Doctoral School in Civil, Environmental and Mechanical Engineering

Topic 1. Civil and Environmental Engineering

Elisa Stella

Analysis of the impact of hydrological alterations and multiple stress factors on the ecological status of Alpine freshwater ecosystems



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Doctoral School in Civil, Environmental and Mechanical Engineering Topic 1. Civil and Environmental Engineering

Doctoral Thesis - June 2018

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Analysis of the impact of hydrological alterations and multiple stress factors on the ecological status of Alpine freshwater ecosystems

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# If you don't know where you are going any road can take you there.

LEWIS CARROLL

### ABSTRACT

reshwater ecosystems are severely undergoing degradation due to the presence of multiple stressors that are undermining their biodiversity. In this sense, quantifying these effects on Alpine regions is challenging, due to the lack of tailored field measurements of hydrological, biological and chemical variables. This work aims to touch some of these aspects, with particular attention to hydrological dynamics and their effects on macroinvertebrates. Field activities have been conducted within the Adige catchment which has been selected as a case study in the FP7 project GLOBAQUA. Collected data have been analyzed by means of statistical tools and results showed a seasonal and spatial variability of biological communities related to hydrological and chemical variables. In particular, it has been observed that richness, diversity and relative composition of macroinvertebrates community are chiefly affected by hydrological alteration and urban pollution. Available literature confirmed that hydrological alteration is one of the most important factors affecting riverine ecosystems. In Alpine regions, most of the hydrological alterations observed are due to hydropower that represents the major source of energy in the Trentino-Alto Adige region. Since the introduction of the free energy market in Italy, hydropower production shows large fluctuations at the daily and larger temporal scales, as the managers aim at producing when the energy price is high. This increased the variability of streamflow downstream the restitution of the power plants. Changing climate is an additional stressor that can enhance the effects of these anthropogenic influences. Thereby, in this work hydrological alterations have been distinguished between those forced by climate change and those caused by the presence of hydropower

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plants and have been analysed in detail. The former have been analysed downstream of the Careser glacier, which has long-term observations of climatic variables, mass balances and streamflow. The main purpose of this study was finding a direct relationship linking biological indicators to streamflow variations related mainly to hydropower operations. Quantifying these effects is challenging due to the fact that the behaviour by which macroinvertebrates respond remains largely unexplained. However, analyses of similarities and independence, performed at the basin scale with data provided by the local Environmental Protection Agencies, showed evident differences in the biological communities between impacted and non-impacted sites. These results bring us to believe that a relationship between biological data and hydrological alteration is expected to exist, but that is not clearly explicated by simple correlations. Giving a quantitative interpretation of this correlation could help hydropower manager to improve and optimize the energy production with a more realistic scenario of the effects on the biological community, with also a perspective of the combined effects caused by the presence of multiple reservoirs within the basin.

### **DEDICATION AND ACKNOWLEDGEMENTS**

n writing this thesis these three years have come in my mind all at once. The good, the bad and the ugly. At the end, the good beats the bad and "saves" the ugly. I owe this good to many people who have been part of these years. First, I would like to thank my supervisor and advisor Prof. Alberto Bellin, for his support and suggestions, to him I owe the opportunity to undertake this path. My gratitude goes also to Valeria Lencioni, for her help in understanding biological concepts. For this I thank Francesca Paoli and Alessandra Franceschini too. Then, all the moments I enjoyed at work were with my colleagues, office mates and friends: Karina, Ines, Stefano, Elena, (I want to include also Ulisse, even though he's never been in the office, nor a colleague), Oscar, Bruno and Erika. I would like to thank Elena also for having been in the field, in the water and in the mud with me, it was the funniest part of this work, moreover she is a good friend. This department gave me the chance to meet also other beautiful people. Among all, my heart goes in particular to Marialaura, who is not only a friend, she's my support and thrust, she's been and still is part of all. Since when Marialaura went back home, Michele (or better Mich, so as not to be mistaken, I know you want to be precise in these things) has being always at my side and my good wingman here in Trento, thank you also for the gas lighter (it's quite a long story). Then I owe Giuseppe a negroni, for having helped me many and many times and occasions, my gratitude will be never enough. In these years I have had the pleasure to meet also Enrico (Raisenfor), a friend, source of many laughs, meaningful discussions and good stories (even though repeated some times). I want also to thank all the members (current, old, friends and co.) of the Zanella house, my turning point during this experience: David, Juanpa, Michele,

Nadia, Sergio, la cantina pirata, Irene, Kim, Stefania, Germana, Vittorio, Filippo and last but not least Paco. I thank David also for putting up with me in my role of "sergeant" and many thanks go to Nadia for the wonderful cover of this thesis! Then, learning theatre is among the most meaningful experiences I had here in Trento, thereby I want to thank all the people I had the pleasure to meet in there. Anna and Vale, then, are a precious part of it and a precious part of my life as well. Even though far from me, and rare the occasions to meet, this work is dedicated also to Carolina, my fighter and little but strong concentration of life, and Enrico, who is much more prudent than me and Carolina, but at the end he has always put up with us (maybe now, being in USA, is easier), I always keep them in my thoughts. Moreover I owe another negroni to Steven, my counterpart and among my dearest and oldest friends. A special and grateful thank is for my parents, who always believe and trust in me, and also for my brother Michele, together with Silvia, who gave me the joy to be aunt of two wonderful children, Alessandro and Kevin, may their curiosity be always the reason, the guide and the path. My thank goes also to Gabriele and Giusy, together with Giada, who welcomed me in their home and family and make me feel a member of it. Lastly, from the bottom of my heart, head and stomach I dedicate this work to Riccardo, who is always by my side, in the good, the bad and the ugly, thanks for all the patience and care in pushing me in everything, you are the freedom I need.

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# CHAPTER

### **INTRODUCTION**

One of the most difficult things during a PhD is to answer people what your research is about, especially to those who are not in the field. The answer cannot be too long nor too specific. However, being too general is also a defect: once, another engineer answered me that he was doing mathematical models, well, great, but "concerning what?" (with the assumption that one has to understand what you are talking about, and in this case to have in mind what's the meaning of mathematical models). During a summer school a professor suggested to try giving the answer as if we had one minute of conversation with a stranger in an elevator, and we have only that minute to explain everything. However, I found that even a minute could result too long for someone, so I reduced my answer to: "I study the ecological impact of hydropower plants and more in general of hydrological alterations". Then, when someone asks me details and I explain that I study the responses of macroinvertebrates the subsequent questions are: "what are macroinvertebrates? Why them?", however most of the time my interlocutor has

already lost his interest at the words "macroinvertebrates" or "hydropeaking" (it was encouraging finding that it doesn't happen only to me). Nevertheless I try to explain that according to the "Water Framework Directive (WFD)" (interlocutor's frown) macroinvertebrates, that are animals that live underwater in our streams and rivers, lack a backbone, and can be seen by the naked eye (yawning), are considered good and convenient indicators of the health of riverine ecosystems as their abundance and richness enables an objective judgment of the ecological condition (snoring).

However, in this circumstance I find these answers too general, thus following I try to be more specific. As hydrologist, the initial purpose of this PhD was to use hydrological models to understand the effects of hydrological alterations on the wild biodiversity, so that to provide a simple tool to improve the management of water resources. Thereby, the initial step was to find a relationship between hydrological variables and available biological indicators. However at this step I found that such relationship is still missing, and moreover that macroinvertabretes don't show unique responses to hydrological alterations [85]. Furthermore, within the objectives of the FP7 European project GLOBAQUA

(http://www.globaqua-project.eu/en/home/, [79]), part of this work has been dedicated on field samplings and on some analyses of the obtained data.

This work focuses on Alpine river systems. In fact they are essential resources in Europe since they provide freshwater for human consumption and for productive activities such as agriculture, livestock and industry [100, 101]. Given the wide range of activities conducted in its catchment, resulting in a multiplicity of stressors, the Adige River basin was selected in the GLOBAQUA project as a case study representative of the Alpine region. The predominant pressures affecting the Adige River are: (i) streamflow and water temperature alterations caused by hydropower production [118, 119]; (ii) land use (mainly agriculture) and industrial activities [21], which are increasingly present downstream; and (iii) nutrients and pollutants released by waste water treatment plants (WWTPs) i.e. effluents, which are expected to show significant temporal variations due to seasonal fluxes in tourists [24]. On this basis, in this study we 1) analysed hydrological responses caused by anthropogenic pressure (i.e. hydropower operations) and climate change, this last has been evaluated for the Careser glacier case study, which holds long-term observations of climatic variables, mass balances and streamflow; 2) explored in general the effects on macroinvertebrates of multiple stressors present within the Adige catchment and then focused on the interaction between anthropogenic hydrological alterations and biological response, both quantitatively and qualitatively and at

the basin scale. Hereunder, in this section, we briefly introduce and contextualize the main issues that have been distinguished in this work.

# 1.1 Hydrological alterations within Alpine systems.

Most river systems across the Alps suffer from streamflow alteration, which induces changes in sediments and organic matter loads, thereby threatening ecosystem integrity [118]. Most of these changes are caused by anthropogenic pressures (e.g. land use, industry, agriculture, water abstraction, etc.), and among these the most impacting is undoubtedly the streamflow alteration due to hydropower exploitation. Changing climate is an additional stressor that can enhance the effects of these anthropogenic influences. Thereby, part of this study focuses on detecting the effects of these alterations that we identify as follows: anthropogenic alterations related to hydropower production, and hydro-climatic changes.

Anthropogenic alterations related to hydropower production. In the 1960s a large number of reservoirs were built in the Alps to enhance hydropower production in an attempt to meet energy demand, particularly in winter months, when streamflow is low. Since the introduction of the free energy market in Italy, hydropower production shows large fluctuations at the daily and larger temporal scales as the producers aim at producing when the energy price is high. This increased the variability of streamflow downstream the restitution of the power plants [105, 118].

In the Adige river basin, water is the major energy source; a large number of reservoirs were built to take advantage of the high hydropower potential of the region. This leads to a significant streamflow alteration in the basin, particularly at intermediate and low flow regimes [118].

Current methods for understanding hydrological dynamics consider five major components of flow regime: magnitude, duration, frequency, timing and rate of change [73]. These components are generally explained by 32 variables deemed "ecologically relevant" (IHA variables, [89]), leading to complexities in finding a general relationship that links streamflow variations to ecological biodiversity.

In the present study hydrological alterations within the Adige catchment have been investigated at daily and sub-daily scales. To simplify the following analyses on hydro-biological interactions, we selected sites that explain various rates of alteration and we compared altered streamflow to the natural regime (i.e. in the absence of water uses) by evaluating basic statistics.

**Hydro-climatic changes.** The relevant role of climate change on the observed alterations of the water cycle in snow-dominated regions is widely recognised, particularly in the European Alps [6, 9, 42, 70].

In Alpine headwaters meltwater from glaciers contributes significantly to downstream river flow in summer [58, 63], when scarce contribution comes from other sources [52]. However, these systems are experiencing rapid degradation all over the world, thereby raising concern on the many implications on downstream services [39, 75, 109]. A number of contributions suggest that recent changes in the precipitation patterns and the contemporaneous rise of temperatures have led to rapid glaciers melting with a consequent alteration of the typical Alpine hydrological regime [10, 62].

In this regard, Alpine glaciers, with observations dating from 1850 [83], have reported among the most negative mass balances [11]. According to the last estimate [82] glaciers and ice patches in the European Alps cover one-third of the area measured in the 1970s (i.e. a loss rate of about  $2\% \text{ y}^{-1}$ ). Further studies [47, 53] observed that current rates of glaciers retreat have strongly increased above long-term averages and according to the most recent estimate are of the order of about -1 m of water equivalent (w.e.) $\text{y}^{-1}$ .

While many studies reported the overall reduction of glacier mass due to climate warming, its consequences on downstream streamflow are less debated. In the Alpine rivers a number of studies focused on assessing future scenarios [7, 35, 54, 61, 107] while few others analysed existing observed data [29, 30, 45, 46, 93]. Thereby gathering all relevant data in order to understand how climate change is affecting the hydrological regime represents a relevant task that may improve the understanding of recent trends, their drivers and impacts on downstream ecosystem services and accordingly to adapt their management.

In this respect, we have analysed a long-term streamflow time series in the Careser creek emerging from the Careser Glacier. With monitoring activities documented since the end of the 19th century, this glacier has been largely investigated [18-20, 115]. Furthermore, records of streamflow are available since the 1920s from the company that uses its meltwater for hydropower purposes. Because of these long-term observations, the Careser has been classified as one of the reference glaciers by the World Glacier Monitoring Service, which provides balances data every two years [117]. We investigated the on-going changes of the hydrological regime by performing a comprehensive analysis of multiscale variations of precipitation, temperature, water discharge and glacier mass. We analysed downstream hydrological variations associated to climatic drivers and we assessed the effects detected over the long-term period. We then discuss our results in relation to the decline of the glacier surface and of the resulting mass balances.

# 1.2 The effects of hydrological and chemical stressors on macroinvertebrate community.

### 1.2.1 Multiple stressors on freshwater ecosystems.

Agricultural, industrial and domestic activities exert pressures on freshwater ecosystems, in some cases impairing their ability to provide essential services [31]. Threats to freshwater biodiversity are grouped under a number of interacting categories such as water over-exploitation, water pollution, flow modification, destruction or degradation of habitat, geomorphological alterations, land use changes and invasion by exotic species and pathogens [4, 34, 80, 103] . Diffuse (e.g., agricultural activities, intensive animal farming) and point (e.g., from urban areas due to the increase in the human population density) pollution are the main sources of contaminants entering freshwater ecosystems. In particular, concerns have been raised regarding pesticides (insecticides, herbicides, fungicides), pharmaceutical products (PhACs) and personal care products (PCPs) [55, 80].

Studies conducted by Lencioni et al. [65, 67] provided basic knowledge on the structure and functional properties of Alpine invertebrate communities. Other studies focused on the effects of specific factors, but studies on the combined effects of a multiplicity of stressors are still lacking in the Alpine region. In this regard, additional effort is required to better understand and quantify the fate of contaminants at the river basin scale [13, 43, 91]. In the same way, the investigation, within a coherent framework, of the impact of micro and emerging pollutants [74], diffused sources of pollution [90] on freshwater ecosystems, is a challenging task. Therefore, the application of a comprehensive approach that allows the effects of multiple stressors to be investigated at a given location may provide essential information to better understand and assess biological responses to this multiplicity of stress factors.

In this context, part of this PhD thesis aimed to identify the relationships between multiple pressures and the response of the invertebrate community at targeted sites, which are representative of a number of scenarios encountered in Alpine rivers, considering the Adige river basin as case study. Specific attention was paid to the middle course of the Adige River, in the province of Trento, and to one of its main tributaries, the Noce River. We expected that (i) seasonal patterns of hydrological and chemical parameters would be observed according to the natural hydrological regime and the different water uses (tourist activities in winter, agriculture in spring-summer) within the river basin; (ii) the richness and diversity of invertebrates would decline along the river network as a consequence of changes in water pollution and hydrological alterations; and (iii) the changes in community composition would show a seasonal pattern related to the temporal pattern of the different pressures.

# 1.2.2 Assessing the role of anthropogenic hydrological alterations on macroinvertebrate biodiversity.

The influence of streamflow on the preservation of ecological biodiversity is an established concept [4, 17, 84]. A number of studies define the role of streamflow as a dominant factor in the characterisation of riverine ecosystems, assessing that many types of flow alteration (e.g. magnitude, frequency, and timing) induce a variety of ecological responses [17]. Generally, flow regime varies geographically in response to climate (precipitation and temperature) and catchment controls on runoff (topography, geology, land cover, position in network). As aforementioned, in Alpine systems flow alterations mainly derive from hydropower plants operations and in particular hydropeaking mostly affects ecosystems of the downstream watercourses, modifying the specific composition and longitudinal zoning of downstream invertebrate populations [22, 23].

A recent review [86] analyses the existing literature based on ecological responses to alterations on the flow regimes, trying to detect any potential relationships that would be especially useful in a management context [3]. It has been observed the relative sensitivities of different ecological groups (i.e. fishes,

8

macroinvertebrates and riparian vegetation) to alteration in flow magnitudes, but robust statistical relationships were not supported. However, while the negative response of fishes has been unambiguously demonstrated, macroinvertebrates showed mixed responses to changes in flow magnitude, with abundance and diversity both increasing and decreasing in response to elevated and to reduced flows. Lloyd et al. (2003, [68]) identified several constraints that limit attempts to derive quantitative relationships and suggested the use of larger dataset in order to cover a broader range of responses and hydrological alterations.

On this basis, and given the fact that in the existing literature most case studies focus on smaller scales (i.e. local or reach scale), this work attempts to investigate the effects of anthropogenic hydrological alterations on benthic invertebrates at the basin scale, considering the Adige river basin as case study, limited to the territories of the Provinces of Trento and Bolzano. We used biological data provided by the local environmental agencies of the two Provinces and performed correlation analyses between biological indicators and hydrological variables, obtained with streamflow data provided by the Hydrological Offices of the Provinces of Trento and Bolzano. We then tested the influence of hydropower operations on the composition of the macroinvertebrate community by performing an analysis of similarity (ANOSIM). We finally verified the influence of seasonality and type of river (depending on the received contribution, glacial or surface, and on the distance from the source).



#### **STUDY SITE, DATA AND METHODS**

### 2.1 Study site: the Adige river basin

The Adige is the second longest river in Italy (409 km) and flows from the Alps into the Adriatic sea at Porto Fossone between Brenta estuary and Po River deltas. It rises from a spring located in proximity of the Resia lake at the elevation of 1.586 m a.s.l. and its catchment drains an area of about 12.100 km<sup>2</sup> in NE Italy (see Figure 2.1), becoming the third largest basin after Po and Tiber basins. The highest peaks in the catchment are above 3.500 m a.s.l. The mean discharge registered at Boara Pisani gauging station (45°06′19″*N*, 11°47′02″*E*) is about 202 m <sup>3</sup>/*s* [5], half of which occurs in the period from June to September, showing the typical behaviour of Alpine catchments.

The Adige river flows through the territories of the Province of Bolzano (62% of the overall river basin surface), the Province of Trento (29%) and the Veneto region (9%). From the source to Merano the Adige flows through the Venosta valley (with a drainage area of 1.680 km<sup>2</sup>), then it heads south and receives the contribution of the Isarco river at Bolzano, and of both the Noce and Avisio rivers just upstream of Trento, where its drainage area rises to 9810 km<sup>2</sup>. Then, towards the end of its course it reaches Verona (drainage area of 11.100 km<sup>2</sup>) and flows through the Padana plain without receiving other significant contributions. For this reason, hydrological analyses have been conducted chiefly in the northern part of the catchment within the territories of the provinces of Trento and Bolzano. The main tributaries of the Adige river are the following: Passirio, Isarco, Noce, Avisio and Fersina. The river basin encompasses almost 550 lakes, most of which have a surface smaller than 1 hectare and are of glacial origin [5]. The biggest natural lake is Caldaro, followed by Anterselva, Braies and Carezza in the Province of Bolzano, Lake Tovel and Terlago in the Province of Trento.

Climate in the Adige river basin is characterised by dry winters, snow and glacier-melt in spring, whereas it is humid during summer and autumn. Annual average precipitation values range between 500 mm in Val Venosta and 1600 mm registered at high altitudes and in the southern part of the basin. Streamflow shows a typical Alpine regime with two peaks, one in spring due to snowmelt and the other in autumn due to cyclonic storms, which are the main cause of flooding events. In winter, when the precipitations within the catchment are chiefly solid, streamflow is minimum. Summers are typically wet and dominated by short and intense convective storms.

The main agricultural areas in the provinces of Bolzano and Trento are located in the Adige, Noce and Venosta valleys and the cultivation comprise mainly fruit trees and grapes [21]. Land use at high elevations is dominated by grass, grazing and forest. Mountains cover an area of around 9.700  $km^2$ , with very wide forest, pioneer vegetation and exposed rocks. The largest urban areas are located in the main valleys, where land use is more

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Figure 2.1: Map of the Adige basin. Black polygons indicate the main sub-basins, the dark blue line highlights the Adige River, whereas the light blue lines represent the tributaries. The lower right inset shows the location of the Adige catchment within the Italian territory.

### heterogeneous.

In the Province of Bolzano, 55% of the catchment area is covered by forest (forests, vegetation areas and agroforestry), whereas 37% by meadows and pastures. The remaining area is subdivided as it follows: 4.3% for permanent crops (i.e., vineyards and orchards); 2% for water; 1.1% urban areas; 0.3% arable land and finally 0.4% industrial and commercial areas. In the Province of Trento, forest (mainly conifers) occupies 68.7%, while 11.5% is covered by rocks, 16.5% is used for agricultural purposes and the remaining 2.8% is constituted by urban areas [96].

The Adige river provides water to 34 large hydropower plants, with a total effective power of 650 MW [5] (Figure 2.1). Water management for hydropower production is performed with 28 reservoirs, 15 in the Province of Bolzano and 13 in the Province of Trento, with an operational storage of 560.59  $\times 10^{6}~m^{3}.$  The two largest reservoirs are Resia and Santa Giustina, with a storage respectively of  $116 \times 10^6$  m<sup>3</sup> and  $172 \times 10^6$  m<sup>3</sup>; 11 reservoirs have a storage between  $10 \times 10^6$  and  $50 \times 10^6$  m<sup>3</sup>, 11 between  $1 \times 10^6$  and  $10 \times 10^6$  m<sup>3</sup>, whereas the remaining 4 have storage smaller than  $1 \times 10^6$  m<sup>3</sup> [5]. In addition to the aforementioned large hydropower plants, about 1050 small hydropower plants are distributed within the river basin [87, 88]. We notice that aggregated values relative to hydropower production are reported differently depending on the information provided by the two managing authorities [87, 88]. In the Province of Bolzano, the mean annual hydropower production amounts to 6940 GWh, while total average nominal licensed water discharge for hydropower production in the Province of Trento is about  $404 \text{ m}^3 \text{s}^{-1}$ .

Besides the presence of hydroelectric power plants the basin is intensively exploited with a large number of small withdrawals associated to a variety of water uses: agricultural, civil and

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industrial. Their average nominal licensed water discharge is relatively low since 70% of the derivations are lower than half a liter per second, while actually 90% is lower than one litre per second. In the Province of Bolzano the second highest water demand, after hydropower, is for agriculture (i.e., irrigation), followed by industrial and other uses, and finally for drinking water supply. Similarly, in the Province of Trento, after hydropower, most of the water is used for civil uses (in this case including drinking, sanitary and zootechnic purposes), followed by agricultural uses, whereas industries ranked third.

# 2.1.1 The Careser Glacier as a case study for assessing long-term climatic variations

The Adige river is fed by many glaciers, which are carefully monitored in the catchment [5], since they strongly influence the annual flow regime and due to the increasing concern related to the observed acceleration in melting caused by rising air temperatures. Analysis of land cover changes as retrieved by CORINE (www.eea.europa.eu/publications/ COR0-landcover) products in 1990 and 2012 showed that glaciers have reduced their surface extent with a negative trend of about 2.3 km<sup>2</sup>year<sup>-1</sup>. According to the last report from the basin authority [5], glaciers cover an area of 212 km<sup>2</sup>, corresponding to 1.9% of the catchment's area at Verona. Among these, the Careser glacier represents a valuable case study because of its long-term observations.

This glacier (Figure 2.2) is located in the south-eastern side of the Ortles-Cevedale group, in the inland of Val di Pejo (north-west of Trento Province, Italy). It occupies a large, south-facing circle surrounded by peaks ranging from 3162 m a.s.l. (Cima Lago Lungo) to 3386 m a.s.l. (Cima Venezia). Its elevation ranges from 2,910 to 3,275 m a.s.l. and its area of about



1.31  $km^2$  (year 2015) is rather flat and subdivided into three main ice bodies and three smaller patches [18, 19, 109].

Figure 2.2: Geographic location of the Careser Glacier (2006).

The current aspect of this glacier is the result of significant changes caused by relevant loss of volume experienced in the last century. In fact, at the end of the 19th century its area covered about 6.8 km<sup>2</sup> and extended to 2600 m a.s.l., where in the 1920s an artificial lake (the Careser Lake) was built for hydropower production.

The glacier is fed mainly during winter by direct precipitation and wind-drifted snow. In the spring and summer seasons meltwater feeds Rio Careser, which drains about 8.5 km<sup>2</sup> into Lake Careser.

In order to monitor hydropower production, the Careser glacier and the downstream creek have been studied since the 1920s, thus a considerable number of data is available. Mass balances have been calculated since 1967 by the University of Padova up to 2001 [41, 113, 115]. Since 2002 the monitoring activity has been carried out by the Comitato Glaciologico Trentino (Societadegli Alpinisti Tridentini)[20].

### 2.2 Data

**Sampling campaigns.** Biological and chemical samplings were performed in two campaigns: the first (following referred to as 1) was held in February and the second (referred to as 2) in July 2015, in order to capture both low and high flow conditions occurring in the winter and summer seasons, respectively. Both winter and summer are tourist seasons, with the highest increase in population in the winter. Seven sites were sampled in each campaign (Figure 2.3): five along the Noce River and the remaining two along the main stem of the Adige River. Locations were selected according to the objectives of the GLOBAQUA project [79] and their main characteristics are described in table 2.1.

Water temperature, pH, dissolved oxygen (DO) and electrical conductivity were measured using an Aquatroll 200 multi-parameter probe, while turbidity was measured using a Ponsel IR optical turbidimeter. River velocity was measured using a Decatur Electronics Europe Inc. radar gun [108], except at sites 2 and 3 where mean water velocity was determined by tracer tests using bromine (in Febrary 2015) and NaCl (in July 2016). At each site, water samples were collected at 50 cm depth at three points (left, right and center of the river section) and mixed immediately after sampling. Water samples for the analysis of PhACs, PCPs and pesticides were stored in 1L gray PE bottles. Samples were transported to the laboratory in a refrigerated isothermal container and stored at -20°C until extraction and analysis. Water samples for ion analyses were



Figure 2.3: Geographic location of the sampling sites were the field campaigns were conducted.

collected in triplicate. The samples were filtered immediately through glass fiber filters (Whatman GF/F) and frozen at -20°C until analysis.

Macroinvertebrate communities were sampled using a pond net (Surber sampler, 500  $\mu$ m mesh size). Six samples were randomly collected at each site after disturbing the streambed one meter upstream of the net by kicking. Samples were preserved with 4% formaldehyde.

**Macroinvertebrates dataset.** Further biological data of aquatic macroinvertebrates were provided by the local environmental

Site	River	X UTM Coord.	Y UTM Coord.	Location	Focus		
1	Noce	648165	5143139	Bresimo	Reference site		
					Emerging		
2	Noce	623630	5124045	Tonale	micropollutants		
					(tourism)		
					Emerging		
3	Noce	639983	5131178	Mezzana	micropollutants		
					(tourism)		
					Downstream S.		
4	Noco	662113	5120547	Mezzo-	Giustina dam.		
4	NUCE	002113	5120547	corona	Water		
					abstraction.		
5	Noce	660322	5113612	Zambana	Hydro-		
J	NUCC	000322	5115012	Zambana	thermopeaking		
					Stressors from		
6	Adige	664228	5117638	Faedo	the upper part of		
					the basin		
					Emerging		
7	Adige	664351	5097306	Mattarello	micropollutants		
•	nuige		0001000	Wattareno	(tourism). Hydro-		
					thermopeaking		

Table 2.1: Main characteristics of the GLOBAQUA sampling locations.

agencies (Agenzia Provinciale per la Protezione dell'Ambiente, APPA) of Trento (APPA TN) and Bolzano (APPA BZ). Within the Adige basin, in the Trentino Alto Adige region, we analysed a total of 300 observations available from 2008 to 2015 and sampled at 75 monitoring sites. All the observations for each site didn't have a specific time pattern, most of the stations in fact had five or less samples taken in different years and in different periods of the year. Both datasets provided data identified at genera level. The classification of the ecological status is calculated according to the STAR-ICM index [16], in which five quality classes are identified: 1 (high status) to 5 (very bad status). Streamflow data. Streamflow data in the Adige River Basin, continuously registered at 58 gauging stations, have been provided by the Hydrological Offices of the Autonomous Provinces of Trento (www.floods.it) and Bolzano (www.provincia.bz.it/hydro). Some of them provide daily historical data for the last two decades, while others have been continuously operated since 1920. High-resolution data are also available with the time step of 10 minutes. For this study, sub-daily analyses were performed with values aggregated at the hourly scale.

In detail, in order to evaluate hydrological alterations caused by hydropower production we considered, where available, the gauging stations close to the sites described in table 2.1 (i.e. Noce at Vermiglio, Malé, S.Giustina and Mezzolombardo, and Adige at S. Michele all'Adige and Villa Lagarina).

Climatic variations evaluated in the Careser creek, downstream of the glacier, were investigated considering daily water balances available at the artificial lake (Enel station) from year 1990 to 2010, from which daily incoming discharges were derived. Other streamflow (Q) time-series were available upstream of the lake (Careser Baia hydrometer). The station of Careser Baia records streamflow at sub-daily intervals, and due to the winter freezing of the stream only summer data are available: from 1976 to 2002 values were taken every 15 minutes, then in 2003 the hydrometer was changed and since then discharge is given every five minutes. This dataset showed many significant gaps, thus some years were not considered in our analyses.

Finally, in the correlation analyses between hydrological alterations and biological data we used 36 gauging stations close to the biological monitoring sites (see Figure 3.15). Climatic data. Precipitation and temperature data, used for assessing the effects of climatic drivers on hydrological alterations on the Careser glacier, were provided by the meteorological offices of the Autonomous Provinces of Trento (www.meteotrentino.it). In particular, for this study, daily data of air temperature (T, since 1939) and precipitation (P, since 1929) were available at the gauging station located at the dam (2600 m a.s.l.); since 1990 both variables are registered every 15 minutes. In order to reconstruct eventual gaps we used data of the close stations of Pejo (1565 m a.s.l.) and Cogolo (1190 m a.s.l.).

**Glacial mass balances.** Long-term observations of winter accumulation ( $B_a$ ), summer ablation ( $B_s$ ) and net mass balance ( $B_n$ ) of the Careser glacier were available since 1966/1967, provided first by the University of Padova (from 1967 to 2001) and then by the Comitato Glaciologico Trentino (since 2002). Values were obtained from measurements taken with ranging rods located between 2900 m a.s.l. and 3200 m a.s.l.

**Gap filling and series reconstruction.** Gaps within the series of temperature and precipitation were filled performing Kriging with external drift [51] on the close stations of Pejo and Cogolo. Where possible, missing values of the streamflow records were reconstructed by performing autoregressive functions implemented in Matlab R2017b [2]. We excluded years with large gaps such that affected the main statistics and we reconstructed missing periods no longer than two days. The parameters of the autoregressive model were calibrated on the seven days previous the gap.

The natural regime (i.e. in the absence of water use), evaluated in the analysis of anthropogenic hydrological alterations, was reconstructed by excluding all water uses within the catchment [8].

**Biological and chemical analyses** Macroinvertebrates sampled during the field campaigns were analysed in the laboratory by the Department of Evolutionary Biology, Ecology and Environmental Sciences of the University of Barcelona. Samples were sieved through a 500- $\mu$ m mesh and macroinvertebrates were sorted, counted and identified under a dissecting microscope (Leica Stereomicroscope). Identification was at the genera or species level for nearly all groups of taxa with the exception of the Oligochaeta and Diptera, which were identified at the family level. For each site, taxonomic richness (S), Shannon diversity (H), and percentage of Ephemeroptera, Plecoptera and Trichoptera (EPT%) were determined. Moreover, in order to assess the biological status, the Extended Biotic index (IBE) (Italian biotic index, [48]) was calculated. The IBE is based on the presence of invertebrates representative of groups of varying sensitivity to pollution and number of taxa [40]. The analyses of chemical compounds were carried out by the Catalan Institute for Water Research (ICRA). An off-line solid phase extraction (SPE) preceded the determination of PhAC concentrations by ultra-high-performance liquid chromatography coupled to triple quadrupole linear ion trap tandem mass spectrometry (UHPLC-QqLIT-MS<sup>2</sup>) [44]. For PCPs, the analyses were carried out using a method based on isotope dilution and online solid phase extraction-high performance liquid chromatography-tandem mass spectrometry (on line SPE-HPLC- $MS^2$ ) [38]. Analyses of the target pesticides were performed using a method based on isotope dilution online solid phase extraction-high performance liquid chromatography-tandem mass spectrometry (SPE-LC-MS/MS)

as described in Palma et al. [81]. Nitrate, sulfate, chloride, sodium, potassium, and calcium were determined by ion chromatography (761 Compact IC, Metrohm). For further details on the laboratory analyses refer to Mandaric et al. [71].

### 2.3 Methods

# 2.3.1 Attribution of hydrological alterations within the Alpine region.

### Setting anthropogenic hydrological alterations.

The hydrological regime was charachterized by means of suitable statistical indicators of water discharge: annual mean, standard deviation and coefficient of variation (FCV), 10th, 25th, 75th and 90th quantiles (Q10, Q25, Q75 and Q90, respectively). The same statistics were also computed for the time series of streamflow (Q) increments between two successive time periods, defined as follows:  $\Delta Q = Q(t_{i+1}) - Q(t_i)$ .

### Setting climatic hydrological alterations.

*Trend Analyses* Trend analyses of precipitation, temperature and streamflow data were performed with linear regressions, moving averages and with the nonparametric Mann-Kendall test (Mk-test, [60, 72]). The MK-test is applied for detection of monotonic increasing or decreasing trends. In contrast to parametric trend tests, it does not require a priori assumptions of the underlying parameter distributions and is suitable in the analysis of multiple datasets [49]. We performed the MK-test combined with the pre-whitening method [112], which removes serial correlation prior to trend analysis to avoid erroneous trend detection due to serial correlation of the time series. We assigned a significance level  $\alpha = 0.05$  to the Kendall's p-value.

*Wavelet and cross wavelet analyses.* Wavelet Transform (WT) can be an useful tool to disentangle the dominant modes of hydrological signals and identify their change of strength in time. In fact, WT is localized in time, and thus makes possible to detect time variations in the modes of variability associated to signal unsteadiness [98].

The continuous wavelet transform  $W_n(s)$  of a discrete signal  $x_n, n = 1, 2, ..., N$ , where N is the length of the series, is defined as follows [98]:

$$W_n(s) = \sum_{n=1}^{N} x_{n'} \Psi^* \left[ \frac{(n'-n)\Delta t}{s} \right],$$
 (2.1)

where  $\Delta t$  is the sampling time step,  $\eta(n' - n)\Delta t$  is the dimensionless time and superscript \* indicates the complex conjugate.

The wavelet function  $\Psi(\eta) = (2\pi s/\Delta t)^{1/2}\Psi_0(\eta)$  is obtained by normalizing a "mother" wavelet  $\Psi_0(\eta)$ . The most commonly used "mother" wavelet for analyzing geophysical signals is the Morlet function:

$$\Psi_0(\eta) = \pi^{-1/4} e^{i\omega_0 \eta} e^{-\eta^2/2}$$
(2.2)

where we assumed  $\omega_0 = 6$  and m = 6.

The distribution of energy among the modes of variability (periods) is described by the wavelet power spectrum (WPS) which is defined as  $|W_n(s)|^2$ . Significant information on the nature of the signal can be obtained by analyzing how signal energy, which is proportional to the amplitude of the fluctuations, varies with time. This can be obtained by integrating the WPS over a desired set of scale.

The Cross Wavelet Transform (XWT) and the Wavelet Coherence (WC) are efficient tools for examining relationships in time frequency space between two variables.

The XWT of two time-series  $x_n$  and  $y_n$  is defined as  $W^{XY} = W^X W^{Y*}$ , where the superscript \* denotes complex conjugation.

In order to evaluate the phase difference between the two signals, we used the circular mean of a set of  $angles(a_i, i = 1, 2, ..., n)$ , defined as [116]:

$$a_m = arg(X, Y)$$
 with  $X = \sum_{i=1}^n cos(a_i)$  and  $Y = \sum_{i=1}^n sin(a_i)$ .  
(2.3)

 $a_m$  was defined over regions with a statistical significance higher than 5% and was then converted in time shift between the two series.

Cross wavelet power reveals areas with high common power, which is detected by the wavelet coherence (WC) that defines where, in the time frequency space, the two variables are highly related. The WC of two time-series is defined as [99]:

$$R_n^2(s) = \frac{\left|S(s^{-1}W_n^{XY}(s))\right|^2}{S(s^{-1}|W_n^X(s)|^2)S(s^{-1}|W_n^Y(s))|^2},$$
(2.4)

where *S* is a smoothing operator.

### 2.3.2 Evaluating the effects of multiple stressors on Alpine macroinvertebrates.

**Multiple Stressor Analyses.** We evaluated the impact of multiple stressors on the biological community using the data sampled during the GLOBAQUA field campaigns. Principal Component Analysis was applied to the hydrological and environmental data. Variables included in the dataset analysed by PCA were selected by means of Spearman correlation applied to paired variables. When the determination coefficient was higher than

0.90, one of the variables forming the pair was removed to avoid autocorrelation and collinearity. This resulted in the selection of the coefficient of variation of water discharge (FCV), water temperature (Temp), nitrate concentration, water conductivity (Cond), water turbidity (Turb), urban and agricultural land use percentages (% urban, % agricult), PCPs, PhACs and pesticides (Pest) as variables to be used in the PCA analysis. Variables were inspected for normality, and when necessary log-transformed using decimal logarithms. Composition and density (individual/m<sup>2</sup>) of the community were evaluated. Taxa present at less than 1% of the total density and only present at one site were excluded. Taxa densities were log-transformed to reduce the influence of extreme observations on the subsequent ordination procedure [94]. A CLUSTER analysis was performed on biological samples, applying the farthest neighbourhood method and Bray-Curtis similarity distance. In addition, Similarity Percentages (SIMPER) analysis of the biological data was performed to determine the contribution of each taxon to the average resemblances between sample groups, and to identify the predominant taxa at each site. Spearman correlations between biological parameters and environmental characteristics were also calculated. In addition, a non-parametric redundancy analysis (dbRDA) was performed to determine the correlation between taxa composition and the environmental variables. RDA is a direct ordination analysis that selects a set of variables (predictors) that best explains the variability of a biological community [12]. All the analyses were performed using PRIMER 6 (Version 6.1.6, Primer-E Ltd Plymouth UK).

**Setting the effects of anthropic hydrological alterations.** In order to have more significant results, in addition to the samples

collected during the field activities, the data provided by the local agencies (APPA TN and APPA BZ) was added in the analyses. The resulting community matrix (sites versus community composition) was first log-transformed and a resemblance matrix was calculated using Bray-Curtis Similarity Index.

With the purpose of analysing the effects of anthropogenic hydrological alterations on the aquatic community at the basin scale only those sites where hydrometric measurements were available were first considered and the Pearson's rank correlation between hydrological and biological variables was performed on the complete dataset, and then on yearly, province and river type subsets (defined below in section 3.2.2). The set of hydrological an biological variables used are described in table 3.5, section 3.2.2.

We then performed a multidimensional scaling ordination (MDS) and the analysis of similarities (ANOSIM, [25]), testing the null hypothesis that there are no assemblage differences between impacted and non-impacted sites. ANOSIM is a non-parametric multivariate analysis which detects changes in the community structure between specific sites (distinguished based on given conditions, depending on the purpose of the study); the resulting statistic test R varies between 0 and 1: if the null hypothesis is verified (i.e. no differences between sites or group of sites) R=0. We performed the one-way ANOSIM by distinguishing three types of impact: downstream the hydropower discharge restitution (Impact 1), upstream the hydropower restitution and downstream the dam (i.e. river disruption and minimum environmental flow, MEF; (Impact 2) and in the absence of anthropogenic hydrological alterations (*Impact 3*). We also tested seasonal independence for each type of impact.



**RESULTS AND DISCUSSIONS** 

# 3.1 Hydrological alterations within Alpine systems

### 3.1.1 Hydrological alterations caused by hydropower plants.

Generally, water discharge in this region is higher in summer than in winter, however there are some exceptions where the natural hydrological regime is altered by hydropower. Indeed in our analyses site 5 (Table3.1) shows this pattern, being located downstream, and at short distance from the restitution of the Mezzocorona hydropower plant.

Based on the analyses of the time series and their statistics, greater variations in discharge between summer and winter seasons were observed for small values of streamflow (i.e. the 10<sup>th</sup> and 25<sup>th</sup> quantiles, Q10, Q25) compared to high streamflows. This was due to the alterations caused by hydropower, which are particularly evident at low flow [118]. Figure 3.1 shows the streamflow (first column) and the duration curves (second

Site	1	2	3	4	5	6	7	
Annual	7 56	21.76	88.16	203.08	246 43	1135.2	1542.6	
$\mathbf{Q}_{\mathbf{max}} \left[ \mathbf{m}^3 / \mathbf{s} \right]$	1.50	21.70	00.10	203.30	240.45	1155.2	1042.0	
Month when	Mou	Sontombor	October	November	November	Juno	June	
Q <sub>max</sub> occurs	way	September	October	November	November	Julie		
Annual	0.25	0.79	5.02	5.04	12 10	68.36	115.2	
$\mathbf{Q}_{\min} [\mathbf{m}^3 / \mathbf{s}]$	0.23	0.75	5.52	5.04	42.43	00.30		
Month when	Anril	February	February	February	Anril	February	February	
Q <sub>min</sub> occurs	npin	rebruary	rebruary	rebruary	npm	rebruary	rebruary	
Annual Mean	0.50	0.14	11 10	0.71	25.7	122.6	200.0	
<b>Q</b> [ <b>m</b> <sup>3</sup> / <b>s</b> ]	0.59	0.14	11.15	9.71	55.7	155.0	209.9	
FCV [/]	1.00	1.93	0.76	0.98	0.58	0.65	0.62	
Variance [m <sup>3</sup> /s] <sup>2</sup>	0.35	0.08	73.05	90.51	434.96	7581.4	16687.9	
Q10 [m <sup>3</sup> /s]	0.25	0.05	3.87	5.06	10.63	55.46	92.8	
Q25 [m <sup>3</sup> /s]	0.32	0.06	5.25	6.54	18.22	70.75	121.63	
Q75 [m <sup>3</sup> /s]	0.63	0.12	14.81	10.34	49.25	171.18	261.64	
Q90 [m <sup>3</sup> /s]	1.09	0.16	22.77	13.99	60.81	243.92	363.53	

Table 3.1: Main hydrological characteristics and variables calculated at each sampling site. In the table AMF is the Annua Mean Flow and FCV is the Flow Coefficient of Variation.

column) at sites 3, 4 and 5. Site 4, which is located between the Mollaro reservoir and the restitution of the Mezzocorona power station, showed a general reduction in streamflow with respect to the natural regime and was not impacted by hydropeaking. Downstream from the reservoir, and before the restitution of the Mezzocorona power station, the river is fed by the constant release of about 2 m<sup>3</sup>/s from the reservoir [88] to guarantee the minimum ecological flow (MEF), which supplements the natural contribution of the residual catchment. The other sites showed no observable alterations in the duration curves with respect to the reconstructed natural streamflow. However, the streamflow record at site 5 (third row, first column) reflects the regularisation effect of the upstream reservoirs (Mollaro and S. Giustina), with a significant reduction of high flows, which was also reflected in the flow duration curve.



Figure 3.1: Streamflow time series (first column) and flow duration curves (second column) for sites 3 (first row), 4 (second row) and 5 (third row). In the first column the black lines indicate water discharge (recorded or computed using the model by Bellin et al. (2016) [8]) in the presence of utilisations and the red lines indicate the reconstructed natural water discharge (in the absence of utilisations). Similarly, in the second column the black lines indicate the flow duration curves obtained in the presence of utilisations and the red lines indicate the flow duration curves of the reconstructed natural flow regime.

The Cumulative Distribution Functions (CDFs) of daily streamflow variation (Q) at sites 4,5 and 7 are shown in Figure 3.2. All the CDFs were rather steep at Q=0, suggesting the more frequent occurrence of small or no changes in water discharge between two successive time periods. Sub-daily variations (green line) were steeper than daily changes (red line), particularly at site 7, revealing that small variations were more frequent at the sub-daily scale, as expected. Sub-daily variations are not presented for site 4 since no measurements were available at this site and streamflow was calculated using the hydrological model at the daily scale. The largest alteration in the CDF as a result of anthropogenic pressure (i.e. hydropower) was observed at site 5, with the daily variations in the natural (reconstructed) streamflow being steeper around zero with respect to the observed (altered) streamflow. For simplicity, only sites with significant differences are reported in Figure 3.2; the others showed a behaviour similar to that of site 7.



Figure 3.2: Cumulative Distribution Functions of water discharge increments  $\Delta Q$  at sites 4 a), 5 b) and 7 c). Black lines refer to the observed (recorded or computed by the model) daily streamflow, the red ones to the reconstructed natural daily streamflow and the green ones to the actual (only recorded) hourly streamflow. Note that at site 4 streamflow was obtained using the model by Bellin et al. (2016) [8], which operates at the daily time scale, and thereby hourly streamflow increments were not available.

# 3.1.2 Detection of hydrological alterations caused by climate change: the Careser Glacier case study.

**Climatic trends** In order to characterise existing climatic trends we analysed temperature and precipitation data by employing a number of derived climatic variables. Thereby, we described precipitation with number of snow and rainy days, while from temperature data we derived the potential melting period. We defined the number of rainy days as the number of days a year when precipitation, in liquid form, exceeded the value of 1 mm. In the same way, the number of snow days were counted when precipitation, in the form of snow, exceeded 1 mm and at the same time temperature was below 2° C. Then, the melting period was assumed to begin after at least seven consecutive days with an average temperature ( $T_{ave}$ ) above 0° C and to end when  $T_{ave}$ <0° C. Each variable has been evaluated yearly from 1940 to 2015.

Table 3.2 reports the main results of MK-test. Liquid precipitation and the melting period (defined in number of days) are significantly increasing over the years. In particular, the melting period shows the highest increasing rate ( $0.55 \text{ days} \cdot \text{y}^{-1}$ ), which means that snow and later ice are potentially melting for an increasingly longer period and could cause relevant reduction of glacier volume. Instead, MK-trend of the number of snow days is not significant and results almost constant over the years.

Table 3.2: Mann-Kendall (MK) test results. MK-Trend detects the increasing (positive) or decreasing (negative) trends of the variables, given in [(number of days)y<sup>-1</sup>]. Mk statistical parameter Z detects the significance of the results. For |Z|>1.95996 resulting trends are significant ( $\alpha = 0.05$ ).

	MK-Trend [-/year]	MK-stat Z
Rainy days	0.32	5.35
Snow days	0.05	1.35
<b>Melting period</b>	0.55	7.63

Figure 3.3 shows time trends of these climatic variables. In detail, the number of rainy days increased up to the middle 1960s, then slightly decreased up to 1990, when it inverted its trend and since then it is increasing again. Instead, the number of snow days shows a fluctuating behaviour, inverting repeatedly its trend. Furthermore, as previously observed, temperatures tend to keep their values above 0° C for a longer period during the year. In fact, in most years since 1990 the potential melting period extends from beginning of May to October (Figure 3.3c).



Figure 3.3: Number of rainy (a) and snow (b) days and melting period (c). The black line represents the calculated moving average over a window of 5 years, while the dashed red line is the linear regression.

**Streamflow analyses** Figure 3.4 shows the wavelet analysis performed on the daily streamflow data registered at the dam from 1990 to 2010 (Enel station). The series has some significant peaks in the ~6 months band, suggesting irregular seasonal behaviour. Besides, we observe relevant high power on the band around the annual scale (i.e. within the range of 256-512 days). However, the time evolution of annual energy, plotted on the bottom of figure 3.4, shows that the signal is decreasing its power (significant, i.e. |Z|>1.95996 for  $\alpha$ =0.05, MK-trend of -9.93 y<sup>-1</sup>, MK-test performed on aggregated annual energy normalized by its overall mean).

In order to understand more deeply these dynamics we calculated the time evolution of annual runoff volume, derived from the daily streamflow data. We aggregated and averaged the annual evolution of the obtained volumes within periods of five years and their curves were compared (figure 3.5). Significant changes in the timing of streamflow were detected, with anticipated and reduced summer runoff. In detail, we can observe that in the beginning of the 90s' the maximum runoff occurred between mid-July to mid-August, then shifted between mid-June to mid-July in the last decade of the available data, with a noticeable decrease in August. Moreover, total runoff volumes decreased from a maximum of about 6.5 millions m<sup>3</sup>, occurring in average in 1991-1995, to 5.5 millions m<sup>3</sup> in average



Figure 3.4: Wavelet transform on daily streamflow registered at the Enel station from 1990 to 2010. The figure on the top shows the results of the wavelet analysis in the time-frequency space (period in days). The plot on the bottom describes the evolution of the signal energy within the range of scales 256-512 days (i.e. around the annual scale).

for the period 2006-2010.



Figure 3.5: Summer runoff volumes. Values were averaged over periods of 5 years and then compared over the period 1991-2010.

Different pieces of information were deduced looking at sub-daily data, recorded at Careser Baia. Since this dataset provides only summer values, we performed the following analyses on a year-by-year basis, thereby by way of example we report results showing a representative case. In this regard on



Figure 3.6: On the plot on the top: sub-daily streamflow data recorded at Careser Baia. Its wavelet transform (period measured in days) is shown in the box on the bottom. The figures show a representative year (2004).

the top of figure 3.6 we observe that at a certain point of the year streamflow changes its hydrological regime and shows pronounced sub-daily fluctuations. This behaviour is clearly detected with the wavelet transform, on the bottom of figure 3.6, in the ~1 day period when the signal suddenly increases its power. Assuming that snow-cover mitigates ice melting induced by temperature, we expect that when snow completely melts the hydrological regime changes leading to impulsive and abrupt fluctuations. Thus, with the help of the wavelet transform, we detected for each year since the 70s' this turning point, and our results are summarised in figure 3.7. A general anticipation of more than a month was detected: from middle August (in 1970) to late June (in 2015). In particular since 1985 oscillations clearly anticipate at increasingly pace. However, due to anomalies in temperature and precipitation events, some years show a complicate behaviour, thus we couldn't detect any changing point in the hydrological regime. For this reason some recent

years are missing in our analysis. Furthermore, missing values in the early years are due to the presence of significant gaps in the original dataset, which prevent the correct application of the wavelet analysis.



Figure 3.7: Change of the hydrological regime. Linear regression has been evaluated overall the years (dashed red line).

At the sub-daily scale we could also investigate the timing of streamflow to temperature. Thereby, we performed the Cross Wavelet (XWT) and the Wavelet Coherence (WTC) analyses of the two variables. We averaged the values of the sub-daily datasets in order to have equal series with hourly intervals. XWT and WTC were conducted year-by-year on summer series from 1990 to 2015, and, as previously, in the figures that follow we show a representative case.

The XWT of Q and T is shown in figure 3.8a. We notice that the two variables have common features in correspondence of the abrupt oscillations of Q. In this band of high common power we notice that Q is in-phase with T (i.e. Q mirrors the behaviour of temperature). Furthermore wavelet coherence (Figure 3.8b) confirms this relationship, showing high correlation around the period of 24 hours and mainly in the regions where the two variables are in-phase.

We then calculated the phase, namely the time shift between Q and T (Figure 3.9). For most years we could observe a clear recurring behaviour in the timing of Q in response to T: in



Figure 3.8: Cross wavelet transform (XWT) (a) and wavelet coherence (WTC) (b) between Q and T. The relative phase relation is shown as arrows (in-phase pointing right, anti-phase pointing left, and T inducing Q by 90° pointing straight down). The figures show a representative year (2004).



Figure 3.9: Time shift (in hours) between Q and T. Only significant (i.e. confidence level of 5%) and positive shifts (i.e. discharge respond to temperature) are shown. As previously, the figure relates to a representative year (the same of figures 3.8 and 3.6).

summary, in the initial part of the melting season, the time shift starts relatively high and then rapidly decreases and flattens out for the rest of the summer. We identified a point after which the delay of Q flattens its trend, as if meltwater from bare ice was the only contribution to runoff, leading to the stabilisation of downstream flow. We detected this point for each year and table 3.3 compares the obtained dates with those found and showed in figure 3.7. Besides a few cases, we generally observe a good correspondence between the two estimates, with a delay of the shift change in a range of one-two weeks with respect of the hydrological change. We can assume these delays are related to the presence of remaining snow patches upstream the gauging station and to the dynamics of glacier melting and retreat (in this regard we refer to the study conducted by Rossini et al., 2018 [92]).

Table 3.3: Comparison between the date when hydrological regime changes and the moment when a change in the Q-T shift behavior was detected. Years with significant gaps or cases in which we couldn't identify a clear change are not reported.

Year	Hydrological Change	Shift Change
1996	12 Jul	25 Jul
1997	6 Aug	18 Aug
1998	17 Jul	29 Jul
1999	15 Jul	3 Aug
2004	6 Jul	9 Aug
2005	20 Jul	4 Aug
2006	14 Jun	28 Jun
2007	7 Jun	24 Jul
2010	2 Jul	22 Jul

**Hydrological changes forced by climatic drivers.** The described results evidenced that the rapid drop of mass of the Careser glacier is strictly related to climatic drivers and to the rapid shrinkage of the glacier itself.

Temperature data showed that since the 1990s the prospective melting period is getting longer, from early May to October. As a consequence timing of streamflow is anticipating and runoff peaks in June (Figure 3.5), about a month earlier than at the beginning of the 1990s, as also reported by Carturan et al., 2007 [20].

According to our assumption, when snow completely melts the hydrological regime changes leading to impulsive and abrupt oscillations of streamflow data. Our results show that this changing point is anticipating and occurs almost in late June (Figure 3.7), when we also registered the peak of runoff, as stated above.

Winter accumulation doesn't weight significantly in the annual mass balance and its contribution doesn't offset the observed changes, as also stated in earlier studies [20, 114]. Thereby, our results are related to the combined effects of the prolonged rise in temperature and the less ice surface. According to Bliss et al. (2014) [11] as glaciers melt at a faster rate in response to climate warming, meltwater provides larger contribution to downstream runoff, which however is then affected by a reduction ensuing glaciers decline [58]. In our case runoff fastened and increased since the beginning of the 1990s, and then has decreased since 2000, as less glacier volume contributes to its formation. In this regard figure 3.10 shows the decline of the glacial volume.



Figure 3.10: Loss of volume from the glacier mass (in million  $m^3$ ). Dashed red line represents the moving average within a time window of 9 years. Data have been taken from mass balances measured and calculated first by the University of Padua, then by Comitato Glaciologico Trentino.

As most Alpine glaciers [83], the Careser glacier has lost most of its volume starting from the early 1980s. Then, at the end of the 80s this trend stabilised over the years, keeping values almost constant, with few exceptions. Starting from 2000 mass losses decreased, as an effect of the significant reduction of glacier surface. Furthermore, according to Carturan et al. (2013) [18] thinning of the Careser glacier strongly accelerated starting from 1980, with mass dropping rates of about -1.3 m w.e.  $y^{-1}$ , resulting in bedrock emergence and fragmentation of the ice body in smaller patches. Consequently, downstream hydrological regime suffered significant alterations and ice meltwater contributed earlier to downstream runoff since the middle 1980s. However, in the last years the climate forcing seems to be overcome by the lack of ice surface, and in some years we couldn't detect a clear turning point in the hydrological regime, even though temperature was above 0°C for almost all the period (Figure 3.11).



Figure 3.11: Summer streamflow data at the Careser Baia recorded in the years 2008 (a), 2010 (b) and 2014 (c).

# 3.2 The effects of hydrological and chemical stressors on macroinvertebrate community.

### 3.2.1 Multiple stressors on Alpine freshwater ecosystems.

**Physical, chemical and biological parameters within the Adige basin: results of the field campaigns** Results of the laboratory analyses of the samples taken during the field campaigns are reported in Table 3.4. The concentrations of PCPs and PhACs detected during the two sampling campaigns were reported in a recent paper [71].



Figure 3.12: PCA analysis for environmental variables. Concentrations are represented with symbols and are labeled with two numbers, the first referring to the site and the second to the season, with 1 indicating winter (orange points) and 2 summer (green points, for example 3.2 indicates the sample taken at site 3 in the summer campaign).

The result of the PCA analysis for the hydrological and chemical data is shown in Figure 3.12. The first two components explain a total variance of 54.8%. The first axis (abscissa) was positively correlated with the coefficient of variation of streamflow, temperature, turbidity, PCP concentration and agricultural land uses. Summer samples at sites 6 and 7 showed the highest correlations. PhACs and nitrate were on the negative side of this axis, as were winter samples at site 2. Axis 2 (ordinate) showed a positive correlation with conductivity, pesticides, and urban and agricultural land uses. Winter samples at site 7 showed the highest positive correlation and concentrations observed in the summer at sites 1 and 3 were on the negative side. Most of the sites (2, 3, 4, 6, 7) moved downwards in the PCA in the summer sampling, reflecting a reduction in the concentration of most

chemical compounds and higher river discharge. Axis 3 (not represented) explained 77.3% of the total variance and confirmed the strength of the correlation between nitrate and PhACs and site 2 in winter on one side; and PCP and pesticides and site 7 on the other. As shown by Mandaric et al. (2016) [71], the joint effect of low streamflow and higher tourist numbers during winter resulted in an overall higher concentration of PPCPs (pharmaceuticals and personal care products). The concentrations of pesticides were also higher in winter although they are applied to crops in spring-summer. In general, higher water discharge in summer caused a global reduction of all pollutants due to higher dilution. A recently published national report on the levels of pesticides in samples collected in 2013-2014 [56] confirmed the diffusion of these pollutants into the river in the province of Trento. Of the 33 substances analysed, boscalid, dimetomorf, fluopicolide and chlorpyrifos were the most frequently found in surface waters.

For what concerns sampled macroinvertebrates, in both field campaigns, the highest species richness was observed at site 1 and the lowest at site 3. At all sites richness was significantly lower in the summer with respect to the winter sampling campaign (Table 3.4). The Shannon diversity index ranged from 1.3 to 2.4 and the most obvious decrease between winter and summer was observed at sites 1 and 6.

#### Effects of multiple stressors on the macroinvertebrate community.

Higher richness and diversity relative to other upstream sites were observed at site 4 in both samplings. This site, located between the Mollaro reservoir and the restitution of the Mezzocorona hydropower plant, is affected by a significant alteration in the natural streamflow, since the reservoir discharges a constant amount of water without any seasonal

IBE Classificati	Diversity Shannon (I	Species Richness (S	Pest	РСР	PhACs	CI	<b>SO4</b>	NO3	Turb	CE	Temp	
on IBE		<u> </u>										
12 Class I	2.3	35	3.4	33.1	447.8	2.4	15.4	2.1	0.01	67.0	1.3	1.1
12 Class I	2.0	29	ω	993.9	10051.3	13.0	13.5	17.9	3.5	77.3	4.1	2.1
10 Class I	1.8	19	22.5	350.8	2313.8	6.01	27.6	4.4	2.15	87.7	3.9	3.1
10 Class I	2.0	30	17.6	2417.2	938.7	6.1	28.2	4.4	3.5	201.6	6.4	4.1
8 Class II	1.3	24	13.4	501.8	1283.4	3.82	8.10	3.0	6.2	202.5	5.7	5.1
11 Class I	2.4	26	6.6	44.6	1443.6	6.5	38.2	3.8	2.6	182	5.8	6.1
11 Class I	1.4	26	33.6	553.06	6014.77	4.8	21.9	2.9	3.15	231.5	7.7	7.1
10 Class I	1.7	20	2.1	43.1	291.04	1.1	34.5	0.4	4.6	125	13.7	1.2
9 Class II	1.65	15	5.1	270.4	3157.0	1.9	9.8	2.7	2.7	132	12.8	2.2
9 Class II	2.2	14	ω	175.1	417.7	1.3	25.8	1.2	62.5	68.4	11.4	3.2
10 Class I	2.3	18	9.7	168.4	292.6	1.37	12.7	2.1	4.0	180	14.7	4.2
9 Class II	2.2	15	25.8	51.9	343.2	1.4	12.9	2.3	4.2	173	13.7	5.2
9 Class II	1.8	15	4.7	208.4	434.5	2.6	32.9	1.7	70	160	15.2	6.2
9 Class II	1.6	16	10.7	11549.9	1263.6	3.3	33.2	1.9	172	72.5	15.7	7.2

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modulation, but is not affected by hydropeaking, which instead impacts site 5. In addition, the constant release of water reduces seasonal temperature variations (the release causes warming in winter and cooling in summer) that may favour the presence of some species (Maiolini et al., 2007). Accordingly, we found higher densities of some taxa, such as Baetis, Simuliidae, Chironomidae and some species of Coleoptera, Thichoptera, and Gasteropoda, while other species (e.g. Capnia sp. and Capnioneura sp.) that are adapted to colder waters were less abundant.

The distribution of sites according to their ecological similarities based on taxonomic composition and density was determined by CLUSTER analysis (Figure 3.13). This analysis revealed four distinct groups of sites. Samples from the first sampling campaign were grouped together, showing high similarity in invertebrate composition and density with respect to the second sampling campaign. In this group, in particular at sites 2, 3 and 4, the taxa that contributed the most to the similarity were Baetis and Chironomidae, Simuliidae and Limoniidae (these taxa accounted for 46% of the total sample, SIMPER). Sites 5, 6 and 7 were mainly characterized by the presence of Gammarus, Baetis, Elmidae and Chironomidae (these taxa accounted for 48% of the total sample). Site 1 differed in community composition with respect to the other sites, with some taxa of Coleoptera and Trichoptera that were present only at this site, which is the least impacted by human activities. The second sampling campaign was representative of the other groups. Only sites 2 and 3 in the upper Noce and 6 and 7 in the main stem of the Adige showed similarities higher than 60%. The taxa that contributed most to the similarities at sites 2 and 3 were Chironomidae, Oligochaeta and Ephemeroptera such as Rhithrogena sp. and Ecdyonurus sp., and Plecoptera such as Capnia sp. and Capnioneura sp. (these

taxa accounted for 57% of the total contribution to similarity). Chironomidae, Psychomyiidae, Baetis and Gammarus (65% of the total contribution) dominated at sites 6 and 7.



Figure 3.13: Dendrogram (Cluster analysis) showing similarity in community composition. The two-digit numbers indicate the site and the season, respectively, with 1 for winter (indicated in red) and 2 for summer (black labels, for example 3.2 indicates the sample taken at site 3 in the summer campaign).

The dbRDA analysis helped explore the relationship between the composition and density of the invertebrate community and environmental characteristics (Figure 3.14). The first principal component separated most of the headwater sites (on the right) from low water (on the left) sites. Only sites 4 and 5 showed a different correlation with this axis according to the sampling period. Lower conductivity, turbidity, flow variability, and pesticide pollution were observed in headwaters. These sites (from 1 to 3) were characterized by a higher number of taxa with the presence of Plecoptera and Trichoptera, which were the taxa that are most sensitive to pollution among those detected in the two sampling campaigns. In summer, the most abundant taxa at sites 4 and 5 were Coleoptera (Helodidae sp.) and Ephemeroptera (Serratella sp.). Higher numbers of Gammarus sp., Hirudinea, Psychomyia sp., Hydropsyche sp., Baetis sp., and

the Dipteran families Chironomidae and Simulidae were present at the downstream sites, which are characterized by a higher percentage of agricultural and urban land uses and a higher concentration of some of the related pollutants: pesticides and PCPs. In addition, at sites 6 and 7, the hydrological indicator included in the analyses (i.e. the coefficient of variation of the daily water discharge) was positively correlated with the presence of Gammarus sp., Hirudinea, and Psychomyia sp (Figure 3.14b). A clear seasonal pattern in the composition of the biological community was indicated by axis 2. In particular, most sites occupied the upper part of the graph in summer and were characterized by poorer community composition (less taxa) compared with the winter sampling (located in the lower portion of the axis). This axis was positively correlated with water temperature and negatively with PhAcs and nitrate concentrations, which were both higher in winter at site 2.



dbRDA1 (31.5% of fitted. 26% of total variation)

Figure 3.14: dbRDA analysis between biological (a) and environmental (b) variables. Winter sampling is indicated in red and summer sampling in black.

Our results show that some pollution and hydrological alterations could explain changes in invertebrate richness, diversity and composition. The highest species richness was detected at site 1 in both sampling campaigns, while a gradual decrease was observed at sites 2 and 3, corresponding to the absence of several sensitive species (Plecoptera, Trichoptera and Coleoptera groups) and an increase in other taxa (e.g. Chironomidae). These changes related to human perturbation at headwaters have been observed in previous studies in other Alpine rivers [66]. Discharge from the WWTP just upstream from site 2 increased the nutrient and urban contaminant concentrations (mainly PhACs), however the biotic index (IBE) was unable to detect any changes in community composition at this site, with respect to reference site 1. This confirms some of the limitations of biotic indexes described in other studies [26].

The macroinvertebrate community changed downstream from site 5, with other taxa dominating as evidenced by the cluster and multivariate analyses (Figures 3.13 and 3.14). Moreover, streamflow alteration due to hydropower seemed to have an effect on community composition at site 5, which is the site most affected by hydropeaking. Here the abundance of some species (e.g. Gammarus) increased while others (i.e., Baetis and Dipterans) declined, and both richness and diversity also decreased. The density of Gammarus was significantly correlated with the coefficient of variation of streamflow (Spearman coefficient = 0.60, p<0.05) while diversity was negatively correlated with this coefficient (Spearman coefficient = -0.40, p<0.05). Because of its ability to enter into the sediment for refuge [33], Gammarus has an advantage, with respect to other species, in tolerating rapid and periodic changes in the river stage due to the intermittency of hydropower production [77].

The present results also evidence the seasonality in invertebrate community composition, as also documented in literature [36]. The two samplings were clearly separated in the cluster and

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dbRDA analyses according to community composition and density. A general decrease in richness and abundance was observed in the summer season, although some taxa (e.g. Serratella and Helodidae) showed higher densities in that period. As suggested by Ward (1994) [104], such a change is related to the change in temperature that constitutes an upper limit for many species and a lower limit for many others. Moreover, as observed in studies conducted in other high mountain rivers [69, 76] changes in community composition in the Adige River were likely related to species life cycle. Pollution is diluted in the Adige River due to higher river discharge in summer; however, hydropeaking effects are constant. This means that in summer the invertebrate community will respond more to its life cycles and at some sites to hydropeaking, with pollution being less important than in winter.

## 3.2.2 Assessing the impact of anthropogenic hydrological alterations on macroinvertebrate biodiversity.

**Correlation analysis** As afore introduced, it could be helpful finding a simple relation that links hydrological alterations to biological status in order to better define some guide lines about the efficiency and sustainability of the hydropower production, still debated from an environmental point of view. Thereby, our first step was to perform simple Pearson's correlations between the two type of indicators.

*Definition of the hydrological and biological variables.* In order to relate hydrological alterations to macroinvertebrates community we used APPA data and GLOBAQUA samples and considered only those sites where we had streamflow records. Thereby we performed the correlation analysis on a reduced dataset of 36 sites (Figure 3.15) with a total of 134 observations. Of those, 14 stations are in the Province of Trento (44 observations) and the remaining 22 in the Province of Bolzano (90 observations).

We performed the Pearson's correlation on the complete dataset, then on subsets by year, by Province (Trento and Bolzano, complete and yearly data) and by the type of river (glacial or surface flow). The latter was determined according to the Environmental Monitoring Plan provided by the local agencies APPA [1], taking into account also the distance from the source. In this work, according also on the type and the number of data available, we distinguished four subdivisions:

**Gl:** *glacial* distant up to 75 *km* from the source (in order to have a significant number of data to perform the analyses);

**ss small:** *surface flow* reaches far from the sources up to *25 km*;

**ss medium:** *surface flow* with a distance to the source between *25* and *75 km*;

**ss big:** *surface flow* reaches distant *more than 75 km* from the source.

Based on the results discussed in section 3.1.1, we chose a set of suitable variables in order to describe hydrological alterations caused by hydropower release. These variables have been selected accordingly to the dates and months of the biological samples. Streamflow variations have been smoothed at the weekly scale (by considering the moving average) to consider also the management operated by the hydropower producers, which usually follows weekly cycles.

Macroinvertebrate community was described by the richness, evaluated ad the genus level, and by the Shannon's index of diversity.



Figure 3.15: Spatial distribution of the biological sites used for the correlation analysis.

All the hydrological and biological variables adopted in the following analyses are described in table 3.5.

Correlation between hydrological and biological variables. Figure 3.16 summarises the correlations between all the variables described for the complete dataset. Since we observed that many variables were distributed in a cone-like shape, we first verified whether our data was affected by heteroscedasticity by running the Engle' s test, implemented with the function available for Matlab R2017b, which resulted verified (i.e. there isn't conditional heteroscedasticity). Looking at the values of the Pearson's coefficients, the Shannon's index of diversity and the genera richness are strongly dependent, showing high correlation ( $R^2$ =0.87). Similarly,

Variable	Definition
$\frac{Q_{m,avg}}{[m^3/s]}$	monthly mean discharge.
Q [m <sup>3</sup> /s]	discharge value at the date of the biological observation.
$\Delta Q \ [m^3/s]$	we evaluated the differences between the maximum and
	minimum values taken in a moving window of 7 days.
	Then, we considered the monthly maximum, evaluated
	at the same month of the biological obsevation.
$\delta Q [m^3/s]$	as above, but the monthly average was considered.
Н	Shannon's index of diversity, calculated over the total
	abundance for each genus sampled.
S	number of genera sampled.

Table 3.5: Set of hydrological and biological variables adopted in the correlation analysis.

normalized maximum and average streamflow alterations (called dq and DQ) are significantly related ( $R^2$ =0.66). Whereas streamflow observed at the sampling date (Q) is poorly related to the other hydrological variables. Generally, we observed low correlation between hydrological and biological variables. The average streamflow alteration is the most positively related to the ecological indicators, namely as the alteration is higher the richness and diversity of the macroinvertebrates community increase, though  $R^2$  has still low values.

Similar results have been obtained within subsets of each type of river (Figure 3.17). Higher appreciable positive values of the correlation coefficient have been observed between the hydrological variations (DQ and dq) and the biological indicators in the cases of small and medium surface flow rivers, though yet their relationship remains low. Glacial and larger surface flow rivers show very low and spread interactions. Indeed, due to their stressful environmental conditions, glacier-fed rivers host extremely simplified communities, highly sensitive to environmental changes [15]. Only few species highly



Figure 3.16: Correlation matrix between hydrological and biological variables. Correlation coefficient: Pearson. Q: dimensionless discharge; DQ: dimensionless maximum discharge alteration; dq: dimensionless average discharge alteration; S = genus richness; H = Shannon diversity. All the hydrological variables have been normalised by the corresponding monthly long term average of streamflow. The Pearson's coefficient is indicated within each box, red values indicate the most relevant bio-hydrological interactions.

specialised to survive cold temperatures, high turbidity, low channel stability, high discharge and low amounts of nutrients colonise these environments [67]. Whereas downstream large rivers are affected by the presence of agricultural and urban areas, therefore hydrological variations jointly interact with



other environmental and physical dynamics and their effects are scarcely discernible.

Figure 3.17: Correlation matrices between hydrological and biological variables distinguished by type of rivers. Correlation coefficient: Pearson. Q: dimensionless discharge; DQ: dimensionless maximum discharge alteration; dq: dimensionless average discharge alteration; S = genus richness; H = Shannon diversity. All the hydrological variables have been normalised by the corresponding monthly long term average of streamflow. The Pearson's coefficient is indicated within each box, red values indicate the most relevant bio-hydrological interactions.

We then subdivided the data by year and verified if we could detect some pattern. Figure 3.18 shows as representative case the annual pattern of correlation coefficients between the maximum hydrological variation (DQ) and the biological diversity (H) for the complete dataset (Figure 3.18a) and the subsets of Trento (Figure 3.18b) and Bolzano (Figure 3.18c). Observations have been provided for a small period, from 2008 to 2015, and our results didn't show relevant trends, even though it could seem different for the subset of Trento, in which R<sup>2</sup> decreases and inverts, however given the limited number of years, this result cannot yet be considered relevant.



Figure 3.18: Annual trends of the Pearson's correlation coefficients for the complete dataset a) and for Trento b) and Bolzano c) data.

**Analysis of similarity** Given the outcomes of the correlations analyses, our hypothesis moved on finding wether there exists independence between impacted and non-impacted sites at the catchment scale. If our null hypothesis is verified, then we cannot confirm that hydropower significantly affects the macroinvertebrates community.

We performed the ANOSIM analysis on the complete dataset and on each province subset (Trento and Bolzano). At this scale the presence of other type of stressors as well of other environmental variables can significantly influence our data, however we didn't have sufficient information to detect where these pressures affected most macroinvertebrates. In this sense, we decided to make a distinction based on the ecological status defined by the STAR-ICMi classification. We first selected those sites classified in high and good status, then we held only those in high status, where we had enough data. We also subdivided the complete dataset by the type of river, according to the classification described in section 3.2.2. In all these cases we compared the biological community distinguishing by the type of impact. Thus, we defined:

**Impact 1:** downstream the restitution of the hydropower plant;

**Impact 2:** upstream the restitution and downstream the dam (effect of river disruption and of the minimum environmental flow, MEF);

**Impact 3:** absence of hydrological alteration caused by hydropower release.

We also verified the influence of seasonality and performed the ANOSIM distinguishing between winter/autumn and spring/summer. In this case we couldn't verify each month or season since the number of data wasn't sufficient for this level of detail.

Table 3.6 summarises the results obtained for all the cases just described. Seasonality on each type of impact doesn't influence significantly our data and in addition Impact 2 has a low significance (p>0.05) because of the small number of data available to analyse in this case. These results could be expected for Impact 1, where flow alterations counteract seasonal variability, however, the independence found in natural conditions could be explained by the number and type of observations we had available, since, for each station, measurements hadn't a specific time pattern, as also mentioned in section 2.2. Moving the attention to the results obtained by distinguishing the type of impact, if we include in our analysis all the observations available, we found that the hydropower impact doesn't affect on biological community, even if we look at the single provinces (Trento and Bolzano). However, hydrological alterations become relevant if we exclude the sites where ecological quality isn't sufficiently good, and this is more evident looking at the datasets of each Province. Thus we can assume that, by excluding other type of stressors, according to our assumptions, the composition of the invertebrates assemblages are affected by the presence of hydropower plants, however this dependence isn't detected by the indicator of ecological quality.

These results have been also observed with the MDS ordination analysis, shown in figure 3.19. Sites with good ecological status have been reported in the first column, while in the second column we analysed only the sites with high ecological status. However, we have to note that Impact 2 hasn't sufficient data and results no more detectable or relevant. Furthermore, the dataset of Trento reduces significantly, thus in this case we assume more reliable the results obtained including sites in good status. The dataset of Bolzano shows clear distinction between downstream impacted (Impact 1) and unaltered (Impact 3) sites, either in the case where only sites in high status were considered and also including the stations where ecological quality is good. Looking at the complete dataset, as we considered better ecological quality we observed that the community of the impacted sites deviated from those unaltered, as also detected with the ANOSIM R coefficient (R=0.399 in the high status case and R=0.208 in the good case, see Table 3.6).

In order to investigate how different environments affect our analyses, we performed the test on each type of river as classified in section 3.2.2, distinguishing again the type of Table 3.6: Results of the ANOSIM analysis. In the top table the influence of seasonality and type of river have been investigated. Gl: glacial, glacier-fed rivers; SS2: ss medium, medium surface flow rivers; SS3: ss big, large surface flow rivers; sites within small surface flow rivers (ss small) presented only one type of impact so this case is not reported. In the second table ANOSIM results for the complete, Trento and Bolzano datasets. All: all the sites; Good: high and good status; High: only high ecological status. R: statistic test coefficient; N.obs: number of observations; significance level p< $\alpha$ =0.05. Seasonality on each type of impact doesn't influence significantly our data. Hydrological alterations become relevant if we exclude the sites where ecological quality isn't sufficiently good, and this is more evident looking at the datasets of each Province. Thus we can assume that, by excluding other type of stressors, according to our assumptions, the composition of the invertebrates assemblages are affected by the presence of hydropower plants, however this dependence isn't detected by the indicator of ecological quality.

		Type of River				
	Impact 1	Impact 2	Impact 3	Gl	SS2	SS3
R	0.102	0.028	0.066	0.158	0.384	0.138
р	0.043	0.382	0.002	0.001	0.002	0.239
N.obs	60	13	135	52	62	15

	Complete dataset			Trento Province			Bolzano Province		
	All	Good	High	All	Good	High	All	Good	High
R	0.077	0.208	0.399	0.061	0.218	0.545	0.194	0.43	0.49
р	0.005	0.001	0.001	0.067	0.001	0.001	0.002	0.001	0.001
N.obs	300	208	107	198	115	38	102	90	69

impact and keeping those sites where the ecological quality was at least good, for the reasons already mentioned. We obtained relevant results for medium surface flow rivers (called SS2 in table 3.6), in accordance with the Pearson's coefficient in the previous paragraph. Whereas observations available along SS3 rivers (or ss big, as previously defined) weren't significantly sufficient for our purpose. We neglected to report results for small surface flow rivers as there were only sites unaltered, thus the ANOSIM by type of impact couldn't be performed. Despite



Figure 3.19: MDS plots for the complete (first row), Bolzano (second row) and Trento (third row) datasets. In the first column the good case is shown, while results obtained considering only sites in high ecological status are in the second column.

the small number of observations available, we deduced that within the glacial rivers hydropower impact seems to affect less the community, which is more impacted going downstream in medium rivers fed by surface flow (SS2). Still, we precise that to better appreciate and distinguish environmental effects we need a larger number of observations and information.

# CHAPTER

#### **CONCLUSIONS**

This PhD thesis explores the effects of multiple stressors on biological community limited to benthic invertebrates fauna within an Alpine catchment, the Adige river basin, and in particular it focuses on their response to hydrological alterations. The latter have been distinguished between those forced by climate change and those caused by the presence of hydropower plants and have been analysed in detail. The main objective at the base of this work was to identify if it is possible to draw a general relationship between flow alteration and ecological responses. Specifically, in this work we 1) analysed hydrological responses caused by anthropogenic pressure (i.e. hydropower restitution) and climate change, this last has been evaluated for the Careser glacier case study, which holds long-term observations of climatic variables, mass balances and streamflow); 2) explored in general the effects on macroinvertebrates of multiple stressors present within the Adige catchment and then focused on the interaction between anthropogenic hydrological alterations and biological response,

both quantitatively and qualitatively and at the basin scale. In order to investigate hydrological variations we considered daily and sub-daily streamflow data and in the case of anthropogenic alterations we compared altered and "unaltered" (i.e. in the absence of water uses) conditions, both related to streamflow values and its variations. Whereas hydro-climatic changes, evaluated in the Careser glacier, were detected by performing a comprehensive analysis of multi-scale variations of precipitation, temperature, water discharge and glacier mass. Therefore we analysed downstream hydrological variations associated to climatic drivers and we assessed the effects detected over the long-term period. Summarising, it was found that:

- Hydropower plants affect low discharges. Daily and sub-daily comparison shows some differences in low values and in general the most affected by hydropower operations. Discharge fluctuation show differences between daily and sub-daily scales. However the most impacted sites have relevant gaps also at daily scales. In general we have effects on low jumps;
- Hydrological changes detected downstream of the Careser glacier were strongly related to the observed climatic variables. In particular, timing of streamflow is anticipating as a consequence of the longer melting period. As the glacier melts at a faster rate in response to climate warming, meltwater provides larger contribution to downstream runoff, which however is then affected by a reduction ensuing glaciers decline. In addition winter accumulation doesn't weight significantly in the annual mass balance and its contribution doesn't offset the observed changes. Sub-daily changes revealed that ice

fragmentation and disappearance is significantly affecting the hydrological regime and streamflow is mainly receiving water in summer from snow-melting and rain events, whereas ice meltwater contributes decreasingly.

As second step, we generally described how multiple stressors present within the Adige river basin affect freshwater ecosystems referred to macroinvertebrates. Then, we explored in detail their responses to hydrological alterations caused by hydropower plants and compared a set of hydrological variables to biological indicators defined by genera richness and diversity. Thereby we investigated the composition of the benthic community in relation to altered and unaltered conditions. In general we concluded that:

- richness, diversity and composition of the macroinvertebrate community responded to changes in the main stressors along the river. The inputs from WWTPs (already detected in headwaters) and a general increase in pollution downstream had more influence in winter when river discharge was lower. Water flow variability due to hydropower operation seemed to favour some taxa (e.g. *Gammarus*) at sites located downstream from the restitution of a large hydropower plant.
- with the data available we couldn't detect a general relationship between flow alterations and benthic macroinvertebrates. However changes in the composition of the impacted communities have been detected with respect to unaltered conditions, denoting their sensitivity to hydrological variations related to hydropower restitution, though robust statistical relationships are not supported yet. Hereunder we intend to discuss these

results in relation to existing literature and the European directive.

### 4.1 Interaction between hydrological alterations and benthic macroinvertebrates: conclusions and challenges

In light of our results, a general relationship that links anthropogenic hydrological alterations to available biological indicators is yet missing. Richness and diversity have not clear and unambiguous responses to hydrological variables despite the altered community seems to differ from that observed in sites unaltered by hydropower release. A recent review [86] highlights that to date in literature no clear patterns emerge for responses of macroinvertebrates or riparian species to changes in flow magnitude, even though numerous case studies (and expert knowledge) document that flow alterations (defined with many variables, e.g. magnitude, frequency, and timing) induce a variety of ecological responses [17, 86]. Given that alteration of flow regimes is typically confounded with other environmental factors, we would not necessarily expect unambiguous relationships between single measures of flow alteration and ecological response, and accordingly it has been suggested an integration of a range of other factors (e.g. channel morphology and anthropogenic modification of in-stream habitat) to hydrological variables [111]. Furthermore, in order to span a broader range of responses it has been proposed the use of larger datasets [68].

Given these considerations, some critical points have been found in our analyses. We focused on hydrological variables and neglected other environmental and geomorphological variables, which are difficult to acquire at the catchment scale and related to our data, besides their inclusion defeats the purpose to find a clear and simple relationship between flow alteration and biological indicators. Furthermore, the network of streamflow gauging stations doesn't cover the broader biological dataset, thus targeted analyses on the dynamics upstream (assumed unaltered) and downstream the hydropower plants were partially incomplete.

Some gaps have been also noticed in the biological monitoring network, in particular downstream dams, in order to explore the effects of river disruption and of the minimum environmental flow, which wasn't significantly detected in this study. Additionally we suggest the need to integrate further investigating sites targeted to clarify the distinction between altered and unaltered flow conditions taking in account comparable environmental characteristics (e.g. according to the classification of the type of impacts mentioned in section 3.2.2). In fact, one approach suggested in literature is to design sampling programs that target existing flow-altered sites across gradients of flow alteration to allow general relationships between flow alteration and ecological response to be inferred [3, 85, 86]. Often, monitoring of sites that have experienced flow alteration is generally not done and so learning opportunities are missed [106]. On these bases, further analyses should be encouraged considering a larger spatial scale (e.g. covering the Alpine scale), according to existing data and improved monitoring network.

In our results it has also been observed that biotic indices doesn't reflect the observed changes in composition of the benthic community related to hydrological impacts. The lack of significant correlation could be related to the high taxonomic level adopted in the classification by the local agencies, in accordance with the Italian legislation. Major efforts on

investigating how taxonomy influences these results may be of interest in assessing environmental risks related to hydropower management and could confirm the importance of taxonomy for biodiversity and environmental conservation [97]. Indeed, the efficiency of the biotic indices currently adopted in accordance with the Water Framework Directive (WFD 200/60/EC, [32]) is being widely discussed in many studies [37, 102]. For what concerns the STAR\_ICMi index, sources of uncertainty are diverse and hard to disentangle and have been boiled down to: ecological, errors and sampling related causes [27, 28, 95]; additionally, a recent study verifies that the index is affected by subsambling, namely it is dependent by the number of individuals sampled, undermining the correct estimation of the ecological status [95], jointly to the lack of a measure of uncertainty [37, 95]. These concepts are included in a more general framework of criticism which is being moved toward some aspects of the European Water Directive, in particular concerning the concept of reference conditions [14, 59, 78, 102], which has been even defined a "mirage" [37], as such conditions hardly exist and also underestimate the continuously evolving nature of environmental systems, not considering the long-term interactions between human and natural systems.

Lastly, for what concerns hydrological changes caused by climatic drivers, understanding how these alterations affect the ecosystem status represents a future challenge, emerged recently with respect to WFD. Indeed, the European water directive does not explicitly mention risks posed by climate change to the achievement of its environmental objectives [110], and researches based on ensuing aquatic responses are moving in this direction lately [50, 57, 64]. In this regard, an overall lack of long-term ecological studies on the Southern Alps has been observed [64]. Accordingly, the availability of targeted

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monitoring activities is still missing, and growing concerns are arising due to the rapid changes that glaciers and their downstream systems are experiencing, as have been highlighted in this study for what concerns hydrological changes, thereby the availability of precious data is seriously undermined.

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Freshwater ecosystems are severely undergoing degradation due to the presence of multiple stressors that are undermining their biodiversity. In this sense, quantifying these effects on Alpine regions is challenging, due to the lack of tailored field measurements of hydrological, biological and chemical variables. This work aims to touch some of these aspects, with particular attention to hydrological dynamics and their effects on macroinvertebrates.

Available literature confirmed that hydrological alteration is one of the most important factors affecting riverine ecosystems. In Alpine regions, most of these alterations are due to hydropower that represents the major source of energy in the Trentino-Alto Adige region. Changing climate is an additional stressor that can enhance the effects of these anthropogenic influences.

The main purpose of this study was finding a direct relationship linking biological indicators to streamflow variations related mainly to hydropower operations. Quantifying these effects is challenging due to the fact that the behaviour by which macroinvertebrates respond remains largely unexplained. However, analyses of similarities and independence, performed at the basin scale with data provided by the local Environmental Protection Agencies, showed evident differences in the biological communities between impacted and non-impacted sites. These results bring us to believe that a relationship between biological data and hydrological alteration is expected to exist, but that is not clearly explicated by simple correlations. Giving a quantitative interpretation of this correlation could help hydropower managers to improve and optimize the energy production with a more realistic scenario of the effects on the biological community, with also a perspective of the combined effects caused by the presence of multiple reservoirs within the basin.

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