

# WATERS



## STATISTICAL WORKSHOP ON GRADIENT STUDIES

Uppsala, 13-15 February 2012

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Anders Grimvall, Stina Drakare & Leif Pihl

### WATERS partners:



WATERS - Waterbody Assessment Tools for Ecological Reference conditions and status  
in Sweden

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## Executive summary

The first statistical workshop in WATERS was held in Uppsala from 13<sup>th</sup> to 15<sup>th</sup> of February 2012 with the aim of discussing statistical principles for the design of the freshwater and marine gradient studies. A total of 25 persons attended the workshop that was partly comprised of four statistical lectures on experimental designs, power analyses and statistical methods for visualising data in addition to group discussion, split into gradient studies for freshwater and marine waters. Based on the statistical lectures and fruitful discussions the designs of the two gradient studies were advanced and became more specific. The workshop laid the foundation for the final designs to be completed in spring 2012. In the assessment of the workshop the participants were overall positive about the outcome of the workshop.

## Svensk sammanfattning

Den första workshopen om statistik inom WATERS hölls i Uppsala 13 till 15 februari 2012. Syftet var att diskutera statistiska principer för designen av de gradientstudier som planeras inom både havs- och sötvattengrupperna. Totalt deltog 25 personer. Mötet bestod av fyra föreläsningar inom experimentell design, styrkeanalys och statistiska metoder för att visualisera data. Därtill fanns generöst med tid för diskussioner inom respektive fokusområde om gradientstudiernas design. Resultatet av blandningen av föreläsningar och diskussioner blev långt framskridna planer på studierna upplägg och design. Workshopen lade därmed grunden för den slutgiltiga designen som kommer att fastställas under 2012. Vid utvärderingen av workshopen var deltagarna på det hela taget mycket nöjda med de tre dagarna i Uppsala.

## Introduction

The first statistical workshop in WATERS was held in Uppsala from 13<sup>th</sup> to 15<sup>th</sup> of February 2012 at the premises of the Swedish University of Agricultural Sciences (SLU). An invitation for participating in the workshop was sent out 2 January to all participants of the WATERS project with the request for interested parties to submit an expression of interest before 1 February 2012. For practical reasons the number of participants was limited to 25 persons. There was a large interest for participating in the workshop and a total of 25 persons attended.

The objective of the workshop was to discuss the design of gradient studies, both on the freshwater and marine side, from a statistical perspective. The workshop included four statistical lectures and presentations of the two gradient studies. The group work/discussions were divided into a freshwater group and a marine group. The group discussions were supported by five trained statisticians.

This summary report contains a short description of all the presentations, the agenda for the workshop and a list of participants.

## Principles for experimental design and analysis of variance

This lecture was given by Claudia von Brömssen from the Swedish University of Agricultural Sciences.

In the beginning the similarities between regression analysis and analysis of variance (ANOVA) was introduced, demonstrating how different data sets can be analysed by both types of analyses. The focus of the lecture was on the different types of experimental designs. The assumptions and general principles underlying different types of designs was presented as exemplified with one-way ANOVA, two-way ANOVA, blocking, repeated measures and split-plot models. The lecture also included a presentation on the interpretation of interactions in ANOVAs. Finally, differences between fixed and random factors were discussed, and the consequences of choosing one or the other for the interpretation of the results.

## Gradient studies and experimental design

This lecture was given by Thorsten Balsby from Aarhus University.

Gradient studies differ from classic experimental designs, as the researcher does not have control of the dependent variables. When planning a gradient study the researcher should try to ensure that there will be variation along the gradients. If gradient studies should make predictions about the response of an ecosystem to a pressure; it is assumed that the

ecosystem response to the pressure should follow the same trajectory in response to increase and decrease of pressures. However, oftentimes ecosystems follow different trajectories in response to increases and decreases of pressures. I presented two case studies on gradients, one from the Everglades to illustrate how to take care of spatial dependencies and why mixed models could do this. The other case study was on uncertainty of estimates of the eelgrass depth limit. In the eelgrass study we parameterized how the uncertainty varied with the depth for number of years, divers, transects and replicates. For a 6-year survey period, we then estimated the depth specific variance for combinations of years, divers, transects and replicates, and how much time each combination would cost. If a maximum of 100 h could be spent on a survey the lowest uncertainty on estimates of the maximum depth limit would be achieved using 2 years, 2 or 3 divers surveying the transect at the same time and 3-7 transects. The recommendation for surveys of eelgrass depth limit would therefore be a survey that lasts at least 2 years, use at least 2 divers and survey at least 3 transects. Usually only one diver has been used in surveys, but by using 2 divers the uncertainty of an estimate of the depth limit could be reduced to about 60 %.

## Statistical power

This lecture was given by Jacob Carstensen from Aarhus University.

Every statistical test has two types of errors: Type I error (probability of  $\alpha$ , typically max 5%) is rejecting the null hypothesis when in fact it is true, and Type II error (probability of  $\beta$ , typically max 20%) is accepting the null hypothesis when in fact it is not true. The power is defined as  $1-\beta$  and describes the probability of actually rejecting the null hypothesis when it is not true, i.e. detecting the difference present. Power can, for simple statistical tests, be calculated explicitly using formulas from statistical textbooks, however, for more complicated analyses the easiest approach to calculate power is through Monte Carlo simulation where the alternative hypothesis (a putative difference) is simulated many times and the proportion of tests that reject the null hypothesis is calculated. Such calculations were shown in SAS using a linear regression as example. The presentation also discussed power when spatial or temporal correlation is present, which reduced power. For experimental designs it can be an advantage to do a “virtual sampling”, that is a computer simulated sampling according to the design, and then analyse the simulated data with the proposed model for the analysis. The presentation also included a general introduction to the principles of experiment design, from formulating the ecological hypothesis, transforming this into a statistical problem formulation with distributional assumptions and hypotheses, which can finally be used to calculate power for a given number of observations.



## Visualization of environmental data

This lecture was given by Anders Grimvall from Havsmiljöinstitutet.

Our interpretation of environmental data is often to a great extent based on graphical illustrations of the collected data, and there is a long tradition to draw different types of plots. During the past years, the need to visualize fairly large datasets has grown stronger. This is partly due to the fact the amount of available data has increased. However, it is also clear that the most burning environmental issues are related to large-scale changes of the environment. Here, chemical and physical water quality data were taken as a point of departure for a discussion of data visualization and how such techniques can make it easier to move from overview to details, and vice versa. In particular it was emphasized that animated scatter-charts can facilitate the detection of trends and change-points in large datasets. Such techniques have two advantages: (i) a large number of charts can readily be produced and inspected; (ii) in a long sequence of charts, each chart can be inspected while the previous chart is still fresh in memory. In some software packages or programming languages (e.g. R) the animation is produced by making a video of a sequence of images. In spreadsheet environments (e.g. Excel), graphs are automatically updated when a macro is employed to step by step modify the contents of a spreadsheet. More information about animation can be found on the websites [www.miljostatistik.org](http://www.miljostatistik.org) and by using the link <http://www.intechopen.com/articles/show/title/visual-detection-of-change-points-and-trends-using-animated-bubble-charts>

## FA3 report on marine gradient study

The marine gradient study was presented by Leif Pihl from Gothenburg University.

Assessment of ecological status according to Water Framework Directive (WFD) is based on four different quality elements (i.e. phytoplankton, macrophytes, benthic invertebrates and fish). The objective of WP 3:5 is to design and conduct a field study where responses of indicators for these four quality elements are compared in eutrophication gradients. Indicators will be compared in relation to their sensitivity and functional response to nutrient loading in two coastal areas, one on the east coast and one on the west coast of Sweden. By this simultaneous comparison in fixed areas, the response of different indicators to pressures could be evaluated.

Dose-response relationships for different indicators will be compared along pressure gradients, where class boundaries and reference conditions will be evaluated. The gradient study will be carried out in areas where background data are available and if possible in cooperation with ongoing monitoring programs and other projects.

The two areas under study will be the fjord system inside Orust and Tjörn on the Swedish west coast and the archipelago of Östergötland in the Baltic. The sampling program will be designed to compare indicators response on two scales: between and within water bod-

ies in areas exposed to different nutrient loadings. On the west coast three water areas in a fjord system will be compared, with an off-shore area as a less disturbed reference. On the east coast studies will be conducted in six areas (3 in the inner and 3 in the middle archipelago), with one common less disturbed reference area in the outer archipelago. When comparing indicators between water bodies, data on nutrient status will mainly be based on information from existing long-term monitoring in each water body. To detect spatial variations in eutrophication status within water-bodies new samples of nutrients or proxy for nutrients (chlorophyll, Secchi-depth) are needed.

During the spring 2012 compilation of available background data will be conducted and a sampling program will be designed for the different indicators and nutrient status. A pilot study will be carried out during the period May-August, with the focus of testing relationships between various indicators and nutrient status along gradients. After evaluating the results from the pilot study the main investigating will be conducted in 2013.

## FA4 report on freshwater gradient study

The freshwater gradient study was presented by Stina Drakare from the Swedish University of Agricultural Sciences.

Assessment of ecological status of lakes and streams according to WFD is based on phytoplankton (only lakes), macrophytes, benthic diatoms, benthic invertebrates and fish. The objective of WP 4.6 is to conduct field studies to measure the precision and sensitivity of these quality elements to selected environmental pressures. Moreover, besides measuring the response of structural metrics to pressures, functional response variables (e.g. leaf-litter decomposition, ecosystem metabolism) will also be included in the gradient studies, which will allow comparison of structural versus functional response to selected pressures. Special focus will be on quantifying redundancy among the different response variables, and false positive and false negative errors to selected pressures. Data from the gradient studies will also be used to validate current classification criteria. The stressor response relationships across different organism groups and encompassing ecosystem function will be used to aid selection of the best organism group/functional metric to address specific stressors.

Three types of stressors will be studied; nutrient enrichment (lakes and streams in S. Sweden), hydro-morphological alteration (streams, N & S Sweden), and forestry or loss of riparian integrity (northern streams). The latter two pressure gradients currently do not have metrics, so these gradients will be used to develop and calibrate new metrics.

Discussions were mainly focussed on identifying regions with known pressures and sites for the gradient study. It was decided to do a GIS-search based on VISS-data to select lakes of L-N1 or L-N2a type, and, if possible, to combine impacted lakes with lakes of low nutrient enrichment from the national monitoring programs. Discussion of the stream gradient studies mainly focussed on choosing a representative pressure like channelization

or continuity for hydro-morphological alteration. Good suggestions of sites from Östergötland for both gradients of nutrient enrichment and hydro-morphological alteration were presented and it was decided to continue with this area in searching for adequate sites.

During 2012 compilation of available background data will be conducted and the sampling program designed in detail. We aim to do the lake gradients 2012 and all stream gradients during 2013.

## Agenda (in Swedish)

<b>Måndag 13/2</b>		
09:30	Fika	-
10:00	Start, allmän information	Ulf Grandin
10:10	Föreläsning. Försöksplanering I	Claudia von Brömssen
11:00	Rast	
11:10	Tankar kring upplägget på sötvattengruppens gradientstudie	Richard Johnson
11:30	Tankar kring upplägget på havsvattengruppens gradientstudie	Leif Pihl
11:50	Organisation av grupparbetet, gruppindelning <ul style="list-style-type: none"> <li>• Sekreterare, Rapportör</li> <li>• Rum: Biosfären, 4051 (14 platser), 4059 (6-7 platser)</li> </ul>	Ulf, Jacob
12:00	Lunch	
13:15	Grupparbete	
14:30	Föreläsning. Försöksplanering II	Thorsten Balsby
15:15	Fika och fortsatt grupparbete	
16:00	Avslutning	

<b>Tisdag 14/2</b>		
09:00	Föreläsning: Statistisk styrka	Jacob Carstensen
10:00	Fika och fortsatt grupparbete	
12:15	Lunch	
13:30	Tvärgruppsdiskussioner för summering om diskussionerna hittills	
14:30	Fika och fortsatt grupparbete, tid för specifika frågor till statistiker	
16:00	Avslutning	

<b>Onsdag 15/2</b>		
09:00	Föreläsning: Visualisering av data	Anders Grimvall
10:00	Redovisning av resultat från havsvattengruppen	
10:30	Redovisning av resultat från sötvattengruppen	
11:00	Eventuella ytterligare frågor	
11:30	Utvärdering av workshoppen /Nästa workshop	Jacob, Ulf
12:00	Lunch	
13:00	Fortsatt grupparbete/Avresa	
15:00	Avslutning	

## List of participants

<b>Deltagare</b>	<b>e-post</b>	<b>Organisation</b>
Anders Grimvall	Anders Grimvall <anders.grimvall@liu.se>	Havsmiljöinstitutet
Stina Drakare	Stina Drakare <Stina.Drakare@slu.se>	IVM, SLU
Kerstin Holmgren	Kerstin Holmgren <kers-tin.holmgren@slu.se>	Akvatiska resurser, SLU
Leif Pihl	Leif Pihl <leif.pihl@marecol.gu.se>	Göteborgs universitet
Martin Karlsson	Martin Karlsson <martin.karlsson@slu.se>	Akvatiska resurser, SLU
Per Olsson	Per Olsson <per.olsson@toxicon.com>	Toxicon AB
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**WATERS is financed by:**



SWEDISH ENVIRONMENTAL  
PROTECTION AGENCY