



SIXTH FRAMEWORK PROGRAMME
SPECIFIC TARGETED RESEARCH OR INNOVATION PROJECT

REBECCA
Relationships between ecological and chemical status
of surface waters

Contract No.: SSPI-CT-2003-502158

REBECCA WP4 Rivers

**Deliverable D6:
Report on existing methods and relationships linking
pressures, chemistry and biology in rivers**

December 2004

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Report on existing methods and relationships linking pressures, chemistry and biology in rivers.

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0 Introduction

The objective of this report is to present the relevant existing knowledge on relationships linking chemical and hydromorphological conditions to biological quality elements in European rivers and to present the methods and indicators used to describe the biological quality elements.

The aims of the report are twofold:

- 1) To give end-users an overview of existing methods and relations
Potential end-users are all institutions and persons involved in the implementation of the Water Framework Directive in relation to rivers.
- 2) To establish a common reference among the REBECCA WP4 partners in their further development of relations and methods
 - to identify the chemical and hydromorphological parameters determining ecological status,
 - to quantify the relationships between pressures, chemical quality elements, and biological quality elements for pressures in different river types in relation to geographical region,
 - to identify key biological indicators for describing the ecological impacts, and develop integrated indicators for estimation of the overall ecological status
 - to estimate necessary pressure reductions to achieve ecological objectives, and
 - to identify knowledge gaps.

The report is structured according to the working approach of REBECCA WP4 on rivers with the work divided into 5 activities. Activities 1, 2, 3 and 4 cover the major pressures impacting rivers: hydromorphological pressures, acidification and toxicity, pollution with organic matter and nutrients causing eutrophication. Activity 5 covers multiple pressures in a geographical context.

The overall organisation of the REBECCA activities has been slightly amended by establishing a new cross cutting work package, WP-toxicity, that is responsible for activities (i.e. on lakes, rivers and marine environments) on toxic compounds and their impacts on the aquatic ecosystems. Therefore, toxic compounds are not covered in this report, unless related to the types of pressures covered.

A general approach of the report is to focus mainly on the primary impacts of the different pressures. The reason for this is that the possibility to establish causal relationships between a pressure and the impact on river biota is considered to be higher for the directly impacted biota. As an example, the relationships between nutrient enrichment and autotrophic organisms are more likely to be operational compared to relations between nutrients and heterotrophic organisms.

The relations, methods and indicators described are found through literature studies, but no new relations or models are developed at this stage. The existing literature and data retrieved specifically for the REBECCA WP4 work is used for the further development of relations and methods within the REBECCA project.

1 Hydromorphological pressures

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1.1 Introduction

With the word ‘hydromorphological pressures’ we understand all changes caused by human influences to either the flow regime (hydro) or the morphology of the stream that affect the stream biota. The most important hydromorphological pressures are:

- building of dams or weirs for hydropower, water supply or other purposes
- channelisation and/or dredging of rivers or streams to improve drainage or for navigation;
- weed cutting to improve drainage
- abstraction of water directly from the stream or from groundwater for water supply or irrigation, or diversion, e.g. for hydropower or irrigation

Other influences that are not described in detail here include urbanisation, afforestation/deforestation, draining of wetlands (tiling), transport and supply of water from outside the river basin to increase river discharge at dry periods, and high discharges of water treatment plants in small river basins.

Literature review

A literature review was carried out using the following procedure:

- 1) ISI Web of Science and Aquatic Sciences and Fisheries Abstracts (ASFA) were used to search for all journal papers which included three words: river OR stream OR watercourse, a hydromorphological search word (see Annex 1, Table 1), and a biological search word (see Annex 1, Table 1). A total of approximately 17,000 references were extracted.
- 2) References from journals considered less likely to hold key publications (e.g. journals covering tropical rivers) were removed. This reduced the number of papers to 11,705. The titles, abstracts and journal details of these papers are kept in an Endnote file named ‘Hydromorph_1946_2004.enl’.
- 3) From this database, we extracted the most important papers on the effects of dams, channelisation, abstractions, weed cutting, etc. on the stream biota (Annex 1, Table 2). The titles, abstracts and journal details are kept in an Endnote file named ‘Hydrolmorphology.enl’, holding 280 references.

1.2 Hydromorphological pressures

1.2.1 Dams and weirs

The term ‘impoundment’ is often used to describe any structure that alters river water levels. Larger height structures (often termed dams) are built to provide a hydraulic head to generate electricity (hydropower) and to store water for irrigation and water supply. Smaller height structures or weirs are built to maintain water levels in low flow periods for agriculture, shipping and recreation and to divert water for supply, hydropower or irrigation. Depending on the height of

the impoundment, the upstream physical environment will change, increasing depths and retention times, and decreasing velocities, this could well have knock-on effects on the ecosystem. Impoundments can also destroy the connectivity of the river, alter the flow regime and lower the sediment transport downstream. Flushing flows may be used to clean out fine sediment from the reservoir and downstream river-bed and also to re-structure the bed generally, as river sections downstream dams usually have lower and less frequent floods than the natural flow regime (Growth & Growth, 2001), although more extreme floods are often unaffected. The less variable flow regime and lower sediment transport often leads to higher water temperatures and clarity. Large dams generally lower the temperature downstream, when cooler water is drawn from lower down in the impounded reservoir. Another typical feature of rivers downstream of hydropower stations are sub-daily, daily and weekly flow fluctuations depending on hydropower pricing. The physical changes also lead to changes in water chemistry.

Plants and animals are affected by the physical and chemical changes. The higher clarity and reduced variability of flow usually lead to higher abundance of periphyton and higher plants (Dessaix & Fruget, 1995; Biggs et al., 1998). In nutrient-rich rivers the occurrence of phytoplankton is usually increased downstream the dam as a result of the phytoplankton production in the reservoir. Benthos communities below dams often show a reduction in species richness, while some species increase in abundance (Fruget, 1991). For example, invertebrate filter feeders (e.g., *Hydrophyche*, *Simuliidae*) often increase in numbers downstream of reservoirs. Migratory fish (e.g., salmon, trout and eel) are especially affected by the breaking of the connectivity of the river. The spreading of other animal groups and plants can also be hampered because of the reservoir (Allan, 1995).

The effects of dams on the stream biota are generally well documented. Approximately 1,500 out of the 11,700 papers in 'Hydromorph_1946_2004.enl' were concerned with this topic (see Annex 1, Table 2).

1.2.2 Channelisation

Rivers and streams are channelised for purposes such as navigation, flood protection, drainage of agricultural land, urbanisation, and, for example in Finland, some rivers have been straightened and narrowed to facilitate transport of logs downstream. The straightening of meandering streams or rivers is often combined with a recurrent dredging of deposited material to maintain the desired river profile. Channelisation changes a naturally meandering stream with hydromorphological variety into a uniform channel with homogeneous bed substrate and relatively uniform water velocity across the stream. The channel is usually constructed to be wider than the natural profile to allow for a larger conveyance, and the water depth and velocity will decrease. Often the flow variability and the light conditions at the streambed will be affected as well.

These physical changes have been associated with changes in the stream flora and fauna (Giller & Malmqvist, 1999; Allan, 1995; Dessaix & Fruget, 1995; Hurtle & Lake, 1983). The reduced variety of habitat will likely result in lower species richness compared with undisturbed streams. Many stream invertebrates are adapted to a life on coarse substrates (stones and gravel) with high current velocities that, in addition to creating numerous microhabitats, ensures an environment rich in oxygen. In addition, channelisation is likely to reduce the number of instream velocity refugia, thereby decreasing the resistance of the biological community to flood events (e.g. for invertebrates: Winterbottom *et al.*, 1997). The availability of fish spawning habitats is dramatically reduced in navigated river channels. In those cases lateral or secondary channels may act as a refugium (Grift, 2001)

1.2.3 Weed cutting

Weed cutting is undertaken in many small and medium sized streams to increase the discharge capacity and prevent the surrounding areas from inundation. River vegetation is rarely found deeper than 1.5-2 metres and weed cutting is thus a phenomenon related to smaller streams. When weeds are cut, the water level drops and the water velocity increases, which again may increase the sediment transport and temperature. Bank stability may also reduce. However, the physical changes and the impacts on the river ecosystem are strongly dependent on the weed cutting method.

The immediate effect of weed cutting is a direct loss of plants, which serve as habitat for invertebrates and refuge for fish. The lower water level also reduces the available space and the habitat diversity. Long-term use of weed cutting changes the composition and structural complexity of the macrophyte community, which becomes poorer in species and spatially more homogeneous. Also substantial changes in composition patterns can develop with an enhanced abundance of fast-growing species with a high dispersal capacity (Baattrup-Pedersen et al., 2003). A reduced diversity and structural complexity of macrophyte communities can affect invertebrate and fish communities negatively. This probably relates to a less spatial and temporal physical heterogeneity i.e. less varied substrate composition and more narrow range of flow velocities with decreasing structural diversity of the macrophyte community (Garner & Bass, 1996). Therefore loss of macrophyte species and homogenisation of communities as a result of weed cutting may have cascading effects on the whole stream biota.

1.2.4 Abstraction and diversion

Water is abstracted from streams and rivers for water supply or irrigation, rivers are diverted for various reasons, and sometimes streams are augmented by water from other catchments to increase the streamflow for agricultural purposes.

In case of abstractions, water is taken either directly from the stream, in which case the flow is reduced immediately, or it is taken from groundwater. A groundwater abstraction will affect the streamflow dependent on the distance of the borehole from the stream and the hydraulic properties of the sediments between the bore and the stream. Often the highest demand occurs in summer, when the flow is naturally low. Thus, the most important effect of abstractions is on the low flow regime, while diversions are more likely to affect the stream at all times. Reducing the flow will lead to lower depth and velocity and an increase in temperature (especially in summer), and the dilution of pollutants will reduce. In some cases when water is diverted or abstracted, the stream may even dry up completely.

A reduction in flow caused by abstraction, or diversion, affects the biota (e.g., Collier, 2002). Usually the growth of filamentous algae is favoured (mainly due to lower velocities) and the invertebrate community will change from one that grazes on thin periphyton films to one that lives amongst thick periphyton mats. Amphibious plants may invade more central parts of the stream from which they are normally kept away by high velocity and depth, and the distribution of plant species will change. Reduced water level and velocity leads to a loss of river habitat, which may affect biota on all levels. Fragmentation of the system will affect especially migratory fish, but also general animal movement, of which little is generally known. A complete drying up, even for a short period, is usually detrimental to the river system unless the biota has developed in such a system. In general, reduced flow will affect oxygen concentrations negatively and this is especially critical for a large proportion of the invertebrate community as most stream invertebrates are unable to regulate their oxygen uptake (e.g., Golubkov & Tiunova, 1989).

1.2.5 Other changes (draining, urbanisation, forestation/deforestation)

Draining, urbanisation and afforestation/deforestation change both the flow and the sediment regime. Both draining (tiling) and urbanisation increase the quick runoff and transport particles to the stream that would not otherwise have been mobilised. Discharge of water treatment plants may have a very large impact on river flows, especially in small river basins. The problems caused by these pressures often relate to both the chemical and the physical changes.

1.3 Indicators/metrics

1.3.1 LIFE Index

The LIFE index (Lotic Invertebrate index for Flow Evaluation, Extence et al., 1999) was formulated to test whether it is possible to link changes in benthic invertebrate community structure with indices of historical river flow at a gauge close to the sample site. The LIFE index can be calculated from species or family-level bio-monitoring data. Every taxon is assigned a velocity preference from I to VI (based on literature data), and five abundance categories are used. Implicit in the 'velocity' preference is preference or avoidance of silty substrates. A matrix is then used to give a combined score for each taxon in the sample of between 1 and 12. The scores for all taxa are added together, and the average score is the LIFE index. It is important to note that the index *is expected to* be sensitive to natural and artificial flow changes; it thus allows an extrinsic hypothesis to be tested. Extence et al., 1999, demonstrated that correlations exist between LIFE score and moving averages of historical flows (e.g. Figure 1). LIFE is currently being used in England and Wales as part of the implementation of Catchment Abstraction Management Strategies and the Water Framework Directive (Soley et al., 2002; Dunbar et al., 2004), although these applications largely concentrate on expected LIFE score, predicted by the RIVPACS model (Clarke et al., 2004; Wright et al., 2000). LIFE is also being tested in the STAR project and its generic sensitivity to flow tested (Dunbar & Clarke, 2004). While the LIFE index has been shown to be influenced by hydrological variability, it can also be affected by habitat degradation (which also affects substrates and velocities), and to a lesser extent, water quality.

A similarly constructed index (MFR - mean flow rank) has been developed by the Environment Agency of England and Wales to relate flows to macrophyte communities, but is currently unpublished, except in Soley et al., 2002.

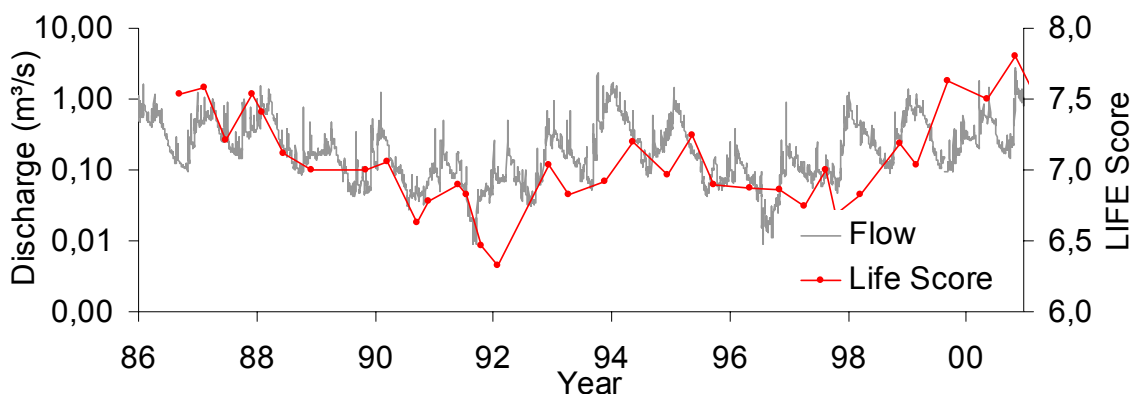


Figure 1.1. Flow and the LIFE score in the Waithe Beck in the UK (data from Extence et al., 1999)

1.3.2 Habitat Suitability Indices (HSIs)

In the context of hydromorphology, it is sometimes possible to link river flow data to community response (e.g. the LIFE methodology). However, this often requires a greater jump than available

data will allow. An alternative is to relate flows to habitat (physical variables) via a hydraulic model (Section 3.5.5) and habitat (physical variables) to ecology via habitat suitability indices (HSIs) or preference indices. Physical HSIs allow relation of habitat preferences for a given species and life stage to hydraulic parameters such as velocity, depth and substrate. Most HSIs have been defined as univariate response functions but more complex multivariate relationships also exist (Parasiewicz & Dunbar, 2001). Habitat preferences take into account the available habitat at the time the data are collected. Examples of HSIs (preference indices) for two life stages of brown trout developed on UK data are given in Figure 2. Note that these HSIs were developed by underwater observation of fish in streams less than 1m deep. Up to this depth there was no evidence of avoidance of deeper water. However, extrapolation of habitat quality in deep water is consequently highly uncertain.

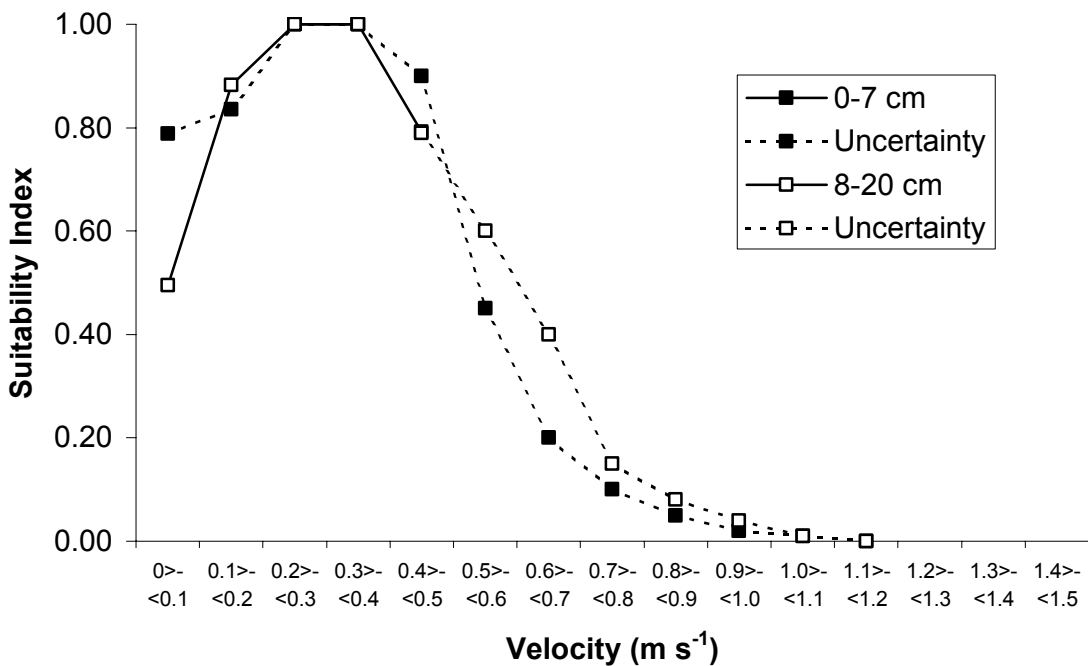
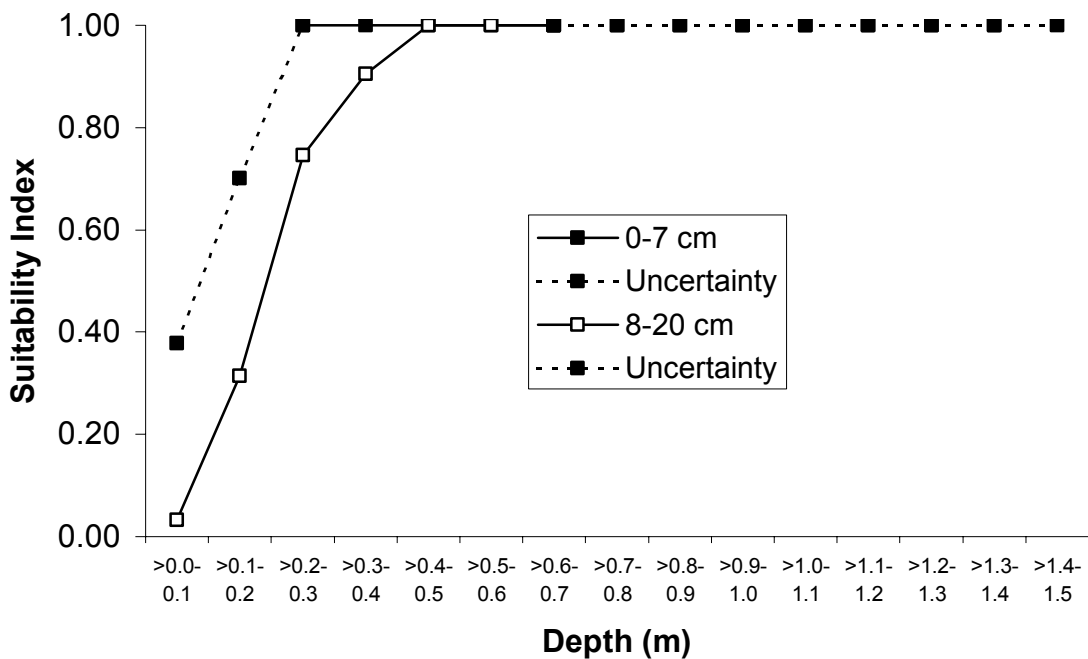


Figure 1.2. Habitat suitability indices (preference indices) for two size groups of juvenile trout (Dunbar et al., 2001).

1.3.3 Other fish indicators

The Fame project (<http://fame.boku.ac.at>) is currently developing fish-based metrics sensitive to various pressures, including hydromorphology. No published outputs are available at this time (but are expected to be available shortly).

1.4 Classification systems

1.4.1 Hydrological

The most well known classification system based on natural, commonly historical flows is the Tennant method (Tennant, 1976), sometimes also called the Montana method, which specifies that 10% of the average flow is the lower limit for aquatic life and 30% of the average flow provides a satisfactory stream environment. The Tennant method was based on hydraulic data from eleven U.S. streams (including streams in Montana) and considerations of what values of velocity, depth and width were needed for sustaining the aquatic life, with a focus on fish.

Historical flows can also be used to define 'an ecologically acceptable flow regime', although one should be careful to distinguish between pre-development historical flows and impacted historical flows, which may over a period of time have altered the composition of biota. Arthington et al. (1992) describe an 'holistic method' that considers not only the magnitude of low flows, but also the timing, duration and frequency of high flows. Such a flow regime would not only sustain biota during extreme droughts, but it would also provide the high flows and flow variability needed to maintain the diversity of the ecosystem. The range of variability approach (RVA) and the associated indicators of hydrologic alteration (IHA) identify an appropriate range of variation, usually one standard deviation, in a set of 32 flow statistics derived from the 'natural' flow record (Richter et al., 1997). The holistic and the RVA methods are conservative and maintain the ecosystem by retaining the key elements of the natural flow regime. They are probably most appropriate for river systems where the linkages between ecosystem integrity and flow requirements are poorly understood or for preliminary ecological assessment. Approaches based on mimicking the natural flow regime can be considered as 'top down' in that all aspects of the flow regime are considered important. The building block method (BBM; King et al., 2001) while coming from a similar philosophy to the Australian holistic methods can be considered 'bottom-up' in that it aims to define standards for individual elements of the flow regime using structured guidance, but also on a case by case basis.

1.4.2 Morphological

Streams can be classified according to their hydromorphology. Different methods and indices are used in different countries. Austria uses the Austrian Habitat Survey (Werth, 1987; Muhar et al., 1996; Muhar et al., 1998), Denmark uses the Danish Stream Habitat Index (Pedersen & Baattrup-Pedersen, 2003), France the SEQ Physique (Agences de l'Eau & Ministère de l'Environnement, 1998), Germany the Ecomorphological Survey for Large Rivers (Fleischhacker & Kern, 2002), and the UK uses the River Habitat Survey (RHS) (Raven et al., 1998). The methods use a number of parameters (channel, bank, floodplain, flow-related) and a scoring system to evaluate the hydromorphological status of streams. Most of these methods are based on a pseudo reference condition, which is identified on the basis of the top x% of sites according to their habitat quality scores. This causes a

problem for the type of rivers of which there are only few, or few with no impacts (e.g., large rivers).

1.5 Models

1.5.1 Habitat models

The Instream Flow Incremental Methodology (IFIM) and other methodologies have been developed as a framework for river management, which are used throughout the world to identify flow regimes for ecological protection (Dunbar et al., 1998; Acreman & Dunbar, in press). Simple methods and more advanced habitat models have been developed as a part of this framework, primarily to evaluate the trade-offs associated with different water management scenarios. However, water managers have also attempted to use habitat models to set environmental 'minimum flows'. Most habitat models use preference indices (see Section 3.5.3), which determine how suitable a given quality element (commonly hydraulic parameters: velocity, depth, substrate) is for certain species and their individual developmental / life history stages. These models then combine the results from hydraulic models with the preference indices to produce values of river area weighted by habitat quality (weighted usable area) as a function of flow for a given species and life stage. Current software include PHABSIM (Physical Habitat Simulation; Bovee, 1982; Milhous et al., 1989) and RHABSIM (River Habitat Simulation) used in the United States, RHYHABSIM (River Hydraulics and Habitat Simulation; Jowett, 1989) used in New Zealand, EVHA (Evaluation of Habitat; Pouilly et al., 1995) in France, CASIMIR in Germany (Jorde, 1997), RSS (River Simulation System; Killingtviert & Harby, 1994) in Norway and HABITAT in the Netherlands (Duel et al., 2003).

Recently, the outputs from conventional habitat models have been generalised and linked to two dimensionless variables, Reynolds number and Froude number (Lamouroux and Capra, 2002). They found that Reynolds number (conveniently scaled to be equivalent to discharge per unit width) explained most of the changes in habitat area within reaches, and that Froude number at median discharge explained most of the differences between reaches. The relationships have been tested on datasets from France and New Zealand (Lamouroux et al., 1999; Lamouroux et al., 2002).

Habitat models have received criticisms both for being too complicated and too simplistic, one of the key issues has been the development and transferability of the preference relationships. However, as few alternatives are available, they remain key tools for assessment of environmental flows, particularly for assessment of more significant hydrological pressures.

1.5.2 Bioenergetic habitat models

Relationships between fish growth and environmental variables such as food intake and temperature have been known for many years; simple models predicting fish growth have also been developed. The concept of a bioenergetic model for fish growth has been added to by several researchers (e.g. Fausch, 1984; Hayes et al. 2000, Guensch et al., 2001; Booker et al., 2004). They added a shorter-term behavioural component in order to use these models to predict profitable physical habitat locations. This concept works well for juvenile trout and salmon which are 'sit and wait' predators intercepting drifting invertebrate prey. Gross energy (E_{gross}) intake is represented by the energy value of drifting invertebrate food delivered through the water column to the fish. The maximum E_{gross} is in turn determined by a capture area model. Costs of feeding (used energy due to movement) are then taken from E_{gross} . The behavioural bioenergetic model uses information on water depths and velocities in order to determine the energy value of different areas of a river channel.

A potential advantage of such an approach is that its process basis should be able to overcome some of the fundamental problems with empirical descriptions of habitat suitability, particularly with regard to transferability of such relationships between rivers. Ultimately it should be possible to place greater confidence on predictions from such models, especially under scenarios that fall outside the range of field measurements.

1.6 Knowledge gaps

A substantial amount of scientific literature has been produced on the effects of hydromorphological pressures on stream biota. Annex 2 gives a list of the number of papers in 'Hydromorph_1946_2004.enl' that have specified hydromorphological keywords in either the title or the abstract.

Despite the huge number of papers published on the topics, we have identified the following knowledge gaps or needs:

- Generic ways of summarising flow regimes and morphological conditions, and their deviations from natural, which can be related to ecological indicators.
- In contrast to say organic pollution of rivers, or eutrophication of lakes, there are currently very few biological indicators that are sensitive to hydromorphology. This is in part due to the difficulties in assembling the required datasets to develop and test the indices.
- No methods have been developed to separate combined hydromorphological effects. Often a change in biology is associated with several hydromorphological changes and there are no methods to predict the effect of each individual hydromorphological factor (e.g., hydrological and morphological).
- There is a lack of knowledge understanding and describing macrophytes as structural features and physical moderators of habitat. Small weedy lowland streams are often the most pressured because they are located in lowland areas where water demand is usually high.

There is a need for:

- Quick methods of assessing critical hydrological pressures and their effects on the biota. Important hydromorphological elements include:
 - 1) Morphology, including depth, velocity and substratum, and its variability.
 - 2) The flow regime, including low flows, average flows and high flows, their timing, magnitude, frequency, duration.
 - 3) Weed cutting and dredging.
- Biological indicators or metrics that reflect the degree of hydromorphological pressure, including the lateral connectivity of streams. The LIFE index is an invertebrate indicator that varies with hydromorphology, but no appropriate indicators or metrics have been developed for other trophic levels.

1.7 References

Note: This is a reference list for the works cited in the above chapter. A full bibliography is found in the Endnote file 'Hydrolmorphology.enl', holding 280 references.

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Annex 1

Table 1. Search words used for the literature review.

Hydromorphological	channel, canal, meander, braided, embankment, point bar, site bar, sinosity, management, maintenance, weed cutting, dredging, physical, scour, substrate, spawning gravel, velocity, erosion, deposition, siltation, stream bed, river bed, run, glide, mire, riffle, pool, backwater, habitat, hydrologic, hydraulic, fluvial, regime, discharge, abstraction, diversion, bankfull, hydro electric, hydro power, dam, reservoir
Biological	macroinvertebrate, invertebrate, insect, arthropod, periphyton, algae, benthos, diatom, filamentous, macrophyte (not riparian and not wetland), higher plant (not riparian and not wetland), submerged plant (not riparian and not wetland), trout, salmon, fish, eel, perch, pike, roach, bream, chum, whitefish, minnow, brook, lamprey, phytoplankton

Table 2. The number of papers of the 11,705 references in 'Hydromorph_1946_2004.enl' with a given keyword in either the title or the abstract (* indicates wildcard)

Keyword	Number of papers
Dam	1498
Channeli*	137
Weed cut*	12
Abstraction	64
Diversion	136
Draining	139
Urbanisation/urbanization	53
*forestation	59

2 Acidification of rivers and lakes

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2.1 Introduction

Acidification has for a long time been a serious pollution problem in different regions of Europe as well as in other parts of the world. Acidification of water implies a pH reduction caused by anthropogenic activities or by natural processes. It is however, important to keep these two causes for low pH separate as they represent waters having different levels of toxicity.

Acidification was regarded as the cause for several fish kills and absence of fish in numerous lakes in Northern Europe prior to 1950. The acknowledgement of the problem and the chemical solution was known already in 1920. Liming was then initiated at hatcheries to enhance survival of trout and salmon (Dahl 1921, 1927; Sunde 1926). However, the cause-effect-chains, linking acid rain to acid water were established first in the 1950s (Dannevik 1959). Acidification became an international issue around 1970 (Almer, 1974). Since then, focus has changed, from pH being regarded as the main factor for environmental changes prior to 1980, to aluminium mobilized from the edaphic environment being identified as the prime stressor in 1978. Focus has also changed from “proving” effects and causal relationships in the 1980’s to present day topics being aimed at identification of reference conditions and confounding factors. At present, acidification is closely linked to climate change and how climate effects both acidification and recovery processes (see Acid Rain conference call; Prague, 2005). While identification of the magnitude of the problem and suggestions for mitigation actions was important in the 1980’s, present focus is more related to recovery or the lack of recovery following deposition reductions (Skjelkvåle et al 2003).

From 1970 damage from acid deposition on forests was observed close to industrial areas in parts of Europe, and in some cases forests died. Rivers in regions with high acid deposition and low acid buffering capacity of the water catchments the reproduction of salmon did not succeed. Acidification has affected numerous compartments of the ecosystem, where fish is especially vulnerable. Acidification has had a large influence on stocks of Atlantic salmon (*Salmo salar*), brown trout (*Salmo trutta*), roach (*Rutilus rutilus*), Arctic char (*Salvelinus alpinus*) and perch (*Perca fluviatilis*). Simultaneously to the decline in the fish populations, changes in all parts of the ecosystem were observed. The decomposition rate of allochthonous material declined dramatically. The communities of macrophytes changed as did the periphyton and macroinvertebrate communities (D’Itri 1982, Raddum & Fjellheim 1984, Farmer 1990). There was a change from more or less acid sensitive to acid tolerant communities. During the 1980’s macroinvertebrate indices were developed to quantify these changes. Later assessment systems were developed also for other kinds of biota.

2.2 Sources of acidification

Acidification implies a reduction in pH. This reduction can be brought about by various pressures, including acid rain, industrial waste discharge, changes in forestry management practices, draining of mire, marsh- and wetlands and by e.g. mining activity (Hyne and Wilson, 1997), but can also be due to natural watershed processes including sulphur reduction and the properties of organic acids (Gensemer and Playle, 1999). Most of the above-mentioned pressures have only local effects as they are generally limited to individual watersheds. Acid rain however, is transboundary, and has a global influence with respect to deposition and is as such regional (see articles from 6th Acid Rain Conference in Japan, Dec. 2000; Water, Air & Soil Pollut. No.130). Acid rain excerpts its im-

impact on water quality through interactions with the soil geo-chemistry, where local variation in impact levels are related to variations in the geo-chemical properties of the bedrock and soils (composition and weathering rates) and to variation in precipitation and deposition patterns. In addition, factors like vegetation cover, climate, and hydrology will increase variation in the chemical response to the acid deposition. Acid rain is mainly related to burning of fossil fuels and the subsequent emission of sulphur dioxide and nitrogen oxides into the atmosphere. In addition, ammonia from agricultural activities contributes to the acid loading through the nitrification process. In air, and in contact with water, the oxides are transformed into sulphuric and nitric acids. The acidifying compounds are emitted into the atmosphere, where they can be transported over large distances, before being deposited as “acid rain”. At a European scale, acidification has negative impact throughout most of northern Europe, including the British Isles and the Alps.

The deposition of acid rain over Europe is at present “low” compared to the “peak” years in the 1970s. This reduction and chemical “recovery” has been brought about through changes in industrial economy the implementation of several international agreements, where the latest multi purpose protocol aims for a cut in SO₂ deposition by 75% and NO_x deposition by 50% within 2010 relative to the emission levels in 1990. Although sulphur deposition has declined in North America and Europe (Bull et al. 2001; Stoddard et al., 1999; 2000; Schöpp et al. 2003), no significant decrease in nitrogen deposition can be expected in the near future (Alewell et al., 2001ab). This situation is demonstrated by the wet deposition of sulphur and nitrogen in southern Norway during the last decades (Figure 1). Future effects relating to deposition of N represents as such an uncertainty. Reduction in acid loading will initiate a recovery process, where the recovery rate is related to changes in the deposition of acids, but also to local variations in soil properties e.g. weathering rates. Areas having more easily weatherable soils (more calcareous) and biogeochemical buffering capacity is predicted to recover prior to regions having more acidic soils (more siliceous). As the resistance or tolerance to acid rain is unevenly distributed in Europe, sensitivity to acid influence varies throughout Europe (Skjelkvåle & Wright 1991). Due to this variation, some, but not all water bodies having acidification problems will show improving water quality by the target year 2016 (Fig.4).

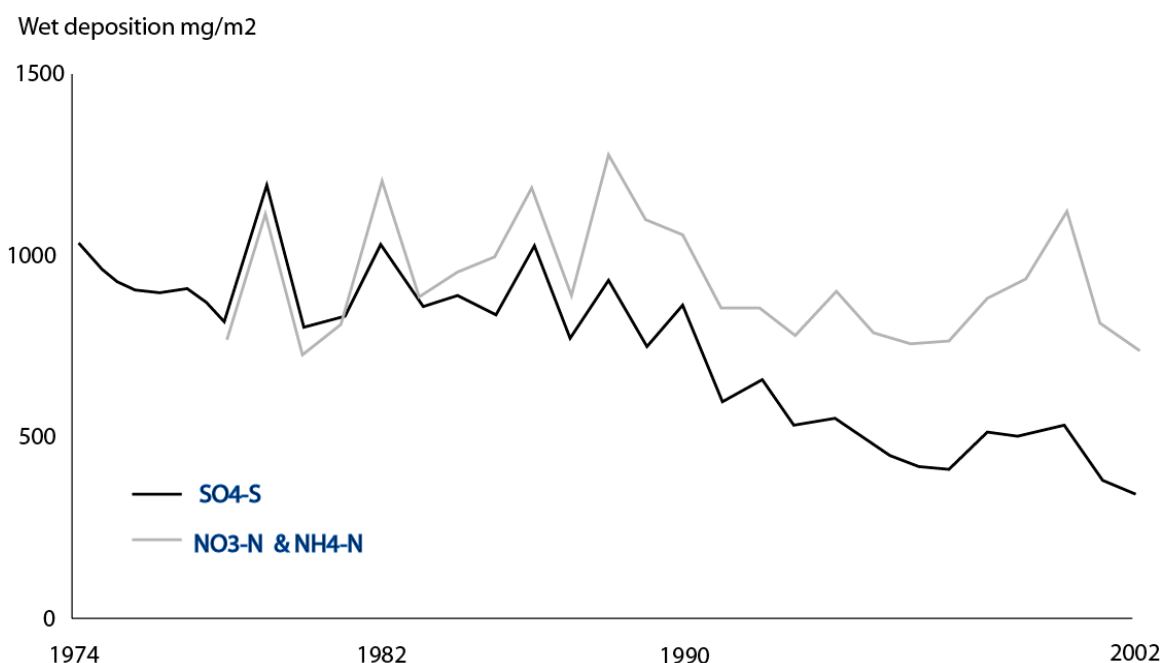


Figure 2.1. Trends in the wet deposition of sulphur and nitrogen shown as averages from 7 representative sites in southern Norway (redrawn from Statens Forurensningstilsyn (SFT), 2003).

2.3 Impacts on water quality

2.3.1 Key chemical water quality elements

The chemical impacts on surface water quality from acid water are well known. Although acid rain often is associated with a SO_4 -increase, water toxicity is related to the combined effects of numerous changes to water chemistry. The immediate effect of the acidification is decreasing pH and alkalinity. Besides increasing H^+ -concentration, acidification also changes cationic and anionic species composition and mobilizes metals from the edaphic to the aquatic environment. Streams draining water sheds with low buffering capacity will have increased levels of aluminium. The increase of aluminium is especially important, as aluminium is toxic for many aquatic organisms, including plants, macroinvertebrates and fish. The solubility of many metals increases substantially when pH changes from alkaline to acidic conditions. For example, as pH is reduced from 8.1 to 4.5, metal concentrations from average rock in water would increase from 0.03 to 500 $\mu\text{g}/\text{l}$ for iron, from 0.003 to 25 $\mu\text{g}/\text{l}$ for copper, from 0.006 to 170 $\mu\text{g}/\text{l}$ for nickel and from 0.004 to 80 $\mu\text{g}/\text{l}$ for zinc (Kramer 1976). Increased deposition of metals follow the deposition of acidifying compounds is also observed. Whether originating from leaching or from deposition, trace metal concentrations are elevated in acidified streams. In Norway and Sweden there has been a decrease in the deposition of metals during the last decades, except may be for copper (Lydersen et al 2002).

There is an intimate relationship between acid rain, geo-chemistry and water quality, where the magnitude of the pH-reductions varies between individual water bodies and between regions, depending on intensity of acid rain, precipitation, and climate. While the reduction in pH can be high in clear-water systems, the reductions in pH are often smaller in water bodies influenced by organic matter. This is partly due to pH being logarithmic. Similarly, during water quality recovery, pH is expected to increase more in water having low organic content than in water bodies where organic acids maintain a low pH. Changes in aluminium concentration and speciation is however more important to the biota than any increase in pH. Aluminium is in water present on different forms varying in size, and in reactive and charge properties. Of these, the form defined as inorganic monomeric (Ali) or as labile (LAI) or cationic aluminium contains the species that are toxic. Different analytical protocols are used at the various analytical laboratories to separate these fractions. The concentration of toxic aluminium reported is unfortunately related to the analytical protocol, hampering comparison of data across studies. The toxicity of LAI is further related to the organic content of water, to temperature and to the concentration of base cations (Gensemer and Playle, 1999). Under certain conditions, and especially within rivers, there can be a large discrepancy between the concentrations of LAI reported after traditional analysis of aluminium and the concentrations actually experiences by the water dwelling organisms (Gensemer and Playle, 1999). Downstream the confluence of acid and lesser acid waters, aluminium will polymerize to lesser or non-toxic forms of aluminium. The transformation rate is time, temperature and pH dependent. As unstable forms can persist for hours, extensive areas of the river can be under influence of this ongoing polymerizing process. Dose-response relationships that are based on in situ fractionation of aluminium (within seconds after sampling) yield better and different relationships from those based on traditional stored water samples. The in situ method is only practical in experiments, but knowledge concerning the kinetics is imperial for the understanding of how water quality can be poor in rivers despite pH being satisfactory (Kroglund et al. 2001a, 2001b).

Reduced acid loading initiates a succession of changes, where soil properties are restored first, then water quality and finally biological properties. During the chemical recovery phase, the base cation concentration is reduced paralleling the reduction in SO_4 and is expected to remain low until weathering recovers the base saturation of soils. Biological recovery however implies only that the water quality has improved sufficiently to permit reestablishment of the acid sensitive species that were present prior to acidification. Reestablishment does not require that necessarily all water

quality components are restored. Different species have different water quality requirements, hence different critical limits. The least sensitive species can be the first to recolonize water bodies that previously were uninhabitable. Due to species and site-specific recolonization rates, biological recovery will be further delayed compared to chemical recovery. So, although acidification to a large extent is a problem of the past, past depositions will continue to have biological effects for decades (Gunn and Sandøy, 2001, 2003). Our ability to detect recovery is also reduced due to poor understanding of what the preacidification biotic composition looked like. Several surrogates have been proposed using time for space substitution and analogues to establish reference conditions. Some regions of Northern Europe are not expected to recover following the implementation of present protocols (see Fig. 4). These predictions lean heavily on the dose-response models that are presently being used, where faulty identification of dose will offset the predicted recovery rate.

Although acidification is often represented by changes in pH, acidification is not a single stressor; but is a combined or multiple stressors, involving more than two components. One should therefore avoid undue attention to changes in H^+ . As there is a large variation in the causal relationships influencing water quality, the relative contribution of the various physio-chemical elements influencing the biological community in the individual water body needs to be identified and separated, e.g. the toxicity relating to H^+ needs to be separated from the effects relating to aluminium or other metals. An understanding of the mechanisms and processes influencing changes in toxicity are imperative to understanding where and when environmental damage/recovery is to be anticipated. Based on this, it is likewise important not to confuse natural acidification with anthropogenic acidification. While both are associated with a pH-reduction, elevated levels of aluminium in its toxic form is mainly encountered in the latter, or in ecosystems influenced by strong acids. Water bodies acidified by natural processes can have low pH due to high content of organic (humic) matter, following oxidation of sulphide rich soils and to some extent by sea salts. Furthermore, nutrient uptake by plants and trees, production of CO_2 by respiration and several other processes can contribute to pH reductions. At the same time, a water body that is acidified due to natural causes can also be additionally acidified due to the deposition of strong acids, and hence contain metals at toxic concentrations, despite pH being close to reference levels. The correct identification of pre-industrial water quality is therefore imperative for a correct identification of biological reference conditions. Naturally acidic water bodies can have a "high ecological status" despite having a low pH and an H^+ restricted biological community. In water bodies affected by acid rain, species restricted by H^+ can colonize the system when pH is sufficiently high, while species restricted by aluminium can only colonize the system when aluminium (and other metal) concentrations are below the toxic thresholds. Species being restricted or those who have benefited from changes in the biological community will first respond when the biological constraints are removed. This complex interaction between chemistry and biology will affect recovery rates.

Acidification can be continuous or more episodic. In the early days of acidification, the toxic periods were probably episodic, to be followed by a more continuous acidification state as the critical limits became more and more exceeded. During the recovery phase, water quality can still be impaired due to episodes. These can be brought about by variations in deposition of acidifying substances, precipitation and by sea salt episodes (Evans et al., 2001; Hindar et al. 2004). The biological community is generally not restricted by annual averages, but by the chemistry during periods having ecological consequence for survival. Timing of episodes to sensitive biological periods is therefore crucial to biological recovery. Furthermore, an episode lasting only for a short period of time can still have minor biological effects, while episodes that have a lesser intensity but longer duration can have large ecological effects.

ANC (acid neutralizing capacity) is frequently used as a surrogate for the dose (being pH and aluminium). ANC expresses the relationship between base cations (SBC) and the strong acid anions (SAA). A low ANC means that the ability to buffer the acidity is small and the lake or soil is sensitive to further acidification. The acidification related reduction in ANC is due to decreased bicarbonate (HCO_3^-) and increased H^+ and cationic Al species (Al^{n+}) (Evans et al. 2001). ANC can

also be reduced due to dilution. The use of ANC has been questioned, especially as it does not predict water quality “correctly” in water bodies heavily influenced by organic matter (Lydersen et al 2004) or in regions affected by sea salt episodes (Harriman et al., 1995; Evans et al., 2001; McCartney et al 2003). In these, the relationship between H^+ and Al differ from the relationships obtained at clear water sites (Lydersen et al., 2004). Ecological status can therefore be incorrectly set for water bodies having a high organic content (naturally acidic) or that are being affected by sea salts. ANC is at the same time closely related to pH, alkalinity and aluminium. ANC is used to predict biological status for individual species and for communities, and to forecast chemical changes permitting biological recovery (see Fig 1). Empirical relationships have been developed for fish (Lien et al. 1996; Bulger et al. 1993; Kroglund et al., 2002), macroinvertebrates (Raddum and Fjellheim 1984; Raddum and Skjelkvåle 1995), and diatoms (Juggins et al. 1995). An ANC threshold of $20 \mu\text{eq l}^{-1}$ indicates water quality required for the good ecological status mandated by the WFD. However, the relationship between ANC, water quality and biological health is not straightforward. As mentioned previously, ANC values depend on both organic content and are affected by sea salts. The relationship between ANC and pH or LAI is also related to TOC (Fig 3; data from the 1000-lakes survey, 1986). Assuming biological responses are related to H^+ , ANC must be higher in water containing organic matter than in water with low organic content to obtain the same biological protection. Furthermore, for a given ANC, there is more LAI present in water containing organic matter than in clear waters. As a consequence of this, to protect the biological community, ANC must be higher in water containing organic matter than in clear water systems. The same ANC value does not provide the same level of protection, irrespective of other water quality traits. These problems arise because a surrogate for the true toxic components is being used. The relationships between H^+ , Al, base cations, SO_4 , NO_x and ANC needs to be re-evaluated, putting focus on confounding factors. In the future, when acid rain is abolished and the water contains no more toxicants, there should no longer be any relationship between ANC and biological status. This implies that targets based on high ANC values can require water to be restored to reference conditions that never existed. This flaw has few present implications, but will have increasing implications for the determination of chemical and biological targets in the future.

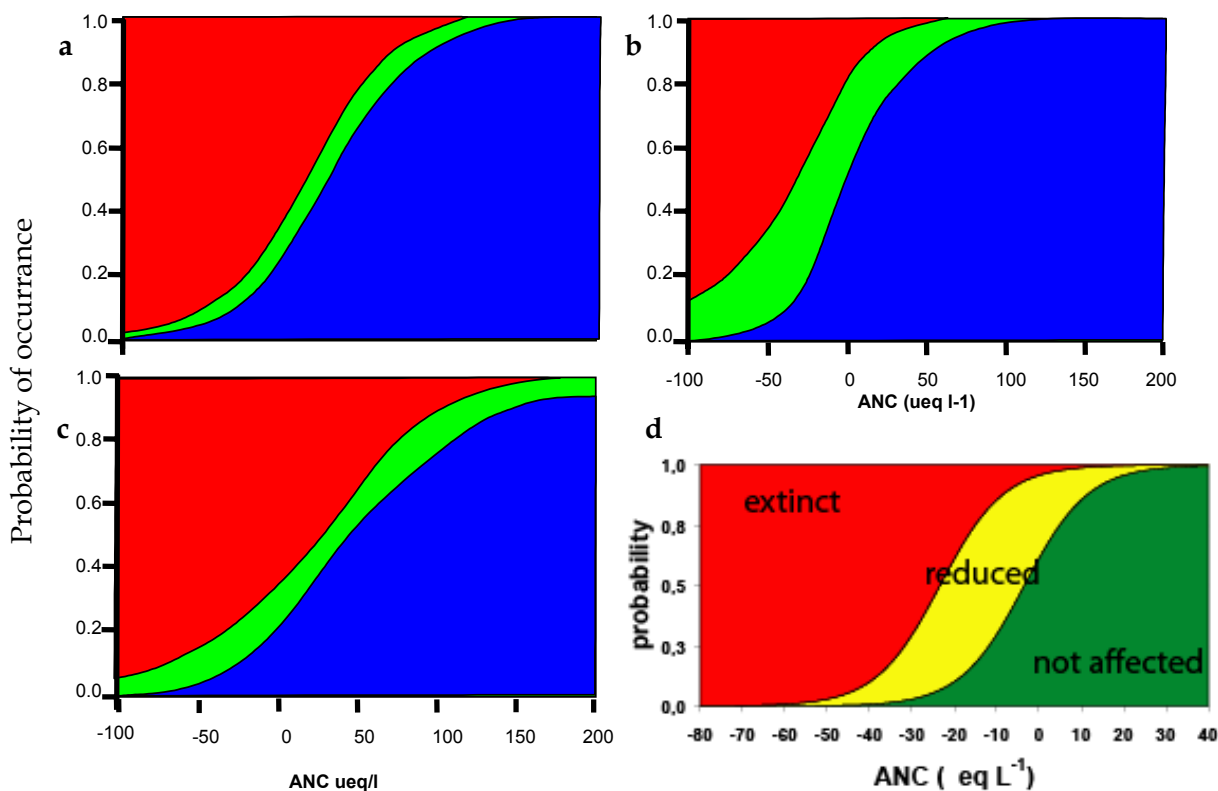


Figure 2.2 Response curves derived using logistic regression and showing the relationship between ANC (ueq/l) and the probability of occurrence of (a) the diatom *Achnanthes minutissima*, (b) the mayfly *Baetis rhodani* and

(c) non-impovertished macroinvertebrate assemblage and (d) brown trout. Figures redrawn from Juggins et al. 1995 and Lien et al, 1996)

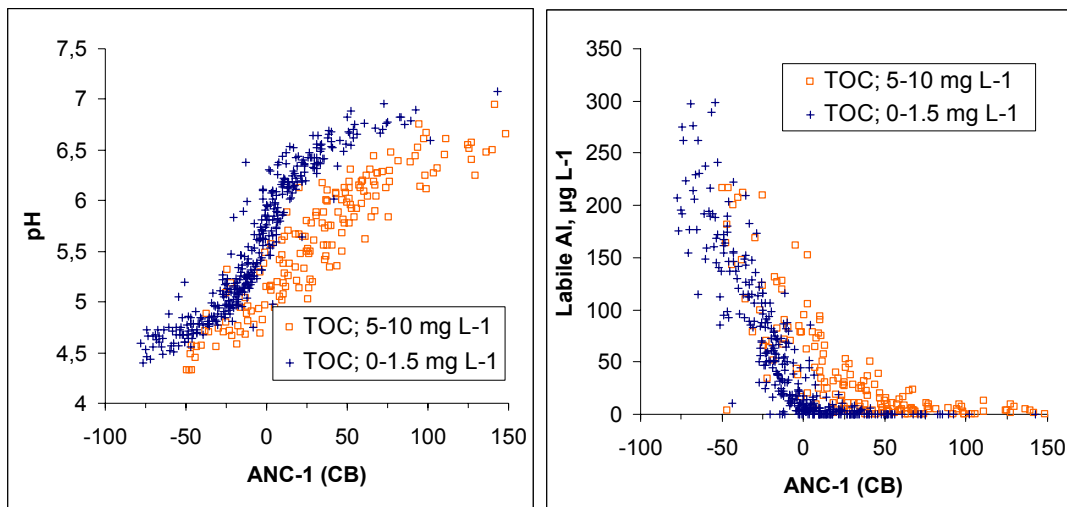


Figure 2.3. Relationships between ANC and (a) pH and (b) inorganic <labile> aluminium µg/l for Norwegian lakes having low (<1.5 mg C/l) and high (>5 mg C/l) organic content. Data from the Norwegian 1000-lakes survey in 1986.

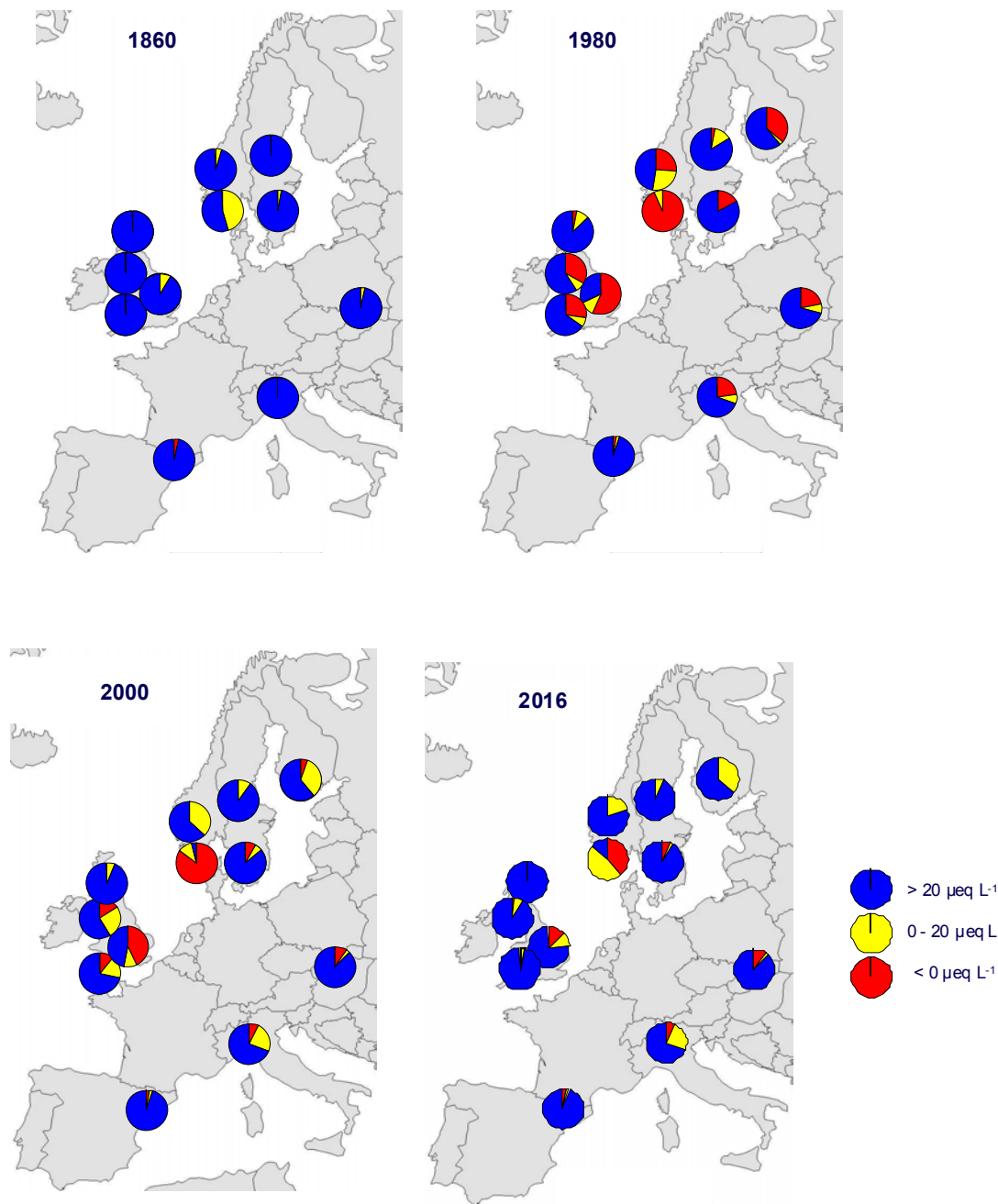


Figure 2.4. The 1860 & 1980 reconstructed, 2000 observed and 2016 predicted surface water ANC concentration for each region expressed in 3 ANC classes (from Moldan et al 2004).

2.3.2 Water quality modelling

There are numerous models based on chemical criteria. Most models rely on the critical load concept. Critical load is defined as the maximum amount of a pollutant that can be deposited on an ecosystem without adverse effects (Nilsson and Grennfelt 1988). Correct setting of critical load requires that the relationships between deposition (chemical pollutant) and biological impact are correctly understood. In the case of acid deposition, the critical load is the maximum deposition of acidity that can be deposited without having adverse effect on the ecosystem. Application of critical loads involves identification of a key organism (or organisms) to be protected, identifying a

“critical limit” for the concentration of a water chemistry indicator and a model to relate deposition rate to the indicator status. Several chemical indices can here be used, e.g. pH, aluminium, ANC, Ca/H⁺-ratio etc. Changes to water quality that have no biological implication are to be interpreted as acceptable. As the chemical and biological sensitivity of acidification varies according to the kind of bedrock and soil type throughout Europe the critical load varies accordingly (Figure 5).

Critical loads and their exceedance are calculated using various static models, such as the steady-state water chemistry model (SSWC) and the first-order acid balance model (FAB) (Posch et al. 1997). Steady-state models indicate that something will happen in the future if e.g. pollution is reduced, but does not indicate the time scale of recovery. Rates in change can be modelled using dynamic models.

Geo-chemistry and soils play a central role in the response of forests, lakes and rivers to acid rain. Dynamic, process-based models integrate and interpret theoretical knowledge from soil science and hydrochemistry with results from experiments and monitoring. Response times are related to the content of mineral- and organic matter, weathering rates etc; all properties that are more or less unique for the individual site. Because of complex nature of interactions, dynamic models are used to predict time rate of changes. Experimental data from ongoing experiments (long time series) are used to validate these models. The main dynamic model used in acidification studies are the MAGIC model.

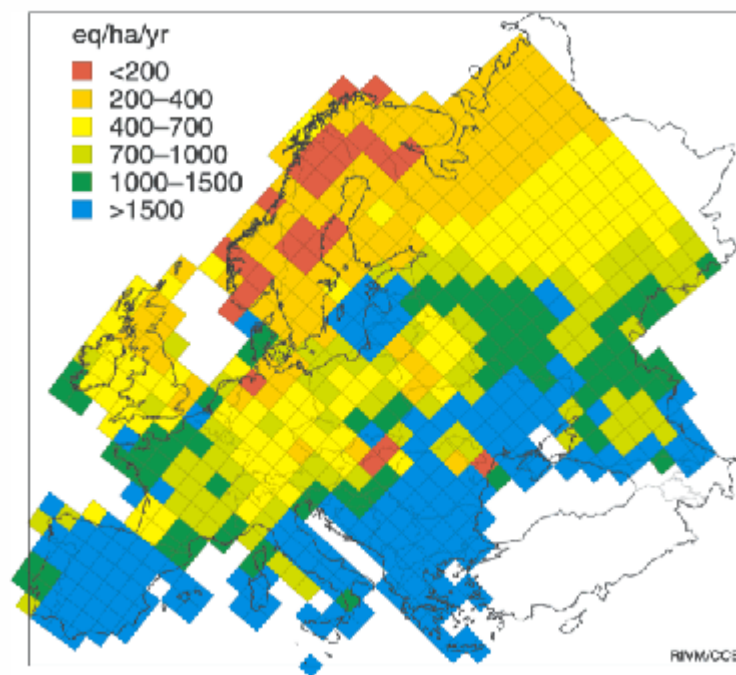


Figure 2.5. Critical loads for acid deposition. Red indicates high sensitivity, blue low.
(from The Swedish NGO Secretariat on Acid Rain; <http://www.acidrain.org/acidification.htm>)

At present there are no dynamic models for biological responses to acidification. The ASRAM-model for Atlantic salmon represents an exception (Korman et al. 1994). Dynamic models can predict the time lags between changes in chemistry and the biological response.

2.4 Impacts of acidification on biota

Acidification can (will) affect all biological components of the aquatic ecosystem, where the severity of the response is related to the intensity and duration of water quality impairment, and the relative sensitivity of the individual species present prior to acidification. During acidification,

species that were restricted due to biological interactions can increase in abundance. As such there is a chemical and a biological component regulating species diversity. Depending on the degree of chemical change and on species specific sensitivity to these changes, the presence of some freshwater organisms is restricted by acidifying water. Other species are mainly affected by indirect causes, e.g. by change in inter- and intraspecific competition and predation. The biological response patterns are as such complicated, depending on both the chemical and biological species composition in the individual water body. Furthermore, toxicity is enhanced or reduced depending on variation in e.g. ionic strength (with particular emphasis on calcium), organic content and with respect to temperature (Rosseland and Staurnes, 1994; Gensemer and Playle, 1999).

2.4.1 Primary producers

Most acidified watercourses lie within landscapes that have hard inert bedrock, have sandy soils or are dominated by peat land. The rivers in such landscapes are generally fast flowing, and phyto-benthos (mainly attached algae) and bryophytes as the main vegetation types (primary producers). Macrophytes are less common, they are normally restricted to moderately to slow flowing sedimentary reaches. The current velocity is usually too high to develop a significant biomass of phytoplankton and phytoplankton will not be assessed in this context.

Impacts of acid water

Several changes appear during acidification. These are reported to affect functional as well as structural properties of primary producers in rivers (Stokes 1986, Elwood & Mulholland 1989, Planas 1996). So far, structural changes are better documented and understood than functional. Some structural changes caused by acidification are well documented: 1) species composition changes, 2) species diversity decreases and 3) vegetation biomass may increase, due to one or a few primary producers that develop abnormally large biomass.

Nutrient availability

Acidified waters are usually nutrient poor, and changes in nutrient availability, that affect primary production, are often observed during acidification (Grahm 1997, Turner et al. 1987, 1991, Fairchild and Sherman 1990, 1992, Lindstrøm 1996, Planas 1996 a.o.). An oligotrophication (depletion of nutrient supply, particularly phosphorus) was hypothesised in acidic waters (Grahm et al. 1974). The extent to which this is a general phenomenon remains to be documented (Olsson & Petterson 1993).

Nitrogen, NO_x has taken on increased importance as a component of acid deposition (Gahnstrøm et al. 1993, Axler et al. 1994, Planas 1996). The consequences of increased nitrogen deposition to acidified freshwater ecosystems, is by no means fully understood. One probable effect is increased production and accrual of primary producers, a phenomenon observed in regions with increased nitrogen deposition (Hermann et al. 1993, Axler et al. 1994, Lindstrøm 2001). This hypothesis is supported by recent studies that identified nitrogen as the primarily limiting nutrient in some oligotrophic watersheds and observed increased algal biomass due to experimental nitrogen addition (Lindstrøm 1996, Cessmann et al. 1992, Francoeur & Biggs 1999, Lindstrøm 2001).

Another possible effect of increased nitrogen is changes in species composition due to increased nitrogen/phosphorus ratios. The phosphorus content in nutrient poor acidified rivers is low, generally less than 5 µ P/L, and the nitrogen/phosphorus ratio used to be around 20-40 before the nitrogen deposition increased. Today, nitrogen/phosphorus ratios around 100-200 are found regularly in these rivers. Some attached algae, mosses and macrophytes are reported to be particularly common in waters with such elevated nitrogen/phosphorus ratios (Lindstrøm et al. 2004). Unfortunately, the lack of proper historical data has so far prevented a test of this theory.

The importance of inorganic carbon as a key element in the build up of organic material is often overlooked. Carbon is the main structural element and is needed in much larger quantities than nitrogen and phosphorus during primary production (Redfield 1958, Kalert 2001). Studies in nutrient poor, acidified rivers revealed that some filamentous green algae produce organic material that, measured by weight, contains >500 times more carbon than phosphorus (Lindstrøm & Johansen 2001). The state in which carbon occurs changes dramatically during acidification, from mainly bicarbonate (HCO_3^-) to free carbon dioxide (CO_2), Figure 6. This change in the relative proportions of inorganic carbon species causes extensive changes in species composition of freshwater primary producers, because some use bicarbonate to produce organic material, while others use carbon dioxide. This is increasingly acknowledged as the main cause for changes in species composition and diversity when a water-body becomes acidified (Roelofs 1983, Stokes 1986, Keeley 1998, Elwood & Mulholland 1989, Shaphiro 1990, Turner et al. 1995, Planas 1996, Vinebrooke & Graham 1997, Madsen et al. 2002, Brouwer et al. 2002).

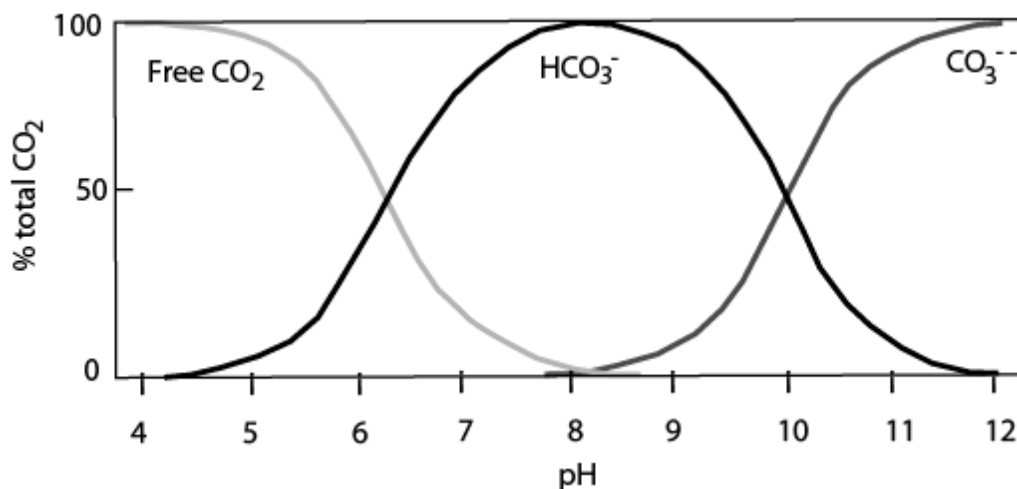


Figure 2.6. Relation between pH and the relative proportions of inorganic carbon species of (CO_2 , HCO_3^- and CO_3^{--}). Slightly modified from Wetzel 1983.

Species composition

In a study of a large number of acidified rivers in Norway, pH optimum and tolerance towards acidity were assessed for benthic algae, bryophytes and macrophytes (Lindstrøm et al. 2004). The study revealed well defined tolerance limits for many taxa (Table 1). Surprisingly many tolerated $\text{pH} < 5.0$ and were classified as acid tolerant. Some even seemed to profit from acidification and increased significantly in frequency and biomass in markedly acidified rivers ($\text{pH} < 5.0$). On the other hand, a large number of taxa were never found below $\text{pH} 6.5$ and some were even limited to rivers with pH permanently above 7.0. Different requirements concerning carbon source is probably the main reason for these differences.

Table 2.1. Tolerance limit towards acidity for 205 taxa of freshwater vegetation based on large datasets collected in Norway (Lindstrøm et al. 2004)

Vegetation type	Total number of taxa assessed	Acid tolerant $\text{pH} < 5$	Weakly acid sensitive $\text{pH} > 5$	Moderately sensitive $\text{pH} > 5.5-6.0$	Strongly sensitive $\text{pH} > 6.5$
Benthic algae	127	34	8	36 (divided in 2 groups)	49
Macrophytes	70	12	9	28	29
Bryophytes	11	5	3	3	Not assessed

Total	208	51	20	67	78
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Species diversity

Species diversity decreases during acidification, this affects diversity at a given locality as well as regional diversity (Eilers et al. 1984, Brodin 1994). The study in Norway demonstrated that the impact on regional diversity was more pronounced than at a given locality (Lindstrøm et al. 2004). In this study, a decrease in regional diversity of approximately 50 % was found for all vegetation types when pH decreased from 6.5 to <5.0. The regional decrease was caused by the disappearance of almost all categories classified as acid sensitive (not tolerating pH <5.0) illustrated for two groups of attached algae in Figure 7. When pH reached 5.0 the remaining groups were predominantly those that were classified as acid tolerant. These acquired significantly increased frequency and occurred in most of the acidified rivers. At a given river locality on the other hand, hydro-morphological and other physical and biological conditions strongly affected species diversity and tended to overrule acidification in controlling species diversity.

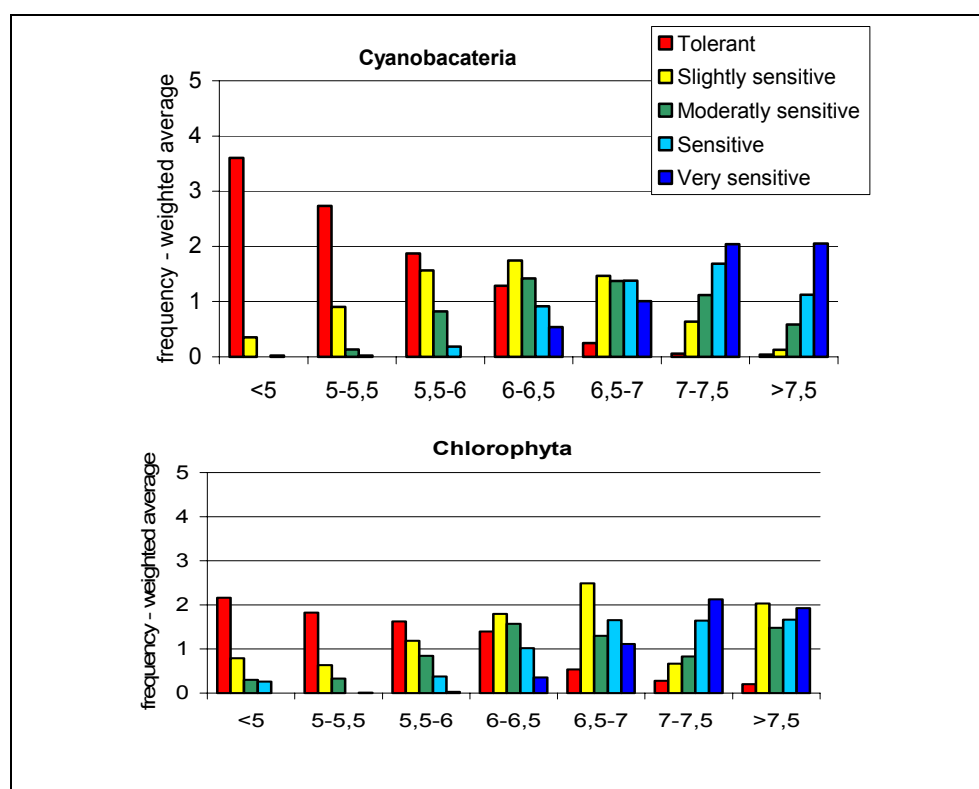


Figure 2.7. Distribution of 5 categories of acid tolerance benthic algae along the pH-gradient. Data based on cyanobacteria and chlorophyta found in 768 algal samples collected in Norwegian rivers (Lindstrøm et al. 2004). Benthic algae or plankton algae?

Biomass

Standing crop may increase and a few, sometimes only one, primary producer is reported to develop mass occurrence in acidified rivers. The most common and striking phenomenon is proliferations of filamentous green algae covering most of the river bed (Stokes 1986, Planas et al. 1989, Wei et al. 1989, Jackson et al. 1990, Turner et al. 1995, Lindstrøm & Johansen 2001, Vinebrooke et al. 2002, a.o.). These mats/clouds usually consist of Zygnemales, mainly one or a few species within the genera Mougeotia and Zygonium. Other types of mass developments are carpets of liverworts (Lazarek 1985, Brandrud 2002, Lindstrøm et al. 2004) and massive stands of bulbous rush (*Juncus bulbosus*) (Grahn 1997, Roelofs 1983, Havas & Hutchinson 1993, Brandrud 2002).

Functional properties

Functional effects of acidification on primary producers are not as well documented as structural. One important effect that has been observed, is enhanced dark respiration in phytobenthos communities (Schindler 1990, Turner et al. 1991 & 1995, Planas 1996). This is a clear stress symptom indicating perturbations of the nutrient cycling, which amongst others affects the production-rate and biomass accrual-rate of the primary producers (Schindler 1990, Turner et al. 1995).

Food web interactions

Modifications in food web interactions have been hypothesised to be important in determining the observed responses of primary producers to acidification. Decreased grazing pressure is given as an explanation to increases in vegetation biomass (Schindler et al 1991, Elwood & Mulholland 1989 a. o.). However, no clear and common relationship between grazers and benthic algae has been found, that explains the structural changes and the rapid changes in algal biomass (mainly increase) that takes place in some acidified waters (Planas 1996).

Aluminium and heavy metals

Some effects of increased aluminium and heavy metals in acidified waters have been observed in a few studies, but are so far not documented to have any strong impact on primary producers (Planas 1996).

Metrics - critical limits

Few indexes or models have been developed that relates directly between the state of the primary producers and the chemical properties of acidified waters.

Reconstruction of the acidification process and the pH development, by counts of diatom frustules in lake sediments, has been carried out for a long time (Renberg & Hellberg 1986, Stevenson et al. 1991). A similar approach has been tried in lakes and rivers, applying frustules of recent diatoms (Cameron et al. 1999).

A close co-variation was found between the state in which carbon occurs in acidic waters (expressed by pH) and the structural properties of primary producers (Lindstrøm et al. 2004). This provided the basis to develop an index that calculates the relationship between a biological element (in this case the primary producers) and a commonly measured chemical variable (pH) (Figure 8). The index calculates the presence of acid sensitive primary producers. Index calculations based on attached algae (Lindstrøm et al. 2004) showed that pH must be steadily 6.3 or more to make sure that the algal community is not impacted by acidification. In other words, the critical limit towards acidification for attached algae in running water is pH 6.3. The study also document that the algal community, with great probability is impacted if pH is 5.6 or lower, as no samples at pH below 5.6 had significant contents of acid sensitive taxa.

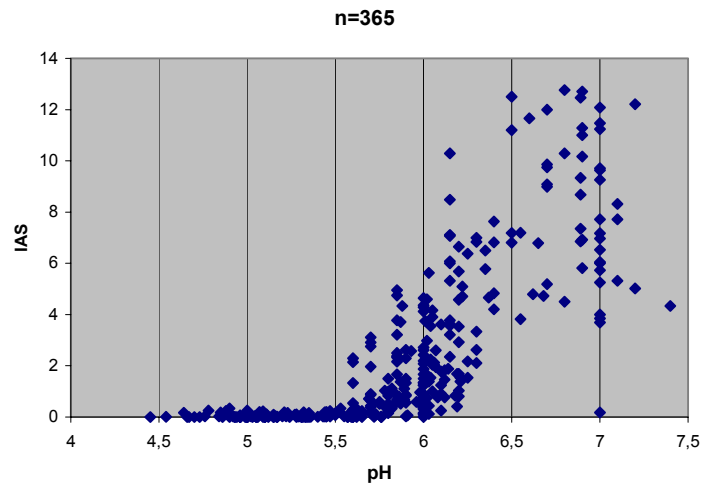


Figure 2.8. Index of Acid Sensitivity (IAS), development along the pH-gradient. Data based on attached algae collected from rivers in Norway (Lindstrøm et al., 2004) Benthic invertebrates

Indices and metrics

Indices based on macroinvertebrates have been developed as assessment tools during the last 2-3 decades in several countries in which acidification of rivers and lakes has been recognised as a significant problem. Acidification indices in Europe are primarily based on presence or absence of indicator species. Lists of species or higher taxa of known tolerance have been produced. The lists often exceed 100 taxa. A large part of these are, however, tolerant species commonly observed both in acidified and not acidified water. Some indices apply a multimetric assessment method including quantitative relations between taxa as well as taxa richness in the index. The scientific data material and experience on which the indices are based are for some regions and water qualities good whereas for other more scarce (Henriksson & Medin 1986, Raddum & Fjellheim 1984, Bækken & Aanes 1990, Lien et al 1996, Guerold et al 1999, Raddum 1999, Wiederholm 1999, Bækken et al., 2000).

Within the EU- project AQEM acidification assessment tools were identified for the countries within the project (AQEM: The Development and Testing of an Integrated Assessment System for the Ecological Quality of Streams and Rivers throughout Europe using Benthic Macroinvertebrates. (Hering et al., 2003). The number of taxa, including the number of EPT taxa (Ephemeroptera, Plecoptera, and Trichoptera), the distribution of locomotion types, the Medin index as well as the German acidification index was judged to be useful tools in assessing the impact of acidification (Henrikson & Medin 1986, Braukmann 1992, Aqem 2003).

In France a biotic index of acidification (B.I.A) has been proposed to assess the acidification in the Vosges Mountains (Guerold et al., 1997, 1999). The method uses a multimetric approach applying data on both taxa richness as well as the macroinvertebrate tolerance to acid water. The index has 10 classes that vary from 0 (unaffected) to 9 (strongly acidified). The index has shown to have a highly significant relation to the mean pH and ANC of the stream water.

The Swedish Medins acidification index is based on a substantial set of macroinvertebrate data from Sweden (Henrikson & Medin 1986, Wiederholm 2000). The tolerance list is mainly based on macroinvertebrate data from humus rich rivers in Sweden. The index is a multimetric index using more than one criterion to make a total assessment of the final acidification status. Besides the presence /absence evaluation of indicator taxa, it also uses the relation between numbers of individuals of the species richness in the assessment (this is unclear). The outcome of this index is put in one of five classes.

A Norwegian index, the Raddums index, is based on a set of data primarily from clear water rivers with low conductivity in the southern and western Norway. From these, an acidification tolerance list has been produced. Each taxon is given a score corresponding to a pH interval and to an acidification class in the index system (Raddum & Fjellheim 1984, Lien et al 1996). The knowledge on which the scores are based varies, for some taxa it is good for others less good. This index basically has four classes. In addition there is an option to make an assessment within the intolerant class (Raddum index 2) based on the relation between the number of individuals of the mayfly genus *Baetis* and the total number of tolerant stonefly individuals. The Raddum index has been used also on an European scale in e.g. the ICP-Water monitoring programme (Raddum 1999, Raddum & Skjelkvåle 1995).

Bækken & Aanes (1990) made some changes in the Raddum index tolerance list primarily by adding some tolerance assessment data from more brown water regions of eastern Norway. Another index was proposed for humic water by Bækken et al (2000), primarily intended for the eastern brown water streams in Norway. It is based on the previous knowledge of Norwegian indices and the Swedish Medins acidification index as well as unpublished Swedish tolerance assessments data by Engblom and Lingdell. The resulting acid water tolerance taxa list categorise the tolerance in four classes.

Impacts of acid water

Because macroinvertebrate communities of running water have shown to be seriously affected by acidification and some species at low levels, the macroinvertebrates have been used as "early - warning" organisms to detect acidification in an early phase (Otto & Svensson 1983, Raddum & Fjellheim 1984, Raddum et al 1988). Numerous works have shown that both structural and functional changes have been observed in the ecosystem of acidified streams (Stoner et al. 1984). The processes of leaf litter breakdown are severely reduced in acidified water (Burton et al 1985). A recent work of Dangles & Guerold (2000) observed the breakdown rate of leaves decreased nine times in acidified streams compared to circum neutral streams. Further, the feeding strategy of the macroinvertebrate community changed; scrapers were eradicated whereas both gathering and filtering collectors were drastically reduced. The effects may be a consequence of the toxicity of both H⁺ and aluminium.

Although emissions of sulphur oxides have been reduced, Guerold et al (2000) have demonstrated that the acidification continues to affect streams in the Vosges Mountains in N-E of France. These streams were in the past believed to function as a refuge biotope for numerous species. It was concluded that the acidification represent a real threat for numerous species of this region. This may apply also for other areas of acidification. However, in most cases there is very limited knowledge of the pre acidification fauna.

Critical load and critical limit of acidifying compounds have been key concepts in the study of biological effects (Nilsson 1986). "Critical limit" usually is defined in relation to the effect on the most sensitive species. This implies that it may vary in different regions of Europe depending on the species composition and their adaptations. As for the critical limit of ANC (acid neutralisation capacity) to macroinvertebrates it has been suggested an interval from about 20 µeq/l to 50 µeq/l (Raddum & Skjelkvåle 2001). The French B.I.A index began to observe impoverished macroinvertebrate communities at ANC values between 50 and 100 µeq/l. However, severe loss of taxa was only observed below mean values 50µeq/l (Guerold et al 1999).

Turnbull et al (1995) in N-E Scotland observed fewer species and less sensitive species with increasing critical loads. It was, however, concluded that even though critical loads provides a good predictor for biotic status it is not as sensitive a parameter as pH or alkalinity. The acidification indices are most often related to pH intervals as it has been observed that changes in the communities are correlated to this parameter. For the Nordic indices the lower limit for the class of intolerant species has been pH 5.5. In the French I.B.A. index unaffected streams were only obtained

with mean pH values above 6.5. The most significant change in the index values occurred between pH 6.5 and 5.5, a pH interval that has not received much attention in the Nordic indices (Guerold et al 1999).

Multivariate techniques have been used to evaluate and confirm the relation between macroinvertebrates and the water chemistry. The ability use macroinvertebrates to predict pH has been proved to be good by Larsen et al (1996). Hamalainen & Huttunen (1996, 1998) used a WA (weighted average) model and tolerance-limit approaches for inferring stream minimum pH from macroinvertebrates. There was a strong correlation between the macroinvertebrates and the minimum pH using the WA approach. The tolerance-limit approach assigned the streams into correct acidity classes, but the inferred pH was not related to the observed minimum pH. Further, the WA models were able to infer a minimum pH above 5.5, which is not covered by most tolerance-limit approaches meaning unclear.

Sandin & Johnson (2000) assessed the Nordic indices for their ability to predict the acid situation in rivers as defined by the critical load and/or pH. There was a rather high frequency of sites where the biological indices did not reflect chemical status. Particularly important was the tendency of failing to identify acidification even when this was predicted by critical loads and/or pH. Some of this mismatch may be explained by the incomplete understanding of the detoxifying role of humic water and a corresponding incomplete implementation of changing tolerance to acid water in the acidification indices. And of course there would be a risk that some estimates/measurements of chemical parameters used in the assessment were incorrect.

Episodic events

Acidification very often is an episodic event. Most often it is connected to the snowmelt in spring or rain storms during the autumn. The importance of acid episodes was recognised also by Weatherly & Ormerod (1991). Even though there are long standing evidences that episodes of acidification in streams is important biologically there are still uncertainties on their effects on invertebrate communities. As was demonstrated in the southern Swiss Alps, there are big differences between acid sensitive streams with similar low-flow chemistry and contrasting episode chemistry (Lepori et al 2003). A special kind of acidification episode has been observed at the south-western coast of Norway. Heavy storms introduced high concentrations of Na⁺ from marine water to the catchments. An ionic exchange occurred increasing the inorganic monomeric aluminium and H⁺ to high concentrations in the stream water, reducing the pH to low levels (Hindar et al 1995). The frequency of episodes, how long they last as well as the degree of acidity are important for their biological effects in the streams. Knowledge on the nature and frequency of episodes and their biological effects is however scarce.

Humus

It seems to be a change in the tolerance of macroinvertebrates to acid water when moving from clear water to brown water. Some species used in classifying clear water streams in the not acidified class can not be used in the same way in brown water sites. In general data from the Nordic countries indicate a higher tolerance to low pH when increasing the TOC (total organic carbon). It is generally assumed that humus detoxifies the water making metals and in particular aluminium less bio-available. However, little work has been done to clarify this relation on macroinvertebrates. More research is needed on the relation between the concentrations of humus, metals, pH and toxicity. It has been shown experimentally that humus may reduce the toxic effect of low pH to the mayfly *Baetis rhodani* and the crustacean *Gammarus lacustris* (Bækken & Aanes 1990).

Natural acid water

Parts of Europe, in particular the northern countries, have regions with naturally acid water due to organic acids. These may be wrongly regarded as temporarily or permanently acidified by anthropogenic sources and consequently be candidates for liming actions (Warfvinge et al 1995, Korte-

lainen & Suakkonen 1995). A recent publication focused on the structure and function of naturally acid streams (Dangles et al 2004). The taxonomic richness of macroinvertebrates as well as the breakdown rate of leaf litter in natural acid streams was not significantly different from the circum neutral streams. Another data set comparing anthropogenic acidified streams to circum neutral streams showed reduced taxonomic richness and reduced leaf breakdown rates in the acidified streams. The authors claim that the findings have implications for the management of streams in areas where considerable effort is spent on liming without consideration of the fact that many of these have acid water as the natural state.

General toxicity of aluminium and heavy metals

Many macroinvertebrates are physiologically unable to tolerate low pH. However, it may be great differences in their tolerance. Differences in the concentration of Ca, Na, CO₂, Al and other metals may influence the mortality (Havas 1981). It has, however, been difficult to isolate the effects of low pH from associated chemical change. Inorganic Al has been shown to be toxic to fish and to a number of macroinvertebrates and is certainly the most important toxic metal in anthropogenic acidified water. The kind of metal species is important with regard to toxicity. Usually the most toxic forms are the inorganic cationic metal species. In a review of publication dealing with the toxicity of Al to macroinvertebrates Herrmann (2001) concludes that it is problematic from the literature to give general guidelines to when Al is toxic. Although many studies consider problems to occur when inorganic Al is in the range of 0.1 to 0.3 mg/l, many exceptions exist with respect to species, Al fraction and speciation, the presence of humus, pH and exposure time.

There are numerous studies, mostly experimental, also regarding the toxic effects from other metals on macroinvertebrates, not regarding the effects of changing pH (Mance 1987). In general these studies show biological effects over large intervals of concentrations for a variety of heavy metals (Figure 2.9).

These data also suffers from large differences in the methods, water qualities (as pH, Ca and TOC) and species, and they are of limited value in assessing the toxicity in dynamic natural environments.

The concentration of Al as well as other metals (Cu, Cd, Ni, Mn, Pb, Zn) was measured in precipitation and surface water in northern England. In the acid water Al concentrations were at toxic levels for fish (LC₅₀) whereas for some of the other metals the free ion concentrations were one-to-four orders of magnitude lower than reported LC₅₀ values for fish (Lawlor & Tipping 2002). As the concentrations were close to LC₅₀ values (short time mortality effects) it may be expected that sensitive species of both fish and macroinvertebrates will be affected by these metal concentrations. Some few in situ studies seem to give two kinds of effects regarding heavy metals in the macroinvertebrate community: 1. Taxa richness are reduced as sensitive species disappear. Mayflies, in particular species of the genera *Heptagenia* and *Ephemera* seem to be sensitive to metal pollution. 2. The abundance of the remaining taxa may be considerably reduced (Leland et.al 1989, Malmqvist & Hoffsten 1999, Clements et al 2000, Deacon et al. 2001, Hirst et al 2002, Courtney & Clement 2002)

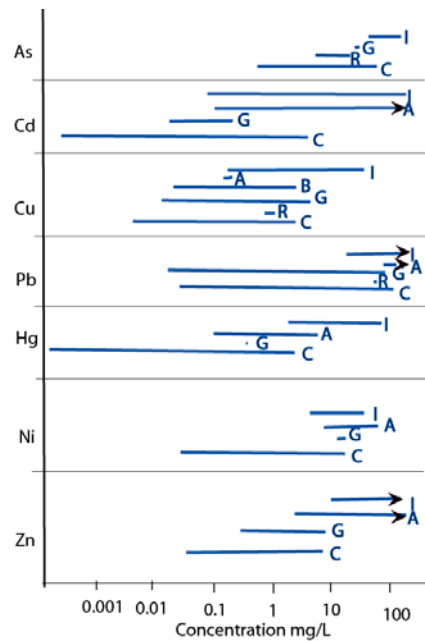


Figure 2.9. The range of observed adverse effect concentrations for freshwater invertebrates (main taxa) from laboratory tests. A: annelids, B: bivalves, C: crustaceans, G: gastropods, I: insects R: rotifers; (after Mance 1987).

2.4.2 Fish

With respect to acidification, fish is generally more sensitive to aluminium than to the pH reduction per se. While Atlantic salmon is unaffected by H⁺ down to pH values lower than 5.4 (Lacroix and Townsend, 1987; Fivelstad et al., 2004), the species is extinct in rivers having average pH values lower than 5.5, and reduced abundance when pH is lower than 5.9, when the water also contains bio available aluminium. Based on such observations, pH alone does not give a satisfactory prediction of fish population status (Kroglund et al., 2002). Understanding the interaction between pH and aluminium, but also between aluminium and fish and aluminium and other water quality constituents is fundamental for the understanding of the mechanisms resulting in population changes. A justification for fish within the WFD is that fish generally raises a large public interest and has a high socio-economic value. Furthermore, acidification effects on fish populations have been documented in all affected areas. The impacted regions differ with respect to several physico-chemical properties, permitting the inclusion of regional factors modifying the most simple dose-response relationships that are presently used.

Acidification has caused a general loss of fish populations in all regions affected by acidification. Salmonids and roach are more sensitive than perch and pike. While acid-sensitive fish species have experienced the greatest losses, even populations of acid-tolerant species are affected in some regions of Europe (Rask and Tuunainen, 1990; Rask et al., 1995; Welsh and Perry, 1997). The number of extinct populations within Norway, Sweden and Finland exceed 10.000. In addition, an even higher number of populations have reduced viability, i.e. show a non-healthy status. Compared to the number of lakes that have lost or reduced fish populations, fish kills are rare. In most lakes and rivers, the effects of acidification are only observed as changes in fish species composition and/or abundance (Rask and Tuunainen, 1990, Hesthagen et al., 1999; Tammi et al., 2003). The fact that fish kills are rare events, despite populations going lost, suggest that density reductions occurs at life stages where kills are not easily observed (e.g. egg and fry), that density reduction is a gradual event (e.g. more related to fish health and susceptibility to diseases, predation etc) more than to acute events. Fish kills have been observed under extreme events and extreme examples of total fish loss are recorded in central and southwest Scotland and north Wales (Harriman and Morrison 1982; Harriman et al. 1987; Hindar et al, 1994). Fish kills (*Salmo salar*) were observed in several

Norwegian rivers in the years between 1910 and 1920, kills that most likely were due to acidification and seasalt episodes (Hindar et al., 2004). Despite massive fish kills, it took more than 40 years for the various affected populations to go extinct. This illustrates that linking dose to response is not necessarily a straight forward process.

Changes to or loss of fish populations are not due to any lack of food. Bottom up relationships have therefore minor importance in acidified systems (Rosseland and Staurnes, 1994; Sparling and Lowe, 1996; Gensemer and Playle, 1999). Fish population health is mainly related to aluminium, where the activity of H^+ will enhance toxicity. Since aluminium is not vital to any aquatic biota, the species have not evolved any defence mechanism directed specially towards this metal. Aluminium exercises its toxic properties by being accumulated onto the fish gill (Rosseland et al., 1994; Sparling and Lowe, 1996; Gensemer and Playle, 1999). Aluminium interacts with the gill of fish affecting both tissue function and properties. The accumulation is directly related to the concentration of *in situ* fractionated LAI (Gensemer and Playle, 1999; Kroglund et al., 2001ab). The relationships are poor when based on water samples where the dynamic nature of aluminium has been stabilized through ageing (Kroglund et al., 2001ab). Aluminium is accumulated onto the mucus layer surrounding the gill, onto cell walls and is over time transported inside the cell. The toxic role and effect depends on concentration, exposure duration and where Al is accumulated. As the speciation and hence toxicity of aluminium is dynamic, aluminium does not fit directly into biotic ligand models as these assume that the metals are present on a free, and stable form (Chapman et al., 2003). The unstable nature of aluminium renders aluminium difficult to use in dose-response models. Aluminium characteristics are more dynamic in rivers due to larger spatial heterogeneity in water quality (water from tributaries enters and mixes with main river water, resulting in local variation in water quality) than in lakes, and more at pH levels around 5.6 or higher than at pH levels lower than 5.2. When acid tributaries enter and mixes with "better" water in the main river, aluminium polymerizes from toxic to non-toxic forms. This time, temperature and pH-related process is less pronounced in lakes having a longer retention time. Aluminium can however be on an unstable form along lake shorelines, when acid surface runoff mixes with lake water having a higher pH. The probability for errors in pH/aluminium relationships are as such related to pH and to water retention time.

Fish mortality is generally related to ionoregulatory and respiratory dysfunction. Before the water quality becomes toxic, the fish demonstrates numerous sub lethal responses (Rosseland and Staurnes 1994, Gensemer and Playle, 1999). While physiological responses resulting in mortality are mainly demonstrated in experiments, sub lethal responses affecting growth, development and recruitment are demonstrated for natural populations (Gensemer and Playle, 1999). Sub lethal responses are only rarely included in population surveys.

No effect concentrations (NEC) are related to species sensitivity and vary according to life stage variation in sensitivity. In addition there is a large strain dependent component in sensitivity for non-anadromous species (Rosseland and Staurnes, 1994; Dalziel et al., 1995). Strain related variance is the cause for some unwanted variance in the empirical dose response models. This cause for variance must be acknowledged, but is not included in any dose-response models at present. pH limits for several inland species are presented in Tammi et al., 2003).

NEC do not vary between strains for Atlantic salmon (Rosseland et al., 2001), but vary grossly between life stages (Rosseland and Staurnes, 1994). Smoltification (a synchronization of physiological and behavioural traits needed to enhance marine survival) is extremely sensitive to all types of metals and micro pollutants. For anadromous species (species migrating between fresh and salt water), fish abundance (measured as number of river returning salmon with respect to smolt production) will be reduced if seawater performance is reduced. This implies that population effects are observed long before the doses are high enough to have any impact measurable as reduced fry density or growth in the freshwater life stages. Metals and micro pollutants affect seawater tolerance by inhibiting the energy transfer enzyme Na-K-ATPase. The activity of this

enzyme can drop substantially after some few hours of exposure (Staurnes, 1993; Staurnes et al 1996; Kroglund and Finstad, 2003). This implies that short-term episodes or exposure to elevated concentration of aluminium downstream acid confluences can affect marine survival despite, relationships that are not picked up in any traditional monitoring programme. ANC, pH and LAI limits for Atlantic salmon are given in Kroglund et al., 2002). Many salmon stocks are affected by episodes. Water quality limits with respect to episodes are given in Kroglund and Rosseland (2004).

ANC is the favoured indicator of acidification pressure for fish, although pH still is frequently used (often due to lack of chemical data). Some problems regarding the use of ANC were addressed in section 2. These confounding factors need to be included into the model to derive indices that can have a broader application than current day models. Many of the fish-response models are based on national data only. An aim within Rebecca would be to organise larger data-sets into a model that could predict changes to ecological status throughout Europe using the same indices. The following knowledge gaps are therefore focused upon:

2.5 Knowledge gaps

It is well known that acidification affects the biota in freshwater including algae, invertebrates and fish. There are a number of gaps in our understanding of relationships between water quality (defined as ANC, pH, aluminium, DOC, base cations etc.), fish status, invertebrate- and periphyton species and communities that need to be elaborated. This is necessary to develop quantitative metrics for biotic elements in an acidified environment and when acid precipitation is reduced. As for metals the knowledge on biological effects is limited for natural dynamic systems. This is particularly relevant when seen in connection to different water types and water qualities. The following gaps are identified:

- The acidification tolerance for more species should be clarified to increase the precision of the metrics used for assessment.
- The effects of H⁺ need to be separated from aluminium.
- Acidification effects to age structure, growth and reproduction need to be separated from natural causes for variation.
- Organic material affects the relationship between toxicants and biotic status, but the relationships are poorly understood. Present day agreed upon ANC limits will not protect fish in water having more than 4 mg DOC.
- How will the relationships between pH, aluminium and cation concentrations (ANC) change with reduced acid deposition.
- How will reduced retention of NO_x affect the ANC/aluminium relationship?
- Will nitrogen, temperature increase and sea salt episodes confound recovery?
- Episodes can have biological importance that is not easily detected. How do repeated episodes with changing water quality affect the biota?
- What are the effects of heavy metals in dynamic natural systems with changing water quality.
- How are the interactions between periphyton, macroinvertebrates and fish changed in acidified systems. What are the toxic effects and what is a secondary biological effect?

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3 Pressures from organic matter

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3.1 Introduction

The objective of this chapter on organic pollution of rivers is to review the existing knowledge on the relationships between the content of organic matter in rivers and WFD relevant biological quality elements of the rivers. The focus is on the impacts of organic matter discharged with wastewater, because this type of input of organic matter is one of the main causes of pollution in rivers.

The reviewed literature includes scientific papers and reports from institutions responsible for environmental monitoring and management, and research. The topic has been studied for more than a century and treated in thousands of publications, but relatively few of them report operational relations between biological quality elements and the quality and concentration of organic matter in the river.

3.2 Role of organic matter in rivers

Organic matter is a natural and necessary component of all ecosystems as food for the heterotrophic organisms. The sources of organic matter to rivers are allochthonous inputs from the river catchment, especially from the riparian areas, and autochthonous inputs, i.e. the production of organic matter through photosynthesis by the river primary producers.

Natural and unimpacted river ecosystems differ partly because of natural differences in quality and quantity of organic matter input and different autochthonous production in various river types. The differences in degradability and nutritional value lead to a favouring of different types of heterotrophic organisms in different streams (see e.g. Servais et al. 1987, Baldy et al. 1995).

Examples of such different types of organic matter are humic substances (humic and fulvic acids) from riparian forests and moors, refractory particulate organic matter (e.g. from some types of leaves from riparian vegetation), protein rich leaves from riparian vegetation, stream macrophytes, attached algae and phytoplankton produced in upstream lakes or in big rivers). Both the nutritive values and the seasonality of the inputs of these different types of organic matter differ from river to river and the biota of the rivers may change accordingly.

Organic substances are thermodynamically unstable in natural waters. Carbon forms the direct linkage in interactions between the inorganic components and living organisms. The carbon cycle is interrelated with all the other element cycles. Fig. 3.1 schematically presents interactions between the production and respiration processes including organic waste introduction.

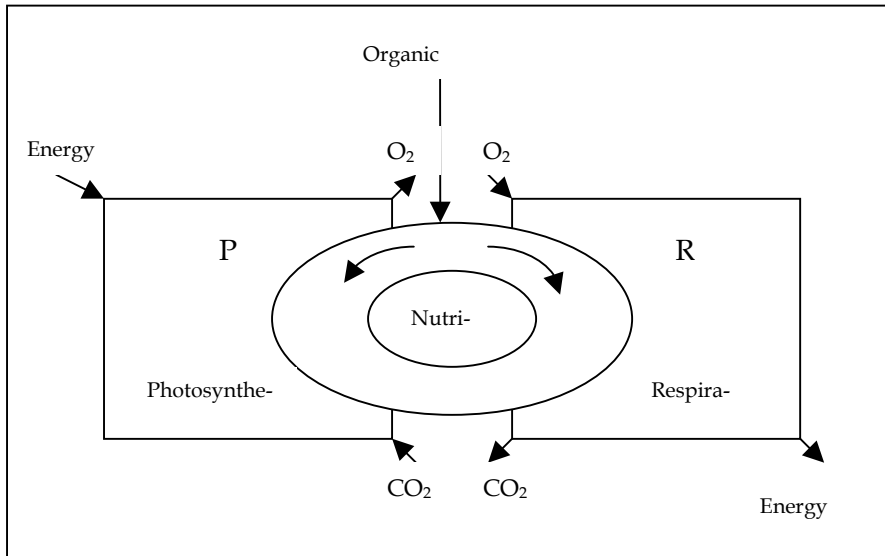


Figure 3.1 Schematic cycle of the organic substances in the aquatic environment (Based on Stumm & Morgan, 1981)

Photosynthetic organisms (mostly algae and plants) synthesize biomass (P = rate of photosynthesis) which is consumed by animals (consumers, herbivores) and biodegraded by bacteria and fungi (decomposers) through respiration (R = rate of respiration). When an excess of organic wastewaters are discharged into the natural waters the ecological balance is disturbed.

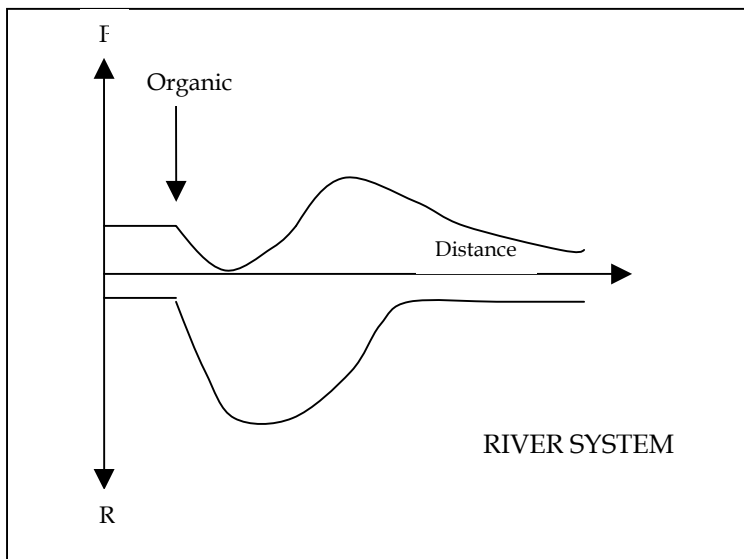


Figure 3.2 Longitudinal distributions of photosynthesis and respiration in a river with a large discharge of organic matter, e.g. wastewater into a river.

Rates of autotrophic and heterotrophic processes may show different longitudinal trends along river systems. Organic pollution due to wastewater into river system greatly affects the natural longitudinal trends of respiration and photosynthesis rates. Respiration is enhanced downstream of an input of organic waste. On the other hand photosynthesis becomes more important far downstream where organic wastes are partially mineralized and suspended solids have settled out (see Fig 3.2). Organic matter in natural waters includes a great variety of organic compounds. Both natural processes and anthropogenic activities may cause changes in the content of organic compounds in the aquatic environment. Anthropogenic changes in riparian vegetation may affect the quantity and quality of the allochthonous inputs of organic matter and affect the river ecosystem favouring a different species selection. Conversely, the inputs of degradable organic matter with

wastewater discharges into rivers have led to dramatic deteriorations of river ecosystems. This is caused by increased availability of food of a different quality for the heterotrophic organisms, by covering the riverbed habitats with sludge and in the extreme cases by deoxygenating the river water.

The significance of organic pollution of European rivers is stressed by the European Environment Agency (EEA) in the Dobris Report and in the EEA report on European Rivers and Lakes (EEA 1991, EEA 1994). EEA also confirms (EEA 2003a, 2003b) that the wastewater discharges of degradable organic matter is reduced significantly in Europe during the 1990ies.

The major pollution sources of organic matter are domestic wastewater, industrial wastewater from food, pulp and paper processing and agricultural discharges from livestock production and fish farming. Besides, occasional discharges such as storm-water run-off from urban areas can be an important pollution source.

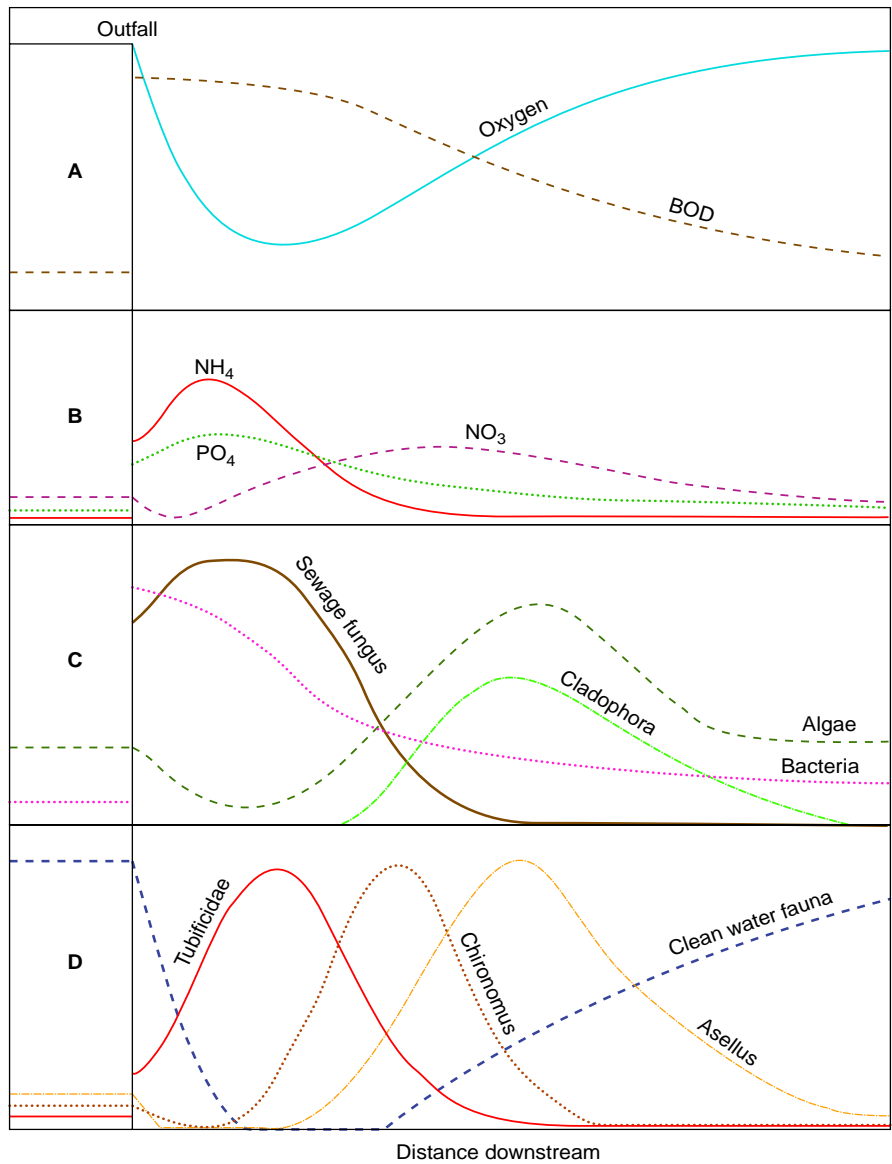
The specific impacts mainly depend on two factors: the resulting increase in the concentration of organic matter in the receiving water and the quality of the organic matter discharged. Furthermore, the ecological damage depends on the hydromorphological characteristics of the river and on the other pressures on the river ecosystem.

Biological wastewater treatment has dramatically reduced the wastewater pollution impact in many European rivers. The organic loading from a wastewater discharge is often reduced by a factor of around 10 when an effective biological treatment is established, but at the same time the quality of the organic matter discharged has changed and therefore also the type of ecosystem impact. In Fig. 2.3, typical changes in water quality downstream of wastewater discharges are shown.

The concentration of organic matter in natural water bodies results from an interplay of net productivity, exudation of organic substances by phytoplankton, and import and export (inflow, outflow, dissolution, sedimentation, etc.) of organic matter.

Reference figures for organic carbon export rates for large temperate rivers are available from the literature (Thurman, 1985). Values of 2.2 ± 0.3 and 1.2 ± 0.4 tonnes $\text{km}^{-2} \text{ year}^{-1}$ have been estimated for dissolved (DOC) and particulate (POC) organic carbon, respectively (Lugo, 1983). In the largest Italian river, the Po, Pettine et al. (1998) found export rates of 1.7 and 1.9 tonnes $\text{km}^{-2} \text{ year}^{-1}$ for DOC and POC. It is also possible to make a reasonable evaluation for the contribution of phytoplankton biomass to the total POC export by rivers. To this end, conversion factors have been proposed to transform chlorophyll data in the corresponding algal POC. Admiraal et al. (1992) applied a factor of 25 to Chl a in the lower river Rhine. Relexans et al. (1988) used a factor of 30 to convert total pigments in the Loire River. A factor of 35 has been used for converting Chl a data in the Loire and other French rivers (Vire, Oise and Garonne) based on the SCOR-UNESCO equations (Relexans & Etcheber, 1982, Dessery et al. 1984). In a more recent investigation in the lower Po River, Pettine et al. (1998) used factors of 20 and 30 for total pigments and Chl a, respectively. By using the above conversion factors, it was estimated that algal POC accounted for about 50 % of total POC in the lower Loire (Billen et al. 1986), for 15-65 % of total particulate carbon (PC) in the lower Rhine (Admiraal et al. 1992). Pettine et al. (1998) calculated an annual average contribution of algal POC of about 15 % to total POC with a peak of about 25 in the most productive periods from April to August.

Furthermore, fulvic and humic acid concentrations in river waters are highly dependent on physico-geographical conditions and are usually in the range of tens to hundreds of micrograms of carbon per litre. In natural conditions fulvic and humic acids can comprise up to 80 per cent of the DOC (Chapman, 1996).



Modified from Hynes (1960)

Figure 3.3 Typical changes in dissolved oxygen, nitrogen and carbon dioxide downstream of a wastewater input to a river (Modified from Hynes, 1960). This pattern is typical for bigger rivers while in small streams the pattern is more variable. The benthic fauna can be more heavily impacted closer to the discharge point than the oxygen minimum due to deposits of organic matter or bacterial growths on the riverbed.

3.3 Characteristics of organic matter in wastewater and in rivers

Rivers consist of water, materials and energy moving from headwaters to mouth. They represent important dynamic components of landscapes since their characteristics are intimately tied to the nature of their watershed (Tockner et al., 2003). Natural acquisition of chemical substances occur by dissolution and by chemical reactions with solids, liquids and gases with which rivers come into contact (Meybeck, 1982). They thus play a major role in global biogeochemical cycles by transporting elements from terrestrial environments to downstream lakes or seas. The quantity and quality of elements exported from watersheds are greatly influenced by physical, chemical and biological processes occurring along river corridors (including riparian zones) (Lepistö et al.,

2001; Stanley & Doyle, 2002; Zhiliang et al., 2003). A number of elements are often present well in excess of any biological demands within the river and may pass unaffected the river corridor. Other elements such as carbon, nitrogen and phosphorus may be present in concentrations sufficiently low to limit primary production of some types of plants, and river corridors may exhibit a large capacity to transform and retain these nutrients (Haycock et al., 1993; Humborg et al., 2000; Hein et al., 2003). Rivers experience substantial annual and interannual variations in their discharge, sediment load and concentration of the various chemical compounds transported. Organic matter ranges from dissolved molecules to particulate aggregates and can be present as living and dead material.

In rivers, natural levels of organic matter are related to in-river production and alloctonous input from river watershed. Human activities alter organic matter natural levels in rivers mainly by (i) modifications of river watershed (urban and agricultural land use) and (ii) direct organic matter releases into river water (domestic and industrial wastewater, livestock and fish farming,) (Polls et al. 1980; Harremoes 1982; Hvitved-Jacobsen 1982). Impacts on chemical river water quality from organic discharges have been described and modelled during the last hundred years (Streeter and Phelps 1925, Hynes 1960). A key research field has been to follow the fate of the organic matter discharged into the river and especially to develop models for the resulting impacts on the concentration of dissolved oxygen in the river water downstream of the discharge point. Decomposition and dilution processes reduce the concentration of the organic matter further downstream of the discharge point and, as the distance from sewage effluent increases, organic matter concentration may possibly reach the same values detected immediately upstream of the discharge point. The first models (Streeter and Phelps 1925) were based on oxygen consumption described by a first order mineralisation of the organic matter and a re-aeration proportional to the oxygen deficit. The model implementation during the last decades have led to comprehensive and complicated models describing the dynamics of the oxygen concentrations in time and space in a river. Examples of such models are the river models established by the IWA (International Water Association) Task Group on River Water Quality Modelling (Shanahan et al. 2001, Reichert et al. 2001, Vanrolleghem et al. 2001) and the modelling framework for simulation of river and stream water quality available at US EPA homepage (Chapra & Pelletier, 2003). These recent river water quality models give a dynamic description of chemical variables such as DO, BOD and ammonium.

Domestic and industrial wastewater discharges are the most important sources of organic pollution of rivers (Table 3.1). If not properly treated, such discharges greatly affect the quantity and quality of organic matter in rivers. Since organic matter represents a trophic resource for several organisms, these changes will affect directly river biota.

Easily degradable organic matter in wastewater usually has a more pronounced and direct impact on the river biota compared to more slowly degradable organic matter. The generally used metric for this easily degradable organic matter is the Biochemical Oxygen Demand (BOD), usually measured oxygen consumption (respiration) during a period of 5 days. Other commonly used parameter to characterize the concentration of organic matter are the COD (Chemical Oxygen Demand) that roughly corresponds to the total amount of organic matter if the oxidation is made with dichromate. Moreover, the contents of organic matter in the water is often measured by total organic carbon analyses (TOC) or separated in Particulate Organic Carbon (POC) and Dissolved Organic Carbon (DOC).

Table 3.1. Concentration (mg/l) of various compounds in wastewaters.

Parameter	Food processing (IRSA)	Urban Runoff (Gray, 1989)	Paper industry (IRSA)	Domestic (Henze et al., 1995)
Suspended Solids	4.2-13000	2-11300	<5-70	120-450

BOD₅	5.2-25000	1-700	90-1150	100-350
COD	2-22400	5-3100	235-2000	210-740
N total	0.05-1313	1.1-6.2	3.2-51.2	20-80
N organic	0.3-510	0.1-16	-	8-30
N-NH₄	0.1-66	0.1-14	-	12-50
Fats	1.2-3000	-	-	30-100
Chlorides	48-5690	2-25000	55-235	200-500*
P total	0.02-520	0.09-4.4	0.1-0.3	4-23
Potassium	8-418	-	-	-
Sodium	60-807	-	-	-
Calcium	57-112	-	-	-
Magnesium	25-50	-	-	40-150
Proteins	210-560	-	-	7-25
Carbohydrates	100-1500	-	-	10-40

*with 100 g/m³ in the water supply

3.3.1 General parameters describing organic matter

As the decomposition of organic matter requires the consumption of oxygen, severe organic pollution leads to deoxygenation of the river water. Therefore dissolved oxygen (DO, mg O₂/l) is probably one of the most important tracers of organic pollution, but its use in traditional monitoring programmes is problematic because of the diurnal variations especially in small streams. The dominance of photosynthesis during the light hours often leads to supersaturating with oxygen during the day, but the high respiration rates during the night may lead to critically low levels of dissolved oxygen. Moreover, bacteria oxidise the ammonium to nitrate contributing to the depletion of oxygen downstream of a sewage effluent without nitrification, and this effect is usually evaluated along with the effects of organic pollution. Downstream of the organic pollution input, the mineralization of organic matter leads to an increase in the concentration of CO₂ and a consequent decrease in pH in the water, along with the oxygen consumption.

The Biological Oxygen Demand (BOD), the Chemical Oxygen Demand (COD) and the Total Oxygen Demand (TOD), the units of which are mg O₂/l, are very important general parameters to measure organic pollution. The BOD-analysis is used to measure the oxygen demand of the microorganisms for the oxidation of organic matter and ammonia after a predetermined period (5, 7 or 25 days). The addition of thiourea to the sample prevents the oxidation of ammonia, making it possible to measure BOD due to organic matter.

Different chemical oxidants can be used to determine the COD. Potassium permanganate is used to determine the chemical oxygen demand (COD_p). However, it gives an incomplete oxidation of organic matter and this analysis should therefore only be used to have a rough measurement of organic matter and to estimate the necessary dilutions in the BOD-analysis. Potassium dichromate is a more thorough oxidant, but the oxidation includes different inorganic materials (NO₂⁻, S₂⁻, S₂O₃²⁻, Fe²⁺, SO₃²⁻). Ammonia, which is released by the oxidation of organic nitrogen, is not oxidized under these conditions. The measured COD-value gives a good picture of the total content of organic matter in the river as the above mentioned inorganic materials normally are not present in significant concentrations in wastewaters (Henze et al., 1995) (Table 3.2). The oxidation at high temperature in the presence of a suitable catalyst gives the total chemical oxygen demand (TOD). Organic compounds not oxidized by the COD-analysis are oxidized by using the TOD analysis. Besides, ammonium is oxidized too. TOD-values are therefore somewhat higher than COD values and can be used for the calculation of the total organic carbon (TOC) in the water (see below).

Table 3.2 Analysis values for the different measuring methods for two different types of wastewater (Henze et al., 1995).

Element	Name	Raw Wastewater	Biologically treated wastewater without
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		(g O ₂ /m ³)	nitrification (g O ₂ /m ³)
C _{BOD}	5-day biochemical oxygen demand	280	25
C _{BOD7}	7-day biochemical oxygen demand	320	30
C _{BOD∞}	Total biochemical oxygen demand	400	35
C _{COD}	Chemical oxygen demand*	600	100
S _{S,COD}	Easily biodegradable matter, measured as COD	60	5
X _{S,COD}	Slowly biodegradable matter	200	10

An extended biological treatment is likely to produce a treated wastewater with a BOD₅ of about 5 mg/l which is far below the limits of 20 mg BOD/l and 75 mg COD/l required by the Urban Waste Water Directive (91/271/EEC).

The use of the concentration of degradable organic matter to classify the pollution level is not of new date. Hynes (1960) refers that in 1898 the British Royal Commission on Sewage Disposal investigated a large number of British rivers and found that the 5 day BOD (BOD₅) of the river water gave a very fair measure of their cleanliness. The commission found that rivers could be classified as in table 3.3.

Table 3.3 river classification based on BOD₅

River class	BOD ₅ (mg O ₂ /l)
Very clean	1
Clean	2
Fairly clean	3
Doubtful	5
Bad	10

More recently, the European Environment Agency (EEA 1991, EEA 1994) has compiled monitoring results from European rivers and found average BOD₅ levels for near pristine rivers of about 1.6 mg/l. EEA concluded that river reaches slightly affected by human activities generally have a BOD below 2 mg/l whereas a BOD exceeding 5 mg/l generally indicates pollution. This is very close to the conclusions made by the British Royal Commission in 1898.

3.3.2 Total organic carbon

In rivers, organic matter is present with a continuous size spectrum, from dissolved small molecules, macromolecules and aggregates to organisms. With an arbitrary definition, the fraction retained on a filter with a pore size of 0.5 to 1.0 µm is defined as particulate matter. Particulate organic matter (POM) can originate from autochthonous production of phytoplankton and from allochthonous sources from soils, riparian vegetation and wastewater. It can also be produced in situ from dissolved organic matter by physico-chemical and biological processes. It has been suggested that rivers carry 10 times more dissolved organic matter (DOM) than POM (Meybeck, 1982). However, Ittekkot & Laane (1991) showed that strong variations in DOC/POC ratios are strongly dependent on the concentrations of suspended matter. In their study rivers carried 10 times more DOM than POM only for concentrations of suspended matter smaller than 15 mg/L. Low DOC/POC ratio values are characteristic for highland rivers and high values for lowland rivers, suggesting an inverse relationship between the suspended matter and the DOC/POC ratios (Meybeck, 1982).

DOC and POC may be combined in a single measurement of total organic carbon (TOC) on unfiltered water. For more than 100 rivers, mainly located in lowland areas, TOC was found to be generally between 2 and 30 mg/l with a median value of 10 mg/l (Meybeck, 1982). Relations between COD and TOC values for rivers have also been established with reference to specific rivers: for

example, Pettine et al. (1985) have fitted COD and TOC data in the Po River to the following linear equation (SD = ± 0.9 mg/l):

$$\text{TOC (mg C/l)} = 0.51 + 0.28 \text{ COD (mg O}_2\text{/l)}$$

However the extrapolation of these results to other rivers should be verified, because this relationship is strongly influenced by the average oxidation state of organic carbon in the river, which is given by $[4(\text{TOC}-\text{COD})]/\text{TOC}$, where TOC and COD are expressed in mol/l of carbon and oxygen, respectively.

Degradation of organic matter by heterotrophic bacteria is one of the major processes controlling the oxygen level of aquatic ecosystems and thus their quality. However, parameters traditionally used in models describing organic matter degradation, such as biochemical oxygen demand (BOD) and chemical oxygen demand (COD) and Total Organic Carbon (TOC), are often insufficient to correctly predict the impact of waste water release on aquatic ecosystems. Ecological models able to better describing riverine ecological processes and developing efficient rehabilitation scenarios would require, as input data, a detailed characterization of the organic matter discharged by treatment plant effluents (biodegradable and refractory parts of dissolved and particulate organic carbon) (Servais et al., 1999).

Dissolved organic matter is operationally defined as all the organic material that passes through a filter of a given size, which can range from 0.2 µm (polycarbonate membranes) to 0.7 µm (GF/F Whatman glass fiber filters). This definition comes from the fact that filtration is the most widely used procedure to separate the particulate from the dissolved matter. The availability of dissolved organic matter to heterotrophic bacteria probably depends on the qualitative characteristics of the organic substrate (biochemical composition and molecular size being the most important), but is also influenced by the availability of inorganic nutrients. Therefore improving the qualitative and quantitative characterization of the dissolved organic matter pool is a necessary step towards a better understanding of the microbial food web and the evaluation of modes of organic carbon transfer among the dissolved, colloidal and particulate pools. The DOM consists largely of humic substances and more labile compounds from the major biochemically important compound classes, such as carbohydrates, steroids, alcohols, amino acids, hydrocarbons and fatty acids. Recently, ultrafiltration techniques have been applied to natural samples to separate and concentrate dissolved organic matter into different molecular size classes. These techniques have stimulated investigations on the chemical composition and bioreactivity of organic matter in the different classes. There are now many experimental results that suggest that colloidal organic carbon (COC) may account for 40-80% of DOC in rivers and findings by Benner et al. (1992) have highlighted that high molecular mass polysaccharides (1nm-0.2µm) made up an important part of colloidal matter.

Servais et al. (1999) analyzed treated and untreated wastewater of various treatment plants in the Paris suburbs. They included high load activated sludge (HLAS), HLAS followed by nitrification, HLAS with an aerobic and an anaerobic stage, HLAS in aerobic conditions and HLAS in aerobic conditions followed by a tertiary nitrification treatment. Their results allowed defining specific loads of organic matter, nutrients and microorganisms. These loads were expressed in g inhabitant⁻¹ d⁻¹. For raw water, the specific loads of TOC ranged between 26.4 and 28.3 g C inh⁻¹ d⁻¹ with particulate organic matter constituting the main part (70±76%) and the biodegradable fraction accounting for 60-75% of TOC. Concerning microorganisms, the average specific load of total bacteria was around 2 g C of biomass inh⁻¹ d⁻¹; the nitrifying biomass represented 0.3-2.5% of the total bacterial biomass. Depending on the type of treatment, the specific load of TOC in treated water ranged between 3 and 10.8 g C inh⁻¹ d⁻¹; it corresponded to removal percentages in the 59-89% range. In addition, these researchers showed interesting relationships between various variables. A significant correlation was observed between the biodegradable fraction of TOC (BTOC) and BOD with an average BTOC/BOD of 0.35. The biodegradable fraction of POC (BPOC) represented 68%

of BTOC in raw water and 43% in treated water. The total biomass of bacteria constituted 8% of the BTOC. These relationships can be used to roughly approximate the composition of the organic matter discharged in the river in terms of BTOC, BDOC, BPOC and bacterial biomass when the BOD of an effluent of urban wastewater (raw or treated) is known. These parameters are essential for modelling the impact of waste water discharge in a river according to ecological models developed by Billen et al. (1993), Even & Poulin (1993) and Even et al. (1998).

3.3.3 Major specific organic components

The impact of the organic matter discharged in the river on its water quality is a function of the sewage effluent composition. Domestic, municipal and industrial wastewaters may differ greatly not only in terms of concentrations but also of the quality of organic matter. Thousands of organic components are contained in wastewaters. In medium strength sewage 75 % of the suspended solids and 40 % of the filterable solids fractions are organic. In settled sewage 50 % of the organic carbon and 35-50 % of organic nitrogen is in solution (Tab. 3.4). Three quarters of the organic carbon are constituted by molecules belonging to the major organic groups: carbohydrates, fats, proteins, amino acids, and volatile acids. The reminder comprises other organic molecules such as hormones, vitamins, surfactant, antibiotics, hormonal contraceptives, purines, pesticides, hydrocarbons and pigments (Gray, 1989). Toxic compounds, even if present in small concentrations, may have significant impacts on river system. Three major biodegradable fractions are included in the organic matter pool and distributed among soluble, colloidal and particulate phases (Henze et al., 1994). The *directly degradable* fraction (mainly acetic acid) is in the soluble form. The *easily degradable* fraction (mainly lower alcohols, lower amino acids and simple carbohydrates) is mainly present in the colloidal matter. The *slowly degradable* fraction is in the particulate suspended matter. In each of these forms, in particular in the suspended organic matter, inert organic matter can be present.

Table 3.4 Organic constituents of domestic wastewater (from Painter, 1983 Cit. in Gray, 1989).

Constituent	In solution		In suspension	
	Concentration mg/l	Proportion as C of total C in solution %	Concentration mg/l	Proportion as C of total C in suspension %
Fats	-		140	50
Carbohydrates	70	31.3	34	6.4
Free and bound amino acids	18	10.7	42	10
Volatile acids	25	11.3		
Non-volatile acids	34	15.2	12.5	2.3
Detergents (ABS)	17	11.2	5.9	1.8
Uric acid	1	0.5	-	-
Creatine	6	3.9	-	-
Amino sugars	-	-	1.7	0.3
Amides	-	-	2.7	0.6
Organic carbon by direct analyses	90	100	211	100

3.4 Biological indicators of organic impact

The enrichment of OM affects flora and fauna living in the river mainly because the increased availability of food of a different quality changes the competitiveness of the different species in the river and because of a deoxygenation caused by increased heterotrophic activities. Further, the habitat of the riverbed can be changed into a coverage of a bacterial film, and the oxygen status in the sediment can be deteriorated even without significant changes in DO in the river water. Severe deoxygenation may lead to a decrease of community diversity with only a few dominant species able to live under low oxygen conditions. Ammonium discharged or released by decomposition processes may be converted, at high pH and at high temperatures, to ammonia that in turn is extremely toxic to biota. Reduced oxygen concentration is one of the effects of organic matter enrichment since river biota can be impacted by organic matter even if the level of dissolved oxygen is unaffected. Moreover the Water Framework Directive focuses on the impacts of pressures on river biota which in turn could be used to assess river quality. In Europe there is a long tradition for the use of biotic indices to characterize river quality, but only a few studies focus on the quantitative relations between biological indicators and pollutants concentrations.

Five key biological elements have been explicitly mentioned in the WFD 2000/60 (in particular in the Guidance Document n.º7 Monitoring under the WFD) to assess impacts of pressures on river quality: benthic invertebrates, macrophytes, benthic algae fish and phytoplankton. Among these five key biological elements, benthic algae and benthic invertebrates are considered priority indicators for the assessment of the impacts of organic pressures on river quality.

- Indices based on the composition and abundance of benthic invertebrates and the presence of sensitive taxa are mainly developed to detect organic pollution.
- Indices based on the composition, abundance of macrophytes and the presence of sensitive taxa are mainly used to detect eutrophication and river hydromorphology (including hydropower effects).
- The composition, abundance and the presence of sensitive taxa of benthic algae are

mainly used as an indicator of productivity but they can be used to detect eutrophication, acidification and river hydromorphology.

- Composition and abundance, sensitive species diversity and age structure of fish community can be used to detect habitat and morphological changes, acidification and eutrophication.
- Composition, abundance and planktonic blooms, and presence of sensitive taxa of phytoplankton are used as indicator of productivity and eutrophication.

There are two currently favored branches of stream assessment (Milner & Oswood, 2000):

- One approach considers multimetric indices, whose goal is to incorporate in a single value different components of the biological community that are sensitive to a broad range of human activities (e.g., Barbour et al., 1999). The multimetric approach has been adopted for the ecological assessment in European rivers (see, e.g., Hering et al., 2004; Dahl et al., 2004; Ofenböck et al., 2004). Multimetric systems, based on a set of variables or metrics representing community structure (taxa richness, relative abundance, dominance), pollution tolerance, functional feeding groups, habitat preferences, and life history strategies, offer robust and sensitive insights into how an assemblage responds to natural and anthropogenic stressors (Klemm et al., 2003). Multimetric indices have been developed for macroinvertebrates, fish and periphyton (Miller et al., 1988; Kerans & Karr, 1994; De Shon, 1995; Hill et al., 2000), and riparian birds (Bryce et al., 2002). Each index should be limited to cover only organisms within one group of key biological elements (phytoplankton, periphyton, macrophytes, invertebrates, fish).
- The other approach relies on multivariate statistical methods to uncover patterns in taxonomic composition. Examples of this second method include, multivariate systems like BMWP (Armitage et al., 1983), BEAST (Reynoldson et al., 2000) and PERLA (Zahrádková et al., 2000).

3.4.1 The Saprobic System

The Saprobien System, which focuses on organic pollution and the associated decrease in dissolved oxygen, was first developed by Kolkwitz & Marsson (1908) and subsequently changed and improved (Rolauuffs et al., 2003). According to the Saprobic System, waterbodies are characterised by the degree of heterotrophy (ratio between heterotrophic and autotrophic processes) with the resulting trophic scale including the groups reported in table 4.1:

Table 3.1 Saprobic system trophic groups.

oligosaprobic	Practically unpolluted	(I)
β -mesosaprobic	Rather slightly polluted	(II)
α -mesosaprobic	Rather heavily polluted	(III)
Polysaprobic	Heavily polluted	(IV)

A determination of the saprobic value is based on the sampling and the identification of species of fauna and flora and a comparison with the Saprobic characteristics established for each species (see e.g. Liebmann (1951) and Sládeček (1973)). The Saprobic System is widely used in Central Europe. Roman figures are often used to indicate the saprobic level. No single species is representative of a single saprobic zone. Rather its distribution will follow a normal curve that stretches

over several zones. The shape and area of this distribution curve defines the *saprobic valency* of the species and the position of the apex is its saprobic value. Various lists of saprobic values have been published for a very large part of the European river biota. Sládeček (1973) compared saprobic indexes and the mean BOD₅ and dissolved oxygen to the upper margin of saprobic classes. In a statistical analysis of 1083 data-sets on saprobity and ammonia content in Slovakian rivers, Rothschein (1986) determined a non-linear relation between the ammonium content and the saprobic class. In the table 4.2 NH₄⁺ values for the upper limits of the saprobic classes are given, derived from data in Slovakian rivers. The NH₄⁺ values represent 90 %-iles.

Table 3.2 Saprobic classes related with BOD₅ (mg/l) DO (mg/l) NH₄⁺ (mg/l) values

Saprobic classes	Saprobic index	BOD ₅ (mg/l)	DO (mg/l)	NH ₄ ⁺ (mg/l)
Xenosaprobity	< 0,5	< 1	> 8	< 0.5
Oligosaprobity	< 1,5	< 2,5	> 6	< 0.9
β-mesosaprobity	< 2,5	< 5	> 4	< 1.7
α-mesosaprobity	< 3,5	< 10	> 2	< 3.3
Polysaprobity	< 4,5	< 50	> 0,1	< 5.9

3.4.2 Phytobenthos

Attached filamentous green algae such as *Cladophora* are a frequent nuisance in shallow rivers polluted by wastewater. Usually the occurrence is mainly linked to the level of inorganic nutrients in the river water (Whitton, 1970), but often enhanced growth is not seen in rivers with elevated nitrate and phosphate concentrations if otherwise unpolluted. Other factors such as organic compounds can be important. Gerloff & Fitzgerald (1976) found that the growth of *Cladophora* was stimulated by the addition of vitamin B1 and B12 and by organic matter from sewage or from dried algae.

Several studies have used periphyton as an indicator of water quality (Collins & Weber, 1978; Leland & Carter, 1984; Ramelow et al., 1987; Biggs, 1989; Mc-Cormick et al., 1996; Kawecka et al., 1996). This algal community, which remains in one place for most of its life cycle, has been accepted as valuable ecological tool in the assessment of water quality. The advantage of using the entire periphyton community over a single species is that species with different responses to contaminants could develop in the community; periphyton community composition gives therefore a more integrated information than an isolated species about water bodies quality. The sensitivity or tolerance of algae to eutrophication and other forms of pollution has led to the development of many indicator systems and indices to assess water quality in rivers (Giorgi & Malacalza, 2002). In particular, methods have been developed for the biological monitoring of European rivers using algae (Whitton et al., 1991; Whitton & Rott, 1996), and several diatom-based indices have been adapted to estimate river water quality (Descy, 1979; Sládeček, 1986; Leclercq & Maquet, 1987; Coste & Aypassorho, 1991; Descy & Coste, 1991; Schiefele & Schreiner, 1991; Prygiel & Coste, 1993; Eloranta, 1995; Kelly & Whitton, 1995; Kelly et al., 1995).

Benthic diatoms are widely used as water quality indicators. In particular, diatom species composition is used to evaluate water quality in terms of organic pollution (Fabri & Leclercq, 1984; Sládeček, 1986; Descy & Coste, 1991), eutrophication (Kelly et al., 1995; Sabater et al., 1996) or acidification (e.g. Steinberg & Putz, 1991; Ter Braak & van Dam, 1989). The short generation time makes diatoms able to respond rapidly to environmental changes (Stevenson & Pan, 1999) and a substantial number of 'diatom indices' have been developed for estimation of water quality in various geographic areas (Potapova et al., 2004). Despite their apparent diversity, most of the 'diatom indices', e.g., the 'Trophic Diatom Indices' of Hofmann (1994) and Kelly (1998), the saprobic index of Sládeček (1973), the 'Pollution Index' of Descy (1979), and the 'Generic Diatom Index' (Coste & Aypassorho, 1991), are in essence numerical models that use species indicator values to predict water quality parameters (Potapova et al., 2004). A first step to develop diatoms based monitoring

tools for rivers is to define diatom taxa ecology. Several authors have determined the ecology of worldwide abundant diatom taxa in correlation with defined chemical, physical and saprobiological parameters (Lange-Bertalot, 1979; Denys, 1991a, b; van Dam et al. 1994; Hofmann, 1994; Rott et al., 1997). Occurrence, species composition and biomass are usually related to the level of inorganic nutrients. Organic pollution also affects the occurrence of benthic diatoms, and for many species saprobic values can be found in the literature (Liebmann, 1951). Fjordingstad (1964) describes a system for the estimation of the pollution levels in streams based on the communities of benthic phyto-microorganisms and identified the relations between saprobic zones and approximate river BOD (Tab. 4.3)

Table 3.3 relations between saprobic zones and approximate river BOD

Saprobic zone	BOD ₅ average (mg/l)	BOD ₅ range (mg/l)
Poly-saprobic	30	15-60
α-mesosaprobic	10	4-30
β-mesosaprobic	5	2-15
γ-mesosaprobic	3	2-6
Oligo-saprobic	2	1-4

Eichenberger & Wuhrmann (1966) studied the effects of addition of sewage to experimental channels in Switzerland and found significant impacts on autotrophic and heterotrophic microbenthos with organic enrichments corresponding to a few mg BOD/l. Both organic matter and nutrients were considered to be responsible for the impacts. Although organic pollution is usually accompanied by eutrophy in the rivers, and could not be clearly separated, the dominance of some taxa may indicate the type of water quality (Hofmann, 1994, 1996). A high abundance of taxa such as *Achnanthe minutissima*, which is highly sensitive towards organic pollution (Steinberg & Schiefele, 1988), or *Amphora pediculus*, which is a eutrophic species, but sensitive to organic pollution (Steinberg & Schiefele, 1988), indicated improvement in quality level among several streams and rivers in southern Poland (Kwandrans et al., 1998). Recently, the use of algae for monitoring rivers has been adopted by some governmental water quality monitoring programmes and environmental impact studies (Guash et al., 1998; Prygiel & Coste, 1993; Whitton et al., 1991; Whitton & Rott, 1996). However, the accuracy of diatom indices for water quality assessments across Europe (Prygiel et al., 1999) is widely limited since it is not possible to clearly distinguish the variations in diatom assemblages caused by natural fluctuations (ecoregional factors) or caused by human-related disturbance.

Phytobenthos indices discussed:

- Trophic Diatom Indices
- Saprobic Index
- Pollution Index
- Generic Diatom Index

3.4.3 *Sphaerotilus natans*

One of the most well known indicators of pollution of streams with easily degradable organic matter is probably “sewage fungus”. This is macroscopic colonies of heterotrophic microorganisms living upon the organic matter discharged into the stream and covering the stream bottom and other substrates. The sheath bacterium *Sphaerotilus natans* is an important constituent, but many other bacteria, fungi and animals contribute to the formation of “sewage fungus” (Hynes 1960). The colonies can have lengths of several tens of centimetres and form a cm thick carpet covering the stream bottom. With readily available organic matter (dissolved sugars, fatty acids and

amino acids) and especially in cold water “sewage fungus” can develop even at low concentrations of organic matter (a few mg BOD/l).

3.4.4 Macroinvertebrates

Carbon, energy, and inorganic nutrients budgets of many streams and rivers are dominated by processes associated with large pools of detritus organic matter both in the dissolved and particulate forms (Webster & Meyer, 1997); heterotrophic microbes, mainly fungi and bacteria, mediate these processes (Meyer, 1994, Findlay et al., 2002). Through the detritus-based food web, OM (organic matter) and microbes become an important food source for a wide range of macroinvertebrates, whose abundance often correlates with the OM standing stocks (Findlay et al., 2002). Macroinvertebrate activity, vice versa, speed up the OM decomposition rates (Cummins, 1973; Fazi & Rossi, 2000). OM indirect effect is the depletion of oxygen content, mediated by microbial oxygen consumption, also affect the macroinvertebrate community structure. Both these direct and indirect impacts of organic matter make the macroinvertebrate community one of the most relevant indicators of organic matter pollution in rivers and streams. The sensitivity of macroinvertebrates to a wide range of impacts makes them a very useful tool for assessing river quality. It is well known that macroinvertebrate communities from different habitats may respond in different ways to anthropogenic impacts (Parsons & Norris, 1996) and taxa from selected habitats might be particularly useful in detecting specific impact types (Kerans & Karr, 1994). Currently, macroinvertebrates are the most commonly used elements for biological classification of rivers in Europe. Although the details in methodologies might vary from country to country, the use of macroinvertebrates to assess the effects of organic pollution of rivers has a long history throughout Europe. More recently methods for using macroinvertebrates as indicators of other pressures including toxic chemicals and alterations in river flows and channel morphology, have or are being developed.

Since the key products of organic matter decomposition are nutrients, eutrophication is very closely linked to organic pollution. Effects of organic pollution and eutrophication on stream benthic fauna are linked to each other by processes of organic matter and nutrients transformations.

The first biotic index based on macroinvertebrates was the Trent Index developed by the Trent River Authority (Metcalf-Smith 1996). As the Saprobic system, the Trent Index mainly targeted organic pollution. In contrast to the saprobic system, the Trent Index uses only selected indicator taxa (i.e. with different sensitivity to reduced oxygen concentrations) in combination with a measure of diversity. The Trent Index has been the ancestor to all biotic indices currently in use in Europe (except those based on the Saprobien system). They differ, however, due to regional differences in macroinvertebrate species composition between regions of Europe and due to the introduction of various modifications relating e.g. to taxonomic level, index calculation etc. Arguing that the Saprobien index was taxonomically too demanding and too restricted in geographic context, researchers in the UK developed a more simplified biotic metric, i.e. the Biological Monitoring Working Party (BMWP) score and the Average (BMWP) Score Per Taxon (ASPT) (Armitage et al., 1983). Considering the special constraints of the Spanish rivers, Alba-Tercedor & Sánchez-Ortega (1988) developed an adaptation of the BMWP and ASPT to the Iberian Peninsula. Currently used methods and metrics include the BMWP and the ASPT (in the United Kingdom), the Belgian Biotic Index (BBI), Italian Extended Biotic Index (IBE) - a modified version of the Extended Biotic Index (Woodiwiss, 1964) - number of Ephemeroptera, Plecoptera and Trichoptera (EPT) taxa, Danish Stream Fauna Index (DSFI) (Skriver et al., 2000), number of Ephemeroptera and Plecoptera (EP) taxa, number of Ephemeroptera, Plecoptera, Trichoptera, Coleoptera, Odonata, and Bivalvia taxa, percentage EPT taxa, number of Ephemeroptera taxa, and number of Plecoptera taxa and the Danish Stream Fauna Index (DSFI).

The recent EU project “The Development and Testing of an Integrated Assessment System for the Ecological Quality of Streams and Rivers throughout Europe using Benthic Macroinvertebrates” (AQEM) developed alternative approaches to assess the impact of organic pollution by defining

stream typespecific multimetric indices (Sandin & Hering, 2004). During the implementation of this project, a multimetric index was tested and different metrics have been developed for the individual stream types, all of which show a relationship with organic pollution in the respective stream types (Hering et al., 2003). For organic pollution assessment, single metrics such as biotic indices and biotic scores are widely used and are usually based on high taxonomic levels, (e.g., BMWP, ASPT, Belgian Biotic Index; Metcalfe, 1989). The saprobic system based on benthic macroinvertebrates identified predominantly to the species level, is preferred in Central Europe.

Macroinvertebrate indices and metrics discussed:

- Biological Monitoring Working Party score (BMWP)
- Average Score Per Taxon (ASPT)
- Belgian Biotic Index (BBI)
- Indice Biotico Estesio (IBE)
- Number of Ephemeroptera, Plecoptera and Trichoptera taxa (EPT)
- Danish Stream Fauna Index (DSFI)

3.4.5 Secondary indicators (macrophytes, phytoplankton, fish)

Little information is available on the impacts of organic pollution on river macrophytes, although macrophytes are widely affected by high levels of organic matter. Two factors are likely to be important: (i) deposits of organic matter and low redox potentials in the sediments will prevent the occurrence of many macrophytes; (ii) increased turbidity and increased occurrence of attached micro-organisms could reduce light availability for macrophytes growth.

It is very unlikely that an organic pollution can impact river phytoplankton without leading to a more significant impact on heterotrophic organisms or benthic diatoms. Phytoplankton is therefore not considered a valuable indicator in the assessments of ecological impacts from discharges of organic pollutants into rivers.

The protection of fish populations in rivers from impacts of organic pollution has traditionally been a key issue in river pollution management. In the Fresh Water for Fish directive (78/659/EEC) much focus is on criteria for dissolved oxygen, BOD and ammonia. The guidance and mandatory criteria from the directive are reported in table 4.4.

Table 3.4 guidance and mandatory criteria from the Fresh Water for Fish directive (78/659/EEC): water quality criteria related to organic pollution.

	Dissolved oxygen (DO) mg/l		BOD ₅ mg/l		Total ammonium mg/l		Free ammonia mg/l	
	Mandatory	Guidance	Mandatory	Guidance	Mandatory	Guidance	Mandatory	Guidance
Salmonid waters	≥7 (100%)	≥9 (100%)	-	≤3	≤1	≤0.04	≤0.025	≤0.005
Cyprinid waters	≥7 (50%)	≥8 (50%) ≥5 (100%)	-	≤6	≤1	≤0.2	≤0.025	≤0.005

These criteria are based on the tolerances of fish to low oxygen concentrations and to ammonia. The criteria have been a major reason for the development of river models describing the impacts of discharges of organic matter with wastewater on the concentrations of DO, BOD and ammonia in rivers from Streeter and Phelps (1925) to Shanahan et al (2001).

This approach based on fish tolerances in relation to oxygen and ammonia concentrations has been successful regarding the direct effects of organic pollution on fishes, but it has not taken into con-

sideration the indirect effects of the other changes of the river ecosystem caused by organic enrichment.

Unfortunately, very little scientific work has been published recently on the relations between organic enrichment and fishes at the ecosystem level. However, it is evident that revisions and new approaches are needed compared to the Fresh Water for Fish directive criteria. Some of the problems are:

- The directive guidance value for BOD is high (6 mg/l) for cyprinid rivers. Often the water velocity in these rivers is low. An elevated level of BOD caused by organic discharges will have a much more profound effect in such a river than in a fast flowing river, where the BOD criteria is more strict (3 mg/l) if it is designated to be a salmonid river.
- Ecosystem impacts leading to changes in fish populations have generally not been considered. These changes can be changes in species and/or increases or decreases in populations caused by the organic enrichment.
- Relations between water quality and the quality of spawning areas for fishes is not clarified.
- Oxygen criteria are certainly relevant in principle, but the monitoring of compliance is very resource demanding to achieve a reasonable coverage, especially because of diurnal and seasonal irregular fluctuation caused by biological processes in the rivers.

It is especially important to identify types of pollution where fish populations are more vulnerable to organic pollution than the other biological quality elements, and subsequently to establish relations between organic enrichment and fishes for such cases.

3.5 Knowledge strengths and gaps

The conclusion from the literature review is that benthic macroinvertebrates and benthic algae are useful biological elements for the evaluations and assessments of organic matter impact on river ecosystems. The strength of this knowledge is summarized in the following box.

Knowledge strengths in the relationships linking organic matter pressures with chemistry and biology in rivers

- Changes in concentration of organic matter in rivers affect the river biota. A substantial amount of literature has demonstrated the consequences of organic enrichment on macroinvertebrates, attached algae and fish in rivers.

- Relations between BOD levels and saprobic indices are well established for rivers polluted by urban wastewater, especially for wastewater, that is not biologically treated, and for increases in BOD levels of more than about 1 mg/l. As the natural background level of BOD in rivers is also around 1 mg/l, minor changes in BOD level caused by wastewater discharges are likely to affect a river ecosystem.
- Organic pollution impacts are not necessarily related to oxygen deficits. Changes in biota caused by degradable organic matter also occur at low increases in the concentration of organic matter and without a substantial oxygen deficit in the river.
- The quality of organic matter discharged is important for the impacts on river biota. The biota of unpolluted rivers reflects the source, type and degradability of organic matter inputs to a river. Therefore, inputs from grassland, deciduous forest, coniferous forest, autotrophic production of organic matter by algae or macrophytes, and changes in riparian vegetation affect the quantity and quality of the organic matter and the species composition in the river. These changes can be dramatic for individual species, but the overall ecosystem changes are likely to be so small, that the overall biological quality can be considered unimpacted. Different is the case of organic inputs from anthropogenic discharges and/or eutrophication with nutrients which have a significant impact on river ecosystems. Wastewater discharges lead to high increases in concentrations of organic matter and significant changes in natural nutrient ratios (C/N/P/Si) in rivers, with serious consequences on the mineralization of organic matter and the composition of associated community.
- Mathematical models are available for the evaluation of chemical water quality. Concentrations of DO, BOD, ammonium and other chemical constituents are well described in rivers as a function of the discharge of pollutants, but the related models usually require a very substantial amounts of input data.
- River ecosystems experience many types of organic inputs. Traditionally, wastewater discharges from towns and industry were the dominant polluters with organic matter. Most of these discharges have been substantially reduced during the last decades, and the significance of other wastewater sources have become more important, e.g. urban storm-water, discharges from agricultural husbandry, fish-farming, wastewater from scattered dwellings and organic inputs to rivers from eutrophied lakes. The quality of the organic matter differs among pollution sources and also the quality of the remaining organic matter in treated wastewater is different from the untreated or insufficiently treated. Therefore, the relationships between organic loading and the biota response in rivers are strongly dependent on the type of wastewater.

So far biotic indices related to these elements have been intensively described. However, the existing knowledge is scattered and it is not operational in relation to the implementation of the Water Framework Directive. In particular, the scientific knowledge about quantitative relationships between biotic indices and the concentrations and/or qualities of organic matter should be improved. It is not possible at present to establish dose-response relationships between organic pollution and the biota. These relationships are likely to change with ecoregions and to be affected by the other possible pressures affecting river ecosystems. Therefore, a first research effort, to fill our knowledge gaps, should be done to better distinguish the variations in communities caused by natural fluctuations (ecoregional factors) from those caused by pressure from organic matter pollution. The effects of organic matter degradation in streams are significantly influenced by local conditions of current velocity, sub-stratum and channel morphology. Moreover, a major knowledge gap is that we do not know how streams are recovering from decreased levels of organic pollution and if they will return to their initial, undisturbed state.

When multiple impacts (e.g. habitat degradation or water pollution) are present meanwhile, it could be important to have habitat specific assessment methods. Particularly for Mediterranean rivers (that are characterized by a highly variable hydrological regime) confounding results may

arise when data derived from the transport and depositional areas are pooled together. The importance of separating transport (riffle) from depositional (pool) areas when combined effects of organic pollution and morphological degradation are present should be considered as a necessary research issue. When developing an instream macroinvertebrate assessment system, it is important to define the habitats which should be sampled at the micro- (e.g. cobbles, leaf litter) as well as at the meso-scale (e.g. riffle, pool). At the mesoscale, for rivers with a high diversity of hydromorphological features, additional care is needed to develop a meaningful and representative sampling strategy (e.g. Buffagni et al., 2000). Many assessment methods currently in use are specific to selected river units, e.g. the IBE method, which samples the "riffle" area only (Ghetti, 1997). The ecological attributes of organic pollution predetermines possible differences between lotic (riffles) and lentic (pools) stream habitats. Effects of organic matter degradation in streams are significantly influenced by local conditions of current velocity, substratum and channel morphology. Many Authors have already demonstrated differences in the community composition of "riffle" and "pool" areas (e.g., Brown & Brussok, 1991; Doisy & Rabeni, 2001). Moreover, the invertebrate fauna of different habitats is known to respond in different ways to water quality variation (Parsons & Norris, 1996). According to Plafkin et al. (1989), individual samples from pools and riffles should not be merged before analysis. In fact, the combination of more than one habitat in the evaluation of biological impairment may produce errors (Parsons & Norris, 1996), suggesting that caution should be exercised when selecting habitats for bioassessment surveys (e.g., Buffagni et al., 2000). The results of the AQUEM project (Hering et al., 2003) concerning differences between depositional and transport units of rivers support such conclusions. The mixing of the data derived from the transport and depositional areas may confuse the effects of the anthropogenic perturbations under analysis. In more general terms, many assessment methods based on macroinvertebrates implicitly assume that differences between pool and riffle areas exist, requiring macroinvertebrates to be collected in riffles only (e.g., IBE). Buffagni et al. 2004 showed that separate assessment of the riffle and pool groups of samples provides additional information, which is useful when combined effects of organic pollution and morphological degradation have to be considered.

An overwhelming scientific knowledge exists on the biological impacts of pollution of river with organic matter. The major gap needed to fill is the lack of operational tools to be able to calculate the needed reductions in wastewater discharges of organic matter to meet biological quality criteria for the different types of rivers in the different parts of Europe.

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4 Nutrients causing eutrophication

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4.1 Introduction

Autotroph (phytoplankton, macrophytes and phytobenthos) levels in rivers are highly responsive to changes in inorganic nutrient concentrations, especially at low nutrient levels. For all these biological elements, increased nutrient concentrations can potentially lead to an overall increase in biomass and the competitive exclusion of nutrient-sensitive, slow-growing species if no other factor intervenes. Large-scale phytoplankton growth is limited to slow-flowing rivers and macrophytes and phytobenthos are limited to areas where light penetration reaches the bed of the river.

Numerous studies have been undertaken to examine the relationships between macrophytes and benthic diatoms and nutrient status in rivers. However, we now need to explore the feasibility and applicability of upscaling the results of these different studies across Europe. This is necessary to assess and develop relationships between nutrient concentrations/loadings and the ecological status of rivers (monitored by periphyton, macrophytes and phytoplankton in rivers), required for the Water Framework Directive. Further understanding of the role of river bed sediment-associated nutrients on macrophytes and phytobenthos is needed as well as a better understanding of the relative importance of nitrogen and phosphorus.

At a more general level, being able to distinguish between natural nutrient enrichment within the river continuum from anthropogenic causes will be of importance in the assessment of biological community and their significance. Recent researches found that nitrogen may be limiting in rivers contrary to common belief that phosphorus is the main limiting nutrient for plant growth in rivers. A priori limitations can be determined using the ratio soluble N/P. The actual methods to determine which of N and P is truly limiting are either experimental enrichments or specific growth rates.

Benthic invertebrates and fish fauna are only indirectly responsive to inorganic nutrient changes, through change of habitat and food resources and therefore analysis for REBECCA will focus primarily on the linkages between autotrophs and nutrients.

The overall major requirements in relation to the implementation of the Water Framework Directive are to develop relationships that provide an indication of the reductions in nutrient concentrations which may be required in different rivers to achieve a good ecological status assessed from the occurrence of macrophytes, periphyton and/or phytoplankton in the river.

4.1.1 Structure of review

Autotroph (phytoplankton, macrophytes and phytobenthos) levels in rivers are highly responsive to changes in inorganic nutrient concentrations, especially at low nutrient levels. For all these biological elements, increased nutrient concentrations can potentially lead to an overall increase in biomass and the competitive exclusion of nutrient-sensitive, slow-growing species if no other factor intervenes. Large-scale phytoplankton growth is limited to slow-flowing rivers and macrophytes and phytobenthos are limited to areas where light penetration reaches the bed of the river.

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The pressure investigated in this chapter is the inorganic nutrients causing eutrophication. This report chapter aims at identifying the relations between inorganic nutrients and biological elements relevant to the Water Framework Directive. It also explores the relations between anthropogenic sources and in-stream inorganic nutrients. It is structured in six sub-chapters detailing:

- How the literature search has been conducted,
- General information on eutrophication and nutrient cycles, and a summary of its impacts on the river biota.
- The sources of nutrients for the rivers, especially anthropogenic sources.
- The impacts of the anthropogenic sources of inorganic nutrients on the water quality of the rivers.
- The existing relationships between nutrient levels and biota, the methods used to establish these relationships and the existing models, indicators and classification derived from them. It focuses on phytoplankton, macrophytes and phytobenthos, as these three elements are the most responsive to nutrient levels. Fish and macroinvertebrates are nonetheless mentioned.
- Summary of the knowledge strengths and the knowledge gaps on the relationships between biota and inorganic nutrients in rivers.

4.1.2 Working approach and information sources

Literature searches were made using the ISI Web of Science database, for peer-reviewed journal articles.

The searches were based on the combination of a keyword for rivers, one for the biological element and one for the eutrophication. The keywords used are as following:

- River keyword: river / stream / watercourse
- Biological element keyword: phytoplankton / periphyton / phytobenthos / diatoms / macrophytes / macroinvertebrates / invertebrates / fish / metric / indices / classification
- Eutrophication key word: eutrophication / nutrients / inorganic nutrients / phosphorus / phosphate / nitrogen / nitrate

This search identified 1,115 articles likely to contain relevant information. Further investigation revealed that, of these articles, 615 were highly relevant to this study. The actual review has been done on about 230 articles, considered as the most relevant.

Table 4.1. Repartition of journal articles in the reference database per type of biota it is focussing on

	Number of articles in database
Phytoplankton	67
Periphyton	236
Macrophytes	115
Macroinvertebrates	116
Fish	63

4.2 General impacts of nutrient enrichment

4.2.1 Eutrophication: definitions

Within the context of European legislation, there are several different definitions of eutrophication. Box 4.1 presents definitions from two EU directives (Urban Waste Water Directive and Nitrates Directive), from the European Environment Agency and the UK Environment Agency. All these definitions agree that eutrophication is enrichment in nutrients, principally phosphorus and nitrogen, leading to an increase in algae and higher plant growth and a disturbance of the ecological balance of the aquatic ecosystem.

Box 4.1. definitions of eutrophication as used in the European Directives and the European Environment Agency

Eutrophication is defined as...

European Directives:

- *Urban Waste Water Treatment Directive (91/271/EEC)*

... The enrichment of water by nutrients, especially compounds of nitrogen and/or phosphorus, causing an accelerated growth of algae and higher forms of plant life to produce an undesirable disturbance to the balance of organisms present in the water and to the quality of the water concerned.

- *Nitrates Directive (91/676/EEC)*

... the enrichment of water by nitrogen compounds, causing an accelerated growth of algae and higher forms of plant life to produce an undesirable disturbance to the balance of organisms present in the water and to the quality of the water concerned;

European Environment Agency:

- *European Environment Agency (1994):*

... Excessive enrichment of waters with nutrients, and the associated adverse biological effects.

... Nutrient enrichment, typically in the form of nitrates and phosphates, often from human sources such as agriculture, sewage, and urban runoff.

... Process by which a lake, a river, part of a sea, etc. is enriched with nitrates, phosphates and other nutrients which favour the growth of algae and often kill other organisms by lack of oxygen.

- *European Environment Agency –(1999)*

... Nutrient enrichment of the aquatic environment leading to increased rate of supply of organic matter; including primary production. This enrichment leads to environmental perturbation and changes ecological quality, and ultimately reduces the utility of the water body.

Environment Agency (UK):

... The enrichment of water by nutrients, stimulating an array of symptomatic changes including increased production of algae and/or higher plants, which can adversely affect the diversity of the biological system, the quality of the water and the uses to which the water may be put.

4.2.2 Nutrients involved in eutrophication

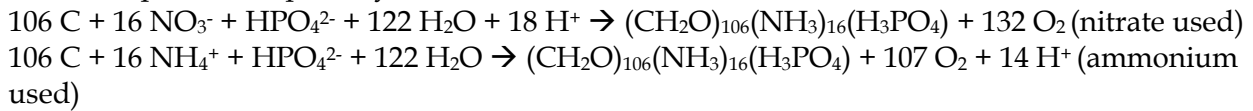
Nutrients required for algae and higher plants growth are:

- Nitrogen
- Phosphorus
- Inorganic carbon
- Silicon (for diatoms)

Nitrogen, phosphorus and inorganic carbon are key elements for photosynthesis, and so algal and plant growth, whereas silicon is indispensable to build diatom frustules.

Box 4.2. General equations of photosynthesis, when nitrate or ammonium is used

General equations of photosynthesis:



Algae and higher plants take up dissolved inorganic forms of these nutrients from the water column and/or sediments. For nitrogen, both nitrate and ammonium can be taken up (see equations of photosynthesis in the 2 cases in box 2) but freshwater algae preferentially uptake ammonium nitrogen when both are present (Hamilton et al., 2001). Nitrate nitrogen can nonetheless be an important source of nitrogen.

The availability of the nutrients depends on a number of factors. Inorganic carbon availability is determined by levels of dissolved carbon dioxide, linked to the balance between photosynthesis, respiration, CO₂ transfers at the air-water interface and carbonate solubility (Neal et al., 1998). It is usually assumed that inorganic carbon is not limiting for the algae and plant growth. However, nitrogen, phosphorus (and silicon for diatoms) can be limiting. Dissolved reactive phosphorus for phosphorus fraction (Ekholm, 1994) and nitrate and ammonium for the nitrogen fraction are the most readily bioavailable nutrients for algae and higher plants. Their concentrations depend on a number of processes. These processes are summarised in the figure 4.1 for the in-stream nitrogen cycle and in figure 4.2 for the in-stream phosphorus cycle.

Figure 4.1. In-stream nitrogen cycle

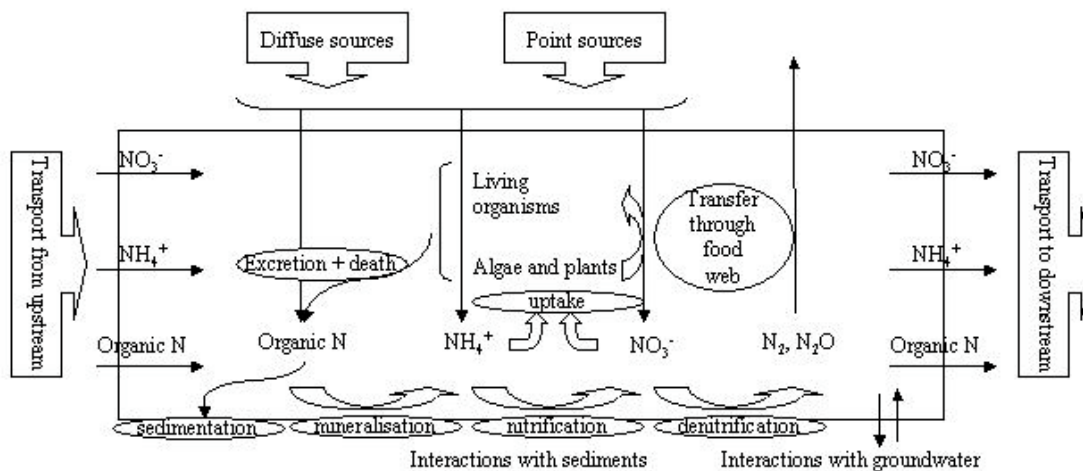
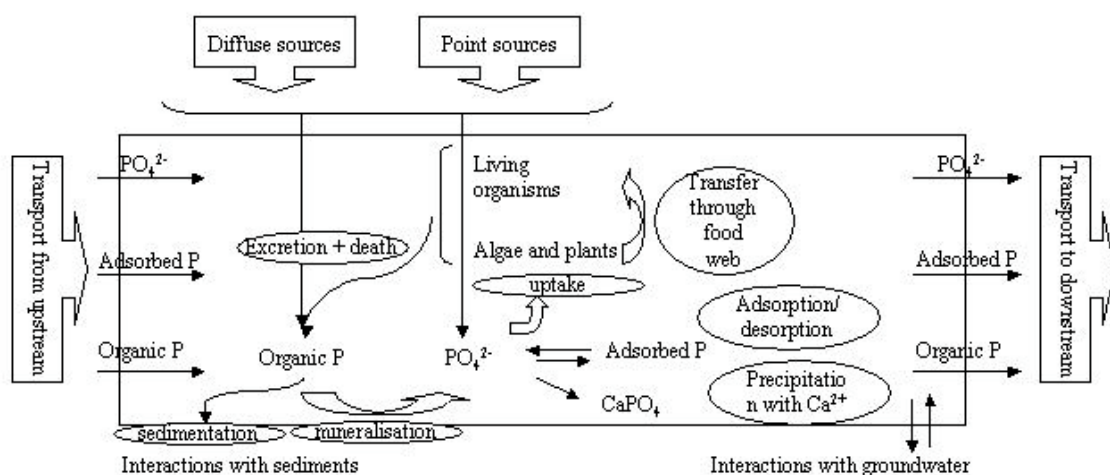


Figure 4.2. In-stream phosphorus cycle



Both these figures demonstrate the close links between organic matter and inorganic nutrients and illustrate a natural background enrichment from upstream to downstream, as organic matter and inorganic nutrients progressively accumulate in the stream and are transported downstream. Movements of organic matter from downstream to upstream can occur with fish and macroinvertebrate movements. Ultimately all major forms of nitrogen and phosphorus within the river are bioavailable.

The natural background enrichment along the river can easily be overwhelmed by anthropogenic inputs diffuse sources or point sources. Few anthropogenic activities remove organic matter and nutrients from streams. Abstraction of water modifies the annual nutrient load but does not affect the nutrient concentration in the water column. Angling, weed cutting and dredging of sediments are other anthropogenic activities that can remove nutrients from streams. However, their significance on the overall nutrient cycling may be limited.

4.2.3 General impacts

Enrichment in inorganic nutrients of a stream can cause the following impacts, as summarised in Nijboer et al.,2004:

- *Increase in primary production rate and mass growth of algae and plant:* increase in nutrients can potentially increase the growth of algae and macrophytes if these nutrients are initially limiting and if the other in-stream conditions allow more growth (e.g. light).
- *Change in communities:* Change in nutrients can lead to a shift from nutrient-sensitive slow-growing species to nutrient-tolerant fast-growing species. Competition for light induced by an increased growth may occur between epiphytes, macrophytes and phytoplankton, modifying the balance of these algae and plants. It reflects on macroinvertebrates and fish, through habitat and food web.
- *Reduction in oxygen:* high nutrient levels favour a high biomass of algae and plants and an increased decomposition rate. If algae and plants produce oxygen during the day, they consume it during the night and an increased biomass means an increase uptake of oxygen at night. It also means an increase in organic matter when the organisms die. Decomposition of organic matter is increased in presence of high nutrient level, there is more organic matter to decompose and decomposition consumes oxygen. All these processes can lead to oxygen depletion.
 - *Toxicity:* in the most extreme cases of eutrophication, combining high level of nutrients and depletion of oxygen, toxic substances like nitrite and ammonia can form. In high concentrations, they are lethal to fish and invertebrates.

The impacts of inorganic nutrient enrichment on water quality will be detailed in chapter 5 and impacts on river biota will be reviewed in detail for phytoplankton, periphyton, macrophytes, macroinvertebrates and fish in chapter 6. Interactions between these biological components (e.g. via predation, competition etc) may greatly influence the impacts of eutrophication.

4.3 Nutrient sources

4.3.1 Natural sources

Nitrogen and phosphorus are naturally present in streams, through transport of terrestrial organic matter, leaching and runoff of terrestrial inorganic nutrients and decomposition of in-stream bio-

logical material. They can also derive from the weathering of bedrock materials and deposition of dusts and salts.

Mainstone et al. (2002) estimated that the background export rates of total phosphorus in four UK rivers would be between 0.1 and 0.2 kg TP ha⁻¹year⁻¹, which means mean total phosphorus concentrations between 3.7 to 24.6 µg.l⁻¹.

4.3.2 Driving forces and pressures from anthropogenic sources

All anthropogenic activities that release organic matter and inorganic nutrients in the environment may cause in-stream enrichment in inorganic nutrients. They can be grouped into four categories: human life, industry, agriculture and transport/power plants. This sub-chapter details the pressures exerted by each anthropogenic activity. The impacts of these “raw” pressures are modulated by the level of activities and the technology in use.

Populations and industry

In western countries, each person releases 1-1.5 kg P.capita⁻¹.year⁻¹ and 2.2 to 5.5 kg N.capita⁻¹.year⁻¹ as wastes by excretion and use of detergents (European Environment Agency, 1999). The wastewaters, which are subject to variable levels of treatment, are then discharged as sewage effluent to rivers. An ensemble of socio-economic factors determines the population in a catchment and its dynamics.

The other main source of phosphorus from households is from the use of detergents. The phosphorus content of detergents has markedly decreased over the last 20 years. European Environment Agency (1999) indicates a decrease of 94% of the consumption of polyphosphates in detergents between 1975 and 1990 in Western Germany. The impact of detergents is driven by the market share of phosphate-free detergents and implementation of regulations on it.

Industries, primarily the food and drink industry, the industries producing phosphorus-based fertilisers, cleaning materials and/or metal finishing (Morse et al., 1993), can discharge significant amounts of phosphorus to surface waters. For example, large industrial companies such as Bayer can emit almost twice as much phosphorus as the whole of Denmark (European Environment Agency, 1999). Emissions from industries are driven by technology used and regulations.

The impacts of wastewaters from households and industries on rivers will then depend on the wastewater treatment, if any, received before being discharged to the river. In Europe, population for which wastewater is treated varies from 50% (southern and eastern Europe) to 80% (nordic and western Europe). Primary and secondary treatments primarily aim to remove organic matter from wastewaters and few inorganic nutrients are actually removed. After primary and secondary treatment, including detergents, 0.68 kg P.person equivalent⁻¹.year⁻¹ is discharged to water (Hilton et al., 2002). More and more, tertiary treatments are used to remove phosphorus and, to a lesser extent, nitrogen. Plants with specific biological treatment and chemical precipitation of phosphorus can remove up to 95% of the phosphorus. Population covered only by primary treatment range from 2 to 15%, population covered by secondary treatment as well as primary treatment from 8 to 60% and population also covered by tertiary treatment from 2 to 70% (European Environment Agency, 1999; Morse et al., 1993).

Implementation of the EU's Urban Waste Water Treatment Directive require the implementation of tertiary treatments that remove 80% of phosphorus, or 70% of nitrogen, or 75% of both nitrogen and phosphorus for sewage treatment works over 10,000 person equivalent that discharge to eutrophic waters or waters at risk of becoming eutrophic. Countries like Denmark where high investments have been done in wastewater treatments do not need to invest as much as countries like France and Greece to control phosphate (Farmer, 2004).

The type of wastewater treatments is driven by regulations on discharges and investments in wastewater treatments.

Agriculture

Agriculture releases nutrients to the environment under different forms: emissions of ammonia to air and leaching and runoff of nitrogen and phosphorus that reach the river. For crop production, inorganic fertilisers and manure can be applied. In Europe (European Environment Agency, 1999), use of nitrogen fertilisers varies from 30-70 kg N in Southern Europe to 180 kg N in the Netherlands in 1990, and 10 to 50 kg P for phosphorus fertilisers. Hilton et al. (2002) estimates the export coefficient of phosphorus by land cover types. It varies from 0 to 0.83 kg P.ha⁻¹.year⁻¹, depending on the type of landuse.

Application of manures varies across Europe from <40 to > 200 kg N and 8.8 to 44 kg P per hectare agricultural land. Surplus of nitrogen and phosphorus can runoff and manure emits ammonia to the air.

Livestock produces manure and ammonia as well. Hilton et al. (2002) estimates the export coefficient of phosphorus to 0.0054 (fowl) to 0.22 (cattle) kg.head⁻¹.year⁻¹. Emissions of ammonia were of 2,500 × 1,000 tonnes in 1994 in Europe and it is estimated that over 90% of these emissions come from agricultural activities (Skeffington, 2002).

The export rate of phosphorus and nitrogen can be affected by climate, geology, topography, soils hydrology, land management, the spatial location of nutrient sources in the catchment (Johnes et al., 1996).

Transport and power plants

Both transport and power plants combust fossil fuels. By-products of these combustions are nitrogen oxides. Once deposited, they can be transported to the rivers. 60 % of the nitrogen oxides in 1995 were coming from transport, 21 % from energy sector and 14 % from the industries (European Environment Agency, 1999).

The emissions are driven by the use of roads and air for transport and energy politics. The OCDE (OCDE - 2000) set targets in its “Environmentally Sustainable Transport” to reduce NOx emissions by 75/80 % by 2030 to achieve its goal on eutrophication.

4.3.3 Relative importance of the difference sources

Nitrogen load

Nitrogen within rivers is derived predominantly from agricultural sources (e.g. Jarvie et al., 1998). In Europe, agricultural sources can represent between 46 and 87% of the total nitrogen load. It depends on the surface of land in agricultural use and how intense it is. Point sources can also play an important role, up to 35 to 43% for some catchments and principally from sewage discharges. Nitrogen from the atmosphere can also represent a significant part of the nitrogen load, for example up to 35% for a Swedish catchment from 1982 to 1987 (European Environment Agency, 1999).

Phosphorus load

Source apportionment of phosphorus can highly vary between catchments. It is usually proportional with human activity. Phosphorus present naturally would represent only 3 to 15 % of the current levels (Farmer, 2004; Morse et al., 1993). In densely populated catchments, 50 to 76% of the phosphorus load was coming from point sources, mainly from sewage discharges unless in highly industrial catchments, and 20 to 40 % from agricultural diffuse sources (European Environment Agency, 1999). The percentage distribution of non-point sources of phosphorus could vary between 2 and 60% for European rivers (Farmer - 2004). As phosphorus source is dominated by ur-

ban areas, an increase in population density can lead to a shift from phosphorus to nitrogen limitation in streams (Garnier et al., 2002).

Quantification of diffuse losses of nitrogen N and phosphorus P

Several quantification tools exist to estimate the diffuse nutrient losses to surface freshwater systems. A EU research project EuroHARP (www.euroharp.org) is evaluating nine of them on 17 catchments. EuroHARP is a research project under the 5th Framework program. It started on the 1st of January 2002 and is running for 4 years. It offers guidance on quantification and reporting of nitrogen and phosphorus losses for different sources of nutrients, for example aquaculture plants, sewage treatment works, industrial plants etc, and their apportionment.

4.4 Nutrient concentrations

4.4.1 Primary effects on water quality

Anthropogenic activities discharge nitrogen and phosphorus nutrients from diffuse and point sources, in soluble and/or particulate form. Anthropogenic activity increases the overall nitrogen and phosphorus concentrations in the water column and possibly in the sediments through the settlement of particles. At a bigger scale, point sources and tributaries can induce spatial variations of concentrations in nutrients. Thus, there will be elevated levels of phosphorus just downstream of major wastewater treatments that will then decrease downstream through increased dilution, algal assimilation and sedimentation (Young et al., 1999). Inputs from tributaries at a lower concentration in nutrients can locally reduce the nutrient concentration in the main stream.

As shown in Hamilton et al. (2001), an increase of ammonium nitrogen leads to an increase in ammonium retention in streams. In the most upstream reaches, the upper layer of sediments is in aerobic conditions. These are conditions favourable to nitrification and decomposition of organic matter. These two phenomena, as well as the uptake of ammonium by algae and plants, use ammonium. Decomposition of organic matter delivers new nutrients to the water column. Nitrification increases the concentration of nitrate. Preferential uptake of ammonium by algae and higher plants reduce ammonium concentration from upstream to downstream. In headwaters there is no denitrification that could reduce nitrogen concentrations, as aeration is sufficient to keep aerobic conditions. Combined with nitrification of ammonium, the nitrate concentration can increase from upstream to downstream.

The lower river reaches are usually deeper, and lack of oxygenation may result in sediments becoming anaerobic. In these circumstances, nitrification is replaced by denitrification. In the absence of additional N sources, nitrate concentrations will then decrease due to denitrification and uptake by algae and plants (Peterson et al., 2001).

Interactions between river water with sediments are particularly important for phosphorus (e.g. House, 2003). Particulate phosphorus can settle on the riverbed. Desorption and adsorption of phosphorus can buffer concentrations of soluble reactive phosphorus in river. So, if the soluble reactive phosphorus concentration in the water column is higher than the equilibrium phosphorus concentration, phosphorus will be adsorbed by the sediment (House et al., 1997). This equilibrium concentration is dependent on the particle size and composition. Qotbi et al. (1996) confirmed the importance of soft sediments as store of bioavailable phosphorus for the Lot (France) and the dependence to particle size and area covered by soft sediments.

4.4.2 Interactions with river ecology

Increase in in-stream inorganic nutrients leads to a potential for higher uptake rates from algae and macrophytes. As summarised in Mainstone et al. (2002), increase in nutrients levels can lead to:

- An increase in plant growth, creating a large standing stock, and changes in the balance of species
- The development of epiphytic, epibenthic, filamentous and planktonic algae, that shade macrophytes

A high population of macrophytes, phytobenthos and phytoplankton can strip soluble reactive phosphorus (Mainstone et al., 2002) and ammonium (Balbi, 2000) out of the water column, even if the nutrient levels are high. All the autotrophs also require oxygen and a high standing stock of macrophytes and algae can lead to decrease in dissolved oxygen.

Increase in biomass of algae and macrophyte leads to increased organic levels of organic matter (when they die). This organic matter can either decompose in situ (very slow flowing rivers) or being carried away, increasing the nutrient levels of the river either locally or downstream. This decomposition requires oxygen, further lowering the dissolved oxygen level in the water column.

Interactions with ecology will be further detailed in section 4.5.

4.4.3 Seasonality of impacts

The impacts of inorganic nutrient enrichment vary with flow rate, temperature and light levels (e.g. Jarvie et al., 2003). Temperature conditions all in-stream processes, biotic and abiotic, whereas light levels primarily influence autotroph growth. Flow rate, temperature and light levels vary annually, from a minimum of temperature and light and a maximum of flow in winter to a maximum of temperature and light levels and a minimum of flow in summer. Inorganic nutrient enrichment has the highest impact in spring/summer, when algal and plant potential growth is at its highest and flow at its lowest.

Flow (river discharge in m^3/s) has a significant impact on all processes and impacts of nutrients sources. First, it determines the contact duration between nutrients and sediments and biological elements, thus determining the amount of nutrients that can be taken up. Second, it can dilute discharges from point sources. Its relation with climatic events will lead to differences in the forms of nutrients, their load and concentrations in the water column. And it can mobilise nutrients stored in sediments and slough algae and macrophytes.

The relative importance of the nutrient sources varies in time. Indeed, load discharges from point sources are usually constant over the year. As flow varies in time, from high flow in winter to low flow in summer months, the contribution of point sources to water concentrations in phosphorus is highest in summer (Mainstone et al., 2002) as they are not as diluted as in winter. It leads to peaks of phosphorus at low flows (Young et al., 1999).

Diffuse sources of phosphorus are more related to climatic events. So, the highest loads in phosphorus from diffuse sources occur in autumn, due to heavy rainfall after the summer months and high runoff, and are minimal in summer when little rainfall leads to little runoff. Diffuse sources will contribute more to the total phosphorus concentrations in autumn. However, it does not mean that autumn inputs of phosphorus from diffuse sources will have an impact on ecology, as it will mostly be as particulate phosphorus.

So, the relative contributions of diffuse and point sources will vary in time. As they do not discharge the same nutrients in the same forms, it can be of importance in assessing the relative ecological impacts of the different sources of nutrients.

The dominance of diffuse sources versus point sources also has important impacts on the nutrient concentration behaviour. Indeed, Jarvie et al. (1998) showed a distinction between agricultural and urban/industrial UK catchments through the link between flow and concentrations in nitrate, nitrite, ammonium and soluble reactive phosphorus. Nitrate concentration is strongly positively correlated with flow in agricultural catchments, due to diffuse agricultural runoff, whereas it is strongly negatively correlated with flow in industrial/urban catchments, where flow dilutes the discharge of nitrate. In all catchments, nitrite, ammonium and soluble reactive phosphorus were strongly and negatively correlated with flow, which underlines the importance of point sources and dilution for these nutrient elements. Balbi (2000) confirmed the positive correlation between nitrogen and discharge, and negative correlations between phosphorus and discharge.

The relative importance of diffuse versus point sources has a clear impact on the ratio between total dissolved inorganic nitrogen and total dissolved phosphorus. In urban and industrial catchments, this ratio was below 11, which, according to Mainstone et al. (2002) indicate periods of potential nitrogen limitations during the growing season, whereas it was over 20, which indicates periods of potential phosphorus limitations. As shown before, the concentration in nitrate increase with high flow (mobilisation of diffuse sources) and the concentration in phosphorus decrease due to dilution. It leads to an increase in the ratio nitrogen/phosphorus with high flows. It underlines the seasonality of phosphorus versus nitrogen limitations in catchments.

Flow can also have significant effect on in-stream nutrient retention. High flow reduces the water residence times thus reducing the time when nutrients interact with bed sediments and/or biota. Flow can also mobilise nutrients that were retained in a reach during summer. It is particularly significant for phosphorus as resuspension can represent 65% of total phosphorus export of a watershed in September (Svendsen et al., 1993).

Flow and rainfall are not the only seasonal events that can influence in-stream concentrations and loads of inorganic nutrients. The seasonality of in-stream processes also conditions the impacts of anthropogenic sources of nutrients on water quality. Temperature is one of the key determinants for in-stream processes. Temperature influences the maximum dissolved oxygen concentrations and the activity of all in-stream organisms. There is more activity in summer months than winter, resulting in higher rates of uptake of oxygen by microorganisms and higher organic matter decomposition rates.

There is also a contrast between the dormant season and active growth season. In dormant seasons, algae, higher plants and microorganisms have a very low uptake of nutrients. So the concentrations of these nutrients increase but do not lead to increase biomass and growth. On the other hand, during active growth season, the uptake of nutrients will be very high, and it will reduce the concentrations in nutrients in water column.

4.4.4 Models of water quality

A wide range of models has been developed. They all include a hydrological compartment. They can represent the in-stream processes and/or the land processes with varying degrees of complexity. The EU project "Benchmarking Models for the Water Framework Directive" (BMW) created a database of water quality models that include their main characteristics and their domain of applicability. This database is available on Internet, after a free registration to the BMW website (<http://www.rbm-toolbox.net/toolbox3/index.php>).

It includes models of in-stream processes for the river domain, for example QUESTOR, QUAL-2E, RIVQM, SOBEK and HERMES, and models of terrestrial processes for nutrients, for the diffuse domain, for example CREAMS, CASCADE, MIKE SHE, and SWAT.

4.4.5 Effects of reduction in nutrient inputs

Few studies have explored the effects of a reduction in nutrient inputs on water quality or the biota. House (2003) indicates that nutrients stored in the sediments could be released in the water column as the equilibrium between water column and sediments. In this case, the concentration of nutrients in water would not decrease. The model developed in Wade et al. (2002a) estimated that for the Thames, the sediments would not release nutrients after implementation of phosphorus-stripping work at the wastewater treatment plant upstream the site of study.

As for impacts on the biota, Jarvie et al. (2002) indicates that implementation of phosphorus-stripping before the discharge of wastewater did not lead to an improvement of biota macrophytes and phytobenthos. Young et al. (1999) analyses this situation as resulting from an insufficient reduction in in-stream nutrients, that keeps an eutrophic status of water. On the other hand, Sosiak (2002) found a decrease in phytobenthic biomass after reduction of phosphorus in discharge of wastewater treatment (even if not directly downstream) and in macrophyte biomass following reduction in nitrogen.

These conflicting experiments underline a lack of knowledge of the effect of nutrient stripping and the recovery that can be expected from the biota.

4.5 Impacts on river biota

Autotrophs are dependent on inorganic nutrients for phosphorus and nitrogen sources. Without these elements, no photosynthesis could occur. Thus, this subchapter will detail the effects of nutrients on macrophytes, phytoplankton and phytobenthos. The effects of inorganic nutrients causing eutrophication on fish and benthic invertebrates will be evoked at the end.

For each of the autotrophs, the effects of inorganic nutrients on biomass and species assemblage are detailed. Interactions with other environmental factors are given, as well as the relative importance of nutrients. The techniques used to establish the relationships are briefly presented with examples of papers in which they have been used. Lastly, the subsequent models, indices and classifications are presented.

Table 4.2. Methods of investigation of the relation between a biological element (macrophytes, phytoplankton, phytobenthos, macro invertebrates, fish and interactions between 2 or more biologic element), number of references in database and examples of key references. RM: river monitoring, LB: laboratory bioassay, IE: in-situ experiment

Biological elements	Methods of investigation	Number of references	Examples of references
Macrophytes	RM	53	(Baatrup-Pedersen et al., 2003, Carbiener et al., 1990, Demars et al., 1998, Grasmuck et al., 1995, Jarvie et al., 2002, Mesters, 1995, Thiebaut et al., 2002)
	LB	8	(Carr et al., 1998, Crossley et al., 2002, Madsen et al., 2002, Prahl et al., 1991, Spencer et al., 2003, Spink et al., 1993, Underwood et al., 1992, Zauke et al., 1982)
	IE	0	
Phytoplankton	RM	30	(Améziane et al., 2003, Balbi, 2000, Foster et al., 1997, Garban et al., 1999, Marker et al., 1997, Noppe et al., 1997, Reynolds, 2003, Ruse et al., 1997, Thebault et al., 1999, Young et al., 1999)
	LB	6	(de Andrade et al., 1998, Guelda et al., 2004, Olguin et al., 2004, Smoot et al., 1998)
	IE	0	
Phytobenthos	RM	100	(Biggs et al., 1998, Dent et al., 1999, Horner et al., 1990, Kelly, 2003, Kiffney et al., 2000, Kjeldsen, 1994, Kwandrans, 2002, Lindstrøm et al., 2004)
	LB	19	(Anderson et al., 1999, Humphrey et al., 1992, Marinelarena et al., 2001, Mulholland et al., 1994, Mulholland et al., 1991, Smoot et al., 1998, Sterling et al., 2000, Walton et al., 1995)
	IE	49	(Biggs - 2000, Biggs et al., 2000, Rosemond, 1993, Scrimgeour et al., 1997, Slavik et al., 2004, Snyder et al., 2002)

Benthic invertebrate fauna	RM	67	(Bargos et al.,- 1990, Buffagni et al., 2000, Camargo et al., 2004, Kiffney et al., 2001, Lazaridou-Dimitriadou et al., 2004, Skoulikidis et al., 2004, Soininen et al., 2004)
	LB	3	(Anderson et al., 1999, Gido et al., 2001, Walton et al., 1995)
	IE	14	(Biggs, 2000, Bowden et al., 1994, Corkum, 1996, Gothe et al., 1993, Keldsen, 1996, Mundie et al., 1991, Perrin et al., 1997)
Fish	RM	20	(Belliard et al., 1999, Eklöv et al., 1998, Miltner et al., 1998, Norton et al., 2002, Rashleigh, 2004)
	LB	3	(Smoot et al., 1998, Sterling et al., 2000)
	IE	5	(Deegan et al., 1997, Harvey et al., 1998, Moss, 1976, Stewart, 1987, Vanni et al., 1990)
Interactions between 2 or more biological elements	RM	3	(Basu et al., 1997, Dangelo et al., 1997, Rosemond, 1994)
	LB	6	(Anderson et al., 1999, Gido et al., 2001, Mulholland et al., 1994, Mulholland et al., 1991, Underwood et al., 1992, Walton et al., 1995)
	IE	17	(Deegan et al., 1997, Flecker et al., 2002, Harvey et al., 1998, Keldsen, 1996, Perrin et al., 1997, Peterson et al., 1993)

4.5.1 Macrophytes

Relationships between nutrients and macrophytes

Macrophytes are large aquatic plants. They can exist under various growth forms, for example emergent, submerged and floated-leaved.

Macrophytes are part of the primary producers in rivers. Their growth is conditioned by the uptake of inorganic nutrients by their roots and their shoots. Inorganic nutrients can be taken up either from the water column or the sediments, or both in varying proportion (Clarke et al., 2001). Nutrients reach the roots by slow diffusion in the sediments (Sand-Jensen et al., 1991). The relative importance of sediments and water column as a source of nutrients depends on the macrophyte species and the nutrient conditions (Crossley et al., 2002, Maine et al., 1999, Spink et al., 1993). In nutrient-limited rivers, enrichment in inorganic nutrients may lead to an increase in biomass and species richness up to a point when competitive exclusion occurs, reducing the species richness, and, for the worst cases of eutrophication, the disappearance of all macrophytes (Mesters, 1995). Enrichment may decrease the ratio between roots and shoots (Barko et al., 1991, Madsen et al., 2002). Flow can interact with nutrients, as a moderate flow improves the renewal of water in contact of the macrophytes, thus favouring the macrophyte growth (Barendregt et al., 2003, Crossley et al., 2002).

However, these effects of nutrients are contested in several studies. (Demars et al., 1998) underlined the difficulty to identify the specific effect of nutrients because of the synergistic effect of a number of physical factors. Holmes et al. (1999) indicated that species diversity in itself is not an indicator of trophic status of rivers as a wide range of diversity can occur at any phosphorus concentration and low to intermediate diversity occur at any phosphorus concentration as well. Environment Agency (2002) pointed out as well that macrophytes were not an indicator of the level of nutrients in lowland rivers and Madsen et al. (2002) did not find an effect of enrichment on the growth rate of two macrophyte species, suggesting that nutrient were not limiting for their growth. The trend may exist but only reflect the parallel evolution of nutrients and morphology of a river from spring to mouth. Indeed, most studies have compared the macrophyte communities and biomass at different sites of a same river (e.g. in Demars et al., 1998, Thiebaut et al., 1998).

Other studies confirmed the prominence of physical characteristics on determining macrophyte biomass and species composition (Baattrup-Pedersen et al., 2003, Barendregt et al., 2003, Demars et al., 1998, Schorer et al., 2000). They underlined the effects of flow and its variations, light (water depth, shading by riparian vegetation, suspended solids, phytoplankton and epiphytes), substrata and weed cutting.

Flow velocity and discharge exerts different influences on macrophytes. First, flow velocity is a mechanical stress that can break macrophytes and form an obstacle to colonisation. Low flow velocity favours colonisation, medium flow velocity favours the biomass growth and high flow velocity retards both (Biggs, 1996). High flow is often associated with suspended solids that shade the macrophytes. Water discharge conditions, combined with the width of the river, the water depth and so the light attenuation and the habitats (Barendregt et al., 2003). Flow discharge can be the most important factor determining the macrophytes' cover and biomass (Flynn et al., 2002).

Light that reaches the macrophytes can be affected by a number of different factors (Holmes et al., 1999) that can affect the submerged macrophytes or all types of macrophytes. Water depth and

suspended solids (already mentioned), phytoplankton and epiphytes affect submerged macrophytes. Riparian vegetation can shade the whole stream thus affecting all types of macrophytes.

Weed cutting has been found to be the main factor that separates communities in small Danish streams (Baattrup-Pedersen et al., 2003). Structure of macrophytes was more heterogeneous and the species richness higher in uncut reaches than reaches where weed-cutting was carried out (Baattrup-Pedersen et al., 2002). At the time of these two studies, the coverage of macrophytes was not affected by weed cutting.

Macrophytes exert a strong influence on their environment, especially when forming dense groups. They can change the redox potential of sediments, thus freeing inorganic phosphorus (Barendregt et al., 2003). Flow can be canalised between the groups of macrophytes, thus affecting in long term the morphology of the river. It is slowed down when flowing through the macrophytes groups. The overall flow of the river is slowed down if macrophytes occur along the whole width of the river. As the flow slows down, organic particles and suspended solids start to deposit, enriching the sediments within the macrophytes groups (Sand-Jensen, 1998). This organic matter is mineralised locally, increasing the concentrations in inorganic nutrients available to the macrophytes (Clarke et al., 2001). When water velocity is very low, dying and dead macrophytes decompose at the same place, further enriching the local environment. As velocity can be very slow within macrophyte stands, the water conditions can be very different from the overall river (Sand-Jensen et al., 1991). When a stream is dominated by macrophytes, river water quality can strongly vary. Thus, diurnal variation of DO and pH of streams dominated by submerged macrophytes were found of greater amplitude than for other streams (Wilcock et al., 2001).

Macrophytes interact with other biota. They can shade the phytobenthos and the phytoplankton. Also, epiphytes, which develop on the macrophytes, can shade the macrophytes, reducing light up to a point that can lead to the death of macrophytes (ten Cate et al., 1991). They can form a barrier to nutrient diffusion, thus reducing the amount of nutrients the macrophytes have access to (Sand-Jensen et al., 1991). Macrophytes are also an important habitat for macroinvertebrates, fish, amphibians and zooplankton (Mesters, 1990). Fish can physically disturb macrophytes, making them sensitive to velocity, and macrophytes can be grazed (Demars et al., 1998). When macrophytes form dense groups, this can protect macroinvertebrates and zooplankton from predation. Grazing of epiphytes by snails have a positive impact on macrophytes (Underwood et al., 1992). However, the high diurnal variability of water conditions like pH and DO (Wilcock et al., 2001) within such macrophytes' groups can adversely affect the fauna.

Existing methods to investigate the relationships

Biomass

Direct measures of macrophyte biomass are not widely used. Evaluation of the macrophyte biomass is more often done by estimating the surface they cover or abundance. If measuring biomass directly, shoots' biomass, roots' biomass or both shoots' and roots' biomass can be measured. There is only a limited relationship between cover and biomass, from various studies on macrophytes in southern England, Hugh Dawson (CEH Dorset) concluded that for the same cover, biomass could vary by a factor of three.

Species composition

Macrophytes composition can be assessed by recording the species/taxa present at a site and possibly the species abundance. Several indices for species richness and evenness have been developed: for example Average Score Per Taxon (H.M.S.O - 1987) based on number of taxa, surface cover class and biomass, distribution factor (Verbreitungsfaktor) based on species abundance per

section, Shannon-Wiener functions (Shannon et al., 1963), Sorensen index and the Simpson index (Simpson - 1952). A few studies have considered the growth form of macrophytes rather than species and taxa (e.g. Ali et al., 1999, Willby et al., 2000).

How to establish the relationships?

The relationships between inorganic nutrients and macrophytes' biomass and composition can potentially be studied through (see table 2):

- Laboratory bioassays
- In-situ enrichment
- Analysis of monitoring data

Laboratory bioassays or in-situ enrichments have not been commonly used to study the effect of nutrient enrichment, contrary to phytobenthos and in-situ enrichment have not been carried out.

Laboratory bioassays are based on artificial channels. They have been used to identify the relative effects of P and N enrichments in sediments on biomass of a few macrophyte species (Carr et al., 1998, Spencer et al., 2003, Spink et al., 1993), the relative contribution of shoots and roots to the uptake of nutrients (Madsen et al., 2002) and the interactions between flow and nutrients (Crossley et al., 2002).

Monitoring data have been far more widely used. Analysis of the data can vary from the simplest to the most complex.

Regression analysis allows making a link between species composition (as diversity indices), abundance or biomass with nutrients or other factors (Carr et al., 2003, Flynn et al., 2002).

Correlations are used to analyse interactions with a wider range of variables, including nutrients, as in Mesters (1995), Szoszkiewicz et al. (2002) and Thiebaut et al. (2002).

More complex analysis can explore species preferences, and group sites and species of similar behaviour as well as linking it with the environmental characteristics, including inorganic nutrients. Such techniques include ordination and clustering techniques (Demars et al., 1998, Holmes et al., 1998, Riis et al., 2000), for example canonical correspondence analysis (Bernez et al., 2004, Ferreira et al., 1999), principal component analysis (Carbiener et al., 1990, Grasmuck et al., 1995, Willby et al., 2000), detrended correspondence analysis (Baatrup-Pedersen et al., 2003) and other multivariate analysis (Chatenet et al., 2000, Ensminger et al., 2000).

Table 4.3. Summary of some of the relations found between macrophytes and inorganic nutrients

Biological element	Water quality element	Relation	Sites	Nutrient range	References
Total % macrophyte cover	SRP	Regression: $y = -120.37 + 77.1 \cdot \log \text{SRP}$ ($R^2 = 0.45$, $p = 0.002$)	River Kennet, UK n=20	Not indicated	(Flynn et al. - 2002)
MTR	Soluble nitrate concentration Soluble phosphate concentration	Correlation: -0.43 ($p < 0.042$) -0.58 ($p < 0.003$)	River Welland, UK	Not indicated	(Demars et al. - 1998)
Shift in species	PO_4^{3-}	1 st ordination axis correlated among other things with: PO_4^{3-} ($R = 0.74$, $p = 0.0015$, $n = 17$)	28 streams, The Netherlands	Not indicated	(Mesters - 1995)
Species richness	N- NH_4^+ P- PO_4^{3-}	Coefficient of rank Spearman correlation 0.457 ($p < 0.05$) 0.611 ($p < 0.001$)	30 sites of Vosges/Alsace floodplain, France	Mean N- NH_4^+ = 31-422 $\mu\text{g/L}$ P- PO_4^{3-} = 6-413 $\mu\text{g/L}$	(Thiebaut et al. - 2002)
Abundance	N- NH_4^+ P- PO_4^{3-}	Coefficient of rank Spearman correlation 0.600 ($p < 0.001$) 0.707 ($p < 0.001$)	30 sites of Vosges/Alsace floodplain, France	Mean N- NH_4^+ = 31-422 $\mu\text{g/L}$ P- PO_4^{3-} = 6-413 $\mu\text{g/L}$	(Thiebaut et al. - 2002)
Species evenness	N- NH_4^+ P- PO_4^{3-}	Coefficient of rank Spearman correlation -0.238 (non significant) -0.780 ($p < 0.0001$)	30 sites of Vosges/Alsace floodplain, France	Mean N- NH_4^+ = 31-422 $\mu\text{g/L}$ P- PO_4^{3-} = 6-413 $\mu\text{g/L}$	(Thiebaut et al. - 2002)
MTR	N- NO_3 N- NH_4 N total SRP P total	Pearson linear correlation coefficient 0.156 -0.299 ($p < 0.05$) -0.368 ($p < 0.05$) -0.260 -0.294 ($p < 0.05$)	48 sites (19 rivers) in lowland Poland	Not indicated	(Szozkiewicz et al. - 2002)
6 phytosociological groups	N- NH_4 N- NO_3 P- PO_4	-0.68 + 0.81 (contextual) - 0.92	29 sites	N- NH_4 = 0-90 $\mu\text{g.L}^{-1}$ P- PO_4 = 1-42 $\mu\text{g.L}^{-1}$ N- NO_3 0.2-7.5 mg.L^{-1}	(Carbiener et al. - 1990)

Use of relationships in models and indices

Models developed

Several dynamic models have been developed. Wright et al. (1986) gave a model of macrophyte biomass in shallow waters, function of light and nutrient limitations. A model of macrophyte and epiphyte biomass has been developed for the River Kennet (Southern England), including SRP (Wade et al., 2002b). The MACRIV model represents macrophytes by its oxygen equivalent and is used to model diurnal variation in oxygen and nutrient cycle (Park et al., 2003). AQUATOX (Park et al., 2004) models macrophyte biomass depending on light availability, current velocity and temperature. It distinguishes the bryophytes from the other macrophytes and bryophytes are dependent on nutrients.

Figure 4.3. Equation of macrophyte biomass in Kennet model (Wade et al., 2002b):

$$d(x_7)/dt = \frac{c_{10} \cdot \theta_M^{(T-20)} \cdot x_7 \cdot x_{12} \cdot R \cdot c_{12}}{(c_{11} + x_{12}) \cdot (c_{12} + x_7)} - c_{14} \cdot x_7 \cdot x_8 \cdot x_1$$

with:

x_7 macrophyte biomass at time t (g C m⁻²)
 c_{10} macrophyte growth rate (day⁻¹)
 $\theta_M^{(T-20)}$ macrophyte temperature dependency
 x_{12} SRP in pore water at time t (mg P L⁻¹)
 R solar radiation at time t (normalised to 0-1 from W.m⁻²)

c_{12} self shading (g C m⁻²)
 c_{11} half saturation of P for macrophyte growth (mg P L⁻¹)
 c_{14} macrophyte death rate (s g C⁻¹ day⁻¹)
 x_8 epiphyte biomass at time t (g C m⁻²)
 x_1 flow out of reach at time t (m³.s⁻¹)

Figure 4.4. Equation of macrophyte biomass in AQUATOX (Park et al., 2004):

$$dBiomass/dt = \text{loading} + \text{photosynthesis} - \text{respiration} - \text{excretion} - \text{mortality} - \text{predation} - \text{breakage}$$

With:

loading loading of macrophytes, usually used as a "seed"

photosynthesis rate of photosynthesis

respiration respiratory loss

excretion excretion or photorespiration

mortality nonpredatory respiration

predation herbivory

breakage loss due to breakage
 Photosynthesis includes a light limitation factor, a temperature limitation factor, habitat limitation factor, and a toxicant effect factor. The light limitation factor includes the shading effect of periphyton and macrophytes. For bryophytes only, it also includes a nutrient limitation factor expressed as following:

$$\text{NutrLimit} = \min(\text{Plimit}, \text{Nlimit}, \text{Climit})$$

Indices and classifications

Sensitivity of a species to inorganic nutrients and its preferences has been derived from expert opinions and statistical analysis as described above.

In Robach et al. (1996), principal component analysis on water chemistry for 129 sites in the Alsace Rhine floodplain and in the Northern Vosges mountains (France) found that the first axis was highly correlated with conductivity, hardness and pH whereas the second axis represented the trophic level with a correlation with phosphate of 0.83 and ammonia nitrogen of 0.78. Annual mean concentrations of phosphate phosphorus and ammonia nitrogen range from 3 to 350 µg/L across the sites, and from 0.5 to 0.75 mg/L for nitrate nitrogen. Another principal component analysis on macrophytes species presence and abundance matched with the previous distribution of water quality characteristics and leads to the determination of 10 phytosociological groups of macrophytes, 4 for acidic waters and 6 in calcareous waters, depending on pH, conductivity, hardness, N-NH₄⁺, P-PO₄³⁻ and N-NO₃⁻.

Lists for macrophyte species can be found in Holmes et al. (1998), Holmes et al. (1999), Zelinka et al. (1961), etc. Preferences of nutrients in sediments have not been successful yet because of the high variability of sediments characteristics (Clarke et al., 2001).

From these trophic preferences have been developed a number of different indices, summarised in (Haury et al., 2000). From the multitude of indices, 2 main indices can be identified: the Mean Trophic Rank (MTR) and the Trophic Index of Macrophytes (Trophie-Index Macrophyten - TIM).

The Mean Trophic Rank has been developed for England and Wales to implement the EC Urban Waste Water Directive: it is used to assess the impact of point sources on the river (Kelly - 1998). It is based on the combination of species at a site and, for each species, its indicator value and its abundance (Holmes et al., 1998, Holmes et al., 1999). 128 species have been identified for this index. They can be split in 4 broad groups and in 10 River Community Types (Holmes et al., 1998). These River Community Types are physically described and the mean MTR value for each of these groups can be used as a reference for physically similar sites. The MTR has been adapted to several countries, including Poland (Szozkiewicz et al., 2002).

Box 4.3. Brief description of Mean Trophic Rank (derived from Holmes et al., 1999)

$$MTR = \frac{\sum CVS}{\sum SCV} \cdot 10$$

where $CVS = SCV \times STR$

and where $SCV =$ Species Cover Value (cover class per taxon, on a scale 1-9)

$STR =$ Species Trophic Indicator (score assigned to species on a scale 1-10: the higher the score, the lower the tolerance to nutrient enrichment)

MTR score	Interpretation
MTR > 65	Unlikely to be eutrophic *
25 - 65	Site likely to be either eutrophic or at risk of becoming eutrophic **
MTR < 25	Site badly damaged by eutrophication, organic pollution, toxicity or is physically damaged

* If the score is less than might be expected for a site of this physical type, the site may be at risk of becoming eutrophic.

** If the score is less than might be expected for a site of this physical type, the site is eutrophic or at risk of becoming eutrophic.

The GIS Macrophytes indices developed in France follow similar reasoning as the MTR (Haury et al. - 1996).

The Trophic Index of Macrophytes (Trophie-Index Macrophyten) has been developed for Bavaria (Germany) to indicate the trophic state of rivers. It is based on a trophic value (determined with SRP and phosphorus in sediments) and a sensitivity index and number of sites where the species occur (Schneider et al., 2003). This index is not directly linked to in-stream nutrient concentrations.

Box 4.4. Brief description of Trophic Macrophyten Index (Schneider et al. - 2003)

$$TIM = \frac{\sum_{i=1}^n IV_i \cdot W_i \cdot Q_i}{\sum_{i=1}^n W_i \cdot Q_i}$$

Where IV_i = indicator value of species i . It is derived from the distribution of species count across the trophic categories. The trophic categories are derived from the amount of phosphorus from pore water and overlying water - 0 to > 1500 $\mu\text{g/L}$ - used by the plant at the sampling site (different from concentrations actually measured).

W_i = weighting factor of species i . It is derived from the tolerance of the species. The higher the less tolerant

Q_i = quantity of species i in the river section

Value of the TIM	Trophic state
$1.00 \leq TIM < 1.45$	Oligotrophic
$1.45 \leq TIM < 1.87$	Oligo-mesotrophic
$1.87 \leq TIM < 2.25$	Mesotrophic
$2.25 \leq TIM < 2.63$	Meso-eutrophic
$2.63 \leq TIM < 3.05$	Eutrophic
$3.05 \leq TIM < 3.50$	Eu-polytrophic
$3.50 \leq TIM < 4.00$	Polytrophic

4.5.2 Phytoplankton

Relationships between nutrients and phytoplankton

Phytoplankton is composed of algae and diatoms in suspension in water column. It can include only 'true' phytoplankton species or also phytobenthic species detached by flow, especially after spates. For the most upstream stretches, hydraulic conditions are not favourable to the development of true phytoplankton and phytoplankton is then dominated by benthic algae (Améziane et al., 2003). If the algal cells sink too deep and are not resuspended, the lack of light lead to their death. In Marker et al. (1997), it is thought that it is the cause of the crash in phytoplankton biomass in summer, as very low flow are not capable of resuspended phytoplankton cells. It could also be a key factor in the phytoplankton composition, with dominance of diatoms (heavy cells) in early spring later replaced by lighter cells (Sand-Jensen et al., 1991) as flow decreases. The generation time of phytoplankton is of several days. It makes it highly reactive to change in conditions of water and light. It also underlines that a significant phytoplankton biomass cannot be reached unless the residence time of water in the river is greater than the generation time. So, only slow-moving rivers, canals and on-river reservoirs can have a significant phytoplankton biomass. Thus Jarvie et al. (2003) showed that phytoplankton was a significant part of primary production in a canal but only account for a small part of the primary production in a shallow river. Due to transport by flow and longer residence times in more sinuous downstream sections, the phytoplankton biomass increases from upstream to downstream (Foster et al., 1997). Discharge of water from reservoirs and lakes can increase the phytoplankton biomass and richness downstream the discharges, with the addition of reservoir-specific species (Améziane et al., 2003).

Nutrient enrichment can lead to a change in biomass and/or a change in species composition. Indeed, an increase in inorganic nutrients (P and/or N) can lead to an increase in biomass (e.g. Vanni et al., 1990), or algal blooms. These algal blooms have been found to occur principally in spring in rivers (Balbi, 2000, Marker et al., 1997) and are considered as a nuisance. Nutrient enrichment can lead to a change in species composition by favouring a selective competition between nutrient-tolerant species and nutrient-sensitive species. It favours the species for which the preferred ratio N/P is nearer to the ratio N/P in the water column (Hecky et al., 1988). Species composition is also affected by the way nutrient enrichment occurs. Indeed, nutrient pulses have been shown to favour a higher species richness, as all species can persist (Hecky et al., 1988).

Silicon concentration does not have an effect on the overall total biomass of phytoplankton. However, it will affect the phytoplankton species composition (Foster et al., 1997, Hecky et al., 1988), especially for diatoms.

The effect of enrichment will however be modulated by the initial nutrient concentrations in the water column. There is no effect of enrichment if the nutrient is not limiting. Thus, Mallin et al. (2004) showed no impact of phosphorus enrichment on phytoplankton biomass (0-161.3 $\mu\text{mol P/L}$, stream concentration: 0.2-4.6 $\mu\text{mol P/L}$) for 2 blackwater streams whereas nitrogen enrichment over 14.3 $\mu\text{mol N/L}$ in nitrate (stream concentration: 0.4-66.4 $\mu\text{mol N/L}$) leads to an increase in phytoplankton, whatever the form of nitrogen. Young et al. (1999) showed as well that there was little correlation ($r=-0.29$) between chlorophyll a and phosphorus concentrations (0.1-6.7 mg $\text{PO}_4\text{-P/L}$) for 3 lowland rivers of the Thames catchment -the River Thames, the River Kennet and the River Blackwater-.

However nutrients may not be the principal determinants of the phytoplankton biomass and composition. Balbi (2000) and Hakanson et al. (2003) underlined that discharge, light and temperature were significant predictors of chlorophyll a concentrations in the water column. Discharge conditions the scouring of phytobenthos and the resuspension of sedimenting phytoplankton cells. Light is affected by the seasons, the shading by riparian vegetation, shading by macrophytes and turbidity of water. Inorganic nutrients were included in the analysis by Balbi (2000) and Hakanson et al., (2003) but were not found as significant as these three factors. Guelda et al. (2004) confirmed the predominance of light on phytoplankton biomass until a threshold of 7 $\text{E.m}^{-2}.\text{d}^{-1}$ is reached. Over this threshold, nutrients can be limiting.

Overall, the seasonal variability of phytoplankton (biomass and assemblage) is linked to seasonal variation in hydraulic and meteorological factors rather than nutrients, nutrients and other abiotic factors explaining the spatial variation in phytoplankton (Ibelings et al., 1998, Noppe et al., 1997, Wehr et al., 1997).

Increase in phytoplankton has an impact on the water quality. Indeed, to increase its biomass, it has taken up nutrients; an increase in phytoplankton biomass is associated with a decrease in nutrients (Balbi, 2000). However, the effect can vary depending on the nutrients considered: Balbi - (2000) showed a decrease in inorganic nitrogen and silicon associated with an increase in chlorophyll a but not a decrease in soluble reactive phosphorus whereas an increase in chlorophyll a was associated with a decrease in silicon and total phosphorus in the River Elbe (Guhr et al., 2004). Garban et al. (1999) emphasised the sharp decrease in both silicon (- 70%), ammonium (- 50 %) and soluble phosphorus (- 40 %) due to the phytoplanktonic bloom. At its death, phytoplankton decomposes and thus release inorganic nutrients to the river. Young et al. (1999) showed that a peak of phosphorus was observed in late summer to autumn after the peak of chlorophyll a in summer, suggesting the role of phytoplankton death. However, it was concomitant with a decrease in flow. As phosphorus is predominantly released by point sources not related with climatic events, a decrease in flow can lead to an increase in phosphorus concentration. The

in flow can lead to an increase in phosphorus concentration. The relative importance of these two phenomena has not been explored (Young et al., 1999). Diatoms can uptake silicon to such an extent that silicon concentration can reach levels adverse to diatoms. They lead to death of diatoms and release of silicon in the rivers, whose concentration subsequently rises in river (Wall et al. - 1998).

Lastly, phytoplankton can interact with other biotic elements, which can modulate its response to eutrophication. Indeed, phytoplankton in shallower rivers can suffer from shading by macrophytes whereas in deeper rivers, the situation is reversed, with phytoplankton able to shade macrophytes and phytobenthos (Balbi, 2000). Indeed, the bloom of phytoplankton is considered to end the peak of phytobenthos (Roos, 1983). Dense macrophytes can slow the flow locally, thus sedimenting phytoplankton, and can accommodate zooplankton. Phytoplankton biomass can also be affected by grazing by zooplankton and filtering by invertebrates and lamellibranches (Basu et al., 1997). This predation can severely reduce the phytoplankton biomass, thus hiding the primary increase of phytoplanktonic biomass.

Existing methods to investigate the relationships

Chlorophyll a

It is the most used parameter to estimate the phytoplankton biomass. It is part of most of the monitoring systems. However, it has several limitations (Balbi, 2000): the phytoplankton species do not have the same ratio biomass/chlorophyll a, it does not distinguish suspended periphytic algae and 'true' phytoplankton and it may measure chlorophyll a from dead algae.

Biomass of phytoplankton estimated from the volume of the algae is more resource demanding but can include quantitative information on the species composition.

Cell count

Several studies (Jarvie et al., 2003, Marker et al., 1997, Stevenson et al., 1995, Wehr et al., 1997) used the cell density, measured by counting the phytoplankton cells in a volume of water. It is a measure of phytoplankton biomass. However, it is not as used as the chlorophyll a concentrations as it is more time consuming.

Species composition

Several studies (e.g. Noppe et al., 1997, Reynolds et al., 2002) have focussed on phytoplankton composition. Assemblages have been determined through principal component analysis. However, assemblages evolve seasonally, and the studies of phytoplankton assemblage have been limited in time and spatial scale.

How to establish the relationships

To establish the relationships between nutrients and phytoplankton biomass and species, three different approaches can be taken (see table 2):

- Expert knowledge. It is usually used to attribute nutrient preferences to algal species.
- In a laboratory, study the responses of phytoplankton species to addition of inorganic nutrients.
- Monitoring of rivers and its conditions and analyse the results to find relations between the phytoplankton biomass and composition and nutrient conditions. This last method was the most used.

Laboratory studies can be applied to samples of river water, with its full range of species or to a limited number of species. All the conditions are set by the protocols and regulated. They have been used to investigate the relative effect of light and nutrients (Guelda et al., 2004), the response to nutrient addition in nutrient-limiting conditions or nutrient-sufficient conditions (Smoot et al., 1998), determine the metabolic rate of phytoplankton (de Andrade et al., 1998) or study the response to nutrient enrichment (Mallin et al., 2004, Mallin et al., 2000, Olguin et al., 2004).

Different methods of analysis can be used to study the link between nutrient conditions and the phytoplankton biomass or composition.

The simplest one is the simple regression between phytoplankton biomass (cell density or chlorophyll a) and either phosphorus concentration (total P or SRP) or nitrogen concentration (total inorganic N, NH_4 , NO_3 ...). This method has been used for example in Basu et al. (1997, Van Nieuwenhuyse et al. (1996) and Young et al. (1999). Multiple regression can also be applied (Wehr et al., 1997). It allows focusing only on nutrient concentrations.

Correlations between phytoplankton parameter and other water quality indicators take into account the effects of several water quality parameters at the same time (not only nutrients). It has been used in Garban et al. (1999), Ibelings et al. (1998), Puirso et al. (1997) and Stevenson et al. (1995).

More refined analysis of the relationship between phytoplankton and water quality include the grouping of the phytoplankton species according to their behaviour and link it with the water quality characteristics. It can be done using (detrended) canonical correspondence analysis, the Two Ways Indicator Species Analysis (TWINSPAN) and principal component analysis (Noppe et al., 1997, Olguin et al., 2004, Potapova et al., 2004, Ruse et al., 1997).

Artificial neural network is another technique that creates a black box model between input data (nutrients, light etc) and output data (assemblage composition, biomass etc) through a training procedure (Recknagel et al., 1997).

Use of relationships in models and indices

Models developed

From analysis above, number of different models can be created. Recknagel et al. (1997) presented different types of models, the analysis they are based on and what they are used to model/

- Empirical models and time-series analysis models
- Deterministic models
- Heuristic word models, fuzzy models and artificial neural network models

To model the overall biomass, empirical and time-series analysis models can be used. Empirical models are based on correlations between chlorophyll a and different factors such as nutrients. They can be used to predict mean seasonal or annual chlorophyll a concentrations. For modelling of chlorophyll a depending on time, time-series analysis models can be used. They are derived from multivariate analysis including limiting and multiple factors.

To model the composition of phytoplankton, different options exist. If the interest is in modeling qualitatively seasonal dynamics of phytoplankton, heuristic models can be used, built on causal knowledge on limiting, physiological and multiple factors. If the interest is in quantifying the pos-

sible occurrence of species, fuzzy models can be used. Artificial neural network is a similar approach used to predict both the timing and the magnitudes of algal species (see Hakanson et al., 2003). Ruse et al. (1997) used Generalised Linear Models to predict the response of phytoplankton taxa to variation in environmental data for the Thames for each season. Best predictions were achieved in spring. In this model, only nitrate concentration was used for the inorganic nutrients, as there were redundancies between nitrate, ammonium, nitrite, orthophosphate and nitrate:orthophosphate ratio.

Hakanson et al. (2003) explores the variability of biomass for 5 phytoplankton groups. It concluded that it is unlikely that a model for mean monthly chlorophyll concentrations in rivers could yield r^2 -values higher than about 0.6.

Table 4.4. Summary of some relations found between phytoplankton and inorganic nutrients

Biological element	Water quality element	Relation	Sites	Nutrient range	References
Chlorophyll a, January to spring chlorophyll maximum	Silicate NH ₃ NO ₃ -N TN SRP N/P ratio	Regression 0.71 (p<0.05) 0.43 (p<0.05) 0.25 (p<0.05) 0.20 (p<0.05) 0.09 (p<0.05) 0.15 (p<0.05)	River Nene (small lowland river), UK 11 sites, up to over 22 years	NH ₄ -N = 0-5.3 mg/L NO ₃ -N = 0.4-20 mg/L NO ₂ -N = 0-0.4 mg/L SRP = 0.1-4.4 mg/L	(Balbi - 2000)
Chlorophyll a, summer period	TP	Quadratic model log(Chl)=-1.65 + 1.99 (log TP) - 0.28(log TP) ² (r ² = 0.67) Chl in mg.m ⁻³ and TP in mg.m ⁻³	Temperate streams (mainly North America, but also Europe, Korea, Iraq and Australia) n = 292	TP = 5 - 1030 mg P/m ³	(Van Nieuwenhuyse et al. - 1996)
Chlorophyll a	NO ₃ SiO ₂	Pearson correlation coefficient: 0.91 (p=0.003) 0.87 (p=0.006)	Canojoharie Creek, Hudson River Basin, US	NO ₃ -N = 0.05-3.3 mg/L SiO ₂ = 1-5 mg/L	(Wall et al. - 1998)

To model the biomass of specific species or functional groups with time, deterministic models can be used. They are based on previous knowledge of trends, seasonality and dependency to limiting, physiological and multiple factors. Several deterministic models have been developed.

Cellular growth rate model depending on external concentrations of nutrients and their limitations follows the Monod model (Hecky et al., 1988, Krogstad et al., 1989).

SWAT2000 (Neitsch et al., 2002): models algal biomass through chlorophyll a, depending on light, nitrogen and phosphorus. The main objective of this model is the modelling of the quantity and quality of water from the fields to the river and the impacts of farming practices.

Figure 4.5. Equation of phytoplankton biomass from SWAT2000 (Neitsch et al., 2002)

$$\Delta a \lg ae = ((\mu_a \cdot a \lg ae) - (\rho_a \cdot a \lg ae) - (\frac{\sigma_1}{depth} \cdot a \lg ae)) \cdot TT$$

where:

$\Delta a \lg ae$ is the change in algal biomass concentration (mg alg/L)

μ_a the local specific growth rate of algae (day⁻¹)

ρ_a the local respiration or death rate of algae (day⁻¹)

σ_1 the local settling rate for algae (m/day)

$depth$ the depth of water in the channel (m)

$a \lg ae$ the algal biomass concentration at the beginning of the day (mg alg/L)

TT the flow travel time in the reach segment (day)

Effects of temperature, light, nitrogen and phosphorus limitations are represented in the growth rate μ_a as following:

$$\mu_a = \mu_{a,20} \cdot 1.047^{(T_{water} - 20)}$$

$$\text{with } \mu_{a,20} = \mu_{max} \cdot FL \cdot \frac{2}{\frac{1}{FN} + \frac{1}{FP}}$$

Where:

$\mu_{a,20}$ the local specific algal growth rate at 20°C (day-1)

μ_{max} the maximum specific algal growth rate (day-1)

FL the algal growth attenuation factor for light

FN the algal growth attenuation factor for nitrogen

FP the algal growth attenuation factor for phosphorus

With:

$$FN = \frac{(C_{NO3} + C_{NH4})}{(C_{NO3} + C_{NH4}) + K_N}$$

$$FP = \frac{C_{solP}}{C_{solP} + K_P} \text{ and}$$

Where:

C_{NO3} the concentration of nitrate in the reach (mg N/L)

C_{NH4} the concentration of ammonium in the reach (mg N/L)

K_N the Michaelis-Menten half-saturation constant for nitrogen (mg N/L)

Where:

C_{solP} the concentration of phosphorus in solution in the reach (mg P/L)

K_P the Michaelis-Menten half-saturation constant for phosphorus (mg P/L)

AQUATOX (Park et al., 2004): models phytoplankton, periphyton, macrophytes, invertebrates and fish depending on nutrients, organic matter, organic toxicant, oxygen, suspended sediments, sediments, temperature and discharges. It is applicable to streams, lakes and reservoirs.

RIVERSTRAHLER (Billen et al., 1994): combines an hydrological component based on the stream order and the AQUAPHY model. The AQUAPHY model models the chlorophyll a concentration and biomass evolution in each stream order depending on light, temperature, flow, ammonium, nitrate and phosphate concentrations. In Garnier et al. (2002), this model is further refined to distinguish between diatoms and chlorophyceae, to include predation by zooplankton and lamelli-branches and to model the interactions of nutrients across the sediment-water interface.

Figure 4.6. Equations of phytoplankton growth and nutrient uptakes in the RIVE model (Garnier et al., 2002)

$$\Delta PHY = \mu_{\max} \cdot \frac{S/PHY}{S/PHY + K_S} \cdot lf \cdot PHY$$

where :

ΔPHY the growth of phytoplankton
 μ_{\max} the maximum growth rate
 PHY the phytoplankton biomass

S the phytoplankton reserve
 K_S the half saturation est
 lf the nutrient limitation factor

With: $lf = \frac{PO_4}{PO_4 + K_{pp}}$ or $lf = \frac{NO_3 + NH_4}{(NO_3 + NH_4) + K_{pn}}$ or $lf = \frac{SiO_2}{SiO_2 + K_{psi}}$ (for diatoms only)

Where:

PO_4 the concentration in PO_4 in the reach
 K_{pp} the half saturation constant for P uptake
 NO_3 the concentration of NO_3 in the reach
 NH_4 the concentration of NH_4 in the reach

K_{PN} the half saturation constant for N uptake
 SiO_2 the concentration in SiO_2 in the reach
 K_{psi} the half saturation constant for Si uptake

The uptakes of nutrients are modelled as follow:

NH_4 uptake =

$$NH_4 \text{ uptake} = \frac{\Delta PHY}{cn} \cdot \frac{NH_4}{NH_4 + NO_3}$$

$$PO_4 \text{ uptake} = \frac{\Delta PHY}{cp}$$

$$NO_3 \text{ uptake} = \frac{\Delta PHY}{cn} \cdot \frac{NO_3}{NH_4 + NO_3}$$

$$SiO_2 \text{ uptake} = \frac{\Delta PHY}{csi}$$

Where:

cn the algal C/N ratio
 cp the algal C/P ratio

csi the algal C/Si ratio

Other models have been developed as well, for example the model developed by Thebault et al. (1999) that models chlorophyll a and biovolume depending on nutrients for the Lot river, coupled with an hydrological module, the STREAM model for steep and shallow streams (Park et al., 1996) and the model of phytoplankton and nutrients by McIntyre et al. (2003).

Indices and classifications

There is no index specific to phytoplankton. Species richness and similarity could be used but is currently not.

Species preferences for nutrient conditions have been explored in lakes by Reynolds et al. (2002) for 31 phytoplankton functional groups; some also occur in rivers and thus nutrient preferences could also be used.

4.5.3 Phytobenthos

Relationships between nutrients and phytobenthos

Whereas phytoplankton is considered to be a good indicator of eutrophication in larger rivers, phytobenthos is a better one for smaller rivers (Piiro et al., 1997) and is the primary energy source in many mid-sized (3rd to 6th order) streams (Stevenson, 1996).

Phytobenthos is a highly diversified biota (Stevenson, 1996), with different types of organisms (diatoms, filamentous algae, blue-green etc), growing on different substrates (rocks, soft riverbed, macrophytes etc) and developing different forms (filamentous, thin or thick mat etc). Phytobenthos is present in every type of rivers where light reaches the riverbed or surface of macrophytes.

As all autotrophs, phytobenthos development depends on availability of inorganic nutrients. An increase in inorganic nutrient (P and N) availability leads to accelerated growth of phytobenthos and an increase in biomass (Johnston et al., 1990, Mulholland et al., 1994, Snyder et al., 2002, Welch et al., 1992), with a shift from slow-growing, nutrient-sensitive species to fast-growing, nutrient-tolerant species (Biggs et al., 1998).

The response of phytobenthos to nutrient enrichment depends on whether the nutrient was limiting and the initial nutrient level. Indeed, Perrin et al. (1997) obtained little phytobenthic response to P addition, a moderate one when adding N, and an even higher one when adding both N and P (P has become limiting once N was added). Stanley et al. (1990) had a rapid response of phytobenthic biomass when the P level was initially low (<0.010 mg/L) and only a moderate one when ambient P was higher (0.015-0.025 mg/L). Flow and its variation can also play an important role on determining the response of phytobenthos. Indeed, the discharge stability over periods of less than a year govern the average phytobenthic biomass and nutrients will control the overall biomass only over prolonged periods of stability with moderate to low flow (Biggs, 1996). So, a small increase in dissolved nutrients will greatly increase the frequency of high biomass events if there are infrequent floods and accrual periods over 100 days (Biggs, 2000).

In term of species richness, nitrogen and phosphorus enrichment have been found to increase the species richness in oligotrophic streams up to a point where competitive exclusion occurs and species richness decreases (Marcus, 1980, Snyder et al., 2002). The way nutrients are delivered, as for phytoplankton (Hecky et al., 1988), impact on how the community assemblage will respond. Nutrient pulses can lead to higher richness, with a mix of species of different nutrient preferences, as found in Finland rivers (Soininen, 2002).

Kjeldsen (1994) underlined a different response of phytobenthos peak biomass to phosphorus enrichment depending on the riverbed substrate: on fine-grained sediments phosphorus was the main driver of the biomass peak, whereas on stony substrate it only gives the maximum potential biomass and the actual peak biomass is determined by a range of other factors.

In Hecky et al. (1988), optimum ratio N/P for species are given. The N/P ratio in the water column favours species whose N/P ratio is the nearest to the ratio of water. However, Snyder et al. (2002) underlined that the ratio N/P only indicates potential N or P or N and P limitations when the ab-

solute concentrations are low. Otherwise, N and P are not limiting and the ratio N/P is not significant.

Estimations of biomass level biomass that hampers the aesthetic quality of rivers have been developed. In Welch et al. (1988), 100 mg chlorophyll a per square metre is considered to be the threshold over which development of phytobenthos, often as filamentous algae, is perceived unfavourably. Dodds et al. (1998) reviewed several studies of such thresholds. They range from 50 to 200 mg chlorophyll a per square metre, and mainly over 100 mg chlorophyll a per square metre. From this threshold, can objectives on nutrients be set (for example Dodds et al., 1997).

Phytobenthos can form mats on the riverbed that range from less than 100 μm to several millimetres thick (Sand-Jensen et al., 1991). Formation of thick mats is progressive when not disturbed by spates. With age, the mats thicken, gradients of nutrients and water quality between the inside of the mat and the water column form. Exchanges between the mat core and the water column occur by diffusion; they depend on the mat thickness and the renewal of water in contact with the mat (function of flow). Diffusion and uptake rate are stimulated by increase in phosphorus in the water column up to a point where they are both limited by the mat thickness (Horner et al., 1990).

The mat formation leads to a change of species, depending on their position within the mat, due to light and water quality changes. The evolution of the mat is described in Sand-Jensen (1983). If the early stages of the mat have a good correlation between water quality and the mat biomass (thin mat, uniformly in contact with the water column), it is not the case for more mature stages. Indeed, the lower parts of the mat are progressively separated from the water column as the mat becomes too thick for good diffusion. The internal processes become more important than exchange with the water column (Mulholland et al., 1994, Sand-Jensen, 1983).

The mat also forms a frontier between the water column and the sediments, where nitrification and mineralisation occur: the mat, especially the lower parts, has access to new sources of nutrients, separate from the water column. This weakens further the link between phytobenthos biomass and water column nutrients.

Nutrient cycling and recycling within the mat also occurs, another source of confusion with the effect of the water column concentrations. This explains why the uptake rate of nutrients in the water column by phytobenthos is inversely proportional with the phytobenthic biomass (Horner et al., 1990). The mat structure can also affect the amount of light that reaches all algal cells. Keldsen (1996) showed that an increase in phosphorus did not lead to an increase in biomass due to self-shading.

Impacts of nutrients may be affected by other variables. Indeed, light, discharge and grazing by snails and benthic invertebrates are three main drivers of phytobenthic assemblage and biomass (Sand-Jensen, 1983).

Kiffney et al. (2000) showed that light was the single best indicator explaining the variation in peak chlorophyll a biomass. Light varies seasonally and the quantity that reaches the riverbed depends on the water depth (Sand-Jensen, 1983)- function of discharge and river width - , suspended solids and shading by riparian vegetation, phytoplankton and macrophytes.

Discharge and its variation conditions the growth forms and composition of phytobenthos and, during spates, scours a significant amount of biomass, transported downstream. The effect of spates on removing biomass depends on the frequency of spates and the substrata. The more frequent the spates, the less biomass is removed, as the mat is thinner and composed of species resis-

tant to scouring. Phytobenthos is more sensitive to spates on loose substrata than stony ones, as it is more unstable (Pringle, 1990). Flow conditions the potential for colonisation by new phytobenthos (Biggs et al., 1998).

Anderson et al. (1999) showed that grazing controlled biomass more than enrichment in soluble reactive phosphate whereas Walton et al. (1995) pointed out that grazing lead to a shift in species whatever the enrichment was and grazing is the third dimension in the habitat matrix developed by Biggs et al. (1998) with an effect similar to spates. The effect of grazing on the phytobenthos biomass and composition is affected by the type of grazers, its abundance and the initial composition of the phytobenthos.

Seasonal variations of irradiance and discharge leads to the exponential increase in spring, as irradiance increases and discharge decreases (Sand-Jensen, 1983), and regulated by a number of other factors including nutrients and shading afterwards.

Most of the analyses have been carried out either on diatoms (when considering species composition) or global biomass (with chlorophyll a concentration per square metre). However, few diatoms live in cold water and at high altitude whereas cyanobacteria and green algae are more common, as demonstrated by the recent work of Lindstrøm et al. (2004) on a Norwegian watercourse.

Phytobenthos interacts with the other biota through different mechanisms. Phytoplankton and macrophytes can shade the phytobenthos, thus preventing its growth. Epiphyton (taxa that develop on macrophytes) interact with macrophytes; there is no epiphyte where there is no macrophyte and a high development of epiphytes can shade macrophytes to a point that macrophytes die (ten Cate et al., 1991). At last, dense growth of phytobenthos can cause a higher instability of oxygen concentrations (higher during the day, lower during the night) that can be unfavourable for other organisms like macroinvertebrates and fish (ten Cate et al., 1991).

Existing methods to investigate the relationships

The algal population and community structure can be assessed with the total biomass and the species composition (Stevenson, 1996).

Biomass

Biomass of phytobenthic communities can be estimated by the chlorophyll a per square metre, the dry mass, the ash-free dry mass, the cell density and the biovolume. Chlorophyll a per square metre is the most commonly used as it is easy to use and not as labour-intensive. All of these techniques have inherent limitations (Stevenson, 1996).

Species composition

Species composition is assessed by recording the number of species/taxa present in the community and their relative abundance. Different indices can be calculated from these measures to reflect the species richness, the species evenness and the similarity of species among communities. The Shannon-Wiener functions have been widely used to reflect the species richness and evenness whereas numerous diversity indices based on the Zelinka and Marvan formula (Zelinka et al., 1961) have been developed to indicate both richness and evenness of communities.

How to establish the relationships

The relationships between inorganic nutrients and phytobenthos can be studied via three different methods (see table 2):

- Laboratory bioassays
- In-situ enrichment experiments
- Monitoring of both phytobenthos and water quality and then using empirical method to make the link.

Laboratory bioassays consist for phytobenthos in studying it in experimental streams. They have been used to analyse the effects of flow and nutrients on phytobenthos biomass (Horner et al., 1990, Humphrey et al., 1992), the effect of phytobenthos on nutrients (Anderson et al., 1999, Marinelarena et al., 2001, Mulholland et al., 1994), the relative effect of substrata enrichment and water column enrichment (Pringle, 1990) or the interactions with grazers (Hill et al., 1992, Kiffney et al., 2001, Walton et al., 1995). Nutrient-diffusing substrata have also been used to study the interactions between nutrient enrichment and light or grazing (Flecker et al., 2002, Rosemond, 1993).

In-situ enrichment experiments can be through a whole river fertilisation or using nutrient-diffusing substrata. Whole river fertilisation has mainly been used in several arctic streams to study the long-term effect of fertilisation on phytobenthos and other biota (Harvey et al., 1998, Slavik et al., 2004) and to other streams (Bernhardt et al., 2004, Biggs et al., 2000). Nutrient-diffusing substrata are more commonly used, to determine which of phosphorus and nitrogen is limiting and the spatial repartition of nutrient limitations (Henry et al., 2003, Scrimgeour et al., 1997) and/or to study the effects of enrichment on phytobenthic biomass (Rinke et al., 2001) and light/shading (Mosisch et al., 2001).

River monitoring is the most commonly method used to analyse the link between phytobenthos biomass and composition and environmental conditions (including nutrient levels). Once the phytobenthos and the water quality have been measured, similar techniques as for phytoplankton and macrophytes can be used, from the simplest to the most complex.

Simple regressions have been used in Biggs (2000) to link the mean monthly chlorophyll a and nutrients and days of accrual (reflect the effect of flow variation). Multiple-regression, sometimes with stepwise selection, have been used as well in Biggs (2000) and Kiffney et al. (2000). Regression are usually used to link biomass with nutrients.

To include a wider range of water quality variables (and not only nutrients), correlations can be used for the phytobenthic biomass or for diatom indices and environmental characteristics for example in Aizaki et al. (1988) Chetelat et al. (1999), Dell'Uomo et al. (1999), Gurbuz et al. (2002), Hill et al. (2001), Kelly et al. (1995a), Kwandrans (2002), Snyder et al. (2002), and Stewart (1987).

More complex analysis can explore species preference and the existence of similar behaviour between taxa and river sites, through ordination and clustering techniques such as TWINSpan (Leland - 1995), canonical correspondence analysis (Griffith et al., 2002, Harding et al., 1999, Munn et al., 2002, Winter et al., 2000), detrended correspondence analysis (Soininen et al., 2002), principal component analysis (Prygiel et al., 1993, Vilbaste et al., 2003) and other methods.

Use of relationships in models and indices

Models developed

Regression models have been developed between the phytobenthic biomass and the concentration in nutrients, for example in Biggs (2000) and Winter et al. (2000).

Table 4.5. Some examples of relations found between phytbenthos and inorganic nutrients

Biological element	Water quality element	Relation	Sites	Nutrient range	References
Chlorophyll a (in mg.m ⁻²)	TP, in µg.L ⁻¹ TN, in µg.L ⁻¹	log Chl a = 0.905 log TP + 0.490 (r ² =0.56, p<0.001) log Chl a = 0.984 log TN - 0.935 (r ² =0.50, p<0.001)	33 sites, 13 rivers of Ontario and Quebec (Canada)	TP = 6 - 130 µg.L ⁻¹ TN = 179 - 2873 µg.L ⁻¹	(Chetelat et al. - 1999)
Mean monthly benthic algal biomass In mg/m ² chlorophyll a	SIN SRP	Log ₁₀ (chla)= 0.109+0.483.log ₁₀ SIN (r ² =0.122, p=0.057) Log ₁₀ (chla) = 0.468 + 0.697 log ₁₀ SRP (r ² =0.226, p=0.008)	30 sites, 25 streams, New Zealand	SIN = 6.2 - 232 mg/m ³ SRP = 1.3 - 31.6 mg/m ³	(Biggs - 2000)
Max monthly benthic algal biomass In mg/m ² chlorophyll a	SIN SRP	Log ₁₀ (chla)= 0.711 + 0.688.log ₁₀ SIN (r ² =0.325, p=0.001) Log ₁₀ (chla) = 1.400 + 0.797 log ₁₀ SRP (r ² =0.295, p=0.002)	30 sites, 25 streams, New Zealand	SIN = 6.2 - 232 mg/m ³ SRP = 1.3 - 31.6 mg/m ³	(Biggs - 2000)
Diatom taxa richness	TN	Spearman correlation: 0.28 (p<0.01, diatoms at genus level) 0.42 (p<0.001, diatoms at species level)	199 streams, Mid-Appalachian streams, USA	Not indicated	(Hill et al. - 2001)
% dominance (diatoms)	TP	Spearman correlation: -0.41 (p<0.01, diatoms at genus level) -0.45 (p<0.001, diatoms at species level)	199 streams, Mid-Appalachian streams, USA	Not indicated	(Hill et al. - 2001)
TDI	NH ₄ -N	Pearson correlation coefficient 0.430 (p < 0.001)	9 sites, for Vistula river, Poland	NH ₄ -N = 0.2 - 3.0 mg/L NO ₃ -N = 0.2 - 3.0 mg/L PO ₄ -P = 0.05-0.55 mg/L	(Kwandrans - 2002)
Diatom richness	NH ₄ -N TP	Spearman correlation coefficient 0.55 0.64	11 rivers in Idaho, USA	NO ₃ -N + NO ₂ -N = 0.002-1.61 mg.L ⁻¹ NH ₄ -N = 0.005-0.037 mg/L	(Snyder et al. - 2002)

Multivariate analysis allowed predicting the occurrence of phyto-benthic genus at a site under near pristine condition (Chessman et al., 1999), following the RIVPACS method, or establishing diatom-nutrient transfer functions (Schonfelder et al., 2002). Welch et al. (1992) developed a model of biomass (chlorophyll a per square metre) depending on nutrients calibrated with laboratory data. However, biomass levels were overestimated due to a high macroinvertebrate grazer density.

In term of dynamic models, the uptake of nutrients by phyto-benthos cells depending on external nutrient concentrations can be modelled with Michaelis-Menten kinetics (Kjeldsen, 1994). The analysis of peak biomass of benthic algae in 10 Danish lowland streams over 5 years by Kjeldsen (1994) outlined that on fine grained sediments, the dissolved inorganic phosphorus explains 61% of the variation of the peak algal biomass of phyto-benthos, according to the following equation: $Y=929.3.x/(49.2+x)$ ($r^2=0.61$), with Y the maximum biomass (mg chlorophyll.m⁻²) and x the dissolved inorganic phosphorus (µg P.L⁻¹, 1 - 273 µg P.L⁻¹). However, it has been applied only to streams where the hydrological conditions were stable. And it only applies to sites with fine-grained sediments: no significant correlation between the peak algal biomass and the dissolved inorganic phosphorus concentration was found on rocky substrata. More recently, Flipo et al. (2004) developed a model of in-stream river processes and planktic and periphytic biomass.

AQUATOX (Park et al., 2004) models the biomass of periphyton per square metres depending on light, nutrient, velocity and substrate availability.

Indices and classification

Numerous attempts at defining the indicator values of individual diatom species have been done, either based on expert knowledge (Kelly et al., 2001, Kelly et al., 1995b, van Dam et al., 1994) or through methods of data analysis (Munn et al., 2002, Schonfelder et al., 2002).

These indicator values have then been used to create a number of indices that include this indicator value and the relative abundance of each taxa: Specific Pollution Sensitivity Index (Cemagref - 1982), Generic Diatom Index (Coste et al., 1991), Descy's Index (Descy, 1979), Sládeček's index, Leclercq & Maquet's index (Leclercq et al., 1987), Trophic Diatom Index (Holmes et al., 1999, Kelly et al., 2001, Kelly et al., 1998) and EPI-D (Dell'Uomo, 1997, Dell'Uomo et al., 1999) are some of them. They usually only consider diatoms. The Periphyton Index of Biotic Integrity (PIBI) developed by Hill (2000) is one of the few indices to include other type of benthic algae. However, it is limited to only 2 blue-green taxa.

A classification of the river sites has been developed for the UK based on the Trophic Diatom Index (TDI) as an indicator of eutrophication (Kelly, 1998). The TDI values range from 0 (indicating very low nutrient concentrations) to 100 (indicating very high nutrient concentrations). It is complemented with the percentage of pollution tolerant valves, to estimate the relative influence of organic pollution to eutrophication.

Figure 4.7. Equation to estimate the Trophic Diatom Index (TDI) (Kelly et al., 2001)

$$TDI = 25 \times \left(\frac{\sum a_j \cdot s_j \cdot v_j}{\sum a_j \cdot v_j} - 1 \right)$$

Where:

- a_j the abundance of species j
- s_j the pollution sensitivity of the species j (1-5)
- v_j the indicator value of the species j (1-3)

In Italy, the EPI-D is considered suitable to diagnose inorganic nutrients, organic matter and minerals impacts on phyto-benthos (Dell'Uomo et al., 1999).

Figure 4.8. Equation to estimate the Eutrophication/Pollution Index based on Diatoms (EPI-D) (Dell'Uomo, 1997, Dell'Uomo et al., 1999)

$$EPI - D = \frac{\sum a_j \cdot f_j \cdot i_j}{\sum a_j \cdot f_j}$$

Where:

- a_j the relative abundance of the species j (1 to 5, increasing abundance)
- f_j the reliability of the indicator (1, 3 and 5: increasing reliability)
- i_j the eutrophication/pollution index characteristics of the species j

Table 4.6. Values of the EPI-D and status of rivers (Dell'Uomo, 1997)

EPI-D	Status	EPI-D	Status
0.0 < EPI-D ≤ 1.0	Excellent	2.0 < EPI-D ≤ 2.2	Moderately polluted
1.0 < EPI-D ≤ 1.5	Good	2.2 < EPI-D ≤ 2.5	Strongly polluted
1.5 < EPI-D ≤ 1.8	Fairly good	2.5 < EPI-D ≤ 3.0	Heavily polluted
1.8 < EPI-D ≤ 2.0	Weakly polluted	3.0 < EPI-D ≤ 4.0	Completely polluted

Lastly, Biggs (2000) developed a classification depending on soluble reactive phosphorus and soluble inorganic nitrogen, and the number of days of accrual in gravel/cobble-bed streams, the boundaries being determined by the 2 following nuisance levels of chlorophyll a 60 and 200 mg chlorophyll a per square metre.

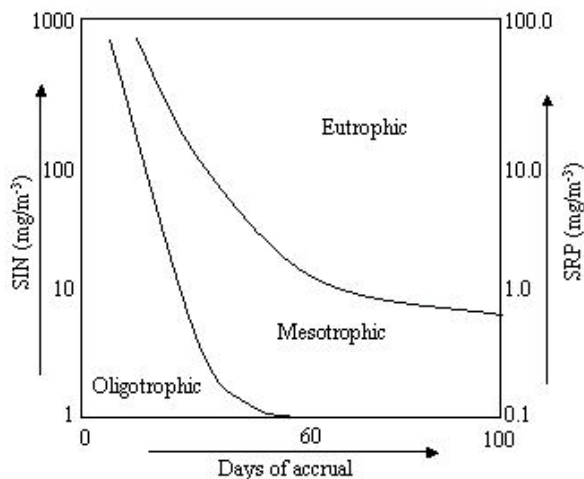


Figure 4.9. Schematic classification as developed by (Biggs, 2000) (simplified)

The boundary between oligotrophic and mesotrophic is set at 60 mg/m² chlorophyll a and the boundary between mesotrophic and eutrophic is set at 200 mg/m² chlorophyll a.

Macroinvertebrates and fish

Because of their longer lifetime, benthic invertebrates and fish are considered as good integrators of multiple pressures over several months (macroinvertebrates) to several years (fish). Macroinvertebrates and fish have been extensively studied; numerous habitat models like RIVPACS have been developed for macroinvertebrates and an important number of indices have been created. However, because of this integration capacity, the specific effect of eutrophication is difficult to distinguish. Thus, Kelly et al. (1998) advised that benthic invertebrates should not be used to assess eutrophication status in rivers.

Inorganic nutrients do not have a direct impact on fish and benthic invertebrates, except for the worst cases of eutrophication when toxic concentrations of ammonia and very low levels of dissolved oxygen occur. These two situations can lead to the death of sensitive species, affecting the species composition and the age structure.

The main impacts of inorganic nutrient enrichment are through the change of habitats and through the food web.

The changes of habitats caused by eutrophication include the smothering of gravel bed by phyto-benthos and an increase (or a disappearance) of macrophytes. Smothering of the riverbed can deprive benthic invertebrates from their habitat, an increase in macrophytes biomass can lead to an increase in macroinvertebrates by limiting predation and exclusion of the bigger fish. As macroinvertebrates have preferences for some macrophyte species (Humphries, 1996), change in macrophyte composition can impact on the macroinvertebrate composition.

In term of biomass, increase in autotrophs biomass can lead to an increase of the invertebrates and fish that feed on them through the food web. It can have a fast impact on the macroinvertebrate biomass (Ramírez et al., 2004) and fry biomass. It can switch the food web from a bottom-up limitation to a top-down one (Deegan et al., 1997). Models of food webs can be of use to analyse the effective effects of eutrophication.

4.6 General conclusions

Qualitative effects of inorganic nutrient enrichment on autotrophs are well understood. In summary, nutrient enrichment leads to a general increase of biomass, if the other environmental conditions allow it, and a change in species from nutrient-sensitive toward more nutrient-tolerant species. In term of species richness, enrichment of oligotrophic streams can lead to an increase in species richness until the enrichment reaches a threshold over which competitive exclusion of nutrient-sensitive species decrease the overall species richness. For all autotrophs, the effects of nutrient enrichment depends on the initial enrichment of the stream: in oligotrophic streams, even a slight enrichment can lead to high increase in biomass and alter the species assemblage, whereas it may not cause any change in an already eutrophic stream.

Knowledge of quantitative effects of nutrient enrichment is more variable. Numerous studies have been carried out, focussing on various biological elements and representations (e.g. chlorophyll a/m², TDI, species richness etc for phytobenthos) and on various inorganic nutrients (SRP, TP, NO₃-N, TN, NH₄-N etc). Quantitative relations between inorganic concentrations and autotrophs have usually been carried out for biomass, a few times on indices. When assessing species assemblages, nutrients may be included but their relative effect is masked by the other habitat variables also included in the analysis. In a number of papers, the nutrient ranges of the sites were not indicated, making it hard to judge the applicability of the relations found to other rivers. And few papers actually consider the nutrients from sediments whereas macrophytes can uptake a significant amount of nutrients from their roots and phytobenthos can be directly in contact with them.

In term of assemblages, most of the scientific efforts have focussed on diatoms and macrophytes; little work has been done on phytoplankton assemblages. From these studies, indices, and classifications based on these indices, have been developed for diatoms and macrophytes. They all have in common the fact that they integrate the relative abundance of each species and their respective tolerance to nutrients.

Phytoplankton is considered to be a good indicator of eutrophication in slow-flowing deep lowland rivers whereas phytobenthos would be a good one for all other types of rivers. The position of macrophytes is not so clear. Indeed, link between macrophytes and inorganic nutrients is not as strong as for phytoplankton and phytobenthos and macrophytes' biomass and composition would be mainly driven by the hydromorphology characteristics of each site along the river. Macroinvertebrates and fish can be influenced by nutrient enrichments. However, they are rarely directly influenced by nutrient concentrations but more through repercussion of changes in the food web and the habitat.

Modelling of biomass of phytoplankton is the most developed modelling. Some models of diatoms' and macrophytes' biomass have also been developed. However, there are very few models for the species composition, especially for phytobenthos other than diatoms and phytoplankton. And the models developed may apply only to restricted areas. Their validity when upscaling should be tested.

It is usually assumed that phosphorus is the limiting nutrient for autotroph growth. Now, this idea has to be reconsidered due to the number of cases where nitrogen has been found limiting. The effects of the different forms of nitrogen and phosphorus and the relative effects of sediments and water column on macrophytes and phytobenthos have received little attention.

When determining species preferences or when analysing the effects of water quality on biota, effects of inorganic nutrients and organic pollution are usually confounded. Distinguishing between these two pressures would be fundamental in determining what to do to achieve a good status.

Few studies have focussed on the recovery of the river after reduction in either phosphorus or nitrogen (or both) sources. This aspect should be considered fundamental in determining the measures Members of State need to implement to achieve a good status.

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5 Combined pressures and geographical context

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5.1 Introduction

The purpose of this activity is the description of relations between a combination of pressures coming from human activities (agriculture, urbanisation...) and ecological status in European rivers, with a largescale approach. However, across Europe, the natural and human geographical contexts are very diverse, and thus the relationship between anthropogenic pressures and ecological status may vary according to the sensitivity of river ecosystems and combinations of pressures. A regional framework related to the dominant ecological processes could be useful to properly identify the strength of the pressures / impacts relationships.

As a first step this chapter is a review of the relevant literature on this kind of models, linking pressures and river ecological status (through biological indicators) at large scales, with emphasis on the possible variability related to the geographical context.

5.1.1 WFD requirements

The EU Water Framework Directive requires the "identification of significant anthropogenic pressures and the assessment of their impacts on water bodies" (Annex II, WFD). These ecological impacts are not only determined by clearly identified point sources discharges, but also by series of complex human influences including diffuse pollution, alteration of water and sediment regimes, hydromorphological changes, connectivity breaks etc. (Borchardt & Richter, 2003).

"Ecological quality is integrating all pressures and showing the overall status of the ecosystem." (EEA 2003b). In most cases various pressures are acting simultaneously, and managers have to define a hierarchy amongst these to identify priority actions. Moreover, these pressures are not evenly distributed on the territory, and decisions makers have to decide where to act in priority. Finally, pressures are generated by different kinds of human activities, and integrated policies to restore river ecosystems must be tailored towards whole socio-economical structures, such as agriculture, industry, urban areas, etc. This means that an integrated approach and policy should include possibilities to have measures differentiated according to the nature of the different water-bodies and to the socio-economic conditions within the catchment.

5.1.2 A large scale view

In this sense, a large-scale view is necessary to take political decisions. A successful implementation of the WFD will require a common framework and policies for large territories, what needs scientific support and tools.

However, from a scientific point of view, the problem is complex for two reasons :

1 - The criteria to identify and assess pressures must be based on available data. But many pressures are difficult to identify and measure, especially when working in large territories. In most cases, we have not the same level of information upon the various kinds of pressures, such as chemical and hydro-morphological, point sources or diffuses, at the local or basin scale. For this

reason, it is often necessary to rise up at the level of the driving forces to get spatially homogeneous information. For example, it is very difficult to determine in a large basin the amount of pesticides and nutrients rejected, the sediment yield due to erosion, the degree of river channelisation etc., but we have access to the percentage of land dedicated to one kind of agriculture that generates these pressures.

2 - In large spatial contexts, the influence of scales when studying ecological processes is a key issue. One important problem is the upscaling of ecological relationships : how a model developed at a local scale can be safely extrapolated to larger areas? Another one is related to the interactions between natural characteristics, that determine the sensitivity of river ecosystems, and anthropogenic alterations: has a given pressure the same impact in different regions?

To address this question, it is necessary to study pressure / impact relationships in geographically homogeneous areas, so as to identify the relative influence of human and natural drivers of ecosystem responses. The proper way for doing that is to work at the level of “**hydro-ecoregions**”, considered homogeneous as regards the natural control factors (geology, relief, climate..) of aquatic ecosystems.

5.2 Combined pressures : Driving forces

Following the DPSIR concepts, the *driving forces* are human activities reflected by land use (agriculture, urbanization) ; these activities generate a combination of *pressures* (pollution discharges, physical structures...) which alter the abiotic components of the ecosystem (hydro-morphology, physico-chemistry,...). These alterations have an impact on biological communities that determine the *ecological status*. Agriculture and urbanization are considered the principal impairment sources in the literature, but the relative influence of combined pressures coming from agricultural or urban land use is not well established.

Agriculture

Agriculture is addressed by the EEA (2003b) as a significant driving force in terms of ecological quality by the way of various pressures : increased sediment yield (especially siltation), nutrients and organic pollution, hazardous substances, as well as indirect hydromorphological modifications like river channelization, habitat fragmentation, water abstractions, etc. "The impact of agriculture on Europe's water will have to be reduced if good surface and groundwater status are to be achieved" (EEA, 2003b).

Especially intensive agriculture is designated by many authors as a cause of degradation of stream habitat (Allan et al., 1997, Roth et al., 1996). In agricultural systems, the loss of vegetation cover may result in altered hydrological regime (Cormier et al. 2000), increase of erosion and hence in sediment yield, etc. Livestock influences especially N in rivers.

The biological effects of agriculture have been noted by several authors (Bis et al. 2000) : decrease in the number of species (taxa richness) and abundance of fish and invertebrates, shift in communities composition for algae (Cuffney et al., 2000).

Urbanization

The primary impacts of urbanization are related to direct point sources pollution, due to raw or treated sewage inputs. Pollution problems are well known by the managers, and most of the financial investments are dedicated to reduce sewage pollution.

However, many others pressures are related to urban areas. Housing, roads and other infrastructures constitute impervious surfaces. Increasing impervious area may result in altered hydrologi-

cal regimes : changes in timing, frequency, duration and magnitude of flood events (Cormier et al., 2000), with hydro-morphological and biological consequences. Untreated storm water runoff may cause also acute problems of organic or toxic pollution. And finally, many channelisation works are dedicated to the flood protection of urbanized areas.

More specific problems are related to large industries, energy production, navigation, tourism, etc. However these problems are generally well known by the managers, and depend on local specific measures, or will induce the designation of heavily modified water bodies with adapted environmental objectives. These pressures concerning generally large rivers are not easily integrated in a large scale view, and deserve a more targeted approach.

In most cases, it is impossible to get at large scales a precise information on these various pressures; but analysing in a first approach the relative impacts of agriculture and urbanization can provide to the decision makers, at the district or state level, useful information for the choice of management priorities.

5.3 Models

We have looked for studies linking biological indicators to pressures by rising up at the driving forces level ; in the case of combined pressures, this level is represented by land use. The aims are to identify 1) the main drivers of ecosystem alteration, 2) their relative importance, 3) the effect of spatial scale on the relationships, 4) the possible effect of the natural geographical context, and 5) the existence of impact thresholds.

A selection has been made taking into account the number of sites considered in the work, what gives us an idea of the spatial coverage, and local studies were not considered. Nevertheless the cases with more than 30-40 studied sites are rare.

5.3.1 Quality element and metrics

The impact of combined pressures is generally looked at through the response of fish or macroinvertebrates, and sometimes algae. Physical habitat state is also often measured insofar as it influences biota.

Generally, authors use multimetric indices able to detect a wide range of alterations. For communities, the multimetric index IBI (Index of Biological Integrity, Karr 1981) is the most commonly used (Allan et al. 1997, Cormier et al. 2000, Cuffney et al. 2000, Lammert and Allan 1999, Stewart et al. 2002, Wang et al. 2001). Benthic macroinvertebrate communities are assessed through classical quality index like BMWP (Bis et al. 2000) and EPT (Stewart et al. 2001), or also by multimetric indices like the ICI (Cormier et al. 2000). Diversity indices (Shannon, Hilsenhoff) are sometimes used (Wang et al. 2001, Wang & Kanehl 2003). For diatom assemblages, Pan et al. (2004) measure several autoecological metrics (based on Lange-Bertalot 1979 and Van Dam 1994), as well as the Trophic Diatom Index. Habitat quality is measured by Habitat Index (Roth et al. 1996), or US-EPA protocols (Dovciak & Perry 2002). Many of these metrics and indices and more are discussed in the other chapters of this report.

5.3.2 Examples of European compilations

At the European level, successive studies of the European Environment Agency attempted to evaluate the global status of river quality and the intensity of main pressures (EEA 1991, 2003b). In the most recent one is developed a draft core set of indicators for water concerning five sectors : agriculture, energy, fisheries, tourism and transport. One indicator is "length of rivers less than good quality in national classifications", but this status is not related to the pressures, and it is

clearly stated that "none of the national classification schemes meets the WFD requirements, so there is not at present information enabling to assess the situation in relation to the objectives of the directive. Different types of schemes cannot be quantitatively compared" (EEA 2003b).

5.3.3 Existing models and relationships

We are not aware, within Europe, of large scale models linking driving forces (land use) to biological indicators, although works could exist in the "grey literature". We review here some selected examples of studies done abroad, even if their validity for European countries is untested.

The methods used are quite diverse, but most of the literature reviewed relies on regressions and multivariate analysis, while more sophisticated statistical models are scarce.

In one of the pioneer works, Steedman (1988) evidenced positive relationships between the fish communities (IBI) and the percentage of forest in the basin, the proportion of channel with riparian forest, and a strong negative impact of watershed urbanization. He emphasised also the necessity of more detailed evaluation of agricultural land use, and streamside vegetation to improve predictive models. These are still open issues.

5.3.4 Spatial scale : Basin vs riparian effect

Land use was found to be a strong predictor of biological and habitat integrity (Allan et al., 1997). But studies of the scale effect (basin vs riparian) led to apparently contradictory conclusions.

In search of "effective scale for stream management" in a large basin, Dovciak & Perry (2002) tested the effectiveness of two landscape classification systems (watershed vs. agroecoregion) in explaining habitat and invertebrate features. They observed that local habitat conditions, which strongly influence invertebrates assemblages, are determined by regional scale landscape factors.

In a literature review, Gergel et al. (2002) aimed to demonstrate how landscape indicators (amount and arrangement of land cover, spatial distribution of patches etc. derived from aerial and satellite images and mapping) complement traditional riverine indicators, resulting in a broader perspective on rivers monitoring. They reviewed various studies relying landscape and biological indicators at different spatial levels (catchment, riparian zone, nearshore), and concluded that "a variety of investigators have tried to determine the spatial extent, or distance from the water body, over which landscape patterns influence water quality or aquatic biota, yet this question remains unresolved".

Sponseller et al. (2001) found in nine headwater basins that "water chemistry was generally related to features at the catchment scale", while temperature, substratum and macroinvertebrates were strongly influenced by the land-cover patterns of a 200m riparian corridor. Stewart et al. 2001 found strong relations between both watershed and riparian corridor land cover versus fish and invertebrates assemblages in 38 streams.

For Roth et al., (1996), the regional share between agricultural and forested areas is the primary determinant of stream conditions (IBI, HI), while riparian vegetation is a weak secondary predictor of stream integrity. On the other hand, Lammert & Allan (1999) found no relationship between regional land use and biological integrity ; only habitat and immediate land use (100 m buffer) predicted biotic integrity. In this study, sites were located close together and regional land use did not differ greatly. So the contrasting results may be the consequence of differing scales of the study design, coupled with the differences in the scale of the various effects (sediment control, etc.) (Allan et al. 1997).

In a study of 172 sites, Meador & Goldstein (2003) concluded that water physico-chemistry and riparian condition may be better indicators of fish community condition than basinwide land use.

In agricultural area, the fish index is closely related to the riparian condition index, but this relationship is weaker in urban areas. They emphasised "the universal importance of riparian zones to the maintenance and restoration of diverse fish communities in streams"

Urban vs agricultural land use

Urban and agricultural land use are the most studied driving forces, and one or the other are considered by some authors as the most impairing.

Negative relationships between the percentage of agricultural area in the watershed and fish communities indices were found by Roth et al. (1996) and Walser & Bart (1999). Cuffney et al. (2000) evidenced impacts on fish, invertebrates and algae along an agriculture intensity gradient. Pan et al. (2004) found weak linkages between agricultural land use and lotic diatoms communities during summer baseflow. Longer temporal scale is integrated by Harding et al. (1998), who related the present-day diversity of stream invertebrates and fish to the influence of past land use (1950).

Negative correlations between urbanisation and invertebrate diversity indices is shown by Stepenuck et al. (2002), who study the invertebrates response through a gradient of imperviousness caused by urbanisation. A similar negative effect of imperviousness is evidenced by Wang & Kanehl (2003). Roy et al. 2003 showed strong negative relationships between urban land cover and invertebrate indices, and described what habitat variables are most important in explaining this effect.

Among the few large scale studies trying to identify the relative impact of agricultural and urban land use, Meador & Goldstein (2003) evidenced complex relationships. High fish community conditions can be found even in area where agricultural land use was relatively high (>50% of the basin). Other factors like urbanization may be more deleterious to stream fishes, and often high levels of agriculture are associated with low levels of urbanization. They concluded that agricultural land use is only a general descriptor, and practices must be taken into account to explain the effective impacts.

Biological response thresholds

In the USA, Cuffney et al. (2000) studied the different forms of biological response for each community (fish, invertebrates and algae) along an agriculture intensity gradient; 25 sites were sampled in 3 different ecoregions. They evaluate the pressures coming from agriculture through multimetric condition indices: pesticide load and NPAI (Non-Pesticide Agriculture Index). Whatever the cause of impairment, different responses were detected: linear relations for fish, and threshold-response for invertebrates and algae. They concluded that "the forms of the biological responses to land use gradients are critically important to the development and implementation of cost-effective mitigation and monitoring procedures".

According to an urbanization gradient, some authors proposed thresholds values corresponding to a significant biological alteration. Stepenuck et al. (2002) found a sharp decline of invertebrates communities above a threshold situated around 8% to 12% of imperviousness. A similar result is evidenced by Wang & Kanehl (2003); beyond 7% of impervious area in the watershed, invertebrate index values tended to be consistently poor. Roy et al. 2003 identified a breakpoint of about 15 - 20% urban land cover above which the invertebrate indices indicated a clear degradation.

Other works are more "conceptual". Addicott et al. (1987) emphasize the importance of environmental patterns, together with spatial and temporal scales of ecological processes. Bryce & Clarke (1996) link landscape-level ecoregions within a basin to bridge the gap between stream habitat and state-level ecoregions classifications. Boulton (1999) reviews the definitions of the river health concept, and how to choose the right indicators at the right scale. The large scale approach is also addressed in Naiman & Turner (2000), who recognise the need for better understanding the processes at the watershed and landscape levels.

5.3.5 Large scale pressure-impact models : a possible approach

At the end of this brief review, there is no model that can be directly applied in Europe, to link combined pressures and river ecological status (through biological indicators) at a large or even regional scale. However, this problem has been addressed by the Cemagref in France, and some preliminary results have interested the managers. We propose here a possible approach which could be generalized in the REBECCA project.

The objectives of the models under development are to relate the ecological status of rivers to indicators of driving forces and pressures, at the state-wide level and for the main hydro-ecoregions. These models are aimed to :

- 1) *Extrapolation* : to extrapolate (spatially, and if possible temporally) the probable ecological status of unmonitored water bodies ; can be used also to evaluate the global pressure level matching the limit of the good ecological status...
- 2) *Diagnosis* : to set a hierarchy between the sources of impact, and identify the dominant "pathologies" of a given region or river type.
- 3) *Restoration* : to point out the factors that can be managed, and where to act in priority.

The first models have been developed with EQR value of the French invertebrate index (IBGN), and the pressures are evaluated through the land cover (from CORINE); a preliminary report is available (Wasson et al. 2004). Two examples are given below.

1 - Spatial extrapolation with decision tree models

The objective is to extrapolate, on the main stems of the hydrographic network, a present-day probability of "good" ecological status with the IBGN index. The method uses the Decision Tree models (Breiman 1984). There are four steps in the model (figure 1):

- The Land cover (simplified in four categories) is calculated for the catchment of each biological station, and a decision tree is developed relating land cover with the ecological status, simplified in two categories : "good" (high and good status) or "bad" (less than good status).
- This decision tree is used to predict the status (good or bad) of the 6100 "Hydrological zones" (subcatchments) according to the land cover upstream of each subcatchment.
- An external validation of this model is made by comparing the status of the subcatchments predicted by the model, and the observed status of the sites situated on the main stem of the corresponding subcatchment.
- From these results, predictive outcomes are used to map the *present probability of good ecological status*, based on the IBGN index, for the main stem of each subcatchment (figure 2).
- The whole process is repeated with a slightly higher good status boundary to identify borderline situations, i.e. the subcatchments that are at the limit of the present "good status" hypothesis (figure 2).

Pressure / impact models : Principle of spatial extrapolation

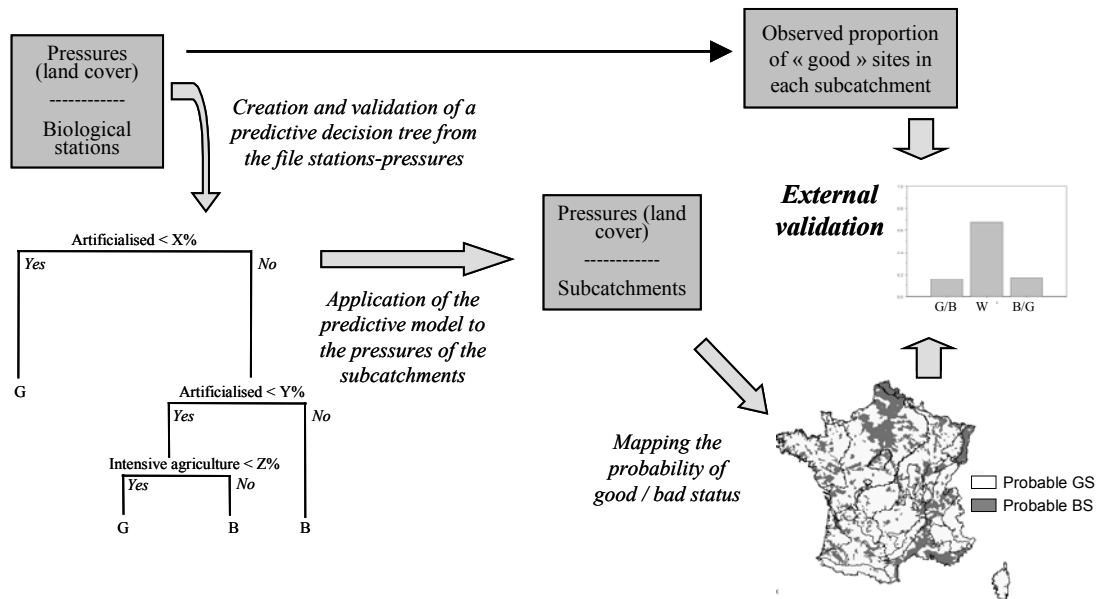


Figure 5.1. Pressure / impact models : methodological approach for the spatial extrapolation. Present probability of "good"/"bad" ecological status based on decision tree models at the subcatchment scale. G : good status ; B = Bad status ; W = well predicted ; G/B = predicted good/observed bad ; B/G = predicted bad/observed good. See text for details.

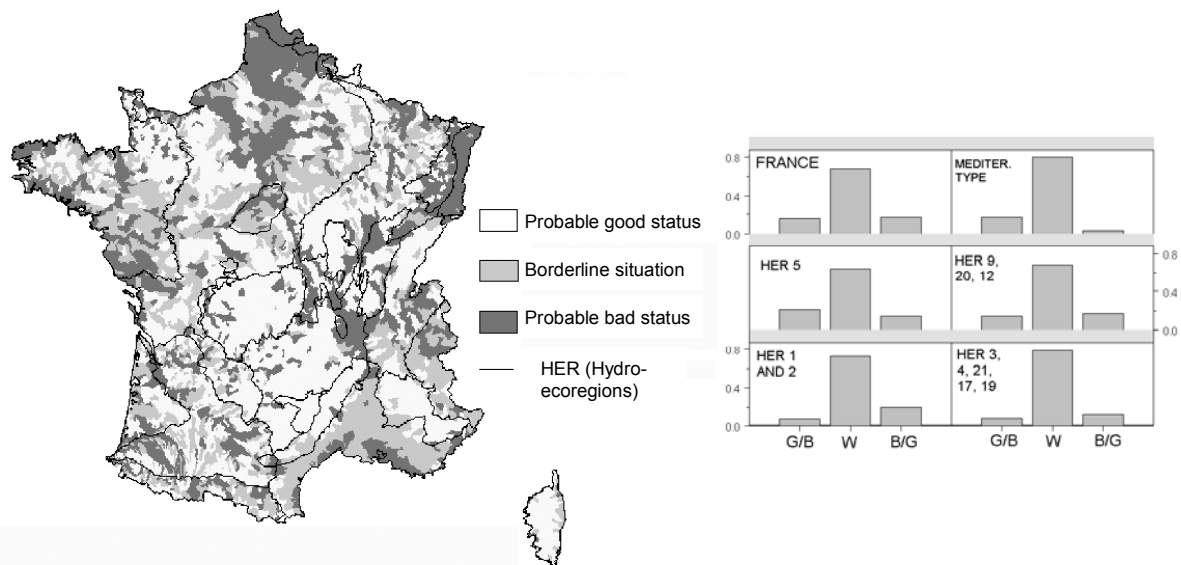


Figure 5.2 : Map of the present probability of "good"/"bad" status and borderline situations at the subcatchment scale, from pressure/impact models relating invertebrate index (EQR-IBGN) to land cover (CORINE). Six different models have been elaborated for five groups of hydro-ecoregions and for the whole France. On the right, external validation (see figure 1 for legend).

2 - Diagnostic of dominant driving forces with PLS regression models

The objective here is to run an explicative analysis to identify the categories of land cover that have the strongest negative or positive correlation with the EQR value of the IBGN index. For each IBGN site, the land cover is evaluated in the corresponding catchment and in a 3 km long riparian buffer. The model used is a Partial Least Square regression (PLS) (De Jong 1993, Tenenhaus 1998).

In the example given in figure 3 for the whole France, the model clearly points out the built up areas in the catchment as the most impacting factor, whereas forested and natural areas in the catchment and "low impact" agriculture (pastures) in the riparian buffer have the most positive effect on the IBGN. Models can be run with more precise land use categories, and at the hydro-ecoregions level to provide a more relevant diagnostic.

These two kinds of models can be run on datasets from different countries in the REBECCA project, provided that the catchments of the biological stations are available as a GIS layer.

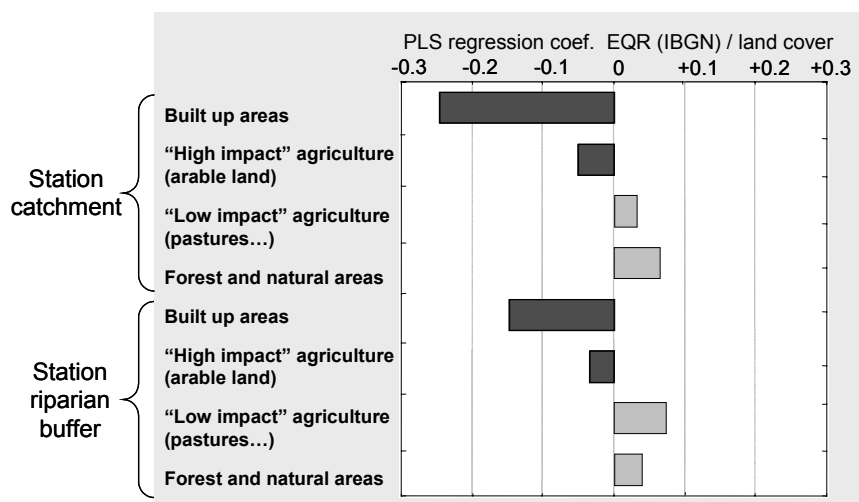


Figure 5.3. Results of the PLS regression for France. Cross-validated, $R^2 = 16.3\%$. Correlation coefficient between EQR (IBGN) and land cover (from CORINE Land Cover) of the station catchment and riparian buffer (analysis on 3640 stations).

5.4 Knowledge gaps

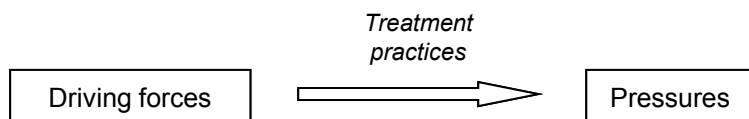
Research at **large spatial scale** is not well developed in general although there is a strong need for it. We have not found clear relations stated among the scientific community at this level. Studies are mostly focused on just one basin, watershed or ecoregion, and there is an important lack of inter-regions comparative studies. However, to deal with large scale decision-making, specially within diverse territories as in Europe, a regional approach would be very useful to identify 1) comparable issues even in regions spatially separated and 2) different regional river "pathologies" that should be managed differently even within a country. This would set a coherent natural framework for river study and management. Ecoregions are so far not widely used as a framework for large-scale studies of pressures/impact relationships. If ecoregional approach may improve the predictive capacity of pressure / impact models is still an open question. Natural constraints could determine both ecosystems sensitivity and typical land use and practices, thus leading to different regional responses. These issues are complex and need further investigations.

The share and the hierarchy of the different driving forces in the impact on stream ecosystems is still uncertain, when we consider the results of the papers reviewed. Although the negative effects of agriculture are generally stated by numerous authors, there is not enough scientific basis to quantify the degree of impairment, to discriminate effects from different kinds of cultures, and to set thresholds (of % of agricultural land use, for example).

When comparing the different forms of land use, some authors found agriculture as the most impairing activity, while others evidenced the strongest negative correlations with urban areas. This

could be due to differences in the methodology (bioindicators, methods, models), but perhaps these results are not contradictory and the hierarchical relations between these driving forces vary according to the level and type of development, or to the ecoregional context.

Ecosystems sensitivity, related to the natural context, may influence the intensity of the impairment. But the national socio-economic context determine the intensity of the pressures generated by a given land use, as well as the policies adopted to alleviate the impacts. Here is a key issue, and an outcome of this study is that the treatment practices must be introduced in the system to better understand the relationships between driving forces and pressures.



For example, the same category of agricultural land use may have different effects in terms of pollution, sediment yield etc. on water ecosystems if practices are intensive or extensive. In the same way, two urban areas of the same size will not have the same impacts depending on waste treatment, and this may depend on the local, regional or national socio-economical context. In this way the treatment practices may increase or reduce the intensity of the pressures. However it is not easy to define relevant indicators of treatment practices.

The different relations that take place at different spatial levels, like riparian corridor vs. basin, need better understanding. It is still difficult to state, when studying impairment causes in a given site, the influence of local riparian corridor, upstream riparian corridor, basin or sub-basin conditions of land use. Again, the papers reviewed present contrasting results; some identify landscape-level factors as the main variables for predicting biological indicators, while others find no relationships beyond the local scale measurements. Are these discordances due to the different human and natural contexts, or to the different methods used? In any case, both scales have to be considered in developing models. The restoration of river corridors is often seen as a key action to improve ecological status, but the actual “buffering capacity” of riparian areas needs to be properly evaluated.

The same happens with the existence of thresholds in the relations between driving forces and ecological status. Some managers are wondering if a good ecological status is attainable in heavily urbanized basins, or in intensively cultivated areas. Some of the models analysed talk about threshold levels, beyond which streams are systematically impaired; a comparison and validation should be made for the European territory. This is a complicated issue as the different biological compartments may not have the same response curve to a given pressure. Another important related question is the feasibility of spatial extrapolation of ecological status on the basis of land use in non monitored water bodies. Answering such questions will be crucial for the implementation of the WFD. However, there is still a strong gap of knowledge and scientific statements.

In general, it seems that the present knowledge is not sufficient to give clear advice to decision makers. This kind of questions are relatively recent, so there is a lack of conceptual models, and still not enough feedback. Large scale analysis comparing relationships through different scales, states, and ecoregions are still necessary.

Note : literature search methodology

Only papers concerning biological components (fish, invertebrates, diatoms) have been selected. Our research has focused on studies at large scales, at least a river basin or watershed, relying land use at the landscape level and biological components of streams. Special attention has been given to documents with a regional approach. The selected key-words for the literature research were

land use, landscape, impact, regionalization, ecoregion, model, scale, watershed, river, fish, invertebrate, bioindicator, riparian corridor. References come first from our own bibliographical database. Other larger data bases were consulted: Web of Science and Science Direct to which the CEMAGREF is subscribed.

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6 General conclusions

The review of literature on relationships between physical/chemical quality elements in rivers and biological quality elements has identified thousands of publications on these issues. Therefore the first main conclusion is that there is a general and very substantial knowledge on the different types of pressures (types of pollution), on the physical and chemical characteristics and on the impacts on river plant and animal life.

For all the important types of pressures studies have been made and described in the literature based on either scientific experiments or environmental monitoring. This has generated a general and widespread understanding of the general ecological impacts of the different types of pressures among both scientists and managers with the authorities.

However, most studies did not cover the entire causal chain including pressures, physical/chemical quality and biological quality. Most studies have been empirical in nature and all three elements (pressure, physical/chemical quality and biology) have almost never been treated in a quantitative manner. In the vast majority of studies one element has been assessed quantitatively while the other two elements have only been qualitatively assessed. Such studies have given us a useful understanding of general relationships, but will not enable us to establish dose-response relationships that could be used as a tool in the implementation of the WFD.

Another difficulty in establishing causal relationships from pressures to the river flora and fauna is that multiple pressures dominate in rivers in populated catchments where most pollution studies have been undertaken. Further, only limited conclusion can be made from laboratory experiments, because they can not easily be up-scaled to entire river ecosystems and catchments, and targeted, large scale scientific pollution experiments have not been made.

For these reasons the quantitative relationships found in literature between the biota/biological quality indicators and physical/chemical parameters are rather few compared to the needs for the authorities implementing the Water Framework Directive. Furthermore, there is an apparent lack of studies investigating recovery processes after a stressor has been reduced or removed. This limits our ability to predict how river quality will be improved after reducing a pressure on the ecosystem.

The next task of the REBECCA WP4 work on rivers is to analyse and describe these relationships based on information found in literature and especially from available monitoring results from rivers covering both biological and chemical/physical quality elements. With the very large number of possible biological quality metrics, the large number of different river types in EU and the different types of pressures this is still a very ambitious task.

With the focus of producing indicators, relationships and classification tools of practical use for the authorities in their implementation of WFD two important aspects of the strategy for this work are to focus on

- pressure-impact relationships with a direct linkage between the physical/chemical quality and the biota and
- indicators and metrics describing the biological quality that can be rather generally applied and not limited to be useful for one or a few types of rivers.