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Food Quality Effects on Zooplankton Growth and Energy Transfer in Pelagic Freshwater Food Webs

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Abstract
Persson, J. 2007. Food Quality Effects on Zooplankton Growth and Energy Transfer in Pelagic Freshwater Food Webs. (Effekter av födokvalitet på djurplanktons tillväxt och på energiöverföringen i födovävar i sjöar). Acta Universitatis Upsaliensis. *Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology* 292. 43 pp. Uppsala. ISBN 978-91-554-6859-0.

Poor food quality can have large negative effects on zooplankton growth and this can also affect food web interactions. The main aims of this thesis were to study the importance of different food quality aspects in *Daphnia*, to identify potentially important differences among zooplankton taxa, and to put food quality research into a natural context by identifying the importance of food quality and quantity in lakes of different nutrient content.

In the first experiment, the RNA:DNA ratio was positively related to the somatic growth rate of *Daphnia*, supporting a connection between P content, RNA content, and growth rate. The second experiment showed that EPA was important for *Daphnia* somatic growth, and 0.9 µg EPA mg C^{-1} was identified as the threshold below which negative effects on *Daphnia* growth occurred.

A field survey identified patterns in the PUFA content of zooplankton that could be explained by taxonomy and trophic position. *Cladocera* enriched EPA and ARA relative to seston, and *Copepoda* primarily enriched DHA. In a whole-lake experiment, gentle fertilization of an oligotrophicated reservoir increased the seston P content and the biomass of high quality phytoplankton (*Cryptophyceae*, high EPA content). This was followed by increases in zooplankton and fish biomasses.

An empirical model based on data from a literature survey predicted that food quantity is most important for zooplankton growth in oligotrophic lakes, and that food quality factors are more important in eutrophic lakes. Thus, zooplankton growth, and energy transfer efficiency in the food web, is predicted to be highest in mesotrophic lakes. The results predict that the strength and nature of food quantity and quality limitation of *Daphnia* growth varies with lake trophic state, and that some combination of food quantity and/or quality limitation should be expected in nearly all lakes.

Keywords: elemental stoichiometry, phosphorus, polyunsaturated fatty acids, EPA, oligotrophic lakes, empirical modelling, Daphnia, Cladocera, Copepoda

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Little by little, one travels far
J. R. R. Tolkien

List of papers

These are the papers on which this thesis is based. They are hereon referred to by their roman numerals.

I   Vrede, T., <u>J. Persson</u> & G. Aronssen (2002) The influence of food quality (P:C ratio) on RNA:DNA ratio and somatic growth rate of *Daphnia. Limnology and Oceanography* **47:** 487-494 [*]

II   Ravet[†], J. L., <u>J. Persson</u>[†] & M. T. Brett. Dietary threshold concentration of three polyunsaturated fatty acids for *Daphnia* somatic growth and reproduction. *Manuscript*

III   <u>Persson, J.</u> & T. Vrede (2006) Polyunsaturated fatty acids in zooplankton: variation due to taxonomy and trophic position. *Freshwater Biology* **51:** 887-900 [‡]

IV   <u>Persson, J.</u>, T. Vrede & S. Holmgren. Responses in crustacean zooplankton populations to food quality and quantity changes after whole lake nutrient enrichment of an oligotrophicated sub-alpine reservoir. *Manuscript*

V   <u>Persson, J.</u>, M. T. Brett, T. Vrede & J. L. Ravet (2007) Food quantity and quality regulation of trophic transfer between primary producers and a keystone grazer (*Daphnia*) in pelagic freshwater food webs. *Oikos, accepted* [‡]

Contents

Abbreviations

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C	Carbon
N	Nitrogen
P	Phosphorus
TP	Total phosphorus
P:C	Phosphorus to carbon ratio (molar)
partC	Particulate carbon
FA	Fatty acid
PUFA	Polyunsaturated fatty acid ($\geq$ 2 double bonds)
ARA	Arachidonic acid, 20:4ω6
DHA	Docosahexaenoic acid, 22:6ω3
EPA	Eicosapentaenoic acid, 20:5ω3
SDA	Stearidonic acid, 18:4ω3
RNA	Ribonucleic acid
DNA	Deoxyribonucleic acid
RGD	Relative Growth rate Decrease

Introduction

Food web efficiency

The transfer of carbon across trophic levels in food webs is a central function in all ecosystems and has been the subject of numerous studies since the pioneering work of Lindeman (1942). Animals that feed on plants or detritus play a critical role in the transfer of carbon (C), energy and nutrients in all food webs (Shurin *et al.* 2002) and it is important to understand the factors that regulate this carbon flow.

McNaughton *et al.* (1989) presented a correlation between primary and secondary production, using data from many different terrestrial ecosystems (fig 1). This highly significant ($p<0.001$) correlation show a clear positive trend, but with an r^2 of 0.36 most of the variation in grazer production is left unexplained. For any given level of primary production there is great variation in the secondary production, almost two orders of magnitude. So if plant production increases there is not necessarily a corresponding increase in consumer production as a result of this.

Figure 1 The relationship between net primary production (plants) and net secondary production (grazers) in a diverse set of terrestrial habitats, from deserts to tropical rain forests. Units are kJ m^{-2} yr^{-1}. Modified from McNaughton *et al.* (1989).
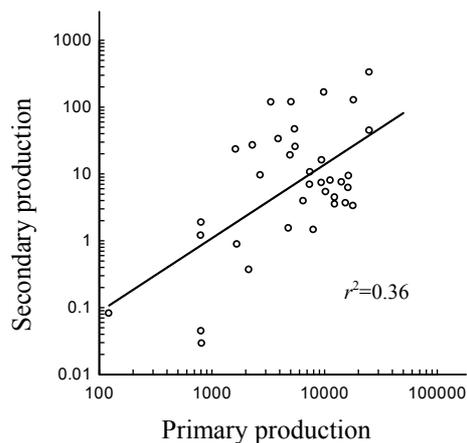
Studies based on large compilations of lake data generally show a positive relationship between phytoplankton biomass and herbivore biomass (McCauley & Kalff 1981; Hanson & Peters 1984; Yan 1986), but as in figure 1 these relationships leaves much variation unexplained. Reasons for this spread in field observations can be many, *e.g.* variation in environmental factors such as temperature and pH, or biological interactions such as competition or predation. But even in lab experiments with constant environment, and neither competition nor predation there was still considerable variation left in the herbivore growth response to a given amount of food (*e.g.* Müller-Navarra 1995; Müller-Navarra & Lampert 1996). This indicates that there was also something within the food itself that varied.

As concluded in the meta-analysis on consumer versus resource control in freshwater pelagic food webs by Brett and Goldman (1997): "To gain a better understanding of food-web interactions it is important to determine which factors regulate the efficiency at which primary production is converted to herbivore biomass". The trophic transfer of energy and nutrients are favoured by conditions that allow for high growth rates (Sterner & Elser 2002), and consumers feeding on nutritionally imbalanced food can be associated with large respiratory losses (Darchambeau *et al.* 2003), which in turn leads to impaired individual growth rates and energy transfer efficiency since the respired energy is lost from the system.

## The pelagic food web and the role of zooplankton

Crustacean zooplankton (hereafter zooplankton) are important from an ecosystem perspective since they provide the major link between the organisms at the base of the food web and the organisms at higher trophic positions (fig 2). Zooplankton can affect the biomass, production, and species composition of the phytoplankton assemblage (Sterner 1989) and they also provide an important food source for juvenile fish (Hrbacek *et al.* 1961; Gliwicz & Pijanowska 1989).

Seston is a collective term for all particulate material present in the free water, which includes protozoa, phytoplankton, bacteria, and detritus. Together these make up what is available for zooplankton to feed on (fig 2). Phytoplankton constitutes the dominating primary producers in the open water of lakes, they incorporate inorganic nutrients using solar energy, providing carbon and energy that can be transferred to higher trophic levels in the pelagic food web. In addition to phytoplankton, energy mobilized by bacteria may contribute to the zooplankton food either directly by consumption of bacteria or indirectly via bacterivorous protozoa. Detritus may also provide an important energy source, especially in lakes where phytoplankton makes up a very small proportion of seston. Depending on the type of lake the importance of the autotrophic phytoplankton pathway relative to the het-

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erotrophic bacterial pathway energy mobilization can vary substantially, and bacterial mobilization of external carbon sources can dominate in many lakes (Jansson *et al.* 2000; Karlsson *et al.* 2002).

Zooplankton thus hold a key position in the pelagic food by making these sources of energy available to higher trophic levels, and there is a need to know how zooplankton growth and trophic transfer efficiency is regulated in order to understand how pelagic food web structure and function is regulated.

Figure 2 The freshwater pelagic food web structure. ➡ indicate fluxes of matter, → fluxes of inorganic nutrients, and ⟹ fluxes of organic nutrients (Vrede 1998).
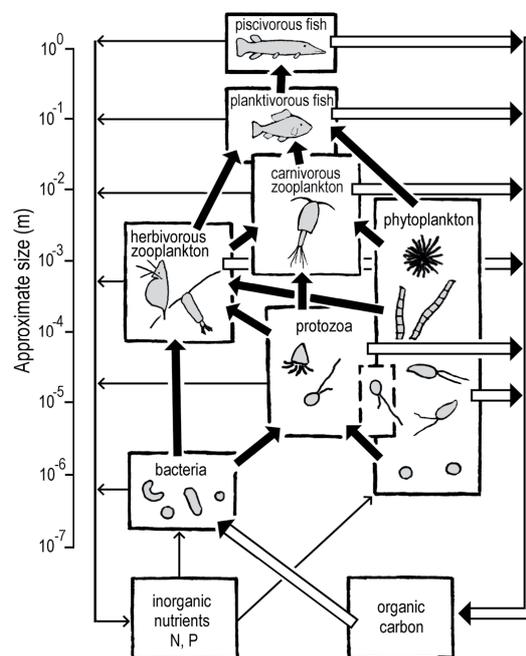
Factors affecting zooplankton

In early studies it was commonly assumed that species succession in the zooplankton community was the result of abiotic factors such as light intensity and water density or viscosity (Hutchinson 1967). Other explanations have highlighted predation pressure (Hrbacek *et al.* 1961; Brooks & Dodson 1965; Gliwicz & Pijanowska 1989), food quantity (Sommer *et al.* 1986; Demott 1989), or the presence of inedible algae (Leibold 1989). Predation from fish is of great importance in the determination of which type of zooplankton that dominate. When many zooplanktivorous fishes are present the predation pressure on large-bodied zooplankton is high. Therefore, species

such as big *Daphnia* decrease and the smaller and fast-growing species increase.

During the last decades it has been convincingly shown that also food quality constraints can be an important regulating mechanism for zooplankton growth (Gulati & DeMott 1997; Hessen *et al.* 2004), and that this can affect the flux of organic matter and nutrients in food webs (Andersen *et al.* (2004).

Food Quality

The idea that nutritional constraints can be an important regulating mechanism for the flux of organic matter and nutrients in food webs was touched upon already by Lotka (1925), but this field of research has recently got a dramatic upswing.

Anything that an organism requires for correct functioning that it cannot make on its own must be supplied from its environment, for animals most commonly via the diet. An animal is thus dependent on its dietary supplies of these substances, which are referred to as essential substances. And the concentration of these essential substances determines the food quality.

Much of the theoretical framework was developed in aquatic systems (Sterner & Elser 2002), and it is well acknowledged that food quality can have a major impact on zooplankton (Hessen 1992; Gulati & DeMott 1997), and on the zooplankton interactions with the surrounding biota via their nutrient regeneration (Elser & Urabe 1999). But food quality is by no means only important in aquatic food webs, terrestrial herbivores are also likely to frequently be nitrogen (N) or phosphorus (P) limited by their food (Elser *et al.* 2000a). It has for example been shown that the seasonal migration of wildebeest and zebras can be driven by the P and N content of the available grass (Ben-Shahar & Coe 1992). A meta-analysis of terrestrial and aquatic systems showed that the biomass turnover rate is higher in terrestrial plants that contain more N or P (Cebrian 1999), the plants of high food quality (higher content of N or P) were more heavily grazed, indicating that they were more desirable for the grazers.

Many factors can potentially constrain secondary production, *e.g.* digestibility (Porter 1975), toxicity (Lampert 1981), colony formation (Lürling & Van Donk 1996), availability of dietary sterols (von Elert & Wolffrom 2001; von Elert *et al.* 2003), amino acids (Laabir *et al.* 1999), P (Hessen 1992; Gulati & DeMott 1997), PUFA (Brett & Muller-Navarra 1997). Among the potentially important food quality parameters, P and certain polyunsaturated fatty acids (PUFA) have so far been identified as the most important essential substances in zooplankton nutrition.

Elemental stoichiometry

Traditional ecosystem ecology uses "one currency models", with C or energy flow between trophic levels. In contrast, elemental stoichiometry (from greek *stoikheion*, element and *–metres*, measurer) considers relative proportions (ratios) of potentially limiting key elements, and therefore offers a multiple-currency approach potentially better suited to help understanding of ecological processes. Ratios commonly used are P:C and N:C, *i.e.* for every C atom in an organism there is a certain number of P or N atoms. This also provides a measure on how diluted P or N is in C, C being the most common element in organisms. The transitions between trophic levels can be viewed as chemical reactions and the components needed higher up in the food web have to be available from the lower trophic levels, from which energy and most of their elements are coming. It is therefore important that the consumers' diet is reasonably well balanced and contains the needed elements. P and N are generally less concentrated in autotrophs than in heterotrophs, and the N and P concentrations are also much more variable in autotrophs (Elser *et al.* 2000a). Hence, from an ecosystem perspective the trophic step from autotrophs to heterotrophs can be a problematic one. When there are large differences between the autotrophs nutrient content relative to what the consumer needs to support its body constituents, this can decoupled this trophic step.

The growth rate hypothesis (GRH) states that there are connections between the life history, the body elemental composition, and the nutritional requirements of an organism (Elser *et al.* 1996). The largest pool of P in crustaceans is in nucleic acids (Vrede *et al.* 1999), mainly in ribosomal RNA which is needed for protein synthesis, a key component of animal growth (Elser *et al.* 1996). A fast growing organism therefore needs a lot of RNA, and a lot of RNA contains a lot of P (ca 10% by weight) that in turn has to be acquired via the diet.

In accordance with GRH, the fast-growing *Daphnia* have a relatively high P content (Main *et al.* 1997), and are therefore more vulnerable to P limitation. There is great variation in where the P limitation threshold for *Daphnia* growth has been reported to occur. But a compilation of many studies indicate that *Daphnia* growth is reduced when the diets P:C ratio was below 0.0033 (Brett *et al.* 2000). P:C ratios below this ratio are found in many natural lakes (Hecky *et al.* 1993; Hassett *et al.* 1997), and P limitation is accordingly likely to occur *in situ*. Other zooplankton taxa such as *Diaphanosoma* and *Bosmina* have lower P contents and are not as vulnerable to low P:C ratios in their diet (Sterner & Schulz 1998). Copepods are also thought to be less prone to P limitation due to their relatively low P content, whereas they could be more vulnerable to N limitation due to their relatively high N content.

Even though GRH states that there should be a covariation of RNA content and somatic growth rate, this has not been backed up by empirical stud-

ies. And whether zooplankton in nutrient poor systems are likely to be af-
fected by elemental food quality constraints is still an open question since
most research on this subject has been carried out in the lab or in relatively
nutrient rich lakes.

Polyunsaturated fatty acids
PUFA are important for maintaining the fluidity of membrane structures and
they also function as precursors for hormones involved in *e.g.* gonad devel-
opment, reproduction, inflammatory and immune responses. PUFA are al-
most exclusively synthesized by autotrophs. Herbivores can convert alpha-
linolenic acid (18:3ω3) to 20:5ω3 (EPA) and 22:6ω3 (DHA; Brett & Muller-
Navarra 1997), but this conversion is inefficient and the animals will grow
better when provided with direct sources (Brett & Muller-Navarra 1997).
Studies by *e.g.* Ahlgren *et al.* (1990) and Müller-Navarra (1995) show that
PUFA composition is important for the growth of freshwater zooplankton.
Apart from the ω3 PUFA, also the ω6 PUFA 20:4ω6 (ARA) is of interest,
although it has so far received limited attention.

   After a substance has been shown to be essential then the question that
naturally follows is at which concentrations it becomes limiting. The earliest
studies commonly used an on/off approach, i.e. the treatments used were
"nothing" or "plenty". But to find out whether a limiting agent is of impor-
tance in natural lakes it is necessary to be able to tell at which levels it can be
limiting. EPA has been shown to be important for zooplankton growth, and
has therefore got most attention among the PUFA (Müller-Navarra *et al.*
2000; Müller-Navarra *et al.* 2004). Two studies that reported on this gave
very contrasting answers (Müller-Navarra *et al.* 2000; Becker & Boersma
2005), and it is therefore still unclear at which concentrations EPA becomes
limiting.

   Fatty acids (FA) can be used as trophic markers since a consumers FA
composition can be influenced by that of their diet (Graeve *et al.* 1994). But
it is not known if there also are systematic differences in FA content between
zooplankton taxa, and if such differences can be connected to their life his-
tory *sensu* GRH. Differences in FA content among taxa could affect their
demands for essential fatty acids in their diet, but it could also affect their
value as food for taxa of higher trophic position?

Applied aspects: compensatory nutrient enrichment

The construction of dams in high altitude lakes for hydroelectric power production often lead to an initial increase in food web production as additional nutrients are leached from the flooded surroundings. This effect generally decreases after a few years and the long term damming effect is a decrease in nutrient content and standing fish stock (Grimard & Jones 1982 and refs therein). The large fluctuations in water level (often 10−20m annually) in water reservoirs cause severe stress on the littoral zone via heavy ice and wave erosion. The eventual loss of littoral biota is thought to be one reason why many of reservoirs eventually become extremely unproductive.

The energy mobilization at the base of the food web may be so low that it restricts the number of trophic levels that can persist in low-productive lakes. It is therefore important that the small amount of energy available is efficiently transferred in the food web. The fish communities in subarctic lakes are dominated by Arctic char (*Salvelinus alpinus*), a species that to a large degree is dependent on energy sources originating from the littoral zone (Karlsson & Byström 2005). Strongly eroded littoral zones are almost devoid of food sources, and this has negative effects on fish abundance and growth.

To restore the production and standing stock of the native fish populations, the pelagic production can be boosted by means of compensatory nutrient addition. Stimulating the pelagic food web, and ultimately fish production, by adding inorganic nutrient is a method that has been practiced successfully in NW America for decades, see Hyatt *et al.* (2004) for a recent review.

The food web stimulation should preferably be done without negatively affecting the native flora and fauna. And to be as ecologically and financially sound as possible, energy should be retained in the food web on the way from phytoplankton to fish in these projects. It is therefore important that the zooplankton growth efficiency is not negatively affected by poor food quality.

Aims of this thesis

The main aims of this thesis were to study the importance of different food quality aspects in *Daphnia*, to identify potentially important differences among zooplankton taxa, and to put food quality research into a natural context by identifying the importance of food quality and quantity in lakes of different nutrient content.

The papers in this thesis address the following specific questions:

Is the RNA:DNA ratio connected to *Daphnia* growth rate? (**I**)

At which threshold concentrations does *Daphnia* get affected by PUFA limitation? (**II**)

Are taxonomy and/or trophic position important for the FA content of zooplankton? (**III**)

Can an ultraoligotrophic lake with low abundance of high food quality be enriched without affecting the food quality negatively? (**IV**)

Can food quantity and quality regulation of *Daphnia* growth be predicted across a lake trophic state gradient? (**V**)

Materials and methods

I & II are experimental studies, **III** is a field survey, **IV** is a field experiment, and **V** is a synthesis paper.

Study organisms

The water fleas (*Daphnia*) are small (< 6 mm) planktonic crustaceans that often dominate zooplankton biomass in freshwaters all over the world. *Daphnia* are keystone species since they are very efficient grazers on phytoplankton, bacteria and protozoa, and also because they are so common. This has made them and the factors influencing them highly interesting through the years. They reproduce asexually during most of their growing season, when all female populations produce identical copies of themselves (clones). Partly due to this they have short generation times of less than a week at optimal conditions and they are quickly able to respond to increases in food abundance. *Daphnia* are unselective filter feeders which mean that as long as the food objects are in the right size spectrum these are almost equally likely to be eaten (Bern 1994). They are easily cultivated in the laboratory. Thanks to this *Daphnia* has been the experimental subject of countless studies the last century. They are also the organisms that have been most extensively used in the research field of this thesis and they are in the focus of attention of **I**, **II** and **V**.

In freshwater environments, most food quality research has hitherto focused on *Daphnia* despite that other crustacean species (*Bosmina*, *Holopedium*, *Eudiaptomus*, Cyclopoids, *etc*.) frequently are quantitatively more important. In addition to *Daphnia*, the systems we studied also contained other species of crustacean zooplankton that are common in oligotrophic lakes. The cladocerans *Bosmina coregoni* s.l., *Holopedium gibberum*, *Bythotrephes longimanus*, and the calanoid copepods *Arctodiaptomus laticeps* and *Heterocope* spp. (*H. saliens* and *H. appendiculata*) were included in the field survey of **III**. *Bosmina*, *Holopedium*, *Arctodiaptomus*, and *Cyclops strenuus* dominated the zooplankton communities in the two lakes studied in **IV**.

The herbivorous cladocerans *Bosmina* and *Holopedium* are filter feeders like *Daphnia* (*e.g.* Hessen 1985) that consume phytoplankton as well as seston particles of similar size, *e.g.* flagellates, ciliates, bacteria and detritus.

Bythotrephes, *Heterocope,* and *Cyclops* are primarily carnivorous raptorial feeders, which feed mainly on protozoa, rotifers and small crustaceans (Monakov 1972; Boudreau & Yan 2003), but phytoplankton may constitute a substantial fraction of their diets (Kling & Fry 1992).

Experiments

I was carried out at the Department of Limnology at Uppsala University, Uppsala. We used the artificial L16 growth medium (Lindström 1991). Stock cultures of *Daphnia galeata* were maintained on *Rhodomonas lacustris*. The food algae *Scenedesmus quadricauda* was grown in chemostats, and by varying the P content of the growth medias and the dilution rate of the chemostats, algae with different P:C ratios (0.0026−0.0256) were obtained. Cultures were grown in a temperature controlled room at 16.5°C and a 18:6 h light:dark cycle. Batch experiments were conducted in bottles and the animals were moved to fresh food medium every 24 h. Juvenile *Daphnia galeata* were fed algae with different P:C ratios and their somatic growth and RNA:DNA ratios were measured. RNA:DNA was measured using a novel single-dye (Ribogreen) fluorometric method.

II was carried out at the Department of Civil and Environmental engineering at the University of Washington, Seattle. We used the artificial L16 growth medium (Lindström 1991). Stock cultures of *Daphnia pulex* were maintained on *Scenedesmus obliquus* in a growth chamber with a constant temperature of 18°C and a 14:10 h light:dark cycle. Monocultures of *Scenedesmus obliquus* and *Microcystis aeruginosa* grown on L16 were used for the experiments. Flow-through experiments were conducted in a 200 L aquarium equipped with 24 partially submerged growth chambers. A peristaltic pump supplied mixtures of phytoplankton and PUFA supplements to the *Daphnia*. Food treatments were prepared fresh daily. The experiments were started with juvenile *Daphnia* and terminated when the animals in any treatment had developed their first clutch. We measured somatic growth rates and counted the numbers of eggs in the brood pouches.

Field studies

Study area

For **III** we collected seston and zooplankton samples from 12 boreal to sub-alpine oligotrophic lakes in north-western Sweden (latitude 63°15' N−64°00' N, longitude 12°35' E−14°05' E). These lakes cover the variation in lake types in this region, *i.e.* oligotrophic and ultra-oligotrophic with low or very low humic content. In **IV** two of these lakes, Mjölkvattnet and Burvattnet, were sampled every two weeks during the growing season of the years 2001−2005 in order to follow temporal changes of sestonic elemental and FA content, as well as the phytoplankton and zooplankton species composition and biomass, before and after the initiation of compensatory nutrient enrichments to Mjölkvattnet. Lake Mjölkvattnet (regulated since 1942) had become oligotrophicated and the growth of the fish population much reduced. The increase of nutrients is expected to increase the abundance of phytoplankton, but a potential problem could be that the quality of the phytoplankton community could decrease due to the whole lake fertilization.

Phytoplankton and zooplankton nutrient content

A known volume of lake water was filtered onto GF/F filters for analysis of sestonic C, N and P, and PUFA. Animals were kept at the *in situ* temperature in lake water for a maximum of a few hours before they were sorted to species or genus under a dissection microscope. The most abundant zooplankton taxa were sampled from each lake. Zooplankton were stored in cryotubes in liquid nitrogen until freeze-dried, after which they were stored under a nitrogen atmosphere in a -70 ºC freezer. Zooplankton samples for C, N, P, and FA analysis were weighed (±1 µg) on a microbalance

C and N were analysed with an elemental analyser, using acetanilide as standard. P concentrations were measured with the molybdate-blue method after hot acid oxidation with 5% potassium persulphate (Menzel & Corwin 1965).

FA were measured as their methyl esters using a gas chromatograph. Individual FA were identified by comparing their retention times with those of standard mixtures, GCL-68A (Nu-Check Prep Inc.) and Fish oil 30 (Larodan Fine Chemicals AB). For occasionally checking single peaks, separate standard FA were added. The FA were quantified by comparing the area of the peaks with the peak area of an internal standard (23:0).

Synthesis study

For **V**, I compiled data to assess the relationships between lake trophic state and food quantity, seston P content, and seston EPA content. Based on these associations we used a Monte Carlo approach to generate distributions of these food quantity and quality factors at specific total phosphorus (TP) levels. These distributions were used to predict the depression of *Daphnia* growth relative to their maximum growth potential (Relative Growth Depression, *RGD*) using published associations (and results from **II**) between *Daphnia* growth and partC particulate carbon), P:C, and EPA:C, respectively. We used two different formulas to calculate the *RGD* due to EPA, one based on field collected seston (Müller-Navarra *et al.* 2000) and one based on EPA supplemented algal cultures (**II**).

We modeled the total *RGD* by combining the growth reducing effects of partC, P, and EPA. This was done assuming that there are multiplicative interactions between the growth decreasing effects of the three food parameters. The biological meaning of this multiplicative model is that food quantity and food quality can, at the same time constrain animal growth. The food quantity being the primary resource which can be used with different efficiency depending on its quality.

Summary of the papers

Is the RNA:DNA ratio connected to *Daphnia* growth rate? (**I**)

The results showed that the RNA:DNA ratio and somatic growth rate of *Daphnia* were positively correlated (fig 3a; r^2=0.94). Below a threshold P:C ratio of approximately 0.005 (C:P 200) the RNA:DNA ratio (and somatic growth) of *Daphnia* decreased with decreasing P:C ratio (fig 3b). The RNA:DNA ratio of the *Daphnia* responded to differences in food quality within 5 h.
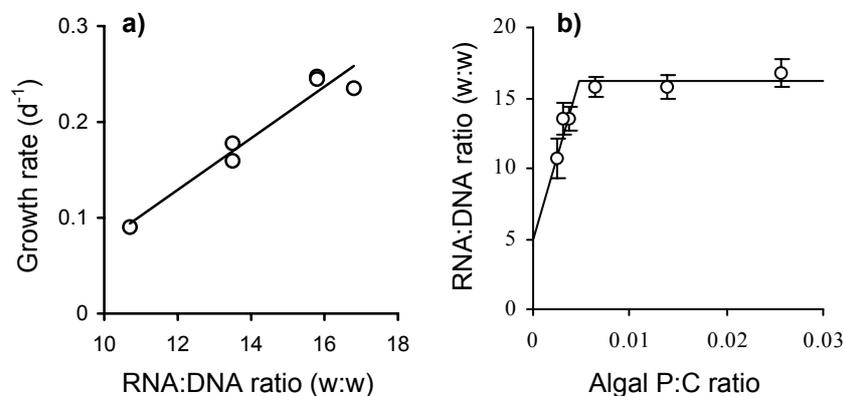
Figure 3 a) Relationship between RNA:DNA ratio and somatic specific growth rate of *Daphnia galeata*. **b)** Relationship between algal P:C ratio and specific growth rate of *Daphnia galeata*. Modified from Vrede *et al.* (2002).

These results supported GRH (Elser *et al.* 1996) by showing a strong positive connection between RNA content and somatic growth. It was previously known that food quantity had an effect on the RNA:DNA ratio and somatic growth in marine copepods (Saiz *et al.* 1998; Wagner *et al.* 1998). This study extended the use of RNA:DNA ratios to freshwater systems and *Daphnia*, and also showed that the method could be used with food quality treatments. RNA:DNA ratio thus offer a method that allows substantial

shortening of experiment duration in these kinds of experiments. To measure the somatic growth rate in a cohort of juvenile *Daphnia* an experiment length of about a week is required to be able to detect the increase in weight. An experiment using RNA:DNA as a response parameter can be considerably shorter, potentially as short as a few hours.

At which threshold concentrations does *Daphnia* get affected by PUFA limitation? (II)

This experiment assessed the effects of varying concentrations of EPA, ARA, and 18:4ω3 (SDA) on *Daphnia* growth and reproduction. We defined the threshold concentration as the first significant ($p<0.05$) decrease in growth rate or egg production relative to dietary PUFA saturation levels, starting from the highest addition. Our results showed that *Daphnia* somatic growth rates declined when dietary concentrations of EPA were less than 0.9 and 0.4 µg EPA mg C^{-1} for the *Microcystis* and *Scenedesmus* treatments respectively, and that the egg production declined at concentrations less than 0.1 and 0.4 µg EPA mg C^{-1} for the same two algal groups. Neither supplements of ARA nor SDA had any effects on *Daphnia* somatic growth. ARA did not affect the egg production either, but for SDA concentrations of less than 0.4 and 0.8 µg mg C^{-1} the egg production decreased for the *Microcystis* and *Scenedesmus* treatments respectively.

We used the EPA results from the *Microcystis* treatment to develop an empirical model that predicts *Daphnia* somatic growth from the EPA concentration (µg mg C^{-1}) of their diet (fig 4).

Figure 4 The EPA threshold results from the *Microcystis* treatment (observed) were used to develop an empirical model ($r^2 = 0.96$) between *Daphnia* growth rate and dietary EPA content. Normalized growth = 1- e$^{(-1.865*(EPA+0.213))}$ Error bars indicate the standard deviation.
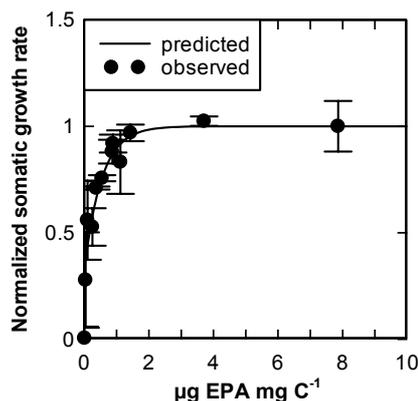
The results indicate that the dietary EPA threshold for *Daphnia pulex* is in the range of 0.4 to 0.9 µg mg C^{-1} for somatic growth rate and 0.1 to 0.4 µg mg C^{-1} for egg production. While the EPA thresholds for both growth rate

and egg production were similar for the *Scenedesmus* treatment, there was a substantial difference in the *Microcystis* growth and reproduction thresholds. This suggests that dietary PUFA thresholds for *Daphnia* are not the same for different phytoplankton taxa. The observed differences could result from the presence or absence of substitutable resources such as EPA precursors (Wacker & Von Elert 2001). The threshold for egg production was lower than for somatic growth, indicating that eggs were prioritized when the access to EPA is low, or that the EPA demand for somatic growth is higher than for egg production.

Müller-Navarra *et al.* (2000) found that *Daphnia magna* experienced a significant decline in somatic growth rate and egg production when the relative EPA content of seston collected from the hypereutrophic Stonegate Pond dropped below 3.1 μg EPA mg C^{-1}. In contrast, Becker and Boersma (2005) reported an EPA threshold for *Daphnia magna* growth as low as 0.04 μg mg C^{-1} in a direct supplementation experiment. If this threshold for *Daphnia* is true, it would imply that EPA limitation probably never occurs in nature. Our EPA threshold model predicted that the EPA threshold limitation level for *Daphnia* is intermediate to the thresholds reported in these two studies. The EPA growth response model presented in this study provides a basic framework for predicting EPA limitation in natural lake systems. We believe that a model of this type is a practical first step towards investigating where and when dietary EPA limitation is likely to occur in aquatic ecosystems.

Are taxonomy and/or trophic position important for the FA content of zooplankton? (**III**)

The phylogenetic origin and/or trophic position explained more of the variation in zooplankton FA content than did the FA content of their food. Despite considerable efforts to explain the variation within each zooplankton taxa using the variation in the seston FA content no such relationships were found in this study.

A discriminant function analysis that sorted the seston and the animal taxa based on the content of PUFA separated the different groups well (fig 5), and only one out of 63 samples was misclassified. The seston samples were well separated from the animals along the first two components, and also the copepods and cladocerans form separate clusters. It is also possible to see that predators, animals of a higher trophic position than herbivores, are farther away from the seston samples than are the more herbivorous animals.

Figure 5 The first two components from the discriminant function analysis, using individual PUFA as covariates and sample type (zooplankton taxa and seston) as grouping variable. Sample type symbols: + Seston; □ *Daphnia spp.*; △ *Bosmina coregoni* s.l.; ◇ *Holopedium gibberum*; ■ *Bythotrephes longimanus*; ○ *Arctodiaptomus laticeps*; and ● *Heterocope* spp. Open symbols are herbivores and closed symbols carnivores. From Persson and Vrede (2006).
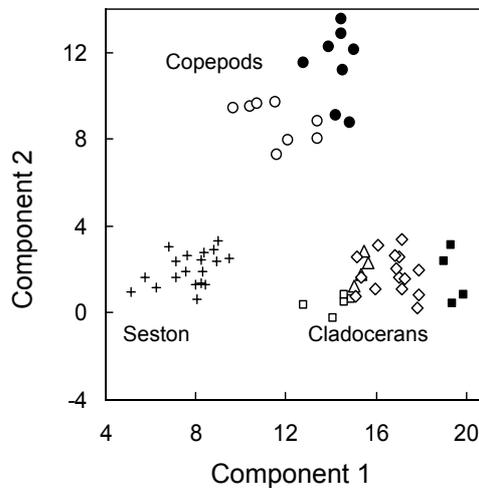
The differences that were important in separating the samples in the discriminant function analysis were that the animals contained higher concentrations of 20- and 22-carbon PUFA relative to 18-carbon PUFA than the seston (fig 6). The major taxonomic difference in FA content, the one that separated out the copepods from the cladocerans, turned out to be that the 20 and 22-carbon PUFA were dominated by DHA in copepods but by EPA in the cladocerans. The predators contained even higher proportions of 20- and 22-carbon PUFA than the herbivorous animals.
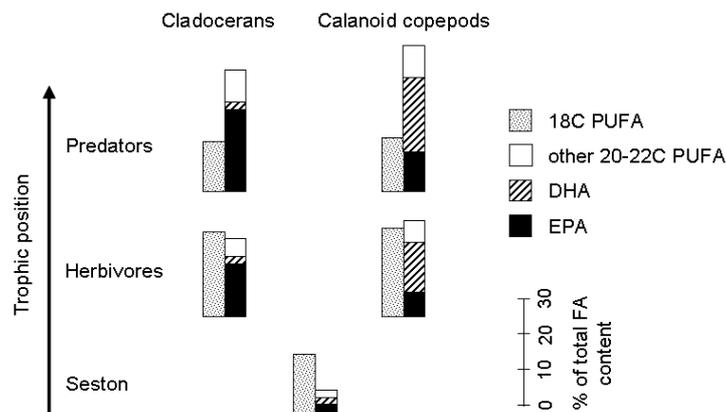
Figure 6 The fate of PUFA in the plankton food web. "Cladoceran herbivores" show the average PUFA percentage of *Bosmina*, *Daphnia*, and *Holopedium*. "18C PUFA" and "20−22C PUFA" are groups of PUFA with 18 and 20−22 carbon atoms long chains respectively. From Persson and Vrede (2006).

The most striking difference among the zooplankton taxa investigated in this study was that the calanoid copepods contained a large fraction of DHA while all the cladocerans were rich in EPA and ARA. Similar differences between *Daphnia* spp. and various copepod species have been noted previously (*e.g.* Farkas 1970; Ballantyne *et al.* 2003) but this study showed that this pattern holds for several previously uninvestigated taxa. Thus, a general pattern emerges with a large difference in FA composition that correlates with phylogenetic origin and/or life history characteristics as well as with the trophic position of the organism (fig 6).

The greater content of EPA and ARA in cladocerans compared with copepods may be related to the cladocerans' higher potential for reproduction. This relationship between EPA and reproductive capacity is speculative, however, and the functions of EPA and ARA in crustaceans remain to be clarified. Compared with cladocerans, copepods have better developed abilities to feed selectively (Butler *et al.* 1989) and escape predators. We hypothesise that these abilities may be a function of a more highly developed nervous system rich in DHA in copepods.

Can an ultraoligotrophic lake with low abundance of high food quality be enriched without affecting the food quality negatively? (**IV**)

The patterns in TP and P:C were clearly different in the two lakes (fig 7) and we interpret these relative changes as being due to the addition of P to Mjölkvattnet. Especially, since the trends in these parameters are all in agreement with decreasing TP in Burvattnet, and stable or slightly increasing TP in Mjölkvattnet due to the fertilization. The stoichiometric food quality factors were stable or improved in Mjölkvattnet during the years of fertilization and stayed at non-limiting levels. In Burvattnet the food quantity (partC) increased as much as in the manipulated lake but the seston P content decreased to levels that could affect zooplankton growth negatively.
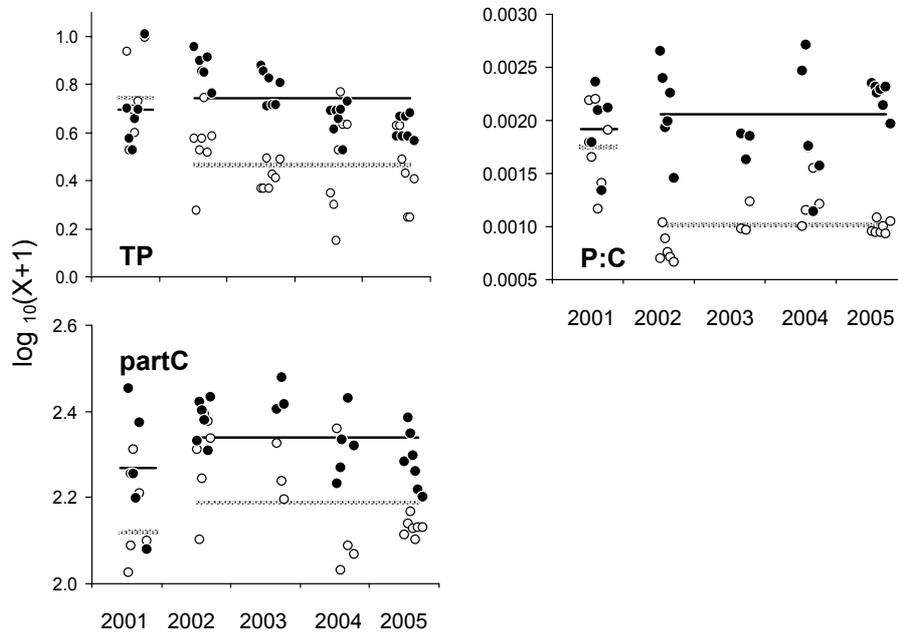
Figure 7 Time series of the $\log_{10}(X+1)$-transformed TP ($\mu g\ l^{-1}$), partC ($\mu g\ l^{-1}$), and P:C ratios (molar) in Mjölkvattnet and Burvattnet. Horizontal lines indicate the means before (2001) and after nutrient additions to Lake Mjölkvattnet (2002-2005). Grey lines and empty symbols represent the reference system Burvattnet, and solid lines and filled symbols represent Mjölkvattnet.

The phytoplankton biomass was similar in both lakes during 2001 (fig 8). Phytoplankton then decreased in Burvattnet during 2002−2005 while the phytoplankton biomass was higher during the years of enrichment in Mjölk-vattnet compared to in 2001. Before fertilization the phytoplankton biomass was dominated by *Chrysophyceae* in both lakes. In Burvattnet the proportion of *Chrysophyceae* decreased in 2002 and the composition then remained similar the following years. In Mjölkvattnet the proportion of *Chrysophy-ceae* decreased in 2002, and continued to decrease in 2003−2005, the *Chrysophyceae* dominance was replaced by a dominance of *Cryptophyceae*.

The biomass of zooplankton was approximately the same in Burvattnet all years and the composition was also relatively similar throughout the study period (fig 8). In Mjölkvattnet there was a gradual increase in zooplankton biomass during the study period.
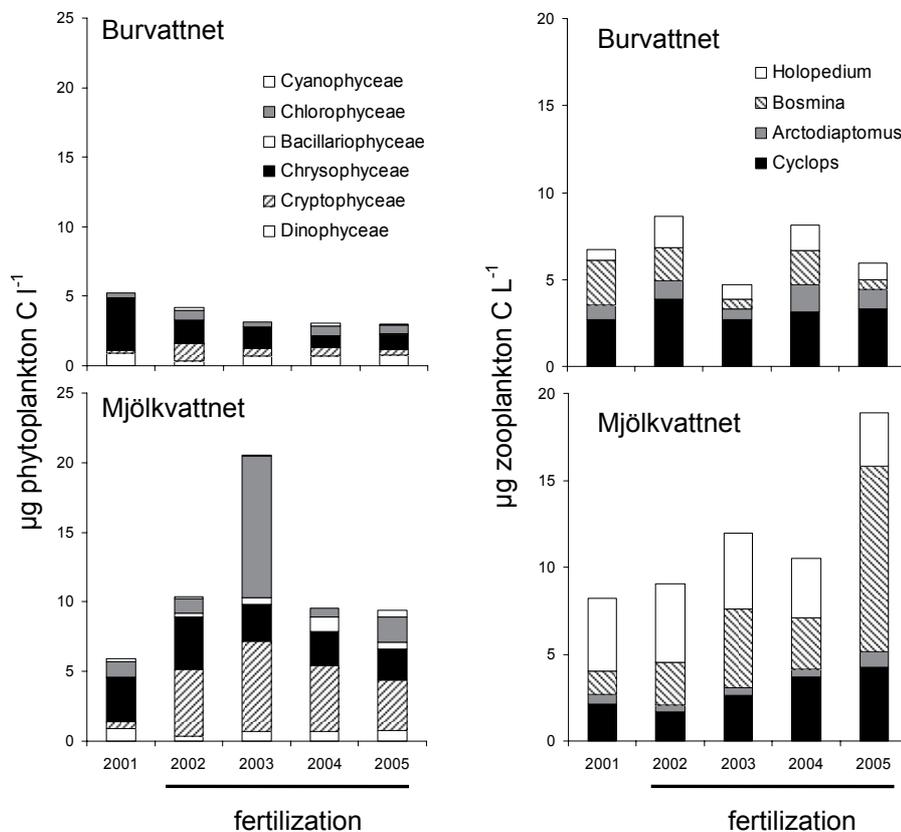
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Figure 8 The biomass and composition of the phytoplankton and zooplankton communities in Burvattnet and Mjölkvattnet.

Chrysophyceae and *Cryptophyceae* that dominated the phytoplankton biomass both have beneficial FA contents and are food of good quality for zooplankton (Ahlgren *et al.* 1990; Brett & Muller-Navarra 1997). Even though we did not measure seston FA after 2003 the phytoplankton composition was such that it is highly unlikely that the FA concentrations changed considerably during the latter years.

The increase in phytoplankton and zooplankton biomass in Mjölkvattnet was followed by increases in the fish stock, of both Arctic char and Brown trout, and the fish stocks decreased in Burvattnet during the same period (Milbrink *et al.* unpublished). The increased fish stock in Mjölkvattnet may have affected the zooplankton community: the biomasses of *Bosmina* and *Cyclops* both increased during the nutrient additions, whereas *Holopedium* decreased over this period (fig 8).

The results from this enrichment study shows that gentle fertilization of oligotrophicated systems can increase the production without causing undesirable shifts in the original species composition in the plankton communi-

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ties, and also without decreasing the lake's aesthetical values. This implicates that this kind of lake treatment provide an acceptable method to restore the fish production to pre-impoundment levels in these types of lakes.

Can food quantity and quality regulation of *Daphnia* growth be predicted across a lake trophic state gradient? (**V**)

Seston partC and P:C ratio were significantly positively correlated to TP (fig 9a, 9b), whereas the EPA:C ratio of seston showed a significant unimodal relationship with TP (fig 9c).
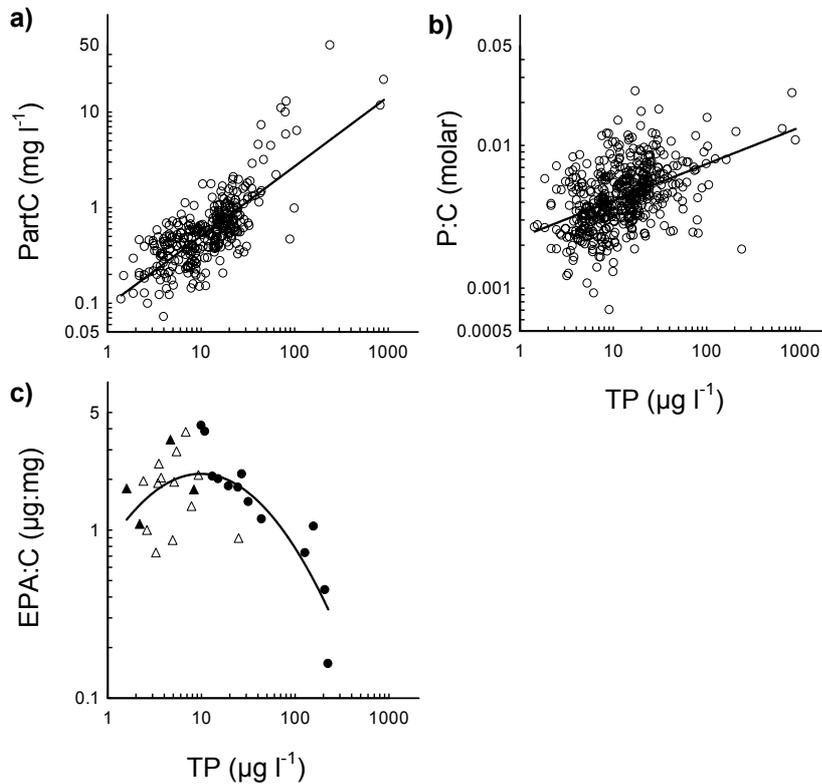

Figure 9 Regressions between TP and seston variables. **a)** For particulate carbon. **b)** For P:C ratio. **c)** For EPA:C ratio, observations presented by ● are from (Müller-Navarra *et al.* 2004), and those by △ or ▲ are from Persson & Vrede (unpublished), empty symbols were 40 μm prefiltered. Modified from Persson et al (2007).

In accordance with the trend in figure 9a, the RGD calculated from the TP regression predicted that the low partC in oligotrophic systems should coin-

cide with a large *RGD* of *Daphnia*. Growth depression due to low food availability decreased with increasing TP and was virtually absent at TP>63 µg l^{-1}. The *RGD* due to P:C was small at TP 2.5 µg l^{-1}, declined at TP 6.3 µg l^{-1}, and decreased to even lower levels at higher TP. When using the EPA content of field seston, *Daphnia* RGD due to low EPA:C was at a minimum at TP 10 µg l^{-1} and gradually increased with decreasing and increasing TP. The EPA:C model based on EPA supplementation experiments predicted maximum *Daphnia RGD* at TP 100 µg l^{-1}, a minimum *RGD* at TP 10 µg l^{-1}, and intermediate *RGD* at TP 2.5 µg l^{-1}.

When using the model based on field seston for the EPA limitation, the combined *Daphnia RGD$_{tot}$* were lowest at TP 16 and 25 µg l^{-1} (fig 10a). When the EPA limitation from **II**, using supplemented algal cultures, was combined the minimum *Daphnia RGD$_{tot}$* were found at TP 25 and 40 µg l^{-1} respectively (fig 10b).

Figure 10 The total relative growth rate depression of *Daphnia* across the gradient of TP. **a)** According to the field seston model for EPA:C growth depression. **b)** According to the EPA addition model based on **II** for EPA:C depression, using the multiplicative interaction between factors. The boxed line is the median observation, the margins of the boxes indicate the 25th and 75th percentiles, and the whiskers indicate the 10th and 90th percentiles. Modified from Persson *et al.* (2007).
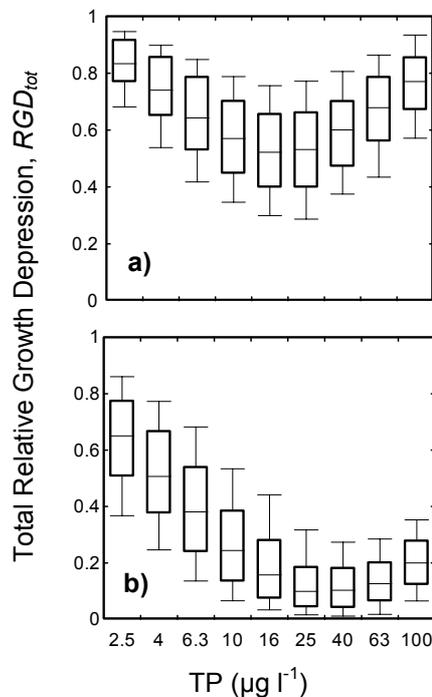
We used the results of a study that identified EPA as the main limiting agent on *Daphnia* growth by correlating the growth of experimental animals with the content of field collected seston, dominated either by diatoms or cyanobacteria (Müller-Navarra *et al.* 2000). Their results assumed that the EPA content is the main difference between the diatoms and the cyanobacteria. By comparing their results with the results of **II** in which we kept all variables constant but the diets content of EPA, we were able to point out that there were great differences in the conclusions when using the results from

these two studies. And that Müller-Navarra et al (2000) most likely included more limiting agents than EPA in what they refer to as EPA limitation.

The model in this study predicts that the strength and nature of food quantity and quality limitation of *Daphnia* growth varies with lake trophic state, and that some combination of food quantity and/or quality limitation should be expected in nearly all lakes. Food availability was predicted to be the dominant constraint on *Daphnia* growth in the most nutrient poor conditions and EPA limitation the dominant constraint in the most productive lakes. Furthermore, *Daphnia* growth rates are expected to be highest in lakes with intermediate trophic status. Our model thus make predictions of patterns that can be tested in empirical investigations, and together with potential future results we believe that this study holds promise to enhance our understanding of how carbon transfer efficiency is linked to lake trophic status, and thus improve our ability to predict effects of environmental changes on ecosystem processes and biogeochemical cycles.

Conclusions and perspectives

Conclusions

The major conclusions that can be drawn from my thesis are:

1. The RNA:DNA ratio was closely connected to growth rate in *Daphnia*. This indicates that there are connections between RNA content, P content, and high growth rates.

2. EPA limitation of *Daphnia* somatic growth occurred at food EPA concentrations below 0.9 μg mg C^{-1}. The threshold for egg production was lower than for somatic growth, indicating that eggs were prioritized when the access to EPA was low, or that the EPA demand for somatic growth was higher than for egg production.

3. There were patterns in the zooplankton PUFA content that was explained by their taxonomy and trophic position. *Cladocera* enriched EPA and ARA relative to seston, and *Copepoda* primarily enriched DHA. The proportion of 20 and 22-carbon PUFA increased with increasing trophic position in the food web.

4. In a whole-lake experiment, gentle fertilization of an oligotrophicated reservoir increased the seston P content and the biomass of high quality phytoplankton (*Cryptophyceae*, high EPA content). Thus, stimulating a higher and efficient energy transfer from phytoplankton to higher trophic levels.

5. Our model predicted that the strength and nature of food quantity and quality limitation of *Daphnia* growth varies with lake trophic state, and that some combination of food quantity and/or quality limitation should be expected in nearly all lakes. Food availability was predicted to be the dominant constraint on *Daphnia* growth in the most nutrient poor conditions and EPA limitation the dominant constraint in the most productive lakes. Furthermore, *Daphnia* growth rates are expected to be highest in lakes with intermediate trophic status.

Perspectives

When we understand more about what determines the composition of organism biomass, we will also be closer to our goal of understanding how substances are transferred in food webs and how ecosystems work. The growth rate hypothesis that was supported by **I** has since been developed further and expanded to biota other than *Daphnia* (Elser *et al.* 2003). We have come a

long way in understanding how nutritional demand of P interacts with RNA content, growth rates, and even intergenetic spacers (Elser *et al.* 2000b). But there are probably elements other than P, or biochemicals that can provide similar connections between nutritional demands and life histories. The results from **III** indicate that there are differences in the PUFA composition of zooplankton that might be explained by their life history. PUFA are likely to be more complicated than P in this case since zooplankton content of PUFA is more flexible (Brett *et al.* 2006) than zooplankton P content. Feeding experiments with several taxa of zooplankton and food with varied PUFA content will be required to tease apart the effects of taxonomy and the effects of the foods composition.

The threshold P:C ratio for *Daphnia* growth reduction is relatively well known (Brett *et al.* 2000) due to the many studies that has addressed this. But only a few studies have estimated the threshold concentrations of PUFA. Of these **II** is the most convincing so far, but many more are needed before we can state that we actually know something regarding the EPA threshold concentration. Some kind of interaction of EPA with other PUFA or some other substance(s), possibly sterols, was indicated in **II** since the threshold for *Daphnia* growth rates were quite different when two different experimental algae were used. Was the lower EPA threshold when using green algae due to that other PUFA alleviated the growth rate depression due to low EPA or were other PUFA converted to EPA? At least some animals probably have the ability to convert substantial amounts of one PUFA to another PUFA. This ability is thought to be limited in *Daphnia*, but very little is known about other cladocerans, and in particular about the copepods. Results in **III** indicate that the copepods in that study might have been able to synthesize DHA and 22:5ω6 from other FA since these were much more concentrated in the copepods than in seston. The results of von Elert (2002) indicated that the *Daphnia galeata* in his study were able to convert 18:3ω3 given in their food to EPA in order to relieve the effect of EPA limitation. But there was no effect of SDA supplementation on the *Daphnia pulex* growth in **II**, despite a clear effect when EPA was given. This indicates that there could be variation in the ability to convert PUFA also within taxa.

In future studies it is important to clarify the physiological role of different PUFA, to what extent PUFA can be used interchangeably, and also to what extent zooplankton can convert one PUFA to another.

We drew our conclusions regarding food quantity and food quality effects in **IV** on knowledge based on *Daphnia* experiments. It has been apparent for a number of years that other taxa than *Daphnia* should preferably also be used in these types of experiments to be able to draw more general conclusions. There have been a few experiments on *Bosmina*, but questions regarding taxa such as *Holopedium gibberum* and copepods have not been sufficiently addressed, despite that these are common in many lakes. These taxa are un-

fortunately not as easily experimented upon as *Daphnia*. *Holopedium* is very difficult to culture, and copepods are also difficult and slow to culture. And in addition to this, copepods are also very selective about what they eat, making supplementation experiments more difficult. But hopefully these obstacles can be overcome, and if we can expand our knowledge of food quality constraints to more taxa than *Daphnia* this would improve our knowledge about other taxa in the field, and also potentially help us understand more about connections between nutritional demands and life histories.

V provide a step towards better understanding of where food quantity and/or food quality are of importance but future studies are required to empirically test both the underlying assumptions of the models and the model predictions:

1. The models in **V** need to be tested. It is not clear how different growth limiting parameters interact with each other, and which kind of interaction is the best descriptor of how food quality factors interact. The empirical support for any of the model used in **V** is at present weak or absent, and there is a need for empirical tests of these alternative models.

2. To test the predictions in **V** regarding limiting agents and magnitude in RGD there is a need for simultaneous food quantity and food quality measurements *in situ*. Complemented with growth experiments with natural seston and factorial manipulations of food quantity and food quality. Food quality being the P and EPA content, but also the sterol and amino acid content, of the diet.

Summary in Swedish (Sammanfattning)

Effekter av födokvalitet på djurplanktons tillväxt och på energiöverföringen i födovävar i sjöar

Eftersom människor, av naturliga skäl, är den art på den här planeten vi känner bäst till så vet vi väldigt mycket om vad som bör finnas i vår diet och vad som bör undvikas. Det är inte bara hos människor som det spelar en roll vad som finns i maten; vad som finns i födan kan ha stora effekter även på andra organismer. Ett storslaget exempel är att de otroligt stora gnu- och zebrahjordarnas årstidsvandringar i Serengeti och Masai Mara, i varje fall till viss del, kan vara styrda av fosfor- och kväveinnehållet i gräset de betar.

I den öppna vattenmassan i sjöar går energin vanligtvis från solen in i växtplankton och sedan vidare via djurplankton upp i högre nivåer i näringskedjan. Det har visat sig att näringsinnehållet i växtplankton kan ha en stor effekt på hur bra djurplankton växer, och riktigt dålig födokvalitet kan till och med göra att djurplankton inte växer alls. Födokvalitén kan alltså ha stora effekter på djurplankton, och eftersom djurplankton utgör en viktig födokälla för fisk så kan födokvalitet ha stor påverkan på hela näringsväven i sjöar.

Den här avhandlingen handlar om hur stor negativ inverkan födokvalitén och födomängden kan ha på tillväxten hos djurplankton i olika sjöar av olika näringsinnehåll, och om viktiga skillnader i kraven på födans kvalitet mellan olika grupper av djurplankton. I arbetet för den här avhandlingen ingår två labexperiment (**I & II**), en serie fältprovtagningar (**III**), ett helsjöexperiment (**IV**), samt en modellstudie (**V**) som är baserad på en litteratursammanställning.

Genom att förstå mer av vad som styr variationen i levande organismers innehåll kommer vi även att veta mer om hur ekosystem fungerar. Resultaten från det första labexperimentet (**I**) visade att tillväxthastigheten i *Daphnia* är tydligt kopplad till mängden RNA. Och eftersom RNA innehåller en stor andel av fosforn i *Daphnia* stödjer resultaten en koppling mellan fosforinnehåll, RNA-innehåll, och tillväxthastighet.

För att vi ska kunna ha en uppfattning om när och var olika tillväxtbegränsande ämnen är viktiga behöver vi veta vid vilka koncentrationer de börjar påverka tillväxten. Det andra labexperimentet (**II**) visade att den fleromättade omega3-fettsyran EPA (20:5ω3) var begränsande för tillväxten hos

Daphnia när koncentrationen i födan var lägre än 0,9 µg EPA per mg kol. Detta betyder att EPA kan vara en viktig begränsande faktor i många sjöar eftersom lägre EPA-koncentrationer är vanligt förekommande.

Fältprovtagningen av djurplankton i Jämtland (**III**) visade att det fanns tydliga skillnader i innehållet av fettsyror mellan djuren och den föda de ätit. Hinnkräftor innehöll framförallt fettsyrorna EPA och ARA (20:4ω6) medan hoppkräftor innehöll mestadels DHA (22:6ω3). På ett liknande sätt som att fosfor hör ihop med snabb tillväxt så finns det eventuellt egenskaper hos dessa djur som hör ihop med de olika fettsyrorna. Vi spekulerar i att EPA och ARA är viktiga för äggproduktion medans DHA är viktigt för de väl utvecklade sinnesorganen hos hoppkräftorna. Det fanns även tydliga skillnader mellan djurgrupper med olika dieter, rovdjur innehöll mer av de längsta fleromättade fettsyrorna än växtätare. Resultaten från fältprovtagningen kan betyda att olika grupper av djurplankton har olika krav på födans innehåll av fettsyror, och även att de är av olika näringsvärde för rovdjur som äter djurplankton.

Jag studerade hur mängden föda och födokvalitén påverkades under fyra år av näringstillsättningar till en extremt näringsfattig vattenreservoar i norra Jämtland (**IV**). Som en följd av att den reglerades blev vattenreservoaren extremt näringsfattig och fiskbeståndet minskade drastiskt i den. Att stimulera produktionen av plankton och fisk genom att tillsätta näring är en metod för att kompensera den negativa effekten av regleringen. I näringfattiga sjöar finns det väldigt lite föda, men den som finns är ofta av relativt bra kvalitet. För att kunna stimulera tillväxten av djurplankton och fisk i dessa är det viktigt att inte födokvalitén blir dålig efter näringstillsatserna. Resultaten visade att vår näringstillsats ökade mängden växtplankton, och ökningen bestod av arter som innehåller stora mängder av viktiga fettsyror. Koncentrationen av fosfor i seston var också den bra efter näringstillsatserna. Att födokvalitén var bra efter näringstillsatserna märktes också på att mängderna av djurplankton och fisk ökade i sjön, vilket indikerar att den tillsatta näringen överfördes effektivt i näringskedjan.

Baserat på en litteratursammanställning över hur mängden av föda, samt innehållet av fosfor och EPA förändras med av näringsinnehållet i sjöar, gjorde vi en modell på hur dessa faktorer påverkar tillväxt i *Daphnia* i sjöar med olika näringsinnehåll (**V**). Den förutsäger att födomängden är den viktigaste faktorn som begränsar djurplanktons tillväxt i näringsfattiga sjöar, att fosforinnehållet i seston är lågt och mest begränsande i mycket näringsfattiga sjöar, samt att EPA samt andra oidentifierade ämnen i seston har de största begränsande effekterna i näringsrika sjöar. Sammantaget visar resultaten från modellen att någon typ av födomängds- eller födokvalitetsbegränsning kan förväntas i alla sjöar. Och energiöverföringen från växtplankton till djur högre upp i näringskedjan i den öppna vattenmassan förutspås vara effektivast i sjöar med en medelhög näringsnivå.

Sammantaget bidrar resultaten från den här avhandlingen med att ge en uppfattning om i vilka typer av sjöar fosfor och EPA är mest troligt att fungera som viktiga tillväxtbegränsande faktorer för djurplankton, samt att sätta födokvalitetseffekterna i relation till födomängdseffekterna. Resultaten visar också att det finns skillnader i fettsyreinnehållet i olika djurplanktongrupper vilket kan ha viktiga konsekvenser för både djurplankton och rovdjur som äter dem.

För att återkomma till mänsklig födokvalitet så är det slående att samma omega-3 fettsyror som är viktiga i människors diet är viktiga även för djurplankton, nämligen EPA och DHA. Och eftersom de omega-3 fettsyror vi äter framför allt kommer från akvatiska system gör det än mer viktigt att veta mer om hur dessa fettsyror produceras och transporteras i dessa system.

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