Microbiological & Heavy Metal Contamination of Watercress in the Wellington Region

Chris Edmonds

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ABSTRACT

Watercress is a popular food and is traditionally served cooked although it is increasingly consumed raw either in salads or as a garnish. Watercress is harvested from waterways both for personal use and for commercial sale in New Zealand. Watercress grows in an aquatic environment and is subject to potential chemical and microbiological contamination from the water/sediment it grows in.

The consumption of watercress infected with enteric pathogens could potentially cause serious gastrointestinal illness (e.g. Campylobacteriosis) and people gathering watercress could also be at risk of infection from contact with contaminated surface waters. Excessive heavy metal contamination of watercress may also cause adverse health effects. The aims of this study were therefore to:

- investigate potential microbiological and heavy metal contamination of watercress and growing waters from eleven streams in the Wellington and Wairarapa regions; and
- assess public health risks regarding harvesting and consumption of watercress from these streams.

The stream sites selected covered a range of urban, semi-urban and rural catchments and a range of water quality and sediment characteristics. The sites selected were representative of catchment types found elsewhere in New Zealand.

All of the sites showed variable but significant levels of *E. coli* in both the watercress and water samples and therefore the potential for enteric waterborne pathogens to be present. *Campylobacter* was detected in the growing waters at all sites (80% of the samples) and in 11% of the watercress samples. The water results also showed levels of *E. coli* well above recommended freshwater recreational contact safety guidelines at most sites. In rural areas there is the potential for fascioliasis in persons consuming infected raw watercress.

Except for zinc on one occasion, heavy metal levels in watercress did not exceed the NZ Food Regulations (1984) at any of the sites. Watercress at urban sites had higher levels of specific heavy metals, relative to other sites. In streams subject to industrial discharges or natural processes e.g. geothermal activity, watercress may potentially bio-accumulate heavy metals to levels in excess of food regulations.

Based on the findings of this study:

- Watercress harvested from any uncontrolled surface water source in New Zealand should not be consumed unless the watercress is thoroughly cooked in boiling water.

- Watercress should not be eaten raw unless it can be demonstrated that the growing waters are strictly controlled and adequately monitored.

- People gathering watercress may be at risk of waterborne illnesses through contact with contaminated water.

- Watercress grown in sediments/water subject to significant heavy metal contamination may bio-accumulate heavy metals to levels in excess of health guidelines.

ACKNOWLEDGEMENTS

I wish to thank Sally Giles and Andrew Bichan (Regional Public Health) for their assistance with this study. I would also like to thank Bella Tua, Erin Kearney, Gail O'Leary Bronwyn Johnson and Catherine Hayes (Health Protection Officers) for helping to collect the samples. Thanks to Toby Regan for GIS input and Rohan Wells (NIWA) for identification of the watercress species. Thanks also to Dr John Love, Greg Devane and Rosemary Whyte from ESR for assistance with analysis and interpretation. Thanks to Dr Richard Hawke my supervisor for assistance and feedback throughout the study.

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1. INTRODUCTION

1.1 Justification

Watercress *(Nasturtium sp*., Figure 1) is an aquatic perennial herb that grows wild along the margins of slow moving rivers, streams, ditches and drains. It is common in New Zealand especially in the North Island (Roy *et al*., 1998). Watercress is a popular food and is traditionally served cooked although it is increasingly being consumed uncooked in salads or as a garnish, and is being sold as a salad vegetable in restaurants/cafes. Watercress is harvested both for personal use and for commercial sale in New Zealand.

Figure 1: Watercress

As watercress grows in an aquatic environment (sometimes completely submerged), it is subject to potential chemical and microbiological contamination from the water/sediment in which it grows. Sites favourable for watercress usually have comparatively small volumetric flows, which do not allow for much dilution of the pollutants they receive and are often either in urban or intensive agricultural areas.

Many streams in New Zealand are subject to faecal contamination from urban and rural runoff (diffuse and point sources), which can introduce enteric pathogens e.g. *Camplyobacter*, into the receiving waters (Ball and Till, 1998). The consumption of watercress infected with enteric pathogens could potentially cause cases of gastrointestinal illness and therefore could be a potential risk factor contributing to New Zealand's high incidence of certain enteric diseases e.g. Campylobacteriosis (Ministry of Health, 1998). Persons gathering watercress could also be at risk of infection from contact with contaminated surface waters.

Despite the fact that salad vegetables are generally regarded as low risk foods they can harbour a range of pathogens. Since these foods are not necessarily cooked before consumption they can pose a health risk to the consumer (Graham, 1999). Watercress may pose a significantly greater risk than other common salad vegetables (e.g. lettuce) as it grows in waterways potentially subject to waterborne contaminants. Other common salad vegetables, would not be exposed to the majority of waterborne pathogens due to their land based or controlled hydroponic growing environments.

Although watercress has not formally been linked to any enteric disease outbreak in New Zealand, many overseas outbreaks of human gastro-enteritis have been linked to the consumption of contaminated fresh vegetables (Beuchat, 1996), and watercress is included in a list of salad vegetables which have caused extensive outbreaks of salmonellosis (ICMSF, 1998).

In terms of heavy metal contamination, a number of studies undertaken on Central North Island Rivers, e.g. the Waikato River, have revealed levels of arsenic in watercress above the World Health Organisation limit for food stuffs. This is attributed to the geothermal activity in the area and the impact of geothermal power stations (Robinson *et al*., 1995; Deely, 1998). Overseas studies have also demonstrated watercress's ability to uptake certain heavy metals from contaminated water/sediments e.g. (Wong, 1985).

1.2 Aims of Study

The aims of this study were therefore:

- to investigate potential microbiological and heavy metal contamination of watercress and growing waters from a number of streams in the Wellington and Wairarapa regions; and
- to assess public health risks regarding harvesting and consumption of watercress from these sites.

1.3 Sources of Microbiological Contamination

There are two main ways in which microorganisms of faecal origin can enter surface waters. First, through point source discharges, such as discharges of sewage, farm effluent or urban stormwater. Second, through diffuse sources of faecal pollution, such as from livestock grazing adjacent to waterways (Ball, 1997).

From overseas studies it is apparent that there is a wide variation in the occurrence of faecal indicators and pathogens in effluent, both from animal and human sources (Ball, 1997). The numbers of pathogens in sewage is a function of their abundance in the population and their survival in the environment. For point source discharges of sewage or animal wastes, the method of wastewater treatment has a large effect on the microbial load, whereas for diffuse pollution sources, factors such as temperature, sunlight and exposure times may be more significant (Ball, 1997). Higher concentrations of pathogens might be expected in the event of disease outbreaks, and seasonal variation can be expected due to the seasonal nature of some human and animal diseases (Ball 1997).

Microorganisms tolerate unfavourable environments to varying degrees. With few exceptions, pathogens and faecal indicator organisms decline upon entry to fresh water environments (Ball, 1997). Factors, which contribute to microbial death rate, include exposure to sunlight, water temperature, predator activity, sedimentation and the chemical components of the water. The responses of various micro-organisms to these factors vary greatly, so the relationship between pathogens and indicators is likely to differ in faecal material which has been resident in a water body for different periods of time due to differing environmental exposures (Ball, 1997). Following rainfall, microbial loading may be significantly increased due to surface run-off, urban storm water, faulty sewage reticulation overflows, and re-suspension of river sediments.

Farm runoff, in particular that from dairy farms, adds significantly to the contamination of waterways (MfE, 1999b). Apart from farm oxidation ponds, which have been shown to contain high levels of bacteria and pathogens, a large amount of animal waste ends up on the paddocks. Depending on a number of factors including, distance to the nearest stream, rainfall intensity, stock numbers etc., faecal material ends up in waterways in variable quantities (MfE, 1999b). Where stock have access to streams and rivers for drinking water, faecal material may be deposited directly into the water.

Urban stormwater may contain waste from domestic animals, which collects on footpaths, gutters and lawns. This contaminated water is washed into the stormwater system during rainfall and ends up in urban streams. Animal waste collected in stormwater can contain disease causing organisms such as *Campylobacter* which may affect human health if ingested.

There are a number of factors which can cause sewage to enter the environment before reaching the treatment facility e.g. broken or leaking pipes, construction activities (MfE, 1999b). During high rainfall the treatment facility may not be able to cope with the volumes of water and sewage entering the system, so pumps are turned off and sewage is discharged directly to the environment. This is made worse where houses have stormwater illegally connected to waste water. Some older sewerage systems do not have completely separated sewage and stormwater pipes. During high rainfall stormwater can enter the sewerage system and cause sewage to overflow into the stormwater pipes. Sewage is then discharged directly to the environment. Emergency overflows can also occur periodically during maintenance of sewerage systems.

1.4 Sources of Heavy Metal Contamination

Heavy metals are potential contaminants in surface waters subject to rural and urban discharges. Heavy metals are known to bind with sediments, particularly finer sediments (Aitken *et al*., 1997), and may be re-mobilised into the water and/or food chain if sediment is re-suspended. Most heavy metals originate from industrial areas, and due to their affinity for sediments are transported through stormwater and sewer systems and deposited in low energy areas in surface waters (Aitken *et al*., 1997). There are a number of heavy metals that could be present in growing waters/sediments from industrial discharges, and other sources including landfill run off, e.g. nickel, cadmium, copper, zinc, chromium, arsenic, lead and mercury.

1.4.1 Nickel

Nickel can enter the environment naturally through weathering of minerals and rocks and run-off from soils (MOH, 1995). Industrial discharges can also contribute to the nickel in water. Nickel is used mainly in the production of stainless steel and other corrosion-resistant alloys and as a catalyst in industrial processes and in oil refining (MOH, 1995). More than 90% of the nickel in the aquatic environment is associated with particulate matter of sediments (ANZECC, 1992).

1.4.2 Cadmium

Cadmium has a wide range of sources and may enter water from industrial and domestic discharges or from street and agricultural runoff (MOH, 1995). Its principal industrial uses are in electroplating other metals or alloys for corrosion protection, in solders and in amalgam used in dentistry. In agriculture, farm run-off of phosphate fertilisers containing cadmium is an important source of diffuse cadmium pollution (MOH, 1995). Exhaust emissions and tyre wear contributes a significant amount of cadmium to street run-off. Cadmium may be accumulated by a number of aquatic organisms, with bio-concentration factors in the order of 100-100,000 (ANZECC, 1992).

1.4.3 Copper

Copper occurs widely in nature in rocks and soils as sulphide and carbonate compounds (MOH, 1995). Copper is used in a range of industries including timber treatment, the manufacture of electrical wiring, electroplating and textiles (MOH, 1995). Plants and animals readily accumulate copper; bio-concentration factors ranging from 100 to 26000 have been reported for various species of phytoplankton, zooplankton, macrophytes, macroinvertebrates and fish (ANZECC, 1992).

1.4.4 Zinc

Zinc can enter the environment from both natural processes (e.g. weathering and erosion) and through anthropogenic means (e.g. zinc production, waste incineration) (ANZECC, 1992). Zinc is used as a coating to prevent corrosion of iron and steel products, and in the manufacture of brass. Zinc oxide is an important component in the manufacture of paint and rubber products including tyres (MOH, 1995).

1.4.5 Chromium

Chromium is present in most soils and rocks and it can enter water naturally from weathering and run-off from soils (MOH, 1995). In natural waters chromium exists in two main inorganic forms, hexavalent (6+) and trivalent (3+) (ANZECC, 1992). Chromium is used in a range of industries, especially leather tanning and timber treatment. Hexavalent chromium compounds are used in the metallurgical industry and in the chemical industry. Trivalent chromium salts are used in textile dying, in the ceramic and glass industry, and in photography (MOH, 1995).

1.4.6 Arsenic

Arsenic can enter the aquatic environment by the weathering of minerals and rocks, run-off from soils, from geothermal fluids or atmospheric deposition (ANZECC, 1992). Arsenic can also enter the aquatic environment through industrial discharges. Arsenic and its compounds are used in the production of semiconductors, pigments, in alloys for lead and copper, insecticides, herbicides and as timber preservatives (MOH, 1995).

1.4.7 Lead

Anthropogenic outputs of lead to the environment outweigh all natural sources (e.g. weathering of sulphide ores, especially galena) (ANZECC, 1992). Lead reaches the aquatic environment through precipitation, fall out of lead dust, street runoff and industrial and municipal wastewater discharges (ANZECC, 1992). It is used in a wide range of industries for example, the manufacture of acid-storage batteries, solder, piping and is used in electroplating and the metallurgy industries (MOH, 1995).

1.4.8 Mercury

Mercury can enter the aquatic environment from the weathering of rocks and minerals, runoff from soils and geothermal activity (MOH, 1995). Industrial discharges can also contribute to the mercury in water. Mercury is used in industry in paint preservatives and pigments, in pulp and paper manufacture and in the production of electrical equipment (MOH, 1995).

To get a good representation of different growing sites it was decided to sample watercress and growing waters from a number of surface water sites in the Greater Wellington Region. To assess heavy metal contamination, the concentrations of the heavy metals referred to above would be measured. These heavy metals are commonly monitored as part of resource consent conditions for wastewater discharges in the Wellington Region. Studies in the lower reaches of Waiwhetu Stream have also confirmed that the sediments are strongly enriched with lead and zinc, and moderately enriched with cadmium, chromium, nickel, copper, and arsenic (Aitken, 1998). To assess potential microbiological contamination, suitable faecal indicator organisms (e.g. *E. coli*) and a common waterborne pathogen (*Campylobacter*) would be tested.

2. STUDY AREA

Surface waters in the Wellington Region including the Kapiti Coast and Wairarapa, were chosen as sample sites for this study. Surface waters in these areas are subject to variable flow and water quality.

The average rainfall for Wellington is approximately 1,230mm/yr, although rainfall varies widely depending on altitude, orographic effects and the paths of rain bearing storms. In the Hutt Catchment for example, rainfall varies from about 1,200mm/yr in low-lying areas to more than 6,000mm/yr in the headwater areas of the Tararua Ranges (McConchie, 2000). Rainfall is distributed relatively evenly throughout the year with only a slight winter maximum (McConchie, 2000).

High water quality in the Wellington Region is only recorded for sites that have limited development in their surrounding catchments (McConchie, 2000). Sites with good water quality are either in, or in close proximity, to forest parks. Sites with poor water quality have comparatively small volumetric flows and are often either in urban or intensive agricultural areas (WRC, 1999).

Within the Wellington region the parent material of the soils is largely derived from a single source rock – greywacke and associated argillite – which form the hills and mountains of the district. In addition to the materials derived from greywacke, other soil parent materials include volcanic ash from the Taupo region, and peat (McConchie, 2000).

2.1 Site Selection Criteria

Eleven streams in the greater Wellington Region (Figure 2) were selected as watercress sampling sites. The sites were intended to cover a range of urban, semi-urban and rural catchments and a range of water quality and sediment characteristics. The sites were also selected on the basis that they were representative of catchment types generally applicable elsewhere in New Zealand. Specific sites were selected based on:

- they are known watercress harvesting sites, and/or
- they are geographically representative of watercress growing areas of the region, and/or
- they are potentially exposed to a range of chemical and microbiological contamination.

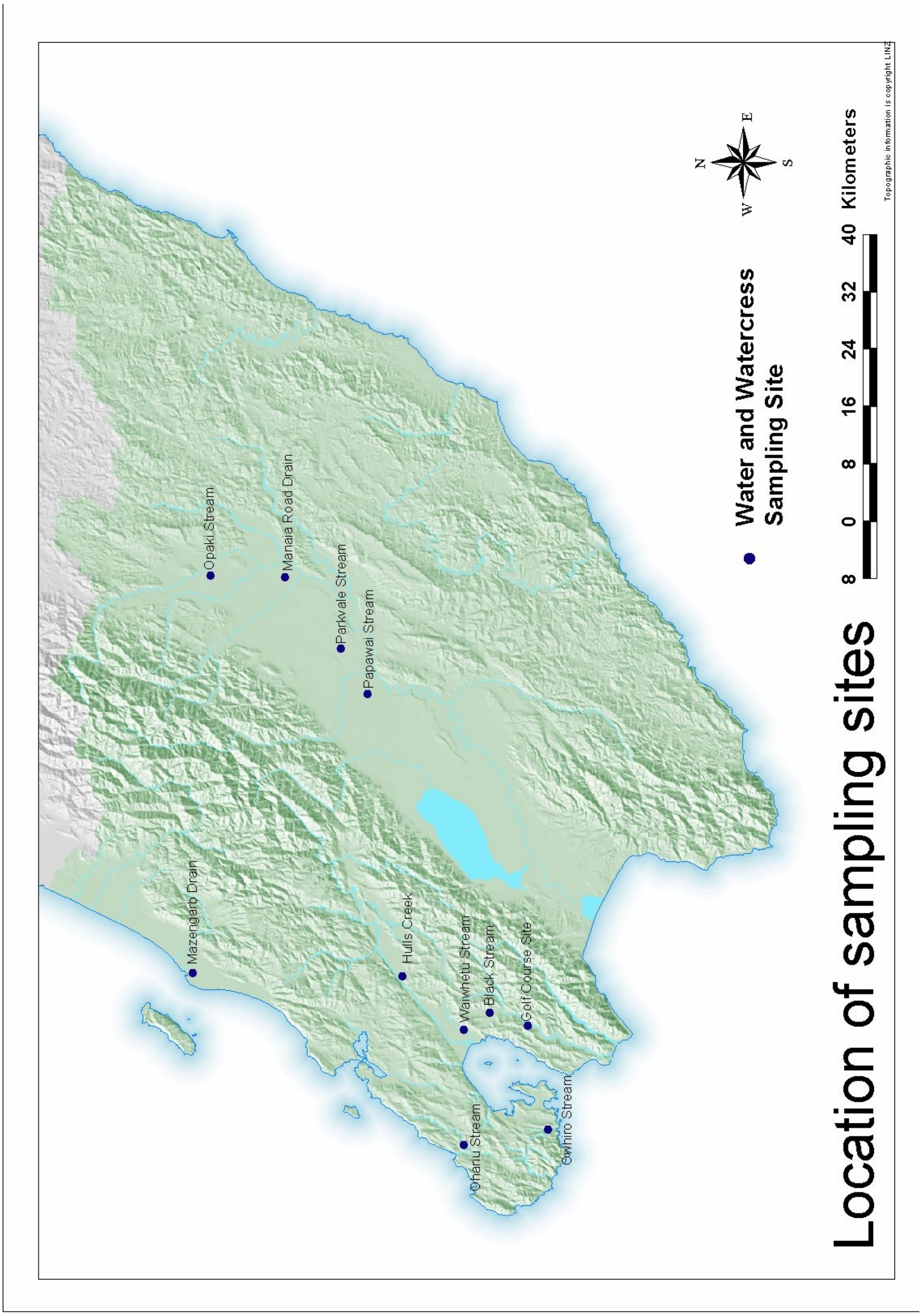
A major limitation to selecting sites was whether watercress grew in sufficient quantity to allow sampling for five weeks. Of the eleven sites where there was sufficient watercress available for this study, five were known to be sites where watercress is gathered on a regular basis either for commercial sale or personal consumption (four in the Wairarapa & one in Wellington which were identified as collection sites after discussions with local iwi, land owners and local authorities).

The sites chosen were:

- Waiwhetu Stream (urban)
- Hulls Creek (urban)
- Mazengarb Drain (semi-urban)
- Owhiro Stream (semi-urban)
- Ohariu Stream (rural)
- Black Stream (urban)
- Wainuiomata Golf Course (bush)
- Papawai Stream (rural)
- Parkvale Stream (rural)
- Manaia Drain (rural)
- Opaki Stream (rural)

The Wainuiomata Golf Course site was selected as a ‘control site’ for heavy metal contamination (the Wainuiomata Water Supply Catchment was the first choice as a control site, but was not considered suitable due to the absence of watercress in sufficient quantities). In terms of a control site for microbiological contamination, it was considered that watercress not grown in controlled conditions would be subject to varying degrees of faecal contamination. It was therefore not considered feasible to select a ‘control site’ for microbiological contamination although the golf course site was considered free from all but feral animal contamination.

Figure 2: Location of Sampling Sites



2.2 Individual Sites

2.2.1 Waiwhetu Stream

The Waiwhetu Stream (Figure 13) is a small, slow-flowing stream which originates in the eastern Hutt hills above the suburb of Naenae (Aitken, 1998). The stream flows from relatively uncontaminated headwaters east of Taita Cemetery, through the suburbs of Naenae, Epuni, Waterloo, Waiwhetu, Gracefield, and Seaview. The catchment land cover is approximately 55% urban, 40% scrub and 5% indigenous forest (Source: WRC Land Cover Database 2001). The estimated mean flow is 293 l/s (Harkness, M. (WRC) pers. comm., January 2001).

The urban portion of the catchment has an extensive stormwater system which discharges directly into the Waiwhetu Stream (Aitken, 1998). Sections of the Waiwhetu Stream have experienced, over many years, industrial waste discharges resulting in heavily polluted sediments (Aitken, 1998). However, the majority of these discharges were located downstream of the selected sampling site. Watercress was not abundant at the sampling site as the stream bank had been recently cleared of weed.

2.2.2 Mazengarb Drain

The Mazengarb Drain (Figure 7) is a drainage channel with a sandy bed, low water velocity and little riparian vegetation. The drain discharges into the estuary zone of the Waikanae River. Surface runoff into the drain includes tertiary treated wastewater from the Paraparaumu Sewage Treatment Plant, run-off from the Paraparaumu Landfill and rural and urban stormwater run-off. The catchment land cover is approximately 15% urban, 76% pastoral, 4% indigenous forest, 2% planted forest and 2% scrub (Source: WRC Land Cover Database 2001). The estimated mean flow is 173 l/s (Harkness, M. (WRC) pers. comm., January 2001).

The Mazengarb Drain has consistently ranked as one of the poorer quality water bodies on the Kapiti Coast (KCDC, 1998). The sampling site was located downstream of both the Sewage Treatment Plant discharge and the landfill branch.

2.2.3 Ohariu Stream

Ohariu Stream (Figure 8) flows down the Ohariu valley (which is a semi-rural catchment) before its confluence with the Makara Stream. The catchment land cover is approximately 1% urban, 90% pastoral, 1% indigenous forest, 3% planted forest and 5% scrub (Source: WRC Land Cover Database 2001). The estimated mean flow is 349 l/s (Harkness, M. (WRC) pers. comm., January 2001).

The stream is likely to be subject to predominantly non point source rural run-off e.g. septic tanks and stock which have access to the stream. The sampling site was located approximately 2km before Ohariu Stream’s confluence with the Makara Stream.

2.2.4 Owhiro Stream

Owhiro Stream (Figure 10) flows through the suburb of Brooklyn, and the Happy Valley area before discharging at the Western end of Owhiro Bay. Owhiro stream is met by Carey's Stream from the West. Careys Stream flows through a culvert under the Happy Valley Landfill and alongside Landfill Road. The majority of the catchment is in gorse scrubland. The catchment land cover is approximately 7% urban, 4% pastoral, 85% scrub and 4% bare ground and landfill (Source: WRC Land Cover Database 2001). The estimated mean flow is 58 l/s (Harkness, M. (WRC) pers. comm., January 2001). Most of the leachate from the landfill is collected and piped to the sewer at Moa Point. The sampling site is located downstream of the Carey's Gully stream confluence, adjacent to Happy Valley Park.

2.2.5 Hulls Creek

Hulls Creek (Figure 5) flows from an upper scrubland hill area and through the Rimutaka Prison farm. Just downstream of the prison farm an eastern tributary meets Hulls Creek and drains the back of the farm and a military rifle range. Stock can access the creek throughout much of this area. Another tributary flows into Hulls Creek just before the floodgates. The catchment of this tributary (rifle range tributary) includes the military rifle range and a small farm area.

Further downstream the Pinehaven and Silverstream Tip Streams discharge into Hulls Creek before it discharges into the Hutt River. The catchment land cover is approximately 34% urban, 9% primarily pastoral, 18% indigenous forest, 11% planted forest, 25% scrub, and 3% bare ground and dumps (Source: WRC Land Cover Database 2001). The estimated mean flow is 317 l/s (Harkness, M. (WRC) pers. comm., January 2001). The sampling site was located immediately downstream of the Pinehaven stream discharge (before the Silverstream Tip Stream confluence).

2.2.6 Black Stream

Black Stream (Figure 3) originates in a bush/gorse catchment and passes through Wainuiomata's urban area before discharging into the Wainuiomata River. The stream is subject to significant urban runoff. The catchment land cover is approximately 32% urban, 4% primarily pastoral, 9% indigenous forest and 55% scrub (Source: WRC Land Cover Database 2001). The estimated mean flow is 211 l/s (Harkness, M. (WRC) pers. comm., January 2001).

The sampling site was located approximately 1 km upstream of its confluence with the Wainuiomata River.

2.2.7 Wainuiomata Golf Course Stream

The stream's catchment (Figure 4) is a small bush/gorse covered valley immediately southwest of the Wainuiomata Golf Course. The catchment is not subject to urban or rural runoff and was considered to have few contaminant sources due to the protected nature of the catchment. The catchment land cover is approximately 10% indigenous forest and 90% scrub (Source: WRC Land Cover Database 2001). There is no data available on flow rates.

The sampling site was located immediately where the stream emerges from the bush/gorse cover.

2.2.8 Papawai Stream

The Papawai Stream catchment (Figure 11) is subject to predominantly rural runoff (mainly dairy farms) and some urban runoff from Greytown. While there are no consented dairy discharges direct to the stream, there are significant non-point discharges. The catchment land cover is approximately 4% urban, 95% primarily pastoral, 1% indigenous forest (Source: WRC Land Cover Database 2001). The estimated mean flow is 300 l/s (Harkness, M. (WRC) pers. comm., January 2001). The sampling site is located upstream of the Greytown Sewage Treatment Plant discharge where Fabians Road crosses over the stream. The site is a known watercress collection site for commercial sellers (Johnson, B. (Choice Health) pers. comm., 2000).

2.2.9 Parkvale Stream

The Parkvale Stream catchment (Figure 12) is predominantly farmland. The stream is subject to mainly dairy farm runoff, similar to the Papawai Stream. The catchment land cover is approximately 1% urban, 97% primarily pastoral, 1% indigenous forest, 1% planted forest (Source: WRC Land Cover Database 2001). The estimated mean flow is 360 l/s (Harkness, M. (WRC) pers. comm., January 2001). The sampling site is located where Para Road crosses the stream. The site is another known collection site for commercial sellers (Johnson, B. (Choice Health) pers. comm., 2000).

2.2.10 Manaia Drain

The Manaia Road Drain's catchment (Figure 6) is predominantly farmland. Relative to the other sites in the Wairarapa it has a comparatively small catchment and a corresponding low discharge. The catchment land cover is approximately 1% urban, 99% primarily pastoral (Source: WRC Land Cover Database 2001). There is no data available on flow although it would be significantly lower than any of the other sites in this study. The sampling site is adjacent to Gladstone Road and is a known collection site for local iwi and the general public (Johnson, B. (Choice Health) pers. comm., 2000).

2.2.11 Opaki Stream

The Opaki Stream (Figure 9) catchment is mainly farmland with predominantly dairy farm runoff, similar to the other Wairarapa streams in this study. The catchment land cover is approximately 4% urban, 95% primarily pastoral and 1% bare ground (Source: WRC Land Cover Database 2001). The estimated mean flow is 250 l/s (Harkness, M. (WRC) pers. comm., January 2001). The sampling site is located immediately north of Masterton where S.H. 2 crosses the stream. The sampling site is a known collection site for local iwi and the general public in the vicinity (Johnson, B. (Choice Health) pers. comm., 2000).

The eleven sites chosen for this study cover a wide range of catchment types, including predominantly rural, urban, scrub, and bush covered, and contain variable contaminant sources. The waterways would be typical of waterways in other areas in New Zealand and therefore results would be expected to be generally applicable to other parts of the country.

Figure 3: Black Stream (Note: pink stars specify sampling range) **1km**

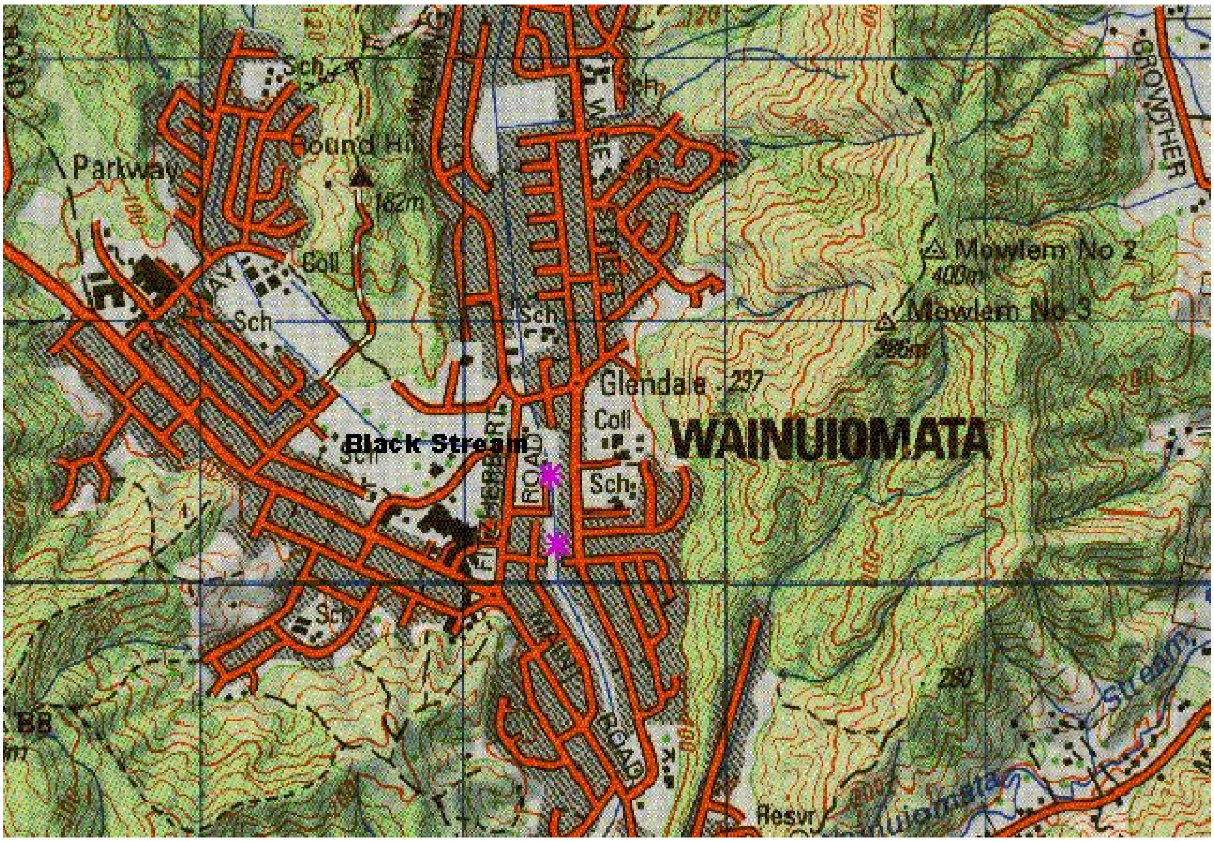


Figure 4: Wainuiomata Golf Course Stream **1km**

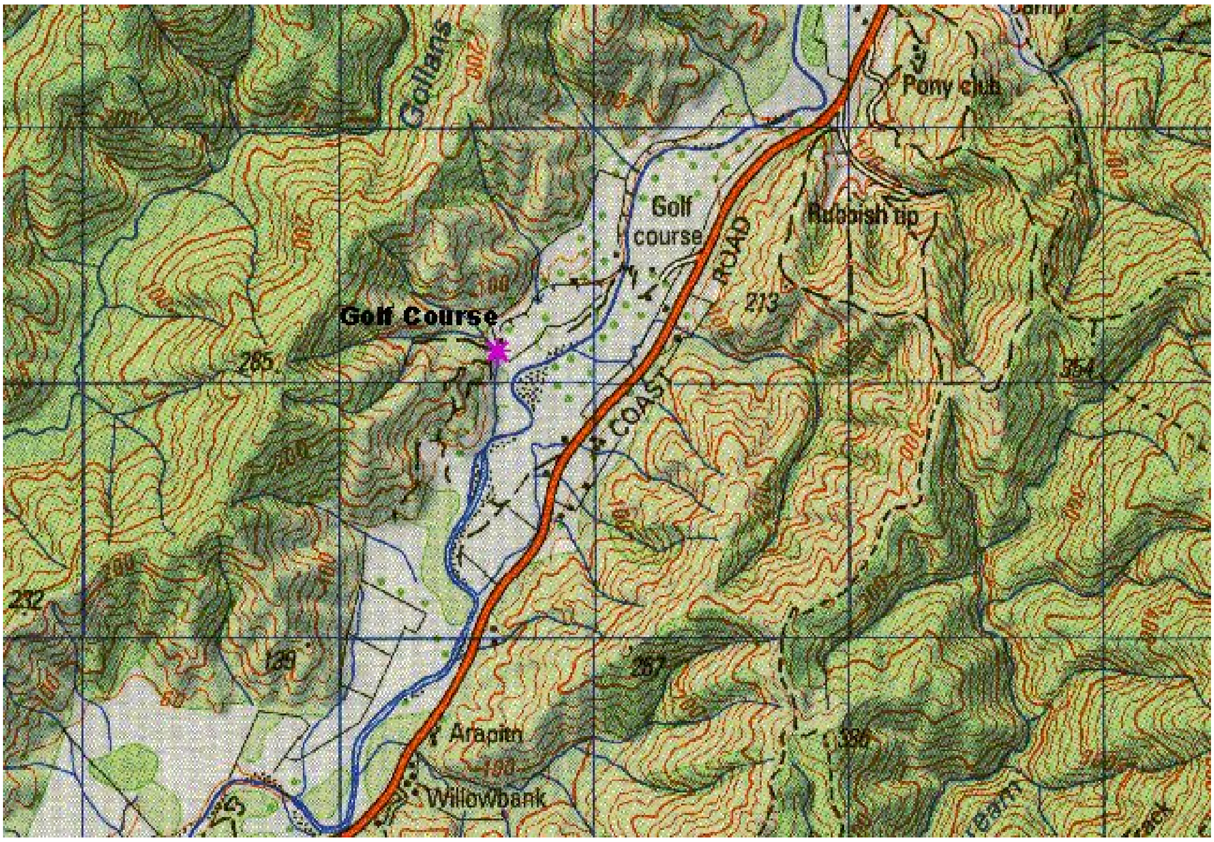


Figure 5: Hulls Creek **1km**

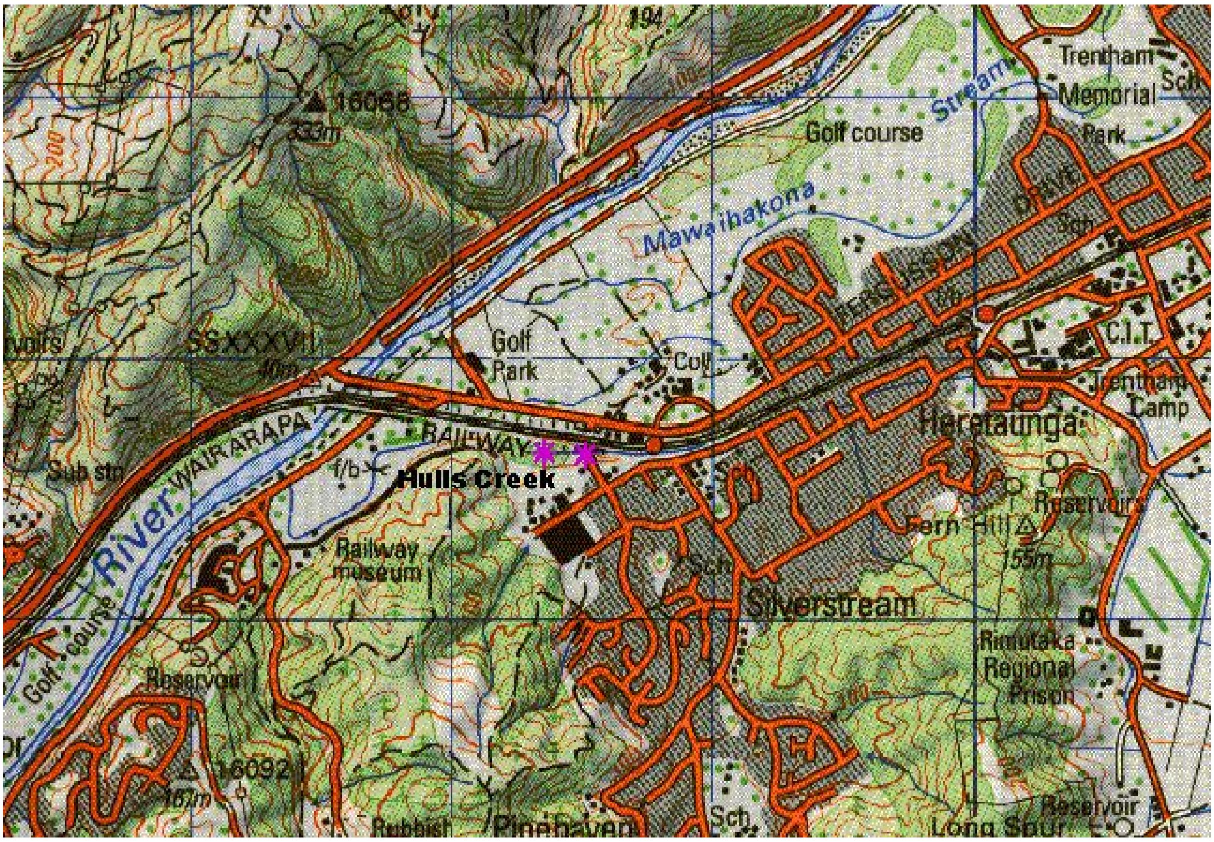


Figure 6: Manaia Road Drain **1km**

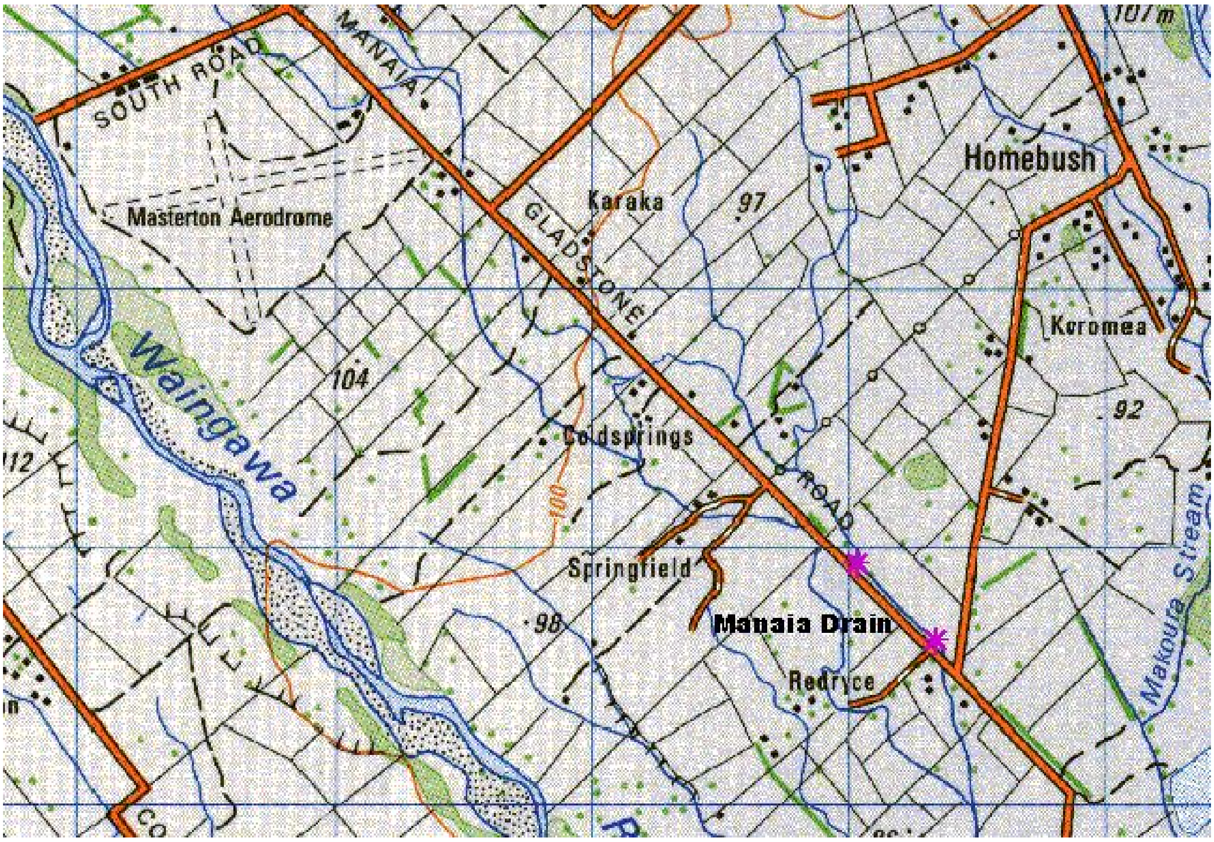


Figure 7: Mazengarb Drain 1km

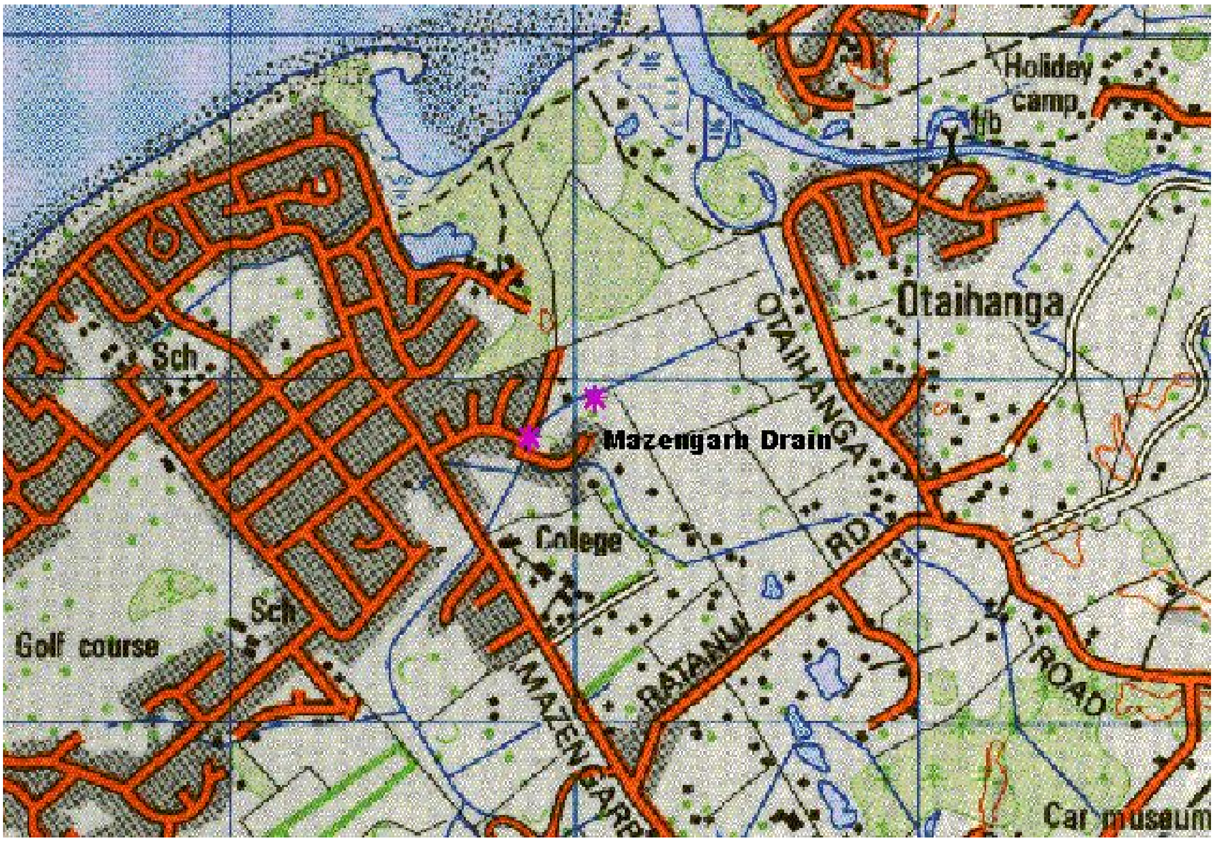


Figure 8: Ohariu Stream 1km

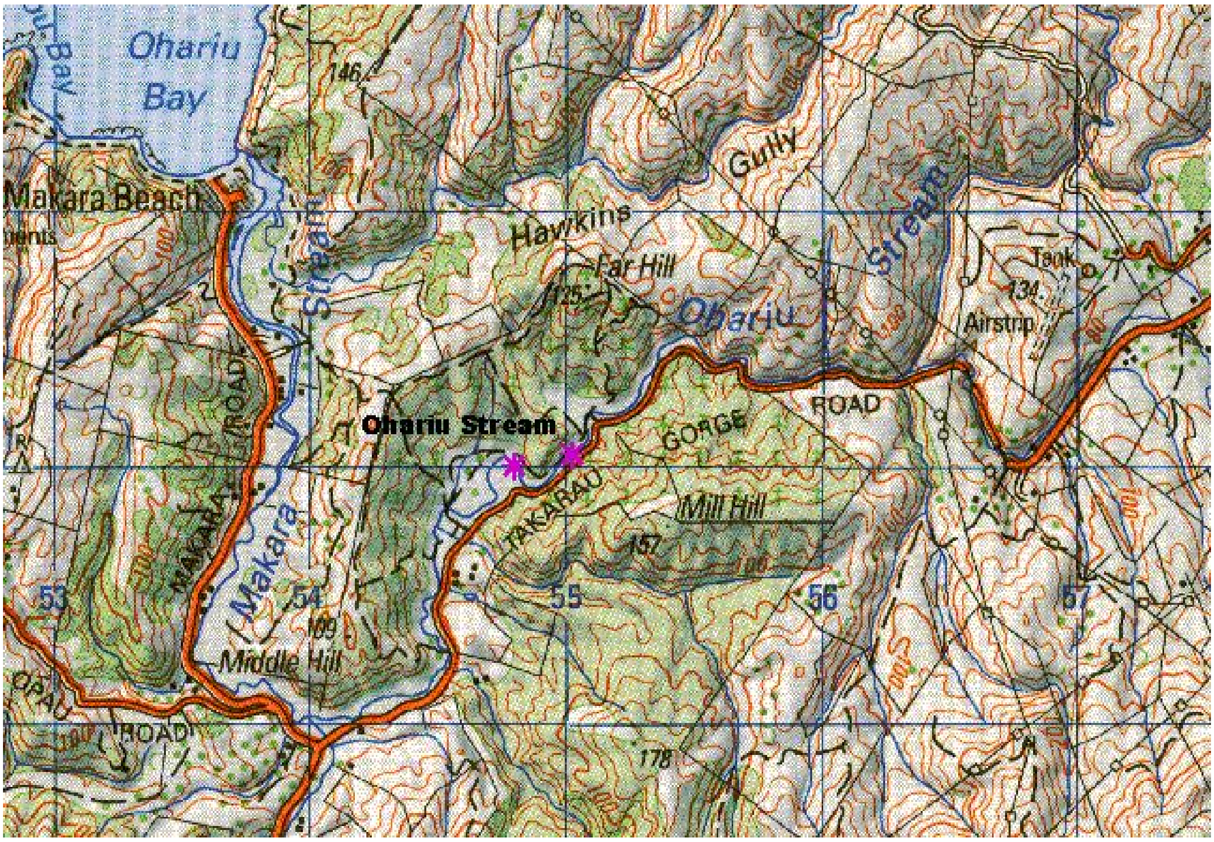


Figure 9: Opaki Stream 1km

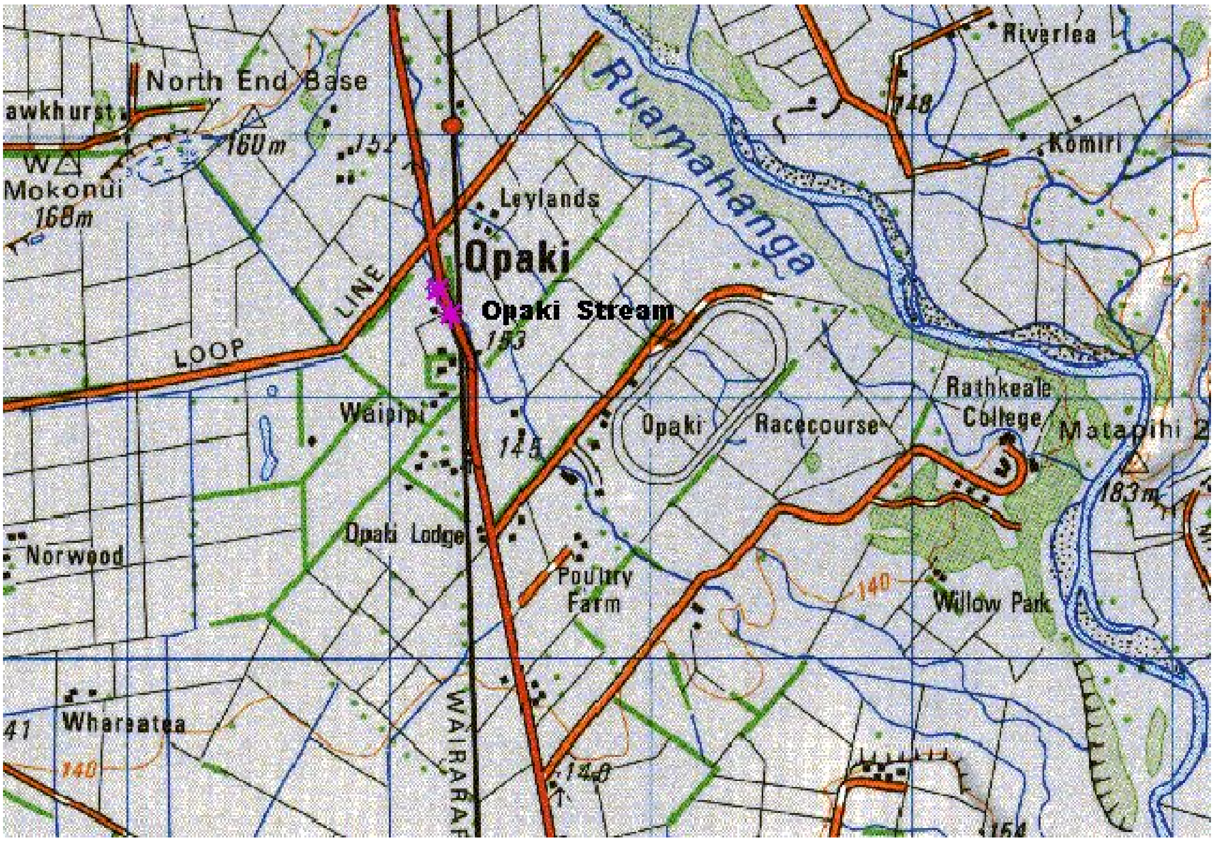


Figure 10: Owhiro Stream 1km

Figure 11: Papawai Stream 1km

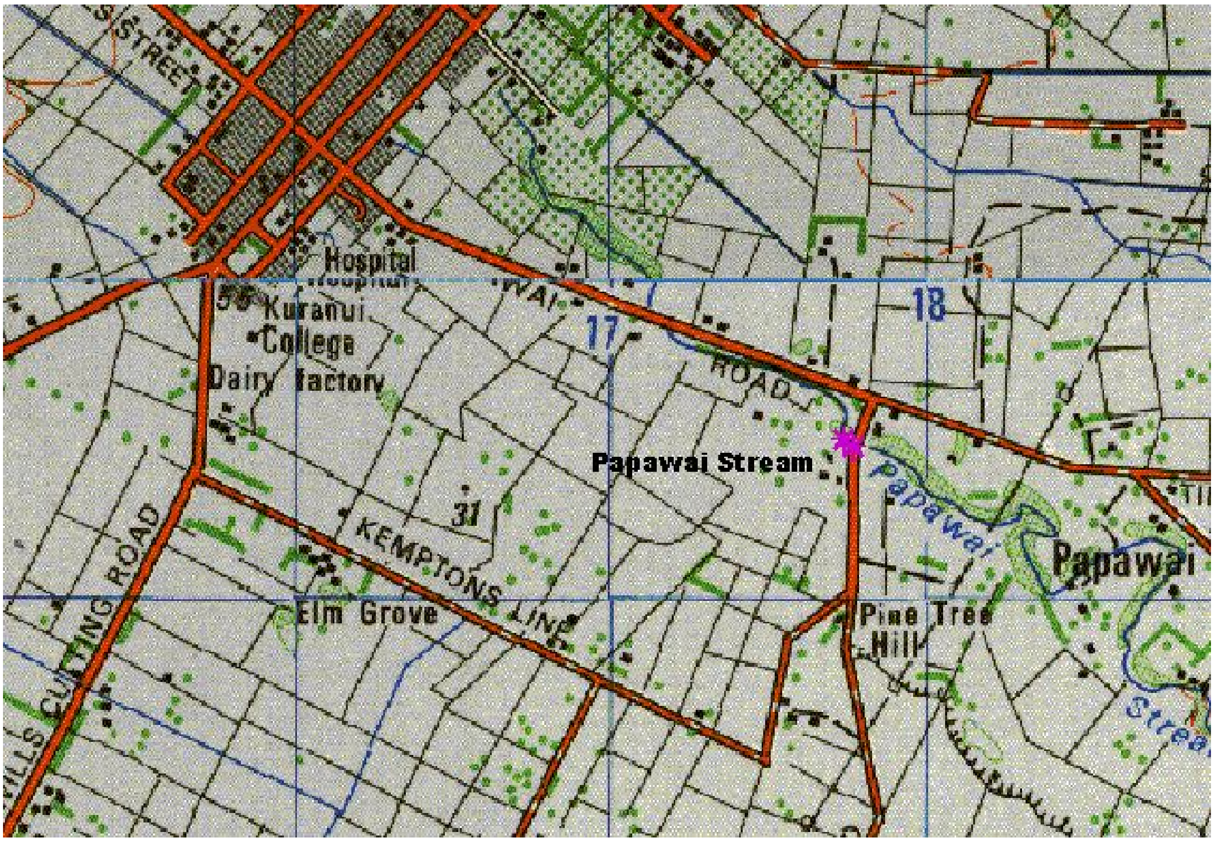


Figure 12: Parkvale Stream 1km

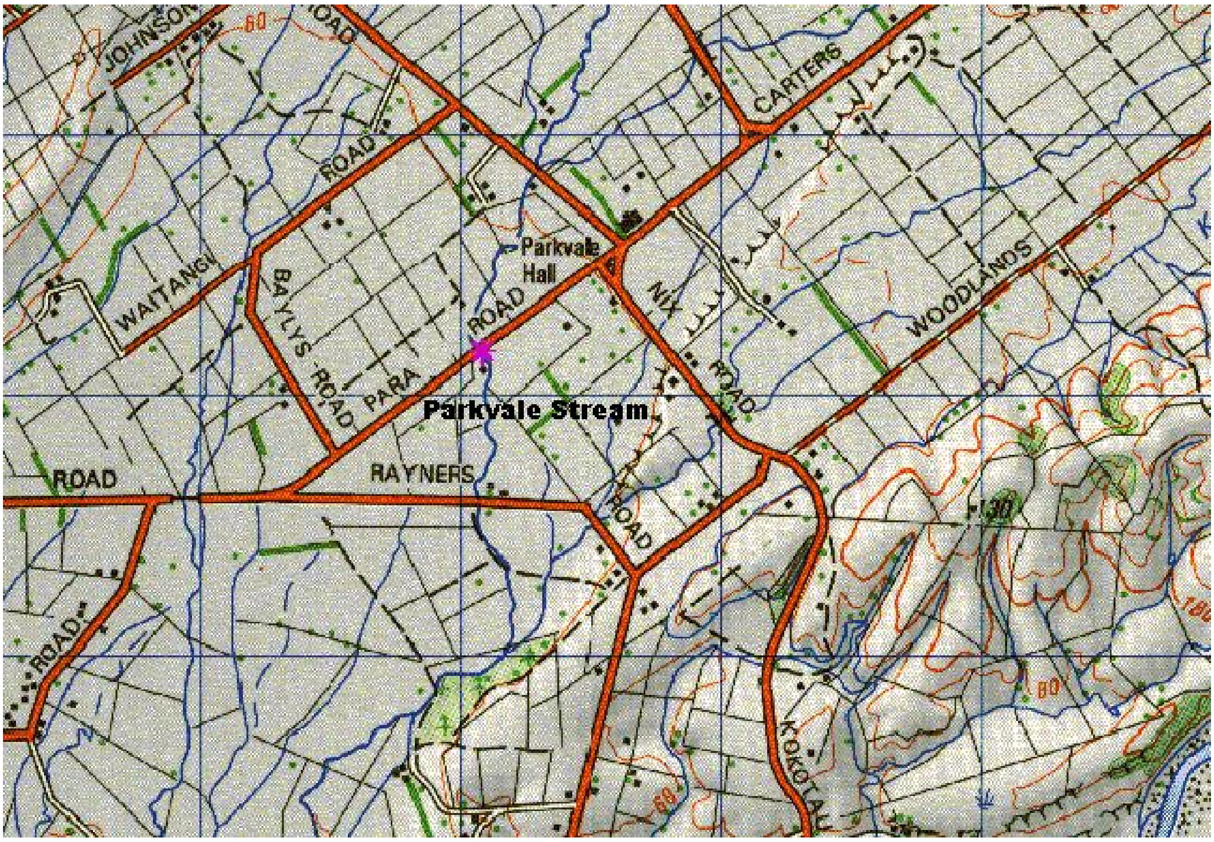
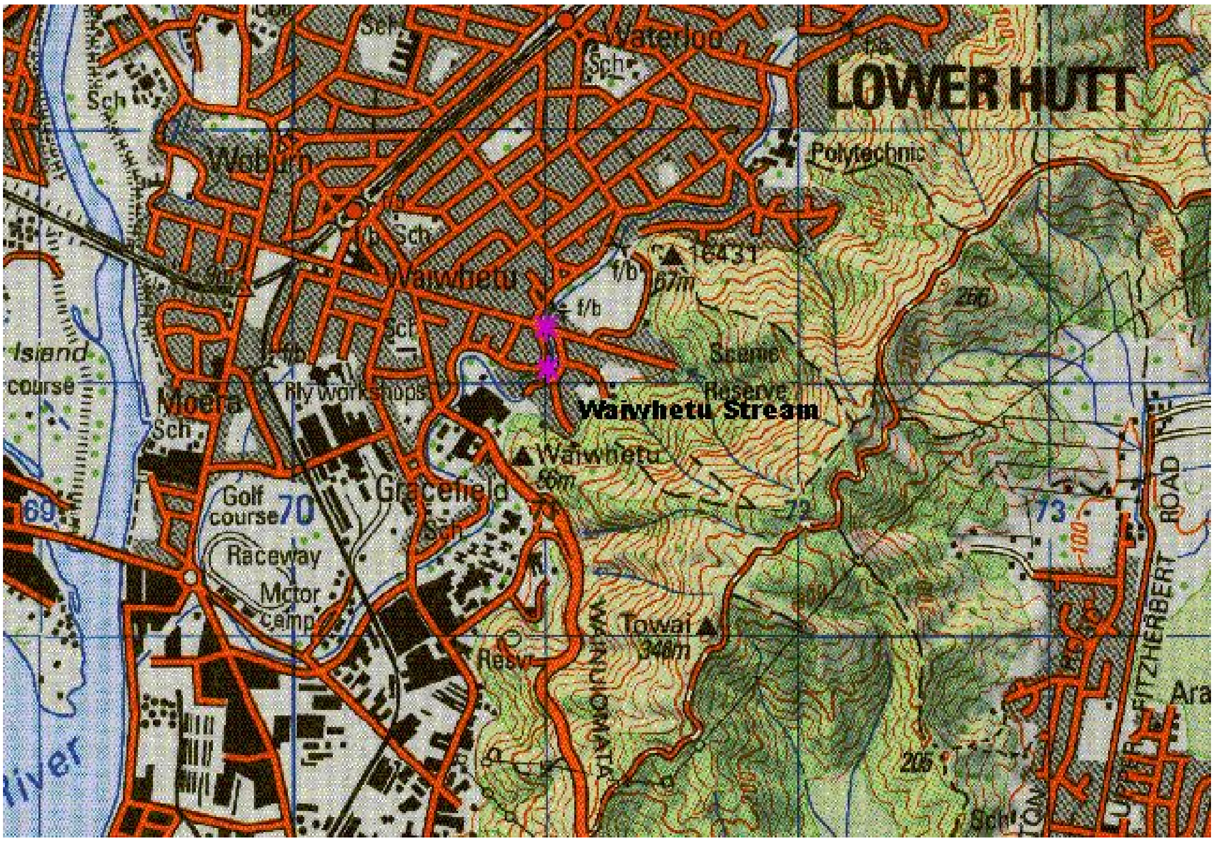


Figure 13: Waiwhetu Stream **1km**



3. ANALYSIS

3.1 Sampling Technique

Watercress was gathered along a variable length of each of the waterways depending on the abundance of the watercress. Sampling commenced in the last week of March 2000 (except for the golf club site) and continued for a total of five weeks. The reason for testing over a five-week period was to assess variability in contaminant concentrations.

In the first week, five 300g watercress samples were collected from each of the sites for individual microbiological analysis in order to be able to compare microbiological results against the Ministry of Health's "*Microbiological Reference Criteria for Food 1995*". The exception was Waiwhetu Stream where only one sample was collected each week as there was insufficient watercress available. For the remaining four weeks, five samples (if available) were taken from each site and combined by the laboratory into one sample for analysis. In summary, a total of 9 watercress samples were analysed from each site except from Waiwhetu where 5 samples were analysed.

Healthy looking watercress was selected randomly from the stream. The whole plant excluding the roots was taken. A fresh pair of sterile disposable gloves was used to collect each sample of watercress. Each sample was placed into an appropriately labelled sterile swirl bag and placed in a chilly bin with slika pads for overnight transportation to ESR for microbiological and heavy metal analysis.

Growing waters were only sampled for microbiological contamination. However, testing of growing waters (and potentially sediments) for evidence of heavy metal contamination would be undertaken if watercress samples showed significant contamination. A 1L water sample was taken from each of the sites simultaneously to watercress sampling. The water sample was collected adjacent to where the watercress was collected. Each sample was collected in a sterile bottle, appropriately labelled and placed in the chilly bin for overnight delivery to ESR.

3.2 Watercress Identification

To identify the species of watercress present at each site, seedheads and/or flowers (where available) were sent to Rohan Wells, a Freshwater Ecologist at NIWA in Hamilton, for identification:

- Papawai Stream: *Rorippa microphylla*
- Parkvale Stream: *Rorippa microphylla & Rorippa nasturtium-aquaticum*
- Manaia Drain: *Rorippa microphylla & Rorippa nasturtium-aquaticum*
- Opaki Stream: *Rorippa nasturtium-aquaticum* (Identified on the basis of flower size only, not seed which is positive ID).
- Mazengarb Drain: *Rorippa nasturtium-aquaticum*
- Owhiro Stream: *Rorippa microphylla*
- Hulls Creek: *Rorippa nasturtium-aquaticum*
- Black Stream: *Rorippa nasturtium-aquaticum & Rorippa microphylla*
- Waiwhetu Stream: *Rorippa microphylla*
- Ohariu Stream: *Rorippa nasturtium-aquaticum*
- Golf Course Stream: (no flowers or seed capsules present to positively identify the species).

Although two species were identified, the morphology of both is similar and therefore a significant variation in contaminant concentrations between the two species of watercress would not be expected.

3.3 Microbiological Analysis

3.3.1 Summary

Microbiological testing of the watercress included bacterial counts for presumptive coliforms, faecal coliforms, *Escherichia coli (E. coli)* and presence/absence tests for *Campylobacter* species. Microbiological testing of the growing waters included bacterial counts for total coliforms, *E. coli* and presence/absence tests for *Campylobacter* species.

Of all the coliforms, *E. coli* is the most specific indicator of faecal contamination readily available (MfE, 1999b). The public health significance often depends on their origin, but generally, *E. coli* levels are taken as an indicator of the degree of faecal pollution, and the potential for enteric pathogens to be present.

E. coli comes from the family of bacteria known as Enterobacteriaceae and is the most common bacteria of this group (MfE, 1999b). It is nearly always found in the gut of humans and animals, usually in high numbers. It can survive for up to four to six weeks in fresh water especially when shaded from sunlight and is a definite indication of recent faecal contamination (MfE, 1999b).

E. coli is the preferred indicator organism for fresh waters and was therefore chosen as the most appropriate indicator organism for testing watercress and growing waters. The probability of *E. coli* multiplying in water is very small so the number detected relates to the original level of faecal contamination (MfE, 1999b).

There are many different types, or strains, of *E. coli*. Many are harmless to humans, but some pathogenic strains of *E. coli* are very serious e.g. *E. coli* 0157, because of the symptoms they cause and the extremely low dosage required to cause these symptoms. *E. coli* 0157, which is transmitted from ruminants, enters the food chain primarily by contaminated foods (Ball and Till, 1998). The significance of waterborne *E. coli* 0157 in New Zealand is not known (Ball and Till, 1998); however, *E. coli* 0157:H7 was detected in two of 531 faecal specimens in a survey of healthy dairy cattle (Buncic and Avery, 1997).

Campylobacter is a common food and water-borne pathogen and is the most frequently notified food-borne disease in New Zealand (Ministry of Health, 1998). There is good evidence to implicate contaminated drinking water in several Campylobacteriosis outbreaks in rural areas of New Zealand (Ball and Till, 1998).

3.3.2 Laboratory Analysis

Microbiological analyses were undertaken by the ESR Christchurch Public Health Laboratory, an accredited IANZ laboratory.

Each sample, or composite of five samples, of watercress was tested for the following bacteria:

- *E. coli* (presumptive coliforms & faecal coliforms were also measured as part of the method for measuring *E. coli*).
- *Campylobacter* species.

Water samples were tested for:

- *E. coli* (total coliforms were also measured as part of the method for measuring *E. coli*).
- *Campylobacter* species.

E. coli:

A 50 g portion of each watercress sample was weighed into a sterile Stomacher bag and homogenised with 100 ml of sterile 0.1% Peptone water. Serial dilutions from 10^{-1} to 10^{-3} were prepared and 5 tube MPNs were set up using Laurel sulphate broth. These were incubated at 35^{o}C for a total of 48 hours. Positive tubes were subcultured to EC Broth and incubated for 24 hours at 44.5^{o}C. Faecal coliforms from these tubes were further confirmed to *E. coli* using EMB agar plates, BGBB and Indole production (limits of detection, <1.8, >2400MPN/g).

Water samples from each watercress growing site were tested for total coliforms and *E. coli* using "Colilert" MPN (limits of detection, <1, >2400 MPN/100ml).

Campylobacter spp:

A 10-g portion of watercress was weighed into 90 ml of "Exeter Broth" and incubated at 42°C in a reduced oxygen atmosphere (produced by using a "Campy-Gen" envelope in a Gas-Pak jar). After 48 ± 2 hours incubation the broth was subcultured onto an "Exeter agar" plate and incubated as for the broth. Suspicious colonies isolated on this plate were confirmed as being *Campylobacter spp* by Gram stain, catalase and oxidase tests.

A 1 litre volume of water from each watercress growing site was filtered through a 45 μ membrane filter which was then placed in 90 ml of "Exeter Broth". This broth was then treated as for the watercress sample.

<u>3.3.2 Results - Watercress samples</u>

Watercress has been classified as a salad vegetable to enable comparison against the *Microbiological Reference Criteria for Food* (1995), section 5.25 (salads), since watercress is commonly eaten as a salad vegetable. The Reference Criteria do not include acceptable levels for *E. coli* but recommends limits for faecal coliforms of n=5, c=2, m=10^2, M=10^3 where:

n = number of samples.
c = maximum allowable number of defective samples.
m = an acceptable level and values above it are marginally acceptable (i.e. 2 samples) or unacceptable (i.e. >2 samples).
M = a level which separates marginally acceptable quality from defective quality. Any sample above M is unacceptable.

Overseas studies suggest that these criteria may be inappropriate for plant material. Bacteria such as *Klebsiella* and *Enterobacter* species which are normal inhabitants of plant material give a positive faecal coliform test, but their presence in foods of plant origin may not necessarily be associated with faecal contamination (Zhao *et al.*, 1996; Mpuchane and Gashe, 1995; Splittstoesser *et al.*, 1980). These authors recommend that the faecal coliform test be replaced by analysis for *E. coli* when testing plant derived material.

A large salad survey in England and Wales in which 2552 samples were examined, used limits for *E. coli* of n=5, c=2, m=10^2, M=10^3 (Little *et al.*, 1997). Therefore, in this study the faecal coliform limits recommended in the Reference Criteria for Foods (5.25) have been replaced by the same limits for *E. coli.*

For the first week all sites (except Waiwhetu) had five samples analysed and could be directly compared to the reference criteria. For subsequent weeks where samples were combined, the result was considered to be an average and was considered non-compliant if >M.

There are no criteria for *Campylobacter* in the *Microbiological Reference Criteria for Food* (5.25) relating to salads. However, the Microbiological Reference Criteria for *Campylobacter* in *Foods – Cooked Ready-To-Eat*, (5.8b) "*Some components not cooked in manufacturing* *process (e.g. sandwiches)*", is 0/10gm *Campylobacter*. This criteria suggests that any level of *Campylobacter* contamination is unacceptable in foods that will not be cooked prior to consumption. The presence/absence test for *Camplylobacter* was therefore considered sufficient to determine the safety of raw watercress for human consumption.

Full results are listed in Appendix 1. Results of microbiological analyses of the watercress samples (*E. coli*) compared to the Microbiological Reference Criteria for Food, Section 5.25 - Salads, are summarised in Table 1. *Campylobacter* results are summarised in Table 2.

Table 1: Microbiological results of watercress samples by site compared with Microbiological Reference Criteria (Salads)

Growing site	Week One (five samples)			Subsequent Four Weeks
	[1]Acceptable	[2]Marginally acceptable	[3]Non-complying	[4]Non-complying
Mazengarb Drain (Semi Urban)		Yes		
Owhiro Stream (Semi Urban)			Yes	2
Hulls Creek (Urban)		Yes		1
Black Stream (Urban)			Yes	1
Waiwhetu Stream (Urban)	N/A	N/A	N/A	
Ohariu Stream (Rural)		Yes		1
Papawai Stream (Rural)			Yes	1
Parkvale Stream (Rural)		Yes		
Manaia Drain (Rural)		Yes		
Opaki Stream (Rural)		Yes		
Golf Course Stream (Bush)			Yes	

[1]Acceptable: *E. coli* count first week (5 samples < 100).
[2]Marginally acceptable: *E. coli* count first week (2 samples >100 and <1000).
[3]Non-complying: *E. coli* count first week (3 or more samples >100 or any sample >1000).
[4]Non-complying: *E. coli* count subsequent 4 weeks (>1000, composite sample).

For the first week, none of the sites met the acceptable reference criteria for salads. Six sites were marginally acceptable and four sites were unacceptable. For the remaining four weeks, the samples failed the criteria on six occasions at five sites. There didn't appear to be any difference between catchment types. For example, the Golf Course Stream, Black Stream, Owhiro Stream and Papawai Stream were all classified non-complying in the first week.

It is also difficult to draw conclusions, or determine differences, between catchments as the variability between watercress samples at each site in the first week of sampling was significant. Figure 14 shows the variability in *E. coli* counts at three sites during the first week. For other sites the variation was also significant. For example, the results for Papawai Stream during the first week ranged from <1.8 to >2400 *E. coli* MPN/g.

Figure 14: Watercress Site *E. coli* Count Variability
5 samples - first week

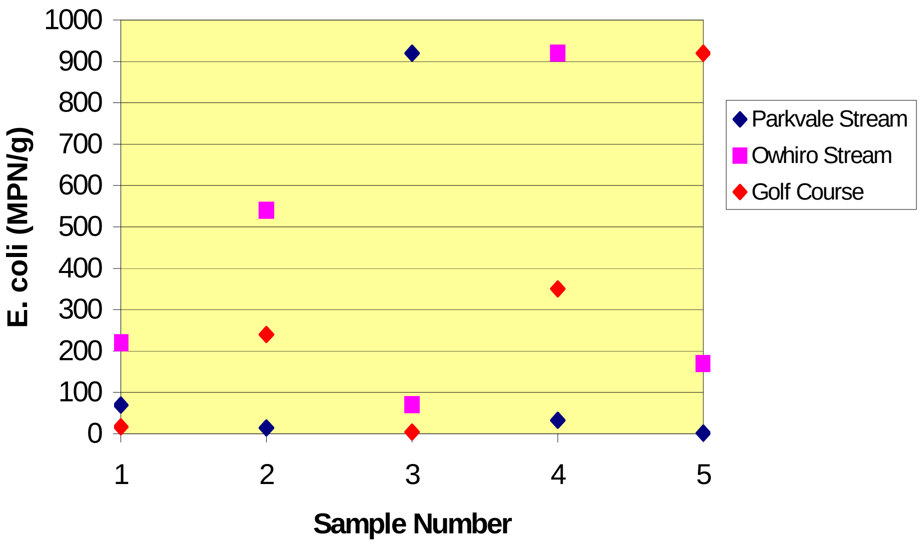


Table 2: *Campylobacter* Results

Analysis	*Campylobacter* Presence (watercress 1st week)	*Campylobacter* Presence (watercress, total)	*Campylobacter* Presence (water)
Mazengarb Drain (Semi Urban)	(3/5)	(4/9)	(4/5)
Owhiro Stream (Semi Urban)	(0/5)	(0/9)	(4/5)
Hulls Creek (Urban)	(0/5)	(0/9)	(5/5)
Black Stream (Urban)	(0/5)	(0/9)	(4/5)
Waiwhetu Stream (Urban)	(0/5)	(0/5)	(3/5)
Ohariu Stream (Rural)	(0/5)	(1/9)	(5/5)
Papawai Stream (Rural)	(0/5)	(1/9)	(5/5)
Parkvale Stream (Rural)	(0/5)	(0/9)	(5/5)
Manaia Drain (Rural)	(0/5)	(0/9)	(1/5)
Opaki Stream (Rural)	(2/5)	(3/9)	(5/5)
Golf Course Stream (Bush)	(0/5)	(1/9)	(3/5)

Campylobacter was present in 11% of the watercress samples. There did appear to be a difference between catchment types with *Campylobacter* being found in rural catchments (i.e. Opaki, Papawai, Ohariu), semi urban catchments (i.e. Mazengarb) and the Golf Course site, but not in samples from catchments with significant urban development. However, there was no clear relationship between *Campylobacter* presence in watercress and presence in growing waters. For example, the urban sites had *Campylobacter* present in the growing waters three times or more, but not in the watercress on any occasion.

An important reason as to why it is difficult to draw conclusions from the data is that the test for *Campylobacter* was only a presence/absence test and therefore gives no indication of the actual numbers in each sample i.e. whether heavily or lightly contaminated. Some degree of *Campylobacter* contamination would be expected in catchments subject to faecal contamination as *Campylobacter* is excreted by a wide variety of animals and birds (MfE, 1999).

Campylobacter was only present in 11% of the watercress samples compared to 80% of the water samples. Because of differences in sample size and physical properties between water and watercress, it is difficult to make a direct comparison. Factors affecting *Campylobacter* presence on watercress would include the surface area of the watercress sample and the resulting water volume retained, and retention and accumulation of bacteria on plant surfaces.

3.3.3 Results - Water samples

No applicable standard or guideline for acceptable levels of *E. coli* in growing waters could be found. Therefore, *E. coli* levels were grouped into classes to give an indication of the degree of faecal contamination at the growing sites.

No applicable standard or guideline for acceptable levels of *Campylobacter* in growing waters could be found. For the purposes of this study, the presence/absence test for *Campylobacter* in growing waters was considered appropriate as any level of *Campylobacter* in growing waters could potentially contaminant watercress, thus exceeding the Microbiological Reference Criteria for *Campylobacter* in foods (i.e. section 5.8b).

Gathering watercress exposes people to the risk of infection, for example, through cuts and abrasions, splashes to the eyes and mucus membranes, hand to mouth activity (e.g. eating and smoking) and exposure to aerosols. While recreational water health guidelines are used primarily for health risk assessment of swimming and other related activities such as diving, the Ministry for the Environment's *"1998 Bacteriological Water Quality Guidelines for Marine and Fresh Water"*, were used to assess potential health risks from water contact through harvesting watercress. The limit of a single sample greater than 273 *E. coli*/100ml (Alert/Amber Mode II) and a single sample greater than 410 *E. coli*/100ml (Action/Red Mode) were used for comparison.

Results of microbiological analyses of the 55 water samples are summarised on an individual site basis in Table 3. Full results are listed in Appendix 2. The Data was analysed for counts of *E. coli*. Counts were aggregated into the groups <100, <1000, >1000, >2400 MPN/g.

Table 3: Microbiological Results of Water Samples Per Site

Analysis	*Campylobacter* Presence	*E. coli* $<10^2$ (MPN/g)	*E. coli* $<10^3$ (MPN/g)	*E. coli* $>10^3$ (MPN/g)	*E. coli* >2400 (MPN/g)
Mazengarb Drain (Semi Urban)	(4/5)			4	1
Owhiro Stream (Semi Urban)	(4/5)		2	3	
Hulls Creek (Urban)	(5/5)		2	1	2
Black Stream (Urban)	(4/5)		1	1	3
Waiwhetu Stream (Urban)	(3/5)		2	1	2
Ohariu Stream (Rural)	(5/5)		2	2	1
Papawai Stream (Rural)	(5/5)	1	3		1
Parkvale Stream (Rural)	(5/5)		3	1	1
Mania Road (Rural)	(1/5)	3	1		1
Opaki Stream (Rural)	(5/5)		3		2
Golf Course Stream (Bush)	(3/5)	2	3		

There didn't appear to be a relationship between the presence of *Campylobacter* and *the E. coli* count at the sites. For example, although the Golf Course site had the consistently lowest *E. coli* levels, *Campylobacter* was still present in 3 out of 5 samples. This is consistent with previous work, for example, it has previously been shown that there is not a good correlation between the number of *E. coli* detected and the presence of *Campylobacter* in surface waters (Savill *et al*., 1999).

Campylobacter was present on only 1 occasion at the Manaia Road site. This may be due to a number of variables including specific catchment characteristics e.g. low concentrations of livestock, minimal flow rates and water depth in the drain compared to the other sites in the study, and variability of *Campylobacter* concentrations/distribution in surface waters. At all the other sites *Campylobacter* was present in 3 or more of the samples. This suggests that *Campylobacter* is potentially present in all catchment types e.g. urban, rural and bush/scrub covered catchments.

The presence of *Campylobacter* in all catchment types would be expected as *Campylobacter* is excreted in the faeces of humans and a wide variety of animals (particularly by cattle at certain times of the year) and birds (poultry often being implicated) (MfE, 1999). The test for *Campylobacter* was also presence/absence and therefore gives no indication of the actual numbers in each sample i.e. whether heavily or lightly contaminated.

The urban streams e.g. Hulls Creek, Waiwhetu Stream and Black Stream, had a greater number of *E. coli* results above 1000 MPN/100mls than the predominantly rural streams. This may be due to the run off characteristics of sealed surfaces. In rural areas soil and plants may help retain microorganisms before they reach surface waters. However, there will be significant variation in *E. coli* levels due to such variables as rainfall and sunlight exposure.

The *E. coli* count did not exceed 1000 MPN/100mls on any occasion at the Golf Course site. This would most probably be due to the bush/scrub cover reducing overland flow, the absence of significant point source discharges and the fact that there are few animals in the catchment.

It is difficult to compare *E. coli* counts in water to watercress because in water it was measured in MPN/100ml while in watercress it was measured in MPN/g. Figure 15 illustrates the high degree of variability between *E. coli* water and watercress counts at Owhiro Stream. While in general at most sites the water *E. coli* count was higher than the watercress count this was not always the case. This variability could be due to a number of factors including the surface area of the watercress sample and the resulting water volume retained. Watercress may also retain residual microbiological contamination from previous contact with contaminated growing water.

Figure 15: Owhiro Stream Water vs. Watercress *E. coli* Counts

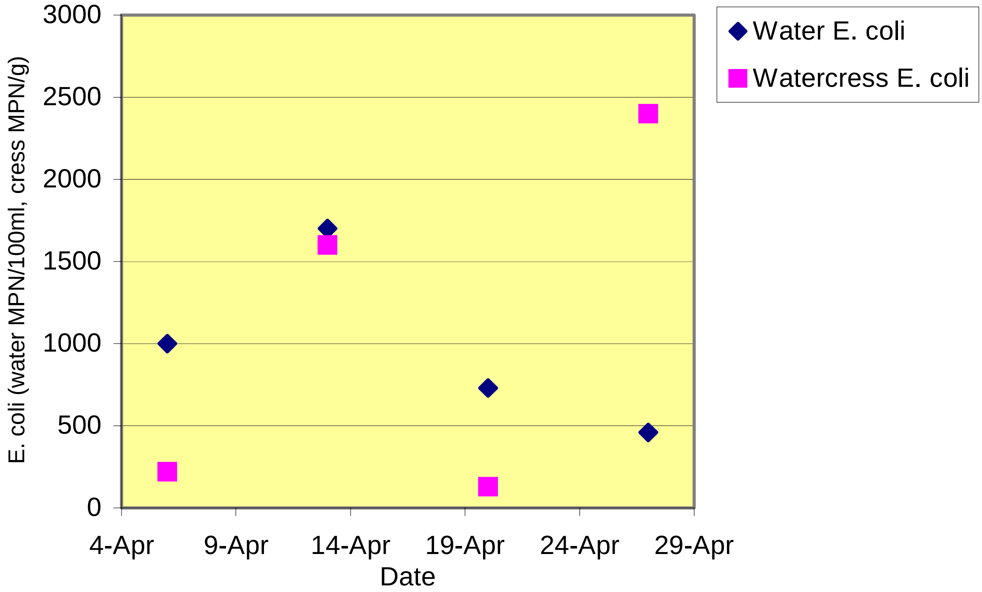


Table 4: ***E. coli*** **Results of Water Samples compared to the MfE 1998 Recreational Freshwater Quality Guidelines**

Analysis	<273 *E.coli*/100ml	>273 *E.coli*/100ml (alert/amber mode II)	>410 *E.coli*/100ml (action/red mode)
Mazengarb Drain (Semi Urban)			5
Owhiro Stream (Semi Urban)			5
Hulls Creek (Urban)	1		4
Black Stream (Urban)	1		4
Waiwhetu Stream (Urban)		1	4
Ohariu Stream (Rural)	1		4
Papawai Stream (Rural)	3	1	1
Parkvale Stream (Rural)	1	1	3
Manaia Drain (Rural)	3		2
Opaki Stream (Rural)	1		4
Golf Course Stream (Bush)	5		0

All the sites, with the exception of the golf course site, exceeded the action/red mode on one or more occasions. The urban streams, in general, exceeded the action/red mode more often than the rural streams, which were more variable with a greater number of measurements under alert/amber mode II. The Golf Course Stream was under the alert/amber mode II on all occasions. The results demonstrate that there is a potential health risk from water contact when collecting watercress from surface waters in most catchment types. Bush/gorse covered catchments may potentially pose less of a health risk.

3.4 Heavy Metal Analysis

3.4.1 Summary

Watercress concentrations of a range of heavy metals were measured (i.e. arsenic, chromium. lead, cadmium, nickel, copper, zinc and mercury), to give an indication of potential contamination. Studies in the lower reaches of Waiwhetu Stream have confirmed that the sediments are strongly enriched with lead and zinc, and moderately enriched with cadmium, chromium, nickel, copper, and arsenic (Aitken, 1998). Five combined watercress samples in total were tested for heavy metals from each site for five weeks.

3.4.2 Nickel

The New Zealand Food Regulations (1984) do not set a limit for nickel in food. Nickel is present in small amounts in most foods between 0-6.5 ppm. Nickel is poorly absorbed from food or drink with the majority being excreted in the faeces (Hamilton *et al*., 1990). Nickel and many nickel compounds are considered carcinogens or probable human carcinogens (Micromedex, 2000).

3.4.3 Cadmium

The New Zealand Food Regulations (1984) set a limit for cadmium in “any food other than shell fish” of 1.0 ppm. Cadmium occurs naturally at low levels in the environment and is present at measurable levels in most foods (MOH, 2000). However, cadmium can have serious health effects, even at relatively low levels of exposure. The renal cortex appears to be the most sensitive target tissue in humans, resulting in chronic kidney failure. Osteomalacia (softening of the bones) is also seen (MOH, 2000).

3.4.4 Copper

The New Zealand Food Regulations (1984) set a limit specifically for copper of 30 ppm in “any other food except animal offal and tea”. Copper is an essential trace element; however, it is toxic at high concentrations. Chronic copper toxicity, known as Wilson’s disease, can result in damage to the kidneys, brain and other organs (Micromedex, 2000).

3.4.5 Zinc

The New Zealand Food Regulations (1984) set a limit specifically for zinc of 40 ppm in “any other food except meat and shellfish”. Zinc is an essential trace element; however, it is toxic at high concentrations. Zinc doses two to three times the RDA lower the body’s copper content, an effect that, in animals, leads to degeneration of the heart muscle. Higher doses affect cholesterol metabolism, alter lipoprotein levels, and appear to accelerate the development of atherosclerosis (Hamilton *et al*., 1990).

3.4.6 Chromium

The New Zealand Food Regulations (1984) do not set a limit specifically for chromium. However, chronic exposure may cause necrosis of the kidneys and liver damage (Micromedex, 2000). Hexavalent chromium is about 10-100 times more toxic than the trivalent form and has been classed as a carcinogen. Total chromium was measured for this study.

3.4.7 Arsenic

The New Zealand Food Regulations (1984) set a limit for arsenic in "any other food" of 2.0 ppm. Arsenic occurs in foods in organic and inorganic forms. The inorganic forms are significantly more toxic than the organic forms. Inorganic arsenic can cause a range of serious acute and chronic health effects, including skin disorders, gastrointestinal complaints heart problems, peripheral vascular disorders and, both central and peripheral neurological damage (MOH, 2000)

3.4.8 Lead

The New Zealand Food Regulations (1984) set a limit specifically for lead of 2.0 ppm in "any other food except tea". Lead interferes with many of the body's systems, particularly the vulnerable tissues of the nervous system, kidney, and bone marrow. The foetus, infants and children are particularly at risk because the body absorbs lead most efficiently during times of rapid growth. (Hamilton *et al*., 1990).

3.4.9 Mercury

The New Zealand Food Regulations (1984) set a limit specifically for mercury of 0.03 ppm in "any other food except feral pigmeat". Mercury may be present in food in two different forms, organic and inorganic mercury. Organic mercury is significantly more toxic than inorganic mercury and more easily absorbed (MOH, 2000). The mercury analyses for this project measured total mercury. Organic mercury is a serious cumulative toxin that can cause severe disruption of the developing central nervous system, resulting in retarded mental and physical development. The foetus and infants are much more sensitive than adults, and are therefore at particular risk. (MOH, 2000).

3.4.10 Laboratory Analysis

Heavy metal analyses were undertaken by Agriquality in Gracefield, Lower Hutt, an accredited IANZ laboratory.

All watercress samples were rinsed with distilled water and any excess water allowed to drain. The samples were then blended to a "soup like" mixture before they were sub-sampled for analyses. A known weight of each sample was digested with concentrated nitric acid sitting in a boiling water bath for 1 hour then made to volume. The content of metals was determined by analysis using ICP-MS operating under standard procedures for this instrument.

Each batch of watercress samples included two samples of a certified reference material, a duplicate from one of the samples and two spiked control samples (low and high levels). Acceptable quality control results were obtained from all batches.

Table 5: Heavy Metal Limits of detection:

Lead	<0.01 (ppm)
Cadmium	<0.01 (ppm)
Zinc	<0.1 (ppm)
Mercury (Total)	<0.02 (ppm)
Arsenic (Total)	<0.25 (ppm)
Copper	<0.05 (ppm)
Nickel	<0.05 (ppm)
Chromium	<0.25 (ppm)

3.4.11 Heavy Metals Results

The sample results were compared to the New Zealand Food Regulations (1984) permissible proportion levels (in ppm) for selected heavy metals. There are no regulation levels for chromium and nickel.

Table 6: Heavy Metal Results

	Nickel	**Cadmium**	**Copper**	**Zinc**	**Chromium**	**Arsenic**	**Lead**	**Mercury**
	(ppm)	**(ppm)**	**(ppm)**	**(ppm)**	**(ppm)**	**(ppm)**	**(ppm)**	**(ppm)**
Permissible levels Food Regulations (1984)		1ppm	30ppm	40ppm		2ppm	2ppm	0.03ppm
Black Stream mean	0.13	0.01	0.82	17.00	0.28	0.25	0.11	0.02
median	0.11	0.01	0.76	13.00	0.25	0.25	0.10	0.02
maximum	0.34	0.01	1.20	26.00	0.40	0.25	0.25	0.02
Golf Course mean	0.24	0.01	0.39	4.70	0.25	0.25	0.02	0.02
median	0.21	0.01	0.41	4.20	0.25	0.25	0.02	0.02
maximum	0.31	0.02	0.49	5.90	0.25	0.25	0.02	0.02
Hulls Creek mean	0.12	0.01	1.01	24.00	0.38	0.25	0.52	0.02
median	0.13	0.01	0.93	18.00	0.44	0.25	0.69	0.02
maximum	0.25	0.02	1.90	**50.00**	0.52	0.27	1.00	0.02
Manaia Road mean	0.07	0.01	0.42	6.50	0.25	0.25	0.02	0.02
median	0.05	0.01	0.44	6.60	0.25	0.25	0.01	0.02
maximum	0.12	0.01	0.56	8.00	0.25	0.25	0.03	0.02
Mazengarb Drain mean	0.06	0.01	0.51	5.10	0.25	0.25	0.07	0.02
median	0.05	0.01	0.48	5.20	0.25	0.25	0.06	0.02
maximum	0.07	0.01	0.67	6.70	0.25	0.25	0.15	0.02

Ohariu Stream mean	0.05	0.01	0.38	2.96	0.25	0.25	0.02	0.02
median	0.05	0.01	0.38	3.10	0.25	0.25	0.02	0.02
maximum	0.05	0.01	0.41	3.50	0.25	0.25	0.03	0.02
Owhiro Stream mean	0.05	0.01	0.76	9.74	0.25	0.25	0.26	0.02
median	0.05	0.01	0.75	9.40	0.25	0.25	0.25	0.02
maximum	0.06	0.01	0.88	13.00	0.25	0.25	0.47	0.02
Papawai Stream mean	0.06	0.01	0.48	3.98	0.25	0.25	0.01	0.02
median	0.06	0.01	0.42	4.10	0.25	0.25	0.01	0.02
maximum	0.08	0.01	0.77	5.00	0.25	0.25	0.02	0.02
Parkvale Stream mean	0.05	0.01	0.32	2.46	0.25	0.25	0.01	0.02
median	0.05	0.01	0.29	2.20	0.25	0.25	0.01	0.02
maximum	0.05	0.01	0.50	3.40	0.25	0.25	0.01	0.02
Waiwhetu Stream mean	0.07	0.02	0.84	11.2	0.25	0.25	0.23	0.02
median	0.05	0.01	0.79	11.00	0.25	0.25	0.22	0.02
maximum	0.10	0.05	1.40	17.00	0.26	0.25	0.33	0.02
Opaki Stream mean	0.05	0.01	0.32	2.88	0.25	0.25	0.02	0.02
median	0.05	0.01	0.28	2.70	0.25	0.25	0.02	0.02
maximum	0.05	0.01	0.44	3.50	0.25	0.25	0.03	0.02

In this study all heavy metal concentrations (except for one zinc result) were well below New Zealand Food Regulation limits. The highest result measured in this project was 50 ppm for zinc in Hulls Creek which just exceeds the limit of 40 ppm.

The highest concentrations for other measured metals were as follows: lead (1.0 ppm (limit 2ppm)), arsenic (0.27 ppm (limit 2 ppm)), chromium (0.52 ppm), copper (1.90 ppm (limit 30ppm)), cadmium (0.05 ppm (limit 1 ppm)), nickel (0.34 ppm). No mercury was detected in any of the samples. Given that these values were well below the New Zealand Food Regulation limits, health risks from consumption of watercress containing these heavy metals at the measured levels is considered minimal.

Despite the low concentrations, there were trends in the concentrations of certain heavy metals, which may reflect the nature of the catchment. Watercress samples from the semi-urban Owhiro Stream and urban Black Stream, Hulls Creek, and Waiwhetu Stream all had higher levels of zinc and lead then other sites which possibly reflects increased urban runoff.

Samples from Hulls Creek at different sampling dates also showed elevated levels of zinc, chromium and lead when compared against samples from other sites. This mixture of metals, all at elevated levels, suggest potential historical/current industrial contamination of Hulls Creek. Samples from Black Stream also appear to have a somewhat elevated metal content with a similar distribution to those from Hulls Creek.

4. DISCUSSION

4.1 Microbiological

4.1.1 Campylobacter

Campylobacter was isolated in 44 (80%) of the water samples and 10 (11%) of the watercress samples. All of the sites had *Campylobacter* present in the growing waters and except for Manaia Road Drain, *Campylobacter* was present in the growing waters at each site on three or more occasions. *Campylobacter* appeared to be uniformly present in waters in rural and semi-urban and urban catchments. Although the Golf Course site had the consistently lowest *E. coli* levels, *Campylobacter* was still present in three out of five samples. All sites were considered unsuitable for gathering watercress for consumption (unless boiled) due to the presence of *Camplyobacter* in the growing waters.

The presence of *Campylobacter* on 11% of the watercress samples is of public health significance. *Campylobacter* has been shown to survive long enough on Ready To Eat (RTE) produce to cause infection (Beuchat, 1996). The *Microbiological Reference Criteria for Food* (*5.8a*) for cooked ready-to-eat foods is 0/10gm *Campylobacter*.

As *Campylobacter* was found in growing waters at all the sites it appeared to have a wide distribution across all catchment types. This would be expected as *Campylobacter* is excreted in the faeces of a wide range of mammals (e.g. humans, sheep, cattle, goats), and birds (poultry often being implicated) (MfE, 1999).

For example, in a survey conducted by Gill and Harris (1982) in New Zealand, *Campylobacter fetus subsp. Jejuni* were detected in the faeces of calves and sheep. Calves were more likely to be infected with *Campylobacter* than adult cattle with 50% of the 50 faeces samples from unweaned calves containing between 100 - 5,000 (mean $1.6x10^3$) cfu *Campylobacter*/g faeces whereas the organism was not isolated from any of the 75 faecal specimens from adult cattle. Conversely, adult sheep were infected more than lambs with respective isolation rates of 14% (10/71) and 2.4% (1/42). Thermophilic *Campylobacter* species were recovered from 22% of 273 dairy cows, with the highest incidence in autumn (Fakir, 1986).

4.1.2 *E. coli*

All the watercress and growing water samples at each site showed variable levels of *E. coli* contamination. Although there are no applicable standards to determine safety of growing waters, most sites showed high levels of *E. coli* in the water with only the Golf course site not exceeding 10^3 (MPN/100ml) on any occasion. High *E. coli* counts would be expected in slow moving streams where watercress grows in sufficient quantities for collection.

Although in this study the Microbiological Reference Criteria (salads) are only used as a comparison and not as a measure of suitability for safe consumption, of the ten sites (excluding Waiwhetu Stream), none of the watercress samples were acceptable on all occasions, six sites were non-complying and four were marginally acceptable. Little *et al*., (1997) describes the results of a large salad survey in England and Wales in which 2552 samples were examined. The survey used the same limits that were used in this study (i.e. *E. coli*, n=5, c=2, m=10^2, M=10^3). The survey found only 1% of salads had *E. coli* counts of 10^2 cfu or more per g. In this study, 46% of watercress samples had *E. coli* counts of 10^2 cfu or more per g. This indicates significant faecal contamination of watercress from contaminated growing waters.

While it is not possible from the tests used to know whether *E. coli* are of human, animal or avian origin, all of these species can act as carriers of micro-organisms that can cause human disease. If faecal contamination (as indicated by *E. coli* results) is present on watercress then human pathogens may also be present posing a risk of infection. The presence of any enteric pathogen on Ready To Eat (RTE) vegetables is a major food safety concern. Pathogens such as *Salmonella*, *Campylobacter*, *Giardia*, *Cryptosporidium* and specific human viruses are potential contaminants of watercress especially in rural situations, where livestock can carry and excrete these pathogens.

Giardia and *Cryptosporidium* are protozoan pathogens widespread in the environment and New Zealand surface waters (Ball and Till, 1998). Humans and a wide range of domestic and feral animals can carry these parasites and when infected can excrete high numbers of cysts/oocysts. Water transmission of infection to humans is well recognised (Ball and Till, 1998). However, *E. coli* concentrations do not correlate well with the more persistent protozoan pathogens and therefore have limited use for predicting health risk from these groups of organisms, especially when faecal indicator concentrations are low (Ball & Till, 1998b). The large numbers of protozoan parasites in New Zealand surface waters may increase this problem (Ball & Till, 1998b).

Viruses such as Adenoviruses, Enteroviruses, Rotaviruses, and Norwalk viruses can cause a wide range of diseases including respiratory infections, skin rashes, conjunctivitis and gastro-enteritis. They can be transmitted via ingestion or contact with polluted water (Ball and Till, 1998).

4.2 Potential Health Risks from Microbiological Contamination of Watercress

There is no formal data available on the incidence of microbial hazards from watercress grown in New Zealand. Therefore, it is difficult to assess the impact of watercress on enteric infection rates in the human population. However, from the national outbreak information that has been reviewed to date (ESR Reports, 1997-2000), there has been no known enteric disease outbreaks linked to the consumption of watercress in this country.

Many overseas outbreaks of human gastro-enteritis (e.g. *Shigella* (USA)), have been linked to the consumption of contaminated fresh vegetables (Beuchat, 1996) and watercress is included in a list of salad vegetables which have caused extensive outbreaks of Salmonellosis (ICMSF, 1998).

There is considerable international information available on the microbiological contamination of salad vegetables. However, studies of general salad vegetables must be interpreted with caution in relation to watercress, as this study indicates that due to the nature of its growing environment it should be considered a more at 'risk food' than salad vegetables generally.

Studies have shown that thorough washing and treatment of produce with chlorinated water can reduce the populations of pathogenic and microorganisms on fresh produce but it cannot eliminate them. Beuchat (1997) suggests that the reduction of risk for human illness associated with raw produce can be better achieved through controlling points of potential contamination e.g. during harvesting, processing and distribution.

The significant variability of *E. coli* counts in watercress found in this study would make it difficult to assess levels of microbiological contamination to determine suitability of raw watercress for human consumption. In regard to raw watercress, controls on the quality of the growing waters would be the most appropriate method of reducing health risks for consumers.

The US FDA "*Guide to Minimise Microbial Food Safety Hazards for Fresh Fruits and Vegetables*" (FDA/CFSAN, 1998) states that "*whenever water comes into contact with fresh produce, its source and quality dictate the potential for pathogen contamination. If pathogens survive on the produce, they may cause foodborne illness*". The Food and Drug Association (1999) suggested that the best means of monitoring sprout production was to test growing water. Testing for pathogens as an indicator of watercress or water quality is not cost effective due to the potential number of pathogens which could be present. It is more appropriate to use a suitable faecal indicator organism e.g. *E.coli*, as an indicator of the growing water quality (ICMSF, 1998; Vanderzant and Splittstoesser, 1992). However, as *E. coli* concentrations may not correlate well with protozoan pathogens in growing waters, strict quality controls on source waters/growing conditions would be required in addition to a suitably strict *E. coli* standard (e.g. New Zealand Drinking Water Quality Guidelines (2000) standard of less than 1 *E. coli*/100mls), to ensure minimal potential for watercress contamination.

A report on the hygienic production of watercress (Public Health Laboratory Service, 1961) recommended that watercress grown wild in the United Kingdom should not be sold for human consumption. The microbiological results from this Wellington study support this recommendation.

4.3 Fascioliasis

Liver flukes (*Fasciola hepatica*) are parasitic in the bile ducts of mammals. Cercariae encyst on aquatic vegetation (e.g. watercress) and the cysts (metacercariae) are then swallowed by the final host (humans, sheep, goats, cattle) while feeding (Dalton, 1998). In humans the typical symptoms resulting from liver fluke infestation are anaemia, inflammation and haemorrhage of the intestinal tract and damage to hepatic tissue (WHO, 1998). Wild watercress is reported as the main source of infection in Europe where there is a high rate of endemic fascioliasis in domestic animals (Dalton, 1998).

Over the last 30 years or so, liver fluke infection of livestock has become considerably more widespread in New Zealand, largely in association with the spread of the exotic snail host, *Lymnaea columella* (Mitchell, 1995). A 1984 nation-wide sheep survey showed the prevalence of liver fluke infection in sheep in the North Island to be 7.5% in the North Island and 1.07% in the South Island. However, the levels varied considerably between regions e.g. South Auckland (12.6%), Taranaki (16.9%), Nelson (18%) and Westland (29%) (Charleston *et al*., 1990). A 1984 survey of cattle livers slaughtered over a six-month period in the Moerewa works in Northland showed a prevalence of about 10% (Kearns, 1987). Movement of infected stock into liverfluke free areas where intermediate snail hosts live is how the infestation spreads from area to area (Mitchell, 1995). Further spread can be expected as more properties with suitable resident snail populations have infected stock grazed on them.

Although there have been no documented cases of fascioliasis in New Zealand related to consumption of watercress gathered from New Zealand waterways, there is definite potential for cases of fascioliasis in New Zealand through consumption of raw watercress. The risk of fascioliasis would be minimised by ensuring that raw watercress for human consumption is only grown under strictly controlled conditions.

4.4 Potential health risks due to water contact while gathering watercress

At many growing locations, gathering watercress exposes people to contact with contaminated water. Gathering watercress exposes people to the risk of infection, for example, through cuts and abrasions, splashes to the eyes and mucus membranes, hand to mouth activity (e.g. eating and smoking) and exposure to aerosols. To give an indication of potential health risks through water exposure, the *E. coli* levels in the growing waters at each site were compared to the 1998 Ministry for the Environment '*Bacteriological Water Quality Guidelines for Fresh Water*'.

As the guidelines were developed on the basis of only a few international studies relating bacteriological indicators to illness in the general public, their suitability for use as fresh water guidelines in New Zealand requires further evaluation (MfE, 1998). Nonetheless, the guidelines are an appropriate reference standard to assess potential health risks. The Ministry for the Environment and the Ministry of Health are undertaking a Fresh Water Microbiological Research Programme that aims to develop more robust fresh water guidelines, but conclusions and recommendations from the study are not yet available.

The measured *E. coli* levels were compared against the single sample exceedence limits in the guidelines. Alert Mode II and Action/Red Modes are triggered when a single bacteriological sample exceeds a predetermined level. Under Alert Mode II, the guidelines recommend that a sanitary survey should be undertaken to identify the sources of contamination. Under the Action Mode the guidelines recommend that the local authority and health authorities should warn the public through the media that the water body is unsafe and arrange for the local authority to erect signs warning the public of a health danger (MfE, 1998).

All of the sites except for the golf course site exceeded the action/red mode level on one or more occasions. Persons gathering watercress from these or similar sites could potentially be at risk of illness from contact (e.g. hand to mouth contact, direct skin contact, aerosols) with contaminated water. Water conforming to the guideline values may still pose a potential health risk to high-risk user groups such as the very young, the elderly and those with impaired immune systems (MfE, 1998). Until recently it was believed that gastro-enteritis was the main health effect from contact with polluted water, but now it is becoming clear that respiratory effects also occur, and may be more prevalent than gastro-enteritis (MfE, 1998). Skin and eye/ear infections are also common health effects (MfE, 1998).

4.5 Potential Health Risks from Heavy Metal Contamination of Watercress

Measured heavy metal concentrations in watercress at all the sites, with the exception of one zinc result, were within applicable food regulations and thus were not considered a health risk. However, some of the site results indicate that there is the potential for contamination in excess of food regulation guidelines in watercress growing at sites subject to current or historical heavy metal contamination. A study of watercress collected from three sites in Hong Kong showed that the levels of iron, zinc and magnesium were higher in watercress collected from heavy metal contaminated sites (iron ore tailings) than cultivated and uncultivated land areas (Wong, 1995).

There are certain waterways in New Zealand where, due to the natural presence of heavy metals such as arsenic, it is not safe to collect watercress e.g. Waikato River System. Studies undertaken in areas of the central North Island, have revealed high levels of arsenic in the watercress, probably due to the geothermal activity in the area and the impact of geothermal power stations (Robinson *et al.,* 1995; Deely, 1997). For example, watercress collected in late February 1994 from the Waikato River near a geothermal power station (Ohaaki near Broadlands) averaged 412 ug/g (dry weight) arsenic (Robinson *et al.*, 1995). The amount of arsenic found in these samples of watercress (even when the concentration is converted to a fresh-weight basis) is well in excess of the WHO limit for arsenic in foodstuffs (2ug/g fresh weight). A sample taken as a control from the Tiritea stream near Massey University had less than 0.001 ug/g d.w. arsenic. The watercress in the Waikato River appears to behave similarly to other aquatic or semi-aquatic macrophytes in that it accumulates arsenic (Robinson *et al.,* 1995).

A companion 1994 Massey University study (Robinson *et al.,* 1996) looked at the relationship between arsenic levels in growing waters and the corresponding level in watercress. It was found that plants grown in arsenic solutions of 0.4 ug/ml or greater, exceeded 2 ug/g, (the WHO limit for arsenic in foodstuffs). The report recommended that humans should not consume watercress growing in water that at any time has 0.05 ug/ml arsenic. Robinson *et al* (1995) showed that arsenic concentrations in geothermal waters that flow into the Waikato River and other lakes of the Taupo Volcanic Zone contain as much as 6 ug/ml arsenic, thus exceeding the recommended limit.

The fact that this Wellington study did not reveal heavy metal levels of concern in the watercress does not mean that watercress grown at other sites in New Zealand would be safe to consume. Overseas and NZ studies have shown the ability of watercress to bio-accumulate heavy metals to levels which could pose health risks. A specific site risk assessment would need to be undertaken to determine potential heavy metal contamination before the safety of consuming watercress gathered from a site could be assessed.

5. CONCLUSIONS

All of the sites showed variable but significant levels of *E. coli* in both the watercress and water samples and therefore the potential for enteric waterborne human pathogens to be present. *Campylobacter* was detected in the growing waters in at least one sample from each of the sites (80% of the samples) and in 11% of the watercress samples.

The results demonstrate that urban, semi-urban, rural and bush/scrub catchments are subject to variable levels of faecal contamination which may include pathogens such as *Campylobacter*. The source of this contamination is likely to be both point and non-point discharges. Watercress gathered from surface waters in these catchment types is at significant risk of contamination by enteric pathogens and therefore, persons consuming raw watercress gathered from these catchment types are at potential risk of gastrointestinal illnesses. There is also the risk of cross-contamination of other foods during preparation. In rural areas there is also the potential for fascioliasis in persons consuming raw watercress due to contamination with *Fasciola hepatica* cysts.

The water sample results also showed levels of *E. coli* well above the alert/red mode action for freshwater recreational contact safety, with all of the sites, except for the Golf Course site, exceeding the action/red mode level on one or more occasions. Persons gathering watercress could potentially be at risk of illness through contact with faecally contaminated water.

Although the study only looked at surface waters in the Wellington Region, it is considered that the significance of the microbiological results can be applied nationally. Catchments in other areas of New Zealand would be exposed to similar sources of faecal contamination either from rural livestock, urban runoff or feral animals such as possums and goats. As *Campylobacter* was found in all of the catchments including the Golf Course Site, it is reasonable to assume that it would be widespread in other New Zealand catchments due to the number of animal species that carry and excrete *Campylobacter*.

As demonstrated by our study, the significant variability of *E. coli* counts in watercress makes it difficult to assess microbiological contamination to determine suitability of raw watercress for human consumption. In regard to raw watercress, controls on the quality of the growing waters would be the most appropriate method of reducing health risks for consumers. However, *E. coli* concentrations do not correlate well with protozoan pathogens such as *Cryptosporidium*. Therefore, using faecal indicators in growing waters alone to determine suitability for human consumption may not accurately represent the risk. Strict quality controls on source water/growing conditions in addition to applying a strict faecal indicator standard to the growing waters (e.g. New Zealand Drinking Water Quality Guidelines (2000) *E. coli* standard) would be required to ensure minimal potential for watercress contamination.

The heavy metal results did not show watercress contamination above the NZ Food Regulations limits at any of the sites, except for zinc on one occasion. However, there were higher levels of specific metals in a number of urban streams, relative to rural streams and the control site. In streams subject to industrial discharges or natural processes, watercress may bio-accumulate heavy metals to concentrations in excess of acceptable regulatory levels thereby potentially causing adverse health effects depending on the amount consumed. For example, there are areas, associated with geothermal activity, in the central North Island, where studies have revealed levels of arsenic in the watercress in excess of WHO guidelines.

6. RECOMMENDATIONS

1. Watercress harvested from any uncontrolled surface water source in New Zealand should not be consumed unless the watercress is thoroughly cooked in boiling water to destroy potential human pathogens.

2. Watercress should not be eaten raw unless it can be demonstrated that the growing environment is strictly controlled and effectively monitored to ensure the water source is of suitable standard e.g. controlled hydroponic cultivation. A suitably strict standard should be applied to the growing waters e.g. New Zealand Drinking Water Quality Guidelines (2000) *E. coli* standard of less than 1 *E. coli*/100mls, to monitor acceptable watercress microbiological quality.

3. Further research should be undertaken to assess the risk of contracting waterborne illnesses through contact with contaminated water when gathering watercress from uncontrolled surface waters.

4. The potential for fascioliasis in New Zealand from consumption of wild watercress should be the subject of a targeted study. This would involve further research on such factors as the geographic distribution of the host snails and the geographic distribution of *Fasciola hepatica* in livestock.

5. There are areas in New Zealand, specifically the central North Island, where studies have revealed high levels of arsenic in the watercress as a result of geothermal activity. In these areas it is recommended that watercress collected from local waterways is not consumed.

6. To minimise potential health risks, watercress should not be gathered for consumption from waterways subject to significant historical/current industrial discharges.

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APPENDIX 1: Results of Microbiological Analysis of Watercress

APPENDIX 2: Results of Microbiological Analysis of Water

APPENDIX 3: Results of Heavy Metal Analysis