



Food Safety in Horticultural Markets:

Report No. 3

Assessment of microbial, pesticide residue and heavy metal contamination of horticultural produce marketed in Harare, Zimbabwe

E. Mangwayana and F. Mapanda

Department of Soil Science and Agricultural Engineering

University of Zimbabwe

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Imperial College
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Overview

Horticultural produce in Harare obtained mainly from peri-urban farmers and the surrounding farmers is thought to be contaminating the food chain and deteriorating the production resource base. This poses severe health risks to consumers of contaminated produce. The sources and levels of heavy metal, pesticide residues and pathogenic contamination were determined in a three year DFID CPHP funded project.

Leafy vegetables and tomatoes which are consumed in large quantities by many families in Harare were sampled from retail outlets in high density areas and from smallholder farms were found to be contaminated by faecal coliforms (*E.coli*) and a few samples contaminated with organ chlorines and organo phosphates though the levels were below maximum permissible limits. However, soil concentrations of heavy metals in smallholdings at sewage farms were low and within EU limits though showed potential to increase beyond acceptable levels in 5 to 60 years time.

Microbial Contamination

Vegetables are consumed daily by an average household in Zimbabwe, this is mainly so in poor families were mainly leafy vegetables and tomatoes for the main part of the daily relish. These vegetables, which are normally sourced mainly from the street vendors and at times from the backyard garden, are at times contaminated with faecal matter. Faecal contamination remains a serious issue for safety assurance of horticultural produce.

The faecal oral route offers opportunities for food borne disease causing micro-organisms such as *Salmonella spp.*, *Shigella spp.*, *Yersinia enterocolitica* and pathogenic strains of *Escherai coli* such as *E.coli* O157:H7. These pathogens are often present in low levels, and require specialist techniques for detection; it is therefore normal to use a more easily detected indicator organism. This indicator organism must be always present in faeces, but not form part of the natural micro- flora of other components of the environment such as water, soil or food commodities. *E.coli* poses all the characteristics hence it was used in this study to act as an indicator of faecal contamination of horticultural produce within main marketing areas of horticultural produce in Harare.

Sample collection

Field surveys were carried out in the dry and wet season in four marketing area in Harare namely Chikwanha, Epworth, Mufakose Glenview. However for Glenview area was dropped in the wet season and replaced by Tafara - Mabvuku after results of the dry season indicated that samples from Glenview were maybe contaminated by produce from sewage farms, which are close to Glenview area. These markets are located in the high-density areas where the majority of Harare urban dwellers stay.

The sites were visited once a week and samples were randomly collected from vegetable markets and street vendors in marketing area. The samples were randomly collected each marketing site through acting like potential buyers, buying two bundles of leafy vegetables and a kilogram of tomatoes to represent a single sample to reduce bias. The

main leafy vegetables marketed were rape, covo and tsunga. From the sampling survey it was noted that most of the vegetables were coming the main Mbare Wholesale market with a few of the vendors mainly in Epworth marketing home-grown vegetables. Per season we collected a total of 320 samples (160 leafy vegetables and 160 tomatoes).

The samples were collected early in the morning and were transported in cooler boxes to the laboratory and analyzed the same day for microbial contamination and part of the sample was retained in the freezer for pesticide analysis.

Microbial analysis of the vegetables

The samples were analyzed for *E.coli* using the British Standard method for microbiological examination of food and animal feeding stuffs (BS 7563: Part O, 1996). *E.coli* is not part of the normal micro flora of fresh produce including vegetables. However, the normal microflora of plant surfaces (stems and leaves) are dominated by other members of the Enterobacteriaceae that are not pathogenic for man. When looking for faecal contamination in fresh produce, a technique is required that is specific for *E.coli* and not susceptible to interference from the normal microflora of plant surfaces. Several techniques have been developed that make use of the fact that *E.coli* is one of the few members of the Enterobacteriaceae that produce the enzyme B-D-glucuronidase to degrade B-D-glucuronic acid. The improved technique where a chromogenic substrate is used was used for this study. The procedure has been accepted by the United Kingdom Public Health Laboratory Service (PHLS, United States Food & Drug Administration (FDA) and major UK supermarkets as a reliable procedure for *E.coli* detection in fresh produce.

Results

Guidelines set by the Public Health Laboratory Service (PHLS) of United Kingdom were used as there are no set guidelines for Zimbabwe. The guidelines are as indicated below.

- 0-20cfu/g = Acceptable
- 21-100cfu/g = Borderline
- 101-10⁴cfu/g = Unacceptable
- 10⁴ = Potential Hazard

Our results (Table 1, 2 , 3 & 4) indicate a potential problem of faecal contamination of horticultural produce at the markets, particularly at Epworth as evidenced by high levels of E.coli which was used as an indicator species.

Table 1: *E.coli* in tomatoes and leafy vegetables obtained from Chikwanha Market.

PHLS Criteria E.Coli CFU/g	Dry Season		Wet Season	
	Leafy Veggies %	Tomatoes %	Leafy Veggies %	Tomatoes %
20 cfu/g satisfactory	7.5	80	2.5	70
21-100 cfu/g borderline	77.5	15	57.5	12.5
101-10 ⁴ cfu/g unacceptable	10	5	22.5	17.5
10 ⁴ cfu/g potential hazard	5	0	17.5	0
TOTAL	100	100	100	100

Chikwanha market is more of a wholesale market where large and small-scale producers market their produce. In the early hours of the morning it acts mainly as a wholesale market which closes at 10am, then thereafter it acts a retail market where produce obtained earlier on from wholesale market is then marketed. The main marketing area is sheltered with basic sanitation facilities therefore there is likelihood of microbial contamination. In terms of produce changing hands in this market, there is mainly the

producer, transporter from the farm and wholesaler hence a small percentage of about 15 to 40% can be classified as unacceptable for human consumption. The marketing structure of this market can be compared to that of Mbare Wholesale vegetable market in Harare.

Table 2: *E.coli* in tomatoes and leafy vegetables obtained from Epworth Market.

PHLS Criteria <i>E.Coli</i> CFU/g	Dry Season			Wet Season		
	Leafy %	Vegs %	Tomatoes %	Leafy %	Vegs %	Tomatoes %
20 cfu/g satisfactory	0		15	0		15
21-100 cfu/g borderline	0		0	0		0
101-10 ⁴ cfu/g unacceptable	5		7.5	5		7.5
10 ⁴ cfu/g potential hazard	95		77.5	95		77.5
TOTAL	100		100	100		100

Epworth marketing area presented the worst-case scenario where the 85 to 100% of produce both leafy vegetable and tomatoes can be classified as unacceptable for human consumption. This can be explained by the haphazard settlement in the area without safe tapped water and water borne sewerage system. The area consisted of mainly street vendors who use well water or stream water to wash and store their vegetables. Street vendors tend to dip their vegetables in containers full of water as protection of their produce from the harsh high daytime temperatures. This contaminated water from stagnant streams and unprotected wells contaminate the vegetables. The other possible source of contamination is during transportation from the main markets where the vegetables are carried in unclean sacks in open trucks and stored overnight at home if they are any leftovers from the previous day.

Efforts are underway by the Epworth Local Board to introduce electricity, safe tapped water and water borne sewage system into the area. There is also need to construct vegetable stalls in the area with basic amenities as a way to reduce cross contamination of produce. The entire Epworth area currently has one area with vegetable market stalls close to Epworth mission area though the vendors are shunning it at present stating that its located far away from their customers. There is therefore need to construct these stalls in consultation with street vendors and at the same time highlight benefits of marketing vegetables under these protective structures.

Table 3: *Ecoli present in tomatoes and leafy vegetables obtained from Mufakose Market.*

PHLS Criteria E.Coli CFU/g	Dry Season			Wet Season		
	Leafy %	Vegs	Tomatoes %	Leafy %	Vegs	Tomatoes %
20 cfu/g satisfactory	5		60	0		57.5
21-100 cfu/g borderline	0		25	2.5		22.5
101-10 ⁴ cfu/g unacceptable	70		15	60		20
10 ⁴ cfu/g potential hazard	25		0	37.5		0
TOTAL	100		100	100		100

In the Mufakose market it's mainly the leafy vegetable that poses a threat to human consumption. However in this market place which is similar to Epworth in the sense that most of the vegetables are marketed by street vendors a large proportion of the tomatoes 78 to 80% of the vegetables fall within acceptable limits as opposed to Epworth area. This difference could be due to the fact there is a clean supply of tap water, which is used to the produce.

Table 4: *E.coli* in tomatoes and leafy vegetables obtained from Glenview & Mabvuku-Tafara Market.

PHLS Criteria	Dry Season			Wet Season		
	Leafy %	Vegs %	Tomatoes %	Leafy %	Vegs %	Tomatoes %
20 cfu/g satisfactory	10		27.5	0		60
21-100 cfu/g borderline	0		0	2.5		12.5
101-10 ⁴ cfu/g unacceptable	5		12.5	57.4		25
10 ⁴ cfu/g potential hazard	85		60	40		2.5
TOTAL	100		100	100		100

The Glenview and Tafara-Mabvuku market area has same characteristics as the Mufakose market area.

In summary in terms of microbial contamination the vegetables at the main market wholesale market is less contaminated than produce in the retail outlets located in the main marketing areas. There is contamination as produce moves down the marketing chain from the wholesaler all the way down to the consumer. There is need to follow produce through the marketing chain to establish stage or stages where the produce is contaminated as a way to reduce microbial contamination.

Heavy metal contamination

Consumers' demand for good quality vegetables is increasing, but the perceptions of 'good quality' are subjective among consumers. To some, undamaged, dark green and big leaves could be considered as 'good quality' vegetables. But, is quality related to safety? This study determined the quality status of leafy vegetables grown on heavy metal-contaminated gardens in terms of their heavy metal uptake and potential human exposure at Mukuvisi, Pension and Crowborough production sites.

Land disposal of sewage and industrial effluent in Zimbabwe enriches soils with heavy metals (Mangwayana, 1995; Oloya and Tagwira, 1996a; Steneva, 1996; Nyamangara and Mzezewa, 1999). But, production of leafy vegetables on such land has proved to be successful in urban Harare with respect to the visual qualities of vegetables. The vegetables appear dark green, 'healthy' and 'attractive' to consumers. However, potential health risks from heavy metal contamination of vegetables could be high since washing or boiling cannot reverse contamination that occurs through root uptake.

Wastewater is one of the major inputs in urban and peri-urban horticulture in Zimbabwe, amid local concerns that it may contain substantial amounts of heavy metals among other pollutants. The quality of soil with respect to contamination by Cu, Zn, Cd, Ni, Cr and Pb was determined at Mukuvisi site, irrigated using river water containing industrial and domestic effluent, and at Pension and Crowborough sites, irrigated using a mixture of treated sewage effluent and digested sludge. Soil samples were collected from selected gardens at each site, digested in aqua regia (concentrated HCl and concentrated HNO₃ in ratio of 3:1, respectively) and analysed for total heavy metal concentrations using the atomic absorption spectrophotometry method.

Soil sampling showed low concentrations of heavy metals in soils at production sites (Figure 1, 2, 3, 4, 5 & 6) suggesting that any risk to consumers is very localized. Comparison of the garden topsoil, subsoil and the controls indicated that the continual

and uncontrolled input of wastewater in horticulture resulted in deterioration of the production sites with respect to heavy metal enrichment of the productive topsoil, thereby preventing long-term production and increasing environmental pollution. Heavy metals do not disintegrate or disappear in the environment and therefore they remain in the soil, and may be introduced into the human food chain through plant uptake among other pathways. The study indicated that some heavy metals, notably Cu, Zn and Cd, had already started exceeding their permissible concentrations in some gardens at Pension and Crowborough production sites.

The estimated annual heavy metal loading rates showed that within 5-60 years, all studied heavy metals would have exceeded their permitted limits in soils, depending on site. This means that the use of wastewater in urban horticulture may enrich soils with heavy metals to concentrations that may pose potential environmental and health risks in the long term.

High potential health risks, especially from Cd intake, exist for daily consumers of leafy vegetables at Mukuvisi and Pension sites in Harare, but the risk to consumers at market is likely to be low.

Table 5: Estimated annual heavy metal loading rates (kg ha⁻¹ year⁻¹) at Mukuvisi and Pension sites, in comparison with the UK annual heavy metal loading rate limits for soils.

Metal	Mean annual loading rates		Annual Loading rate limits
	Mukuvisi	Pension	
Cu	2.3	20	7.5
Zn	7.0	-	15
Cd	6.7	195	0.15
Ni	2.3	99	3
Cr	2.3	195	15
Pb	5.0	80	15

Heavy metal annual loading rates (Table 5), estimated from the volumes of wastewater used in a year and the mean concentrations of heavy metals in wastewater were above the UK annual loading rate limit for Cd at Mukuvisi, and for all studied metals at Pension site. The loading rates indicated that within an average period of less than 5-60 years from year 2000, all soils would have reached their maximum permitted limits (UK) for all studied heavy metals, with Cd being the earliest to exceed its limit of 3 mg kg^{-1} .

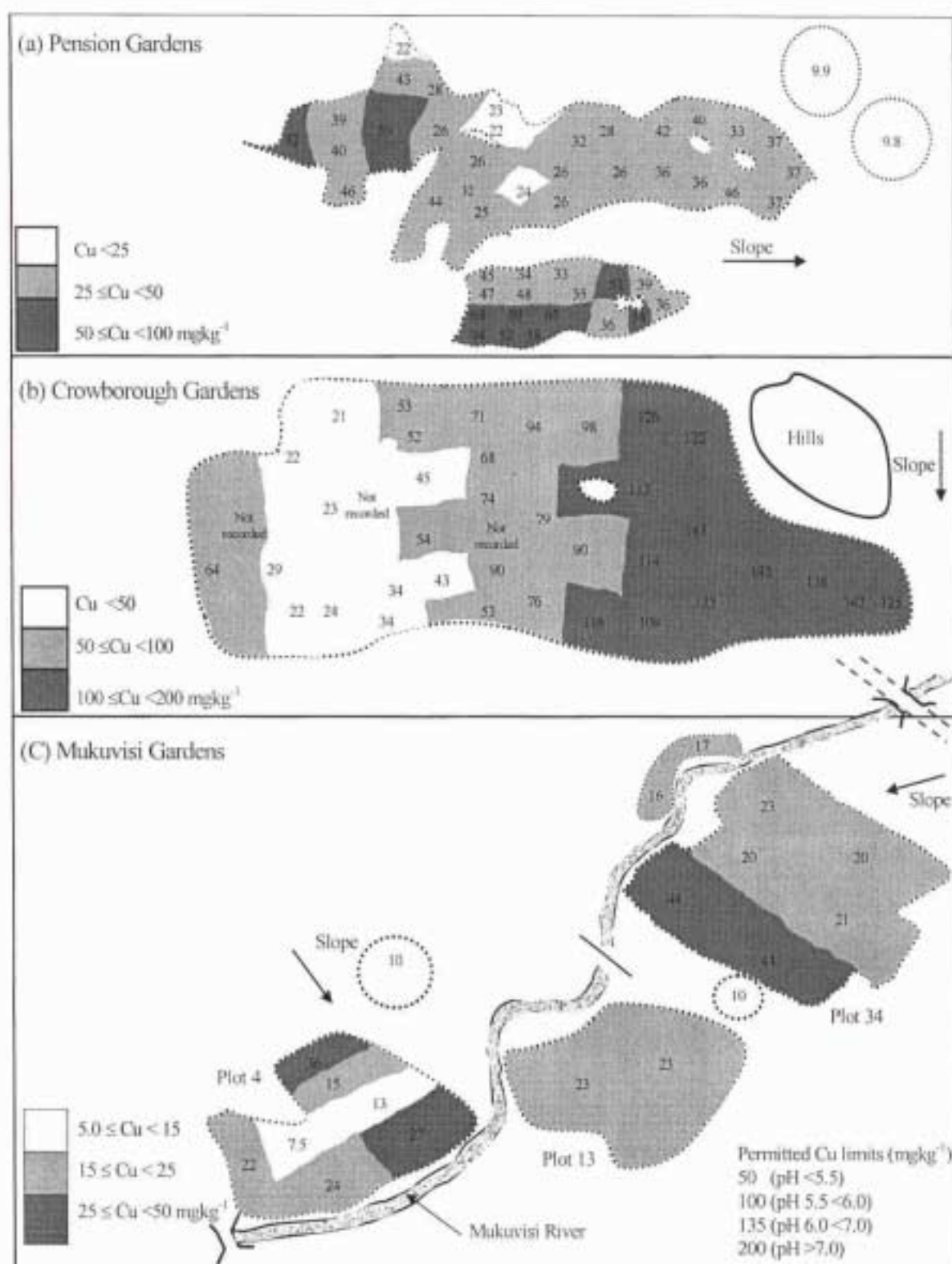


Figure 1: Soil Cu distribution in Pension (a), Crowborough (b) and Mukuvisi (c) gardens. Control soils Cu are shown (circled), but no suitable controls were found for Crowborough.

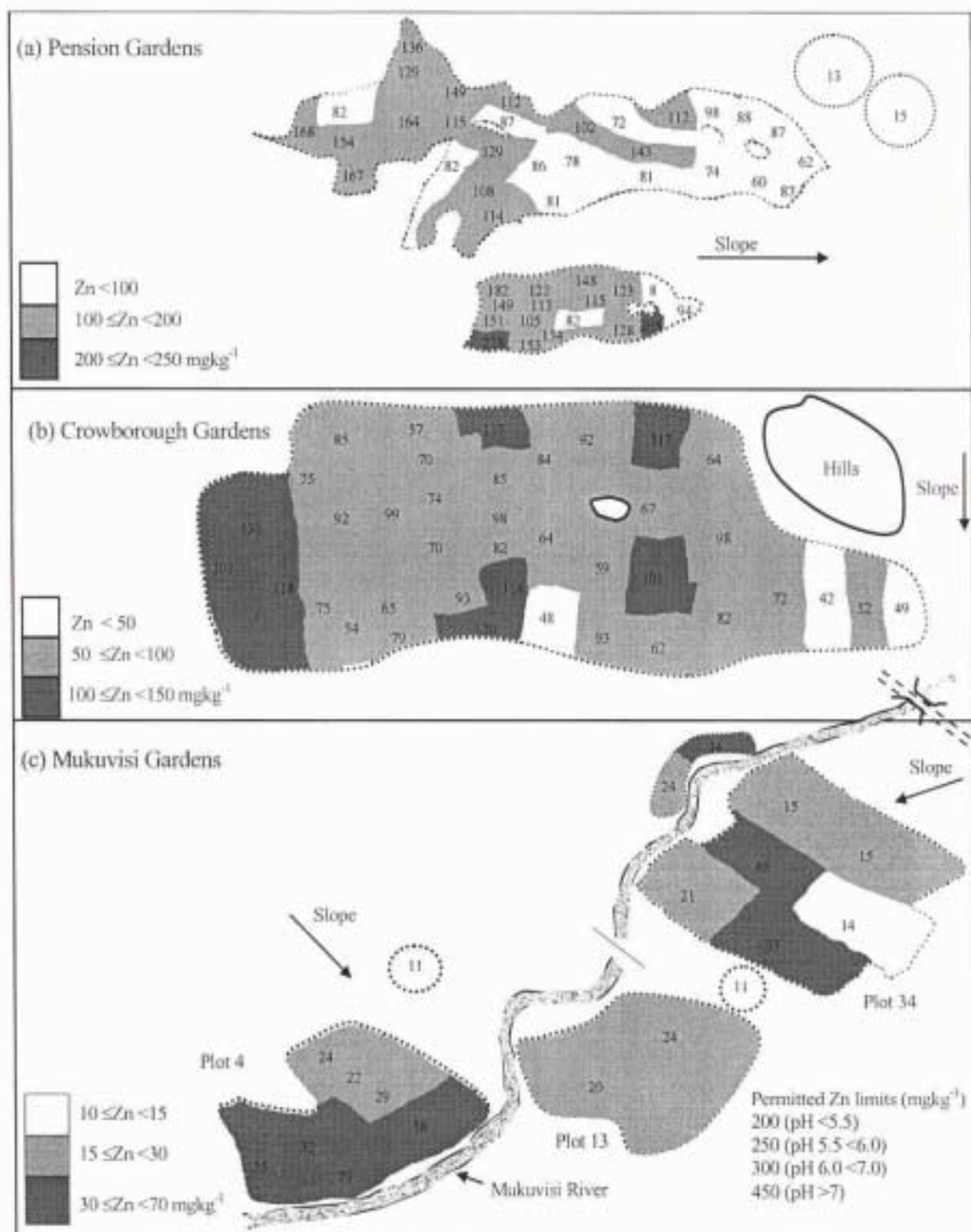


Figure 2: Soil Zn distribution in Pension (a), Crowborough (b) and Mukuvisi (c) gardens. Control soils Zn are shown (circled), but no suitable controls were found for Crowborough.

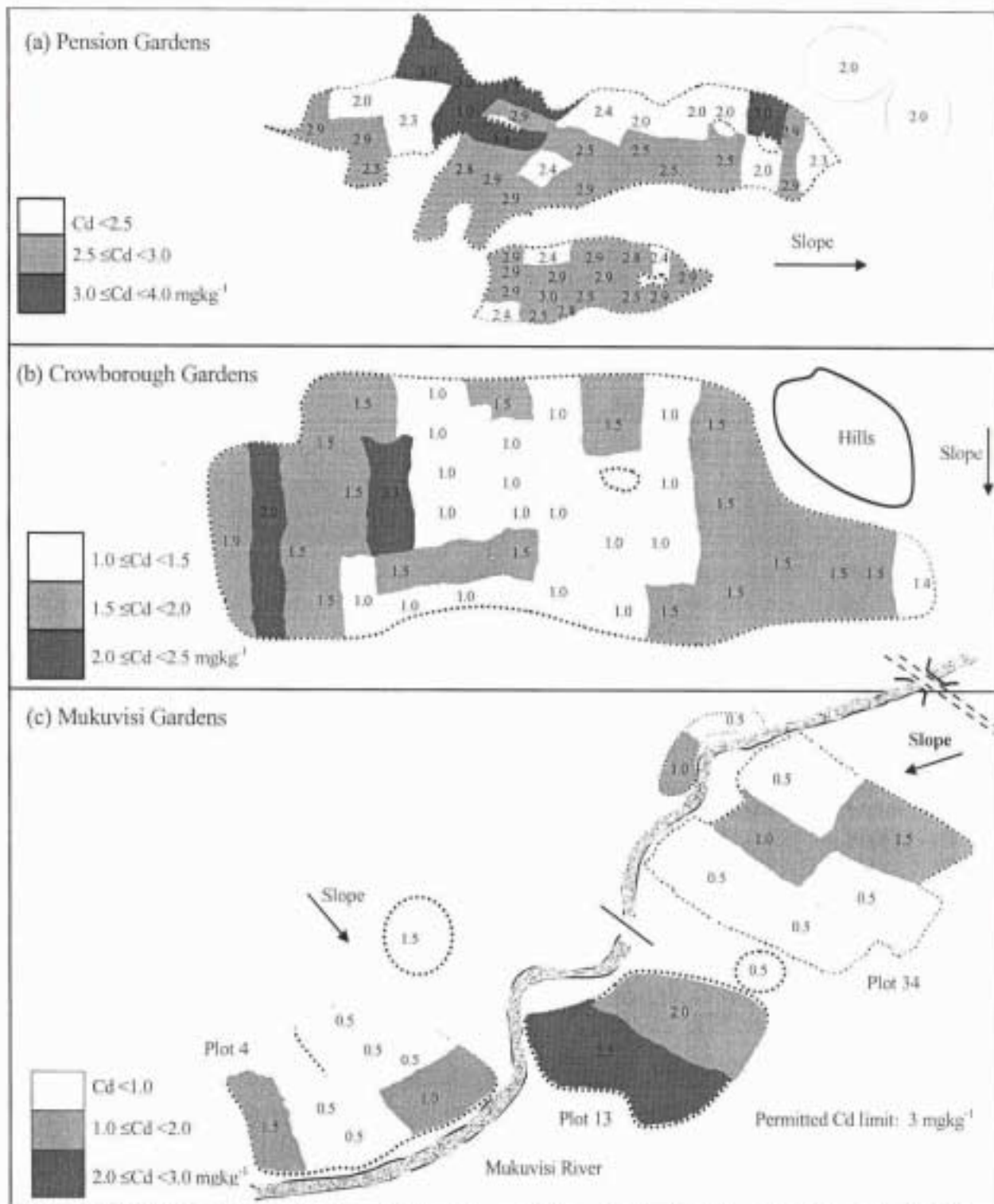


Figure 3: Soil Cd distribution in Pension (a), Crowborough (b) and Mukuvisi (c) gardens. Control soils Cd are shown (circled), but no suitable controls were found for Crowborough.

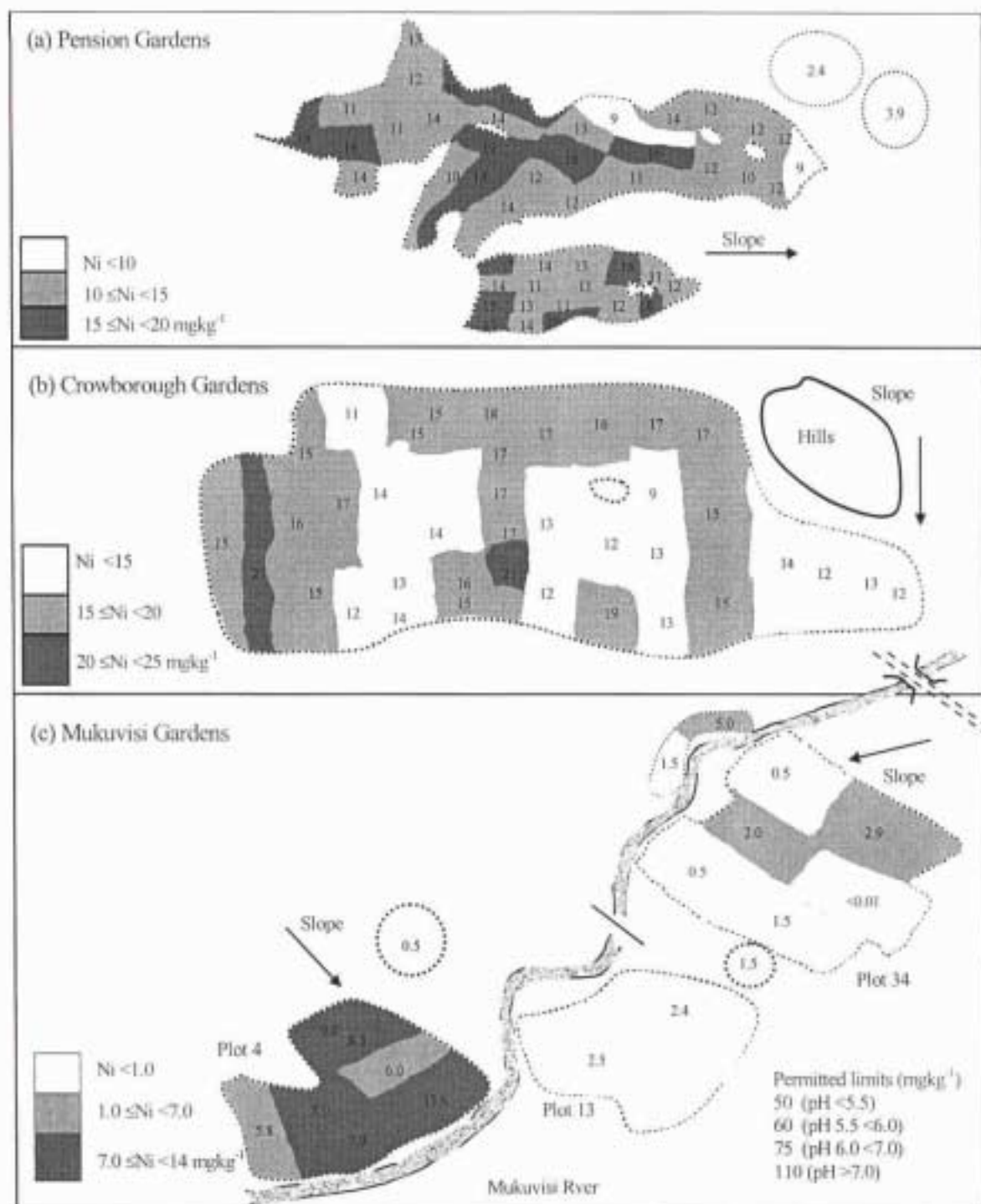


Figure 4: Soil Ni distribution in Pension (a), Crowborough (b) and Mukuvisi (c) gardens. Control soils Ni are shown (circled), but no suitable controls were found for Crowborough.

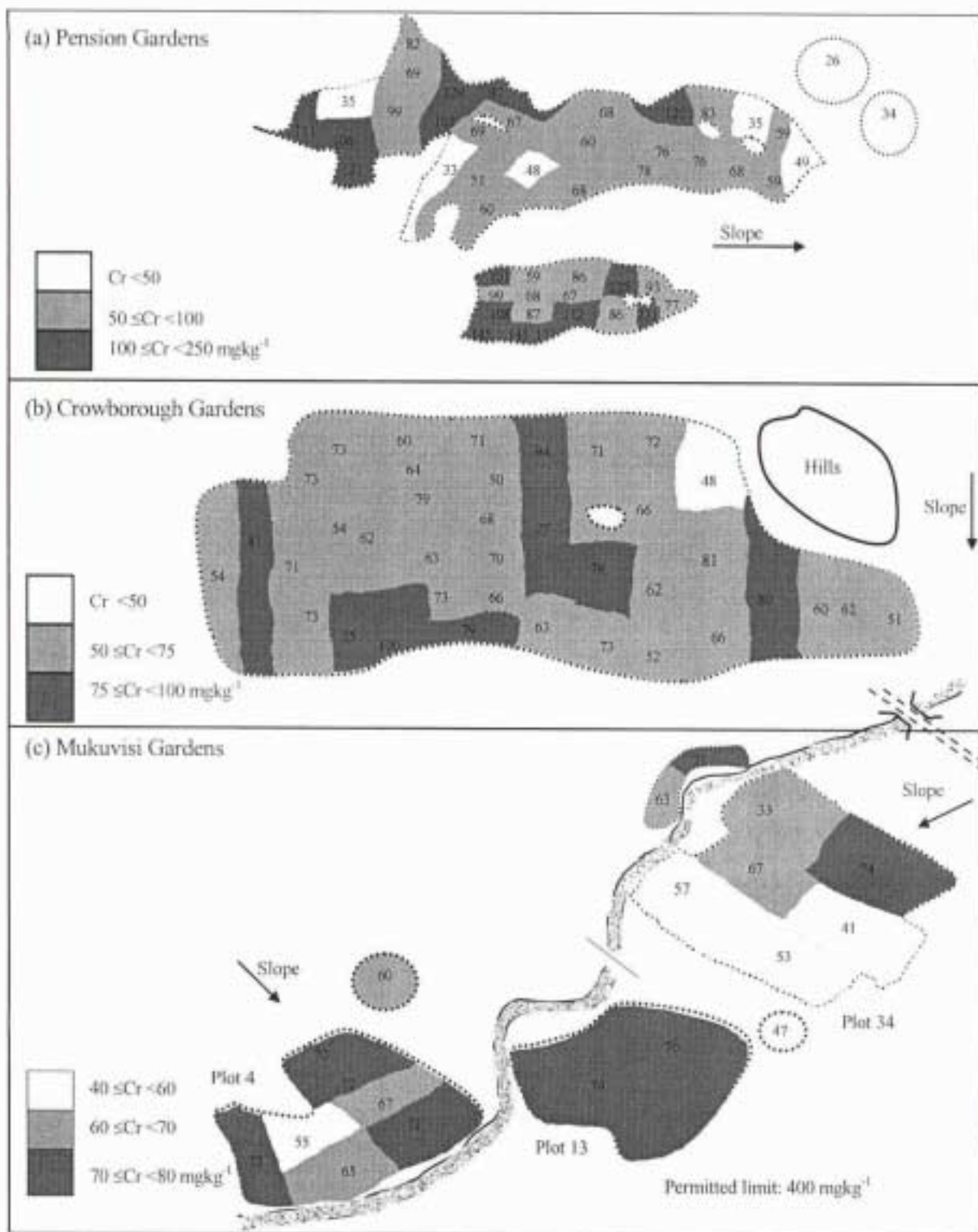


Figure 5: Soil Cr distribution in Pension (a), Crowborough (b) and Mukuvisi (c) gardens. Control soils Cr are shown (circled), but no suitable controls were found for Crowborough.

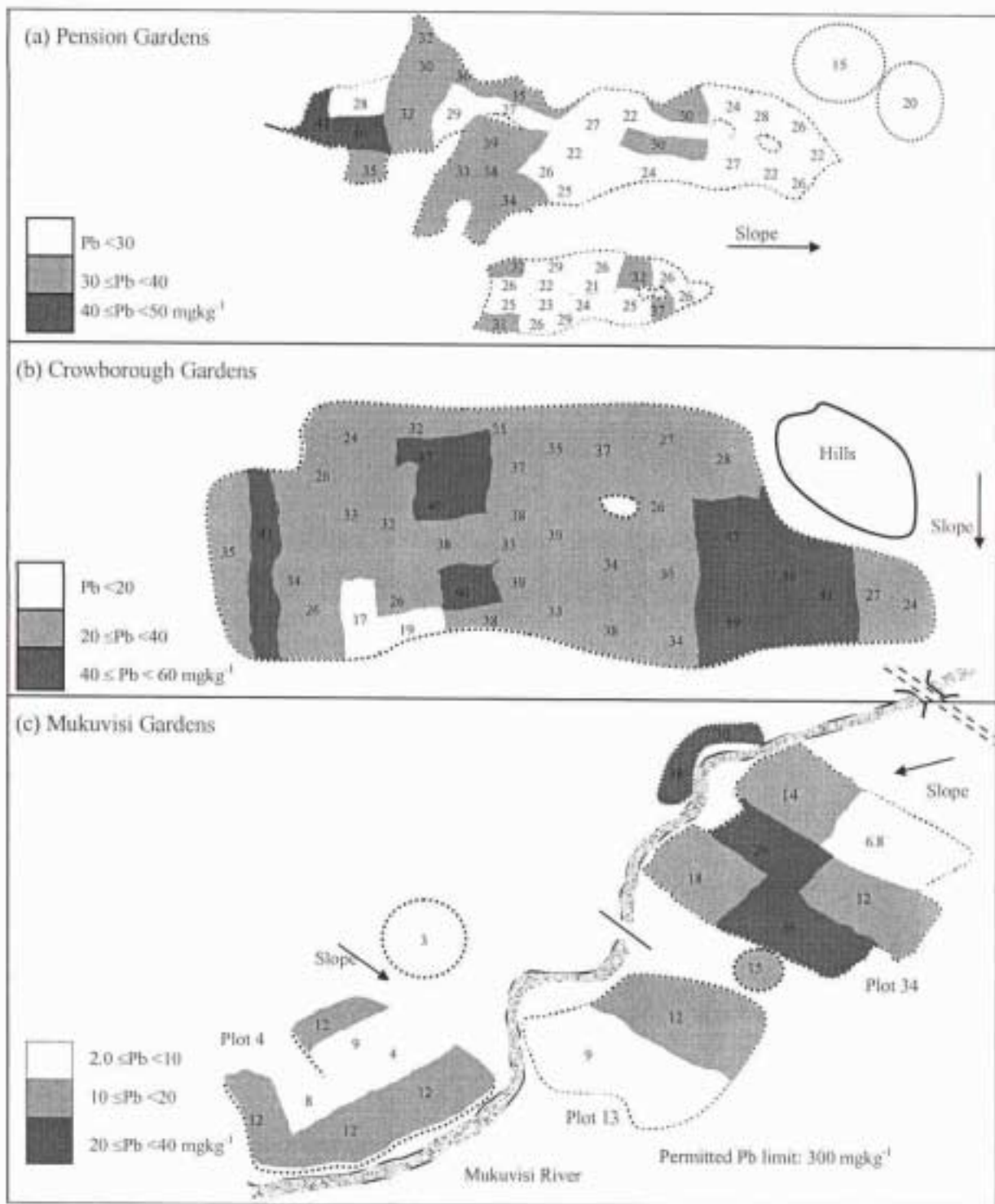


Figure 6: Soil Pb distribution in Pension (a), Crowborough (b) and Mukuvisi (c) gardens. Control soils Pb are shown (circled), but no suitable controls were found for Crowborough.

Assessment of Contamination of Leafy Vegetables

Leafy vegetables are essential to a well balanced diet for their Fe content, and in diets with no milk they play a significant role in supplying Ca (Bencini *et al.*, 1991). The quality of leafy vegetables (*Brassica juncea* and *B. napus*) grown on heavy metal contaminated soils was determined with respect to Cu, Zn, Cd, Ni, Pb and Cr at Mukuvisi, Pension and Crowborough sites. Vegetable leaf samples were collected from selected gardens at each site, as well as from a greenhouse study at three harvesting stages using *B. juncea* as a test crop. Dry combustion was used to destroy organic matter in the vegetables prior to acid-digestion in aqua regia, and analysis using the atomic absorption spectrophotometry method.

There was significant evidence of pre-harvest contamination of leafy vegetables by heavy metals, taken up from contaminated soils. Cadmium and Pb concentrations in leaves exceeded FAO/WHO permitted concentrations (Figure 7) (0.2 mg kg^{-1} and 0.3 mg kg^{-1} fresh weights, respectively) in some of the leafy vegetables at all studied sites. The potential risks of exposure to all studied heavy metals (except Cu and Zn) from consumption of contaminated vegetables, relative to the minimum risk levels, were high for a low income adult (60 kg body wt.) and very high for children. Children were especially vulnerable because their bodies cannot tolerate as much, and because of increased exposure when playing at contaminated sites (WHO, 1999).

Among the studied heavy metals, Cu and Zn are essential elements for both plants and animals, while Cd, Ni, Pb and Cr are non-essential elements. Although the total concentrations of Cu and Zn in soil and in vegetables were higher compared to the concentrations of other heavy metals, the daily intakes of Cu and Zn from studied vegetables alone were below the WHO minimal dietary requirement ($1.4 \text{ mg Cu day}^{-1}$ and $15\text{-}20 \text{ mg Zn day}^{-1}$, WHO (1993)) for an adult, and would be supplemented by other diets supplying Cu and Zn.

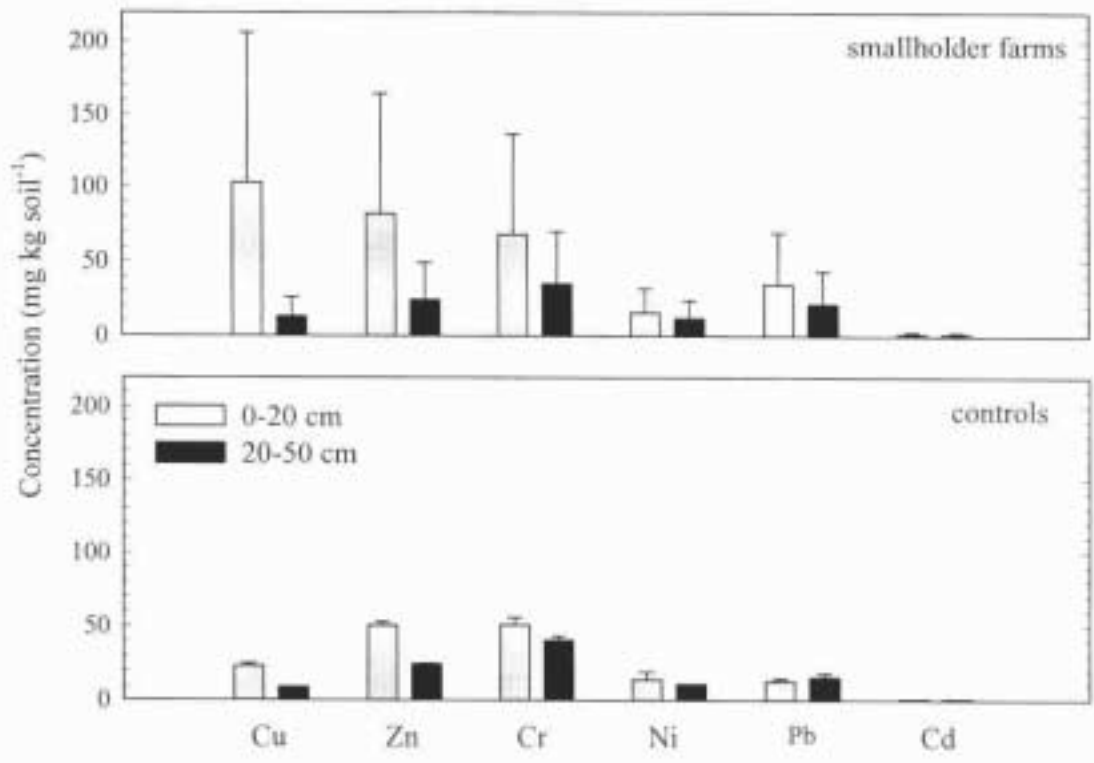
Table 6: The bio-concentration factors (CFs) for *B. juncea* and *B. napus* and the daily intake rates (mg day^{-1}) of studied heavy metals at Mukvisi and Pension sites in Harare.

Bio-Concentration Factors								
Site	Garden	Species	CF_{Cu}	CF_{Zn}	CF_{Cd}	CF_{Ni}	CF_{Pb}	CF_{Cr}
Mukvisi	4	<i>B. napus</i>	0.16	0.7	4.6	0.5	0.4	0.07
	4	<i>B. juncea</i>	0.18	1.2	2.7	0.4	0.4	0.10
	34	<i>B. napus</i>	0.06	0.7	1.7	-	0.03	0.07
	Mean CF value			0.13	0.9	3.0	0.5	0.3
Pension	3-4	<i>B. napus</i>	0.03	0.8	0.3	0.3	0.1	0.01
	1-2	<i>B. juncea</i>	0.03	0.8	0.5	0.2	0.1	0.02
	5-6	<i>B. juncea</i>	0.06	1.3	0.2	0.5	0.1	0.02
	8-11	<i>B. napus</i>	0.02	1.2	0.4	0.4	0.2	0.04
	25-26	<i>B. juncea</i>	0.08	2.3	0.6	0.4	0.2	0.04
	27-28	<i>B. juncea</i>	0.08	1.8	0.5	0.4	0.2	0.05
	20-22	<i>B. juncea</i>	0.08	2.6	0.6	0.5	0.2	0.09
Mean CF value			0.05	1.6	0.4	0.4	0.2	0.04
Daily Intake (DI) Rates								
Metal	Average contents in leaves (mg kg^{-1} fresh wt.)		Daily intakes (mg day^{-1})		MRL ^a	PSL ^b		
	Mukvisi	Pension	Mukvisi	Pension				
Cu	0.2	0.2	0.04-0.05	0.04-0.05	1.2	30		
Zn	3.2	15	0.58-0.71	2.66-3.26	18	60		
Cd	0.2	0.1	0.04-0.04	0.02-0.02	0.012	0.06		
Ni	0.3	0.5	0.05-0.06	0.08-0.10	0.12	0.3		
Pb	0.3	0.4	0.05-0.06	0.07-0.09	0.18	0.214		
Cr	0.5	0.3	0.08-0.10	0.05-0.06	90	100		

^aMinimum Risk Level (mg day^{-1}) for a 60 kg body weight adult

^bProvisional Safe Levels (mg day^{-1}) for a 60 kg adult, derived from Provisional Tolerable Weekly Intakes (PTWI) of Cu, Cd and Pb (FAO/WHO); Tolerable Daily Intake (TDI) for Ni (WHO); Adequate Intake (AI) for Cr (MAFF) and Provisional Maximum Tolerable Daily Intake (PMTDI) for Zn (FAO/WHO)

Figure 7. Concentrations of heavy metals in soil from smallholder vegetable plots at Crowborough Sewage Farm.



Pesticide contamination.

Vegetables particularly leafy vegetables are prone to attack by several pests hence the need to apply pesticides to achieve better yield and quality during the entire period of growth and sometimes even fruiting stage. These pesticides that are absorbed by the vegetables can be hazardous to human if the vegetables consumed contains some pesticide residues. The major aim of this study on pesticide residues was to know the status of marketable vegetables in four main marketing areas of Harare in respect of prescribed maximum residue limit (MRL) values of the applied organochlorine & organophosphate pesticides applied.

Sampling strategy

The sampling strategy used to assess microbial contamination was also adopted for assessment of pesticide residue contamination.

Pesticide residue analysis

A representative sample (25g) was extracted using the acetone extraction method after maceration in a mechanical blender, Nakura et al. (1994). The extracted samples were cleaned up using column chromatography packed with florisil for organochlorines and silica gel for organophosphates packed between layers of anhydrous sodium sulphate.

The cleaned samples were analyzed on Varian gas chromatography (GC) equipped with capillary columns using electron capture (ECD) and nitrogen-phosphorous detectors (NPD).

Results

The analyzed samples indicated that some of the horticultural produce mainly leafy vegetables contained some organochlorine & organophosphate residues though the levels detected are below the Codex minimum. The data is given in Tables 7, 8, 9 & 10 and is compared with MRL values of each pesticide where possible.

Chikwanha Market

Table 7: Pesticide residues in produce from Chikwanha market in Chitungwiza. (40 samples were analyzed)

Name of pesticide	No. of samples with residues		Range µg/g	MRL. µg/g
	Leafy Veggies	Tomatoes		
DDD	2	1	0.002-0.003	NR
DDT	3	0	0.002-0.005	3.5
Deldrin	5	2	0.01 – 0.03	NR
Dimethoate	5	4	0.02 – 0.08	0.50
Parathion	4	0	0.01 – 0.02	NR
Primiphos	4	3	0.01 –0.02	1.00
Di-thiocarbamate	0	2	0.4 – 0.9	5.00

NB: *MRL = Minimum residue level*
NR = No existing CODEX MRL

Epworth Market

Table 8: Pesticide residues in produce from Epworth market in Harare. (40 samples were analyzed)

Name of pesticide	No. of samples with residues		Range µg/g	MRL µg/g
	Leafy Veggies	Tomatoes		
DDD	2	2	0.002-0.004	NR
DDT	2	0	0.003	3.5
Deldrin	6	4	0.01 - 0.04	NR
Dimethoate	7	1	0.01 - 0.08	0.50
Parathion	1	2	0.005 - 0.9	NR
Primiphos	3	3	0.01 - 0.08	1.00
Di-thiocarbamate	0	5	0.03 - 1.8	5.00

NB: *MRL* = Minimum residue level
NR = No existing CODEX MRL

Mufakose Market

Table 9: Pesticide residues in produce from Mufakose market in Harare. (40 samples were analyzed)

Name of pesticide	No. of samples with residues		Range µg/g	MRL µg/g
	Leafy Veggies	Tomatoes		
DDD	1	2	0.002-0.003	NR
DDT	2	0	0.003-0.004	3.5
Deldrin	6	5	0.01 - 0.05	NR
Dimethoate	5	2	0.01 - 0.03	0.50
Parathion	3	2	0.01 - 0.03	NR
Primiphos	4	5	0.01 - 0.02	1.00
Di-thiocarbamate	1	4	0.07 - 4.5	5.00

NB: *MRL* = Minimum residue level
NR = No existing CODEX MRL

Glenview / Tafara-Mabvuku Market

Table 10: Pesticide residues in produce from Glenview & Tafara Mabvuku market in Harare. (40 samples were analyzed)

Name of pesticide	No. of samples with residues		Range µg/g	MRL µg/g
	Leafy Veggies	Tomatoes		
DDD	3	3	0.002-0.005	NR
DDT	1	1	0.002-0.003	3.5
Deldrin	8	3	0.01 – 0.03	NR
Dimethoate	7	2	0.01 – 0.1	0.50
Parathion	3	0	0.01	NR
Primiphos	5	0	0.02 –0.04	1.00
Di-thiocarbamate	1	4	0.3 –2.4	5.00

NB: *MRL = Minimum residue level*

NR = No existing CODEX MRL

At all the four sites the main pesticide residue contained in both the leafy vegetables and tomatoes was that of dimethoate though this pesticide is classified as non-persistent. Its high occurrence could be explained by its availability on the local market and its relative low cost.

Comparing all the four marketing sites it can be noted that the marketed vegetables are contain some pesticides residues like deldrin, DDT and DDT derivatives like DDD which is not supposed to applied on vegetables according to Zimbabwean regulations. Deldrin is used mainly in cotton production but due to its relative availability on the market and its noted effectiveness when it comes to pest control farmers tend to use it. DDT is a banned substance but still available on the Zimbabwean market mainly to control malaria and tsetse fly and it was also extensively used in agriculture prior to 1993, Zaranyika (1996). The detected residues of DDD and DDT are maybe residual amounts in the soil environment, which is later taken up by plants since DDT has a high bioaccumulation potential.

Based on the results obtained in this survey it not possible to conclusively determine the risk of pesticide residue contamination from production through to consumption maybe because the sampling strategy we adopted was not sufficient.