

4. Methods of Investigation

Biological Methods to assess Water Quality

Examples:

- **Physiological methods**
= experimental - ecological methods
- **Ecological methods**
= descriptive - analytic field methods

4. Methods of Investigation

Biological Methods to assess Water Quality

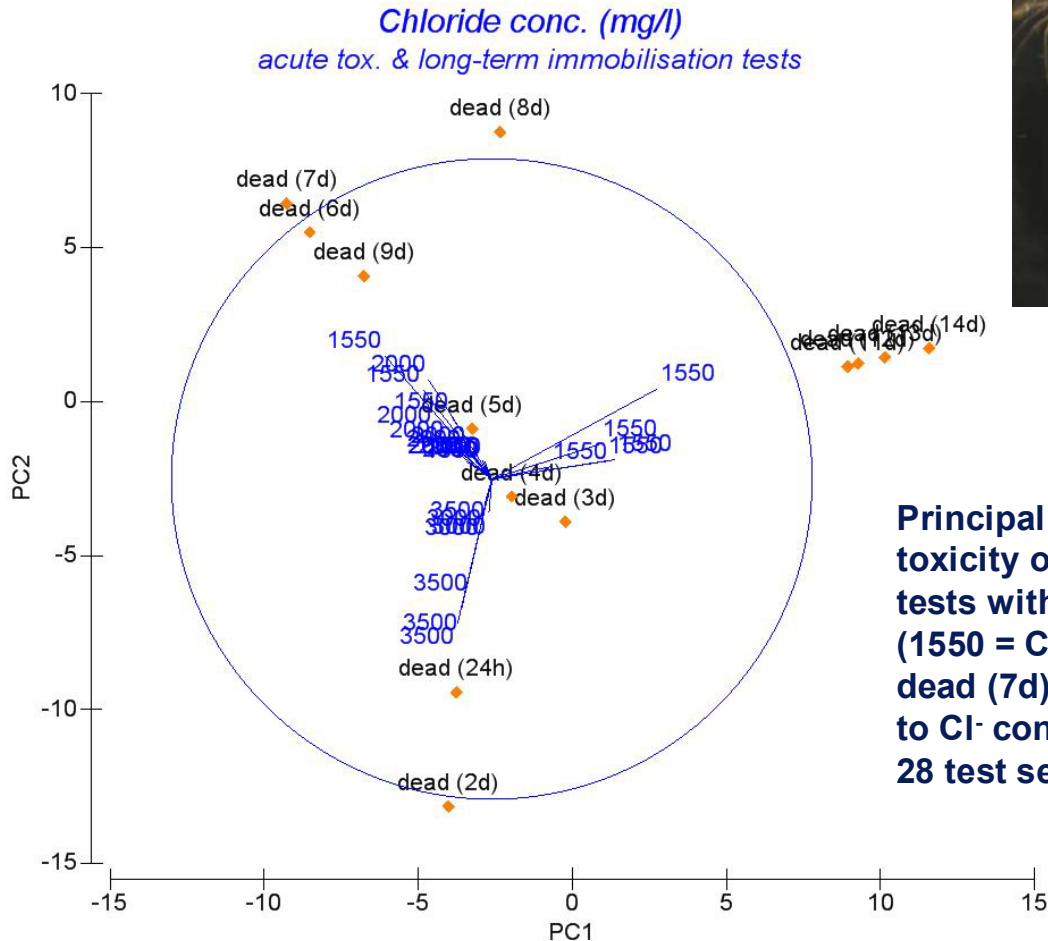
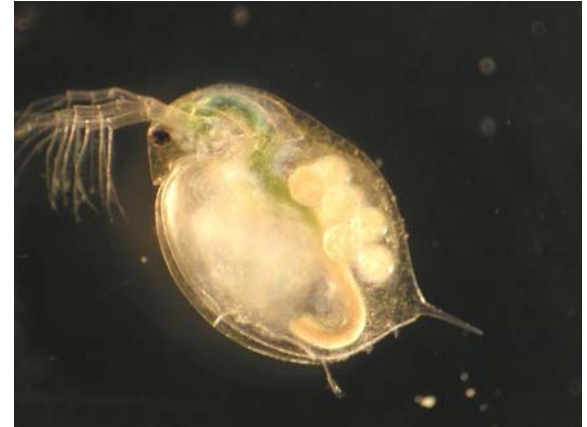
Physiological Methods = Biotests

- **Activity of particular metabolic features of the sample**
(*e.g.: Luminescent Bacteria Inhibition Test*)
- **Effect of the sample on the bioactivity of exposed organism**
(*e.g.: Toxicity Test with Daphnia magna*)

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Physiological Methods = *Toxicity Test with Daphnia magna*

- Acute immobilisation test (24 h)
- Reproduction test (21 day exposure)

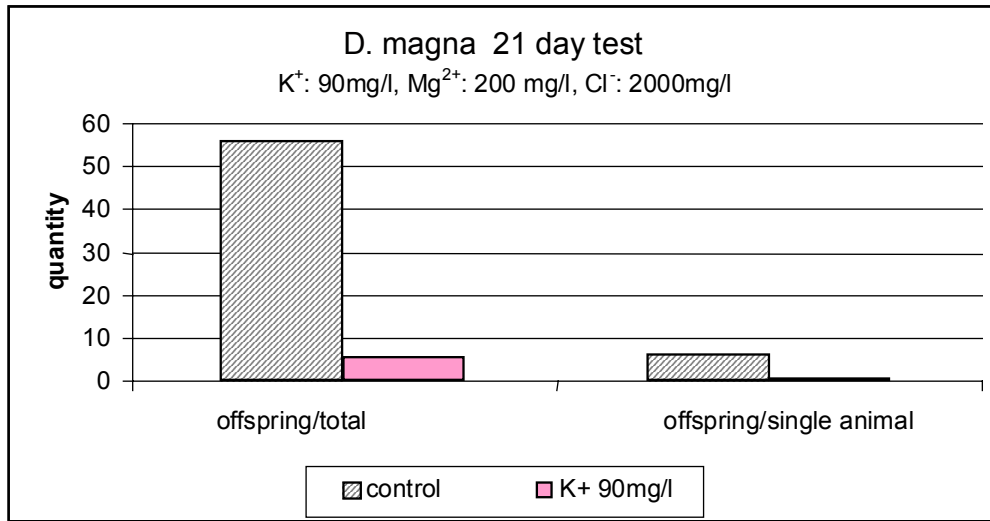


Principal component analysis of the toxicity of chloride concentrations in tests with *Daphnia magna*. (1550 = Cl⁻ conc. 1550 mg/l; dead (7d) = correlation of dead *Daphnia* to Cl⁻ conc. after 7 days). 28 test series applied.

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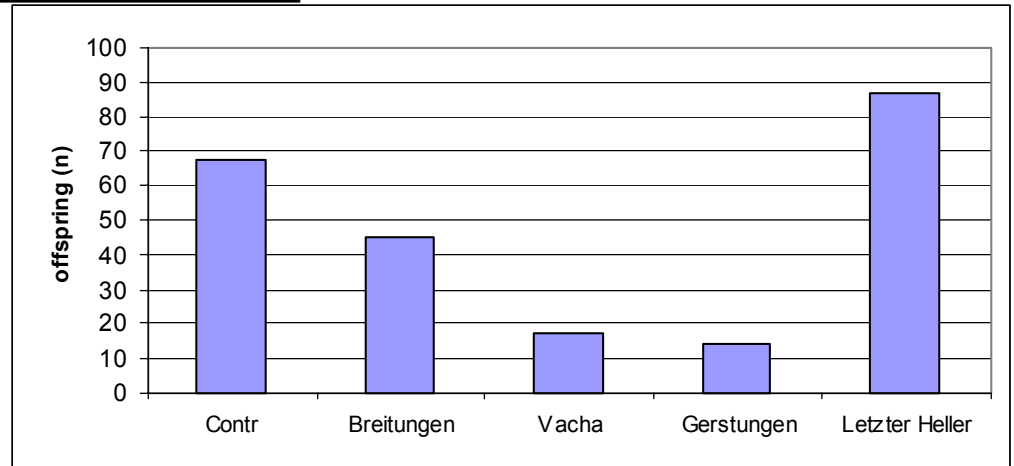
Physiological Methods = *Toxicity Test with Daphnia magna*

- **Reproduction test (21 day exposure)**



Comparison of the total number of offspring and the offspring per animal in the control and exposure groups.

Total number of offspring in 21-day test with water from different sections of the Werra.



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Biological Methods to assess Water Quality

Ecological Methods = Bioindication

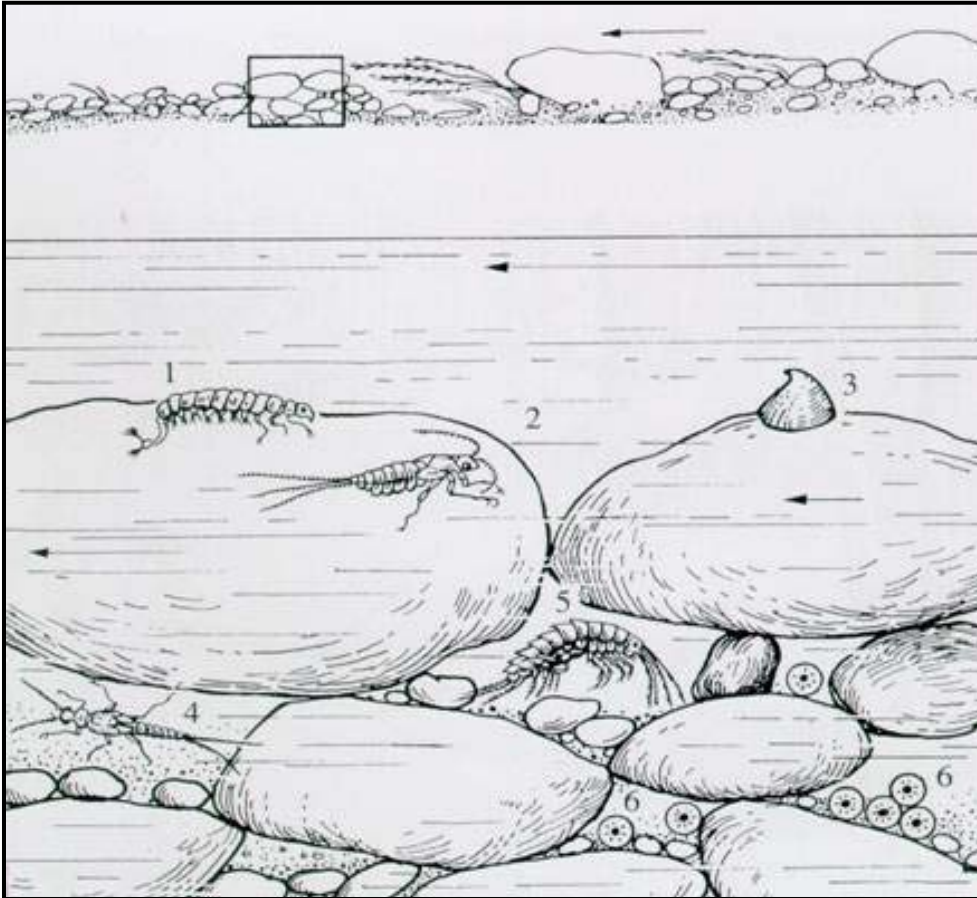
- Community of organism reflects the situation of the ecosystem
- Single Taxa have specific optima and tolerance ranges for selected environmental factor

= Biondicators

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Biological Methods to assess Water Quality

Ecological Methods = Bioindication with Macroinvertebrates



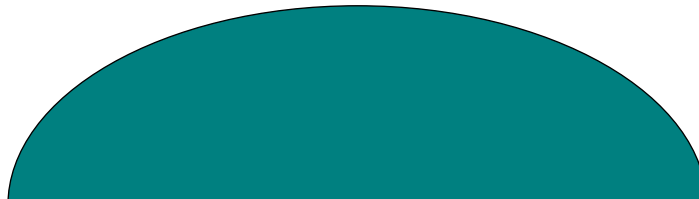
1. Caddisfly larvae
2. Mayfly larvae
3. Limpet (*Ancyclus fluviatilis*)
4. Stonefly larvae
5. Amphipod (*Gammarus* spp.)
6. Trout-eggs

4. Methods of Investigation

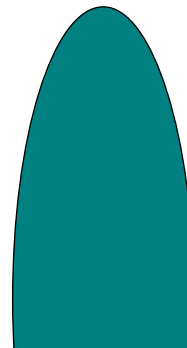
Ecological Methods = Bioindication with Macroinvertebrates

Tolerance Range:

◆ Euryoecious



◆ Stenoecious

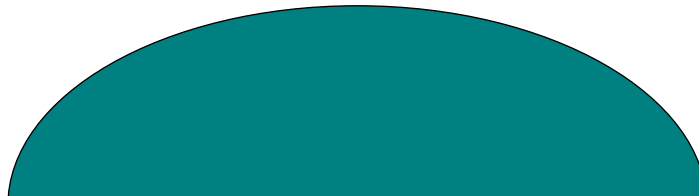


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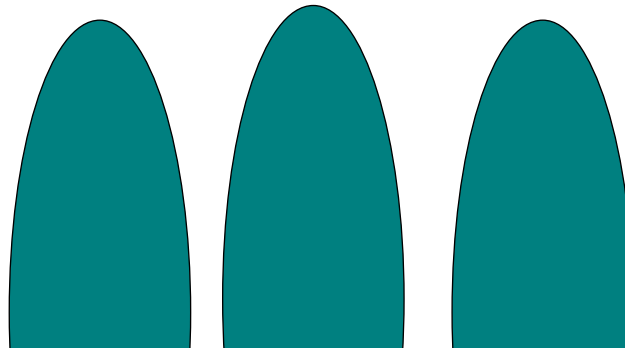
Ecological Methods = Bioindication with Macroinvertebrates

Tolerance Range:

◆ Euryoecious



◆ Stenoecious



4. Methods of Investigation

Ecological Methods = Bioindication with Macroinvertebrates

- Stenoecious organisms

Stonefly Larvae (Plecoptera)



4. Methods of Investigation

Ecological Methods = Bioindication with Macroinvertebrates

- Stenoeccious organisms

Mayfly Larvae (Ephemeroptera)



4. Methods of Investigation

Ecological Methods = Bioindication with Macroinvertebrates

- Stenoeccious organisms

Caddisfly Larvae (Trichoptera)



Plectrocnemia conspersa



Brachycentrus montanus

4. Methods of Investigation

Ecological Methods = Bioindication with Macroinvertebrates

- Stenoecious organisms

Freshwater shrimps (Amphipoda)



4. Methods of Investigation

Ecological Methods = Bioindication with Macroinvertebrates

- Stenoecious organisms

Worm (Oligochaeta)



4. Methods of Investigation

Ecological Methods = Bioindication with Oligochaeta

On and in Sediments	Sedimentfeeders: Naididae most times on sediments or in fine particulate organic matter; Tubificidae usually headfirst in sediment.
Epiphytic	especially Naididae
Soft substrates (silt, organic sediments)	Tubificidae with hair bristles, Naididae settle close to the sediment-surface (Aufwuchs-feeder), tube-building species (<i>Dero spp.</i>) invade the substrate up to 2 cm.
Sand, Gravel	Tubificidae without hair bristles, <i>Psammoryctides spp.</i> in the gravel interstices.

Oligochaeta always colonise the oxidised zones of the sediment, Tubificidae often occur in the lower boundary layer to the anoxic zone.

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Feeding habits of Oligochaeta

Tubificidae: with the anterior body buried in the sediment, great quantities of material are transported through the gut to the sediment-surface. With increasing amounts of organic substances in the sediment, the population density of tubificids increases as well.

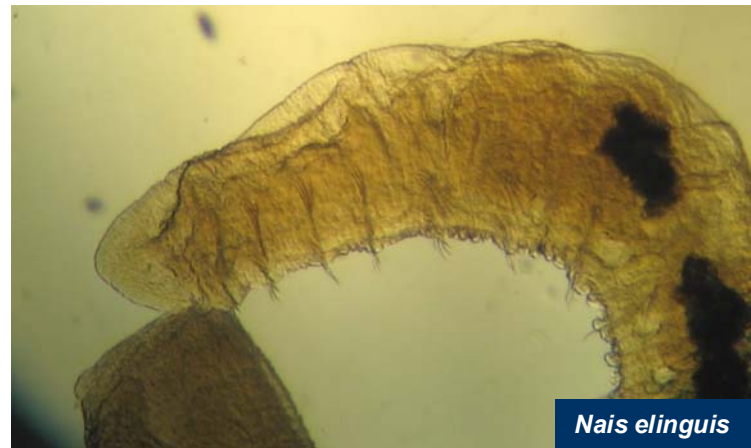
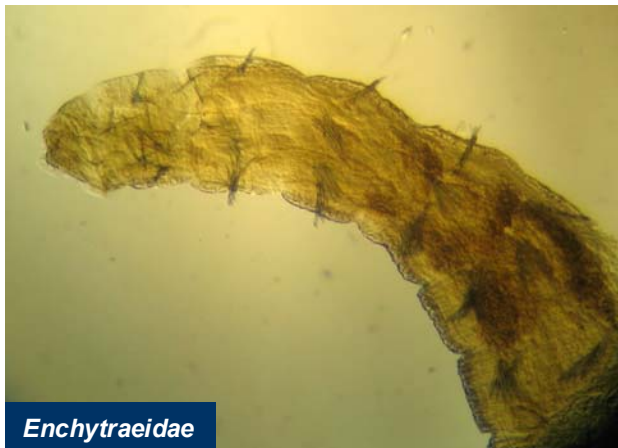


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Feeding habits of Oligochaeta

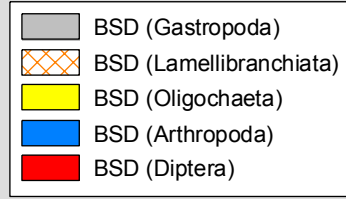
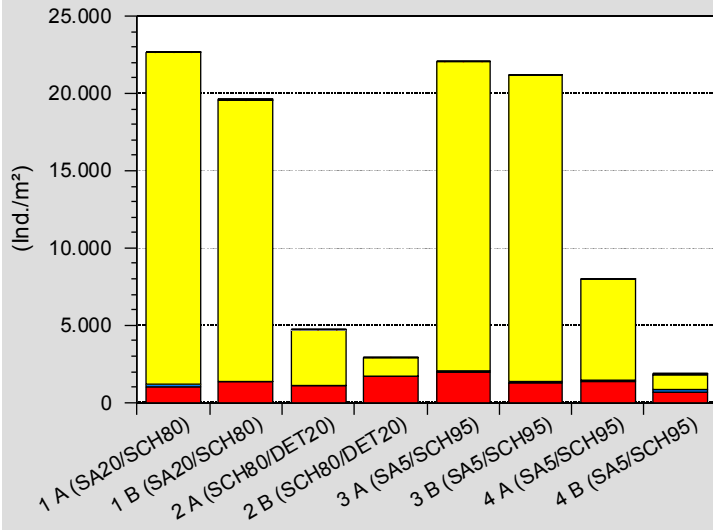
Tubificidae: the feeding-zone is at a depth of 1-3 cm. They don't feed directly at the sediment-surface or more than 6 cm below. The oligochaete habitat within the sediment consists of this feeding-zone and a subjacent migration area. Oligochaetes migrate horizontally to find new areas.

Sediment- and detritus-feeders show a low nourishment utilisation around 2 % - 50 %. Tubificidae assimilate only 4,1% of the incorporated sediment (Brinkhurst u. Austin 1979), in contrast to 60 % - 97 % in case of pure bacterial diet (Berry 1976). The microorganisms on the particles are the real food to these sediment-feeders.

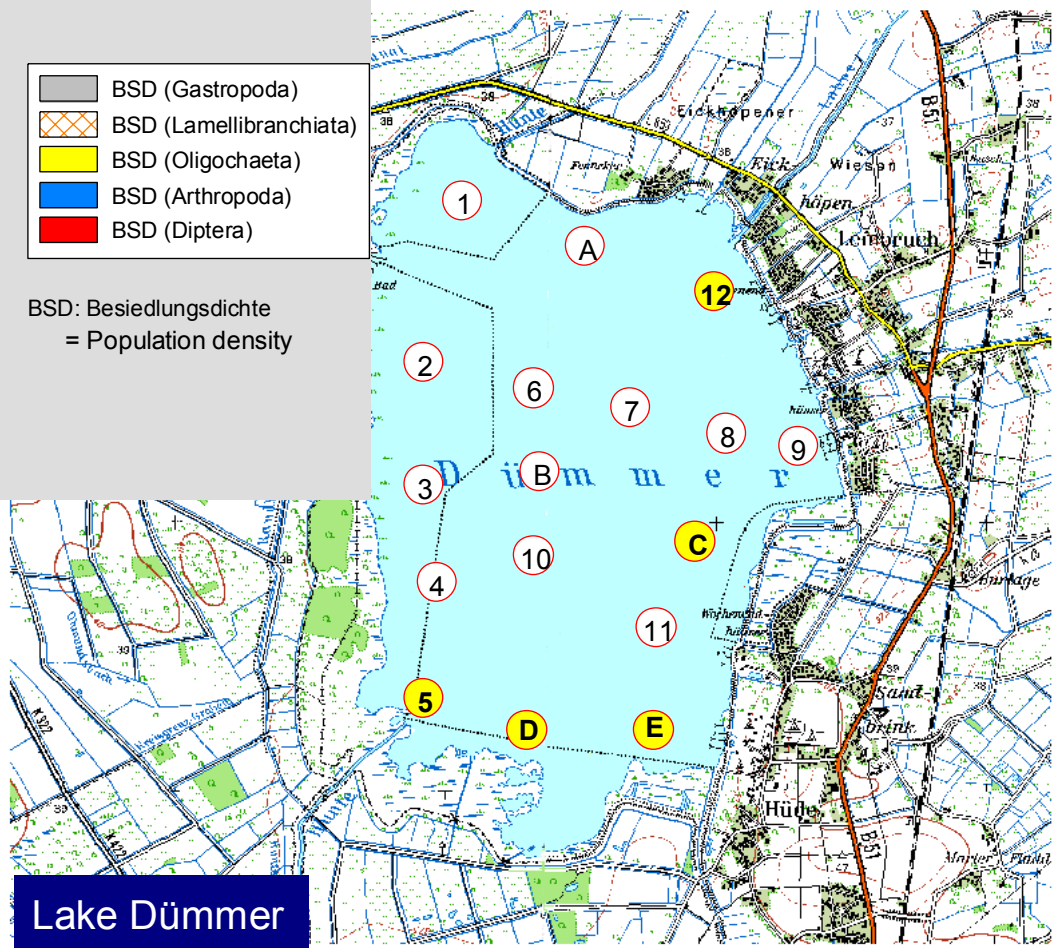


4. Methods of Investigation

Population densities

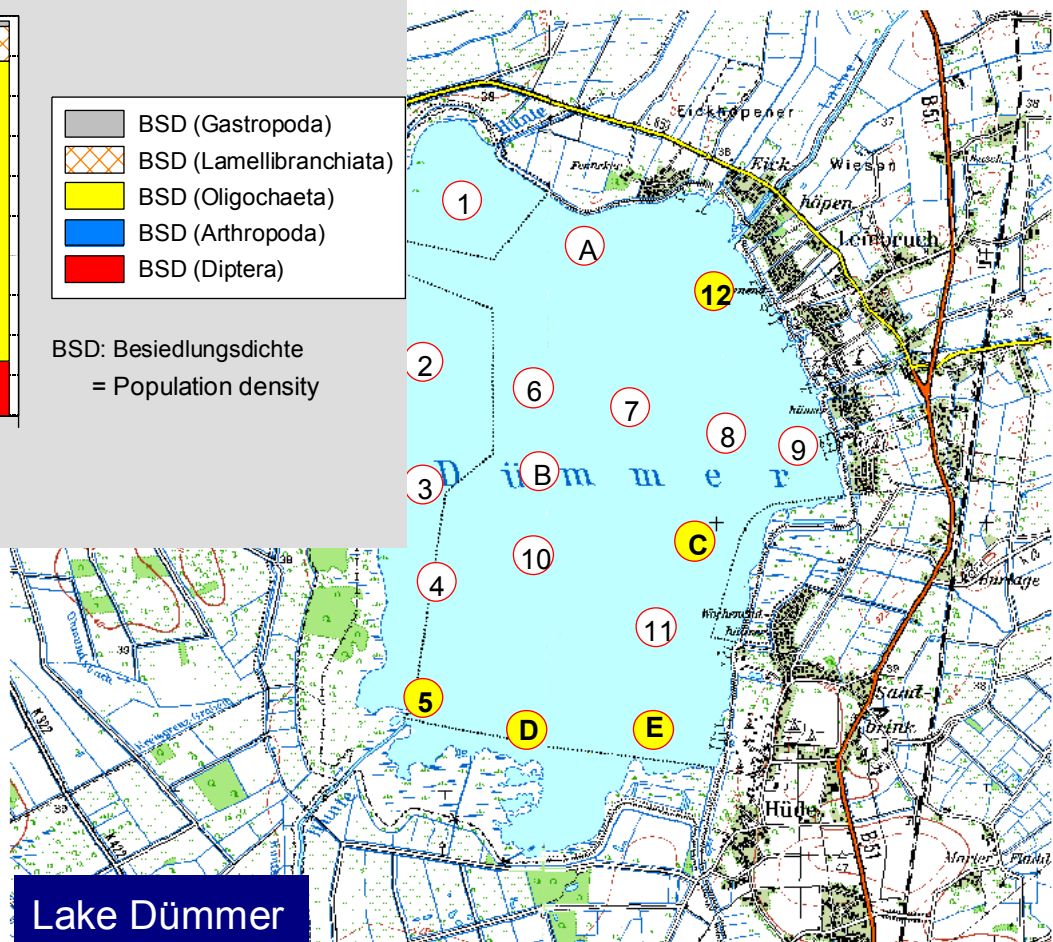
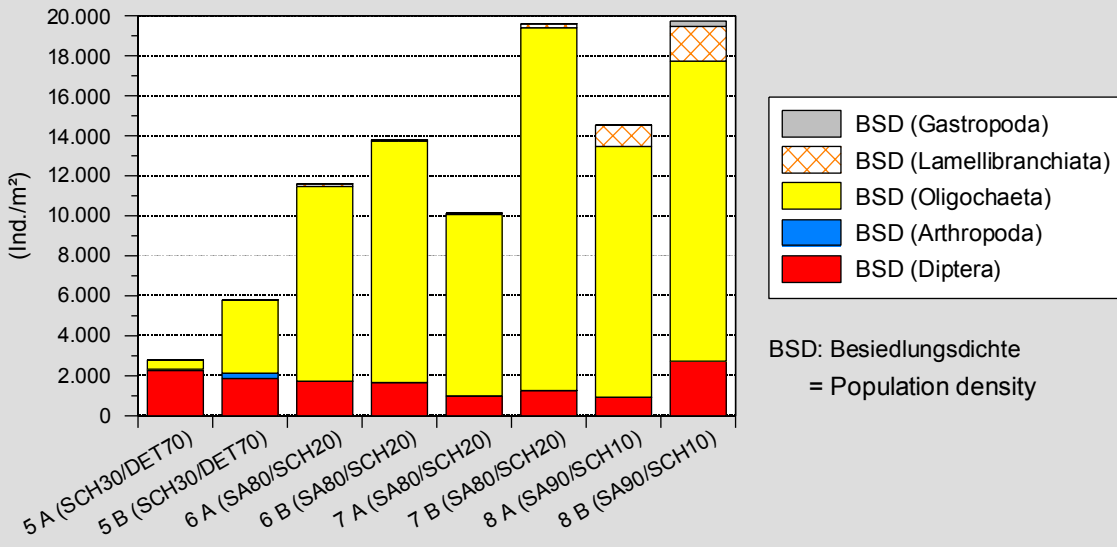


BSD: Besiedlungsdichte
= Population density



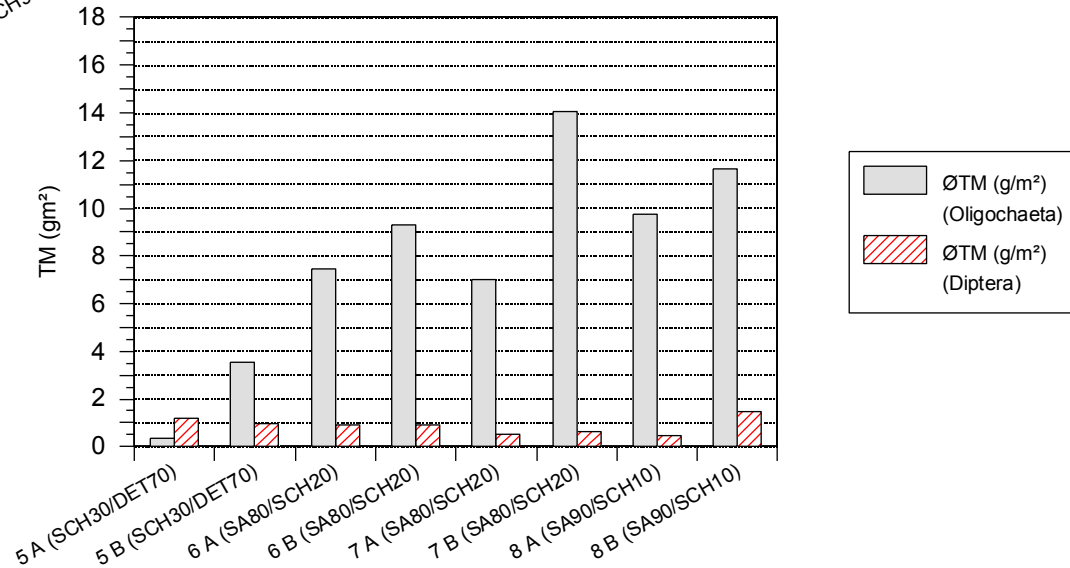
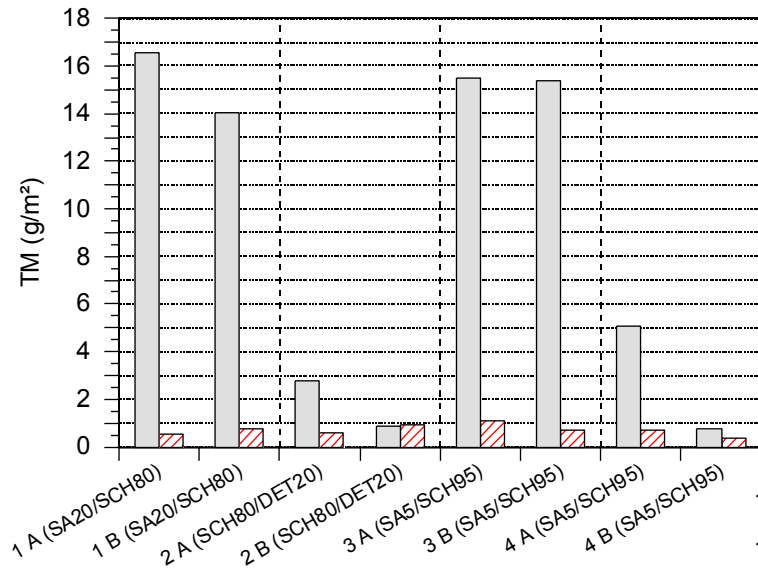
4. Methods of Investigation

Population densities



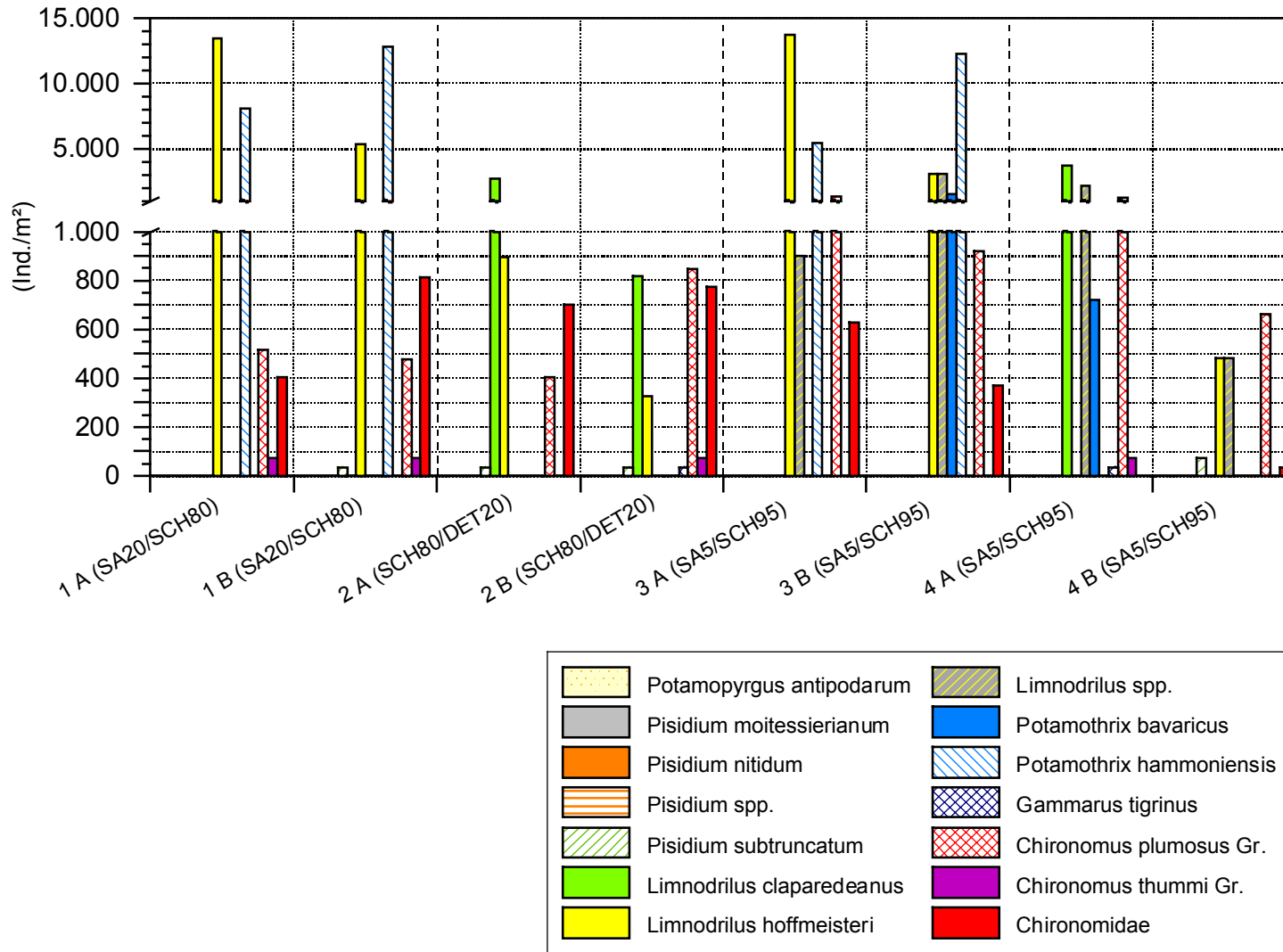
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Dry matter of Oligochaeta and Chironomidae in 8 sites at Lake Dümmer (Nov. 2004)



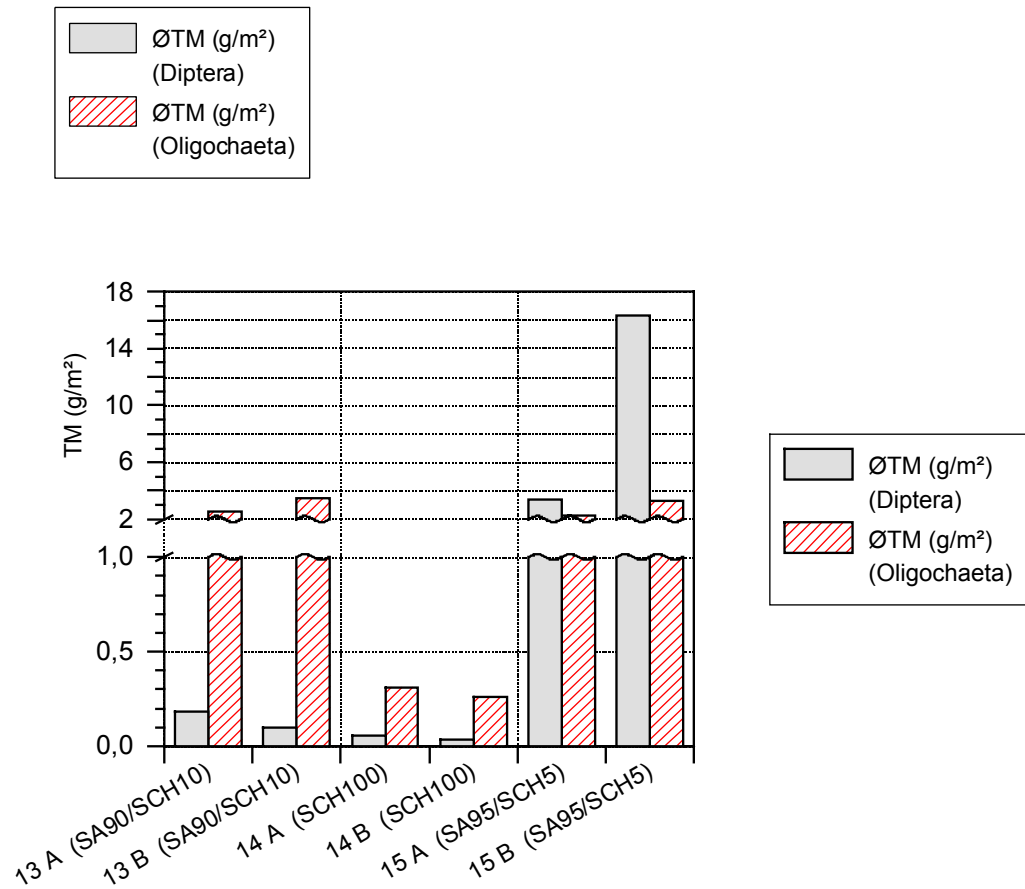
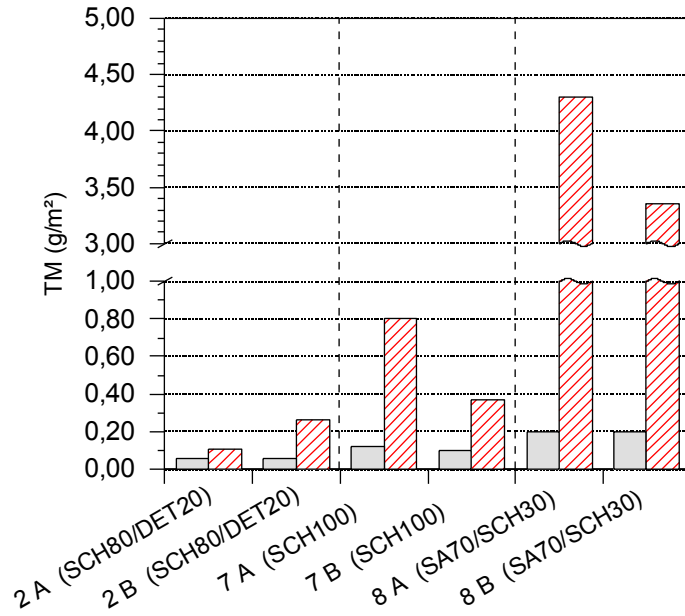
4. Methods of Investigation

Dominant Taxa in 4 sites at Lake Dümmer (Nov. 2004)



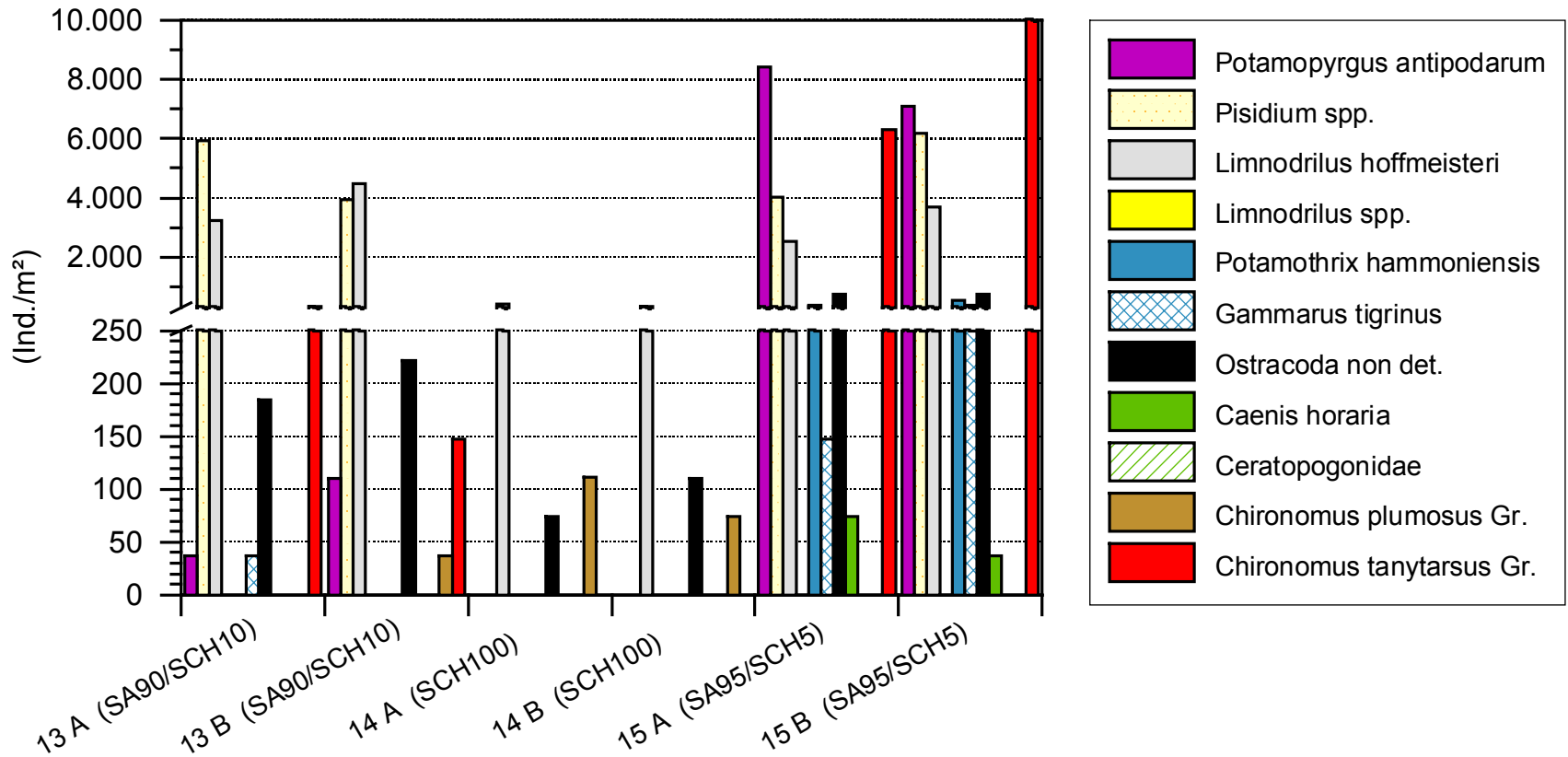
4. Methods of Investigation

Dry matter of Oligochaeta and Chironomidae in 6 sites at Steinhuder Meer (Nov. 2004)



4. Methods of Investigation

Dominant Taxa in 3 sites at Steinhuder Meer (Nov. 2004)



4. Methods of Investigation

Abundance of Oligochaeta in different lakes (Grigelis 1980; Bätthe 2004)

Waters	Population densities	Dry matter
Dümmer 2004	Mean: 4.600 Ind./m ² Maximum: 15.000 Ind./m ²	Mean: 2,9 g/m ² Maximum: 11,6 g/m ²
Steinhuder Meer 2004	Mean: 1.900 Ind./m ² Maximum: 10.000 Ind./m ²	Mean: 1,7 g/m ² Maximum: 7,75 g/m ²
Lake in Moldavia 1979	Minimum: 480 Ind./m ² Maximum: 5.640 Ind./m ²	Minimum: 0,68 g/m ² Maximum: 18,84 g/m ²
Tsimlyanskaya reservoir 1979	Sand: 3.629 Ind./m ² Mud: 19.000 Ind./m ² Other: 5.841 Ind./m ²	Sand: 5,41 g/m ² Mud: 19,20 g/m ² Other: 7,36 g/m ²
Lake Paravani 1979	Mean: 15.348 Ind./m ²	Mean: 10,10 g/m ²

4. Methods of Investigation - Oligochaeta: significance for water resources management

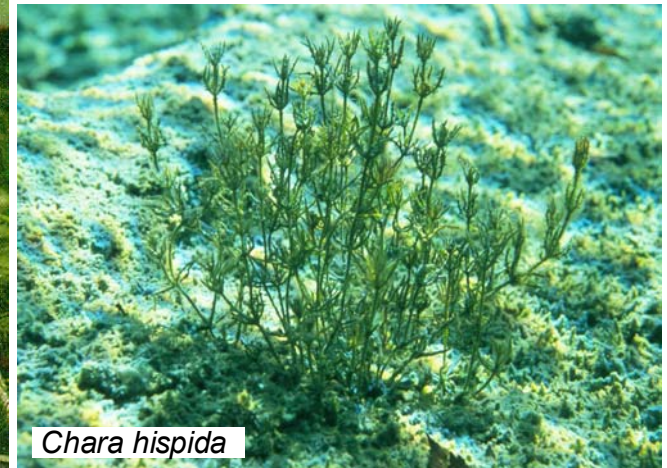
Oligochaeta in different lake-types (Milbrink, G. 1980) (each species = 10 points for detection)
 (total phosphor: **oligotrophic**: 0-20 µg/l; **mesotrophic**: 20-40 µg/l; **eutrophic**: >40 µg/l)

	Oligotrophy	Mesotrophy	Eutrophy
1. <i>Stylodrilus heringianus</i> Claparède	9	1	
2. <i>Rhynchelmis limosella</i> Hoffmeister	8	2	
3. <i>Bythonomus lemani</i> Grube	8	2	
4. <i>Peloscolex velutinus</i> (Grube)	8	2	
5. <i>Peloscolex ferox</i> (Eisen)	8	2	
6. <i>Lamprodrilus isoporus f. variabilis</i> Svetlov	(8)	(2)	
7. <i>Limnodrilus profundicola</i> (Verrill)	7	3	
8. <i>Psammoryctides barbatus</i> (Grube)	5	5	
9. <i>Rhuacodrillus falciformis</i> Bretscher	(5)	(5)	
10. <i>Rhyacodrillus coccineus</i> (Vejdovský)	4	6	
11. <i>Psammoryctides albicola</i> (Michaelson)	4	5	1
12. <i>Aulodrilus pigueti</i> Kowalewski	3	5	2
13. <i>Bothrioneurum vej dovskyanum</i> Stolc	2	5	3
14. <i>Aulodrilus limnobius</i> Bretscher	2	5	3
15. <i>Tubifex ignotus</i> (Stolc)	2	4	4
16. <i>Potamotheix moldaviensis</i> (Vejdovský and Mrázek)	2	4	4
17. <i>Limnodrilus udekemianus</i> Claparède	1	4	5
18. <i>Potamotheix bavaricus</i> (Oschmann)		5	5
19. <i>Potamotheix bedoti</i> (Piguet)		5	5
20. <i>Potamotheix vej dovskyi</i> (Hrabê)		5	5
21. <i>Ilyodrillus templetoni</i> (Southern)		5	5
22. <i>Potamotheix heuscheri</i> (Bretscher)		4	6
23. <i>Aulodrilus pluriseta</i> (Piguet)		4	6
24. <i>Limnodrilus cervix</i> Brinkhurst		4	6
25. <i>Limnodrilus claparedeanus</i> Ratzel	1	3	6
26. <i>Potamotheix hammoniensis</i> (Michaelson)	1	3	6
27. <i>Tubifex tubifex</i> (Müller)	5		5
28. <i>Limnodrilus hoffmeisteri</i> Claparède	1	2	7

4. Methods of Investigation

Ecological Methods = Bioindication with Macrophytes

- Long live cycles
- Slow reaction time concerning changes of the environment
- A large number of species have specific optima and tolerance for the phosphate concentration
- Assessment of trophic situation with macrophytes



4. Methods of Investigation

Ecological Methods = Bioindication with Diatoms

- Short live cycles
- Fast reaction time concerning changes of the environment = very sensitive
- A large number of species with specific optima and tolerance for the phosphate concentration or acidity of the aquatic medium
- Assessment of trophic situation with diatoms
- Assessment of acidity status with diatoms



4. Methods of Investigation

Ecological Methods = Bioindication

Combination of Bioindicators to assess the Trophic Situation

Macrophytes are long-term indicators

- **Investigation runs for one or more years to assess the trophic situation and its changes**
- **Macrophytes indicate long-term conditions of trophic level**

Diatoms are short-term indicators

- **Investigation runs for several months to assess the trophic situation and its changes**
- **Diatoms indicate short-term conditions of trophic level (3-4 weeks period)**

4. Methods of Investigation

Ecological Methods = Bioindication

Indicator-Group 1,0	Indicator-Group 1,5	Indicator-Group 2,0
Chara hispida Chara polyacantha Chara strigosa Potamogeton coloratus Utricularia stygia	Chara aspera Chara intermedia Utricularia minor	Chara delicatula Chara tomentosa Potamogeton alpinus
Indicator-Group 2,5	Indicator-Group 3,0	Indicator-Group 3,5
Chara contraria Chara fragilis Nitella opaca Nitellopsis obtusa Potamogeton gramineus Potamogeton natans Potamogeton x zizii	Chara vulgaris Myriophyllum spicatum Potamogeton filiformis Potamogeton perfoliatus Utricularia australis	Myriophyllum verticillatum Potamogeton berchtoldii Potamogeton lucens Potamogeton pusillus
Indicator-Group 4,0	Indicator-Group 4,5	Indicator-Group 5,0
Hippuris vulgaris Lagarosiphon major Potamogeton pectinatus	Elodea canadensis Elodea nutallii Potamogeton compressus Potamogeton crispus Potamogeton obtusifolius Ranunculus circinatus Ranunculus trichophyllus	Ceratophyllum demersum Lemna minor Potamogeton mucronatus Potamogeton nodosis Sagittaria sagittifolia Spirodela polyrhiza Zannichellia palustris

- **Ecological tolerance of species leads to indicator-group classification**
- **Macrophytes show correlation between phosphor concentration and species appearance**
- **Diatoms show correlation between phosphor concentration (different ranges) and species appearance**
- **Monitoring of macrophytes or diatoms allows calculation of indices [MI (macrophyte-index), DI (diatom-index), TIM (trophie-index-macrophytes) Melzer (2000), Trophieindex Coring (1999), Trophieindex Rott et al. (1998), etc.]**

4. Methods of Investigation

Ecological Methods = Bioindication

TP (total phosphorous) categories

TP [$\mu\text{g/l}$]	Degree of Pollution	Trophy	Colour
$x < 10$	very low	oligotroph	dark-blue
$10 \leq x < 15$	low	oligo - mesotroph	light-blue
$15 \leq x < 20$	moderate	mesotroph 1	dark-green
$20 \leq x < 30$	moderate - significant	mesotroph 2	light-green
$30 \leq x < 40$	significant	eutroph 1	yellow
$40 \leq x < 55$	heavily	eutroph 2	orange
$x \geq 55$	very heavily	eutroph 3	red

4. Methods of Investigation

Ecological Methods = Bioindication

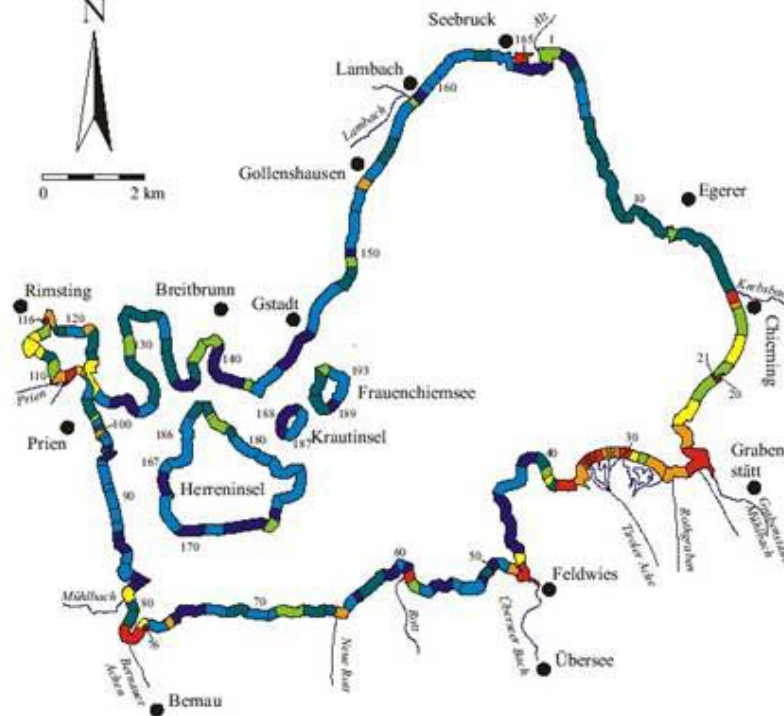
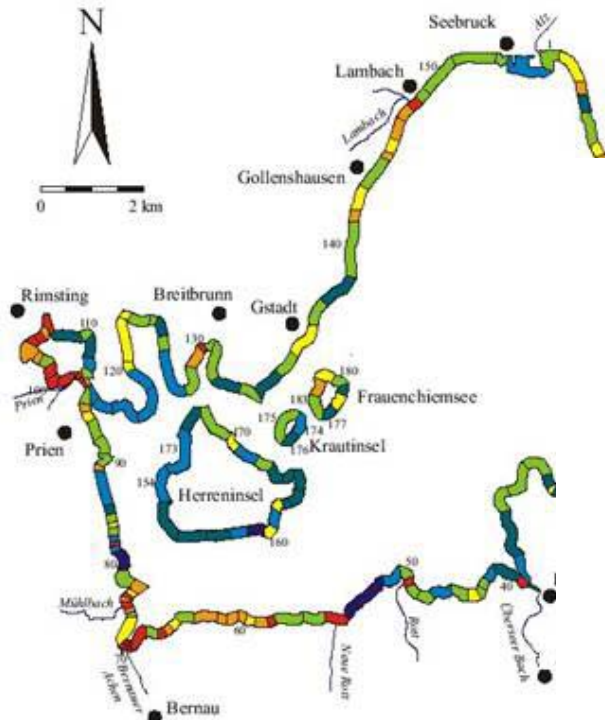
Index categories (harmonising Diatom- and Macrophyte-Indices)

TP [$\mu\text{g/l}$]	Diatom-Index	Macrophyte-Index	Colour
$x < 10$	$1,00 \leq x < 1,90$	$1,00 \leq x < 2,40$	dark-blue
$10 \leq x < 15$	$1,90 \leq x < 2,45$	$2,40 \leq x < 2,70$	light-blue
$15 \leq x < 20$	$2,45 \leq x < 2,95$	$2,70 \leq x < 2,95$	dark-green
$20 \leq x < 30$	$2,95 \leq x < 3,70$	$2,95 \leq x < 3,30$	light-green
$30 \leq x < 40$	$3,70 \leq x < 4,20$	$3,30 \leq x < 3,55$	yellow
$40 \leq x < 55$	$4,20 \leq x < 4,65$	$3,55 \leq x < 3,90$	orange
$x \geq 55$	$4,65 \leq x < 5,00$	$3,90 \leq x < 5,00$	red

4. Methods of Investigation

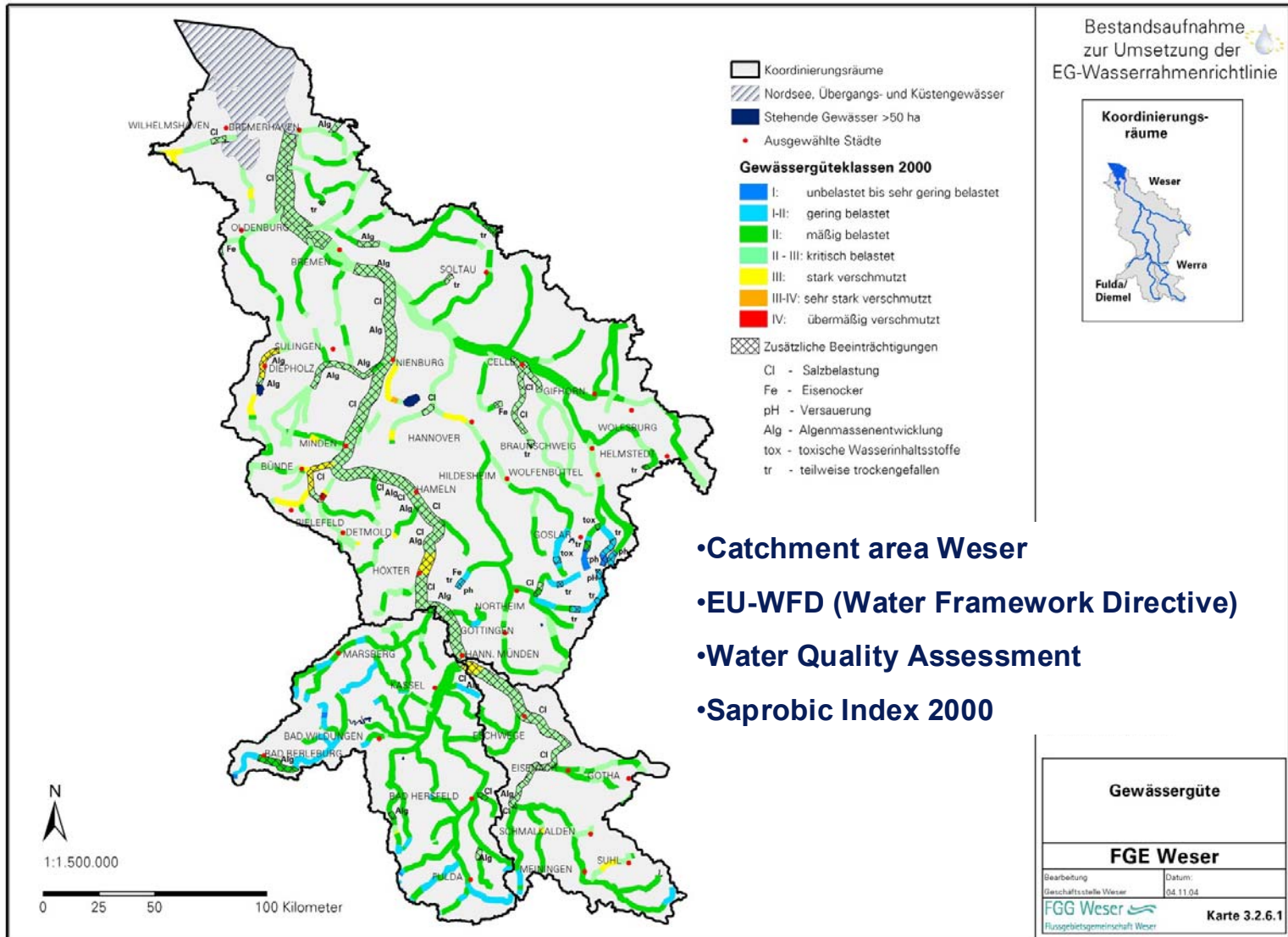
Ecological Methods = Bioindication

Macrophyte-Index Lake Chiemsee 1985 - 1998



Chiemsee 1998
 Karte 2
 Nährstoffbelastung nach
 Makrophytenindex

4. Methods of Investigation



- Catchment area Weser
- EU-WFD (Water Framework Directive)
- Water Quality Assessment
- Saprobic Index 2000

4. Methods of Investigation

Guidelines for Quality Assurance in Sampling Methods

Examples:

- **EN 25667-2 (1993): Water quality - Sampling - Part 2: Guidance on sampling techniques (ISO 5667-2: 1991)**
- **JAMP guidelines on Quality Assurance for biological monitoring in the OSPAR area (ASMO 2002)**
- **Manual for Marine Monitoring in the COMBINE Programme of HELCOM, PART B. General Guidelines on quality assurance for monitoring in the Baltic Sea: http://www.helcom.fi/groups/monas/CombineManual/en_GB/main/**

4. Methods of Investigation

Examples:

Guidelines "Sampling and Abiotic Parameter"

- DIN 38 402 Teil 12: Probenahme aus stehenden Gewässern (A 12)
- DIN 38 402 Teil 15: Probenahme aus Fließgewässern (A 15)
- AQS – Merkblatt zu den Rahmenempfehlungen der Länderarbeitsgemeinschaft Wasser für die Qualitätssicherung bei Wasser-, Abwasser- und Schlammuntersuchungen (LAWA AQS-Merkblatt P-8/3; Lfg. V/99)
- DIN 38 404 Teil 4: Physikalische und physikalisch-chemische Kenngrößen (Gruppe C) - Bestimmung der Temperatur (C4)
- DIN 38 404 Teil 5: Physikalische und physikalisch-chemische Kenngrößen (Gruppe C) - Bestimmung des pH-Wertes (C5)
- DIN EN 25 814: Bestimmung des gelösten Sauerstoffs – Elektrochemisches Verfahren (ISO 5814 : 1990)
- DIN 38 408 Teil 23: Gasförmige Bestandteile (Gruppe G) Bestimmung des Sauerstoffsättigungsindex (G 23)
- DIN EN 27 888: Bestimmung der elektrischen Leitfähigkeit (ISO 7888 : 1993)

4. Methods of Investigation

Examples:

Guidelines „Limnological Investigations“

- ATT – Arbeitsgemeinschaft Trinkwassertalsperren e.V. Arbeitskreis Biologie (1998): Erfassung und Bewertung von Planktonorganismen. ATT Technische Informationen Nr. 7, 151 S., Siegburg
- DIN EN 13946 (2003-09): Wasserbeschaffenheit - Leitfaden zur Probenahme und Probenaufbereitung von benthischen Kieselalgen in Fließgewässern; Deutsche Fassung EN 13946: 2003
- DIN EN 14407 (2004-10): Wasserbeschaffenheit - Anleitung zur Bestimmung, Zählung und Interpretation von benthischen Kieselalgen in Fließgewässern; Deutsche Fassung EN 14407: 2004
- DIN EN 28265 (1994-03): Wasserbeschaffenheit; Probenahmegeräte für die quantitative Erfassung benthischer Makro-Invertebraten auf steinigem Substraten in flachem Süßwasser (ISO 8265: 1988); Deutsche Fassung EN 28265: 1994

In Preparation: ISO TC 147/SC 5 N (2009) Water quality - Guidance on the selection of sampling methods and devices for benthic macroinvertebrates in fresh waters.

In Preparation: DIN EN 14996 (2006-08) Wasserbeschaffenheit – Anleitung zur Qualitätssicherung biologischer und ökologischer Untersuchungsverfahren in der aquatischen Umwelt.

4. Methods of Investigation

Phytoplankton/Zooplankton

Relevant in stagnant waters (lakes, impounded river sections)

Guidelines:

- Methods for quantitative assessment of phytoplankton in freshwaters. Part I: Orlik, Blomqvist, Brettum, Cronberg & Eloranta. Rapport No. 4860, ISBN: 91-620-4860-0. Part II: Blomqvist & Herlitz. Rapport No. 4861. ISBN: 91-620-4861-9
- JAMP eutrophication monitoring guidelines – chlorophyll a (ASMO 1997)
- JAMP eutrophication monitoring guidelines – phytoplankton species composition (ASMO 1997)
- Manual for Marine Monitoring in the COMBINE Programme of HELCOM, PART C. Programme for monitoring of eutrophication and its effects, Annex C-4 Phytoplankton chlorophyll-a: http://www.helcom.fi/groups/monas/CombineManual/AnnexesC/en_GB/annex4/
- Manual for Marine Monitoring in the COMBINE Programme of HELCOM, PART C. Programme for monitoring of eutrophication and its effects, Annex C-5 Phytoplankton primary production: http://www.helcom.fi/groups/monas/CombineManual/AnnexesC/en_GB/annex5/
- Manual for Marine Monitoring in the COMBINE Programme of HELCOM, PART C. Programme for monitoring of eutrophication and its effects, Annex C-6 Phytoplankton: species composition, abundance and biomass: <http://www.helcom.fi/stc/files/CombineManual/PartC/AnnexC6.pdf>

4. Methods of Investigation

Phytoplankton/Zooplankton

Relevant in stagnant waters (lakes, impounded river sections)

Equipement:

- Plankton net (mesh size depends on chosen method for particular question)
- Visibility-Disc (Secchi-Disc)
- Ruttner sampler (Standard water sampler)
- IWS (Integrated Water Sampler)
- Boat with sonar, cable winch, GPS
- PU-bottles, glass-bottles
- sample protocol
- Fixation solution (e.g.: Ethanol, Logul's solution, Formalin)

4. Methods of Investigation

Phytoplankton/Zooplankton



4. Methods of Investigation

Phytoplankton/Zooplankton



Visibility-Disc (Secchi-Disc)



Ruttner-Sampler; Secchi-Disc; Oxymeter (cable length 30 m)

4. Methods of Investigation

Phytobenthos

Relevant in stagnant and running waters

Guidelines:

- EN 13946 (2003): Water quality - Guidance standard for the routine sampling and pretreatment of benthic diatoms from rivers.- TC 230 WG 2 TG 3, "Macrophytes and algae".
- EN 14407 (2004): Water quality - Guidance standard for the identification, enumeration and interpretation of benthic diatom samples from running waters. - TC 230 WG 2 TG 3, "Macrophytes and algae".

Equipement:

- **Denture brush, scalpel (lancet)**
- **shallow bowl**
- **IWS (Integrated Water Sampler) for additional phytoplankton investigation**
- **PU-bottles**
- **sample protocol, GPS**
- **Fixation solution (e.g.: Ethanol)**

4. Methods of Investigation

Macrobenthos

Relevant in running and stagnant waters

Guidelines:

- CEN/TC 230/WG 2/TG 1 N101a (2005): Water quality – Guidance on the selection of sampling methods and devices for benthic macroinvertebrates in freshwaters. - 28 pp.
- JAMP eutrophication monitoring guidelines – benthos (ASMO 1997)
- Manual for Marine Monitoring in the COMBINE Programme of HELCOM, PART C. Programme for monitoring of eutrophication and its effects, Annex C-8 Soft bottom macrozoobenthos:
http://www.helcom.fi/groups/monas/CombineManual/AnnexesC/en_GB/annex8/

Equipement:

- Handnet (acc. to Böttger or "aqem", kicksampler, surber-sampler)[mesh size: 500 µm]
- Plastic tray (rectangular, 1/8 m², white)
- Sieves (different mesh sizes)
- Gumboots, Waders
- PU-bottles (different sizes, 1 Litre, 100 ml, 50 ml)
- sample protocol, GPS, tweezer (pincette), magnifier, pencil
- Fixation solution (e.g.: Ethanol)

4. Methods of Investigation

Macrobenthos

Sampling site and Equipement



4. Methods of Investigation

Macrobenthos

Substrates/Sampling sites



Megalithal > 40 cm



Macrolithal >20 - 40 cm



Mesolithal >6 - 20cm

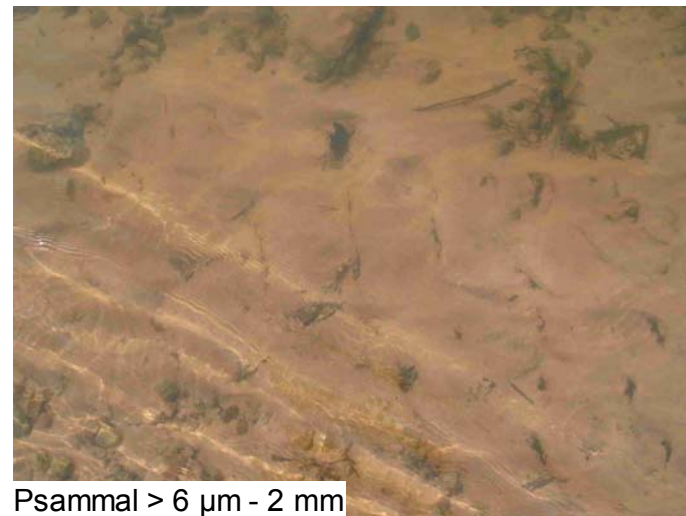
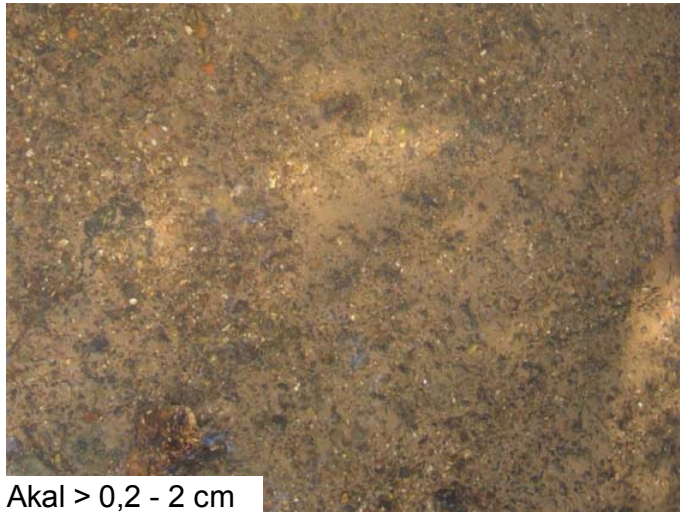


Microlithal >2 - 6cm

4. Methods of Investigation

Macrobenthos

Substrates/Sampling sites



4. Methods of Investigation

Macrobenthos

Substrates/Sampling sites



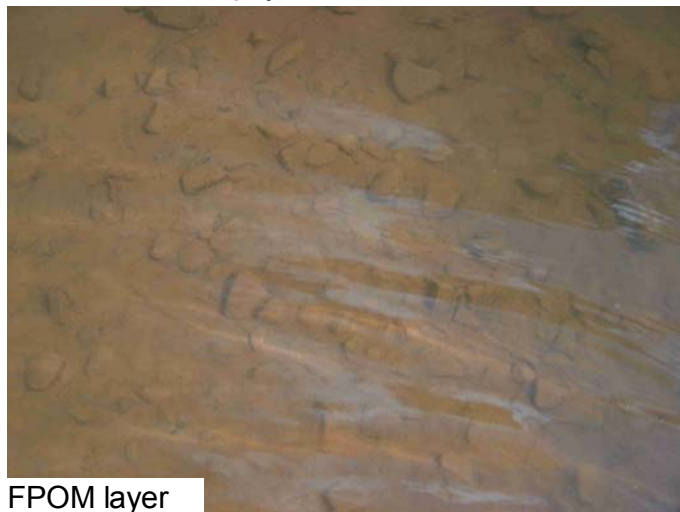
technolithal



submersed macrophytes



emergent macrophytes

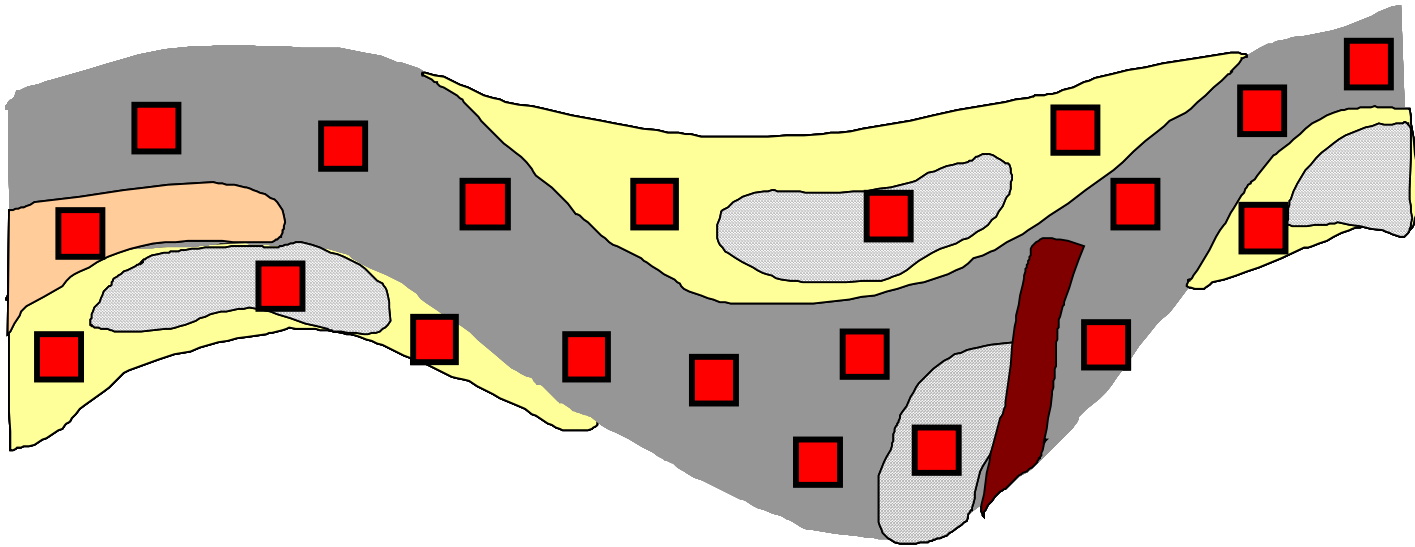








FPOM layer

4. Methods of Investigation

Macrobenthos

Substrates/Sampling sites



	Mesolithal (55% = 11 subsamples)		CPOM (15% = 3 subsamples)
	Akai (5% = 1 subsamples)		Xylal (<5% = 0 subsamples)
	Psammal (25% = 5 subsamples)		subsamples

4. Methods of Investigation

Macrobenthos

Samples

- **quantitative**: based on defined area and all available substrates (e.g. 1 m²)
- **qualitative**: based on defined time and all available substrates (e.g. 20 min.)
- **abundance**: individuals counted (per area) (ind./m²) or
- **individuals estimated in abundance classes (1 – 7)**

sampling protocol

4. Methods of Investigation

Chemicophysical Parameters

Parameters:

oxygen, temperature, pH-value, electrical conductivity

Measuring devices:

oximeter, pH-meter, conductivity meter, thermometer

Preparations:

- check power supply
- clean device and sensors
- calibrate sensors

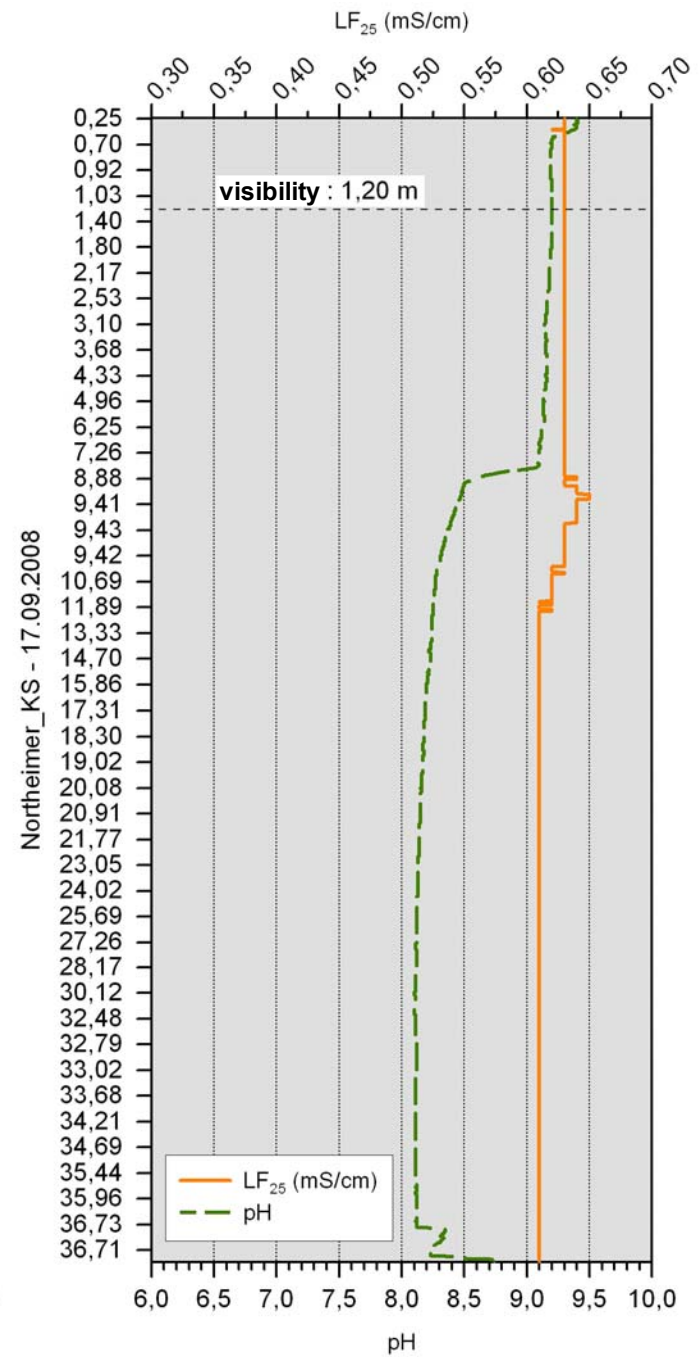
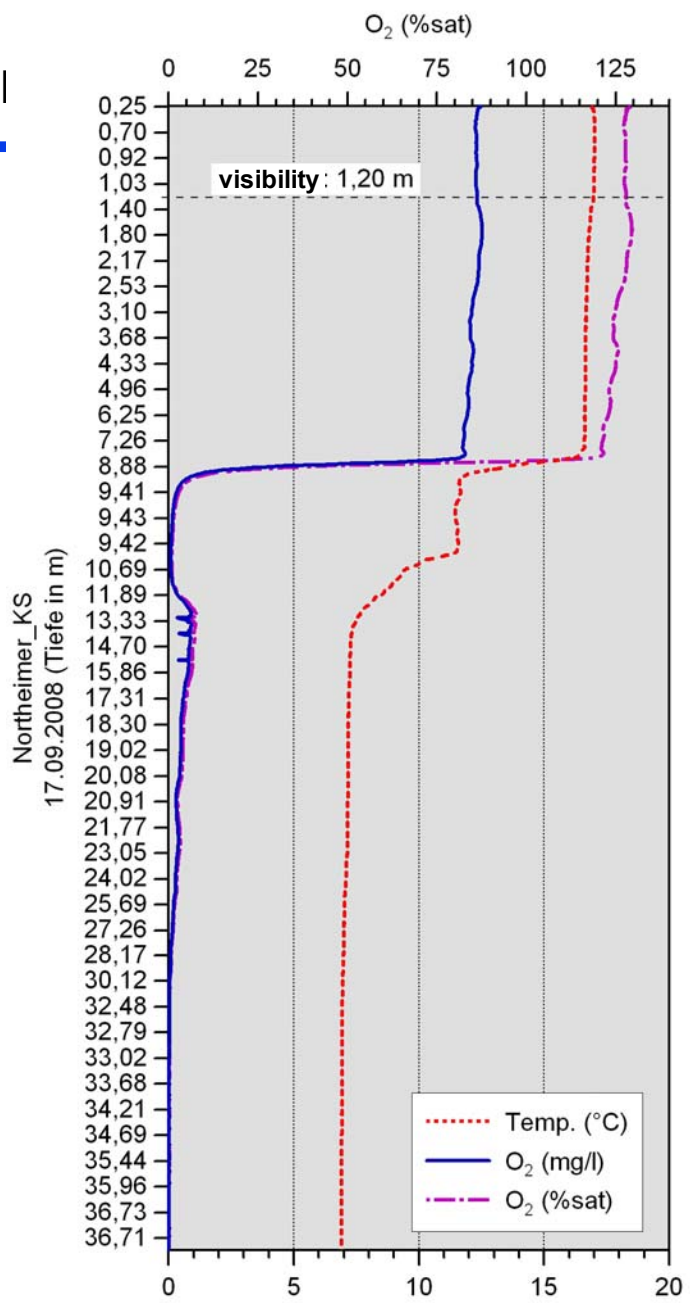
At sampling site:

- calibrate oxygen sensor (air pressure adjustment)
- measurement always in current water (hand- or automatic stirrer)

4. Methods of |

depth profile:

- Temperature
- Oxygen-concentration
- O₂-Saturation
- Conductivity
- pH-Value

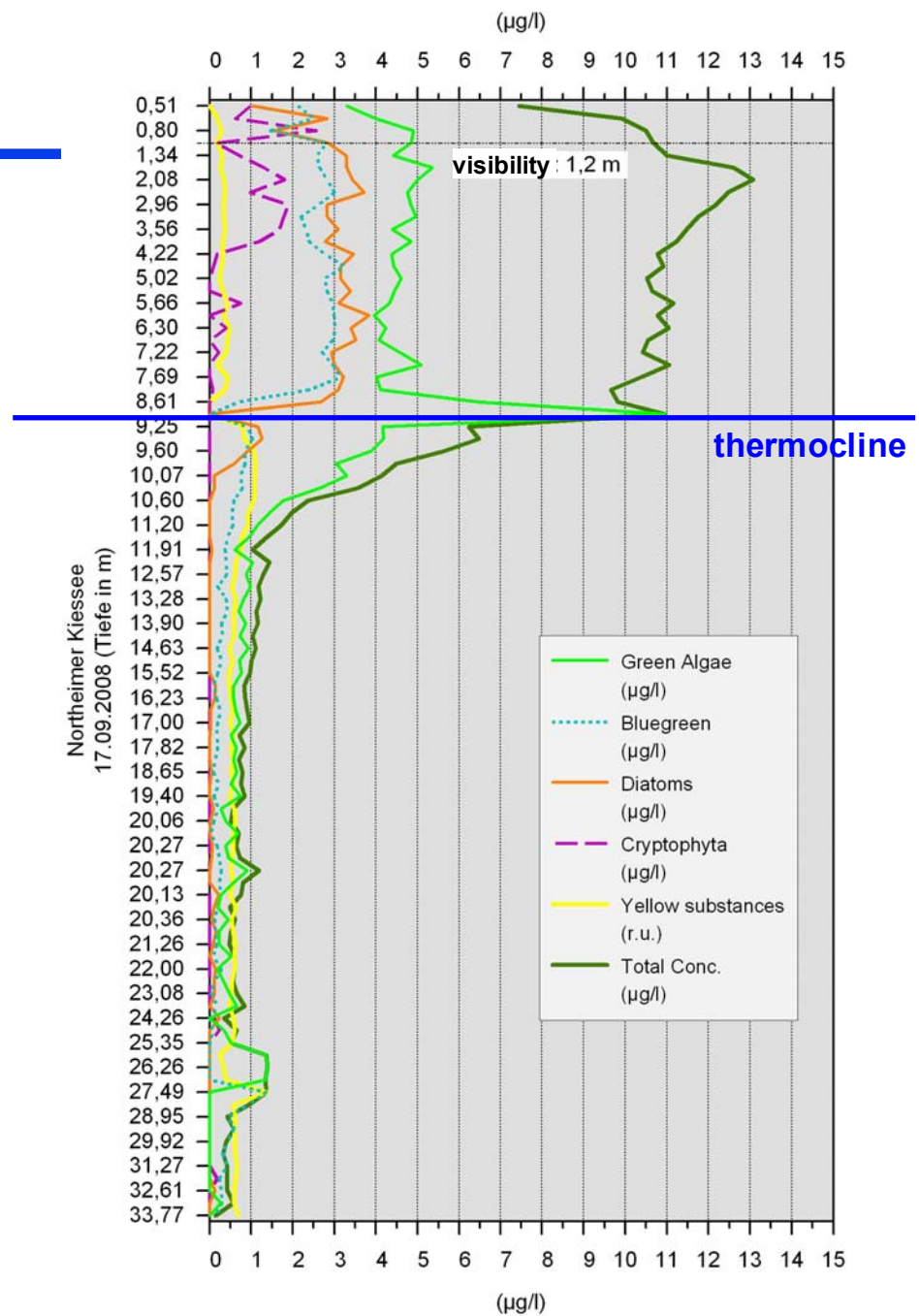


4. Methods of Investigation

Phytoplankton:

(algal classes)

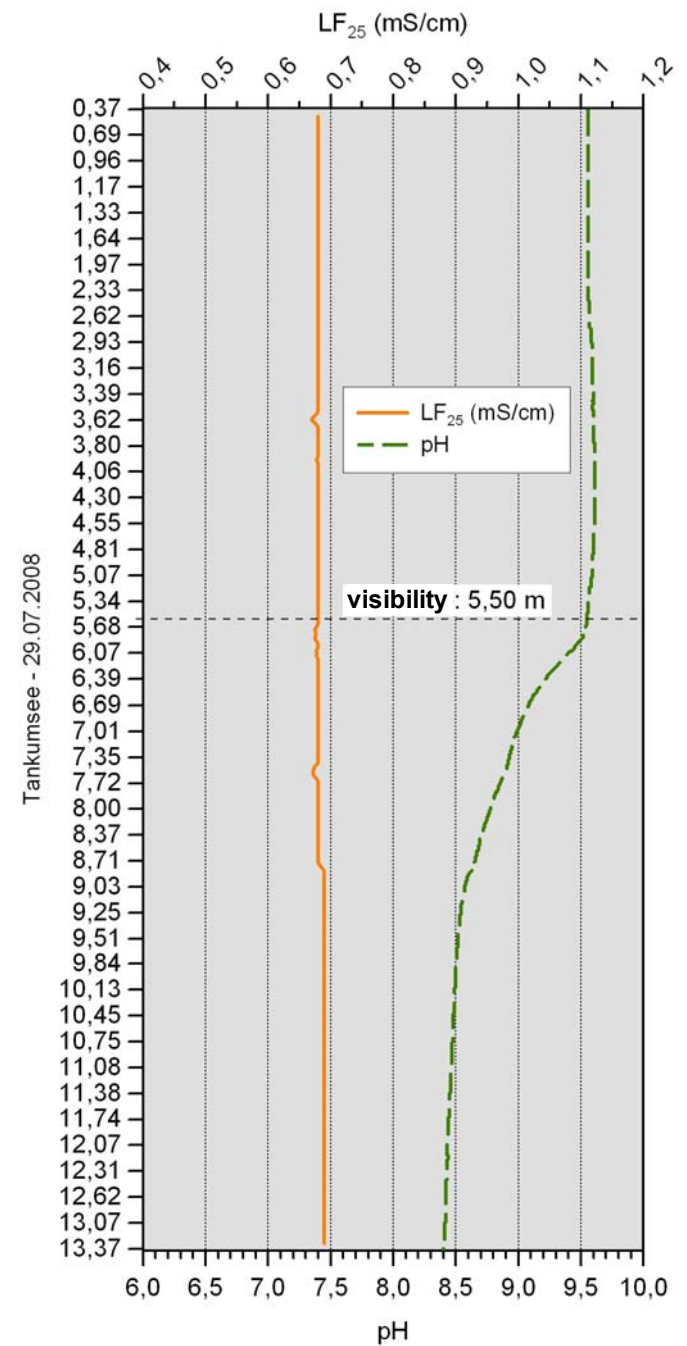
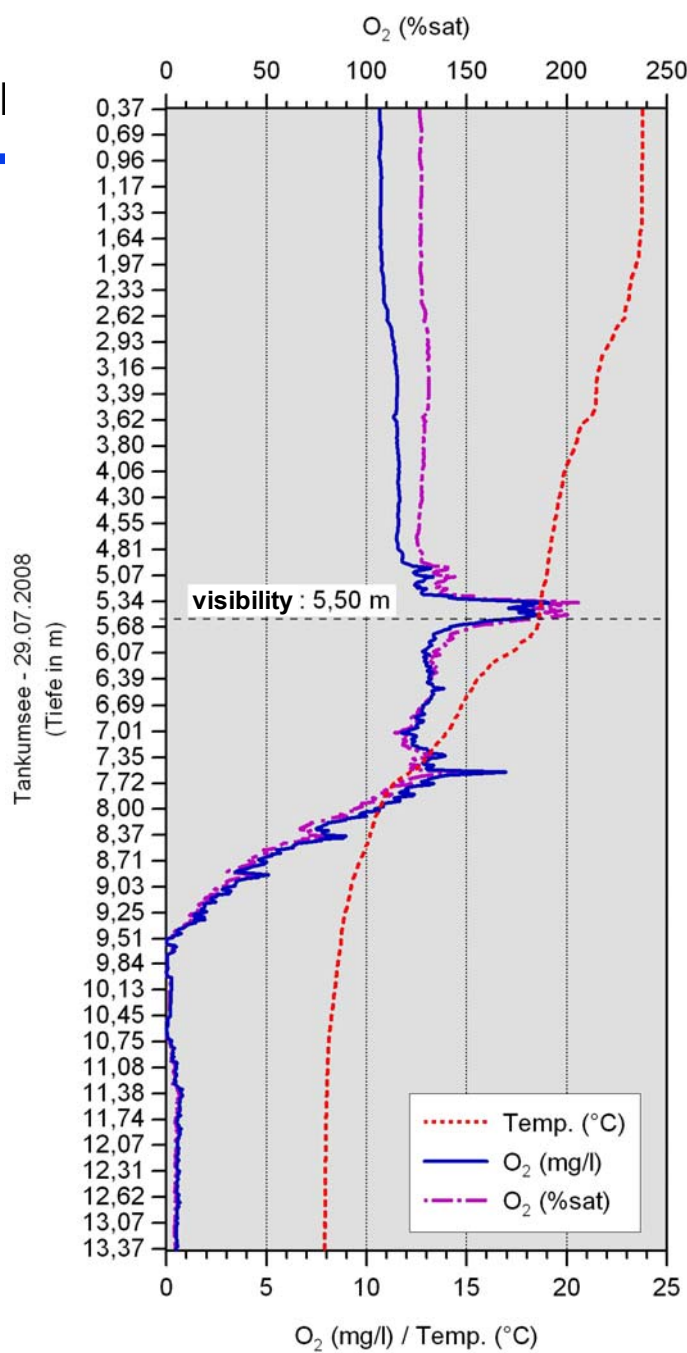
- Total Conc.
- Green Algae
- Bluegreen Algae
- Diatoms
- Cryptophyta
- Yellow substances



4. Methods of |

depth profile:

- Temperature
- Oxygen-concentration
- O₂-Saturation
- Conductivity
- pH-Value



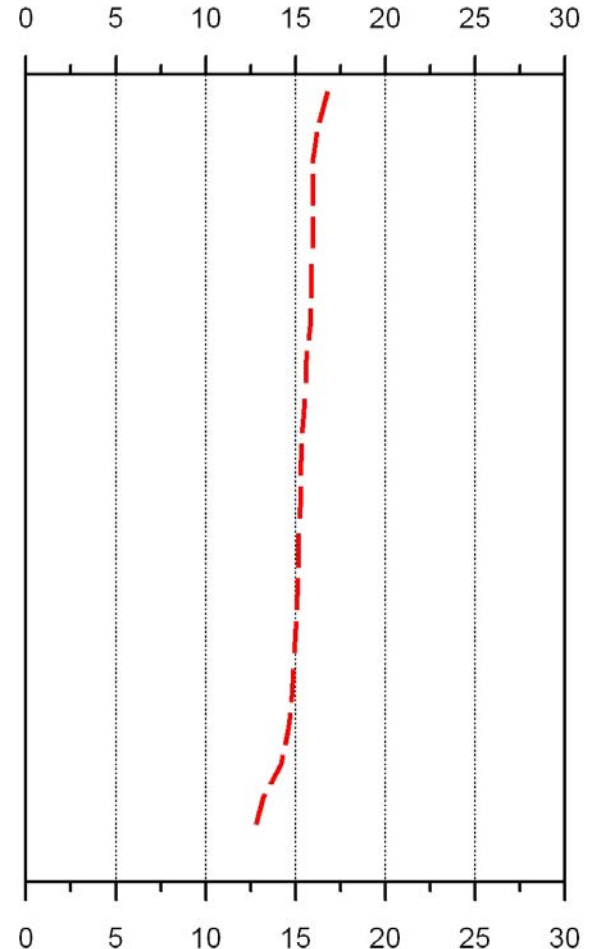
4. Methods of Investigation

depth profile

orthograde oxygen chart (Odertalsperre; oligo- mesotroph)

Odertal-Barrage 17.09.03
(O₂ %sat.)

Odertal-Barrage 17.09.03
(°C)



4. Methods of Investigation

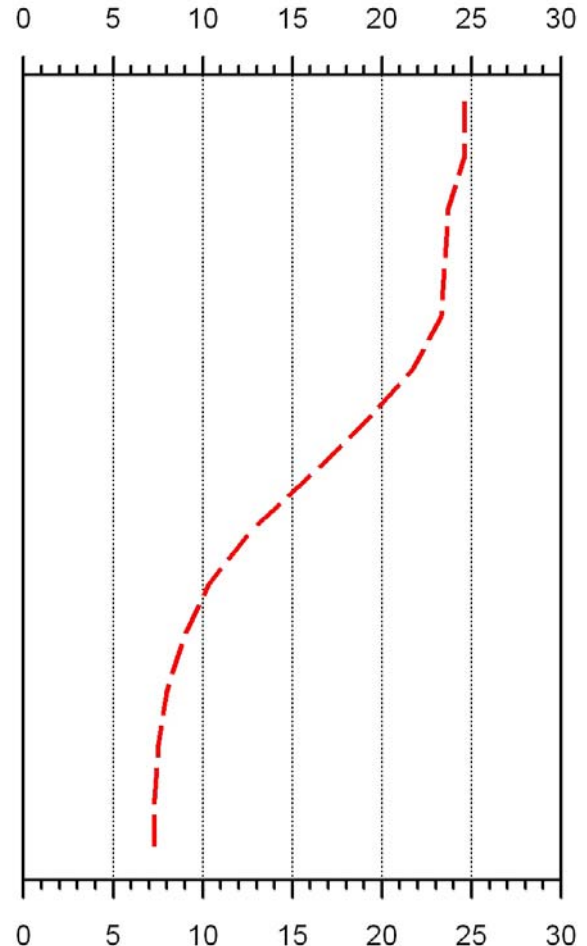
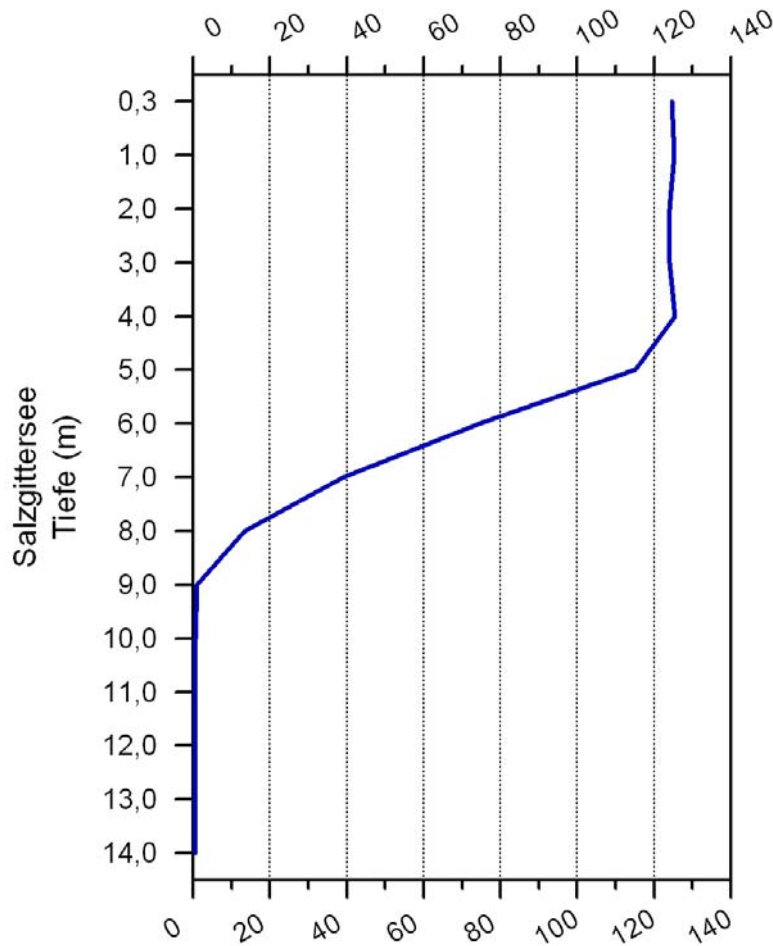
depth profile

klinograde oxygen chart

(Salzgittersee; eutroph)

Salzgittersee
31.07.03 (O₂ %sat.)

Salzgittersee
31.07.03 (°C)



4. Methods of

depth profile
Bodensee
(28.03.2008)

CTD 145
(Sea&Sun
Technologies)

