BENTHIC MACROINVERTEBRATES OF TEMPERATE, SUB-ANTARCTIC STREAMS:
THE EFFECTS OF ALTITUDINAL ZONING AND TEMPERATURE ON THE
PHENOLOGY OF AQUATIC INSECTS ASSOCIATED TO THE
ROBALO RIVER, NAVARINO ISLAND (55°S), CHILE

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The Cape Horn Biosphere Reserve, within the remote Sub-Antarctic ecoregion is a reservoir of expressions of biological and cultural diversity. Although it is considered one of 24 wilderness areas remaining in the world, it is not free from local and global threats, such as invasive species, and climate change. Field biologists and philosophers associated to the Sub-Antarctic Biocultural Conservation Program and the Omora Ethnobotanical Park, have worked to describe the region’s biocultural diversity, linking ecological and philosophical research into education, ecotourism, and conservation, through a methodology called field environmental philosophy (FEP), which integrates ecological sciences and environmental ethics through a 4-step cycle consisting of: 1) interdisciplinary research; 2) composition of metaphors; 3) design of field activities with an ecological and ethical orientation; and 4) implementation of in situ conservation areas. In this context, the purposes of this dissertation were to: 1) provide a comprehensive review of publications regarding the conservation status of aquatic and terrestrial insects at a global scale and with an emphasis in southern South America; 2) study the distribution of benthic macroinvertebrates through the sharp altitudinal gradient of the Róbalo River watershed; 3) describe the life histories of *Gigantodax* sp (Simuliidae: Diptera) and *Meridialaris chiloeense* (Leptophlebiidae: Ephemeroptera) in the Róbalo River and to assess the potential effects of climate change on their phenology; and 4) to apply FEP methodology in order to better understand and communicate the intrinsic and instrumental values of freshwater invertebrates in the Cape Horn Biosphere Reserve.
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By

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CHAPTER 1
INTRODUCTION

The biological richness of the Cape Horn archipelago and the temperate, Magellanic Sub-Antarctic ecoregion has remained underestimated. The majority of the research conducted in the region has focused on vertebrates and vascular plants, and omitted other classes of organisms, such as non-vascular flora (Rozzi et al. 2010) and freshwater macroinvertebrates. In an effort to overcome this problem, national and international scientists at the Omora Ethnobotanical Park, have been extensively researching the biodiversity of non-vascular (i.e. bryophytes, lichens) vegetation of Cape Horn. This has allowed them to characterize this region as an area that contains more than 5% of the world’s bryophytes on less than 0.01% of the Earth’s land surface. In addition, according to recent floristic and taxonomic studies, more than 50% of liverwort and bryophyte species are endemic to the temperate rainforests of southern South America (Rozzi et al. 2008b). This information provided strong support for the creation of the Cape Horn Biosphere Reserve in 2005 and showed how less conspicuous taxonomic groups, such as bryophytes, can motivate the protection of whole ecosystems (Rozzi et al. 2008).

Although the community structure of freshwater invertebrates has been the subject of much research in river systems (Miserendino and Pizzolon 2000), an equivalent effort has not been made to study the fauna of aquatic benthic macroinvertebrates of Sub-Antarctic freshwater environments. Potential benefits of research on macroinvertebrates include rapid assessments of biological resources and the detection of possible pollutants in freshwater environments. Pristine environments in remote areas are suitable for the detection of environmental changes and some studies have been carried out to assess the effects of global climate change on high-latitude freshwater ecosystems (Hauer et al. 1997, Miserendino and Pizzolon 2000). In this context, it
becomes especially important to continue with the characterization of freshwater benthic macroinvertebrate taxa in order to develop reference lists of the fauna associated to the different ecosystems of the Sub-Antarctic Magellanic ecoregion. This information is necessary in order to ask basic questions about the ecological dynamics of Sub-Antarctic watersheds and to study the effects of global change in the region, such as the increase of the temperatures (climatic change) in the last few decades, the introduction of exotic species (i.e. North American beaver, bison), and cultural homogenization. Moreover, the characterization of freshwater benthic macroinvertebrates could allow for the establishment of new conservation criteria for the Cape Horn Biosphere Reserve and its watersheds.

Furthermore, the Cape Horn Biosphere Reserve, embedded within the remote Sub-Antarctic ecoregion is a reservoir of expressions of biological and cultural diversity and a frontier on the verge of globalization. Although it is considered one of 24 wilderness areas remaining in the world, it is not free from local and global threats, such as salmon farming, invasive species, and climate change. In this context, field biologists, philosophers, artists, and local actors associated to the Sub-Antarctic Biocultural Conservation Program and the Omora Ethnobotanical Park (OEP), have worked together to describe the region’s biological and cultural diversity, linking ecological and philosophical research into education, ecotourism, and biocultural conservation, through a methodology called Field environmental philosophy. This methodology integrates ecological sciences and environmental ethics through a 4-step cycle consisting of: 1) interdisciplinary ecological and philosophical research; 2) composition of metaphors and communication of simple narratives; 3) design of field activities with an ecological and ethical orientation; and 4) implementation of in situ conservation areas.

In this context, the purposes of this dissertation were to:
1. Provide a comprehensive review of publications regarding the conservation status of aquatic and terrestrial insects at a global scale and with an emphasis in southern South America

2. Study the distribution and functional feeding structure of benthic macroinvertebrates through the sharp altitudinal gradient of the Róbalo River watershed

3. To describe the life history of *Gigantodax* sp (Simuliidae: Diptera) in the Róbalo River and to assess the potential effects of climate change on its phenology

4. To describe the life history of *Meridialaris chiloeense* (Leptophlebiidae: Ephemeroptera) in the Róbalo River and to assess the potential effects of climate change on its phenology, and

5. To apply Omora Park’s Field Environmental methodology in order to better understand and communicate the intrinsic and instrumental values of freshwater invertebrates in the Cape Horn Biosphere Reserve
CHAPTER 2
THE CONSERVATION STATUS OF AQUATIC INSECTS IN THE LITERATURE

Abstract
A comprehensive review of publications regarding the conservation of aquatic and terrestrial insects at a global scale and with an emphasis on southern South America is provided. Three prominent conservation journals were reviewed (Conservation Biology, Biodiversity and Conservation, and Biological Conservation) and found that 5% of all the works published between 1995 and 2008 focus on the conservation of freshwater insects. The highest percentage of publications on the conservation of aquatic insects comes from Europe (2.3%), while the lowest percentage comes from South America (0.1%). To assess the trends of aquatic insect research in southern South America, a literature search using Zoological Records, Biological Abstracts, and Current Contents was conducted. The literature was reviewed; finding that there has been a significant increase in the number of publications related to the taxonomy orders Ephemeroptera, Plecoptera, Trichoptera, Odonata, and Diptera (family Chironomidae) since 1975.

Introduction
Despite the ecological importance of invertebrates in freshwater ecosystems, they are rarely considered or given priority on conservation discussions. This view persists despite several recent studies that demonstrate a growing number of freshwater extinctions (Ricciardi and Rasmussen 1999). Along with other freshwater invertebrates, aquatic insects are an indispensable part of the food web and nutrient cycling in stream ecosystems and are an important component in the diet of many fish, amphibians, birds and mammals (Morse 2009).
They are of special importance in terms of conservation and protection of freshwater ecosystems because of their high sensitivity to environmental stress and/or ability to withstand changes in the environmental conditions due to the specie-specific variations in tolerance that insects have developed for a wide range of environmental conditions. Biological assessments of aquatic environments have been practiced since the early 1900s (Wallace 1996), and the study of aquatic insects has greatly influenced the development of basic ecological concepts. Many methods have been developed to assess stream quality using aquatic insects, ranging from assessing physiological and morphological changes of individuals to various measures of community structure (Wallace 1996). Moreover, aquatic insects have been used extensively as indicators of the level of pollution in the waters that we drink and use for recreation and many practical uses (Hynes 1970; Hellawell 1978; Pyle et al. 1981; Wallace 1996; Abellán et al. 2005; Morse 2009).

More than 950,000 species of insects have been described, comprising 72% of the total identified animal species on earth (Hoffman Black and Vaughan 2003). Although some of the most conspicuous insects of running waters, such as the Ephemeroptera, have attracted the interest of naturalists and taxonomists since the seventeenth century, until the past few decades little was known about freshwater invertebrates in virtually all taxonomic groups and regions (Allan and Flecker 1993). Due to the uneven level of knowledge in taxonomic and ecological surveys, it has been hard to obtain an approximate number of the aquatic insect species described until now, but some surveys indicate that the number of aquatic insects described ranges from 90,097 – 90,107 species, with an estimation of almost 214,000 predicted species, making up 20% of all species of insects described (Morse 2009). Because aquatic insect diversity has been incompletely catalogued, it has been hard for conservation scientists to determine levels of imperilment for this group, therefore the benefits and risks of aquatic insect biodiversity can not
be fully appreciated until that biodiversity is known (Morse 2009). Many species of aquatic 
insects remain unknown to science, and many have been collected and recognized as undescribed 
(Morse 2009). Unlike the situation seen in freshwater fishes, where several recent studies have 
concluded that on a worldwide basis at least 20 percent of the world’s freshwater fish species are 
either threatened or extinct, there does not seem to be a real consensus on an imminent extinction 
crisis in regard to aquatic insects, although this is not meant to imply that certain species are not 
at risk (Polhemus 1993).

Despite the extensive amount of research in the fields of limnology and stream ecology, 
many authors believe that biodiversity is at greater risk in freshwater systems than in any other 
ecosystems (Abellán et al. 2005). Previous studies suggest that the leading causes of aquatic 
insect endangerment are habitat destruction, displacement by introduced species, alteration of 
habitat by chemical pollution, acid rain, water impoundments, and wetland draining amongst 
others (Pyle et al. 1981; Polhemus 1993; Hoffman Black and Vaughan 2003). Furthermore, a 
study carried out by the Natural Heritage Program (U.S), determined that 43% of stoneflies, 19% 
of tiger beetles and butterflies, and 17% of dragonflies and damselflies are critically imperiled or 
imperiled in the U.S. (Hoffman Black and Vaughan 2003) and that all five groups of species 
considered in the study really on freshwater habitats for their survival. It is estimated that 
100,000 of every million species of insects could be extinct by 2050 because of habitat loss, yet 
insect conservation remains the awkward “kid sister” to vertebrate conservation (Dunn 2005b). 
This taxonomic bias is clearly seen in conservation initiatives and research. Invertebrate research 
is highly underrepresented and vertebrate research is highly overrepresented in terms of number 
of species described for each group (Clark and May 2002). Moreover, funding and research 
activities of the majority of conservation organizations, focus mainly on bird and mammal
conservation (Clark and May 2002). More efforts should be placed on the identification and discovery of aquatic insects because of the unprecedented rate at which species are being lost. Without more research, education, and funding directed towards aquatic insect conservation, an inevitable crisis is faced in the proper protection of freshwater environments, as a high abundance and diversity of aquatic insect taxa helps assure the processing of large amounts of nutrients (Cummins and Klug 1979) and it assures a high diversity and quality of freshwater resources.

In lights of the current biodiversity crisis, which is undeniably an insect biodiversity crisis (Dunn 2005b), a comprehensive review of publications is provided regarding the conservation of aquatic and terrestrial insects at a global scale and with an emphasis on southern South America in order to assess a possible bias in conservation research regarding these organisms. Three prominent conservation journals (*Conservation Biology*, *Biodiversity and Conservation*, and *Biological Conservation*) were reviewed, and the number of articles published from 1995-2008 regarding aquatic and terrestrial insects was recorded. A focus was placed on the status of research in southern South America, as the percentage of articles published was the lowest for all areas of the world investigated.

**Methods**

Global Conservation of Aquatic and Terrestrial Insects

To evaluate the conservation status of aquatic and terrestrial insects, data from three international conservation journals were collected: *Conservation Biology* (CB), *Biodiversity and Conservation* (B&C), and *Biological Conservation* (BC). These journals were selected because of their high 2009 impact factor (ISI Web of Knowledge: CB = 4.6, B&C = 2.0 and BC= 3.9)
their long standing in conservation research, their involvement in the promotion of conservation biology, and because they provide a good representation of the global scientific literature in conservation biology (Fazey et al. 2005). All reviews, letters, contributed papers, and short notes from 1995 to 2008 for the selected journals were surveyed. The articles were examined and catalogued in search for a focus on aquatic and terrestrial insect conservation and selected them based on their title, keywords, and abstract (total $n = 7780$; comprised of CB = 3060, B&C = 1696, and BC = 3024 articles). After selecting the articles, the articles were organized into the following world regions: Africa, Asia & Middle East, Europe, South America, and North America. In order to assess global conservation trends, a one-way Analysis of variance (ANOVA) and a Student-Newman-Keuls (SNK) test ($\alpha 0.05$) (Team 2010) were conducted.

Research Trends in Southern South America

A literature search using Zoological Records, Biological Abstracts, and Current Content was conducted. A total of 747 articles, reviews, letters, and short notes published from 1975 to 2010 were examined. To assess the research trends regarding aquatic insects in South America, focus was placed on the Orders Ephemeroptera, Plecoptera, Trichoptera, Odonata, and Diptera (Chironomidae). The Family Chironomidae was emphasized amongst other dipterans because its members play an important role in energy transfers in ecosystems and constitute a major food source for many organisms (Engel 1988, Balci and Kennedy 2002). After selecting the articles, they were organized into decades (according to publication year) and into the categories of taxonomy, ecological processes, bioassessment, biogeography, and paleobiology. To examine differences between research trends and decades, R software (2010) was used to conduct a one-
way parametric Analysis of variance (ANOVA) and a Student-Newman-Keuls (SNK) test ($\alpha = 0.05$).

Results

Global Conservation of Aquatic and Terrestrial Insects

Overall, the percentage of works published regarding the conservation of aquatic and terrestrial insects, compared to the total amount of publications reviewed, was low for the three journals selected (5% and 27% respectively). The highest percentages of articles published for both, aquatic and terrestrial insects, were 17% and 2.6% for the journal Biodiversity and Conservation, while the lowest percentages were 0.2% and 4% for the journal Conservation Biology (Fig. 1).

At a global scale, the percentage of publications in relation to the conservation of aquatic insects is significantly higher in Europe (2.3%) (One-way parametric ANOVA, $P = 0.03$; Student-Newman-Keuls test, $\alpha = 0.05$), while Africa, Asia and the Middle East, Australia and Oceania, and North America, range from 0.5-0.6%. The percentage of articles published in South America is significantly lower (Student-Newman-Keuls test, $\alpha = 0.05$) than other regions of the world, with only 0.1% of all of the publications focusing on the conservation of aquatic insects (Fig. 2). Although terrestrial insect conservation seems to be a more prominent topic amongst conservation biologists, there are no significant differences between the different regions of the world (One-way parametric ANOVA, $P = 0.15$, $\alpha = 0.1$). The highest percentage of publications is observed for Europe and North America (8% and 7% respectively). Africa, Asia and the Middle East, Australia and Oceania, and South America range from 2.2%-3.4% of publications regarding the conservation of terrestrial insects (Fig. 3). Coleoptera and Lepidoptera are the
most represented Orders amongst the conservation literature, as 58% of all of the articles focus on the conservation of these organisms.

Research Trends in Southern South America

In general, there has been a significant increase in the number of publications in the orders Ephemeroptera, Plecoptera, and Trichoptera since 1975. The highest number of publications has been related to the orders Odonata and Diptera (Chironomidae), with a total of 95 and 90 publications respectively (Fig. 4). The percentage of publications related to the taxonomy and ecological processes of the Orders under study are significantly higher than other research trends (Fig. 5, One-way parametric ANOVA, P < 0.0001, α 0.05), and the majority of the works have been published in the journals *Aquatic Insects, Gayana, Odontologica, Revista de la Sociedad Entomológica Argentina, Studies on Neotropical Fauna and Environment*, and *Zootaxa*.

Discussion

Insects as a group receive only minimal legislative protection, presumably because of their small size and distant evolutionary relationship to humans (Metrick and Weistzman 1996, Bossart and Carlton 2002) and in general, invertebrate research is highly underrepresented (Clark and May 2002). By reviewing three of the most prominent conservation journals, it was possible to determine that 1) aquatic insects are underrepresented in conservation literature and 2) at a global scale, Europe presents the higher percentage of publications related to the conservation of aquatic insects (2.3%), while the percentage of publications for South America is the lowest (0.1%) in the world.
These results suggest that there is a bias in conservation. Various authors have established that vertebrates, especially endotherms, are over-represented in relation to the number of species described (Bonnet 2002, Clark and May 2002). According to the International Union for the Conservation of Nature and Natural Resources (IUCN) 2009 Red List, zero percent of all of the species of insects described (1,004,898) are threatened (Adler and Footit 2009, IUCN 2010) and they appear to be specially sensitive to human transformation of habitat, resulting in a fast decrease in their number of species, declining faster than birds or vascular plants (Travis 2003, Samways 2009). It is believed that this percentage is the direct result of the low amount of publications that specifically focus on the description and identification of new insect species and their conservation.

Throughout history, Europe (and specially England) has been the epicenter where the majority and most sophisticated ecological studies applied to insect conservation problems have been developed (Pyle et al. 1981). In 1925, the Insect Protection Committee of the Royal Entomological Society of London came into being and it issued its first endangered species list in 1946 (Pyle et al. 1981). Furthermore, during the 1960s and 1970s, insect conservation gained a firmer footing and scientists of the British Nature Conservancy held a symposium on invertebrate conservation (Pyle et al. 1981).

The percentage of works published in South America regarding the conservation of aquatic insects is significantly lower than other areas of the world. Knowledge of scientific literature pertaining to the taxonomic works available for South America was provided in a bibliography entitled *Biota acuática de Sudamérica Austral*. This volume edited by S. H. Hurlbert (Hurlbert 1977), with sections written by recognized taxonomic experts provided a comprehensive compendium for the works published through the mid 1970’s. However, a
considerable number of taxonomic and ecological works have been published since then. By reviewing the literature from 1975-2005 of the works conducted in southern South America (Patagonia, Sub-Antarctic ecoregion), it was determined that the majority of research carried out today is related to the taxonomy and systematics of aquatic insects. This is very important, as before insects and other invertebrates can be protected, it is important to know, at least, what species are present, if populations are stable or declining, and the habitat needs of these populations (Hoffman Black and Vaughan 2003). Although the amount of taxonomical works is increasing in southern South America, particularly in Chile and Argentina, the percentage of authors who focus in the study of invertebrates is low. According to a study about the taxonomists currently working in Chile, there is a significant mismatch between biological diversity and the number of taxonomists per group (Simonetti 1997). Vertebrates in Chile (1700 species), that account for only 11% of the fauna, are studied by 42% of the animal taxonomists, while insects (about 12000 species), comprising 61% of the animal species in Chile, receive attention by 21% of total Chilean taxonomists (Simonetti 1997).

In the long run, more emphasis needs to be placed on invertebrate surveys, systematics, taxonomy, and population ecology, so that these species can be identified, catalogued, and their life histories understood (Hoffman Black and Vaughan 2003). It is necessary to concentrate biological research and public education on “star species” when these are available in threatened habitats, in the manner that has proved so successful in vertebrate conservation (Wilson 1987). This has already been put into practice by using insects that have been catalogued as charismatic or umbrella species. Terrestrial insects such as butterflies, dragonflies and beetles have been widely studied and are appealing to the general public. Most scientists would agree that lepidopterans and odonates are considered more charismatic than other orders, and taxonomic
representations on conservation tracking lists are closely linked with these broad designations of relative charisma (Bossart and Carlton 2002). These designations promote dramatic discrepancies on conservation initiatives. For example, large and showy dragonflies and damselflies, which have a long established, significant professional and amateur following, occur on tracking lists at a frequency of 26 times greater than that expected by their numbers alone, whereas Diptera are 13 times less likely to be listed as a species of concern (Bossart and Carlton 2002). Furthermore, several Invertebrate Specialist Groups have been established within the IUCN’s Survival Service commission, including Lepidoptera, Odonata, and cave invertebrates (Pyle et al. 1981). A similar effort has not yet been developed for orders of aquatic insects such as Hemiptera, Trichoptera, Ephemeroptera, or Plecoptera.

In order to help overcome the conservation bias reported in this article, three ideas are proposed that could help increase the number of publications and conservation initiatives regarding aquatic insects:

a) Focus research on “understudied” regions of the world, such as southern South America – as shown in the results of this article, research has been highly underrepresented in the Southern Hemisphere. If efforts are concentrated in studying regions of the world such as southern South America, the number of aquatic insect species described would dramatically increase. For example, Chile’s Magellanic Sub-Antarctic Archipelago constitutes a very pristine temperate forest ecosystem (Moorman 2006), which has been catalogued as one of the 24 most pristine areas left in the world (Mittermeier et al. 2003). Furthermore, the central and southern regions of Chile have been reported as a hotspot of biodiversity for freshwater invertebrates (CONAMA 2008). This region has been designated as one of 25 hotspots of biodiversity of freshwater invertebrates in the world (Myers et al. 2000, CONAMA 2008) and it is clearly
isolated from the rest of South America by a series of geographic barriers, allowing for a great level of endemism and diversity at the species level. Moreover, southern South America has been reported as having some of the cleanest freshwater sources in world. Thus, increasing the amount of research that focuses on freshwater insects in this region of the world, would allow for the development of conservation initiatives that would ensure the protection of one the greatest sources of freshwater in the planet and one of the last pristine areas left in the world.

b) Increase the amount of funding available for taxonomical research focused on the description and identification of new aquatic insect species - Taxonomical studies should be emphasized, as the biggest problem in determining candidate insects and other related arthropods for endangered status is our lack of knowledge of the biodiversity, distribution, habits, and abundance of endemic insects (Pyle et al. 1981). After this is achieved, “star” aquatic insect species can be designated as targets for conservation and will allow us determine the level of imperilment of many species that could be at risk, thus allowing for the implementation of conservation initiatives and legislations that could help us protect whole watersheds.

c) Increase the amount of public education programs which focus on field experiences and direct encounters with aquatic insect biodiversity and their habitats – Today, direct encounters with nature are becoming increasingly rare, due to that a large portion of our knowledge about nature is mediated by mathematical equations and models, technology, and established ecological theory (Rozzi et al. 2005). The easiest way to resolve this problem is through educational activities that include field experiences and “face-to-face” interactions with aquatic insects and their surroundings. Furthermore, it is necessary to start environmental education programs at the pre-school and elementary school levels by making children familiar with cultural, ecological, and the spiritual values (Hargrove 2008) regarding the biodiversity of
Efforts to Overcome the Publication Bias on Aquatic Insect Biodiversity in South America: The Case of the Omora Ethnobotanical Park

Although the percentage of publications regarding the conservation of aquatic insects in South America is significantly low, researchers, philosophers and artists at the Omora Ethnobotanical Park (OEP), located in Navarino Island (55°S), Chile, have been actively researching the ecology and natural history of these organisms in order to promote their conservation through research, education and ecotourism activities. The OEP is embedded within the Cape Horn Biosphere Reserve, an area in which the biological richness of the Cape Horn archipelago and the temperate, Magellanic Sub-Antarctic ecoregion has remained underestimated. The majority of the research conducted in the region has focused on vertebrates and vascular plants, and omitted other classes of organisms, such as non-vascular flora (Rozzi et al. 2010) and freshwater macroinvertebrates. In an effort to overcome this problem, national and international scientists at OEP, have been extensively researching the biodiversity of non-vascular (i.e. bryophytes, lichens) vegetation of Cape Horn. This has allowed them to characterize this region as an area that contains more than 5% of the world’s bryophytes on less than 0.01% of the Earth’s land surface. This information provided strong support for the creation of the Cape Horn Biosphere Reserve in 2005 and showed how less conspicuous taxonomic groups, such as bryophytes, can motivate the protection of whole ecosystems (Rozzi et al. 2008c). Today, research, education, ecotourism, and conservation activities are taking place in order to elucidate the diversity of aquatic insects inhabiting the rivers and streams of the Sub-Antarctic ecoregion. These activities are taking place through a 4-step cycle that links research,
conservation, and education: This 4-step cycle includes: 1) the generation of new scientific knowledge, 2) communication of this knowledge through the invention of metaphors, 3) ecologically guided field activities, and 4) *in-situ* conservation (Rozzi et al. 2008c). As more information about the diversity of aquatic insect biodiversity in the Sub-Antarctic ecoregion is generated, new educational experiences are being developed in which children learn about freshwater biodiversity and its importance for the conservation of whole watersheds. This will lead to a long-term process in which future generations will understand the importance of these organisms for the conservation of Sub-Antarctic ecosystems.

Chapter References


Fig. 1. Percentage of articles regarding the conservation of freshwater and terrestrial insects published between 1995 and 2008 in the journals Conservation Biology, Biodiversity and Conservation, and Biological Conservation.

Fig. 2. Percentage of conservation of freshwater insect articles published between the years 1995 and 2008 in the journals Conservation Biology, Biodiversity and Conservation, and Biological Conservation. The percentage of works published in Europe is significantly higher than other regions of the world, while the percentage of works published in South America is significantly lower (One-way parametric ANOVA with SNK analysis, P = 0.03, α=0.05).
Fig. 3. Percentage of conservation of terrestrial insect articles published between the years 1995 and 2008 in the journals Conservation Biology, Biodiversity and Conservation, and Biological Conservation.

Fig. 4. Average number of publications (± 1 SD) between the years 1975 and 2010 for the orders Plecoptera, Trichoptera, Ephemeroptera, Odonata, and Diptera (Chironomidae). Lines over bars indicate a significantly different number of publications (One-way parametric ANOVA, P < 0.0001, α 0.05).
Fig. 5. Number of publications in relation to taxonomy, ecological processes, biogeography, and bioassessment. Letters “a” and “b” represent a significantly different number of publications for each research trend (One-way parametric ANOVA, P < 0.0001). A SNK analysis indicated that publications related to taxonomy were significantly higher than publications involving ecological processes, while publications related to biogeography and bioassessments were not significantly different from each other (α 0.05).
CHAPTER 3

BENTHIC MACROINVERTEBRATE DISTRIBUTION AND FUNCTIONAL FEEDING STRUCTURE ALONG THE ALTITUDINAL GRADIENT OF A SUB-ANTARCTIC FLUVIAL SYSTEM IN THE CAPE HORN BIOSPHERE RESERVE, CHILE (55°S)

Abstract

The distribution, community, and functional feeding structure of benthic macroinvertebrates in the Róbalo River were studied during the austral summers of 2008, 2009 and 2010. The river, located in the Cape Horn Biosphere Reserve (55°S), provides drinking water to the world's southernmost town, Puerto Williams. It is embedded in an altitudinal gradient with habitats characteristic of the Cape Horn region, with evergreen, mixed and deciduous forests, and Andean highlands. The river was divided into 5 sites separated by 100 m of altitude. Invertebrates collected were identified and classified according to functional feeding groups. Habitat and physical-chemical characteristics were described at each altitudinal location. PCA shows that benthic communities are strongly affected by altitude, temperature, and substrate diversity. Furthermore, temperature profiles indicate that Sub-Antarctic watersheds are characterized by sharp thermal gradients, in which cumulative degree-days (CDD) per year sharply increase through relatively short altitudinal gradients (0-600 meters above sea level). This is notably contrasted with northern hemisphere streams, where similar accumulation of yearly degree-days are reached at significantly higher altitudes, as been shown for streams in the Rocky Mountains, ranging from 0-1500 m.a.s.l.

Introduction

The Sub-Antarctic Magellanic ecoregion has been identified as one of the 24 wilderness areas remaining on the planet (Mittermeier et al. 2003), given that more than 70% of its original
vegetation cover is conserved in an area > 10,000 km² that lacks terrestrial connectivity, industrial and urban development, and harbors one of the lowest human population densities within temperate latitudes (Rozzi et al. in press). Furthermore, great topographic and climatic barriers isolate this austral South American forest biome from the nearest tropical forests by 1500-2000 km, provoking a geographic isolation that has generated remarkably high levels of endemism and a unique biodiversity of vertebrates, invertebrates and non-vascular and vascular plants (Rozzi et al. in press). For example, close to 90% of woody plant species, 33 % of woody genera, and about 60% of the bryophyte species (mosses and liverworts) are endemic to this austral biome (Rozzi et al. in press); and in terms of freshwater invertebrates, there are many genera and supra-genera level endemisms (Ringuelet 1961, Miserendino and Pizzolon 2000). Despite its unique biodiversity and high percentages of endemism, the biological richness of the Cape Horn archipelago and the Magellanic Sub-Antarctic ecoregion has remained understudied, often falling outside of national and international biodiversity prioritization schemes. Historically, the majority of the research conducted in the region has focused on higher taxa such as bird assemblages (Anderson and Rozzi 2000, Ibarra 2007, Pizarro et al. 2010) and vascular plants (Pisano 1977) and omitted other types of “under-perceived” small organisms, such as benthic macroinvertebrates, which are often difficult to study.

Although the community structure of freshwater invertebrates has been the subject of substantial research in northern Patagonian river systems (Miserendino and Pizzolon 2000), an equivalent effort has not been made to study the fauna of aquatic benthic macroinvertebrates of Sub-Antarctic freshwater environments. For this reason, it becomes extremely important to describe the diversity of freshwater benthic macroinvertebrates, as several authors have found that cold-adapted stream fauna of southern Patagonia and the Sub-Antarctic ecoregion has
undergone a high degree of speciation and endemism (Illies 1969, Miserendino and Pizzolon 2000). These taxa have also proven useful in Cape Horn to describe ecosystem-level processes, including the effects of invasive North American beavers on ecosystem function of Sub-Antarctic streams (Anderson and Rosemond 2007a).

Furthermore, the recognition of variables for which steepness and altitude are surrogates in aquatic ecosystems, is implicit in the long-standing recognition of the importance of factors such as temperature and current speed (Hynes 1970, Palmer et al. 1994). The gradient of environmental conditions that occurs as a function of altitude offers excellent opportunities to investigate factors which influence the diversity, composition, and abundance of stream organisms (Ward 1986). For example, temperature plays a major role in the distribution and abundance of lotic zoobenthos (Hynes 1970, Ward 1986) and it becomes especially important in streams with a marked altitudinal gradient.

In this context, the purposes of this study were to: 1) Provide a concise description of the benthic fauna associated to the altitudinal gradient of a Sub-Antarctic fluvial system in the Cape Horn Biosphere Reserve, Chile (55°S) and 2) To assess the relationships between richness, density, and distribution of benthic macroinvertebrates with physical and chemical variables along the altitudinal gradient.

Materials and Methods

Study Area

The Róbalo River (S 54° 59.647’, W 067° 40.878’) is located on Navarino Island (55°S), and it provides drinking water to the city of Puerto Williams, Capital of the Chilean Antarctic Province and the world’s southernmost town. It has an extension of approximately 12 km and
runs through the marked altitudinal gradient of the Dientes of Navarino Mountain Range, where it drains into the Beagle Chanel. Navarino Island lies south of the Beagle Channel and north of Cape Horn (Fig. 6). The archipelago became the Cape Horn Biosphere Reserve (CHBR) in 2005. It hosts the world’s southernmost-forested ecosystem and encompasses all of the islands south of the Beagle Channel, as well as the Chilean portion of Tierra del Fuego Island located south of the highest peaks in the Darwin Mountain Range (Rozzi et al. 2006). The study site lies within this biosphere reserve and the Omora Ethnobotanical Park, part of Chile’s network of Long Term Socio-Ecological Research Sites (LTSER).

The forests of this region are composed primarily of three members of the genus *Nothofagus* (Pisano 1977) and are part of the South American temperate forest biome, occurring along a narrow but latitudinally extensive strip of land between 35° and 55°S (Armesto et al. 1998). This region, represents a singularly isolated biome at the southern end of the continent, as great orographic and climatic barriers separate the biome from the nearest tropical forests, by 1500 to 2000 km (Rozzi et al. 2000). The north is delimited by the Mediterranean-type scrublands and the hyper-arid Atacama Desert, to the east lies the Patagonian steppe, and westward and southward lies the Pacific Ocean (Rozzi et al. 2012). This insularity has promoted the evolution of cold-adapted stream fauna that has undergone a high degree of speciation and endemism (Illies 1969, Miserendino and Pizzolon 2000).

The regional climate of the area is strongly affected by the westerly storm tracks coupled with precipitation induced by the high western flanks of the Andean Cordillera (McCulloch et al. 1997). It also depends on polar weather fronts from the south Pacific (Rozzi et al. 2006). The city of Puerto Williams is found within the influence of the *Dfk’c Transandeon with steppe degeneration* climatic zone. It represents a transition between rain climates of the western coast
of Magallanes and the steppe climates of the eastern side of the mountain range (Rozzi et al. 2006). The uniformity of the seasonal variation of precipitation as well as its thermal characteristics, allow for the growth of deciduous forests. The average annual temperature and precipitation recorded for Puerto Williams is 6.0°C and 467.3 mm respectively. Precipitation is less polluted than for any other of the world’s temperate forests (Likens 1991) and the warmest month has an average temperature of 9.6°C (January-March), while the coldest month averages 1.9°C (May-August).

The river, which has an approximate length of approximately 12 km, ascends from sea level to 600 meters above sea level (m.a.s.l). For this study, 5 sampling sites, separated by approximately 100 m of altitude, were selected according to vegetation, habitat, and landscape changes along the watershed (Fig. 7, Table 1). Sampling of benthic macroinvertebrates was carried out during the austral summers of 2008, 2009, and 2010.

Following is a description of the five sites studied:

- **Site I (586 m.a.s.l)**

  This site encompasses the headwater region of the Róbalo River. The reach has an average width of 2.7 meters and the substrate consists mainly of rock gravel and riffle habitats. The source of energy of the area is primarily autochthonous, due to the lack of riparian vegetation along the reach. The vegetation of the area is dominated by high-Andean communities, such as the lichen *Neuropogon* sp.

- **Site II (486 m.a.s.l)**

  The substrate of this site is consists primarily of large boulders and riffle habitats. The reach has an average width of 2.3 meters and the vegetation consists mainly of cushion plants.
such as *Bolax gummifera*. This is the first part of the river that feeds into a small lake (Laguna del Salto), to later flow down to the Beagle Channel.

- **Site III (380 m.a.s.l)**

  The vegetation at this altitude is dominated by stunted (krummholz) low deciduous southern beech forests (*Nothofagus antarctica*). The river flows into a second lake (Laguna Tortuga) and the substrate is primarily composed of large boulders and riffle habitats. The reach has an average width of 2.5 meters.

- **Site IV (240 m.a.s.l)**

  Located downstream from Róbalo Lake, this site is characterized by the presence of high deciduous southern beech forests (*N. pumilio*). The substrate is primarily composed of woody debris, riffles, and sand. The reach has an approximate reach of 20 meters. This site encompasses an abandoned dam constructed by the exotic and invasive, North American Beaver (*Castor canadiensis*). The riparian environment was greatly modified by *C. canadiensis*, which reduced forest canopy up to 30 meters away from streams (McCulloch et al. 1997).

- **Site V (120 m.a.s.l)**

  The riparian vegetation is dominated by evergreen and deciduous mixed forests (*N. betuloides* and *N. pumilio*). The substrate is primarily composed of large boulders, riffles, and woody debris.

**Field Methods**

*Habitat Characterization*

- **Physical** – Physical characteristics were recorded for each altitudinal location. At each site, a 100-meter reach was selected and divided it into 10 transects of 10 meters each. Dominant in-stream substrate cover (%) was recorded at each transect, as well as average stream...
depth (m) and width (m). The riparian environment along each reach was evaluated by recording the bank slope (°), riparian vegetation composition (%), and percent tree canopy cover (%). The bank slope and percent tree canopy cover were obtained by using a clinometer and a concave spherical densiometer, respectively.

- **Chemical** – Water chemistry parameters were recorded at each site and during each sampling event. Dissolved oxygen (mg/L), water temperature (°C) and conductivity (μS) were recorded three times at each reach using a YSI 8.5 meter. From January of 2009 to April of 2010, average, maximum, and minimum daily temperatures (°C) were recorded every 5 hours at each sampling site using Hobo dataloggers (Model HOBO® U22 Water Temp Pro v2). Historical water and air temperature data (2006-2011, °C) for site V (120 m.a.s.l) were obtained from a permanent meteorological site (managed by Chile’s Dirección General de Aguas (DGA)) located at this altitude. Although water and air temperature data are readily available for general access through DGA’s website, daily water temperature data for large periods of times is sometimes missing and not always consistent. For this reason, water temperature data (°C) was estimated from March of 2006 until March of 2009, by determining the relationship between water to air temperature (°C) through simple linear regressions, following Crisp and Howson (1982). As explained before, temperature data for the remaining period was obtained from Hobo data loggers deployed at site V (120 m.a.s.l). With this information, Cumulative Degree-Days (CDD) from 2006-2011 were calculated for this altitude. CDD were computed, following (Snyder et al. 2001) by summing the daily average temperature (°C) by month and for a period of 1 year. Cumulative Degree-Days are reported, as well as minimum and maximum (°C) water temperature, because CDD provide a better resolution of temperature significance on aquatic insect distributions and phenology.
Substrate – Substrate characterization was carried out using Wolman pebble counts to quantify particle size distribution (Harrelson et al. 1994). At each transect, 100 randomly selected points were chosen and the diameter of the substrate type was measured. These values were then used to determine substrate diversity by applying a Shannon-Weiner Diversity Index to the abundance of particular size classes of substrate found during the pebble counts (Anderson and Rosemond 2007b).

**Benthic Macroinvertebrate Sampling and Community Analysis**

Benthic macroinvertebrates were collected during the austral summers (January) of 2008, 2009 and 2010. Three samples were taken from riffle habitats at each site using a Surber net (0.09m²) with a 243μm mesh. Each sample was stored in 70% ethanol and transported to the laboratory for processing. A malaise trap was also set at each site to obtain adult representatives of the organisms, needed as an aid to identify to the lowest taxonomic level possible. The traps were deployed during the late evening and left out throughout the night at each sampling site and period, except for site I (586 m.a.s.l), as the weather and habitat conditions did not allow for a proper setting of the trap. Benthic macroinvertebrates were identified to the lowest taxonomic level, and functional feeding groups (FFGs), i.e. collector-gatherer, filterer, predator, shredder, and scraper, were determined using the best available literature (Merritt and Cummins 1996, Miserendino and Pizzolon 2000). Samples collected with a Surber net were used to estimate community composition and density (individuals/m²); while organisms obtained from the malaise traps were used to complement existing taxonomic lists of benthic macroinvertebrates associated to the watershed of the Róbalo River.
Benthic macroinvertebrate density (organisms/m²) was graphed using the beanplot method, as described by Kamstra (2008). This method was preferred over a boxplot, because a boxplot and its variant make use of the median of a group of data points, while the beanplot method uses the average of the group and also shows an overall average, giving the information in the form of a density trace (Kampstra 2008).

Statistical Analyses

All tests to determine statistical significance were set at α level of 0.05, and all assumptions were tested before proceeding with parametric analysis. Normality was tested using Shapiro-Wilks tests and homogeneity of variances was tested using an Hartley’s $F_{\text{max}}$ ratio test. If assumptions for normality or homogeneity of variances were not met, a log transformation was used log (x + 1). All data was analyzed using R software, version 2.9.2.

Differences between physical-chemical parameters through the altitudinal gradient (five sites) and years (2008, 2009, and 2010) were assessed with a two-way Analysis of variance (ANOVA). When significance was achieved, a Tukey HSD post hoc test was used.

To determine the effect of elevation on the community structure of benthic macroinvertebrates along the altitudinal gradient of the Róbalo River, richness and Pielou’s Evenness Index ($J$) (Pielou 1969) were calculated for each site, using Biodiversity Pro 5.0 software. Richness, evenness, density, and FFGs were compared in terms of altitude and sampling year using two-way ANOVAs. When significant results were obtained, a Tukey HSD post hoc test was applied. We used simple linear regressions to examine the relationship between substrate diversity ($H'$ of pebble counts) and benthic macroinvertebrate richness along the altitudinal gradient.
A principal components analysis (PCA) was conducted to examine the linear combinations of environmental variables (substrate diversity H’, temperature, year, and conductivity) that best explained the distributions of benthic macroinvertebrates along the altitudinal gradient. Dissolved oxygen and pH were not included in the PCA analysis, as there were not significantly different through the altitudinal gradient. A multiple Max R stepwise regression procedure was used to identify the best set of environmental variables that explained variation in benthic macroinvertebrate taxa. Taxa included in PCA were those that made up more than 90% of the total density at each site.

Results

Habitat Characterization

Sites at lower altitudes (120, 240, 380 m.a.s.l) had a higher percentage of vascular canopy cover than sites at higher altitudes (486 and 586 m.a.s.l), while the percentage of non-vascular herbaceous cover increased with altitude (Table 2). Substrate diversity (Shannon’s H’) increased towards the mouth of the Róbalo River, and it was higher at site V (120 m.a.s.l).

Stream width (m), depth (cm), and bank slope (°) increased with decreasing altitude (Table 2). The yearly water temperature (°C) range was higher for sites at lower altitudes. The highest water temperature (17°C) was recorded for site V (120 m.a.s.l). Cumulative Degree-Days (CDD) significantly increased as altitude decreased, with CDD at site V (120 m.a.s.l) being 4x higher than the CDD recorded for site II (486 m.a.s.l) (Table 2, Fig. 8). The regression between mean air temperature (°C) and mean water temperature (°C), obtained from DGA’s weather site at site V (120 m.a.s.l) from 2006-2010 suggests that the relationship between the two is approximately linear over most of the observed temperature range (y=-0.36 + 0.99x,
R²=0.89). With this information, we were able to estimate mean water temperature data (°C) for dates with missing information from DGA’s weather site, and estimate CDD from 2006-2011 at site V (120 m.a.s.l) in the Róbalo River watershed.

In terms of physical-chemical parameters, conductivity (μS), dissolved oxygen (mg/L) and temperature (°C) were significantly different along the altitudinal gradient and between sampling years (two-way ANOVA, P < 0.0001, α 0.05). A Tukey HSD test indicated that conductivity was significantly higher at sites at 120, 240, and 386 m.a.s.l, while water temperature (°C) was significantly lower during 2008, and it significantly increased with decreasing altitude. Dissolved oxygen (mg/L) was significantly higher at higher altitudes (486 and 586 m.a.s.l) (two-way ANOVA, P = 0.0004, α 0.05) (Table 3).

Benthic Macroinvertebrate Assemblages Along the Altitudinal Gradient of the Róbalo River

A total of 42 benthic macroinvertebrate taxa were identified along the altitudinal gradient of the Róbalo river watershed. These taxa included 24 genera and 7 species, with the remaining taxa being identified to order or family level.

**Benthic Macroinvertebrate Richness**

Site III (380 m.a.s.l) had the highest total richness (34), while site I (586 m.a.s.l) had the lowest total richness (Table 8). Benthic macroinvertebrate richness was significantly different between years and altitude, with altitude accounting for 53% of the total variation and year accounting for 12% of the total variation (2 way-ANOVA, P < 0.0001 (F=13.17) and P = 0.006 (F=6.10) respectively, α 0.05) (Fig. 9). Overall, benthic macroinvertebrate richness was significantly different between the years 2009 and 2008 (P = 0.0073), while there were no
significant differences between the years 2009 and 2010 (P = 0.829). Furthermore, richness was significantly higher at site III (380 m.a.s.l) than at all other sites (P < 0.0001) (Fig. 9). Richness at sites at sites II and I (486 and 586 m.a.s.l) was not significantly different (P = 0.892), while richness at site I (586 m.a.s.l) was significantly lower than sites IV and V (240 and 120 m.a.s.l) (P = 0.007 and P = 0.0006 respectively) (2-way ANOVA with Tukey HSD multiple range test, α 0.05).

Finally, benthic macroinvertebrate richness increased as a function of substrate diversity (H’) through the altitudinal gradient of the Róbalo River watershed (Fig. 10).

_Benthic Macroinvertebrate Evenness (Pielou’s J)_

Pielou’s evenness index (J) was overall, significantly lower during the year 2008, and, during the same year, it increased with decreasing altitude. There were no significant differences between the years 2009 and 2010 (Fig. 9, 2-way ANOVA with Tukey HSD multiple range test, α 0.05). During 2009, Pielou’s J, was significantly lower at site V (120 m.a.s.l), and significantly higher at site I (586 m.a.s.l), while during 2010, the opposite pattern was observed (Fig. 9, 2-way ANOVA with Tukey HSD multiple range test, α 0.05).

_Benthic Macroinvertebrate Density_

Benthic macroinvertebrate density (individuals/m²) was overall, higher at site IV (240 m.a.s.l), than at any other sites, and lower at site II (486 m.a.s.l), with density varying from 200-400 organisms/m². In general, density was more evenly distributed at site V (120 m.a.s.l) than at other sites, while density had a bimodal distribution at site I, with density varying from 95-300 organisms/m² (Fig. 11). This pattern is also evidenced by Pielou’s evenness Index (Fig. 9).
Furthermore, the order Diptera represents the order with the highest density of organisms, ranging from 100-1000 individuals/m², with site IV, accounting for the highest density reached through the altitudinal gradient (Fig. 12). In general, if site IV (a former beaver dam), was not considered in the analyses, Dipterans would increase in density with increasing altitude. The order Trichoptera has the lowest density of organisms through the altitudinal gradient, with a maximum of 220 and 150 individuals/m² at sites IV and III (240 and 380 m.a.s.l), respectively, and in general, its density remains constant through the gradient (between 50-150 individuals/m²) (Fig. 12). The order Ephemeroptera exhibits a similar pattern to the Trichoptera, but it is represented by higher number of individuals/m², with a maximum of 420 individuals at site IV (240 m.a.s.l). Lastly, the density of Plecoptera decreases with increasing altitude, with the highest density achieved at site IV (240 m.a.s.l), with a maximum of 320 individuals/m² (Fig. 12).

**Benthic Macroinvertebrate Functional Feeding Groups (FFG)**

Collector-gatherer, filterer, predator, and scraper densities (individuals/m²) were significantly influenced by altitude; filterer and shredder densities by year; and collector-gatherer and filterer, by the interaction of altitude and year (Two-way ANOVA, α 0.05, Table 3).

In general, collector-gatherer density varied through the altitudinal gradient, with densities being significantly higher at sites III (380 m.a.s.l) and V (586 m.a.s.l). Filterer densities significantly decreased with increasing altitude, with densities at site II (240 m.a.s.l) being significantly higher than all other sites. Predator, scraper, and shredder densities remained relatively uniform through the altitudinal gradient, with densities at site II (486 m.a.s.l), being
significantly higher than all other sites (two-way ANOVA with Tukey HSD test, α 0.05) (Table 4).

Environmental Relationships

The results of the PCA analysis for the species that make up > 90% of the total density of organisms in 3 years of sampling, and the four environmental variables selected (substrate diversity (H’), altitude, conductivity, and temperature), showed that 47.7% of the variance in species density was accounted by the first two principal components. The results indicate that *Udamocercia* (Notonemouridae: Plecoptera) and Orthocladiinae (Chironomidae: Diptera) have a significant positive relationship with altitude (Pearson’s correlation of 0.41 and 0.54, respectively, α 0.05, two-tailed test), while *Meridialis* (Leptophlebiidae: Ephemeroptera), *Rheochorema* (Hydrobiosidae: Trichoptera), *Edwarsina* (Blephlariceridae: Diptera), *Limonia* (Tipulidae: Diptera), *Hemerodromia* (Empididae: Diptera), *Gigantodax* (Simuliidae: Diptera), and *Luchoelmis* (Elmidae: Diptera) have a significant negative relationship with altitude (Pearson’s correlations > -0.40, α 0.05, two-tailed test) (Fig. 13). Furthermore, *Pelurgoperla* (Gripopterygidae: Plecoptera) has a significant positive relationship with year, conductivity, and temperature (Pearson’s correlations 0.32, 0.37, and 0.52 respectively, α 0.05, two-tailed test). Orthocladiinae (Chironomidae: Diptera) and *Hemerodromia* (Empididae: Diptera) are negatively correlated with substrate diversity (Pearson’s correlations -0.47 and -0.51 respectively), while *Gigantodax* (Simuliidae: Diptera) shows a positive relationship with this parameter (Pearson’s correlation=0.40) (Fig. 13). Finally, Max $R^2$ analysis further showed that most of the taxa included in the analysis are significantly correlated with altitude and temperature (Tables 6 and 7).
Discussion

Benthic Macroinvertebrate Richness, Evenness, and Functional Feeding Groups

In general, richness increases towards the mouth of the Róbalo River, with the highest richness being reached at site III (380 m.a.s.l). This result is related to substrate heterogeneity, as substrate diversity (as expressed by Shannon’s H’) is higher at this location (Figs 9 and 10). From the zoogeographical point of view, the system studied belongs to the Sub-Antarctic ecoregion, which has a strong dominance of Austral or Notogeic, and also Nearctic and Brasilic elements (Miserendino and Pizzolon 2000), and as a result of this isolation, the area presents high levels of endemism at the genus and species level. The macroinvertebrate fauna of the Róbalo river, resembles that of other Patagonian streams (Miserendino and Pizzolon 2000) as well as some similar streams in the Northern hemisphere and New Zealand (Winterbourn 1978, Miserendino and Pizzolon 2000) in which dipterans, plecopterans, ephemeropterans, and trichopterans are the predominant groups (Ward 1986). The invertebrate faunas of many stony streams in New Zealand and Australia are also dominated by these four orders of insects, with many of the same families being prominent (Miserendino and Pizzolon 2003) and with New Zealand and South America having the strongest affinities at the family level. For example, many of the families (Leptophlebiidae, Austroperlidae, Gripopterygidae, Notonemouridae, and Hydrobiosidae) present at the Róbalo River, and other Patagonian streams (Miserendino and Pizzolon 2000, 2003), are also present in New Zealand streams. However, several families of freshwater invertebrates that are widespread or common in Patagonia and Sub-Antarctic streams, are not found in New Zealand, and include Perlidae, Baetidae, Glossosomatidae, Limnephilidae, and Hyallelidae (Miserendino and Pizzolon 2003). In this study, the order Diptera was the most diverse and abundant through the altitudinal gradient (14 genera), followed by Trichoptera (8
genera), Plecoptera (6 genera), and Ephemeroptera (3 genera).

Although the Cape Horn Biosphere Reserve is one of the 24 most pristine areas remaining in the world (Mittermeier et al. 2003), it is an area replete with exotic invasive species, such as the North American beaver (*Castor canadensis*) (Anderson and Rosemond 2007b), which was introduced to the area in 1946. Today, the North American beaver has invaded almost 100% of all rivers and streams in Navarino Island. In this context, the altitudinal gradient of the Róbalo River is not free from this invasion, as one of the study sites (site IV, 240 m.a.s.l), was heavily impacted by an abandoned beaver dam. In contrast with other Patagonian streams and Northern hemisphere streams (Miserendino and Archangelsky 2006a), total density of benthic macroinvertebrates did not increase with decreasing altitude, instead, total density was significantly higher at site IV (240 m.a.s.l). This result is more than likely due to the effect of the beaver, as beaver engineering in ponds, create taxonomically simplified, but more productive benthic macroinvertebrate assemblages in the Cape Horn Biosphere Reserve (Anderson and Rosemond 2007b). Furthermore, macroinvertebrate richness, diversity and functional feeding groups were significantly lower at this site, further complementing previous studies associated to monitoring the effects of the exotic invasive north American beaver on benthic macroinvertebrate community assemblages. As it was expected, density of filterers significantly increased towards the mouth of the Róbalo River, and in contrast with other Patagonian streams, collector-gatherers showed a fluctuating density pattern through the altitudinal gradient, rather than an uniform increasing pattern towards the mouth of the river (Miserendino and Pizzolon 2000) (Table 5). Predators and scrapers remained constant through the altitudinal gradient, while shredder’s density was significantly higher at sites IV and V (240 and 120 m.a.s.l).
Benthic Macroinvertebrate Assemblages Along the Altitudinal Gradient of the Róbalo River

Water temperature is one of the major factors determining the distribution and life histories of aquatic insects along gradients of latitude and elevation (Vannote et al. 1980). Furthermore, temperature is greatly affected by climatic changes along gradients of altitude or latitude. In this study, we found that the Róbalo river watershed is characterized by a sharp altitudinal gradient, in which the cumulative degree-days per year, sharply increase by a magnitude of 4x (from 501-2037 CDD) from its headwaters to its mouth (Fig. 8), through a relatively short altitudinal gradient (12 km, 0-600 m.a.s.l). This is notably contrasted with northern hemisphere streams, where the same accumulation of yearly degree-days are reached at significantly higher altitudes, as has been shown for streams in the Rocky Mountains, where the same CDD are recorded for altitudinal gradients ranging from 0-1500 m.a.s.l (Hauer et al. 1997).

Benthic macroinvertebrate distributions are highly correlated with elevation and the thermal characteristics along the gradient. This result is similar to studies carried in the northern hemisphere (McDonald Creek, Glacier National Park, 48°N), where the same results are found through an altitudinal gradient reaching 2,912 m of elevation (Hauer et al. 2000). In this context, we propose the Róbalo River as a reference site of study for other Sub-Antarctic streams within the Cape Horn Biosphere Reserve, as PCA and MaxR² results, show clear and significant relationships with the parameters just described, as well as with substrate diversity and year of sampling (Fig. 13, Tables 6 and 7). In this way, we can conclude that density and distribution patterns among similar species and across taxonomic groups, as found in this study, can be used to predict distribution and abundance patterns of benthic macroinvertebrates at other watersheds within the Cape Horn Biosphere Reserve. Thus, the Róbalo River provides an ideal setting for long-term monitoring of benthic macroinvertebrate community dynamics through a sharp Sub-
Antarctic altitudinal gradient. Furthermore, because of the sharp response of benthic macroinvertebrate communities to altitude and temperature, this study emphasizes the value of macroinvertebrates as ideal indicators of thermal modifications that may be associated with change in land, riparian vegetation or climate change scenarios (Hauer et al. 2000).

Chapter References


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Fig. 6. Ecoregion of Sub-Antarctic Magellanic forests (dark gray). Navarino Island is located north of Cape Horn and south of the Beagle Channel. The Cape Horn Biosphere Reserve and the Omora Ethnobotanical Park are located within this ecoregion (Rozzi et al. 2008).
Fig. 7. Altitudinal gradient and sampling sites through the Róbalo river watershed. This altitudinal gradient includes the characteristic habitat types of the Cape Horn region. From the shoreline upwards these habitats are: evergreen forests dominated by *Nothofagus betuloides*; mixed evergreen-deciduous forests dominated by *N. betuloides* and *N. pumilio*; deciduous forests dominated by *N. pumilio* and *N. antarctica*; alpine tundra dominated at lower levels by cushion plants, e.g., *Bolax gummifera*; and at upper levels by lichens such as *Neuropogon* sp. Fig. modified from (Rozzi et al. 2010). Sampling sites are represented with roman numerals I – V and range from 120 – 586 m.a.s.l.

Fig. 8. Total Cumulative Degree-Days (CDD) obtained for a period of 14 months from the headwaters to the mouth (sites II-V) of the altitudinal gradient of the Róbalo River watershed.
Fig. 9 Mean richness and evenness (Pielou’s J Evenness Index) (±SE) of benthic macroinvertebrates along the altitudinal gradient of the Róbalo River watershed for the years 2008, 2009, and 2010.
Fig. 10. Regression of substrate diversity (Shannon-Wiener diversity Index $H'$) and benthic macroinvertebrate richness along the altitudinal gradient of the Róbalo river watershed. Data was combined for all three sampling years.

Fig. 11. Beanplot of altitudinal patterns of density (mean number of ind./m$^2$) for benthic macroinvertebrates along the altitudinal gradient of the Róbalo River watershed. Each “bean” consists of a density trace, which is mirrored to form a polygon shape (Kampstra 2008) and the individual observations are shown as small white lines, which in this case correspond to the years 2008, 2009 and 2010. The dotted line indicates the general median, while the solid line indicates the average for each group.
Fig. 12. Beanplots of altitudinal patterns of density (mean number of ind./m²) for the major aquatic insect orders along the altitudinal gradient of the Róbalo River watershed. As in the previous graph, each “bean” consists of a density trace (Kampstra 2008), which is mirrored to form a polygon shape and the individual observations are shown as small white lines. The dotted line indicates the general median, while the solid line indicates the average for each group.

Fig. 13. Principal components analysis (PCA) for the distribution of macroinvertebrates that contribute 90% of the abundance along the altitudinal gradient of the Róbalo river watershed, in relation to conductivity (μS), dissolved oxygen (DO, mg/L), substrate diversity (H’), year of sampling, and altitude (m.a.s.l).
Table 1  Sampling sites selected to perform characterization of benthic macroinvertebrate fauna associated to the Róbalo River watershed (55°S)

<table>
<thead>
<tr>
<th>Site</th>
<th>Description</th>
<th>Altitude (m)</th>
<th>GPS Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Headwaters</td>
<td>586.0</td>
<td>S 54° 59.674', W 067° 40.878'</td>
</tr>
<tr>
<td>II</td>
<td>Laguna del Salto</td>
<td>486.0</td>
<td>S 54° 59.520', W 067° 40.869'</td>
</tr>
<tr>
<td>III</td>
<td>Laguna de la Tortuga</td>
<td>380.0</td>
<td>S 54° 59.225', W 067° 40.858'</td>
</tr>
<tr>
<td>IV</td>
<td>Former Beaver dam</td>
<td>240.0</td>
<td>S 54° 58.289', W 067° 39.926'</td>
</tr>
<tr>
<td>V</td>
<td>Downstream</td>
<td>120.0</td>
<td>S 54° 57.106', W 067° 38.606'</td>
</tr>
</tbody>
</table>

Table 2  Description of physical habitat variables along the altitudinal gradient of the Róbalo River watershed, Navarino Island (55°S). Means (±SE) are provided for stream depth and width, as well as bank slope. Annual degree-days along the altitudinal gradient were compared with a one-way ANOVA. Values within each altitude with different letters were significantly different with Tukey HSD test (α 0.05)

<table>
<thead>
<tr>
<th>Altitude (m.a.s.l)</th>
<th>120</th>
<th>240</th>
<th>380</th>
<th>486</th>
<th>586</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-Vascular herbaceous cover (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Left bank</td>
<td>7.6</td>
<td>10.3</td>
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<td>29.4</td>
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<tr>
<td>Right bank</td>
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<td>4.4</td>
<td>0</td>
<td>9.9</td>
<td>38.5</td>
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<tr>
<td>Vascular herbaceous cover (%)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Left bank</td>
<td>54.8</td>
<td>36.8</td>
<td>44.9</td>
<td>49.2</td>
<td>12.8</td>
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<tr>
<td>Right bank</td>
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<td>48.5</td>
<td>52.2</td>
<td>29.5</td>
<td>19.4</td>
</tr>
<tr>
<td>Substrate diversity (H')</td>
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<td>1.9</td>
<td>1.9</td>
<td>1.8</td>
<td>1.5</td>
</tr>
<tr>
<td>Substrate type (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Very Fine Gravel (0.2 - 0.4 cm)</td>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Fine Gravel (0.4 - 0.8 cm)</td>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Medium Gravel (0.8 - 1.6 cm)</td>
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<td>1</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Coarse Gravel (1.6 - 3.2 cm)</td>
<td>25.7</td>
<td>20.4</td>
<td>5</td>
<td>11</td>
<td>0</td>
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<tr>
<td>Very Coarse Gravel (3.2 - 6.4 cm)</td>
<td>31.7</td>
<td>33.7</td>
<td>24</td>
<td>22</td>
<td>0</td>
</tr>
<tr>
<td>Small Cobble (6.4 - 9.0 cm)</td>
<td>11.9</td>
<td>21.4</td>
<td>17</td>
<td>16</td>
<td>0</td>
</tr>
<tr>
<td>Medium Cobble (9.0 - 12.0 cm)</td>
<td>9.9</td>
<td>5.1</td>
<td>15</td>
<td>17</td>
<td>0</td>
</tr>
<tr>
<td>Large Cobble (12.8 - 18 cm)</td>
<td>7.9</td>
<td>11.2</td>
<td>10</td>
<td>10</td>
<td>8</td>
</tr>
<tr>
<td>Very Large Cobble (18.0 - 25.6 cm)</td>
<td>0</td>
<td>6.1</td>
<td>2</td>
<td>3</td>
<td>12</td>
</tr>
<tr>
<td>Small Boulder (25.6 - 51.2 cm)</td>
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<td>1</td>
<td>1</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>Large Boulder (102.4 - 204.8 cm)</td>
<td>0</td>
<td>0</td>
<td>25</td>
<td>21</td>
<td>77</td>
</tr>
<tr>
<td>Stream width (m)</td>
<td>8.5(3.3)</td>
<td>8.8(2.1)</td>
<td>2.8(0.9)</td>
<td>2.3(0.9)</td>
<td>2.6(0.4)</td>
</tr>
<tr>
<td>Stream depth (cm)</td>
<td>29.0(5.2)</td>
<td>28.3(5.8)</td>
<td>13.4(2.6)</td>
<td>7.7(2.8)</td>
<td>6.8(1.1)</td>
</tr>
<tr>
<td>Bank slope (°)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left bank</td>
<td>31.7(6.7)</td>
<td>24.0(2.0)</td>
<td>12.7(11.2)</td>
<td>16.1(4.9)</td>
<td>6.6(1.3)</td>
</tr>
<tr>
<td>Right bank</td>
<td>18.6(12.6)</td>
<td>14.3(7.9)</td>
<td>22.2(9.7)</td>
<td>4.8(4.6)</td>
<td>10.0(3.7)</td>
</tr>
<tr>
<td>Yearly temperature range (°C)</td>
<td>0 - 17.0</td>
<td>0 - 16.0</td>
<td>0 - 12.7</td>
<td>0 - 10.0</td>
<td>NA</td>
</tr>
<tr>
<td>Total cumulative degree-days (&gt;0°C)</td>
<td>2037D</td>
<td>1631C</td>
<td>971B</td>
<td>501A</td>
<td>NA</td>
</tr>
</tbody>
</table>
Table 3 Mean physical-chemical parameters values (±SE) throughout the altitudinal gradient of the Róbalo River watershed. Means (±SE) were compared with a two-way ANOVA. Values are based on data obtained during austral summers of 2008, 2009, and 2010 (n=16). Values within each category with different letters were significantly different with a Tukey HSD test (α 0.05).

<table>
<thead>
<tr>
<th>Year</th>
<th>Parameter</th>
<th>120</th>
<th>240</th>
<th>386</th>
<th>486</th>
<th>577</th>
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</thead>
<tbody>
<tr>
<td>2008</td>
<td>D.O (mg/L)</td>
<td>10.8 (0.3)</td>
<td>10.5 (0.3)</td>
<td>11.3 (0.3)</td>
<td>12.5 (0.3)</td>
<td>12.6 (0.3)</td>
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<tr>
<td></td>
<td>Conductivity (μS)</td>
<td>56.1 (2.7)</td>
<td>45.6 (9.1)</td>
<td>42.3 (2.0)</td>
<td>29.3 (6.2)</td>
<td>28.3 (1.1)</td>
</tr>
<tr>
<td></td>
<td>pH</td>
<td>8.3 (0.3)</td>
<td>8.8 (0.3)</td>
<td>8.3 (0.3)</td>
<td>8.6 (0.3)</td>
<td>8.9 (0.3)</td>
</tr>
<tr>
<td></td>
<td>Temp (°C)</td>
<td>8.6 (0.2)</td>
<td>4.7 (0.3)</td>
<td>5.4 (0.6)</td>
<td>5.5 (1.0)</td>
<td>4.4 (0.1)</td>
</tr>
<tr>
<td>2009</td>
<td>D.O (mg/L)</td>
<td>11.8 (0.3)</td>
<td>11.8 (0.3)</td>
<td>14.0 (0.3)</td>
<td>13.7 (0.3)</td>
<td>13.6 (0.3)</td>
</tr>
<tr>
<td></td>
<td>Conductivity (μS)</td>
<td>54.2 (8.2)</td>
<td>54.6 (7.0)</td>
<td>41.3 (6.5)</td>
<td>36.9 (1.9)</td>
<td>33.8 (3.4)</td>
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<tr>
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<td>pH</td>
<td>8.4 (0.3)</td>
<td>8.3 (0.3)</td>
<td>8.6 (0.3)</td>
<td>8.5 (0.3)</td>
<td>8.4 (0.3)</td>
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<tr>
<td></td>
<td>Temp (°C)</td>
<td>13.6 (0.4)</td>
<td>11.9 (0.9)</td>
<td>11.2 (0.9)</td>
<td>12.8 (0.8)</td>
<td>10.5 (0.7)</td>
</tr>
<tr>
<td>2010</td>
<td>D.O (mg/L)</td>
<td>8.9 (0.3)</td>
<td>9.3 (0.3)</td>
<td>8.7 (0.3)</td>
<td>10.6 (0.3)</td>
<td>10.9 (0.3)</td>
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<tr>
<td></td>
<td>Conductivity (μS)</td>
<td>54.2 (1.7)</td>
<td>54.6 (7.0)</td>
<td>44.4 (11.5)</td>
<td>31.6 (3.9)</td>
<td>25.7 (3.7)</td>
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<td>pH</td>
<td>8.4 (0.3)</td>
<td>8.3 (0.3)</td>
<td>8.6 (0.3)</td>
<td>8.5 (0.3)</td>
<td>8.3 (0.3)</td>
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<td></td>
<td>Temp (°C)</td>
<td>14.5 (0.7)</td>
<td>11.3 (0.6)</td>
<td>10.3 (0.1)</td>
<td>9.9 (0.3)</td>
<td>6.8 (0.4)</td>
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Table 4 Two-way ANOVA of the effects of altitude (m.a.s.l) and year (2008, 2009, 2010) on the density (individuals/m²) of benthic macroinvertebrate Functional Feeding Groups (FFG) through the altitudinal gradient of the Róbalo River (significant values in bold, α 0.05).

<table>
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<tr>
<th>Source</th>
<th>df</th>
<th>FFG</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F</th>
<th>p</th>
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<tr>
<td>Altitude</td>
<td>4</td>
<td>Collector Gatherer</td>
<td>1.15</td>
<td>0.28</td>
<td>6.02</td>
<td>0.001</td>
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<td></td>
<td></td>
<td>Filterer</td>
<td>22.03</td>
<td>5.51</td>
<td>23.38</td>
<td>&gt;0.001</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Predator</td>
<td>3.52</td>
<td>0.88</td>
<td>6.64</td>
<td>&gt;0.001</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Scraper</td>
<td>1.51</td>
<td>0.38</td>
<td>2.66</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Shredder</td>
<td>1.64</td>
<td>0.41</td>
<td>0.66</td>
<td>0.62</td>
</tr>
<tr>
<td>Year</td>
<td>2</td>
<td>Collector Gatherer</td>
<td>0.15</td>
<td>0.07</td>
<td>1.66</td>
<td>0.22</td>
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<tr>
<td></td>
<td></td>
<td>Filterer</td>
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<td>1.17</td>
<td>4.98</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Predator</td>
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<td>0.23</td>
<td>1.72</td>
<td>0.19</td>
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<td>0.36</td>
<td>2.51</td>
<td>0.09</td>
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<tr>
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<td></td>
<td>Shredder</td>
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<td>2.12</td>
<td>3.41</td>
<td>0.04</td>
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<tr>
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<td>8</td>
<td>Collector Gatherer</td>
<td>0.91</td>
<td>0.11</td>
<td>2.37</td>
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<td>0.03</td>
<td>0.25</td>
<td>0.97</td>
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<tr>
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<td>Scraper</td>
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<td>0.11</td>
<td>0.76</td>
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<td>Shredder</td>
<td>2.36</td>
<td>0.29</td>
<td>0.47</td>
<td>0.86</td>
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</table>
Table 5 Mean density (individuals/m²) values (±SE) of benthic macroinvertebrate functional feeding groups through the altitudinal gradient of the Róbalo River. Values are based on benthic macroinvertebrate samplings during the austral summers of 2008, 2009, and 2010. Values within each category with different letters were significantly different with a Tukey HSD test (α 0.05)

<table>
<thead>
<tr>
<th>Functional Feeding Group</th>
<th>120</th>
<th>240</th>
<th>380</th>
<th>486</th>
<th>586</th>
</tr>
</thead>
<tbody>
<tr>
<td>Collector Gatherer</td>
<td>180.2(43.3)bC</td>
<td>388.5(76.3)b</td>
<td>457.5(64.0)ab</td>
<td>211.1(50.7)b</td>
<td>484.9(23.4)a</td>
</tr>
<tr>
<td>Filterer</td>
<td>370.1(45.2)b</td>
<td>896.6(41.0)c</td>
<td>259.5(54.2)b</td>
<td>38.4(12.3)a</td>
<td>62.7(4.3)a</td>
</tr>
<tr>
<td>Predator</td>
<td>45.3(6.5)a</td>
<td>223.6(37.8)b</td>
<td>85.0(12.2)a</td>
<td>41.6(6.8)a</td>
<td>49.8(5.9)a</td>
</tr>
<tr>
<td>Scraper</td>
<td>63.8(7.4)a</td>
<td>111.9(38.5)b</td>
<td>47.2(19.2)a</td>
<td>44.2(15.1)a</td>
<td>54.9(9.4)a</td>
</tr>
<tr>
<td>Shredder</td>
<td>79.9(13.4)b</td>
<td>96.8(17.2)b</td>
<td>37.1(8.3)a</td>
<td>30.4(9.6)a</td>
<td>34.4(19.6)a</td>
</tr>
</tbody>
</table>

Table 6 Results of principal component analysis. Eigen values for the first 4 principal components are provided

<table>
<thead>
<tr>
<th>Principal Components</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
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<tr>
<td>Axes</td>
<td>Eigen values</td>
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<td></td>
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</tr>
<tr>
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<td>5.00</td>
<td>3.25</td>
<td>2.22</td>
<td>1.70</td>
</tr>
<tr>
<td>Variability percentage variance</td>
<td>27.80</td>
<td>18.05</td>
<td>12.33</td>
<td>9.42</td>
</tr>
<tr>
<td>Cumulative percentage variance</td>
<td>27.80</td>
<td>45.84</td>
<td>58.18</td>
<td>67.60</td>
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</table>
Table 7 Max $R^2$ regression summary of benthic macroinvertebrates in response to five selected variables (altitude, temperature, year, substrate, and conductivity) through the altitudinal gradient of the Róbalo River. Models were selected based on overall $p$ values $<0.05$ and taxa that exhibited an $R^2$ value $> 0.30$ for each proposed model

<table>
<thead>
<tr>
<th>Order</th>
<th>Family/Subfamily</th>
<th>Genus</th>
<th>$R^2$</th>
<th>$p$</th>
<th>1$^{st}$ variable</th>
<th>Partial $R^2$</th>
<th>2$^{nd}$ Variable</th>
<th>Partial $R^2$</th>
<th>3$^{rd}$ Variable</th>
<th>Partial $R^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diptera</td>
<td>Simuliidae</td>
<td>Gigantodax</td>
<td>0.74</td>
<td>$&lt;0.0001$</td>
<td>Altitude</td>
<td>-0.71</td>
<td>Substrate</td>
<td>0.47</td>
<td>Temperature</td>
<td>0.34</td>
</tr>
<tr>
<td>Diptera</td>
<td>Chironomidae/Orthocladiinae</td>
<td></td>
<td>0.68</td>
<td>0.000</td>
<td>Altitude</td>
<td>0.69</td>
<td>Substrate</td>
<td>-0.48</td>
<td>Temperature</td>
<td>-0.34</td>
</tr>
<tr>
<td>Diptera</td>
<td>Blephariceridae</td>
<td>Edwarsina</td>
<td>0.63</td>
<td>$&lt;0.0001$</td>
<td>Altitude</td>
<td>-0.57</td>
<td>Temperature</td>
<td>0.43</td>
<td>Substrate</td>
<td>0.33</td>
</tr>
<tr>
<td>Plecoptera</td>
<td>Griopterygidae</td>
<td>Pelugorperla</td>
<td>0.60</td>
<td>$&lt;0.0001$</td>
<td>Temperature</td>
<td>0.56</td>
<td>Year</td>
<td>0.39</td>
<td>Altitude</td>
<td>-0.19</td>
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<td>Altitude</td>
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<td>Substrate</td>
<td>-0.50</td>
<td>Year</td>
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<td>Nesameletidae</td>
<td>Metamonius</td>
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<td>0.010</td>
<td>Year</td>
<td>0.42</td>
<td>Altitude</td>
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<td>Temperature</td>
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<td>Trichoptera</td>
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<td>Rheochorema</td>
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<td>0.005</td>
<td>Altitude</td>
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<td>Substrate</td>
<td>0.20</td>
<td>Temperature</td>
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</tr>
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<td>Altitude</td>
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<td>Luchoelmis</td>
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<td>Substrate</td>
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<td>Substrate</td>
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<td>Conductivity</td>
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<td>Meridialaris</td>
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<td>Substrate</td>
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<td>0.33</td>
<td>Year</td>
<td>-0.20</td>
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Table 8 Total list of taxa described throughout the altitudinal gradient of the Róbalo River watershed during the course of the study. FFG refers to “Functional Feeding Group”: par-pred: parasite-predator; cg= collector-gatherer; sc= scraper; fil=filterer; sh= shredder; pred=predator. “x” denotes presence at a particular altitudinal location

<table>
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<th>Class</th>
<th>Order/suborder</th>
<th>Family</th>
<th>Sub-Family</th>
<th>Genus</th>
<th>Species</th>
<th>FFG</th>
<th>Altitude (m.a.s.l)</th>
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<td>sp.</td>
<td>cg</td>
<td>x</td>
<td>x</td>
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<td>sp.</td>
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CHAPTER 4
LIFE HISTORY OF *Gigantodax* sp. ENDERLEIN 1925 (SIMULIIIDAE: DIPTERA) IN A SOUTHERN SOUTH AMERICA SUB-ANTARCTIC STREAM AND THE POTENTIAL EFFECTS OF CLIMATE CHANGE

Abstract

The life history of the world’s southernmost blackfly (*Gigantodax* sp. Enderlein 1995 (Simuliidae: Diptera)) was studied in the altitudinal gradient of a Sub-Antarctic fluvial system (Rôbalo river, 55° S, Cape Horn Biosphere Reserve, Chile) from August 2009 to August 2010. This study provides the first comprehensive life history description for any aquatic insect in the Sub-Antarctic ecoregion of southern South America. Results indicate that the life history of *Gigantodax* sp. ranges from univoltine (headwaters, 560 meters above sea level (m.a.s.l)) to multivoltine (mouth, 0 m.a.s.l) through the altitudinal gradient studied. We propose *Gigantodax* sp. as a suitable indicator in the context of climate change studies, as a small rise (2°C) in temperature could switch its life history to primarily multivoltine, provoking serious changes on benthic macroinvertebrate community structure and geographic distributions, amongst others. Furthermore, this multivoltine pattern differs with the prevalence of univoltine life cycles recorded for Simuliidae and other freshwater taxa at equivalent latitudes in North America, further evidencing the marked contrasts exhibited by Northern and Southern Hemisphere temperate and sub polar biomes.

Introduction

Black flies (Simuliidae: Diptera) are holometabolous insects that are key in both aquatic and terrestrial ecosystems. The 2000 nominal species of black flies are worldwide in distribution and occur in all continents except Antarctica (Currie and Adler 2008). Larvae occur in huge
numbers under favorable conditions, attaining population densities of up to a million
individuals/m$^2$ (Currie and Adler 2008). They feed on particular organic matter and play an
important role in the processing of organic matter. Because of their filter-feeding habit, black
flies are of great importance in terms of environmental monitoring of freshwater contamination,
particularly because immature stages (larvae and pupae) are highly susceptible to both organic
and inorganic pollution. The genus *Gigantodax* (Diptera: Simuliidae) is the largest genus of
Neotropical Prosimuliini, with 68 species extending along the Andean mountain system from
Mexico (Wygodzinsky and Coscarón 1989) to Cape Horn. Its distribution ranges from sea level
to high altitudes (4700 m in the tropics), but it generally prefers altitudes from 700 to 2000 m
(Wygodzinsky and Coscarón 1989). The Sub-Antarctic islands of Cape Horn, located within the
Magellanic Sub-Antarctic ecoregion, host the world’s southernmost populations of *Gigantodax*
sp.

The Magellanic Sub-Antarctic ecoregion (48°-56°S) is the southernmost portion of the
temperate forests of southwestern South America, which extend approximately 30°-56°S along
Chile and Argentina (Ippi et al. 2009). It is considered one of the 24 most pristine areas left in
the world (Mittermeier et al. 2003), and it is one of the only areas where temperate forests
remain unfragmented and virtually unaltered (Dunn 2005a). It encompasses all of the islands
south of the Beagle Channel, as well as the Chilean portion of Tierra del Fuego Island located
south of the highest peaks in the Darwin Mountain Range (Anderson 2006). The unique flora
and fauna of this ecoregion have diverse origins, in particular the two divergent Neotropical and
share a circum-Antarctic taxonomic affinity with those of New Zealand, South Africa, and
Australia, given that they all once constituted the Gondwana continent (Moorman et al. 2006). In
this context, the fluvial systems of the Sub-Antarctic ecoregion in southern Chile are home to the world’s southernmost populations of several species and families of aquatic insects, such as members of the families Scirtidae, Elmidae, Dytiscidae (Coleoptera); and Blephlariceridae, Ceratopogonidae, Empididae, Thaumaleidae, Tipulidae, and Simuliidae (Diptera), (Moorman et al. 2006, Ashworth et al. 2011).

The islands located within this ecoregion and the Cape Horn Biosphere Reserve (CHBR, 55oS), have relatively limited seasonal variation, with a mean air temperature that ranges between 1 and 10°C, buffered by the surrounding ocean. As a consequence, many Sub-Antarctic invertebrates show extensive overlap of generations, often with little or no seasonal structure to the life-cycle, and no true diapause in response to specific cues (Convey 1996).

Life-history information is of fundamental importance for virtually all ecological studies of freshwater invertebrates (Bossart and Carlton 2002). The effects of thermal variation on life histories and habitat selection are particularly important to consider because freshwater insects are able to persist over a temperature range of < 20°C to almost 50°C (Merrit et al. 2008). In addition, many stream insects use temperature or temperature change as an environmental cue for emergence (Rozzi et al. 2000a, Bossart and Carlton 2002). The larval developmental period of a population can often be predicted using summation of thermal units (i.e. degree days) (Sweeney 1984). Degree-days (DD) models are useful in predicting biological events in the development of plants and poikilothermic animals (Pedigo et al. 1996). By calculating total number of DD that are accumulated each day, and comparing this total number of DD required for an event to occur (e.g., egg hatch, emergence), we can estimate the date on which a certain event occurred and predict future events, such as emergence patterns (Pedigo et al. 1996).
The thermal diversity to which aquatic insects are exposed varies spatially and temporally and is subject to anthropogenic alteration (Ward and Standford 1982). Anthropogenic impacts, such as climate change, have already affected the phenology of various taxa, covering all trophic levels (Pisano 1977, Parmesan 2003, Parmesan 2006, Christopher Hassall 2007, Richter and Suhling 2008). Such shifts in phenology may affect the synchrony of key activities with the supply of food or habitat, thus causing a temporal de-coupling or mismatch that may even lead to population crashes or extinctions (Parmesan 2006, Richter and Suhling 2008). Several examples of ecological consequences of recent climate change have been described, including changes in population size and shifts in geographic range, as well as community and ecosystem-level effects (Annette Menzel 2006, Parmesan 2006, Isabelle Durance 2007, Parmesan 2007, Richter and Suhling 2008). Thus, the necessity of predicting the potential effects of any temporal mismatch of life cycles with environmental parameters, such as temperature under different climate change scenarios, becomes essential when assessing the effects of temperature change over organisms, populations, and ecosystems.

In this context, the purpose of this study was to describe the phenology of *Gigantodax* sp throughout the altitudinal gradient of a South American Sub-Antarctic fluvial system, characteristic of the of the Magellanic Sub-Antarctic ecoregion, in order to identify whether it exhibits univoltine, multivoltine or both types of life histories in this region of the world. Growing degree-days (GDD) were obtained from water temperature data collected throughout the altitudinal gradient. Voltinism patterns through the gradient were predicted as well as the effects of environmental warming on *Gigantodax* sp.’s life cycle and the possible implications for Sub-Antarctic stream ecosystems.
Materials and Methods

Study Area

The Róbalo River (S 54° 59.647’, W 067° 40.878’, Navarino Island) is located within the Cape Horn Biosphere Reserve (CHBR) and it provides drinking water to Puerto Williams, the Chilean Capital of the Antarctic Province and the world’s southernmost town. It runs through the marked altitudinal gradient of the Dientes of Navarino Mountain Range, where it drains into the Beagle Chanel. Navarino Island lies south of the Beagle Channel and north of Cape Horn and it has an approximate area of 2500 km² (Fig. 14). The forests of the island consist primarily of three members of the genus *Nothofagus* (Pisano 1977) and are part of the South American temperate forests biome, which occurs along a narrow but latitudinally extensive strip of land between 35° and 55°S (Armesto et al. 1998). These forests are composed of mixed evergreen *Nothofagus betuloides* (Coihüe de Magallanes), high deciduous southern beech, *N. pumilio* (Lenga), and the deciduous *N. antarctica* (Ñirre) (McCulloch et al. 1997a). The CHBR encompasses all of the islands south of the Beagle Channel, as well as the Chilean portion of Tierra del Fuego Island located south of the highest peaks in the Darwin Mountain Range (Rozzi et al. 2006). The study site lies on the north Coast of Navarino Island, which houses the city of Puerto Williams, and the Omora Ethnobotanical Park, part of Chile’s network of Long Term Socio-Ecological Research Sites (LTSER). Furthermore, it is the Southern hemisphere’s latitudinal equivalent of northern British Columbia, Canada and Southern Alaska (USA).

The river is characteristic of the streams of the island (clear waters and partially forested) and it has an approximate length of approximately 12 km, ascending from 0 meters above sea level (m.a.s.l) to 600 m.a.s.l. We selected five sampling sites ranging from locations near its mouth at 120 m.a.s.l to its headwaters at 587 m.a.s.l. The sites were selected based on the marked
vegetation patterns observed through the gradient (Fig. 15). It is important to note that this altitudinal gradient is highly representative of Sub-Antarctic ecosystems, as it exhibits all of the habitat types that can be observed within this ecoregion in a short walking distance and relatively low altitudes. Benthic macroinvertebrates were sampled at each of these sites during the austral summers of 2008, 2009, and 2010, and one permanent site, at 120 m.a.s.l, was selected to be continuously sampled for a period of 12 months (details explained below), starting in August of 2009.

Climate

The regional climate of the area is strongly affected by the westerly storm tracks coupled with precipitation induced by the high western flanks of the Andean Cordillera (McCulloch et al. 1997b). It also depends on polar weather fronts from the south Pacific (Rozzi et al. 2006a). The town of Puerto Williams is found within the influence of the Dfk’c Transandeian with steppe degeneration climatic zone. It represents a transition between rain climates of the western coast of Magallanes and the steppe climates of the eastern side of the mountain range (Rozzi et al. 2006a). The uniformity of the seasonal variation of precipitation as well as its thermal characteristics, allow for the growth of deciduous forests. The average annual temperature and precipitation recorded for Puerto Williams is 6.0°C and 467.3 mm respectively. Precipitation is less polluted than for any other of the world’s temperate forests (Likens 1991) and the warmest month has an average temperature of 9.6°C (January-March), while the coldest month averages 1.9°C (May-August). Compared to similar northern latitudes (such as Sub-Arctic and Arctic environments of northern North America), where the climate is continental in nature and it is characterized by harsh and extreme environments (temperature ranges from -14°C to 5°C)
(Strathdee and Bale 1998), the Sub-Antarctic tip of South America is characterized by cool but not extreme climate year-round, with relatively low seasonal variation in temperatures due to oceanic damping (Convey 1996).

Field and Laboratory Methods

Physical – Chemical Parameters

Water chemistry parameters were recorded at each site and during each sampling event. Dissolved oxygen (mg/L) and conductivity (μS) were recorded using a YSI 8.5 meter. From January of 2009 to January of 2011, water temperature (°C) was recorded every 5 hours at each sampling site throughout the altitudinal gradient. Recordings were obtained using Hobo® data loggers (Model HOBO® U22 Water Temp Pro v2). Differences between sites were tested using one-way ANOVAs. When significant results were obtained, a Tukey HSD post hoc test was applied (R Software version 2.9.2). All data was tested for normality (Shapiro-Wilk w statistic) and inequality of variances (Hartley’s $F_{max}$ ratio test) before proceeding with ANOVA ($\alpha 0.05$).

Benthic Macroinvertebrate Sampling

Benthic macroinvertebrates were sampled at each of the sites during the austral summers of 2008, 2009, and 2010, and one permanent site (at 120 m.a.s.l) was sampled continuously for a period of 12 months starting in August of 2009. For the purposes of this study, results obtained from the sampling carried out at site V (120 m.a.s.l) are presented, and voltinism patterns are predicted based on water temperature (°C) data obtained for the remaining sites.

In order to study the phenology of Gigantodax sp., larval insects were collected from site V (120 m.a.s.l) every 2 weeks. Three samples were taken from riffle habitats using a Surber net
(0.09m²) with a 243μm mesh. Each sample was stored in 70% ethanol and transported to the laboratory for processing.

**Determination of Gigantodax sp. Larval Instars and Voltinism**

*Gigantodax* sp. larval instars were determined by measuring head capsule lengths (HCL) (mm) and widths (HDW) (mm) with an Olympus SHZ dissecting microscope coupled with a Tucsen TCA 5.0C (5mp) camera with TSview digital imagining processing software. Instars were separated by producing a scatter plot of HCL against HCW (Wang and Kennedy 2004) and distinguishing characteristics for each instar were described. HCL was measured from the anterior margin of the frons to the posterior margin of the head’s sclerites, while HCW was measured as the distance of the head capsule across the eyes. Identification to the genus level was done using (Wygodzinsky and Coscarón 1989).

The relative abundance of each larval instar and the total number of pupae (per sampling period) were used to estimate cohort lines and voltinism. Because at least 3 *Gigantodax* sp. larval morphotypes, were identified, *Gigantodax* sp. Specimens were reared out in the laboratory until pupation was reached, in order to match each larval morphotype to its appropriate pupae. To estimate the starting point of a generation, the time period at which proportionally higher relative abundance of first larval instars was recorded. Approximate emergence of adults was estimated by noting the time period at which a proportionally higher number of fifth instar larvae and total number of pupae was obtained.

**Determination of Cumulative Degree Days and Growing Degree Days**

Average daily water temperature (°C) was calculated for each site using the information
retrieved from the Hobo data loggers. Information from site I (586 m.a.s.l) was not retrieved due to lost equipment, therefore temperature (°C) data for sites II-V (486-120 m.a.s.l) are presented. Cumulative Degree- Days (CDD) for each site were computed by summing the daily average temperature (°C) by month and for a period of 1 year, following (Snyder et al. 2001). Growing Degree-Days (GDD) were estimated by summing the daily average temperature (°C) starting at the period at which a proportionally higher number of first instar larvae were observed, and finishing at the period at which a proportionally higher number of fifth instar larvae was observed.

Estimation of Voltinism through the Altitudinal Gradient

Cohort lines and voltinism through the altitudinal gradient were estimated by obtaining the CDD for each site by month. Using the results of the approximate GDD obtained from site V (120 m.a.s.l), the number of generations (cohorts) that could be possible for each site related to its total number of CDD during a one-year period were estimated.

Results

Physical – Chemical Parameters

Dissolved oxygen (mg/L) and pH were not significantly different between sites throughout the altitudinal gradient (one-way ANOVA, P = 0.53 and 0.89, α 0.05), while conductivity (μS) was significantly higher at sites IV and V (240 and 120 m.a.s.l, one-way ANOVA with Tukey HSD analysis, P = 0.0003, α 0.05) (Table 9).

Maximum water temperatures (°C) were reached during austral summers (December – March) and ranged between 10-17°C from the headwaters to the mouth of the Róbalo River
watershed, while minimum temperatures fell below 0°C during austral winters (June-August). Total cumulative degree-days (CDD) changed dramatically every 100 meters of altitude through the altitudinal gradient, and ranged from 501 to 2037 CDD, from the headwaters to the mouth (Fig. 16).

Determination of *Gigantodax* sp. Larval Instars and Voltinism

- Larval instars: Five distinct larval instars were obtained for *Gigantodax* sp. by plotting head capsule length and width (mm) (Fig. 17). Head capsule lengths ranged from 0.37 mm (first instar) to 0.96 mm (fifth instar), while head capsule widths ranged from 0.29 mm (first instar) to 0.84 mm (fifth instar) (Fig. 17).

- Voltinism: A significantly higher abundance of *Gigantodax* sp. 1st larval instars were observed during December, March, and June (Fig. 18), while a significantly higher abundance of 5th larval instars were observed during February, May, and December. The highest relative abundances of 5th larval instars coincide with the highest numbers of total pupae collected, therefore, by counting the amount of degree-days that it takes for one generation of *Gigantodax* sp. to reach the highest abundance of pupae, it is estimated that it takes approximately 336 degree-days for one generation of *Gigantodax* sp. to complete. With this information, it is concluded that at site V (120 m.a.s.l), where the total CDD per year are 2037, *Gigantodax* sp. has a multivoltine life cycle, with 3 generations per year and with possible adult emergence times during February, May, and December (Fig. 18).

Estimation of Voltinism through the Altitudinal Gradient

As stated before, CDD range from 501-2037 from headwaters to the mouth of the Róbalo
River watershed. With this information, it is estimated that *Gigantodax* sp.’s life history varies from univoltine (1 generation per year), at the river’s headwaters, to multivoltine (3 generations per year) at its mouth (Fig. 19).

**Discussion**

**Multivoltine Life Cycles in South American Sub-Antarctic Streams**

Although life history studies have been carried out for many years in the Northern Hemisphere and other countries of the Southern Hemisphere (i.e. New Zealand), little is known about the life histories of aquatic insects in Patagonia (Beltran and Miserendino 2011) and the Sub-Antarctic ecoregion of southern Chile. This is the first study to report on the life history of any freshwater insect inhabiting Sub-Antarctic South American streams. Our results indicate that *Gigantodax* sp. (Diptera: Simuliidae) has a life history, which varies from univoltine to multivoltine (3 generations per year) from the headwaters to the mouth of the sharp altitudinal gradient of the Róbalo River Watershed. These results directly contrast with other studies carried out in the Southern and Northern Hemisphere at similar latitudes (Table 10). Most aquatic insects of high latitudes and altitudes exhibit a seasonal emergence pattern, as the interaction of temperature, photoperiod, and local climate conditions helps to determine aquatic insect seasonal phenology in temperate regions (DeWalt et al. 1994). Generally, where climates are cold, rapid development is difficult and species with several generations per year are unusual (Danks 2007). Indeed, many Arctic/Antarctic and alpine species have life cycles that last more than one year (Danks 2007). Comparing life history patterns from high and intermediate latitudes in both, the north and south hemisphere (Table 10), we can see that multivoltine life histories are most frequent at lower latitudes (i.e. 34°N), while univoltine, semivoltine, and merovoltine life
histories are most frequent at higher latitudes (i.e. 50°N, 42°S). For example, univoltine and semivoltine life cycles have been described for aquatic insects inhabiting other Sub-Antarctic islands, as in the case of the diving beetle *Lancetes angusticollis* (Curtis) (Coleoptera: Dytiscidae), which exhibits a semivoltine life cycle in South Georgia, one of the coldest Sub-Antarctic islands, with temperatures that range from -5 – 10°C during the summer (Rodney and Convey 1998). Furthermore, the stoneflies *Notoperla fasciata* and *N. magnaspina* (Plecoptera: Gripopterygidae), exhibit semivoltine and merovoltine life cycles in Patagonian streams, where temperatures range from 1°C – 12°C (Beltran and Miserendino 2011). In our study, we were able to determine that *Gigantodax* sp. (Diptera: Simuliidae) exhibits a life cycle that varies from univoltine to multivoltine throughout a sharp altitudinal gradient typical of Cape Horn’s ecosystems. The presence of a multivoline life cycle could be explained through the lack of seasonality experienced on most Sub-Antarctic islands in comparison with maritime and continental Antarctic zones. This may be the most significant influence on the life history of aquatic insects (Convey 1996) inhabiting these areas, where the summers are cool and the winters are not specially cold (Danks 2007) due to oceanic damping. In this sense, cool temperatures year-round, combined with a low or non-existent risk of extreme winter lows, could allow for activity and development to be continuous (Convey 1996). Furthermore, as a result of this limited seasonal variation, many Sub-Antarctic invertebrates show extensive overlap of generations, often with little or no seasonal structure to the life cycle, and no true diapauses in response to specific cues (Convey 1996).

*Gigantodax* sp. as an Indicator of Climate Change in South American Sub-Antarctic Streams

Although climate change and the increase in the predicted frequency of extreme climatic
events have the capacity to change aquatic community structure as well as the geographical
distribution and the development and timing of life histories of individual species (Ryan and
Ryan 2006, Winterbourn et al. 2008), studies on the potential effects of climate change on the
freshwater fauna the Sub-Antarctic islands of Cape Horn have not been carried out. Several
studies have shown that aquatic ecosystems are as vulnerable to global change as terrestrial and
marine ecosystems (Heino et al. 2009), although the evidence is unbalanced across ecosystems
and information from streams is scarce (Durance and Ormerod 2007). Lentic (i.e. lakes and
ponds) and lotic (i.e. streams and rivers) ecosystems are considered to be most sensitive to land
use change, exotic species, and climate change at a global scale, with aquatic ecosystems at high
latitudes being more strongly threatened by climate change (Heino et al. 2009). The
temperatures of streams and rivers vary more rapidly than those of lakes, and apart from spring-
fed streams, water temperatures in running waters are strongly influenced by changes in air
temperature and are usually higher or lower than air temperatures when the latter are low or high
respectively (Elliott 1991). Therefore, temperature changes as a consequence of climate change
would be expected to be more marked in streams and rivers than in other freshwater habitats
(Elliott 1991). This has a direct impact on the phenology of river insects, as they have evolved
under the constraints of a thermally fluctuating system and show little or no ability to
compensate or acclimate to changing environmental temperatures (Sweeney et al. 1992).

In this context, we hypothesize on the effects of global warming on the life history of
Gigantodax sp. and propose it as an indicator in the context of climate change studies, as a 2°C
rise in temperature, as seen in other Sub-Antarctic islands, such as New Zealand and South
Georgia (see (Rodney and Convey 1998, Winterbourn et al. 2008), would cause a switch on the
annual degree-days accumulated along the sharp altitudinal gradients observed through the
course of many of southern South American Sub-Antarctic streams. As we can see in Fig. 20, an increase in 2°C, would switch the annual degree-days from approximately 500 DD at 486 m.a.s.l to almost 1200 at the same altitude, and through the range from 2000 DD to 2800 DD. This switch on the DD accumulated through the altitudinal gradient studied, would have great impacts on the life history *Gigantodax* sp. As we concluded earlier, the total degree-days needed for *Gigantodax* sp. to complete its life cycle are approximately 338 DD and the number of generations varies radically from univoltine (at the headwaters of the Róbalo river watershed) to multivoltine (3 generations per year) at locations close to the mouth of the river. With an increase in the annual accumulated DD such as the one predicted in Fig. 20, the life cycle of *Gigantodax* sp. would switch to completely multivoltine throughout the altitudinal gradient. This could have great ecological implications, not only for *Gigantodax*, but also for many other species inhabiting these Sub-Antarctic streams. In this context, we could expect changes in the trophic structure of benthic macroinvertebrate communities, geographic distribution switches, or even local extinction of various species of aquatic insects, as the cold-stenothermal species inhabiting these streams may not be able to meet their low temperature thresholds for proper egg/larval growth and development to occur (Winterbourn et al. 2008).

**Conclusion**

Previous studies have shown that aquatic insects inhabiting high latitude streams or rivers usually exhibit univoltine, semivoltine, or merovoltine life histories. In this study, we found that the life history of *Gigantodax* sp. varies from univoltine to multivoltine throughout a sharp altitudinal gradient typical of southern South America Sub-Antarctic stream ecosystems. These results differ from other studies carried out on the Sub-Antarctic islands of South Georgia or
New Zealand, where the life histories of the aquatic insects studied are mainly univoltine or merovoltine.

We consider *Gigantodax* sp. a suitable indicator of global climate change in southern South American Sub-Antarctic streams for at least 4 reasons: 1) as a poikilothermic animal, the rate, magnitude of growth, development and overall behavior respond significantly to thermal change on a diel, seasonal, and annual basis (Ward and Standford 1982, Sweeney et al. 1992), 2) it is highly abundant in Sub-Antarctic streams, 3) it is found at altitudes that vary from sea level to high Andean zones, and 4) its life cycle varies from univoltine to multivoltine throughout a short altitudinal gradient, which allows us to closely monitor its phenology and the possible switches (phenological, population ranges, trophic community interactions, etc) that may occur due to a global warming scenario in the short and long term. Finally, the Róbalo River Watershed and the Dientes of Navarino mountain range host a representative mosaic of Cape Horn archipelago’s Sub-Antarctic habitats (Rozzi et al. 2010b), which can be reached throughout a relatively short distance and altitude. Thus, this mosaic of Sub-Antarctic habitats, which is characterized by sharp changes in water temperature, as reflected by the total cumulative degree-days in Fig. 16, provides a perfect scenario for the long term monitoring of water temperature regimes and the effects of global warming on the freshwater fauna of the Sub-Antarctic ecoregion of southern South America.

Chapter References


Pisano, E. 1977. Fitogeografia de Fuego-Patagonia Chilena I. Comunidades vegetales entre las latitudes 52 y 56S. Anales del Instituto de la Patagonia 8:121-250.


Fig. 14. Ecoregion of Sub-Antarctic Magellanic forests (dark gray). Navarino Island is located north of Cape Horn and south of the Beagle Channel. The Cape Horn Biosphere Reserve and the Omora Ethnobotanical Park are located within this ecoregion (Rozzi et al. 2008c).
Fig. 15. Altitudinal gradient and sampling sites through the Róbalo river watershed. This altitudinal gradient includes the characteristic habitat types of the Cape Horn region. From the shoreline upwards these habitats are: evergreen forests dominated by *Nothofagus betuloides*; mixed evergreen-deciduous forests dominated by *N. betuloides* and *N. pumilio*; deciduous forests dominated by *N. pumilio* and *N. antarctica*; alpine tundra dominated at lower levels by cushion plants, e.g., *Bolax gummifera*; and at upper levels by lichens such as *Neuropogon* sp. Fig. modified from (Rozzi et al. 2010b). Sampling sites are represented with roman numerals I – V and range from 120 – 586 m.a.s.l.
Fig. 16. Total Cumulative Degree-Days (CDD) obtained for a period of 14 months from the headwaters to the mouth (sites II-V) of the altitudinal gradient of the Rôbalô River watershed.

Fig. 17. Scatter plot of *Gigantodax* sp. head capsule length and width (mm) indicating larval instars. Mean length and width (mm) ± SD are indicated below each instar category.
Fig. 18. Relative abundance of larval instars and total number of pupae of *Gigantodax* sp. from the Róbalo River watershed, at site V (120 m.a.s.l), from August of 2009 through August of 2010. Bar thickness represents the relative proportion of individuals in each instar. Bold, curved lines indicate proposed cohorts derived from cumulative degree-days data obtained from the field.
Fig. 19. Predicted cohort lines for Gigantodax sp. (shown by bold, curved lines) throughout the altitudinal gradient of the Róbalo River watershed, based upon data collected at site V (120 m.a.s.l) and cumulative degree days per month and total accumulated in a 12 month period. As it is shown, Gigantodax sp.’s life cycle ranges from univoltine to multivoltine from the headwaters to the mouth of the river. Each cycle takes approximately 338 degree-days to complete.
Fig. 20. Linear regression between altitude (m.a.s.l) and their annual degree- days (solid line) along the altitudinal gradient of the Róbalo river watershed. The dashed and dotted lines shows the relationship if a 2°C and 4°C warming occurs. Fig. modified from (Sweeney et al. 1992)

Table 9 Mean physical-chemical parameters values (±SE) throughout the altitudinal gradient of the Róbalo River watershed. Values are based on data obtained during austral summers of 2008, 2009, and 2010 (n=16). Values within each category with different letters were significantly different with a Tukey HSD test (α 0.05)

<table>
<thead>
<tr>
<th>Altitude (m.a.s.l)</th>
<th>Parameter</th>
<th>586</th>
<th>486</th>
<th>380</th>
<th>240</th>
<th>120</th>
<th>F</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Conductivity (μS)</td>
<td>27.4(5.1)(^A)</td>
<td>29.4(3.7)(^A)</td>
<td>31.0(5.6)(^A)</td>
<td>50.5(9.5)(^B)</td>
<td>60.9(11)(^B)</td>
<td>15.7</td>
<td>0.0003</td>
</tr>
<tr>
<td></td>
<td>Dissolved Oxygen (mg/L)</td>
<td>12.2(1.4)(^A)</td>
<td>12.1(1.6)(^A)</td>
<td>11.1(2.6)(^A)</td>
<td>10.3(1.2)(^A)</td>
<td>10.3(0.7)(^A)</td>
<td>0.83</td>
<td>0.53</td>
</tr>
<tr>
<td></td>
<td>pH</td>
<td>8.3 (0.3)(^A)</td>
<td>8.3(0.1)(^A)</td>
<td>8.3(0.2)(^A)</td>
<td>8.2(0.3)(^A)</td>
<td>8.2(0.7)(^A)</td>
<td>0.26</td>
<td>0.89</td>
</tr>
</tbody>
</table>
Table 10 Comparison of voltinism across different orders and families of aquatic insects from high and intermediate latitude sites in the northern and southern hemisphere

<table>
<thead>
<tr>
<th>Location (Country)</th>
<th>Lat. (°)</th>
<th>Temperature range (°C)(^a)</th>
<th>Degree-Days(^b)</th>
<th>Order (Family)</th>
<th>Voltinism(^c)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Southern Yukon (Canada)</td>
<td>63°N</td>
<td>-21°C – 22°C</td>
<td>No information provided</td>
<td>Diptera (Simuliidae)</td>
<td>Univoltine</td>
<td>(Currie 1997)</td>
</tr>
<tr>
<td>Eastern North America (United States)</td>
<td>31° - 50°N</td>
<td>-37°C – 15°C</td>
<td>Varies from 6500 at 31°N to 1500 at 50°N</td>
<td>Ephemeroptera (Leptophlebiidae)</td>
<td>Generally univoltine</td>
<td>(Sweeney et al. 1992)</td>
</tr>
<tr>
<td>Central Georgia (USA)</td>
<td>34°N</td>
<td>4°C – 26°C</td>
<td>No information provided</td>
<td>Coleoptera (Dytiscidae)</td>
<td>Multivoltine</td>
<td>(White and Barman 2000)</td>
</tr>
<tr>
<td>Oklahoma (USA)</td>
<td>34°N</td>
<td>3.5°C – 29°C</td>
<td>No information provided</td>
<td>Ephemeroptera (Caenidae)</td>
<td>Multivoltine</td>
<td>(Taylor and Kennedy 2006)</td>
</tr>
<tr>
<td>Chubut Province (Argentina)</td>
<td>42°S</td>
<td>1°C – 12°C</td>
<td>No information provided</td>
<td>Plecoptera (Gripopterygidae)</td>
<td>Semi and merovoltine</td>
<td>(Beltran and Miserendino 2011)</td>
</tr>
<tr>
<td>Waitaker River (New Zealand)</td>
<td>42°S</td>
<td>8.5°C – 17°C</td>
<td>No information provided</td>
<td>Ephemeroptera (Leptophlebiidae)</td>
<td>Univoltine, bivoltine</td>
<td>(Towns 1983)</td>
</tr>
<tr>
<td>South Georgia (United Kingdom)</td>
<td>54°S</td>
<td>-5°C – 10°C</td>
<td>No information provided</td>
<td>Coleoptera (Dytiscidae)</td>
<td>Semivoltine</td>
<td>(Rodney and Convey 1998)</td>
</tr>
<tr>
<td>Róbalo River Watershed, Cape Horn (Chile)</td>
<td>55°S</td>
<td>-4°C – 20°C</td>
<td>Varies from 500 at 586 m.a.s.l to 2037 at 120 m.a.s.l</td>
<td>Diptera (Simuliidae)</td>
<td>Univoltine and multivoltine</td>
<td>This study</td>
</tr>
</tbody>
</table>

\(^a\) Yearly temperature ranges including cold and warm seasons
\(^b\) Relative to 0°C and refers to degree-days accumulated in freshwater systems
\(^c\) Voltinism relative to specific location. Number of generations per year is subject to change at varying temperatures, habitats, and latitude
CHAPTER 5
LIFE HISTORY OF *Meridialaris chiloeense* (Demoulin) (EPHEMEROPTERA: LEPTOPHELEBIIIDAE) THROUGH THE ALTITUDINAL GRADIENT OF A SOUTHERN SOUTH AMERICA SUB-ANTARCTIC STREAM

Abstract

The life history of one of the world’s southernmost mayflies, *Meridialaris chiloeense* (Demoulin) (Leptophlebiidae: Ephemeroptera), was studied through the altitudinal gradient of a Sub-Antarctic fluvial system (Róbalo river, 55°S, Cape Horn Biosphere Reserve, Chile) from August 2009 to August of 2010. The results indicate that the life history of *Meridialaris chiloeense* ranges from merovoltine (headwaters, 560 meters above sea level) to univoltine (mouth, sea level) through the altitudinal gradient studied. We provide historical water temperature data for the river and propose *Meridialaris chiloeense* as a suitable indicator in the context of climate change studies, as a small rise in temperature could switch its life cycle to multivoltine, provoking serious changes on benthic macroinvertebrate community structure and geographic distributions, amongst others.

Introduction

Ephemeroptera nymphs are a conspicuous component of the macroinvertebrate fauna in mountain streams (Hollmann and Miserendino 2006). Most mayfly nymphs consume epilithic algae and fine particulate organic matter (Cummins and Klug 1979, Miserendino et al. 2001), and are an important component in the diet of many fish, amphibians, birds and mammals (Morse 2009). Furthermore, they are highly susceptible to pollution and have been extensively used as indicators water quality (Hynes 1970, Hellawell 1978, Pyle et al. 1981, Wallace 1996, Abellán et al. 2005, Morse 2009).
Mayflies are hemimetabolous insects and the length and number of life cycles per year depends largely on the geographic location and size of the species, with large burrowers in temperate climates taking over two years to mature, while tropical species may have several generations in a year (Barber-James et al. 2008). Although in the last decade, the knowledge of Ephemeroptera has gained attention in Argentina and Chile (Hollmann and Miserendino 2006), only a few studies have been conducted on their life histories in the streams and rivers of the Sub-Antarctic ecoregion of southern South America. Most aquatic insects of high latitudes and altitudes exhibit a seasonal emergence pattern, as the interaction of temperature, photoperiod, and local climate conditions helps determine aquatic insect seasonal phenology in temperate regions (Weathers et al. 2000). Generally, where climates are cold, rapid development is difficult and species with several generations per year are unusual (Danks 2007). Indeed, many Arctic/Antarctic and alpine species have life cycles that last more than one year (Danks 2007). Comparing life history patterns from high and intermediate latitudes in both, the north and south hemisphere, we can see that multivoltine life histories are most frequent at lower latitudes (i.e. 34°N), while univoltine, semivoltine, and merovoltine life histories are most frequent at higher latitudes (i.e. 50°N, 42°S). For example, the mayflies *Metamonius aniceps* (Nesameletidae) and *Meridialaris chiloeensis* (Leptophlebiidae), exhibit bivoltine and univoltine life histories, respectively, in mountainous Patagonian streams (42°S) (Hollmann and Miserendino 2006), while other members of the family Leptophlebiidae exhibit a mixture of poorly synchronized, univoltine and bivoltine life histories with overlapping generations and cohorts in New Zealand streams (Towns 1983). Furthermore aquatic insects inhabiting other Sub-Antarctic islands, as in the case of the diving beetle *Lancetes angusticollis* (Curtis) (Coleoptera: Dytiscidae), exhibit
semivoltine life cycles in South Georgia, one of the coldest Sub-Antarctic islands, with temperatures that range from -5 – 10°C during the summer (Rodney and Convey 1998).

In this context, the lack of seasonality experienced on most Sub-Antarctic islands in comparison with maritime and continental Antarctic zones could be the most significant influence on the life history of aquatic insects (Convey 1996) inhabiting these areas. In this sense, cool temperatures year-round, combined with a low or non-existent risk of extreme winter lows, could allow for activity and development to be continuous (Convey 1996). Furthermore, as a result of this limited seasonal variation, many Sub-Antarctic invertebrates show extensive overlap of generations, often with little or no seasonal structure to the life cycle, and no true diapausa in response to specific cues (Convey 1996).

The order Ephemeroptera is represented by 42 families, with a little over 3000 described species in 40 genera (Barber-James et al. 2008). According to the actual knowledge of this order in Chile, there are 57 species included in 25 genera and 7 families (Camousseight 2006). Although the region of Magellan (Chile), located within the Sub-Antarctic ecoregion in southern South America, has a relatively poor Ephemeroptera fauna (4 families, 9 genera, and 16 species (Vera-Palacios 2007)) , the family Leptophlebiidae is very diverse and widely distributed (Hollmann and Miserendino 2006). With seven described species in Chile (Camousseight 2001), the genus Meridialaris shows a broad distribution range (26° to 55°S). Meridialaris chiloense (Demoulin) is widely distributed and develops dense populations in Andean-Patagonian streams (Villanueva and Modenutti 2004, Hollmann and Miserendino 2006).

In this context, the purpose of this study was to describe the phenology of Meridialaris chiloense throughout the altitudinal gradient of a South American Sub-Antarctic fluvial system, characteristic of the of the Magellanic Sub-Antarctic ecoregion. Growing degree-days (GDD)
were obtained from water temperature data collected throughout the altitudinal gradient. We
predicted voltinism patterns through the gradient and in a historical perspective and discuss on
the effects of environmental warming on *Meridialaris chiloeense* life cycle and the possible
implications for Sub-Antarctic stream ecosystems.

Materials and Methods

Study Area

The Róbalo River (S 54° 59.647′, W 067° 40.878′, Navarino Island) is located within the
Cape Horn Biosphere Reserve (CHBR) and it provides drinking water to Puerto Williams,
Capital of the Chilean Antarctic Province, and the world’s southernmost town. It runs through
the marked altitudinal gradient of the Dientes of Navarino Mountain Range, where it drains into
the Beagle Chanel. Navarino Island lies south of the Beagle Channel and north of Cape Horn
and it has an approximate area of 2500 km² (Fig. 21). The forests of the island consist primarily
of three members of the genus *Nothofagus* (Pisano 1977) and are part of the South American
temperate forests biome, which occurs along a narrow but latitudinally extensive strip of land
between 35° and 55°S (Armesto et al. 1998). These forests are composed of mixed evergreen
*Nothofagus betuloides* (Coihüe de Magallanes), high deciduous southern beech, *N. pumilio*
(Lenga), and the deciduous *N. antarctica* (Ñirre) (McCulloch et al. 1997a). The CHBR
encompasses all of the islands south of the Beagle Channel, as well as the Chilean portion of
Tierra del Fuego Island located south of the highest peaks in the Darwin Mountain Range (Rozzi
et al. 2006). The study site lies on the north Coast of Navarino Island, which houses the city of
Puerto Williams, and the Omora Ethnobotanical Park, part of Chile’s network of Long Term
Socio-Ecological Research Sites (LTSER). Furthermore, it is the Southern hemisphere’s
latitudinal equivalent of northern British Columbia, Canada and Southern Alaska (USA).

The river is characteristic of the streams of the island (clear waters and partially forested) and it has an approximate length of approximately 12 km, ascending from 0 meters above sea level (m.a.s.l) to 600 m.a.s.l. We selected 5 sampling sites ranging from locations near its mouth at 120 m.a.s.l to its headwaters at 587 m.a.s.l. The sites were selected based on the marked vegetation patterns observed through the gradient (Fig. 22). It is important to note that this altitudinal gradient is highly representative of Sub-Antarctic ecosystems, as it exhibits all of the habitat types that can be observed within this ecoregion in a short walking distance and relatively low altitudes. We sampled benthic macroinvertebrates at each of these sites during the austral summers of 2008, 2009, and 2010, and selected one permanent site, at 120 m.a.s.l, which was sampled continuously for a period of 12 months (details explained below), starting in August of 2009.

Climate

The regional climate of the area is strongly affected by the westerly storm tracks coupled with precipitation induced by the high western flanks of the Andean Cordillera (McCulloch et al. 1997b). It also depends on polar weather fronts from the south Pacific (Rozzi et al. 2006). The city of Puerto Williams is found within the influence of the *Dfk‘c Transandeanc with steppe degeneration* climatic zone. It represents a transition between rain climates of the western coast of Magallanes and the steppe climates of the eastern side of the mountain range (Rozzi et al. 2006). The uniformity of the seasonal variation of precipitation as well as its thermal characteristics, allow for the growth of deciduous forests. The average annual temperature and precipitation recorded for Puerto Williams is 6.0°C and 467.3 mm respectively. Precipitation is
less polluted than for any other of the world’s temperate forests (Likens 1991) and the warmest month has an average temperature of 9.6°C (January-March), while the coldest month averages 1.9°C (May-August). Compared to similar northern latitudes (such as Sub-Arctic and Arctic environments of northern North America), where the climate is continental in nature and it is characterized by harsh and extreme environments (temperature ranges from -14°C to 5°C (Strathdee and Bale 1998), the Sub-Antarctic tip of South America is characterized by cool but not extreme climate year-round, with relatively low seasonal variation in temperatures due to oceanic damping (Convey 1996).

Field and Laboratory Methods

*Physical – Chemical Parameters*

Water chemistry parameters were recorded at each site and during each sampling event. Dissolved oxygen (mg/L) and conductivity (μS) were recorded using a YSI 8.5 meter. From January of 2009 to January of 2011, water temperature (°C) was recorded every 5 hours at each sampling site throughout the altitudinal gradient. Recordings were obtained using Hobo data loggers (Model HOBO® U22 Water Temp Pro v2). Differences between sites were tested using one-way ANOVAs. When significant results were obtained, a Tukey HSD *post hoc* test was applied (R Software version 2.9.2). All data was tested for normality (Shapiro-Wilk w statistic) and inequality of variances (Hartley’s $F_{max}$ ratio test) before proceeding with ANOVA ($\alpha \ 0.05$).

*Benthic Macroinvertebrate Sampling*

We sampled benthic macroinvertebrates at each of the sites during the austral summers of 2008, 2009, and 2010, and selected one permanent site (at 120 m.a.s.l), which was sampled
continuously for a period of 12 months starting in August of 2009. For the purposes of this study, we present the results obtained from the sampling carried out at 120 m.a.s.l and predict on the voltinism patterns based on water temperature (°C) data obtained for the remaining sites.

In order to study the phenology of *Meridialaris chiloeense*, larval insects were collected from site V (120 m.a.s.l) every 2 weeks. Three samples were taken from riffle habitats using a Surber net (0.09m²) with a 243μm mesh. Each sample was stored in 70% ethanol and transported to the laboratory for processing.

* Determination of *Meridialaris chiloeense* and Voltinism

The developmental stage of mesothoracic wing pads was used to help elucidate the life history of *Meridialaris chiloeense*, and distinguishing characteristics for each developmental stage were described (Taylor 2001). Wing pad stages are shown in Fig. 23. An Olympus SHZ dissecting microscope coupled with a Tucsen TCA 5.0C (5mp) camera with TSview digital imagining processing software were used in the process of characterizing the developmental stages of *Meridialaris chiloeense*.

The relative abundance of each nymph developmental stage was used to estimate cohort lines and voltinism. Genus and species identifications were carried out using the best available literature (Pescador and Peters 1986b, Domínguez and Fernández 2009). To estimate the starting point of a generation, the time period at which a proportionally higher relative abundance of first developmental stage nymphs was recorded. Approximate emergence of adults was estimated by noting the time period at which we obtained proportionally higher number of fifth developmental stage nymphs.
Determination of Cumulative Degree Days and Growing Degree Days

Average daily water temperature (°C) was calculated for each site using the information retrieved from the Hobo® data loggers. Temperature information from site I (586 m.a.s.l) was not retrieved, therefore temperature (°C) data for sites II-V (486-120 m.a.s.l) is presented. Cumulative Degree-Days (CDD) for each site were computed by summing the daily average temperature (°C) by month and for a period of 1 year, following (Snyder et al. 2001). Growing Degree-Days (GDD) were estimated by summing the daily average temperature (°C) starting at the period at which we observed a proportionally higher number of first developmental stage nymphs and finishing at the period at which we observed a proportionally higher number of fifth developmental stage nymphs.

Estimation of Voltinism through the Altitudinal Gradient

Cohort lines and voltinism were estimated through the altitudinal gradient by obtaining the CDD for each site by month. Using the results of the approximate GDD obtained from site V (120 m.a.s.l), the number of generations (cohorts) that could be possible for each site related to its total number of CDD during a one-year period were estimated.

Estimation of Historical Water Temperature (°C) and Voltinism

As explained before, Cumulative Degree-Days (CDD) are computed by summing the daily average temperature (°C) during a determined time period (Snyder et al. 2001). Historical CDD (2006-2011) for site V were obtained, by obtaining water and air temperature data (°C) from a permanent meteorological site (managed by Chile’s Dirección General de Aguas (DGA)) located at this altitude. Although water and air temperature data are readily available for general access through DGA’s website, daily water temperature data for large periods of times is
sometimes missing and not always consistent. For this reason, water temperature data (°C) was estimated from March of 2006 until March of 2009, by determining the relationship between water to air temperature (°C) through simple linear regressions, following Crisp and Howson (1982). As explained before, temperature data for the remaining period was obtained from Hobo® data loggers deployed at site V (120 m.a.s.l). After obtaining estimated mean daily water temperature (°C) data, CDD per month and year were calculated, and possible emergence and hatching dates of *Meridialaris chiloeense* at 120 m.a.s.l from 2006 to 2011 were estimated.

Results

Physical – Chemical Parameters

Dissolved oxygen (mg/L) and pH were not significantly different between sites throughout the altitudinal gradient (one-way ANOVA, $P = 0.53$ and $0.89$, $\alpha 0.05$), while conductivity (µS) was significantly higher at sites IV and V (240 and 120 m.a.s.l, one-way ANOVA with Tukey HSD analysis, $P = 0.0003$, $\alpha 0.05$) (Table 1).

Maximum water temperature (°C) was reached during austral summers (December – March) and ranged between 10-17°C from the headwaters to the mouth of the Róbalo River watershed, while minimum temperatures fell below 0°C during austral winters (June-August). Total cumulative degree-days (CDD) changed dramatically every 100 meters of altitude through the altitudinal gradient, and ranged from 501 to 2037 CDD, from the headwaters to the mouth (Fig. 24).

Determination of *Meridialaris chiloeense* Nymph Developmental Classes and Voltinism

- Nymph developmental classes: Five distinct nymph developmental classes were
obtained for *Meridialaris chiloeense* by describing the developmental stage of the mesothoracic wing pads (Taylor and Kennedy 2006) (Fig. 23). Table 12 provides a detailed description of each developmental class.

- **Voltinism:** A proportionally higher abundance of *Meridialaris chiloeense* 1\textsuperscript{st} developmental stage nymphs were observed during April (Fig. 25), while a proportionally higher abundance of 5\textsuperscript{th} developmental stage nymphs were observed from January to March. By counting the amount of degree-days that it takes for one generation of *Meridialaris chiloeense* to reach the highest abundance of 5\textsuperscript{th} developmental stage nymphs, we can estimate that it takes approximately 1185 degree-days for one generation of *Meridialaris chiloeense* to complete (Fig. 25).

**Estimation of Voltinism through the Altitudinal Gradient**

Cumulative Degree-Days (CDD) range from 501-2037 from headwaters to the mouth of the Róbalo River watershed. Thus, it is estimated that *Meridialaris chiloeense*’s life history varies from merovoltine, with a generation taking approximately 25 months to complete at the river’s headwaters, to univoltine (1 generation per year) at its mouth (Fig. 26).

**Estimation of Historical Water Temperature (°C) and Voltinism**

The regression between mean air temperature (x, °C) and mean water temperatures (y, °C) obtained from DGA’s weather site V (120 m.a.s.l) from 2006-2010 (Fig. 27), suggests that the relationship between the two is approximately linear over most of the observed temperature range (r$^2$=0.89, y = -0.36 + 0.99x). With this information, mean water temperature data (°C) for dates with missing information from DGA’s weather site were estimated, along with the CCD
from 2006-2011 at site V (120 m.a.s.l) in the Róbalo river watershed. Furthermore, historical hatching and emergence dates for *Meridialaris chiloeense* at site V (120 m.a.s.l) from 2006 to 2011 were estimated. As noted in Fig. 28, CDD vary in approximately 160 CDD between 2006 and 2009, while there is a variation of 208 CDD with 2010. This variation has a direct effect on the emergence dates of *Meridialaris chiloeense*: in years with a higher accumulation of degree-days (2006, 2008, 2010), emergence begins in early January, while during years with a lower accumulation of degree-days (2007, 2009), emergence begins in late January and early February.

**Discussion**

*Meridialaris chiloeense* Life Cycle in South American Sub-Antarctic Streams

Although life history studies have been carried out for many years in the Northern Hemisphere and other countries of the Southern Hemisphere (i.e. New Zealand), little is known about the life histories of aquatic insects in Patagonia (Beltran and Miserendino 2011) and the Sub-Antarctic ecoregion of southern Chile. This is study is one of the few report on the life history of any freshwater insect inhabiting Sub-Antarctic South American streams. Our results indicate that *Meridialaris chiloeense* (Ephemeroptera: Leptophlebiidae) has a life history, which varies from merovoltine to univoltine, from the headwaters to the mouth of the sharp altitudinal gradient of the Róbalo River Watershed. These results are similar to other studies carried out in Northern Patagonian streams (Hollman et al. 2006) but contrasting in the fact that *Meridialaris chiloeense* presents a bivoltine life cycle at 42°S. Most aquatic insects of high latitudes and altitudes exhibit a seasonal emergence pattern, as the interaction of temperature, photoperiod, and local climate conditions helps to determine aquatic insect seasonal phenology in temperate regions (Weathers et al. 2000). Generally, where climates are cold, rapid development is
difficult and species with several generations per year are unusual (Danks 2007). Indeed, many Arctic/Antarctic and alpine species have life cycles that last more than one year (Danks 2007). Comparing life history patterns from high and intermediate latitudes in both, the north and south hemisphere, we can see that multivoltine life histories are most frequent at lower latitudes (i.e. 34°N), while univoltine, semivoltine, and merovoltine life histories are most frequent at higher latitudes (i.e. 50°N, 42°S). For example, univoltine and semivoltine life cycles have been described for aquatic insects inhabiting other Sub-Antarctic islands, as in the case of the diving beetle *Lancetes angusticollis* (Curtis) (Coleoptera: Dytiscidae), which exhibits a semivoltine life cycle in South Georgia, one of the coldest Sub-Antarctic islands, with temperatures that range from -5 – 10°C during the summer (Rodney and Convey 1998). Furthermore, the stoneflies *Notoperla fasciata* and *N. magnaspina* (Plecoptera: Gripopterygidae), exhibit semivoltine and merovoltine life cycles in Patagonian streams, where temperatures range from 1°C – 12°C (Beltran and Miserendino 2011). The type of life cycle exhibited by *Meridialaris chiloeense* in this study, could be explained through the lack of seasonality experienced on most Sub-Antarctic islands in comparison with maritime and continental Antarctic zones. This may be the most significant influence on the life history of aquatic insects (Convey 1996) inhabiting these areas, where the summers are cool and the winters are not specially cold (Danks 2007) due to oceanic damping. In this sense, cool temperatures year-round, combined with a low or non-existent risk of extreme winter lows, could allow for activity and development to be continuous (Convey 1996). Furthermore, as a result of this limited seasonal variation, many Sub-Antarctic invertebrates show extensive overlap of generations, often with little or no seasonal structure to the life cycle, and no true diapauses in response to specific cues (Convey 1996).
Meridialaris chiloeense as an Indicator of Climate Change in South American Sub-Antarctic Streams

Although climate change and the increase in the predicted frequency of extreme climatic events have the capacity to change aquatic community structure as well as the geographical distribution and the development and timing of life histories of individual species (Ryan and Ryan 2006, Winterbourn et al. 2008), studies on the potential effects of climate change on the freshwater fauna the Sub-Antarctic islands of Cape Horn have not been carried out. Several studies have shown that aquatic ecosystems are as vulnerable to global change as terrestrial and marine ecosystems (Heino et al. 2009), although the evidence is unbalanced across ecosystems and information from streams is scarce (Durance and Ormerod 2007). Lentic (i.e. lakes and ponds) and lotic (i.e. streams and rivers) ecosystems are considered to be most sensitive to land use change, exotic species, and climate change at a global scale, with aquatic ecosystems at high latitudes being more strongly threatened by climate change (Heino et al. 2009). The temperatures of streams and rivers vary more rapidly than those of lakes, and apart from spring-fed streams, water temperatures in running waters are strongly influenced by changes in air temperature and are usually higher or lower than air temperatures when the latter are low or high respectively (Elliott 1991). Therefore, temperature changes as a consequence of climate change would be expected to be more marked in streams and rivers than in other freshwater habitats (Elliott 1991). This has a direct impact on the phenology of river insects, as they have evolved under the constraints of a thermally fluctuating system and show little or no ability to compensate or acclimate to changing environmental temperatures (Sweeney et al. 1992).

In this context, we hypothesize on the effects of global warming on the life history of Meridialaris chiloeense and propose it as an indicator in the context of climate change studies, as a 2°C rise in temperature, as seen in other Sub-Antarctic islands, such as New Zealand and South
Georgia (see (Rodney and Convey 1998, Winterbourn et al. 2008), would cause a switch on the annual degree-days accumulated along the sharp altitudinal gradients observed through the course of many of southern South American Sub-Antarctic streams. This switch on the DD accumulated through the altitudinal gradient studied, would have great impacts on the life history *Meridialaris chiloeense*. As we concluded earlier, the total degree-days needed for *Meridialaris chiloeense* to complete its life cycle are approximately 1185 DD and the life history varies radically from merovoltine (at the headwaters of the Róbalo river watershed) to univoltine, at locations close to the mouth the of the river. With an increase in the annual accumulated DD, the life cycle of *Gigantodax chiloeense* would switch to completely univoltine throughout the altitudinal gradient. This could have great ecological implications, not only for *Meridialaris chiloeense*, but also for many other species inhabiting these Sub-Antarctic streams. In this context, we could expect changes in the trophic structure of benthic macroinvertebrate communities, geographic distribution switches, or even local extinction of various species of aquatic insects, as the cold-stenothermal species inhabiting these streams may not be able to meet their low temperature thresholds for proper egg/larval growth and development to occur (Winterbourn et al. 2008).

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Fig. 21. Ecoregion of Sub-Antarctic Magellanic forests (dark gray). Navarino Island is located north of Cape Horn and south of the Beagle Channel. The Cape Horn Biosphere Reserve and the Omora Ethnobotanical Park are located within this ecoregion (Rozzi et al. 2008).
Fig. 22. Altitudinal gradient and sampling sites through the Róbalo river watershed. This altitudinal gradient includes the characteristic habitat types of the Cape Horn region. From the shoreline upwards these habitats are: evergreen forests dominated by *Nothofagus betuloides*; mixed evergreen-deciduous forests dominated by *N. betuloides* and *N. pumilio*; deciduous forests dominated by *N. pumilio* and *N. antarctica*; alpine tundra dominated at lower levels by cushion plants, e.g., *Bolax gummifera*; and at upper levels by lichens such as *Neuropogon* sp. Fig. modified from (Rozzi et al. 2010). Sampling sites are represented with roman numerals I – V and range from 120 – 586 m.a.s.l.
Fig. 23. Mesothoracic wing pad developmental stages for *Meridialaris chiloeense*: 1) absence of wing pads and well-defined compound eyes; 2) absence of wing pads; 3) clear wing pads on the first thoracic segment; 4) wing pads with veins, expanding from the first thoracic segment to the mid portion of the second thoracic segment; 5) darkened wing pads, expanding to the mid portion of the third thoracic segment.
Fig. 24. Total Cumulative Degree-Days (CDD) obtained for a period of 14 months from the headwaters to the mouth (sites II-V) of the altitudinal gradient of the Rôbalo River watershed.
Fig. 25. Relative abundance of *Meridialaris chiloeense* nymph developmental classes from the Róbalo River watershed, at site V (120 m.a.s.l), from August of 2009 through August of 2010. Bar thickness represents the relative proportion of individuals in each developmental class. Bold, curved lines indicate proposed cohorts derived from cumulative degree-days data obtained from the field.
Fig. 26. Predicted hatching and emergence dates for *Meridialaris chiloeense* (shown by bold, curved lines) throughout the altitudinal gradient of the Róbalo river, based upon data collected at 120 m.a.s.l and cumulative degree days per month and total accumulated in a 12 month period. As it is shown, *Meridialaris chiloeense*’s life cycle ranges from merovoltine to univoltine from the headwaters to the mouth of the river. Each cycle takes approximately 1185 degree-days to complete.
Fig. 27. Plot of mean monthly water temperature ($y, ^\circ C$) against mean monthly air temperature ($x, ^\circ C$) for the Róbalo River at 120 m.a.s.l, from 2006-2009.
Fig. 28. Historical Cumulative Degree Days (CDD) from 2006-2011. The Fig. shows estimated historical hatching and emergence dates for *Meridialis chiloense* at site V (120 m.a.s.l) from 2006 to 2011. As noted in the Fig., CDD vary in approximately 160 CDD between 2006 and 2009, while there is a variation of 208 CDD with 2010. This variation has a direct effect on the emergence dates of *Meridialis chiloense*: in years with a higher accumulation of degree-days (2006, 2008, 2010), emergence begins in early January, while during years with a lower accumulation of degree-days (2007, 2009), emergence begins in late January and early February.
Table 11 Mean physical-chemical parameters values (±SE) throughout the altitudinal gradient of the Róbalo River watershed. Values are based on data obtained during austral summers of 2008, 2009, and 2010 (n=16). Values within each category with different letters were significantly different with a Tukey HSD test (α 0.05)

<table>
<thead>
<tr>
<th>Altitude (m.a.s.l)</th>
<th>Parameter</th>
<th>586</th>
<th>486</th>
<th>380</th>
<th>240</th>
<th>120</th>
<th>F</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Conductivity (μS)</td>
<td>27.4(5.1)^A</td>
<td>29.4(3.7)^A</td>
<td>31.0(5.6)^A</td>
<td>50.5(9.5)^B</td>
<td>60.9(11)^B</td>
<td>15.7</td>
<td>0.0003</td>
</tr>
<tr>
<td></td>
<td>Dissolved Oxygen (mg/L)</td>
<td>12.2(1.4)^A</td>
<td>12.1(1.6)^A</td>
<td>11.1(2.6)^A</td>
<td>10.3(1.2)^A</td>
<td>10.3(0.7)^A</td>
<td>0.83</td>
<td>0.53</td>
</tr>
<tr>
<td></td>
<td>pH</td>
<td>8.3 (0.3)^A</td>
<td>8.3(0.1)^A</td>
<td>8.3(0.2)^A</td>
<td>8.2(0.3)^A</td>
<td>8.2(0.7)^A</td>
<td>0.26</td>
<td>0.89</td>
</tr>
</tbody>
</table>

Table 12 Description of *Meridialaris chiloense* nymphal developmental classes

<table>
<thead>
<tr>
<th>Developmental Class</th>
<th>Description</th>
</tr>
</thead>
</table>
| I                   | a) Unpigmented, lack gills, and lightly sclerotized.  
b) Head lacks setae and well-developed compound eyes. Presence of 3 ocelli, one medial and two lateral to the margins of the head.  
c) Absence of wing pads. |
| II                  | a) Pigmented, presence of slender, feather-like gills  
b) Presence of setae and well-developed compound eyes on the lateral margin of the head.  
c) Absence of wing pads. |
| III                 | a) Pigmented, presence of feather-like gills.  
b) Presence of setae and well-developed compound eyes.  
c) Presence of clear wing pads on the first thoracic segment. |
| IV                  | a) Pigmented, presence of feather-like gills.  
b) Presence of setae and well-developed compound eyes.  
c) Presence of wing pads with veins which expand from the first thoracic segment to the mid portion of the second thoracic segment |
| V                   | a) Pigmented, presence of feather-like gills.  
b) Presence of setae and well-developed compound eyes.  
c) Presence of darkened wing pads, which expand to the mid portion of the fourth thoracic segment. |
CHAPTER 6

APPLYING FIELD ENVIRONMENTAL PHILOSOPHY AT THE OMORA ETHNOBOTANICAL PARK: AQUATIC INVERTEBRATES OF THE SOUTHERNMOST WATERSHEDS

Abstract

The Cape Horn Biosphere Reserve, embedded within the remote Sub-Antarctic ecoregion is a reservoir of expressions of biological and cultural diversity and a frontier on the verge of globalization. Although it is considered one of 24 wilderness areas remaining in the world, it is not free from local and global threats, such as salmon farming, invasive species, and climate change. In this context, field biologists, philosophers, artists, and local actors associated to the Sub-Antarctic Biocultural Conservation Program and the Omora Ethnobotanical Park (OEP), have worked together to describe the region’s biological and cultural diversity, linking ecological and philosophical research into education, ecotourism, and biocultural conservation, through a methodology called Field environmental philosophy. This methodology integrates ecological sciences and environmental ethics through a 4-step cycle consisting of: 1) interdisciplinary ecological and philosophical research; 2) composition of metaphors and communication of simple narratives; 3) design of field activities with an ecological and ethical orientation; and 4) implementation of in situ conservation areas.

We applied this four step cycle to the study of the diversity and life histories of the freshwater aquatic invertebrates associated to the Róbalo River, which provides drinking water to Puerto Williams, capital of the Chilean Antarctic Province and the world’s southernmost town. We focused on the question: How to better understand and communicate the intrinsic and instrumental values of freshwater invertebrates of the Róbalo and the Cape Horn Biosphere Reserve?
Introduction

The Sub-Antarctic ecoregion of southern South America has been identified as “hotspot” for bryophyte diversity (Rozzi et al. 2008c). It also harbors a mosaic of unique cultural and biological diversity (Rozzi et al. in press), and it is considered one of 24 wilderness areas remaining in the world (Mittermeier et al. 2003). However, this remote region is not free from local, regional, and global threats, such as salmon farming, invasive exotic species, massive tourism, and global climate change. In this context, the Cape Horn Biosphere Reserve (CHBR), which is embedded within the Sub-Antarctic ecoregion of southern Chile, emerges as one of the last “wild” places on earth, in an increasingly urbanized world (Rozzi et al. 2010e). The CHBR has an extension of 4,884,274 ha of marine and terrestrial ecosystems (Rozzi et al. 2007) and it is the home of the Omora Ethnobotanical Park (OEP, located in Navarino Island, 55ºS), one of Chile’s first Long Term Socio-Ecological Research Sites (LTSER).

To address these issues, ecologists, philosophers, and artists have worked together at OEP to describe and better understand the biocultural diversity of the region through an innovative methodology called Field environmental philosophy (FEP), which integrates ecological sciences and environmental ethics into education, ecotourism, and biocultural conservation through an interconnected and interrelated four-step cycle (Rozzi et al. 2010b). The FEP methodology has been incorporated in the formal curriculum of the University of Magallanes (UMAG, Punta Arenas, Chile) Master’s program in Sub-Antarctic Conservation, in an experience, which allows graduate students to develop ecological and philosophical research and integrate it into innovative education, ecotourism, and biocultural conservation activities (Rozzi et al. 2010b). Furthermore, UMAG’s graduate program in Conservation Sciences
(Masters of Science in Sub-Antarctic Conservation), has established a closed collaboration with the University of North Texas (UNT) and the Institute of Ecology and Biodiversity (IEB, Chile).

Field Environmental Philosophy’s Four-Step Cycle

Omora Park’s FEP emphasizes ecologically and philosophically guided field experiences in local habitats, socio-cultural communities, and regional institutions, designed to stimulate the perception of and valuation toward biological and cultural diversity in specific places and moments (Rozzi et al. in press). In this way FEP integrates environmental ethics and ecological research into innovative biocultural education and conservation activities through and interrelated four-step cycle:

Step 1 - Interdisciplinary research: Students conduct ecological, environmental philosophy, and ethnoecological research about Sub-Antarctic biodiversity and at the same time learn, and aim to better understand the diversity of perceptions, names, ecological practices and cosmologies of the region.

Step 2 - Composition of metaphors and communication through simple narratives: students compose metaphors with 2 main objectives: a) generate tools that allows for communication with the general public, and b) integrate ecological and philosophical discoveries, through analogical thinking which allows them to conceptually synthesize facts and values into education, ecotourism, and biocultural conservation activities.

Step 3 - Design field activities with an ecological and ethical orientation: the education program of Omora Ethnobotanical Park emphasizes “face-to-face” encounters with the inhabitants of the ecosystems in which we live, as well as with their unique habitats and habits.
The development and implementation of these activities stimulate the valuation and better understanding of the regional biocultural diversity.

Step 4 - Implementation of physical spaces for in situ conservation: the implementation of physical spaces for in situ conservation is important for at least three reasons: a) contributes to the preservation of regional biodiversity; b) protects the habitats and ecological interactions within ecological communities, and c) allows for the in situ observation and better understanding of ecological interactions, therefore, foresting a sense of responsibility and respect towards biocultural diversity.

This methodology was successfully implemented by the Omora Ethnobotanical Park’s research team, which determined that the Cape Horn Biosphere Reserve represents a “hotspot” of bryophyte diversity, hosting more than 5% of the world’s species of mosses and liverworts, on less than 0.001% of the earth’s surface (Rozzi et al. 2008c). Following the FEP methodology, researchers also investigated the aesthetic value of bryophytes (Rozzi et al. 2006) and composed the metaphor and interpretative trail “Miniature Forests of Cape Horn.” They also created an ecologically and ethically guided educational and ecotourism activity called “Ecotourism with a Hand Lens” and implemented an interpretative trail and conservation area for the Miniature Forests of Cape Horn at Omora Ethnobotanical Park (Rozzi et al. 2008a, Rozzi et al. 2010e).

Field Environmental Philosophy and Freshwater Invertebrates in the Cape Horn Biosphere Reserve

In this work, we studied the diversity and life histories of freshwater invertebrates of the Róbalo river watershed, which provides drinking water to Puerto Williams, the Capital of the Chilean Antarctic Province and the world’s southernmost town. We also studied the ecological and ethical values of river ecosystems as stated in works by Aldo Leopold, and derived from the
Yahgan traditional ecological knowledge. We then followed Omora’s Field environmental philosophy to better understand their intrinsic and instrumental values and contribute to their conservation within the Cape Horn Biosphere Reserve (CHBR). The CHBR, located within the Sub-Antarctic ecoregion (Fig. 29) in southwestern South America (55°S) (Fig. 30), contains the world’s cleanest freshwater sources, as it does not receive industrial pollutants and the precipitation chemistry in the region reveals one of the lowest concentrations of nitrate ever recorded (Likens 1991b, Weathers et al. 2000). Historically, most of the research in the region has focused in easily perceived groups such as vertebrate species and vascular plants, omitting other types of organisms such as non-vascular plants and freshwater invertebrates, which are usually under perceived and are challenging to study (Rozzi et al. 2008).

This study investigates for the first time, with a FEP methodology, the freshwater invertebrates of the CHBR. Freshwater invertebrates play essential roles in food webs (Morse 2009), and have been widely used as water quality indicators (Wallace 1996). Despite the ecological importance of invertebrates in freshwater ecosystems, they have not been the targets of research within the Sub-Antarctic ecoregion, although various authors have established that they present a high degree of endemism and speciation (Miserendino and Pizzolon 2000) and have been fundamental in the studies about the effects of the introduced North American beaver in the CHBR (Anderson et al. 2006).

To confront the previously described challenges, we translated the results of our ecological research into conservation, education, and ecotourism activities to communicate and better understand the intrinsic and instrumental values of freshwater invertebrates in the Cape Horn Biosphere Reserve by using the FEP methodology and adapting our research to its four-step cycle (Fig. 31):
• Step 1 - Interdisciplinary research: Benthic Macroinvertebrates of the Cape Horn Biosphere Reserve

The Róbalo River (S 54° 59.647’, W 067° 40.878’, Navarino Island) runs through the marked altitudinal gradient of the Dientes of Navarino Mountain Range, where it drains into the Beagle Channel. The river is characteristic of the streams of the island (clear waters and partially forested) and it has an approximate length of 12 km, ascending from 0 to 600 meters above sea level (m.a.s.l). It is embedded within a diverse and sharp gradient of habitats characteristic of the Cape Horn region: evergreen, mixed, and deciduous forests dominate low altitudes (from the coast to approximately 450 m.a.s.l), while lichens such as *Neuropogon* sp. and cushion plants, such as *Bolax gummifera*, dominate the high Andean zone. Since 2003, scientists at the Omora Ethnobotanical Park (OEP) have been studying freshwater invertebrates in the Róbalo river in order to answer questions related biogeographic affinities, community structure, and the potential effects of exotic species such as the North American beaver *Castor canadensis* (Anderson et al. 2006, Moorman et al. 2006).

In the context of ecological research, we contributed to freshwater invertebrate research in the CHBR by studying their diversity, distribution, and life histories within the altitudinal gradient of the Róbalo river watershed during the austral summers of 2008, 2009 and 2010. For the first time, we provide an altitudinal gradient profile of the communities of benthic macroinvertebrates along the Róbalo River. We sampled freshwater invertebrates at 5 different altitudes, identified them, recorded their abundances, and described the physical-chemical characteristics of their habitat at each altitude. We determined that certain families of Dipterans, such as the Chironomidae (non-biting midges) are more abundant and diverse at higher altitudes, while others, such as the Blephlariceridae (net-winged midges), are only found at lower altitudes. Furthermore, we found that the distribution and abundance of stoneflies belonging to the family
Notoneouridae and mayflies of the family Nesemeletidae, vary significantly with altitude, temperature, and type of Substrate along the river. Moreover, other taxa such as *Gigantodax* (Diptera: Simuliidae), *Rheochorema* sp. (Trichoptera: Hydrobiosidae), and *Tanypodinae* (Diptera: Chironomidae) are found throughout the whole altitudinal gradient.

Additionally, we found that the life histories of certain aquatic insects, such as the dipteran *Gigantodax* sp., range from one generation per year (univoltine) to multiple generations per year (multivoltine) through the altitudinal gradient studied. This discovery is relevant at local, regional, and global scales for at least 3 reasons: 1) for the first time, we provide a comprehensive altitudinal zoning and life history description for any aquatic insect in the Sub-Antarctic ecoregion of southern South America, 2) we identified aquatic insects, such as *Gigantodax* sp., as potential indicators of climate change in the Sub-Antarctic ecoregion. As we hypothesized, a small increase in temperature could switch the life history of *Gigantodax* sp. to primarily multivoltine, provoking serious changes on benthic macroinvertebrate community structure and geographic distributions, amongst others, and 3) our results directly contrast with other studies carried out in the Southern and Northern Hemisphere at similar latitudes, as most studies suggest that aquatic insects of high latitudes exhibit univoltine life cycles.

Regarding environmental philosophy research, we focused on the work of Aldo Leopold. In *The Sand County Almanac*, Leopold dedicates an essay to the Wisconsin River, in which he integrates ecological, aesthetic, and ethical values of the river ecosystem. In *The Green Pasture* (1949), he integrates both terrestrial and aquatic component of the river and its valley:

Some paintings become famous because, being durable, they are viewed by successive generations, in each of which are likely to be found a few appreciative eyes.

I know a painting so evanescent that it is seldom viewed at all, except by some wandering deer. It is a river who wields the brush, and it is the same river who, before I can bring my friends to view his work, erases it forever from human view. After that it exists only in my mind's eye.
Like other artists, my river is temperamental; there is no predicting when the mood to paint will come upon him, or how long it will last. But in midsommer, when the great white fleets cruise the sky for day after flawless day, it is worth strolling down to the sandbars just to see whether he has been at work.

The work begins with a broad ribbon of silt brushed thinly on the sand of a receding shore. As this dries slowly in the sun, goldfinches bathe in its pools, and deer, herons, killdeers, raccoons, and turtles cover it with a lacework of tracks. There is not telling, at this stage, whether anything further will happen. (page 51, Leopold 1949a)

In this passage, Leopold emphasizes the dynamic and seasonal character of the Wisconsin River (Wisconsin, United States), an attribute that is even more marked within the sharp altitudinal gradient of the Róbalo River. In this excerpt from *The Green Pasture*, Leopold also offers a holistic view of the river ecosystem, its life, its communities and inhabitants. These components are seldom viewed in formal education and tourism, but if we stop for just a few minutes, we will discover that the river is not just water, but instead it is alive, and its life depends on all of the members that live within and outside of its waters. By integrating these concepts into formal education and tourism, school children, tourists, and other visitors can undergo an ethical transformation that involves the understanding of key ecological concepts.

Regarding the ethnoecological research, the focus is in the Yahgan traditional ecological knowledge, contained in the narrative of Omora. The Yahgan people are the world’s southernmost ethnic group (McEwan et al. 1997). Nomadic hunters, fishers, and gatherers, the Yahgans canoed the channels of sub-Antarctic archipelago region south of Tierra del Fuego, leaving behind a remarkable Amerindian archeological and cultural legacy (Guisindeo 1961, Rozzi et al. 2006). Omora Ethnobotanical Park takes its name from the Yahgan story of the hummingbird *Omora (Sephanoides sephanoides)* who helped create the rivers and streams of the Cape Horn Region. Hummingbirds are admired by many Amerindian cultures, specially the Yahgans, in which *Omora* is an occasional visitor considered to be a man, and at the same time,
a small man or spirit who maintains social and ecological order, as demonstrated in the following story:

In ancestral times, when birds were still humans, a great drought occurred in the region of Cape Horn and its inhabitants were dying of thirst. The cunning fox known as Ciláwaia found a lake, and without telling anyone, he built a solid fence around it so that no one could enter. Hidden as such, he drank lots of water, concerned only with himself. After some time, the other people discovered this lake and asked the fox for a little bit of water. Ciláwaia did not listen to them and expelled them from the lake. The condition of the people got worse and when they were about to dye, they contacted Omora.

Although this little hummingbird was an occasional visitor, he was always willing to help and very soon arrived. When he arrived, the people told him about their sufferings and Omora rapidly took the flight towards the fox to confront him. Omora asked Ciláwaia to share the water with the other people and Ciláwaia refused. Omora became furious and without replaying to the fox, returned to the settlement. He reflected and rapidly rose again, took his sling and flew back to the fox. After asking the fox again to share the water with a negative answer, Omora furiously fired a sharp stone with his sling, killing the fox with the first throw.

The other people had been watching and happily arrived to the place and started to drink the water, satiating their thirst. They drank so quickly that the lake became empty and the birds that arrived late found a few drops with which they moistened their throats. Then, the wise owl Sirra, Omora’s grandmother told the birds to collect mud from the bottom of the lake and fly to the peaks of the mountains to fling the mud over them. The birds listened to Sirra and their balls of mud gave birth to springs that became watercourses that spouted from the mountains, forming small streams and large rivers that lowed through the ravines. Today all of those watercourses still flow down from the mountains and provide exquisite water. Since that time, no one has died of thirst. (extracted and edited from pages 168-169, Rozzi et al 2010c)

Omora’s story emphasizes the importance of biological diversity for the long-term sustainability of the ecosystem services provided by the river, as a source of drinkable water for humans and other non-human animals (Rozzi et al. 2006b, Rozzi et al. 2010c). Additionally, this story concur with contemporary scientific understanding about the need for conservation of bird communities in the Cape Horn region, as birds contribute to ecological processes that are essential for the maintenance and conservation of hydrological processes (Rozzi et al. 2006b). Furthermore, from the perspective of contemporary environmental ethics, the ethical imperative implicit in the Yahgan narrative is consistent with the notion of instrumental value (Rozzi 2011).
By helping the birds and other animals obtain water, Omora is also conserving the source of drinkable water for humans, as they maintain the integrity of the vegetation and watershed habitats in order to sustain long-term flows of fresh drinkable water in Cape Horn (Rozzi et al. 2010d). In this way, by protecting the birds, Omora also protects streams and rivers that flow through the mountains of Cape Horn, and at the same time, the Yahgan view concurs with our scientific findings through the understanding of the river ecosystem as a biotic community, which entails interactions among terrestrial and freshwater biota. At the same time, this view also relates to Leopold’s perspective of highlighting the dynamic character of the river ecosystem. In this context, our study adds to Leopold and the Yahgan perspective, by explicitly emphasizing the presence of the freshwater water fauna as part of the river’s community of life.

- Step 2. Composition of metaphors and communication through simple narratives: “The River as a Community of Life” and “Trichoptera: The Underwater Constructors”

The Greek root of the word metaphor means “to transfer or carry”, and it implies a mapping between two domains, a transfer of one domain onto another to facilitate meaning (Proctor and Larson 2005). Metaphors allow people to understand abstract and perplexing subjects such as invasive species (Larson 2005), evolutionary biology, and environmental ethics (Rozzi et al. 2010b). Furthermore, they have been proposed as important tools when linking different disciplines together through interdisciplinary efforts (Oelschlaeger and Rozzi 1998). Recently, biological metaphors have been proposed as tools that aid in the visualization and clarification of complex scientific and day-to-day concepts, as they are drawn from everyday language and hence cannot be isolated from the social context (Larson 2006). Under this perspective, metaphors are considered cultural messengers, as they do not only constitute a
purely linguistic expression, but also represent a fundamental cognitive structure for human beings (Rozzi et al. 2010b).

For example, through the implementation of the metaphor “The Miniature Forests of Cape Horn”, scientists working at Omora Ethnobotanical Park have been successful at transferring and communicating their discoveries about the high diversity of non-vascular plants to decision makers and the general public. This metaphor provides a similitude between the group of biotic communities composed by mosses, liverworts, lichens, and the invertebrate inhabitants which live amongst them, with the biotic communities composed by large trees, shrubs, and epiphytes which are inhabited by birds and other organisms (Rozzi et al. 2010b). The composition and implementation of this metaphor, has inspired the development of multiple activities, which integrate ecological sciences, arts, and environmental ethics at a local, regional, national, and international scale. In this way, the use of this metaphor appeals to a large myriad of social groups, for whom the existence of non-vascular plants was previously almost entirely unknown (Rozzi et al. 2010b).

In this way, metaphors are not only effective in the communication processes with the general public, but are also effective in the synthesis and creation of novel ethical and ecological concepts (Rozzi et al. 2010a). In this context, during the year 2010, we conducted workshops with pre-school and middle school children, educators, and ecotourism operators from Punta Arenas and Puerto Williams (Chile) in the development of two novel metaphors: “The River as a Community of Life” and “Trichoptera: the Underwater Constructors.”

During these workshops, participants explored and experienced face-to-face interactions with the freshwater invertebrates inhabiting the streams and rivers of the Cape Horn Biosphere Reserve, helping in the development of the presented metaphors (Fig. 32). After and during the
field experiences, school children described aquatic invertebrate communities as “insect towns” and noticed that these “towns” resembled the communities in which they lived. Thus, as a group, we composed “The River as a Community of Life.” Furthermore, children and adults pointed out that certain insects, such as the Trichoptera (caddisflies), build their own homes from the materials found at the bottom of the stream. Participants felt that this habit closely resembled their own habit of constructing and designing their houses, and therefore we developed the metaphor “Trichoptera: the Underwater Constructors.” Appendix A provides a summary of the total number of participants and results produced during the development of the workshops.

The composition of metaphors as tools for communication goes far beyond the simple classification of an aquatic community as “a community of life” or of an insect order as “underwater constructors.” Through these metaphors, participants were able to understand important ecological concepts, such as community and population ecology, food web dynamics and nutrient cycling, and how the different inhabitants play a crucial role in the maintenance of this community of life. For example, these metaphors communicate ecological knowledge, which emphasizes the fundamental role played by the Trichoptera within the nutrient cycles of streams and rivers (Cummins and Klug 1979, Vannote et al. 1980). When these insects build their “houses” from materials derived from the forests around the rivers or streams, they help to decompose organic materials, which are then used as food by other inhabitants of the river’s “community of life.” In this sense, habitats (where we live), habits (how we live), and inhabitants (who we are) constitute an ecosystem unit even for the small animal communities (Rozzi et al. 2008a).

The face-to-face encounter with the Trichoptera and its habits promotes a better understanding and an appreciation for the intrinsic value of these organisms. While their
“building” habit evokes a similitude with our own habit of designing and constructing our homes, through our direct encounter we can also identify with how our own lifestyles can be impacted or modified by what the other-non-human transmits (Diehm 2003), thus we should not consider this similarity as a mere anthropomorphism of the Trichoptera. When we experience face-to-face encounters with these small “constructors”, we experience an opportunity for an ethical transformation: we cease to see the other than-human not only as an object of study, but also, we begin to appreciate and accept it as another subject, with whom we share the great diversity of habitats and habits in which we co-inhabit. In this way, we can see beyond the “object-trichoptera-insect” view and its “utility” as a water quality indicator, and move closer to a deeper understanding of its intrinsic value.

As we marvel ourselves with the constructions of these insects and begin to understand their role in the maintenance of freshwater ecosystems, we could propose the Trichoptera as a charismatic group for the conservation of Sub-antarctic watersheds. Thus, through the field experiences and the direct encounters with these small insects, we can better appreciate their intrinsic value and their ecological importance, while at the same time, promoting the conservation of Sub-Antarctic watersheds and their inhabitants.

- Step 3. Design of field activities with an ecological and ethical orientation – “Underwater with a Hand Lens”

Since the year 2000, the OEP education and environmental ethics program conducts the Omora Environmental Science Workshop at the school in Puerto Williams Liceo Donald McIntyre Griffits. This workshop has been a valuable addition to the school’s science education and has been formalized as part of its permanent curriculum. Furthermore this workshop emphasizes environmental ethics through field experiences and direct encounters with other
living beings (Rozzi et al. 2005). This approach counter balances the excess of “mediated information” (i.e. mathematical equations, audiovisual technology, and historically pre-imposed scientific narratives) prevailing in formal and informal education in today’s schools, even in remote regions such as Cape Horn, where curricula are “blind” to the regional diversity (Rozzi et al. 2005). For a decade, the workshop has been conducted with 5th and 6th grade students, who have experienced direct encounters and face-to-face interactions with the vast biodiversity of mosses, lichens, and birds of the Sub-Antarctic ecoregion. During 2010, we added a novel emphasis to the workshop, which was extended to 7th and 8th grade students, and it focused on the diversity and importance of freshwater invertebrates inhibiting Cape Horn’s rivers.

Throughout the year, weekly field experiences were carried out at the Róbalo (within Omora Park) and Ukika Rivers. Students learned about the ecological importance of freshwater invertebrates, how to identify them, and how to appreciate and better understand their intrinsic and instrumental values. As the workshops progressed, with participation by the students and their teacher, we used the metaphors previously described in order to teach students about freshwater invertebrates and developed the ecologically and ethically guided field activity “Underwater with a Hand Lens.” This activity aims to aid in the process of discovering, appreciating, and valuing the different invertebrates and community of co-inhabitants, their habitats, and habits. Underwater with a Hand Lens includes three main steps (Fig. 33):

1. Observation of Habitats: Visitors observe and identify the different macro and microhabitats within the river. They learn to distinguish between large and small scale habitats. Students and visitors first recognize and describe macro components of the river and the riparian forest ecosystem. Visitors better understand how terrestrial and aquatic ecosystems interact by noting how leaves, sticks and large pieces of wood fall in its waters to become the food and
home of the invertebrates. After distinguishing the main components of the riparian forest as an essential part of the river’s ecosystem, visitors pay attention to the microhabitats within the river. They learn to distinguish between riffle areas, pools, woody debris, or leaf packs that may serve as habitats for freshwater invertebrates. Familiarizing with the heterogeneity of habitats at different scales is essential for the understanding of the “community of life”, as the invertebrate inhabitants use different habitats of these systems during their life cycles. In this context, visitors observe the immature stages in the water (larvae or nymphs), as well as their adult forms, flying and mating in the forest.

2. Face-to-face encounters with the underwater inhabitants: Visitors go to river’s shore and pick up a submerged rock, recording the exact place in which they found it. The rock is placed in a container with water and the visitors observe how the invertebrates, the “underwater inhabitants”, begin to move and swim within the container. With the help of a hand lens, visitors carefully observe the diversity of the river’s live inhabitants, their micro-habitats, and relate it to the macro-habitats observed in the previous step.

3. Valuing and respecting the habitat, habits, and inhabitants of the rivers’ of Cape Horn: After observing the invertebrates, visitors place the rock on the exact same area in which they found it, and with it, return all invertebrates alive to their microhabitats in the river.

The third step of the activity, which involves returning the rock to the exact same place to where it was found, represents an essential action towards an ethical, educational, and eco-tourism experience, that transforms the understanding and attitudes of visitors toward the river, and the community of freshwater invertebrates. The rock is the home of many invertebrates and the place where it was found is not trivial. By returning the rock to its place of origin, participants value and respect the habitat and habits of the underwater inhabitants. They cease to
see the invertebrates, the other-than-human inhabitants, as a mere objects of study and begin to appreciate them as subjects, as living beings with whom we coexist and share our habitats and lives.

Through this activity, visitors also gain an experiential understanding of the metaphor “The River as a Community of Life.” They begin to understand the river as “more than just water”, by coming closer to its habitats, inhabitants and their habits. By experiencing a face-to-face encounter with the freshwater invertebrates of the Róbalo River, visitors experience a further ethical transformation, which allows them to better understand the instrumental and intrinsic values of the freshwater invertebrates. According to Callicott (1995), the intrinsic value of a thing is its value for its own sake, as an *end* in itself. Some values are instrumental (means to an end) to something else, but some values must be *intrinsic*, which means that they must be valuable in themselves and not as means to other ends (Callicott 1995). In contrast, the instrumental value of a thing does not lie within that thing in itself, but it lies in the utility that it posses as *means* to and *end* (Primack et al. 2006).

This experience helps visitors to better understand the intrinsic and instrumental values of freshwater invertebrates in the rivers of Cape Horn. Toward that aim, we have read and analyzed another essay by Aldo Leopold *Thinking like a Mountain*. In this essay, Leopold integrates both the instrumental and intrinsic values of biodiversity. Regarding the first, he refers to the howl of a wolf and describes what it means for other living beings, humans included. Leopold describes a mountain ecosystem and its trophic interactions in terms of its habitats (the mountain, the farm, and the pine forests), inhabitants (the wolf, the deer, the coyote, the farmer, and the hunter), and their habits surrounding the howl of the wolf. To the farmer, the wolf represents “a threat of red
ink at the bank”, to the deer, “it is a reminder of the way of all flesh”, and to the hunter “a challenge of fang against the bullet.”

Regarding the intrinsic value, Leopold refers to the moment of the dying wolf, and experienced an ethical transformation:

We reached the old wolf in time to watch a fierce green fire dying in her eyes. I realized then, and have known ever since, that there was something new to me in those eyes – something known only to her and the mountain. (Leopold 1949b)

At that moment, Leopold saw that the wolf shared a lot with him. The wolf was a living being, just like him, she had two eyes, and through those eyes, the wolf could communicate, learn, and feel, just like he could. As Leopold killed the wolf, he discovered that there was “something new in her eyes, something known just to the wolf and the mountain” (Leopold 1949b), and he perceived the importance of the wolf for itself, its family, and the mountain. He no longer saw the wolf (or other predators) as a negative influence for the deer population (or the ranchers); instead he saw it as part of a whole, as part of a “community of life.” In this sense, Leopold experienced a change of vision from the instrumental to the intrinsic value of the wolf, which was only accomplished by his direct encounter with the dying wolf.

In the case of “Underwater with a Hand lens”, participants can experience a similar ethical transformation, because they can understand that the river ceases to “only” be the supplier of drinking water to Puerto Williams, they also see “face to face” with the little invertebrates how it is the “home” or “oikos” of the underwater inhabitants. By carefully observing them and their home, participants relate to these inhabitants as other living beings, with whom they share their habitats (the river, the forests), and living habits along or within the watershed of the Róbalo River. Thus, just as Leopold experienced the face-to-face encounter with the wolf, through “Underwater with a Hand Lens” participants experience a direct encounter with the
underwater inhabitants of the Róbalo River. They perceive first hand the other non-human living beings and better understand the *intrinsic value* of each them (Rozzi 2007) as part of the river’s community of life.

- Step 4. In-situ conservation: “The Underwater Inhabitants of the Rivers of Cape Horn”, interpretative trail at OEP.

As it was previously mentioned, the Sub-Antarctic Magellanic ecoregion is one of the 24 most pristine areas remaining in the world (Mittermeier et al. 2003), but it is not free from global and local threats such as global climate change, the introduction of invasive exotic species, and tourism. Today, the remote Sub-Antarctic archipelago is critically threatened by massive tourism, as for the global citizen, this remote region represents one the last “wild” destinations, and it is experiencing an explosive growth in the number of foreign visitors (Rozzi et al. 2010d). Additionally, the Regional Development Plan for the Chilean Region of Magallanes and Antarctica has identified tourism as one of the five main priorities for economic development, resulting in a doubling number of tourists in the Magellanic region and an increase of one order of magnitude in Puerto Williams during the last decade (Rozzi et al. 2010d). In this context, the regional government has encouraged the development of new forms of sustainable eco-tourism to offer an alternative for social well-being, as well as for the conservation of indigenous ecological knowledge and pristine environments, such as Sub-Antarctic habitats.

In this context, Omora Park researchers have developed new and innovative forms of eco-tourism, such as *Tourism with a Hand Lens* (Rozzi et al. 2010b) and the newly inaugurated trail *The Miniature Forests of Cape Horn* (Rozzi et al. 2010d), which involve the participation of the local community, particularly the Yahgan Indigenous Community, educators and pre-school, school, and university students.
As an effort to contribute to the development of sustainable forms of eco-tourism in the Magallanes region, and to promote the in situ conservation of freshwater invertebrates and their habitats, we designed and built a new interpretative trail at Omora Ethnobotanical Park, which introduces a novel theme and type of eco-tourism in Chile: the conservation of freshwater insects. We also implement the metaphors “The River as a Community of Life” and “Trichoptera: the Underwater Constructors”, as well as the ecologically and ethically guided field activity “Underwater with a Hand Lens.” Additionally, in order to further integrate Yahgan traditional ecological knowledge into eco-tourism activities within the region, we integrate the story of Omora as an essential part of the trail’s narrative, by communicating its history and meaning for the conservation of Sub-Antarctic watersheds.

In this way, the main objectives of the trail “The Underwater Inhabitants of the Rivers of Cape Horn” are to promote the observation and valuing of:

a. Large and small patterns: visitors observe and learn about the geographical and historical context in which the Róbalo River is immersed in (i.e. Dientes de Navarino Mountain Range, Beagle Channel, impacts by invasive species, microhabitats);

b. Ecological processes: visitors distinguish between the river’s headwaters and its mouth, understanding concepts such as primary and secondary productivity, and organic matter decomposition)

c. Biodiversity components: visitors learn how to identify invertebrates, vertebrates, and plants within the riparian forest.

These objectives are fulfilled by guiding visitors through 4 interpretative sites, within a 850-meter trail (Fig. 35, Appendix B). Following is a description of each interpretative site, along with its total length (in meters):
Station 1: Riparian Forests (50 m). This site is immersed within the riparian forest of the Róbalo River. Visitors distinguish between the different components of a watershed by learning how the riparian forest is an essential component for its maintenance, which provides habitats and nutrients for the different inhabitants of the river. Furthermore, visitors are invited to identify the different species of trees, such as *Nothofagus betuloides* and *N. pumilio*, and lichens, such as *Usnea* spp., as important components of the Sub-Antarctic forest (Appendix B, Interpretative trail brochure, pages 3-4, Fig. 36).

Station 2: The Bridge (15 m). Visitors are invited to cross the Róbalo River in order to experience a “direct encounter” with the impact of the exotic and invasive North American beaver (*Castor canadensis*), on Sub-Antarctic watersheds. The beaver was introduced to the Sub-Antarctic ecoregion in the 1940’s and since then it has invaded the majority of its watersheds. Furthermore, the guide provides the story of the bridge, in which biologists, artists, philosophers, and engineers associated to the Omora Ethnobotanical Park, worked together to build a bridge that “never touches” the river in order to protect the “house” of the freshwater invertebrates (Appendix B, Interpretative trail brochure, pages 5-6).

Station 3: Vertebrate inhabitants (230 m). In this site, visitors learn about birds and insects that act as links between terrestrial and freshwater habitats. As they walk through the forest, visitors discover small and large insectivorous birds, such as the, Magellan wood-pecker (*Campephilus magellanicus*), the fire-eyed diucon (*Xolmis pyrope pyrope*), the wren (*Troglodytes aedon chilensis*), and the thorn-tailed rayadito (*Aphrastura spinicauda spinicauda*), which are often seen feeding on the larvae of terrestrial beetles (Cerambycidae: Coleoptera), as well as adult aquatic insects, such as dragonflies (Aeshnidae: Odonata) and crane flies (Tipulidae: Diptera). Furthermore, as visitors approach the river’s banks, they learn about how
most adult aquatic insects that emerge from streams live briefly in the nearby riparian zone, where their adult activities, such as mating, dispersal, and feeding, influence their distribution and the distribution of birds in the riparian forest (Appendix B, Interpretative trail brochure, pages 7-8).

Station 4: The underwater inhabitants (80 m). After leaving site 3, visitors walk by the shores of the Róbalo River, observing and describing the different macro (riparian forest, beaver dams, etc) and micro-habitats (riffles, woody debris, leaf packs, pools, etc). As they walk through the shore, visitors approach an observatory platform (Fig. 5), in which the ecologically and ethically guided activity “Underwater with a Hand Lens” (previously described in Step 3) is performed. The guide provides the visitor with a set of hand lenses, identification keys, plastic spoons, soft-touch forceps, and a plastic container in order to carry out the activity (Fig. 5). Visitors learn about the great diversity of freshwater invertebrates that live in the Róbalo River, as well as about their importance for the maintenance of its water quality. As it was previously described, “Underwater with a Hand Lens” aims to a) aid in the process of discovering, appreciating, and valuing the different invertebrates and community of co-inhabitants, their habitats, and habits, through a “face-to-face” encounter, and b) value and respect the home of the underwater inhabitants.

Finally, the design and development of the trail was possible due to the collaboration between biologists, engineers, artists, and philosophers associated to the Omora Ethnotabotanical Park, and it was financed with the financial support of Chile’s Corporación de Fomento de la Producción (Innova-CORFO). In this way, the new structures, such as the bridge and the “Underwater Inhabitants” sites represent an in situ conservation area for the watershed of the Róbalo River and its vertebrate and invertebrate inhabitants.
Discussion

The integration of freshwater invertebrates into Omora’s Field environmental philosophy methodology allowed us to successfully translate our findings into workshops with Puerto Williams’ 7th and 8th grade students, who conducted research associated to our findings and obtained Chile’s National Science Award for middle schools (Fig. 34). Finally, based on this activity and the metaphors previously described, students designed and conducted a research study entitled “Richness and abundance of aquatic insects in rocks and woody debris associated to the Ukika River, CHBR.” This study was inspired by the direct encounters experienced with the underwater inhabitants, as they noticed that most of them lived on submerged rocks. During data collection, students did not collect invertebrates for further identification in the laboratory (collecting would include preserving them in 70% ethanol), instead, they spent more time in the field, carefully identifying them, and consciously placing them back to the exact same place in which they were found. Thus, during this study, the students were able to successfully integrate ecological research and environmental ethics into a research project which was presented to the XI Annual National Science and Technology Conference for Chilean Schools, organized by Chile’s EXPLORA CONICYT (Comisión Nacional de Investigación Científica y Tecnológica) program, obtaining the first place and the National Science Award for best middle school research projects (Fig. 36).

Furthermore, we implemented a new interpretative trail at OEP, which allows visitors to explore the importance of the Róbalo river watershed and its inhabitants, participating in the ecologically and ethically guided field activity “Underwater with a Hand Lens.” By following this methodology, we have witnessed and recorded transformative ethical experiences by students, researchers, and other participants, who are able to translate their discoveries into
ethical and responsible actions that stimulate, in turn, new questions research, metaphors and sustainable activities in the Cape Horn Biosphere Reserve (Fig. 37).

Chapter References


Fig. 29. Ecoregion of Sub-Antarctic Magellanic forests (dark gray). Navarino Island is located north of Cape Horn and south of the Beagle Channel. The Cape Horn Biosphere Reserve and the Omora Ethnobotanical Park are located within this ecoregion (Rozzi et al. 2008c).
The Cape Horn Biosphere (CHBR) has an extension of 4,884,274 ha of marine and terrestrial ecosystems (Rozzi et al. 2007). The map illustrates the core (green), buffer (pink), and transition (yellow) zones of the CHBR. The Omora Ethnobotanical Park, is one of the three founding sites of Chile’s first Long Term Socio-Ecological Research network, and serves as an interdisciplinary research center and nature reserve for the CHBR. It is located in Navarino Island (55°S), 3 km south of Puerto Williams, the capital of Chilean Antarctic Province, and the southernmost town in the world (Source of the Cape Horn Biosphere Reserve Initiative 2005).
Fig. 31. Four-step cycle of the Field Environmental as it applies to freshwater invertebrates of the Cape Horn Biosphere Reserve in order to integrate ecological sciences and environmental ethics into biocultural conservation, education, and ecotourism. Each step is indicated in blue, the method used in green, and the results obtained are indicated in black. The black arrows indicates that the four steps are interconnected and interrelated with each other (Modified and adapted from Rozzi et al. 2008c).
Fig. 32. Workshops carried out with middle school (A) and pre-school students (B) from the local public school in Puerto Williams, Liceo Donald McIntyre Griffits. Picture C depicts a Trichoptera family as viewed by a 5-year-old student.

Fig. 33. Steps of the ecologically and ethically guided field activity “Underwater with a Hand Lens.” A student or a local ecotourism operator at “The Underwater Inhabitants of the Rivers of Cape Horn” interpretative trail within Omora Ethnobotanical Park guides the activity. The guide provides identification keys for the major groups of invertebrates, hand lenses, soft touch forceps and small nets to retrieve the rocks from the bottom of the river.
Fig. 34. Seventh and eight grade students from Liceo Donald McIntyre Griffits, Puerto Williams, Chile, conducting and presenting their research about aquatic insects associated to the Ukika River, Puerto Williams, Navarino Island, Chile. The students participated in Chile’s 2010 regional and national science and technology fair, organized by Explora and Conicyt (The National Commission for Scientific and Technological Research), obtaining the National Science Award for elementary schools category. From left to right (above picture), Felipe Leyton (8th grade), Francisco Olivares (8th Grade), Tamara Contador, and Carlos Saavedra (7th grade).
**Fig. 35.** Satellite image of Omora Ethnobotanical Park’s main interpretative trails. The Underwater Inhabitants of the Rivers of Cape Horn is represented by a white line. Stations are indicated with the numbers 1 through 5. Refer to Appendix A for a detailed description of each site.

**Fig. 36.** Bridge built in the interpretative trail The Underwater Inhabitants of the Rivers of Cape Horn. The bridge was built in an interdisciplinary effort with the help of engineers, artists, philosophers, and biologists associated to the Omora Ethnobotanical Park.
**Fig. 37.** Applying Field environmental philosophy at the Omora Ethnobotanical Park: Aquatic Invertebrates of the Southernmost Watersheds. This Fig. summarizes the implementation of Omora’s FEP to the study of freshwater invertebrates associated to the Róbalo river watershed, Puerto Williams (Navarino Island, 55°S).
APPENDIX A

DESCRIPTION AND NUMBER OF PARTICIPANTS
<table>
<thead>
<tr>
<th>Step</th>
<th>Number of participants</th>
<th>Indicators/Results</th>
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| 1) Ecological and Philosophical Research       | Multiple researches associated to Omora Park, University of North Texas and Universidad de Magallanes, Chile. | • Description of diversity and altitudinal zoning of freshwater invertebrates along the RRW  
• Description of life histories of selected aquatic insects |
| 2) Composition of metaphors and communication through simple narratives | Liceo Donald Mc.Intyre Griffits:  
• Pre-school: 30 students  
• 7<sup>th</sup> and 8<sup>th</sup> grade: 33 students  
• Teachers and parents: 17 participants  
• Multiple researchers associated to Omora Park, University of North Texas and Universidad de Magallanes, Chile. | • Composition of two novel metaphors:  
  o The River as a Community of Life  
  o Trichoptera: the underwater constructors  
• Communication and implementation of metaphors through workshops, school and community activities in Puerto Williams, Chile. |
| 3) Design of field activities with an ecological and ethical orientation | Liceo Donald Mc.Intyre Griffits:  
• 7<sup>th</sup> and 8<sup>th</sup> grade: 33 students  
• Multiple researchers associated to Omora Park, University of North Texas and Universidad de Magallanes, Chile. | • Design and implementation of ecologically and ethically guided field activity “Underwater with a Hand Lens” |
| 4) Implementation of physical spaces for in situ conservation | Liceo Donald Mc.Intyre Griffits:  
• 7<sup>th</sup> and 8<sup>th</sup> grade: 33 students  
• Design and implementation by engineers, artists, biologists, and philosophers associated Omora Ethnobotanical Park | • Design and implementation of new interpretative trail within Omora Park “The Underwater Inhabitants of the Rivers of Cape Horn” |
Benthic macroinvertebrate field identification guide, postcards, and brochure published during the development of this dissertation. The brochure presented, will be used as a guide for visitors of The Underwater Inhabitants of the Rivers of Cape Horn trail at Omora Ethnobotanical Park. The postcards will be distributed at a regional level in Punta Arenas, and at a local level in Puerto Williams.