The morphology of the threespine stickleback and its relation to predators and parasites, from three lakes in western Norway

Thesis

Candidatus scientiarum

Sturla Austad

Department of Zoology

University of Bergen

Norway

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Abstract

We studied the three-spined stickleback (Gasterosteus aculeatus) in three different lakes, with respect to morphology (Dorsal spine numbers and pelvis), distribution and relationship to the parasites Schistocephalus solidus and Glugea anomala and selective predation by fish predators. From these investigations we concluded the following. 1) We found possible indications that fish predators may have an effect on the population of S. solidus and G. anomala, and the morphological traits of the stickleback. 2) The stickleback's morphology related to parasites in Lake Liavatnet, showed that the pelvis reduced group had the lowest total volume of G. anomala cysts. May be due to fish predation pressure, or possibly because the group are hybrids made from introgression in Lake Liavatnet, which makes them less susceptible to G. anomala than the two other groups. The stickleback population from Lake Nesavatnet, we found a difference in prevalence from G. anomala and dorsal spines, as three spines we more infected than two spines. For S. solidus, we found that number of infections was higher and total volume was larger for two dorsal spines than for three. The result was not significant, but was opposite of the result from G. anomala 3) No relationship was indicated between G. anomala and S. solidus, as neither total volume nor number of infections was impaired by the presence of the other parasite.

1. Introduction

Concept of speciation is the joint evolution from one to two or more groups (Coyne, 2004). Arguments for forces which leads to speciation has been distinct (Chandler, 1989). MacColl (2010), deliberate reproductive isolation in a diverging population is central for speciation. Conditions for reproductive isolation is commonly known to occur, if populations are to be spatially isolated and fail to reproduce with each other, or they produce hybrids which are inferior for survival (MacColl, 2010). However, multiple forces can interact within this process. A specie might be separated due to intra or inter specific competition, which could emerge into two coexisting species, from one common ancestor (Schluter, 1992). When subspecies inhabit different ecological environment, barriers against migration might appear (Hendry, 2004), and reproductive isolation might be enhanced. These ecological barriers might occur through change of habitat and geographic isolation or different selections against hybrids, and they contribute to stir the acceleration of this process. The result could lead to a dramatic decline in gene flow between the two subspecies.

Interactions between species could be connected through higher trophic levels (Vamosi, 2004). Divergence between sympatric species could favour specific defensive traits, because the habitat favours specific modes of defence (Abrams, 2000).
Most research which has been done on the role of natural enemies which promote variability between populations, is done on predators (MacColl, 2010), while the effect of parasites is poorly understood. If parasites expose their host significantly different between local populations, divergent selection could eventually lead to reproductive isolation between species (MacColl, 2009). This is due to genotypes in their native environment exhibiting greater fitness than genotypes in novel environment, which could cause selection against migrated species.
Gow (2007), Eizaguirre (2011) and Rundle (2003) support MacColl, suggesting parasitism could contribute as a continuous force of specie divergence, and it deserves further attention.

Evidence which parasites can act as a selective force against migrants is suggested by MacColl (2010). He mentioned multiple evidences for this process:
When a new specie occur in recipient communities, their failure is often explained by "The biotic resistance hypothesis" (Maron, 2001). The biotic resistance hypothesis is based on

introduced species which often fail to invade communities because strong biotic interactions with native species hinder their establishment and growth. Most research on this area has been done on herbivores and plants, and indicates that native herbivores can affect plant invasion by reducing plant growth, seed set and survival.
Ricklefs (2005), suggest invasion success for invaders hinges on co evolutionary relationships with predators and pathogens.
Research done on parasite-host interactions, indicates the greatest gene flow rate of hosts versus parasites, is the strongest predictor for local adaptation (Jason D.H, 2008). Local adaptation might therefore give the parasites an advantage against migrants.
Research which has been mentioned (Maron, 2001, Jason D.H, 2008), and occurrences of parasites such as avian pox virus and malaria which has devastated populations (MacColl, 2010), suggest parasitism to be an ecological factor for selection against migration when colonization takes place (MacColl, 2010).

Clarke (2011), suggest colour patterns of closely-related species or subspecies for *Gasteroteus aculeatus* (Carl Von Linne, 1758) can be compared, and give insights of how local populations adapt to their environment. Millinski (1990), suggest a hypothesis from sexual theory, where the female sexes observe male ornaments as a trait for male quality, for example, as resistance against parasites. Braithwaite (2000) results indicate female stickleback select mates based on their red colouration.

The threespine stickleback is often used as a model of exploring behavioural biology, and there has been an accelerating increase in number of papers published off this specie (Huntingford, 2009). McKinnon (2002) mention the stickleback as important in natural modelling of speciation research, and has found natural and sexual selection to be central as a driving force of reproductive isolation in this specie complex.
Vamosi (2002, 2004) and Rundle's (2003) research on limnetic and benthic three-spined stickleback under their influence of predation was likely to have a divergent effect on the two sub species regarding defensive armour, and habitat choice.
MacColl (2009) indicated different parasite burdens between sympatric three-spined stickleback species. Therefore it is reason to suspect stickleback would evolve local defensive traits toward the parasites it is exposed to (Abrams, 2000).
MacColl (2010) continued research on parasitic burden on the three-spined stickleback, indicated difference of parasitic burdens. His experiments with marine sticklebacks in fresh

water environment compared with fresh water sticklebacks, was clear, marine fish were more susceptible to fresh water parasites than native freshwater fish. As a consequence of infection, marine stickleback showed reduced growth compared to non infected sticklebacks. MacColl (2010), mention growth as related to reproductive success in fish, and concludes parasites therefore, to reduce fitness on marine fish.

Two of the major parasites for the threespine stickleback are the cestode *Schistocephalus solidus* (Müller, 1776) and the microsporidian *Glugea anomala* (Moniez, 1887). The lifecycles for these two parasites differ, as *S. solidus* has three hosts before it enables to reproduce (Giles, 1983; Hammerschmidt, 2005) and *G. anomala* only has one host (the stickleback) (Millinski, 1985). The parasites also differ on the effect they have on their host, the threespine stickleback. The cestode *S. solidus* changes the behaviour of its host by suppression of normal anti-predator behaviour (Milinski, 1985; Ness, 1999). The microsporidian *G. anomala* changes the behaviour of its host by intensifying the normal anti-predator behaviour, making the host forage at longer distances from predators than uninfected sticklebacks (Milinski, 1985).

In our experiment, we investigate the threespine stickleback (hereafter stickleback), in three different fresh water lakes which is habited with parasites or predators. One of the lakes is connected through creeks to the two other lakes. Based on this, we chose to investigate the following questions. How may fish predators and parasites influence the stickleback's morphology in these three lakes? Is there any relation between the stickleback's morphology and parasite prevalence and parasite burden? Are the two parasites *G. anomala* and *S. solidus*, when present in the same host, inflicted by each other?

2. Material and methods

2.1 Study area

Our study area is located on the west coast of Norway, in Hordaland County. In the rural district of Sveio we find the three closely located lakes were we did the sampling (Picture 1). The first sampling site is *Lake Nesavatnet*. It has the position 59°33'14.00"N, 5°26'16.56"E, and has an altitude of 16 meters above sea level. The lake has a surface area 0.66 km^2. The lake has no upstream connecting lakes, and an altitude barrier of six meter prevent introgression of sticklebacks from the downstream lake.

Lake Liavatnet, has the position 59°33'03.90"N, 5°24'54.10"E. It has an altitude of 10 meters above sea level, and has an surface area of 0.23 km^2. Lake Liavatnet is downstream from Lake Nesavatnet, separated by a 100 meter long river. Lake Liavatnet drains into Lake Vigdarvatn. The two lakes are on approximately the same altitude and are connected by a 50 meter slow running river. This makes it easy for sticklebacks and other fish to migrate between the two lakes. Lake Vigdarsvatn is not included directly in this study, but information of its stickleback population and other fish populations are available.

Lake Kvernavatnet, has the position 59°33'18.03"N, 5°27'15.68"E. It has an altitude of 16 meters above sea level and a surface area of 0.05 km^2. Lake Kvernavatnet is located near Lake Nesavatnet, but they are not connected, and Lake Kvernavatnet has no other upstream lakes.

Picture 1: Overview of the three lakes. Kvernarvatnet (59°33'18.03"N, 5°27'15.68"E) to the right, Nesavatnet (59°33'14.00"N, 5°26'16.56"E in the middle and Liavatnet (59°33'03.90"N, 5°24'54.10"E) to the left (Google earth).

2.2 Sampling

Sampling was done during August and September in the year 2010. In Lake Nesavatnet, samples were collected from two different sites. In Lake Kverna- and Liavatnet, samples were collected from one site.

The sticklebacks were collected using a plastic fry trap (Breder, 1960). After sampling, the samples were preserved in 70% ethanol.

2.3 Predatory fish in the three lakes

The investigation for fish predators was done during the same hours of the day in both Lake Kverna- and Nesavatnet. This was done by Tom Klepaker and Per Jakobsen, using 12 gill nets varying in mesk size (22mm and 54mm in diameter).

In Lake Kvernavatnet, it was found two fish species, trout and arctic char. Trout is a well known predator for sticklebacks (Vamosi, 2002). Arctic char may also be a possible predator. However, as the arctic char is small in size, it may not be as effective predator on sticklebacks. The sample (Table 1) may therefore indicate that the predation level in Lake Kvernavatnet has an influence on the stickleback population.

In Lake Nesavatnet, it was found three arctic chars. It was not registered any stickleback in their stomach content. This suggests the predation level on stickleback in Lake Nesavatnet may be low.

Table 1: Number of fish and stomach content in Kvernavatnet

Kvernavatnet	Trout	Arctic char
Number of specimen found	64	3
Specimens found with their stomach contents analysed	45	2
Fish predators with sticklebacks found in their stomach contents	22	0

In Lake Liavatn, we have no catch data on fish predators. But from Støle (2000), we have data from Lake Vigdarvatnet. Since the two lakes are at the same altitude, and only separated by a short river, it is reasonable to assume that the fishes found in Lake Vigdarvatn also are found in Lake Liavatn. The earlier investigations of fish species in Lake Vigdarvatnet, showed it was populated with brown trout, arctic char, rainbow trout and Atlantic salmon.

2.4 Scoring spine and pelvic morphology

To determine the stickleback's pelvic score, we used the same method as Bell (1993). The method is used to count the pelvic score which is done by investigating each lateral side of the stickleback. Each lateral side has a pelvic score varying from 0-4 points, depending whether the stickleback has a normal or reduced pelvic. A normal pelvis would mean that the stickleback has a fully developed pelvis, which equals a full combined pelvic score (CPS=8, picture 3.1). A reduced pelvic could mean that the stickleback have an asymmetric pelvic (CPS=1,3,5 or 7). This occurs when one of the lateral sides lack a spine or a plate. A reduced pelvic can also be symmetrical (CPS= 2, 4 and 6; see picture 2).

Deciding the dorsal spine number of the stickleback was done by counting the number of spines on the stickleback's dorsal side. The normal spine number on a stickleback is three, but can vary from different stickleback populations.

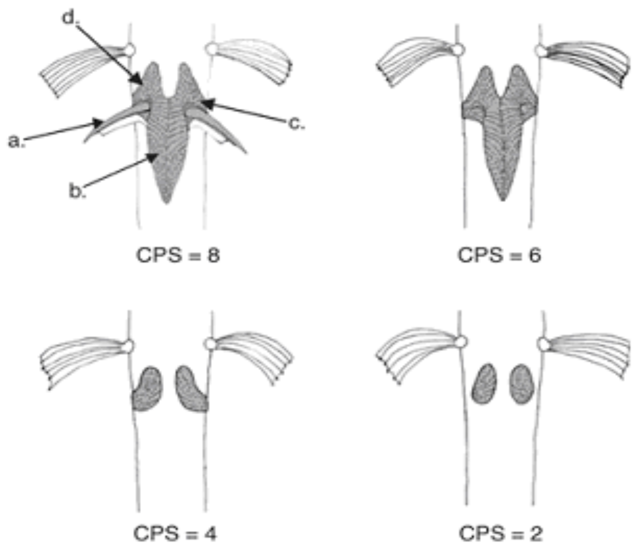

Figure 2: Displays a complete pelvic score and the reduction of a pelvis *(Klepaker, Østbye 2008).*

a = pelvic spine, b = posterior process, c = ascending branch, d = anterior process.

2.5 Separating the 0+ and 1+ generations in the sample

Before we could start analyzing the stickleback's morphology- and parasites, we had to separate the different age classes in the samples.
Length frequency analysis was used to separate the two year classes (presumably 0+ and 1+) in the samples. We used the standard fish length to measure the sticklebacks. We focused on the 1+ generation. By focusing on just 1+ generation you can pay attention to less variables which can disrupt your data, for instance the 0+ and 1+ generation can differ in amount of infections (Arnold, 2003).

From the figure of length distribution from Lake Nesavatnet (Figure 6.1 a) it was hard to decide where the cut-off point of the two generations occurred. However, by analyzing the figure from Lake Liavatnet (figure 6.2 a), we decided to separate the 0+ from 1+ generation at 30 mm.

When we do a rough separation of the two generations, error of mistakes can occur. Besides analyzing the figures wrong, the parasites can have an influence on the fish growth. For instance *G. anomala* might reduce the fish growth by reducing its food intake, and take nutrition from its body (Ward, 2005). *S. solidus*, is known to give its hosts gigantism (Arnott, 2000), which means that the host actually grows larger when infected with the parasite. This suggests my separation of the 0+ and 1+ generation is not a 100% accurate. However, it gives a good overview, consider I did not have time to check the fish ear stones, the otoliths, for accurate age analyzes.

2.6 Laboratory analyzes

Procedure for data registration for specimens in Lake Nesavatnet:

1) I dried off the ethanol, and weighed the fish.
2) Investigated the stickleback's pelvis and counted the number of spines on the fish dorsal side (DSN).
3) Counted the number of ectodermic cysts of *G. anomala*, on the fish's body surface, using a dissecting microscope.
4) Photographed all the cysts, using a Nikon D100 model, which had been set up on a rack.
5) Dissected the fish using a scissor, starting from behind the gills, and then opening it on the ventral side.
6) Investigated the fish for *S. solidus*, if the fish had been infected, I would take the plerocercoids out and put them in a marked plexi-glass with ethanol.
7) I looked for any endodermic cysts of *G. anomala*.
8) I cut off the internal organs except the sexual organs, to put them in the jar of ethanol.
9) Identified the sex for each individual.

10) Put the plerocercoids in a glass ruler which puts pressure on their body preventing them to get curled up when the picture were taken.
11) Finally, I put the parasites, fish and the internal organs in the same glass of ethanol, and marked the sample for identification.

Procedure for data registration for specimens in Lake Liavatnet:
The same procedure was done as on sample one and two, except from opening the stickleback due to lack of time during my thesis and no sign of internal infection from *S. solidus*.

Procedure for data registration for specimens in Lake Kvernavatnet:
Pelvic score and dorsal spine number was measured the same was as earlier samples. However, the stickleback was examined for all types of parasite species. This was done on twelve specimens

2.7 Computer analyses and volume calculation

After all the pictures were taken, we needed to measure the diameter of the different plerocercoids and cysts. Here I used a computer program, called Image J. With this program I could get accurate measurements of the length of the fish, circuit of the *S. solidus* and the diameter of the *G. anomala*.
To calculate the volume of the *G. anomala*, I measured the longest diameter on each cyst, and the diagonal diameter. The cyst do not have a perfect circular shape, therefore, by taking the average of those two, we would get a more accurate measurement of the diameter.
We calculated the volume of the cysts by using the formula for volume of a sphere,

$$V = \frac{4\pi r^3}{3}$$

By using the computer program, we measured the circuit around the plerocercoids. This technique allowed us to calculate the volume by using the formula (Michaud, 2006),

$$V = e^{0,279} \cdot A^{1,385} \cdot 10^{-9}$$

A= area of the plerocercoids (μm^2)

2.8 Statistical analysis

The data were analysed using STATISTICA- and SPSS statistical programs. Following analyses are used:

- One way-ANOVA: The method is used to analyse variance when you have a dependent variable with three or more categories (Crawley, 2007).
- Independent sample t- test: For comparing two independent sample means using Student's t test (Crawley, 2002).
- Chi square test: For testing hypotheses involving count data in cross tables (Crawley, 2007).
- Tukey HSD-test: Used when dealing with multiple comparisons (ANOVA) (Crawley, 2002)
- Fisher test, two by two contingency tables: Count data when there are two explanatory variables and both have just two categories (Crawley, 2007).

Excel was used to make the diagrams.

3. Results

3.1 Morphology of the sticklebacks from the different lakes:

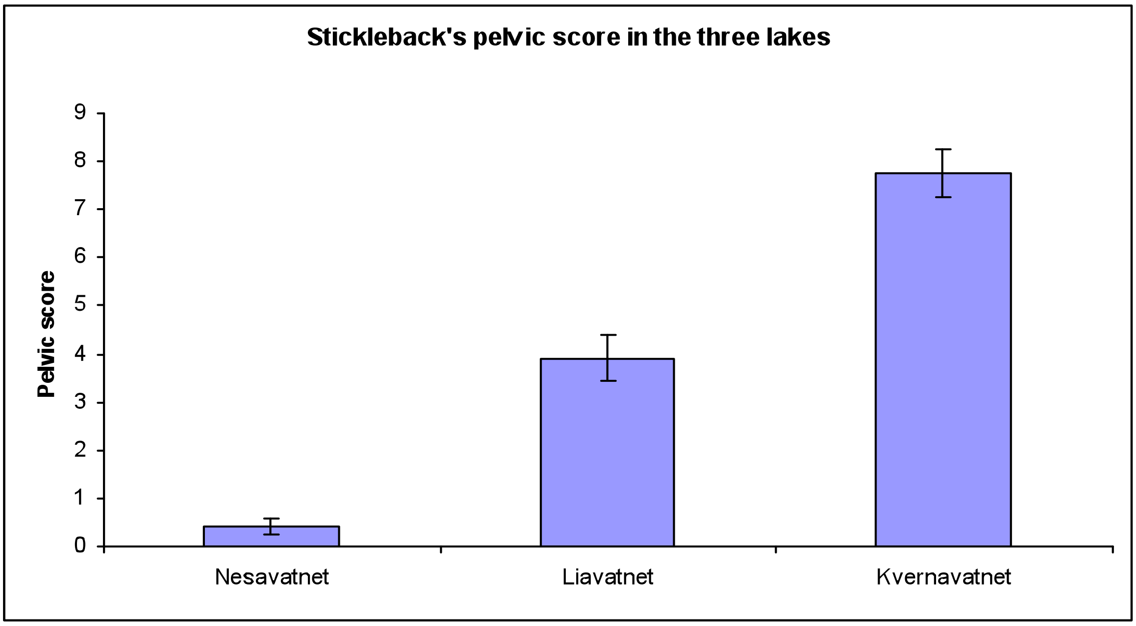

Figure 3: Stickleback's average pelvic score distributed between the three lakes with 95% confidence intervals

Figure 3 shows the average pelvic score (CPS) for the sticklebacks in the three lakes. There is a significantly difference between the three populations in the development of the pelvis (Anova, $F_{2,275} = 99.90$ $p= <0.001$).

Figure 4 gives an overview of the distribution of the CPS in the three populations. In Lake Nesavatnet, the majority of the specimens have a pelvic score equal to zero, meaning the pelvis is totally lost. No CPS higher than two is found, so in this population all specimens lack pelvic girdle and spines (Figure 2). The population from Lake Liavatnet show more variation in CPS. About a quarter of the sample have a CPS of 8, corresponding to a normal pelvis with two pelvic spines. One fifth lack any pelvic structure (CPS=0). The rest have an intermediate CPS, but CPS=1 and CPS= 2 are the most common. In Lake Kvernavatnet, all except one specimen has a CPS=8, a normal pelvic structure.

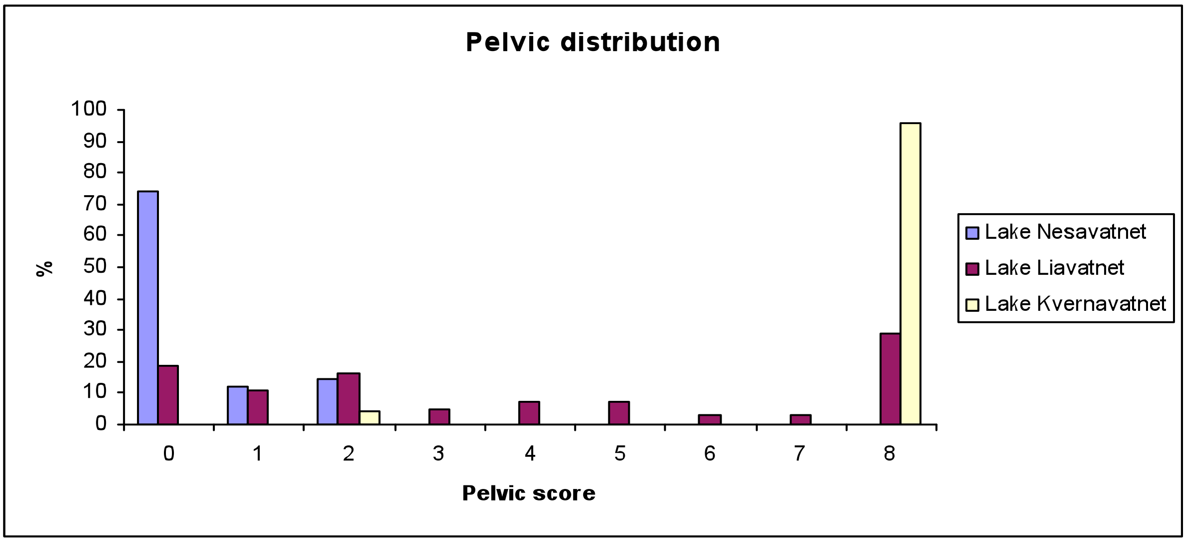

Figure 4: Pelvic distribution for the sticklebacks from the different lakes.

The number of dorsal spines also vary between the populations (Anova, F2,275 = 57.998 p= <0.001). Figure 5 show that the population in Lake Kvernavatnet has least variation, almost all specimens has three dorsal spines. In Lake Nesavatnet, more than 80 percent have lost one the three spines. Most common is loss of first or second spine. In Lake Liavatnet, a majority (80 percent) have three spines, while 20 percent have 2 spines.

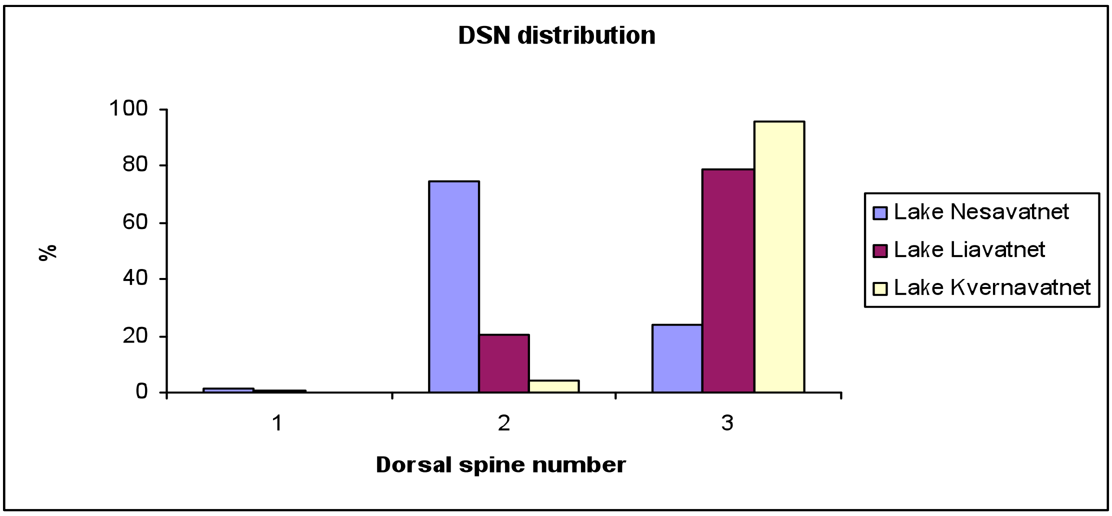

Figure 5: Dorsal spine number distribution for the sticklebacks from the different lakes.

3.2 Parasite prevalence and number of infections on the sticklebacks

Table 2 show the prevalence of *S. solidus* and *G. anomala* in the three lakes.
Lake Nesavatnet showed prevalence of both parasites. The microsporidian was found in over 73% of the specimens, and the plerocercoids were found in over 53% of the specimens. Prevalence of both parasites on the same stickleback was found in 37% of the specimens. Nearly 11% of the specimen had no sign of either *G. anomala* or *S. solidus*.
In Lake Liavatnet, we found no sign of the cestode *S. solidus*. Yet, the microsporidian *G. anomala* was found in 68% of the sticklebacks.
Lake Kvernavatnet, shows sign of infection from neither *G. anomala* nor *S. solidus*.

Table 2: Prevalence of the two parasites in the three lakes

Infection	Lake Nesavatnet (%)	Lake Liavatnet (%)	Lake Kvernavatnet(%)
G. anomala	36.7	68,0	0,0
S. solidus	16.7	0,0	0,0
Double infection	36.9	0,0	0,0
No infection	10.7	32,0	100,0
Total number of specimens	**84**	**169**	**25**

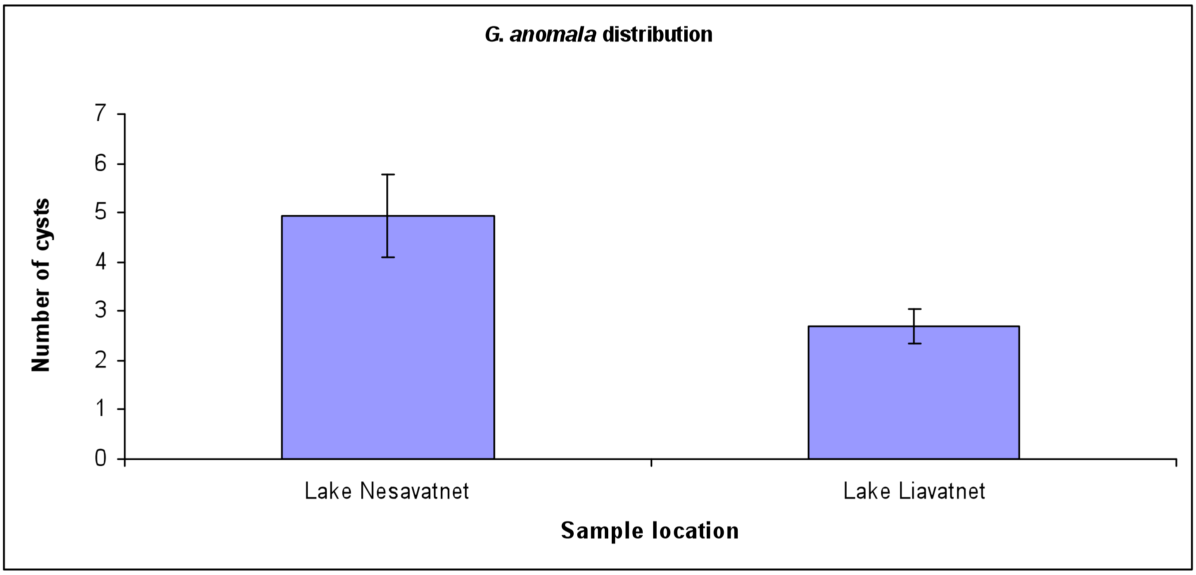

Figure 6: Average number of *G.anomala* cysts on infected sticklebacks from the different lakes with 95% confidence intervals.

Figure 6 shows the average number of *G. anomala* cysts on infected sticklebacks from the two lakes. The average number of cysts from the parasite varied between the two lakes (Anova, $F_{2,275} = 20.634$ $p= <0.001$). Sample from Lake Nesavatnet had the highest number of infections with an average of almost five cysts. Lake Liavatnet was close to three cysts per stickleback. Yet, the total volume of cysts from Lake Lia-and Nesavatnet was the same (Independent t-test, t2 =-2.09 , p= 0.835).

3.3 Parasite interaction

In the samples from the three lakes, it was only in Lake Nesavatnet that infection from both *S. solidus* and *G. anomala* occured. Investigating possible interaction between the two parasites in this population, we compared the number and total volume of plerocercoids and cysts respectively in single and double infected sticklebacks. A single infection means that the stickleback is infected by either *S. solidus* or *G. anomala*. A double infection is when the stickleback is infected by both *S. solidus* and *G. anomala*.

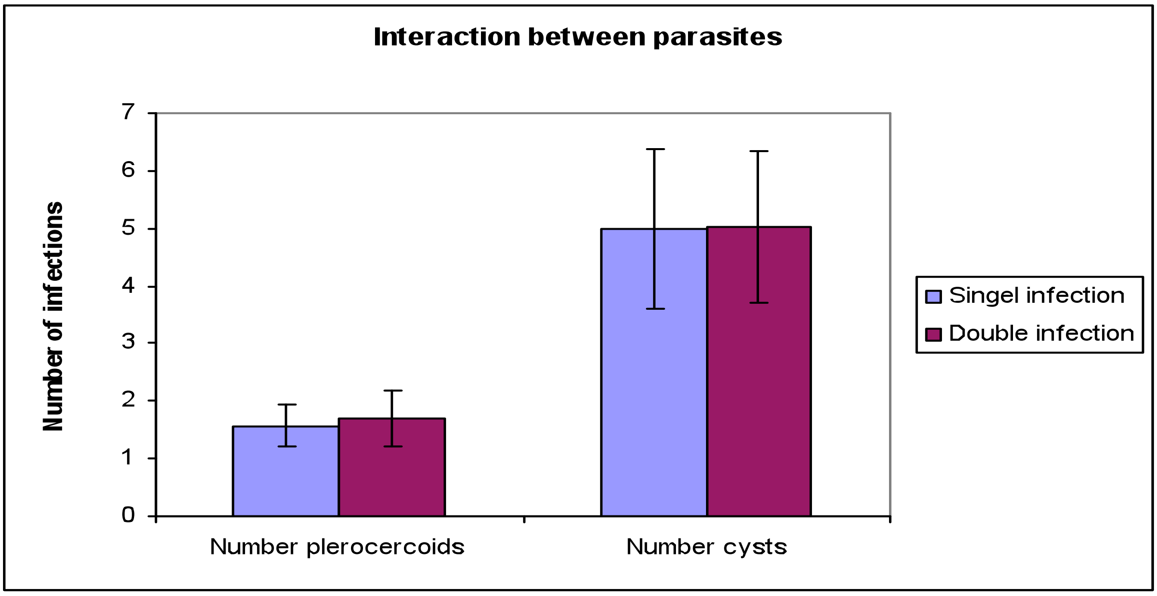

Figure 7: Average number of G.anomala cysts and S.solidus plerocercoids when sticklebacks were single and double infected with 95% confidence intervals.

Figure 7 show the number of cysts and plerocercoids when the stickleback is either single or double infected. We see that the number of plerocercoids when also infected with *G. anomala*, was slightly higher than if *S. solidus* was found alone in the stickleback. Yet, the difference in number of infections was not significant (Independent t-test, t_{43} = *-0.377, p= 0.708*). Both in single and double infected sticklebacks we found an average between 1.5 and 2 plerocercoids.

The mean number of *G. anomala* cysts when infected with *S. solidus*, was the almost the same as if it was only *G. anomala* in the stickleback (Independent t-test, t59 =-0.035, p= 0.973). Both types of infections were close to five cysts.

We then looked for differences between single and double infected sticklebacks with respect to total volume of cysts and plerocercoids (Table 3).

The total volume of plerocercoids under the prevalence of *G. anomala*, was less than if it was only *S. solidus* in the stickleback (Table 3). But, due to large variation, the difference in volume was not significant (Independent t-test, t43=0.672, p= 0.505). Both types of infections had an average total volume around 300 mm^3. The total volume of *G. anomala* cysts when infected with *S. solidus*, was the nearly same as if it was only *G. anomala* in the stickleback. Both types of infection had an average volume between 16 and 17 mm^3 and the slight difference in means was not significant (Independent t-test, t59=0.03, p= 0.976).

Table 3: Average volume of G.anomala cysts and S.solidus plerocercoids when sticklebacks were single or double infected.

	G.anomala			***S. Solidus***		
	N	Mean Total Vol. cysts	Std. Deviation	N	Mean Total Vol. Pl.	Std. Deviation
Singel infection	30	16.5 mm^3	2.45	14	314 mm^3	104
Double infection	31	16.4 mm^3	2.44	31	283 mm^3	157

3.4 Parasite relation to stickleback morphology

We see from the previous sections that the morphology and parasite prevalence differ between the sticklebacks in the three lakes. We therefore wanted to see if there are any relations between the pelvic and dorsal spine morphology and the prevalence, number and total volume of two parasites.

In the sample from Lake Kvernavatnet had almost all sticklebacks had a completely developed pelvic structure and three dorsal spines, and none of the two parasites were found. In Lake Liavatnet, the specimens showed the most variable distribution of pelvic morphs, and also dorsal spine number varied (figures 3 and 4). In this population only *G. anomala* was found and had a prevalence of 68 percent. To simplify analyses, the intermediate CPS (2-7) were grouped together, producing three pelvic morph groups; pelvis loss, pelvis reduced and pelvis normal. Comparing prevalence, we found that the pelvic reduced group had a lower prevalence (60 percent) compared to the pelvic loss group (72 percent) and the pelvic normal group (76 percent). But this difference was not large enough to produce a significant difference between the three pelvic morph groups (Chi-square test, df = 2, P= 0.158). Comparing number of cysts in infected fish, shows that the pelvic loss group has the highest mean number (3.1), followed by the pelvic reduced group (2.7), while the pelvic normal group had the lowest number of cysts (2.2). But this difference was not significant (Anova, F2,112 =1.194, P= 0.31).

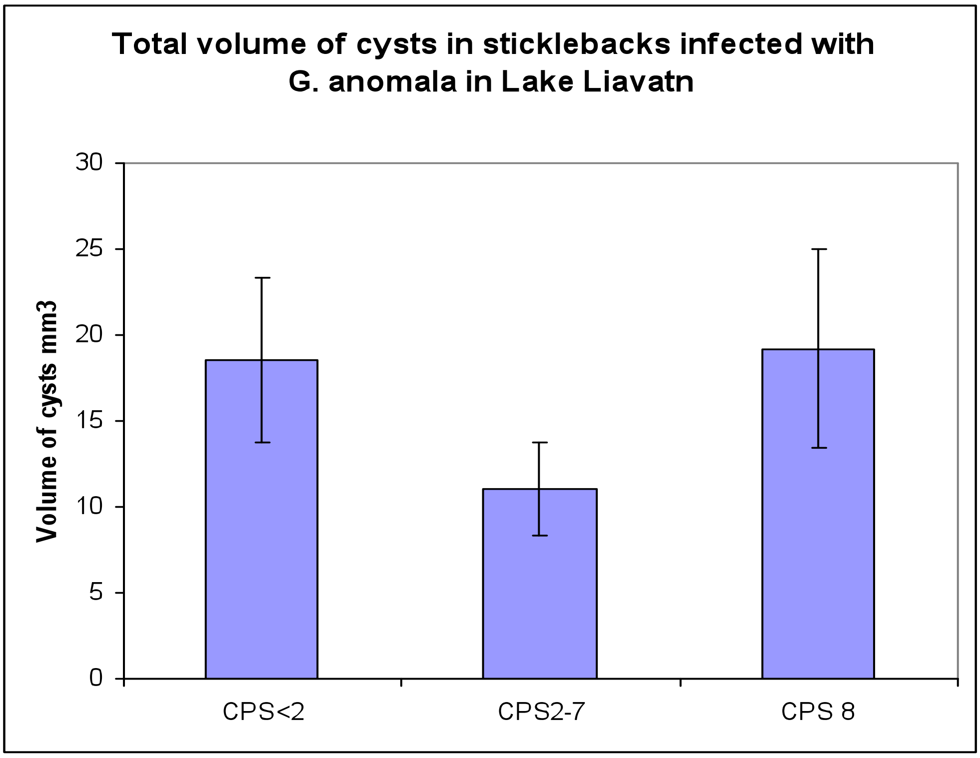

Figure 8: Total volume with 95 % confidence intervals of cysts in sticklebacks infected with G. anomala in Lake Liavatn

Comparing the total volume of the cysts in the three groups shows that the volume in the pelvic reduced group is lower than the two other groups (Fig. 8). This difference is significant (Anova, F2,112 = 3.81, P = 0.025). The difference is produced by a combined effect of higher number of cysts and larger volume of each cyst in the pelvic loss group, and a larger volume of each cyst in the pelvic normal group.

In Lake Liavatnet we find sticklebacks with 2 or 3 dorsal spines. No relation spine number and infection was found, either regarding prevalence (Fisher's exact test, df = 1, P= 0.385), number of cysts (Independent t-test, t_{112}= 0.144, p= 0.886).
In Lake Nesavatnet no sticklebacks with normal pelvis was found, and the majority had a complete loss of pelvic structure. Only CPS found was 0, 1 and 2. This population was under prevalence of both *G. anomala* and *S. solidus*. Two dorsal spines are most common, along with a smaller group with three spines. We will now analyse the parasite relation with CPS and dorsal spine numbers for both parasites.

We then investigated the stickleback's morphology related to *G. anomala*. Regarding pelvic structure, the prevalence is higher in the two lowest CPS groups, but the difference is not significant (Chi-square test, df = 2, P= 0.253). The number of cysts is highest and total volume of cysts is largest in the CPS 2 group, but the differences did not come out significant (Number of cysts: Anova, F2,81 =0.343, P= 0.711; Total volume: Anova, F2,58 =2.104, P= 0.131)

For dorsal spine number, the prevalence is higher in specimens with three spines (90 percent) than for two spines (66 percent), and the difference is significant (Fisher's exact test, df = 1, P= 0.047). The number of cysts is higher and the total volume of the cysts is larger in fish with three spines compared to fish with two, but the differences are not significant (Number of cysts: Independent t-test, t_{57}= 0.66, p= 0.510; Total volume of cysts: Independent t-test, t_{57}= 1.95, p= 0.057).

We then investigated the stickleback's morphology related to *S. solidus*. No difference between the three CPS groups regarding prevalence is found (Chi-square test, df=2, P= 0.438). The number of plerocercoids is lowest and total volume of plerocercoids is smaller in the CPS 2 group, but the differences between the groups are not significant. (Number of plerocercoids: Anova, F2,42 =1.95, P= 0.220; Total volume of plerocercoids: Anova, F2,42 =1.056, P= 0.357). For dorsal spine number, the prevalence does not differ between two and three spines (Fisher's exact test, df = 1, P= 0.444). The number of plerocercoids is higher and the total volume of the plerocercoids is larger in fish with two spines compared to fish with three, but the differences are not significant (Number of cysts: Independent t-test, t_{42}= 1.37, p= 0.177; Total volume of cysts: Independent t-test, t_{42}= 2.00, p= 0.052).

Stickleback's symmetry related to parasites in Lake Nesavatnet. We can observe a difference between the difference CPS groups. CPS=1 represent an asymmetric pelvis while the two other groups are symmetrical, we see that the proportion of not infected sticklebacks are lower and the proportion of double infected sticklebacks are higher in asymmetric sticklebacks (Table 4). However, due to small groups, the difference between sticklebacks with symmetric and asymmetric pelvis is not significant (Chi-square test, df = 6, P= 0.444).

Table 4: Parasite distribution related to stickleback's symmetry.

		CPS		
		0	1	2.0
No infection	Number	6	0	3
	%	9.7	0.0	25.0
G. anomala infection	Number	23	3	4
	%	37.1	30.0	33.3
S. solidus infection	Number	11	1	2
	%	17.7	10.0	16.7
Double infection	Number	22	6	3
	%	35.5	60.0	25.0
Total number		62	10.0	12.0

4. Discussion

4.1 Interaction between *S. solidus* and *G. anomala*

S. solidus and *G. anomala* are two parasites which are associated with the three-spined stickleback (Millinski, 1985; MacColl, 2009; Scharsack, 2007). Yet, the interaction between the two parasites when under the host, the stickleback*,* has not been paid much attention on except Arme (1967) which looked at the number of infections between the two parasites. When two manipulative parasites with conflicting life cycles meet, there is a potential conflict between them (Cezilly, 2000). When manipulative parasites infect the same intermediate host, but differ in their final host, Outreman (2002) and Millinski (1984) suggest that selection should favour the establishment of a negative association between these manipulators. Some parasites are known to develop anti-larval immunity (concomitant immunity), which means they create a barrier against continual infection and restricting burden size in their host (Brown, 2001).This strategy may occur in conspecific competition. However, since *G. anomala* and *S. solidus* are common parasites in three-spine stickleback, the possibility of the parasites developing a competing strategy towards each other may have occurred (Outreman, 2002).

Our result from Lake Nesavatn in this study show that the cestode *S. solidus* was not affected by the microsporidian *G. anomala*. Single infections by *S. solidus* did not differ significantly in number of infections - or total volume, from double infections by *S. solidus* with *G. anomala*. Single infections by *G. anomala* was neither significantly different in number of infections- nor total volume from double infections with *S. solidus*.

We did not find any relationships between the number of *G. anomala* cysts and the intensity of *S. solidus*, in Lake Nesavatnet, which correspond with Arme result (1967). What explains this result is yet unknown, but might be due to several factors. For instance, the parasites may be immunological hidden to each other. It may also be the case that they do not conflict enough to evolve concomitant immunity towards each other.

4.2 Morphology, parasites and predation on the stickleback

The results show that the three lakes have populations of stickleback which differ in pelvic structure and dorsal spine numbers. We see that sticklebacks in Lake Nesavatnet, which was the population with the highest prevalence and number of infections by both *G. anomala* and the only population with prevalence of *S. solid*us, also had the most reduced pelvic structure and the lowest mean dorsal spine number. We also observe a difference in number of *G. anomala* cysts between sticklebacks in Lake Nesavatnet and the populations from the two other Lakes.

The stickleback population in Lake Liavatnet, are polymorphic for pelvic structure; a proportion has a normal developed pelvis, another proportion has a reduced pelvis similar to those in Lake Nesavatn and the last proportion is intermediate between the two others. The sticklebacks were also second in number of cysts from *G. anomala*, and prevalence of *G. anomala* was equal to sticklebacks in Lake Nesavatnet. No *S. solidus* infections were found.

The stickleback population from Lake Kvernavatnet, which clearly had the highest pelvic score, were not infected by any of the two parasites in focus here and moreover, infections of other parasites was also lower than in Lake Nesavatnet. These results indicate different ecological characters for each lake, possibly because habitat selection can differ and favour other modes of defence (Abrams 2000). Therefore, we will discuss which selection pressures may influence the stickleback populations.

Lake Kvernavatnet is an isolated lake which prevent any gene flow and hence the population is undisturbed by introgression from other populations. We have data indicating a high level of predation from trout. Trout is a well known predator on stickleback (Vamosi, 2002), which can influence anti predator traits of sympatric sticklebacks (Vamosi, 2004). High selection pressure from trout could explain why Kvernavatnet separates from the two other lakes, with all (except one specimen) having a normal pelvic structure. A stickleback population which is under high predation pressure is less likely to reduce investment in pelvic armour. Armour like spine and plates may increase probability to survive predator encounters, by making themselves more difficult to handle and ingest (Grand, 2000). Additionally, Grand (2000) found a negative correlation between armour and chance to give up food to avoid predator.

This suggests that the stronger pelvic structure the stickleback equipped with, the longer it will maintain feeding when confronted by a predator.

In the open water the probability of stickleback feeding on copepods is higher (MacColl, 2009; Vamosi, 2002). MacColl (2009) suggests this would increase the stickleback probability to transference of cestode, *S. solidus*, from the copepod. This is because copepods are *S. solidus* first host during its life cycle (Giles, 1983; Hammerschmidt, 2005) . However, our results show no infection of S.solidus in our sample from Lake Kvernavatnet.

An experiment done by M. Millinski (1985) indicated that sticklebacks which was infected with *S.solidus*, ate closer to cichlids, fled less far and recovered more quickly after an attack, than uninfected stickleback. The same response behaviour occurs through interaction with avian predators (Giles, 1983). Barbers (2004) experiment indicated stickleback's behaviour to be normal during the first seven weeks after infection by *S. solidus*. After 9-15 weeks, the plerocercoids have reached its infective size, which is 50mg. During this period the stickleback gets slower to reach for cover and to make a directional response. The manipulation of *S.solidus* increases the host's vulnerability to predation when the host has at least one competent plerocercoid, which weighs more then 50mg (David C Heins, 2002; Barber, 2004). Scharsack (2007) question whether the monoamine neurotransmitters, which might cause this behaviour, are produced by the parasite, or are a result of a chronic stress reaction by the host.
S. solidus is also affecting the sticklebacks shoaling behaviour (Barber, 1995). The advantage of shoaling, are known to be protection against predation and enhanced food detection. However, it also increases food competition since they are in a group (Barber, 1995). Barbers (1995) experiment indicated that sticklebacks infected with *S. solidus* spend less time in a shoal, than non-infected sticklebacks. Therefore, he concluded it could have consequences for the parasite on the transmission to the final host.

Experiments done by Ness (1999) indicate that infection from *S.solidus* changes behaviour of the host to suppression of normal anti-predator behaviour. The suppression was greatest in demelanized fish, which points to rapid change of colouration of the hosts' body, making it more visible to predators. Simulated attacks on infected stickleback by trout showed less response, fled shorter distances and recovered more rapidly then uninfected ones.

Consequences of the effect which the parasite has on its host, Ness (1999), suggest the parasite's life cycle might enhance vulnerability to predators which are not suitable hosts. Cezilly (2010) support the suggestion from Ness (1999), were *S. solidus* populations may get reduced in lakes where predatory fish are common.
Investigation over a period of five years, showed a rapid decline of stickleback infected by *S.solidus* as a likely effect of introduced Atlantic salmon (Jakobsen, 1987). Therefore, a possible explanation in Lake Kvernavatnet is that the abundant trout population diminished the *S. solidus* population to the level where it was not able to sustain it self.

No infections of the microsporidian *G.anomala* was found in Lake Kvernavatnet. However, *G. anomala* has a different life cycle from *S. solidus* which makes it independent of multiple hosts in order to reproduce (Millinski, 1985). Therefore it might be easier for the parasite to survive under different habitats, as *G.anomala* is known to habitat salt/brackish water (MacColl, 2010) and the benthic and limnetic zone in fresh water (MacColl, 2009). The behaviour it inflicts the stickleback is also very different from how *S.solidus* influence their host.

An experiment has shown infected stickleback which forage at longer distances from predators, than uninfected fish (M. Milinski, 1985). It has also been shown through experiments that infected sticklebacks show higher tendency to shoal then uninfected sticklebacks. Also, while shoaling the fish spends most time in the group, at the front position in the shoal (Ward, 2005). By joining a group, the *G. anomala* or host decreases it chance to get eaten (Barber, 1995). By occupy the front position of the shoal, it also increases the availability of food (Ward, 2005). Earlier findings suggest that *G.anomala* may increase their host chance to survive. However, an infection could cause severe damage to respiratory organs, sensory function and locomotory movements (Arme, 1967), which could make them more vulnerable to predators. As for *S. solidus*, it is difficult to conclude if predators have terminated the *G.anomala* population, due to no earlier research. The parasite might never have existed in Kvernavatnet, though it seems probable as the neighbour lakes are habited with *G. anomala*. If a population have existed there, a possible scenario may be that the predation pressure has led to a decline of the *G. anomala* population where it could no longer be sustainable.

Lake Liavatnet is not an isolated lake as Lake Kverna- and Nesavatnet. Being connected to two other lakes (Lake Nesavatnet and Lake Vigdarvatnet) makes Lake Liavatnet a connection point for where sticklebacks from different populations meet. Most of the specimen in Lake Nesavatnet did not have any pelvic armour, and none have a fully developed pelvis (highest CPS=2) in the lake. Klepaker (Unpublished data) investigation on sticklebacks from Lake Vigdarvatnet, show that this population has a normal complete pelvis and the normal three dorsal spines. The reason for the polymorphic distribution of the pelvis among the stickleback population in Lake Liavatnet may be the introgression from both Lake Nesavatnet with strongly reduced armour and Lake Vigdarvatnet with normal armour and hybridization between them.

We know from earlier investigations that Lake Vigdarvatnet is populated with brown trout, arctic char, rainbow trout and Atlantic salmon (Støle, 2000). Since Lake Vigdarvatnet and Lake Liavatnet are connected by a small slow running river (Picture 2, appendix), it is likely to assume Lake Liavatnet has the same fish predators as Lake Vigdarvatnet. Moreover, it is unlikely that fish eating birds, the final host of the tapeworm *S.solidus* (Smyth 1949), are absent in the Lake Liavatn so other ecological factors must explain the absence of *S.solidus* in Lake Liavatn.

With only one sample we can not say for certain that *S. solidus* is not prevalent in Lake Liavatnet. However, since Lake Liavatnet likely is populated with fish predators, fish predation can be a plausible cause for the absence of sticklebacks infected by *S. solidus* in Lake Liavatnet, as it were in Lake Kvernavatnet. But this raises the question why *S. solidus* is present in Lake Vigdarsvatnet (Klepaker, Unpublished data). A possible scenario could be that in Lake Liavatnet the predation from piscivorous fish is slightly higher than in Lake Vigdarvatnet. Therefore, *S. solidus* disappears first and *as G. anomala* is more predation resistant (Milinski, 1984), manages to survive. However, it is not present data which support this assumption. Yet, we find an almost equal prevalence of *G. anomala* in Lake- Liavatnet and Nesavatnet, even though *G.anomala* infections in Lake Liavatnet had a significantly lower number of cysts on each infected stickleback. We did not find any fish predators in our sample in Lake Nesavatnet, although there has been registered arctic char in the lake. Hence, a possible scenario may be the selection pressure from fish predators in Lake Liavatnet, is strong enough to reduce the *S. solidus* population. While the impact on *G. anomala* is lower.

Compared with the sticklebacks in Lake Nesavatnet, it may not be strong enough to reduce the prevalence of *G. anomala* significantly, but possibly the intensity on the sticklebacks.

Living in the benthic zone, armour can be less beneficial as invertebrates can use spines and lateral plate to get better a grasp of the stickleback's body (Vamosi, 2004). However, in the limnetic zone, sticklebacks get more exposed to fish predators, and the armour helps by increasing post-capture escapes (Hoogland, 1956; Vamosi, 2002). Sticklebacks in Lake Liavatnet contain both none pelvic (CPS=0) and complete pelvic (CPS=8). When different morphology traits can give reproductive success in different habitats, it may indicate that sticklebacks are facing different habitat-specific selection, which can occur by living in the benthic and limnetic zone of a lake (Vamosi, 2004). Based on research from Vamosi (2002; 2004), a possible explanation to the polymorphic population in Lake Liavatnet is a division in benthic and limnetic forms. However, we have no further present information to support this explanation for the polymorphic distribution of pelvic morphs in Lake Liavatnet.

Lake Nesavatnet has no inflowing brooks from other lakes, which makes gene flow from other populations unlikely. As the stickleback population is an allopatric population, the stickleback's morphology can give a good indication for the ecological forces in the lake (Vamosi, 2004). Lake Nesavatnet, as mentioned earlier, has a probably low predation pressure from fish on stickleback. Predatory fish are known to restrict odonates to littoral cover where salmon consume nymphs across open substrate (Reimchen, 1979; Bell, 1993). If Lake Nesavatnet has few predatory fish, the reduced selection press may allow greater densities of macro invertebrates to spread out of their littoral habitat. Armour can be a liability as invertebrates use spines and lateral plates to get a better grasp on the stickleback (Vamosi, 2002). Then a combined effect of low fish predation and high macro-invertebrate predation may have contributed to the significantly lower pelvic score and dorsal spine number for the sticklebacks in Lake Nesavatnet compared to the two other lakes. In addition to pelvic reduction, stickleback population in Lake Nesavatnet also differed from the two other populations by having a significantly fewer dorsal spines, which also support the assumption of a relaxed predation from fish predators.

Experiments done on marine and freshwater sticklebacks, which differ in body armour show that low armour sticklebacks grew faster when juveniles, than heavy armour sticklebacks (Marchinko, 2007). Both marine and freshwater sticklebacks were raised in fresh water.

Faster growth rate seemed to have a positive effect on the stickleback. Faster growing stickleback had greater lipid stores and lower metabolism, which may be why they had higher rates of overwinter survival. They also attained larger body length which is correlated with reproductive potential and decreased predation attacks (Marchinko, 2007). In fresh water communities, benthic organisms are known to have a faster growth rate than limnetic organisms (Vamosi, 2002). This may be due to less investment in body armour. If the loss of pelvic and dorsal spine can increase growth as a result of reduced energy costs and give the same advantages as Marchinko (2007) suggested, it may work as a selective force in reduced armour. These studies (Reimchen 1979; Vamosi, 2004; Marchinko, 2007) show that if the sticklebacks in Lake Nesavatnet do not get any additional protection from armour, it is beneficial from developmental reasons to reduce pelvis and spine numbers.

Lake Nesavatnet, was heavily infected by the cestode *S. solidus* with more than half the samples were infected. The lake also had the most profound *G. anomala* prevalence, and in addition a higher number of cysts per fish compared to the two other lakes in the study. Such heavy infection, some fish with more than ten cysts, probably will have a dysfunctional effect on their body (Arme, 1967), which may increase the predation risk. This, combined with studies that show that fish predation lower the prevalence of *S. solidus* (Jakobsen, 1987), indicate an lake with low fish predation. This may be a plausible scenario in Lake Nesavatnet. Moreover, in a lake which is heavily populated with the parasites in focus and as sticklebacks are not known to have any behavioural defence against *S.solidus* (Wedekind, 1996). It is possible there could be a selection from female stickleback's mate choice regarding MHC (Major histocompatibility complex) genes (Eizaguirre, 2011). The worms are affected by the stickleback MHC genetic make up, by inhibiting the parasite growth (Scharsack, 2007).

MacColl (2010) introduced marine sticklebacks to fresh water community of parasites and compared the infection rate with fresh water stickleback. The experiment indicated that marine fishes which were introduced to freshwater were more susceptible to freshwater parasites than the native freshwater fish. *G. anomala* infections were not significantly different between the two stickleback populations. This may be due to the fact that the parasite is abundant in both marine- and fresh water environment, suggesting that the marine water stickleback already have developed resistance mechanism against the parasite (MacColl, 2010). This indicates that resistance mechanism against *G.anomala* may be common trait in different stickleback populations. It may be a plausible explanation for why sticklebacks in

Lake Nesavatnet and Lake Liavatnet had the same prevalence of *G. anomala* infections. When comparing lakes it is less plausible that ecological factors promoting or inhibiting infections are the same.

4.3 Parasite relation to stickleback morphology

Armour morphology of sticklebacks is documented to relate to predation (Grand, 2000), but less is known about any relation between armour morphology and parasites.
In our study, we observe that the stickleback population in Lake Kvernavatnet has a normal pelvis (except one specimen). The population in Lake Liavatnet has a polymorphic pelvic distributed, with the dominating part being a normal- and fully reduced pelvic. Sticklebacks from Lake Nesavatnet, was dominated by a fully reduced pelvic.

When we look at the parasite distribution for the stickleback in the different lakes, we see that Lake Kvernavatnet does not have any infection from the two parasites. Population in Lake Liavatnet, has a high prevalence of *G. anomala*, but no infection from *S. solidus*. Sticklebacks from Lake Nesavatnet have a high prevalence of both parasites in focus.
Here, we see overall pattern of morphology and parasite prevalence. Sticklebacks in Lake Kvernavatnet have normal pelvic and no indication of the two parasites in focus. On the opposite side of the scale, we have the sticklebacks from Lake Nesavatnet, with an almost completely reduced pelvis and high prevalence of parasites. Sticklebacks from Lake Liavatnet has a polymorphic pelvic and a parasite distribution which is lower than sticklebacks from Lake Nesavatnet, but higher than sticklebacks from Lake Kvernavatnet. As we have pointed out earlier, predation from fish may have a large part of the reason for this outcome. However, we wanted to find out if there may be a relation between the stickleback's morphology and parasite prevalence within the populations.

For the sticklebacks from Lake Liavatnet, the prevalence, number of cysts and total volume of *G. anomala* were investigated to see if there were any relations to the stickleback's morphology. We divided the sticklebacks from Lake Liavatnet in three groups, CPS= 0-1, 2-7 and 8. When comparing these three groups, we saw that total volume of cysts with CPS=2-7 was lower than the two other groups. This group had also the lowest prevalence, and was second in number of cysts. These differences were not significant, but this can be explained by small group sizes. If this difference is real, different explanaitions are possible. Our present result may indicate that if there is introgression in Lake Liavatnet from Lake Vigdar- and Nesavatnet. These hybrids may represent a better distribution of MHC genes, which could result in a better immune system against *G. anomala*.

In addition to Lake Liavatnet, we studied the relation between morphology and parasites on sticklebacks in Lake Nesavatnet. As mentioned earlier, the sticklebacks from Lake Nesavatnet were infected by both parasites in focus. We divided the sticklebacks in three groups after pelvic score (CPS= 0,1 and 2). The prevalence of the two parasites was inverse with respect to pelvis score. S. solidus was most prevalent in sticklebacks with CPS=2 and least prevalent in CPS=0. G. anomala was least prevalent in CPS=2. However, none of these differences came out statistically significant. A possible reason for that the dominating proportion of the stickleback's had completley reduced ,and two other groups in the sample were low in numbers, making any conclusions of differences between groups uncertain. So, larger samples are necessary to assess any relation between pelvic morphs and the parasites in Lake Nesanvatnet.

The parasite relation to dorsal spine number was also studied, in Lake Nesavatnet. Here, we came out with a significant value for prevalence of *G. anomala*. Three dorsal spines were more susceptible to parasite infection than two dorsal spines. Number of cysts and total volume were also higher in three dorsal spines, but the result was not significant.
For the parasite, *S. solidus*, none of the result came out significant. However, for number- and total volume of plerocercoids, the value was higher for two dorsal spines. As for the pelvic morphs, the two parasites distribute inversely with respect to dorsal spine numbers. Even if the results are far from conclusive, they point in an interesting direction of parasite – stickleback armour relation, worth a further study.

5. Conclusion

As ecology has been seen as an important factor in speciation (Orr, 1998), we tried to see what possibilities each lake presented to give the stickleback their different morphological traits. The sticklebacks in two of the lakes (Nesa- and Kvernavatnet) are reproductive isolated from other populations. The morphology of the two populations are quite opposite of each other. Sticklebacks in Lake Kvernavatnet have a complete pelvis and three dorsal spines, and the population in Lake Nesavatnet has mostly lost pelvis and just two dorsal spines. The last population, in Lake Liavatnet, is a mix of different pelvic morphs from pelvic loss to normal pelvis. It is likely to explain this mix from gene flow from both Lake Nesavatnet (pelvis loss) and Lake Vigdarsvatnet (normal pelvis), and that the intermediate pelvic morphs represent hybrids. This explanation is strengthen by the dorsal spine number in Lake Liavatnet being intermediate to the two other lakes

When looking at how fish predators may have shaped this different distribution in morphology between the three populations, we found indications of stickleback being under predation by trout in Lake Kvernavatnet. In Lake Nesavatnet we found no fish predators present, which may be interpreted as a low predation pressure on stickleback from fish. These interpretations also concur with the armour morphology of the sticklebacks in the two lakes. However, in the third lake in the study, Lake Liavatnet, the stickleback population is more polymorphic with respect to armour morphology. Since the fish predators from Lake Vigdarvatnet are likely to "migrate" to Lake Liavatnet and moreover Liavatnet has spawning brooks for trout, we should expect that the sticklebacks in Lake Liavatnet would resemble those in Lake Vigdarvatnet, who have a normal pelvis. But, since the Lake Liavatn population have a significant proportion of pelvic reduced specimens, this may indicate that the stickleback population in Lake Liavatnet are not as exposed to fish predators as presumed. But if fish predation is relaxed, we should expect to find prevalence of the cestode *S. solidus.* However, we found no sign of the parasite in our sample from the lake. A possible explanation for the presence of a large proportion of low armour sticklebacks in Lake Liavatnet can be that the gene flow entering Lake Liavatnet from Lake Nesavatnet is so high that it overrides the effect of predation. The absence of *S. solidus* in our specimens is difficult to explain by other factors than the predation pressure from non-host predators (fish) in the lake.

More data needed before we can draw clear conclusions. Among there is a lack of evidence regarding the role of predators. Both predators and sticklebacks should have been sampled at different sites in the lakes during season and samples from more than one year would have been included, regarding the role of morphology and parasitism.

We also examined the hypotheses whether there was a relation between morphology and parasite prevalence for sticklebacks in Lake Lia- and Kvernavatnet. The stickleback population in Lake Liavatnet, the pelvic reduced group had the lowest total volume of G. anomala cysts. May be due to fish predation pressure, or possibly because the group are hybrids made from introgression in Lake Liavatnet, which makes them less susceptible to G. anomala than the two other groups. The stickleback population from Lake Nesavatnet, we found a difference in prevalence with respect to morphology, but failed to prove these to be statistically significant. However, these results may indicate a relation between the two parasites which has not been look at, but more research is needed to conclude whether there are any interactions between the two parasites and the stickleback's morphology.

The last aim of the study was to test if *G. anomala* and *S. solidus*, when present in the same host, inflicted on each other. Comparing the parasites in single- and double infected sticklebacks produced no indication of parasite inflection. Neither number of infections (cysts and plerocercoids) nor total parasite burden (volume) differed when the parasite was single infector compared to sharing the host with the other parasite. This indicates that the two parasites have not developed concomitant immunity against each other, at least not in the studied lake. However, more research is needed to conclude whether there are any other interactions between the two parasites, or if such interaction varies between different populations of parasites and sticklebacks.

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Appendix

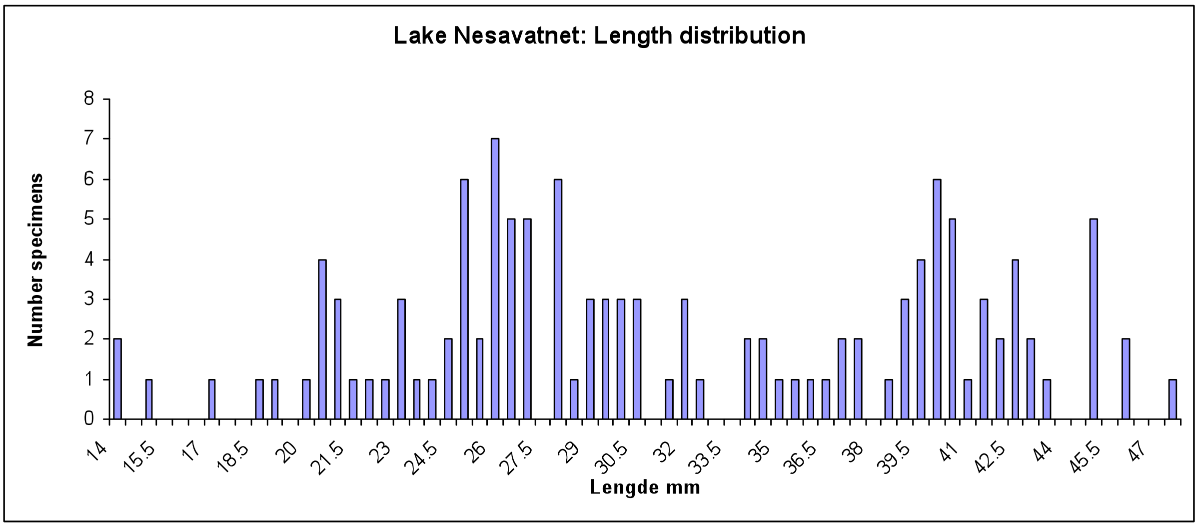

Figure 9: Length distribution of sticklebacks in Lake Nesavatnet

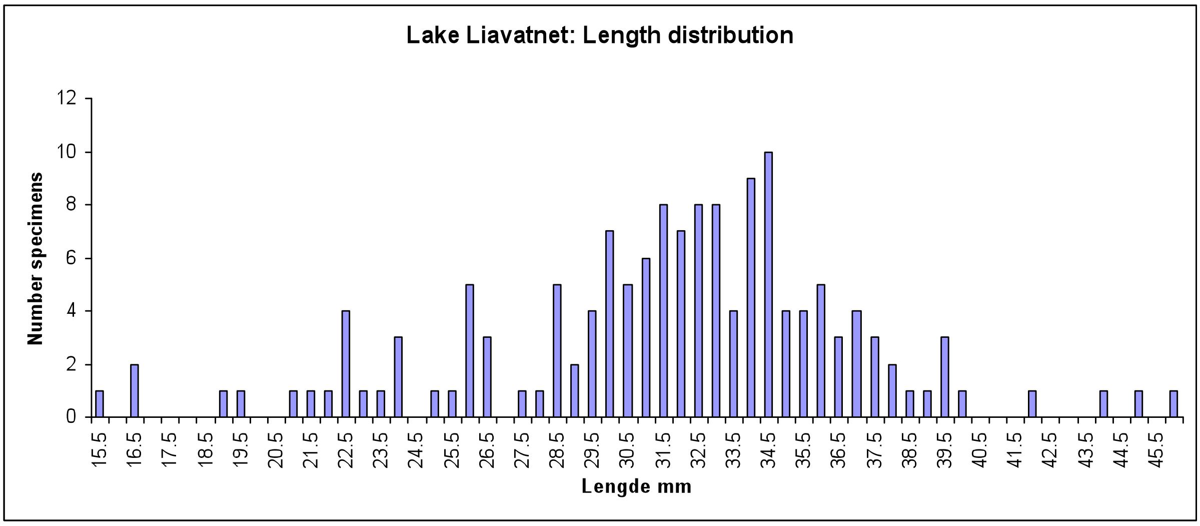

Figure 10: Length distribution sticklebacks in Lake Liavatnet

Picture 2: Connection point between Lake Vigdar- and Liavatnet.

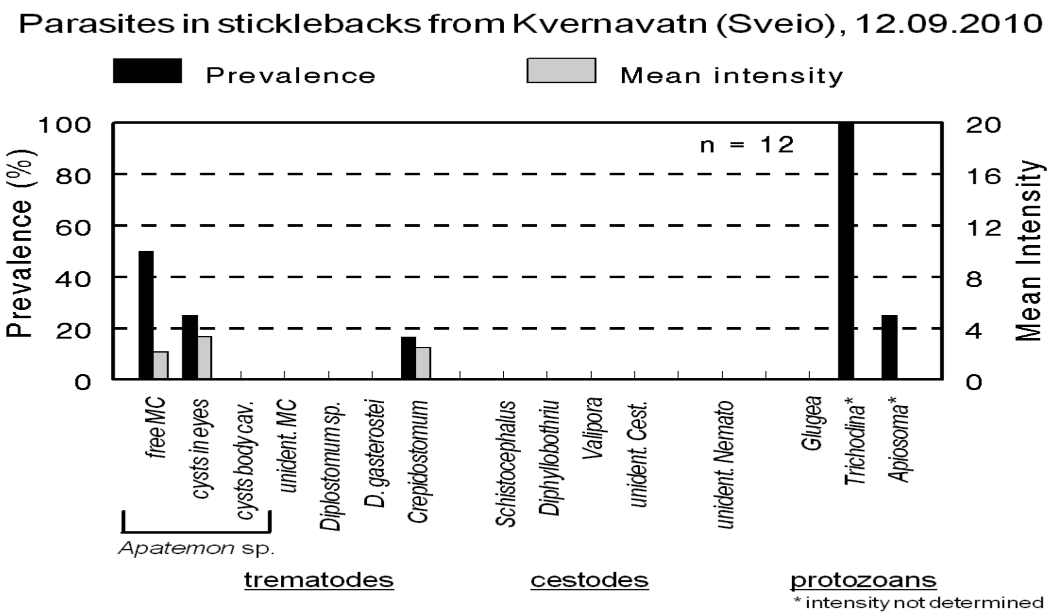

Figure 11: Show the diversity of parasites found in Lake Kvernervatnet, investigated by Martin Kolbe

Table 5: Raw-data for the threespine stickleback morphology, including sample number.

nummer	sample	Lengde	Vekt	Kjønn	infeksjon	CPS	vs	hs	DSN	N/U	dist cps
1	1	37.4	0.61	0	3	0	0	0	2	0	0
2	1	43.5	1.17	1	3	0	0	0	3	0	0
3	1	40.5	1.05	0	2	0	0	0	2	0	0
4	1	44.1	0.82	0	3	0	0	0	2	0	0
5	1	41.6	0.89	1	3	1	1	0	2	0	0
6	1	49.5	1.2	0	3	0	0	0	2	0	0
7	1	46.8	1.35	1	2	0	0	0	2	0	0
8	1	45.9	1.05	1	1	0	0	0	2	0	0
9	1	45.3	1.27	0	3	0	0	0	2	0	0
10	1	45.5	1.17	0	3	0	0	0	2	0	0
11	1	39.9	0.78	0	3	0	0	0	2	0	0
12	1	45.5	1.48	0	3	0	0	0	2	0	0
13	1	47.6	1.2	0	2	2	1	1	2	0	0
14	1	42.9	1.11	0	3	0	0	0	2	0	0
15	1	45.4	1.35	0	2	0	0	0	2	0	0
16	1	39.0	0.8	0	2	0	0	0	2	0	0
17	1	40.0	0.91	0	2	0	0	0	2	0	0
18	1	40.0	0.82	0	3	0	0	0	3	0	0
19	1	41.9	0.93	0	3	1	1	0	2	0	0
20	1	42.4	0.82	0	3	1	0	1	2	0	0
21	1	46.8	1.95	0	3	0	0	0	2	0	0
22	1	39.9	0.65	1	1	0	0	0	2	0	0
23	1	38.3	0.5	1	1	0	0	0	1	0	0
24	1	39.5	0.76	0	3	2	1	1	3	0	0
25	1	40.3	0.87	0	2	1	1	0	2	0	0
26	1	43.1	1.13	0	3	0	0	0	3	0	0
27	1	42.5	1.04	0	3	0	0	0	2	0	0
28	1	38.4	0.8	0	3	1	1	0	3	0	0
29	1	42.0	0.87	0	3	0	0	0	3	0	0
30	1	41.4	0.94	0	2	0	0	0	2	0	0
31	1	41.6	1	1	3	1	1	0	2	0	0
32	1	39.6	0.79	1	3	0	0	0	2	0	0
33	1	43.5	1.27	0	2	0	0	0	2	0	0
34	1	41.8	0.95	0	2	0	0	0	2	0	0
35	1	37.9	0.69	0	3	0	0	0	2	0	0
36	1	40.4	0.98	0	3	0	0	0	2	0	0
37	1	39.1	0.74	0	3	0	0	0	3	0	0
38	1	39.9	0.78	0	3	2	1	1	1	0	0
39	1	42.5	1.07	0	2	0	0	0	2	0	0
40	1	41.5	0.93	0	3	0	0	0	2	0	0
41	1	40.5	0.78	0	3	0	0	0	2	0	0
42	1	41.2	0.94	0	2	2	1	1	2	0	0
43	1	40.6	0.94	0	2	0	0	0	2	0	0
44	1	40.7	1.06	0	3	0	0	0	2	0	0
45	1	37.0	0.48	0	1	0	0	0	2	0	0
46	1	34.5	0.45	0	1	0	0	0	2	0	0
47	1	36.3	0.46	0	1	1	1	0	3	0	0

48	1	34.8	0.42	1	0	0	0	0	2	0	0
49	1	35.6	0.69	0	3	1	1	0	2	0	0
50	1	34.8	0.49	1	1	2	1	1	3	0	0
51	1	37.2	0.57	0	1	0	0	0	3	0	0
52	1	36.7	0.5	0	0	0	0	0	2	0	0
53	1	32.5	0.47	0	2	0	0	0	2	0	0
54	1	35.0	0.39	0	1	0	0	0	2	0	0
55	1	31.9	0.31	0	1	1	0	1	3	0	0
56	1	33.2	0.51	1	3	2	1	1	3	0	0
57	1	35.6	0.47	0	1	0	0	0	2	0	0
58	1	33.5	0.36	0	1	0	0	0	2	0	0
59	1	35.0	0.38	0	1	0	0	0	3	0	0
60	1	31.8	0.32	0	3	0	0	0	3	0	0
61	1	31.8	0.28	0	1	0	0	0	3	0	0
62	1	33.3	0.31	1	1	0	0	0	3	0	0
63	1	34.5	0.33	0	0	0	0	0	2	0	0
64	1	32.5	0.29	0	0	0	0	0	2	0	0
65	2	37.8	0.48	0	0	2	1	1	3	0	0
66	2	37.5	0.48	0	1	2	1	1	3	0	0
67	2	32.5	0.32	0	1	0	0	0	2	0	0
68	2	32.2	0.36	0	1	0	0	0	2	0	0
69	2	36.2	0.46	0	1	0	0	0	2	0	0
70	2	34.4	0.38	0	1	0	0	0	2	0	0
71	2	40.7	0.68	1	0	2	1	1	2	0	0
72	2	40.0	0.55	0	0	0	0	0	2	0	0
73	2	38.7	0.53	0	1	0	0	0	2	0	0
74	2	31.8	0.27	0	1	0	0	0	2	0	0
75	2	37.7	0.47	0	0	0	0	0	2	0	0
76	2	39.4	0.58	0	1	1	1	0	2	0	0
77	2	37.0	0.47	0	1	2	1	1	2	0	0
78	2	36.9	0.46	1	1	0	0	0	2	0	0
79	2	31.6	0.24	0	0	2	1	1	3	0	0
80	2	34.3	0.38	0	1	0	0	0	2	0	0
81	2	32.0	0.33	1	1	2	1	1	2	0	0
82	2	33.2	0.33	0	1	0	0	0	2	0	
83	2	36.5	0.39	0	1	0	0	0	2	0	
84	2	33.2	0.39	0	1	0	0	0	3	0	
85	3	36.809	0.27		1	2	1	1	3	0	
86	3	33.894	0.25		1	1	1	0	2	0	
87	3	34.71	0.3		0	4	2	2	2	0	
88	3	36.459	0.28		1	8	4	4	3	1	
89	3	33.341	0.26		0	5	4	1	2	0	
90	3	37.027	0.37		1	1	0	1	3	0	
91	3	36.852	0.41		1	8	4	4	2	1	
92	3	38.601	0.39		0	2	1	1	3	0	
93	3	37.246	0.35		1	8	4	4	2	1	
94	3	38.339	0.39		1	0	0	0	3	0	
95	3	33.53	0.3		1	8	4	4	2	1	
96	3	38.936	0.43		1	7	3	4	3	0	
97	3	35.789	0.33		1	2	1	1	3	0	
98	3	33.025	0.33		1	0	0	0	3	0	
99	3	36.481	0.34		1	1	0	1	3	0	
100	3	36.395	0.46		0	0	0	0	3	0	
101	3	35.88	0.38		1	2	1	1	2	0	

102	3	35.416	0.36	1	2	1	1	3	0
103	3	34.681	0.37	1	8	4	4	3	1
104	3	34.652	0.33	1	1	0	1	3	0
105	3	31.135	0.23	0	0	0	0	3	0
106	3	33.341	0.33	0	2	2	0	3	0
107	3	35.229	0.35	1	0	0	0	3	0
108	3	35.978	0.35	1	3	2	1	3	0
109	3	31.423	0.26	1	1	1	0	3	0
110	3	33.268	0.28	1	8	4	4	3	1
111	3	34.479	0.31	1	8	4	4	2	1
112	3	30.847	0.21	1	0	0	0	3	0
113	3	32.951	0.25	1	8	4	4	3	1
114	3	34.537	0.35	1	0	0	0	3	0
115	3	33.787	0.25	1	8	4	4	3	1
116	3	36.641	0.3	0	2	1	1	3	0
117	3	31.366	0.2	1	0	0	0	3	0
118	3	36.959	0.37	0	8	4	4	3	1
119	3	34.998	0.29	1	2	1	1	3	0
120	3	34.537	0.24	0	2	1	1	3	0
121	3	36.151	0.32	1	5	4	1	2	0
122	3	33.759	0.24	0	1	1	0	2	0
123	3	30.184	0.2	1	7	4	3	2	0
124	3	35.604	0.31	1	8	4	4	3	1
125	3	36.151	0.29	1	0	0	0	2	0
126	3	37.391	0.35	1	8	4	4	3	1
127	3	35.142	0.3	0	4	3	1	3	0
128	3	36.728	0.31	0	2	1	1	3	0
129	3	38.962	0.42	0	7	4	3	3	0
130	3	38.27	0.44	1	1	1	0	3	0
131	3	37.651	0.49	1	1	1	0	3	0
132	3	36.151	0.39	1	4	4	0	3	0
133	3	37.074	0.32	0	8	4	4	3	1
134	3	36.497	0.34	1	4	4	0	3	0
135	3	30.76	0.21	0	0	0	0	3	0
136	3	41.254	0.53	1	8	4	4	2	1
137	3	33.009	0.3	1	8	4	4	3	1
138	3	37.189	0.35	0	8	4	4	3	1
139	3	34.422	0.29	1	2	2	0	3	0
140	3	32.634	0.23	1	2	2	0	3	0
141	3	34.249	0.31	1	5	4	1	2	0
142	3	32.173	0.21	1	4	4	0	3	0
143	3	32.432	0.2	1	0	0	0	3	0
144	3	35.287	0.25	0	5	4	1	3	0
145	3	34.652	0.35	1	3	2	1	3	0
146	3	35.604	0.33	0	1	1	0	2	0
147	3	32.634	0.27	1	1	1	0	3	0
148	3	34.681	0.3	1	8	4	4	3	1
149	3	32.231	0.21	0	0	0	0	3	0
150	3	37.276	0.38	1	8	4	4	1	1
151	3	33.672	0.31	1	8	4	4	3	1
152	3	39.697	0.41	1	0	0	0	3	0
153	3	34.162	0.25	0	5	4	1	3	0
154	3	36.699	0.36	0	8	4	4	3	1
155	3	34.56	0.34	0	1	1	0	3	0

156	3	30.359	0.21	1	8	4	4	3	1
157	3	36.533	0.38	0	4	3	1	2	0
158	3	30.669	0.26	1	8	4	4	3	1
159	3	38.851	0.45	1	6	4	2	3	0
160	3	31.921	0.26	0	6	3	3	3	0
161	3	30.521	0.2	0	0	0	0	3	0
162	3	32.675	0.25	0	4	2	2	3	0
163	3	38.396	0.37	1	0	0	0	2	0
164	3	33.968	0.35	1	2	1	1	2	0
165	3	31.239	0.26	1	8	4	4	3	1
166	3	32.136	0.22	0	5	1	4	2	0
167	3	31.418	0.23	1	0	0	0	3	0
168	3	36.242	0.39	1	8	4	4	3	1
169	3	33.142	0.25	0	8	4	4	3	1
170	3	34.327	0.29	1	0	0	0	2	0
171	3	38.492	0.42	1	8	4	4	3	1
172	3	33.285	0.3	1	5	4	1	3	0
173	3	41.81		1	3	3	0	3	0
174	3	37.379	0.4	1	8	4	4	3	1
175	3	35.224	0.3	1	8	4	4	2	1
176	3	32.029	0.25	1	8	4	4	3	1
177	3	34.01	0.29	1	4	4	0	3	0
178	3	35.404	0.34	1	8	4	4	3	1
179	3	33.142	0.25	0	3	3	0	3	0
180	3	30.76	0.22	0	5	4	1	3	0
181	3	31.382	0.18	0	0	0	0	3	0
182	3	32.819	0.28	0	8	4	4	3	1
183	3	35.668	0.3	0	7	3	4	3	0
184	3	35.44	0.31	1	1	1	0	2	0
185	3	34.542	0.3	0	8	4	4	3	1
186	3	37.63	0.35	0	8	4	4	2	1
187	3	36.589	0.32	1	8	4	4	3	1
188	3	35.404	0.36	1	0	0	0	3	0
189	3	31.454	0.28	1	8	4	4	2	1
190	3	40.646	0.65	1	0	0	0	3	0
191	3	31.862	0.26	1	0	0	0	2	0
192	3	42.083	0.61	0	6	4	2	3	0
193	3	34.973	0.31	0	8	4	4	3	1
194	3	36.661	0.36	1	0	0	0	3	0
195	3	36.698	0.32	0	3	2	1	3	0
196	3	32.567	0.26	1	3	2	1	3	0
197	3	35.727	0.3	1	8	4	4	3	1
198	3	31	0.19	0	2	1	1	3	0
199	3	33.86	0.3	1	5	4	1	2	0
200	3	33.489	0.23	1	2	1	1	2	0
201	3	35.584	0.28	1	8	4	4	3	1
202	3	33.501	0.23	1	5	3	2	3	0
203	3	31.335	0.21	1	2	1	1	3	0
204	3	36.876	0.34	0	0	0	0	3	0
205	3	31.071	0.23	1	7	4	3	3	0
206	3	34.452	0.34	1	2	1	1	2	0
207	3	35.332	0.36	1	2	1	1	3	0
208	3	34.004	0.31	1	8	4	4	3	1
209	3	33.106	0.31	0	2	1	1	2	0

210	3	40.12	0.44	1	2	1	1	3	0
211	3	34.363	0.29	1	8	4	4	2	1
212	3	36.374	0.3	0	1	1	0	3	0
213	3	33.369	0.27	1	3	3	0	3	0
214	3	33.537	0.33	1	0	0	0	3	0
215	3	36.302	0.29	1	0	0	0	3	0
216	3	33.465	0.27	0	8	4	4	2	1
217	3	32.484	0.24	0	2	1	1	3	0
218	3	38.241	0.4	1	5	4	1	3	0
219	3	34.614	0.3	0	0	0	0	3	0
220	3	31.407	0.26	1	0	0	0	3	0
221	3	31.933	0.21	1	8	4	4	3	1
222	3	36.014	0.32	1	1	1	0	3	0
223	3	31.765	0.2	1	5	4	1	3	0
224	3	31.478	0.24	1	8	4	4	3	1
225	3	32.053	0.24	0	2	1	1	3	0
226	3	33.86	0.27	1	6	3	3	3	0
227	3	37.451	0.47	1	8	4	4	3	1
228	3	40.323	0.44	1	3	2	1	3	0
229	3	31.67	0.23	0	2	1	1	3	0
230	3	32.029	0.24	0	0	0	0	2	0
231	3	35.44	0.34	0	8	4	4	3	1
232	3	36.445		1	0	0	0	3	0
233	3	33.537	0.31	1	2	1	1	3	0
234	3	34.183	0.24	1	1	1	0	3	0
235	3	33.106	0.26	1	4	4	0	3	0
236	3	34.219	0.21	1	0	0	0	3	0
237	3	39.282	0.37	1	8	4	4	2	1
238	3	37.954	0.38	1	4	4	0	3	0
239	3	35.26	0.25	0	0	0	0	3	0
240	3	31.406	0.17	0	3	3	0	3	0
241	3	31.694	0.21	0	2	1	1	3	0
242	3	32.723	0.23	1	2	1	1	3	0
243	3	33.968	0.23	1	1	0	1	2	0
244	3	33.321	0.37	1	0	0	0	3	0
245	3	33.681	0.29	1	1	1	0	2	0
246	3	31.921	0.17	1	2	1	1	3	0
247	3	34.219		1	4	1	3	3	0
248	3	33.573	0.24	1	8	4	4	3	1
249	3	31.634	0.18	0	6	3	3	3	0
250	3	32.891	0.26	0	8	4	4	3	1
251	3	32.567	0.25	1	2	1	1	3	0
252	3	31.454	0.22	1	1	1	0	3	0
253	3	34.974	0.34	1	8	4	4	3	1
254	4	36.5			8	4	4	3	1
255	4	38.5			2	1	1	3	0
256	4	31.0			8	4	4	3	1
257	4	32.0			8	4	4	3	1
258	4	32.5			8	4	4	3	1
259	4	30.5			8	4	4	3	1
260	4	36.5			8	4	4	3	1
261	4	31.0			8	4	4	3	1
262	4	30.5			8	4	4	3	1
263	4	40.0			8	4	4	3	1

264	4	37.5	8	4	4	3	1
265	4	32.0	8	4	4	3	1
266	4	35.0	8	4	4	3	1
267	4	40.5	8	4	4	3	1
268	4	29.0	8	4	4	3	1
269	4	28.5	8	4	4	2	1
270	4	33.0	8	4	4	3	1
271	4	35.5	8	4	4	3	1
272	4	41.0	8	4	4	3	1
273	4	32.0	8	4	4	3	1
274	4	29.5	8	4	4	3	1
275	4	29.5	8	4	4	3	1
276	4	29.0	8	4	4	3	1
277	4	28.0	8	4	4	3	1
278	4	30.0	8	4	4	3	1

Table 6: Raw-data for the parasite *G. anomala*. V= Volume in mm^3.

number	sample	1V	2V	3V	4V	5V	6V	7V	8V	9V	10V	11V	12V	13V	14V	15V	16V	17V
1	1	3.1	2.1															
2	1	2.5	1.4	1.3	0.3	0.1												
3	1																	
4	1	0.3	0.1	0.1														
5	1	6.0	0.7															
6	1	5.6																
7	1																	
8	1	2.5																
9	1	2.7	2.6	2.0	2.0	1.1												
10	1	4.2	2.6	1.4														
11	1	4.2	2.3	2.0	1.9	1.8	1.6	1.3	1.2	1.1	0.9	0.8	0.5	0.3				
12	1	0.5	0.0															
13	1																	
14	1	8.2	6.3	3.0	1.7	0.9												
15	1																	
16	1																	
17	1																	
18	1	12.1	###	9.7	7.7	6.1												
19	1	3.3	0.5	0.3														
20	1	5.8	5.5	4.6	4.6	4.2	3.0	2.8	2.8	2.3	2.0	1.9	0.4					
21	1	4.8	2.6	0.6														
22	1	7.6	4.6	2.4	2.1	1.6												
23	1	4.9	3.9	3.3	2.9	2.8	2.4	1.7	1.5	0.3								
24	1	9.2	7.0	6.5	5.9	4.7	2.5	2.1	0.7	0.3								
25	1																	
26	1	6.5	4.6	1.9														
27	1	8.3	7.3	4.6	2.2	1.7	1.4											
28	1	2.3																

29	1	**8.4**	**1.4**	**0.5**												
30	1															
31	1	**10.6**	**7.7**	**6.4**	**2.4**											
32	1	**8.8**	**6.7**	**6.0**	***5.4***	***4.5***	2.8	2.1	**0.4**							
33	1															
34	1															
35	1	**7.8**	**6.6**	**5.9**	**5.1**	1.3										
36	1	**3.0**	**2.7**	**2.6**	**2.0**											
37	1	8.2	**7.4**	6.8	**3.5**	***3.2***	**2.6**	**0.6**								
38	1	***8.8***	6.4	***5.0***	***4.7***	4.4	**4.2**	***2.8***	**0.5**	**0.4**	**0.1**					
39	1															
40	1	**7.0**	**4.8**	**2.0**	**0.1**											
41	1	**0.5**														
42	1															
43	1															
44	1	**16.8**	**6.1**	**3.1**	**1.2**											
45	1	**15.2**	**6.4**	**3.5**	**0.6**											
46	1	***9.3***	***7.9***	***6.2***	**3.0**	**2.7**	2.7	**1.9**	***1.7***	**1.7**	1.2	***0.2***				
47	1	**10.2**	**7.4**	**5.1**	4.6	**2.8**	1.3									
48	1															
49	1	**3.2**	**3.2**	**2.6**	2.3	**1.8**	0.6									
50	1	***12.1***	**7.2**	6.8	**6.0**	**3.8**	**1.7**	0.0								
51	1	**9.9**	**8.6**	**7.7**	**3.0**	5.2										
52	1															
53	1															
54	1	**7.6**	**7.1**	**5.8**	**0.7**	0.0	0.0	***0.0***	***0.0***	***0.0***	***0.0***	***0.0***	***0.0***	***0.0***	**0.0**	
55	1	**0.1**														
56	1	**2.4**	**1.7**	0.9	**0.7**	**0.2**	***0.1***	0.1	***0.1***	**0.1**	***0.1***	***0.0***	**0.0**	***0.0***	***0.0***	***0.0***
57	1	**6.0**	**4.3**	1.7	**0.8**	**0.4**										
58	1	**10.1**	**9.0**	**7.2**	**1.8**											
59	1	**5.9**	**5.3**	5.3	**2.6**	**2.1**										
60	1	**4.0**	**1.6**													
61	1	**6.4**	**1.5**													
62	1	**0.9**														
63	1															
64	1															
65	2															
66	2	**34.3**	**5.5**													
67	2	**5.1**	**2.3**	1.1	**0.8**	**0.6**	0.1									
68	2	**11.3**	**7.5**													
69	2	**5.5**	**1.5**	**0.2**	**0.1**											
70	2	**0.9**	**0.7**	**0.2**	0.2	**0.1**										
71	2															
72	2															
73	2	**7.9**	**2.5**	**1.8**	0.3	**0.2**	0.1									
74	2	**4.6**	**1.4**	**0.5**	**0.2**	0.1										
75	2															
76	2	**5.6**	**0.6**	**0.5**	**0.3**											
77	2	8.2	**3.7**	**2.6**	**2.5**	**1.5**										
78	2	**0.9**														
79	2															
80	2	**2.5**														
81	2	**1.8**	**1.1**	**0.5**	0.2	**0.1**										
82	2	**10.0**	**3.1**	**0.5**	**0.2**	0.1										

83	2	1.5	1.4															
84	2	*17.0*	6.2	5.1	3.5	*2.6*	2.4	*2.0*	1.6	*0.6*	*0.4*	0.4	0.4	*0.4*	0.2	0.2	0.1	*0.1*
85	3	1.7	0.6															
86	3	11																
87	3																	
88	3	4.2	0.5															
89	3																	
90	3	9.9																
91	3	19	12															
92	3																	
93	3	5.1	4.1															
94	3	37																
95	3	11	10	6.2	3													
96	3	1.9																
97	3	0.2																
98	3	10																
99	3	1.5																
100	3																	
101	3	2.5	2.5	0.2														
102	3	1.2																
103	3	17	8	7.2	5	4.6	3.9	1.8	1.2	0.3								
104	3	6.3	3.1	1.4														
105	3																	
106	3																	
107	3	8.6	4.6	4.3	3													
108	3	9.6	6.1															
109	3	2.6																
110	3	23	2.8															
111	3	3.1	0.9															
112	3	5.9																
113	3	3.9	3.2	2.9	3													
114	3	12	5.3	3.6	3	3.1	1.2	2.8										
115	3	18	11															
116	3																	
117	3	6.1																
118	3																	
119	3	2.8	0.2															
120	3																	
121	3	5.9	4.7	2.8	3	2.3	1.9	1.4	1.4	0.8	0.7	0.4	0.3	0.1				
122	3																	
123	3	8.3	0.4															
124	3	2.5	0.4															
125	3	12	9	5.6	3	2.4												
126	3	3.4																
127	3																	
128	3																	
129	3																	
130	3	15	6.4	4.7	3	3	2.1	2.1	1.8	1.2	1.1	1						
131	3	5.9	4.7	4.5														
132	3	7.1	6.3															
133	3																	
134	3	3.4	2.9	2.5	2	1.4	1.3	1.2										
135	3																	
136	3	6.1	3.4															

137	3	**6**	**3.8**		
138	3				
139	3	**7.8**	**7**	**5.4**	**4**
140	3	**3.4**	**3**	**0.7**	
141	3	**14**	**13**	**9**	
142	3	**9.7**			
143	3	**15**	**12**		
144	3				
145	3	**9**	**8.9**	**4.2**	**3**
146	3				
147	3	**8.9**	**3.2**		
148	3	**9**	**8.1**		
149	3				
150	3	**3.7**			
151	3	**34**	**26**		
152	3	**13**	**5.7**	**2.4**	
153	3				
154	3				
155	3				
156	3	**4**			
157	3				
158	3	**28**	**7.9**		
159	3	**5.8**			
160	3				
161	3				
162	3				
163	3	**9.4**	**4.3**		
164	3	**5.3**	**1.8**		
165	3	**23**			
166	3				
167	3	**17**	**6.1**		
168	3	**18**			
169	3				
170	3	**22**			
171	3	**5.7**	**2.2**		
172	3	**15**			
173	3	**4.7**	**3**	**1.5**	
174	3	**0.3**	**0.3**		
175	3	**3.8**	**2.9**	**1.8**	
176	3	**8.8**	**4.9**		
177	3	**7**			
178	3	**3.9**			
179	3				
180	3				
181	3				
182	3				
183	3				
184	3	**9.4**	**4.3**		
185	3				
186	3				
187	3	**4.5**			
188	3	**0.5**	**0.1**		
189	3	**29**			
190	3	**13**	**3.8**	**1.4**	**1**

191	3	**22**	**13**	**11**							
192	3										
193	3										
194	3	**14**	**12**	**9.7**	**7**	**4**	**2.8**	**2.6**			
195	3										
196	3	**4**	**4**	**2.9**	**2**						
197	3	**41**									
198	3										
199	3	**9**									
200	3	**14**									
201	3	**6.9**	**4.9**	**2.7**	**2**	**0.7**	**0.4**	**0.3**	**0**		
202	3	**9.4**									
203	3	**2.3**									
204	3										
205	3	**9.6**	**6.9**	**4.4**	**4**	**2.3**	**1.7**				
206	3	**0.7**									
207	3	**8.9**	**4.5**	**4.3**	**3**	**2.8**	**2.1**	**1.8**	**0.7**	**0.6**	
208	3	**19**	**11**								
209	3										
210	3	**8.8**	**5.3**								
211	3	**11**	**3.6**								
212	3										
213	3	**12**									
214	3	**6**	**4.5**	**3.7**	**4**	**3.3**	**3.3**	**3.2**	**2.3**	**1.5**	**1**
215	3	**6.4**	**4.2**	**3.8**	**3**	**2.4**	**0.7**	**0.7**	**0.6**	**0.3**	
216	3										
217	3										
218	3	**9.8**	**7.7**								
219	3										
220	3	**5.2**	**1.5**								
221	3	**8**									
222	3	**2.5**	**2.3**								
223	3	**9.7**	**3.2**	**2.9**							
224	3	**24**	**9.8**								
225	3										
226	3	**4.4**	**3.1**	**1.6**	**2**	**0.3**					
227	3	**37**	**33**	**4.4**							
228	3	**3.6**	**3.2**								
229	3										
230	3										
231	3										
232	3	**0.3**									
233	3	**5.6**	**3.4**	**0.5**							
234	3	**7.6**									
235	3	**9.5**	**4.6**	**2.1**							
236	3	**2.3**									
237	3	**13**	**9.5**	**6.9**	**6**						
238	3	**6.1**									
239	3										
240	3										
241	3										
242	3	**2.3**	**0.3**	**0.3**							
243	3	**7**	**1.4**								
244	3	**22**	**13**	**12**	**9**	**7.2**	**4**	**4**	**3.6**	**3**	**2.3**

245	3	**6.7**	
246	3	**2.1**	**0.2**
247	3	**0.2**	
248	3	**0**	
249	3		
250	3		
251	3	**3.6**	**0.1**
252	3	**11**	
253	3	**2.5**	
254	4		
255	4		
256	4		
257	4		
258	4		
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272	4		
273	4		
274	4		
275	4		
276	4		
277	4		
278	4		

Table 7: Raw-data for the parasite *S. solidus*. V= Volume in mm^3

number	sample	V1	V2	V3	V4	V5	V6
1	1	118.9					
2	1	216.9					
3	1	256.0	130.5				
4	1	182.6					
5	1	158.8	157.0				
6	1	300.7					
7	1	293.9					
8	1						
9	1	337.2					
10	1	196.2	142.4	123.4			
11	1	166.8					
12	1	235.5	228.9	198.2			
13	1	235.0					

14	1	274.3	258.3				
15	1	199.3	186.9	131.7			
16	1	260.4					
17	1	132.7	88.0				
18	1	178.4					
19	1	190.1	121.1				
20	1	219.0	93.8				
21	1	165.3	160.1	157	116.3	112.0	98.5
22	1						
23	1						
24	1	193.7					
25	1	259.0	214.1				
26	1	198.0	195.8				
27	1	110.7	59.6				
28	1	203.9					
29	1	263.2					
30	1	298.4					
31	1	265.8					
32	1	119.9	97.9				
33	1	225.0	212.3				
34	1	221.5					
35	1	174.0					
36	1	284.2					
37	1	199.2					
38	1	201.6					
39	1	278.7	58.0				
40	1	232.5					
41	1	149.7					
42	1	202.2					
43	1	183.8	138.1				
44	1	300.6					
45	1						
46	1						
47	1						
48	1						
49	1	114.9	103.7	46.6	45.8	36.2	12.9
50	1						
51	1						
52	1						
53	1	194.4					
54	1						
55	1						
56	1	112.6	96.0				
57	1						
58	1						
59	1						
60	1	53.6					
61	1						
62	1						
63	1						
64	1						
65	2						
66	2						
67	2						

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