Habitat determinants and predatory interactions of the endemic freshwater crayfish (Koura, *Paranephrops planifrons*) in the Lower North Island, New Zealand.

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Chapter 1:

Introduction:

Paranephrops planifrons and *P. zealandicus* collectively known as koura or New Zealand freshwater crayfish are endemic to New Zealand. *P. planifrons* are found in the North Island and the north and west of the South Island while *P. zealandicus* are found to the south-east of the South Island and on Stewart Island (Parkyn *et al*, 2002). A compilation of information from the National Institute of Water and Atmosphere (NIWA) New Zealand Freshwater Fish Database (NZFFD) carried out by McDowall (2005) shows that koura have been recorded throughout New Zealand.

Since European arrival in New Zealand large areas of the country have been deforested for agricultural purposes. This land clearance and intensification of land use has lead to declines in biodiversity and water quality compared with streams in less developed or more pristine catchments (Niyogi *et al*, 2007). Although reductions in diversity have been noted; abundance and biomass of some fish species has increased in pasture streams (Hicks & McCaughan, 1997), and koura have managed to maintain similar annual production between forested and agricultural streams even though densities in agricultural streams are generally lower (Parkyn *et al*, 2002). The similarity in annual production is brought about by higher temperatures in agricultural streams leading to faster growth rates compared to koura found in forested streams.

Although the above would seem to support the idea that land use change has not had detrimental effects on koura populations, it has been shown that koura in pasture streams are more likely to have reduced populations and take longer to recover after major floods (Parkyn & Collier, 2004). In the study done by Parkyn and Collier (2004) it took three years for one pasture stream to start to show recovery from a 1 in 28 year flood. Therefore the effects of pastoral land use on koura populations may not be obvious over short time scales such as other invertebrate community changes, but may need to be monitored over long time scales (Parkyn & Collier, 2004). Parkyn (2000) found that in pastoral streams cobbles and in-stream vegetation were important habitats while in streams with riparian vegetation tree roots, undercut banks, and large substrate were important. Usio and Townsend (2000) found *P. zealandicus* were negatively associated with trout presence and suspended sediment concentrations, and were positively associated with wood cover. Young of Year (YOY) koura also differed in their requirements where they were found to be positively associated with substrate which was larger in size. Adult koura were negatively associated with current velocity and positively associated with depth, leaf cover, and percentage sand (Usio & Townsend, 2000). Changing land use has the ability to change all of these aspects which are important for koura habitat.

The arrival of humans has extended beyond the changes in land use and resulted in the introduction of freshwater fish species such as the brown trout (*Salmo trutta*). Brown trout are present in the IUCN's list of 100 of the World's Worst Invasive Alien Species, being recorded at number 83 (Lowe *et al*, 2000). They were first introduced into New Zealand in 1867 (Olsson *et al*, 2006) and are now one of the few freshwater species in New Zealand which have full legal protection, with rules governing their capture. The introduction of brown trout has been suggested as one of the reasons for a decline in the abundance of many native fish species (Olsson *et al*, 2006). These reductions are thought to have occurred through predation and competition for food and space (Townsend & Crowl, 1991; McIntosh *et al*, 1992; McIntosh, 2000a). Koura have not been exempt from the effects of brown trout, with a negative association found between the presence of brown trout and the presence of koura (Olsson *et al*, 2006; Usio & Townsend 2000). This negative association is known to occur through direct effects such as predation (Gibbs, pers comm.) and indirectly through reduced feeding as shown in Chapter 3. Usio and Townsend (2000) found that above a barrier which prevented fish moving upstream koura were present, but below the barrier koura were either absent or found in very low densities.

Koura are known to be negatively associated with the presence of brown trout, as noted above. In other countries, however, some crayfish species have been shown to predate on fish species. *Orconectes virilis* have been found to predate on eggs and sac fry of lake trout (*Salvelinus namaycush*) and rainbow trout (*Oncorhynchus mykiss*) (Savino & Miller, 1991).

Frequently used methods for sampling koura presence/absence and population dynamics are electro-fishing at night (Usio and Townsend, 2000), electro-fishing during the day (Hicks, 2003), and visual inspection of the site including looking under rocks (Hicks, 2003). Rabeni *et al* (1997) evaluated direct observation at night, baited traps, quadrat sampling, handnetting at night, mark-recapture technique, estimating abundances using depletion technique and electro-fishing to see which gave the best population estimates of koura. Electro-fishing was found to be the most successful of these techniques (Rabeni *et al*, 1997). This method is useful for streams in which electro-fishing can be carried out such as open fast flowing streams, but is not practical for streams which are too small or densely covered in vegetation preventing the carrying and use of the required equipment.

This thesis looks at combining the factors discussed above into a set of field and controlled experiments. Therefore it has been separated into the following objectives:

1. Koura have been found throughout New Zealand and many habitat variables have been used to explain their distribution. In this chapter we aim to identify what habitat variables explain the distribution of koura in streams of the lower North Island.

2. The introduction of brown trout into New Zealand has had large ramifications for the functioning of freshwater environments within New Zealand. Brown trout are known to consume koura and we will attempt to quantify the amount and the size class that is taken. Crayfish species found overseas have also been recorded consuming juvenile fish and fish eggs, we attempt to see if the same also occurs with koura consuming either introduced or native fish species.

3. Brown trout are known to have a negative association with the presence of koura. This is thought to occur through direct consumption of koura, previous studies have shown that koura are not able to detect chemical cues emitted from brown trout. In this chapter we will attempt to look at the effect that brown trout have on the rate at which koura break

down leaf matter, this will be compared with the effect of a native predator, the long fin eel.

4. When estimating the population size of animals many different tools have been developed over the years. Tools for sampling koura populations all have some form of bias which tends to favour a certain demographic of the population. In this chapter we will attempt to use leaf packs and the rate of leaf breakdown as a surrogate for the density of koura found at a site.

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Habitat determinants of freshwater crayfish (koura, *Paranephrops planifrons*) in the Lower North Island, New Zealand.

Abstract:

A study in the Lower North Island located *Parenephrops planifrons* (koura) at 73 sites out of 104 sites visited (appendix 1). There was a significant difference in habitat variables between the sites which had koura present and those where they were absent. Examples of sites are shown in Appendix 3. Habitat variables important for classifying koura habitat included riparian cover, predators, winter equilibrium temperature and presence of in-stream habitat in the form of vegetation, litter cover and the stream sequence composition. Regression trees built could accurately describe the data but the kappa statistic was low.

Introduction:

Since European arrival in New Zealand land use has changed from a forested landscape to one modified for agricultural use. This land use change has frequently been blamed for detrimental effects on native fish communities, including the extinction of the southern grayling (*Prototroctes oxyrhynchus*) and the decline in galaxiid abundance (Hicks, & McCaughan, 1997; McDowall, 1990). However, the production and distribution of the New Zealand freshwater crayfish (koura) do not appear to have been affected (Parkyn, 2000; Parkyn, Collier, & Hicks, 2002). Two described species of koura are currently endemic to New Zealand. *Paranephrops zealandicus* (White, 1847) are restricted to the south-east of the South Island and Stewart Island, while *Paranephrops planifrons* (White, 1842) are found in the North Island and the north and west of the South Island (Parkyn *et al*, 2002).

Parkyn *et al*, (2002) found densities of koura in native streams were higher than those in agricultural streams. However, the increased temperature and greater reliance on invertebrates for food in agricultural streams lead to similar annual production in both types of streams (Parkyn, 2000; Parkyn *et al*, 2002). This increased production has occurred because koura in agricultural streams grow faster, even though this faster growth rate results in decreased longevity.

Usio and Townsend, (2000) found *Paranephrops zealandicus* Young of Year (YOY) density in a forested catchment was negatively associated with trout presence and suspended solid concentrations, while being positively associated with wood cover and coarse substrates. Adult koura were found to be negatively associated with trout presence, suspended solid concentrations and, current velocity and positively associated with wood cover, depth, leaf cover and percentage sand.

Similar studies have been carried out on *Paranephrops planifrons* in the North Island, focusing on the Waikato region. Most of these studies however, have focused on the difference between agricultural and native forested streams, looking at production levels, the use of different habitats within the stream and their ability to withstand disturbance (Parkyn & Collier, 2004; Parkyn *et al*, 2002; Parkyn, Collier, & Hicks, 2001; Hicks & McCaughan, 1997).

In this study I examined the distribution and biotic and abiotic habitat variables that determine where koura occur in the lower North Island.

Methods:

Field methods:

Study sites:

The study region consisted of the lower half of the North Island, New Zealand. Field sites were chosen on the basis that they were representative of the different land use patterns which occur in the lower North Island and also sites which allowed ease of access both during the day-time and for night-time surveys. New Zealand topographical maps were consulted to find appropriate sites, although no decision was made about site selection until the site was visited. GPS coordinates using a Garmin Etrex were taken at each of the sites, giving New Zealand grid references and altitude.

Day habitat survey:

In each 100m study reach water clarity was measured using a black disk. At five equidistant points in riffles across the study reach a small stone was taken for periphyton analysis. Five quorer sediment samples were taken in riffles to estimate the abundance of sediment covering the stream bed. A background sample of the stream water was taken first, to allow for correction of background suspended sediment in the water column. Quorer samples were taken by driving a bottomless bucket into the substrate as far as possible. Measurements of the bucket depth were recorded (this included the water and substrate depth) and then the water depth was measured (from the top of the substrate to the water level). The substrate was then disturbed for 30 seconds and 500mls of water taken from inside the quorer. Quorer and perihyton samples were kept on ice for transport to the laboratory, where they were frozen for later analysis.

The weather conditions on the day were recorded along with evidence of livestock access. Visual estimates (classified as rare, common, or abundant) of diatoms, macropytes, bryophytes, filamentous and mat algae were made. Riparian land use was recorded for, the first twenty metres from the stream, and then for the surrounding land use after these twenty metres. The stream bank composition (rocky, soil) was also recorded as well as any evidence of erosion. A visual estimate was made of the stream make-up, with the percentage of pool, riffle, run and shutes recorded. A qualitative habitat analysis was made using forms from the Auckland Regional Council. Conductivity and temperature were measured using an Orion handheld meter, and pH was measured using an Orion handheld meter. Water velocity was measured by releasing fluorescein dye and timing how long it took to travel 50m. Five measurements of depth and wetted width were made at equidistant points along the 100m stream reach. Slope was measured as fall across five to 20m using a piece of string and builders level. Substrate composition was assessed using the Wolman walk (Wolman, 1954) and picking up 75 particles and fitting them into size categories. The retention of coarse organic matter in the stream was measured by releasing 20 surrogate leaves (rosella red 225 gsm card, 105mm x 15mm) into the stream. Leaves were left for 30 minutes and the distance travelled recorded. Any leaves that travelled greater than 100m were recorded as having travelled 100m and any missing leaves were excluded. Duncan stability index measurements were taken at three equidistant points along the 100m stretch of the stream, as outlined in Duncan *et al* (1999).

Night time survey:

Koura were sampled by spot lighting the previously sampled 100m stretch. All koura that were sighted were counted, and the oribit carapace length (OCL) measured. The type of substrate on which the koura were found was also noted. All fish (native or exotic) observed were caught, identified, and measured before being released back into the stream. The night time temperature of the stream and the time of the visit were recorded.

Lab methods:

Periphyton:

Photosynthetic pigments were extracted from stones by sub-merging them in 90% acetone for 24 h at 5 ^{0}C in darkness. Absorbance was read at 750, 665 and 664 nm on a Varian Cary 50 Conc. UV-Visible spectrophotometer^{TM} (Varian Australia Pty Ltd, Mulgrave, Australia) before and after 0.1 M HCl was added. We calculated the amount of chlorophyll-*a* (μg cm^{-2}) on each cobble as described by Steinman & Lamberti (1996), and corrected for stone surface area (upper half) calculated using length, width and depth of each cobble following Graham, McCaughan & McKee (1988).

Sediment cover:

Sediment samples were filtered through pre-ashed Whatman CFC 40 μm filter papers. A suspension of 300ml was filtered, samples were dried for 24 hours at 105^{0}C to constant weight. They were then ashed at 450^{0}C for four hours, and re-weighed.

FWENZ data:

Data was also taken for each of the sites from the Fresh Water Environmental New Zealand variables and included in some of the data analysis.

Statistics:

Presence/absence of koura was mapped using ARCview GIS 3.3. Data was analysed using WEKA 3.4 to form regression trees. The data was separated into two different groupings; the first grouping contained only the variables that had been measured when we visited each of the sites and the second grouping contained these measured variables as well as the data taken from the FWENZ database.

Results:

A total of 104 sites were sampled for koura (Fig 1), with koura being present at 73 sites and absent from 31 sites (Fig 1). The number of koura seen at a site varied from 0 to 165.

An ANOSIM analysis showed that there was a significant difference between the sites which had koura present and the sites which lacked koura. The regression trees below show which of the habitat variables were important in the difference between koura present and absent sites.

The regression tree containing only site variables (Fig 2) identified 8 variables as important to explaining the distribution of koura. These variables being the percentage of riparian cover made up of planted forest, the percentage of stream bed made up of shutes, conductivity, the percentage of vegetation 20m back from the stream edge made up of native scrub, the presence of small trout, the percentage of litter cover on the streambed, the percentage of stream bed made up of pools and the percentage of undercut banks in the study reach. Using a cross validation of the 104 sites, 65 of these sites were correctly classified and 39 incorrectly classified, with a kappa statistic of 0.0602. If the data is used as a training set it correctly classified 93 and incorrectly classified 11 of the sites, with a kappa statistic of 0.74. Both the cross validation and training set method give identical looking regression trees.

The regression tree which contained both site variables and FWENZ data identified 9 variables as being important to explain the data (Fig 3). These 9 variables were current wintertime equilibrium temperature, the percentage of riparian cover made up of planted forest, the abundance of macrophytes, the abundance of bryophytes, conductivity, the presence of eels, the presence of small trout, the percentage of vegetation 20m back from the stream edge made up of scrub and percentage of riparian area in a land cover database (LCDB) category. Using a cross validation of 10% of the data, the tree correctly classifies 61 of the sites and incorrectly classifies 43 of the sites, with a kappa statistic of 0.0027. If the data is used as a training set it correctly classifies 95 of the sites and incorrectly classifies 9 of the sites, with a kappa statistic of 0.7913. Both the cross validation and training set method give identical looking regression trees.

Discussion:

Habitat variables which are currently measured relating to the characteristics of streams and rivers seem to aid little in the identification of characteristics which are important to predict koura presence and absence from sites when attempting to build predictive models. Even though the results of the ANOSIM test show a significant difference between present and absent sites, the cross-validation technique in Weka provides us with little confidence in the model produced.

When the data was used as a training set for the regression trees it still incorrectly classified 11 of the sites when using only habitat variables collected at each of the sites. It incorrectly classified 9 of the sites when the habitat variables from the FWENZ database were also included. The use of the data as a training set allows us to breakdown only the variables which are important for the sites we sampled, but the data is of little use if we want to predict other sites where koura could be found.

The cross-validation technique creates a regression tree which will allow us to predict sites which have koura present or absent. However the cross validation technique used for koura in this study has a kappa statistic of 0.0602, incorrectly classifying 39 sites for habitat variables measured at each of the sites. The kappa statistic is even lower at 0.0027 if the FWENZ habitat variables are included with the measured habitat variables, incorrectly classifying 43 sites. The use of these trees is therefore unlikely to aid us in the identification of sites which have koura present or absent.

When looking at both of the regression trees, using the data as training information, the interaction with both trout and eels has shown to be important in detecting the presence of koura. Eels are known predators of koura and it has been found that koura are able to detect the presence of eels through chemicals emitted through the eels' skin (Shave *et al*, 1994). A negative relationship between koura and trout presence has been identified in other streams (Townsend, 2003; Usio & Townsend, 2000; Olsson *et al*, 2006; Jansma, 1995). The size distribution of koura is also known to be affected by the presence of trout, with a higher percentage of the population being made up of large koura (OCL greater than 35mm) in the presence of trout (Usio and Townsend, 2000). Trout are also known to consume koura within Lake Taupo, with the consumption of koura changing the flesh colour and shape of the trout (Gibbs, DoC, pers comm.). The interesting point in this survey is that it is the presence of small trout (under 15cm) which is an important factor. The presence of more than 7 small trout is likely to result in the absence of koura from the site. The presence of multiple small trout and lack of koura raises an interesting point; are the small trout consuming/competing with koura?

Perch (*Perca fluviatilis*) have been found to only consume crayfish (*Pacifastacus leniusculus*) once they reach 15cm in length (Nystrom *et al*, 2006); if this is similar for brown trout then predation is unlikely to be responsible for the absence of koura. Although this relationship was found in perch, a negative association was found between trout and koura when trout averaged 132mm +/- 41mmSD (Usio & Townsend, 2000), meaning that Young of Year (YOY) and small koura are predated on by juvenile trout. It has also been found where trout are larger than 15cm, that trout and galaxiid species do not co-exist (Townsend, 2003). This finding may indicate that koura are not immune from predation even in the presence of juvenile trout. The presence of trout may also lead to indirect non-consumptive effects (Hill & Lodge, 1999; Mather & Stein, 1993), affecting the feeding of ability koura (this thesis, Chp 3) by reducing activity (Blake & Hart, 1993, Mather & Stein, 1993). Thus koura abundance would be reduced even after they have reached the size at which predation is no longer a risk (Usio & Townsend, 2000) due to increased mortality from lack of food (Stein & Magnusson, 1976; Hill & Lodge, 1999).

Riparian cover in both of the regression trees is shown to be an important habitat variable. The presence of forested riparian vegetation generally coincides with pools (due to pool forming elements of trees), undercut banks (Parkyn & Collier, 2004) and the presence of litter in the stream. All of these variables are present in at least one of the regression trees but the sites at which they occur split the importance of the variables. For example, the presence of planted forest (this excluded naturally occurring forest) in the measured habitat variables is the first variable to distinguish between the sites. Those sites with planted forest all had koura present, however for sites without planted forest, it was the presence of waterfalls which was important. The presence of waterfalls is likely to mean a lack of trout as they create a natural barrier to trout movement upstream (Usio & Townsend, 2000). Fifteen of the sites which had waterfalls had koura present. One factor which did prove to be important across the data was the availability of some sort of vegetation in the stream whether it be macrophytes, bryophytes or leaf litter. This may provide a food source for adult koura who are thought to be generally detritivorous (Parkyn *et al*, 2001), or alternatively it may provide refuges for koura to hide from predators.

The results from this survey fail to show some habitat variables which have been shown as being important in other studies. This may be due to the fact that koura were not split into different size classes and I worked with data looking at the presence or absence of koura, not the density of koura. A negative relationship between koura and sediment was found by Usio and Townsend (2000) but was not found in this study, with koura being present at sites with suspended sediment concentrations ranging from 0.60 to 160 mg/litre. Usio and Townsend (2000) did however, base all of their study in a single forested catchment. The upper sites of Usio and Townsend's (2000) study had a waterfall which excluded fish of any type, but it is likely that the stream had higher sediment concentrations further downstream due to erosion potential in a larger area of the catchment. This happens to also coincide with the presence of trout below the waterfall in the lower half of the study area. This may mean that the two measured variables (sediment concentrations and trout presence) cannot be treated separately as the presence of trout was not consistent across all the sediment concentrations. My study has been carried out across multiple catchments and land uses, taking into account sites without trout (with and without barriers) and is likely to be more representative of streams across the bottom half of the North Island.

Current wintertime equilibrium temperature plays an important role in differentiating between the presence and absence of koura when the FWENZ data is included. None of the sites had koura present if the wintertime equilibrium temperature was below 3.8^0C and 33 of the sites only had koura present if the wintertime equilibrium temperature was above 4.9^0C. Higher temperatures have been shown to decrease the intermoult frequency of koura, although they have no effect on the size increase at moulting. The warmer temperatures are likely to lead to faster growth over a season as the intermoult period decreases; koura can therefore moult more times in a season resulting in higher growth rates with higher temperatures (Parkyn, 2000). Given the above information from the regression tree which included the FWENZ data, it would appear that koura do not occur in streams with wintertime equilibrium temperatures below 3.8^0C.

Conclusion:

Koura were found at a number of sites which differed in stream characteristics and the surrounding land use. Of the variables measured those that came out as being important for the prediction of koura presence included the riparian vegetation present at a site, the presence of predators and instream factors such as the presence of bryophytes, macrophytes and leaf litter. The use of regression trees did not provide a model which we could use for predicting koura presence or absence from sites as the significance provided by the model in this analysis doesn’t provide a high level of confidence. For the variables which were shown to be important, most related in some form to the complexity of habitat provided in streams, such as the availability of stream vegetation and the composition of the streambed (for example, number of pools, shutes, etc). This is likely to coincide with the presence of predators being seen as a factor influencing koura presence. The greater the complexity of the habitat, the more likely that koura are able to hide from predators such as eels and trout. This study agrees with findings from other studies about factors important in koura habitat for example, in-stream habitat from macrophytes, bryophytes and leaf litter. However, while other studies have considered the individual habitat requirements of the different life stages of koura this study did not.

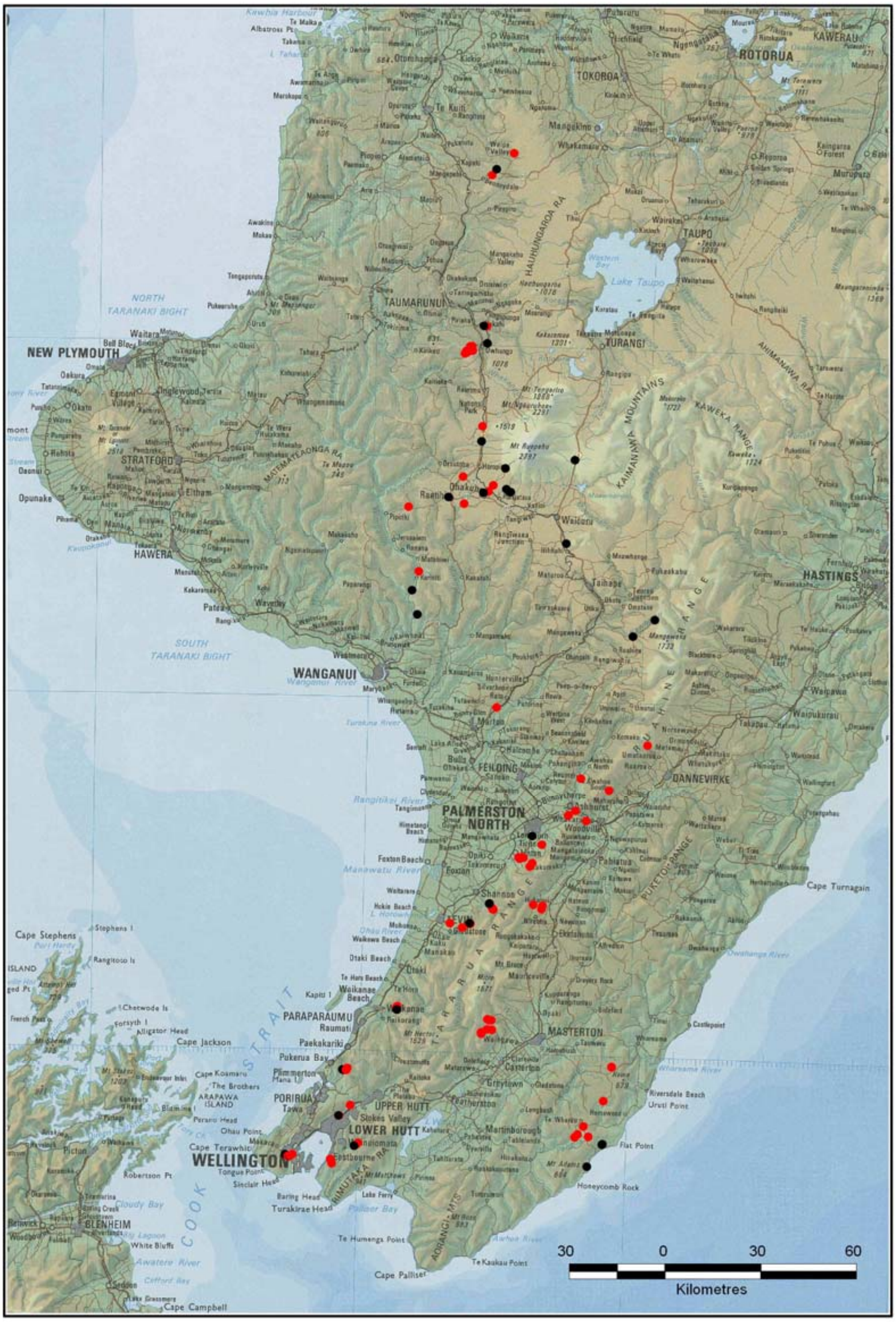


Figure 1: Presence (red circles) and absence (black circes) of koura over the 104 study sites within the Lower North Island.

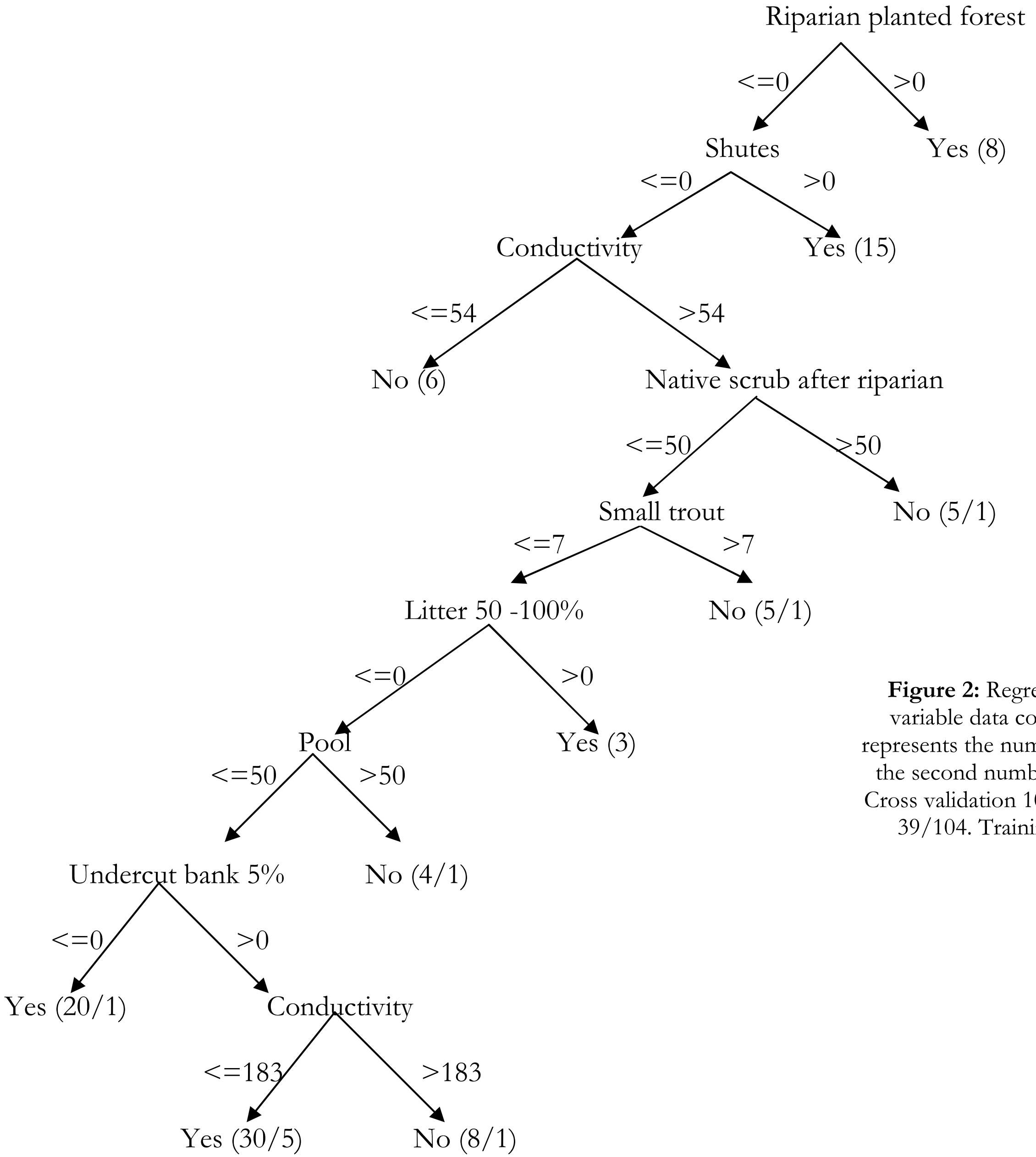
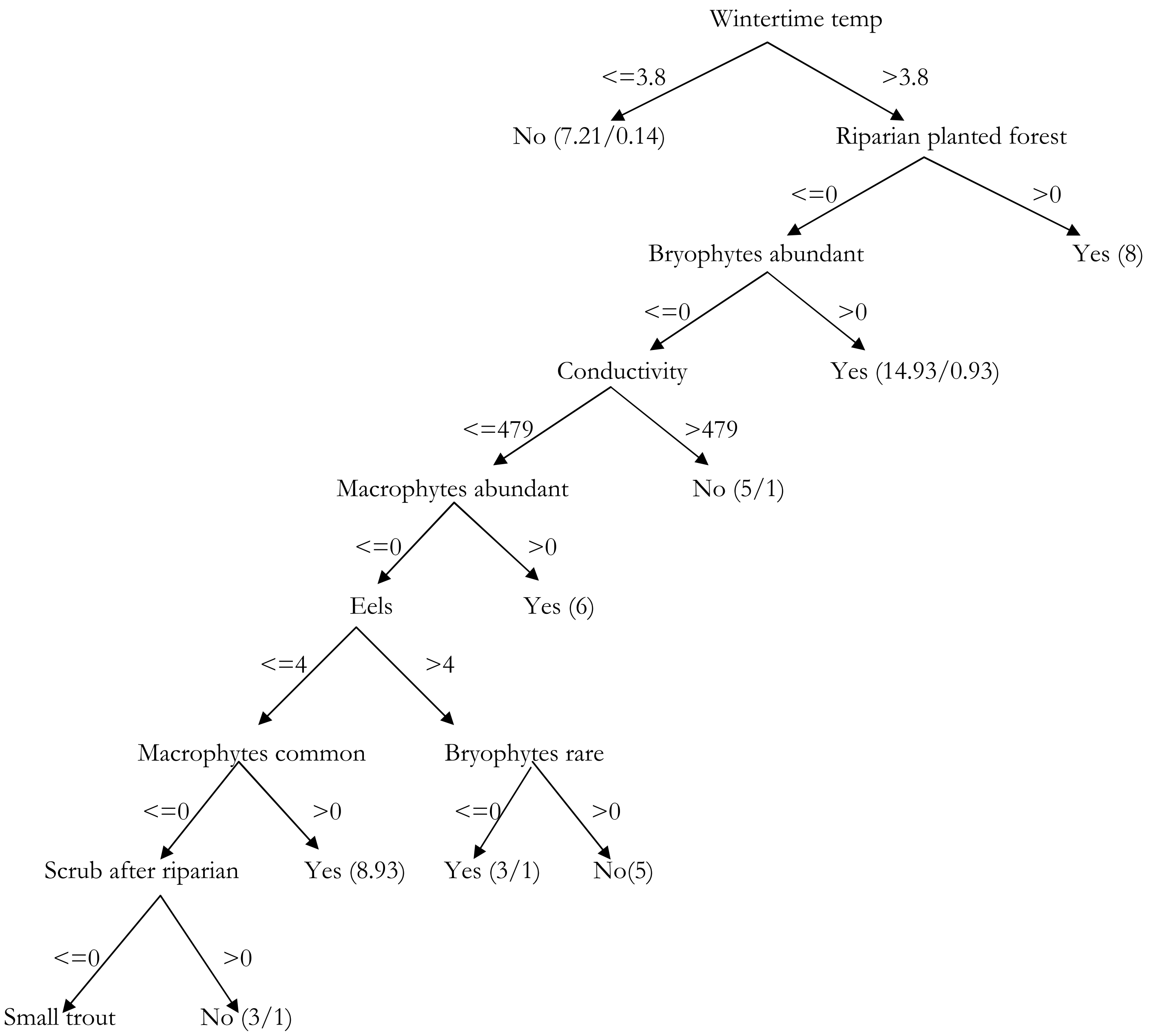


Figure 2: Regression tree showing breakdown of the sites using habitat variable data collected at each of the sites. The first number in brackets represents the number of sites at which the variable is correctly classified and the second number the number of sites at which it is incorrectly classified. Cross validation 104 - Kappa statistic 0.0602, Correct = 65/104, Incorrect = 39/104. Training set - Kappa statistic 0.74, Correct 93/104, Incorrect 11/104.



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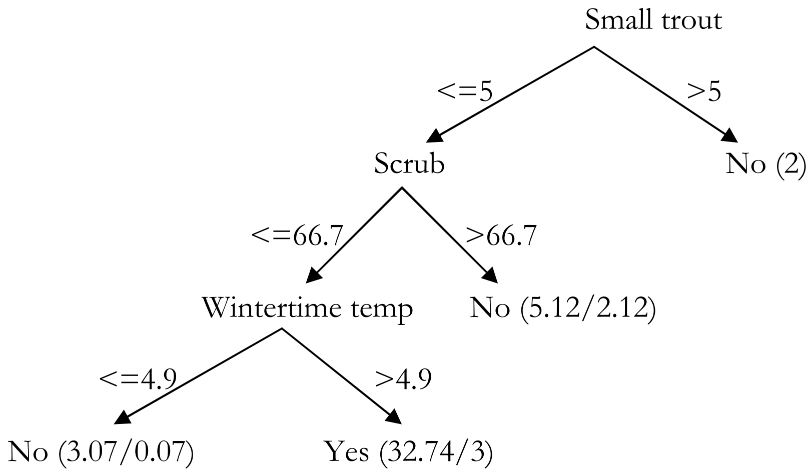

Figure 3: Regression tree showing breakdown of the sites using habitat variable data collected at each of the sites and from the FWENZ database. The first number in brackets represents the number of sites at which the variable is correctly classified and the second number the number of sites at which it is incorrectly classified. Cross validation 10% - Kappa statistic 0.0027, Correct = 61/104, Incorrect = 43/104. Training set - Kappa statistic 0.7913, Correct 95/104, Incorrect 9/104

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Appendix 1

Sites sampled in the lower North Island.

Site	Easting	Northing	P/A	Stream name
1	2668926	5986745	P	Gollans Stream
2	2669261	5985500	P	Butterfly Creek
3	2668836	5986860	P	Unnamed tributary of Butterfly Creek
4	2731391	6078481	P	Unnamed tributary of Kahuterawa Stream
5	2732040	6079533	P	Unnamed tributary of Kahuterawa Stream
6	2743520	6094713	P	Unnamed tributary of Manawatu River
7	2743355	6094450	P	Unnamed tributary of Manawatu River
8	2745574	6096006	P	Manawatu Gorge unnamed tributary of Manawau River
10	2749000	6092783	P	Manawatu Gorge unnamed tributary of Manawau River
11	2654575	5988368	A	Unnamed tributary of Silver Stream
12	2656996	5988424	P	Unnamed stream of Central Wellington
13	2713325	6240747	P	Unnamed tributary of Kakahi Stream
14	2713462	6240656	P	Unnamed tributary of Kakahi Stream
15	2711497	6240639	P	Kawautahi Stream
16	2711768	6240290	P	Unnamed tributary of Kawautahi stream
17	2713338	6242094	P	Unnamed tributary of Kakahi Stream
18	2712740	6241494	P	Unnamed tributary of Kakahi Stream
19	2712769	6242035	P	Unnamed tributary of Kakahi Stream
20	2735173	6085343	P	Turitea Stream
21	2729092	6081321	P	Unnamed tributary of Nguturoa Stream
22	2729326	6081245	P	Unnamed tributary of Nguturoa Stream
23	2728116	6080888	P	Unnamed tributary of Nguturoa Stream
24	2747297	6106212	P	Makohine Stream
26	2727786	6081337	P	Unnamed tributary of Nguturoa Stream
27	2732109	6088095	A	Unnamed tributary of Turitea Stream
28	2724019	6197216	A	Mangaehuehu Stream
29	2725108	6196333	A	Waiharuru Stream
30	2725424	6196333	A	Unnamed tributary of Waiharuru Stream
31	2716502	6217040	P	Waimarino Stream
32	2716210	6212334	A	Unnamed tributary of Makatote River
33	2719932	6198410	P	Unnamed tributary of Mangawhero River
34	2718366	6196453	P	Unnamed tributary of Mangawhero River
35	2716855	6196205	A	Unnamed tributary of Mangawhero River
36	2705925	6194801	A	Unnamed tributary of Makotuku River
37	2710698	6192745	P	Mangahowhi Stream
38	2710399	6201201	P	Unnamed tributary of Makotuku River
39	2723624	6203757	A	Mangawhero River
40	2717950	6248252	P	Unnamed tributary of Kakahi Stream
41	2716915	6248425	A	Unnamed tributary of Kakahi Stream
42	2711051	6239757	P	Unnamed tributary of Kawautahi Stream
43	2711859	6240572	P	Unnamed tributary of Kawautahi Stream
44	2717986	6243018	A	Unnamed tributary of Whakapapa River
46	2689480	6034461	P	Unnamed tributary of Waikanae River
47	2689892	6034725	P	Waikanae River
48	2689657	6033670	A	Unnamed tributary of Waikanae River

Site	**Easting**	**Northing**	**P/A**	**Stream name**
49	2689657	6033670	A	Unnamed tributary of Waikanae River
50	2673046	6014628	A	Horokiri Stream
51	2672629	6014701	A	Unnamed tributary of Horokiri Stream
52	2655616	5987810	P	Unnamed tributary of Karori Stream
53	2655971	5987838	A	Unnamed tributary of Karori Stream
54	2656142	5987924	P	Unnamed tributary of Karori Stream
55	2675058	6003585	P	Unnamed tributary of Hutt River
56	2671592	6000502	P	Speedys Stream
57	2671425	6000494	A	Unnamed tributary of Speedys Stream
58	2674117	6015300	P	Unnamed tributary of Horokiri Stream
59	2673917	6014641	P	Unnamed tributary of Horokiri Stream
60	2719568	6065229	P	Arapeti Stream
61	2719887	6064989	P	Unnamed tributary of Tokomaru River
62	2719767	6065114	P	Unnamed tributary of Tokomaru River
63	2706389	6060611	P	Unnamed tributary of Koputaroa Stream
64	2712778	6060455	P	Unnamed tributary of Makahika Stream
65	2712554	6060335	P	Unnamed tributary of Makahika Stream
66	2712469	6060689	A	Unnamed tributary of Makahika Stream
67	2719563	6065419	P	Unnamed tributary of Tokomaru River
68	2718735	6066889	A	Mangatangi Stream
69	2718718	6066715	A	Unnamed tributary of Mangatangi Stream
70	2710148	6059302	P	Unnamed tributary of Makahika Stream
71	2718440	6030291	P	Gentle Annie Creek
72	2718069	6030704	P	Atiwhakatu Stream
73	2719231	6030331	P	Unnamed tributary of Atiwhakatu Stream
74	2746285	5994443	P	Unnamed tributary of Pahaoa River
75	2745389	5993641	P	Pahaoa River
76	2748142	5996939	P	Unnamed tributary of Andersons Stream
77	2749681	5993798	P	Unnamed tributary of Pahaoa River
78	2716015	6026556	P	Unnamed tributary of Mangatarere Stream
79	2716108	6026028	P	Unnamed tributary of Mangatarere Stream
80	2717922	6027352	P	Unnamed tributary of Mangatarere Stream
81	2719375	6027206	P	Unnamed tributary of Mangatarere Stream
82	2756973	6015447	P	Makahaka Stream
83	2756928	6015540	P	Unnamed tributary of Makahaka Stream
84	2754412	6004899	P	Totara Stream
85	2749199	5984455	A	Unnamed coastal stream
86	2753847	5991313	A	Unnamed tributary of Arawhata Stream
87	2754107	5991437	A	Arawhata Stream
88	2677576	5991928	P	Wainuiomata River
89	2677023	5991456	P	Wainuiomata River
90	2676277	5991053	A	Wainuiomata River
91	2719527	6296076	P	Unnamed tributary of Mangaaruhe Stream
92	2720901	6297842	A	Mangaaruhe Stream
93	2726472	6302910	P	Unnamed tributary of Otamaroa Stream
94	2735047	6065145	P	Unnamed tributary of Mangatainoka River
95	2735322	6066102	P	Mangaraupiu Stream
96	2732322	6066401	P	Unnamed tributary of Mangaraupiu Stream
97	2696533	6171333	P	Pararoa Stream
98	2694385	6165413	A	Unnamed tributary of Whanganui River

Site	**Easting**	**Northing**	**P/A**	**Stream name**
99	2696043	6157719	A	Whauteihi Stream
100	2756206	6102367	P	Unnamed tributary of Coppermine Creek
101	2768162	6116532	P	Tamaki River west Branch
102	2770425	6155917	A	Unnamed tributary of Kawhatau River
103	2763648	6150766	A	Mangakukeke Stream
104	2742698	6180060	A	Unnamed tributary of Hautapu River
105	2745572	6206407	A	Unnamed tributary of Mangaio Stream
106	2721016	6128661	P	Unnamed tributary of Porewa Stream
107	2693416	6191750	P	Unnamed tributary of Koreronui Stream

Appendix 2

Definitions of the abbreviations used in figures 3 and 4.

Abbreviation	**Definition**
Riparian planted forest	The riparian zone from the stream edge back 20m consists of planted forestry e.g. pine forestry.
Shutes	The percentage of the 100 metre stream study reach which is made up of shutes.
Conductivity	A measure of the total ions in the water in the study reach.
Native scrub after riparian	The percentage of habitat made up of native scrub, after the immediate riparian zone (starting at 20 metres from the water edge).
Small trout	The number of trout under 15cm encountered while spot lighting the study reach at night.
Litter 50 – 100%	The study reach of the stream has a covering of leaf litter between 50 to 100% of the streambed.
Pool	The percentage of the 100 metre stream study reach which consists of pools.
Undercut bank 5%	The amount of bank that is undercut in the 100 metre study reach.
Wintertime temp	Current wintertime equilibrium temperature. Taken from FWENZ database.
Bryophytes abundant	The amount of bryophytes present in a study reach. One of three categories – rare, common, abundant.
Macrophytes abundant	The amount of macrophytes present in a study reach. One of three categories – rare, common, abundant.
Eels	The number of eels seen in the study reach when night spotting.
Macrophytes common	The amount of macrophytes present in a study reach. One of three categories – rare, common, abundant.
Bryophytes rare	The amount of bryophytes present in a study reach. One of three categories – rare, common, abundant.
Scrub after riparian	The percentage of habitat made up of native and introduced scrub, after the immediate riparian zone (starting at 20 metres from the water edge).
Scrub	Percentage of riparian area in LCDB category (scrub)

Appendix 3

Examples of some of the sites sampled.

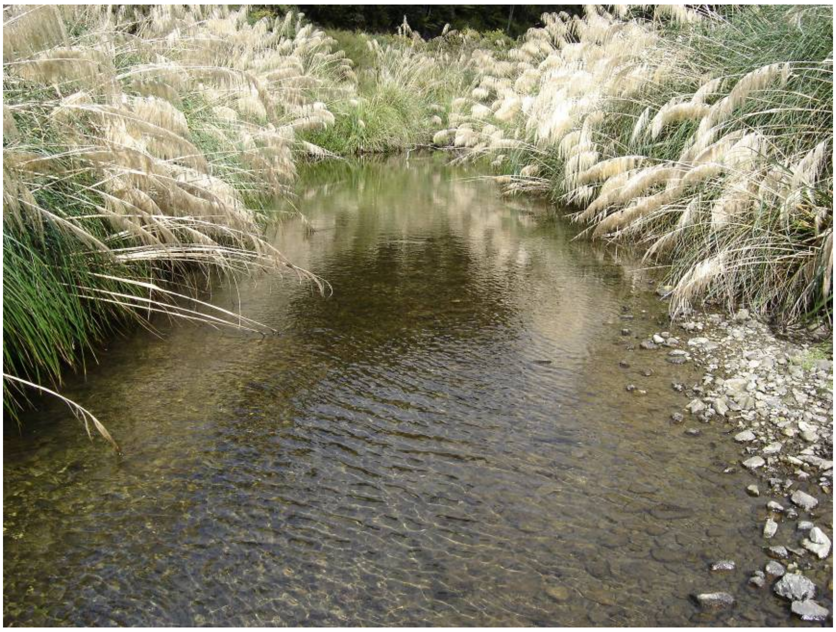

Plate 1: Site 88 - Wainuiomata River. Koura present.

Plate 2: Site 92 - Mangaaruhe Stream. Koura absent.

Plate 3: Site 30 - Unnamed tributary of the Waiharuru Stream. Koura absent.

Plate 4: Site 23 - Unnamed tributary of the Nguturoa Stream. Koura present.

The predatory interactions between introduced brown trout (*Salmo trutta*), the endemic freshwater crayfish (koura, *Paranephrops planifrons*) and Cran's bully (*Gobiomorphus basalis*).

Abstract:

A study in the Manawatu region attempted to establish the predatory interactions between the introduced brown trout (*Salmo trutta*), the endemic freshwater crayfish (koura, *Paranephrops planifrons*) and the endemic Cran's bully (*Gobiomorphus basalis*). The study failed to show adult brown trout predating on koura. Koura were not found to actively predate on live juvenile brown trout or Cran's bullies, but did scavenge upon dead juvenile brown trout. The capture of live fish by koura is energy expensive and provides a plausible reason as to why live fish were not consumed but dead trout were scavenged. The failure of brown trout to consume koura is likely related to high summer temperatures, koura size class and elevated stress throughout the experimental period.

Introduction:

Brown trout (*Salmo trutta*) were introduced into New Zealand in 1867. From their introduction until 1921, sixty million brown trout have been raised and released into New Zealand streams (Townsend, 1996).

Studies in New Zealand have shown that trout affect vertebrate and invertebrate species in freshwater environments. Declines in galaxiid abundance in New Zealand and Australia have been brought about by predation from large introduced trout (McIntosh, 2000a; Crowl, Townsend & McIntosh, 1992), with interspecific competition leading to the displacement of native galaxiid species from their microhabitats (Bonnett & McIntosh, 2004). Invertebrate species have been affected by being directly preyed on as a food source. This has also lead to indirect effects, for example a study by Greig and McIntosh (2006) found that trout predation on *Zelandopsyche ingens* (an obligate shredder) lead to a reduction in the density of this species, which in turn lead to a reduction in the breakdown of coarse particulate organic matter and reduced production of fine particulate organic matter.

Olsson *et al*, (2006) found that abundances of *Paranephrops planifrons* and galaxiids in eighteen West Coast streams in the South Island of New Zealand were significantly lower in streams with trout. In another study by Shave, Townsend, & Crowl (1997) the anti-predator behaviour of *Paranephrops zealandicus* was tested against a native predator (the longfin eel *Anguilla dieffenbachii*) and the introduced brown trout. Koura were able to detect chemical cues released from eels but not from trout, and it was thought that koura may be at greater risk from the introduced predator due to their inability to detect trout non-contact chemical cues.

An overseas study on *Camarus bartonii*, a freshwater crayfish, in the headwater streams of Allegheny Plateau, United States of America found that large crayfish made up a higher proportion of the crayfish population at sites with high fish biomass (Seiler & Turner, 2004). This has been proposed to be consistent with the hypothesis that fish predation on small crayfish may be limiting crayfish population size in some streams (Seiler & Turner, 2004). It is effects such as these that have placed the brown trout onto the International Union for Conservation of Natures (IUCN's) list of 100 of the World's Worst Invasive Alien Species, with brown trout being ranked at number 83 and rainbow trout (*Oncorhynchus mykiss*) ranking higher at number 63 (Lowe *et al*, 2000).

In contrast to the predation of brown trout on native stream fauna, overseas crayfish species have been found to consume the eggs and juveniles of some fish species (Savino & Miller, 1991; Rahel, 1989; Bryan, Robinson, & Sweetser, 2002). *Orconectes virilis* predated on eggs and sac fry of lake trout (*Salvelinus namaycush*) and rainbow trout (*Oncorhynchus mykiss*), although the rate of predation depended on the substrate size in the enclosures in which the experiments were carried out (Savino & Miller, 1991).

The Little Colorado spinedace (*Lepidomeda vittata*), native to North American streams, is trout-like in feeding habits, habitat use and behaviour and was found to be predated on by an introduced crayfish (*Orconectes virilise*). When introduced rainbow trout (*Oncorhynchus mykiss*) were added to the experiment the avoidance behaviour of the spinedace was even more pronounced (Bryan *et al*, 2002).

Introduced signal crayfish (*Pacifastacus leniusculus*) and the Swedish native crayfish *Astacus astacus* have been found to consume the eggs and tadpoles of seven amphibian species in "out of stream" experiments. Video recordings of the experiment showed that crayfish

used their claws and walking legs to catch the tadpoles (Axelsson, Nystrom, Sidenmark, & Bronmark, 1997).
Koura are known omnivores (Parkyn *et al*, 2001), however no studies have been conducted to see if they predate on introduced and native fish species at different life stages.

With the above factors in mind this chapter has a two part focus:

- The first looking at the interaction between adult brown trout and koura. Allen and Claussen stated that "the larger the fish, the larger the animal which it eats, and the more easily are hard-shelled forms consumed" (1960, pg 80). Large lake brown trout have been known to consume koura (Gibbs, pers comm.) and in this experiment we intend to see if stream sourced adult brown trout affect the density and size distribution of a koura population in their natural environment.
- The second part intends to look at koura predation on live juvenile brown trout and adult cran's bullies as well as looking at scavenging on dead juvenile brown trout.

Methods:

Brown trout predation on koura:

The study was carried out in the Kahuterawa and Turitea streams, Palmerston North, New Zealand (fig 1). Both streams originate in the Tararua Ranges and flow south-east towards the Manawatu River.

Trout were obtained from Manawatu streams and rivers by spotlighting with a lightforce portable spotlight powered by a 12 volt dry cell battery. Trout varied in fork length size from 40 to 60cm. Trout were placed into purpose built stream enclosures until they were required for the experiment.

Koura were obtained from the Mangaore catchment by spotlighting with a lightforce portable spotlight, powered by a 12 volt dry cell battery. All sizes of koura were captured except for berried females. Koura were kept in a constant temperature room (12^0C) until they were required for the experiment.

Three enclosures were used each measuring 6 metres in length, 0.70 metres wide and 0.50 metres in height. The outside frame of the enclosures consisted of concrete strengthening steel welded together and then covered with green wind shade cloth around the three largest sides. The top of the enclosure was covered with wind shade cloth independent of the sides to allow ease of access while the experiment was running. At one end of the enclosures the wind shade cloth was doubled up to provide a place for trout shelter. Both two ends of the enclosures were made of 2.54mm stainless steel mesh to allow water flow and the entry of some stream invertebrates.

The enclosures were placed in the streams at the end of a pool and the start of a riffle (or vice versa). This allowed areas of slower moving water within the enclosures. Substrate representative of the streambed was placed in the enclosures 24 hours prior to the start of the experiments to allow colonisation by invertebrates.

At the beginning of the experiment 6 koura per square metre were added (27 in total) to the enclosures, with trout added to two of the enclosures and the other enclosure left as a control. Koura oribit carapace length (OCL) was recorded prior to the start of the experiments to ensure that size distribution was equal across the enclosures. Initially the experiment was left to run for 96 hours, however this was increased to 168 hours after the initial run.

The experiments were run 5 times, the first two in the Kahuterawa Stream and the remaining 3 in the Turitea stream.

All trout and koura were returned to their original place of capture at the conclusion of the experiments.

Koura predation on brown trout and cran's bullies:

This study was carried out in the Ngururoa Stream, Linton, Manawatu, New Zealand (fig 2). The stream originates in the Tararua Ranges and flows south-west towards the Manawatu River through farmland.

A total of 20 juvenile brown trout ranging in fork length size from 4 to 5.9 cm were collected by electro-fishing from Karori Wildlife Sanctuary, Wellington. Eight koura (minimum oribit carapace length (OCL) of 3.0cm) were obtained by spotlighting with a lightforce portable spotlight powered by a 12 volt dry cell battery from an unnamed tributary of the Kawautahi Stream, Owhango.

Koura and trout were placed in enclosures 20cm by 20cm wide and 30cm high. The enclosure consisted of a plastic container that had 10 by 15 cm holes in the four sides and bottom, with 2.54mm stainless steel woven mesh covering the holes. Aluminium fly screen mesh was placed over the top of the enclosures to prevent koura climbing and trout jumping out. The enclosures were placed in the stream and anchored to a warrath standard (Appendix 1). Prior to the start of the experiment substrate from the stream bed was placed in the bottom of each of the enclosures and left for 24 hours to allow macroinvertebrates to colonize.

Each of the animals was randomly allocated to one of the following three treatment enclosures in the stream.

1) Trout only (trout control).
2) Koura only (koura control).
3) Koura and trout together (experimental trial).

Each trial was replicated five times. Trout were kept at a density of two per enclosure and one koura per enclosure. Trout fork length and koura OCL measurements were made at the start of the experiment.

Enclosures were examined daily to ensure that the mesh had not become blocked. The trial was run for 96 hours, after which the enclosures were removed from the stream. Any remaining koura and trout were recorded.

Water depth was measured in each enclosure using a ruler and temperature measured using an Orion 122 pocket meter at the conclusion of the experiment.

The above experiments were repeated using native adult Cran’s bullies and dead juvenile brown trout at a density of two fish per enclosure.

Results:

Brown trout predation on koura:

The initial experiment which ran for 96 hours in the Kahuterawa Stream had all koura accounted for at the conclusion of the experiment consequently, the length of time was increased to 168 hours for the remaining experiments. The second experiment was ended early due to vandalism and the three remaining experiments were carried out in the Turitea stream. The last three experiments had all koura recovered from the enclosures at the conclusion of the experiments.

Koura predation on brown trout and cran's bullies:

There was no difference between the depth ($t = 1.41$, $df = 3$, $p = 0.26$), the length of koura ($t = -0.23$, $df = 7$, $p = 0.24$), the length of live and dead trout ($t = -0.42$, $df = 11$, $p = 0.68$ and $t = 1.56$, $df = 9$, p-value = 0.15 respectively), and the length of bullies ($t = 1.63$, $df = 9$, p-value = 0.14) in the trial and control enclosures across all the experiments.

Live trout:

Out of the five trial replicates only one trout died after 96 hours and it was assumed that the koura had predated on it due to the koura being seen to eat the trout when the trial was finished. All the remaining study animals were recovered.

Live Cran's bullies:

All study animals were recovered at the conclusion of the experiment.

Dead trout:

Out of the five trial replicates four trout (from two enclosures only) were consumed after 96 hours. None of the trout in the control enclosure had evidence of scavenging and all of the koura (in both the experimental and control enclosures) survived.

Discussion:

Koura have been shown to be negatively associated with the presence of trout in streams (Usio & Townsend, 2000; Olsson *et al*, 2006). Although negative correlations have been found between koura abundance and trout presence there has been little attempt to examine the effect that trout directly have on koura populations. Large crayfish have been found to make up a larger proportion of crayfish (*Cambarus bartonni*) populations in sites which have high fish biomass (Seiler & Turner, 2004). Usio and Townsend (2000) found that in the presence of brown trout a large proportion of the koura population were in the large size class (greater than 35mm OCL), compared to a koura population that had a higher proportion of smaller koura (less than 35mm OCL) in the absence of brown trout. However, the opposite has also been found with predator biomass having no effect on the mean crayfish (*Pacifastacus leniusculus*) size in lakes (Nystrom *et al*, 2006). Gut contents of large brown trout taken from streams were found to contain, on average, terrestrial invertebrates (60%), small crayfish (27%) and isopods (27%). In large perch (*Perca fluviatilis*) caught from lakes, crayfish made up 46% of stomach contents (Nystrom *et al*, 2006). Trout in Lake Taupo, New Zealand are known to consume koura with their shape and flesh colour being an indication that they have consumed koura while in the lake (J.Gibbs, Department of Conservation, *pers comm.*).

Koura are known to make up the majority of shredder biomass and one study found that they made up 99% of the shredder biomass (Usio & Townsend, 2001). *Paranephrops spp* are generally carnivorous as juveniles and as they mature the consumption of detritus becomes more important in the diet (Parkyn *et al*, 1997), although growth only comes about by utilizing the energy that is derived from consuming invertebrate prey and not by utilizing the detritus (Parkyn *et al*, 2001). The fact that juveniles are mostly carnivorous makes them more susceptible to predation as they must hunt prey to eat. Adults however, are detritivorous in nature allowing them to potentially find refuges by hiding in leaf packs while consuming detritus. Larger crayfish are also more likely to be able to defend themselves when being attacked (Allen & Claussen, 1960). This is likely to help explain the pattern found in a study by Seiler and Turner (2004), where a larger size of crayfish was found in areas that had a high biomass of predatory fish.

Part one of this chapter failed to show results similar to the previously mentioned studies. In this study brown trout did not actively predate on koura. The reasons for this are varied and are considered below.

- The experiment was carried out in February when stream temperatures were high within the study streams. Spot measurements taken during the day in the Turitea varied from 17 to 19.5^{0}C. Brown trout are known to stop feeding at 21^{0}C (Death, 2002). With temperatures nearing the known maximum for feeding it is possible that feeding rates were greatly reduced during the experiment. Also other food sources were available in the form of invertebrates in drift and these may have been taken rather than actively hunting prey.

- The length of time experiments were run for may not have been long enough. This is the reason why the experiment was increased from 96 hours to 168 hours. However, this time frame may still not have been long enough to allow the trout to adjust to their new environment and then commence feeding as normal.
- The substrate placed in the enclosures will not have been as tightly packed as the substrate found in the streams. This may have allowed the koura to hide more effectively than would normally be the case in their natural environment. This increased ability to hide would reduce the chance that they had of being predated on.
- There was a lack of Young of Year (YOY) koura in the enclosures. Due to the time of year and the risk of YOY escaping from the enclosures, they were not used in these experiments. This may have meant that the size of koura encountered in the experiments were too large for the brown trout to be able to consume.
- A combination of the above factors may have been responsible.

Although no measurements were taken of the substrate in these experiments, in other experiments it has been found that the interaction between substrate size and predation is important in order to be able to predict crayfish abundance (Nystrom *et al*, 2006). It has also been found in *Paranephrops zealandicus* that Young of Year (YOY) are positively associated with coarse substrate. While adults and YOY are positively associated with wood cover (Usio & Townsend, 2000), both of these factors provide refuges to allow koura to hide from both native and introduced predators. Feeding habits of koura and trout may mean that they are less likely to encounter each other. Koura are generally nocturnal (Usio & Townsend, 2000), although they can be seen out foraging during the day (*per obs*). Brown trout generally feed during the day although they have been shown to forage actively at night (Zimmerman & Vondracek, 2007), increasing the chance of brown trout coming across koura as their feeding habits overlap. Even with these factors taken into consideration the above experiment still failed to show any brown trout predation on koura in the 168 hour time period.

The introduction of brown trout into New Zealand in 1867 for recreational fishing means that brown trout and koura haven't evolved together over long time scales. This lack of co-evolution is thought to account for the lack of a behavioural response when koura have been subjected to trout chemical cues (Shave *et al*, 1994), especially when it was compared to the behaviour of koura exposed to chemical cues from the native predator, the longfin eel (Shave *et al*, 1994). The inability of koura to detect trout chemical cues works in two ways; the inability of juvenile koura to detect trout chemical cues places them at greater risk of predation, and the inability of adult koura to detect juvenile trout may mean that they miss potential prey species. The results found in the above experiment provide evidence for this. The lack of trout chemical detection would mean that koura would have to actively seek trout through visible cues or through opportunistic movements. This lack of active predation on trout by crayfish was also found in a study by Stenroth and Nystrom (2003) on signal crayfish (*Pacifastacus leniusculus*), where the survival of trout was positively correlated with the velocity of the water flowing through the enclosures, and had nothing to do with the presence of *P. leniusculus*. This trial was also run for a month, with greater potential that the crayfish and trout would encounter each other. *Astacus astacus* (native noble crayfish) and *Pacifastacus leniusculus* (signal crayfish) in a long-term study were also found to have no effect on fish species at 64 sites studied in Sweden, although they had frequently been mentioned as species predating and competing with fish (Degerman *et al*, 2007).

Procambarus clarkii, a scavenger crayfish, was found to prefer animal food when it consisted of dead, dying or immobilised fish (Lowery & Mendes, 1977). Commercial fishermen have reported damage to fish caught in gill nets due to *P. clarkii* feeding on fish that were trapped in the nets (Lowery & Mendes, 1977). Renai and Gherardi (2004), also found that *P. clarkii* preyed faster on dead trout fry than it did on live ones. A previous study has found that because crayfish are unable to float and are poor swimmers, more mobile aquatic life are not captured effectively as crayfish food (D'Abramo & Robinson, 1989). Therefore dead or injured trout are taken due to the energy costs that are associated with having to predate on prey that is highly mobile (Renai & Gherardi, 2004). At the conclusion of my study a trout was consumed by a single koura (per obs). The trout and koura were in close proximity as they were being transported back to the lab in the same container. In a study conducted on signal crayfish

(*P. leniusculus*), it was discovered that fish remains were found frequently in their stomachs. There was, however, no indication of whether the fish were dead or alive when they were consumed as the crayfish were taken from the wild and then their stomach contents analysed (Lowery & Mendes, 1977).

The results in my study reinforce the findings of overseas studies on different crayfish species. Koura in my experiments scavenged on the remains of the dead trout in forty percent of the enclosures. Although this result is not significant it shows that if koura happened to come across dead matter, and it requires little energy expenditure to obtain it they will consume it. Parkyn *et al* (2001) found through stable isotope analysis that aquatic invertebrates were incorporated into body tissue of *Paranephrops planifrons*, while detritus was not utilised for growth. This gives weighting to the fact that protein derived from animal sources is important for growth and that a diet consisting only of leaf detritus will not sustain crayfish populations.

The koura used in my experiment had not previously been exposed to trout as the unnamed tributary from which they were collected is too small for trout to occupy. Lack of co-evolution may be given as a plausible reason for the lack of actively predating on brown trout however, this conflicts with the findings in the bully trials. If co-evolution was important for koura feeding then one would expect koura to predate on bullies as they occur together in streams, are both endemic to New Zealand and are both benthic in their habitat use, meaning they are more likely to encounter each other. The fact that they did not predate on bullies in this study reinforces that the lack of co-evolution is an unlikely reason and it is more likely to be due to energy expenditure in catching prey. Similar studies carried out overseas have found the opposite result in that adult crayfish predated on eggs and juveniles of brown trout (Rubin and Svensson, 1993). Although crayfish appeared to actively predate on trout in this trial, there is no mention of other food sources being made available for them to feed on. Food was also withheld for 48 hours prior to the start of the experiment. Therefore, the results found in this experiment may not be so relevant to the natural environment.

Adult crayfish are thought to go through an ontogenetic shift in the food they consume, with detritus becoming more and aquatic invertebrates less important as they become larger. This has been reported in a range of crayfish species (Mason, 1975; Reynolds, 1979; Goddard, 1988; France 1996a). In a study involving *Paranephrops zealandicus*, larger koura were found to contain a higher proportion of terrestrial detritus in their stomachs (Hollows *et al,* 2002). Although these studies have found a lack of animal matter in the stomach of crayfish, a study by Guan and Wiles (1998), found that 30% of amorphous material in the stomach was thought to have come from animal origin due to its oily paste and its occurrence with hard bodied animal material. This substance was also lacking in the stomachs of those crayfish which had only consumed detritus material. Momot (1995) suggested that studies on crayfish stomach contents often overestimated the indigestible material (plant fragments, detritus) and underestimated the presence of easily digestible soft-bodied metazoans. Therefore, animal predation may be more important in crayfish than previously thought, but continues to be underestimated using current study methods.

Conclusion:

The recent introduction of trout to New Zealand has had negative impacts on the freshwater fauna found in waterways in New Zealand. The first experiment in this chapter failed to show a negative relationship between the presence of trout and the abundance of koura. Brown trout may in fact not predate on koura however, this is highly unlikely given the results of other studies and the gut content of brown trout that have been caught by fishermen (Usio & Townsend, 2000; Seiler & Turner, 2004; Nystrom *et al*, 2006; Townsend, 2003; Olsson *et al*, 2006; Gibbs, pers comm.). The reasons for this lack of predation are likely due to a combination of environmental factors such as higher water temperatures, koura size range and elevated stress levels during the experiment.

For crayfish growth it is recognised that animal protein is required (Momot, 1995). Although it has been found that adult koura stomachs are dominated by leaf detritus in forested catchments, the opposite has been found in pasture streams where invertebrates were found to dominate the stomach material (Parkyn *et al*, 2001). It is this consumption of animal protein that leads to growth in koura as shown from stable isotope analyses (Parkyn *et al*, 2001).

The fact that koura did not actively predate on brown trout in the above trial does not disagree with the findings that animal protein is important for growth; it may be that koura did not see brown trout as a potential protein source. The reasons for this vary, koura may not see brown trout as a food source as they have not co-evolved due to the trouts' recent introduction to New Zealand. Alternatively the hunt for brown trout may be too energy-expensive for koura and it is therefore uneconomical for them to predate. The energy-expensive theory is supported by the fact that the koura did not predate on the Cran's bullies in the experiments. These species were frequently found together when carrying out fieldwork (*per obs*) and would therefore have co-evolved together. The bullies may have evolved methods to detect the present of the koura, or it may be that bully capture requires too much energy to be expended by the koura and is not worth the reward when they catch the prey. This is further supported by koura consuming dead trout in forty percent of the stream enclosures in the above experiments.

This study did not examine the contents of the koura stomachs at the conclusion of the experiment; it is therefore not possible to conclude what they consumed while the trial was in progress, although natural food sources found in streams would have been available to them.

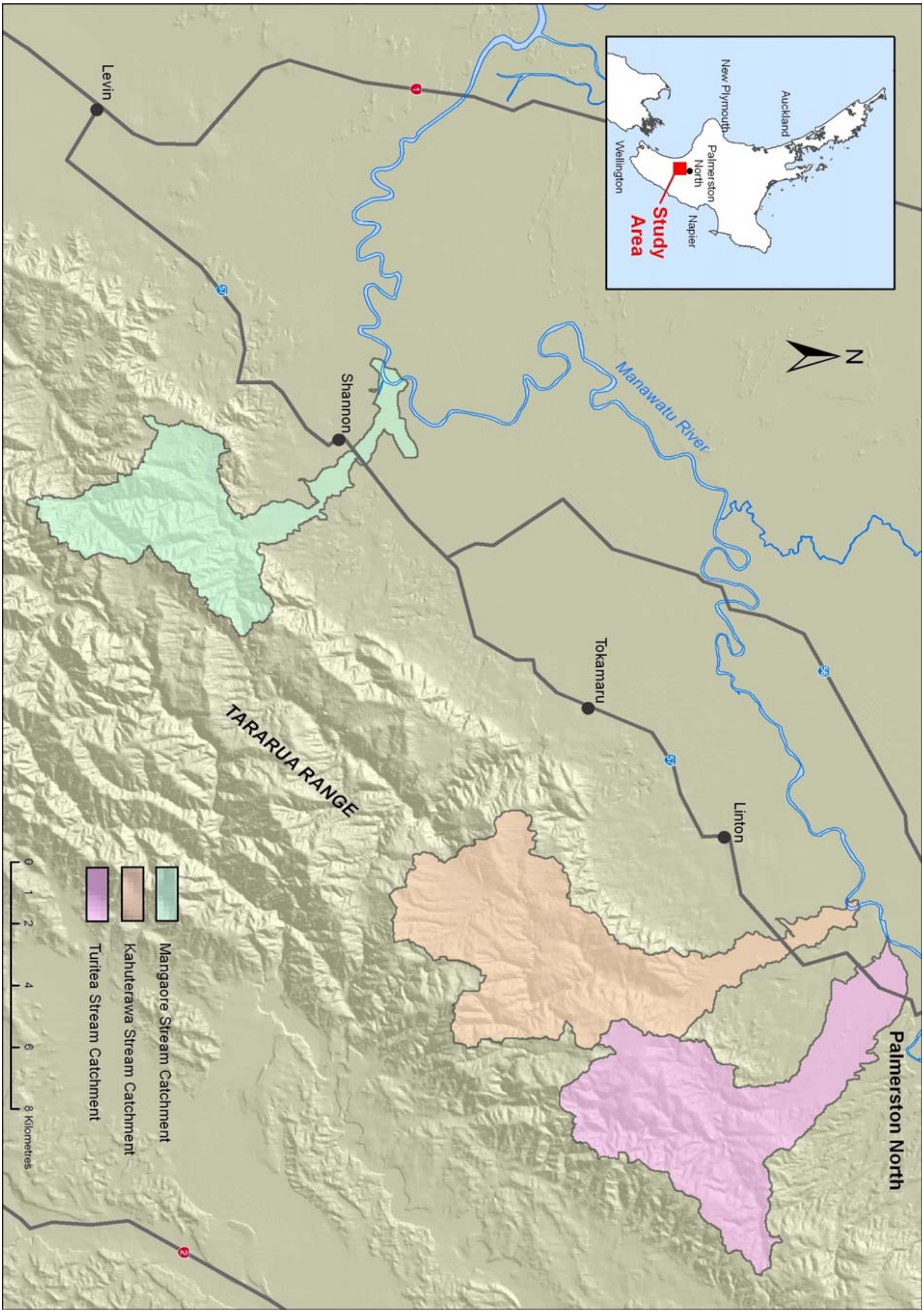

Figure 1: Map showing the catchment boundaries of the Mangaore, Kahuterawa and the Turitea Stream, Manawatu.

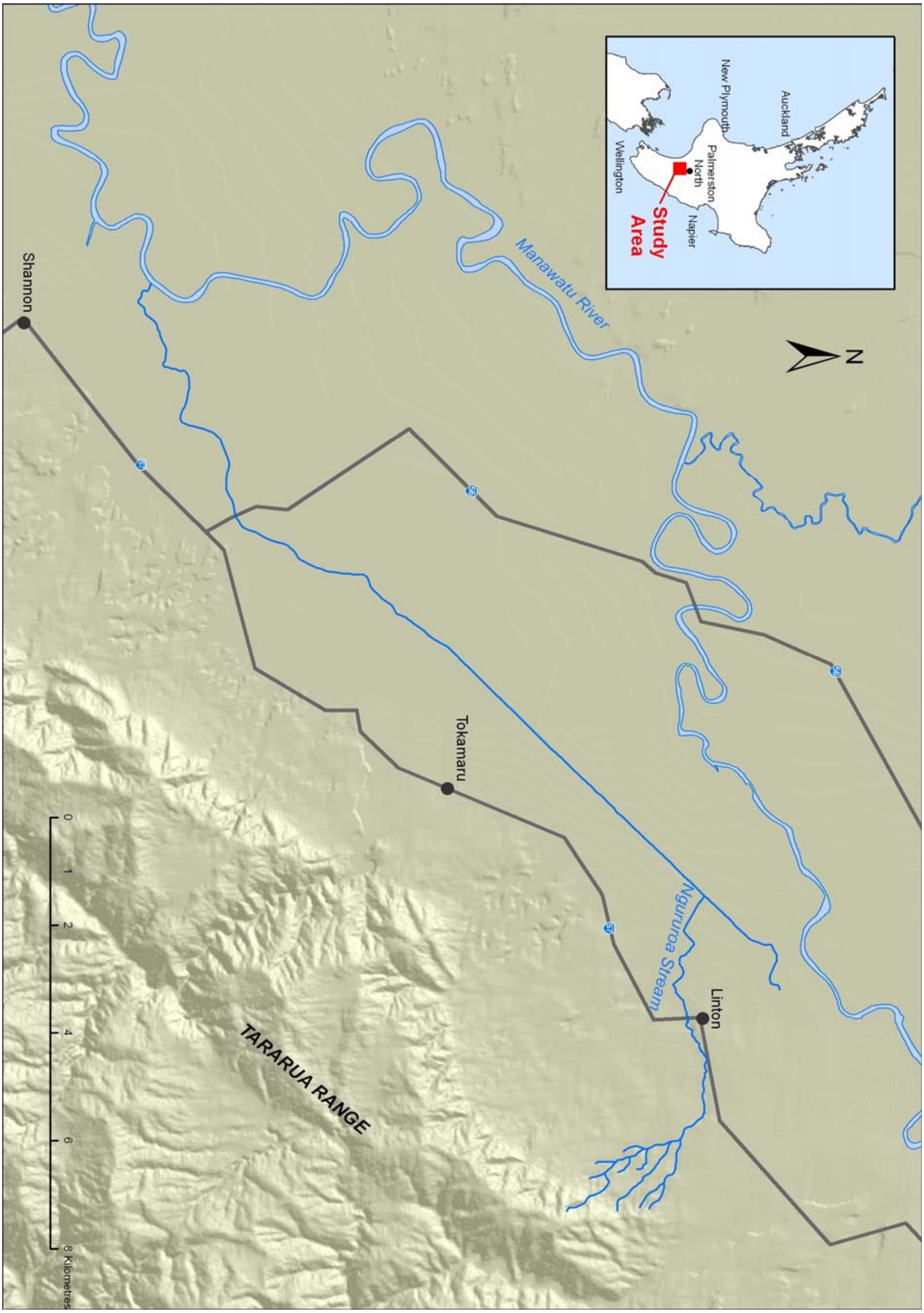

Figure 2: Map showing the Ngururoa Stream, Manawatu.

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Appendix 1

Plate 1: Enclosure used for koura predation trials.

Plate 2: Example of stream set up of enclosures used for koura predation trials. Note: actual streams used for this experiment were smaller in size.

The effects of eel (*Anguilla* spp) and brown trout (*Salmo trutta*) presence on the rate of leaf breakdown by freshwater crayfish (koura, *Paranephrops planifrons*).

Abstract:

The presence of predators significantly reduces the ability of *Paranephrops planifrons* (koura) to process leaf matter. The presence of brown trout (*Salmo trutta*), an introduced predator, reduces the processing of leaf matter more than that of a native predator *Anguilla* spp. The reduced breakdown of leaf matter is likely to have significant flow-on-effects to in-stream processes considering koura can make up 99% of the shredder biomass in some streams. This reduction in shredding will reduce the amount of food that is available for other invertebrates such as filter feeders, which rely on coarse and fine organic matter for food.

Introduction:

Koura have been found to greatly reduce the amount of leaf matter in artificial channels (Parkyn *et al*, 1997). This breakdown of leaf matter by koura is important for consumers further down the food chain that rely on Coarse (CPOM) and Fine Particular Organic Matter (FPOM) for food. Consequently, koura play a key role in structuring stream food webs (Parkyn *et al*, 1997). The presence of koura predators has the ability to change the dynamics of this process by changing the way in which koura behave. These changes are predicted to work in two potential ways; firstly, by density-mediated effects through predator-induced changes in the abundance of consumers, and secondly, by trait-mediated effects through alterations in the consumers' behaviour (Greig & McIntosh, 2006).

The introduction of the brown trout (*Salmo trutta*) in 1867 (Olsson *et al*, 2006) has been implicated in the reduction of a number of native fish species in New Zealand (Glova, 2003; Townsend, 2003). The presence of brown trout has also caused changes in habitat use by some fish and invertebrate species; inanga have been found to occupy pools less in the presence of brown trout (Glova, 2003), while mayflies *Deleatidium spp.* and *Nesameletus spp.*, which commonly graze on microscopic algae, were found to feed more at night in streams that had trout compared with troutless streams (Townsend, 2003). The introduction of brown trout has therefore affected the habitat use and feeding behaviour of native species, but it has also affected the instream process of leaf litter breakdown. The abundance of *Zelandopsyche ingens*, an obligate shredder, was found to be reduced in the presence of brown trout with consequent reduction in the amount of CPOM and FPOM available for other species (Greig & McIntosh, 2006). This reduction in CPOM was a result of shredding invertebrates being consumed.

The endemic koura (*Paranephrops* spp) is negatively associated with the presence of brown trout (Usio & Townsend, 2000; Olsson *et al*, 2006). Studies on *Camarus bartonii*, a freshwater crayfish, in the headwater streams of Allegheny Plateau, United States of America, found that large crayfish made up a higher proportion of the crayfish population at sites with high fish biomass (Seiler & Turner, 2004). This is consistent with the hypothesis that fish predation on small crayfish may be limiting crayfish population size in some streams (Seiler & Turner, 2004).

Koura (*P. zealandicus*) are not able to detect chemical cues emitted by trout, meaning that they are more susceptible to trout predation (Shave *et al*, 1994). The presence of the longfin eel (*Anguilla dieffenbachii*) however, is detected by koura via the presence of chemical cues emitted from the eels' skin (Shave *et al*, 1994). As a result koura behaviour changes to reflect the presence of the eel predator.

The following experiment is designed to examine the effects of two top predator species (brown trout and eel species) on the rate of leaf breakdown by koura. The ability of koura to detect eels should result in koura feeding less in the presence of eels when compared to feeding in the presence of brown trout.

Methods:

The experiments were conducted in three troughs holding 600 litres of water. Each of the troughs had a surface area of 3 m^2. Fifty kilograms of substrate ranging from small cobbles through to large cobbles was placed at one end of each of the troughs (Appendix 1).

The troughs were filled with bore water. A pump in each of the troughs circulated water at 25 litres per minute within each trough. The pump outflow was placed at the top of the rocks to replicate a waterfall in a stream, and the pump itself was placed at the opposite end of the waterfall to allow for a straight flow of water (similar to flow in a stream).

Each of the troughs had three leaf packs placed on the bottom with rocks on the ends to hold them in position. The leaf packs were constructed using plastic mesh (5mm holes), 15cm by 9cm in size and filled with 10gms (+/- 10%) of wet willow (*Salix matsudana*) leaves. The willow leaves were collected from the substrate of the Turitea Stream, meaning that the leaves had been colonized by microbes. Average dry weights of an additional three leaf packs served as initial dry weight of leaf packs, similar to Usio and Townsend, (2001).

Koura were collected from tributaries of the Mangaore Stream, Shannon, New Zealand (fig 2). All sizes of koura were collected except for Young of Year (YOY), as they are known to be carnivores at this age. Egg-bearing and koura with missing pinchers were also avoided. The koura were kept in tanks at constant room temperature (12^0C) at Massey University until they were required.

Trout ranging in fork length from 45 to 57cm and eels ranging in length from 65 to 110 cm were caught from the Turitea Stream (fig 2) by spotlighting. Trout and eels were put in purpose built enclosures that were kept in the stream until they were required.

Twenty one koura were placed in each of the troughs (i.e. seven per m^{-2}). The size distribution of koura placed into each of the three troughs was kept similar. One adult trout or eel was also added to two of these enclosures; the third trough being left without trout or eel to act as a control.

The koura, trout and leaf packs were added to the troughs at the same time and bird netting was placed over the top of the troughs to prevent koura and trout from escaping or being predated on by birds. Each experiment was left to run for 7 nights, after which the koura, trout and leaf packs were removed from the enclosures. Leaf packs were taken from the field and frozen for later analysis.

The experiments were repeated six times with new study animals and leaf packs each time and the three enclosures were randomly reallocated throughout the experiments. All trout, eels and koura were returned to their original place of capture upon completion of the experiments.

Leaf pack processing:

The contents of each leaf pack were removed and the leaf matter dried at 35^{0}C for 7 days, weighed and then ashed at 500^{0}C for 3 hours and reweighed.

Statistics:

A One Way Analysis of Variance (ANOVA) with a Tukey HSD All Pairwise comparison test was carried out using Statsitix 8.1 to see if there was any difference between the leaf processing rates in the different treatments.

Results:

There was no koura, trout or eel mortality.

There was a significant difference in the breakdown of leaf matter between all three treatments ($F_{2,15} = 38.6$, $P < 0.001$) (Fig 1). The Tukey HSD also showed that there was a significant difference between all three treatments. The control treatment had significantly more leaf matter processed than treatments with trout or eels present. Also koura processed significantly more leaf matter in the presence of eels than in the presence of trout.

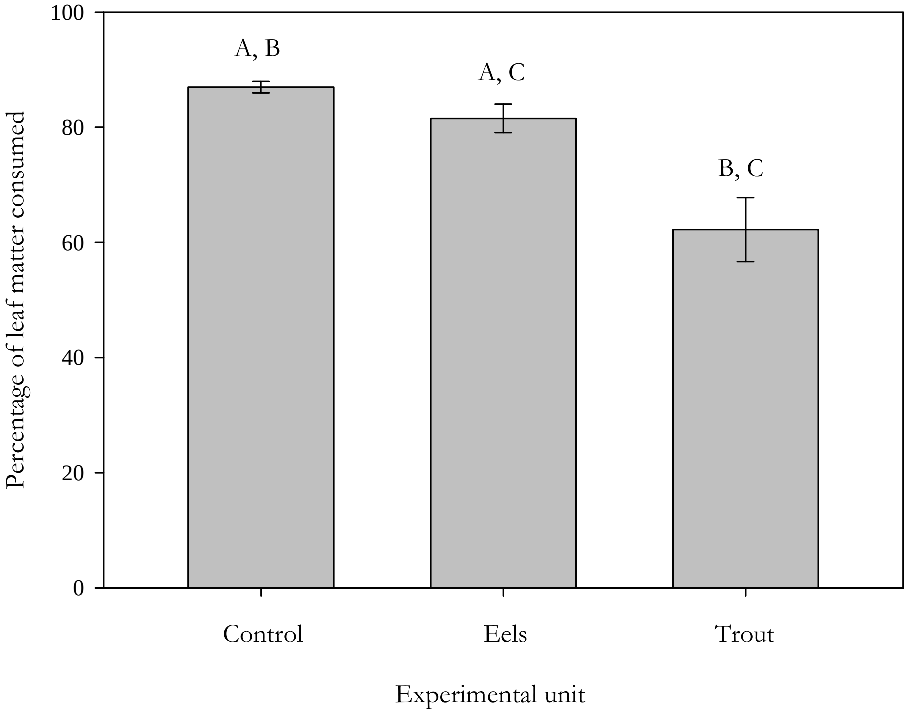


Figure 1: The average percentage of leaf matter consumed by koura at a density of 7 per square metre, in the presence of an introduced and native predator. The corresponding letters indicate a significant difference between the treatments.

Discussion:

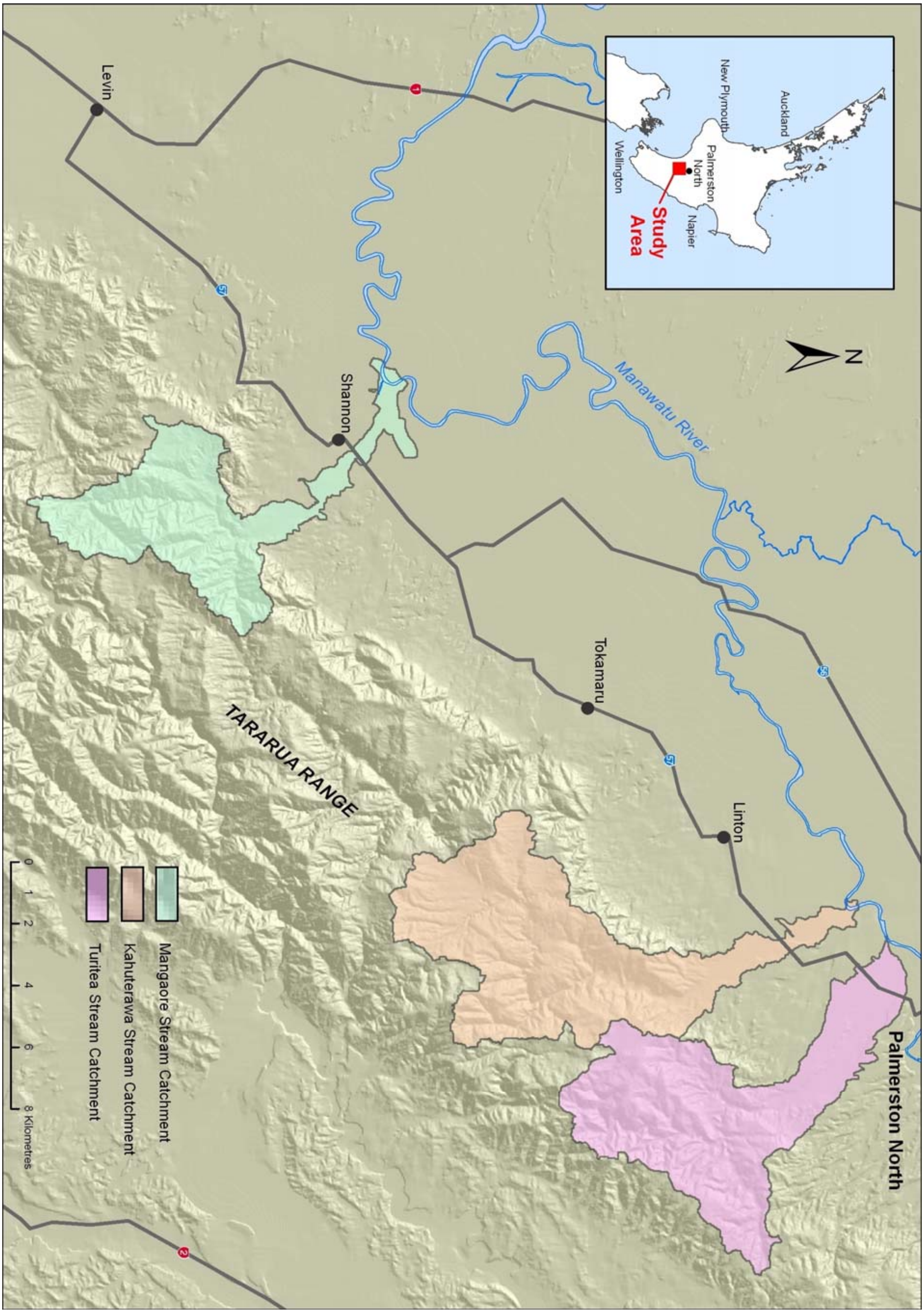
The introduction of brown trout into New Zealand waterways has resulted in the reduction of some fish species (Townsend, 2003). Koura have not been exempt from the effects of brown trout and there is a negative association between trout presence and koura density (Usio & Townsend, 2000; Olsson *et al*, 2006). This effect is also seen in streams which have trout barriers, with koura being almost absent or in low numbers below the barriers and being found at higher densities above the barriers (Usio & Townsend, 2000). Brown trout have been found to consume crayfish (*Pacifastacus leniusculus*), with small crayfish having been found to make up 27% of the stomach contents of stream-living brown trout (Nystrom *et al*, 2006). My experimental results show that the feeding behaviour of koura was reduced by the presence of brown trout, even though they are not able to detect chemical cues that are emitted by brown trout (Shave *et al*, 1994). This change in feeding behaviour was not due to predation on the koura, as all koura were recovered at the conclusion of the experiments. It was achieved by brown trout affecting the feeding behaviour of koura, as the trout and koura were always in the same enclosure. This may not accurately reflect the natural situation as trout move around in the stream, meaning that koura may be able to come out and feed when the risk of predation from trout has been reduced.

Koura have been identified as performing a vital role in the processing of Particulate Organic Matter (POM) (Usio & Townsend, 2001), with *P. zealandicus* comprising 99%, on average, of the total biomass of shredder invertebrates in Silver Stream, New Zealand (Usio & Townsend, 2001). The second highest biomass was the stonefly *Austroperla cyrene* at only 0.62%. The role that koura play in streams may have therefore been underestimated, and my study shows that the introduction of brown trout may change the cycle of POM production within streams in New Zealand. Many studies have focused on the negative correlation between the presence of brown trout and koura (Usio & Townsend, 2000; Olsson *et al*, 2006), but fail to consider the effect that brown trout may be having on koura feeding behaviour. Although it has been shown that koura are able to detect the native predator *Anguilla dieffenbachii* through chemical cues, they were unable to do the same for the introduced brown trout (Shave *et al*, 1994). This study shows that even though koura could not detect the brown trout through chemical cues, trout presence affected koura feeding behaviour. This was most likely through visual cues and the close proximity that koura and brown trout were kept in during the experiments.

This study's findings are similar to Nystrom (2005), who found that although the presence of fish did not always lead to lethal effects, fish presence may have indirect effects by causing reductions in growth by affecting behaviour and feeding (Nystrom, 2005). The growth of koura in this study was not assessed but it would be interesting to see if the koura in the control maintained weight better than those in the experimental troughs. Reduced activity by crayfish in the presence of predators has been found (Blake & Hart, 1993; Mather & Stein, 1993), to lead to decreased food intake and lower growth rates, and eventually higher mortality rates (Stein & Magnusson, 1976; Hill & Lodge, 1999). The reduction in the processing of leaf matter was not through a reduction in the number of koura in the enclosures, but was indirect being affected by the way in which koura behaved. This is the opposite from the case of *Zelandopsyche ingens* where the reduction in FPOM production was due to the predation of trout on the shredder (Greig & McIntosh, 2006). The change in feeding behaviour was more pronounced in the presence of the brown trout than of the native predator. Since koura are able to detect eels via chemical cues it was originally thought that eels would affect koura behaviour more so than trout. It could be possible that koura have developed mechanisms which allow them to feed while still being in the presence of eels, such as using the strength of chemical cues to determine how close the predator is. The effect of the reduction in the amount of CPOM and FPOM on the food chain was not considered, but could potentially have significant effects on the food supply for species that rely on CPOM and FPOM for food.

Conclusion:

The presence of brown trout affects the feeding behaviour of koura, and therefore the production of POM. Koura are predated on directly however, in this study the effects were indirect resulting from a change in the feeding behaviour of koura. This change in feeding behaviour is more pronounced in the presence of brown trout, an introduced predator, compared to eels, a native predator of koura, despite the fact that koura have the ability to detect eels through chemicals emitted in the skin mucous.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Napier
Wellington
Study Area
Mangaore Stream Catchment
Kahuterawa Stream Catchment
Turitea Stream Catchment
0 1 2 4 6 8 Kilometres

Figure 2: Map showing the catchment boundaries of the Mangaore, Kahuterawa and the Turitea Stream, Manawatu.

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Appendix 1

Plate 1: Experiment setup of troughs.

Plate 2: Example of trough used in experiment. Mesh covered the troughs to stop both animals escaping and aerial predation.

Plate 3: Close up of individual trough. Water is pumped to the top of the rocks (see top of picture).

Plate 4: Close up of individual trough. Orange tags represent the placement of the leaf litter packs.

Development of a leaf pack tool for estimating koura (*Paranephrops spp.*) population density.

Abstract:

The use of leaf packs to estimate koura population density was investigated in an controlled environment in the Manawatu. There was a significant difference in the amount of leaf matter that was consumed between the enclosures that contained koura and those that did not however, there was no difference in the amount of leaf matter that was consumed between the enclosures with high and low densities of koura. Leaf decomposition rate is therefore not likely to prove an effective sampling method.

Introduction:

Estimates of population sizes are frequently made using mark-recapture methods (Jones & Coulson, 2006; Olsen & Vollestad, 2001; Bueno, Shimizu & Rocha, 2007; Jensen, 1992; McCartney, Armstrong, Gwynne, Kelly, & Barker, 2006). The use of mark-recapture methods relies on a number of assumptions about the population that are not necessarily met in natural environments and can therefore make estimating population size difficult and potentially unreliable. Some of these assumptions include:

- Closure by boundary, animals do not move in or out of the area that is being trapped.
- The size of the population is constant over the period of investigation: no recruitment (birth or immigration) or losses (death or emigration) occur.
- No variation in capture probabilities as a function of capture history (e.g. trap happy or shy animals).

Many of the current practices for mark-recapture have been developed for vertebrate species with recent developments looking at terrestrial invertebrates however, few studies look directly at stream invertebrates.

Population estimates have been attempted with the endemic freshwater crayfish (*Paranephrops planifrons* or koura) in some streams (Rabeni *et al*, 1997) using a variety of different sampling techniques. Mark-recapture was found to be useful in this study if numerous visits were going to be made to a site. This used in conjunction with electro-fishing was found to result in the highest abundance estimates (Rabeni *et al*, 1997). The use of a variety of survey techniques will therefore give an indication of the population dynamics within a study population. This is fine in long term studies at a particular site, but more difficult for one-off surveys. All survey methods involving koura have a bias of some sort. Electro-fishing relies on the stream being clear to prevent the sampler becoming tangled in the vegetation and baited traps tend to favour aggressive males which prevent other individuals from entering the traps (Rabeni *et al*, 1997).

The purpose of this study was to try and develop a quick population estimate tool. This tool relies on the feeding rates of koura providing an estimate of the population density within study reaches. Development of such a tool may allow study of koura populations in streams for which conventional methods may not be appropriate.

Methods:

The experiments were conducted in three troughs holding 600 litres of water. Each of the troughs had a surface area of 3 m^2. Fifty kilograms of substrate ranging from small cobbles through to large cobbles was placed at one end of each of the troughs (refer Chapter 3 Appendix 1 for setup).

The troughs were filled with river water taken from the Manawatu River. A pump in each of the troughs circulated water at 25 litres per minute within each trough. The pump outflow was placed at the top of the rocks to replicate a waterfall in the stream, and the pump itself was placed at the opposite end of the trough to allow for a straight flow of water (similar to flow in a stream).

Each of the troughs had three leaf packs placed on the bottom with rocks on the ends to hold them in position. The leaf packs were constructed using plastic mesh (5mm holes), 15cm by 9cm in size and filled with 10gms (+/- 10%) of wet willow (*Salix matsudana*) and polar (*Populus alba cv. Nivea*) leaves. The willow and polar leaves were collected from the substrate of the Turitea Stream, meaning that the leaves had been colonized by microbes.

Average dry weights of an additional three leaf packs served as initial dry weight of leaf packs, similar to Usio and Townsend, (2001).

Koura were collected from tributaries of the Mangaore Stream, Shannon, New Zealand (fig 2). All sizes of koura were collected except for Young of Year as they are recorded as mostly eating carnivore matter. Egg-bearing and koura with missing pinchers were also avoided. The koura were kept in tanks at constant room temperature (12^0C) at Massey University until they were required.

Three different koura densities were used in each of the experiments; High density had 27 koura or 9 koura per square metre, low density had 9 koura or 3 koura per square metre and the control had no koura.

The koura and leaf packs were added to the water at the same time and bird netting was placed over the top of the troughs to prevent koura from escaping or being predated on by birds. Each experiment was left to run for 7 nights then the koura and leaf packs were removed from the enclosures. Leaf packs were taken from the field and frozen for later analysis.

The experiments were repeated three times with new study animals and leaf packs each time and the three enclosures were randomly reallocated throughout the experiments. All koura were returned to their original place of capture upon completion of the experiments.

The contents of the leaf pack were removed and the leaf matter was dried at 35^0C for 7 days, weighed and then ashed at 500^0C for 3 hours and reweighed.

Statistics:

A One Way Analysis of Variance (ANOVA) with a Tukey HSD All Pairwise comparison test was carried out using Statistix 8.1 to see if there was a difference between the leaf processing rates in the different treatments.

Results:

At the conclusion of the trials all koura were recovered alive from the enclosures.

There was a significant difference between the treatments ($F_{2,15} = 21.2$, $P < 0.001$) (Fig 1). The Tukey HSD showed that there was a significant difference between the high and low density treatments compared with the control treatment but there was no significant difference between the high and low density treatments themselves. Figure 1 shows the average percentage (+/- 1SE) of leaf matter consumed over the trial in the various density enclosures.

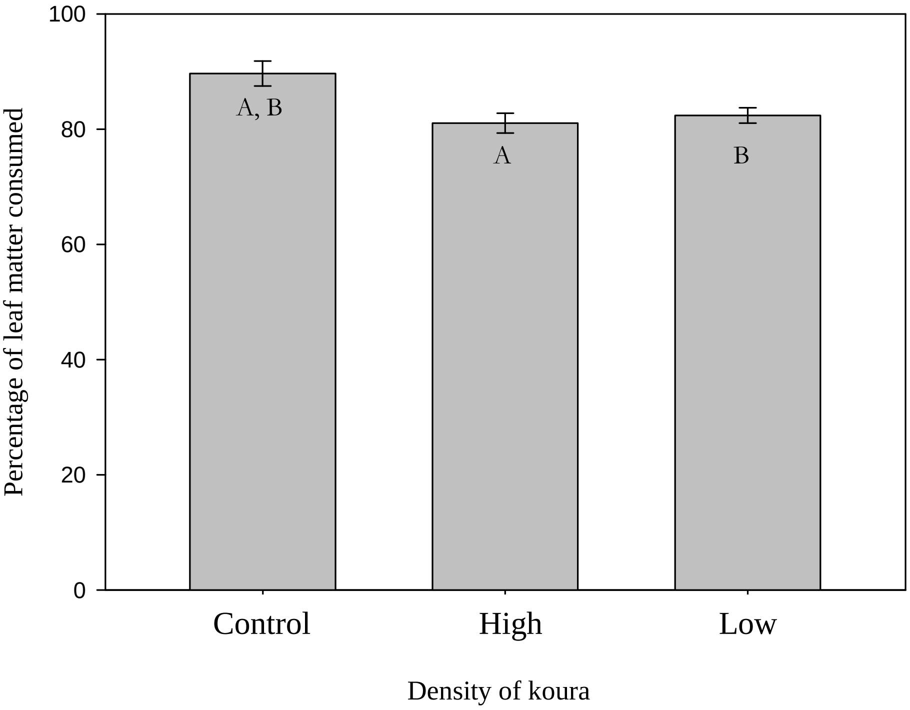

Figure 1: The average percentage (+/- 1SE) of leaf matter consumed in the various density trials. The corresponding letters indicate a significant difference between the different treatments.

Discussion:

The presence of koura resulted in leaf breakdown. The density of koura within the enclosures failed to show any effect on the rate at which the leaves were processed. The difference in density between the two treatments is large with three times the number of koura in the high density enclosures compared to the low density enclosures. With such a difference in density one would expect a difference in the amount of leaf litter that was consumed. Two possible reasons for no difference between the densities are:

- The time the experiments were run for was not long enough to allow the koura to adapt to this food source. This is highly unlikely though, given that in Chapter 3 experiments showed a significant difference between treatments which all contained a lower density of koura than the high density treatment in this experiment but were run for the same amount of time.
- A dominant koura may have been using the leaf packs as a territory preventing other koura from being able to get close to the food supply. However, one would expect this to be consistent across low and high densities, although high densities may mean that koura are more defensive of their territories preventing others from accessing food.

Using leaf pack breakdown rates is of little use as a tool for estimating the population size of koura within a stream. The use of this tool would also fail to consider the presence of juvenile koura, which are carnivorous (Parkyn *et al*, 1997). It would also fail to take into consideration the effect that the presence of predators may have on the feeding rate of koura in the stream being studied (Chapter 3). Conventional sampling methods give more accurate estimates of population dynamics than the method considered here.

It is possible that this tool may be beneficial in cases when conventional methods cannot be used, for example, in tannin-stained streams or where only presence/absence information is required. However, even then, electro-fishing or setting traps may give a more accurate description and be performed in a more timely manner as there is no need to make leaf packs and spend time analysing the amount of leaf matter consumed. The effect that invertebrates, other than koura, have on the processing of leaves was not considered in the above experiments. However, if it had been found that leaf packs in controlled environments could indicate koura densities then this would have formed the next part of the experiments.

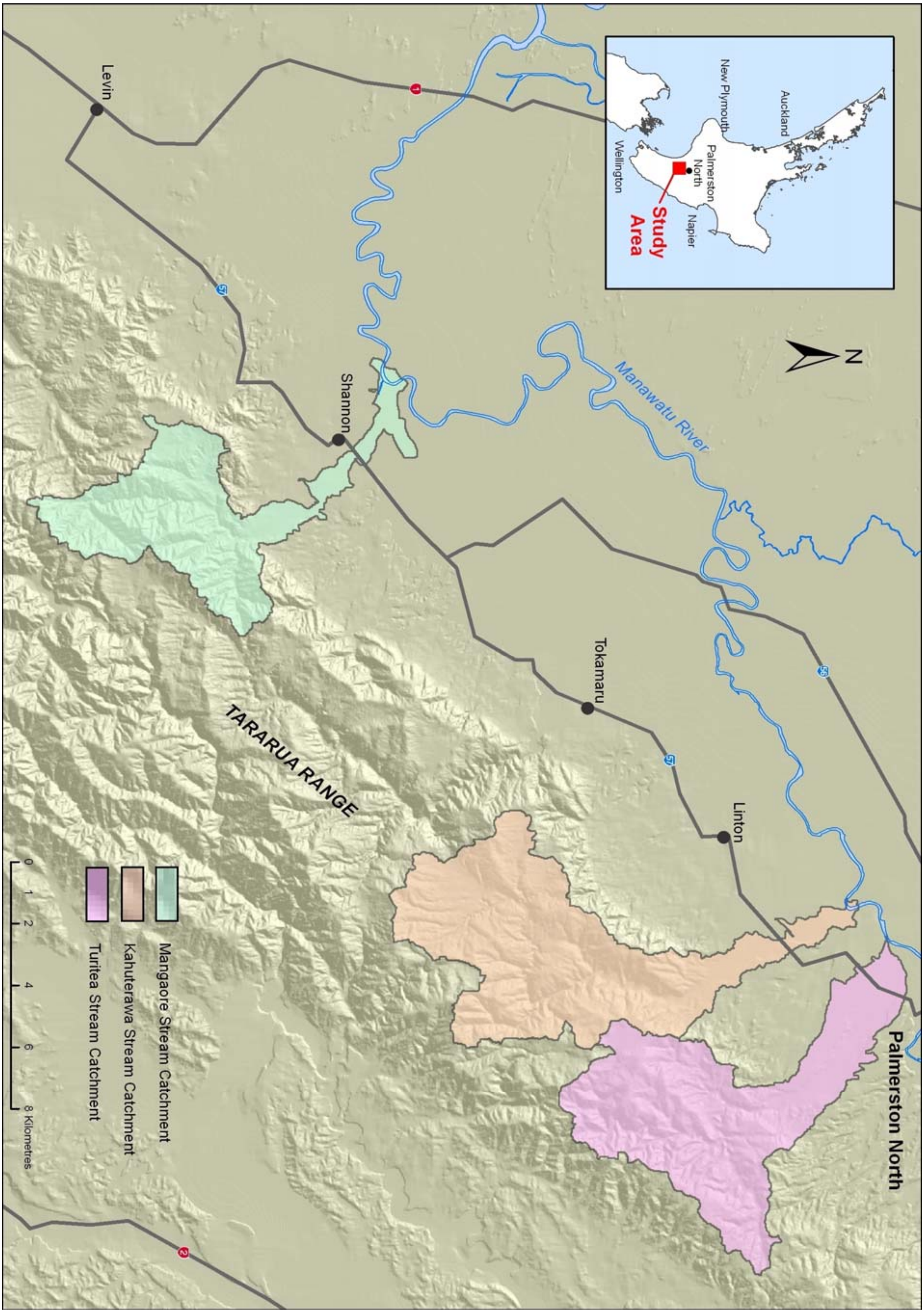

Figure 2: Map showing the catchment boundaries of the Mangaore, Kahuterawa and the Turitea Stream, Manawatu.

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General Discussion:

Habitat variables were not found to be good predictors of the presence or absence of koura at new sites. If the information collected is used as training data the regression trees are able to learn most of the sites and correctly classify them. When looking at the regression trees which have both GIS information and on site habitat variables there are similarities in the variables which are seen as being important. The presence of eels and trout were important in distinguishing sites with and without koura, with these species being known predators of koura. Interestingly, the presence of small trout was most closely linked with koura presence/ absence. In Chapter Three I showed that the presence of trout affects the rate at which koura feed on leaf matter. Therefore trout effects on koura can be both direct and indirect. Other habitat variables relate to riparian vegetation, the amount of vegetation found in the study area and the pool-riffle-run sequence of the study site in question. The presence of pools and shutes was seen as an important factor aiding in the differentiation between sites with koura present or absent. Pools tend to capture more leaf litter and also allow the growth of aquatic plants, both important factors for the presence of koura. Furthermore shutes tend to prevent the movement of trout into areas where koura currently lack this predation threat. The current wintertime equilibrium temperature was also important in predicting koura presence/absence and this is likely to relate to the fact that koura rely on temperature of the stream to be active, metabolize food and grow. Temperatures which are too low are likely to lead to death of koura.

The introduction of brown trout into New Zealand has had adverse effects on the wildlife found within streams. Koura have been found to be negatively associated with the presence of brown trout. Experiments in this thesis failed to show the consumption of koura by brown trout kept in enclosures. Observations made from Lake Taupo show that the shape and the flesh colour of brown trout that consume koura change. Possible reasons for this include:

- Stream temperatures climbing towards the level at which brown trout are known to stop feeding.
- The availability of other food sources in the form of invertebrates in drift which may have been taken rather in preference to actively hunting prey.

- The experimental timeframe may not have been long enough to allow brown trout to adjust to the new conditions.
- The substrate placed in the enclosures wasn't as tightly packed as the stream bed allowing koura to hide and therefore decreasing the probability that brown trout would predate on them.
- The absence of Young of Year (YOY) koura in the enclosures. The koura in the enclosures may not have been representative of the normal size of koura predated on in the wild.
- A combination of the above factors may have been responsible.

Compared to the effects that brown trout have on koura in some species of crayfish, it has been found that crayfish actively predate on eggs and fry of trout. This predation by crayfish on trout in these experiments was governed by the substrate size in the enclosures. This thesis found that koura do not actively predate on juvenile brown trout or adult Crans' bullies and even in the presence of dead brown trout the consumption rate was low. The energy required for koura to be able to capture live fish prey is likely to be high due to their slow movement and hence the reason we saw a lack of predation on live fish in these experiments but the scavenging on dead fish.

As discussed briefly above the presence of a predator has been shown to affect the rate at which koura are able to process leaf matter into Coarse and Fine Particulate Organic Matter (CPOM and FPOM, respectively). The presence of koura has been shown to be important in the processing of leaf matter, with koura making up 99% of the shredder biomass in Silver Stream in Otago. The results from this study show that the effects of brown trout are indirect by reducing the feeding activity of koura and therefore reducing the production of CPOM and FPOM. An interesting point found in this study was the fact that the feeding activity of koura was significantly different in the presence of eel compared to brown trout. Eels have been shown to emit chemicals through their skin which koura are able to detect and consequently modify their behaviour in the presence of this predator species. This was not found to be the case for brown trout, although in this study it was the presence of brown trout which had greatly reduced the amount of leaf matter that was consumed by koura. Maybe in the presence of eels koura can modify behaviour depending on the strength of the chemicals emitted, strong chemical cues meaning the predator is close and weak chemical cues that the predator is a small threat.

In the last chapter I trialled a new method for establishing the presence or absence of koura in a stream. However, the continued use of conventional sampling methods for establishing koura presence/absence in streams and rivers will be more successful than using leaf packs as a surrogate. The use of leaf packs would fail to consider the size distribution of the population as the koura would not be captured to measure, and the Young of Year (YOY) koura would not consume the leaf matter. The use of multiple methods may give a more accurate picture of the koura population within a study site.