than that of the grass along the MR. The other sites did not exhibit significant variations.

The selenium concentration was found to be highest in the grass samples from the soil along WFS site (Figure 12-5); this concentration was 3.79 times higher than that of the grass samples from the soil along MR site. The selenium concentration in the grass from the irrigation WFS site was 2.28 times higher than that of the grass from the irrigation MR site.

The practically significant differences between the elemental concentrations in the grass samples from the irrigation WFS and irrigation MR sites; and the elemental concentrations in the grass samples from the soil along WFS and soil along MR sites were illustrated in Table 5-14.

Table 5-14: The practically significant difference between the elemental concentrations in the grass samples from the irrigation WFS and irrigation MR sites; and the elemental concentrations in the grass samples from the soil along WFS and soil along MR sites

Irrigation WFS vs. irrigation MR			Grass along WFS vs. grass along MR		
Element	**Effect size**	**Practical significance**	**Element**	**Effect size**	**Practical significance**
Cobalt	1.97	Large	Cobalt	12.35	Large
Nickel	3.56	Large	Nickel	61.69	Large
Copper	1.66	Large	Copper	2.86	Large
Zinc	0.41	Medium	Zinc	40.13	Large
Selenium	1.56	Large	Selenium	2.66	Large
Cadmium	2.00	Large	Cadmium	12.00	Large
Lead	13.00	Large	Lead	2.70	Large
Uranium	527.00	Large	Uranium	801.20	Large

The results obtained from the comparative analysis of the elemental concentration in grass samples from the irrigation WFS and irrigation MR sites revealed that all elemental concentrations except for that of zinc had a difference that was practically significant, with the uranium concentration having the largest effect size.

The results obtained from the comparative analysis of the elemental concentrations in grass samples from the soil along WFS and soil along MR sites revealed that all elemental concentrations had a difference that was practically significant. Uranium, nickel and zinc concentrations having the largest effect sizes. Considering that a large effect size is achieved at a value equal to or greater than 0.8, the uranium concentration therefore had a massive difference in both comparisons.

The practically significant differences between the elemental concentrations in the grass samples from the wetland WFS and control WFS sites; and the elemental concentrations in the grass samples from the soil along WFS and control WFS sites were illustrated in Table 5-15.

Table 5-15: The practically significant difference between the elemental concentrations in the grass samples from the wetland WFS and control WFS sites; and the elemental concentrations in the grass samples from the soil along WFS and control WFS sites

Wetland WFS vs. Control WFS			Grass along WFS vs. Control WFS		
Element	**Effect size**	**Practical significance**	**Element**	**Effect size**	**Practical significance**
Cobalt	57.0	Large	Cobalt	235.33	Large
Nickel	18.4	Large	Nickel	116.34	Large
Copper	NA	NA	Copper	2.53	Large
Zinc	NA	NA	Zinc	28.90	Large
Selenium	NA	NA	Selenium	7.54	Large
Cadmium	NA	NA	Cadmium	0.89	Large
Lead	NA	NA	Lead	0.69	Medium
Uranium	6.9	Large	Uranium	25.76	Large

The results obtained from the comparative analysis of the elemental concentrations in grass samples from the wetland WFS and control WFS sites revealed that only the cobalt, nickel and uranium concentrations had differences that were practically significant, with the cobalt concentration having the largest effect size. The results obtained from the comparative analysis of the elemental concentrations in the grass samples from the soil along WFS and control WFS sites revealed that all the elemental concentrations except for the lead concentration had a difference that was practically significant. The cobalt, nickel and zinc had the largest effect sizes.

5.6 Cattle

Trace elements exist in the animal body's cells and tissues in a variety of functional and chemical combinations (Underwood & Suttle, 2001). Soil, water and vegetation are the primary sources of all elements found in animal tissues. Livestock usually derive a high proportion of their trace elements from the feed and vegetation that they consume (Underwood & Suttle, 2001).

In this section, the elemental concentrations of tissue samples from cattle grazing in the experimental group and cattle grazing in the control group were illustrated, compared with each other and that the elemental concentrations normally found in cattle. The elemental concentrations that were found in the respective tissue samples from both groups of cattle exhibited a large standard deviation. This does not necessarily indicate a low repeatability, as the cattle do not all graze on the exact same pastures. Certain heavy metals have a low retention time and may be excreted from the body within a few days. For example, it takes 11 days for half of the uranium to leave the bones, and 2-6 days for the kidneys (ATSDR, 2011). For this reason, grazing habits of the cattle prior to slaughtering play a significant role in elemental concentrations within the respective tissue samples. As the cattle were not restricted to graze solely on homogenous pastures, large standard deviations within the elemental concentrations were to be expected. Liver, kidney, spleen, muscle tissue, bone and hair samples were collected from cattle during the slaughtering process at a local abattoir.

5.6.1 Elemental Concentrations commonly found in Cattle

The elemental concentrations found in cattle samples were illustrated in Table 5-16. These ranges were found predominantly in Pulse (1994) except for the cobalt concentration found in the muscle tissue, which was found in ATSDR (2004), and the uranium concentration found in liver, kidney and muscle tissue, which was found in ATSDR (2011).

Table 5-16: Normal elemental concentrations found in cattle liver, kidney and meat samples as provided by Pulse (1994)

Element	Liver (fresh weight)	Kidney (fresh weight)	Meat (fresh weight)
Cobalt (mg/kg)	0.02 – 0.085	0.071	0.001[a]
Nickel (mg/kg)	0.1 – 0.6	0.15 – 0.50	0.35 – 0.4
Copper (mg/kg)	25 - 100	4.0 – 6.0	1.2 – 1.5
Zinc (mg/kg)	25 - 100	18 - 25	20 – 247
Selenium (mg/kg)	0.25 – 0.5	1.0 – 1.5	0.07 – 0.15
Cadmium (mg/kg)	0.02 – 1.0	0.05 – 1.5	0.004 – 0.024
Lead (mg/kg)	0.1 – 1.0	0.2 – 2.0	0.04 – 0.15
Uranium (mg/kg)	0.026[b]	0.07[b]	0.014[b]

[a] Found in Jorhem & Sundström 1993 as quoted by ATSDR (2004).
[b] Found in EPA (1985:j) as quoted by ATSDR (2011).

The elemental concentrations that were found in the samples from the cattle in the experimental group (Table 13-1) and control group (Table 13-2) can be found in Appendix E. The data provided in Table 5-16 was for the elemental concentrations of fresh weight samples, unlike the samples in this study which were 100 % dry weight. The sample preparation used in this study revealed that liver samples contained 66.28% water, kidney samples contained 67.43 % water and the muscle tissue samples contained 74.62 % water. Compensating for the dry weight, a comparison to Table 5-16 revealed that:

- The nickel, cadmium and lead concentration that were found in the cattle liver, kidney and muscle tissue samples from both the experimental and control groups were found to be within the ranges normally found in cattle.

- Cobalt concentrations found in the liver and muscle tissue samples of cattle from both the experimental and control groups exceeded the normal ranges, and the cobalt concentrations found in the kidney samples from the experimental group exceeded the normal range.

- The copper concentration found in the kidney samples from the cattle in the experimental group exceeded that of the normal concentration range.

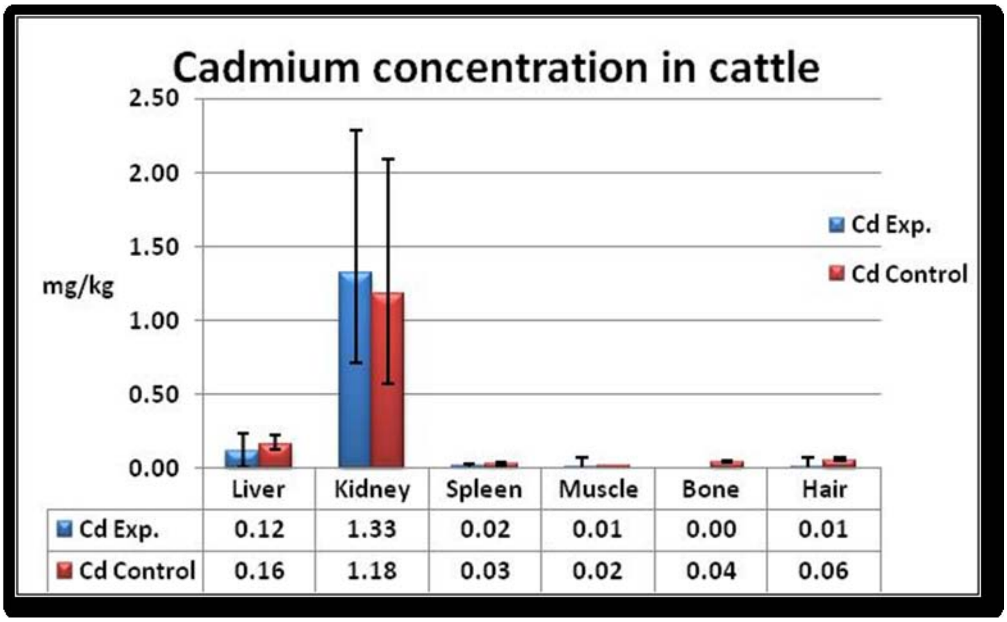
- The zinc concentration found in the liver and kidney samples from the cattle from the experimental group, and the kidney samples from the cattle from the control group exceeded the normal range.

- The selenium concentration found in the liver, kidney and muscle tissue samples in the cattle from the experimental group, and the kidney samples from the cattle in the control group exceeded the normal range.

- The uranium concentration found in the liver, kidney and muscle tissue samples in the cattle from the experimental group exceeded the normal range.

5.6.2 Elemental Concentrations found in the Cattle from the Experimental and Control Groups

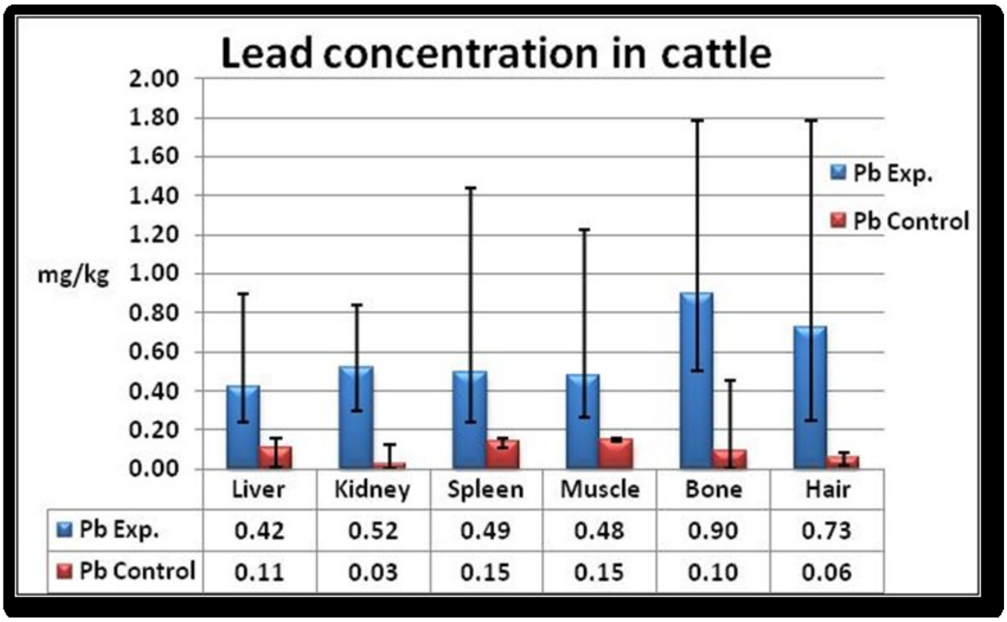
The cadmium concentration of the different cattle tissue samples were illustrated in Figure 5-20. The control and experimental groups were found to be very similar, and in all cases except the cadmium concentration found in the kidney of the control samples were higher than that of the experimental samples. The highest concentration was found in the kidney samples, as predicted in the literature review (ATSDR, 2008).



	Liver	Kidney	Spleen	Muscle	Bone	Hair
🟦 Cd Exp.	0.12	1.33	0.02	0.01	0.00	0.01
🟥 Cd Control	0.16	1.18	0.03	0.02	0.04	0.06

Figure 5-20: Cadmium concentration in cattle tissue samples. Control group in red, experimental group in blue

The lead concentration of the different cattle tissue samples were illustrated in Figure 5-21. Lead accumulated throughout all sampled tissues, as predicted by the literature (Spitz & Trudinger, 2009). The experimental group was found to contain the highest concentrations in all tissue sample comparisons, with the highest concentration found in the bone, 9 times higher in the experimental group.



	Liver	Kidney	Spleen	Muscle	Bone	Hair
🟦 Pb Exp.	0.42	0.52	0.49	0.48	0.90	0.73
🟥 Pb Control	0.11	0.03	0.15	0.15	0.10	0.06

Figure 5-21: Lead concentration in cattle tissue samples. Control group in red, experimental group in blue

The uranium concentration in the different cattle tissue samples were illustrated in Figure 5-22. This proved to be a very interesting result due to the large differences that were observed between the control and experimental groups, and the array of unanticipated tissue concentrations. Uranium was expected to accumulate predominantly in the bone, liver and kidney samples (ATSDR, 2011). Conversely, the highest uranium concentrations in the cattle samples from the experimental group were found in the liver, kidney, spleen and muscle samples. A comparison of the two groups reveals that the uranium concentration in the experimental group was 126.75 times higher in the liver, 4350 times higher in the kidney, 47.75 times higher in the spleen, 31.6 times higher in the muscle tissue, 60 times higher in the bone and 129 times higher in the hair that that of the control group.

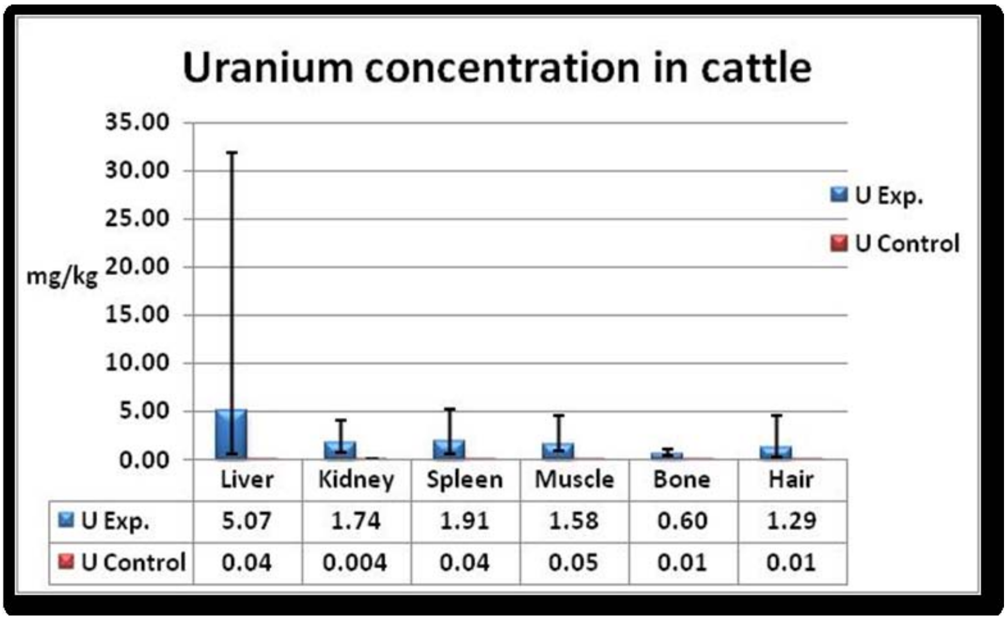


Figure 5-22: Uranium concentration in cattle tissue samples. Control group in red, experimental group in blue

The graphs of the other elements of interest can be found in Appendix E.

The cobalt concentrations in the respective samples (Figure 13-1) did not differ a great deal between the two groups. The concentrations in the liver, bone and hair were higher in the control group and the concentrations in the kidney, spleen and muscle were higher in the experimental group.

The nickel concentrations in the respective samples (Figure 13-2) were all found to be higher in the experimental group, with kidney (387.5 times higher), bone (167.5 times higher) and hair (76.5 times higher) samples showing the greatest difference.

The copper concentrations in the respective samples (Figure 13-3) did not differ a great deal between the groups, and the highest concentrations were found in the liver samples. All samples except hair were found to be higher in the experimental group.

The zinc concentrations in the respective samples (Figure 13-4) were all found to be higher in the experimental group, with the highest concentrations found in the liver (2.9 times higher), muscle tissue (1.3 times higher), and kidney (1.9 times higher) samples.

The selenium concentrations in the respective samples (Figure 13-5) were all found to be higher in the experimental group, with the highest concentrations found in the liver (1.5 times higher) and spleen (3.2 times higher) samples.

The practically significant differences between the elemental concentrations in the cattle samples from the control and experimental groups were illustrated in Table 5-17 and Table 5-18.

Table 5-17: The practically significant difference between the elemental concentrations in liver, kidney and spleen samples from cattle in the control and experimental group

	Liver	Liver	Kidney	Kidney	Spleen	Spleen
Element	**Effect size**	**Practical significance**	**Effect size**	**Practical significance**	**Effect size**	**Practical significance**
Cobalt	NA	NA	9.59	Large	1.15	Large
Nickel	1.80	Large	4.19	Large	309.20	Large
Copper	0.17	Small	21.33	Large	2.51	Large
Zinc	20.25	Large	10.85	Large	22.24	Large
Selenium	18.87	Large	51.03	Large	3.85	Large
Cadmium	NA	NA	116.95	Large	NA	NA
Lead	5.59	Large	17.53	Large	8.26	Large
Uranium	547.57	Large	103.82	Large	233.01	Large

Table 5-18: The practically significant difference between the elemental concentrations in muscle tissue, bone and hair samples from cattle in the control and experimental group

	Muscle	Muscle	Bone	Bone	Hair	Hair
Element	**Effect size**	**Practical significance**	**Effect size**	**Practical significance**	**Effect size**	**Practical significance**
Cobalt	8.75	Large	NA	NA	NA	NA
Nickel	14.92	Large	4006.20	Large	3419.66	Large
Copper	4.96	Large	3.13	Large	NA	NA
Zinc	1.50	Large	0.98	Large	7.57	Large
Selenium	21.73	Large	22.35	Large	58.15	Large
Cadmium	NA	NA	NA	NA	NA	NA
Lead	43.66	Large	35.57	Large	3.17	Large
Uranium	569.94	Large	940.93	Large	614.44	Large

Cobalt illustrated a practically significant difference in the kidney, spleen and muscle tissue. Nickel illustrated a practically significant difference in all the samples with bone and hair samples differing substantially. Copper illustrated a practically significant difference in kidney, spleen, muscle tissue and bone samples. Cadmium only had a practically significant difference in the kidney samples. Zinc, selenium, lead and uranium all illustrated a practically significant difference in all the samples. The effect sizes for uranium were colossal in all the sample comparisons.

5.7 Cattle Lick and Fertilizer

Although mining may be the primary contributor to metal or elemental pollution in the WFS, there are other contributors such as industries, sewage treatment plants and agricultural activities. Other sources that were identified to be possibly contributors to the elemental load in this study were fertilizers used on the planted pastures and the mineral supplement that was provided to the Cattle. These two sources were identified, sampled (one sample of each) and analysed for elemental concentrations. They did not form part of the detailed investigation and therefore their results, as presented in Table 5-19, will only be discussed briefly.

Table 5-19: Elemental concentrations of the mineral supplement and fertilizer

Element	Mineral supplement	Fertilizer
Co mg/kg	2.08	0.04
Ni mg/kg	3.40	1.50
Cu mg/kg	73.03	5.82
Zn mg/kg	357.25	18.37
Se mg/kg	2.70	0.34
Cd mg/kg	0.49	0.42
Pb mg/kg	0.13	8.58
U mg/kg	31.58	0.04

All elements of interest were found at a variety of concentrations in both samples. The two most interesting concentrations were that of uranium (31.58 mg/kg) in the mineral supplement and that of lead (8.58 mg/kg) in the fertilizer. The lead concentration in the fertilizer could be a contributing factor to the high lead concentration in the irrigated pastures from the WFS (Figure 5-18), and the high lead concentration that was found in the soil samples from the irrigated pastures from the MR (Figure 5-13). These two samples attest the validity of the statement that agricultural practices could be a possible source of contamination.

6 Data Interpretation

6.1.1 *Soil-to-plant Transfer Factors*

So far the elemental concentrations in soil and grass have been established for the various sites. As mentioned previously the migration and accumulation of elements from soil to plants is complex and influenced by many factors. It is not possible to accurately determine the elemental concentrations in soil which are available for uptakes by plants, but the factor of soil elements that transfer into the plants could be calculated by estimation the concentration ratio for elemental concentrations in soil and grass. This ratio is referred to as the "Transfer Factor" and describes the amount of elements expected to enter a plant from its substrate, under equilibrium conditions (Sheppard and Sheppard, 1985).

The Transfer Factor (*TF*) was expressed as:

$$TF = \frac{\text{Element concentration of plant (mg/kg dry weight)}}{\text{Element concentration of soil (mg/kg dry weight)}}$$

The results for all the elements in the different sites are given in Table 6-1.

Table 6-1: Transfer factors for each element in the respective sites

Element	Irrigation WFS	Irrigation MR	Grass along WFS	Grass along MR	Wetland WFS	Control WFS
Cobalt	0.13	0.02	0.04	0.02	0.41	0.08
Nickel	0.42	0.05	0.05	0.03	0.26	0.07
Copper	1.15	0.33	0.25	0.15	0.32	0.41
Zinc	3.12	1.23	0.44	1.03	0.86	2.83
Selenium	2.30	0.98	0.87	2.61	0.33	1.79
Cadmium	1.27	0.16	0.44	0.28	0.38	1.82
Lead	0.46	0.02	0.07	0.06	0.01	0.10
Uranium	7.27	0.05	0.36	0.07	0.12	2.53

From the results presented in Table 6-1 it can be concluded, that there are some important differences between the transfer factors of each element in each site. The highest *TF* for most elements were found in the irrigation WFS site, which is remarkable considering that this site did not have particularly high elemental concentrations in the soil, in comparison with other sites. The delineating factor is the water from the WFS which is used to irrigate this site. Elemental uptake by plants primarily occur by means of soil-root transfer, but there is another mechanism of uptake; foliar uptake, which could explain the higher *TF* in the irrigation WFS site (Marschner, 1997). The mechanism of foliar uptake can be described as the uptake of elements from water that is applied to the above ground portion of the plant. To mention a few examples to support this method of thinking, the Transfer Factors for uranium (Figure 6-1) and lead (Figure 6-2) will be discussed.

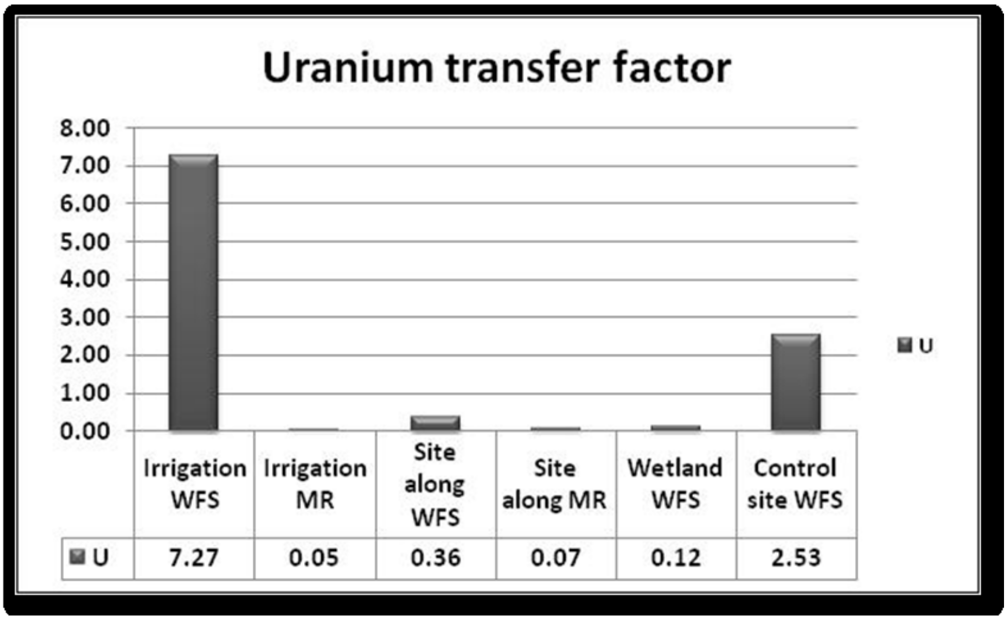


Figure 6-1: Transfer factor for uranium in the various sites

The uranium concentrations in soil and grass were graphically represented in Figure 5-14 and Figure 5-19. The highest transfer factor was found in the irrigation WFS site, where the uranium concentration in the grass samples was found to be 0.46 times higher than the uranium concentration in the soil samples. This transfer factor is much higher than that of the other sites as is evident from the results in Figure 6-2.

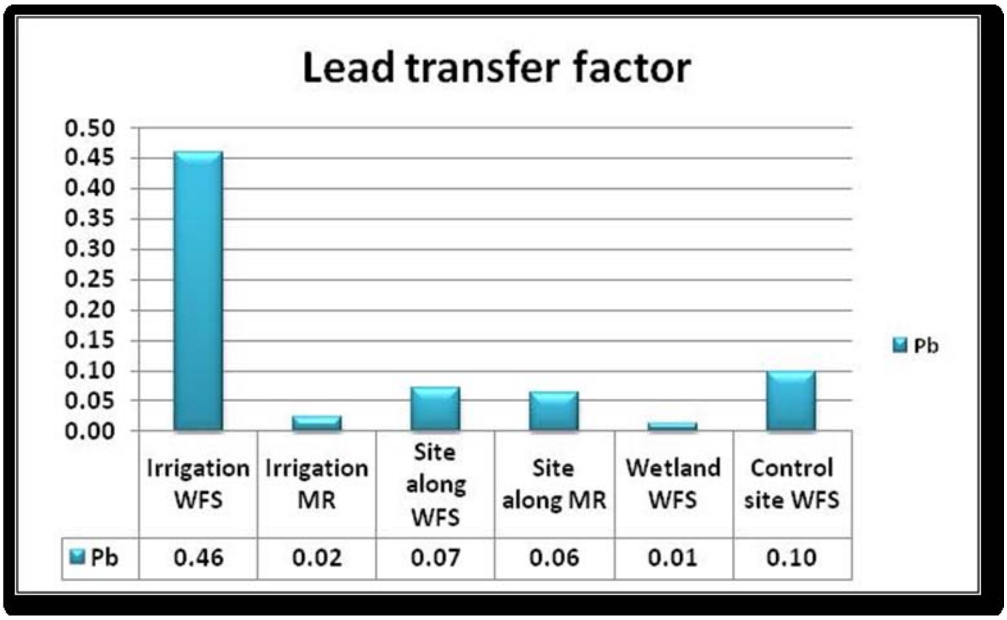


Figure 6-2: The transfer factor for lead in the various sites

The fact that the *TF* for most of the elements were highest in the irrigation WFS site suggests that foliar uptake does play a significant role in the elemental uptake for grass irrigated from the WFS. This could be proved by considering the lead concentration in soil (Figure 5-13) and grass (Figure 5-18) samples from both the irrigation WFS site and the irrigation MR site. The lead concentration in the soil samples from the irrigation MR site was 3.5 times higher than that of the irrigation WFS site, and the lead concentration in the grass samples from the irrigation WFS site was 5.9 times higher than that of the irrigation MR site. Although there are various aspects that play significant roles in the uptake of elements by plants, the delineating factor between these two sites has certainly got to be the elemental concentrations of the water used for irrigation.

The rest of the *TF* graphs for the elements of interest can be found in Appendix D.

6.2 Major Route of Exposure

As mentioned earlier, trace elements exist in the animal body's cells and tissues in a variety of functional and chemical combinations, and soil, water and vegetation are the primary sources of all elements found in animal tissues (Underwood & Suttle, 2001). According to Dickinson *et al*., (2007) a mature beef cow will consume 12.375 kg of grass per day, 38 litres of water per day and about 0.7 kg of soil per day (Mayland *et al*., 1997) and (Mayland *et al*., 1975).

In order to estimate the elemental ingestion for cattle grazing in the experimental site, the following scenario was envisaged:

- Water ingestion: An assumption was made that half the water cattle ingest daily would be undisturbed WFS water and the other half disturbed water from the WFS.
- Grass and soil ingestion: An assumption was made that cattle would graze a third of the day at each of the following sites: site along the WFS, pasture irrigated from the WFS and the control WFS site.

The elemental concentrations for the different samples and sites were used to calculate the total elemental ingestion per day as seen in Table 6-2.

Table 6-2: Elemental ingestion from soil, grass and water

Element	Soil ingestion	Grass ingestion	Water ingestion	Total Ingestion
Co (mg/day)	46.71	37.21	7.93	91.85
Ni (mg/day)	366.73	346.40	48.93	762.05
Cu (mg/day)	29.47	189.82	3.90	223.18
Zn (mg/day)	201.95	1720.47	82.95	2005.37
Se (mg/day)	1.10	23.20	1.23	25.52
Cd (mg/day)	0.10	1.02	0.16	1.28
Pb (mg/day)	12.01	31.49	0.62	44.12
U (mg/day)	28.55	247.47	4.06	280.08

The major route of ingestion for all elements of interest except cobalt and nickel was found to be via grass, as shown Figure 6-3 which illustrates the percentile of total uranium ingestion by means of grass, soil and water.

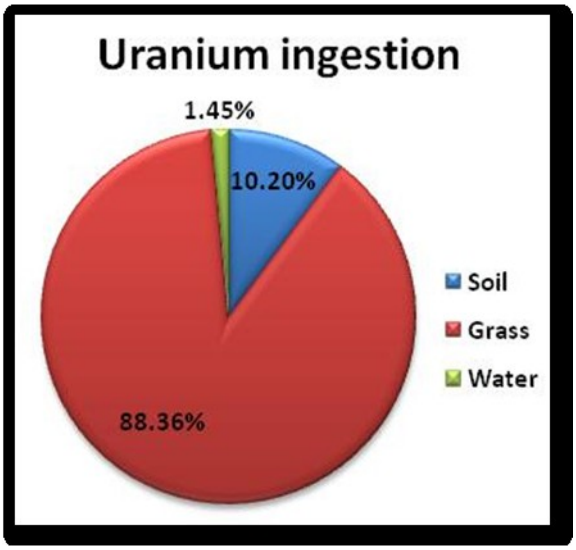


Figure 6-3: Uranium ingestion percentages for soil, grass and water

The major route of ingestion for nickel and cobalt was found to be from soil, the nickel ingestion percentiles were illustrated in Figure 6-4. Elemental ingestion by means of water appears to be less significant when compared to grass and soil; this could be attributed to the low elemental concentrations that were found in the water samples.

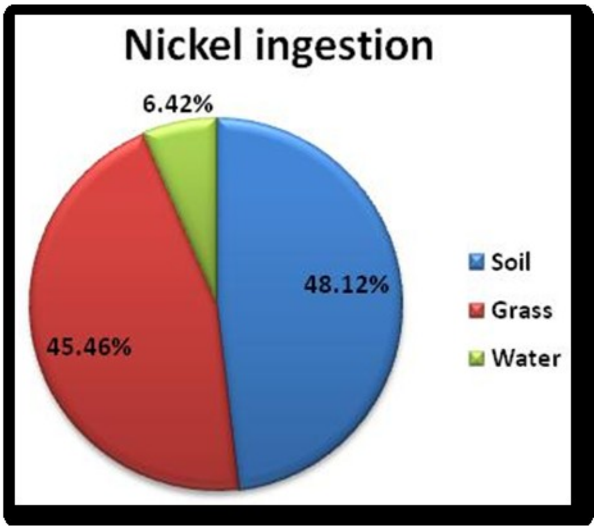


Figure 6-4: Nickel ingestion percentages for soil, grass and water

Although this scenario was not based on actual ingestion and grazing facts, it could serve as an indication of elemental ingestion rates and produce means to reduce elemental ingestion if it should be required. The ingestion percentile graphs for the other elements of interest can be found in Appendix F.

6.3 Cattle consumption model for the Wonderfonteinspruit related to human health risk

Cattle feed not only impacts on animal health but more importantly on human health (Van der Fels-Klex *et al.*, 2011). It is therefore crucial to analyse the elemental concentrations found within cattle feed in areas exposed to contaminants and in turn relate the results to the impacts on human health. It is expensive and impractical to have analysis done on each animal when they are slaughtered and introduced into the consumer market.

This aim of this section was to develop a cattle consumption model based on a control and experimental group in an area where the experimental group was exposed to elevated levels of contaminants emanating from the Gold mining industry in South Africa. The term cattle consumption model refers to the intake of soil, grass and water of the animal. In turn the predictive cattle consumption model was used to perform a human health risk assessment based on the EPA (Environmental Protection Agency) methodology.

6.3.1 Methodology

Measured field data from both the control and experimental groups were fit to a consumption model to predict concentrations in the liver, kidney, spleen, muscle tissue, bone and hair of the animal based on the intake of soil, grass and water. The predictive model was then employed to perform a human health risk assessment based on the animal intake and the human consumption for various scenarios. A schematic of the modelling process was shown below in Figure 6-5.

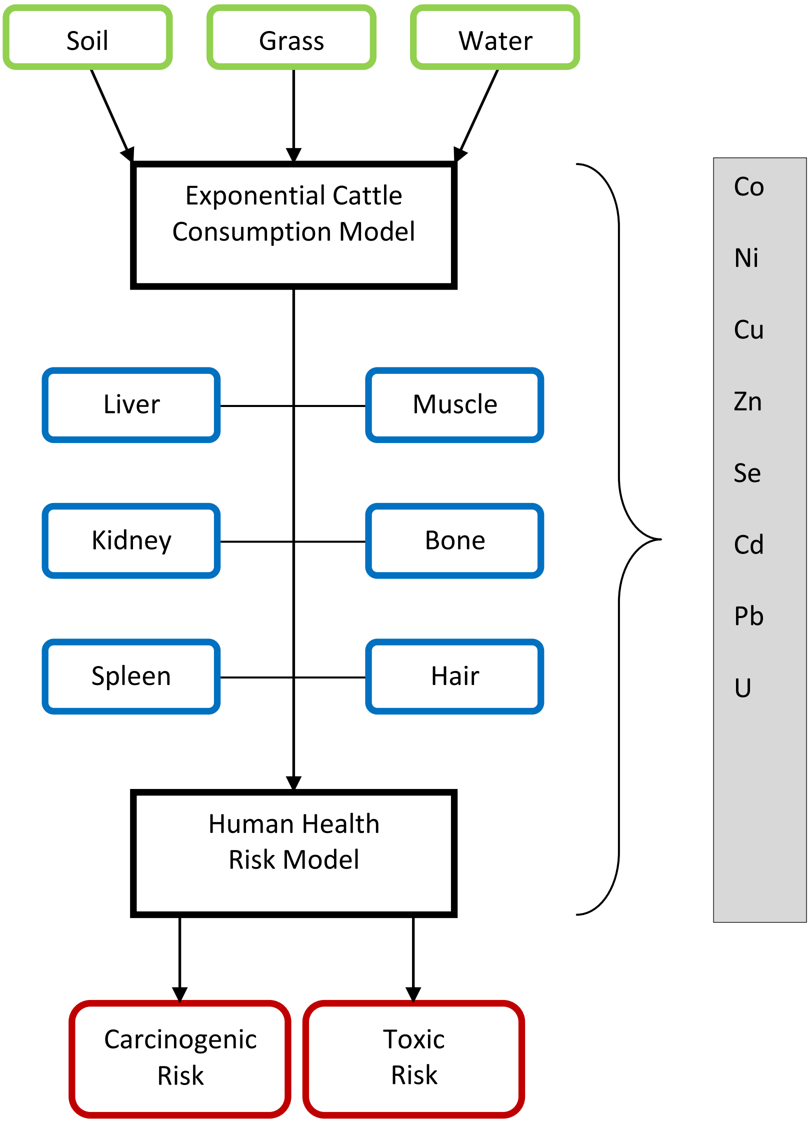


Figure 6-5: Schematic of the modelling process

6.3.2 *Available Data*

The data that was collected from the control and experimental groups was presented in Table 6-3 and Table 6-4 respectively. Note that multiple samples were analysed for each element and only the average values are presented here. Furthermore the animal age group were approximately 6 years for both the control and experimental groups and all concentrations expressed in *mg/kg* represent the dry weight concentrations.

Table 6-3: Control group data

		Co	Ni	Cu	Zn	Se	Cd	Pb	U
Soil	(mg/kg)	39.5500	153.0000	46.0750	28.9750	0.5040	0.1050	25.5250	0.7150
Grass	(mg/kg)	0.8733	5.5725	10.2000	32.5250	0.6950	0.0400	0.9000	0.0400
Water	(mg/L)	0.0000	0.0019	0.0038	0.0028	0.0069	0.0000	0.0005	0.0008
Liver	(mg/kg)	0.3321	0.3018	159.3400	104.2900	0.6759	0.1636	0.1064	0.0425
Kidney	(mg/kg)	0.1073	0.0017	18.1165	78.0500	6.5710	1.1820	0.0261	0.0039
Spleen	(mg/kg)	0.0394	0.2140	4.8640	98.6400	1.0179	0.0299	0.1459	0.0426
Muscle	(mg/kg)	0.0062	0.2044	3.3095	154.7400	0.3895	0.0174	0.1515	0.0485
Bone	(mg/kg)	0.3809	0.0181	0.2428	64.1650	0.3379	0.0408	0.0964	0.0090
Hair	(mg/kg)	0.4198	0.0201	7.5415	96.8300	0.3205	0.0579	0.0583	0.0099

Table 6-4: Experimental group data

		Co	Ni	Cu	Zn	Se	Cd	Pb	U
Soil	(mg/kg)	62.2800	488.9722	39.2898	269.2685	1.4626	0.1343	16.0148	38.0730
Grass	(mg/kg)	3.0067	27.9917	15.3389	139.0278	1.8740	0.0823	2.5447	19.9972
Water	(mg/L)	0.2060	1.2708	0.1012	2.1545	0.0319	0.0041	0.0160	0.1053
Liver	(mg/kg)	0.3717	0.4794	181.7144	307.4890	2.6919	0.1396	0.3347	5.0679
Kidney	(mg/kg)	0.2637	0.5765	23.3517	158.4403	10.8869	1.6414	0.3616	1.7125
Spleen	(mg/kg)	0.0946	0.5221	6.1874	137.3542	3.1350	0.0244	0.3972	1.8769
Muscle	(mg/kg)	0.0586	0.5734	5.0493	221.5512	2.0467	0.0172	0.3963	1.5514
Bone	(mg/kg)	0.3941	3.1775	2.4424	89.4480	1.1064	0.0151	1.0842	0.5889
Hair	(mg/kg)	0.2811	1.4304	6.1600	160.9846	1.5122	0.0327	0.5206	1.2875

6.3.3 Cattle Consumption Model

Two general approaches to modelling accumulation in animal organs are found in literature namely the linear accumulation and the steady state model using an exponential relationship (Van der Fels-Klex *et al.*, 2011). The linear model assumes an irreversible accumulation with no excretion path and usually represents worst case scenarios. The formulation of the linear model is given in equation (1) (Van der Fels-Klex *et al.*, 2011).

$$C_t = BTR \times DI \times t \qquad (1)$$

Where:

C_t = Concentration at time t (mg/kg)

BTR = Biotransfer rate

DI = Daily Intake (mg/day)
t = Time (days)

The exponential model assumes a steady state scenario where accumulation takes place, but an excretion path is also present. The formulation for the exponential model is given in equations (2) to (3) (Van der Fels-Klex *et al.*, 2011).

$$C_t = C_0 e^{-\lambda t} + C_{ss}\left(1 - e^{-\lambda t}\right) \quad \text{with} \quad \lambda = \frac{\ln(2)}{T_{1/2}} \tag{2}$$

$$C_{ss} = BTF \times DI \times F_{abs} \quad \text{with} \quad BTF = \frac{COR}{W\lambda} \tag{3}$$

Where:

C_t = Concentration at time t (mg/kg)
C_0 = Initial concentration (mg/kg)
λ = Elimination time constant
T1/2 = Half-life time (days)
C_{ss} = Steady state concentration (mg/kg)
BTF = Biotransformation factor (day/kg)
DI = Daily Intake (mg/day)
F_{abs} = Absorption in alimentary canal (%)
COR = Carry over rate
W = Weight of organ (kg)
t = Time (days)

The predictive cattle consumption model not only includes organs, but also muscle tissue, bone and hair even though the health risk assessment will only be conducted on liver, kidneys and muscle tissue as the main sources of human ingestion. Making use of the similarity between equations (1) and (3) and applying an exponential response to the model in analogy to equation (2) the following general equation was formulated:

$$C_t = C_0 e^{-\lambda t} + (BTR \times DI)\left(1 - e^{-\lambda t}\right) \quad \text{with} \quad \lambda = \frac{\ln(2)}{T_{1/2}} \tag{4}$$

Where:

C_t = Concentration at time t (mg/kg)

C_0 = Initial concentration (mg/kg)

λ = Elimination time constant

T1/2 = Half-life time (days)

BTR = Biotransformation factor (day/kg)

DI = Daily Intake (mg/day)

t = Time (days)

6.3.4 *Model Calibration*

The initial concentration at day zero of the animals tested was not available for this study and was assumed zero for the purposes of this study. An initial concentration is however expected in a new-born calf whose mother has been subjected to contaminated feed and water. Due to the fact that two groups of data exist (control and experimental) requires the simultaneous solution of two equations in two unknowns (*BTR* and *λ*) for each component of interest. The equation set for a single element was shown in equation (5).

$$C_{t_{con}} = BTR \times DI_{con} \times \left(1 - e^{-\lambda t}\right) \tag{5}$$

$$C_{t_{exp}} = BTR \times DI_{exp} \times \left(1 - e^{-\lambda t}\right)$$

The solution of the equation set in (5) was achieved using MATLAB® with an initial *BTR* guess as the average *BTR* obtained using the linear model in equation (1) for each element of interest.

The observed vs. simulated results for the control group and experimental group was shown in Figure 6-6 and Figure 6-7 respectively. From the results it appears that the predictive model estimates the experimental group better than the control group. Note that concentrations were presented on a log scale in Figure 6-6 and Figure 6-7 and the error bars indicate the minimum and maximum of the samples analysed with respect to the average value measured.

The correlations between observed and simulated values when the control and experimental groups are combined were shown in Figure 6-8. The lowest correlations were obtained from bone (74%) and hair (86%) which was not used in the health risk assessment. The three sources used in the health risk assessment with their associated correlations are as follows:

- Liver (98%)
- Muscle tissue (93%)
- Kidney (87%)

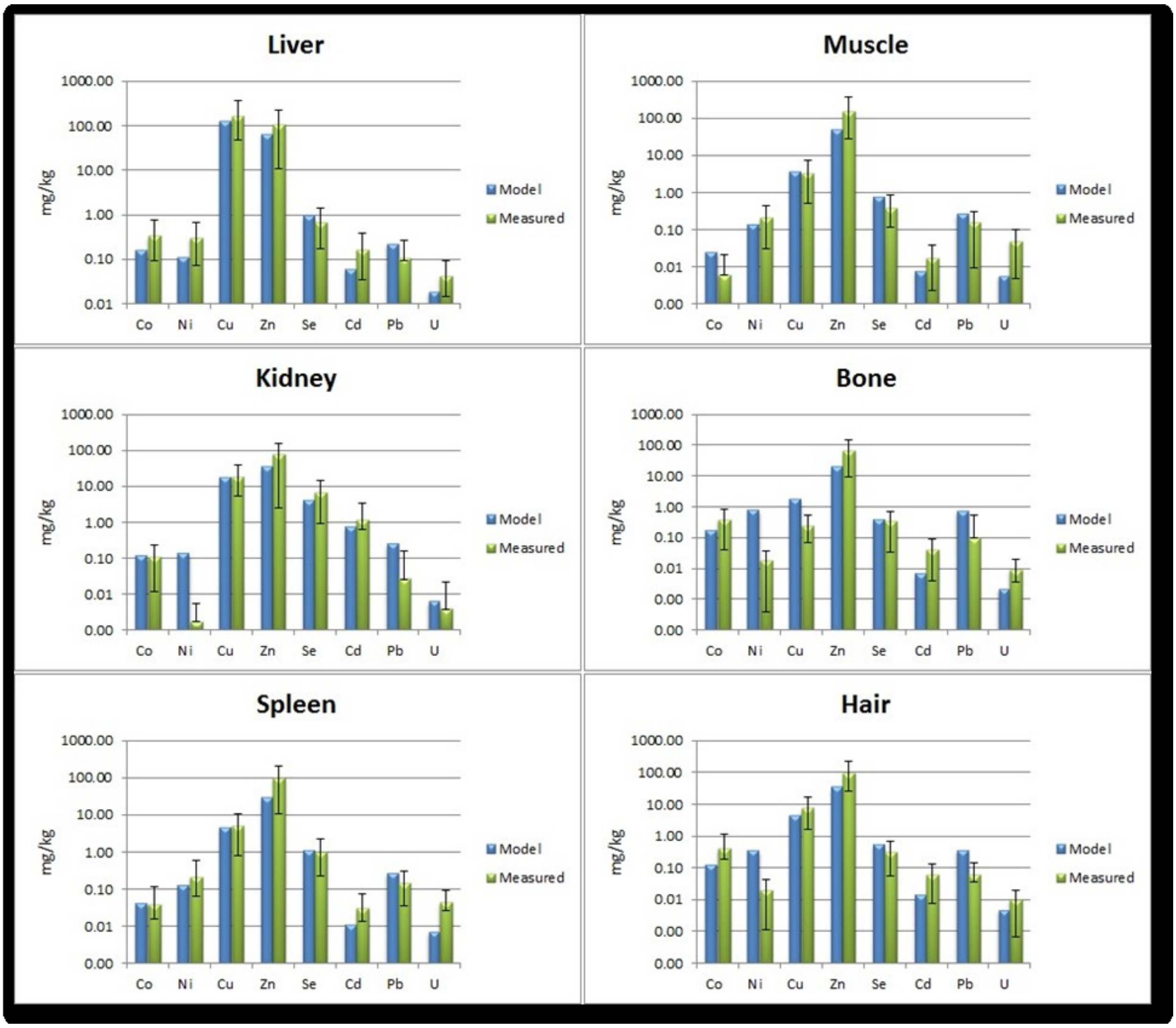


Figure 6-6: Control group - measured vs. modelled values

All model predictions for the control group lie between the measured minimum and maximum values with the exception of those listed in Table 6-5.

Table 6-5: Model predictions outside the minimum-maximum range measured

	Co	Ni	Cu	Zn	Se	Cd	Pb	U
Liver								
Kidney		x					x	
Spleen						x		x
Muscle	x							
Bone		x	x				x	x
Hair	x	x					x	

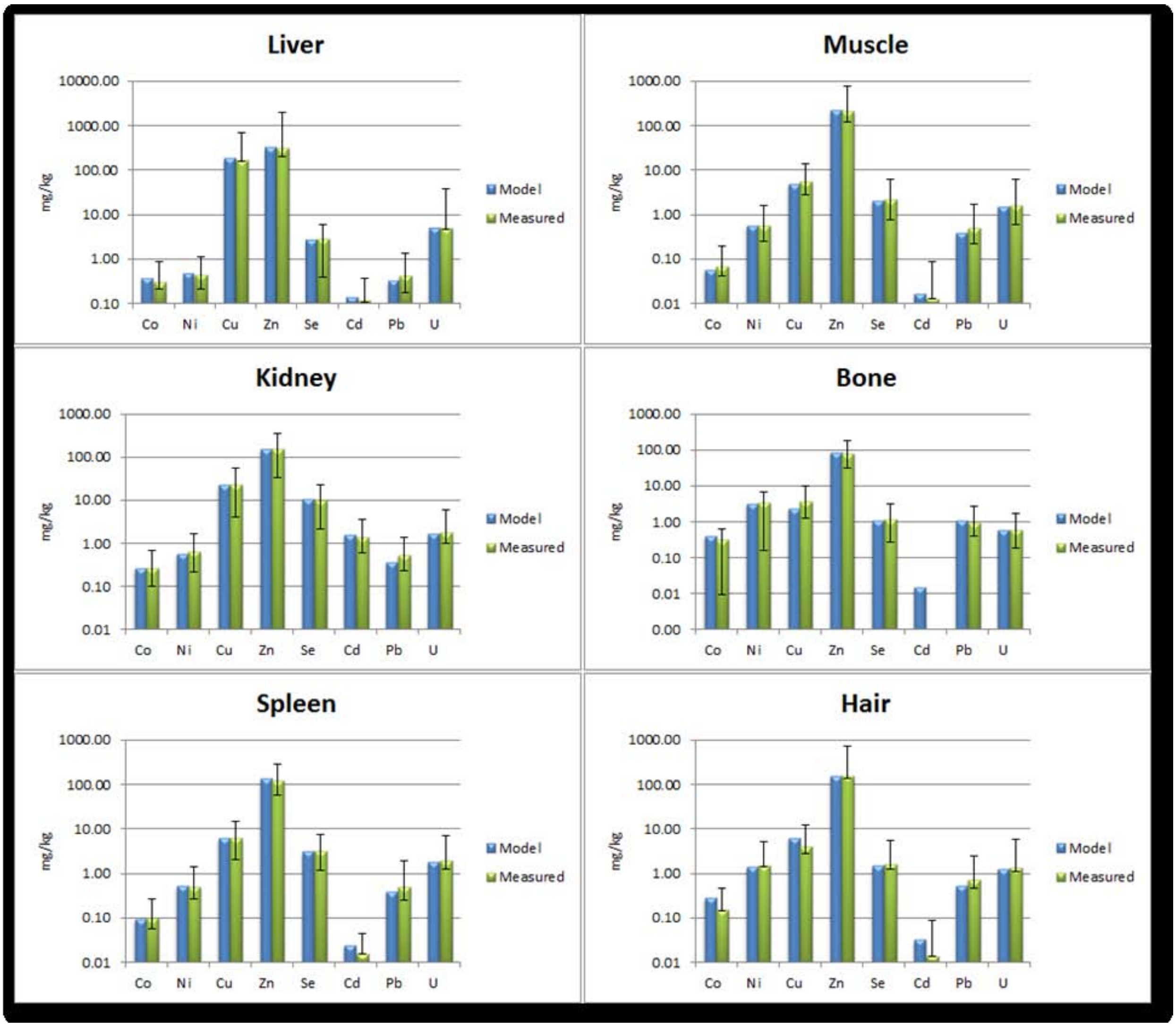


Figure 6-7: Experimental group - measured vs. modelled values

All model predictions for the experimental group lie between the measured minimum and maximum values with the exception of those listed in Table 6-6.

Table 6-6: Model predictions outside the minimum-maximum range measured

	Co	Ni	Cu	Zn	Se	Cd	Pb	U
Liver								
Kidney								
Spleen								
Muscle								
Bone						x		
Hair		x						

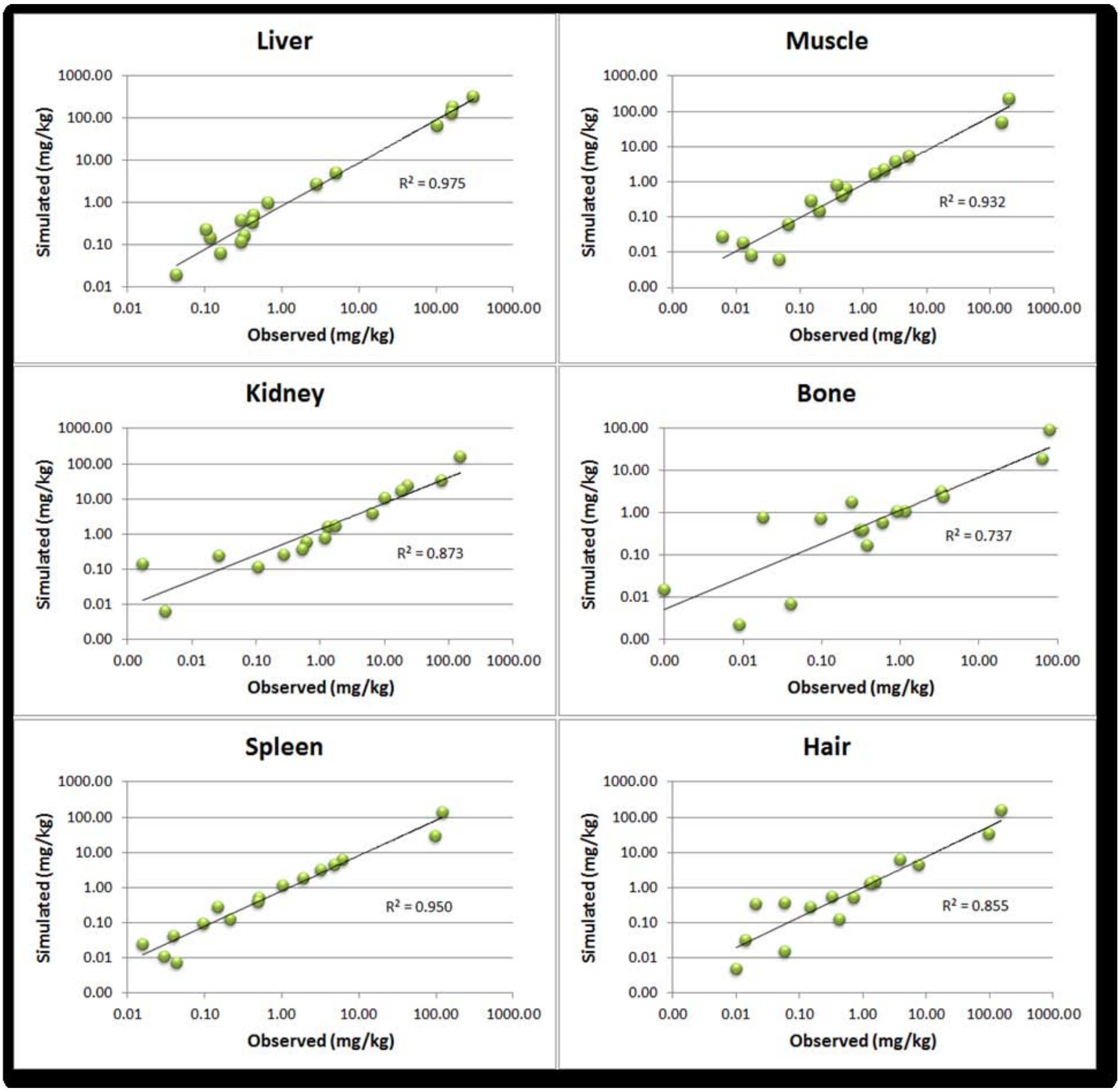


Figure 6-8: Correlation between observed and simulated values for both control and experimental groups

6.3.5 Non-unique Solution

Of particular importance is to note that the solution of two equations in two unknowns are non-unique, hence the initial guesses to ensure a solution in the correct solution space (Grosan *et al.*, 2008). Example responses of kidneys to cadmium were shown in Figure 6-9 to illustrate this point. The model can be fit to obtain the same concentration at year 6 for various half-life values. The only way to achieve a fit with a high confidence is to consider similar data sets at different times to better describe the curve. Unfortunately additional datasets were not available for this study to achieve the above mentioned.

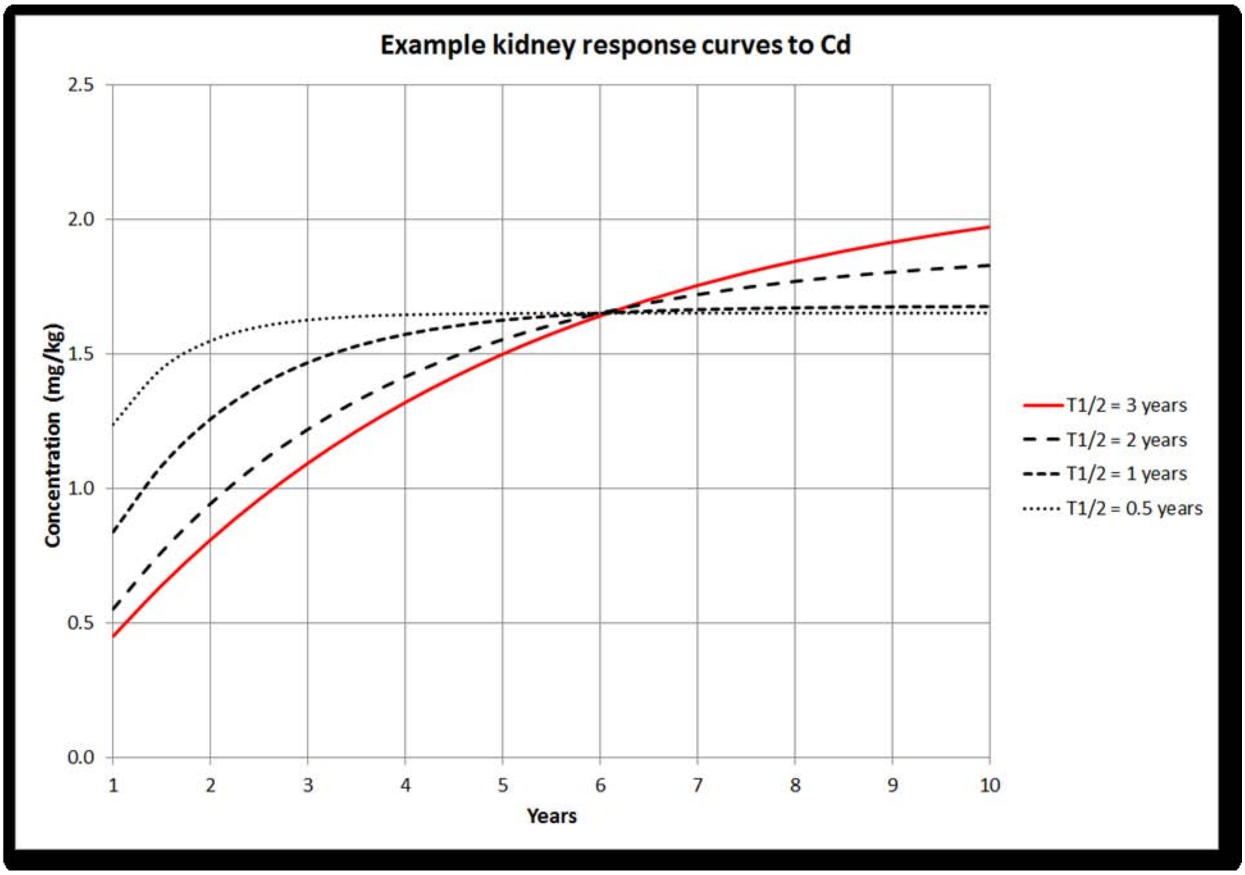


Figure 6-9: Example kidney response to cadmium

It has already been shown that sufficient correlation between measured and observed values were obtained from the predictive model, calibrated for data at year 6. This is seen as an upper limit for the risk assessment as 6 years are the general age at which these animals are slaughtered.

6.3.6 Model Fitness

The influence of the intake of water was proven to be negligible based on measurements of cadmium in drinking water (Van der Fels-Klex *et al*., 2011). The predictive model fitness was tested by setting the intake of water and soil to zero, leaving grass as the only source in the cattle consumption model. The model fitness under these conditions, expressed as the correlation between observed and simulated values were presented in Figure 6-10.

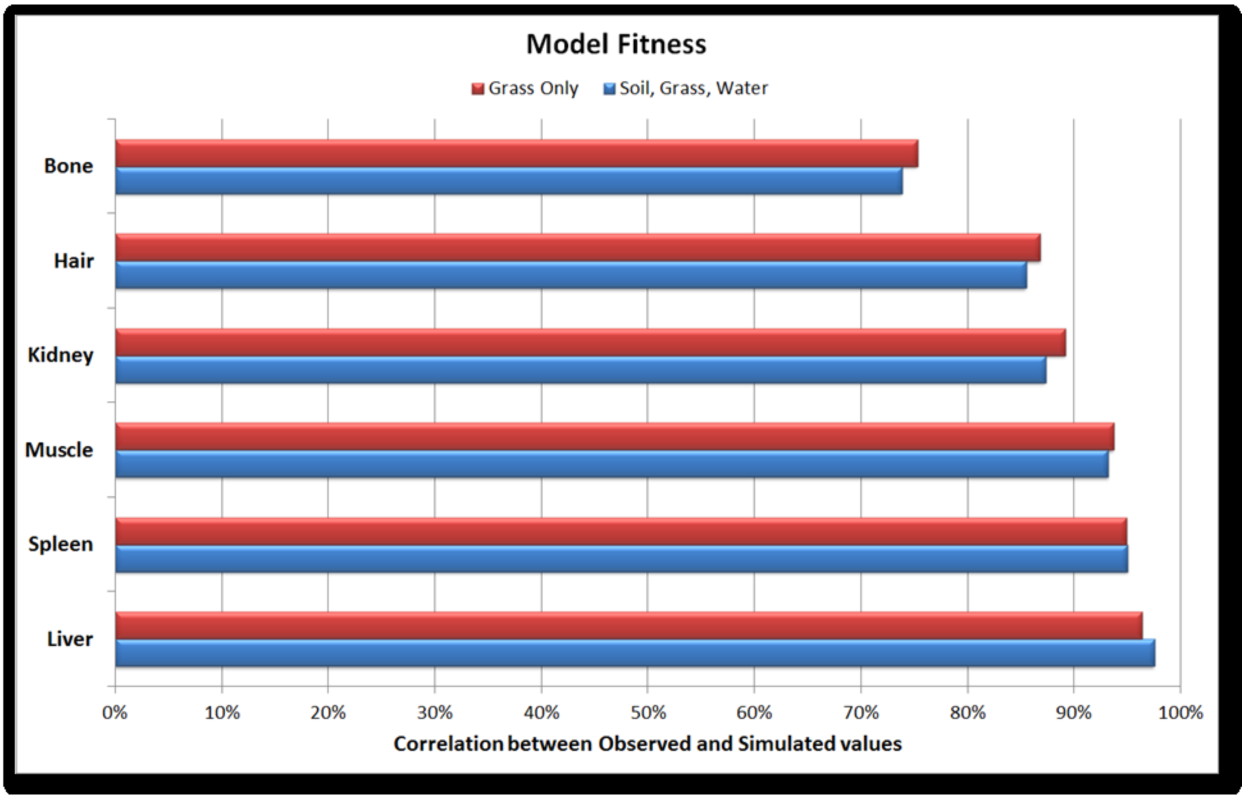


Figure 6-10: Model fitness when grass is used as the only source

It was clear from Figure 6-10 that only small changes in model correlation have occurred and only liver and spleen samples have shown a decrease in correlation whereas all the other components actually showed an increased correlation. This confirms that the predictive model can be used effectively with grass as the only source.

6.3.7 Risk Assessment Model

The EPA (Environmental Protection Agency) methodology for carcinogenic and toxic risk was implemented. Toxic and carcinogenic assessments take into account the following routes of exposure: ingestion, inhalation and dermal sorption. For the purpose of this paper only the ingestion pathway was considered.

6.3.8 Methodology

Before calculating the risks associated with carcinogenic and toxic assessments, the total dose, average daily dose and lifetime average dose have to be defined. The equations used to define risks associated with human exposure to a contaminant were generally based on those specified in the EPA "Risk Assessment Guidance for Superfund" (EPA, 1989).

For each pathway, the total dose that will reach a human has to be calculated. The total dose was defined as:

$$Dose = C \times IR \times ED \qquad (6)$$

Where:

Dose = Total Dose (mg)

C = Maximum concentration (mg/kg)

IR = Initial concentration (kg/day)

ED = Exposure Duration (days)

The average daily dose (ADD) was determined by dividing an estimate of the total dose accrued during the exposure duration from a pathway by an averaging time or an expected lifetime:

$$ADD = \frac{Dose}{BW \times ED} \qquad (7)$$

Where:

BW = Average body weight over exposure period (kg)

Carcinogenic Risk

Carcinogenic risk assessments are determined over a human's lifetime. Therefore the lifetime average daily dose (LADD) is calculated as:

$$LADD = \frac{Total\ Dose}{BW \times Lifetime} \qquad (8)$$

The carcinogenic risk calculation was based on a Poisson model:

$$Risk = 1 - e^{-LADD \times CPF} \approx LADD \times CPF \qquad (9)$$

Where:

CPF = = Cancer Potency Factor *(mg/kg/day)*

The potency factor is the slope of the percentage of animals developing cancer versus the dosage level of a particular chemical. The slope of this curve is then extrapolated to the low doses expected to be encountered by humans who may be exposed to the same chemical. Carcinogenic risk is expressed as a probability of the occurrence of cancer cases e.g. a calculated risk of 10^{-6} is interpreted as the occurrence of one case in a million. The measure of 10^{-6} as an acceptable risk is sited in various literature. According to Kelly (1991) this figure should be redefined on scientific, social and economic basis. For the purpose of this paper the 10^{-6} is recognised as an acceptable risk estimate.

Toxic Risk

The toxic risk is calculated as:

$$Risk = \frac{ADD}{RfD} \tag{10}$$

Where:

RfD = = Reference Dose *(mg/kg/day)*

The reference dose is an estimation of daily exposure to the population (including sensitive subgroups), likely to be without an appreciable risk of deleterious effects during a lifetime. Toxic risks compare the average daily dose of a pollutant to a reference dose calculated for that specific pollutant. Once the average daily dose is equal to or greater than the reference dose, the risk of a person suffering toxic effects due to exposure to the particular pollutant is 99%.

6.3.9 *Cancer Potency Factors (CPF) and Reference Doses (RfD)*

Available sources consulted on CPF and RfD with the associated values for the elements in question were shown in Table 6-7.

Table 6-7: Available Cancer Potency Factors and Reference Doses

	Co	**Ni**	**Cu**	**Zn**	**Se**	**Cd**	**Pb**	**U**
Cancer Potency Factor (CPF)								
IRIS	-	-	-	-	-	-	-	-
ATSDR	-	-	-	-	-	-	-	-
RAIS	-	-	-	-	-	-	0.0085	-
Reference Dose (RfD)								
IRIS	-	0.002	-	0.300	0.005	0.001	-	0.003
ATSDR	-	-	0.900	-	0.005	-	-	0.006
RAIS	0.003	0.020	0.040	0.300	0.005	0.001	-	0.003

Sources:

IRIS – Integrated Risk Information System (U.S. EPA, 2012)

ATSDR – Agency for Toxic Substances and Disease Registry (ATSDR, 2011)

RAIS – Risk Assessment Information System (The University of Tennessee, 2009)

6.3.10 Results

The input data (soil, grass and water) as given in Table 6-3 and Table 6-4 were applied to the predictive cattle consumption model using the following exposure rates (Table 6-8) assuming the age of the cattle to be six years.

Table 6-8: Exposure rates of cattle

Soil	(kg/day)	0.75
Grass	(kg/day)	12.38
Water	(ℓ/day)	38.50

The predicted elemental concentrations of the muscle, liver and kidneys were then used in the risk assessment model. Both the human toxic and carcinogenic risks were calculated for the consumption of muscle, liver and kidneys for both the control and experimental groups. Table 6-9 lists the assumed inputs for the risk model.

Table 6-9: Inputs for the human risk model

Average human weight	(kg)	70
Average lifetime	(years)	70
Exposure duration	(years)	50

The resultant safe daily intake to avoid toxic risk was shown in Figure 6-11. A pronounced difference exists between the control and experimental groups with regard to the toxic risk. On average South Africans consume 49 kg of meat per annum which translates to 0.13 g/day and includes all types of meat (FAO, 2010.) Using this figure for daily ingestion of meat, a person will be safe from toxic risks for both the control and experimental groups.

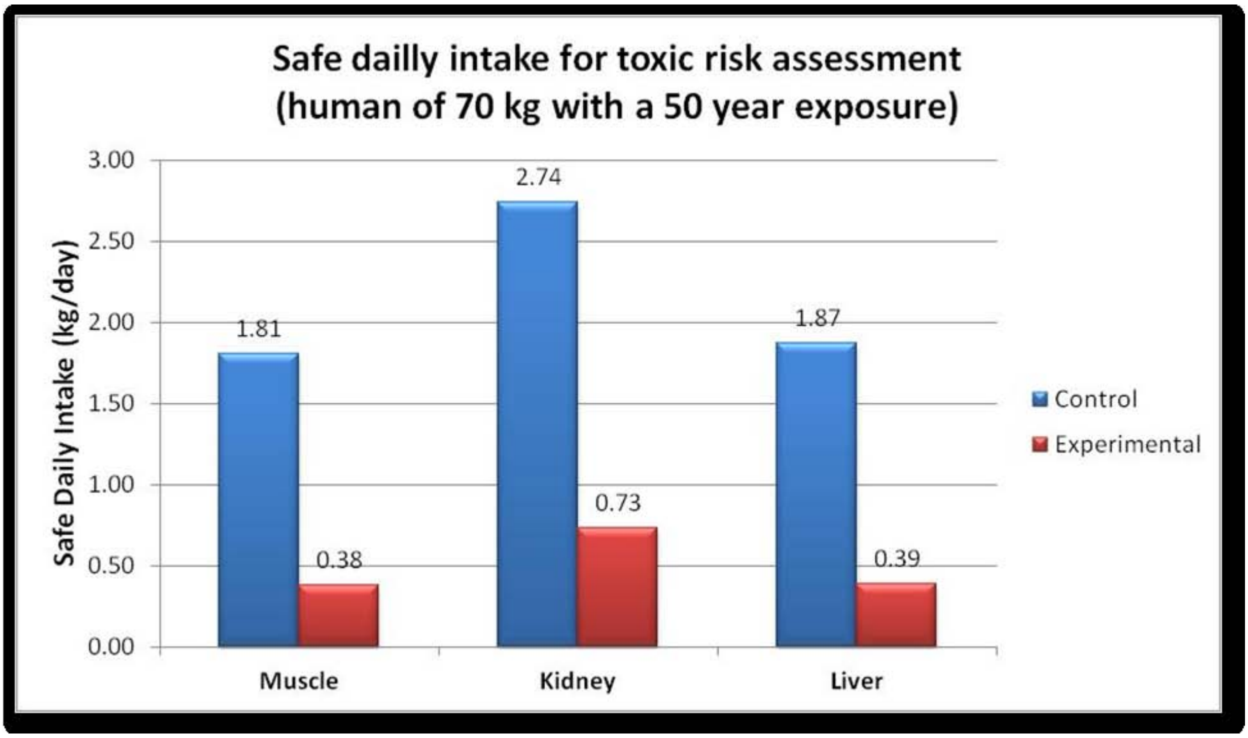


Figure 6-11: Safe daily intake for toxic risk assessment

The only Cancer Potency Factor which could be found in the literature was that of lead as indicated in Table 6-7. There is no conclusive proof that lead causes cancer in human beings and the EPA has therefore determined that lead is a probable human carcinogen (ATSDR, 2007). For this reason, the carcinogenic risk assessment was found to be irrelevant.

6.3.11 Conclusion of risk assessment

It was shown that a predictive cattle consumption model was developed and calibrated from data gathered from a control and experimental group. Animal matter analysed for both groups were related to the cattle age of six years. Although good correlations between observed and simulated values were achieved, the exiting model fit was non-unique. To obtain a more precise model fit a similar dataset is required for both groups, but at a different age.

The predictive model also showed that if only grass were to be used as input, there were no significant changes in the correlation between observed and simulated values. This has a huge advantage in terms of costs associated with laboratory analyses as the analysis of grass will be sufficient for using the model.

A human health risk assessment was performed based on the results of the cattle consumption model. It was shown that no toxic risk exits for both the control and experimental groups if an intake rate of 0.13 kg of meat per day is assumed. Furthermore. Figure 6-11 clearly indicates that an intake rate of up to 0.38 kg of meat per day also has no toxic risk for both groups, which strongly suggests that there is no risk to the human food chain.

7 Conclusions and Recommendations

7.1 Conclusion

The hypothesis; it is expected that toxic and trace elements do transfer and accumulate from the WFS into the surrounding environment and into the cattle grazing in the area was proved to be correct as trace elements were found to transfer and accumulation beyond the stream edge was detected and into soil, grass and livestock.

The collection, analysis and comparison sediment samples from both the Wonderfonteinspruit and Mooi River reveal cobalt, nickel, copper, zinc, selenium, cadmium, lead and uranium to be elements of interest.

The target water quality ranges (TWQR) for livestock watering, as set by DWAF 1996, were exceeded by the average nickel and lead concentrations found in the disturbed WFS water samples.

The study revealed that the major route of ingestion for all the elements of interest, excluding nickel and cobalt was via the ingestion of grass. The major route for nickel and cobalt ingestion was via soil ingestion. The elemental concentrations from water ingestion were found to be a less significant.

A human health risk assessment was performed based on the results of the cattle consumption model. It was shown that no toxic risk exits for both the control and experimental groups if an intake rate of 0.13 kg of meat per day was assumed. Furthermore, Figure 6-11 clearly indicates that an intake rate of up to 0.38 kg of meat per day also has no toxic risk for both groups, which strongly suggests that there is no risk to the human food chain.

7.2 Recommendations

Due to the profound transfer and accumulation of uranium into soil, grass and cattle it is essential that a target water quality range for livestock watering be established.

If unacceptable elemental concentrations within the cattle tissue samples continues (due to ongoing pollution), access to the soil and grass along the WFS and the irrigated pasture should be excluded. This may however have a significant financial impact for farmers in this area as the grazing in these sites play a profound role, especially during dry or low rainfall periods.

The sediment samples from the WFS contained significantly higher concentrations of cobalt, nickel, copper, zinc, selenium, cadmium, lead and uranium, which could pose a serious risk to downstream users. The Coetzee report also recommends that the dam is hazardous in its current state and would require a well-planned remedial action.

The extraction of gold from these sediments should be considered when plans are made to remove the sediment. A feasibility study would be required as the transportation and processing of the sediment may be costly due to the distance from a processing plant.

It is important to note that this study has focused on the impacts on cattle, however to determine the full impact of the mining activities on the WFS, a detailed conceptual model of the receiving environment must be developed including hydrological, geohydrological, meteorological and soil investigations.

Samples could be collected from other livestock species such as sheep, goats and pigs as well as tissue samples from cattle in different age groups in order to determine and compare other exposure pathways and risk assessments.

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