UNIVERSITEIT VAN PRETORIA
UNIVERSITY OF PRETORIA
YUNIBESITHI YA PRETORIA

Effects of TDS and Br on the accumulation of water-borne potentially hazardous chemical constituents As and Pb in broilers

By

Mankabidi Cuthbert Mamabolo

Submitted in partial fulfillment of the requirements for degree

MSc (Agric) in Animal Production Physiology

in the

Department of Animal and Wildlife Sciences
Faculty of Natural and Agricultural Sciences
University of Pretoria
Pretoria

February 2009

DECLARATION

I declare that the dissertation hereby submitted in partial fulfillment for the requirements of the MSc (Agric) Animal Production Physiology at the University of Pretoria has not been submitted by me for other degree at any other institution

Name and Surname: Mankabidi Cuthbert Mamabolo

Signature: MMamabolo

Date: 09/02/2009

ACKNOWLEDGEMENTS

- Firstly, I would like to give praise and honour to GOD almighty for His loving kindness towards me
- It gives me special pleasure to thank my research supervisors Prof NH Casey and Dr JA Meyer. Your guidance and insight critical comments have been invaluable
- Many thank to Mr R Coertze for advices and statistical analysis
- I would also like to thank my wife Mpho for all her support throughout this study
- Finally, I would like to thank my Parents (Maribe and Cecilia) for their support and encouragement all these years

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABBREVIATIONS

As	Arsenic
ARC	Agricultural Research Council
CIRRA	Constituent Ingestion Rate Risk Assessment
ECF	Extracellular Fluid
GRF	Glomerulus's Filtration rate
PHC	Potentially Hazardous Constituents
ICF	Intracellular Fluid
ISCW	Institute of Soil Climate Water
KCl	Potassium Chloride
MAC	Maximum Acceptable Concentration
MLC	Maximum Concentration Level
NaCl	Sodium Chloride
PPM	Parts Per Million
RCLPS	Rural Communal Livestock Production System
WHO	World Health Organisation
WQC	Water Quality Constituents
WQGIS	Water Quality Guideline Index System

US EPA United State Environmental Protection Agency

ABSTRACT

The occurrence of potentially hazardous chemical constituents (PHCC) in subterranean water and the divergent potential for bioaccumulation in different production systems, made it necessary to investigate the effect of alleviator treatment on the accumulation of PHCC in broiler tissues. Local poultry producers are faced with the opportunities to expand their poultry products to overseas countries, therefore, the quality measures must be in place to comply with strict quality control systems like the European Union standards. The effect of water quality on the health and production parameters in broilers and layers has recently been questioned as to its compliance with both the export and local markets. The effects of water quality constituents (WQC) are a function of the type and character of the WQC, the intake rates and exposure to WQC, the type of animal and its physiology and the demands of the environment. Where the livestock and humans use the same water source, the livestock can be an effective indicator species of the risk posed to humans.

The study evaluated the effectiveness of TDS and Br as possible alleviators of PHCC accumulation in broiler tissues grown under intensive production system. Broilers received four types of treatment: control (<500 mg/L TDS; < 0.005 mg/L Br + As + Pb), elevated elements (As=0.1 mg/L; Br=1 mg/L; Pb=0.1mg/L), elevated elements + 1500 mg/L TDS and control + 1500 mg/L TDS in drinking water from one day to 42 days old. The accumulation of PHCC in broiler tissue did not exceed maximum allowable concentrations (EU – MAC) during a short period of exposure. The groups that received TDS retained the lowest PHCC accumulation.

These results suggest that TDS plus bromide in broilers' drinking water could alleviate arsenic and lead accumulation in broilers' tissues. However, the alleviation was not always significant in all the tissues.

Key words: Broilers, TDS, PHCC, As, Pb and Br

CHAPTER 1

LITERATURE REVIEW

1. INTRODUCTION

1.1 Poultry production

The demand for poultry meat in South Africa is increasing because of the high demand for healthy protein sources. The poultry industry is a major part of the agricultural sector and is under pressure for profitability on account of the narrow profit margins caused by high feed costs. The contribution of the poultry industry to the agricultural sector is estimated to be 16% (SAPA, 2007).

The trends in total per capita consumption of meat in South Africa are illustrated in Table 1.1. Poultry meat consumption has been increasing by about 7% per annum, which is better by far than any other meat sector. The increase is the result of rising living standards, which are encouraging consumers to eat higher protein diets, develop healthier eating habits and adopt a more convenient type of diet (Esterhuizen, 2007).

Table 1.1 Per capital consumption of meat in South Africa (Adapted from Esterhuizen, 2007)

Years	Beef	Poultry	Pork	Mutton/lamb
1975	23.12	9.54	4.22	
1980	22.19	13.21	3.86	6.1
1985	20.93	16.58	4.04	6.54
1990	17.82	18.85	3.86	5.3
1995	17.74	16.25	3.44	4.52
2000	14.01	19.21	3.18	3.52
2005	13.91	21.24	3.06	3.28
2015(Projected)	14.49	24.95	3.46	3.81

The largest poultry-producing province in South Africa is the Western Cape (Table 1.2) (Casey *et al.*, 2001).

Table 1.2 Gross production value of poultry amongst the nine Provinces of South Africa

Province	Production
Western Cape	27.1
North West	10.96
Gauteng	13.15
Limpopo	2.28
Mpumalanga	12.64
KwaZulu –Natal	27.17
Eastern Cape	5.22
Free State	1.2
Northern Cape	0.28

Any decline in the health and feed efficiency of poultry will reduce profit margins drastically. Poultry management includes chick management, growth management, pre-processing management, nutrition, hygiene, health, housing and environment (Casey *et al.*, 2001). Efficiency in the industry has improved over a number of years, and there is no room for substandard production performance. Further, the industry is under threat of diseases like Newcastle Disease and Avian Influenza (SAPA, 2007).

There are increasing opportunities for local poultry producers to expand their poultry products to overseas countries, so quality measures must be in place to comply with strict quality control systems like the European Union (EU) standards. The effect of water quality on the health and production parameters in broilers and layers has recently been questioned as to its compliance with both the export and local markets (Casey *et al.*, 2001).

1.2 Rural communal livestock production system

The rural communal livestock production system model caters for the complex requirements in balancing risk and hazard identified in the area of environmental toxicology and the significant role of water quality in improving animal and human health. Shared use of water by humans and animals prohibits the application of treatment to the common water source because of its potentially adverse effect on humans. Another area of concern is the effect of the quality of animal products on human health. The accumulation of constituents like lead (Pb), and cadmium (Cd) in renal cortex tissue in intensive commercial systems does not present a significant consumer hazard, as concentration and dilution within the urban diet effectively provide sufficient safety. However, the same is not true of rural production systems, as one community alone consumes its entire poultry production (Casey *et al.*, 2001).

The production system in rural areas is seldom as brief and well- defined as a commercial system. The exposure period is longer, with a high ingestion rate due to poor environmental conditions like high temperatures. Potentially hazardous constituents in the water may find their way by a number of routes into sensitive user-groups, such as reproductively active women and children. Exposure may be directly via drinking water, or indirectly via the consumption of animal products because of their exposure to the contaminated water source (Casey *et al.*, 2001).

1.3 Water quality

Knowledge of water quality is important for poultry production, as it gives producers the management information required to prevent the potentially adverse problems that may be caused by water quality constituents. These are mostly related to production parameters, health, the quality of the livestock products and watering used in poultry production (Coetzee *et al.*, 2000).

When bacterial contamination of borehole water occurs, it is usually a sign that the surface water is entering the subterranean water supply and steps should be taken to correct the problem. The water can also be chlorinated to eliminate bacterial contamination. The build-up of nitrates or nitrites can also be a major problem for both animals and humans. Such contamination is usually an indication of run-off from animal wastes or fertilizers leaking into the water system or from natural subterranean water. Poultry can tolerate the standards set for humans (Leeson & Summers, 2005).

1.3.1 Water quality constituents

Water quality constituents adversely affect animal performance, growth and reproduction by reducing water intake and subsequently feed intake. The reduction in feed intake results in reduced daily gain and a poor feed conversion ratio. These conditions are undesirable for both intensive and extensive production systems. The accurate and frequent water-quality monitoring is important for early detection and responds to the problems

(Meyer, 2008). Water quality should be monitored every six months at least. Chemical contaminants such as lead (Pb), arsenic (As) and selenium (Se) are the most serious of the problems affecting water quality (Leeson & Summers, 2005).

Water-quality constituents that are potentially hazardous, with a high incidence, are salinity, chloride, sulphate, arsenic, copper, sodium, calcium, fluoride, molybdenum, magnesium nitrate, nitrite and toxic algae. Those that are potentially hazardous with a low incidence are cadmium, chromium, mercury, lead, zinc, selenium, boron, aluminium, cobalt, iron, nickel, vanadium, manganese, pesticides and pathogens (Leeson & Summers, 1997). A compilation of water quality guidelines for poultry watering from various sources is presented in Table 1.3, while Table 1.4 presents concentration of water minerals above which problems may occur.

When chickens ingest a high concentration of heavy metals, water consumption is significantly decreased. There is a linear relationship between increased concentration of the chemical mixture in the drinking water and the decreased body weight of hens. Chemical mixtures of lower concentration decrease egg production and weight gain (Vodela *et al.*, 1997).

Table 1.3 A compilation of water quality guidelines for poultry watering from various sources

WQC	Maximum Acceptable Level	Effects	Reference
Alkalinity	See pH		
Aluminium	0.25mg/L 0.2 mg/L 5mg/L	Reduced growth, rickets.	Kempster *et al.*, 1981 Zimmerman, 1995 Mancl *et al.*, 1991
Ammonia	2mg/L	Dissolve copper from piping and appliances.	Kempster *et al.*, 1981
Antinomy	0.006mg/L	Emetic and a cardio-toxin.	Zimmerman, 1995
Arsenic	0.2mg/L 1mg/L 0.05mg/L	Toxic substance, affects normal physiological processes of the body.	Cater, 1985, Keshavarz, 1987 & Mancl *et al.*, 1991 Kempster *et al.*, 1981 Vohra, 1980 & Zimmrman, 1995
Bacteria	Total = 100/mL Coliform = 50/mL	Infections/solve problem with 1mg/L chloride, for 3 minutes and pH 8. Respiratory disease, bloody dropping.	Schwartz, 1994 & Waggoner *et al.*, 1994 Schwartz, 1994 & Waggoner *et al.*, 1994
Barium	1mg/L 2mg/L	Reduce growth, death.	Vohra, 1980 Zimmerman, 1995
Beryllium	0.004mg/L	Not a priority pollutant, it may be carcinogenic.	Zimmerman, 1995
Bicarbonate	98mg/L 500mg/L	Alone not a problem sometimes are desirable if sulphate or sodium are present	Keshvarz, 1987
Boron	5mg/L	Not a priority pollutant.	Mancl *et al.*, 1991
Cadmium	50mg/L 0.01mg/L 0.005mg/L 0.05mg/L	Excess causes severe health effects. Reduced growth, decreased egg production.	Kempster *et al.*, 1981 Vohra, 1980 Zimmerman, 1995 Mancl *et al.*, 1991
Calcium	402mg/L 600mg/L 200mg/L	Desirable if sodium is present. Non-toxic, clog up pipes.	Kempster *et al.*, 1981 Cater, 1985, Keshavarz, 1987 Vohra, 1980
Chloride	250mg/L 1500mg/L 200mg/L 600mg/L	Detrimental when combined with 50mg/L of Na. Increase water intake, wet litter.	Schwartz, 1994, Waggoner *et al.*, 1994, Ernst, 1989 & Zimmerman, 1995 Cater, 1985 Keshavarz, 1987 Vohra, 1980
Chromium	5mg/L 0.05mg/L 0.1mg/L 1mg/L	May contribute to hardness of water, low toxicity, nutritionally essential, absence causes diabetes Reduce growth.	Kempster *et al.*, 1981 Vohra, 1980 Zimmerman, 1995 Mancl *et al.*, 1991
Cobalt	1mg/L	Nutritionally essential, toxic in excess, reduce growth.	Kempster *et al.*, 1981
Colour	15 colour units	Aesthetic, should be colourless. Red brown = due to iron. Bluish = due to copper.	Zimmerman, 1995

Copper	0.06mg/L 2mg/L 2.5mg/L 0.5mg/L 1.5mg/L 0.6mg/L 1.3mg/L	Bitter, causes liver damage, reduce growth, mortalities, exudative diathesis, muscular dystrophy, gizzard erosion.	Schwartz, 1994 & Waggoner *et al.*, 1994 Kempster *et al.*, 1981 Good, 1985 Carter, 1985, Keshavarz, 1987 & Mancl *et al.*, 1991 Vohra, 1980 Ernst, 1989 Zimmerman, 1995
Cyanide	0.2mg/L		Zimmerman, 1995
Fluoride	2mg/L 0.9-1.7mg/L 0.06-0.08mg/L 4mg/L	Lower feed intake and growth rate.	Carter, 1985, Keshavarz, 1987 & Mancl *et al.*, 1991 Vohra, 1980
Hardness	>180 = hard <60 = soft	Ca and Mg in sulphate form affect performance only with regard to its calcium content. Blocks water system. >200mg/L Ca leads to excessive deposit and scale formation.	Schwartz, 1994 & Waggoner *et al.*, 1994
Iron	0.3mg/L 1.2mg/L 6mg/L 0.1mg/L	Aesthetic-clog pipes, bad taste and odour up to 25mg/L no effect on performance, may contribute to hardness.	Schwartz, 1994 & Waggoner *et al.*, 1994 Ernst, 1989 Kempster *et al.*, 1981 Keshavarz, 1987 Vohra, 1980
Lead	0.02mg/L 0.1mg/L 0.5mg/L 0.05mg/L 0.015mg/L	Toxic element, affects normal physiological processes of body. Decrease performance, reduce egg size and palatability.	Schwartz, 1994 & Waggoner *et al.*, 1994 Ernst, 1989 Carter, 1985, Keshavarz, 1987 & Mancl *et al.*, 1991 Kempster *et al.*, 1981 Vohra, 1980 Zimmerman, 1995
Magnesium	125mg/L 350mg/L 50mg/L 150mg/L (if 250mg/L sulphate is present)	Decrease performance if 50mg/L of sulphate is present, intestinal irritation, laxative effect, watery droppings, hardness, taste, lethargy.	Schwartz, 1994 & Waggoner *et al.*, 1994 Ernst, 1989 Carter, 1985 Keshavarz,1987 Vohra, 1980
Magnesium sulphate	200-400mg/L	Lower egg production. High sulphate is detrimental to water with high chloride (> 100mg/L) content, increased salinity of water.	Kempster *et al.*, 1981
Manganese	4.6mg/L 0.05mg/L 0.6mg/L	May contribute to hardness and turbidity deposits in pipes and bitterness of water, mortalities.	Kempster *et al.*, 1981 Carter, 1985 & Vohra, 1980 Keshavarz,1987
Mercury	10mg/L 0.002mg/L 0.01mg/L	A toxic element with no beneficial physiological function, reduce egg production, reduce growth.	Kempster *et al.*, 1981 Vohra, 1980 & Zimmerman Mancl *et al.*, 1991
Molybdenum	10mg/L	Reduced growth, poultry more tolerant than ruminants.	Kempster *et al.*, 1981

Nickel	0.001mg/L 1mg/L	>1000mg leads to minor toxic effects. Reduced growth.	Zimmerman, 1995 Mancl *et al.*, 1991
Nitrates	25mg/L 200mg/L 20mg/L 10mg/L	Reduce growth. Increase mortality.	Schwartz, 1994, Waggoner *et al.*, 1994 Ernst, 1989 & Mancl *et al.*, 1991 Kempster *et al.*, 1981 Good, 1985, Keshavarz, 1987 Zimmerman, 1995
Nitrites	4mg/L 1mg/L 3mg/L	High toxicity, decreased Vit A in liver and thyroid enlargement	Schwartz, 1994 & Waggoner *et al.*, 1994 Ernst, 1989 Zimmerman, 1995 Mancl *et al.*, 1991
pH	>6.0 2-10 >5.9	Lower performance, lower egg quality, and lower effectiveness of vaccines. Solve with mild solution of NaOH. Acid water –corrode pipes.	Schwartz, 1994 & Waggoner *et al.*, 1994 Kempster *et al.*, 1981 Good, 1985
Phosphate	5mg/L 0.7mg/L	Alone no problem, high levels indicate sewage contamination.	Kempster *et al.*, 1981 Carter, 1985
Salinity	3000mg/L	Unsuitable for poultry. NaCl, Na2SO4, MgSO4.	Mancl *et al.*, 1991
Selenium	0.05mg/L 0.01mg/L	Toxicity substance, affect normal physiological processes in the body. Reduced growth.	Kempster *et al.*, 1981, Zimmerman, 1995 & Mancl *et al.*, 1991 Vohra, 1980
Sodium sulphate	1200mg/L	Increase in diarrhoea. Increase salinity.	Kempster *et al.*, 1981
Sodium	200mg/L 50mg/L 75mg/L	Not detrimental if 500mg/L of bicarbonate is present. Reduced performance if 50 mg/L SO4 or 14mg/L chloride is present. Increased water intake, wet litter. Reduced egg production and growth.	 Ernst, 1989 Keshavarz, 1987
Sodium chloride (NaCl)	1500mg/L	Later sexual maturity. Higher mortalities, higher salinity of water, reduced growth.	Kempster *et al.*, 1981
Sulphate	250mg/L 60mg/L 400mg/L (if Na & Mg are present 300mg/L)	Reduced performance if 50mg/L of magnesium or 50mg/L of Na is present, laxative effect, wet litter, reduced egg production.	Schwartz, 1994 & Waggoner *et al.*, 1994 & Ernst, 1989 Keshavarz, 1987 Vohra, 1980 Mancl *et al.*, 1991
TDS	3000mg/L	Saline or brackish water.	Zimmerman, *et al.*, 1981
Vanadium	0.1mg/L	Nutritionally essential. Reduce growth, depressed albumin quality. Reduced body weight. Depressed hatchability.	Mancl *et al.*, 1991
Zinc	1.5mg/L 2.5mg/L 15mg/L 25mg/L	Astringent taste, may contribute to hardness. Lower growth, decreased fertility, skin disease, muscular dystrophy, reduced bone ash (0.5mg/L).Se in diets.	Schwartz, 1994 & Waggoner *et al.*, 1994 & Ernst, 1989 Carter, 1985 & Keshavarz, 1987 Vohra, 1980 Mancl *et al.*, 1991

Table 1.4 Concentration of water minerals above which problems may occur in poultry (Leeson and Summer, 1997)

Minerals	mg/L
Total soluble salts (Hardness)	1500
Chloride	500
Sulphate	1000
Iron	500
Magnesium	200
Potassium	500
Sodium	50
Arsenic	0.01
Ph	6.0-8.5

1.3.1.1 Arsenic

The name arsenic is derived from *arsenikon*, the Greek word meaning 'potent'. Arsenic is like sugar and is tasteless, which is why it was popular as a means of assassination during the middle ages. However, it was also used to cure ulcers at 1% of potassium arsenite solution of flower solution. Arsenic-containing drugs are still used to treat African trypanosomiasis at the meningoencephalitic stages (Malachowski, 1990).

Arsenic occurs in either inorganic or organic compounds. Arsenites, arsenate and elemental arsenic are inorganic arsenic compounds, whereas arsine and its derivatives are organic arsenic compounds (Winship, 1984).

In the terrestrial environment, arsenic is found mainly in rocks and soils. Arsenic contained in surface and ground water is usually a mixture of arsenite and arsenate. Arsenic is highly distributed in food, and high quantities are present in seafood products (Wade *et al.*, 1999).

Arsenic is not easily dissolved in water, so if it is found in a water supply, it has usually come from mining or metallurgical operations or from runoff of agricultural areas where arsenic-containing materials were used as industrial poisons. Arsenic and phosphate easily substitute each other chemically, which means that commercial-grade phosphate can contain some arsenic. It is highly toxic and has been classified by the United State Environmental Protection Agency (US EPA) as a carcinogen. The current maximum concentration level (MLC) for arsenic is 0.05 mg/L, which was derived from toxicity considerations rather than carcinogenicity (WHO, 2001).

The primary absorption of arsenic by the body is through ingestion, inhalation and percutaneously. Arsenic distributes rapidly into the erythrocytes and binds to the globin portion of haemoglobin. Redistribution to the liver, kidney, spleen, lungs and gastrointestinal tract of the body is prevalent within 24 hours of ingestion. Toxicity occurs through impairing cellular respiration by inhibiting mitochondrial enzymes and uncoupling oxidative phosphorylation via inhibition of the sulfhydryl group containing cellular enzymes and substitution of phosphate with arsenate in "high energy" compounds (Winship, 1984).

Symptoms of toxicity usually appear 30 minutes after acute ingestion of arsenic, but onset may be delayed if it was ingested with food. Acute toxicity symptoms are usually gastrointestinal distress, hypotension and tachycardia (Gorby, 1988). The initial symptoms of acute arsine gas exposure are non-specific in humans, and include headaches, weakness, nausea, vomiting and abdominal pain (Malachowski, 1990).

1.3.1.2 Bromide

The bromide (Br) is an inorganic compound found in surface water and ground water. It is caused by sea intrusion, the impact of connate, or industrial and oil-field brine discharge (Glaze & Weinberg, 1999). Its relatively minor contribution to the composition of water, and the absence of any adverse health-effect attributed to it, has resulted in its receiving only scant attention (Stumm & Morgan, 1981). When oxidized, it can result in the formation of organic and inorganic bromine. The bromate (BrO_3) is the highest oxidation state of the bromide (Symons, 1999). Ozone oxidizes bromide to form hypobromite. Hypobromite continues to be oxidized to form bromate, or forms an unidentified species, possibly BrO_2, which regenerates bromide (Glaze & Weinberg, 1999). The reaction is as follows:

$$O_3 + Br \rightarrow O_2 + OBr-$$

$$O_3 + OBr- \rightarrow 2O_2 + Br-$$

$$2O_3 + OBr- \rightarrow 2O_2 + BrO_3-$$

Bromide concentration in water depends primarily on the geochemistry of the materials with which the water has come into contact, with most episodes of freshwater contamination leading to increased salinity, probably occurring with a concomitant increase in bromide (Kjensmo, 1997). In localized areas, intensive application of brominated pesticides can produce a major contribution to the bromide level (Wegman *et al.*, 1983).

The nutritional requirement for bromide in chicken was postulated by Huff *et al.* (1957). Day-old chicks were reared for 31 days on diets supplemented with bromide, as NaBr, at 8 mg/L and 15 mg/L. Improvements in growth attributed to the supplemental bromide ranged from 8% to 10%. Growth responses to bromide supplementation have been observed in chickens, but the essentiality, biological function, or modes of action of the element have not yet been unequivocally proved (Mertz, 1970). The maximum tolerable levels of bromide in growing chickens are 5 000 mg/L (NAC, 1980).

1.3.1.3 Lead

Lead (Pb) is the Anglo-Saxon word for the element initially known by the Latin *plubum*, which serves as the root of plumbism, meaning "lead poisoning" (Lide, 1994). In 1904, Gibson concluded that lead paints in the homes were responsible for poisoning children. However, lead was not banned in America until 1977. Paints applied before 1950 often contained high amounts of lead carbonate, and about 74% of privately-owned housing built in the United States before 1980 contained lead-based paint.

When lead (Pb) is found in fresh water this usually indicates contamination from metallurgical wastes or from industrial poisons containing Pb. Lead in drinking water comes primarily from the corrosion of the Pb solder used to put the copper piping together. When Pb is present in the body, it can cause serious damage to the brain, kidneys, nervous system and red blood cells. The United States Environmental Protection Agency (US EPA) considers Pb to be a highly toxic metal and a major health threat. The current level of Pb allowable in drinking water is 0.05 mg/L (WHO, 2001).

Lead (Pb) poisoning occurs mainly in cattle and sheep and is usually acute. Pigs, goats and chickens are moderately resistant (DWA & F, 1996). McGowan and Donaldson (1987) found that when chickens were intoxicated with 2000 ppm dietary Pb there was a significant decrease in plasma cysteine, taurine and cystathionine concentration. Lead present in chickens' diets increases the riboflavin requirement (Donaldson, 1986).

Water softeners can reduce lead (Pb) accumulation considerably, while activated carbon filtration can also reduce Pb to a certain extent. Reverse osmosis can remove 94% to 98% of the Pb in drinking water at the point-of-use. Distillation also removes the Pb from drinking water. Dietary zinc partially alleviates Pb toxicity by decreasing the absorption rate of Pb. Diets that are high in sulphur-containing amino acids alleviate Pb toxicity through an increase in excretion rate (DWA & F, 1996)

1.3.1.4 Sodium chloride and total dissolved solids (TDS)

1.3.1.4.1 Sodium

Excess levels of sodium (Na) have a diuretic effect. The normal Na level in water is about 32 mg/L. Studies indicate that an Na level of 50mg/L is detrimental to broilers' performance if the sulphate level is also 50mg/L or higher and the chloride level is 14mg/L or higher (Carter & Sneed, 1996;Vohra, 1980).

Table 1.5 Guideline to the use of saline water for livestock and poultry (NRC, 1971)

Total Soluble Contents of Water (mg/L)	Comments
Less than 1000	This water has a relatively low level of salinity and should not present a serious problem to any class of livestock or poultry.
1000 - 2999	These waters should be satisfactory for all classes of livestock and poultry. They may cause temporary and mild diarrhoea in livestock not accustomed to them or watery dropping in poultry (especially at the high levels), but should not affect their health or performance.
3000 - 4999	These waters should be satisfactory for livestock, although they could possibly cause temporary diarrhoea or initially be refused by animals not accustomed to them. They are poor water for poultry, often causing watery faeces, increased mortality and decreased growth, especially in turkeys.
5000 - 6999	These waters can be used reasonably safely for dairy and beef cattle, sheep, swine and horse. It may be safer to avoid their use for animals approaching high levels of pregnancy, where reduced growth and production or increased mortality probably occurs.
7000 - 10000	These waters are unfit for poultry and probably for swine. There may be considerable risk if they are used for pregnant or lactating cows, horses, sheep, the young of these species, or any animals subjected to excessive heat stress or water loss.

1.3.1.4.2 Chloride

When too much chloride (Cl) is consumed, it has a detrimental effect on metabolism. A Cl level of 14mg/L is considered normal for well water. Studies have shown that a level of 14mg/L in drinking water can be detrimental to broilers if combined with 50mg/L of sodium. Cl levels as high as 25 mg/L are not problematic if the Na level is in the normal range (Carter & Sneed, 1996).

1.3.1.4.3 Total dissolved solids (TDS)

Total dissolved solid (TDS) or salinity is a measure of the quality of various inorganic salts dissolved in water. Saline water may detrimentally affect animal health and thus performance by rendering the water unpalatable. Palatability is also influenced by the type of salts present and not just the level of salinity. Magnesium sulphate is more harmful than sodium chloride or sodium sulphate (DWA & F, 1996).

Treatment to reduce salinity is possible, but would require regular maintenance and may not be economically feasible for remote watering locations. Suggested salinity level limits for different classes of livestock are outlined in Table 1.5.

Table 1.6 Water intakes and water/ feed ratios (g/bird/day) in broilers given different levels of NaCl (Coetzee, 2006)

Line	Days	Water Intake (g/bird/day)			Water /Feed Ratio		
		0.4%NaCl	0.8%NaCl	1.6%NaCl	0.4%NaCl	0.8%NaCl	1.6%NaCl
1	0-2	26.5	30.1	31.4	2.69	3.02	3.24
	2-4	38.9	42.5	48.4	2.43	2.55	3.01
	4-8	54.0	61.3	74.8	2.16	2.37	2.87
	8-12	73.3	84.7	107.5	2.09	2.26	2.85
	12-16	100.8	110.5	144.0	2.09	2.24	2.83
2	0-2	17.6	22.1	21.2	2.17	2.46	2.63
	2-4	30.7	36.8	40.3	2.15	2.44	2.83
	4-8	48.9	57.6	69.1	1.96	2.28	2.85
	8-12	68.3	78.4	103.0	1.89	2.14	2.80
	12-16	94.6	110.4	137.9	1.90	2.17	2.77

1.3.2 Water quality guideline

Water quality for livestock can be assessed by using a water quality guideline index system (WQGIS) for livestock, with emphasis on subterranean water sources and corresponding variables. The objective of such a system is to provide the following:

- a flexible management tool for decision-making on water quality;
- a means of incorporating site-specific information in risk assessment;
- supporting information on the various components and their interactions in biological systems required for decision-making;
- a water quality guidelines index system that can be updated as new research information becomes available.

The system, as described by Meyer (1998), uses a model approach that takes into consideration the type of livestock and production system, the environment and the ingestion rate of single or multiple water quality constituents to identify potentially hazardous constituents (those in excess of the recommended guideline) and constituents of concern (those within 10% of the recommended upper limit), which are then used for risk assessment. The ingestion of a specific water quality constituent (WQC) is directly linked to water intake. The system assumes that, if water intake can be predicted, the ingestion rate of a specific WQC can be calculated.

WQGIS has two application levels, generic and specific, each with its specifications. The generic application level is a static water quality guidelines application level that uses single-value comparisons. The specific application level is the preferred application for poultry watering, as it incorporates the site-specific influence on water ingestion (Casey *et al.*, 2001).

The aims of water quality formulations are:

- to identify the livestock production system and source of water;
- to identify relevant variables and their effect on livestock water quality norms;
- to develop water quality constituent guidelines for the respective livestock production systems.

The chemical composition of water is important for the success of livestock production (poultry). The chemical composition of water should not be merely to prevent adverse effects, but also to achieve optimum production (Meyer, 1998).

1.3.3 Poultry health

The poultry health deals with adverse effects on the health of poultry considering the presence of single or multiple water quality constituents (WQC) in the water at concentrations ranging from those that may precipitate a deficiency and to those that may induce toxicity. The adverse effects of WQC may be direct or indirect as far as the poultry is concerned. An example of a direct effect may be a high concentration of bacteria or potentially hazardous constituents (PHC) in the water that can have an adverse effect on the normal physiological processes of the body, resulting in substandard performance, poor health and reduced immunity. This may be caused by single or multiple PHC. Indirect effects may be high concentration of minerals in the water, leading to clogging of the water system and subjecting the birds to water deprivation that impacts negatively on performance. Flooding of the drinker can lead to wet litter that may lead to leg problems (Coetzee, 2006).

1.3.4 Product quality

Product quality deals with the potential hazard to the consumer of poultry products (tissues). The origin of the water potentially hazardous constituents (PHC) is normally the water source that the poultry have ingested. Product quality is very important in rural subsistence production systems, where poultry may be a significant item in the household diet. The risk is increased in the rural context owing to the prevalence of sensitive user groups. These may be pregnant women, infants, children, the elderly and immunity-compromised people who maybe HIV positive. Additional unfavourable environmental conditions such as heat and localised geochemical anomalies may further increase the risk of ingesting of PHC from multiple sources (Casey *et al.*, 2001).

1.4 Water

All life is intimately associated with water (Swenson & Reece, 1993). Water is the most critical nutrient to be consciously supplied to poultry, yet in most instances it is taken completely for granted (Leeson & Summer, 2005). Water is one of the most important components of the body, and has a multiplicity of functions. It circulates and transports substances, provides turgidity, is a medium for chemical reactions and contributes to regulating body temperature. An animal's water requirements are influenced by its physiology and environment. These include conditions like the physiological state of the animal, its stage of physiological development, gestation, physical exertion and ambient temperature-humidity indices. The condition of the feed may also increase the need for a high water intake, for example, a dry ration, dry matter intake, or inorganic salt intake (Swenson & Reece, 1993).

An inverse relationship exists between body weight and water turnover in the bird, which reflects the higher relative rate of the metabolism, the drinking rate, and the evaporation rate of small birds. The body-water content of adult males appears to be higher than that of females, which means that males have less fat in their bodies. Fat contains less water than lean body tissues (Skadhauge, 1981) and the proportion of body-water content decreases with increasing fat (Weiss, 1958).

Table 1.7 Distribution of body fluids in white leghorn female chickens (North & Bell, 1990)

		Percentage of Body Weight				
Age (weeks)	Weight (g)	Total Body Water	Intracelluar Water	Extracellular Water		
				Interstitial	Plasma	Total
1	55.1	72.4	11.4	52.2	8.7	61.0
2	108.4	71.6	21	42.3	7.3	50.6
3	175.3	70.5	24.6	39.1	6.8	45.9
4	241.8	68.4	24.1	38.3	6.0	44.3
6	372.3			36.8	5.9	42.7
8	527.3	68.7	26.6	36.1	6.1	42.2
16	1137.3	64.8	34.8	24.8	5.2	30.0
32	1759.5	57.3	31.1	21.7	4.6	26.2

Young birds have relatively more lean body tissues than adults and their total body water is also higher than that of older birds. The distributions of fluid in intracellular and extracellular compartments also vary with age (Table 1.7), with the higher water content found in 1-week old chicks and the lowest in those approaching maturity (Medway & Kate, 1959).

Table 1.8 Water consumption per 1000 leghorn pullet chicks per day (North & Bell, 1990)

Age in Days	Water Intake (L)
1	8.3
2	9.5
3	10.6
4	12.1
5	13.6
6	15.5
7	17.4
8	19.6
9	22.3
10	25.4
11	28.8
12	32.9
13	37.9
14	43.5

The body loses water in urine, sweat, expired gases and faeces. This lost body water is periodically compensated for by the continuous ingestion of water. There appears to be a mechanism ensuring that water lost is compensated for by water ingested (Swenson & Reece, 1993).

1.4.1 Water intake

A bird's water intake comes from the water drunk, moisture in the feed and metabolic water within its body (Casey *et al.*, 2001). The amount of water consumed increases with increases in air temperature, live weight and the rate of egg production. Higher salt and protein in the rations also increase the water intake (Medway & Kare, 1959). Feeding ingredients high in potassium, such as molasses or Soya bean meal, or calcium /phosphorus sources contaminated with magnesium result in increased water intake. This has no negative effect on the bird, but it increases water excretion, causing wet litter (Leeson & Summer, 2005).

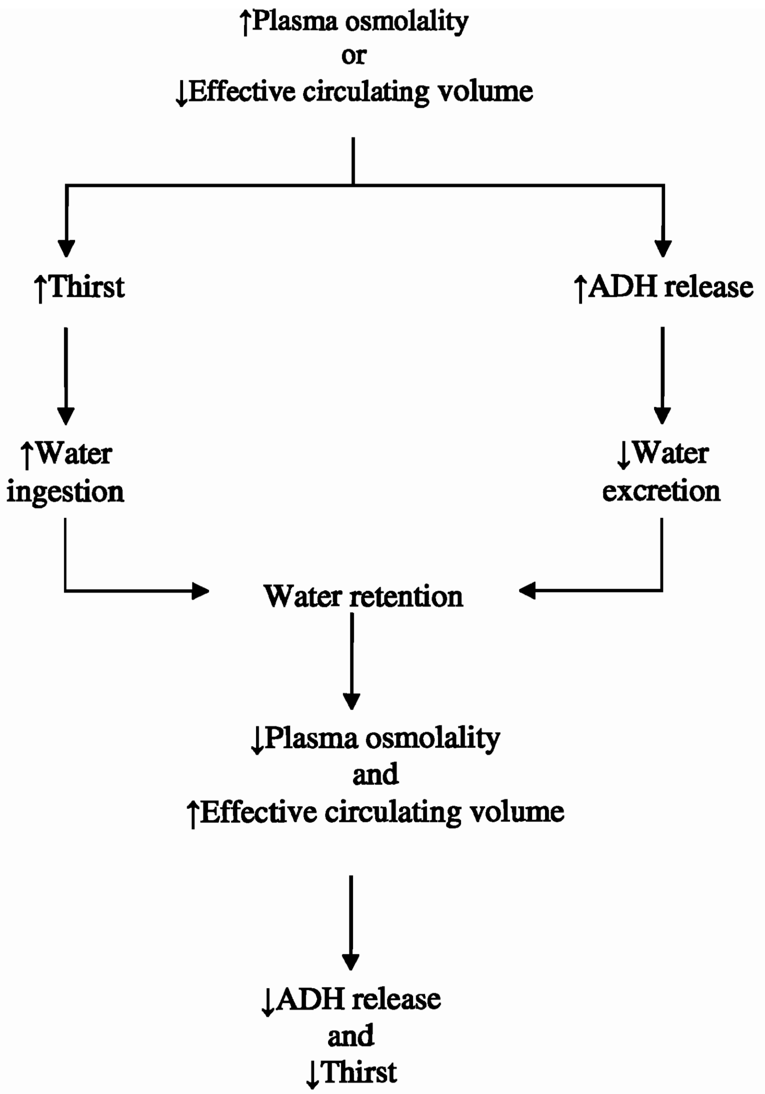

Figure 1 Cycle of events for the relief of hyperosmolality and hypovolemia (Adapted from Swenson & Reece, 1993)

Under normal physiological conditions for adult birds, water intake and output are controlled to maintain a constant level of water in the body. A positive water balance is found in the growing bird to accommodate growth. Drinking water is freely supplied under most commercial conditions, so dehydration should not occur. The adverse effects of shortterm reduced water intake are often the results of a concomitant reduction in feed intake (Leeson & Summers, 2005).

1.4.2 Water loss

The body loses water in urine, from the skin, and in expired gases and faeces (Swenson & Reece, 1993). Water loss in birds occurs predominantly with pulmocutaneous and evaporative water loss. There is an inverse relationship between evaporative water loss and body weight in the absence of temperature stress. Most of the evaporative water is lost from the respiratory system and air sacs, but relatively little through the skin and plumage (Casey *et al.*, 2001). It is generally expected that water consumption, like evaporative loss, increases with increasing environmental temperature. Birds store heat in a hot environment, and consequently undergo a rise in body temperature, resulting mainly from their inability to produce adequate evaporative cooling by panting (Bartholomew & Cade, 1963).

The amount of water lost in a one-day-old chick, expressed as a ratio of body weight, is appreciably higher than that of the adult. This high loss is believed to be, in part, the result of marked respiratory loss due to rapid breathing during the first day. This is thought to be owing to the changeover from allantoic respiration to pulmonary respiration, which results in relative anorexia. Evaporative water loss decreases as chickens mature (Casey *et al.*, 2001).

1.4.3 Electrolytes balance

The kidney contributes to the regulation of the total osmotic concentration of the body fluids as well as to the relative concentration of the various ions in the body fluids by controlling the volume of the water and the concentration of ions entering the urine. The blood plasma is a convenient sample of the body fluid, and the clear liquid, which is voided with the uric acid and faecal materials, can be treated as a sample of urine (Skadhauge, 1976).

Birds theoretically show an optimal electrolyte balance that enables maximum growth and the use of feed under thermoneutrality, in which the water/ electrolyte balance in the body is kept within narrow limits. The effect of the ionic balance of the diet on the performance of broilers may be related to variations in the acid-base balance. Osmoregulation is achieved by the homeostasis of these ions at the intra- and extracellular levels (Borges *et al.*, 2004).

The amount of chloride in the urine increases directly with the salinity of the drinking solution. The adaptation of birds to a dry climate involves decreases in relative evaporative water loss (mechanism not known), decrease in glomerulus's filtration rate (GRF), and high urine osmolality in the dehydrated state in conjunction with cloacal resorption parameters, allowing the urine to enter the cloaca without further water loss. Birds can tolerate a 7% to 13% increase in plasma osmolality (Skadhauge, 1976).

Maiorka *et al.* (2004) evaluated the effect of different levels of sodium (Na) and electrolytes (Na, potassium and chloride) in male Ross broilers' diets. There was a quadratic effect of increasing Na levels on feed and water consumption. The weight gain and feed conversion were improved significantly. The Na level that maximized water consumption in chickens was 0.45%, 0.40% for weight gain and 0.38% for feed conversion. The dietary electrolyte balances that improved broiler performance and feed consumption were 174 meq/kg and 163 meq/kg improved weight gain. These results show that both Na and electrolytes affect the broilers' performance.

1.5 Alleviator treatment

The alleviator treatment alleviates the occurrence of a particular water quality constituent (WQC) in the animals' tissues. In order to implement a financially viable alleviator treatment, certain questions should first be asked:

- To what extent does the presence of other WQC mitigate or exacerbate adverse effect due to a single WQC?
- To what extent does high WQC concentration contribute significantly to alleviating existing trace minerals deficiencies in the diet of the communities involved?
- To what extent does WQC present in animal products such as milk, meat and organs add to the dose intake already experienced by humans from the WQC present in water?

- To what extent can alleviator treatment be effective in livestock used to mitigate adverse effects in humans?
- What about the financial viability of building the necessary structures enabling a single water source to receive multiple treatments, each specific to a different water user?

These considerations and uncertainties serve as motivations for the development and implementation of constituent ingestion rate risk assessments designed specifically for rural communal livestock production systems (Casey *et al.*, 2001). These writers (1998) concluded that the palatability of water may be improved by the addition of either sodium sulphate or sodium chloride in the drinking water, depending on the position of the zone of preferences. A chemical treatment of 25mg/L Boron + 425 mg/L NaCl + 600 mg/L $MgSO_4$ + 600 mg/L Na_2SO_4 + 375 mg/L $CaCl_2$, with a SO_4/TDS ratio of 0.296 and a TDS of 3000 mg/L, was significantly beneficial, in terms of the final weight reached, as an alleviator treatment administered in the drinking water of Afrikaner steers exposed to a water fluoride of 20 mg/L showed (Casey *et al.*, 1998).

1.6 Aim of the study

The aim of the study was to test the effects of total dissolved solids (TDS) and Bromide (Br) on the accumulation of water-borne potentially hazardous chemical constituents Arsenic (As) and Lead (Pb) in the drinking water of broilers at concentrations that exceed the recommended guideline levels of 0.01mg/L.

CHAPTER 2
MATERIAL AND METHODS

2.1 Animals

The animals used in this experiment were 336 mixed Ross (genotype 788) broiler chickens purchased from a commercial hatchery in Pretoria on the day of hatching. The chickens were vaccinated against Newcastle Disease at the hatchery before they were delivered to the poultry unit on the Hatfield Research Farm of the University of Pretoria. No further vaccinations were carried out during the rearing stage and no medications were administered via drinking water to control the diseases. The veterinarian's service was available should any problem arise.

2.2 Housing

The experiment was conducted in an environmentally-controlled broiler house on the University of Pretoria research farm. The house had 28 pens, arranged in two rows of 14 pens on each side of the house. All 28 pens were used for this experiment, each containing 12 chickens. Each pen was fitted with a volumetrically-graded water cylinder to accurately measure water intake, two round pan feeders and nipple drinkers.

2.3 Nutrition

The animals all received the same standard commercial broiler feed purchased from a reputable commercial feed company. The chickens were fed ***ad libitum*** in two phases from the first day until the last day of the experiment to simulate the normal feeding practices in a manual, rural production unit. Days 1-16 were the starter phase, followed by days 16-42, the finisher phase. This feeding regime simplifies feeding in a non-mechanized small-scale rural production system (Casey & Meyer, 2001), and is still within acceptable feeding procedure (Aviagen, 2002). Raw material compositions used in the diets are presented in Table 2.1 and nutrients compositions are in Table 2.2. Nutrients used are recommended for achieving good feed conversion ratio (FCR).

2.3.1 Starter and finisher diet

Table 2.1 Raw material compositions of starter and finisher (%) diets

Raw Materials	Starter	Finisher
Soya Oilcake Meal	28.60	17.03
Sunflower Oilcake Meal	2.45	2.88
Synthetic Lysine	0.29	0.30
Synthetic Methionine	0.21	0.14
Synthetic Threonine	0	0.02
Poultry By Product Meal	2.45	3.90
Yellow Maize	60.34	71.42
Blended Oil	1.92	1.48
Limestone	1.63	1.38
Monocalcium Phosphate	1.34	0.68
Sodium Chloride	0.49	0.43
Growth Promoter	0.03	0.03
Premix	0.22	0.20
Coccidiostat	0.05	0.05
Total (%)	100.01	99.92

Table 2.2 Nutrients compositions of starter and finisher (%) diets

Nutrients	Starter	Finisher
Protein (g/kg)	20.90	17.41
Fat (g/kg)	5.24	5.17
Fibre (g/kg)	3.26	3.23
Ash (g/kg)	6.17	4.74
% ME (MJ/ kg)	12.95	13.42
Digestible Lysine (g/kg)	1.10	0.92
Digestible Methionine (g/kg)	0.49	0.38
Digestible Methionine & Sulphur (g/kg)	0.77	0.64
Digestible Threonine (g/kg)	0.70	0.59
Digestible Tryptophan (g/kg)	0.20	0.15
Digestible Arginine (g/kg)	1.29	1.04
Digestible Isoleucine (g/kg)	0.80	0.64
Digestible Leucine (g/kg)	1.65	1.41
Digestible Valine (g/kg)	0.89	0.75
Digestible Histamine (g/kg)	0.49	0.40
Ca (g/kg)	1.00	0.80
Available Phosphorus (g/kg)	0.43	0.31
Na (g/kg)	0.20	0.18
Cl (g/kg)	0.35	0.32
K (g/kg)	0.85	0.66

2.4 Treatments

The treatments were formulated to simulate the concentrations of selected water quality constituents (WQC) in water from boreholes used to water livestock in rural communal systems. The chemicals used for producing different water treatments were AR-grade. The treatments were prepared at the NutriLab laboratory at the University of Pretoria.

The treatments were:

- Control = <500 mg/L TDS + < 0.005 mg/L (Br + As + Pb)
- Treatment 1 = As = 0.1 mg/L + Br = 1 mg/L and Pb = 0.1 mg/L
- Treatment 2 = Treatment 1+ 1500 mg/L TDS
- Treatment 3 = Control + 1500 mg/L TDS

The water containing chemicals delivered a final concentration of 0.1mg As/L as arsenic-trioxide (As_2O_3), 0.1mg Pb/L as lead nitrate Pb $(NO_3)_2$ and 1 mg Br/L as sodium bromide (NaBr). The lowered As level was selected in an attempt to identify an acceptable hazardous margin in excess of the recommended guideline limit (0.01 mg/L). Bromide was included because it had been repeatedly observed in rural groundwater at elevated concentrations and because of possible protective effects on Potentially Hazardous Chemical Constituents (PHCC) exposure. Sodium chloride (NaCl) was chosen as a potential alleviator based on the positive results obtained in cattle and sheep (Casey et al., 2001) and because high Total Dissolved Solids (TDS) concentrations had been observed in some rural communities.

2.5 Parameters monitored

Parameters recorded were live weight, water intake, feed intake, lighting and temperature. These factors are important components of good management as well as playing an important role in statistical analysis.

2.5.1 Body weight

The initial weight of the chickens was recorded on day 1 of the experiment, after which they were weighed every week. All the chickens were weighed per pen for the duration of the experiment by being placed into a plastic container.

2.5.2 Water intake

The water was administered via the nipple drinker from day 1. Each pen is fitted with a special water cylinder specifically installed for this kind of experiment. These water cylinders facilitate recording the water intake per pen. Water intake was measured every morning from the first until the last day of the experiment. This was carried out consistently at the same time of day to minimize errors caused by variation in water intake at different times of day.

2.5.3 Feed intake

Feed intake is one of the important parameters in chicken management (Aviagen, 2002). Recording it helps predict feed conversion ratio (FCR), which is a good measurement of how well the chickens convert the feed into meat/growth. Feed intake was measured weekly, from the first day of the experiment.

2.5.4 Lighting

Lighting was provided for 24 hours during the first two weeks and thereafter for 18 hours (six hours of darkness) per day throughout the experiment. The lighting programme used was the result of consultation with the rural production projects. The light intensity (lux) was not measured during the experiment.

2.5.5 Temperature

The temperature (^{o}C) measurements in the house were taken three times a day, in the morning (8:00), mid-day (12:00) and evening (18:00). Weather changes were also recorded, because they could affect the chickens' feed and water intake. Humidity was not measured.

2.6 Tissue sampling and analysis

2.6.1 Sample size

Two out of twelve broiler chickens per pen were slaughtered on day 42 of the trial at the Meat Science Abattoir of the University of Pretoria. Only two broiler chickens per pen were slaughtered because of limited manpower and financial constraints, and the rest of the chickens were to be sold to cover the financial obligations of the experiment. Birds were killed according to the normal industrial practice (NDA, 2000) of stunning them and then severing the jugular. The feathers were removed and various body parts collected from each carcass: thighs, breasts, livers, hearts and kidneys.

2.6.2 Organs sampled

The hearts and the kidneys were sampled in the same way. Within the treatments, replicates one and two, three and four, and five, six and seven were pooled together, amounting to three final replicates per treatment. Pooling the samples in this way ensured that sufficient sample mass was obtained for purposes of analysis.

The liver samples were not pooled together within replicates; seven replicates per treatment were observed.

The thighs and the breasts were sampled in the same way. Their larger sample size made it easy to sample the left and the right thighs and breasts separately. The left and right thighs were later pooled together for observation results; the same was done with the breasts. Seven replicates per treatment were thus observed.

The samples were stored in a freezer and later delivered to the Agricultural Research Council-Institute of Soil Climate Water (ARC ISCW) for analysis for the levels of the treatment elements As, Br, Pb, Se, Mo, Cd, Mn, Cr and I. The ARC ISCW conducted the laboratory analyses of all the samples according to the technique prescribed by the ARC ISCW (ARC, 1998). Each observation set was ground and analyzed using standard inductively coupled mass spectrometry ICP-MS techniques to determine trace element concentrations. The samples were digested using microwave-assisted acid digestion of siliceous and organically-based matrices, as summarized in the next paragraph.

A representative sample of up to 0.5 g is digested in 9 mL of concentrated nitric acid and 3 mL hydrofluoric acid for 15 minutes using microwave heating with a suitable laboratory microwave system. The vessel is sealed and heated in the microwave system. The temperature profile is specified to permit specific reactions, reaching 180 ± 5°C in less than 5.5 minutes. It remains at 180 ± 5°C for approximately 9.5 minutes for the completion of the specific reaction. After cooling, the vessel contents are filtered, centrifuged or allowed to settle and then decanted, diluted to volume, and analyzed by the appropriate ICP-MS method (ARC ISCW, 1998).

ICP-MS measures ions produced by a radio-frequency inductively coupled plasma. Analyte species originated in a liquid are nebulised and the resulting aerosol is transported by argon gas into the plasma torch. The ions produced by high temperature are entrained in the plasma gas and introduced, by means of an interface, into a mass spectrometer. The ions produced in the plasma are sorted according to their mass-to-charge ratios and quantified with channel electron multiplier interferences (ARC ISCW, 1998).

In addition to analysing the tissues for the treatment elements, the tissues were also scanned for the microminerals Se, Mo, Cd, Cu, Mn and Cr. These microminerals were in the diet or may have been in the water and it was considered a good opportunity to observe whether TDS may have had an effect on their concentrations in the tissues.

2.7 Statistical analysis

Broiler chickens and treatments were randomly allocated in the house. The SAS (Statistical Analyses System®) software system was used to determine the significance of differences between treatments for WQC analysis. General linear estimate and hypothesis tests were used for regression analysis.

The significance of the differences between treatments were determined by means of the Bonveroni test at a $P< 0.05$ significance level.

CHAPTER 3
RESULTS

The results of the selected elements analyzed in the respective chicken tissues and organs are presented under different sections.

3.1 Liver

T1 had elevated concentrations of As, Br and Pb that yielded the highest significant levels of Br and Pb amongst all the treatment groups (P<0.05) (Table 3.1). The concentrations of As, Mn, Cr and I differed non-significantly. Concentrations of Cu for T1 were significantly similar to those in the Control, or municipal water treatment, whilst the levels of Mo and Cd were comparable to T2, which is the alleviator treatment. In T2, the levels of Se, Mn, As, Br and Pb were significantly (P<0.05) reduced, as compared with T1. Levels of Cu in T2 were significantly similar to T3. Liver results for T3 retained the lowest means for Cr, As, Cd and I (P<0.05).

Table 3.1 Elements in liver tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Element[1]	**Control (C)**	**T1**	**T2**	**T3**
As	0.3710 ^{a} (0.052)	0.3980 ^{a} (0.088)	0.3860 ^{a} (0.056)	0.3560 ^{a} (0.048)
Pb	0.2450 ^{ab} (0.104)	0.3120 ^{a} (0.035)	0.2720 ^{b} (0.022)	0.2420 ^{ab} (0.077)
Se	0.9930 ^{ab} (0.211)	1.1270 ^{a} (0.147)	0.9870 ^{b} (0.096)	1.110 ^{a} (0.160)
Mo	0.6390 ^{a} (0.176)	0.8550 ^{b} (0.169)	0.790 ^{b} (0.100)	0.8220 ^{b} (0.156)
Cd	0.0950 ^{a} (0.010)	0.1090 ^{ab} (0.046)	0.090 ^{ab} (0.011)	0.0840 ^{b} (0.006)
Br	10.610 ^{a} (3.449)	14.890 ^{b} (4.542)	11.920 ^{ab} (3.890)	12.030 ^{ab} (4.120)
Cu	3.890 ^{a} (1.122)	4.4980 ^{a} (0.775)	5.4340 ^{b} (3.174)	4.1590 ^{b} (0.571)
Mn	3.5730 ^{a} (0.580)	4.2140 ^{a} (0.616)	3.8010 ^{a} (0.444)	3.5560 ^{a} (0.571)
Cr	1.2180 ^{a} (0.146)	1.160 ^{a} (0.285)	1.0540 ^{a} (0.232)	1.1170 ^{a} (0.255)
I	0.2480 ^{a} (0.015)	0.2520 ^{a} (0.273)	0.2520 ^{a} (0.022)	0.2340 ^{a} (0.042)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the P< 0.05 level.

There is a clear trend for the alleviator treatment (T2) to retain the lower analyzed elements levels in the liver as compared to elevated treatment (T1). The Control and T3 retained more or less similar levels of analyzed elements in the liver, with the control treatment yielding slightly higher observed results. The results suggest that Br and NaCl may have been playing a role in the T3 obtaining slightly lower mean values compared to those of the Control.

3.2 Kidney

T1 retained the highest significant levels of As and Br amongst all the treatments (P<0.05) (Table 3.2). T3 retained the lowest significant kidney levels for Pb and the Control retained the highest levels (P<0.05). There was significantly little difference amongst most of the analyzed kidney elements, but there is a suggestion that the alleviator treatment (T2) tends to retain the lower element results than elevated treatment (T1). The retention of kidney elements between control and T3 was significantly similar.

Table 3.2 Elements in kidney tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Element[1]	Control	T1	T2	T3
As	0.3465 ^{b} (0.0225)	0.4515 ^{a} (0.0542)	0.3538 ^{b} (0.0285)	0.3573 ^{b} (0.0304)
Pb	0.0914 ^{a}(0.0249)	0.0652 ^{b}(0.0086)	0.0880 ^{a} (0.0189)	0.0388 ^{c} (0.0104)
Se	0.9040 ^{b}(0.0858)	1.1218 ^{a} (0.1959)	1.1323 ^{a} (0.1774)	1.0365 ^{ab}(0.1611)
Mo	0.9346 ^{a} (0.0506)	1.0583 ^{a} (0.1073)	0.9920 ^{a} (0.1290)	0.9355 ^{a} (0.0828)
Cd	0.0976 ^{a} (0.0107)	0.1041 ^{a} (0.0361)	0.0905 ^{a}(0.0212)	0.0830 ^{a} (0.0166)
Br	6.6110 ^{b} (0.6216)	11.2188 ^{a}(1.5233)	6.9408 ^{b} (0.7694)	6.0407 ^{b}(1.2642)
Cu	3.3778 ^{a} (0.1663)	3.4425 ^{a} (0.1101)	3.3878 ^{a} (0.2357)	3.5021 ^{a} (0.3939)
Mn	2.2205 ^{a} (0.1683)	2.3015 ^{a} (0.1907)	2.2298 ^{a} (0.0493)	2.3520 ^{a} (0.2022)
Cr	0.6395 ^{a} (0.0916)	0.7502 ^{a} (0.1101)	0.7150 ^{a} (0.1105)	0.7113 ^{a} (0.1785)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the P< 0.05 level.

3.3 Heart

A trend similar to those in the liver and kidney results were observed in the heart elements results (Table 3.3). Alleviator treatment (T2) attained the lowest significant results for As amongst the four treatments and lower results for Br in comparison to T1. The treatments with total dissolved solids (TDS), which are T2 and T3, mostly retained the lowest observed results for the analyzed elements in general.

Table 3.3 Elements in heart tissue (mg/kg DM) of broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Element[1]	**Control**	**T1**	**T2**	**T3**
As	0.2395 ^{b}(0.0394)	0.2990^{a} (0.0221)	0.2123 ^{c} (0.0228)	0.2403^{b} (0.0240)
Pb	0.1295^{a}(0.0591)	0.1327^{a}(0.0468)	0.0781^{b}(0.0149)	0.0675^{b} (0.0169)
Se	0.0505^{b} (0.0599)	0.8138^{a}(0.2184)	0.6243^{b}(0.0611)	0.5176^{b}(0.0618)
Mo	0.0678^{b}(0.0074)	0.0757^{ab}(0.0085)	0.0908^{a}(0.0266)	0.0726^{ab}(0.0097)
Cd	0.0463^{b}(0.0071)	0.0397^{b} (0.0064)	0.0590^{a} (0.0102)	0.0390^{b}(0.0042)
Br	6.0163^{c}(1.3868)	20.524^{a}(1.0072)	13.0090^{b}(2.5291)	6.00270 ^{c} (1.5138)
Cu	3.6450^{a}(0.5973)	3.9021^{a}(1.2132)	3.7065^{a}(0.2266)	3.6310^{a}(0.4127)
Mn	0.6502^{a}(0.1284)	0.5653^{a}(0.0380)	0.6061^{a}(0.0188)	0.6273^{a}(0.0601)
Cr	0.5793^{b}(0.56581)	0.6076^{b} (0.0064)	0.7115^{a} (0.0793)	0.7833^{a}(0.0042)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the $P< 0.05$ level

3.4 Thigh

Concentrations of elements in the right thigh are presented in Table 3.4. T1 retained the highest significant elements results for Br amongst the four treatments, while other treatments retained the same levels ($P<0.05$). There was no significant difference for As, Pb, Mo, Cd, Cu, Mn, Cr and I amongst all the treatments. The Se levels for T1 and T2 were significantly comparable ($P<0.05$). T2 obtained the lowest of the thigh results for most of the elements analyzed compared with T1, although these were not always significant.

Table 3.4 Elements in the right thigh tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Elements[1]	**Control**	**T1**	**T2**	**T3**
As	0.2440 [a] (0.048)	0.2933 [a] (0.059)	0.2860 [a] (0.057)	0.2670 [a] (0.039)
Pb	0.1020 [a] (0.040)	0.2270 [a] (0.132)	0.1200 [a] (0.050)	0.1620 [a] (0.076)
Se	0.553 [a] (0.127)	0.496 [ab] (0.121)	0.4750 [ab] (0.091)	0.4324 [b] (0.087)
Mo	0.0390 [a] (0.011)	0.0470 [a] (0.008)	0.0440 [a] (0.011)	0.0460 [a] (0.014)
Cd	0.0310 [a] (0.016)	0.030 [a] (0.016)	0.0340 [a] (0.020)	0.0310 [a] (0.029)
Br	9.4230 [a] (3.260)	14.530 [b] (6.618)	9.850 [a] (4.251)	8.890 [a] (4.485)
Cu	0.702 [a] (0.202)	0.6960 [a] (0.162)	0.650 [a] (0.146)	0.7530 [a] (0.198)
Mn	0.1539 [a] (0.042)	0.1599 [a] (0.055)	0.1338 [a] (0.037)	0.1388 [a] (0.198)
Cr	0.8021 [a] (0.169)	0.8517 [a] (0.187)	0.8573 [a] (0.173)	0.7915 [a] (0.100)
I	0.1021 [a] (0.007)	0.0144 [a] (0.008)	0.0123 [a] (0.008)	0.0088 [a] (0.006)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the $P < 0.05$ level.

In the left thigh tissue As and Se were lower in the T2 ($P<0.05$), while T3 retained the lowest values for As. In general, treatments with TDS (T2 & T3) retained the lowest elements results as compared with non-TDS (Control & T1).

Table 3.5 Elements in left thigh tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Elements[1]	**Control**	**T1**	**T2**	**T3**
As	0.2360 [ab] (0.063)	0.2560 [a] (0.072)	0.2110 [ab] (0.049)	0.1970 [b] (0.042)
Pb	0.1620 [a] (0.080)	0.2460 [b] (0.154)	0.1110 [a] (0.035)	0.1710 [ab] (0.158)
Se	0.3930 [a] (0.102)	0.5010 [a] (0.177)	0.3160 [b] (0.047)	0.3810 [a] (0.083)
Mo	0.0380 [a] (0.011)	0.0840 [a] (0.123)	0.0340 [a] (0.006)	0.0370 [a] (0.004)
Cd	0.0130 [a] (0.021)	0.0160 [a] (0.026)	0.0570 [a] (0.174)	0.0080 [a] (0.013)
Br	6.050 [a] (3.424)	13.3940 [b] (5.106)	4.6750 [a] (2.806)	6.7640 [a] (2.499)
Cu	0.680 [a] (0.220)	0.6610 [a] (0.155)	0.7130 [a] (0.221)	0.7460 [a] (0.170)
Mn	0.0320 [ab] (0.051)	0.0610 [a] (0.053)	0.0840 [ab] (0.129)	0.0160 [ab] (0.170)
Cr	0.590 [a] (0.081)	0.7860 [b] (0.243)	0.5610 [a] (0.125)	0.5450 [a] (0.104)
I	0.0010 [a] (0.005)	0.0250 [a] (0.062)	0.0010 [a] (0.001)	Nd

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the $P < 0.05$ level. Nd = not detected below 0.001 mg/kg

A pattern can be seen in both the right and left thigh showing that T2 tends to retain the lowest tissue results. T1 attained the highest result for the analyzed elements, but this was not always significant.

The results for each chicken were pooled together to eliminate the differences between the left and right thighs (Table 3.6). The thigh tissue results for the elevated elements treatment (T1) retained the highest significant values for Pb, Se, Br and Cr amongst all the treatments (P<0.05). All the treatments retained the same thigh tissue results for Mo, Cd, Cu, Mn and I.

Table 3.6 Pooled results of both left and right thighs (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Elements[1]	**Control**	**T1**	**T2**	**T3**
As	0.2400^{ab} (0.055)	0.2740^{a} (0.067)	0.2490^{ab} (0.064)	0.2320^{a} (0.053)
Pb	0.1320^{a} (0.069)	0.2370^{b} (0.140)	0.1160^{a} (0.042)	0.1380^{a} (0.127)
Se	0.4360^{a} (0.096)	0.5350^{b} (0.165)	0.3950^{a} (0.107)	0.410^{a} (0.086)
Mo	0.0390^{a} (0.011)	0.0650^{a} (0.087)	0.0390^{a} (0.010)	0.0420^{a} (0.011)
Cd	0.0220^{a} (0.020)	0.0230^{a} (0.022)	0.0460^{a} (0.121)	0.020^{a} (0.025)
Br	7.7370^{a} (3.695)	13.9640^{b} (5.810)	7.2630^{a} (4.387)	7.8270^{a} (3.713)
Cu	0.6910^{a} (0.207)	0.6780^{a} (0.156)	0.6820^{a} (0.186)	0.7500^{a} (0.180)
Mn	0.0930^{a} (0.077)	0.110^{a} (0.073)	0.177^{a} (0.096)	0.0770^{a} (0.068)
Cr	0.6960^{a} (0.169)	0.8190^{b} (0.215)	0.7090^{ab} (0.211)	0.6680^{a} (0.161)
I	0.0070^{a} (0.008)	0.0190^{a} (0.043)	0.0060^{a} (0.008)	0.0040^{a} (0.006)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the P< 0.05 level.

3.5 Breast

The results for Br, Mo and Cu yielded significant differences results among the treatments (Table 3.7). T1 and T2 retained similar significant level results (P<0.05), and C and T3 were significantly similar for Br results amongst all the treatments. Bromide results were significantly similar between treatment T1 and T2 and also between C and T3 (P<0.05). The same trend was observed in the right thigh results, although it was not always significant. Cd, Mn and I were not detected, on account of their occurrence at a concentration level lower than the detectable levels of 0.001mg/kg in all the groups.

Table 3.7 Elements in right breast tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Elements[1]	**Control**	**T1**	**T2**	**T3**
As	0.1930^{a} (0.045)	0.2020^{a} (0.084)	0.2060^{a} (0.074)	0.1830^{a} (0.067)
Pb	0.0860^{a} (0.075)	0.090^{a} (0.096)	0.0440^{a} (0.030)	0.0470^{a} (0.042)
Se	0.4620^{a} (0.304)	0.5800^{a} (0.325)	0.5910^{a} (0.384)	0.4290^{a} (0.239)
Mo	0.0110^{a} (0.007)	0.0190^{b} (0.010)	0.0160^{ab} (0.007)	0.0170^{b} (0.003)
Br	5.0340^{a} (4.891)	7.5410^{b} (5.605)	7.270^{b} (4.748)	4.9790^{a} (4.107)
Cu	0.6330^{ab} (0.103)	0.6180^{a} (0.079)	0.6540^{ab} (0.068)	0.6890^{b} (0.057)
Cr	0.8420^{a} (0.114)	0.8630^{a} (0.084)	0.830^{a} (0.119)	0.8410^{a} (0.070)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the P< 0.05 level.

Left breast tissue element levels mean and standard deviation results are presented in Table 3.8. The results for Cr and Pb differed significantly amongst the treatments, with T1 retained the highest observed results levels for these elements. These showed a trend that clearly illustrates that T1 retained the highest elements, as compared with T2, that is similar in the liver, kidney, heart and thigh results.

Table 3.8 Elements in left breast tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Elements[1]	**Control**	**T1**	**T2**	**T3**
As	0.1080^{a} (0.030)	0.1320^{a} (0.036)	0.1240^{a} (0.032)	0.110^{a} (0.047)
Pb	0.1590^{a} (0.033)	0.2960 ^{b}(0.175)	0.2170^{ab}(0.210)	0.1620^{a} (0.083)
Se	0.4080^{a} (0.097)	0.5060^{a} (0.144)	0.4520^{a} (0.145)	0.3930^{a} (0.184)
Mo	0.0460^{a} (0.007)	0.0530^{a} (0.009)	0.0480^{a} (0.010)	0.0470^{a} (0.017)
Cd	0.0080^{a} (0.016)	0.0070^{a} (0.008)	0.0020^{a} (0.004)	0.0020^{a} (0.003)
Br	5.0980^{a} (4.193)	7.8650^{a} (4.555)	7.1380^{a} (4.829)	5.5290^{a} (4.403)
Cu	0.4070^{a} (0.059)	0.4350^{a} (0.041)	0.4390^{a} (0.057)	0.3830^{a} (0.128)
Mn	0.0190^{a} (0.039)	0.0290^{a} (0.032)	0.0130^{a} (0.030)	0.0010^{a} (0.128)
Cr	0.7940^{a} (0.087)	0.8880^{b} (0.119)	0.810^{ab}(0.117)	0.7110^{a} (0.266)
I	0.0050^{a} (0.004)	0.0120^{a} (0.015)	0.0030^{a} (0.004)	0.0030^{a} (0.005)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the P< 0.05 level.

Table 3.9 Pooled breast tissue (mg/kg DM) from broilers exposed to As, Pb, Br and TDS in the drinking water under intensive production conditions

	Means and SD (mg/kg DM basis)			
Elements[1]	**Control**	**T1**	**T2**	**T3**
As	0.1510^{a} (0.057)	0.1670^{a} (0.072)	0.1650^{a} (0.069)	0.1460^{a} (0.067)
Pb	0.1230^{ab} (0.068)	0.1930^{a} (0.174)	0.1300^{ab} (0.171)	0.1040^{ab} (0.087)
Se	0.4350^{a} (0.222)	0.5430^{a} (0.249)	0.5220^{a} (0.292)	0.4110^{a} (0.209)
Mo	0.0280^{a} (0.019)	0.0360^{a} (0.019)	0.0320^{a} (0.018)	0.0320^{a} (0.019)
Cd	0.0040^{a} (0.012)	0.0030^{a} (0.006)	0.0010^{a} (0.003)	0.0010^{a} (0.002)
Br	5.0660^{a} (4.455)	7.7030^{a} (4.998)	7.2040^{a} (4.684)	5.2540^{a} (4.173)
Cu	0.5200^{a} (0.141)	0.5260^{a} (0.112)	0.5460^{a} (0.125)	0.5350^{a} (0.184)
Mn	0.0090^{a} (0.029)	0.0140^{a} (0.027)	0.0060^{a} (0.022)	0.0005^{a} (0.002)
Cr	0.8180^{ab} (0.102)	0.8750^{a} (0.101)	0.8200^{ab} (0.116)	0.7760^{ab} (0.202)
I	0.0020^{a} (0.004)	0.0060^{a} (0.012)	0.0010^{a} (0.003)	0.0010^{a} (0.004)

[1]Means within a row for each element bearing the same superscripts do not differ significantly at the P< 0.05 level.

A pattern can be observed for many of the elements in all the tissues and organs analyzed. Although this is not always significant, it was found that non-As-Br-Pb recipient groups C and T3 present with similar means, while groups T1 and T2, which received As-Br-Pb, tended to be dissimilar, with T1 tending to present with higher means than T2. In most of the tissues, there were no significant differences between C and T3, but T3 tended to present with lower means. Groups receiving the TDS treatment, which was achieved by adding NaCl, tended to return lower means than those of their direct comparison groups (C - T3, and T1-T2). Pooled thighs results shows that As, Br, Pb and Se returned significantly higher results for treatment (T1), while the alleviator treatment (T2) retained the lower means. The same trend is observed in the breast results for As, Br and Se, although this is not always significant at the P< 0.05 level. Chromium also followed the same trend among these pooled tissues, with T1 differing significantly from C, and T3 from both C and T1.

3.6 Live weight

The weekly mean and standard deviation body weight results for each treatment group shown in Table 3.10 illustrates that there is no significant difference between the treatments (P<0.05). No abnormal health-related problems were seen during the period of the experiment.

Table 3.10 Live weight (kg) of broilers exposed to As, Pb, Br and TDS in the drinking water over the production cycle

	Means and SD (kg/bird)[2]			
Observation[1]	**Control**[3]	**T1**[3]	**T2**[3]	**T3**[3]
LW 1 (week 1)	0.30[a] (0.006)	0.298[a] (0.013)	0.302[a] (0.012)	0.306[a] (0.010)
LW 2 (week 2)	0.636[a] (0.019)	0.633[a] (0.011)	0.62[a] (0.016)	0.638[a] (0.020)
LW 3 (week 3)	1.110[a] (0.037)	1.119[a] (0.021)	1.11[a] (0.023)	1.117[a] (0.045)
LW 4 (week 4)	1.694[a] (0.072)	1.727[a] (0.041)	1.70[a] (0.044)	1.702[a] (0.062)
LW 5 (week 5)	2.230[a] (0.082)	2.334[a] (0.087)	2.323[a] (0.036)	2.310[a] (0.077)
LW 6 (week 6)	2.90[a] (0.107)	2.901[a] (0.128)	2.887[a] (0.080)	2.891[a] (0.137)

[1]Observations for live weight based on weekly individual recordings for bird in each replicate.

[2]Means for live weight calculated from observations of 7 replicates per group with 12 birds per replicate.

[3]Means within a row for replicates bearing the same superscript do not differ significantly (P < 0.05 level).

3.7 Feed intake

There were no significant differences found for feed intake between treatments or in replicates within treatments (Table 3.11). The results suggest that the levels of selected elements administered or occurring in municipal water do not have any influence on the chickens' growth. However, these results may mean that the exposure period for this experiment was too short, especially in the context of rural communal chicken farming. In rural communal farming, chickens are raised under stressful conditions and are given poor drinking water and feed.

Table 3.11 Feed intake (mg/bird/d) of broilers exposed to As, Pb, Br and TDS in the drinking water over the production cycle

	Means and SD (mg/bird/day)[2]			
Observation[1]	**Control**[3]	**Treatment**[3]	**T2**[3]	**T3**[3]
FI 1 (week 1)	32.8[a] (0.804)	31.2[a] (4.617)	32.7[a] (0.938)	32.8[a] (0.810)
FI 2 (week 2)	39.4[a] (4.184)	37.2[a] (2.529)	38.2[a] (2.480)	39.3[a] (3.151)
FI 3 (week 3)	105.4[a] (11.947)	114.2[a] (6.664)	113.8[a] (3.802)	115.3[a] (8.876)
FI 4 (week 4)	157.5[a] (6.249)	159.1[a] (4.598)	152.6[a] (4.055)	156.6[a] (10.222)
FI 5 (week 5)	178.6[a] (10.989)	186.9[a] (28.235)	176.6[a] (10.614)	177.5[a] (6.674)
FI 6 (week 6)	162.5[a] (12.658)	163.8[a] (12.452)	159.5[a] (13.557)	163.3[a] (10.190)

[1]Observations for feed intake based on weekly replicate recordings.

[2]Means for feed intake calculated from observations of 7 replicates per group with 12 birds per replicate.

[3]Means within a row for groups bearing the same superscript do not differ significantly (P < 0.05 level).

3.8 Water intake

The water intake observed in Table 3.12 indicates that the TDS treatments (T2 and T3) significantly increased water intake from the second week throughout the course of the exposure period. No significant differences were observed in the TDS treatment groups (T2 and T3) or in the non-TDS treatment groups (C and T1). Average increases in water intake between the TDS and non-TDS groups rose from an initial 7% in week 1 to 13% in week 5, followed by a reduction in the last weeks to ca. 9%.

Table 3.12 Water intake (mL/bird/d) of broilers exposed to As, Pb, Br and TDS in the drinking water over the production cycle

	Means and SD (mL/bird/day) [2]			
Observation[1]	**Control C[3]**	**T1[3]**	**T2[3]**	**T3[3]**
WI 1 (week 1)	93 [ab] (0.009)	92 [a] (0.002)	99 [ab] (0.012)	103 [a] (0.001)
WI 2 (week 2)	153 [a] (0.012)	150 [a] (0.005)	173 [b] (0.011)	174 [b] (0.008)
WI 3 (week 3)	180 [a] (0.005)	176 [a] (0.007)	203 [b] (0.009)	197 [b] (0.009)
WI 4 (week 4)	238 [a] (0.006)	232 [a] (0.007)	260 [b] (0.013)	272 [b] (0.013)
WI 5 (week 5)	298 [a] (0.013)	288 [a] (0.012)	326 [b] (0.013)	320 [b] (0.016)
WI 6 (week 6)	310 [a] (0.012)	301 [a] (0.012)	340 [b] (0.035)	338 [b] (0.021)

[1]Observations for water intake based on weekly replicate recordings.

[2]Means for water intake calculated from observations of 7 replicates per group with 12 birds per replicate.

[3]Means within a row for replicates bearing the same superscript do not differ significantly ($P < 0.05$ level).

The resulting average intakes for As and Pb were 0.0236 mg/d/bird for T1, 0.0262 mg/d/bird for T2, and for Br 0.236 mg/d/bird for T1 and 0.262 mg/d/bird for T2 over the trial period. The total ingestion of As and Pb over the duration of the trial was 1.325 mg for T1 and 1.469 mg for T2 respectively for each element, with 13.257 and 14.697 mg of Br being ingested for T1 and T2 respectively over the trial period. Correlations between feed and water intakes were most significant for group T3 (0.90 for week 2 and 0.95 for week 6), but generally weak or negative (although non-significant) for the other groups, with an exception observed for group C on week 5 (0.92).

CHAPTER 4
DISCUSSION

An investigation into rural communal water quality by Casey and Meyer (2000) showed both animals and humans may be at risk when exposed to problematic water sources. The effects of WQC are a function of the type and character of the WQC, the intake rates and exposure to WQC, the type of animal and its physiology and the demands of the environment. The risk is to both livestock and human users of the respective water source. It was shown furthermore that where livestock and humans use the same water source, the livestock can be an effective indicator species of the risk posed to humans. The challenge is therefore to identify WQC in a water source that may be Constituents of Concern (COC) or Potentially Hazardous chemical Constituents (PHCC).

The guidelines for WQC are merely guidelines and not absolute figures because of the variance of factors that may influence the impact that these may have on the animal or human physiology. In this respect factors must be considered that would debilitate physiological conditions. Among the factors are the inorganic constituents in the water that may induce trace mineral imbalances, the differences in tolerance of animals due to the age (Young chickens are more susceptible than older chickens) and the interaction between WQC can have positive or negative effects. Cu can alleviate Mo but will in turn induce scouring disease in cattle (Dick & Bull, 1991). Hartmans and van Ryssen (1997) reported that addition of Se predisposes sheep to Cu poisoning, whereas increasing dietary Se reduced Cu induced chick mortality.

A number of elements can alleviate adverse effects of excessive Se intake, namely As, Hg, Au, Cu and Cd (Underwood & Suttle, 1999). The minerals in the water must be taken into consideration when formulating the diets for animal in order to avoid over supplementation. Cu poisoning can be reduced by the antagonistic effect with Mo and Zn. The relative concentration of various WQCs must therefore be seen in relation to the type of effect, deficiency or toxicity most likely to occur. In human sensitive use group such as pregnant woman and young children are more susceptible to WQCs than other user groups.

The shared utilization of water sources by animals and humans prohibits the application of certain alleviator treatments, on account of their potentially adverse effects on humans (Casey & Meyer, 2001). Concerns about the use of poor water quality in the poultry sector were presented by Casey *et al.*, (1998), based on the results of water quality investigation across the whole of South Africa. Poultry production has narrow profit margins, which mean that any decline in the health and efficiency of feed conversion will reduce profit margins drastically (Casey & Meyer, 2001).

As containing compounds are sometimes included in chicken feeds to act as growth promoters (van Ryssen, 2000). Wallinga (2006) surveyed the use of As in America and found that roxarsone, which contains As, is intentionally fed to chickens to control coccidostat. Tissues obtained from retail stores showed As levels ranging from non-detectable to 21.2 parts per billion (ppb), with a limit of detection of 2 ppb. Lasky *et al.* (2004) determined concentrations of total arsenic in poultry samples and found that individuals who consume 60 g/day of poultry could ingest 1.38-5.24 g/day of inorganic arsenic from the ingestion of poultry alone (tolerance level is 2 g/kg/day).

The 0.1 mg/L of As concentration administered in the treatments is a safe exposure in the context of intensive broiler production, because it retained values below the maximum acceptable concentration (MAC) of 1 mg/kg DM. In this context, the experiment indicated the threshold range for tissue accumulation of As to range from 0.1 mg/kg to 0.6 mg/kg in all the treatments.

Birds generally have a higher Pb tolerance than mammals (Humphreys, 1991), owing to their red blood cells (RBCs) having nuclei that store and retain Pb (Anders *et al.*, 1992). Blood concentrations of Pb associated with clinical signs of Pb toxicosis in birds (10.0-32.9 ppm wet weight) are about 10-100 times higher than those found in mammals (0.20-0.63 ppm) (Humphreys, 1991).

In this trial, the results for Pb in tissues are significantly higher in the elevated elements treatments and lower in the alleviator treatments ($P<0.05$). The accumulation of Pb in different tissues yielded different results. The livers had a higher accumulation of Pb than that of soft tissues, kidneys and hearts. Amongst the soft tissues, the thighs retained higher Pb results than the breasts. The kidney and heart tissues yielded approximately the same Pb results. Administration of Pb at 0.1mg/L level in Pretoria municipal water for broilers reared under the intensive production system for seven weeks does not retain Pb higher than the MAC levels in tissues. However, the interaction of Pb with other elements occurring in localised regions should be taken into consideration when making recommendations.

The most significant observation for this trial was not that the broilers receiving elevated selected elements in the drinking water accumulated these elements at a significantly higher concentration than did their counterparts, but that the concentration attained within a short production period did not exceed the MAC for these elements in poultry production destined for human consumption. Tissue comparisons yielded more sensitive results for thigh samples than for breast samples. The trends in both tissue types were the same, in that they indicated a positive accumulation of the treatment for As, Pb and Br, and also for Se and Cr. The alleviator treatment returned promising results for both tissue types, with significantly lower tissue concentrations in almost all the treatments.

The significantly lower values observed for T3 (TDS treatment) as compared to T1 (elevated elements) suggest that the alleviator potential of a volume-loaded hypertensive chemical may be significant. But alleviator treatment must not be applied in the absence of key PHCC, as the dynamics of the alleviator treatment may precipitate a marginal deficiency of those elements with a narrow range between essentiality and toxicity, as evidenced by the Se results. An additional perspective, by implication, is that TDS must be considered in the site-specific formulation of water quality guidelines for poultry when present at concentrations exceeding 100 mg/L in terms of potential increases in tolerable ranges of other trace elements.

All classes of livestock can use saline water with <300mg/L NaCl. A further increase in salinity requires the guideline to be used in conjunction with observation of the reactions of livestock to a saline water source (Ahmed, 1989). Elsenbroek *et al.* (2003) improved the health and reproductive status of Bonsmara cows exposed to potentially hazardous water by giving them a TDS alleviator treatment at 3000mg TDS/L in drinking water. Holele (2006) found that TDS may reduce negative effects attributed to chronic selenosis by alleviating whole blood concentration increases.

The results of the elements in the liver indicate that element treatments elevated the concentrations of most of the elements analyzed; the TDS alleviator treatments reduced the concentrations of these elements in the presence of Br, which arguably plays a part in alleviation. Further study is needed to assess the role of Br in alleviating elements in broilers. The effect of cultural cooking within exposure groups of slaughter-based production needs further investigation, as this may show more adverse effects of Br, which may outweigh any of its potential benefit.

It is thus relevant to assess not only more than single WQC ranges with regard to water quality norms, but also production systems. Based on the results obtained, it would appear that the treatment levels applied for this trial would be acceptable if observed in the rural communal environment in terms of health, production and product quality norms, the important prerequisite being similar environmentally-controlled housing facilities and quality rations. These findings support the need for rural communities to be provided with adequate facilities with which to implement poultry production, specifically under circumstances that see them reliant on groundwater to provide water for livestock watering. Rural production differs from intensive production systems in that, in intensive systems, the accumulation of constituents like Pb in renal cortex tissue does not present a significant consumer hazard, seeing that concentration and dilution within the urban diet effectively provide for sufficient safety. However, this argument does not hold good for rural production because of the small population group (Casey & Meyer, 2001).

Palatability factors play a significant role in satiety for salt appetite (Jalowiec *et al.*, 1966; Schulkin, 1982), as animals select the more salty-tasting solutions. TDS treatments had significantly higher water intake ($P<0.05$) than the non-TDS treatment. The results support the idea that salt induces thirst, leading to increased water intake (Swenson & Reece, 1993). The fact that the water intake was higher in TDS treatments and their tissue elements levels accumulation was lower suggests that higher water intake in the presence of As-Pb and Br results in a lower accumulation of elements in chickens.

A similar live weights and feed intake was observed for comparable groups (non-TDS treatments as opposed to TDS treatments); health effects linked to induced trace element toxicities and imbalances appear to be acceptable within the short exposure period in broiler production. The results suggest that inclusion of As, Pb and Br over a short period in broilers does not affect growth and feed intake significantly.

When drinking water containing the PHCC is also used in cooking practice, the risk is increased further. This applies more to some elements (such as Cd), which are sufficiently heat-stable to survive the cooking process, and present with nephrotoxicity in the human (Underwood & Suttle, 1999). The cultural-cooking practice may increase the probability of ingestion of an unacceptable concentration of PHCC, as the same water may be used for cooking and food preparation. Soft tissues known to have a high concentration of heavy metals are routinely consumed.

Although no obvious problems were observed regarding meat production, the tissue results do suggest that the chemical treatments interfered with other elements that were not administered. Effects of this nature will be of concern primarily when there is a long-exposure period and in production systems where affected trace elements are marginal in supply, a situation that may arise within rural communal production systems.

CHAPTER 5
CONCLUSION

This experiment demonstrated that an alleviator treatment that had been successful in ruminant species also significantly reduced the concentrations of some PHC in broiler tissues. This experiment also indicated that an arsenic exposure concentration reduced from 0.9 to 0.1 mg/L may result in acceptable product quality in the presence of high bromide, a frequently observed geochemical anomaly in the rural subterranean groundwater investigated. The experiment demonstrated that, prior to commencing with agricultural initiatives that improve food security within rural communities, due consideration must be given to the potential accumulation of PHCC in animal tissues. Furthermore, safe dosages, limiting essential trace elements and safe alleviator treatments, can be formulated on a site-specific basis. Applications to the household level and to potential export markets also require a site-specific approach.

The central point of departure remains a comprehensive water quality investigation in which the relevant norms for the proposed use of the water are investigated, followed by the site-specific risk factors that should be assessed in order to most appropriately utilize and manage the available water.

CHAPTER 6

6.1 CRITICAL EVALUATION

- The chickens were slaughtered only at the end of the trial. If representative samples of each treatment had been slaughtered weekly, valuable information could have been obtained on the rate of As and Pb deposition in the soft tissues.
- The exposure period was too short. An extra two or three weeks would probably have provided more information on the retention of elements directly related to rural communal farming.
- Drinking water samples should have been taken every week and analyzed so as to confirm treatment mixture.
- The stocking density in the pen was not equivalent to either rural or commercial production; 12 chickens were merely placed per pen.

6.2 FUTURE RESEARCH PROPOSAL

- The results of this experiment suggest that bromide, together with sodium chloride, alleviates the accumulation of elements in tissues. Bromide should be tested separately to clearly show its effect in alleviation, because these results may mean simply that sodium chloride alleviates the accumulation of bromide.
- The primary key risk events pertaining to cultural cooking practices using the same source of water (drinking and food preparation) and safe water should be identified.
- The interaction of heavy metals in drinking water and their forms of excretion.
- In-depth assessment of the physiological operation of alleviator treatment.
- The correlation of PHCC between feathers and soft tissues. This will help to estimate the accumulation of PHCC in soft tissues from the feathers.

CHAPTER 7

REFERENCES

AHMED, M.H., FARID, M.F.A., SAWKET, S.M. & HASSAN, N.I., 1989. Effects of water deprivation on feed utilization and mineral balances in sheep drinking natural saline well water. *J. Arid. Environ.* 26: 205-266.

ALLOWAY, B.J., 1995. Heavy metal in soils, 2nd ed. Blackie, Glasgow.

BARTHOLOMEW, G.A. & CADE, T.J., 1963. The water economy of land birds. *The Auk*, 80: 505-539.

ANDERS, E., DIETZ, D.D., BAGNELL, C.R., GAYNOR, J., KRIGMAN, M.R., ROSS, D.W., LEANDER, J.D. & MUSHAK, P., 1982. Morphological, pharmacokinetic, and hematological studies of lead-exposed pigeons. *Environ. Research*, 28: 344–363.

ARC ISCW., 1998. Institute for Soil Climate and Water. *Agricultural Research Council. Water Analysis Division*. Pretoria R.S.A

AVIAGEN., 2002. *Ross Broiler manual.* http://www.aviagen.com/broilermanual/broilermanual.htm

BLAKE, J.P. & HESS, J.B., 2001. Evaluating water quality for poultry. http://www.aces.edu/pubs/docs/A/ANR-1201/

BORGES, S.A., FISCHER DA SILVER, A.V., MAIORKA, A., HOOGE, D.M. & CUMMINGS, K.R., 2004. Physiological response of broiler chickens to heat stress and dietary electrolytes balance (Sodium plus Potassium minus Chloride, milliequivalents per kilogram. *Poult Sci.*, 83 (9): 1551-1558.

BORGES, S.A., FISCHER DA SILVER A.V., MAIORKA, A. & OSTRENSKY, A., 2004. Electrolyte balance in broiler growing diets. *Poult Sci* 83 (10): 623-628.

CARTER, T.A., 1985. Drinking water quality for poultry. *Poult Dig.*, 2: 50.

CASEY, N.H. & MEYER, J.A., 2001. Rural communal livestock production system and wildlife production system. *WRC report*, 857/1/01: 11.

CASEY, N.H., MEYER, J.A. & COETZEE, C.B., 2001. Poultry production system and water quality for ostrich production, *WRC report*, 857/2/01:2.

CENTERS FOR DISEASE CONTROL., 1991. Preventing lead poisoning in young children: a statement by the Centers for disease control. Atlanta GA: *US Dept of Health and Human Service*, Oct.

COETZEE, C., 2006. The development of water quality standards for poultry production in Southern Africa. Ph.D. Thesis Univ. of Pretoria. South Africa.

DEPARTMENT OF WATER AFFAIRS AND FORESTRY (DWAF), 1996. South African water guidelines. Agricultural water use: livestock watering. 2nd edition.

DONALDSON, W.E., 1986. Interaction of lead toxicity and riboflavin status in chickens (Gallus domesticus), Comparative Biochemistry and Physiology Part C: *Comparative Pharmaco toxico*, 85.1: 1-3.

ELSENBROEK, J.H., MEYER, J. & MYBURG, J., 2003. Haemorrhagic diarrhea and reproductive failure in Bonsmara cattle resulting from anomalous heavy metal concentration in soils, forages and drinking water associated with geochemical anomalies of toxic elements on the farm Puntlyf, South Africa. *J. Phys., IV Franc*, 107.

ERNST, R., 1989. Water quality guidelines. *Poult Inter*., 6: 40-42.

ESTERHUIZEN, D., 2007. South African, Republic of poultry and products voluntary report. USDA foreign agricultural service gain report. Gain report nr. SF7042.

GLAZE, W.H. & WEINBERG, H.S., 1999. Identification and occurrence of ozonation by-products in drinking water. *AWWARF Denver, CO*.

GOOD, B., 1985. Water quality affects poultry production. *Poult Dig*., 3: 108-109.

GORBY, M.S., 1988. Arsenic poisoning. *West J Med.*, 149: 308-315.

GRILL, H.J. & MISELIS, R.R., 1986. Lack of ingestive compensation to osmotic stimuli in chronic decerebrate rats. *AM J Physiol.*, 240: 81-86

HARTMANS, F. & VAN RYSSEN, J.B.J., 1997. Metabolism of selenium and copper in sheep with and without sodium bicarbonate supplementation. *J. Agric Science*, 128: 351-384.

HUMPHREYS, D.J., 1991. Effects of exposure to excessive quantities of lead on animals. *Brit Vet J.*, 147: 18–30.

HOLELE, K.K.O., 2006. Effect of TDS content on the Selenium status and certain growth parameters of mutton sheep. *M Inst agrar.* Thesis, University of Pretoria, South Africa.

JALOWIEC, J.E., CRAPANZANO, J.E. & STRICKER, E.M., 1966. Specificity of salt appetite by hypovelemia. *Psychonom. Sci.*, 31: 351-365.

KEMPSTER, P.L., HATTINGH, W.H.J. & VAN VLIET, H.R., 1981. Summarised water quality criteria. Department of Agriculture and Water affairs, Hydrological Research Institute. Technical Report TR 108.

KESHAVARZ, K., 1987. Proper water management for poultry. *Poult Dig.*, 1: 12-22.

LASKY, T., SUN, W., KARDY, A. & HOFFMAN, M.K., 2004. Mean total arsenic concentration in chicken 1989-2000 and estimated exposure for consumers of chicken. *Environ Health Perspect.*, 112: 18-21.

LEESON, S. & SUMMER, J., 2005, Commercial Poultry Nutrition, 3rd ed. *University books Ontario,* Canada.

LIDE, D.R., 1994. Lead and Mercury. In: *The CRC handbook of chemistry and physics, 75th ed.* Boca Raton, FL: CRC Press, 416-418.

MAIORKA, A., MAGRO, N., BARTELS, H.A.S., KESSLER, A.M. & PENZ, J.R., 2004. Different sodium levels and electrolyte balance in pre-starter diets for broilers. *Bra. Jr of Poult Sci.*, 6: 143-146.

MALACHOWSKI, M.E., 1990. An update on arsenic. *Clin Lab Med.*, 10: 459-471.

MANCL, M., SAUIS, M. & WAGERT, L., 1991. Private drinking water supplies. *NRAES-47*, Cooperative Extension. Ithaca. NY 14853.

MCGOWAN, C. & DONALDSON, W.E., 1987. Effect of lead toxicity on the organ concentration of glutathione and glutathione-related free amino acids in chickens, *Toxico let,* 38: 265-270.

MEYER, J.A., 1998. Modelling for the prediction of water quality guidelines for livestock in South Africa. Ph.D. Thesis, University of Pretoria, South Africa.

NATIONAL ACADEMY OF SCIENCE., 1980. Mineral tolerance of Domestic Animals. http://www.nap.edu/openboo/0309030226/html/88.html.

NATIONAL RESEARCH COUNCIL (NRC), 1974. Nutrients and toxic substances in water for livestock and poultry. National Academy of Science, Washington, D.C. USA.

NDA. Meat inspection Manual Poultry part 3. http://www.nda.agric.za/vetweb/Food%20Safety/FS_PO_Manual/1.2%20PART%20III%20PMI%20%20MEAT%20INSPECTION%20MOD.pdf

NICHOLSON, F., CHAMBERS, B., ALLOWAY, B., HIRD, A., SMITH, S. & CARLTON-SMITH, C., 1998. An inventory of heavy metal inputs to agricultural soils in England and Wales. In: Proceedings of the 16th world congress of soil science, Montpellier, France.

NORTH, M.O. & BELL, D.P., 1990. Commercial chicken production manual, 4th ed, Van

Nostrand Reinhold, New York.

OLDFIELD, B.J., BADOER, E., HARDS, D.K. & MCKINLEY, M.J., 1994. Fos production in retrogradely labelled neurons of the lamina terminalis following intravenous infusion of either hypertonic saline of angiotention II. *Neuroscience*, 60: 255-62.

PALOUCEK, F.P., 1993. Lead poisoning: Pharmacists can play a role in preventing and treating lead toxicity. *Am Pharm.*, NS33:81-90.

SCHULKIN, J., 1982. Behaviour of sodium deficient rats: The search for salty taste. *J. Comp. Physiol.*, 96: 628-634

SCHWARTZ, D.L ., 1994. Water quality. VSE 81c. Penn. State U. In Proceedings AVMA Annual Conference. 7.

SKADHAUGE, E., 1976. Water conservation in xerophilic birds. *Israel Journal of Med Sci.*, 12: 732-739,

SMITH, M.O. & TEETER, R.G., 1987. Potassium balance of the 5 to 8-week-old broiler exposed to constant heat or cycling high temperature stress and the effect of supplementing potassium chloride on body weight gain and feed efficiency. *Poult Sci.*, 66: 487-492.

STUMM, W. & MROGAN, J.J., 1981. *Aquatic Chemistry.* An introduction emphasising chemical equilibria in natural waters. John Wiley & Sons, New York.

STURKIE, P.D., 1986. *Avian physiology.* 4th ed.

SWENSON, M.J. & REECE, W.O., 1993. Duke's physiology of domestic animals, 11th ed. Comstock publishing association, Cornell University press

SYMONS, J.M., 1999. The drinking water dictionary. *American Water Works Association.* Denver, CO:N 47.

UNDERWOOD, E.J. & SUTTLE, N.F., 1999. The mineral nutrition of livestock. 3rd

edition, CAB international. ISBN 0 85199 128 9.17-66

VAN RYSSEN, J.B.J., 2000. Poultry litter as a feed ingredient for ruminant: the South African situation. *S A J anim sci.*, 2.1: 2.

VODELA, J.K., LENZ, S.D., RENDEN, J.A., MCELHENNEY, W.H. & KEMPPAINEN, B.W., 1997. Drinking water contamination (Arsenic, Cadmium, Lead, Benzen, and Trichloroethylene) 2. Effects on reproduction performance, egg quality, and embryo toxicity in broiler breeders. *Poult Sci.*, 76: 1493-1500.

WADE, M.J., DAVIS, B.K., CARLISLE, J.S., KLEIN, A.K. & VALOPPI, L.M ., 1993. Environmental transformation of toxic metals. *Occup. Med.*, 8: 575-601.

WAGGONER, R.E., 1986. Tyson Foods. Inc. as quoted by Keshavarz, 1987.

WALLINGA, M.D., 2006. Playing chicken avoiding arsenic in your meat. http://www.healthobservatory.org/library.cfm?refid=80529

WHO., 2001. Arsenic compounds, environmental health criteria 224, 2nd ed. Geneva: *World Health Organisation.*

WINSHIP, K.A., 1984. Toxicity of inorganic arsenic salts. *Adv Drug react Ac Pois Rev.*, 3: 129-160.

YANG, R.S.H. & RAUCKMAN, E.R., 1987. Toxicological studies of chemical mixture of environmental concern at the national toxicology program: health effect of ground water contamination. *Toxico,* 47: 15-34.

ZIMMERMAN, N.G., 1995. Relationship of drinking water quality and broiler performance. Proc. Delmarva Broiler Housing & Flock supervisors' conf. University of DE & MD Cooperative Extension and DPI, Inc.