

Between a Bass and a Hard Place: The Fragmented
Distribution of an Endangered Redfin in the Heuningnes
River System of the Cape Fold Ecoregion.

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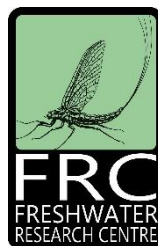
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Plagiarism Declaration

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Glossary

Acronyms:

CFE: Cape Fold Ecoregion. A freshwater ecoregion is “a large area encompassing one or more freshwater systems with a distinct assemblage of natural freshwater communities and species.” (Abell, 2008).

HRS: Heuningnes River System. Encompasses the Heuningnes River and its two major tributaries: the Kars River and the Nuwejaars River.

ODRS: Olifants-Doring River System. Encompasses the Olifants and Doring Rivers, as well as associated tributaries.

ANP: Agulhas National Park. Occupies the Southern extent of the Heuningnes River System (Russel and Impson, 2006).

VIF: Variance Inflation Factor. VIF provides a measure of multi-collinearity among variables in a multiple regression model.

YOY: Young of year. Fish born within the past year which have not yet reached one year of age.

Abstract

Native freshwater fish are globally in decline due to anthropogenic impacts, including changes to water quality, over-abstraction, climate change, and introduction of non-native species. The Cape Fold Ecoregion (CFE) of South Africa is recognised as a hotspot for freshwater fish endemism. It is also recognised as one of six hotspots for freshwater fish invasions. Hence, 36% of native and 66% of endemic freshwater fish are considered threatened (vulnerable, endangered, or critically endangered). The Heuningnes redbin (*Pseudobarbus sp Burchelli Heuningnes*) is one such fish. It is considered endangered (SANBI Red List of Threatened Species) and currently occupies a small number of tributaries within the Heuningnes River System (HRS). The Heuningnes redbin co-occurs with Cape kurpers and Cape galaxias, some of which are currently being described as new and unique lineages. As such, the HRS is of critical conservation significance. This study analysed a data set collected by Keir Lynch in March of 2018, that consists of 22 water quality, geographic, and habitat-related environmental variables. The goal was to determine whether the distribution of native fish is independent of the distribution of non-native fish, as well as what environmental variables best predict the presence and abundance of Heuningnes redbin in the HRS. The data suggest non-native species appear to be excluding native species from invaded reaches. Moreover, redbins prefer warmer, more acidic water with complex habitats in the form of woody debris and gravel substrate, allowing them to hide from predators. Their abundance is negatively correlated with phosphorous, which may indicate that they are affected by pollution from agricultural runoff. Moving forward, conservation practitioners should consider non-native eradication projects, through either chemical or mechanical removal. Additionally, they should also consider creating a buffer of indigenous vegetation, which could mitigate the impacts of agricultural effluents and provide benefits for the currently endangered renosterveld. Additional surveys are necessary to determine the exact extent bass have invaded the system, as well as to obtain an accurate measure of how different land uses are affecting the distribution of native fishes.

1. Introduction

1.1 Threats to freshwater systems

The biodiversity crisis is most clearly characterized by freshwater ecosystems (Tickner et al., 2020). In recent decades there has been a growing realization that freshwater systems are among the most threatened on the planet (Ricciardi and Rasmussen, 1999; Dudgeon et al., 2006; van der Walt et al., 2019; Almond et al., 2022). These systems are threatened by a range of anthropogenic threats, including reduction of water quality and quantity, habitat degradation, overexploitation, and introduction of non-native species (Dudgeon et al., 2006; Tickner et al., 2020). Domestic and industrial point sources introduce excessive nutrient enrichment, toxic chemicals and other compounds, such as endocrine disruptors, reducing water quality (Smith, 2003). Additionally, abstraction for domestic and agricultural activities reduce the quantity of water available for freshwater ecosystems (Dudgeon et al., 2020). Non-native species have been accidentally introduced, or deliberately stocked for angling purposes (Dudgeon et al., 2006). Furthermore, climate-change related impacts, such as changes in temperature, patterns of precipitation, and flow dynamics indirectly exacerbate these threats (Tickner et al., 2020). In essence, “The trade-offs between human use of water, and the water needed for nature, have increasingly been skewed in favour of the former” (Dudgeon, 2020).

Meanwhile, freshwater systems host disproportionately high numbers of species relative to their surface area (Szabolcs et al., 2022). Freshwater species make up 9.5% of known animal species on Earth (Dudgeon, 2019). Freshwater fish account for almost 25% of all vertebrate species (Hughes, 2021). Indeed, of all known species of fish, 51% live in freshwater (Hughes, 2021). Nevertheless, freshwater hotspots receive less attention than their terrestrial counterparts (Myers et al., 1990). Freshwater systems contribute critical services to human well-being through provision of food and water for direct consumption and agricultural irrigation, while simultaneously performing service functions, such as water purification and flood regulation (Carrizo et al., 2017).

Roughly 3% of Earth’s water is fresh, two-thirds of which is frozen at the poles and the remaining third mostly deep underground, leaving between 0.1 and 0.3% available to sustain humans and ecosystems, and serve as habitat for freshwater

organisms (Gleick, 1996). This tiny fraction of water is increasingly monopolised by humans, despite the overwhelming evidence that this has resulted in declines of freshwater biodiversity. Population trends for monitored freshwater species have declined on average by 83% since 1970, compared to an average decline of 69% among terrestrial species (Almond et al., 2022). Many fish species have become extinct, or are highly endangered, particularly in rivers within arid and semi-arid regions, where heavy human demand is placed on freshwater resources (Collares-Pereira and Cowx, 2004).

Rivers and wetlands are naturally isolated and heterogenous ecosystems. These conditions have contributed to the incredible biodiversity found in fresh water systems, but also result in a high proportion of freshwater species being endemic, rare, or range-restricted, making them particularly vulnerable to human-induced perturbations (Magurran, 2009). The functional and compositional heterogeneity of global freshwater systems means that their responses to anthropogenic threats are not uniform across space. Freshwater fish face different threats in different parts of the world. For example, drought is considered the major threat to threatened fish species in the Mediterranean (75%; Smith and Darwall, 2006). In South America, both non-native invasives and over-harvesting resulting from industrial aquaculture are considered the most immediate threats to threatened native species (>30%; Bezerra et al., 2019), while invasive species are affecting close to 85% of threatened species in South Africa (Darwall et al., 2009a).

1.2 Freshwater fish in South Africa

Freshwater fish are the most imperilled taxonomic group in South Africa, with 36% of native species, and 66% of endemic species, listed as threatened, referring to vulnerable, endangered or critically endangered on the IUCN Red List of Threatened Species (Skowno et al., 2018). Freshwater fish are declining due to the pressures of habitat loss, pollution, poor management of dams, and invasive species (Skowno et al., 2018). These anthropogenic impacts have led to the fragmentation of remnant populations, which tend to be confined to mountain tributaries that are not disturbed, degraded, or invaded (Clark et al. 2009; Ellender et al. 2011). Of the various and pervasive threats facing freshwater fish in South Africa, invasive fish are considered the most serious (Cowx 2002). South Africa is in fact one of six listed global hotspots

for fish invasions, with at least 55 fish species introduced into non-native areas since 1726 (Ellender and Weyl, 2014).

The Western Cape province of South Africa is a Mediterranean-like climatic region, recognised as a hotspot of freshwater fish endemism (Ellender et al., 2017). The Mediterranean climate region of this province is encapsulated by the Cape Fold Ecoregion (CFE). The geological and climatic history of the region, as well as the frequency of small and isolated rivers, leads to remarkable levels of endemism (Darwall et al., 2009b). The endemic freshwater fish of the CFE typically have very narrow distribution ranges, with most restricted to single river systems, or single tributaries within river systems (Chakona & Swartz 2013; Chakona et al. 2013; Chakona et al. 2018). Surveys have highlighted the fact that the mainstem populations of many CFE endemics have already been extirpated (van der Walt et al. 2016).

Jordaan et al. (2020) indicated that 84% of native freshwater fish taxa are considered to be under-protected. Similar to the rest of South Africa, non-native fish are the most immediate threat to endemic fish in the CFE (Darwall et al., 2009b). A total of 15 non-native freshwater species have become established in the region (Jordaan et al., 2020). The densities and diversity of native freshwater fishes are negatively affected through competition with and predation by non-native species (Ellender & Weyl 2014). Invasive bass (*Micropterus spp.*) in particular have eradicated native fish populations in the CFE from downstream reaches of rivers (Shelton et al., 2014).

The widespread occurrence and pervasiveness of threats to native fish in the CFE highlight the urgent need for on-the-ground conservation interventions to prevent imminent freshwater fish extinctions (Strayer and Dudgeon, 2010). Moreover, the benefits of conserving these fish will have benefits for other freshwater organisms and hence freshwater ecosystems as a whole. Unfortunately, freshwater conservation capacity in South Africa is severely limited (Impson et al., 2007). It is of critical importance that comprehensive monitoring and surveying programs are developed in order to inform effective conservation interventions. Such data are often lacking, however, due to limited funding and appropriate ecological management (Impson, 2016). Therefore, there is a need to strategically focus limited

resources on high priority interventions to conserve threatened Cape fish and their habitats.

1.3 Freshwater fishes of the Heuningnes River System

Molecular markers have been utilized in recent years to assess the genetic diversity of freshwater fish (Chakona et al., 2013; Ellender et al., 2017; Swartz et al., 2009; Swartz et al., 2004). These studies have shown that the diversity of freshwater fish in the CFE has been underestimated. For example, Swartz et al. (2007a, 2009, 2014) studied the phylogenetic relationships of the *Pseudobarbus* genus and the distribution of genetic lineages in the CFE. Three historically-isolated lineages in *P. burchelli* were identified (Swartz et al. 2007a; 2014). Ellender et al. (2017) describe the lineages as *Pseudobarbus burchelli*, restricted to the Tradouw and Huis Rivers in the Breede River System (Tradouw redfin); *Pseudobarbus sp. "burchelli Breede"* (Breede redfin), limited to the headwater tributaries in the Breede, Duiwenhoks, and Goukou River systems; and *Pseudobarbus sp. "burchelli Heuningnes"* (Heuningnes redfin) which is restricted to the Heuningnes River System (HRS) (Figure 1). The deeply divergent and strongly supported lineages in Tradouw, Breede and Heuningnes redfin will result in these being described as distinct species (Ellender et al. 2017; Swartz et al. 2014).

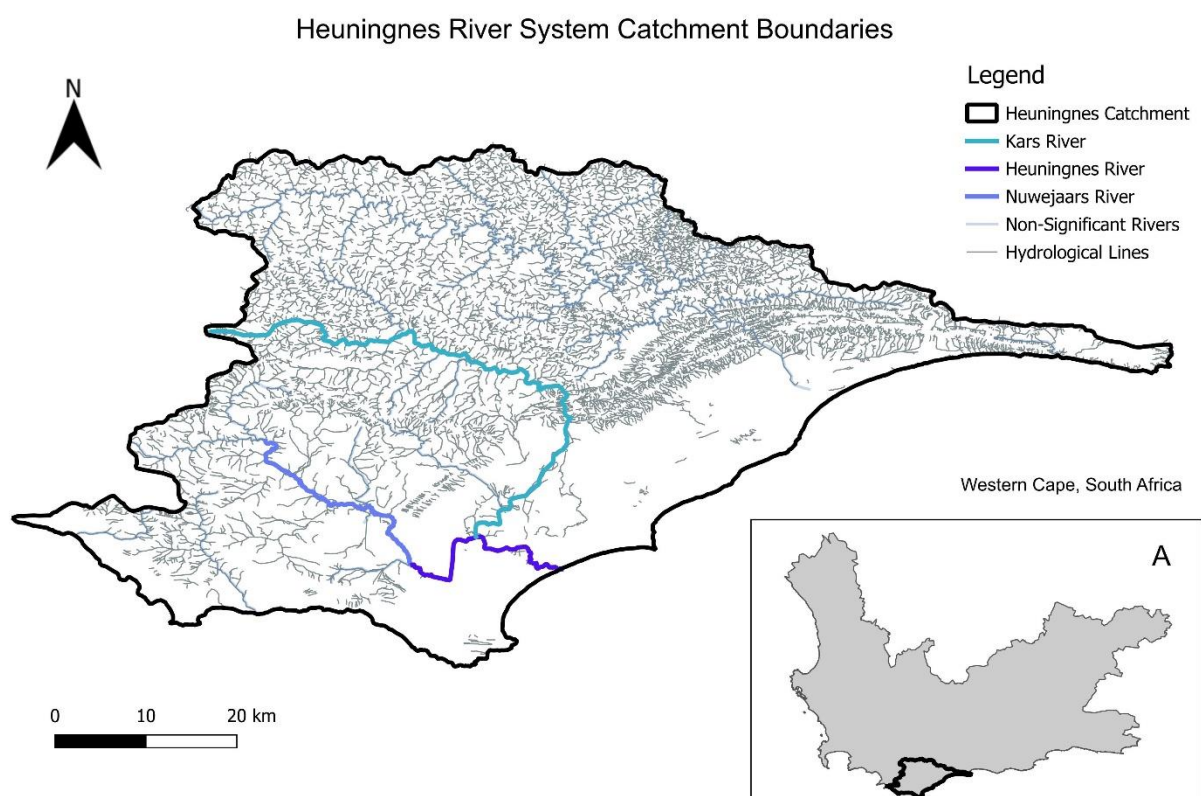


Figure 1: The Western Cape province of South Africa (A), with the HRS at its southern extent expanded to show tributaries.

Russel and Impson (2006) performed a survey of the HRS and found three species of non-native freshwater fish, namely Common carp (*Cyprinus carpio*), bluegill sunfish (*Lepomis macrochirus*), and Spotted bass (*Micropterus punctulatus*). The latter two are widespread and common. Common carp and Spotted bass were introduced into this river system as angling and food fish in the late 19th and early 20th centuries, while Bluegill sunfish were introduced shortly after bass as a fodder fish, when it became apparent that bass had eliminated the smaller indigenous fish species (Russel and Impson, 2006). Where these invasive species occur, their impact on native fish has likely been severe. Non-native fish consume the eggs of native fish, degrade habitat by both removing aquatic plants and increasing turbidity, and compete with and prey upon native fish, leading to poor recruitment and localised extirpations (Russel and Impson, 2006).

The Heuningnes redbin (*Pseudobarbus sp. burchelli heuningnes*) is listed as endangered on the South African National Biodiversity Institute (SANBI) Red List of

South African Species (SANBI Red List, 2016). Two other native fish co-occur with this species, including two Cape galaxias (*Galaxia zebratus* and *Galaxia sp.nov. zebratus heuningnes*) and two Cape kurpers (*Sandelia capensis* and *Sandelia sp.nov. capensis heuningnes*). Cape kurper and Cape galaxia are common and widespread throughout the CFE (Skelton, 2001). The Heuningnes kurper and galaxia, however, are unique and endemic to the catchment (Ellender et al., 2017). As these populations are in decline, it is imperative to develop urgent conservation actions for the Heuningnes redbin (Swartz et al. 2014).

1.4 Study objectives

In order to effectively manage unique and threatened species like the freshwater fishes inhabiting the HRS, and prioritise associated conservation efforts, it is necessary to understand spatial distribution patterns, population status, species-habitat relationships and key threats (Huang & Frimpong 2015). As of yet, no studies have quantitatively evaluated the factors that best explain variations in the abundance of the Heuningnes redbin and co-occurring fish species. This study aims to examine patterns of native fish presence and abundance throughout the HRS in relation to the presence of other fish species and environmental variables, in order to assess the relative influence of these variables on fish presence and abundance.

In order to do so, this report will attempt to address the following questions:

1. What are the distributions of the Heuningnes redbin and co-occurring native fish species in the HRS and how do these relate to the distributions of non-native fish?
2. What environmental factors best explain the distribution of the Heuningnes redbin?
3. How can this information be used to guide future conservation interventions?

2. Methods

2.1 Study area

This study was conducted in the HRS, located roughly 220 km East of Cape Town in the Overberg Region in the Western Cape province of South Africa. The catchment lies between 34° 20' and 34° 50' S and 19° 33' and 20° 17' E, covering an area of 1,442 km² (Banda et al., 2023). Elevation ranges between sea level and approximately 840 m. The climate is characterized as Mediterranean, experiencing hot, dry summers with temperatures up to 27° C between November and March, and cold, wet winters with temperatures below 10° C between May and August (Mkunyana et al., 2019). Average annual rainfall in the catchment is 500 mm/year (Mtengwana et al., 2021).

The catchment contains the Heuningnes River, along with two major tributaries, the Kars and the Nuwejaars Rivers (Figure 1). The Nuwejaars River drains into the Soetendansvlei, a small freshwater lake, whose outlet joins the Kars River to form the Heuningnes River. Terrestrial vegetation within the catchment is extremely diverse, comprising over 1700 species, including Restioid fynbos, Dune fynbos, Limestone fynbos, Acid sand Proteoid fynbos, Ericaceous fynbos, and Renosterveld (Russel and Impson, 2006). The catchment is mostly natural vegetation, but agriculture is the second most dominant land use, with dryland farming and livestock production creating a patchy landscape (Banda et al., 2023). Urban development covers only a small proportion of land area.

2.2 Study species

2.2.1 Native species

The HRS is home to at least five native fish species, three of which are currently undergoing taxonomic revision: Heuningnes redbfin (*Pseudobarbus sp burchelli heuningnes*), Cape galaxias (*Galaxias zebratus*), Heuningnes galaxias (*Galaxias sp. nov. zebratus heuningnes*), Cape kurper (*Sandelia capensis*), and Heuningnes kurper (*Sandelia sp.nov. capensis heuningnes*). Both Cape galaxias and Cape kurper are considered Data Deficient according to the IUCN Red List of Threatened Species (Swartz et al., 2007b; Chakona, 2018). Redfin minnows (*Pseudobarbus burchelli*) as a whole are considered critically endangered by the IUCN Redlist

(Jordaan and Chakona, 2017), while the Heuningnes redbfin (Figure 2) is considered endangered according to the SANBI Red List (Chakona and Swartz, 2013). Both the Heuningnes kurper and Heuningnes galaxia are not formally described species, but are currently undergoing taxonomic revision and may be unique lineages endemic to the HRS (Chakona et al., 2013). For the purpose of this study, Cape kurpers and Cape galaxias were not distinguished from Heuningnes kurpers and Heuningnes galaxias because these lineages were not distinguished at the time of sampling. Little is known about the physiology and ecology of any of these species.



Figure 2: Image of Heuningnes redbfin. Courtesy of Fynbos Fish Trust.

Piscivorous invasive fish are considered the single most important threat facing all three native species. Where mountain streams generally have natural instream physical barriers, such as waterfalls, that prevent the spread of alien fish, coastal systems such as the Heuningnes lack such barriers (Chakona and Swartz, 2013). Additional threats include habitat destruction through bulldozing, abstraction of water for agricultural purposes and pollution from agricultural and forestry effluents.

2.2.2 Non-native species

Three non-native fish species are also found within the HRS: Spotted bass (*Micropterus punctulatus*), Bluegill sunfish (*Lepomis macrochirus*), and Common carp (*Cyprinus carpio*). Spotted bass are popular angling fish, and likely the main culprit responsible for the extirpation of native fish species in this region (Chakona and Swartz, 2013; Figure 3). Adults are primarily piscivorous, but also consume frogs and crabs (Brown et al., 2009). They thrive in areas where bluegill are also able to establish, because they utilise the smaller bluegills as an important food source (van der Walt et al., 2016).



Figure 3: Image of Spotted bass. Courtesy of Sean McVey.

Bluegill sunfish are another predatory fish, also introduced from North America (Skelton, 2001). Bluegills prefer warm, shallow, well-vegetated waters, but tolerate a wide range of water conditions (Skelton, 2001). In South Africa, they are found in the Northern Cape, Western Cape, Eastern Cape, KwaZulu-Natal, Mpumalanga and Free State and are now considered a pest, as they breed prolifically, compete with native fish for food, and prey on the young of native fish (Skelton, 2001).

Common carp were introduced to South Africa in the 18th century (Skelton, 2001). They are hardy and tolerant of a wide variety of conditions and are omnivorous, consuming a wide range of plant and animal matter. They are known to increase suspended solids in the water column, reducing water transparency and macrophyte coverage (Zambrano et al., 2006), and are considered a pest due to their destructive feeding habits.

2.3 Data collection

The data used in this study were collected from within the HRS by Keir Lynch in March of 2018. Sites were selected according to where standing water was year-round, being areas that are able to support populations of freshwater fish (Keir Lynch, pers. comm.; Figure 4).

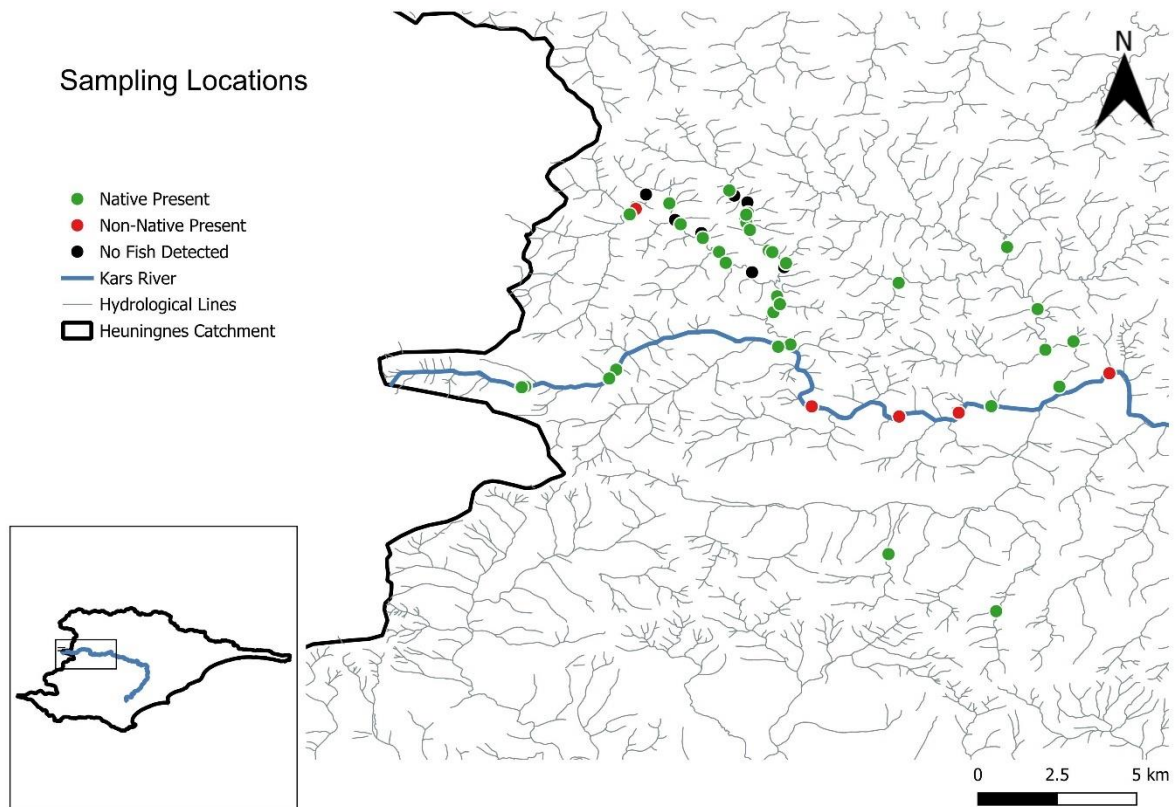


Figure 4: Sampling sites in the upper Kars River of the HRS. Sites where native fish were present are marked in green, sites where non-native fish were present are marked in red, and sites where no fish were detected are marked in black.

Once the sites were selected, co-ordinates were recorded using a GPS. Sampling was then undertaken using a two wing fyke net (Figure 5). The fyke nets were installed after 17h00, and collected before 08h00 the next morning, and were placed facing upstream so that any downstream flow of water would keep the pockets of the nets open. The position of the fyke nets depended largely on the shape and depth of the pools and presence of natural obstructions. One fyke net haul was conducted per site. Any freshwater fish caught in a survey were identified to species and grouped as native or non-native. Sampled fish were immediately returned to the river after processing.

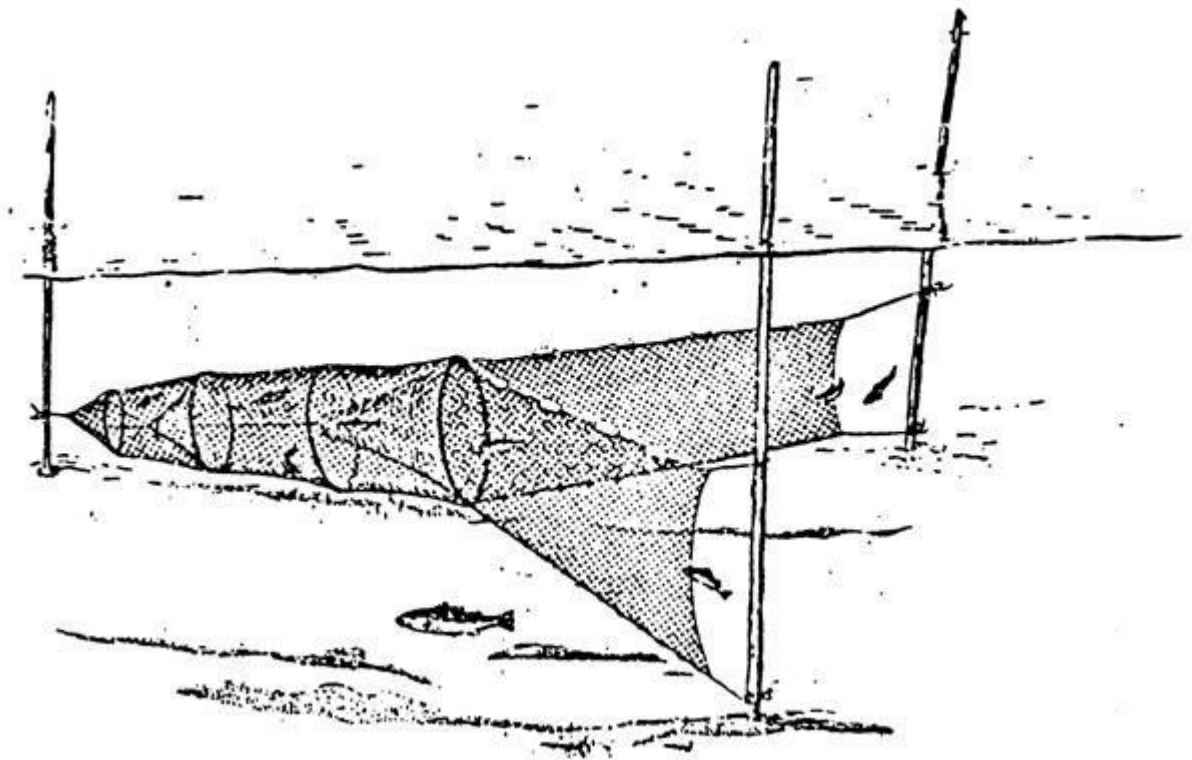


Figure 5: Diagram of a fyke net, courtesy of the Food and Agriculture Organisation of the United Nations (2023).

A range of environmental variables were sampled at each site and were split into three categories: Water quality and geographic variables (Table 1), and habitat variables (Table 2). Water quality variables were collected by taking a sample of water after fyke nets were installed, and analysed the same evening using a DR900 colorimeter.

Table 1: Water quality and geographic variables measured in 2018 at each of the 56 sites sampled by Keir Lynch.

<i>Variable Category</i>	<i>Variable</i>	<i>Unit</i>	
<i>Water Quality</i>	pH	Log X	
	EC	μS/cm	
	DO	% Saturation	
	Temp	° Celsius	
	Ammonia	mg/L	
	Phosphorus		
	Nitrite		
	Nitrate		
	Total Iron		
	Phosphonate		
	TDS		
Inorganic Nitrogen			
<i>Geographic</i>	Elevation		Meters a.s.l.
	Flow		m.s ⁻²
	Slope	%	

Habitat variables were collected using one of two methods. The first method was used to measure two variables, namely undercut banks and river width (Table 2). Firstly, five transects were placed evenly along the width of the site. Each transect was then used to assign one point to a variable class, adding up to a total of five points per variable per site. For example, the river bank of site one was undercut throughout the entirety of the site. Therefore, the class “no undercut bank” was scored one for each of the five transects, for a total score of five. In the case of Site 8, however, two transects covered an undercut bank and three transects did not. Hence, “undercut bank” was assigned a score of two, and “no undercut bank” was assigned a score of three, for a total of five (Figure 6a).

Table 2: Habitat variables measured in 2018 at each of the 56 sites sampled by Keir Lynch. Each variable is divided into between two and five classes. Bank and Width total five points, while the remaining classes sum up to 30 points.

		Variables						
		Substrate	Debris	Bank	Macrophytes	Canopy	Depth (cm)	Width (m)
Classes	Silt-sand	Woody Debris	Under cut	None	Open	Shallow (0 – 50)	Narrow (0 – 3)	
	Gravel	No Woody Debris	No Undercut	Scarce	Partial	Moderate (50 – 100)	Moderate (3 – 6)	
	Cobble	-	-	Moderate	Closed	Deep (100 – 180)	Wide (6 – 10)	
	Boulder	-	-	Abundant	-	Very Deep (>180)	Very Wide (>10)	
	Bedrock	-	-	-	-	-	-	

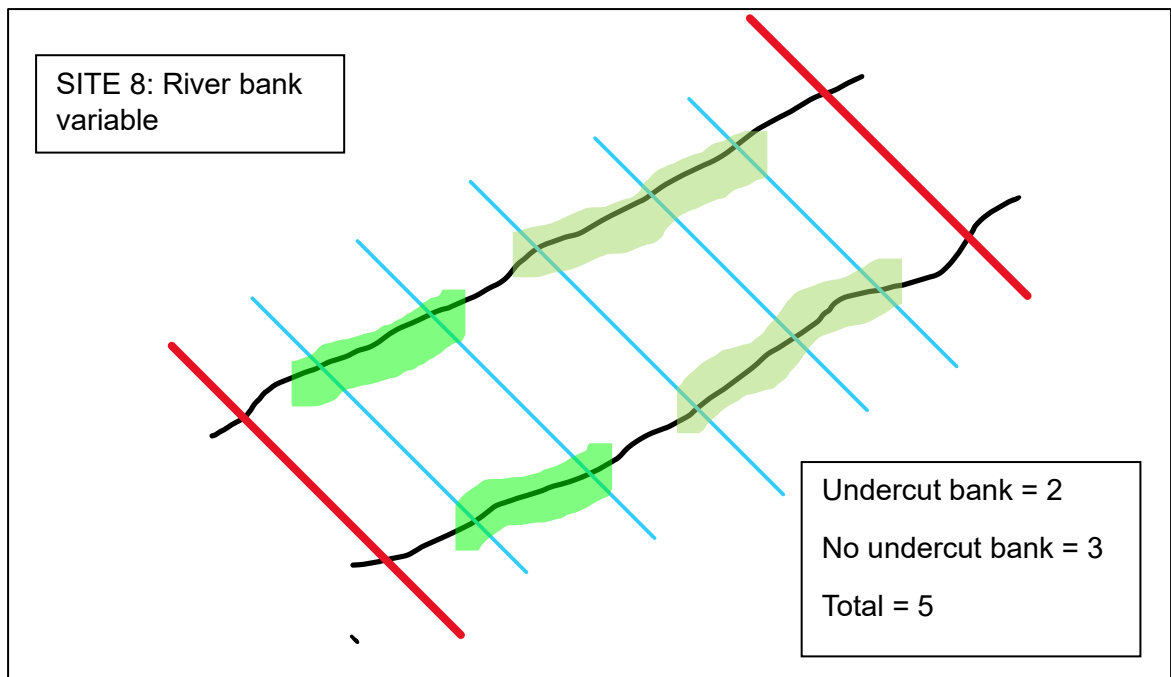


Figure 6a: Diagram of the survey method used for river bank and river width. The red lines on either end delineate the site boundaries. The black lines indicate the river bank. The blue lines across the width of the river represent the five transects. The light green shading along the river bank indicate an “Undercut bank”, while the darker green shading indicates “No undercut bank”, as is explained in the example above.

The second method was used to measure the remaining variables (Table 2). Firstly, five transects were placed evenly along the width of the site. Those transects were then divided into six equal sections, each worth one point. Each point was then assigned to one of the variable classes, resulting in a total of 30 points per variable per site. For example, river depth is divided into four classes: shallow, moderate, deep, and very deep. The depth at Site 1 was mostly moderate, given that shallow was assigned four points, and moderate was assigned 26 points, while deep and very deep were not assigned any points (Figure 6b).

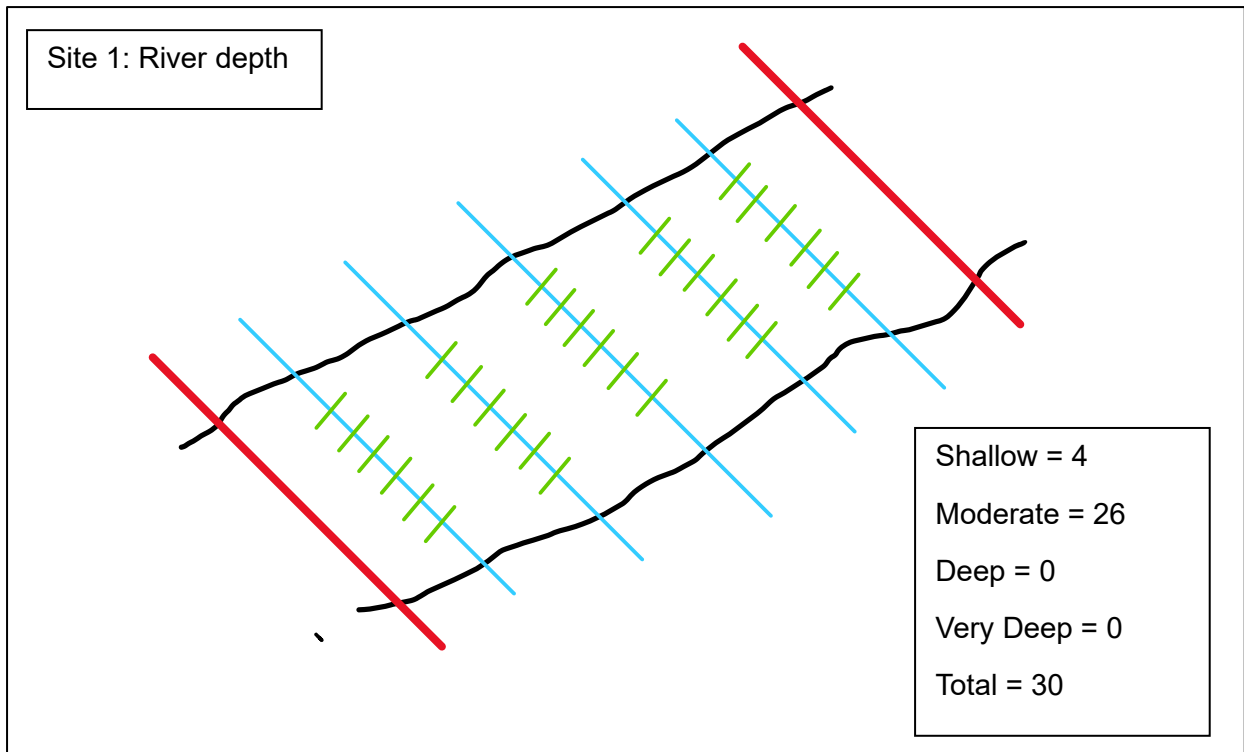


Figure 6b: Diagram of the second survey method used for the remaining river variables. The red lines on either side delineate the site boundaries. The black lines indicate the river bank. The blue lines across the width of the river represent the five transects, while the green lines represent the six points along the transect where the river depth was measured according to the example provided above. 26 points were moderate depth (50 – 100 cm), while four points were shallow (0 – 50 cm).

2.4 Data Analysis

2.4.1 Data preparation

Before analysing the data, they were standardised by replacing missing values with zeros and checking for unexpected outliers or other unusual entries. This dataset did not contain any unexpected outliers.

To simplify the analysis, the proportional values associated with habitat variables were converted into percentages. No further transformations were necessary.

2.4.2 Spatial variation in native abundance according to the presence of non-native species

Pearson's Chi-square test for independence (R package 'stats' version 4.3.2; R core team 2023) was used to assess whether there was a relationship between the presence of native fish in a site and the presence of non-native fish in that same pool.

2.4.3 Identifying top predictors of redbfin abundance

This technique was used to assess whether the presence and abundance of Heuningnes redbfin varies randomly or was correlated with environmental factors. The aim being to understand the primary factors influencing Heuningnes redbfin presence and abundance.

A Generalised Linear Mixed Model (GLMM; R package 'GLMM' version 1.4.4; Fournier et al., 2012) was used to determine the best predictors of redbfin presence and abundance (the response variable). A GLMM extends linear regression to handle a wide range of response variables and data distributions, allowing for the modelling of relationships between explanatory and response variables that follow non-normal distributions. Testing the correctness of a GLMM involves evaluating its performance and assessing how well it fits, using information criteria such as Akaike Information Criterion (AIC).

Four models were considered, accounting for different distributions and regression techniques (Table 3). Among the various models considered, the one built

using backward stepwise regression and negative binomial distribution stood out for having the lowest AIC and delta AIC values, indicating the best model fit.

Table 3: Table of the candidate models ranked from most to least informative based on their AIC and delta-AIC values

Model	AIC	Delta AIC
Negative Binomial Distribution and Stepwise Regression	213.24	0
Negative Binomial Distribution	254.17	40.93
Poisson Distribution and Stepwise Regression	878.10	664.7
Poisson Distribution	478608.57	478395.33

Generally, an assumption of GLMMs is an appropriate estimation of variance, which can be checked by comparing the magnitude of the model residuals against the theoretical variance. If this statistic is too large, then the variance is over-dispersed. In this case, a negative binomial distribution was fit to the model in order to address overdispersion under a Poisson distribution (Linden and Mäntyniemi, 2011).

Backward stepwise regression is a variable selection technique that selects a subset of predictor variables from a larger set of potential variables, based on their statistical significance and contribution to the model. The initial set of predictors included water quality, geographic, and river variables. At each step, an analysis of deviance table was generated and the variable with the highest p-value greater than 0.1 removed. All variables with a p-value lower than 0.1 were retained in the model.

Multi-collinearity among the predictor variables was checked for by using variance inflation factor (VIF) calculation. Higher VIF values indicate stronger multi-collinearity. No multi-collinearity was found in the final model.

3. Results

3.1 Surveys

A total of 56 sites were surveyed in March of 2018. Three native freshwater fish were recorded, namely Heuningnes redbfin (*Pseudobarbus sp burchelli heuningnes*), Cape kurper (*Sandelia capensis*), and Cape galaxia (*Galaxias zebratus*). Additionally, two non-native fish species were recorded: Spotted bass (*Micropterus punctulatus*), Bluegill sunfish (*Lepomis macrochirus*; Table 4). See supplementary materials for all site coordinates and environmental variable measurements, as well as descriptive statistics for each variable per site.

Table 4: The number of surveys where native and non-native fish species were recorded

Native		Non-native		
<i>Pseudobarbus</i>	<i>Sandelia</i>	<i>Galaxias</i>	<i>Micropterus</i>	<i>Lepomis</i>
16	27	8	4	3

3.2 Pearson's test

A 2x2 contingency table was created with the results of the fish surveys in order to perform a Pearson's Chi-square test for independence (Table 5).

Table 5: Contingency table used to perform Pearson’s Chi-square test for independence. One survey detected both native and non-native fish. Five surveys detected non-native fish, and no native fish. Native fish were detected in 39 surveys where non-native fish were not detected. No fish were detected in a total of 11 surveys.

	Native Present	Native Absent
Non-Native Present	1	5
Non-Native Absent	39	11

The results of the test indicate that in pools where non-native fish were present in fyke net surveys, native fish were more likely to be absent ($X^2 = 7.10$; $DF = 1$; $p < 0.05$). Hence, the null hypothesis that the distribution of native and non-native fish occur independently of each other and at random is rejected.

3.3 Influence of environmental factors on redbfin abundance

The initial GLMM was fitted with the relative abundance of Heuningnes redbfin as the response variable, and included water quality, geographic, and river habitat variables as explanatory variables. The model built using a negative binomial distribution using classical stepwise regression returned an AIC value of 213.24.

This model retained temperature, woody debris, substrate, pH, dissolved oxygen, elevation, and phosphorous as the most significant explanatory variables of H. redbfin presence (Table 6). Specifically, ‘no woody debris’ was the most significant predictor from the woody debris set of variables. Similarly, gravel substrate was the most significant class predictor from the substrate variable. The remaining variables were ultimately excluded from the model, indicating that they were not strong predictors of H. redbfin presence and abundance.

Table 6: Results of generalised linear mixed model (GLMM) selection examining predictors of H. redfin abundance. All explanatory variables with $p < 0.100$ were retained in the model. Only those with $p < 0.05$ were considered significant predictors of H. redfin abundance (= $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$). Explanatory variables with $p > 0.10$ were removed from the model in a stepwise fashion, with the least significant variable being removed at each step.*

<i>Response Variable</i>	<i>Explanatory Variables</i>	<i>Estimate</i>	<i>±SE</i>	<i>X²</i>	<i>p-value</i>
<i>H. redfin abundance</i>	Temp	0.460	0.084	5.546	<0.001***
	No Woody Debris	-10.525	1.326	-7.935	<0.001***
	Gravel Substrate	4.493	1.075	4.181	<0.001***
	pH	1.141	0.265	4.304	<0.001***
	DO	0.043	0.014	2.976	0.003**
	Elevation	0.016	0.006	2.455	0.014*
	Phosphorous	-0.951	0.406	-2.345	0.019*

The variance inflation factor (VIF) calculation indicated no multi-collinearity issues were detected among the set of significant predictors (Table 7). VIF values greater than five indicate multicollinearity issues.

Table 7: Variance inflation factor calculation for the statistically significant variables identified by the GLMM.

Variable	VIF
pH	2.86
DO	1.18
Temperature	1.65
Phosphorous	1.12
Elevation	2.65
Gravel Substrate	1.03
No Woody Debris	1.16

4. Discussion

This study analysed data from surveys conducted at 56 river sites within the HRS in order to determine a) whether non-native fish influence the distribution of native fish, b) what environmental variables best explain the presence and abundance of Heuningnes redbfin, and how this information can be used to guide freshwater conservation planning in the area. The data suggest that native and non-native fish are not distributed independently of each other, which further suggests that non-native fish displace native fish in the HRS. Additionally, seven statistically-significant predictors of Heuningnes redbfin abundance were identified, namely water temperature, pH, woody debris, substrate type, elevation, dissolved oxygen, and phosphorous. Other environmental variables measured were not significantly correlated with Heuningnes redbfin abundance.

4.1 Influence of non-native fish on native fish distribution

The distributions of native and non-native fish distributions were largely mutually exclusive. Indeed, all three native fish were absent from fyke-nets set at sites where either Spotted bass or Bluegill sunfish were present (with the exception of one site), suggesting that these non-native species may be excluding native fish from invaded reaches. This result is consistent with several previous studies in South Africa that have all found native fish to be absent or scarce in invaded reaches (Woodford et al., 2005, Russel and Impson, 2006; Shelton et al., 2015; van der Walt et al., 2019; Cerrilla et al., 2022). In the Rondegat River, for example, four out of five native fish species were found to be absent from bass-invaded reaches (Woodford et al., 2005).

It is not clear whether environmental conditions in bass invaded sites might also explain the absence of redfins due to the low sample size of bass invaded sites, given that bass were only detected in six surveys. Increased sampling at more invaded sites would allow for an analysis that could confirm the absence of native fish is caused by the presence of bass. Additionally, these results cannot be used to distinguish which of the non-native species plays a larger role in excluding or predated native fish due to the spatial overlap of both non-native fish species in sampled sites. Similar studies conducted in the CFE, like the Olifants-Doring River System (ODRS) for example, indicate that bass (*Micropterus*) species are having the largest impact on native cyprinid populations, as compared to other non-native

species (van der Walt et al., 2016). Future studies in the HRS could compare the response of native fish to different non-native species, for example in river reaches where Spotted bass are present and Bluegill sunfish are absent, or vice versa, if such sites exist.

Given that non-native fish were negatively associated with the distribution of native fish, it is expected that where the range of non-native species expands, the range of native species will contract, following the trends of similar river systems in the CFE (Shelton et al., 2015; van der Walt et al., 2019; Cerilla et al., 2022). It is challenging, however, to find pre-existing data from the HRS to support this hypothesis, because the majority of studies conducted in the area focus on the phylogeny and evolutionary history of different fishes. As such, there are no quantitative data available on the historical or contemporary patterns of abundance and distribution of native freshwater fishes before the introduction of non-native freshwater fishes (Clark et al., 2009). Given the presence of bass in the upper reaches of the Kars River, it is unlikely that there are barriers preventing further invasion of non-native species. It is critical we ascertain an accurate measure of the extent of invasion in this system, in order to guide future conservation interventions.

4.2 Environmental predictors of redbfin presence

Redfin abundance was generally associated with complex habitats in higher elevations, and sites with better water quality. The class 'No woody debris' was negatively correlated with redbfin abundance (-10.525; $p < 0.001$), indicating that redbfins were generally less abundant where woody debris was lacking. This result could be attributed to the fact that woody debris increases habitat complexity and provides cover for redbfins and other native fish to hide from predators (Russel and Impson, 2006). Redfin abundance also increased at higher elevations (0.016; $p = 0.014$), where the water was more oxygenated (0.043; $p = 0.003$), and less impacted by agricultural pollutants, such as phosphorous (-0.951; $p = 0.019$). Higher elevations in the HRS also corresponded with less invaded reaches, with the exception of one site where Spotted bass were found in the upper reaches of the Kars River (Figure 3).

While the impact of non-native freshwater fishes has been extensively researched, there is a lack of knowledge regarding stressors such as pollution, water

abstraction and habitat degradation on CFE freshwater fishes (Ellender et al. 2017). However, extensive research on the effects of land use and land cover change on water quality has been conducted in South Africa (Kibena et al., 2014; Namugize et al., 2018; Mudaly and van der Laan, 2020; Mararakanye et al., 2022). Nutrient and pollutant loads in streams and rivers come from agricultural activities, and urban and industrial water runoff (Mararakanye et al., 2022). Expanding agriculture has been associated with increasing nitrate (NO_3^-), nitrogen (N), and phosphorous (P) concentrations in river systems (Molina-Navarro et al., 2018). According to Mudaly and van der Laan (2020), phosphorous is one of the major drivers of eutrophication, which is also known to reduce dissolved oxygen (Hanjaniamin et al., 2023). Given that phosphorous (-0.951 ; $p = 0.019$) and dissolved oxygen (0.043 ; $p = 0.003$) are also correlated with the presence and abundance of Heuningnes redfins, it is possible that agricultural effluents are affecting their distribution. Further research, however, is required to statistically assess the extent to which land use is affecting the distribution of redfins.

The correlation with gravel substrate could be an indicator of spawning preference, assuming redfin's share similar traits with the Drakensberg minnow (*Pseudobarbus quathlambae*) (Skelton, 2001). It should be noted, however, that there is significant uncertainty involved in making this correlation, primarily due to limited knowledge and understanding of redfin reproductive biology and spawning behaviour. Future research efforts could be undertaken to assess which environmental conditions are favourable to spawning and larval development. Collecting continuous and fine-scale environmental data during the spawning period of the redfin and observing the conditions under which they spawn, could provide invaluable information for future conservation of this species.

Water temperature and pH were also positively correlated with redfin abundance, indicating that redfin were more abundant at warmer, less acidic sites. Sites where redfin were present ($\bar{x} = 22.36 \text{ }^\circ\text{C}$ [$20 - 26 \text{ }^\circ\text{C}$]) were warmer on average than sites where redfin were absent ($\bar{x} = 20.12 \text{ }^\circ\text{C}$ [$12 - 28 \text{ }^\circ\text{C}$]). According to Reizenberg et al. (2019), thermal tolerances of fishes in the CFE are observed to increase from West to East. These results suggesting a higher thermal range for Heuningnes redfins is consistent with Reizenberg et al. (2019), as they found that Eastern cape redfin (*Pseudobarbus afer*) were found to show greater resilience to

temperature as compared to Western *Pseudobarbus* species, such as Fiery redbfin (*Pseudobarbus phlegethon*). Eastern cape redbfin thermal tolerance may be in part a result of large fluctuations in flow and temperature (Reizenberg et al., 2019). That being said, thermal resilience among other physiological characteristics is likely to differ amongst populations, and further highlights the need for a greater understanding of the physiology of Heuningnes redbfins.

Water temperature in this dataset was measured once per site, as opposed to using a temperature logger, which would have provided a daily average temperature by measuring regularly throughout the day. Given that water temperature in a river normally follows a diurnal cycle, using a point reading rather than a temperature logger, could have biased the model output (Sinokrot and Stefan, 1993). Future studies should endeavour to sample water temperature using a temperature logger, in order to obtain a more accurate indication of temperature preferences for this species. Moreover, seasonal sampling would account for changes in flow and temperature throughout the different seasons, and ensure datasets are more robust and comprehensive. Additionally, regular sampling over a number of years would allow for subsequent analyses to account for inter-annual changes in environmental variables. The lack of such long-term data is currently a limitation of this study. Seasonal and inter-annual sampling would also provide valuable insight into the potential effects associated with climate change, which could grow to be a major threat to native fish in the HRS (Reizenberg et al., 2019).

5. Conservation and sampling recommendations

5.1 Invasive eradication

The results of this study indicate that non-native fish appear to be excluding native fish species from invaded river reaches in the study area. As such, bass eradication or control within the study area should be considered. In the Rondegat River, for example, a pilot study conducted by CapeNature used the piscicide Rotenone to eradicate Smallmouth bass (*Micropterus dolomieu*) in a 4km stretch of river, thus promoting the security of native fish populations (Woodford et al., 2013). The intervention was considered a success due to the complete eradication of non-native fish species, and subsequent increase in native biodiversity (Weyl et al., 2014).

However, the application of piscicide should be considered with caution, due to the potential for collateral impacts on native fish, as well as amphibian and invertebrate fauna (Woodford et al., 2013).

Mechanical removal of invasive species has also shown some success in the CFE. The Thee River in the ODRS was invaded by Spotted bass, and in 2010 a mechanical removal project was initiated (van der Walt et al., 2019). Almost 400 Spotted bass were removed from the upper reaches of the river, 75% of which were captured with gill nets or hand nets, and the remainder removed using a combination of spearguns, seine nets and electrofishing. After a second removal project in 2014, subsequent surveys in 2015, 2016, and 2017 did not detect any bass, and the population was considered to have been extirpated (van der Walt et al., 2019). Mechanical removal projects can fail because of difficulties in removing young-of-year (YOY), or because of compensatory responses to harvesting by adult fish, such as hiding or evading humans (van der Walt et al., 2019; Zipkin et al., 2008). A better understanding of the distribution of non-native fish in the HRS is needed to determine if these factors would impede eradication projects.

Importantly, both of these projects identified or constructed a barrier downstream, above which they were able to eradicate non-native species. In doing so, they also created a refuge for threatened native fishes. Furthermore, there is no point in performing eradication projects if re-introductions cannot be prevented. Identifying a barrier that could prevent re-invasion of non-natives is therefore a crucial component. Barriers also allow for native species to increase their distribution while further strategies are developed to remove invasive fish from downstream reaches. Van der Walt et al. (2016) demonstrated that a natural or artificial barrier with at least an 80cm drop is sufficient to prevent the upstream migration and invasion of bass species in the CFE.

It is therefore highly recommended that comprehensive sampling throughout the entirety of the HRS is undertaken. Invaded sites should be sampled especially, because it would provide greater insight into the distribution of both native and non-native fish, and allow for a more nuanced and meaningful analysis of the factors explaining their distribution. Data from increased sampling could also be used to determine high priority sites for eradication, facilitate the selection of the best

eradication method, minimise the risk to endangered native fishes, and identify existing barriers or sites where artificial barriers can be constructed (Chakona and Swartz, 2013).

5.2 Maintaining habitat quality

This study found that temperature and pH, alongside the habitat variables woody debris and gravel substrate are the most statistically significant predictors of Heuningnes redbin presence and abundance in the HRS. In short, the data suggest redbins prefer complex habitats to hide from predators, and warmer, slightly acidic water. It is unclear, however, what their maximum temperature threshold is, which has important implications for the future conservation of this species under climate change scenarios. Climate change is expected to increase water temperatures into the future, which may in turn exacerbate thermal stress over the warm, dry summer periods (Reizenberg et al., 2019). To adequately conserve this and other threatened species, it is imperative to have accurate knowledge regarding their ecology, life history, abundance, and physiological tolerances (Moyle et al. 2013).

This study also identified potential links between land use, specifically agriculture, and the distribution of redbins, however the results were not conclusive. Redbins were negatively associated with phosphorous, which is a common pollutant from agricultural runoff (Molina-Navarro et al., 2018). Future studies should focus on analysing the extent to which land use, specifically agriculture, affects the distribution of native fish in the HRS. To know whether redbins are more abundant in areas with indigenous vegetation, or in agricultural land, is critical for the conservation of this species. If agricultural runoff is degrading habitat and water quality, and therefore making certain parts of the HRS less suitable for native fish, conservation practitioners should consider interventions that maintain water quality in this system. Petersen et al., (2022), for example, conducted a study that evaluated the relationship between contrasting land uses and riparian vegetation, and changes in nutrient dynamics and water quality. They found that indigenous vegetation demonstrated the lowest runoff volume, as well as the lowest total nitrogen and phosphorous loads as compared to non-native riparian vegetation and agricultural vegetation (Petersen et al., 2022). Creating a buffer of indigenous vegetation between river banks and agricultural land could reduce the volume and impact of

agricultural effluent runoff on native fish distribution in the HRS. Moreover, this intervention could have benefits for renosterveld, which is a type of vegetation native to the HRS, and one of the most critically endangered habitats in the CFE (Topp and Loos, 2019).

6.0 Conclusion

In conclusion, this study has produced and analysed some of the first data describing the predictors of the presence and abundance of the endangered Heuningnes redbin in the Heuningnes River System of the Cape Fold Ecoregion of South Africa. The data suggest non-native species appear to be excluding native species from invaded reaches. Moreover, Heuningnes redbins prefer warmer, more acidic water with complex habitats in the form of woody debris and gravel substrate, allowing them to hide from predators. Redfin abundance was negatively associated with phosphorous, which suggests they may be negatively impacted by degrading water quality due to agricultural effluents. Conservation practitioners should consider non-native eradication projects, through either chemical or mechanical removal. However, regular and comprehensive surveying in the study area is required to identify the potential for collateral damage to native flora and fauna, as well as existing barriers or opportunities for barrier construction to prevent re-invasion post eradication. Additionally, restoring indigenous vegetation such as renosterveld in order to create a buffer between agricultural land and river systems may reduce the impact of agricultural effluents on redbins in the HRS. Future surveys are required to accurately determine the current distribution of non-natives in the system, and identify existing refugia for native fish. Moreover, further sampling and use of spatial analysis techniques should take place in order to accurately measure the extent to which land use affects the distribution of native fish. Finally, gaining a greater understanding of their ecology, life history, and physiological tolerances will provide critical information for the future conservation of this endangered species.

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Supplementary Material

Supplementary Material 1: Water quality variables measurements sampled at each of the 56 sampling sites. Coordinates are measured using decimal degrees South (DDS) and decimal degrees East (DDE).

Site No.	Coordinates		Water Quality Variables											
	DDS	DDE	pH	EC	DO	Temp	Ammonia	Phosphorous	Nitrite	Nitrate	Total Iron	Phosphonate	TDS	Inorganic Nitrogen
1	-34.36753	19.76871	8.22	14.72	86.6	22.1	0.00	0.90	0	0.5	0.59	0.067	17	0.50
2	-34.36563	19.77097	8.24	7.11	99.4	21.9	0.02	0.28	0	0.7	0.22	0.031	16	0.72
3	-34.40106	19.81823	8.21	16.88	98.6	26.4	0.01	0.19	2	3.0	0.24	0.050	15	5.01
4	-34.41214	19.8241	7.55	6.54	126.4	24.9	0.03	0.55	0	1.2	0.22	0.047	7	1.23
5	-34.37391	19.79351	7.39	4.58	107.7	21.5	0.88	0.15	0	0.0	1.98	0.023	41	0.88
6	-34.37576	19.79417	8.60	6.17	97.4	20.4	0.03	0.20	3	0.0	0.00	0.000	42	3.03
7	-34.38038	19.79959	6.69	5.10	107.5	21.4	0.94	0.29	42	1.0	0.16	0.014	39	43.94
8	-34.39563	19.8195	8.03	11.33	96.0	21.4	0.02	1.52	0	1.1	0.52	0.111	26	1.12
9	-34.39835	19.82051	8.01	11.91	92.9	21.9	0.03	0.04	2	0.5	0.33	0.101	22	2.53
10	-34.3637	19.78252	7.74	7.25	96.9	21.0	0.02	0.12	2	0.3	0.07	0.010	33	2.32
11	-34.37086	19.78647	7.82	7.01	70	20.0	0.00	1.34	4	0.0	0.04	0.011	21	4.00
12	-34.36939	19.78448	7.55	5.76	97.1	20.1	0.88	0.05	0	0.0	0.10	0.098	53	0.88
13	-34.38736	19.81091	7.13	3.92	123.0	25.3	0.15	1.93	0	0.0	3.16	0.000	41	0.15
14	-34.38412	19.80189	7.39	5.26	95.3	21.0	0.41	0.04	1	1.3	0.80	0.490	14	2.71
15	-34.36066	19.77449	8.26	5.07	73.5	17.9	0.00	0.30	3	0.0	0.16	0.013	11	3.00
16	-34.36753	19.80865	8.07	13.49	97.53	23.9	0.04	0.06	4	0.5	0.11	0.023	26	4.54
17	-34.36853	19.80847	8.09	12.93	91.4	23.7	0.06	0.09	6	0	0.16	0.000	35	6.06
18	-34.37031	19.80895	7.87	17.28	89.5	24.3	0.05	0.13	8	1.4	0.47	0.000	36	9.45
19	-34.37284	19.81017	7.84	7.19	99.5	20.9	0.03	2.51	0	3.2	0.02	0.000	17	3.23
20	-34.36337	19.80933	8.37	17.88	108.1	21.6	0.06	2.51	3	0	0.13	0.000	34	3.06
21	-34.36699	19.80951	8.4	14.39	107.8	20.7	0	0.06	3	0.6	0.53	0.000	19	3.60
22	-34.38425	19.82259	7.89	20.5	94.1	21.2	0.08	0.08	3	1.3	0.38	0.038	18	4.38
23	-34.37991	19.81664	8.06	17.63	96.5	22.4	0.06	0.05	7	0	0.65	0.030	51	7.06
24	-34.38045	19.81786	8.37	17.9	99.5	22.8	0.13	2.2	3	0.6	0.41	0.052	19	3.73
25	-34.38561	19.82192	8.1	17.37	98.2	26.3	0.03	0.06	0	0.9	0.47	0.311	15	0.93
26	-34.36104	19.80481	7.89	16.22	98.4	17.9	0	0.05	1	0	0.19	0.149	11	1.00
27	-34.35926	19.80309	7.71	13.11	99.4	18.1	0.02	0.38	2	1.1	0.50	0.000	38	3.12
28	-34.41296	19.81985	6.69	1.75	74.4	24.2	0.24	0.00	0	0.0	3.01	0.045	206	0.24
29	-34.4208	19.76426	5.15	0.27	84.7	15.0	0.11	1.67	10	1.1	3.01	0.020	69	11.21

30	-34.4238	19.76184	5.23	0.24	88.5	15.3	0.04	1.44	14	3.4	1.39	0.000	41	17.44
31	-34.42698	19.73257	5.03	0.23	92.3	16.9	0.02	0.32	10	1.6	0.34	0.123	18	11.62
32	-34.4266	19.73307	4.54	0.22	92.9	14.9	0.01	0.68	8	2.1	0.17	0.000	18	10.11
33	-34.42681	19.73177	4.20	0.23	92.4	14.5	0.00	0.37	7	2.2	0.20	0.025	17	9.20
34	-34.3675	19.76889	9.30	8.53	77.4	22.3	0.11	0.23	2	0.1	0.06	0.580	89	2.21
35	-34.36563	19.77097	9.98	4.55	74.1	21.9	0.20	0.99	2	0	0.29	0.520	97	2.20
36	-34.37088	19.78639	9.63	7.69	79.2	22.1	0.05	0.13	3	0	0.04	0.180	82	3.05
37	-34.37561	19.79401	9.96	12.78	85.7	21.1	0.05	0.01	5	2.5	0.00	1.060	43	7.55
38	-34.39831	19.82048	9.64	12.54	89.5	19.7	0.05	0.08	10	0.2	0.00	0.780	26	10.25
39	-34.41214	19.8241	9.47	10.60	91.9	25.13	0.13	0.17	10	2.6	0.00	0.000	162	12.73
40	-34.36761	19.80892	9.38	7.00	73.7	18.4	0.07	0.22	5	1.3	0.38	0.070	12	6.37
41	-34.38033	19.81783	9.59	6.46	74.8	17.5	0.06	0.09	4	0.4	0.30	0.190	35	4.46
42	-34.41292	19.81987	7.57	13.65	43.9	24.9	0.10	0.00	0	0	0.03	2.890	88	0.10
43	-34.4334	19.83141	8.84	11.81	85.2	18.8	0.51	0.00	1	0.2	0.00	2.450	751	1.71
44	-34.43686	19.86142	9.67	9.20	79.5	19.0	0.51	2.51	87	4.3	0.55	1.840	123	91.81
45	-34.43556	19.88188	9.30	4.22	84.4	20.1	0.00	0.10	5	0.5	0.00	0.480	44	5.50
46	-34.43333	19.89307	9.80	10.14	96.6	18.7	0.91	0.60	9	1	0.05	0.150	28	10.91
47	-34.42202	19.93362	9.55	8.90	81.1	18.7	0.05	0.07	2	1	0.00	0.100	51	3.05
48	-34.391	19.86125	9.35	6.98	58.6	25.0	0.07	0.06	5	1.7	0.23	0.130	10	6.77
49	-34.37871	19.8985	8.97	13.37	73.9	25.9	0.08	0.08	4	0.7	0.00	0.000	18	4.78
50	-34.4	19.90896	9.28	10.61	95.97	27.87	0.03	0.14	3	0.9	0.00	0.000	11	3.93
51	-34.41396944	19.91163611	10.25	14.59	84.1	20.2	0.08	0.07	8	1.6	0.07	0.910	13	9.68
52	-34.4111	19.92128	9.84	17.3	85.83	21.4	0.00	0.00	3	1.2	0.51	0.000	45	4.20
53	-34.42661	19.91644	9.67	19.92	56.9	25.0	0.00	0.05	11	0.5	0.15	0.180	34	11.50
54	-34.42683	19.73183	4.59	0.76	85.1	11.7	0.04	2.60	15	1.7	1.04	0.000	17	16.74
55	-34.48407	19.85777	5.36	0.65	85.8	12.1	0.00	0.13	12	1.4	0.13	0.019	11	13.40
56	-34.50368	19.89474	4.40	0.43	70.0	14.6	0.00	0.08	7	1.3	0.04	0.197	5	8.30

Supplementary Material 2: Geographic variables measured at each of the 56 sites.

Site No.	Coordinates		Geographic Variables		
	DDS	DDE	Elevation	Flow	Slope
1	-34.36753	19.76871	192	0	1.24
2	-34.36563	19.77097	193	0	3.14
3	-34.40106	19.81823	132	0	2.21
4	-34.41214	19.8241	126	0	1.58
5	-34.37391	19.79351	160	0	5.44
6	-34.37576	19.79417	163	0	4.92
7	-34.38038	19.79959	155	0	2.89
8	-34.39563	19.8195	141	0	6.44
9	-34.39835	19.82051	133	0	0.70
10	-34.3637	19.78252	180	0	1.20
11	-34.37086	19.78647	164	0	2.48
12	-34.36939	19.78448	167	0	3.66
13	-34.38736	19.81091	144	0	2.67
14	-34.38412	19.80189	152	0	2.46
15	-34.36066	19.77449	184	0	1.32
16	-34.36753	19.80865	178	0	2.43
17	-34.36853	19.80847	180	0	3.96
18	-34.37031	19.80895	177	0	6.08
19	-34.37284	19.81017	173	0	3.52
20	-34.36337	19.80933	186	0	3.44
21	-34.36699	19.80951	182	0	0.03
22	-34.38425	19.82259	156	0	6.86
23	-34.37991	19.81664	162	0	0.39
24	-34.38045	19.81786	161	0	1.81
25	-34.38561	19.82192	157	0	0.47
26	-34.36104	19.80481	196	0	1.81
27	-34.35926	19.80309	199	0	1.30
28	-34.41296	19.81985	124	0	1.28
29	-34.4208	19.76426	209	0.4	8.20
30	-34.4238	19.76184	219	0.4	5.13

31	-34.42698	19.73257	290	0.9	36.86
32	-34.4266	19.73307	283	0.8	20.43
33	-34.42681	19.73177	306	0.8	34.67
34	-34.3675	19.76889	184	0	1.58
35	-34.36563	19.77097	190	0	3.14
36	-34.37088	19.78639	156	0	2.48
37	-34.37561	19.79401	125	0	4.92
38	-34.39831	19.82048	119	0	0.70
39	-34.41214	19.8241	114	0	1.32
40	-34.36761	19.80892	123	0	2.43
41	-34.38033	19.81783	112	0	1.81
42	-34.41292	19.81987	114	0	1.28
43	-34.4334	19.83141	96	0	2.49
44	-34.43686	19.86142	71	0	0.49
45	-34.43556	19.88188	71	0	2.00
46	-34.43333	19.89307	93	0	0.90
47	-34.42202	19.93362	58	0	0.52
48	-34.391	19.86125	134	0	5.44
49	-34.37871	19.8985	117	0	1.74
50	-34.4	19.90896	89	0	1.08
51	-34.41396944	19.91163611	126	0	0.50
52	-34.4111	19.92128	77	0	2.34
53	-34.42661	19.91644	76	0	1.20
54	-34.42683	19.73183	303	0.5	34.67
55	-34.48407	19.85777	196	0.3	5.79
56	-34.50368	19.89474	205	0.3	2.47

Supplementary Materials 3: Habitat variables, specifically substrate and debris, measured at each of the 56 sites

Site No.	Habitat Variables								
	Coordinates		Substrate Variables					Debris Variables	
	DDS	DDE	Silt-Sand Substrate	Gravel Substrate	Cobble Substrate	Boulder Substrate	Bedrock Substrate	Woody Debris	No Woody Debris
1	-34.36753	19.76871	0.600	0.000	0.000	0.400	0.000	0.667	0.333
2	-34.36563	19.77097	0.733	0.033	0.000	0.233	0.000	0.500	0.500
3	-34.40106	19.81823	0.500	0.400	0.000	0.100	0.000	0.233	0.767
4	-34.41214	19.8241	0.267	0.100	0.000	0.000	0.633	0.300	0.700
5	-34.37391	19.79351	0.867	0.133	0.000	0.000	0.000	0.600	0.400
6	-34.37576	19.79417	0.533	0.133	0.000	0.100	0.233	0.533	0.467
7	-34.38038	19.79959	0.867	0.067	0.000	0.000	0.067	0.500	0.500
8	-34.39563	19.8195	0.667	0.167	0.000	0.067	0.100	0.533	0.467
9	-34.39835	19.82051	0.867	0.033	0.000	0.033	0.067	0.467	0.533
10	-34.3637	19.78252	0.867	0.067	0.000	0.000	0.067	0.300	0.700
11	-34.37086	19.78647	0.400	0.067	0.000	0.000	0.533	0.333	0.667
12	-34.36939	19.78448	0.833	0.033	0.000	0.100	0.033	0.167	0.833
13	-34.38736	19.81091	0.900	0.067	0.000	0.033	0.000	0.233	0.767
14	-34.38412	19.80189	0.600	0.133	0.000	0.067	0.200	0.300	0.700
15	-34.36066	19.77449	0.667	0.267	0.000	0.033	0.033	0.467	0.533
16	-34.36753	19.80865	0.733	0.033	0.000	0.033	0.200	0.300	0.700
17	-34.36853	19.80847	0.200	0.567	0.000	0.033	0.200	0.433	0.567
18	-34.37031	19.80895	0.067	0.500	0.000	0.000	0.433	0.200	0.800
19	-34.37284	19.81017	0.700	0.033	0.000	0.067	0.200	0.467	0.533
20	-34.36337	19.80933	0.500	0.333	0.000	0.067	0.100	0.067	0.933
21	-34.36699	19.80951	0.500	0.200	0.000	0.100	0.200	0.133	0.867
22	-34.38425	19.82259	0.333	0.100	0.000	0.033	0.533	0.133	0.867
23	-34.37991	19.81664	0.833	0.000	0.000	0.167	0.000	0.233	0.767
24	-34.38045	19.81786	0.200	0.100	0.000	0.067	0.633	0.133	0.867
25	-34.38561	19.82192	0.500	0.000	0.000	0.000	0.500	0.000	1.000
26	-34.36104	19.80481	0.067	0.633	0.000	0.067	0.233	0.233	0.767
27	-34.35926	19.80309	0.533	0.367	0.000	0.033	0.067	0.000	1.000
28	-34.41296	19.81985	0.433	0.567	0.000	0.000	0.000	0.700	0.300
29	-34.4208	19.76426	0.200	0.733	0.000	0.067	0.000	0.167	0.833

30	-34.4238	19.76184	0.767	0.000	0.000	0.233	0.000	0.400	0.600
31	-34.42698	19.73257	0.000	0.067	0.033	0.400	0.500	0.333	0.667
32	-34.4266	19.73307	0.300	0.033	0.000	0.000	0.667	0.600	0.400
33	-34.42681	19.73177	0.100	0.000	0.000	0.367	0.533	0.467	0.533
34	-34.3675	19.76889	0.433	0.000	0.000	0.367	0.200	0.400	0.600
35	-34.36563	19.77097	0.667	0.000	0.000	0.000	0.333	0.200	0.800
36	-34.37088	19.78639	0.233	0.400	0.000	0.000	0.367	0.500	0.500
37	-34.37561	19.79401	0.500	0.133	0.000	0.033	0.333	0.233	0.767
38	-34.39831	19.82048	1.000	0.000	0.000	0.000	0.000	0.367	0.633
39	-34.41214	19.8241	0.433	0.000	0.000	0.200	0.367	0.533	0.467
40	-34.36761	19.80892	0.600	0.133	0.000	0.000	0.267	0.067	0.933
41	-34.38033	19.81783	0.133	0.000	0.000	0.000	0.867	0.333	0.667
42	-34.41292	19.81987	0.533	0.167	0.000	0.267	0.033	0.467	0.533
43	-34.4334	19.83141	0.833	0.000	0.000	0.167	0.000	0.400	0.600
44	-34.43686	19.86142	1.000	0.000	0.000	0.000	0.000	0.467	0.533
45	-34.43556	19.88188	0.633	0.167	0.000	0.200	0.000	0.133	0.867
46	-34.43333	19.89307	1.000	0.000	0.000	0.000	0.000	0.033	0.967
47	-34.42202	19.93362	0.633	0.367	0.000	0.000	0.000	0.333	0.667
48	-34.391	19.86125	0.933	0.000	0.000	0.000	0.067	0.367	0.633
49	-34.37871	19.8985	1.000	0.000	0.000	0.000	0.000	0.233	0.767
50	-34.4	19.90896	0.600	0.000	0.000	0.000	0.400	0.300	0.700
51	-34.41396944	19.91163611	0.767	0.000	0.000	0.100	0.133	0.000	1.000
52	-34.4111	19.92128	0.567	0.000	0.000	0.000	0.433	0.000	1.000
53	-34.42661	19.91644	0.600	0.267	0.000	0.000	0.133	0.000	0.933
54	-34.42683	19.73183	0.167	0.300	0.000	0.000	0.533	0.533	0.467
55	-34.48407	19.85777	0.167	0.000	0.000	0.600	0.233	0.367	0.633
56	-34.50368	19.89474	0.667	0.000	0.000	0.000	0.333	0.300	0.700

Supplementary Material 4: Habitat variables, specifically river bank, macrophytes, and canopy, measured at each of the 56 sites

Site No.	Habitat Variables										
	Coordinates		River Bank Variables		Macrophyte Variables				Canopy Variables		
	DDS	DDE	Undercut Bank	No Undercut Bank	No Macrophyte	Scarce Macrophytes	Moderate Macrophytes	Abundant Macrophytes	Open Canopy	Partial Canopy	Closed Canopy
1	-34.36753	19.76871	0.000	1.000	0.633	0.233	0.000	0.133	0.533	0.300	0.167
2	-34.36563	19.77097	0.200	0.800	0.533	0.433	0.000	0.033	0.533	0.267	0.200
3	-34.40106	19.81823	0.000	1.000	0.333	0.667	0.000	0.000	0.667	0.300	0.033
4	-34.41214	19.8241	0.000	1.000	0.000	0.467	0.367	0.167	0.633	0.367	0.000
5	-34.37391	19.79351	0.000	1.000	0.700	0.267	0.033	0.000	0.900	0.100	0.000
6	-34.37576	19.79417	0.000	1.000	0.533	0.200	0.267	0.000	0.500	0.400	0.100
7	-34.38038	19.79959	0.000	1.000	0.667	0.333	0.000	0.000	0.667	0.333	0.000
8	-34.39563	19.8195	0.400	0.600	0.633	0.367	0.000	0.000	0.433	0.467	0.100
9	-34.39835	19.82051	0.200	0.800	0.433	0.500	0.067	0.000	0.367	0.567	0.067
10	-34.3637	19.78252	0.000	1.000	0.767	0.233	0.000	0.000	0.867	0.133	0.000
11	-34.37086	19.78647	0.000	1.000	0.267	0.300	0.433	0.000	0.567	0.400	0.033
12	-34.36939	19.78448	0.000	1.000	0.933	0.067	0.000	0.000	1.000	0.000	0.000
13	-34.38736	19.81091	0.000	1.000	0.867	0.133	0.000	0.000	0.933	0.067	0.000
14	-34.38412	19.80189	0.000	1.000	0.700	0.300	0.000	0.000	0.600	0.167	0.233
15	-34.36066	19.77449	0.000	1.000	0.133	0.233	0.167	0.467	0.767	0.133	0.100
16	-34.36753	19.80865	0.000	1.000	0.700	0.267	0.000	0.033	0.733	0.233	0.033
17	-34.36853	19.80847	0.000	1.000	0.767	0.233	0.000	0.000	0.533	0.267	0.200
18	-34.37031	19.80895	0.000	1.000	0.800	0.200	0.000	0.000	0.833	0.167	0.000
19	-34.37284	19.81017	0.000	1.000	0.500	0.500	0.000	0.000	0.567	0.267	0.167
20	-34.36337	19.80933	0.000	1.000	1.000	0.000	0.000	0.000	1.000	0.000	0.000
21	-34.36699	19.80951	0.000	1.000	0.500	0.433	0.067	0.000	0.900	0.100	0.000
22	-34.38425	19.82259	0.200	0.800	0.833	0.167	0.000	0.000	0.767	0.200	0.033
23	-34.37991	19.81664	0.000	1.000	0.967	0.033	0.000	0.000	0.700	0.167	0.133
24	-34.38045	19.81786	0.000	1.000	1.000	0.000	0.000	0.000	0.833	0.167	0.000
25	-34.38561	19.82192	0.000	1.000	1.000	0.000	0.000	0.000	1.000	0.000	0.000
26	-34.36104	19.80481	0.200	0.800	0.800	0.200	0.000	0.000	0.900	0.100	0.000
27	-34.35926	19.80309	0.000	1.000	0.700	0.267	0.033	0.000	0.900	0.100	0.000
28	-34.41296	19.81985	0.200	0.800	0.633	0.300	0.067	0.000	0.433	0.367	0.200
29	-34.4208	19.76426	0.000	1.000	0.333	0.233	0.400	0.033	0.600	0.300	0.100

30	-34.4238	19.76184	0.000	1.000	0.833	0.167	0.000	0.000	0.933	0.067	0.000
31	-34.42698	19.73257	0.000	1.000	0.533	0.300	0.167	0.000	0.467	0.433	0.100
32	-34.4266	19.73307	0.600	0.400	0.500	0.367	0.133	0.000	0.400	0.433	0.167
33	-34.42681	19.73177	0.000	1.000	0.600	0.367	0.033	0.000	0.500	0.500	0.000
34	-34.3675	19.76889	0.000	6.000	1.000	0.000	0.000	0.000	0.333	0.500	0.167
35	-34.36563	19.77097	0.000	1.000	0.400	0.167	0.400	0.033	0.767	0.233	0.000
36	-34.37088	19.78639	0.000	1.000	1.000	0.000	0.000	0.000	0.400	0.500	0.100
37	-34.37561	19.79401	0.000	1.000	1.000	0.000	0.000	0.000	0.700	0.300	0.000
38	-34.39831	19.82048	0.000	1.000	0.833	0.167	0.000	0.000	0.667	0.333	0.000
39	-34.41214	19.8241	0.000	1.000	1.000	0.000	0.000	0.000	0.600	0.400	0.000
40	-34.36761	19.80892	0.000	1.000	1.000	0.000	0.000	0.000	1.000	0.000	0.000
41	-34.38033	19.81783	0.000	6.000	1.000	0.000	0.000	0.000	1.000	0.000	0.000
42	-34.41292	19.81987	0.000	6.000	1.000	0.000	0.000	0.000	0.267	0.500	0.233
43	-34.4334	19.83141	0.000	6.000	1.000	0.000	0.000	0.000	0.667	0.333	0.000
44	-34.43686	19.86142	0.000	1.000	1.000	0.000	0.000	0.000	0.667	0.333	0.000
45	-34.43556	19.88188	0.400	0.600	1.000	0.000	0.000	0.000	0.667	0.333	0.000
46	-34.43333	19.89307	0.000	1.000	1.000	0.000	0.000	0.000	0.667	0.333	0.000
47	-34.42202	19.93362	0.000	1.000	1.000	0.000	0.000	0.000	1.000	0.000	0.000
48	-34.391	19.86125	0.000	1.000	0.367	0.233	0.333	0.067	1.000	0.000	0.000
49	-34.37871	19.8985	0.000	1.000	0.300	0.433	0.200	0.067	1.000	0.000	0.000
50	-34.4	19.90896	0.000	1.000	0.700	0.300	0.000	0.000	1.000	0.000	0.000
51	-34.41396	19.91163	0.000	1.000	0.000	1.000	0.000	0.000	1.000	0.000	0.000
52	-34.4111	19.92128	0.000	1.000	1.000	0.000	0.000	0.000	1.000	0.000	0.000
53	-34.42661	19.91644	0.000	1.000	0.233	0.600	0.167	0.000	1.000	0.000	0.000
54	-34.42683	19.73183	0.000	1.000	0.667	0.333	0.000	0.000	0.567	0.400	0.033
55	-34.48407	19.85777	0.000	1.000	0.600	0.400	0.000	0.000	0.700	0.300	0.000
56	-34.50368	19.89474	0.000	1.000	0.667	0.333	0.000	0.000	0.667	0.333	0.000

Supplementary Materials 5: Habitat variables, specifically river width and river depth, measured at each of the 56 sites.

Site No.	Coordinates		Habitat Variables							
	DDS	DDE	River Depth				River Width			
			Shallow Water Depth (0 - 50)	Moderate Water Depth (51-100)	Deep Water Depth (100 - 180)	Very Deep Water Depth (<180)	Narrow River Width (0 - 3)	Moderate River Width (3 - 6)	Wide River Width (6 - 10)	Very Wide Width (<10)
1	-34.36753	19.76871	0.133	0.867	0.000	0.000	0.000	0.400	0.600	0.000
2	-34.36563	19.77097	0.067	0.467	0.467	0.000	0.000	0.000	1.000	0.000
3	-34.40106	19.81823	0.500	0.467	0.033	0.000	0.000	0.200	0.800	0.000
4	-34.41214	19.8241	0.400	0.467	0.133	0.000	0.000	0.400	0.600	0.000
5	-34.37391	19.79351	0.500	0.400	0.100	0.000	0.000	0.200	0.400	0.400
6	-34.37576	19.79417	0.300	0.367	0.333	0.000	0.000	0.000	0.400	0.600
7	-34.38038	19.79959	0.433	0.567	0.000	0.000	0.000	0.400	0.600	0.000
8	-34.39563	19.8195	0.233	0.533	0.233	0.000	0.000	0.000	1.000	0.000
9	-34.39835	19.82051	0.067	0.333	0.300	0.300	0.000	0.000	0.800	0.200
10	-34.3637	19.78252	0.633	0.300	0.067	0.000	0.000	0.200	0.600	0.200
11	-34.37086	19.78647	0.667	0.300	0.033	0.000	0.000	0.000	1.000	0.000
12	-34.36939	19.78448	0.600	0.400	0.000	0.000	0.000	0.400	0.400	0.200
13	-34.38736	19.81091	1.000	0.000	0.000	0.000	0.000	0.800	0.200	0.000
14	-34.38412	19.80189	0.333	0.567	0.100	0.000	0.000	0.200	0.800	0.000
15	-34.36066	19.77449	0.600	0.400	0.000	0.000	0.000	0.000	1.000	0.000
16	-34.36753	19.80865	0.233	0.667	0.100	0.000	0.000	0.200	0.800	0.000
17	-34.36853	19.80847	0.233	0.533	0.200	0.033	0.000	0.200	0.800	0.000
18	-34.37031	19.80895	0.533	0.467	0.000	0.000	0.000	1.000	0.000	0.000
19	-34.37284	19.81017	0.200	0.800	0.000	0.000	0.000	0.000	1.000	0.000
20	-34.36337	19.80933	0.200	0.667	0.133	0.000	0.000	0.400	0.600	0.000
21	-34.36699	19.80951	0.400	0.533	0.067	0.000	0.000	0.200	0.800	0.000
22	-34.38425	19.82259	0.467	0.533	0.000	0.000	0.000	1.000	0.000	0.000
23	-34.37991	19.81664	1.000	0.000	0.000	0.000	0.000	1.000	0.000	0.000
24	-34.38045	19.81786	0.433	0.267	0.267	0.033	0.000	0.600	0.400	0.000
25	-34.38561	19.82192	0.733	0.267	0.000	0.000	0.000	0.200	0.800	0.000
26	-34.36104	19.80481	0.467	0.533	0.000	0.000	0.000	0.800	0.200	0.000
27	-34.35926	19.80309	0.567	0.433	0.000	0.000	0.000	0.400	0.600	0.000
28	-34.41296	19.81985	0.433	0.567	0.000	0.000	0.000	0.800	0.200	0.000

29	-34.4208	19.76426	1.000	0.000	0.000	0.000	1.000	0.000	0.000	0.000
30	-34.4238	19.76184	0.900	0.100	0.000	0.000	1.000	0.000	0.000	0.000
31	-34.42698	19.73257	0.500	0.500	0.000	0.000	0.600	0.400	0.000	0.000
32	-34.4266	19.73307	0.400	0.400	0.200	0.000	0.200	0.800	0.000	0.000
33	-34.42681	19.73177	0.133	0.533	0.200	0.133	0.200	0.600	0.200	0.000
34	-34.3675	19.76889	0.633	0.367	0.000	0.000	0.000	1.000	0.000	0.000
35	-34.36563	19.77097	0.267	0.567	0.100	0.000	0.000	0.200	0.800	0.000
36	-34.37088	19.78639	0.000	0.633	0.367	0.000	0.200	0.200	0.600	0.000
37	-34.37561	19.79401	0.333	0.567	0.100	0.000	0.000	0.200	0.400	0.400
38	-34.39831	19.82048	0.000	0.000	0.100	0.900	0.000	0.000	0.000	1.000
39	-34.41214	19.8241	0.433	0.267	0.267	0.033	0.400	0.000	0.200	0.400
40	-34.36761	19.80892	0.167	0.300	0.533	0.000	0.000	0.400	0.600	0.000
41	-34.38033	19.81783	0.667	0.133	0.133	0.067	0.200	0.200	0.600	0.000
42	-34.41292	19.81987	0.000	0.000	1.000	0.000	0.200	1.000	0.000	0.000
43	-34.4334	19.83141	0.933	0.067	0.000	0.000	0.000	0.800	0.200	0.000
44	-34.43686	19.86142	0.400	0.600	0.000	0.000	0.000	0.000	1.000	0.000
45	-34.43556	19.88188	0.333	0.667	0.000	0.000	0.000	0.200	0.800	0.000
46	-34.43333	19.89307	0.000	0.367	0.167	0.467	0.000	0.000	0.000	1.000
47	-34.42202	19.93362	0.000	0.067	0.933	0.000	0.000	0.000	0.400	0.600
48	-34.391	19.86125	0.033	0.933	0.033	0.000	0.000	0.000	0.800	0.200
49	-34.37871	19.8985	0.233	0.767	0.000	0.000	0.000	0.400	0.600	0.000
50	-34.4	19.90896	0.000	0.300	0.700	0.000	0.000	0.000	1.000	0.000
51	34.413969	19.91163	0.100	0.833	0.067	0.000	0.200	0.600	0.200	0.000
52	-34.4111	19.92128	0.000	0.033	0.967	0.000	0.000	0.000	0.000	1.000
53	-34.42661	19.91644	0.200	0.800	0.000	0.000	0.000	0.200	0.800	0.000
54	-34.42683	19.73183	0.100	0.200	0.367	0.433	0.200	0.400	0.400	0.000
55	-34.48407	19.85777	0.800	0.167	0.033	0.000	1.000	0.000	0.000	0.000
56	-34.50368	19.89474	0.867	0.133	0.000	0.000	0.400	0.400	0.200	0.000

Supplementary Materials 6: Native and Non-Native fish abundance at each of the 56 sampled sites

Site No.	Coordinates		Native Fish			Non-Native Fish		
	DDS	DDE	Heuningnes redbfin	Cape kurper	Cape galaxias	Spotted bass	Bluegill sunfish	Common carp
1	-34.36753	19.76871	58	138	0	0	0	0
2	-34.36563	19.77097	0	0	0	0	0	0
3	-34.40106	19.81823	103	0	0	0	0	0
4	-34.41214	19.8241	0	0	0	0	0	0
5	-34.37391	19.79351	0	0	0	0	0	0
6	-34.37576	19.79417	23	7	0	0	0	0
7	-34.38038	19.79959	10	0	0	0	0	0
8	-34.39563	19.8195	5	19	0	0	0	0
9	-34.39835	19.82051	3	9	0	0	0	0
10	-34.3637	19.78252	4	1	0	0	0	0
11	-34.37086	19.78647	0	0	0	0	0	0
12	-34.36939	19.78448	0	0	0	0	0	0
13	-34.38736	19.81091	0	0	0	0	0	0
14	-34.38412	19.80189	1	0	0	0	0	0
15	-34.36066	19.77449	0	0	0	0	0	0
16	-34.36753	19.80865	0	125	0	0	0	0
17	-34.36853	19.80847	0	5	0	0	0	0
18	-34.37031	19.80895	0	7	0	0	0	0
19	-34.37284	19.81017	0	1	0	0	0	0
20	-34.36337	19.80933	0	0	0	0	0	0
21	-34.36699	19.80951	0	30	0	0	0	0
22	-34.38425	19.82259	0	1	0	0	0	0
23	-34.37991	19.81664	0	2	0	0	0	0
24	-34.38045	19.81786	1	1	0	0	0	0
25	-34.38561	19.82192	0	0	0	0	0	0
26	-34.36104	19.80481	0	0	0	0	0	0
27	-34.35926	19.80309	0	1	0	0	0	0
28	-34.41296	19.81985	132	5	0	0	0	0
29	-34.4208	19.76426	0	0	19	0	0	0
30	-34.4238	19.76184	0	0	46	0	0	0

31	-34.42698	19.73257	0	0	5	0	0	0
32	-34.4266	19.73307	0	0	4	0	0	0
33	-34.42681	19.73177	0	0	7	0	0	0
34	-34.3675	19.76889	27	18	0	0	0	0
35	-34.36563	19.77097	0	0	0	3	0	0
36	-34.37088	19.78639	225	110	0	0	0	0
37	-34.37561	19.79401	10	0	0	0	0	0
38	-34.39831	19.82048	5	0	0	0	0	0
39	-34.41214	19.8241	1	0	0	0	0	0
40	-34.36761	19.80892	0	2	0	0	0	0
41	-34.38033	19.81783	0	0	0	0	0	0
42	-34.41292	19.81987	3	6	0	0	0	0
43	-34.4334	19.83141	0	0	0	1	0	0
44	-34.43686	19.86142	0	0	0	5	41	0
45	-34.43556	19.88188	0	0	0	1	0	0
46	-34.43333	19.89307	0	1	0	0	115	0
47	-34.42202	19.93362	0	0	0	0	2	0
48	-34.391	19.86125	0	32	0	0	0	0
49	-34.37871	19.8985	0	5	0	0	0	0
50	-34.4	19.90896	0	98	0	0	0	0
51	-34.41396944	19.91164	0	3	0	0	0	0
52	-34.4111	19.92128	0	2	0	0	0	0
53	-34.42661	19.91644	0	2	0	0	0	0
54	-34.42683	19.73183	0	0	5	0	0	0
55	-34.48407	19.85777	0	7	3	0	0	0
56	-34.50368	19.89474	0	0	2	0	0	0

Supplementary Materials 7: The range and mean of water quality and geographic variables sampled at each of the 56 sites.

<i>Variable Category</i>	<i>Variable</i>	<i>Range</i>	<i>Mean</i>
<i>Water Quality</i>	pH	4.20 - 10.25	7.99
	EC	0.22 - 20.50	9.29
	DO	43.87 - 126.40	89.05
	Temp	11.70 - 27.87	20.79
	Ammonia	0 - 0.94	0.13
	Phosphorus	0 - 2.60	0.52
	Nitrite	0 - 21.00	4.84
	Nitrate	0 - 4.30	0.98
	Total Iron	0 - 3.16	0.44
	Phosphonate	0 - 2.89	0.27
	TDS	5 - 751.00	51.45
<i>Geographic</i>	Inorganic Nitrogen	0.10 - 91.81	7.56
	Elevation	58 - 306	158.45
	Flow	0 - 0.90	0.08
	Slope	0.03 - 36.86	4.68