EFFECTS OF STAGNATION ON CRUSTACEANS IN SANDY LOWLAND STREAMS

Bachelor Thesis
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Major: International land and water management

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Preface:

This report, *effects of stagnation on Crustaceans in sandy lowland streams*, was written as a Bachelor thesis report for Van Hall Larenstein University of Applied sciences. It was produced under the supporting of Dr. Anna Besse-Lototskaya and Agata van Oosten-Siedlecka from Department of Freshwater Ecology, Centre for Ecosystem Studies, Alterra. The report seeks to develop a better understanding of the influence of decreasing water flow for common Crustaceans that live in sandy lowland streams. This study used an artificial stagnation condition to examine variation of stream living Crustaceans.

I believe this study will make a substantial contribution to developing an understanding of the impact of stagnation on common Crustaceans in sandy lowland streams in the Netherlands. It provides a basic analysis and it enables readers to comprehend the changes of Crustaceans under stagnation condition.

I would like to thank my supervisor, Dr. Anna Besse-Lototskaya and Agata van Oosten-Siedlecka for the valuable advice and support they has given me in the writing of this report. I would also like to thank my teachers, Mr. Hans van den Dool for his encouragement and guidance. Very special thanks to Dorine Dekkers for her helping on lab experiment. My deepest thanks go to my parents, for their love, understanding and support.

Signature:______________________
Summary

Crustaceans that are very common in streams were often used as the testing of water quality standards. Therefore, it is effective to select Crustaceans as an indicator to examine the influence of decreasing water flow. Climate change induced the extremely dry summer in the Netherlands. Water flow decrease dramatically and water temperature, water quality and dissolved oxygen concentration are changed due to the decreasing water flow. Thus, the Crustaceans which live in the stream will be affected. The effect of stagnation on the Crustaceans assemblages from two sandy lowland streams in the Netherlands (1 high nutrient stream, 1 low nutrient stream) was measured on summer time of 2011. From above, the effect of stagnation on Crustaceans is worth to study as a Bachelor thesis.

In order to explain the influences of stagnation on Crustaceans, field experiment and laboratory experiment were carried out during the summer 2011. In the experiment, Crustaceans samples and water samples were taken once a week or once two weeks. Meanwhile, water level and dissolved oxygen concentration were continuously measured. Thus, the relation between water quality, water temperature and dissolved oxygen concentration and Crustaceans was very clear by comparing the variation of them.

According to the result, the response of Crustaceans for Water temperature, pH and EC that was influenced by water flow was not significant due to the narrow variation range of them. However, Crustaceans was correlated with dissolved oxygen. The composition of Crustaceans in high nutrient stream and low nutrient stream is marked different.
CONTENT

List of Figures ............................................................................................................. I

List of Tables ............................................................................................................... II

Introduction .................................................................................................................. 1

1 Background Information ......................................................................................... 3
  1.1 Streams ................................................................................................................. 3
  1.2 Crustacea ............................................................................................................ Error! Bookmark not defined.

2 Methods ................................................................................................................... 6
  2.1 Research design ................................................................................................. 6
  2.2 Experiment .......................................................................................................... 6
    2.2.1 Site selection ................................................................................................. 6
    2.2.2 Experiment Design ....................................................................................... 6
    2.2.3 Measurements .............................................................................................. 9
  2.3 Data analysis ...................................................................................................... 12

3 Results ..................................................................................................................... 13

4 Interpretation ........................................................................................................... 22

5 Conclusions ............................................................................................................ 24

Reference .................................................................................................................... 25

Annex .......................................................................................................................... 27
List of Figures

Figure 1 Distribution of rain during the summer on 2011 and the location of Hierdensebeek and Groote Molenbeek .................................................................5
Figure 2 Schematic site situation ........................................................................7
Figure 3 Experiment structure on Hierdensebeek .............................................8
Figure 4 Experiment structure on Groote Molenbeek.......................................8
Figure 5 Surber sampler with 25*25 cm opening ..........................................12
Figure 6 Daily precipitation and daily mean water level of control and stagnation stretch on Hierdensebeek ........................................................................14
Figure 7 Mean dissolved oxygen concentration (DOC) and min water level in BI, BC, AI and AC .....................................................................................14
Figure 8 Mean calcium, sodium and iron ion concentration in BC, AC, BI and AI ........................................................................................................14
Figure 9 The amount of different Crustacean species per sample and the average DOC of the sampling day on impact stretch of Hierdensebeek .................................................................15
Figure 10 The amount of different Crustacean species per sample and the average DOC of the sampling day on control stretch of Hierdensebeek .................................................................16
Figure 11 pH value and EC value on sampling day of Hierdensebeek ..............16
Figure 12 Daily precipitation and daily mean water level of control and stagnation stretch on Groote Molenbeek ........................................................................18
Figure 13 Mean dissolved oxygen concentration (DOC) and min water level in BI, BC, AI and AC of Groote Molenbeek. (Left) Figure 14 Mean calcium, sodium and iron ion concentration in BC, AC, BI and AI of Groote Molenbeek. (Right) ..................................................18
Figure 15 The amount of different Crustacean species per sample and the average DOC of the sampling day on impact stretch of Groote Molenbeek .................................................................20
Figure 16 The amount of different Crustacean species per sample and the average DOC of the sampling day on control stretch of Groote Molenbeek .................................................................21
Figure 17 Average water flow velocity on sampling day and Asellus aquaticus amount per sample on impact stretch of Groote Molenbeek .................................................................21
Figure 18 pH value and EC value on sampling day of Groote Molenbeek ........22
Figure 19 Occupation percentage of different species on Hierdensebeek (left) and Groote Molenbeek (right) ........................................................................22
List of Tables

Table 1 Timing of the stagnation experiments ..........................................................9
Table 2 Sampling schema of Groote Molenbeek (2011)...........................................11
Table 3 Sampling schema of Hierdensebeek.............................................................11
Table 4 Mean, Maximum and Minimum Temperature before dam closed and after dam closed on control and stagnation stretch of Hierdensebeek........................................14
Table 5 The total amount of different species of Crustacean on Hierdensebeek........15
Table 6 Mean, Maximum and Minimum Temperature before dam closed and after dam closed on control and stagnation stretch of .........................................................18
Table 7 The total amount of different species of Crustacean on Groote Molenbeek. .......19
Introduction

Changes in the climate system state have never stopped, although the release of greenhouse gas could be stable. (Kwadijk and Rotmans 1995) And it will induce the varying of temperature and the precipitation pattern with extremely intensive rain in winter and long period of dry summer. According to the climate modeling studies that were done by KNMI, the average, maximum and minimum air temperature is predicted to rise in the winter and the summer dry period is longer over 50 years. In addition, the overall trend of the mean precipitation, wet day frequency and especially the precipitation on a wet day in winter months present an upward trend meanwhile the wet day frequency of the summer will decline and more evaporation will occur. (Hurk, Klein Tank et al. 2006)

Hydrological processes are directly dependent on climate conditions. (Kwadijk and Rotmans 1995) Study shows that the relatively small climate changes could have an amplified effect on stream flow (Idso and Brazel 1984). In the extreme summer dry period, the river can hardly be recharged from precipitation and ground water. The discharge could decline dramatically. With a long term of extreme high evaporation rate and low recharge, the low flow or even drought will occur. (Schubert, Suarez et al. 2004) Heicher (Heicher 1993) find out numbers of possible environmental effects caused by low flow. With the overall reduction in flow, the influence of winds, bank storage, spring seepage, tributary streams and the warming effects of the sun increases.

A low flow is a seasonal phenomenon, and an integral component of a flow regime of any river (Heicher 1993). The obvious characters of low flow are the considerable reduction of stream discharge which may lead to increased sedimentation. Moreover, low flow or even no flow will induce the increase of the water temperature. (Chadwick 2008) Stream flow reduction may lead to changes in the relative abundance of algae, allochthonous material and organics. Under reduced flows in summer, biological oxygen demand and phosphorus levels would increase, whereas ammonia levels would fall due to higher nitrification rates. This gives rise to increased nitrate concentrations as ammonia decays to nitrate. The authors concluded that there could be enhanced growth of algal blooms in rivers and reservoirs which could affect dissolved oxygen levels (Whitehead, Wilby et al. 2009).

Crustacean is one of macroinvertebrates that have no backbone and can be seen with the naked eye, and they are sensitive to different chemical and physical conditions. Most crustaceans are free-living aquatic animals which include such familiar animals as crabs, lobsters, crayfish, shrimp, krill and barnacles. The subphylum Crustacean comprises almost 67,000 described species. (Zhang 2011) One of the most common species in the Netherlands streams is Gammarus pulex. (Karaman and Pinkster 1977) Gammarus tigrinus and Asellus aquaticus are also common in study streams. (Di Lascio, Rossi et al. 2011; Kotta, Orav-Kotta et al. 2011) In addition, they are used for detecting the water quality for their sensitivity for water quality changing. Therefore, it is necessary to investigate the response of Crustaceans for stagnation. Environmental modifications can alter Crustacean communities. Firstly, temperature is an essential factor that will influence the reproduction of Crustacean. Many studies proved that
Gammarus pulex will not reproduce when the temperature is lower than 5 °C (Heinze 1932; Hynes 1954; Hynes 1955) and Gammarus tigrinus will not reproduce if the water temperature is lower than 0.5 °C (Pinkster, Smit et al. 1977). Secondly, high level of nutrients in the form of nitrogen and phosphorus that could be increase in stagnation condition (Chang, Evans et al. 2001) activates excessive algal growth. They will consume oxygen that already decreased due to dropping water level. In these low oxygen conditions, some oxygen sensitive Crustacea species will die. (Meijering 1991)

In this paper, crustaceans will be studied only for the sensitivity of the water temperature, water quality and dissolved oxygen concentration changes. The amount and community of crustacean are worth to be used on analyzing the response to the climate change. Firstly, water temperature has a big influence on the amount of crustacean in water. The amount of some crustaceans is usually bigger than the amount of the others species of crustaceans, because they have shorter generation times on suitable water temperature. Moreover, oxygen that may be influenced by water level, water temperature and water velocity (Chadwick 2008) is essential for some species of crustacean (Meijering 1991). Furthermore, current water velocity also directly affects the amount of some crustaceans. Some of them could survive easier than other taxa in the swifter water because they are prominent deposit feeders among the macroinvertebrates in swifter streams (Meijerin. Mp 1971). It is necessary to note that pH and EC could influence the Crustacean, but the tolerance ability of Crustaceans on these aspects is quite strong. (Hynes 1955; Roux 1970; Pinkster, Smit et al. 1977)

From above, the responses of Crustaceans on water temperature, water quality and dissolved oxygen concentration changes that due to decreasing water flow are worth to investigate as a Bachelor thesis. This study was done as a base for a Bachelor thesis Land and water management at the Department of Freshwater Ecology, WUR/Alterra.

The goal of this thesis is 1) to explore the changes in environmental conditions – dissolved oxygen content, discharge, current velocity, water temperature, water quality and substrate composition- during low flow and stagnation in stream 2) to explain the effects of these changes on some common species of Crustacean in sand lowland streams and 3) to indicate the difference between effects in high nutrient and low nutrient stream

Main question:
What are the effects of reduced flow and stagnation on crustacean in sandy lowland streams?

Sub questions:

1) What and how fast will be the changes in stream water (dissolved oxygen content, discharge, current velocity, water temperature, water quality, and substrate composition) once stagnation conditions occur?
2) How large do these changes have to be trigger the changes in crustacean composition?
3) Is there any difference of the response of crustacean between high level nutrients and low level nutrients streams?

Here, the effects of temperature and low flow on Crustaceans of sandy lowland streams are studied. Effects of increased temperature are tested in a field survey that began in the summer of 2011. The focus is on direct effect on Crustaceans through changes in the water temperature and indirect
through influence on the habitat patterns. Influence of the low flow is investigated in field experiments. Changes in environmental conditions and their influence in the Crustaceans are monitored. Results will provide the basis for quantified mitigation strategies. The surveys and experiments are carried out at two locations in the Netherlands. This paper divided into five parts. Part one, background information, provides the brief introduction of Hierdensebeek and Groote Molenbeek and also the characters and preference of *Gammarus pulex, Gammarus tigrinus and Asellus aquaticus*. Part two, methodology, introduce the detail of whole research: site selection, field experiment, laboratory experiment, and data analysis. The results are present in part three. The variation tendency of different aspect and comparing of different aspect with Crustacean amount are shown in this part. Interpretation which is the fourth part discusses and explains the result that present in part three. Conclusion is the last part. In this part, the research question is answered.

1 Background Information

1.1 Streams

Two streams are selected as the experiment catchment in the Netherlands. One is Hierdensebeek and another is Groote Molenbeek, the experiment is carried out in the locations that present in Figure 1. Comparing with past years, summer that with very often occurred overcast day is very wet and cool in 2011 in the Netherlands. Hierdensebeek owns the highest precipitation (475-500 mm) while Groote Molenbeek owns low annual rainfall (300-325 mm).

1.2 Crustacean

In both experiment streams, five species from crustacean are found: *Gammarus pulex, Gammarus tigrinus, Asellus aquaticus, Proasellus banyulensis* and *Proasellus meridianus*. The amount of *Gammarus pulex Gammarus tigrinus and Asellus aquaticus* accounts for the majority of the total amount of Crustacean. Therefore, *Gammarus pulex Gammarus tigrinus and Asellus aquaticus* will be introduced. Furthermore, more than 50% of *Gammarus sp.* is still in juvenile level. Thus, *Gammarus pulex and Gammarus tigrinus* were classified in *Gammarus sp.* level in analysis. *Amphipod Gammarus pulex (Linnaeus 1785)*, which is one of the main shredders in running water of temperate regions (Boyero, Pearson et al. 2012), is a numerically dominant species in small woodland streams in north Western Europe. (Meijerin.Mp 1971; Karaman and Pinkster 1977; Pinkster, Smit et al. 1977) *Gammarus pulex* or *G. pulex* is influenced by water temperature (Sutcliffe, Carrick et al. 1981), food (Graca, Maltby et al. 1993; Franken, Batten et al. 2006) and habitat (Adams, Gee et al. 1987; Peeters and Gardeniers 1998; Franken, Batten et al. 2006) Moreover, *G. pulex* is the most active migrator of the *Gammarus* species of running waters (Meijerin.Mp 1971). It is sensitive for low oxygen condition. (Meijering
Many studies prove that *G. pulex* in freshwater habitats from early spring until the end of the year and it will continuously reproduce when the temperature is higher than 5 °C. (Heinze 1932; Hynes 1954; Hynes 1955) Moreover, *Gammarus pulex* usually generate in fresh or slightly oligohaline waters and is also able to reproduce within very high salinities condition (up to 3000 mg Cl/L) (Hynes 1955; Roux 1970; Pinkster, Smit et al. 1977) According to the laboratory study that done by Friberg and Jacobsen (Friberg and Jacobsen 1994), *Gammarus pulex* (G. pulex) prefer to eat conditioned alder leaves rather than five other foods which include conditioned beech leaves, fresh beech leaves, Sitka spruce needles, a fresh macrophyte, and fresh filamentous green algae. Additionally, Graca, a et al find out that the food preference of G. pulex was not correlated with fungal biomass, leaf disc toughness leaf decomposition or nitrogen content. (Graca, Maltby et al. 1993) 

*Gammarus tigrinus*, which originated from the eastern shores of northern America, has rapidly invaded most of the oligohaline waters and gradually replace the local species in the Netherlands. (Pinkster, Smit et al. 1977) It has a much greater reproductive capacity than *G. pulex* or any other indigenous *Gammarus* species for several reasons: First, the reproductive cycle is comparative shorter with a resting period in the cold winter month. (Below 0.5 °C) In some high water temperature locations, ovigerous females can be found whole year. Moreover, *Gammarus tigrinus* that own a faster incubation time is able to reproduce many (up to 16) generations one season and local species can only produce one to four generations. In addition, it’s very short time to reach sexual maturity. (Pinkster, Smit et al. 1977) Compare with *Gammarus pulex*, the competitive strength of *Gammarus tigrinus* is less in extremely variable habitats and fresh running water condition.

*Isopods Asellus aquaticus* occur in epigean lotic and lentic habitats. *Asellus aquaticus*, which is one year life-cycle macroinvertebrates, has two complete generation every year, with gravid females usually present from February to June and July to September, giving rise to spring and autumn cohorts. (Adcock 1979; Bloor 2010) *Gammarus pulex* and *Asellus aquaticus* generally occupy different zones in rivers: the former occurs in upper reaches but is `replaced' by the latter in lower reaches.
Figure 1 Distribution of rain during the summer on 2011 and the experiment location of Hierdensebeek and Groote Molenbeek
2 Methods

2.1 Research design

The following methods of data collection were used: literature review and experimental research. Literature review was used for exploring the predictable climate change influences on crustacean. The field experiment was carried out during the experiment stagnation conditions that were simulated in the streams. During the experiment, crucial environmental parameters were measured and macrofauna samples collected. The field experiment was carried out from May to October in 2011. Only small part of laboratory experiment was finished last year. Unfortunately, I did not join in the field experiment. But most part of laboratory experiment was done by me. Data that were used in this paper were partly available and partly done by me.

2.2 Experiment

The effects of low flow and stagnation on ecosystem in sandy lowland streams were tested in a field experiment.

2.2.1 Site selection

Two streams were chosen: low nutrient stream called Hierdensebeek (<0.15 mg P/l) and one high nutrient called Groote Molenbeek (>0.15mg P/l). The selection is based on the following rules:
- Permanent, lowland stream (high priority criterion).
- Width ranges between 2-5 m (depth is too variable as parameter to be included).
- The site is situated in a half-open landscape with deciduous vegetation.
- The stretches their selves are partly not shaded.
- Current velocity ranges between 15-25 cm/s.
- The slope ranges 0.5-1 m/km (altitude lower priority; plateau like situation).
- The substrate consists of sand or gravel (low priority!).
- Discharge is characterized as “less” dynamic.
- Location (Latitude and longitude)
Both streams are matching those conditions.

2.2.2 Experiment Design

Site construction

A control and an impact stretch were selected in both streams. At both streams, a control and an
impact stretch with the comparable condition were selected. The control stretch located on the upstream of impact stretch (Figure 2). The impact stretch was set-up at the downstream part of control stretch. The major habitat in both impact stretch and control stretches were comparable. The minimum length of control stretch and impact stretch are 34m and 68m, respectively. Both control stretch and impact stretch experiments were conducted at the same time in one stream.

![Schematic site situation](image)

Figure 2 Schematic site situation

A by-pass was settled next to the impact stagnation stretch. The old meander or a ditch like channel could be used as the by-pass. It was used for redirecting the water flow. A weir was situated at the head of the impact stretch and was closed on the low flow/stagnation period. A low dam which was installed at the end of impact stagnation stretch can inhibit the water flow back from downstream entering the impact stretch. Weir and the low dam could be constructed using sand bags.

**Hierdensebeek**

Figure 3 present the experiment structure on Hierdensebeek. Experiment phase on Hierdensebeek, which is a low nutrient stream (> 0.15mg P/l), was 90 meters in total: control stretch, stagnation stretch and drought stretch (not included in this study) were 30 meters, respectively. The habitat pattern is equal in both control and stagnation stretches. The width on both stretches was around 3 meter. The dam was constructed by sand bag. The abundant channel that located next to the impact stretch was used as by-pass to redirects the excess water.
Groote Molenbeek

Figure 4 present the experiment structure on Groote Molenbeek. Experiment phase on Groote Molenbeek, which is a high nutrient stream, was 90 meters in total: control stretch, stagnation stretch and drought stretch (not included in this study) was 30 meters, respectively. The habitat pattern is equal in both control and stagnation stretches. The width on both stretches is around 3 meter. The weir that was built by water board already exists. The abundant channel on that next to the impact stretch is used as by-pass which redirects the excess water.
Experiment

The water in the stagnation stretch does not flow or hardly flows within the stagnation period. Water feeding of stagnation parts aims to maintain the stagnation situation. The volume of water charging is depended on the water loss due to infiltration and evaporation. Additionally, it is necessary to construct an additional by pass or use a pump that can drain the excess water if the stream is located in a serious seepage area or after a heavy rain. On Groote Molenbeek, there were several big event of rainfall that occurred during the middle of August, and it lead to a big flow that flow over the dam. On Hierdensebeek, the dam was broken in the beginning of September due to the peak discharge. These are normal phenomenon and then the water dropped to the level that we expected. So it is not necessary to construct an additional by pass. Furthermore, it is essential to prevent the animals lose during the drainage. Both control and stagnation stretches were divided into ten sampling sections and five spare sections. There is at least two meters space between the most upstream/downstream sections and each wire/dam.

Timing

The low flow experiments were last for eighteen weeks for Hierdensebeek and twenty one weeks for Groote Molenbeek: five weeks before the stagnation phase on both streams and thirteen weeks during the low flow phase for Hierdensebeek or sixteen weeks during the low flow phase for Groote Molenbeek. (Table 1) As the information that mentioned before, the field experiment was carried out before my Bachelor research so I did not attend in those field works. The starting date of stagnation phase is 6\textsuperscript{th} June, 2011 on Hierdensebeek. The dam closed on 4\textsuperscript{th} July, 2011. Before the dam closed, samples were taken once a week. After the dam closed, first two samples were taken once two weeks. However, the difference is not significant. Later on, samples were collected once three weeks. In week ten, the dam was broken for the peak discharge of the stream. On Groote Molenbeek, it starts sampled on 20\textsuperscript{th} May, 2011 and the dam closed on 27\textsuperscript{th} June, 2011. Before the dam closed, samples were taken weekly. Then first two samples were taken once two weeks and turn to once three weeks for rest samples. Stream was cleaned in September and all macrophytes were removed.

Table 1 Timing of the stagnation experiments

<table>
<thead>
<tr>
<th>2011</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>August</th>
<th>September</th>
</tr>
</thead>
<tbody>
<tr>
<td>week number</td>
<td>22</td>
<td>23</td>
<td>24</td>
<td>25</td>
<td>26</td>
</tr>
<tr>
<td>Experiment</td>
<td>before phase</td>
<td>low flow and Stagnation phase</td>
<td></td>
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<td></td>
</tr>
</tbody>
</table>

2.2.3 Measurements

Continuous measurements:
Discharge, water temperature and oxygen in both control and impact stretches were measured continuously, with twenty min interval. The air temperature logger was installed at the same time.

**Discharge**

At each experimental site, discharge was measured continuously in control and impact stretches. The nearest gauge station was used and a continuous water level recorder was installed in the control and both impact parts. The water level loggers, which used for measuring water velocity over different (preferably weekly) hydrological conditions (flood, low water, etc…) that occur during the experiment, was calibrated.

**Water temperature**

Two temperature loggers (HOBO UA-002-64 logger or similar logger) were used for continuously measuring water temperature with twenty min interval at both control and stagnation stretches. On control stretch, the logger was settled in well mixed water. Meanwhile, the logger was placed in the middle of the stagnation stretch. And it should be located within a shaded spot to avoid heating up. During the experimental period, loggers should be checked regularly to secure that they have not been covered by material, overgrown by algae, were still operational and measure correctly.

**Air temperature**

Air temperature was measured by HOBO UA-002-64 logger (or similar logger). Every logger should be placed 150 cm above the ground, in a shaded area. It also should be checked regularly as the water temperature logger.

**Oxygen**

Two temperature loggers (HOBO UA-002-64 logger or similar logger) have been used for continuously measuring oxygen with twenty min interval at both control and stagnation stretches. On control stretch, the logger was settled in well mixed water. Meanwhile, the logger was placed in the middle of the stagnation stretch. During the experimental period loggers should be checked regularly to secure that they have not been covered by material, overgrown by algae, were still operational and measure correctly.

**Interval measurements:**

**Water chemistry (Federation. 1995)**

Water samples were taken together with the macrofauna sampling. Therefore, there were ten sampling occasions. During the before phase of both streams, one water samples were taken at each occasion on control stretch and one at each occasion on impact stretch. In the stagnation phase, two water samples were taken at each occasion per stream (one at the stagnation stretch and one at control stretch).

During the experiment, pH, EC, O₂, t-P, t-N, ammonium, nitrate, dissolved inorganic phosphorus (PO₄-P), calcium, chloride, iron and sodium were analysed by standard colorimetric-related methods.
Every sampling bottle was completely filled and then closed under the water. All water samples were directly placed in freezer with -18°C after returning to the lab.

**Substrate cover estimation**

Substrate cover was estimated at control and stagnation stretch. At each stretch, three 5 m long sections were set for the substrate estimation and the substrate cover was estimated at same sections: 5 times in the before phase (weekly) and 5 times in the stagnation phase (2 weekly).

The method of estimation was chosen by the partner and depends on the size of the stream and turbidity of the water. The STAR substrate classification was used.

**Macrofauna sampling**

Macrofauna sampling was implemented every week before the stagnation phase and once two weeks or once three weeks during the stagnation phase (*Table 2*).

**Table 2** Sampling schema of Groote Molenbeek (2011)

<table>
<thead>
<tr>
<th>Groote Molenbeek</th>
<th>2011</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>August</th>
<th>September</th>
<th>October</th>
</tr>
</thead>
<tbody>
<tr>
<td>Week</td>
<td>-5</td>
<td>-4</td>
<td>-3</td>
<td>-2</td>
<td>-1</td>
<td>0</td>
<td>1 2 3 4 5</td>
</tr>
<tr>
<td>Planned</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>● ● ● ● ●</td>
</tr>
<tr>
<td>Stagnation</td>
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<td>B</td>
<td>B</td>
<td>B</td>
<td>S</td>
<td>S</td>
<td>S S S</td>
</tr>
<tr>
<td>Samples</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2 2 2</td>
</tr>
<tr>
<td>Period</td>
<td>Before phase</td>
<td>low flow and stagnation phase</td>
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</tbody>
</table>

**Table 3** Sampling schema of Hierdensebeek

<table>
<thead>
<tr>
<th>Hierdensebeek</th>
<th>2011</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>August</th>
<th>September</th>
<th>October</th>
</tr>
</thead>
<tbody>
<tr>
<td>Week</td>
<td>-5</td>
<td>-4</td>
<td>-3</td>
<td>-2</td>
<td>-1</td>
<td>0</td>
<td>1 2 3 4 5</td>
</tr>
<tr>
<td>Planned</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>●</td>
<td>● ● ● ● □</td>
</tr>
<tr>
<td>Stagnation</td>
<td>B</td>
<td>B</td>
<td>B</td>
<td>B</td>
<td>S</td>
<td>S</td>
<td>S □ S</td>
</tr>
<tr>
<td>Samples</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>2 2 2</td>
</tr>
<tr>
<td>Period</td>
<td>Before phase</td>
<td>low flow and stagnation phase</td>
<td></td>
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</tr>
</tbody>
</table>
In before phase, macroinvertebrates samples were taken from one randomly selected (beforehand) section of control and stagnation stretches weekly. Every sample was made up of three Surber samples (25*25 cm). Three Surber samples are taken from three different domain substrates that are presented in the randomly selected (beforehand) section. That means every Surber sample belongs to three different domain substrates. Those three Surber samples are combined in one composite sample.

In the stagnation phase, macroinvertebrates samples are taken from one randomly selected (beforehand) section of control and stagnation stretch. First two samples were taken two weekly and then turned to once three weeks. In Hierdensebeek, dam was broken on week 10 due to extremely high discharge. So samples taking were delayed for two weeks. Every sample is made up of three Surber samples (25*25 cm). Surber samples are taken by Surber Sampler that are used for quantitative analysis of benthic stream organisms and can be used in shallow streams that less than 18” deep with a range of bottoms from silt to large cobble. Net frame made from stainless steel, opening 25 x 25 cm, with Nylon webbing, net bag with 500 microns mesh size, length approx. 70 cm. Handle joint with 20 mm inner diameter.

Samples are taken in different section each visit. So, it ensures that every section was not sampled twice. Depth, current velocity and substrate type were measured and described at each Surber. The width of every sampled section was measure and the substrate cover percentage of every sampled section is noted down.

2.3 Data analysis

After the data collection, the data are analyzed using Excel and archived and organized by Access. Water level (discharge) and precipitation are display in on figure to show the relationship between them. The amount of Crustaceans per sample is present by table. Access was used for organizing the temperature data, water level data and dissolved oxygen concentration data. All continues measurements with 20min interval were transformed to average per day. Mean, Maximum and Minimum Temperature before dam closed and after dam closed on control and stagnation stretch were calculate and present by table. Mean DOC with standard error on control stretch and impact stretch in different period (before dam closed and after dam closed) were show together with minimum water level in different phase. Thus, the relationship will be clear. DOC was continues measured with 20 min interval, the mean, maximum and minimum DOC on the sampling day were used and present together with amount of Crustaceans to investigate the relationship between them. The pH value and EC value were measured in laboratory by water sample on sampling day and represent on line chart to observe the variation tendency. Concentration of iron ion, sodium and calcium ion with standard error were calculated and represented on figure. It is used for investigate the
reason of EC value changes. The occupation percentage of different species in different streams was represented by pie chart to observe the Crustacean community difference between high nutrient and low nutrient stream.

3 Results

3.1 Hierdensebeek

The dam that located between control stretch and stagnation stretch was closed on 04th July, 2011 at 15:50. There is a slightly increase in water level of both stagnation stretch and control stretch that was presented by the blue line and green line, respectively. (Figure 6) And stagnation parts water level remains stable while the control stretch water level shows a slight decline, this situation lasted for a few days till the big event of precipitation occurred 13th July, 2011. Compared with the situation on impact stretch, the influence that caused by the rain on impact stretch was not as serious as the situation on control stretch. Obviously, the heavy rain made the discharge of control stretch rocket up. Then, the discharge of both control and stagnation stretches changed almost along the precipitation. But the range of variation on control part was bigger than it on stagnation part. There was only one exception occurred around 10th Sep. On both control and stagnation stretches, the discharge level after dam closed is higher than before.

The DOC and water level was positively related (Figure 7). Figure 7 represent the relationship between dissolved oxygen concentration and water level. Before dam closed, the mean of dissolved oxygen concentration (DOC) on stagnation stretch was 5mg/L. After the dam closed, the DOC mean value decreased lower than 4 mg/L. Meanwhile, the the mean value of water level on impact stretch before dam closed is also lower than it after dam closed. On control stretch, mean water level increased slightly and the DOC also increased at the same time. The fluctuations of temperature were not very high on both before and after phases. (Table 4) The variation range was from 9.571 to 18.711 °C on control stretch before dam closed and the mean. The variation range on stagnation stretch was 9.571 to 20.901 °C. After the dam closed, the temperature range on both control stretch and stagnation stretches were from around 10 to 17 °C.

The amount of different species of Crustacean was shown in Table 5. Before the dam closed, the amount of *Asellus aquaticus*, *Proasellus banyulensis* and *Gammarus sp.* on different stretch per week are comparable and the difference was not significant. After 4th July (dam closed), the amount of *Asellus aquaticus* and Gammarus sp.* on stagnation stretch was quite smaller than it on control stretch. Especially *Gammarus sp.*, the amount on control parts was several times, a dozen times or even scores of times above the amount on stagnation stretch. On stagnation stretch, the amount of *Gammarus sp.* was comparatively higher than other weeks after dam closed. But the amount of *Asellus aquaticus* and *Proasellus banyulensis* did not changes irregularly
Figure 6 Daily precipitation and daily mean water level of control and stagnation stretch on Hierdensebeek

Table 4 Mean, Maximum and Minimum Temperature before dam closed and after dam closed on control and stagnation stretch of Hierdensebeek: BC=Before dam closed on control stretch; BI=Before dam closed on impact stretch; AC=After dam closed on control stretch; AI=After dam closed on impact stretch.

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Figure 7 Mean dissolved oxygen concentration (DOC) and min water level in BI, BC, AI and AC. (Left)

Figure 8 Mean calcium, sodium and iron ion concentration in BC, AC, BI and AI. (Right)
Table 5 The total amount of different species of Crustaceans per sample on Hierdensebeek. I=Impact stretch, C=Control stretch

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Figure 9 The amount of different Crustacean species per sample and the average DOC of the sampling day on impact stretch of Hierdensebeek
Figure 10 The amount of different Crustacean species per sample and the average DOC of the sampling day on control stretch of Hierdensebeek

Figure 11 pH value and EC value on sampling day of Hierdensebeek

Figure 8 indicates the concentration of iron ion, sodium and calcium ion during the stagnation period. The iron ion on control and stagnation stretch was increased after dam closed. At the same time, the sodium ion on control stretch was declined while the sodium ion on impact stretch was slightly increased. Calcium ion concentration was declined in control stretch but increased on impact stretch and the total amount of Ca$^{2+}$ on impact stretch was higher than it on control stretch.

For the narrow range of temperature, pH and EC variation, it cannot influence the amount of
Crustacean, the relationship between them were not analyzed. The DOC varied enough to trigger changes of Crustaceans amount, so it was analyzed. The relation between DOC and *Gammarus sp.* amount was presented in Figure 9 and Figure 10. On stagnation stretch, the highest number of *Gammarus sp.* was appeared on 6th June and 4th July when the biggest number of minimum DOC level occurred. The lowest number of *Gammarus sp.* was appeared on 23rd August when the lowest DOC occurred on impact stretch. (Figure 9) The situation on control stretch was similar with stagnation stretch. Comparing the control stretch water level which was presented in Figure 6 and DOC in Figure 10, it is obvious that an increase of DOC on control stretch occurred on 18th July and the water level increased at same day. Meanwhile, the numbers of *Gammarus sp.* increase rapidly. Then the DOC and *Gammarus sp.* decreased, and water level declined also. After dam closed, DOC on control stretch was stabilized the same level before the dam closed or even higher. Meanwhile, the amount of *Gammarus sp.* was represented a similar tendency. On impact stretch, the DOC level start decreased and reached the lowest point and the decrease last 3 weeks. All in all, DOC level on control stretch was higher than it on impact stretch and the amount of *Gammarus sp.* was much bigger on control stretch than it on impact stretch.

**Groote Molenbeek**

The dam which located between control and impact stretch was closed on 27th June, 2011. Water level logger which installed on the control stretch was missing, so data of water level on control stretch was not available. Water level variation tendency was shown on Figure 12. Water level on impact stretch was decreasing sharply. After a small event of rain (11.1 mm on 29th June), impact stretch water level continuously dropped to 0.27 m. Then the stream was recharged by precipitation and rebounded to a certain level (around 0.4m) till the middle of August. Two big event of rain induced the overflow of the dam and it resulted the substantial increase of water level. The water level was nearly increased to the peak level for the overflow. Afterwards, it declined to normal level that equals the water level before the overflow. The water level after dam closed was only half the water level before dam closed in most time.

Temperature fluctuated within certain range from 8.0-22.3 °C for control stretch and 6.8-23.6 °C for impact stretch. Mean temperature on both stretches were remaining around 17 °C during whole experiment. The minimum temperature on impact stretch was about 2 degree lower than it on control stretch.(Table 6) The DOC strongly correlate with water level on impact stretch. (Figure 13) Mean water level on impact stretch decreased from 0.6 m to 0.4 m after dam closed. Meanwhile, DOC have dramatically fallen on impact stretch. (drop from 9.3 to 2.98 mg/l) (see Figure 12 and Figure 13)
Figure 12 Daily precipitation and daily mean water level of control and stagnation stretch on Groote Molenbeek

Table 6 Mean, Maximum and Minimum Temperature before dam closed and after dam closed on control and stagnation stretch of

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Figure 13 Mean dissolved oxygen concentration (DOC) and min water level in BI, BC, AI and AC of Groote Molenbeek (Left) Figure 14 Mean calcium, sodium and iron ion concentration in BC, AC, BI and AI of Groote Molenbeek. (Right)
Table 7 The total amount of different species of Crustaceans per sample on Groote Molenbeek. I=Impact stretch, C=Control stretch

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<tr>
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Low flow had influences on pH and electrical conductivity. (Figure 18) The pH value on control stretch scarcely fluctuated in most time and it maintain within 6.5-7.5 range. On impact stretch, it waved from 6 to 8 with only on sharp drop on 20th June when chloride dropped. As for electrical conductivity (EC), the variation tendency was the same on control and impact stretch before dam closed. After the dam closed, EC on stagnation stretch reached the peak and the control stretch EC value reached the peak a few days later. Both of them dropped after they have reached the peak. It fluctuated within 400-600 during the experiment period. (Figure 18)

Figure 14 indicate the concentration of iron ion, sodium and calcium ion during the stagnation period. The iron ion on control was slightly increased after dam closed while iron ion on stagnation stretch rapidly increased from about 0.01 mg/l to 0.05 mg/l (Figure 14). In the same time, the sodium ion on control stretch was declined while the sodium ion on impact stretch was slightly increased. Calcium ion concentration was increased on impact stretch and control stretch. The total amount of Ca$^{2+}$ on impact stretch was much higher than it on control stretch after dam closed.

The amount of different Crustacean species per sample are shown above. (Table 7) Before the dam closed, the amount of Gammarus sp. on different stretch per week were comparable and the difference was not significant. Asellus aquaticus numbers on control stretch was twice the numbers on impact stretch. Proasellus maridianus amount was quite small and the changes do not mean anything. After 27th June (dam closed), the amount of Asellus aquaticus on impact stretch increased sixfold and it was comparable with the Asellus aquaticus amound on control stretch. In contrast, the amount of Gammarus sp. on stagnation stretch was nearly disappeared after dam broke and the amount on control stretch rocked up from 49 to 216.

There was a relation between DOC and Gammarus sp. amount (Figure 15 and Figure 16). The minimum DOC level presented the same tendency with the amount of Gammarus sp. The numbers of Gammarus sp. was comparable on both stretches before dam closed for the
similar condition of the stream. Afterwards, on stagnation stretch, the lowest number of *Gammarus sp.* was appeared on 7th September and 27th September when the lowest minimum DOC level occurred. The situation on control stretch was similar with stagnation stretch during stagnation phase. After dam closed, DOC on control stretch was stabilized the same level before the dam closed or even higher. Meanwhile, the amount of *Gammarus sp.* was represented the similar tendency. DOC level on control stretch was stabilized the same level before the dam closed or even higher after dam closed and the *Gammarus sp.* amount was represented in a similar tends. When it comes to the impact stretch, it is necessary to note the tendency of minimum DOC. It dropped to negative value and stay around this level after dam closed. Oxygen were richer on control stretch than it on impact stretch and *Gammarus sp.* were preferred to live in control stretch.

The relation between average water velocity and *Asellus aquaticus* amount was very strong. (Figure 17) The velocity dramatically decreased from 0.25 m/s (Figure 17) to almost zero due to the dam closed. At the same time, it is important to note that the amount of *Asellus aquaticus* has doubled and redoubled.

![Graph showing the amount of different Crustacean species per sample and the average DOC of the sampling day on impact stretch of Groote Molenbeek](image)

Figure 15 The amount of different Crustacean species per sample and the average DOC of the sampling day on impact stretch of Groote Molenbeek
Figure 16 The amount of different Crustacean species per sample and the average DOC of the sampling day on control stretch of Groote Molenbeek

Figure 17 Average water flow velocity on sampling day and Asellus aquaticus amount per sample on impact stretch of Groote Molenbeek
Hierdensebek, as a low nutrient stream, was suitable for *Gammarus sp.* to survive. *Gammarus sp.* is the most common Crustacea and it occupied 95% in total. There is only 5% of Crustacean are *Asellus aquaticus*, and *Proasellus banyulensis* appeared.

In high nutrient stream, Groote Molenbeek, *Asellus aquaticus* was dominating over the stream with 78% occupancy. *Gammarus sp.* was occupied 22% in total and only 4 *Proasellus maridianus* appeared.

### 4 Interpretation

**Changes in streams**
Water level is decided by the recharging water from upstream, ground water and also precipitation. Right after the dam closed, there was a slight increase instead of sharp decrease that was expected on both control and stagnation stretches on Hierdensebeek. That is because there was a dam between the stagnation and drought sections. When the dam that located on upstream was closed, the water level in the stagnation reached the level of the dam between the sections, because the slope in the stream was quite steep. The decreasing water flow induced many consequences and there is no doubt that oxygen was heavy influenced by water flow. The positive correlation between oxygen and water flow was strong according to the experiment result. On Hierdensebeek, the concentration of dissolved oxygen stabilized around 8 mg/l on control stretch and 3 mg/l on stagnation stretch with approximately 0.35 and 0.25 m water level, respectively. The DOC could lower than zero when water level is around 0.25 m on Groote Molenbeek.

Water temperature was also influenced by the water flow. The high water level could induce the higher minimum water temperature. On Groote Molenbeek, water level dropped from 0.6 m to 0.3 m due to dam closed. For this reason, the minimum temperature slightly declined and the vary range of temperature was wider in stagnation period than the vary range of temperature before stagnation. The maximum temperature increasing might be resulted from air temperature rising. However, the variation range was quite narrow before dam closed and the lowest temperature on stagnation phase was still above 5 °C.

Chemical process in stream was influenced by water flow and water recharge. For both streams, the total amount of calcium, sodium and iron ion on impact stretch rapidly rose and it was much higher than the amount on control stretch during the stagnation period. The concentration of those ions on surface water and precipitation was not high and they are rich in groundwater. It is possible that there was water recharge on impact stretch form groundwater. (Chadwick 2008) For the sodium ion, calcium ion and iron ion changing, EC value decreased. Water level varied within wide limits while the pH value fluctuates within a narrow range (Hierdensebeek pH range is 6-9; Groote Molenbeek pH range is 6-8).

Changes in Crustaceans composition

As the characters that was mentioned in the beginning, Gammarus sp. are sensitive for oxygen (Meijering 1991) and it can normally survival and develop with quite low pH (pH:4.7)(Chadwick 2008), low water temperature (G. pulex: above 5°C; G. tigrinus: 0.5 °C) and very high salinity concentration water (Hynes 1955; Roux 1970; Pinkster, Smit et al. 1977). Thus, the amount change of Gammarus sp. did not correlate with pH value, EC value and temperature within the narrow range of theirs variation. Therefore, dissolved oxygen concentration variation dominated the variation of Gammarus sp. Furthermore, the minimum DOC level has essential influence on the amount of Gammarus sp.. The lowest number of Gammarus sp. was appeared on 23rd August whereas the maximum DOC is higher than some other weeks and the minimum DOC level reached the lowest point on the same day. Isopods Asellus aquaticus occur in epigean lotic and lentic habitats in most time. From the result, it is obviously that oxygen could not influence the amount of Asellus aquaticus but water velocity could. The amount of Asellus aquaticus in stagnant water was much higher than it in flow water. The amount could be very large when the water hardly flows.

Difference between high level nutrients and low level nutrients streams
There are several differences between different species between high nutrient stream and low nutrients stream. (Figure 19) Firstly, the total amount of animals is quite different. The total amount of Crustaceans is over ten thousands on low nutrients stream while Crustaceans numbers is just over two thousands on low nutrients. Secondly, in high nutrient stream, Groote Molenbeek in this case, *Asellus aquaticus* occupied the biggest fraction; *Gammarus sp.* and *Proasellus banyulensis* appeared. Conversely, *Asellus aquaticus* occupied only 5% and *Gammarus sp.* is the most common species in low nutrient stream which is Hierdensebeek in this case. *Proasellus maridianus* which did not appear in high nutrient stream appeared in low nutrient stream.

## 5 Conclusions

Due to stagnation, water temperature, dissolved oxygen concentration, pH and EC value were influenced. The fluctuation range of temperature was not big. The maximum and mean water temperature did not change a lot but the minimum temperature decreased a lot. The variation range of temperature was much bigger after dam closed than it before dam closed. The change is not occurred immediately but in a long period. Comparing with temperature, the DOC changes was drastic. The minimum DOC dramatically and immediately declined after water level started decrease. The minimum DOC could reach and stay around negative value due to decreasing water level. The pH value did not fluctuate widely. There was a slightly rise of pH value within two weeks after water level declined and then it remains in that level. The EC value presented an increase within three weeks after water level decrease, and then slipped back. The decrease of EC may be caused by the ground water recharge for the ion concentration increase in the stream. The water velocity shown a slowly drop in a long period.

The variation ranges of pH EC and temperature were too narrow to trigger the changes on Crustacean. Only DOC and water velocity variation could trigger the change of Crustacean. There is a strong positive correlation between *Gammarus sp.* and DOC; and a negative correlation between *Asellus aquaticus* and water velocity. According to the literature, *Gammarus sp.* and *Asellus aquaticus* can normally grow, develop and reproduce if 1) the temperature is higher than 5 centigrade; 2) pH value is higher than 4.7; 3) chloride ion concentration is lower than 3000 mg Cl⁻/l.

The decreasing water flow could influence amount of Crustacean in different aspect, for example oxygen, temperature, pH value and EC value. But the effects could only be significant if the changes on oxygen, temperature, pH and EC in a certain level. *Gammarus sp.* is very sensitive on dissolved oxygen concentration and *Asellus aquaticus* is sensitive on water velocity. In high nutrient stream, *Gammarus sp.* is very common while *Asellus aquaticus* is very common in low nutrient stream.
Reference


Annex

Project plan of thesis report (See separate annex)