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MULTIMETRIC ASSESSMENT OF FRESHWATER MACROINVERTEBRATE COMMUNITIES IN FLANDERS, BELGIUM

Thesis submitted in fulfilment of the requirements for the degree of
Doctor (PhD) in Applied Biological Sciences

Dutch translation of the title:
Multimetrische beoordeling van macro-invertebratengemeenschappen in zoetwater in Vlaanderen, België

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Woord vooraf

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Wim Gabriels
Gent, 11 november 2007.

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List of abbreviations

A	Flemish lake type “Alkaline” (“*Alkalisch*”)
AERMC	Agence de l’Eau Rhône-Méditerranée-Corse
AFNOR	Association Française de Normalisation
AHES	Average Hellenic Evaluation Score
AQEM	the development and testing of an integrated Assessment system for the ecological Quality of streams and rivers throughout Europe using benthic Macroinvertebrates
ASPT	Average Score Per Taxon
BBI	Belgian Biotic Index
Bg	Flemish river type “Large stream” (*Grote Beek*)
BgK	Flemish river type “Large stream Kempen” (*Grote Kempense Beek*)
BIN	Belgian Institute for Normalisation (*Belgisch Instituut voor Normalisatie*)
Bk	Flemish river type “Small stream” (*Kleine Beek*)
BkK	Flemish river type “Small stream Kempen” (*Kleine Kempense Beek*)
BMWP	Biological Monitoring Working Party Score
Bzl	Flemish lake type “Very slightly brackish” (*Zeer Licht Brak*)
C	Flemish lake type “Circumneutral” (*Circumneutraal*)
CB-GIG	Central-Baltic Geographical Intercalibration Group
EBI	Extended Biotic Index
ECOSTAT	European Working Group “Ecological Status”
EPT	Ephemeroptera, Plecoptera, and/or Trichoptera taxa richness
EQI	Environmental Quality Index
EQR	Ecological Quality Ratio
GEP	Good Ecological Potential
HES	Hellenic Evaluation Score
HESII	Hellenic Evaluation Score Interpretation Index
ICMi	Intercalibration Common Metric index
IBGN	Indice Biologique Global Normalisé
ISO	International Organization for Standardization
ITC	Index of Trophic Completeness

JRC	Joint Research Centre
LBI	Lake Biotic Index
LQI	Lincoln Quality Index
MEP	Maximal Ecological Potential
MMIF	Multimetric Macroinvertebrate Index Flanders
MTS	Mean Tolerance Score
NST	Number of Sensitive Taxa
P	Flemish river type “Polder watercourse” (*Polderwaterloop*)
PTI	Potamon Typie Index
R-C1	Intercalibration common river type “Small Lowland Siliceous Sand”
R-C4	Intercalibration common river type “Medium Lowland Mixed”
REFCOND	guidance on establishing Reference Conditions and ecological status class boundaries for inland surface waters
Rk	Flemish river type “Small river” (*Kleine Rivier*)
Rg	Flemish river type “Large river” (*Grote Rivier*)
RIVPACS	River Invertebrate Prediction And Classification System
Rzg	Flemish river type “Very large river” (*Zeer Grote Rivier*)
SIGNAL	Stream Invertebrate Grade Number-Average Level Score
SWD	Shannon-Wiener Diversity Index
TAX	Taxa richness
TC	Tolerance Class
TS	Tolerance Score
VLAREM	Flemish Regulations concerning Environmental Licences (*Vlaams Reglement betreffende de Milieuvergunning*)
VMM	Flemish Environment Agency (*Vlaamse Milieumaatschappij*)
WFD	European Water Framework Directive
Z	Flemish lake type “Acidic” (*Zuur*)

General introduction

On the 22nd of December 2000, the European Water Framework Directive (WFD; EU, 2000) entered into force. This can be considered as a milestone in the history of European water policy, providing an overall framework for integrated water management based on a catchment approach. The main objective is to achieve a good status for all surface and groundwaters in the European Union by the end of 2015. For natural surface waters, the good status is defined as the combination of a good ecological and a good chemical status. Characterisation of the ecological status is based on a number of biological quality elements as well as hydromorphological, chemical and physico-chemical elements supporting these biological elements (EU, 2000).

To assess the status of the biological quality elements, member states must use a classification method that takes into account a number of parameters depending on the quality element (EU, 2000). The biological elements to be considered depend upon the category of surface waters. For the categories “rivers” and “lakes”, one of these elements is “benthic invertebrate fauna” (EU, 2000), generally referred to as macroinvertebrates. For assessing the status of this quality element, the parameters “taxonomic composition and abundance”, “ratio of disturbance sensitive to insensitive taxa” and “diversity” must be taken into account. The quality index must be expressed as an Ecological Quality Ratio (EQR), which can be defined as the ratio of the actual status to the reference conditions. In other words, the EQR can take any value between zero, corresponding to a bad ecological status, and one, corresponding to a very good ecological status. This EQR range must be divided into five classes reflecting bad, poor, moderate, good and high status, respectively (EU, 2000).

Application of the Belgian Biotic Index (BBI; De Pauw and Vanhooren, 1983) in routinely river monitoring schemes by the Flemish Environment Agency (VMM) since 1989 confirmed the reliability and robustness as a quality assessment method. However, some difficulties arise with regard to the potential application of the BBI as a macroinvertebrate assessment method for WFD implementation in Flanders. A first difficulty in this context is that it is not a type-specific method. All types of rivers are evaluated by means of one single score system. Also, the BBI is originally intended as an assessment system for watercourses (De Pauw and Vanhooren, 1983) and hence an index for stagnant waters is still missing. Furthermore, the abundance, one of the mentioned parameters imposed by the WFD, is not taken into account in the BBI calculation.

The **overall objective** of this study can be defined as follows:

"In order to establish a monitoring network for biological quality assessment based on macroinvertebrates in river and lakes in Flanders, an assessment method is needed that complies with all mentioned technical requirements of the WFD. Moreover, this assessment method should be practical in use and cost-efficient. This assessment method can be the currently used BBI, an adaptation of this index or a completely new index, on the condition that it meets all stated requirements. Class boundaries must be established to divide the index range into the five quality classes imposed by the WFD. Furthermore, the boundaries between the classes "high" and "good" and between "good" and "moderate" should be harmonised with those of the other member states in the course of the intercalibration exercises organised by the European Commission to ensure that ecological status is assessed in a comparable way across all member states."

This thesis is organised in five major chapters:

Chapter 1 presents a general review of biological assessment methods of freshwaters based on macroinvertebrates and the requirements of the European Water Framework Directive on this subject. Different technical aspects such as sampling, identification, index calculation and defining reference conditions are addressed.

Chapter 2 examines strengths and weaknesses of the method currently used for surface water quality evaluation in Flanders, the BBI, with regard to its potential application for implementing the European Water Framework Directive. In addition, two more general problems are discussed which are associated with biological water quality assessment methods. These problems are caused by possible changes in taxonomical status of species, and by the introduction of alien species, respectively. The possible bias that may be introduced by these phenomena is analysed by means of the example of the BBI calculation using VMM sampling data and recommendations are given to avoid such problems. The chapter concludes with a final recommendation with regard to the modification and application of the BBI in the context of the WFD.

In **Chapter 3**, an index is proposed for assessing the status of rivers and lakes in Flanders based on macroinvertebrates in order to comply with the WFD. This index is developed taking into account the existing experience with the BBI, scientific literature, analysis of the macroinvertebrate database of the VMM and a consultation of a panel of macroinvertebrate experts. Furthermore, a preliminary division into quality class boundaries is proposed.

In **Chapter 4**, the characteristics of the proposed method are further explored. The overall index and its composing metrics are studied with regard to their mutual relation, their response to ecological degradation and their relation with the BBI. This is examined by means of a data set of macroinvertebrate samplings throughout Flanders that was collected by the VMM in the framework of the routine monitoring network since 1989.

Chapter 5 describes the Flemish contribution to the European intercalibration exercise for river macroinvertebrates based on VMM sampling data. The aim of this exercise is to ensure that standards for good and high biological status of rivers based on macroinvertebrates are comparable to those of other European member states. This intercalibration is necessary because different member states each use a different assessment system and as a result, the standards are not necessarily comparable. The intercalibration exercise examines whether the class boundaries moderate-good and good-high are comparable with other member states within a region. Member states that have boundaries which are not comparable will be asked to adjust these boundaries. When the intercalibration exercise results in the acceptance of the boundaries of a method for a particular region (in this study thus Flanders), the method can be formally accepted by the European Commission as a “WFD-proof” method for assessing the status of a water body based on macroinvertebrates.

Chapter 1. Ecological assessment of freshwater based on macroinvertebrates - a review

Incorporating redrafted sections of:

De Pauw, N., Gabriels, W. & Goethals, P.L.M. (2006). River monitoring and assessment methods based on macroinvertebrates. In: Ziglio, G., Siligardi, M. & Flaim, G. (eds.). Biological monitoring of rivers. Applications and perspectives. John Wiley & Sons, Chichester, West Sussex, UK. p. 113-134.

and

Gabriels, W., Goethals, P., Adriaenssens, V., Heylen, S. & De Pauw, N. (2003). Development of a score or index for macroinvertebrates for the Flemish rivers and lakes according to the European Water Framework Directive and testing of REFCOND. Final Report (in Dutch). Laboratory of Environmental Toxicology and Aquatic Ecology, Ghent University, Belgium. 72 p. + appendices.

After sleeping through a hundred million centuries we have finally opened our eyes on a sumptuous planet, sparkling with colour, bountiful with life. Within decades we must close our eyes again. Isn't it a noble, an enlightened way of spending our brief time in the sun, to work at understanding the universe and how we have come to wake up in it? This is how I answer when I am asked – as I am surprisingly often – why I bother to get up in the mornings. To put it the other way round, isn't it sad to go to your grave without ever wondering why you were born? Who, with such a thought, would not spring from bed, eager to resume discovering the world and rejoicing to be a part of it?

Richard Dawkins (1998)

1. Biological assessment of freshwater based on macroinvertebrates - a review

1.1 Biological assessment

1.1.1 The necessity of ecosystem monitoring

The earth's ecosystems are strongly affected by anthropogenic pressures (Vitousek et al., 1997; Millennium Ecosystem Assessment, 2005). Humanity's use of natural resources increased from an estimated 70% of the regenerative capacity of the global biosphere in 1961 to approximately 120% in 1999 (Wackernagel et al., 2002). These increasing pressures result in destruction, fragmentation and degradation of natural habitats (Baillie et al., 2004) and a reduction of global biodiversity at unprecedented rates (Pimm et al., 1995; Balmford et al., 2003; Loh et al., 2005; Millennium Ecosystem Assessment, 2005). This biodiversity crisis will dramatically affect human well-being (Chapin et al., 2000; Balmford and Bond, 2005; Hooper et al., 2005; Díaz et al., 2006). Figure 1.1 summarises interactions between human activities and ecosystem degradation (modified after Vitousek, 1997). However, a global crisis may still be averted if radical steps are taken towards sustainability (e.g. Pimm et al., 2001; Diamond, 2005).

To achieve sustainable development without depleting ecological capital, it is essential to periodically monitor the state of the environment and provide early-warning indicators of dysfunction, as well as timely identification of probable sources of stress (Rapport and Singh, 2006). The assessment of ecosystem status involves the articulation of the linkages between human activity, regional and global environmental change, reduction in ecological services and the consequences for human health, economic opportunity and human communities (Rapport et al., 1998).

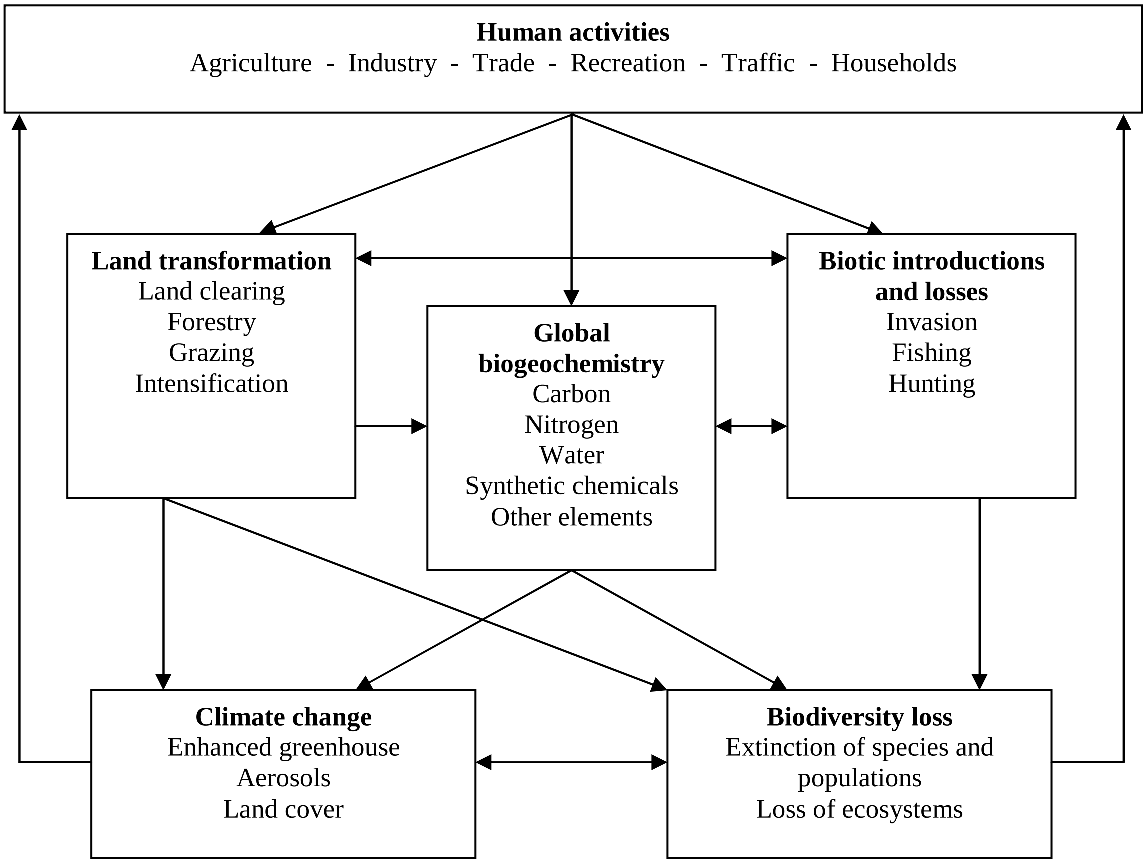

Fig. 1.1 Overview of direct and indirect human alterations of the global ecosystems (modified after Vitousek et al., 1997).

1.1.2 The necessity of ecosystem monitoring in freshwaters

The contribution of freshwaters in ecological degradation is disproportionately high (Dudgeon et al., 2006). Although freshwaters make up only 0.01 percent of the total volume of the world's water and approximately 0.8 percent of the total surface of the Earth, it supports at least 100 000 species out of an estimated 1.8 million, or almost 6 percent of all described species (Dudgeon et al., 2006). However, biodiversity losses are far greater in freshwaters than those in the most affected terrestrial ecosystems (Sala et al., 2000; Dudgeon et al., 2006).

The European Water Framework Directive (WFD; EU, 2000) asserts that in order to achieve its objectives, monitoring the development in water status on a systematic and comparable basis throughout the European Community is necessary in order to provide a sound basis for member states to develop programmes of measures aimed at achieving these objectives (EU, 2000).

1.1.3 Biological versus physical-chemical monitoring of freshwaters

Monitoring the quality of a freshwater ecosystem should not rely on physical-chemical analyses alone. Karr (1996) suggests that biological monitoring and biological criteria provide the most robust approach to track the status of our waters, because waterways that cannot support healthy biological communities are unlikely to support human society for long.

Biological monitoring can provide more information on the state of an ecosystem than physical-chemical monitoring alone. The biotic component of an aquatic ecosystem can be considered as the “memory” of an ecosystem, integrating a wide range of ecological effects over time, while chemical analyses only provide information on the chemical water composition at the moment of sampling (De Pauw and Hawkes, 1993). In certain cases biological communities already respond before analytical detection allows for (De Pauw and Hawkes, 1993). For these reasons, physical-chemical and biological monitoring should be considered as complementary instruments for ecological monitoring (De Pauw and Hawkes, 1993).

1.1.4 Concepts of freshwater ecosystem quality

Moog and Chovanec (2000) define ecological integrity as “the maintenance of all internal and external community processes and attributes, interacting with their environment in such a way that the biotic community corresponds to the natural state of the relevant aquatic habitat.” Ecological integrity requires the attainment of its three elements: physical, chemical, and biological integrity (Barbour et al., 2000).

Based on earlier definitions by Cairns (1977) and Frey (1977), Karr and Dudley (1981) define biological integrity as “the ability of an ecosystem to support and maintain a balanced, integrated, adaptive community of organisms having a species composition, diversity, and functional organisation comparable to that of a natural habitat of a region.”

The scope of river health is generally defined wider than that of ecological integrity. Many authors argue that for assessing river health, in addition to ecological criteria, also human values, uses and amenities should be included (e.g. Meyer, 1997; Boulton, 1999; Karr, 1999; Norris and Hawkins, 2000). Ecological criteria include sustainability, resilience to stress, and ecological integrity (Boulton, 1999), while human values include goods and services such as water supply for industry and agriculture, drinking water, recreation and production of fish for consumption (Meyer, 1997). The analogy between this definition of ecosystem health (in comparison to ecological integrity) and the concept of human health is obvious. While the physical integrity of a person might indicate nothing more than survival, a person that is considered healthy is generally thought to be capable of productive activities (e.g. being able to work).

The objective of a healthy freshwater ecosystem can therefore be summarised as follows (Figure 1.2): the ecosystem should be characterised by physical-chemical, morphological and biological conditions that makes it possible to maintain an aquatic community with structure and function comparable to that of a similar system in undisturbed conditions, and resilient to stress (i.e. a good ecological integrity) and it should be able to support all necessary goods and services to society.

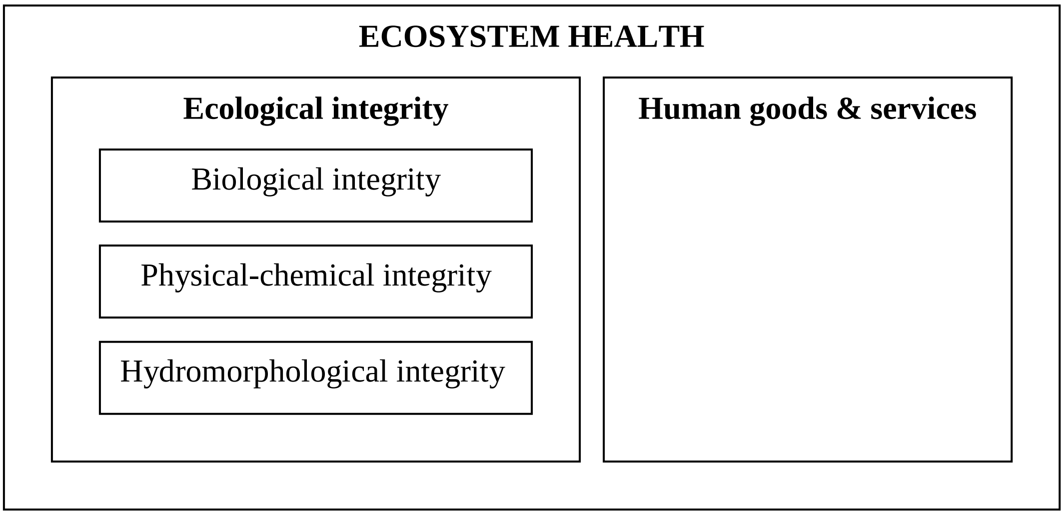

Fig. 1.2 Overview of different concepts of freshwater ecosystem quality.

The assessment of ecosystem health is generally approached by comparing the examined site to the characteristics that would occur at a similar site in the absence of human disturbance (Norris and Thoms, 1999). For this purpose, the concept of reference conditions was introduced. Reference conditions can be described as the best available conditions that can be expected at similar sites, and are described based on observations at several similar sites (Hughes et al., 1986; Hughes, 1995; Reynoldson et al., 1997; see further paragraph 1.6).

Finally, it should be stressed that assessment of ecological integrity or ecosystem health is not necessarily synonymous with determining conservation value. Conservation value is a concept used for ranking potential conservation targets (see e.g. Angermeier and Winston, 1997; Chadd and Extence, 2004; Dunn, 2004; Linke et al., 2007).

1.1.5 The use of biological quality indices

The history of biological water quality assessment spans almost a century. Earlier systems were purely descriptive or qualitative and mainly based on the presence or absence of indicator species, primarily related to discharges of domestic sewage, i.e. organic pollution. Since the early 1950s however ecologists felt the need to convey their complex biological data in a numerical form such as indices or scores (e.g. Beck, 1955; Pantle and Buck, 1955). On the other hand, many ecologists remain sceptical towards the development and use of a biological quality index, because it reduces the complexity of a biological community to a single numerical value, which can easily be misused (Seegert, 2000). However, a biological quality index appears to be more acceptable to policy-makers than raw biological survey data expertly interpreted. A quality index is easy to comprehend, reproducible and allows for compliance checking. By allowing for an effective communication of the condition of biological systems, the use of a quality index can transform biological monitoring from a scientific exercise into an effective tool for environmental decision-making (Karr, 1999).

In order to be useful as a major tool for everyday management of water quality, a biological quality index should meet the following characteristics (Extence et al., 1987):

1. The system should be based on established methods and it should be possible to calculate results retrospectively for historical data;
2. The method should be as simple as possible to use, both in the field and in the laboratory;
3. Non-specialists should be able to easily appreciate the meaning of any grading or index rating;

4. The index should use as much information as practically possible from the sample, as it is the whole community and not just “key groups” which respond to variations in water quality;
5. The index should be applicable to all river types, whether they be fast or slow flowing, habitat-rich or habitat-poor;
6. It should be possible to associate index values with water quality classes, and existing or potential river stretch uses, and thus check for compliance with targets;
7. The index should be cost effective.

1.2 Macroinvertebrates and biological assessment

1.2.1 Definition of macroinvertebrates

Macroinvertebrates are not a systematic unit but a diverse assemblage of taxa, grouped together based on taxonomic restrictions, size and habitat. Generally, macroinvertebrates are considered as those invertebrate animals inhabiting the aquatic environment that are large enough to be caught with a net or retained on a sieve with a mesh size of 250 to 1000 µm, and thus can be seen with the unaided eye (e.g. Sládeček, 1973b; Cummins, 1975; De Pauw and Vanhooren, 1983; Rosenberg and Resh, 1993; Ghetti, 1997; Tachet et al., 2002).

Alternatively, macroinvertebrates are sometimes referred to as benthic invertebrate fauna (e.g. EU, 2000) or macro(zoo)benthos (e.g. Aagaard et al., 1997; Verneaux et al., 2004; Böhmer et al., 2004a; Martel et al., 2007), stressing their tendency towards a benthic existence. The majority of the aquatic macroinvertebrates indeed have a benthic life and inhabit the bottom substrates (sediments, debris, logs, macrophytes, filamentous algae, etc.). However, some representatives of the macroinvertebrates, also serving as bioindicators, are pelagic and freely swimming in the water column, or pleustonic and associated with the water surface (Tachet et al., 2002). Hence the more general term macroinvertebrates is used throughout this thesis.

Macroinvertebrate communities may include a large variety of taxa, the majority usually being arthropods (De Pauw et al., 2006). All taxa belonging to the macroinvertebrates are included in one of the following groups (De Pauw and Vannevel, 1991):

Phylum Porifera (Sponges)

Phylum Cnidaria

Phylum Bryozoa (Moss animals)

Phylum Plathelminthes (Flatworms)

Phylum Annelida (Segmented worms):

- Subclass Polychaeta (Bristleworms)
- Subclass Oligochaeta
- Subclass Hirudinea (Leeches)

Phylum Mollusca:

- Class Gastropoda (Snails)
- Class Bivalvia (Clams and Mussels)

Phylum Arthropoda:

- Class Arachnida
- Class Branchiopoda
- Class Maxillopoda
- Class Malacostraca
    - Order Amphipoda
    - Order Isopoda
    - Order Decapoda
- Class Entognatha:
    - Order Collembola (Springtails)
- Class Insecta:
    - Order Ephemeroptera (Mayflies)
    - Order Odonata (Dragonflies and Damselflies)
    - Order Plecoptera (Stoneflies)
    - Order Hemiptera (True Bugs)
    - Order Neuroptera (Net-winged insects)
    - Order Megaloptera
    - Order Coleoptera (Beetles)
    - Order Trichoptera (Caddisflies)
    - Order Lepidoptera (Butterflies and Moths)
    - Order Diptera (True flies)
    - Order Hymenoptera

Many of the insects, such as Ephemeroptera and Trichoptera, only have a semi-aquatic life cycle, with aquatic larvae and terrestrial adults. Groups belonging to the zooplankton, such as copepods, cladocerans and ostracods are usually not considered as macroinvertebrates. Some authors also exclude watermites from macroinvertebrates.

Macroinvertebrates perform a variety of functions in freshwater ecosystems (Vannote et al., 1980; Wallace and Webster, 1996; Covich et al., 1999). These functions include detritus decomposition (Wallace and Webster, 1996), the release of bound nutrients into solution, by feeding activities, excretion, and burrowing into sediments (Covich et al., 1999), regulation of abundance, location and size of their prey (Covich et al., 1999), supplying food to aquatic and terrestrial consumers (Covich et al., 1999; Malmqvist, 2002), and promoting nutrient transfer to overlying open water of lakes or adjacent riparian zones of streams (Covich et al., 1999).

1.2.2 Macroinvertebrates as biological indicators

Macroinvertebrates are the most commonly used biological indicator group for assessment of freshwater quality (Woodiwiss, 1980; Hellawell, 1986; De Pauw and Hawkes, 1993; Rosenberg and Resh, 1993; Metcalfe, 1989; Hering et al., 2004).

Macroinvertebrates are complementary to other indicator groups for biological assessment and evaluations based on different biological indicator groups should therefore be used in conjunction. As to macroinvertebrates, a number of specific advantages and disadvantages can be identified (Table 1.1).

The reasons for macroinvertebrates being so popular in bioassessment are numerous (e.g. De Pauw et al., 2006). Macroinvertebrates are visible to the human eye and relatively easy to sample (De Pauw et al., 2006). They are ubiquitous and abundant throughout the whole river system in the crenal, rhitral as well as the potamal part (Illies, 1961).
Macroinvertebrates play an essential role in the functioning of the river continuum food web (e.g. Vannote et al., 1980; Cummins, 1992, Giller and Malmqvist, 1998). Having relatively long life cycles and being confined for most part of their life to one locality on the river bed, they act as continuous monitors, integrating water quality over a longer period of time, so they do not have to be sampled very frequently (De Pauw and Hawkes, 1993). They also

constitute a taxonomically very heterogeneous group, showing a broad spectrum of responses to each form of stress, including physical-chemical pollution and morphological changes of the aquatic habitat (Rosenberg and Resh, 1993).

Table 1.1 Advantages and disadvantages of macroinvertebrates for assessing biological water quality (summarised after De Pauw and Hawkes, 1993; Rosenberg and Resh, 1993; De Pauw et al., 2006).

	Advantages	**Disadvantages**
Operational issues	-Visible to the human eye -Easy to collect -Ubiquitous	-Sometimes difficult to identify -Quantitative sampling is difficult
Interpretational issues	-Ecologically relevant -Relatively long life cycles (integrating water quality over time) -Taxonomically diverse, integrating a wide range of stressors	-Variations due to non-quality related factors -Seasonal variations -Geographical variations

Using macroinvertebrates as monitors of river (water) quality however also has its limitations (e.g. De Pauw et al., 2006). A first difficulty is the possibility of wrong identifications of the sampled organisms, because identification is sometimes difficult, in particular for early life stages of insect larvae (Rosenberg and Resh, 1993). Quantitative sampling is difficult due to their non-random distribution in the river bed (De Pauw et al., 2006).

Factors other than water quality, such as current velocity and nature of the substratum, are also important determinants of benthic communities. Since these factors differ along the river in different zones, different communities become established at different sites with the same water quality (Giller and Malmqvist, 1998). Because of the seasonality of the life cycles of some macroinvertebrates, e.g. insects, they may not be found at some times of the year (e.g. Linke et al., 1999; Tachet et al., 2002). An other limitation is their restricted geographic distribution, the incidence and frequency of occurrence of some species being different in rivers throughout the region. Furthermore, because of their geographic distribution, species at the edge of their natural distribution range are theoretically more sensitive to additional stress, e.g. pollution, than those at the centre of their distribution. It would therefore not be possible to have a universal system of biological assessment based on the response of the same species/taxa (Sandin et al., 2000).

1.2.3 Implications of the European Water Framework Directive on monitoring freshwater macroinvertebrate communities

The European Water Framework Directive (WFD; EU, 2000) aims at preventing deterioration of the status of all bodies of surface water and achieving good water status for all waters by the end of 2015. For natural surface waters (rivers, lakes, transitional waters and coastal waters), “good status” is determined by a “good ecological status” and a “good chemical status”. Ecological status is determined by biological quality elements, supported by hydromorphological and physico-chemical quality elements. The biological quality elements should show low levels of alteration resulting from human activities, in other words, their status may only slightly deviate from that normally associated with the surface water body type under undisturbed conditions. These “undisturbed” conditions are called the “reference conditions” (EU, 2000).

These reference conditions describe a very good ecological status of the surface water body type. They must be identified for all biological quality elements (status of water flora including phytoplankton, benthic invertebrate fauna, and fish fauna), hydromorphological quality elements supporting the biological elements (hydrological regime, river continuity and morphological elements) and physical-chemical conditions supporting the biological elements (general elements and specific pollutants) (EU, 2000).

For each quality element a quality classification must be developed that integrates a number of relevant parameters according to the normative definitions. For the element benthic invertebrate fauna, the appropriate parameters for the categories rivers and lakes are presented in Table 1.2. This quality index must be in agreement with an ecological quality coefficient representing relative proportion of the index compared to the reference conditions. The ecological quality coefficient is expressed as a value between zero and one, where zero corresponds to a bad ecological status and one to a very good ecological status. The interval between zero and one is divided into five classes reflecting bad, poor, moderate, good and high status, which are assigned a colour code of, respectively, red, orange, yellow, green and blue (Fig. 1.3) (EU, 2000).

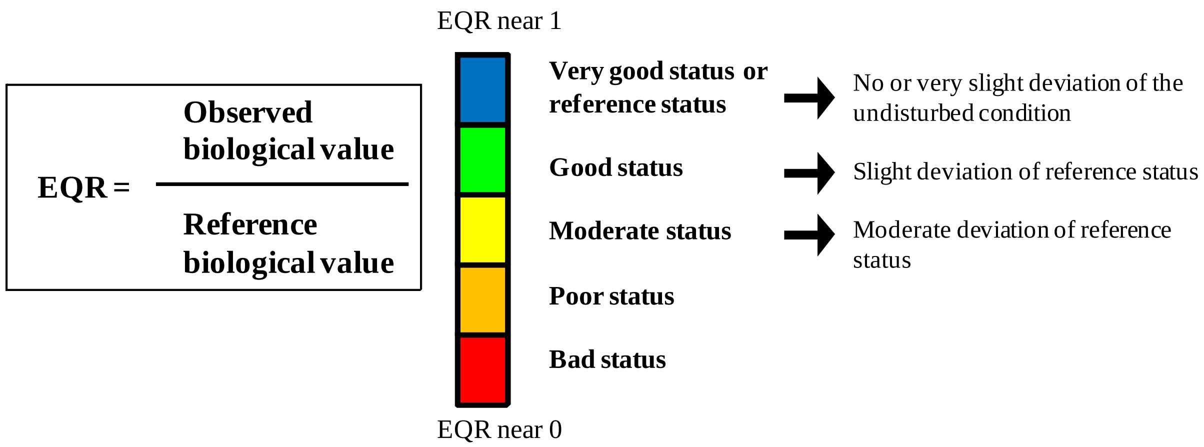

Figure 1.3 Principles for classification of high, good and moderate ecological status of surface waters based on anthropogenic alterations with Ecological Quality Ratios (EQR) (after Wallin et al., 2003).

Table 1.2 presents an overview of the normative definitions prescribed by the WFD for the quality classes “high”, “good” and “moderate” for the biological quality element “benthic invertebrate fauna” for the categories rivers and lakes.

For the classes “poor” and “bad”, the WFD does not prescribe any normative definitions, except for a general description that applies all quality elements at once: “Waters achieving a status below moderate shall be classified as poor or bad. Waters showing evidence of major alterations to the values of the biological quality elements for the surface water body type and in which the relevant biological communities deviate substantially from those normally associated with the surface water body type under undisturbed conditions, shall be classified as poor. Waters showing evidence of severe alterations to the values of the biological quality elements for the surface water body type and in which large portions of the relevant biological communities normally associated with the surface water body type under undisturbed conditions are absent, shall be classified as bad.” (EU, 2000)

The final score of the ecological status or the quality class assigned to a location is determined by the lowest value of the appropriate biological and physical-chemical quality elements

(Wallin et al., 2003). This is generally referred to as the “one-out-all-out principle”. These scores must be mapped per river basin district using the appropriate colour codes (EU, 2000).

Figure 1.4 summarises the necessary steps for assessing the ecological status of a given body of natural surface water according to the European Water Framework Directive.

The subsequent paragraphs of this chapter review the state-of-the-art of all relevant aspects involved in the assessment of a given freshwater site based on the present macroinvertebrates: sampling methods (paragraph 1.3), identification of the sampled organisms (paragraph 1.4), quality indices based on the obtained data (paragraph 1.5), and defining of reference conditions (paragraph 1.6), followed by some general conclusions (paragraph 1.7).

Depending on the assessment method and the objective, all macroinvertebrate groups, or only one or more groups (e.g. oligochaetes, chironomids, gammarids, mayflies) are taken into consideration. However, only methods that take the entire macroinvertebrate community into account will be discussed here. Nijboer et al. (2005) found that the use of subsets of indicator taxa or single taxonomic groups resulted in high classification error, and concluded that taxonomic redundancy is limited, which supports the use of all taxa in characterising a macroinvertebrate community, in particular in areas with high habitat diversity.

One additional aspect that should be mentioned is the assessment of water bodies as a whole. The WFD requires that the quality of an entire water body is reported, while all existing national macroinvertebrate monitoring schemes are currently based on single locations. Most likely, this will be accomplished by using one or more samples as surrogates for the complete water body. Macroinvertebrate sampling protocols or indices for entire lakes, rivers or river stretches have thus far not yet been proposed.

Table 1.2 Normative definitions for the biological quality element “benthic invertebrate fauna” for the categories rivers and lakes (EU, 2000).

Quality status	**Rivers**	**Lakes**
High status	The taxonomic composition and abundance correspond totally or nearly totally to the undisturbed conditions. The ratio of disturbance sensitive taxa to insensitive taxa shows no signs of alteration from undisturbed levels. The level of diversity of invertebrate taxa shows no sign of alteration from undisturbed levels.	The taxonomic composition and abundance correspond totally or nearly totally to the undisturbed conditions. The ratio of disturbance sensitive taxa to insensitive taxa shows no signs of alteration from undisturbed levels. The level of diversity of invertebrate taxa shows no sign of alteration from undisturbed levels.
Good status	There are slight changes in the composition and abundance of invertebrate taxa from the type-specific communities. The ratio of disturbance-sensitive taxa to insensitive taxa shows slight alteration from type-specific levels. The level of diversity of invertebrate taxa shows slight signs of alteration from type-specific levels.	There are slight changes in the composition and abundance of invertebrate taxa compared to the type-specific communities. The ratio of disturbance sensitive taxa to insensitive taxa shows slight signs of alteration from type-specific levels. The level of diversity of invertebrate taxa shows slight signs of alteration from type-specific levels.
Moderate status	The composition and abundance of invertebrate taxa differ moderately from the type-specific communities. Major taxonomic groups of the type-specific community are absent. The ratio of disturbance-sensitive to insensitive taxa, and the level of diversity, are substantially lower than the type-specific level and significantly lower than for good status.	The composition and abundance of invertebrate taxa differ moderately from the type-specific conditions. Major taxonomic groups of the type-specific community are absent. The ratio of disturbance sensitive to insensitive taxa, and the level of diversity, are substantially lower than the type-specific level and significantly lower than for good status.

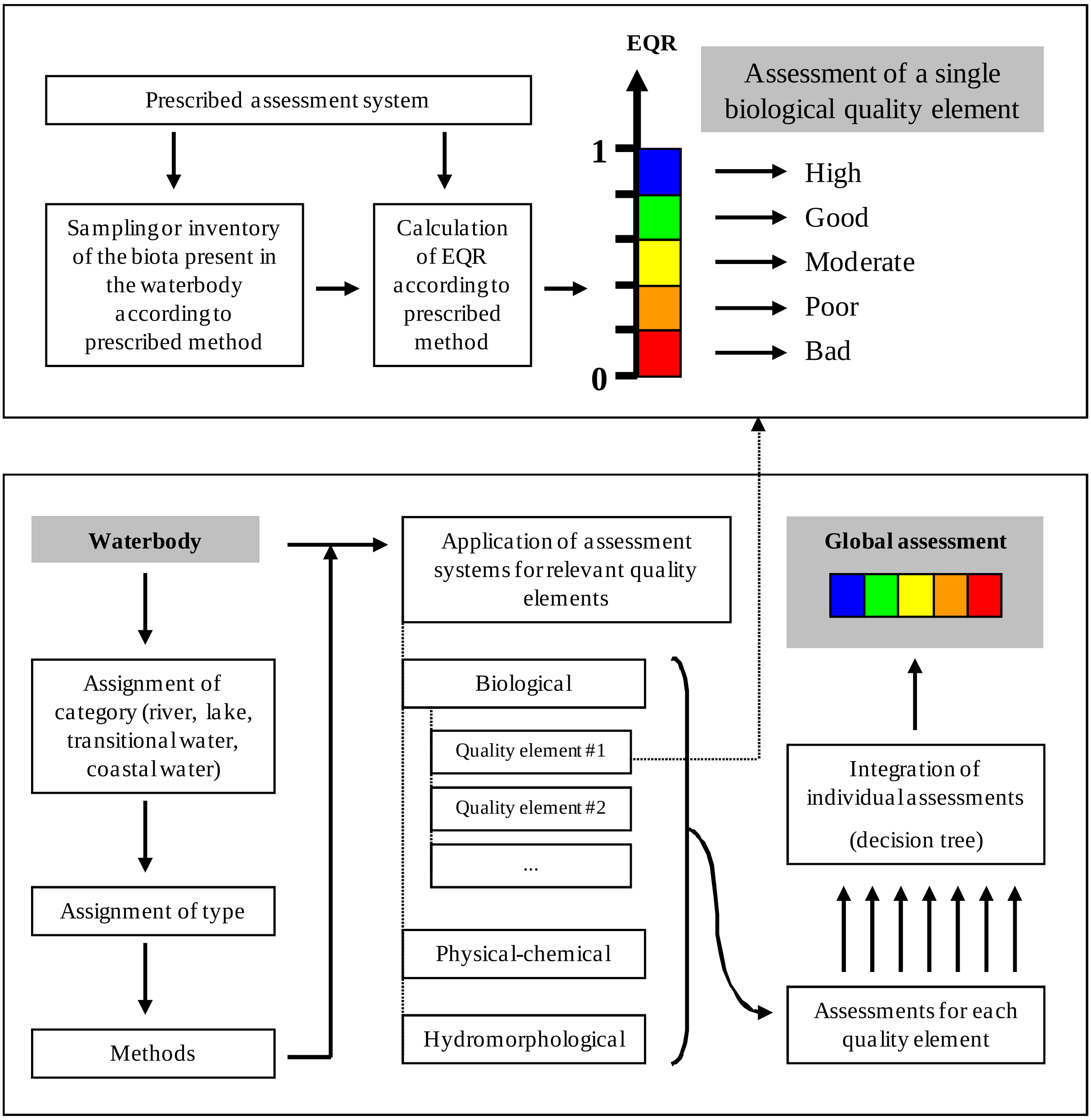

Fig. 1.4 Summary of necessary steps for assessing the ecological status of a given water body according to the European Water Framework Directive.

1.3 Sampling methods

The devices that can be used to collect macroinvertebrates are quite diverse. Commonly used tools in river monitoring programmes are different types of nets (e.g. handnet, surber net, dredges), grabs (e.g. Van Veen, Ponar), core samplers, artificial substrates or colonisation samplers (e.g. standard Aufwuchs sampler, bag sampler, Hester-Dendy), drift and emergence samplers. Overviews of these instruments and convenient handling can be found in Schwoerbel (1970), Mason (1981), Rosenberg and Resh (1982), Ghetti (1997) and Clesceri et al. (1998). Important for the nets are the mesh size (between 250 µm and 1 mm) (e.g. ISO, 1985) for the grabs and cores, the volume and sediment surface sampled (e.g. ISO, 1988) and for the artificial substrates, the material, volume and colonisation (exposure) time (e.g. Rosenberg and Resh, 1982; ISO, 1993; De Pauw et al., 1994).

The selected sampling method depends on the physical characteristics of the aquatic environment (depth, current velocity, sediment structure), the objective of the assessment method (qualitative, (semi-)quantitative), compartments of the ecosystem to be sampled (river bed, sediment, riffles, pools, water column, banks, aquatic vegetation) and the taxonomic groups considered in the assessment method (either the entire macroinvertebrate community or only specific groups; the latter approaches are however not discussed here).

For an overall assessment of running waters, all habitats, or specific habitats are sampled for periods roughly proportional to their extent on the site (e.g. Woodiwiss, 1980; De Pauw and Vanhooren, 1983; Ghetti, 1997). Each site is indeed composed of a mosaic of biotopes (e.g. Fontoura and De Pauw, 1994; Tachet et al., 2002; Hering et al., 2004), i.e. areas where the environmental conditions are uniform and clearly defined and which need to be examined. Different sampling tools may be used for exploring different habitats. Samples may be completed by hand picking of macroinvertebrates on hard substrates (stones, debris, plants) along the banks. Most sampling methods for assessments with macroinvertebrates are qualitative or semi-quantitative in approach which means that the sampling effort is linked to examining the site during a fixed period of time, e.g. 3 to 5 minutes depending on the size of the river (De Pauw and Vanhooren, 1983) and within a certain stretch, e.g. 10 to 20 meters (De Pauw and Vanhooren, 1983).

In shallow rivers, it is common practice to apply the kick sampling with a handnet (e.g. Woodiwiss, 1980). For routine biological monitoring based on biotic indices or scores handnets with a maximum mesh size of 0.5 to 0.75 mm are used, 0.5 mm for surveillance with more complete records of taxa, and 0.25 mm for special surveys requiring complete taxa lists. The latter size will ensure capture of instar stages and very small organisms, which may prove of value in water quality assessment. For ease of sampling and the sensitivity of the species present, riffles, i.e. shallows with a swift current over an eroding substratum, are often chosen. Kick sampling may be done zigzagwise, in transects or stretchwise or by taking separate kick samples in different habitats (e.g. multi-habitat sampling; Hering et al., 2004). In certain assessment methods quantitative samples are recommended and taken with a surber net in respectively the riffles and the pools (e.g. Tuffery and Verneaux, 1968).

For deeper waters that cannot be sampled with a handnet, artificial substrates (colonisation samplers) or grab samplers are recommended (ISO, 1993) to collect macroinvertebrate samples. Commercial (e.g. multiplate or Hester-Dendy samplers) as well as self-made artificial substrates (e.g. basket, box or bag samplers) can be used. Artificial substrates are submerged at a sampling site during a certain period of time in order to be colonised by macroinvertebrates and afterwards collected for analysis. These substrates can be made up of different materials (wood, steel, stone, plastic). Basket, box or bag samplers consist of a container (e.g. polypropylene bag; steel box, barbecue basket) and a filling material, that may consist of crushed rock, pieces of brick, marbles or even natural stones (pebbles) from the river bed. Cheap and universally present brick material provides good results (e.g. De Pauw et al., 1986). Important is the placement of the substrates, not in static areas but in the main flow of the river. To prevent drifting, substrates may need to be weighted. One should however prevent samplers to sink in the sediment, leading to a disturbed colonisation by macroinvertebrates. An exposure time of three to four weeks is the recommendable practice for obtaining a satisfactory assessment (e.g. De Pauw et al., 1994; Ghetti, 1997; Clesceri et al., 1998). Samples of the bottom and sediments can be directly collected by means of grabs. Grab samplers are selected in function of the consistency of the sediment. Some are useful for soft beds (e.g. Eckman dredge), others for more sandy or hard beds (e.g. Van Veen grab, Ponar grab). Depending on the method, a fixed area or volume, which means a certain number of grabs, must be sampled. For example, 20 litres of sediment randomly taken with a Van Veen grab are used for sediment quality assessment with the Biotic Sediment Index (BSI; De Pauw and Heylen, 2001).

In large and deep rivers, a combination of different sampling methods may have to be applied to obtain a complete picture of the macroinvertebrate communities present (e.g. handnet along the shallow river banks, examination of stones along the banks, dredging net and/or grabs or artificial substrates in the deeper parts of the river).

In conclusion, it is clear that many useful sampling methods exist but the use of a standardised sampling protocol for the application of an assessment method is an absolute requirement for obtaining comparable results.

1.4 Identification level

In order to calculate a quality index or score, the sampled macroinvertebrates must be identified up to a predefined taxonomic level. This level can vary from order, family or genus up to species level.

Various authors recommend identification to species level to ascertain a detailed insight in the community composition, avoiding information loss due to lumping of taxa, and showing a strong assemblage-environment relationship (e.g. Resh and McElravy, 1993; Stubauer and Moog, 2000; Verdonschot, 2000; Lenat and Resh, 2001; King and Richardson, 2002; Adriaenssens et al., 2004).

On the other hand, species identification is time-consuming and expensive. On top of that, information loss when identifying to genus or even family level is often small, and according to several authors it is therefore not necessary to descend to the species level (e.g. Warwick, 1988a, 1988b; Bowman and Bailey, 1997; Ghetti, 1997; Olsgard et al., 1998; Dolédec et al., 2000; Gayraud et al., 2003). Another problem associated with species level identifications is the increasing uncertainty that arises with an increasing level of detail. Ellis (1985) acknowledged this when defining taxonomic sufficiency as the level to which the organisms should be identified in order to balance the need to indicate the biological community versus accuracy of the identifications. Species identification requires more taxonomic skill and is not always possible due to a lack of suited identification keys or unavailability of keys for immature stages.

When deciding upon the taxonomic level, all aspects mentioned above should be taken into consideration. The identification level chosen is often the result of a practical trade-off between taxonomic precision and time constraints and financial resources (e.g. Guérold, 2000; Adriaenssens et al., 2004) (see further Chapter 2).

According to Guérold (2000) and Roach et al. (2001) family level is sufficient for detecting perturbations on the macroinvertebrate community, but a more detailed level of identification is necessary for ecological interpretation. Williams and Gaston (1994) proposed the use of higher-taxon categories as surrogates for species in rapid biodiversity surveys. Karr and Chu (1999) consider genus level to be sufficient for developing a multimetric index and also family level to be acceptable in case of limited time and/or financial resources.

Equally important for the successful application of assessment methods based on macroinvertebrates is the availability of suited identification keys up to the required level (e.g. De Pauw and Vannevel, 1991; Schmedtje and Kohmann, 1992; Tachet et al., 2002).

Whatever taxonomic level is used for a biotic water quality index, the level should be fixed with the method description because (1) many methods can only be calculated when using the predefined level, e.g. when taxon-specific tolerance values are defined and (2) taxonomic level can affect index calculation (e.g. Guérold, 2000; Schmidt-Kloiber and Nijboer, 2004).

The problem of invasive species is also important with regard to taxonomic identification level for biological assessment. Due to the invasion of an exotic species, an endemic species of the same genus or family might disappear, yet this will remain unnoticed if taxa are identified to the genus or family level, respectively. This is an important problem with regard to the increasing invasion of macroinvertebrate species in Europe, mainly from the Ponto-Caspian region, but also from other regions (e.g. Van Den Brink et al., 1991; Bij de Vaate et al., 2002; Nguyen and De Pauw, 2002). This will be explored further in Chapter 2.

Besides considerations with regard to biomonitoring, also bioconservation issues should be taken into account. Monitoring aquatic invertebrates is not only important for water quality assessment, but also for the study of biodiversity and conservation. If no information is collected at species level, no information will be available concerning species distributions. The Flemish Nature Report 2003 (Dumortier et al., 2003) emphasises that of all species occurring in Flanders, only an estimated ten percent is documented in Red Lists. For an early identification of biodiversity loss the development and regular revision of more Red Lists is required (Dumortier et al., 2003). The development of a Red List should therefore be considered for all macroinvertebrate groups.

1.5 Indices based on macroinvertebrates

A large and still increasing number of quality indices has been developed based on macroinvertebrate communities. Overviews can be found in e.g. Metcalfe (1989), De Pauw and Hawkes (1993), Rosenberg and Resh (1993), Verdonschot and Dohet (2000), Sandin et al. (2000) and De Pauw et al. (2006). Here, only assessment methods taking the entire macroinvertebrate community into account will be discussed. Indices based on single groups of macroinvertebrates, e.g. chironomids or oligochaetes, will not be discussed here, unless they are components of an overall macroinvertebrate assessment system. Also assessment methods based on organism-level indicators (biochemical, physiological, morphological deformities, behavioural responses, and life-history responses) are not considered here.

The majority of freshwater assessment systems based on macroinvertebrates was developed for rivers, while a far smaller number of systems was developed for lakes, reservoirs, wetlands, sediment, or for general use.

Indices that are used for assessing biological quality of freshwater based on macroinvertebrates can be classified into the following categories (modified from Resh and Jackson, 1993; Thorne and Williams, 1997; Verdonschot, 2000; De Pauw et al., 2006):

1. Single metric indices:
    - Non-taxonomic metrics;
    - Sensitivity/tolerance metrics;
    - Functional metrics;
    - Similarity metrics.
2. Combined indices:
    - Biotic indices (table-based);
    - Biotic indices (formula-based);
    - Multimetric indices.
3. Stressor-specific indices.

The cited categories of assessment methods will be discussed in the following paragraphs. An overview of existing freshwater assessment methods that can be applied with macroinvertebrates, organised in the cited categories, is provided in Table 1.3. Although the sequential order of the three categories does not reflect the historical development of assessment approaches, it was found useful for the purpose of this review, since combined approaches make use of two or more single properties, and stressor-specific indices can usually be regarded as modifications of systems from the two previous categories.

Throughout this overview, a distinction will be made between metrics and indices. A metric may be any community descriptor that can be directly calculated based on the available data, while an index is considered to be a variable that is (or has been) used for assessment, based on an assumed relation of the variable with ecological degradation. Consequently, an index may integrate one or more metrics, whereas a single metric is not necessarily used for the purpose of assessment.

1.5.1 Single metric indices

The category of single metric indices includes all variables that represent single characteristics of a community that can be directly calculated using its basic features. Some of these metrics have also been incorporated into combined indices. However, only those metrics that have reportedly been used as a separate assessment index for macroinvertebrates will be discussed here.

Non-taxonomic metrics

Non-taxonomic metrics, usually referred to as diversity indices, include all variables that are based on measured properties of a community without differentiating among taxon-specific characteristics. In contrast to all other types of metrics that will be discussed here, these metrics may therefore be applied to any type of community because they do not require autecological knowledge.

Diversity indices use one or more of three components of community structure: richness, evenness and abundance (Washington, 1984). Hurlbert (1971) emphasises that although richness and diversity are often positively correlated, an increase in species diversity can be accompanied with a decrease in species richness.

Diversity indices are based on the principle that disturbance of the water ecosystem leads to a reduction in diversity (Hellawell, 1986; De Pauw et al., 2006). Typical examples that have frequently been applied using macroinvertebrate communities are the Shannon-Wiener index (Shannon and Weaver, 1949), the Simpson index (Simpson, 1949), Brillouin's diversity index (Brillouin, 1951), the Margalef index (Margalef, 1958) and the Evenness index (Hill, 1973). A comprehensive review of evenness indices can be found in Beisel et al. (2003).

The Sequential Comparison Index (SCI; Cairns et al., 1968) can also be regarded as a diversity index. This index is based on the number of runs (subsequent series of organisms of the same taxon when all individuals are observed in a random order) in relation to the total number of organisms.

The advantages of diversity indices lie in the fact that they are easy to use and calculate, are applicable to all kinds of watercourses and have no geographical limitations. They are best used for comparative purposes. Having no clear endpoint or reference level is however the main problem; the diversity in natural undisturbed waters can indeed vary considerably, and moreover, all species have an equal weight. This is probably the reason why not one country in Europe has been adopting a diversity index as a national standard for biological water quality assessment (see Ghetti and Ravera, 1994; Nixon, 2003). Because diversity indices are not sensitive to changes in species composition, Camargo (1992b) argues that diversity indices cannot replace biotic indices (see further) and both index types should be used complementarily in ecological studies.

Sensitivity/tolerance metrics

Sensitivity/tolerance metrics are based on the observed principle that macroinvertebrate taxa will disappear with increasing levels of disturbance (De Pauw et al., 2006). MacKenthun (1969) observed the following stepwise disappearance of macroinvertebrate groups subsequent to increasing pollution: stoneflies (Plecoptera), mayflies (Ephemeroptera), caddisflies (Trichoptera), scuds (Amphipoda), aquatic sowbugs (Isopoda), midges (Diptera) and bristle worms (Oligochaeta).

The first index of this type was described by Chutter (1972) for watercourses in South Africa. Chutter's Biotic Index is calculated as an abundance-weighted mean of the scores of all present taxa. For this index, the taxon scores vary for some taxa with the presence of other taxa (Chutter, 1972). Many indices of this type however are derivatives of a biotic score system such as the BMWP (see further). The Average Score Per Taxon (ASPT; Armitage et al., 1983) for instance is calculated as the mean of the tolerance scores of all present taxa (Armitage et al., 1983). Similarly, an IASPT (Iberian ASPT; Alba-Tercedor and Sánchez-Ortega, 1988) can be derived from the IBMWP (Iberian BMWP; Alba-Tercedor and Sánchez-Ortega, 1988). Cook (1976) introduced the Averaged Chandler Score, an ASPT for the Chandler Score. Average score systems applied in New Zealand are the Macroinvertebrate Community Index (MCI; Stark, 1985), the abundance-weighted Quantitative MCI or QMCI (Stark, 1993), and the Semi-Quantitative MCI (SQMCI) which uses abundance classes being transformed into actual abundances (Stark, 1998). The North Carolina Biotic Index (NCBI; Lenat, 1993) is similar to Chutter's index. The Potamon Typie Index (PTI; Schöll and Haybach, 2000) is calculated as the mean of the squares of the indicator values of all taxa. These indicator values range from 1 to 5, and consequently the PTI ranges from 1 to 25. Rossaro et al. (2007) developed an index for Italian lakes called the Benthic Quality Index Modified (BQIM).

Sensitivity metrics have the advantage that they are easy to use and interpret. Because it is an average value, it does not depend on the number of taxa. For this reason, the ASPT is less sensitive to sampling effort (Armitage et al., 1983) and it is also less prone to be affected by seasonal variation than the BMWP (Armitage et al., 1983; Hawkes, 1998).

Functional metric indices

Functional metrics are based on ecological traits of the present community, such as functional feeding mechanism or food type or moving behaviour. Functional metrics have rarely been used as single indicators for biological quality based on macroinvertebrates, although they are frequently used as composing metrics in multimetric indices (see further). Two indices have been described that are entirely based on functional characteristics: the Rhithron-Ernährungstypen-Index (RETI; Schweder, 1990) and the Index of Trophic Completeness (ITC; Pavluk et al., 2000; Bij de Vaate and Pavluk, 2004).

The ITC was developed for assessment of river ecosystem status based on analysis of the trophic structure of benthic macroinvertebrate communities. Approximately 300 species of macroinvertebrates were characterised by trophic criteria plant-animal ratio in the diet, feeding mechanism, food size, food acquisition behaviour, energy- and mass transfers. Based on this characterisation the species were divided into twelve trophic groups. Assuming that in an undisturbed benthic macroinvertebrate community each trophic group should be present, each missing group represents a deterioration in the benthic community (Pavluk et al., 2000).

Similarity metrics

In this approach the assessment is based on the degree of similarity of the community composition to a pre-defined target community. This target, usually called the reference, can be based on actual samplings, expert knowledge, historical data or predictive models, or a combination of these. Methods for deriving reference conditions are further discussed in paragraph 1.7; the present section provides an overview of existing methods for quantifying the similarity between the observed and the target community.

The most straightforward method of comparing an observed to a reference community is to calculate the proportion of the assessment value obtained with any index system between both communities. This is called the Ecological Quality Ratio (EQR) according to the Water Framework Directive (EU, 2000; Wallin et al., 2003). This ratio is expressed as a numerical value between zero and one, where zero represents a very bad ecological status and one a very good ecological status (Wallin et al., 2003):

$$\text{EQR} = \frac{\text{index value of observed community}}{\text{index value of reference community}}$$

An example of an EQR is the Environmental Quality Index (EQI) based on the “River Invertebrate Prediction And Classification System” (RIVPACS) developed in the UK (Wright et al., 1993; Wright, 2000). This system produces a site-specific prediction of the macroinvertebrate taxa that should be present under undisturbed conditions based on a number of physical-chemical features of the examined site. These predicted reference conditions can then be compared with the observed macroinvertebrate communities by calculating the EQI, which is in accordance with the WFD. In fact, the RIVPACS philosophy was an important influence in the drafting process of the WFD (Logan and Furse, 2002). The RIVPACS-EQI can be calculated with different metrics or indices, for example the BMWP, the ASPT or the number of taxa (Wright, 2000). Based on the RIVPACS approach, similar systems have been developed in other countries (see paragraph 1.6.2).

Many index systems however implicitly incorporate a comparison with the reference conditions because the upper bound of the assessment scale is assumed to reflect the reference community. When calculating a multimetric index (see further) the comparison with reference conditions is usually performed for each individual composing metric separately.

Other methods for quantifying the similarity between two sites are Jaccard’s similarity index (Jaccard, 1908), the Sørensen index (Sørensen, 1948), the Bray-Curtis similarity index (Bray and Curtis, 1957) and the Coefficient of Community Loss (Courtemanch and Davies, 1987). These indices are based on the number of common and unique taxa that are found at the compared sites.

1.5.2 Combined indices

Combined indices integrate characteristics of two or more single metric types into one overall measure. In this way, the assessment incorporates more ecological information in comparison to the single metric indices. A large number of combined indices has been proposed thus far.

Among the combined indices, the biotic approach integrates features of the sensitivity and the diversity approach. Two different types can be distinguished within the biotic approach: the table-based biotic indices and the formula-based biotic indices, often referred to as biotic scores. More recently, other combined indices have been developed, called multimetric indices. The difference with the biotic approach is that all metrics are calculated separately and subsequently integrated into an overall index, whereas the biotic index systems provide an overall evaluation without explicitly assessing all separate components.

Biotic indices (table-based)

In the biotic index approach the index is deduced from a table that takes into account the number of taxa and the sensitivity of the most sensitive taxon encountered. The first index of this type was the Trent Biotic Index (Woodiwiss, 1964), later extended to an Extended Biotic Index (EBI; Woodiwiss, 1978).

Some examples of biotic indices, all based directly or indirectly on the Trent Biotic Index, are the French Indice Biotique (Tuffery and Verneaux, 1968), the Indice Biologique Global Normalisé (IBGN; AFNOR, 1992), and its modification for large rivers, the Indice Biologique Global Adapté (IBGA; AERMC, 1997). In Belgium, the Belgian Biotic Index (BBI; De Pauw and Vanhooren, 1983; BIN, 1984; see further Chapter 2) was developed. In Italy, an Italian modification of the EBI was proposed called the Indice Biotico Esteso (IBE; Ghetti and Bonazzi, 1980), and in Denmark an index known as the Viborg Index (Andersen et al., 1984) has been used, which was later modified to become the Danish Fauna Index (DFI) and ultimately the Danish Stream Fauna Index (DSFI; Skriver et al., 2001). An extensive overview of biotic indices that have been described to date can be found in Table 1.3.

The Macroindex (Perret, 1977) is based on a similar table but where most biotic indices use number of taxa, the Macroindex uses the ratio insect/non-insect taxa. It is therefore the only table-based index that is not based on sensitivity and richness but on sensitivity and a composition metric, although it can be assumed that the ratio insect/non-insect taxa is correlated with diversity.

The advantages of the table-based biotic indices are their simplicity and straightforwardness in use and interpretation. Unlike most formula-based biotic indices, the results of a table-based biotic index are restricted to a predefined interval range (often 0-10 or 0-20), which facilitates communication of results.

A disadvantage of this type of indices is the rigid character of a table, which complicates the possibility to make regional or typological adaptations. This problem may be overcome by using an ecological quality ratio by setting a type-specific reference value, but this possibility is practically restricted to indices using a sufficiently differentiated scale (e.g. the 0-20 scale of the IBGN).

Biotic indices (formula-based)

In the biotic score system a predefined score is allocated to each taxon. These individual taxon-scores depend on their sensitivity to pollution. For calculating the score of a site, all individual taxon scores of the encountered taxa are summed. The best-known example of a biotic score is the BMWP (Biological Monitoring Working Party) score (Chesters, 1980) and its revised version (National Water Council, 1981). A modified version of the BMWP, the IBMWP (formerly known as BMWP'), was developed for the Iberian Peninsula (Alba-Tercedor and Sánchez-Ortega, 1988; Alba-Tercedor et al., 2002). An Australian score of this type is the Stream Invertebrate Grade Number-Average Level score (SIGNAL; Chessman, 1995) and its revised version SIGNAL 2 (Chessman, 2003). A method similar to the BMWP, but with scores for each taxon being different over five abundance classes, was already published in 1970 by Chandler. Adaptations of the BMWP were also formulated for Thailand ($BMWP^{THAI}$; Mustow, 2002), Hungary (MMCP; Csányi, 1998) and Poland (BMWP-PL; Kownacki et al., 2004).

Cao et al. (1997) proposed a new index by multiplying the Averaged Chandler Score by the logarithm of the number of species. This modified Averaged Chandler Score can therefore be regarded as a score system where species richness is downweighted relative to sensitivity when compared to the original Chandler Score.

Like the table-based biotic indices, the formula-based indices are transparent and easy to apply. However, the main disadvantage of most formula-based biotic indices is that they lack a clear target because the more taxa-rich the sample is, the higher the index value; which hampers a straightforward interpretation.

A special case of the formula-based biotic index is the Lake Biotic Index (Verneaux et al., 2004), which is based on a calculation formula that incorporates measures based on tolerance as well as measures based on richness, but in contrast to the BMWP-type indices, the LBI values are constrained to an interval of 0-20 (Verneaux et al., 2004).

Multimetric indices

In multimetric systems, several metrics representing different characteristics of the community are combined into one index value or score which is an expression of the overall quality. It is assumed that incorporating more descriptors, will result in an index being more diagnostic of ecosystem health. The individual metrics included can be any kind of the single metrics discussed earlier, but also combined indices such as the BMWP are sometimes used in a multimetric system. Also, metrics representing only a part of the macroinvertebrate community, e.g. chironomids or Trichoptera can be used in combination with other metrics in order to represent the whole community. The metrics can be combined in several ways, e.g. by (weighted) averaging or by assigning scores to the individual metrics and subsequently calculating the sum or the (weighted) average of these scores.

The first index that was explicitly labelled “multimetric” was developed for fish communities by Karr (1981). Later, multimetric indices were also developed for other indicator groups, such as diatoms (e.g. Fore and Grafe, 2002) or plants (e.g. DeKeyser et al., 2003), but also for macroinvertebrate communities. Examples of multimetric indices based on macroinvertebrates for rivers are described by Barbour et al. (1992, 1999), Kerans and Karr (1994), Fore et al. (1996), Thorne and Williams (1997), Paller and Specht (1997), Stribling et al. (1998), Karr and Chu (1999), Major et al. (2001), Mebane (2001), Royer et al. (2001), Blocksom (2003), Butcher et al. (2003a) and Klemm et al. (2003). Examples of multimetric indices based on macroinvertebrates for lakes are Gerritsen et al. (2000b), Lewis et al. (2001) and Blocksom et al. (2002). Within the context of the implementation of the WFD, the European project AQEM developed a strategy and methodology for the establishment of multimetric assessment systems for different streams in Europe based on macroinvertebrates (Hering et al., 2004).

A number of macroinvertebrate indices that were developed in the past demonstrate all properties of a multimetric index although they were not described as such. These indices are therefore also included in this section in Table 1.3. For example, the Hellenic Evaluation Score Interpretation Index (HESII; Artemiadou and Lazaridou, 2005) is an index that equals the average of the quality class (1-5) of the HES (Hellenic Evaluation Score) on the one hand and that of the AHES (Average Hellenic Evaluation Score) on the other hand. HES and AHES are indices of the BMWP and the ASPT-type, respectively. The HESII should therefore also be seen as a multimetric index consisting of two metrics that are combined using a score system. Similarly, the Lincoln Quality Index (LQI) is the average of a 1-7 rating based on the ASPT and a 1-7 rating based on the BMWP. First described by Extence et al. in 1987, the LQI is therefore actually the earliest example of a multimetric index for macroinvertebrates, together with the Invertebrate Community Index (ICI), published in the same year by Ohio EPA.

Metric types that can be incorporated in a multimetric index include (see Resh and Jackson, 1993; Thorne and Williams, 1997; Verdonschot and Dohet, 2000): any of the four previously discussed single metric indices (diversity, sensitivity, functional or similarity metrics), but also combined indices as well as single metrics that are based on specific groups of macroinvertebrates (e.g. numbers or percentages of taxa belonging to certain groups or relative abundances).

An important advantage of multimetric indices is their flexibility. They can be easily adapted to a regional situation, by taking into account the most appropriate metrics and by evaluating each metric to an appropriate target. The flexibility of this type of index is probably the reason why the majority of the indices that were developed in recent years were multimetric indices.

1.5.3 Stressor-specific indices

The index systems discussed in the previous sections were all developed to provide a general indication of ecological degradation caused by any kind of stressor. However, several indices have been developed for identifying specific kinds of degradation. Considering the calculation system, these indices can generally be assigned to one of the previously discussed index types, except that they were developed for the purpose of identifying the presence of a specific source of stress. For instance, the Nutrient Biotic Index for Nitrate (Smith et al., 2007) is computationally identical to an abundance-weighted ASPT-type system (e.g. Chutter's Biotic Index), but the tolerance scores are assumed to reflect tolerance to nitrate enrichment.

Some authors argue that the potential of multimetric index systems for identifying causes of degradation is limited, although they are able to distinguish disturbed from undisturbed sites (e.g. Winger et al., 2005). According to Chessman and McEvoy (1998), the development of stressor-specific indices is a realistic possibility, at least for certain types of disturbance, although for some specific stressors it may require a more detailed level of identification (e.g. genus or species level).

Types of stressors for the detection of which a macroinvertebrate-based system has been developed, include saprobity, nutrient enrichment and acidification.

Saprobic indices are based on the sensitivity of the present indicator organisms to organic pollution. Historically, the saprobic approach was actually the first biological river assessment system ever developed. The saprobic system was introduced by Kolkwitz and Marsson (1909) and later adapted by several authors (e.g. Liebmann, 1962; Sládeček, 1973a). Each species has a specific dependency of organic substances and thus of the dissolved oxygen content. This tolerance is expressed as a saprobic indicator value. The advantage is that a quick classification of the investigated community can be made on a universal scale. A major problem is the identification of the organisms up to species level. The saprobic index calculation also requires the assessment of abundances. The indicator system furthermore implies more knowledge than actually exists: pollution tolerances are subjective and based on ecological observations and are rarely confirmed by experimental studies (e.g. De Pauw et al., 2006).

Although many of the assessment indices that were later developed are considered to assess general degradation, many of these systems, e.g. the biotic indices, were considerably influenced by the saprobic approach, either directly or indirectly.

Other examples of stressor-specific indices based on macroinvertebrate include indices for detecting the impact of acidification (e.g. Sandin et al., 2004), for indicating organic pollution (e.g. Dahl and Johnson, 2004; Dahl et al., 2004), or for nutrient enrichment (e.g. Smith et al., 2007).

In conclusion, stressor-specific indices can be an interesting complementary tool for the indices for general degradation, because they can be helpful for cause allocation under conditions of deterioration. Nonetheless, they cannot replace the applicable methods for identifying the stressor in question (e.g. hydromorphological surveys or physical-chemical measurements).

Table 1.3. Overview of freshwater assessment methods based on macroinvertebrates

Method	Evaluated systems	Region	Reference
1. Single metrics			
1.1 Non-taxonomic metrics			
Shannon-Wiener Index (H’)	General	Any	Shannon and Weaver, 1949
Simpson’s dominance Index (D)	General	Any	Simpson, 1949
Brillouin index	General	Any	Brillouin, 1951
Margalef Index	General	Any	Margalef, 1958
Menhinick Index	General	Any	Menhinick, 1964
Pielou’s evenness index	General	Any	Pielou, 1966
Sequential Comparison Index (SCI)	General	Any	Cairns et al., 1968
Probability of Interspecific Encounter (PIE)	General	Any	Hurlbert, 1971
Hill’s evenness index	General	Any	Hill, 1973
Camargo’s Diversity Index	General	Any	Camargo, 1992a
1.2 Sensitivity/Tolerance metrics			
Chutter’s Biotic Index	Running waters	South Africa	Chutter, 1972
Averaged Chandler Score	Running waters	Lothian Area, Scotland, UK	Cook, 1976
Average Score Per Taxon (ASPT)	Running waters	UK	Armitage et al., 1983
Macroinvertebrate Community Index (MCI)	Running waters	New Zealand	Stark, 1985
Iberian ASPT (IASPT) (formerly known as ASPT’)	Running waters	Iberian peninsula	Alba-Tercedor and Sánchez-Ortega, 1988
Average Biological Monitoring Water Quality (a-BWMQ)	Running waters	Iberian peninsula	Camargo, 1993
North Carolina Biotic Index (NCBI)	Running waters	North Carolina, USA	Lenat, 1993
Quantitative Macroinvertebrate Community Index (QMCI)	Running waters	New Zealand	Stark, 1993
Stream Invertebrate Grade Number Average Level (SIGNAL)	Running waters	Eastern Australia	Chessman, 1995
Nepalese Biotic Score (NEPBIOS)	Running waters	Nepal	Sharma and Moog, 1998
Semi-Quantitative Macroinvertebrate Community Index (SQMCI)	Running waters	New Zealand	Stark, 1998
Potamon Typie Index (PTI)	Large running waters	Germany	Schöll and Haybach, 2000
Swan Wetlands Aquatic Macroinvertebrate Pollution Score (SWAMPS)	Wetlands	Swan Coastal Plain, Australia	Chessman et al., 2002
$ASPT^{THAI}$	Running waters	Thailand	Mustow, 2002
SIGNAL 2	Running waters	Australia	Chessman, 2003
Average Hellenic Evaluation Score (AHES)	Running waters	Central and Northern Hellas	Artemiadou and Lazaridou, 2005

Table 1.3 (continued)

Invertebrate Species Index (ISI)	Running waters	Southeast Queensland, Australia	Haase and Nolte, 2007
Benthic Quality Index Modified (BQIM)	Lakes	Italy	Rossaro et al., 2007
1.3 Functional metrics			
Index of Trophic Completeness (ITC)	Running waters	Europe and Russia	Pavluk et al., 2000
Rhithron-Ernährungstypen-Index (RETI)	Running waters	Germany	Schweder, 1990
1.4 Similarity metrics			
Jaccard’s similarity index	General	Any	Jaccard, 1908
Sørensen index	General	Any	Sørensen, 1948
Bray-Curtis similarity index	General	Any	Bray and Curtis, 1957
Species Deficit Index	General	Any	Kothé, 1962
Pinkham-Pearson similarity index	General	Any	Pinkham and Pearson, 1976
Coefficient of Community Loss	General	Any	Courtemanch and Davies, 1987
Camargo’s Ecotoxicological Index	General	Any	Camargo, 1990
AUSRIVAS EQI	Running waters	Australia	Davies, 2000
RIVPACS EQI (ASPT)	Running waters	UK	Wright, 2000
RIVPACS EQI (BMWP)	Running waters	UK	Wright, 2000
RIVPACS EQI (NFAM)	Running waters	UK	Wright, 2000
BEAST EQI	Lake sediment	Great Lakes, North America	Reynoldson et al., 2000
SWEPACS EQI	Running waters	Sweden	Sandin, 2001
PERLA EQI	Running waters	Czech Republic	Kokeš et al., 2006
Mondego Model	Running waters	Mondego river basin (Portugal)	Feio et al., 2007
2. Combined indices			
2.1 Biotic indices (table-based)			
Trent Biotic Index (TBI)	Running waters	Trent River Catchment, UK	Woodiwiss, 1964
Graham’s Index	Running waters	Lothian Area, Scotland, UK	Graham, 1965
Indice Biotique (IB)	Running waters	France	Tuffery and Verneaux , 1968
Macroindex	Running waters	Switzerland	Perret, 1977
Extended Biotic Index (EBI)	Running waters	Trent River Catchment, UK	Woodiwiss, 1978
Indice Biotico Esteso (IBE)	Running waters	Italy	Ghetti and Bonazzi, 1980
Belgian Biotic Index (BBI)	Running waters	Belgium	De Pauw and Vanhooren, 1983; BIN, 1984

Table 1.3 (continued)

BILL	Running waters	Spain	Prat et al., 1983
Viborg Index	Running waters	Denmark	Andersen et al., 1984
Indice Biologique Global (IBG)	Running waters	France	AFNOR, 1985
Indice Biologique Global Normalisé (IBGN)	Running waters	France	AFNOR, 1992
Indice Biologique Global Adapté (IBGA)	Large running waters	France	AERMC, 1997
FBILL	Running waters	Spain	Prat et al., 1999
Danish Fauna Index (DFI)	Running waters	Denmark	Kirkegaard et al., 1992
Biotic Sediment Index (BSI)	River sediment	Flanders, Belgium	De Pauw and Heylen, 2001
Biotic Index for PAMPean rivers and streams (IBPAMP)	Running waters	Pampean plains, Argentina	Rodriguez Capítulo et al., 2001
Danish Stream Fauna Index (DSFI)	Running waters	Denmark	Skriver et al., 2001
2.2. Biotic indices (formula-based)			
Beck’s Biotic Index	Freshwater	Florida, USA	Beck, 1955
Chandler Score	Running waters	Lothian Area, Scotland, UK	Chandler, 1970
Biological Monitoring Working Party Score (BMWP)	Running waters	UK	Chesters, 1980
Revised BMWP	Running waters	UK	National Water Council, 1981
Iberian BMWP (IBMWP) (formerly known as BMWP’)	Running waters	Iberian peninsula	Alba-Tercedor and Sánchez-Ortega, 1988
Total Biological Monitoring Water Quality (t-BMWQ)	Running waters	Iberian peninsula	Camargo, 1993
Modified Averaged Chandler Score	Running waters	Trent River Catchment, UK	Cao et al., 1997
Magyar Makrozoobenton Család Pontrendszer (MMCP)	Running waters	Hungary	Csányi , 1998
$BMWP^{THAI}$	Running waters	Thailand	Mustow, 2002
BMWP-PL	Running waters	Poland	Kownacki et al., 2004
Lake Biotic Index (LBI)	Lakes	France	Verneaux et al., 2004
Hellenic Evaluation Score (HES)	Running waters	Central and Northern Hellas	Artemiadou and Lazaridou, 2005
2.3 Multimetric indices			
Lincoln Quality Index (LQI)	Running waters	East of England, UK	Extence et al., 1987
Invertebrate Community Index (ICI)	Running waters	Ohio, USA	Ohio EPA, 1987
RIVAUD	Running waters	Western Switzerland	Lang et al., 1989
Mean Point Score (MPS)	Running waters	Texas, USA	Twidwell and Davis, 1989
Rapid Bioassessment Protocol (RBP)	Running waters	USA	US EPA, 1989; Barbour et al., 1992, 1999
Benthic Index of Biotic Integrity (B-IBI)	Running waters	Tennessee Valley, USA	Kerans and Karr, 1994

Table 1.3 (continued)

RIVAUD 95	Running waters	Western Switzerland	Lang and Reymond, 1995
Florida Stream Condition Index (FSCI)	Running waters	Florida, USA	Barbour et al., 1996
Benthic Index of Biotic Integrity (B-IBI)	Running waters	Oregon	Fore et al., 1996
Invertebrate Community Index - St Lawrence (ICI-SL)	Lakes	St Lawrence River ecoregion, Canada	Pinel-Alloul et al., 1996
Hester-Dendy Multimetric Index (HDMI)	Running waters	South Carolina, USA	Paller and Specht, 1997
Multimetric system of bioassessment	Running waters	Thailand, Ghana, Brazil	Thorne and Williams, 1997
Benthic Index of Biotic Integrity (Benthic IBI)	Running waters	Maryland, USA	Stribling et al., 1998
BalkaN Biotic Index (BNBI)	Running waters	Serbia	Simić and Simić, 1999
West Virginia Stream Condition Index (WV-SCI)	Running waters	West Virginia, USA	Gerritsen et al., 2000a
Florida Lake Condition Index (LCI)	Lakes	Florida, USA	Gerritsen et al., 2000b
Lake Bioassessment Integrity Index (LBII)	Lakes	New England, USA	Lewis et al., 2001
Alaska Stream Condition Index (ASCI)	Running waters	Alaska, USA	Major et al., 2001
Macroinvertebrate Biotic Index (MBI)	Running waters	Idaho, USA	Mebane, 2001
River Macroinvertebrate Index (RMI)	Running waters	Idaho,USA	Royer et al., 2001
Lake Macroinvertebrate Integrity Index (LMII)	Lakes and reservoirs	New Jersey, USA	Blocksom et al., 2002
Stream Macroinvertebrate Index (SMI)	Running waters	Idaho,USA	Jessup and Gerritsen, 2002
Invertebrate Index of Biological Integrity (Invertebrate IBI)	Wetlands	Minnesota, USA	US EPA, 2002
Macroinvertebrate-based index of biotic integrity (IBI)	Running waters	West-central Mexico	Weigel et al., 2002
Virginia Stream Condition Index for Non-Coastal Streams (VA-SCI)	Running waters	Virginia, USA	Burton and Gerritsen, 2003
Benthic Community Index	Running waters	Northern Lakes and Forests Ecoregion,USA	Butcher et al., 2003a, 2003b
Macroinvertebrate Biotic Integrity Index (MBII)	Running waters	Mid-Atlantics Highlands, USA	Klemm et al., 2003
Mississippi Benthic Index of Stream Quality (M-BISQ)	Running waters	Mississippi, USA	MDEQ, 2003
Kentucky Macroinvertebrate Bioassessment Index (Kentucky MBI)	Running waters	Kentucky, USA	Pond et al., 2003
Invertebrate Community Index	Running waters	Alabama, USA	Bennett et al., 2004
Multimetric Index (MMI)	Running waters	Germany	Böhmer et al., 2004a
IM9	Running waters	Southern siliceous basins in Portugal	Pinto et al., 2004
AQEM	Running waters	Europe	Hering et al., 2004
Hellenic Evaluation Score Interpretation Index (HESII)	Running waters	Central and Northern Hellas	Artemiadou and Lazaridou, 2005
Potomac Basin-wide Index of Benthic Integrity (B-IBI)	Running waters	Potamac River Basin (USA)	Astin, 2007
Serra dos Órgãos Multimetric Index (SOMI)	Running waters	Serra dos Órgãos, Brazil	Baptista et al., 2007
Intercalibration Common Metric Index (ICMi)	Running waters	Europe	Buffagni et al., 2007

Table 1.3 (continued)

Multimetric Macroinvertebrate Index Flanders (MMIF)	Rivers and lakes	Flanders, Belgium	Gabriels et al., 2007
Vermont Lake Condition BioIndex	Lakes	Vermont, USA	Kamman, 2007
Maatlatten riveren en meren voor de Kaderrichtlijn Water (WFDi)	Rivers and lakes	The Netherlands	van der Molen and Pot, 2007a, 2007b
3. Stressor-specific indices			
3.1 Organic pollution			
Biologically Effective Organic Loading (BEOL)	Running waters	Germany	Knöpp, 1954
Saprobic index	Running waters	Germany	Pantle and Buck, 1955
K_{135} (saprobic)	Running waters	The Netherlands	Gardeniers and Tolkamp, 1976
K_{12345} (saprobic)	Running waters	The Netherlands	Gardeniers and Tolkamp, 1976
Hilsenhoff Biotic Index (HBI) (organic pollution)	Running waters	Wisconsin, USA	Hilsenhoff, 1977
Hilsenhoff Biotic Index (HBI) (1982 version) (organic pollution)	Running waters	Wisconsin, USA	Hilsenhoff, 1982
Improved Hilsenhoff Biotic Index (HBI) (organic pollution)	Running waters	Wisconsin, USA	Hilsenhoff, 1987
Hilsenhoff Family-level Biotic Index (HFBI) (organic pollution)	Running waters	Wisconsin, USA	Hilsenhoff, 1988
Modified Hilsenhoff Biotic Index (HBI) (organic pollution)	Running waters	Wisconsin, USA	Hilsenhoff, 1998
Organic pollution index	Running waters	Southern Sweden	Dahl and Johnson, 2004
3.2 Nutrient enrichment			
Nutrient Biotic Index for Nitrate (NBI-N)	Running waters	New York State, USA	Smith et al., 2007
Nutrient Biotic Index for total Phosphorus (NBI-P)	Running waters	New York State, USA	Smith et al., 2007
3.2 Acidification			
Acidification index	Running waters	Germany	Braukmann and Biss, 2004

1.6 Defining reference conditions for macroinvertebrates

An essential and critical point in biological assessment is the establishment of reference conditions, which constitutes the conceptual basis of an assessment method (e.g. Hughes et al., 1986; Hughes 1995; Reynoldson et al., 1997; Kennard et al., 2006; see also subheading "similarity metrics" of paragraph 1.5.1).

The reference conditions for a given site can be defined as the best available conditions that can be expected at a similar site, and are derived from observations at several similar sites (Hughes et al., 1986; Hughes, 1995; Reynoldson et al., 1997). However, in several regions, in particular in Europe, reference conditions do not exist anymore and must be derived using alternative methods (De Pauw et al., 2006). Several alternative methods for defining the reference state have been proposed, including the use of historical data, paleoecological data, predictive models, and expert judgment (Hughes, 1995; Wallin et al., 2003).

The WFD explicitly requires that biological assessment methods take reference conditions into account by using an EQR. Reference conditions are defined by the WFD as the values of the biological quality elements that are characterised by a very good ecological status. These values should correspond to those normally associated with that type of water body under undisturbed conditions and show no or only very minor evidence of distortion. Furthermore, they must be described separately for each type of water body (EU, 2000; see also paragraph 1.2.3). This implies that the WFD considers that reference conditions are not necessarily totally undisturbed, pristine conditions. They may include very minor disturbance, which means that human pressure is allowed as long as there are no or only very minor ecological effects (Wallin et al., 2003).

Specifically for macroinvertebrates, the WFD defines the very good ecological status using the following criteria (see Table 1.2):

- the taxonomic composition and abundance correspond totally or nearly totally to the undisturbed conditions;
- the ratio of disturbance sensitive taxa to insensitive taxa shows no signs of alteration from undisturbed levels;

- the level of diversity of invertebrate taxa shows no sign of alteration from undisturbed levels.

Wallin et al. (2003) identified the following options for establishing reference conditions according to the WFD:

- spatially based reference conditions using data from monitoring sites;
- reference conditions based on predictive modelling;
- temporally based reference conditions using either historical data or paleoreconstruction or a combination of both;
- a combination of the above approaches;
- where it is not possible to use these methods, reference conditions can be established by expert judgment.

1.6.1 Spatially based reference conditions

Spatially based reference conditions are derived from field samplings at locations that are considered to be in reference state. An advantage of this method is that it is region-specific (Wallin et al., 2003). On the other hand, it is expensive and time-consuming if it can not be based on existing sampling data (Wallin et al., 2003). Moreover, it is not possible to find reference locations in some regions, such as Flanders.

An additional difficulty is the establishment of criteria to choose reference locations. Wallin et al. (2003) proposed a set of "pressure screening" criteria to decide whether a water body is a suitable reference site (see also CB-GIG, 2006).

1.6.2 Reference conditions based on predictive modelling

Using modelling techniques, site-specific predictions can be made regarding community composition under reference conditions. The advantage of the use of models for predicting reference conditions is that they generate site-specific predictions. The main difficulty is that it requires field data for calibrating the models (Wallin et al., 2003). Because the data used for calibrating the models are collected at reference sites, this technique is not an alternative to spatially based reference conditions for regions where reference locations are lacking.

The earliest model for predicting reference conditions was developed in the UK, called RIVPACS (River Invertebrate Prediction and Classification System; Wright, 2000). This system predicts which macroinvertebrate taxa will be present at a watercourse in the absence of disturbances, based on a number of abiotic variables such as geographical coordinates, stream width and depth, slope, discharge, and substratum composition. This model was developed based on a large database of a broad variety of sites that are all considered as reference (Wright, 2000; Clarke et al., 2003).

Later, similar systems were developed for Australia (AUSRIVAS: Australian River Assessment Scheme; Davies, 2000), the Great Lakes in North America (BEAST: Benthic Assessment of Sediment; Reynoldson et al., 2000), Sweden (SWEPACS; Sandin, 2001), the Czech Republic (PERLA; Kokeš et al., 2006) and Portugal (Feio et al., 2007).

1.6.3 Temporally based reference conditions

Temporally based reference conditions may be derived using either historical data or paleoreconstruction or a combination of both (Wallin et al., 2003). The use of historical records can be interesting because it does not require field work and is therefore less expensive, but data are usually unavailable or incomplete and the quality and confidence of the available data is often unknown (Wallin et al., 2003).

The use of paleoreconstruction can be an interesting alternative as well. The advantage of this method is that it is site-specific and can incorporate both physicochemical and biological data. On the other hand this is practically restricted to lakes only and requires high investment costs. It is also restricted to certain biological indicators (Wallin et al., 2003). For macroinvertebrate communities, it has to date not been possible to define reference conditions based on historical data or paleoreconstruction, and it seems unlikely that it will be possible in future.

1.6.4 Reference conditions based on expert judgment

In cases where none of the previously discussed methods is suitable for establishing reference conditions, expert knowledge will be required to describe reference conditions (Wallin et al., 2003).

The advantage of using expert judgment is that all available sources of information can be incorporated into the final description of reference, including spatial, temporal, and model-based knowledge (Wallin et al., 2003). The main weakness of this method is its arbitrary character, and hence the description of reference may be biased (Wallin et al., 2003).

A list with possibly occurring macroinvertebrates in different types of Flemish watercourses and stagnant waters with high ecological quality was published by De Loose et al. (1995). However, the authors did not describe how this list was produced or how it should be used to estimate community composition at a single site under reference conditions (e.g. using probability of occurrence per taxon). It is therefore not feasible to use this list as such in the description of reference state for Flanders.

1.7 Conclusions

The overview presented in this chapter clearly demonstrates that even after a century of endeavour, the interest in the development of biological freshwater assessment methods worldwide is still expanding and in full evolution (Karr and Chu, 2000; De Pauw et al., 2006). Whereas in many developing countries a serious start is now being given to introducing and developing biological methods for freshwater assessment, in numerous developed countries on the other hand, the existing methods applied since many years are now in the process of being optimised and internationally standardised (Heiskanen et al., 2004; De Pauw et al., 2006), while additional research is being carried out towards the development of stressor-specific assessment systems (e.g. Sandin et al., 2004). A significant evolution seems to be a common shift towards the application and development of multimetric indices based on score systems related to reference conditions (EQRs) (Hering et al., 2004). In addition to expert knowledge, also multivariate data analysis and modelling techniques have begun to play a more crucial role in the development, evaluation and optimisation of these indices.

Chapter 2. Strengths and weaknesses of the Belgian Biotic Index method for implementing the European Water Framework Directive in Flanders, Belgium

Incorporating redrafted sections of:

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...we perceive that, relative to the animal kingdom, we should chiefly devote our attention to the invertebrate animals, because their enormous multiplicity in nature, the singular diversity of their systems of organization, and of their means of multiplication, ..., show us, much better than the higher animals, the true course of nature, and the means which she has used and which she still unceasingly employs to give existence to all the living bodies of which we have knowledge.

Jean-Baptiste de Lamarck (1803)

2. Strengths and weaknesses of the Belgian Biotic Index method for implementing the European Water Framework Directive in Flanders, Belgium

2.1 Introduction

Macroinvertebrates have a long history of application in water quality assessment, resulting in a large variety of indices, many of them being country- or region-specific (see Chapter 1). Its application in routinely river monitoring schemes by the Flemish Environment Agency (VMM) for more than a decade confirmed the reliability and robustness of the Belgian Biotic Index (BBI; De Pauw and Vanhooren, 1983) as a water quality assessment method.

When the WFD was introduced in 2000, the question emerged whether the BBI can be used in this context. The WFD imposes a number of criteria that the used assessment systems should comply with (see Chapter 1). It should therefore be examined to what extent the BBI meets these requirements, in order to decide whether it can be applied within the WFD monitoring schemes, or that a revision of the BBI or a different method should be used. In the latter case, the results of the examination of the BBI can provide valuable information on how to decide on the new or revised method.

This chapter initiates with a detailed overview of the calculation of the BBI. Then it is examined whether the BBI fulfils the necessary requirements. Subsequently, some more general problems related to taxonomic resolution in water quality assessment by means of macroinvertebrates are identified using the example of the BBI calculation with data from the VMM sampling dataset. The chapter concludes with a final recommendation with regard to the application of the BBI in the context of the WFD.

2.2 The Belgian Biotic Index

2.2.1 Background

The Belgian Biotic Index (BBI) is a standardised method to assess biological quality of watercourses based on the macroinvertebrate community. The BBI combines characteristics of the indices proposed by Woodiwiss in the UK (1964; Trent Biotic Index) and Tuffery and Verneaux in France (1968; Indice Biotique). The method is based on aquatic macroinvertebrates sampled with a standard handnet, as in the method of Woodiwiss (1964) and the calculation of the biotic index using the table as proposed by Tuffery and Verneaux (1968). Some adaptations were made concerning the sampling method and the taxonomic level of identification.

Based on ample field testing during the preceding years, the method was first described by De Pauw and Vanhooren (1983) and has been adopted as a standard method by the Belgian Institute for Normalisation (BIN, 1984). Since its first publication, the method has been extensively used to assess water quality in Belgium but also abroad (De Pauw and Hawkes, 1993). Since 1989, the Flemish Environment Agency (VMM) annually assesses around thousand sites throughout Flanders (Belgium) by means of the BBI.

Bervoets et al. (1989) proposed, along with some modifications in sample processing, to include taxa represented by only one individual in BBI calculation, but this modification was never incorporated into routinely monitoring schemes of the VMM.

2.2.2 Calculation method

Table 2.1 summarises the taxonomic levels of identification for the BBI as proposed by De Pauw and Vanhooren (1983). Only taxa of which at least two individuals are found in the sample, are taken into account. The calculation of the BBI is based on Table 2.2. When all macroinvertebrates from a sample are identified, a list is made of all taxa of which at least two individuals were encountered. For all these taxa, the tolerance class can be found in the left column of Table 2.2, next to the appropriate indicator group, except for taxa that lack a tolerance class (those taxa are counted only for taxa richness). For the lowest tolerance class encountered, the class frequency is determined, i.e. the number of taxa within this lowest tolerance class. Subsequently, the taxa richness class is determined by counting the number of taxa of which at least two individuals are found in the sample. The taxa richness class assigned can be 0-1, 2-5, 6-10, 11-15, or 16 and higher. Then, the BBI value can be found in the cross-table, in the row with the lowest tolerance class and its associated class frequency, and in the column with the correct taxa richness class (Table 2.2).

The column with indicator groups in Table 2.2 contains some modifications in comparison to the original table of De Pauw and Vanhooren (1983), which will be discussed further in this paper. BBI values correspond to water quality classes with their associated formal valuation, which are summarised in Table 2.3 (De Pauw and Vanhooren, 1983).

Table 2.1. Identification levels of macroinvertebrate taxa for calculating the BBI (De Pauw and Vanhooren, 1983).

Taxonomic group	**Determination level of systematic units**
Plathelminthes	Genus
Oligochaeta	Family
Hirudinea	Genus
Mollusca	Genus
Crustacea	Family
Plecoptera	Genus
Ephemeroptera	Genus
Trichoptera	Family
Odonata	Genus
Megaloptera	Genus
Hemiptera	Genus
Coleoptera	Family
Diptera	family, excl. Chironomidae:
	Chironomidae, group *thummi-plumosus*
	Chironomidae, group *non thummi-plumosus*
Hydracarina	presence

2.2.3 Example

Consider a macroinvertebrate sample that consists of nine different taxa, each being represented by at least two individuals, and two of these have a tolerance class of three, and no taxa have a tolerance class of one or two. By combining the column corresponding with 6-10 taxa and the row with a tolerance class of three and a class frequency of 1-2, the sample would be assigned a BBI of 5. This can be seen in Table 2.2.

Table 2.2. Calculation of the BBI (De Pauw and Vanhooren, 1983). The first column gives the tolerance class for taxa belonging to the corresponding indicator groups of the second column. The third column is the class frequency, or the number of taxa within one tolerance class. Columns 4-8 give the BBI value based on the total number of taxa, the lowest tolerance class, and, the class frequency within the lowest tolerance class. Proposed modifications of indicator groups discussed in this chapter are indicated in bold.

Tolerance class		**Class frequency**	**Number of taxa**				
#	Indicator groups		**0-1**	**2-5**	**6-10**	**11-15**	**≥ 16**
1.	Plecoptera; Heptageniidae	≥ 2	-	7	8	9	10
		1	5	6	7	8	9
2.	Cased Trichoptera	≥ 2	-	6	7	8	9
		1	5	5	6	7	8
3.	Ancylidae; ***Acroloxus*;**	> 2	-	5	6	7	8
	Ephemeroptera (excl. Heptageniidae)	1-2	3	4	5	6	7
4.	*Aphelocheirus*; Odonata; Gammaridae; Mollusca (excl. **Ancylidae**; ***Acroloxus***; Sphaeriidae; ***Corbicula***)	≥ 1	3	4	5	6	7
5.	Asellidae; Hirudinea; Sphaeriidae; Hemiptera (excl. *Aphelocheirus*)	≥ 1	2	3	4	5	-
6.	Tubificidae; Chironomidae, group *thummi-plumosus*	≥ 1	1	2	3	-	-
7.	Syrphidae-Eristalinae	≥ 1	0	1	1	-	-

Table 2.3. Water quality classes corresponding to the BBI values (De Pauw and Vanhooren, 1983).

Quality class	BBI	Colour code	Valuation
I	9 -10	Blue	Lightly polluted or unpolluted
II	7 - 8	Green	Slightly polluted
III	5 - 6	Yellow	Moderately polluted
IV	3 - 4	Orange	Heavily polluted
V	0 - 2	Red	Very heavily polluted

2.3 Evaluation of the applicability of the BBI in the context of the WFD

For the implementation of the WFD in Flanders, an assessment method for rivers and lakes based on macroinvertebrates is needed. Table 2.4 presents an inventory of the requirements that the projected method should comply with and evaluates the compliance of the BBI with these criteria. Ten WFD-related and two additional practical criteria based on an audit of the BBI method (Heylen et al., 1999) are evaluated.

Out of the ten WFD-related criteria, six are completely fulfilled by the BBI. The index provides an indication of general degradation status based on the macroinvertebrate community, taking into account taxonomic composition, sensitivity and diversity, the results can be assigned to five quality classes and it is applicable to rivers. Although it can be discussed what is exactly meant in the WFD by the parameter “taxonomic composition”, it can be assumed that the combined use of indicator groups and class frequencies is sufficient to comply with this parameter.

The two practical criteria are also fulfilled. The BBI is very practical and cost-efficient in use for routine monitoring as well as for individual studies, as long as the necessary equipment and trained staff is available. Admittedly, the application of the index has its cost, but in comparison to other macroinvertebrate assessment methods the BBI is among the cheaper methods, because the equipment is not more than standard and the identification level is limited (which saves on manpower).

Table 2.4. Inventory of WFD-related and practical requirements for a biological assessment method based on macroinvertebrates for rivers and lakes, and appraisal of BBI compliance. ++: perfectly compliant; +: sufficiently compliant; -: not compliant or compliance is debatable; --: problematic.

Requirements	**BBI compliance**
WFD-based requirements	
Indicates status of general degradation based on macroinvertebrates	++
Takes into account taxonomic composition	++
Takes into account abundance	-
Takes into account species sensitivity	++
Takes into account diversity	++
Is expressed as an EQR based on reference	-
Can be classified into quality classes	++
Type-specific	--
Applicable to rivers	++
Applicable to lakes	-
Other requirements	
Practical in use	++
Cost-effective	+

Four criteria are negatively evaluated in Table 2.4, all of them among the WFD-related requirements. Among the parameters that should be taken into account, abundance is not incorporated in BBI calculation. A minimum abundance of two individuals is required for inclusion of a taxon in the index calculation, but the abundance as such is not incorporated in the index calculation. A second problem concerns the EQR approach. The index does not explicitly take a description of reference conditions into account, although one might argue that a BBI value of ten is implicitly considered as reference conditions. In that case, the EQR could be obtained by simply dividing the BBI values by ten. Furthermore, it is not a type-specific method. All watertypes are evaluated by means of the same criteria. It is known however that the composition of the macroinvertebrate communities changes progressively from headwater stream to river (Vannote et al., 1980). Also, the BBI was intended as an assessment system for watercourses (De Pauw and Vanhooren, 1983) and hence an index for stagnant waters is still missing.

Overall, it can be concluded that significant problems arise with regard to the application of the BBI as an assessment method in the context of the WFD. On the other hand, the practical experience that was developed by the VMM through the exploitation of a large monitoring network based on the BBI is highly valuable and should be preserved. Also, the general reliability, robustness and cost-efficiency of the method are important qualities for a biological assessment index, also with regard to WFD monitoring. In conclusion, it should be considered to develop an index that incorporates these qualities into a WFD-compliant system.

2.4 Inconsistencies in biological assessment methods occurring over time

The BBI is based on a taxonomic level that is a practical trade-off between taxonomic precision on the one hand and time constraints and financial resources on the other hand. However, a difficulty that arises with identification levels other than species, is caused by possible changes in taxonomy over time, giving rise to inconsistencies in index calculation. A given genus may be split up into two or more genera or a species can be assigned to a different genus. These changes may alter the value of the biotic indices calculated based on the given taxa, respectively because the number of taxa (of a level higher than species) has changed or a taxon is replaced by another one (having a different tolerance class). This will be demonstrated by a simple example of Belgian Biotic Index calculation of a virtual sample.

Similar problems can occur due to the invasion of exotic species. Newly-occurring taxa raise discussions whether or not to include them in the existing index, which may imply defining a tolerance class for the new taxon, as used in most biotic index methods. This problem has risen for at least one exotic genus in Belgium, as will be discussed later.

2.4.1 Inconsistencies due to taxonomic modifications

De Pauw and Vannevel (1991) published keys in Dutch for identification of aquatic macroinvertebrates, for each group up to the appropriate BBI level. Since the publication of these identification keys, taxonomy of some groups of macroinvertebrates was changed, resulting in genera splitting up into more than one genus. Examples are the gastropod genera *Lymnaea*, *Stagnicola*, *Radix* and *Galba*, formerly all considered as *Lymnaea* species; the gastropod *Physella*, formerly belonging to the genus *Physa*; and *Aquarius najas* (De Geer, 1773), formerly belonging to the genus *Gerris*. As a result, two samples containing the same species and the same number of individuals for each species could result in a different index depending on whether the current state-of-the-art in taxonomy is followed for identifying the organisms or the taxonomic levels *sensu* De Pauw and Vannevel (1991) are used.

This is demonstrated with a simple example of a BBI calculation for two virtual samples (Table 2.5). The two approaches produce different BBI values in both examples. Table 2.5 (panels A and B) gives a list of species with their respective abundances and tolerance classes. Subsequently, the BBI is calculated following both approaches. In the first example (Table 2.5, panel A), identification of the sample following the keys of De Pauw and Vannevel (1991) will result in a decrease of taxa richness with two units, and a decrease of the BBI with one unit, because the genera *Aquarius* and *Radix* are assigned to other genera (*Gerris* and *Lymnaea*, respectively). In the second example (Table 2.5, panel B) the actual taxa richness decreases with one unit, but for BBI calculation it increases with a unit because two individuals are only counted when representing the same taxon since two is the minimal abundance for inclusion in BBI calculation. As a result, the BBI increases with two units in this case.

Both approaches can be justified since the original publication of the BBI (De Pauw and Vanhooren, 1983) only indicates the levels of identification (Table 2.1). Application of the BBI *sensu stricto* today would therefore imply using the current levels of identification, although only using the same taxonomic identification keys at all time would lead to stable results, i.e. a time-independent calculation of BBI values.

Table 2.5. Calculation of the BBI of two virtual samples.

Panel A. Example resulting in a decreased BBI due to taxonomic changes

Species	**Abundance**	**Tolerance class**	**Taxa according to the current state-of-the-art taxonomy (each at the applicable level)**	**Taxa according to the taxonomy as applied in De Pauw and Vannevel (1991) (each at the applicable level)**
Tubifex tubifex	100	6	Tubificidae	Tubificidae
Chironomus riparius	45	-	Chironomidae *non thummi-plumosus*	Chironomidae *non thummi-plumosus*
Erpobdella octoculata	4	-	*Erpobdella*	*Erpobdella*
Lymnaea stagnalis	5	4	*Lymnaea*	*Lymnaea*
Radix peregra	2	4	*Radix*	
Gerris lacustris	4	5	*Gerris*	*Gerris*
Aquarius najas	2	5	*Aquarius*	
Total number of taxa			7	5
Lowest tolerance class			4	4
Tolerance class frequency			2	1
BBI			**5**	**4**
Water quality class			**III (yellow)**	**IV (orange)**

Panel B. Example resulting in an increased BBI due to taxonomic changes

Species	**Abundance**	**Tolerance class**	**Taxa according to the current state-of-the-art taxonomy (each at the applicable level)**	**Taxa according to the taxonomy as applied in De Pauw and Vannevel (1991) (each at the applicable level)**
Tubifex tubifex	100	6	Tubificidae	Tubificidae
Chironomus riparius	45	-	Chironomidae *non thummi-plumosus*	Chironomidae *non thummi-plumosus*
Erpobdella octoculata	4	-	*Erpobdella*	*Erpobdella*
Lymnaea stagnalis	1	4	*(Lymnaea)*	*Lymnaea*
Radix peregra	1	4	*(Radix)*	
Gerris lacustris	2	5	*Gerris*	*Gerris*
Sialis lutaria	10	-	*Sialis*	*Sialis*
Total number of taxa			5	6
Lowest tolerance class			5	4
Tolerance class frequency			1	2
BBI			**3**	**5**
Water quality class			**IV (orange)**	**III (yellow)**

The first and second column of both panels a and b list the species and their respective abundances, the third one the tolerance classes, the fourth one the taxa according to the current state-of-the-art taxonomy (each at the applicable level), and the fifth one the taxa according to the taxonomy as applied in De Pauw and Vannevel (1991) (each at the applicable level). At the bottom of the fourth and fifth column the BBI and the respective water quality class is indicated for both approaches.

An estimation of the percentage of actual samples for which both approaches provide different results was not possible since the identifications of the VMM are only recorded at the lumped levels (e.g. *Lymnaea* including *Stagnicola*, *Radix* and *Galba*). In order to obtain a rough indication, both approaches were compared for *Anisus*, a genus that was split before the publication of the identification keys of De Pauw and Vannevel (1991) and hence all actual taxa are recorded in the VMM data set. The recorded taxa are *Anisus*, *Armiger*, *Bathyomphalus*, *Gyraulus*, *Hippeutis*, *Planorbis* and *Segmentina*. 284 samples from the VMM data set contained at least two individuals of at least two of the seven taxa. BBI was calculated for these samples when distinguishing the seven taxa and calculated again after summing the abundances of the seven taxa into one taxon, *Anisus*. For 34 samples (12,0 %), summing the taxa resulted in a BBI decrease of one unit. The other samples were not affected.

Since there is no reason to assume that taxonomic modifications will not proceed in future, this problem can only be overcome by using a fixed list of taxa at all time (or, more correctly, a semi-fixed list; see further). The establishment of a common list of taxa was already recommended by Woodiwiss in 1980. For the German saprobic index, a fixed taxon list is already in use (DIN, 1990).

2.4.2 Inconsistencies due to the introduction of exotic species

Adverse effects of invasive species on ecosystems have been discussed by several authors (e.g. Lodge, 1993; Cairns and Bidwell, 1996; Mack et al., 2000; Torchin et al., 2003). Invasion of exotic macroinvertebrate genera in Europe is increasing (e.g. Van den Brink et al., 1991; Bij de Vaate et al., 2002). These invasions cause controversy on the subject of index-based biological assessment, strongly related to the question whether or not a fixed taxa list is used. An important aspect of this controversy is the higher potential number of taxa present in monitoring samples due to these introductions, which may cause an increase in index number when using an index dependent on taxa richness. Though alpha diversity, expressed as number of taxa, may have risen, this will only be reflected in index calculation provided the new taxon is included in the list for index calculation. On the other hand, introduction of exotic species might as well cause a decrease of alpha diversity, which is masked due to a higher taxonomic identification level. For example, the invader *Dikerogammarus villosus* (Sowinsky, 1894) (Crustacea, Gammaridae) might outcompete a number of native gammarid species (e.g. Bij de Vaate et al., 2002), but this will not influence the results of the index calculation at family level of a given sample since Gammaridae are still present.

Nguyen and De Pauw (2002) reported the invasion of the Asian clams *Corbicula fluminea* (Müller, 1774) and *Corbicula fluminalis* (Müller, 1774) (Mollusca, Corbiculidae) in the

Belgian section of the river Meuse, and some of the connected canals in the early 1990s and the continuing colonisation of *Corbicula* species in Belgian watercourses. They could not establish a correlation between the clam density or proportion and the quality of the sediment. Since no tolerance class is defined for *Corbicula*, this may cause inconsistencies in BBI calculations due to a lack of consensus on how to deal with this phenomenon. The VMM encounters this genus more and more frequently in its biological samples. The question emerged whether or not this exotic genus should be included in BBI calculation, and if so, which tolerance class to use. A strict interpretation of the tolerance class as described by De Pauw and Vanhooren (1983) would lead to the inclusion of *Corbicula* in the standard list with a tolerance class of 4, being a non-sphaeriid mollusc, and thus being quite tolerant. By means of two calculation examples it is demonstrated that this may cause differences in index calculation (Table 2.6).

Table 2.6 (panels A and B) gives a list of taxa with their respective abundances. Then the BBI is calculated according to three different approaches. In the first approach, *Corbicula* is neglected, in the second it is included without tolerance class (“-”) and in the third it is included with a tolerance class of 4. Note the difference between a tolerance class “-” and the absence of a tolerance class. With a “-” tolerance the taxon is only taken into account for taxon richness, while in the absence of a tolerance class the taxon is not included at all. The first example (Table 2.6, panel A) is a sample actually taken by the VMM on 6 May 1998 at a sampling site in the Albert Canal at Genk. In this case, the inclusion of *Corbicula* leads to an increase of the BBI from 6 to 7. The VMM reported the BBI of this sampling site as 7, and consequently this site met the basic water quality conditions (BBI=7) thanks to *Corbicula*. In the second example (Table 2.6, panel B), a virtual sample, it is demonstrated that the three approaches can as well lead to three different BBI values.

Eighteen samples from the data set of the VMM contained *Corbicula* individuals. In twelve of these samples, at least two individuals were counted and hence *Corbicula* was included in the BBI calculation of these samples. For one sample (Table 2.6, panel A), the BBI was affected when *Corbicula* was discarded. The number of samples was however statistically insufficient and therefore conclusions on the probability of affecting the BBI could not yet be drawn. Nguyen and De Pauw (2002) found that including *Corbicula* species in the Biotic Sediment Index (BSI; De Pauw and Heylen, 2001), altered biological sediment quality classification in 52 % of the cases.

Table 2.6. Calculation of the BBI of a real (panel A) and a virtual (panel B) sample.

Panel A. sample taken by the VMM on 6 May 1998 at sampling site nr. VMM-820000 in the Albert Canal at Genk:

Taxa	**Abundance**	**Tolerance class without inclusion of *Corbicula***	**Tolerance class if *Corbicula* is included without tolerance class**	**Tolerance class according to De Pauw and Vanhooren (1983) *sensu stricto***
Naididae	2	-	-	-
Tubificidae	11	6	6	6
Chironomidae *non thummi-plumosus*	11	-	-	-
Helobdella	1			
Erpobdella	11	5	5	5
Gammaridae	11	4	4	4
Atyidae	11	-	-	-
Asellidae	1			
Cambaridae	2	-	-	-
Bithynia	11	4	4	4
Ancylus	2	3	3	3
Dreissena	11	4	4	4
Sphaerium	11	5	5	5
Corbicula	2		-	4
Valvata	2	4	4	4
Physa	2	4	4	4
Pisidium	2	5	5	5
Ecnomidae	11	-	-	-
Total number of taxa		15	16	16
Lowest tolerance class		3	3	3
Tolerance class frequency		1	1	1
BBI		**6**	**7**	**7**
Water quality class		**III (yellow)**	**II (green)**	**II (green)**

Panel B. virtual sample:

Taxa	**Abundance**	**Tolerance class without inclusion of *Corbicula***	**Tolerance class if *Corbicula* is included without tolerance class**	**Tolerance class according to De Pauw and Vanhooren (1983) *sensu stricto***
Tubificidae	100	6	6	6
Chironomidae *thummi-plumosus*	45	6	6	6
Asellidae	20	5	5	5
Erpobdella	4	5	5	5
Gerris	2	5	5	5
Corbicula	50		-	4
Total number of taxa		5	6	6
Lowest tolerance class		5	5	4
Tolerance class frequency		3	3	1
BBI		**3**	**4**	**5**
Water quality class		**IV (orange)**	**IV (orange)**	**III (yellow)**

The first column lists the taxa, the second one the abundances, the third one the tolerance classes if *Corbicula* is not included, the fourth one the tolerance classes if *Corbicula* is included without tolerance class (“-”), and the fifth one the tolerance classes according to De Pauw and Vanhooren (1983) *sensu stricto*.

In order to obtain a more reliable indication of the frequency of BBI alteration if an alien taxon would be discarded, the same calculation was performed for *Dreissena*, another alien bivalve that was already included in the taxa list of De Pauw and Vannevel (1991), with a tolerance class of 4. *Dreissena* is already present in Belgian waters for a longer time and consequently more data were available for comparing calculations. 421 samples from the VMM data set contained at least two *Dreissena* individuals. The BBI was calculated for all samples and recalculated after exclusion of *Dreissena*. For 100 samples (23,8 %), BBI values decreased when *Dreissena* was excluded. 98 of these (23,3 %) decreased with one BBI unit and two (0,5 %) with two units.

Biodiversity loss that is not evident at the taxonomic level of the biotic index used, is a matter of bioconservation and not of biological assessment of water quality. Therefore the new genus should be included in the taxa list since it has become part of local biodiversity. A biotic index, *in casu* the BBI, is partly based on a rapid biodiversity survey (expressed as number of taxa) as an indicator of the water quality, not of the ecosystem stability. Furthermore, species that invaded our regions at earlier times were already included in water quality assessment and are nowadays commonly accepted. Therefore, it is recommended to include *Corbicula* in the standard taxa list, despite its potential harmful effects. To obtain insight in the adverse effects of the invasion of this genus, more detailed studies - at species level - are necessary.

The VMM has already added the genus *Corbicula* to its standard list for calculating the BBI, however without assigning a specific tolerance class to it (“-”). In this way, *Corbicula* only affects the BBI through the number of taxa and not through its tolerance class, which is also the case for e.g. the taxa of Plathelminthes and most Diptera.

A number of exotic species of Ponto-Caspian origin are invading European watercourses (e.g. Bij de Vaate et al., 2002). Many of these species such as *Dikerogammarus villosus*, belong to a taxon (*in casu* Gammaridae) that is already in the list, while others will have to be included in the list as new taxa, for the same reasons as *Corbicula*. Some of these are very likely to be encountered in Flemish watercourses in the near future. Anticipating this, two Ponto-Caspian taxa should already be added to the list: Ampharetidae (Polychaeta) and Janiridae (Crustacea).

The presence of *Hypania invalida* (Grube, 1860) (Polychaeta, Ampharetidae) was recently reported in the river Meuse (Vanden Bossche et al., 2001). Although not yet encountered in

VMM samples, this may be expected in the near future, especially in the Flemish stretch of the river Meuse. Therefore, Polychaeta should be added as a new group, including one taxon, Ampharetidae, with tolerance class "-", the identification level being set at family (as for Oligochaeta). Another Ponto-Caspian invader, *Jaera istri* Veuille, 1979 (Crustacea, Janiridae) has also recently been encountered in the river Meuse (Usseglio-Polatera and Beisel, 2003), although not collected in VMM samples so far. Consequently, the list of Crustacea should be extended with the family Janiridae, with tolerance class "-".

2.4.3 List of taxa taken into consideration

There is indeed a growing need to ensure that the BBI-values remain comparable in future, which implies not altering the method itself, but rather clarifying the problems that emerge, to ensure its future application without being inconsistent with the past and current practice. Altering the method itself would imply making old and new applications incomparable; in other words, it would be a different index. The aim of this paper with regard to the BBI was to identify the problems that arose since 1991 and propose solutions to these problems.

Initially, a checklist by Vanhooren et al. (1982) was commonly used as a reference base for taxonomy of the systematic levels used in the BBI calculation. Some additional taxa were added later, e.g. due to the separation of the mollusc genus *Anisus* into *Anisus*, *Armiger*, *Bathyomphalus*, *Gyraulus*, *Hippeutis*, *Planorbis* and *Segmentina*.

Although the original description of the BBI method dates from 1983, the situation in 1991 was chosen as point of reference. At that moment, the aforementioned modifications were already established and commonly accepted. The situation in 1991 was chosen as point of reference for two reasons. The first reason is that at that time a large-scale monitoring network in Flanders was being initiated by the VMM, with the already cited modifications. The second reason is that the keys of the Pauw and Vannevel (1991) are nowadays widely used and accepted as standard reference for taxonomic identification levels with regard to the BBI.

In the previous paragraphs it has been shown that taxonomic modifications and alien invasions may both lead to biased BBI calculations. Although a change of one or two units in

BBI (on a 0-10 scale) may seem insignificant, it is not. A small change in BBI may also lead to a change in the quality class (cf. Table 2.3). This may become (legally) crucial when this quality class boundary is also a quality standard, e.g. the boundary between the ecological quality classes “good” and “moderate”, the target imposed by the WFD (EU, 2000). Moreover, a standardised assessment method should be unambiguously applicable and produce unbiased results at all times. This underpins the need for establishing a fixed taxa list. Because more exotic taxa can be expected to invade Belgian watercourses in the future, a fixed taxa list may need to be extended later with those taxa. Therefore a proposal for a fixed taxa list should be more likely called a semi-fixed list, leaving the possibility to add new taxa at a later time.

Table 2.7 is a proposal for a semi-fixed list to be used to calculate the Belgian Biotic Index in order to eliminate the discussed calculation inconsistencies. This list contains 221 taxa and can be considered as a semi-fixed list, in the sense that the taxa already in the list cannot be altered (e.g. split up or lumped), but that the list may be extended with possible future invaders when necessary. The list is based on the taxa identification *sensu* De Pauw and Vannevel (1991) with the addition of the Polychaeta family Ampharetidae, the Mollusca genus *Corbicula* and the Crustacea family Janiridae. The notation “s.l.” (*sensu lato*) was added to those taxa that comprise one or more taxa in addition to the one actually mentioned. In the case of Hydracarina the notation s.l. already appeared on the original list of De Pauw and Vannevel (1991) because Hydracarina s.l. comprises *Hydrozetes* in addition to Hydracarina s.s. (*sensu stricto*). Because the Belgian Institute for Normalisation has adopted the BBI as a standard method (BIN, 1984), it is recommended that its method description (NBN T92-402) be extended by including this new semi-fixed taxa list.

Taxa belonging to groups such as Bryozoa, Hydrozoa, Nemertea, Nematoda, Ostracoda and Porifera are not included in the new taxa list. Taxa from these groups are not frequently encountered in macroinvertebrate samples. These groups already did not appear on the original list in De Pauw and Vannevel (1991), and their addition would cause new inconsistencies between BBI calculations, since they may have been present in older samples. This problem does not arise with new, exotic taxa since they were not yet encountered in the older samples. For this reason, the mentioned groups of taxa were not added to the list.

Comparison of the tolerance classes of Table 2.7 with the indicator groups from Table 2.2 reveals some inconsistencies as well. *Acroloxus*, having a tolerance class 3, is not included in the appropriate column in Table 2.2. This is due to the fact that according to Vanhooren et al. (1982), *Acroloxus* belonged to the family Ancylidae, which is included in Table 2.2 among tolerance class 3. Since *Acroloxus* is now considered as belonging to a separate family (Acroloxidae), it should be included there as well. Furthermore, not only Sphaeriidae should be excluded from the Mollusca mentioned in tolerance class 4, but also *Corbicula*, Ancylidae and *Acroloxus*. All mentioned inconsistencies were corrected and indicated in bold in Table 2.2.

The proposal for future application of the BBI is therefore as follows:

(1) application of the taxa list from Table 2.7 with the associated tolerance classes;

(2) calculation of the index value based on all taxa of which more than one individual was found, using Table 2.2;

(3) determination of water quality class by means of Table 2.3.

Table 2.7. Proposed semi-fixed taxa list of aquatic macroinvertebrates for calculating the Belgian Biotic Index in order to avoid inconsistencies. The first column lists the taxa, the second one the associated tolerance classes. *Lymnaea* s.l. = *Lymnaea* or *Stagnicola* or *Radix* or *Galba*; *Physa* s.l. = *Physa* or *Physella*; *Pseudamnicola* s.l. = *Pseudamnicola* or *Mercuria*; Hydracarina s.l. = Hydracarina or *Hydrozetes*; *Gerris* s.l. = *Gerris* or *Aquarius*; TC = Tolerance Class.

Taxon	**TC**
Plathelmintes	
Bdellocephala	-
Crenobia	-
Dendrocoelum	-
Dugesia	-
Phagocata	-
Planaria	-
Polycelis	-
Polychaeta	
Ampharetidae	-
Oligochaeta	
Aelosomatidae	-
Branchiobdellidae	-
Enchytraeidae	-
Haplotaxidae	-
Lumbricidae	-
Lumbriculidae	-
Naididae	-
Tubificidae	6
Hirudinea	
Cystobranchus	5
Dina	5
Erpobdella	5
Glossiphonia	5
Haementeria	5
Haemopis	5
Helobdella	5
Hemiclepsis	5
Hirudo	5
Piscicola	5
Theromyzon	5
Trocheta	5
Mollusca	
Acroloxus	3
Ancylus	3
Anisus	4
Anodonta	4
Aplexa	4
Armiger	4
Bathyomphalus	4
Bithynia	4

Bythinella	4
Corbicula	-
Dreissena	4
Ferrissia	3
Gyraulus	4
Hippeutis	4
Lithoglyphus	4
Lymnaea s.l.	4
Margaritifera	4
Marstoniopsis	4
Myxas	4
Physa s.l.	4
Pisidium	5
Planorbarius	4
Planorbis	4
Potamopyrgus	4
Pseudamnicola s.l.	4
Pseudanodonta	4
Segmentina	4
Sphaerium	5
Theodoxus	4
Unio	4
Valvata	4
Viviparus	4
Acari	
Hydracarina s.l.	-
Crustacea	
Argulidae	-
Asellidae	5
Astacidae	-
Atyidae	-
Cambaridae	-
Chirocephalidae	-
Corophiidae	-
Crangonyctidae	-
Gammaridae	4
Grapsidae	-
Janiridae	-
Leptestheriidae	-
Limnadiidae	-
Mysidae	-
Palaemonidae	-
Talitridae	-
Triopsidae	-
Ephemeroptera	
Baetis	3
Brachycercus	3
Caenis	3
Centroptilum	3
Cloeon	3
Ecdyonurus	1
Epeorus	1
Ephemera	3
Ephemerella	3
Ephoron	3
Habroleptoides	3
Habrophlebia	3
Heptagenia	1
Isonychia	3
Leptophlebia	3
Metreletus	3
Oligoneuriella	3
Paraleptophlebia	3
Potamanthus	3
Procloeon	3
Rhitrogena	1
Siphlonurus	3
Odonata	
Aeshna	4
Anax	4
Brachytron	4
Calopteryx	4
Cercion	4
Ceriagrion	4
Coenagrion	4
Cordulegaster	4
Cordulia	4
Crocothemis	4
Enallagma	4
Epitheca	4
Erythromma	4
Gomphus	4
Ischnura	4
Lestes	4
Leucorrhinia	4
Libellula	4
Nehalennia	4
Onychogomphus	4
Ophiogomphus	4
Orthetrum	4
Oxygastra	4
Platycnemis	4
Pyrrhosoma	4
Somatochlora	4
Sympecma	4
Sympetrum	4
Plecoptera	
Amphinemura	1
Brachyptera	1
Capnia	1
Chloroperla	1
Dinocras	1
Isogenus	1
Isoperla	1
Leuctra	1
Marthamea	1
Nemoura	1
Nemurella	1
Perla	1
Perlodes	1
Protonemura	1
Rhabdiopteryx	1
Taeniopteryx	1
Hemiptera	
Aphelocheirus	4
Arctocorisa	5
Callicorixa	5
Corixa	5
Cymatia	5
Gerris s.l.	5

Glaenocorisa	5
Hebrus	5
Hesperocorixa	5
Hydrometra	5
llyocoris	5
Mesovelia	5
Micronecta	5
Microvelia	5
Naucoris	5
Nepa	5
Notonecta	5
Paracorixa	5
Plea	5
Ranatra	5
Sigara	5
Velia	5
Megaloptera	
Sialis	-
Coleoptera	
Dryopidae	-
Dytiscidae	-
Elminthidae	-
Gyrinidae	-
Haliplidae	-
Hydraenidae	-
Hydrophilidae	-
Hygrobiidae	-
Noteridae	-
Psephenidae	-
Scirtidae	-
Trichoptera	
Beraeidae	2
Brachycentridae	2
Ecnomidae	-
Glossosomatidae	2
Goeridae	2
Hydropsychidae	-
Hydroptilidae	2
Lepidostomatidae	2
Leptoceridae	2
Limnephilidae	2
Molannidae	2
Odontoceridae	2
Philopotamidae	-
Phryganeidae	2
Polycentropodidae	-
Psychomyidae	-
Rhyacophilidae	-
Sericostomatidae	2
Diptera	
Athericidae	-
Blephariceridae	-
Ceratopogonidae	-
Chaoboridae	-
Chironomidae *non thummi-plumosus*	-
Chironomidae *thummi-plumosus*	6
Culicidae	-
Cylindrotomidae	-
Dixidae	-
Dolichopodidae	-
Empididae	-
Ephydridae	-
Limoniidae	-
Muscidae	-
Psychodidae	-
Ptychopteridae	-
Rhagionidae	-
Scatophagidae	-
Sciomyzidae	-
Simuliidae	-
Stratiomyidae	-
Syrphidae-Eristalinae	7
Tabanidae	-
Thaumaleidae	-
Tipulidae	-

Sampling macroinvertebrates and calculating the BBI is a rigorous task and should be performed with the highest possible care and precision. Along with the calculation method, many other sources of variability exist, such as seasonality (e.g. Hughes, 1978; Furse et al., 1984, Rosillon, 1989; Linke et al., 1999; Humphrey et al., 2000; Reece et al., 2001), operator (e.g. Humphrey et al., 2000) and sampling variation (e.g. Clarke, 2000; Clarke et al., 2002). Due to all these sources of variability, it is difficult to attain a high precision for the BBI. Nevertheless, these other sources of errors are an additional incentive for using a calculation method that is as rigorous as possible.

2.5 Conclusions

Lack of consensus on how to deal with taxonomic modifications and invasions of exotic species may lead to inconsistencies in biotic index calculation. This problem could be overcome by using a semi-fixed taxa list. A semi-fixed list of macroinvertebrate taxa including a tolerance class for each taxon is proposed in order to avoid inconsistencies in the calculation procedure of the Belgian Biotic Index. This list is based on the taxa identification *sensu* De Pauw and Vannevel (1991) with the addition of the Polychaeta family Ampharetidae, the Mollusca genus *Corbicula* and the Crustacea family Janiridae. It is hoped for that this list may lead to a harmonisation of the BBI (and derived indices) calculation practice so that the BBI values can still be compared unambiguously in the future.

With regard to the implementation of the WFD, it is concluded that the Belgian Biotic Index does not comply with a number of WFD-requirements, but the valuable practical experience with this method at the VMM, and the general reliability, robustness and cost-efficiency of the BBI are interesting qualities that are also highly important for WFD monitoring. In conclusion, it should be considered to develop an index that incorporates these qualities into a WFD-compliant system. In case a new index is developed, the recommendations with regard to the use of semi-fixed taxa lists raised in this chapter evidently still hold.

Chapter 3. Development of an index for assessing biological water quality in different types of rivers and lakes in Flanders (Belgium)

Incorporating redrafted sections of:

Gabriels, W., Goethals, P.L.M. & De Pauw, N. (2007). The Multimetric Macroinvertebrate Index Flanders (MMIF) for assessing biological water quality in different types of rivers and lakes in Flanders (Belgium). Limnologica: submitted.

and

Gabriels, W., Goethals, P.L.M. & De Pauw, N. (2007). Development of a multimetric assessment system based on macroinvertebrates for rivers in Flanders (Belgium) according to the European Water Framework Directive. Verhandlungen der Internationale Vereinigung für Theoretische und Angewandte Limnologie 29(5): 2279-2282.

Rivers cannot continue to meet society's needs, or the needs of living things, if humans continue to regard river management as a purely political or engineering challenge. The flow of rivers is part of a greater flow, the planet's water cycle, which sustains not only the flow of water but the entire web of life. Ultimately, the condition, or health, of the aquatic biota is the best means of understanding and controlling humans' impact on the Earth's watercourses and on the whole water cycle.

James Karr and Ellen Chu (2000)

3. Development of an index for assessing biological water quality in different types of rivers and lakes in Flanders (Belgium)

3.1 Introduction

According to the European Water Framework Directive (WFD; EU, 2000), a good status should be achieved for all natural surface waters in the European Union by the end of 2015. A good surface water status is more specifically defined as the attainment of both a good ecological and chemical status. Assessment of the ecological status is based on a number of biological quality elements as well as hydromorphological, chemical and physico-chemical elements supporting these biological elements. To assess the status of the biological quality elements, member states must choose or develop a classification method, taking into account a set of parameters depending on the quality element (EU, 2000).

The biological elements that must be taken into account depend upon the category of surface waters. For the categories “rivers” and “lakes”, one of the relevant elements is the “benthic invertebrate fauna” (EU, 2000), commonly referred to as macroinvertebrates. For this quality element, the parameters “taxonomic composition and abundance”, “ratio of disturbance sensitive to insensitive taxa” and “diversity” should be taken into account. The quality index must be in agreement with an Ecological Quality Ratio (EQR) showing relative proportion of the index compared to the reference conditions. This EQR has a value between zero and one, where 1 corresponds to a (maximal) very good and 0 a (minimal) bad ecological status. The interval between 0 and 1 is divided into 5 classes reflecting high, good, moderate, poor and bad status, respectively (EU, 2000).

To overcome the technical shortcomings of the BBI with regard to the WFD implementation (see Chapter 2), the development of a new, type-specific multimetric index was envisaged. A multimetric index describes the state of an ecosystem by means of several individual variables (metrics). These metrics each represent a different component of ecosystem quality and are combined into one index value. Multimetric indices were first developed for fish communities (e.g. Karr, 1981; Fausch et al., 1984) and later also for other indicator groups, including macroinvertebrates (e.g. Kerans and Karr, 1994; Thorne and Williams, 1997; Böhmer et al., 2004b). An important advantage of multimetric indices is that they are flexible and can easily be adjusted by adding or removing metrics or fine-tuning the metric scoring system. In several European member states, multimetric indices have been developed or are under development for application within the WFD. For example, within the AQEM project (the development and testing of an integrated Assessment system for the ecological Quality of streams and rivers throughout Europe using benthic Macroinvertebrates), multimetric indices were developed for 28 types of streams throughout Europe (Hering et al., 2004).

In this chapter, a multimetric, type-specific index for assessment of rivers and lakes in Flanders based on macroinvertebrates is proposed. This index, called the Multimetric Macroinvertebrate Index Flanders (MMIF), combines the robustness of the BBI with the versatility of multimetric indices, allowing for an adaptation of scoring criteria for each river or lake type to reflect the relative distance to reference conditions. A preliminary concept of this index was described by Gabriels et al. (2006). This chapter provides an overview of the final version of the MMIF and its development process for all types of rivers and lakes in Flanders, using the existing experience with the BBI, scientific literature, analysis of the macroinvertebrate database of the VMM and a consultation of a panel of macroinvertebrate experts.

3.2 Multimetric macroinvertebrate index method development

3.2.1 Typology

The MMIF is a type-specific index, which means that index calculation depends on the type of river or lake a sampling site belongs to. A typology for all categories of water bodies (rivers, lakes, transitional and coastal waters) in Flanders was developed by Jochems et al. (2002) according to the WFD requirements as a framework for the development of assessment methods based on the relevant biological quality elements. For the river category, one adaptation was applied to the typology of Jochems et al. (2002): the river order according to

Strahler (1952) is presently not used as a criterion. The reason for this was that on the one hand determining Strahler order in a consistent manner proved to be unfeasible due to its dependence on map resolution, and on the other hand catchment area was considered sufficiently representative for the size of the watercourse. For lakes, no adaptations were introduced to the typology, but for the purpose of the MMIF, the ten lake types defined by Jochems et al. (2002) were clustered into four more general types.

An overview of the Flemish types of rivers and lakes as used within the MMIF, including their abbreviations and determining properties are presented in Table 3.1. The abbreviations are those used by the VMM and are based on the Dutch names of the types. An overview of the hydro-ecoregions in Flanders according to Jochems et al. (2002) is presented in Figure 3.1.

Table 3.1. Main characteristics of different types of rivers and lakes in Flanders (Belgium), as defined for application of the MMIF (based on Jochems et al., 2002).

River types	**Abbreviation**	**Hydro-ecoregion**	**Catchment area**
Small stream	Bk	Sand/sandy loam/loam	< 50 km²
Small stream Kempen	BkK	Kempen	< 50 km²
Large stream	Bg	Sand/sandy loam/loam	50-300 km²
Large stream Kempen	BgK	Kempen region	50-300 km²
Small river	Rk	Any	300-600 km²
Large river	Rg	Any	600-10000 km²
Very large river	Rzg	Any	> 10000 km²
Polder watercourse	P	Polder	Not applicable
Lake types	**Abbreviation**	**Properties**	
Alkaline	A	pH > 7.5	
Circumneutral	C	7.5 > pH > 6.5; no clay	
Acidic	Z	pH < 6.0; only sand/sandy loam/loam	
Very slightly brackish	Bzl	Na > 250 mg/L; no sand/sandy loam/loam	

3.2.2 Sampling

It is recommended to carry out macroinvertebrate samplings either during spring or autumn in order to avoid the more extreme conditions, both of hydrological regime and of temperature, to ensure a sufficiently reliable classification of water quality at a sampling site.

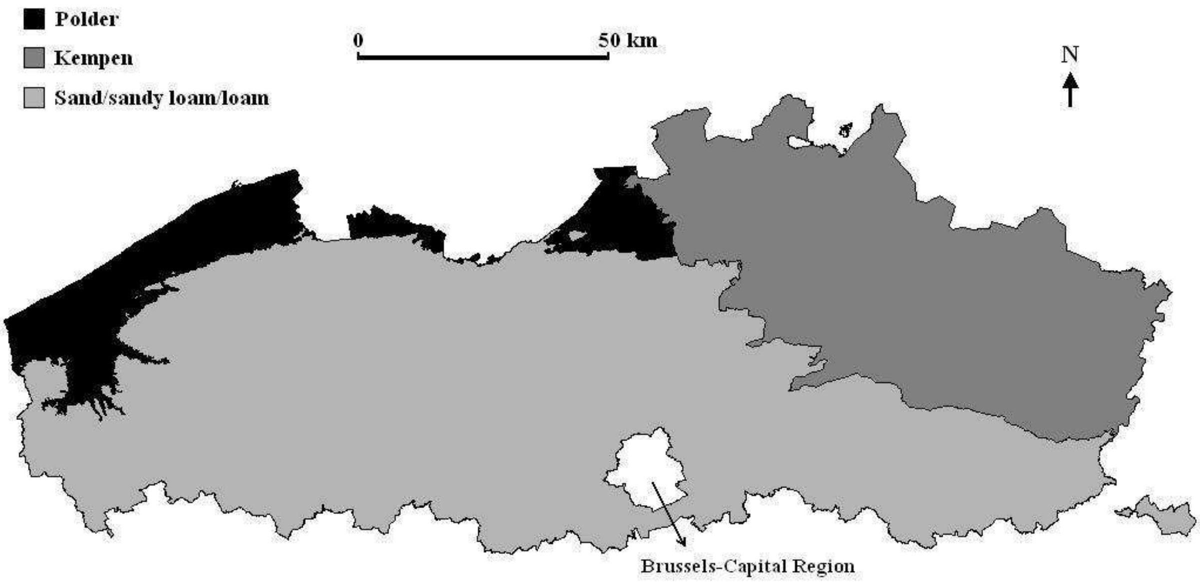

Figure 3.1. Map of Flanders (Belgium) with indication of the hydro-ecoregions (after Jochems et al., 2002) cited in Table 3.1 for distinguishing river and lake types.

Macroinvertebrates are sampled using a standard handnet, as described by De Pauw and Vanhooren (1983) and BIN (1984). This handnet consists of a metal frame of approximately 0.2 m by 0.3 m to which a conical net is attached with a mesh size of minimum 300 and maximum 500 µm. The frame is attached to a 2 m long shaft with two handles enabling it to be handled in a similar way as a scythe. With the handnet, a stretch of approximately 10-20 meters is sampled during 3 minutes for watercourses less than 2 m wide or up to 5 minutes for larger rivers. Sampling effort is proportionally distributed over all accessible aquatic habitats. This includes the bed substrate (stones, sand or mud), macrophytes (floating, submerging, emerging), immersed roots of overhanging trees, and all other natural or artificial substrates, floating or submerged in the water. Each aquatic habitat is explored, either with the handnet or manually, in order to collect the highest possible diversity of macroinvertebrates. For this purpose, kicksampling is performed by vertically positioning the handnet on the bed, and turning over bottom material located immediately upstream by foot or hand. In addition to the handnet sampling, animals are manually picked from stones, leafs or branches (De Pauw and Vanhooren, 1983). For lakes, macroinvertebrates are sampled using the same method, distributing the sampling effort among the representative parts of the water body.

If a site is too deep to be sampled with the handnet method, macroinvertebrates can alternatively be sampled using the so-called Belgian artificial substrates as described by De Pauw et al. (1986; 1994). These substrates are composed of a plastic netting filled with medium-sized (4-8 cm) pieces of brick, with a total volume of approximately 5 L. Per sampling site, three substrates are placed in the water, anchored with a rope to a fixed point located on the bank. The substrates should not be placed in open water but along the banks, in protected sites among the vegetation near the surface, in unprotected sites exposed to surface turbulence, in deeper water. After an exposure time of at least 3 weeks, the substrates are lifted from the water and transferred into a closed container (De Pauw et al., 1986).

3.2.3 Sorting, identification and counting

All collected material is thoroughly examined for presence of macroinvertebrates. Identification is carried out according to the taxonomic levels defined by De Pauw and Vanhooren (1983). This means family, genus or an intermediate level for all taxa (except for watermites, which are considered as a single taxon). The identification levels are summarised in Table 3.2.

A list of all taxa taken into consideration for the MMIF is presented in Appendix 1. This list, consisting of 223 taxa, is based on the list that was presented in Chapter 2 for calculation of the BBI in Flemish watercourses.

After identification, the total number of individuals of each taxon is recorded. If more than ten individuals of the same taxon are encountered, the total abundance can be estimated instead.

Table 3.2. Taxonomic identification levels of macroinvertebrates (De Pauw and Vanhooren, 1983; Gabriels et al., 2005).

Taxon	Identification level
Plathelminthes	genus
Polychaeta	family
Oligochaeta	family
Hirudinea	genus
Mollusca	genus
Crustacea	family
Plecoptera	genus
Ephemeroptera	genus
Trichoptera	family
Odonata	genus
Megaloptera	genus
Hemiptera	genus
Coleoptera	family
Diptera, Chironomidae	group (*thummi-plumosus* or non *thummi-plumosus*)
Diptera, other	family
Hydracarina s.l.	presence (i.e. counted as one taxon)

3.2.4 Metric selection

A preliminary index system was developed, based on an identical set of metrics with type-specific scoring criteria. To this end, a preliminary set of metrics was proposed based on existing experience with the BBI, a literature review, analysis of the VMM data, and expert judgement. This draft list of metrics, together with a set of proposed reference values per metric for each type of river and lake, and a set of tolerance scores ranging from 1-10 for each taxon, was submitted to a panel of macroinvertebrate experts. These experts are listed in Appendix 2. After receiving their remarks, a new list of metrics, reference values, and tolerance scores was established in order to integrate all assembled expert knowledge. The new values were submitted to the same panel again in order to further refine the developed index (see Gabriels et al., 2004 for details). This resulted in a final list of five metrics, a set of type-specific reference values for each metric, and a list of tolerance scores. In this way, the final set of metrics, the reference values and tolerance scores reflected all available knowledge, including existing experience with the BBI, literature review, expert knowledge, and data analysis.

The tolerance scores, ranging from 10 for very pollution sensitive to 1 for very pollution tolerant taxa, are included Appendix 1. The metrics comprised in the MMIF are Taxa Richness (TAX), Number of Ephemeroptera, Plecoptera and/or Trichoptera Taxa (EPT), Number of other (i.e. non-EPT) Sensitive Taxa (NST), the Shannon-Wiener Diversity index (SWD), and the Mean Tolerance Score (MTS). The metrics, abbreviations used, and their calculation methods are summarised in Table 3.3.

Table 3.3. Overview of metrics taken into account in the MMIF.

#	Abbreviation	Name	Calculation
1.	TAX	Taxa Richness	Total number of present taxa
2.	EPT	Number of EPT Taxa	Number of present Ephemeroptera, Plecoptera and/or Trichoptera taxa
3.	NST	Number of Sensitive Taxa	Number of present taxa with tolerance score > 5, not including Ephemeroptera, Plecoptera and Trichoptera
4.	SWD	Shannon-Wiener Diversity	$-\sum_{i=1}^{S} p_i \ln p_i$ (Shannon and Weaver, 1949) with S = taxa richness p_i = relative abundance of taxon i
5.	MTS	Mean Tolerance Score	The mean of the tolerance scores of all present taxa

3.2.5 Description of reference state

For each type of river and lake, a set of reference values for all five metrics was determined using the previously discussed procedure. An overview of the reference values for all metrics for all types of rivers and lakes is presented in Table 3.4.

Table 3.4. Overview of the expert-based reference values that were used to calculate the type-specific criteria (cf. Table 3.1 for abbreviations).

	Rivers								Lakes			
	Bk	**BkK**	**Bg**	**BgK**	**Rk**	**Rg**	**Rzg**	**P**	**A**	**C**	**Z**	**Bzl**
TAX	34	34	38	38	40	42	44	37	33	35	28	30
EPT	7	8	8	9	9	9	10	8	6	8	5	5
NST	9	9	10	10	12	12	12	10	10	10	8	9
SWD	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3	3.2
MTS	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6	6	6	6

3.2.6 Index calculation

Based on the references, a scoring system was developed for each metric consisting of threshold values needed for assigning a score ranging from zero to four (four being assigned to the metric values that were nearest to the reference value). These criteria were developed by equally dividing the interval between an expert-based target reference value and a value corresponding to a bad ecological quality into five smaller intervals. The resulting scoring criteria are summarised in Appendix 3. These five metric scores are summed and subsequently divided by 20 to obtain the final index, ranging from zero for a very poor ecological quality to one for a high biological quality.

When displaying index results for MMIF, the type of river or lake should always be specified because the calculation method is type-specific.

3.2.7 Ecological Quality Ratio and quality class boundaries

As described above, the MMIF is calculated as the sum of the 5 scores divided by 20, resulting in a final index ranging from 0 to 1. This means that the maximum MMIF value of 1 can only be obtained when all metric values are near the type-specific reference value for that metric. For this reason, the range of the MMIF index can be considered as an EQR scale.

The normative definitions of the different quality classes as prescribed by the WFD (see Table 1.2) do not unambiguously lead to EQR values for the class boundaries. The descriptions are rather vague, including qualifications such as “slight changes”, “substantially lower”, etc. It is however assumed here that each quality class should be assigned a similar bandwidth on the EQR scale. Preliminary quality class boundary values were therefore constructed by equally dividing the total range of MMIF values into five classes, as summarised in Table 3.5.

Table 3.5. Preliminary WFD quality class intervals proposed for the MMIF interval range.

MMIF	Evaluation of quality	Colour code
0.80 – 1.00	high quality	blue
0.60 – 0.75	good quality	green
0.40 – 0.55	moderate quality	yellow
0.20 – 0.35	poor quality	orange
0.00 – 0.15	bad quality	red

3.3 Example of index calculation

An example of MMIF calculation is presented in Table 3.6. A random sample was extracted from the VMM dataset. The sample was taken at the Dijle river, a watercourse belonging to the type “small river” (Rk).

The upper part of Table 3.6 lists all taxa found in the sample along with the abundance and the tolerance score. In the subsequent columns (EPT and NST), taxa belonging to one of these groups are marked “1”. The lower part of Table 3.6 lists the calculation of the metrics, scores and the overall index. In this sample, 14 taxa were found, including 1 EPT taxon and no NST taxa at all. The SWD equals 2.06 and the MTS 3.79. Scores are assigned to the metric values according to the criteria for the type Rk. Subsequently, these scores are summed, resulting in 8 and hence the MMIF equals 0.40, which corresponds to the lower limit of the quality class “moderate”, or a yellow colour code according to the WFD quality classes.

Table 3.6. Example of MMIF calculation for a sample taken in the Dijle river by the VMM.

Taxon	Abundance	TS	EPT	NST
Lumbriculidae	11	2	-	-
Naididae	2	5	-	-
Tubificidae	11	1	-	-
Erpobdella	40	3	-	-
Glossiphonia	10	4	-	-
Helobdella	8	4	-	-
Lymnaea s.l.	8	5	-	-
Hydracarina s.l.	2	5	-	-
Asellidae	42	4	-	-
Chironomidae gr non *thummi-plumosus*	6	3	-	-
Culicidae	1	3	-	-
Psychodidae	1	3	-	-
Simuliidae	1	5	-	-
Baetis	5	6	1	-
Total	148		1	0

Index calculation	Value	Score (Rk)
TAX	14	2
EPT	1	1
NST	0	0
SWD	2.06	3
MTS	3.79	2
Sum of scores		8
MMIF (Rk)		0.40
Quality class		moderate

3.4 Discussion

3.4.1 Period of sampling

Seasonal variations are important in macroinvertebrate community composition (e.g. Furse et al., 1984; Rosillon, 1989; Linke et al., 1999; Bêche et al., 2006). Consequently, the period of sampling might affect the evaluation of a sampling site. However, not all metrics necessarily differ significantly between seasons. For example, Šporka et al. (2006) found that EPT metric values did not markedly differ between seasons because in any single month a reasonably representative selection of the three EPT groups was always present.

Still, seasonality should not be neglected when developing a monitoring and/or assessment system. Often this is addressed by constraining the time frame of sampling (Linke et al., 1999). Although this strategy may result in missing information on the overall community at a site (Linke et al., 1999), it can be assumed to be sufficient for water quality assessment purposes. On the other hand, for the purpose of a large-scale monitoring network, it is advisable to choose a timeframe that is sufficiently large to visit all sampling sites in time. Sampling in summer is less reliable due to strong seasonal influences on many metrics, while sampling in winter is inappropriate for logistical reasons (e.g. Šporka et al., 2006). Constraining the sampling period to spring and autumn is therefore a pragmatic and reasonable option.

3.4.2 Taxa list

The MMIF taxa list was based on the list of 221 taxa proposed in Chapter 2 for the BBI, but it contains two additional taxa. The crustacean family *Sphaeromatidae* was added because they can be encountered in slightly brackish stagnant waters. Furthermore, the mollusc genus *Physa* s.l. was split into *Physa* s.s. and *Physella*, resulting in a list of 223 taxa for MMIF calculation. An additional adaptation was the substitution of the taxon name Grapsidae with Varunidae. The only species within the taxon Varunidae that is likely to be found in Flemish freshwaters is an Asian invasive species, the Chinese mitten crab *Eriocheir sinensis* H. Milne Edwards, 1853. This species belongs to the Varunidae, which was formerly considered as a subfamily of Grapsidae. In view of the currently accepted status of this subfamily as a separate family (see e.g. Schubart et al., 2000; Martin and Davis, 2001), the family name for *Eriocheir sinenis* should be Varunidae instead of Grapsidae (Clark, 2006). This taxonomic adaptation however does not affect the index calculation.

In Chapter 2 it was pointed out that, in order to ensure comparable calculations over time, taxonomic modifications should not be adopted in existing taxa lists. But since the MMIF is a new index, adaptations to the cited taxa list can be made as long as they are sustained in future. Both *Physa* s.s. and *Physella* can therefore be included in the proposed taxa list. Other genera that were actually split up into two or more genera (e.g. *Lymnaea*) were maintained as a single genus because their separation was not considered to improve the sensitivity of the index system. Such genera are indicated with "s.l.".

As argued in Chapter 2, the list of taxa used for the BBI calculation should be "semi-fixed", i.e. all included taxa can not be altered at a later stage, but the list should be revised on a regular basis to allow for the inclusion of newly encountered (exotic) taxa. This principle should be applied for the MMIF as well, with an appropriate tolerance score assigned to the newly included taxa.

3.4.3 Metrics used

The final selection of metrics was based on a number of considerations: they should be useful for all Flemish water body types, they should represent a variety of metric categories, they should all have been successfully used throughout Europe to assess water quality, and they should reflect a number of criteria imposed by the WFD.

An identical set of metrics was used for all types while the scoring thresholds were type-specific. This resulted in a straightforward and transparent index calculation method while typological differences were still accounted for. A similar approach can be found in Butcher et al. (2003a), who differentiated the Benthic Community Index by varying the threshold values of a number of metrics linearly with the natural logarithm of watercourse width.

Multimetric indices combine several metrics into a single evaluation. In this way, it is assumed that several aspects of ecosystem functioning, or different measures of ecological integrity, are combined into a more holistic evaluation. Also, combining several metrics is generally assumed to enhance reliability and robustness of an index, because accidental outliers of one metric can be smoothed by the other metrics. Buffagni et al. (2005) argue that multimetric systems are more suitable than single metrics to assess ecological quality and to describe biological communities. Metrics can be classified into several categories, each based on different principles of ecological quality assessment (after Resh and Jackson, 1993; Thorne and Williams, 1997; Verdonschot, 2000; De Pauw et al., 2006; see also Chapter 1): richness or diversity metrics; sensitivity metrics; similarity metrics; metrics based on functions, such as feeding groups; and metrics that combine two or more of these categories, such as biotic indices.

Considering the metrics included in the MMIF, TAX (Taxa Richness) and SWD (Shannon-Wiener Diversity index) can be classified among the richness or diversity metrics, MTS (Mean tolerance Score) among the sensitivity metrics and EPT (EPT Richness) and NST (Number of Sensitive Taxa) among both of these categories. Similarity metrics are not explicitly included, although each individual metric could alternatively be seen as the extent to which it resembles to the reference status, expressed as the expert-based reference value from Table 3.4. Functional feeding group metrics were not used (see further). This examination of metric types illustrates the similarity between the MMIF and the BBI, the index on which the MMIF development was largely based. While the BBI may be seen as a hybrid method using taxa richness on the one hand and sensitivity of the encountered taxa on the other hand, the MMIF uses both properties in a number of metrics.

Metrics of richness or diversity are widely used as indicators of ecological integrity. Diversity metrics are based on the assumption that disturbance of the water ecosystem or communities under stress leads to a reduction in diversity (De Pauw et al., 2006). Richness is widely used in water quality assessments based on macroinvertebrates because it integrates a wide range of environmental effects. For example, Carlisle and Clements (1999) demonstrated the superiority of taxa richness measures in terms of sensitivity, variability, and statistical power when it came to detecting metal-pollution effects. The majority of macroinvertebrate indices that are used for indicating general degradation of aquatic ecosystems include some measure of taxa richness. In the MMIF, included metrics based on richness and diversity are respectively TAX and SWD. The metric SWD is a diversity index that combines diversity and evenness of the encountered community.

Sensitivity metrics are also widely used in water quality assessments based on macroinvertebrates. In comparison to richness or diversity metrics, metrics based on sensitivity offer the advantage that taxon-specific information can be included. These metrics are based on the principle that different taxa respond in various ways to disturbance. This principle has been included in most assessment systems based on macroinvertebrates. The MTS is similar to the British ASPT (Average Score Per Taxon; Armitage et al., 1983), but with the identification levels and tolerance scores defined in Table 3.2 and Appendix 1, respectively.

The metrics EPT and NST can be both assigned to the category metrics of richness or diversity as well as to the category sensitivity metrics. They are a measure of taxonomic richness within the overall macroinvertebrate richness. Both groups are composed of taxa that are sensitive to various sources of disturbance.

Functional feeding group metrics were not used, because the identification level was considered insufficient to reliably assign each taxon to a functional feeding group. Moreover, Karr (1999) questions the use of functional feeding group metrics for macroinvertebrates. Assigning invertebrates into functional feeding groups is, according to this author, often guesswork. Relative abundance of predators is the only macroinvertebrate functional feeding group that seems moderately reliable (Karr, 1999). Palmer et al. (1996) could not demonstrate a pattern in functional feeding group distribution and water quality in a South African River, although individual species had a strong relationship with water quality variables. Also, Fore et al. (1996) concluded that feeding ecology metrics failed to distinguish the most and least disturbed sites.

3.4.4 Reference values

The WFD requires that biological assessment methods are based on the establishment of type-specific reference conditions (EU, 2000). Reference conditions allow for an evaluation of a site by comparing its characteristics with those that are assumed to occur in the absence of anthropogenic pressures (e.g. Bailey et al., 1998; Wallin et al., 2003; Rawer-Jost et al., 2004; Chaves et al., 2006).

Due to a high population density, associated with significant anthropogenic pressures such as intense agricultural land use, industrial activities, and urban wastewater discharges (see e.g. VMM, 2006), surface waters in reference status are extremely scarce in Flanders and most likely not present at all. As a result, a description of reference status in Flanders cannot be based on sampling data with a sufficient level of confidence. Consequently, an alternative method was needed for developing reference values. The type-specific reference values used for MMIF are based on expert judgement.

When it is not possible to develop type-specific reference conditions, based on models or data of other similar systems, or on a combination of these methods, the WFD allows member states to use expert judgement to establish such conditions (EU, 2000; Wallin et al., 2003).

3.4.5 Compliance with the WFD

The WFD requires that ecological status assessments based on benthic invertebrate fauna should consider “taxonomic composition and abundance”, “ratio of disturbance sensitive to insensitive taxa” and “diversity”. The parameter “taxonomic composition and abundance” is related to TAX, EPT, NST and SWD. Among these, abundance is used for calculation of the metric SWD. The parameter “ratio of disturbance sensitive to insensitive taxa” is related to EPT, NST and MTS. The parameter “diversity” is related to TAX, EPT, NST and SWD. It can therefore be concluded that the MMIF complies with these requirements. Other requirements include the type-specific approach, the establishment of reference conditions, and the setting of quality class boundaries. These were also included in the index system, as was previously discussed in the sections above.

The monitoring strategy for macroinvertebrates in Flanders has until now been focussed on the BBI calculation. Because sampling method and identification levels are identical for MMIF and BBI, the present monitoring approach does not need important modifications, with the exception of the distinction that now must be made between the genera *Physa* s.s. and *Physella*. An other adaptation that must be implemented to allow MMIF calculation is that abundance counts need to be recorded, while presently only abundance classes are recorded. For BBI calculation, abundances were not necessarily required, except for the distinction between taxa represented by only one, or more than one individual, respectively.

The assessment required by the WFD is a general ecological assessment. Like the BBI, the MMIF index provides a general indication of ecological degradation, based on the overall community, integrating the effects of a wide range of ecological pressures. However, it may also be useful to develop pollution specific index systems (Chessman and McEvoy, 1998).

Example are macroinvertebrate indices that were developed to detect the impact of acidification (e.g. Sandin et al., 2004) or organic pollution (e.g. Dahl and Johnson, 2004). This was however beyond the scope of this study.

3.4.6 Future developments

This new MMIF method is nowadays accepted by the VMM as a standard to report ecological status of rivers and lakes based on macroinvertebrates in a WFD context. Future developments include further adjustment of the method to incorporate new experience in sampling, identifying and calculation of the index. Also, the taxa list should be revised on a regular basis as previously discussed. Furthermore, an intercalibration with other national macroinvertebrate assessment methods in order to harmonise quality class boundaries is also required by the WFD (EU, 2000). Another question that yet needs to be tackled, is how to assess artificial and heavily modified water bodies.

Finally, it should also be stressed that the macroinvertebrates are only one of the biological quality elements that are prescribed by the WFD. Moreover, the assessment of several biological quality elements, should also be combined with the evaluation of hydromorphological and physico-chemical properties, in order to attain an integrated ecological assessment (Goethals and De Pauw, 2001).

3.5 Conclusions

A new index for biological assessment based on macroinvertebrates has been developed for different types of rivers and lakes in Flanders (Belgium) and complying as such with the European Water Framework Directive. This index, the Multimetric Macroinvertebrate Index Flanders (MMIF), is a type-specific, multimetric index. It combines the robustness of the BBI and the long-term experience in Flanders with the flexibility of multimetric indices, while taking into account the technical requirements of the WFD. According to ecological experts and comparison with BBI data, the MMIF provided reliable results when applied on existing datasets. Consequently, one can state that the MMIF is applicable for reporting about the status of surface waters in Flanders as required by the WFD. Experience in using this new index will probably lead to adjustments in the future, within a local as well as international perspective.

Chapter 4. Comparison of different metrics and indices for ecological assessment of freshwater in Flanders, Belgium

Few will doubt that humankind has created a planet-sized problem for itself. No one wished it so, but we are the first species to become a geophysical force, altering Earth's climate, a role previously reserved for tectonics, sun flares, and glacial cycles. We are also the greatest destroyer of life since the ten-kilometer-wide meteorite that landed near Yucatan and ended the Age of Reptiles sixty-five million years ago. Through overpopulation we have put ourselves in danger of running out of food and water. So a very Faustian choice is upon us: whether to accept our corrosive and risky behavior as the unavoidable price of population and economic growth, or to take stock of ourselves and search for a new environmental ethic.

Edward Osborne Wilson (1998)

4. Comparison of different metrics and indices for ecological assessment of freshwater in Flanders, Belgium

4.1 Introduction

In Chapter 3, a new index is proposed to assess freshwater macroinvertebrate communities in Flanders, Belgium, according to the European Water Framework Directive. This type-specific multimetric index consists of five metrics, including taxa richness (TAX), number of Ephemeroptera, Plecoptera and/or Trichoptera taxa (EPT), number of other sensitive taxa (NST), the Shannon-Wiener diversity index (SWD) and the mean tolerance score of all encountered taxa (MTS). This set of metrics incorporates the taxonomic composition and abundance of the macroinvertebrate community as well as sensitivity and diversity. It is generally assumed that a more reliable biological evaluation is obtained when using multiple criteria (metrics) and type-specific criteria (e.g. Kerans and Karr, 1994; Verdonschot and Moog, 2006). This chapter explores the robustness of the overall MMIF index in comparison to each individual metric. For this, the variability and comparability of the single metrics and the overall index are examined. To this end, the Flemish Environment Agency (VMM) dataset is used, comprising a large number of macroinvertebrate samplings throughout Flanders representing a wide range of ecological pressures.

4.2 Data set

4.2.1 Data source

The Flemish Environment Agency (VMM) has been collecting macroinvertebrate samples within a monitoring network covering the region of Flanders since 1989. This network was established to monitor the biological water quality by means of the BBI. All samples were collected and identified according to the BBI procedure. An extract of this dataset was obtained from the Flemish Environment Agency in March 2006, including all samples that were already processed and recorded at that moment. Overall, this dataset comprised 11 417 samples.

4.2.2 Data pre-processing

In order to improve data consistency, each sample was examined for compliance with a number of criteria. These criteria include completeness of available properties for each sample, restrictions with regard to category (only freshwater samples), sampling season and year (the samples taken in 1989 were excluded because sampling and processing were still in the optimisation process). Removal of non-compliant samples resulted in a reduced subset of the data for further analysis. Table 4.1 provides an overview of this sample selection procedure. For the majority of sampling locations, the type of water body (sensu Table 3.1) was recorded. Samples for which this information was not available, were also removed from the dataset, as can be seen in Table 4.1. The resulting dataset consisted of 7 132 samples. Among the remaining samples, the most recent sample at each location was selected in order to avoid time-dependency of samples. This resulted in a final dataset of 2 238 samples taken at different locations in freshwaters in Flanders in the period between 1990 and 2005, for all of which a minimal set of characteristics is documented.

Table 4.1 Overview of the subsequent steps of sample selection applied to enhance data consistency in the VMM dataset.

Criterion	**Rationale**	**Number of samples:**	
		Compliant	**Remaining**
None	Initial number of samples		11 417
All key properties should be documented (sampling date, location and taxa abundances)	Enable all data processing	11 411	11 411
Sample should be taken in rivers or lakes	Remove irrelevant data	10 951	10 945
Sample should be taken in 1990 or later	Homogenize data	11 137	10 682
Sample should be taken in period May-September	Reduce seasonal variation	8 499	7 975
River or lake type documented	Enable MMIF calculation	9 824	7 132
Most recent sample from each location only	Avoid pseudoreplication		2 238

For most samples, the dataset included the absolute abundance count of the encountered taxa, but in some cases the abundance class according to De Pauw and Vannevel (1991) was recorded instead. To enable the necessary calculations to be made, the classes were transformed into numeric values as follows:

class A (1): 1;
class B (2-10): 2;
class C (11-50): 11;
class D (51-100): 51;
class E (101-1000): 101;
class F (1001-10000): 1001;
class G (10001 and more): 10001.

A transformation of semi-quantitative into numeric values may introduce a bias into data analysis. However, these numeric values are only used for semi-quantitative purposes in this chapter: the abundance values are used for calculating the Shannon-Wiener diversity index, which is based on a logarithmic transformation of abundance values.

Table 4.2 provides a summary of all properties included in the final dataset.

Table 4.2. Overview of basic characteristics included in the VMM dataset of macroinvertebrate samples.

Geographical attributes
Unique sampling site identification code (VMM-code)
Date of sampling
Name of watercourse/lake
Lambert-X-coefficient
Lambert-Y-coefficient
Type of water body
Biological attributes
Abundance counts of 223 macroinvertebrate taxa (semi-quantitative or qualitative)

4.3 Materials and methods

4.3.1 Index calculation

Based on the recorded macroinvertebrate abundances and the water types, the five composing metrics and the MMIF were calculated according to the guidelines in Chapter 3. In addition, the BBI was calculated for all samples as well.

4.3.2 Distribution of the MMIF and its composing metrics

Subsequently, the distribution of the values of the MMIF index and its composing metrics are examined. Correlation coefficients are calculated and compared between each pair of two individual metrics and between each individual metric and the overall index.

4.3.3 Comparison of the MMIF and the Belgian Biotic Index

Furthermore, a regression is calculated between the MMIF index and the BBI. Based on this regression, the class boundaries of both indices are compared. In addition, the frequency distribution of both indices into the five quality classes are compared.

4.4 Results

4.4.1 Distribution of the MMIF and its composing metrics

Table 4.3 presents the minima, maxima and main percentile values of the metrics.

Table 4.4 summarises all correlation coefficients between each pair of the individual metrics and between each metric and the overall index. The significance of the observed correlations is also indicated in the table. All indicated correlations are significant at the $p<0.001$ level. It can therefore be concluded that these correlations are all very meaningful. The correlation coefficients for each combination of individual metrics vary from 0.251 to 0.699, while the five correlation coefficients between each individual metric and the overall index vary from 0.533 to 0.875. For each individual metric, the correlation coefficient with the overall index is higher than the correlation coefficient with any other individual metric, with only one exception: NST is more strongly correlated to TAX (R^2 = 0.699) than to MMIF (R^2 = 0.674).

Table 4.3. Distribution of the values of the MMIF and its composing metrics in the VMM dataset. pN = N-th percentile value.

	Min	**p05**	**p25**	**p50**	**p75**	**p95**	**Max**
TAX	1	3	7	12	17	27	45
EPT	0	0	0	0	1	4	9
NST	0	0	0	1	2	6	14
SWD	0.00	0.41	1.12	1.67	2.14	2.68	3.46
MTS	1.00	2.00	3.25	3.90	4.43	5.03	6.00
MMIF	0.00	0.05	0.20	0.35	0.50	0.70	1.00

Table 4.4. Correlation coefficients (R^2) between the different composing metrics of the MMIF index and the overall index. The asterisks denote the significance of the observed correlations: *: p<0.05; **: p<0.01; ***:p<0.001.

R^2	**TAX**	**EPT**	**NST**	**SWD**	**MTS**	**MMIF**
TAX	-	-	-	-	-	-
EPT	0.403***	-	-	-	-	-
NST	0.699***	0.251***	-	-	-	-
SWD	0.603***	0.288***	0.355***	-	-	-
MTS	0.631***	0.380***	0.446***	0.512***	-	-
MMIF	0.875***	0.533***	0.674***	0.708***	0.790***	-

4.4.2 Comparison of the MMIF and the Belgian Biotic Index

Figure 4.1 shows the regression of the MMIF versus the BBI applied to the VMM dataset. Because both indices are characterised by a limited number of possible values (11 for BBI, 21 for MMIF) and the total number of samples is high (n = 2238), the data points in Fig. 4.1 are represented by means of a bubbleplot, in which the size of the data points is proportional to the number of times this BBI-MMIF combination occurs within the dataset. It can be observed from the graph that the majority of the samples is characterised by a combination of index values near the regression line. The obtained R^2 of 0.79 indicates that both indices are fairly well correlated.

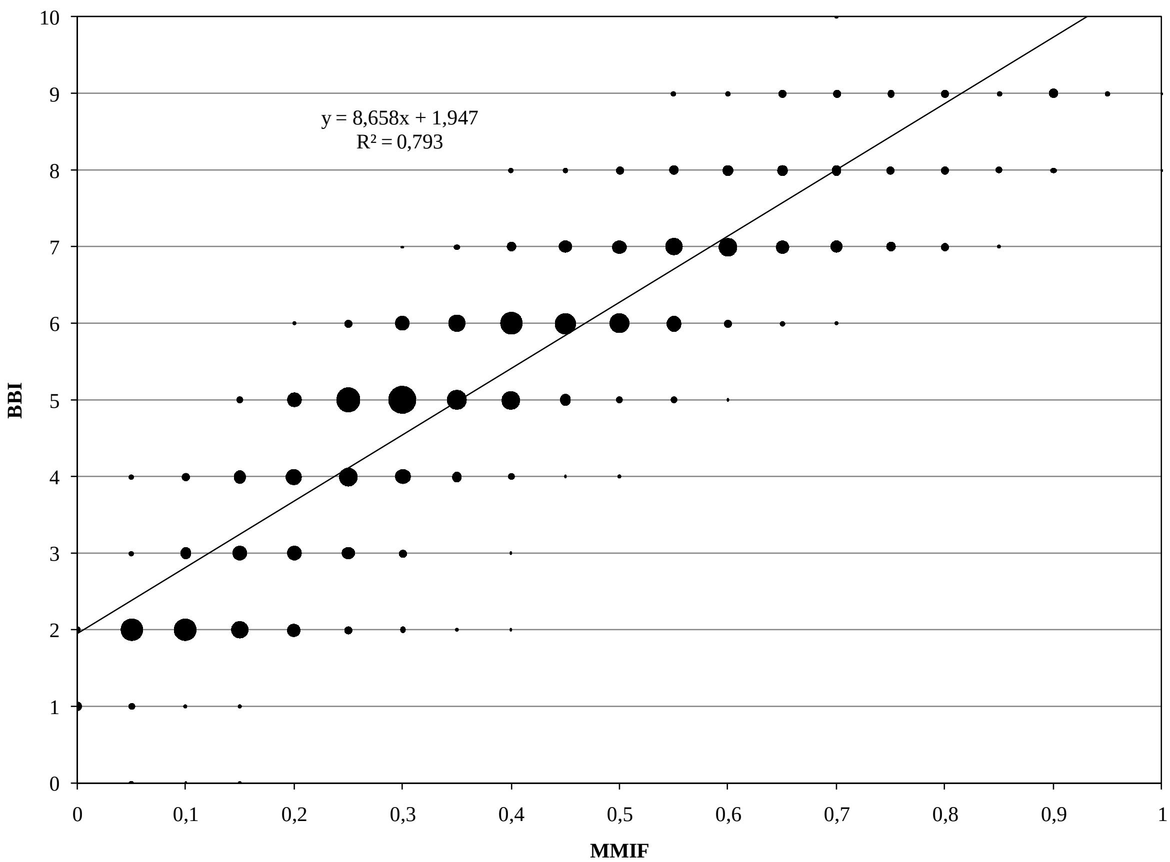

Figure 4.1. Bubbleplot with regression line of the MMIF versus the BBI applied to the VMM dataset.

Using the obtained regression equation, the proposed MMIF class boundaries are transformed into BBI values to provide an indication of the comparability of both sets of class boundaries (Table 4.5). The frequency distributions of both indices into the five quality classes were also compared. For this comparison, all samples were assigned to the quality class based on both calculated indices. For the BBI, the quality class was assigned according to De Pauw and Vanhooren (1983; see Chapter 2), for the MMIF, the preliminary quality class boundaries were applied as proposed in Table 3.5. The resulting class frequency distribution is presented in Fig. 4.2.

Table 4.5. Conversion of preliminary MMIF class boundary values into values on the BBI scale according to the regression based on the VMM dataset.

Class boundary	**MMIF**	**Converted MMIF (on BBI-scale)**
High-good	0.80	8.87
Good-moderate	0.60	7.14
Moderate-poor	0.40	5.41
Poor-bad	0.20	3.67

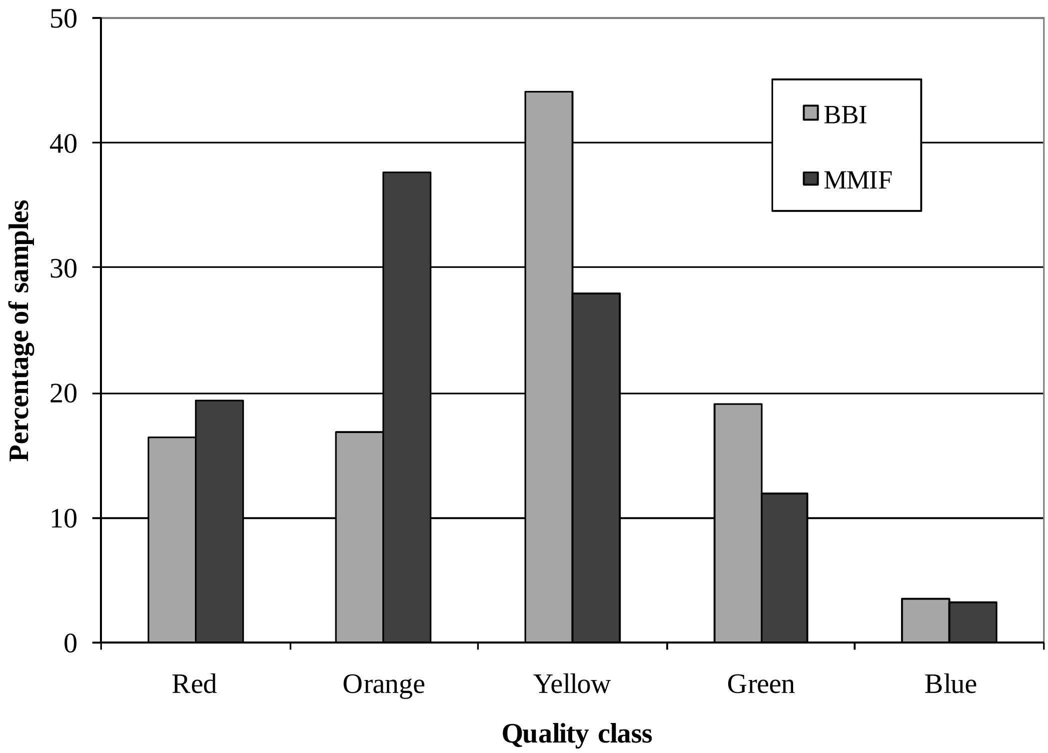

Figure 4.2. Frequency distribution of the five quality classes within the BBI and the MMIF index based on the VMM dataset.

4.5 Discussion

It can be concluded from Table 4.4 that between each pair of two individual metrics a weak to moderate relationship exists, but the differences are sufficiently important to justify the combined use of all metrics. A moderate to good correlation exists between the overall index and each individual metric.

The observation that the overall index is generally more strongly correlated to the individual metrics than metrics are correlated to other metrics, highlights the robustness, and hence the practicability, of the overall index as a result of combining several individual metrics. This supports the general assumption that the combined use of several criteria leads to a more robust biological evaluation in comparison to the use of single criteria (e.g. Kerans and Karr, 1994; Verdonschot and Moog, 2006).

The conversion of preliminary MMIF class boundary values into BBI values enables a comparison of the proposed lower boundary for good quality (the WFD target for 2015) with the existing Flemish legislation. Based on the regression obtained in Fig. 4.1, an MMIF of 0.60 corresponds to a BBI of approximately 7.14 (Table 4.5). The current Flemish BBI standard as defined in VLAREM legislation (Flemish Government, 1995) is 7, or the lower bound of the green quality class for BBI. It can be concluded that both standards are fairly well comparable, the MMIF being slightly more severe. However, the conversion of class boundaries based on a linear regression should be interpreted with care, considering the distribution of points around the regression trendline (Fig 4.1).

Although both indices exhibit a roughly similar class frequency distribution (Fig. 4.2), the distribution of the MMIF values can be characterised as slightly smoother, while the BBI shows a relatively strong peak in the yellow class, which contains more than twice the percentage of any other BBI class. The total percentage of samples that are assigned to one of the two highest classes (green and blue) is 15.1 % for MMIF and 22.7 % for BBI. This confirms the previous observation that both standards are relatively well comparable, although MMIF is somewhat more severe. This can also be observed when the modes of the frequency distributions are compared: for MMIF this is the orange class, while for BBI it is one class higher, the yellow class.

Both the comparison of the BBI standard with the proposed MMIF standard using the regression from Fig. 4.1 as the comparison of the class frequency distributions lead to a similar conclusion. The BBI and MMIF boundaries are roughly comparable, but the MMIF is slightly more severe. However, it should be stressed that the BBI quality class boundaries (De Pauw and Vanhooren, 1983) were not developed in order to reflect WFD normative definitions (EU, 2000), preceding the publication of the latter by almost two decades.

It should be emphasised that a quality standard based on a biological quality index, in the case of the WFD the good-moderate class boundary for a given biological quality element, is an arbitrarily chosen value. However, if different member states would use class boundaries that are not harmonized, there would be a risk that some member states would be disadvantaged and others privileged, which would potentially lead to a severe disequilibrium in economic resources allocation (Buffagni et al., 2007). It is therefore necessary that class boundaries are harmonised across member states (Buffagni et al., 2007).

4.6 Conclusion

The data set explored in this chapter clearly suggests that the MMIF is an useful and robust method to evaluate biological quality based on macroinvertebrate communities. The different composing metrics offer complementary, but not contradictory, evaluation schemes which are all integrated in the overall assessment index. Moreover, the MMIF seems to be sufficiently comparable to the BBI. In other words, the quality assessment provided by the MMIF is at the same time compliant with the WFD (see Chapter 3) and compatible with the BBI assessment approach.

Concerning the range of qualities covered, the BBI evaluation seems to be less severe when the distribution of obtained index values over the interval range is compared for both indices. Although the whole range of quality classes is covered, the highest MMIF values seem to be less frequently attained. This is in agreement with the EQR approach, which should provide an evaluation where the highest possible index value is considered to reflect near-pristine conditions (Wallin et al., 2003). When the quality class boundaries are compared, the proposed lower bound for MMIF seems to be quite similar to the currently applicable Flemish standard for the BBI, although slightly more severe.

Chapter 5. Harmonisation of MMIF quality class boundaries based on the European intercalibration exercise for river macroinvertebrates

Incorporating redrafted sections of:

CB-GIG (2006). Central-Baltic GIG. In: Van den Bund, W. (ed.). WFD intercalibration Technical report Intercalibration technical report - Part 1 Rivers, Section 2 Benthic Macroinvertebrates. JRC, Ispra, Italy.

and

Gabriels, W. (2007). Proposal for adjusting the Flemish class boundaries according to the intercalibration exercise for river macroinvertebrates. Flemish Environment Agency, Aalst, Belgium. 20 p. + appendices.

Because we are the cause of our environmental problems, we are the ones in control of them, and we can choose or not choose to stop causing them and start solving them. The future is up for grabs. We don't need new technologies to solve our problems; while new technologies can make some contribution, for the most part we just need the political will to apply solutions already available. Of course, that's a big "just". But many societies did find the necessary political will in the past. Our modern societies have already found the will to solve some of our problems, and to achieve partial solutions to others.

Jared Diamond (2005)

5. Harmonisation of MMIF quality class boundaries within the European intercalibration exercise for river macroinvertebrates

5.1 Introduction

The European Water Framework Directive (EU, 2000) requires that member states develop an assessment system for all types of rivers, lakes, transitional and coastal waters, based on a number of biological elements, including macroinvertebrates (see paragraph 1.2.3). For assessing macroinvertebrate communities in rivers and lakes in accordance with the WFD, Flanders has developed and proposed a multimetric index called the MMIF (see Chapter 3).

In order to establish boundary values for the water quality classes that are comparable along member states, an intercalibration exercise was envisaged by the WFD for each quality element and for each category of water body. These intercalibration exercises were further split up into a number of geographical regions. A number of these regional intercalibration exercises is currently completed. For river macroinvertebrates, the relevant regional intercalibration exercise for Flanders was coordinated by the so-called Central-Baltic Geographical Intercalibration Group (CB-GIG, 2006).

This chapter describes the Flemish contribution to this intercalibration exercise with the MMIF, using sampling data from the VMM. The purpose of this contribution is to compare the Flemish boundary values to those of the other participating countries and regions and, if necessary, to adjust them in order to be included in the intercalibration decision of the European Commission (EU, 2007). Inclusion in this intercalibration decision implies the approval of the MMIF method as the Flemish method to assess macroinvertebrates in rivers and lakes in the context of the WFD. A similar exercise for lake macroinvertebrates has not yet been initiated at present (EU, 2007).

The exercises aim to harmonise the boundaries between the classes high and good and between the classes good and moderate. For the remaining class boundaries, between moderate and poor and between poor and bad, the WFD does not require an intercalibration exercise and for this reason they are not dealt with during these exercises.

Because environmental policy is a regional issue in Belgium, the implementation of the WFD and hence the development of biological assessment methods is tackled separately by each region (Gérard et al., 2006). For this reason, Belgian regions contribute individually to these intercalibration exercises, in contrast to all other member states, who participate as a whole (CB-GIG, 2006). For brevity, this distinction between countries and regions will not be made in this chapter. The use of "countries", "member states", and related terminology should therefore be interpreted as "countries and/or regions" throughout this chapter. Similarly, "national" should be interpreted as "national and/or regional".

5.2 Materials and methods

5.2.1 General approach

The CB-GIG intercalibration exercise for river macroinvertebrates essentially consists of a regression of each participating member states' method against a common, generally applicable index. This regression is carried out separately by each member state using a national dataset. Based on the obtained regression equation, each country converts its national boundary values into values on the scale of the common index for subsequent comparison. The mean value of the converted national boundaries is calculated and national boundaries that are within a certain range of this mean value are considered to be comparable (CB-GIG, 2006).

Figure 5.1 presents the different steps involved in the intercalibration procedure (CB-GIG, 2006). First, the participating member states collate a national set of river macroinvertebrate samples, all collected at locations that can be assigned to a type from a predefined set of common intercalibration river types, described by the European Working Group "Ecological Status" (ECOSTAT, 2004). In order to be included, these datasets have to meet the WFD (EU, 2000) requirements as well as a number of quality requirements set by the CB-GIG (2006). From these data, reference sites are identified and screened against the pressure criteria agreed by the CB-GIG (2006). From the compliant reference sites, metric reference values are derived, which are used to calculate the Intercalibration Common Metric index (ICMi; Buffagni et al., 2005, 2007; CB-GIG, 2006), an index that is generally applicable to all national datasets. A regression between the national EQR and the ICMi for each country enables the conversion of national class boundaries into values on the common ICMi scale, in order to be compared (CB-GIG, 2006).

5.2.2 Data included

Each participating member state provides a national data set of macroinvertebrate samples. In order to be included in the calculation of the harmonisation band (see further), these datasets have to meet the following acceptance criteria (CB-GIG, 2006):

-for each sample, abundance values of all macroinvertebrate families relevant for ICMi must be available, as well as the result of the national index, and allocation of the samples to one of the common types (see further);

-the national assessment protocol must be WFD-compliant (including sampling method, EQR calculation, and the method that was used to set quality class boundaries);

-a minimum of 2 different reference sites must be included;

-a minimum of 6 reference samples must be included;

-a minimum number of samples per quality class according to national classification must be included:

-high: minimum 4 samples (including reference samples);

-good: minimum 4 samples (including reference samples);

-moderate: minimum 4 samples;

-poor and bad: no required minimum number.

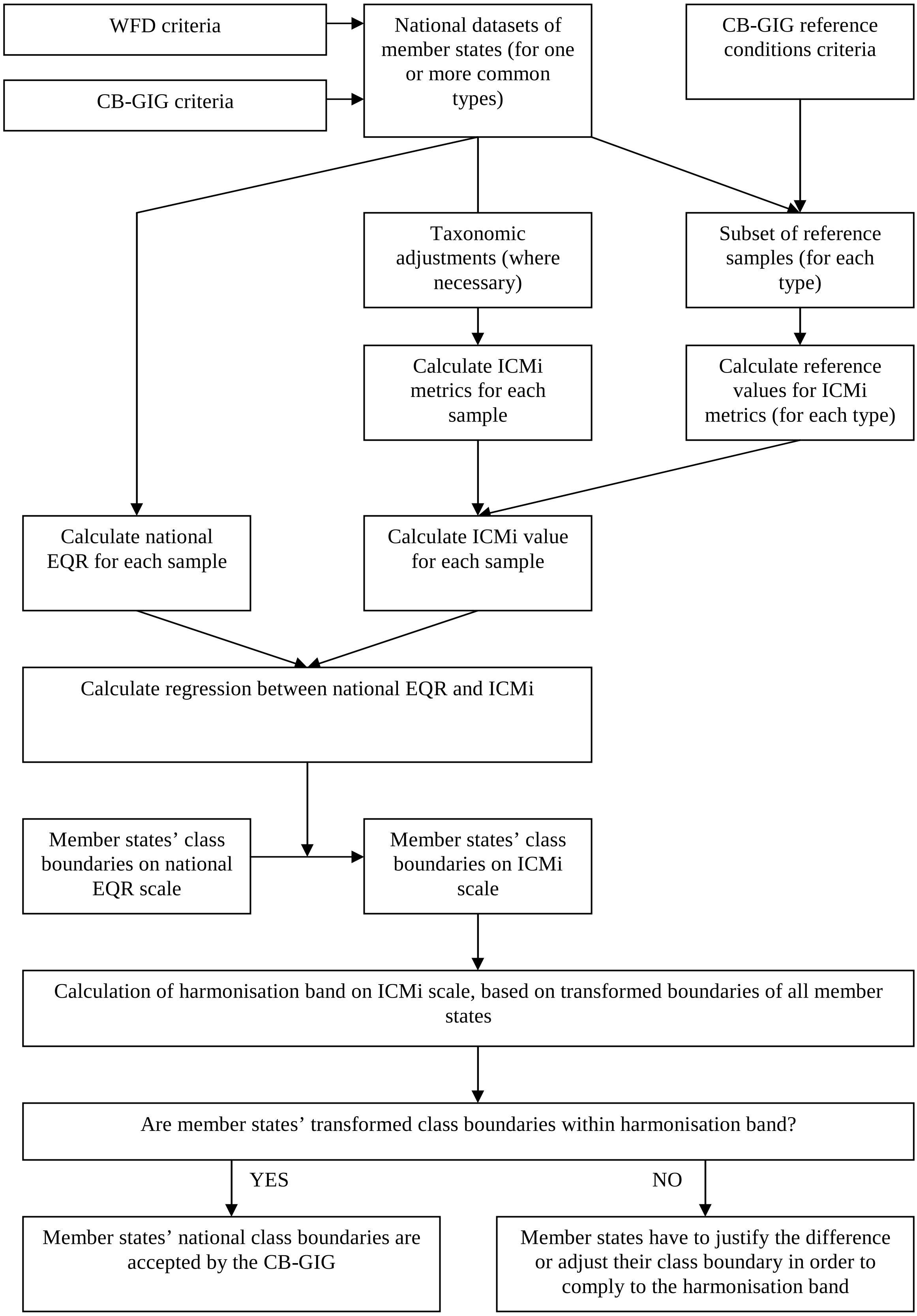

Figure 5.1. Graphical representation of the different steps involved in the intercalibration exercise for river macroinvertebrates conducted by the CB-GIG.

5.2.3 European river types

Each sampling location must be assigned to one of the six European river types that were defined by ECOSTAT (2004) for the purpose of this exercise (Table 5.1).

Table 5.1. Overview of common river types defined for the Central-Baltic intercalibration exercise (ECOSTAT, 2004).

Type	River characterisation	Catchment area (of stretch)	Altitude and geomorphology	Alkalinity (meq/L)
R-C1	Small lowland siliceous sand	10-100 km²	Lowland, dominated by sandy substrate (small particle size), 3-8 m width (bankfull size)	> 0,4
R-C2	Small lowland siliceous - rock	10-100 km²	Lowland, rock material, 3-8 m width (bankfull size)	< 0,4
R-C3	Small mid-altitude siliceous	10-100 km²	mid-altitude, rock (granite) - gravel substrate, 2-10 m width (bankfull size)	< 0,4
R-C4	Medium lowland mixed	100-1000 km²	Lowland, sandy to gravel substrate, 8-25 m width (bankfull size)	> 0,4
R-C5*	Large lowland mixed	1000-10000 km²	Lowland, barbel zone*, variation in velocity, max. altitude in catchment: 800 m, >25 m width (bankfull size)	> 0,4
R-C6	Small, lowland, calcareous	10-300 km²	Lowland, gravel substrate (limestone), 3-10 m width (bankfull size)	> 2

*mixed cyprinids, with some salmonids

5.2.4 Reference conditions

Reference sites are initially selected by the member states using the “REFCOND” guidance (“Guidance on establishing reference conditions and ecological status class boundaries for inland surface waters”; Wallin et al., 2003). However, these reference sites must also comply with a number of additional criteria agreed by CB-GIG (2006). These criteria are listed in Appendix 4. Member states were asked to screen selected reference sites against agreed catchment landuse limits, and when proposed reference sites were over agreed limits, a validation with physico-chemical parameter thresholds at the site scale was necessary or strongly recommended. Member states were also asked to complete a check list to indicate which of the CB-GIG defined reference criteria were used for the screening exercise and the sources of information that were available to the member state for this process. Fig. 5.2 shows the subsequent steps in the screening procedure for potential reference sites (CB-GIG, 2006).

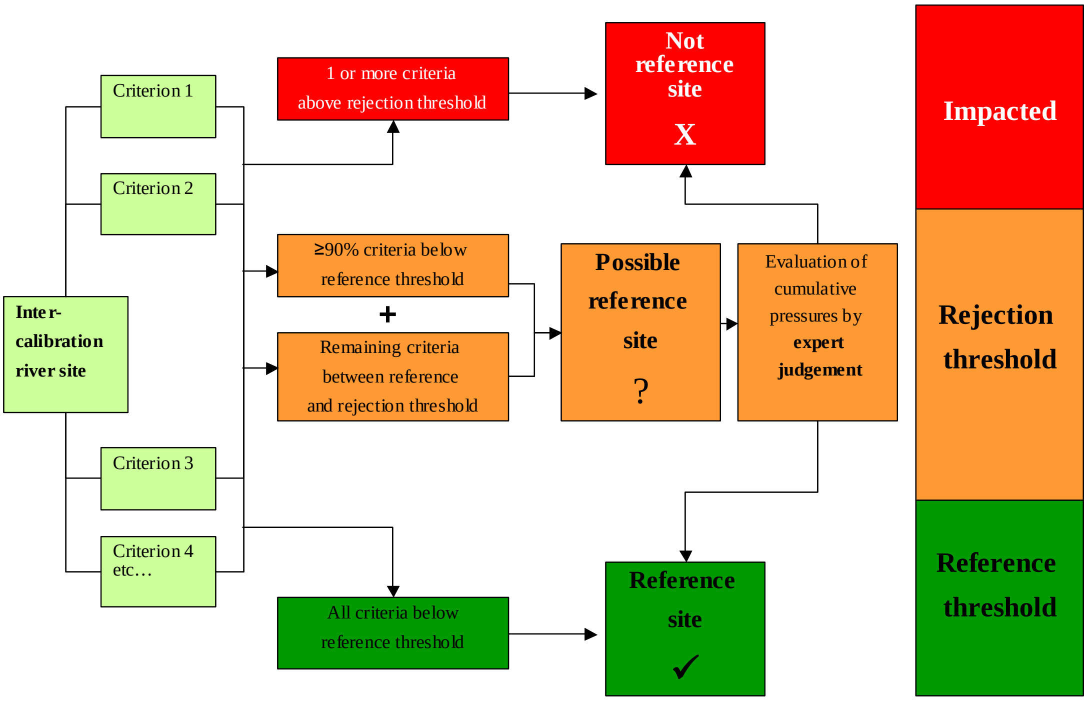

Figure 5.2. Flow chart of the screening procedure for potential reference sites within the intercalibration exercise conducted by the CB-GIG for river macroinvertebrates (reproduced from CB-GIG, 2006).

5.2.5 Calculation of the Intercalibration Common Metric index (ICMi)

First, the taxonomic identification level of the national datasets is transformed to family level, where necessary, in order to enable the calculation of the ICMi (Buffagni et al., 2005, 2007). Based on these data, the six metrics of the ICMi are calculated. An overview of the metrics used in the ICMi is presented in Table 5.2, along with an overview of calculation methods and relative weights (Buffagni et al., 2005, 2007; CB-GIG, 2006).

The ICMi metrics are normalised to a 0-1 scale by dividing them by the type-specific reference conditions. The ICMi equals the weighted sum of the normalised metrics. The weights were assigned to the metric according to the metric type to which they belong in order to give an equal total weighting to each of the three conceptual groups (Table 5.2; CB-GIG, 2006).

Table 5.2. Metrics included in the ICMi with their calculation method and respective weights (Buffagni et al., 2005, 2007; CB-GIG, 2006).

Metric type	Metric	Calculation	Reference	Weight
Tolerance	ASPT	Mean of tolerance scores of all encountered taxa	Armitage et al., 1983	0.333
Abundance/ habitat	Log Sel EPTD	Log_{10} (1 + sum of Heptageniidae, Ephemeridae, Leptophlebiidae, Brachycentridae, Goeridae, Polycentropodidae, Limnephilidae, Odontoceridae, Dolichopodidae, Stratyomidae, Dixidae, Empididae, Athericidae and Nemouridae)	Buffagni et al., 2005	0.266
	1-GOLD	1 - (relative abundance of Gastropoda, Oligochaeta and Diptera)	Pinto et al., 2004	0.067
Richness and diversity	Number of taxa	Total number of encountered families	e.g. Ofenböck et al., 2004	0.167
	EPT taxa	Total number of encountered families belonging to Ephemeroptera, Plecoptera and/or Trichoptera	e.g. Ofenböck et al., 2004; Böhmer et al., 2004a	0.083
	Shannon-Wiener diversity index	$-\sum_{i=1}^{S} p_i \ln p_i$ with S = taxa richness p_i = relative abundance of taxon *i*	Shannon and Weaver, 1949	0.083

5.2.6 Harmonisation of class boundaries

In order to be included in the harmonisation band, the procedure adopted by a member state to derive national boundary values must be WFD-compliant. For each member state, the EQRs from the national assessment method are correlated with the corresponding EQRs from the ICMi. A regression is performed and the regression equation and R^2 value are calculated. By means of the regression equation, national boundary values are transformed into ICMi values. This comparison is carried out only for the class boundary between “high” and “good”, and for the class boundary between “good” and “moderate”. The R^2 of the correlation between the national EQR and the ICMi should be 0.5 or higher (CB-GIG, 2006).

A GIG mean boundary value on the ICMi scale is calculated from those national boundary values that meet all of the acceptance criteria. This mean boundary value is used to establish the so-called 5% harmonisation band, which is agreed by the CB-GIG as the acceptable range for national boundaries. This harmonisation band is defined as the mean boundary value plus or minus 0.05 on the ICMi scale.

For each member state, the converted class boundary value can be compared to the harmonisation band. The following outcomes are possible (CB-GIG, 2006):

- The member state ICMi boundary lies within the harmonisation band: in this case, no action is required; the boundary is considered comparable.
- The member state ICMi-EQR boundary does not lie within the harmonisation band. Here, there are two possibilities:
    - If the member state agrees to harmonise, for ICMi boundary values that occur below the harmonisation band, the national boundary should be adjusted in such a way that the equivalent ICMi boundary falls within the harmonisation band. For ICMi boundary values that occur above the harmonisation band, the member state is not obliged to adjust its national boundaries.
    - If the member state does not agree to harmonise, it should justify why it does not accept the GIG mean boundary. In this case, the member state has to provide a scientific explanation that the boundary differs from the GIG boundary and the harmonisation band.

5.3 Results

5.3.1 Harmonisation band

Reference sites

Table 5.3 lists the number of reference sites identified for each member state per common intercalibration river type.

Table 5.3. Number of reference "sites" (data points) selected by member states according to the CB-GIG defined criteria for each common intercalibration river type (CB-GIG, 2006).

	R-C1	R-C2	R-C3	R-C4	R-C5	R-C6	Total
Austria			25				25
Flanders	0			0			0
Wallonia			20				20
Czech Republic			7				7
Germany	6		20	6			32
Denmark	5			9		7	21
Estonia				6	5	5	16
Spain		16	35	10	10	6	77
France	23	50	107	21		42	243
Ireland		116		13	9	66	204
Italy	32						32
Lithuania				6		10	16
Luxembourg			39	18		26	83
The Netherlands	0			0			0
Poland	8						8
Sweden		14					14
United Kingdom	25	16		30		19	90
Total	**99**	**212**	**253**	**119**	**24**	**181**	**888**

Countries included in harmonisation band calculation

Data from nine countries were included in the calculation of the high-good and good-moderate boundaries (Austria, Wallonia, Germany, Spain, France, UK, Italy, Luxembourg, Ireland). These member states occur to the left of the red line in Figures 5.3 and 5.4. Data from eight countries (Lithuania, the Netherlands, Poland, Sweden, Flanders, the Czech Republic, Denmark, Estonia) were not included in the calculation of the GIG boundaries (member states to the right of red line in Figures 5.2 and 5.3) for reasons including (CB-GIG, 2006):

- the national boundaries were not agreed yet;
- no fully developed national assessment method was available yet;
- reference values were chosen using an approach that differs to that outlined by the CB-GIG (described in paragraph 5.2.4). Flanders and the Netherlands have used alternative approaches for calculating reference values;
- data quality issues (insufficient number of sites or reference sites; poor regression between the national system and the ICMi).

High-good boundary

The results of the "all-types combined" comparison for the high-good boundary (Figure 5.3) indicate that (CB-GIG, 2006):

- twelve countries fall within the harmonisation band: Austria, Wallonia, Germany, Spain, UK, Italy, Luxembourg, Ireland, Lithuania, the Netherlands, the Czech Republic and Estonia;
- four countries lie below the high-good harmonisation band: France, Poland, Sweden and Flanders;
- one country lies above the high-good harmonisation band: Denmark.

Good-moderate boundary

The results of the "all types combined" comparison for the good-moderate boundary (Figure 5.4) indicate that (CB-GIG, 2006):

- nine countries fall within the harmonisation band: Austria, Wallonia, France, UK, Italy, Luxembourg, the Netherlands, Denmark, Poland;
- five countries lie above the good-moderate harmonisation band: Germany, Spain, Ireland, Lithuania and the Czech Republic;
- three countries lie below the good-moderate harmonisation band: Sweden, Flanders, Estonia.

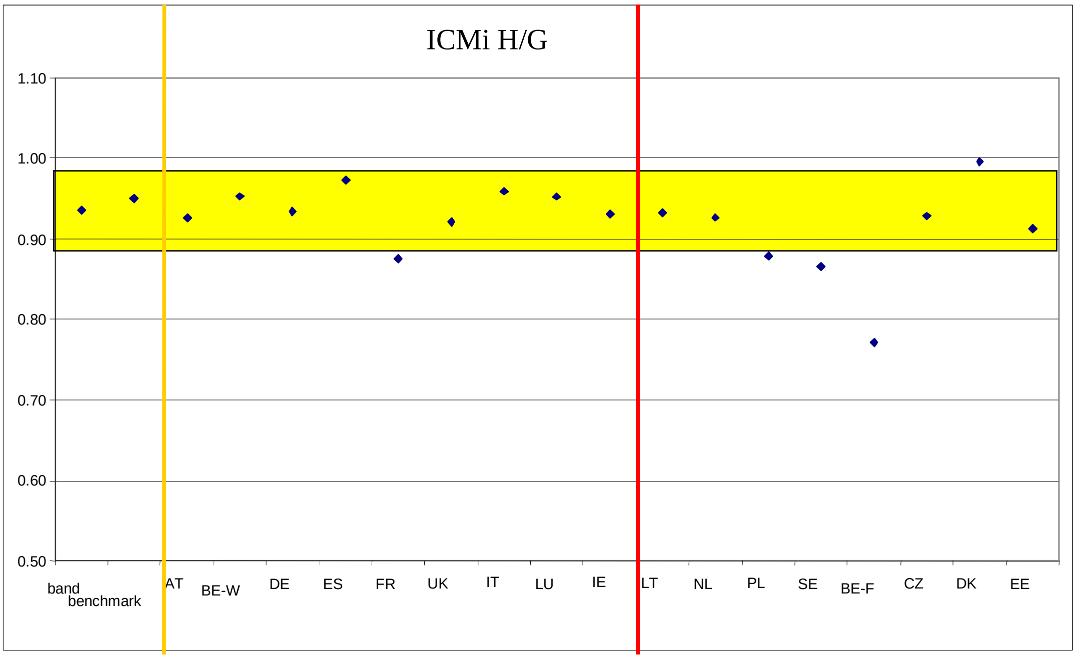

Figure 5.3. Results of the "all types combined" comparison with the ICMi values of member states for the high-good boundary. "Band" represents the GIG high-good boundary value. Only the values of the member states to the left of the red line contributed to the calculation of the GIG boundary. The yellow "harmonisation band" represents a plus or minus 0.05 interval on the ICMi scale around the GIG boundary value (CB-GIG, 2006).

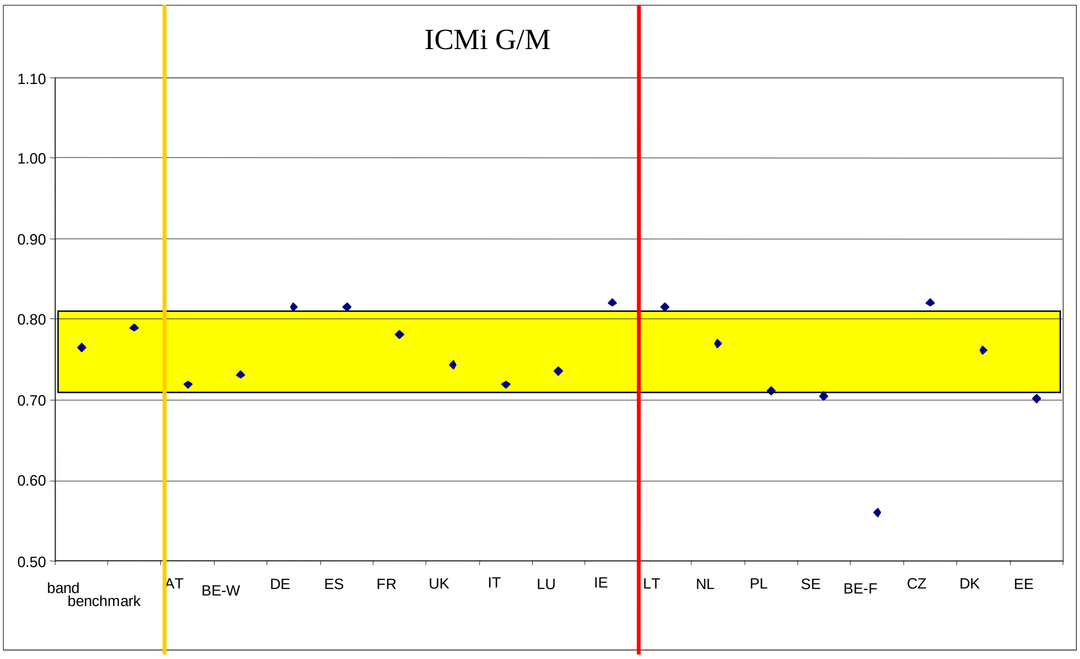

Figure 5.4. Results of the "all types combined" comparison with the ICMi values of member states for the good-moderate boundary. "Band" represents the GIG good-moderate boundary value. Only the values of the member states to the left of the red line contributed to the calculation of the GIG boundary. The yellow "harmonisation band" represents a plus or minus 0.05 interval on the ICMi scale around the GIG boundary value (CB-GIG, 2006).

5.3.2 Flemish results within the CB-GIG intercalibration exercise

The initial contribution of Flanders to the CB-GIG exercise for river macroinvertebrates (Gabriels, 2006) is included in the results discussed in paragraph 5.3.1. Two major problems emerged with regard to this contribution. First, the derivation of reference values for the ICMi metrics was not possible using field data because no reference sites are present in Flanders. An alternative method was used to derive reference values, based on regressions between the reference values for MMIF metrics (based on expert judgment) and ICMi values (Gabriels, 2006). However, the CB-GIG steering group asked all member states lacking actual reference sites to demonstrate that their reference values are comparable to those obtained using the CB-GIG method. This was a compulsory requirement to be included as a member state in the EU intercalibration decision. A second problem concerned the boundary values. When converted into ICMi values, both the high-good and the good-moderate boundary of the Flemish method were clearly below the harmonisation band. As explained in Figure 5.1, all member states that have boundaries below the harmonisation band were asked to either adjust their boundary values or provide a scientifically sound justification why their boundary values are below the band.

For this reason, an alternative and more transparent way of deriving reference conditions was used in order to have reference values comparable to those of the other member states (Gabriels, 2007). This method was loosely based on the one used by the Netherlands (Van Riel, 2006). For the boundary values, it was examined what boundary values could be used that comply with the harmonisation band (Gabriels, 2007).

5.3.3 Alternative proposal for the Flemish contribution

This paragraph deals with the aforementioned update of the Flemish contribution (Gabriels, 2007). Not only the differences with the initial contribution (Gabriels, 2006; paragraphs 5.3.1-5.3.2) but a complete overview including all relevant information on the final contribution will be presented here.

River types

The common types included in the intercalibration exercise relevant for Flanders were R-C1 and R-C4.

Within R-C1, two Flemish types are included:

- Bk: “Small Stream” (see Chapter 3)
- BkK: “Small Stream in the Kempen region” (see Chapter 3)

Within R-C4, three Flemish types are included:

- Bg: “Large Stream” (see Chapter 3)
- BgK: “Large Stream in the Kempen region” (see Chapter 3)
- Rk: “Small River” (see Chapter 3)

The remaining three Flemish river types, Rg (“Large river”), Rzg (“Very large river”) and P (“Polder watercourse”) can not be assigned to one of the common types and are therefore not considered in this exercise.

Data set

All data were obtained from the Flemish Environment Agency (VMM) monitoring database. Initially, a representative number of samples was extracted at random from the database for both R-C1 and R-C4.

To the initial R-C4 dataset, 19 sampling sites were added, more specifically data for the regional river type BgK (“large stream in the Kempen region”), because this type was slightly under-represented in the R-C4 dataset. Furthermore, 26 sites from the R-C4 dataset and 15 sites from the R-C1 dataset were characterised by an ASPT score below 2. This would result in negative values when subtracting 2 prior to normalisation (see further, paragraph 5.3.3). In order to comply with all data requirements, it was decided to exclude these sites from the dataset.

The dataset submitted for R-C1 comprised 193 samples, including 90 samples from the regional type Bk (“Small Stream”) and 103 from the regional type BkK (“Small Stream in the Kempen Region”). For R-C4, the dataset comprised 185 samples, including 130 samples from the regional type Bg (“Large Stream”), 24 from the regional type BgK (“Large Stream in the Kempen Region”) and 31 from the regional type Rk (“Small River”). Table 5.4 summarises the numbers of samples within each quality class according to the MMIF for both European river types.

As can be seen in Table 5.4, both datasets comply with the criterion of comprising at least four samples within the quality classes high, good and moderate.

Table 5.4. Number of samples within each quality class according to the regional classification method, for the dataset submitted by Flanders for the intercalibration exercise for common river types R-C1 and R-C4.

National classification	R-C1	R-C4	Total
High	11	4	15
Good	27	14	41
Moderate	56	39	95
Poor	84	104	188
Bad	15	24	39
Total	193	185	378

Taxonomic adjustments

For MMIF calculation, no further adjustments were necessary. For calculation of the ICMi, some adjustments were made. Data were all available at MMIF identification levels. In order to enable ICMi calculation, taxa were merged to family level where necessary. Watermites were removed from the dataset because these are not included in ICMi calculation.

Reference conditions

In order to enable a regression between MMIF and ICMi, both indices must be expressed as an EQR scale as previously explained.

The MMIF is considered as an EQR scale, and consequently, the maximum value (1.00) can be used as a “surrogate” for the reference value (Gabriels, 2006). Note that the metric target reference values (and hence the scoring systems) are type-specific, so the reference value of 1.00 can be considered as type-specific although the range of MMIF values is identical for all national types.

Because actual reference sites do not exist in Flanders (Gabriels, 2006), reference data could not be extracted from field data. However, since not the actual biological data (taxa lists) of reference sites are required for calculation of the ICMi, but only the corresponding metric values, this problem can be overcome by defining reference values for each ICMi metric.

In earlier contributions to the CB-GIG intercalibration exercise, Flanders has tested a variety of methods for deriving these reference values (see Gabriels, 2006). At present, none of these methods were approved yet by the CB-GIG steering group. An alternative method that was proposed by the Netherlands, using the 75^{th} percentile of high class sites (van Riel, 2006), was recently approved by the steering group. It was therefore investigated whether this method could also be applied for the Flemish data.

Due to the limited number of sites in the dataset that are in high status class according to the MMIF, the 75^{th} percentile values were taken of the sites in high status class for the types R-C1 and R-C4 combined (15 in total; see Table 3). The obtained values are presented in Table 5.5.

When evaluating these resulting values for ICMi metrics, a number of considerations should be taken into account:

- First, actual reference sites are absent in Flanders. Actual data on taxonomic composition are therefore not available. This alternative method however takes into account the 75^{th} percentile of the metric values of sites that are in highest class, and this for each individual metric separately. The resulting values therefore represent values that are the best available for each metric while avoiding possible outliers;

- The predefined reference values for similar MMIF metrics can be compared to the proposed ICMi reference values. This is however difficult due to differences in identification levels between MMIF and ICMi;
- The proposed ICMi reference values can be compared to the reference values of other member states. However, differences of biological data among member states due to biogeographical particularities and dissimilarities in sampling methods and laboratory processing, may lead to erroneous conclusions on such comparisons. In particular, the typical lowland conditions in Flanders, predominantly characterised by relatively low current velocities, should be kept in mind. This limits the geographical comparison, suggesting the Netherlands as member state with the most similar natural conditions to Flanders.

Table 5.5. Proposed reference values for ICMi metrics for the Flemish river types, based on the 75th percentile of high status samples calculated for each metric separately.

ICMi metric	Proposed reference value for Flanders
ASPT	4.798
Shannon-Wiener Index	2.886
EPT families	6.000
Total family richness	27.000
Portuguese GOLD-index	0.703
Sel EPTD	0.943

Table 5.6 shows the 75th percentile values of the ICMi metrics in high class samples in the data from the Netherlands for R-C1 and R-C4 (from the CIRCA website for CB-GIG river macroinvertebrate intercalibration - July 2006). These were used as reference values by the Netherlands (and reportedly accepted by the CB-GIG steering group).

In Figure 5.5, the proposed reference values for Flanders are graphically compared to the reference values reported by those neighbouring countries that contributed to the intercalibration for R-C1 and/or R-C4. These countries are the Netherlands, Germany, France and the UK. For the UK, these values are not the ones actually used for ICMi calculation because type-specific reference values were used instead.

Table 5.6. 75th percentile values of ICMi metrics in high class samples in the data from the Netherlands within R-C1 and R-C4.

European type	**R-C1**	**R-C4**
ASPT	5.357	5.227
Shannon-Wiener Index	1.488	2.144
EPT families	5	5
Total family richness	19	26
Portuguese GOLD-index	0.986	0.887
Sel EPTD	1.699	1.342

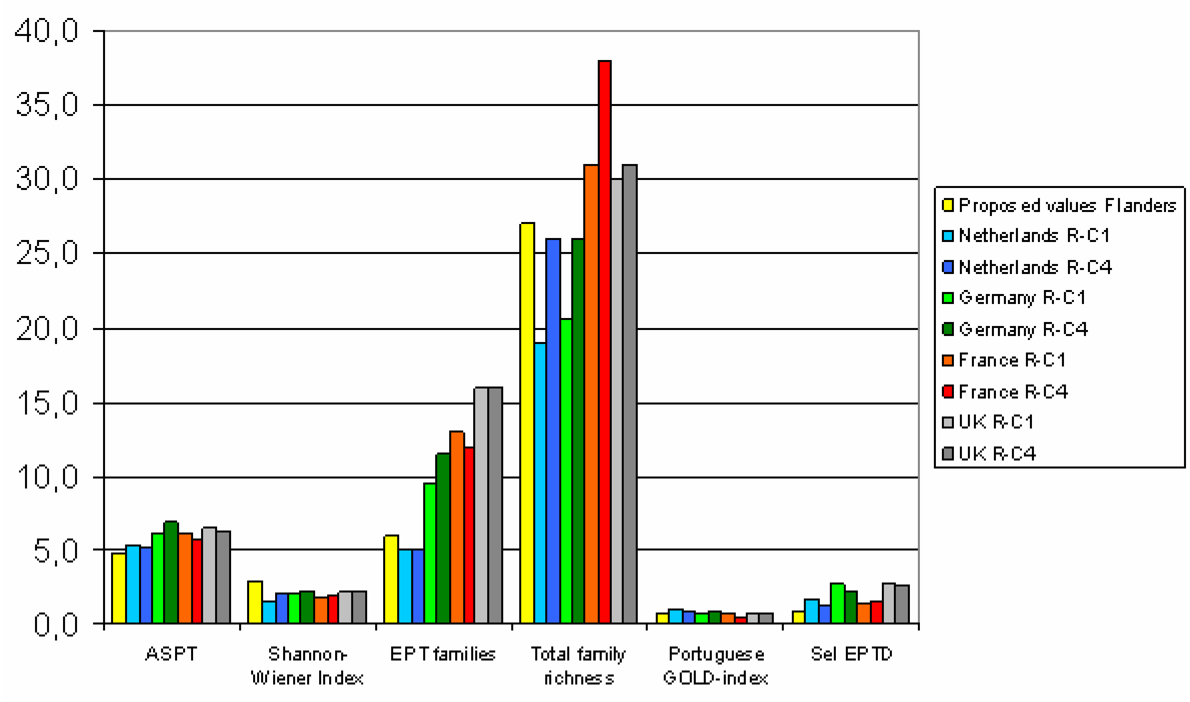

Figure 5.5. Graphical comparison of metric reference values of Flanders (combined) and all neighbouring countries that contributed to the R-C1 or R-C4 intercalibration.

It can be observed in Figure 5.5 that, although differences exist between metric values for all countries, the Flemish values are not systematically higher or lower in comparison to the other countries. The metric values that are relatively low for Flanders are ASPT, EPT families and Sel EPTD. These metrics are known to be highly associated with EPT taxa richness. In the Netherlands, EPT taxa are naturally rare (Van Riel, 2006). Among the cited countries, the Netherlands are most similar to Flanders from an ecological and a biogeographical point of view. The Flemish and Dutch lowland conditions result in predominantly low current velocities. It is therefore reasonable to assume that the situation in Flanders is very similar to the Netherlands for these three metrics. Among all countries compared in Figure 5.5, the Dutch data are most similar to the Flemish data for these metrics. For the other three metrics, no dramatic differences exist between Flanders and the other countries, except for the Shannon-Wiener index, which is somewhat higher than for the other countries. Furthermore, differences among metric reference values (in some cases higher, in some cases lower) between countries become less important when all metrics are combined into a single index. With all these considerations in mind, the proposed values can be considered as representative for reference status for the included national types.

Comparison of Flemish results with the harmonisation band

The ASPT values were subtracted by 2 prior to normalisation. Normalisation was carried out by dividing the metric values by the reference value for this metric. Subsequently, the ICMi (Buffagni et al., 2005, 2007; CB-GIG, 2006) was calculated and compared to the MMIF, for R-C1 and R-C4 separately and also for both types combined.

Regression lines between MMIF and ICMi were calculated. Regressions were calculated for R-C1 (Figure 5.6), R-C4 (Figure 5.7) and for all data combined (Figure 5.8). The R^2 values were in all three cases above 0.60: 0.685 for R-C1, 0.829 for R-C4 and 0.738 for the combined regression.

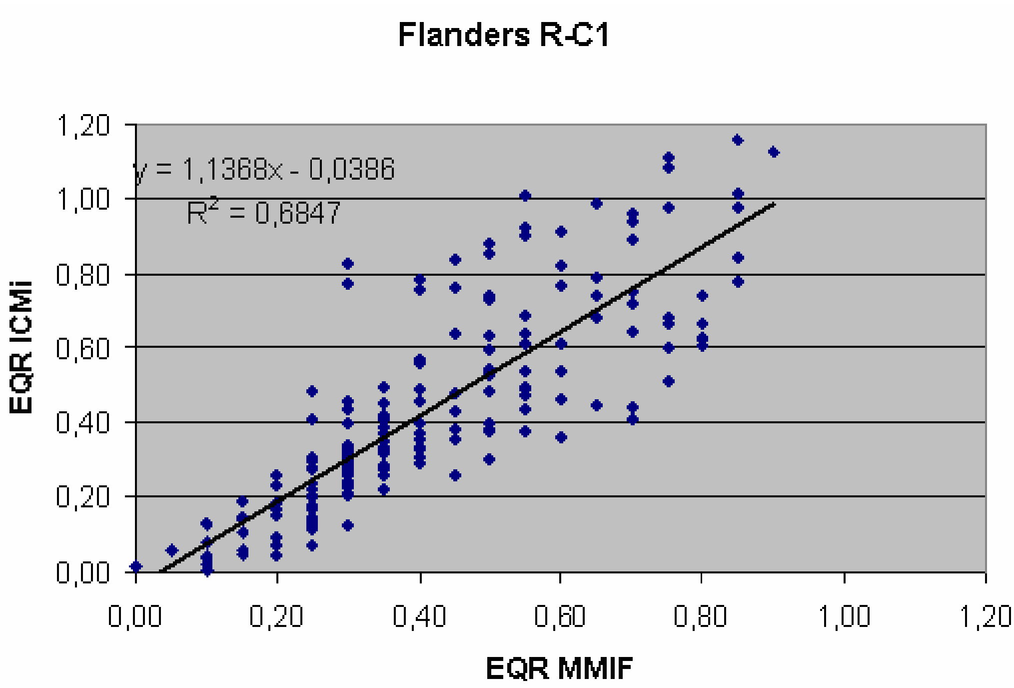

Figure 5.6. Scatterplot with regression line for ICMi versus MMIF applied to Flemish data for R-C1.

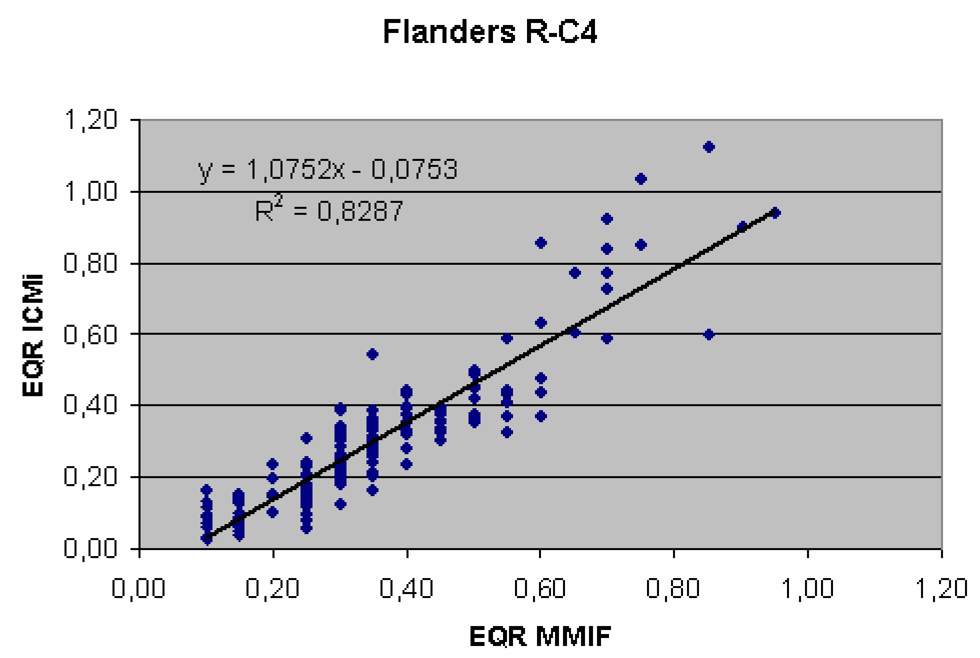

Figure 5.7. Scatterplot with regression line for ICMi versus MMIF applied to Flemish data for R-C4.

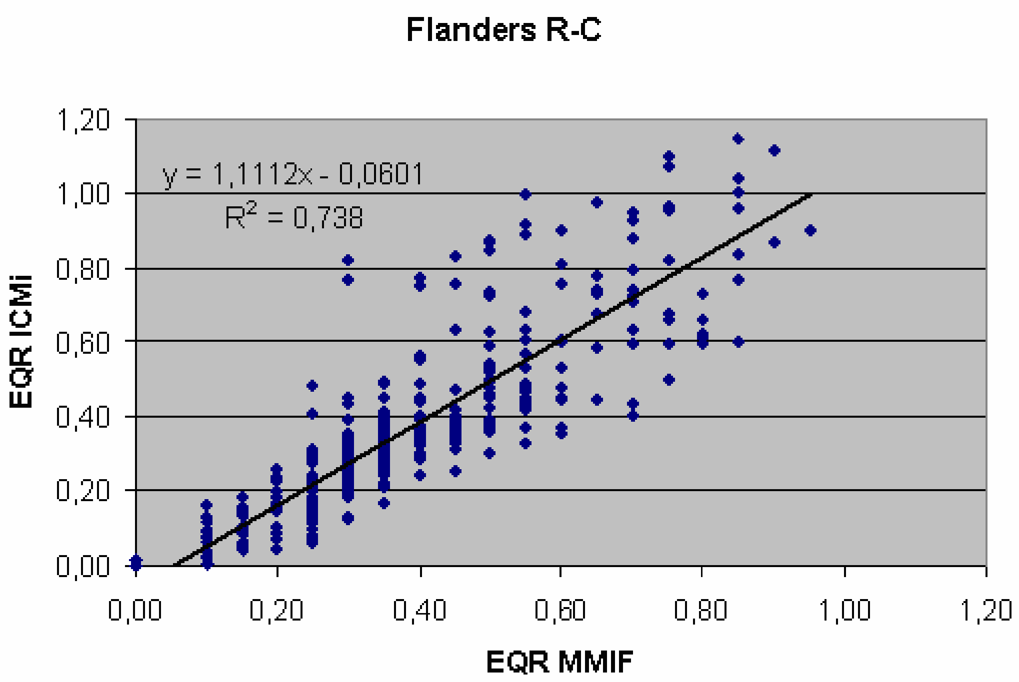

Figure 5.8. Scatterplot with regression line for ICMi versus MMIF applied to Flemish data for R-C1 and R-C4 combined.

Transformation of boundary values

The MMIF boundary values were transformed into ICMi values for the overall intercalibration (R-C1 and R-C4 combined) using the obtained regression equation. The results of these transformations are presented in Table 5.7.

Table 5.7. Transformation of MMIF class boundary values into ICMi values for the combined intercalibration (including R-C1 and R-C4) using the Flemish reference values for ICMi metrics.

Boundary	**MMIF**	**EQR ICMi**
high-good	0.80	0.829
good-moderate	0.60	0.607
moderate-poor	0.40	0.384
poor-bad	0.20	0.162

Compliance with harmonisation band

The national boundaries, when transformed into ICMi values, should be included in the harmonisation band in order to be considered comparable to those of the other member states. For the high-good boundary, this interval is [0.88 - 0.98] and for the good-moderate boundary the interval is [0.71 - 0.81] (see paragraph 5.3.1). The originally proposed class boundaries for the MMIF index are 0.60 for good-moderate and 0.80 for high-good. These class boundaries, when transformed into ICMi values (Table 5.7), are below the harmonisation band for both high-good and good-moderate.

Distribution of MMIF values

Due to its calculation method using metric scores, the range of values of the MMIF is not continuous but takes a number of discrete values with an interval step of 0.05. For instance, the MMIF can be equal to 0.75 or 0.80 but not 0.77. This consideration should be taken into account when establishing the boundary values.

Proposal to adjust MMIF class boundaries

In order to obtain MMIF boundary values that are within the harmonisation bands, an alternative proposal is calculated. When the MMIF values of 0.70 and 0.90 are transformed according to the previously obtained regression calculation, the obtained values were 0.718 and 0.940, respectively. Both values fall within the respective harmonisation bands (Fig. 5.9).

In conclusion, if the Flemish boundary values for both the high-good boundary and the good-moderate boundary are raised with 0.10, these class boundaries result in ICMi values that are included in the CB-GIG harmonisation band. It is therefore proposed to set the boundary values for MMIF to 0.70 for the good-moderate boundary and to 0.90 for the high-good boundary.

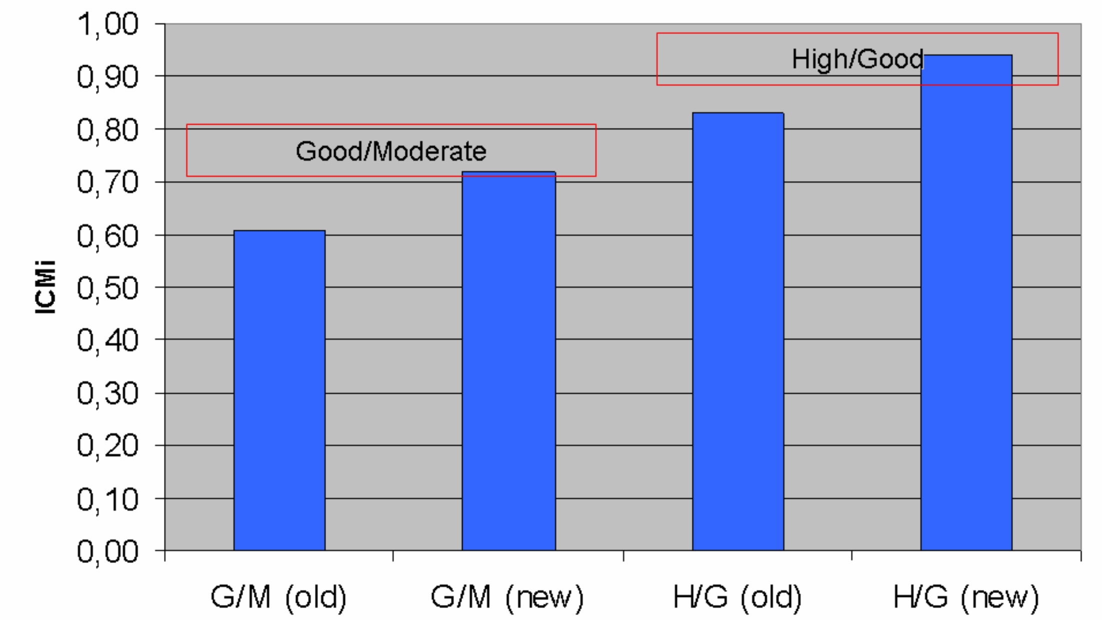

Figure 5.9. Comparison of old and new MMIF boundary values, transformed into ICMi values, with the CB-GIG harmonisation band for the good-moderate boundary and the high-good boundary.

5.3.4 Decision of the CB-GIG steering group on the alternative proposal for Flanders

The alternative proposal was later communicated to the CB-GIG steering group (Gabriels, 2007). This proposed adjustment of the MMIF class boundaries for rivers was accepted by the steering group of the CB-GIG for river macroinvertebrates on 5 April 2007 (CB-GIG, 2007). A document summarising the alternative approach and proposal for adjusting the Flemish boundaries (Gabriels, 2007) was subsequently added to the technical report (previously referred to as CB-GIG, 2006) as an appendix. After discussion in the ECOSTAT working group, the adjusted values were adopted in the draft version of intercalibration decision of the European Commission (EU, 2007), of which the official publication is foreseen for the end of 2007.

5.4 Discussion

5.4.1 CB-GIG river macroinvertebrates intercalibration approach

Several aspects of the procedure adopted by the CB-GIG to compare class boundaries can be criticised. The geographical range is rather broad and includes countries as remote as Estonia, Sweden, Ireland and (Northern) Spain. Also, the common types described are not entirely compatible with national types. The screening criteria for reference sites can give rise to various interpretations and the collection of physical-chemical and geographical data for this purpose is not standardised. The macroinvertebrate samples are collected according to national standards and sampling effort may therefore be quite different. The ICMi index is based on family-level identifications, which is not the case for several countries (including Flanders). Many national indices do not correlate very well with all ICMi metrics, and some national indices have a poor correlation with the overall ICMi index as well. The harmonisation band spans 10 percent of the total EQR range. Each step in the intercalibration process therefore adds to the uncertainty of the results, although the precise uncertainty of each step is difficult to quantify. The reliability of the outcome of the exercise is therefore doubtful. However, most of these uncertainties were difficult, if not impossible to avoid in view of the available data and timeframe. Given the scope and range of the exercise, the outcome of the exercise was therefore nearly the best possibly achievable (see also Biggs, 2006).

Differences in sampling methods are not necessarily problematic because it is assumed that reference site screening criteria are applied by all member states in the same way. From this philosophy, because the metrics are standardised based on metric values observed at reference sites, differences in sampling effort do not affect the standardised metrics.

The choice of the identification level of family was a pragmatic one. The intercalibration exercise is based on existing data sets and consequently, the identification level needed to be applicable with all data sets. Family level was the minimum level applied by all member states.

To motivate the choice of the 5% harmonisation band, CB-GIG (2006) cite the report of the Alpine GIG (2006), who provide a review of factors that add variation to the results when boundary values on the ICMi scale are obtained by transforming national boundary values using the regression formula. These factors include data limitations, natural variability and the simplification principles of the ICMi (Alpine GIG, 2006). Due to these sources of variation, the status assessment is more significant in the middle of a status class compared to the transitional zone of the neighbouring status classes. This “insecure” zone of assessment is assumed to be a quarter of the status class width (Alpine GIG, 2006). When all five quality classes are assumed to take an equal part of the entire EQR scale, a quarter of a quality class equals 5 % of the EQR scale or 0.05 units of the ICMi-EQR scale. A more detailed estimation of accuracy and precision is still lacking in most countries (CB-GIG, 2006).

5.4.2 Flemish contribution to the CB-GIG river macroinvertebrates intercalibration

The MMIF correlates well with the ICMi. The difficulties were the lack of reference values and the initial boundary values being below the harmonisation band.

The problem of the reference conditions was overcome by using for each metric the 75th percentile of values from sites that are in high class according to the MMIF and subsequent evaluation and comparison to reference values of other member states. It is concluded that these values are an acceptable alternative to be used for calculating ICMi metrics. The proposed reference values can therefore be considered as suitable for comparing and harmonising class boundaries. After calculation of the regression between MMIF and ICMi, the originally proposed boundary values were below the harmonisation band. When adjusting the MMIF boundary values to 0.70 for good-moderate and to 0.90 for high-good, the regression results in ICMi values that are included within the CB-GIG harmonisation band. In other words, these proposed boundary values should be considered as comparable to the other member states’ boundary values according to the CB-GIG criteria (Fig. 5.1).

5.4.3 Class boundaries not covered by the intercalibration exercise

The intercalibration exercise coordinated by the JRC only covers the boundaries high-good and good-moderate. This complies with the WFD, which specifies that the intercalibration exercises should be carried out for those boundaries (EU, 2000).

An increase in the boundaries for high-good and good-moderate, while the moderate-poor and the poor-bad boundary remain unchanged, results in a relatively wide moderate class. It is therefore proposed to adjust the moderate-poor and the poor-bad boundary in parallel to the high-good and the good-moderate boundary, i.e. an increase of 0.10 MMIF units. In this way, an equal bandwidth is maintained for the three "central" classes, i.e. good, moderate and poor. The resulting set of quality classes and their associated colour codes are presented in Table 5.8. The differences between the original boundaries and the adjusted boundaries are visualised in Fig. 5.10.

Table 5.8. Relation between MMIF index values and quality classes.

MMIF	Evaluation of quality	Colour code
0.90 – 1.00	high quality	blue
0.70 – 0.85	good quality	green
0.50 – 0.65	moderate quality	yellow
0.30 – 0.45	poor quality	orange
0.00 – 0.25	bad quality	red

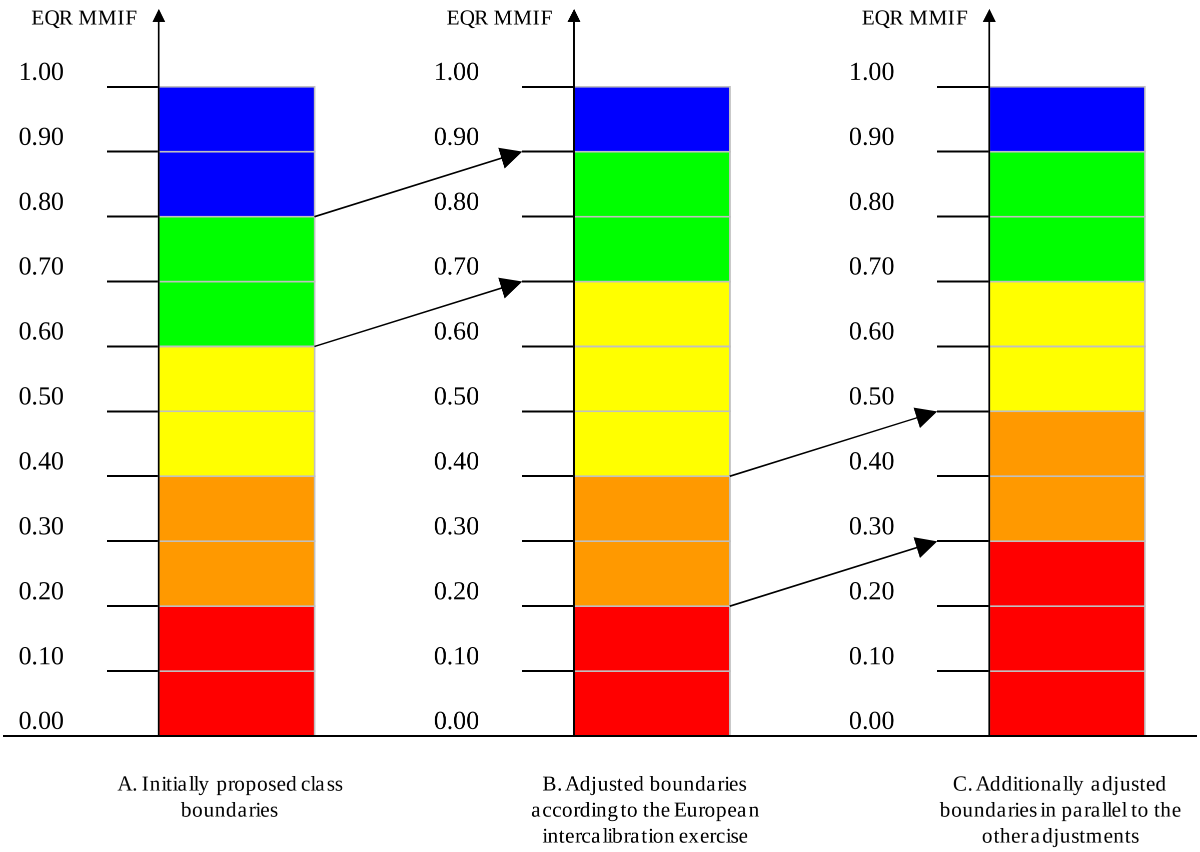

Figure 5.10. Visual representation of the initial MMIF boundary values (A), the boundaries following the adjustments according to the European intercalibration exercise (B), and the boundaries following additional adjustments in parallel to the other adjustments (C).

5.4.4 Scope of the intercalibration exercise and future perspectives

As already mentioned, the adjusted MMIF class boundaries will be included in a Commission Decision on the intercalibration exercise by the end of 2007. However, the intercalibration exercises were not yet completely carried out as envisaged by the WFD. For many quality elements and many categories, no or only a partial exercise was carried out (e.g. EU, 2007). Because these results are incomplete, a new round of intercalibration exercises should be initiated in order to achieve more exhaustive results. With regard to the scope of the MMIF, an intercalibration exercise was merely carried out for river macroinvertebrates, and only for five out of the eight different Flemish rivers types.

The reasons for this incompleteness of the intercalibration results are various. The timeframe to carry out the intercalibration scheme was rather limited. Also, many member states have not yet developed a set of national biological assessment methods, or have not yet collected a sufficiently large amount of monitoring data. To overcome this, many member states that participated in the exercise used an existing data set. Biggs (2006) argues that these incomplete results are the responsibility of the member states, and that more resources should be made available to ensure that a full intercalibration exercise can be carried out including all water body categories and all quality elements.

General discussion and recommendations

Introduction

The overall aims of this study can be summarised as follows:

- examine whether the currently used biological assessment method for watercourses based on macroinvertebrates, the Belgian Biotic Index, is compatible with the WFD requirements;
- if necessary, develop a modified or new assessment method that meets all WFD requirements and at the same time meets all practical constraints;
- demonstrate that the quality standard based on the existing or new assessment method is comparable to those of the other member states, or modify this standard in such a way to achieve this goal.

Results of the individual parts of this study were covered in detail in the previous chapters. In the following paragraphs, these objectives will be discussed from a more general perspective. To conclude, a number of recommendations are provided for further research in this field.

Using biological methods for water quality assessment

A biological quality assessment index provides a general indication of the status of a quality element in the water system. As a basic component of the ecosystem and closely interconnected with all other components, a biological community can serve as a representative part of the entire system (Karr and Chu, 2000). However, it should be emphasised that it can not be the purpose of a biological quality assessment index to provide a single and definitive judgment of the overall status of a water system. A single value cannot replace obtained insight in ecological functioning (e.g. Hynes, 1994). It should therefore be avoided to use them as the only source for conservation planning, because a quality index value does not deliver information on specific ecological questions such as invasive species, population fragmentation or conservational status of species.

When comparing macroinvertebrates with other biological indicator groups, such as diatoms, macrophytes or fish, it has generally been observed that they respond differently to different types of ecological stress (e.g. Triest et al., 2001a, 2001b; Hering et al., 2006) and are therefore complementary in their use for assessment. For this reason, a quality index should always be interpreted in complement with other methods of evaluation, not only physical-chemical and hydromorphological assessment, but also other biological indicator groups. This holistic view of ecological assessment was rightly incorporated in the WFD.

Regional adaptations are necessary to improve assessment systems (e.g. Thorne and Williams, 1997; Seegert, 2000; De Pauw et al., 2006), because water systems show important natural differences (Verdonschot and Nijboer, 2002). As a consequence, the importance (and related value) of the water system conditions can differ significantly among regions (leading to different scores due to its uses and values). Therefore most regions opt for a local assessment system, leading to the use of particular monitoring and assessment methods, as is presently for instance the case in most European member states (De Pauw et al., 2006).

Provided that all limitations and uncertainties of biological assessment indices (e.g. Seegert, 2000) are recognized, they constitute a very useful tool for evaluating the effects of management measures, for decision making and for promoting environmental awareness.

Compliance of the new Flemish biological assessment index with the WFD requirements

It has been argued in Chapter 3 that the new assessment index for macroinvertebrates in rivers and lakes in Flanders complies with all technical requirements of the WFD. Furthermore, the results of the first round of the intercalibration exercise (CB-GIG, 2006; Gabriels, 2007), more specifically the harmonised class boundary values, will be published by the end of 2007 as a Commission Decision (EU, 2007; see Chapter 5). With this publication, the methods can be considered as formally endorsed by the EU. However, as pointed out in Chapter 5, three Flemish river types and all Flemish lake types have not yet been subject to an intercalibration exercise, because the relevant types and categories were not yet covered during the first intercalibration round. These gaps will hopefully be filled during the next intercalibration round(s). Meanwhile, it can be concluded that all feasible tasks have thus far been carried out successfully for meeting the WFD requirements in Flanders with regard to freshwater macroinvertebrates.

Practicability of implementing the new Flemish biological assessment index in the existing monitoring networks

It should be clear that practical and financial constraints were an important consideration when developing this index. At the same time, the index needed to comply with the requirements of the WFD, to be scientifically sound and to be cost-effective for implementation in a routine monitoring scheme. It would however be unacceptable to sacrifice either one of these objectives, because all objectives need to be fulfilled simultaneously.

An interesting aspect with regard to the relation between the BBI and MMIF index is that they are both based on the same sampling and identification protocols. In this way, both indices can be calculated based on a single sample. This means that historical data can be evaluated by means of the MMIF as well, on the condition that abundance data are available and the genera *Physa* s.s. and *Physella* are distinguished. Consequently, it will in future be possible to make use of the same data to communicate quality evolution by means of an already widely known and accepted index on the one hand and to report results to the European Commission in the context of the WFD on the other hand.

Furthermore, the extensive know-how on logistics, sampling, sample processing and identification of macroinvertebrates that has been acquired by the VMM through the exploitation of the BBI monitoring network since 1989, can be perfectly incorporated into the monitoring efforts for the MMIF. The only difference concerns the counting of the animals and the calculation and interpretation of the index.

Comparing biological quality class boundaries across European member states

As discussed in Chapter 3, the normative definitions of the different quality classes (EU, 2000) leave much room for interpretation and consequently do not allow to unambiguously identify the "correct" boundary value on a given EQR scale. If this would have been possible, there would not have been a need for an intercalibration exercise. In this sense, the descriptions provided by the WFD should rather be seen as a rough guidance for setting the standard, whereas the intercalibration is necessary to ensure that the member states' standards are actually comparable.

Still, many uncertainties remain with regard to the outcome of the intercalibration decision, as discussed in Chapter 5. Despite all these practical and technical constraints, the outcome can be characterised as the currently best possibly achievable result. Furthermore, the intercalibration exercise should be seen as a gradual process that should be repeated in future when more field data will have been collected. However, more financial and technical resources should be made available to be able to refine the currently obtained results (Biggs, 2006).

In addition, many water systems are also characterised by multiple anthropogenic uses and related stresses (Verdonschot and Nijboer, 2002), which limit the development of the present biological community. For this reason, the WFD introduced the concept of Maximal Ecological Potential (MEP) and Good Ecological Potential (GEP) to be used instead of the reference conditions to assess the ecological state of a water body (Borja and Elliott, 2007). This MEP can be regarded as the best available ecological conditions under certain hydromorphological pressures that can not be mitigated without severe environmental or socio-economical consequences (EU, 2000). The MEP is an alternative target to be used instead of the reference conditions and should be defined for each artificial or heavily modified water body separately (EU, 2000). These heavily modified and artificial water bodies constitute the majority of the Flemish water bodies. However, a definitive register of water bodies to which this regime applies is not available yet. Also, the GEP must be defined for each heavily modified and/or artificial water body separately. An intercalibration of the ecological targets for these water bodies is not foreseen by the WFD. It is therefore doubtful whether monitoring results for artificial and heavily modified water bodies will be comparable among member states.

Estimation of the overall ecological status of water bodies in Flanders based on the WFD standards

The Flemish BBI standard of 7 as defined in VLAREM legislation (Flemish Government, 1995) is presently only met in approximately 30 percent of sampling locations (Peeters et al., 2006). Considering the relative equivalence between this BBI standard and the originally proposed MMIF class boundary for good quality (as demonstrated in Chapter 4), it can be concluded that the number of sampling locations complying with the adjusted MMIF standard, incorporating the results of the European intercalibration exercise, will be considerably less. In addition, due to the fact that a one-out-all-out approach is used for evaluating the overall ecological quality of a water body, it is clear that currently only a small minority of the natural water bodies can be expected to meet the good ecological status.

However, these intercalibrated standards apply to natural water bodies only. As previously mentioned, for artificial and heavily modified water bodies an alternative target must be set, which has at present not yet been finalised. It is therefore currently not possible to estimate the total number of water bodies in Flanders for which the European standard based on macroinvertebrates will be met.

Recommendations for further research

A biological assessment index is never intended to be used for infinity. An index should reflect the continuously evolving scientific insights, financial constraints, societal needs, and legislation. It is therefore appropriate that such an index will be re-evaluated when it has served as a routine monitoring tool for a longer period of time. Furthermore, pressures acting on water systems, such as morphological degradation and species invasions become increasingly important and will affect assessment results using existing methods/systems in an inconsistent manner (Friberg et al., 2006; see also Chapter 2). As a result of climate change, even reference conditions may change, which should also be taken into account in the assessment schemes (Nõges et al., 2007). In order to reflect these developments, the revision and improvement of methods should be an ongoing process (Friberg et al., 2006). These future adjustments of the method can incorporate new experience in sampling, identifying and calculation of the index. Also, the taxa list should be revised on a regular basis as proposed in Chapter 2.

On the other hand, as argued in Chapter 2, care should be taken not to present results based on different methods as comparable values, because the revision of an index implies that results of the original method should be seen as being based on a different method. In the present study, this has been systematically acknowledged by distinguishing between BBI and MMIF, although both indices are closely correlated, as was demonstrated in Chapter 4.

The whole information chain from data collection (including sampling) towards the final assessment should be standardised and subjected to quality assurance and quality control. An integrated uncertainty analysis of this information chain, from the perspective of the information needs of decision makers is necessary to assure that data collection and assessment is based on methods with the required precision and accuracy (Karr and Chu, 1999). Decision makers should thus also be aware of the level of uncertainty of the used methods and the impact of this uncertainty on the reliability of their planning and restoration actions.

The use of reference conditions has been one of the major innovations in biological water quality assessment of the last decade (Hering et al., 2004). Systems using site-specific reference conditions such as RIVPACS (Wright, 2000) are usually capable of comparing observed and reference values for a number of metrics (e.g. BMWP or ASPT), but, surprisingly, no multimetric index has thus far been proposed or tested using several metrics based on site-specific reference conditions. The development of such an index might be an interesting innovation in biological water quality assessment because it would combine the advantages of site-specific reference conditions on the one hand and the use of multiple assessment criteria on the other hand.

As required by the WFD, the MMIF provides, like the BBI, an overall evaluation of general ecological degradation, without distinguishing the source of stress. However, it may also be useful to develop pollution specific index systems (e.g. Chessman and McEvoy, 1998; Dahl and Johnson, 2004). To achieve this, it will however be necessary to collect a large and comprehensive set of data on macroinvertebrates as well as on all possible sources of degradation.

Further research should make maximal use of the currently available data analysis and modelling techniques. These techniques are becoming increasingly useful in the study of the biogeographic distribution of macroinvertebrates and their ecological preferences and interactions (e.g. Goethals, 2005; Recknagel, 2006). This would not only be very useful for ecological quality assessment, but also for facilitating water management decisions (Goethals, 2005).

For instance, assessment methods could be developed in a more reliable way by gathering information on ecological preferences of taxa and consequently on their vulnerability, habitat specificity and synecology. These insights would also deliver valuable information for decision support in river restoration management (Goethals and De Pauw, 2001), and also on cause detection of river deterioration (cf. the actual development of stressor specific metrics).

Such techniques can also be applied to develop habitat suitability models (e.g. Goethals, 2005; Goethals et al., 2007; Mouton et al., 2007) or to simulate migration patterns of possible recolonisation after habitat restoration measures (e.g. Dedecker et al., 2007). An other application is to predict reference communities based on abiotic properties of a site, which can be used in an assessment scheme (e.g. Wright, 2000).

Because the operation of a monitoring network generates a valuable amount of data on the one hand and ecological research generates useful knowledge to optimise monitoring strategies on the other hand, both disciplines can positively benefit from a more intense integration (Heylen et al., 1999; Goethals, 2005). Moreover, a wider availability of ecological data would greatly benefit ecological research (Parr, 2007). To facilitate the development of international databases on river ecology, the exchange of data collection and handling methods of, for instance, macroinvertebrates but also of physical, chemical and hydromorphological river characteristics will be of major importance (Goethals, 2005).

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Summary

One of the objectives of the European Water Framework Directive (WFD; EU, 2000) is to attain a good status for all surface waters in the European Union by the end of 2015. To this end, the ecological status of natural surface waters should be assessed, based on a number of biological quality elements which depend on the category of surface water (rivers, lakes, transitional waters or coastal waters). For each of these elements, member states must choose or develop a classification method, taking into account a set of parameters depending on the quality element and on the surface water category (EU, 2000). The assessment system must be differentiated among the types of water bodies within a category. The method must be in agreement with an Ecological Quality Ratio (EQR) showing relative proportion of the index compared to the reference conditions. This EQR ranges from zero to one, respectively corresponding to a bad and a very good ecological status. This interval is divided into five classes reflecting bad, poor, moderate, good and high ecological status (EU, 2000).

For the categories rivers and lakes, one of the relevant biological quality elements is the “benthic invertebrate fauna” (EU, 2000), commonly referred to as macroinvertebrates. For this quality element, the parameters “taxonomic composition and abundance”, “ratio of disturbance sensitive to insensitive taxa” and “diversity” should be taken into account.

In Flanders, Belgium, the Belgian Biotic Index (BBI; De Pauw and Vanhooren, 1983), based on macroinvertebrates, has been applied in routine monitoring schemes by the Flemish Environment Agency (VMM) since 1989, confirming the reliability and robustness of this biological quality index. However, with respect to the application of this index for rivers and lakes within a WFD context, not all technical requirements are met by the BBI. The abundance, which is one of the relevant parameters imposed by the WFD, is not taken into account in the BBI calculation, and this index is not explicitly based on a reference condition approach. Furthermore, it is not a type-specific method, in other words, all types of rivers are evaluated by means of the same criteria, and the BBI was intended as an assessment system for watercourses only, not for lakes (De Pauw and Vanhooren, 1983). Two general problems are identified that are associated with taxonomic resolution in water quality assessment based on macroinvertebrates. These are explored by means of analysis of the BBI index calculation method. A first difficulty is caused by possible changes in taxonomy over time, giving rise to

inconsistencies in index calculation. A second problem is due to the introduction of exotic species. Both problems can introduce a bias in calculation of the index. To avoid this problem in future assessment methods, it is proposed to use a taxa list to which no changes are made except for the addition of newly introduced exotic species.

Since the BBI does not meet all the requirements of the WFD, a new index, the Multimetric Macroinvertebrate Index Flanders (MMIF) for assessing rivers and lakes is proposed. This index is developed using the database of macroinvertebrate samples provided by the VMM. The MMIF is calculated based on macroinvertebrate community data obtained using the same sampling and identification procedure as the BBI. The index calculation is a type-specific multimetric system based on five equally weighted metrics, which are taxa richness, number of Ephemeroptera, Plecoptera and/or Trichoptera taxa, number of other sensitive taxa, the Shannon-Wiener diversity index, and the mean tolerance score. The final index value is expressed as an EQR ranging from zero for bad status to one for high status. The MMIF combines the robustness of the BBI and the long-term experience in Flanders with the flexibility of multimetric indices, while at the same time taking into account the technical requirements of the WFD.

To ensure that boundary values for the national biological assessment methods are comparable along all member states in Europe, regional intercalibration exercises were envisaged by the WFD for each quality element and for each category of water body. For river macroinvertebrates, the relevant regional intercalibration exercise for Flanders was coordinated by the so-called Central-Baltic Geographical Intercalibration Group (CB-GIG, 2006). Flanders contributed to this intercalibration exercise, using VMM sampling data, in order to compare the Flemish boundary values to those of the other participating countries and regions and, if necessary, to adjust them to ensure inclusion in the intercalibration decision of the European Commission (EU, 2007). This exercise for river macroinvertebrates essentially consisted in a regression of each participating member states' national method against a common, generally applicable index. This regression is carried out separately by each member state using a national dataset. Based on the obtained regression equation, each country converts its national boundary values into values on the scale of the common index for subsequent comparison. The mean value of the converted national boundaries is calculated and the rescaled national boundaries should be within a certain range of this mean value in order to be considered as comparable (CB-GIG, 2006). Two major problems emerged with regard to the Flemish contribution. First, the derivation of reference values for the metrics of the common index was not possible using field data because no reference sites are present in Flanders. To overcome this problem, an alternative and transparent way of deriving reference values was used in order to have reference values comparable to those of the other member states (Gabriels, 2007). The second problem concerned the boundary values, which clearly fell below the comparability interval. Therefore, alternative boundary values were proposed. This adjustment of the MMIF class boundaries for rivers was formally accepted by the steering group of the intercalibration exercise (CB-GIG, 2007). The adjusted values were adopted in the draft version of the intercalibration decision of the European Commission (EU, 2007), of which the official publication is foreseen for the end of 2007.

Samenvatting

Een van de doelstellingen van de Europese Kaderrichtlijn Water (KRW; EU, 2000) is het bereiken van een goede toestand voor alle oppervlaktewateren in de Europese Unie tegen eind 2015. Met dit doel moet de ecologische toestand van natuurlijke oppervlaktewateren beoordeeld worden, gebaseerd op een aantal biologische kwaliteitselementen, die variëren naargelang de oppervlaktewatercategorie (rivieren, meren, overgangswateren of kustwateren). Voor elk van deze elementen moeten lidstaten een beoordelingsmethode kiezen of ontwikkelen, die een geheel van parameters in rekening brengt afhankelijk van het kwaliteitselement en de oppervlaktewatercategorie (EU, 2000). Het beoordelingssysteem moet gedifferentieerd zijn naar de types waterlichamen per categorie. De methode moet in overeenstemming zijn met een Ecologische Kwaliteitsratio (EKR) die de relatieve verhouding van de index in vergelijking met de referentietoestand weergeeft. Deze EKR varieert van nul tot één, waarbij nul overeenkomt met een slechte en één met een zeer goede ecologische toestand. Dit interval wordt ingedeeld in vijf klassen, die respectievelijk een slechte, ontoereikende, matige, goede en zeer goede toestand weerspiegelen (EU, 2000).

Eén van de relevante biologische kwaliteitselementen voor de categorieën rivieren en meren is de “benthische ongewervelde fauna” (EU, 2000), doorgaans macro-invertebraten genoemd. Voor dit kwaliteitselement moeten de parameters “taxonomische samenstelling en abundantie”, “verhouding tussen voor verstoring gevoelige taxa en ongevoelige taxa” en “diversiteit” in rekening gebracht worden.

In Vlaanderen, België, wordt de Belgische Biotische Index (BBI; De Pauw en Vanhooren, 1983), gebaseerd op macro-invertebraten, reeds sinds 1989 gebruikt in de routine-meetnetten van de Vlaamse Milieumaatschappij (VMM), waardoor de betrouwbaarheid en robuustheid van deze biologische kwaliteitsindex bevestigd is. Wat de toepassing van deze index in rivieren en meren ten behoeve van de KRW betreft, wordt door de BBI evenwel niet aan alle technische vereisten voldaan. De abundantie, één van de relevante parameters opgelegd door de KRW, wordt niet in rekening gebracht in de berekening van de BBI, en deze index is niet expliciet gebaseerd op een referentietoestand-benadering. Daarenboven is het geen typespecifieke methode, met andere woorden, alle types rivieren worden aan de hand van dezelfde criteria beoordeeld, en was de BBI oorspronkelijk enkel bedoeld als een beoordelingssysteem voor waterlopen, en niet voor meren (De Pauw en Vanhooren, 1983). Twee algemene problemen worden geïdentificeerd die samenhangen met het taxonomische niveau in waterkwaliteitsbeoordeling gebaseerd op macro-invertebraten. Deze worden verder verkend door middel van een analyse van de BBI-berekeningsmethode. Een eerste moeilijkheid wordt veroorzaakt doordat na verloop van tijd wijzigingen in taxonomie kunnen worden doorgevoerd, die inconsistenties in indexberekeningen teweegbrengen. Een tweede probleem is te wijten aan de introductie van exotische soorten. Beide problemen kunnen een systematische afwijking in de berekening van de index veroorzaken. Om dit probleem op te vangen wordt voorgesteld om een taxalijst te gebruiken waarin geen wijzigingen worden aangebracht tenzij de toevoeging van nieuw geïntroduceerde exotische soorten.

Vermits de BBI niet voldoet aan alle vereisten van de KRW, wordt een nieuwe index, de Multimetrische Macro-invertebratenindex Vlaanderen (MMIF) voor het beoordelen van rivieren en meren voorgesteld. Deze index is ontwikkeld met behulp van de databank van macro-invertebratenstalen die ter beschikking werd gesteld door de VMM. De MMIF wordt berekend op basis van macro-invertebratenstalen die genomen worden volgende dezelfde staalname- en identificatieprocedure als de BBI. De indexberekening is een typespecifiek multimetrisch systeem gebaseerd op vijf even zwaar gewogen deelmaatlatten, namelijk de taxarijkdom, het aantal Ephemeroptera, Plecoptera en/of Trichoptera taxa, het aantal andere gevoelige taxa, de Shannon-Wiener diversiteitsindex en de gemiddelde tolerantiescore. De totale indexwaarde wordt uitgedrukt als een EKR variërend van nul voor een slechte toestand tot één voor een zeer goede toestand. De MMIF koppelt de robuustheid van de BBI en de lange-termijnervaring in Vlaanderen aan de flexibiliteit van multimetrische indices, waarbij tevens voldaan wordt aan de technische vereisten van de KRW.

Om te garanderen dat klassengrenzen van de nationale biologische beoordelingsmethoden onderling vergelijkbaar zijn tussen de Europese lidstaten, waren door de KRW regionale interkalibratie-oefeningen voorzien voor elk kwaliteitselement en voor elke waterlichaamcategorie. Voor macro-invertebraten in rivieren werd de relevante regionale interkalibratie-oefening voor Vlaanderen gecoördineerd door de zogenaamde Centraal-Baltische Geografische Interkalibratiegroep (CB-GIG, 2006). Vlaanderen nam deel aan deze interkalibratie-oefening, met staalnamegegevens van VMM, om de Vlaamse klassengrenzen te vergelijken met deze van de andere deelnemende landen en ze zonodig aan te passen om te verzekeren dat ze in de interkalibratiebeschikking van de Europese Commissie (EU, 2007) opgenomen zou worden. Deze oefening voor macro-invertebraten in rivieren bestond in wezen uit een regressie van de nationale methode van elke van de deelnemende lidstaten tegen een gemeenschappelijke, algemeen toepasbare index. Deze regressie wordt apart uitgevoerd door elke lidstaat op basis van een nationale gegevensset. Uitgaande van de bekomen regressievergelijking zet elk land haar nationale klassengrenzen om in waarden op de schaal van de gemeenschappelijke index om ze vervolgens te kunnen vergelijken. De gemiddelde waarde van de omgeschaalde nationale klassengrenzen wordt berekend en de omgeschaalde nationale klassengrenzen moeten binnen een zeker interval rond deze gemiddelde waarde liggen om als vergelijkbaar beschouwd te worden (CB-GIG, 2006). Twee belangrijke problemen deden zich voor met betrekking tot de Vlaamse bijdrage. Ten eerste was het afleiden van referentiewaarden voor de deelmaatlatten van de gemeenschappelijke index niet mogelijk op basis van veldgegevens omdat er geen referentiesites voorkomen in Vlaanderen. Om dit probleem te ondervangen werd een alternatieve en transparante manier om referentiewaarden af te leiden gebruikt om referentiewaarden te hebben die vergelijkbaar zijn met die van de andere lidstaten (Gabriels, 2007). Het tweede probleem betrof de klassengrenzen, die duidelijk onder het vergelijkbaarheidsinterval vielen. Daarom werden andere klassengrenzen voorgesteld. Deze aanpassing van de MMIF klassengrenzen voor rivieren werd formeel aanvaard door de stuurgroep van de interkalibratie-oefening (CB-GIG, 2007). De aangepaste waarden zijn opgenomen in de ontwerpversie van de interkalibratiebeschikking van de Europese Commissie (EU, 2007), waarvan de officiële publicatie voorzien is voor eind 2007.

Curriculum vitae

I. Personal data

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II. Education

-Secondary school: Latin-Mathematics (O.L.V. v. Lourdesinstituut Ekeren, Belgium, 1994)
-Candidate Bio-engineer (RUCA Antwerp, 1996)
-Bio-engineer option environmental technology, with distinction (Ghent University, 2000)

III. Work experience

-November 2005 until present:
Flemish Environment Agency (VMM), Department Monitoring and Research (AMO)
Supporting an international intercalibration exercise for biological assessment methods and assisting in the implementation of the national biological surface water monitoring and assessment schemes for the European Water Framework Directive

-April 2004 until October 2005:
Flemish Environment Agency (VMM), Department Ecological Control (AELT)
Coordinating the delineation of the individual and collective wastewater treatment zones in municipalities in the region of West Flanders

-September 2000 until April 2004:
Ghent University, Department Applied Ecology and Environmental Biology
Scientific assistant at the research unit Aquatic Ecology in the framework of various research projects

IV. Other activities

Supervision of practical exercises

Assisting in supervising the practical exercises of the courses “Aquatic Ecology” (students Bio-engineer) and “Biological water quality assessment” (students Bio-engineer and MSc Environmental Sciences and Technologies), Faculty of Agricultural and Applied Biological Sciences, Ghent University, 2000-2004

Supervision of script students

-Eleni Dakou, BSc Biology, Erasmus student at Ghent University from the Aristoteles University of Thessaloniki, 2001-2002
-Andy Dedecker, Bio-engineer, Ghent University, 2000-2001
-Peter Hermans, MSc Environmental Sciences and Technologies, Ghent University, 2000-2001

Jury member of script students

-Adjei-Boateng Augustina Asi, MSc in Environmental Sanitation, Ghent University, 2003-2004
-Yatta Samuel Laku Lukaw, MSc in Environmental Sanitation, Ghent University, 2003-2004
-Veerle Stuer, MSc in Environmental Science and Technology, Ghent University, 2002-2003

Promoter of script students

-Kathelijne De Ridder, MSc in Environmental Science and Technology, Ghent University, 2002-2003 (promoters: ir. Wim Gabriels and Prof. dr. N. De Pauw)

Organisation of workshops

-Co-organiser of the ISC (International Scheldt Commission) workshop of the working group PA4 (good status fresh surface waters) – physico-chemistry and biology, Hotel Eurovillage, Brussels, 11 May 2006
-Organiser of the Seminar PhD students research group Aquatic Ecology (AECO), Ghent University, 18 June 2003

Peer reviews for international journals

-Limnologica: 1 article (2007)
-PLoS ONE: 1 article (2007)
-Hydrobiologia: 2 articles (2004-2007)

Member of steering groups of research projects

-Research project "Onderbouwing maatregelenprogramma voor de Kaderrichtlijn Water" (2007): coordinator: Ann Huysmans (Flemish Environment Agency); contractor: Witteveen+Bos
-Research project "Ecologisch potentieel Meren" (2006 - 2007): coordinator: Gaby Verhaegen (Flemish Environment Agency); contractors: Research Institute for Nature and Forest (INBO) and Ghent University
-Research project "Verkennend onderzoek over eutrofiëring in Vlaanderen" (2005 - 2006): coordinator: Kor Van Hoof (Flemish Environment Agency); contractor: Ecosystem Management Research Group (ECOBE), University of Antwerp

-Research project "Afstemmen van referentiecondities en evaluatiesystemen in functie van de Kaderrichtlijn Water, partim hydromorfologie - applicatiestudie hydromorfologische beoordeling van waterlopen" (2005 - 2006): coordinator: Gaby Verhaegen (Flemish Environment Agency); contractor: Haskoning Belgium NV

V. Scientific awards

-Poster award of the "Happy Hour" meeting of B-IWA (Belgian Committee of the International Water Association), 10 june 2002
Title: Development of a Decision Support System for integrated water management in the Zwalm river basin, Belgium
Authors: T. D'heygere, V. Adriaenssens, A. Dedecker, W. Gabriels, P.L.M. Goethals & N. De Pauw

VI. Publications

Articles in international peer-reviewed journals (included in the Science Citation Index)

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Triest, L., Adriaenssens, V., Belpaire, C., Breine, J., D'heere, E., Gabriels, W., Goethals, P., Simoens, I. & De Pauw, N. (2001). Vergelijking van bio-indicatoren voor de ecologische evaluatie van waardevolle bovenstroomse beektrajecten. VLINA 00/08 rapport. Ministry of the Flemish Community, AMINAL, D/2001/3241/335, Brussels, Belgium. 149 p. + appendices + 4 maps.

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VII. Presentations

Oral presentations

(as presenter or as co-author; presenter is underlined; in case of co-presentations more than one author is underlined)

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Gabriels, W. & Maeckelberghe, H.: MEP GEP: waarover gaat het? In: “Toelichting voor de waterbeheerders. Wat is uw rol in het vaststellen van Maximaal Ecologisch Potentieel en Goed Ecologisch Potentieel?”, auditorium, VMM, Aalst, Belgium, 24 April 2007.

Amerlinck, Y., Benedetti, L., Goethals, P.L.M., Nopens, I., Dakou, E., Gabriels, W. & De Pauw, N.: Coupling of models for an integral ecological water quality management. In: B-IWA (Belgian Committee of the International Water Association) “Happy Hour” meeting, Brussels, Belgium, 22 March 2007.

Goethals, P., Gabriels, W., Messiaen, M., Mouton, A., Maes, A., Dakou, E. & De Pauw, N.: Trends in macro-invertebratendiversiteit in Vlaamse oppervlaktewateren In: WSK studiedag “Aquatische Biodiversiteit”, Ontmoetingscentrum Zebrastraat, Ghent, Belgium, 7 March 2007.

Gabriels, W., Goethals, P., De Pauw, N. & Verhaegen, G.: Ontwikkeling en interkalibratie van de ecologische indicatoren voor de KRW in Vlaanderen. In: Studiedag “Een goede kwaliteit voor onze oppervlaktewaters: zorgen voor morgen?”, Auditorium “Hof Van Liere”, University of Antwerp, Belgium, 21 November 2006.

Maeckelberghe, H. & Gabriels, W.: Resultaten projectgroep PA4 - goede toestand zoet oppervlaktewater. In: SCALDIT slotseminarie, Mechelen, Belgium, 24 October 2006.

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Appendices

Appendix 1. List of taxa included in the Multimetric Macroinvertebrate Index Flanders (MMIF) calculation and their tolerance scores (TS)

Taxon	TS
Plathelminthes	
Bdellocephala	5
Crenobia	7
Dendrocoelum	5
Dugesia	5
Phagocata	5
Planaria	6
Polycelis	6
Polychaeta	
Ampharetidae	3
Oligochaeta	
Aelosomatidae	2
Branchiobdellidae	2
Enchytraeidae	2
Haplotaxidae	4
Lumbricidae	2
Lumbriculidae	2
Naididae	5
Tubificidae	1
Hirudinea	
Cystobranchus	4
Dina	4
Erpobdella	3
Glossiphonia	4
Haementeria	4
Haemopis	4
Helobdella	4
Hemiclepsis	4
Hirudo	4
Piscicola	5
Theromyzon	4
Trocheta	4
Mollusca	
Acroloxus	6
Ancylus	7
Anisus	5
Anodonta	6
Aplexa	6
Armiger	6
Bathyomphalus	5
Bithynia	5
Bythinella	8
Corbicula	5
Dreissena	5
Ferrissia	7
Gyraulus	6
Hippeutis	6
Lithoglyphus	6
Lymnaea s.l.	5
Margaritifera	10
Marstoniopsis	5
Myxas	7
Physa s.s.	5
Physella	3
Pisidium	4
Planorbarius	5
Planorbis	6
Potamopyrgus	6
Pseudamnicola s.l.	5
Pseudanodonta	6
Segmentina	6
Sphaerium	4
Theodoxus	7
Unio	6
Valvata	6
Viviparus	6
Acari	
Hydracarina s.l.	5
Crustacea	
Argulidae	5
Asellidae	4
Astacidae	8
Atyidae	7
Cambaridae	6
Chirocephalidae	6
Corophiidae	5
Crangonyctidae	4
Gammaridae	5
Janiridae	5
Leptestheriidae	6
Limnadiidae	6
Mysidae	5
Palaemonidae	5

Taxon	TS
Sphaeromatidae	4
Talitridae	5
Triopsidae	6
Varunidae	4
Diptera	
Athericidae	7
Blephariceridae	7
Ceratopogonidae	3
Chaoboridae	3
Chironomidae:	
-non *thummi-plumosus*	3
-*thummi-plumosus*	2
Culicidae	3
Cylindrotomidae	3
Dixidae	6
Dolichopodidae	3
Empididae	3
Ephydridae	3
Limoniidae	4
Muscidae	3
Psychodidae	3
Ptychopteridae	3
Rhagionidae	3
Scatophagidae	3
Sciomyzidae	3
Simuliidae	5
Stratiomyidae	4
Syrphidae	1
Tabanidae	3
Thaumaleidae	3
Tipulidae	3
Megaloptera	
Sialis	5
Coleoptera	
Dryopidae	6
Dytiscidae	5
Elminthidae	7
Gyrinidae	7
Haliplidae	6
Hydraenidae	6
Hydrophilidae	5
Hygrobiidae	5
Noteridae	5
Psephenidae	6
Scirtidae	7
Hemiptera	
Aphelocheirus	8
Arctocorisa	5
Callicorixa	5
Corixa	5
Cymatia	6
Gerris s.l.	6
Glaenocorisa	5
Hebrus	6
Hesperocorixa	5
Hydrometra	6
llyocoris	5
Mesovelia	6
Micronecta	6
Microvelia	7
Naucoris	6
Nepa	6
Notonecta	5
Paracorixa	5
Plea	6
Ranatra	6
Sigara	5
Velia	7
Odonata	
Aeshna	6
Anax	6
Brachytron	7
Calopteryx	8
Cercion	7
Ceriagrion	7
Coenagrion	6
Cordulegaster	9
Cordulia	7
Crocothemis	7
Enallagma	7
Epitheca	7
Erythromma	7
Gomphus	7

Taxon	TS
Ischnura	6
Lestes	7
Leucorrhinia	7
Libellula	7
Nehalennia	7
Onychogomphus	7
Ophiogomphus	7
Orthetrum	7
Oxygastra	7
Platycnemis	7
Pyrrhosoma	7
Somatochlora	7
Sympecma	7
Sympetrum	7
Ephemeroptera	
Baetis	6
Brachycercus	7
Caenis	6
Centroptilum	7
Cloeon	6
Ecdyonurus	9
Epeorus	10
Ephemera	8
Ephemerella	8
Ephoron	9
Habroleptoides	8
Habrophlebia	8
Heptagenia	10
Isonychia	7
Leptophlebia	8
Metreletus	7
Oligoneuriella	7
Paraleptophlebia	8
Potamanthus	8
Procloeon	7
Rhitrogena	10
Siphlonurus	7
Trichoptera	
Beraeidae	9
Brachycentridae	9
Ecnomidae	6
Glossosomatidae	9
Goeridae	9
Hydropsychidae	6
Hydroptilidae	8
Lepidostomatidae	9
Leptoceridae	8
Limnephilidae	8
Molannidae	9
Odontoceridae	9
Philopotamidae	6
Phryganeidae	9
Polycentropodidae	6
Psychomyidae	7
Rhyacophilidae	8
Sericostomatidae	8
Plecoptera	
Amphinemura	9
Brachyptera	10
Capnia	10
Chloroperla	10
Dinocras	10
Isogenus	10
Isoperla	10
Leuctra	9
Marthamea	10
Nemoura	8
Nemurella	8
Perla	10
Perlodes	10
Protonemura	9
Rhabdiopteryx	10
Taeniopteryx	10

Appendix 2. List of consulted experts to review the development of the Multimetric Macroinvertebrate Index Flanders (MMIF)

Annick De Winter	VMM	Ghent, Belgium
Boudewijn Goddeeris	KBIN	Brussels, Belgium
Marianne Greijdanus-Klaas	RIZA	Lelystad, the Netherlands
Joost Mertens	VMM	Ghent, Belgium
Jean-Pierre Vanden Bossche	DGRNE	Gembloux, Belgium
Rudy Vannevel	VMM	Aalst, Belgium
Thierry Vercauteren	PIH	Antwerp, Belgium
Thierry Warmoes	VMM	Leuven, Belgium

Appendix 3. Overview of scoring criteria for the Multimetric Macroinvertebrate Index Flanders (MMIF)

	Rivers								**Lakes**			
Type	**Bk**	**BkK**	**Bg**	**BgK**	**Rk**	**Rg**	**Rzg**	**P**	**A**	**C**	**Z**	**Bzl**
Score	**TAX**											
0	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5	≤ 5
1	≤ 12.25	≤ 12.25	≤ 13.25	≤ 13.25	≤ 13.75	≤ 14.25	≤ 14.75	≤ 13	≤ 12	≤ 12.5	≤ 10.75	≤ 11.25
2	≤ 19.5	≤ 19.5	≤ 21.5	≤ 21.5	≤ 22.5	≤ 23.5	≤ 24.5	≤ 21	≤ 19	≤ 20	≤ 16.5	≤ 17.5
3	≤ 26.75	≤ 26.75	≤ 29.75	≤ 29.75	≤ 31.25	≤ 32.75	≤ 34.25	≤ 29	≤ 26	≤ 27.5	≤ 22.25	≤ 23.75
4	> 26.75	> 26.75	> 29.75	> 29.75	> 31.25	> 32.75	> 34.25	> 29	> 26	> 27.5	> 22.25	> 23.75
Score	**EPT**											
0	0	0	0	0	0	0	0	0	0	0	0	0
1	≤ 1.75	≤ 2	≤ 2	≤ 2.25	≤ 2.25	≤ 2.25	≤ 2.5	≤ 2	≤ 1.5	≤ 2	≤ 1.25	≤ 1.25
2	≤ 3.5	≤ 4	≤ 4	≤ 4.5	≤ 4.5	≤ 4.5	≤ 5	≤ 4	≤ 3	≤ 4	≤ 2.5	≤ 2.5
3	≤ 5.25	≤ 6	≤ 6	≤ 6.75	≤ 6.75	≤ 6.75	≤ 7.5	≤ 6	≤ 4.5	≤ 6	≤ 3.75	≤ 3.75
4	> 5.25	> 6	> 6	> 6.75	> 6.75	> 6.75	> 7.5	> 6	> 4.5	> 6	> 3.75	> 3.75
Score	**NST**											
0	0	0	0	0	0	0	0	0	0	0	0	0
1	≤ 2.25	≤ 2.25	≤ 2.5	≤ 2.5	≤ 3	≤ 3	≤ 3	≤ 2.5	≤ 2.5	≤ 2.5	≤ 2	≤ 2.25
2	≤ 4.5	≤ 4.5	≤ 5	≤ 5	≤ 6	≤ 6	≤ 6	≤ 5	≤ 5	≤ 5	≤ 4	≤ 4.5
3	≤ 6.75	≤ 6.75	≤ 7.5	≤ 7.5	≤ 9	≤ 9	≤ 9	≤ 7.5	≤ 7.5	≤ 7.5	≤ 6	≤ 6.75
4	> 6.75	> 6.75	> 7.5	> 7.5	> 9	> 9	> 9	> 7.5	> 7.5	> 7.5	> 6	> 6.75
Score	**SWD**											
0	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2	≤ 0.2
1	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 1.025	≤ 0.9	≤ 0.95
2	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.85	≤ 1.6	≤ 1.7
3	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.675	≤ 2.3	≤ 2.45
4	> 2.675	> 2.675	> 2.675	> 2.675	> 2.675	> 2.675	> 2.675	> 2.675	> 2.675	> 2.675	> 2.3	> 2.45
Score	**MTS**											
0	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2	≤ 2
1	≤ 3.125	≤ 3.125	≤ 3.125	≤ 3.125	≤ 3.125	≤ 3.125	≤ 3.125	≤ 3.075	≤ 3	≤ 3	≤ 3	≤ 3
2	≤ 4.25	≤ 4.25	≤ 4.25	≤ 4.25	≤ 4.25	≤ 4.25	≤ 4.25	≤ 4.15	≤ 4	≤ 4	≤ 4	≤ 4
3	≤ 5.375	≤ 5.375	≤ 5.375	≤ 5.375	≤ 5.375	≤ 5.375	≤ 5.375	≤ 5.225	≤ 5	≤ 5	≤ 5	≤ 5
4	> 5.375	> 5.375	> 5.375	> 5.375	> 5.375	> 5.375	> 5.375	> 5.225	> 5	> 5	> 5	> 5

Appendix 4. Screening criteria for potential reference sites in the Central-Baltic intercalibration exercise for river macroinvertebrates (CB-GIG, 2006)

Suggested template
REFCOND-Guidance
High status or reference conditions is a state in the present or in the past corresponding to very low pressure, without the effects of major industrialisation, urbanisation and intensification of agriculture, and with only very minor modification of physico-chemistry, hydromorphology and biology.
Suggestion for GIG
Totally unaffected sites do not exist anymore (at least due to the world wide atmospheric deposition). As “close-to-pristine” state is unlikely to be encountered, (except perhaps in some national parks), the concept of "pristine state" is not relevant in practice for the definition of reference conditions for the Central Baltic GIG.
If an historic database has to be used, this should be from a time period without intensive industries, hydraulic engineering and agriculture.
Selection criteria for reference sites are based on “anthropic pressures”, that must be “null or very low”; the problem is to define a very low pressure level that leads to insignificant or very low impact at the ecosystem level. “Insignificant impact” could be understood as “hardly distinguishable from natural (spatial and temporal) variability” at the level of the biological elements. A first validation of “very low impact” should be assessed at the level of abiotic parameters (physico-chemistry and hydro-morphology).
In the first stage, biological elements are not considered as selection criteria.
In the second stage, those sites whose aquatic communities exhibit statistically low biological values are carefully checked for pressures, and dubious sites are eliminated. The checking process must consider possible errors in evaluating the pressures, and in sampling methods for biological communities.
If, after checking, no significant pressure or possible error is encountered, these sites are considered as representative of the type’s natural variability.
However, any samples falling outside the range of “good ecological status” should not be included in the calculation of the reference value for the biological quality element considered.
Impacts on rivers or within the catchment should not affect the original characteristics, so that the aquatic community is only altered minimally. Type-specific communities and conditions should be represented.
A river stretch that is considered for the selection of a reference site must be situated within one national type. It must have biological populations representative of the type.

Pressures likely to affect the reference site must be evaluated at the three relevant spatial scales: the **catchment** of the site, the **reach** scale (i.e. the water body), and the **reference site** itself.
Proposed minimum length for the river reaches are: > 1 km for small rivers (stream order 1- 3), > 5 km for medium-size r. (stream order 4 - 5), > 10 km for large rivers (stream order > 6).
For each pressure criteria, two thresholds are defined:
a “reference” threshold, below which a site is considered as “probably reference”;
a “rejection” threshold, corresponding to a high probability of significant impact, above which a site is eliminated.
Sites that have all criteria below the reference threshold are considered as reference sites; sites having most criteria below the reference threshold and only some parameters between the reference and rejection threshold are “possible reference sites”. For these sites, only a few possible pressures (i.e less than 10% of the criteria) should exceed the reference threshold level. If a site exceeds the rejection threshold on any one criterion it should be eliminated. These sites should be retained only after carefully checking the cumulative effects of the pressures using local expertise.
Impacts on rivers or within the catchment area should have only local effects to be considered in Reference State.
It is proposed to use the CORINE Land Cover (CLC) classification for the evaluation of the land use in the catchment and riparian area. However, the land cover represents a “driving force” more than a “pressure”, and thus must be understood as representing a probability of impact. The CORINE land cover classification should be regarded with some caution. The following definitions are proposed :
*<u>Artificial land use</u> : the sum of all the categories of **CLC class 1**. (Urban areas continuous and discontinuous, industrial and commercial zones, communication infrastructures and networks, mines, etc..)*
*<u>Intensive agriculture</u> : the sum of the CLC categories corresponding to a high potential impact from agricultural activities: arable land (including irrigated land), permanent crops (with associated annual crops), vineyards, orchards, olive groves, complex cultivation patterns, - **CLC codes : 2.1, 2.2, 2.4.1, 2.4.2**.*
*<u>Low intensity agricultural areas</u>: the sum of the CLC categories corresponding to a lower potential impact from agricultural activities: pastures, land principally occupied by agriculture, with significant areas of natural vegetation, agro-forestry areas - **CLC codes : 2.3.1, 2.4.3, 2.4.4**.*
*<u>Semi-natural areas</u>: Forest and natural areas, wetlands, water bodies - codes CLC codes : **3.1.1, 3.1.2, 3.1.3, 3.2, 3.3, 4 and 5.***
Point source pollution
Other effluents/discharges (Urban pollution)
REFCOND-Guidance
No or very local discharges with only very minor ecological effects.
Suggestion for GIG
Only minor impairments of the physical and chemical conditions, this means: Near-natural background values

No or very local discharges with only very minor ecological effects.
No known industrial cause of particular pollution (e.g. NaCl, thermal pollution, etc…)
The following criteria can be used to validate very low levels of point source pollution:
Very low level of urbanisation, evaluated by the percentage area of artificial areas in the catchment CLC class 1: see line 26). The following thresholds can be used :
"Reference" threshold : < 0.4% of artificial land use in the catchment area. (Between 0.4% and 0.8%: see line 19-22)
"Rejection" threshold : 0.8 % of artificial area in the catchment.
Above 0.8%, a validation with physico-chemical parameters at the site scale is necessary.
See separate table for chemical reference values.
For small streams: no known point source discharge, or very localised impact with self purification.
For larger streams and rivers : very low point source discharge level. If point sources are present, a validation with chemical parameters is necessary. ***See separate table for chemical reference values***.
Alternatively, the saprobiological water quality class (according to types or ecoregions) can be used to validate "very minor" ecological effects. If this criteria is used, it must be explained.
Specific synthetic pollutants
REFCOND-Guidance
Pressures resulting in concentrations close to zero or at least below the limits of detection of the most advanced analytical techniques in general use (A selection process for relevant pollutants in a river basin is presented as an example of best practice in section 6 of the guidance document from Working Group 2.1, IMPRESS).
Suggestion for GIG
Substances mentioned in Annex X and/or in annex VIII of the WFD should have concentrations at least below the limits of detection of the most advanced analytical techniques in general use
Measured values of other anthropogenic, synthetic substances should be below quality objectives or near natural background concentrations, except for those from atmospheric sources.
The impact of atmospheric pollution on reference river stretches must not be detectable (e.g. depletion of the aquatic community due to acidification)
Spec. non-synthetic pollutants
REFCOND-Guidance
Natural background level/load (see reference above)
Suggestion for GIG
Only minor impairments of the physical and chemical conditions, this means: Near-natural background values– if this can be can estimated; if not, the limit of detection (quantitative) can be used tentatively.
No known discharge of specific non-synthetic pollutants upstream in the river.
If no chemical data are available, the following criteria can be used to validate the very low level of general toxic pressures:

- For small streams : no known toxic pollution discharge.
- For larger streams and rivers : no suspected toxic pollution discharge; if (actual or ancient) toxic pollution sources exist in the basin, ratio PEC / PNEC < 1.
In agricultural areas, sites with a known pollution risk by pesticides (according to existing risk maps) are avoided.
Diffuse source pollution
Land-use intensification: Agriculture, forestry
REFCOND-Guidance
Pre-intensive agriculture or impacts compatible with pressures pre-dating any recent land-use intensification. Pressures pre-dating any recent intensification in airborne inputs that could lead to water acidification.
Suggestion for GIG
The share of anthropogenic land use in the catchment area (agriculture, afforestation) must be small and shows only local effects. In the case of type-specific floodplains, lateral and vertical connectivity has to be maintained. The reference sites must have a wide riparian buffer zone with type specific riparian vegetation.
The land use upstream of the reference site must comply with the following criteria *(land use definition see lines 26-29)*
Intensive agriculture : <20% of the catchment area as reference threshold. Rejection threshold: > 50% of intensive agriculture in the catchment. However, in flat lowlands agricultural landscapes, sites with 20% to 50% of intensive agriculture can be considered only if :
1) there is no significant risk of soil erosion
2) the valley floors are mainly occupied by low intensity agricultural area (mainly pastures) and /or semi-natural areas, and riparian corridors are globally preserved at the reach and site scales. (*See Riparian vegetation criteria line 98)*
Between 20%and 50% of intensive agriculture, a validation with physico-chemical parameters at the site scale is strongly recommended.
See separate table for chemical reference values.
Cattle breeding: only non-intensive (outdoor) cattle breeding; < 1.25 animal (cattle) units per ha of the catchment area.
Vineyards, orchards: < 1% of the catchment area, and not situated in the riparian zone.
Irrigated fields ≤ 10%
Forestry : < 30% tree plantations (coniferous, Eucalyptus..).
If tree plantations > 30% in the catchment, even with no sign of acidification, the riparian corridor must be protected and composed of the type specific natural vegetation. **See Riparian vegetation criteria**
Acidification : no sign of acidification due to coniferous plantation (on siliceous bedrock).
pH > 6. If pH < 6 , it is necessary to determine if the site is naturally acid.
Eutrophication : no sign of plant proliferation (macrophytes, algae).
Eutrophication : if possible validate with chemical values
See separate table for chemical reference values.
Riparian zone vegetation

REFCOND-Guidance
Having adjacent natural vegetation appropriate to the type and geographical location of the river.
Suggestion for GIG
definition of the riparian zone: the minimum width of the riparian zone (or corridor) to be considered is 30m for small streams (order 1-3), 50m for medium size rivers (order 4 - 5) and 100 m for larger rivers (order ≥ 6)
<u>At the reach scale:</u>
In agricultural landscape (Intensive agriculture between 20% and 50%), intensive agriculture land cover < 10% of the reach. Riparian corridor land use > 90% semi natural or low intensity agricultural areas.
In non agricultural landscape (Intensive agriculture < 20%): valley floor and riparian corridor occupied by semi natural or low intensity agricultural areas.
Artificial areas: < 10% of the reach.
<u>At the site scale :</u>
The riparian zone of the site is entirely bordered by the type specific natural vegetation or semi-natural land cover, with the possible exception of access to the river site. *(land use definition see lines 26-29).*
Riparian vegetation zone continuity: uninterrupted or with few interruptions (access to the site).
The lateral connectivity between river and riparian corridor is maintained along the site.
No direct impact of cattle trampling.
Morphological alterations
River morphology
REFCOND-Guidance
Level of direct morphological alteration, e.g. artificial instream and bank structures, river profiles, and lateral connectivity compatible with ecosystem adaptation and recovery to a level of biodiversity and ecological functioning equivalent to unmodified, natural water bodies
Suggestion for GIG
The type-specific hydromorphological conditions are maintained (including the elements mentioned in annex V of the WFD), leading to the conservation of all types of associated physical habitats.
The natural morphological dynamic is maintained, with no or very minor anthropogenic influence. Slightly altered morphological conditions have a high potential to return to natural flow conditions without human action in near future .
<u>At the basin scale:</u>
<u>Sediment transport:</u> No dams which significantly modify the sediment regime (sediment retention) leading to morphological alterations, evidenced by signs of incision of the river bed (e.g. incision > 0.2m * stream order, bare bed rock appearing…).
<u>Migration barriers for fish reference sites</u> : this issue has to be addressed specifically by the fish experts for the definition of reference conditions for fishes.

<u>Suggestion for fish reference conditions</u> :
"Continuity" for fish should be related to the maintenance of river and stream continuity to facilitate movement of type specific species that should be present in reference state - for example, fish should have access to spawning grounds (which may be in upper reaches) as well as nursery areas (which may be back waters) and amphidromous species should have access to the sea.
If this condition is not fulfilled and some migratory species have disappeared, these species should be added to the type -specific list of fish species.
<u>1) at the reach scale</u> *(if no general mapping of morphological alterations exists, an expert evaluation is required for the selected reach)*:
Flow impedance: < 10% of the reach is affected by flow impedance, due to hydraulic effects of weirs, sluices, etc… *The % of the reach affected by flow impedance can be evaluated by the ratio of the sum of weirs' heights (in meters) to the total difference in height (slope * lenght, in meters) between the upper and lower end of the reach.*
Channelisation: < 10% of the reach is affected by “hard works” (like modification of longitudinal and / or transverse profiles, narrow embankment, loss of lateral connectivity…), otherwise, bed and banks composed of natural materials
Stabilisation: < 20% of the reach is affected by “soft works” (like bank protection on one side, distant dikes, bank maintenance, not affecting the longitudinal and / or transverse profile, and lateral connectivity globally maintained…).
If both types of works are combined (lines 134 and 135) < 10% of the reach must be affected.
Siltation: reaches with anomalous siltation suspected, due to agricultural soil erosion, should be avoided (expert judgment).
Connection to groundwater: Total lateral and vertical connection to groundwater.
Substrate conditions: Correspond to related typology
River profile and variation in width and depth: Correspond to related typology
River continuity: At the reach scale, the continuity of the river is not disturbed by anthropogenic barriers and allows undisturbed migration of aquatic organisms (including resident fish populations).
River continuity: At the reach scale, the continuity of the river is not disturbed by anthropogenic barriers and allows free sediment transport.
<u>2) at the site scale</u> :
The site is not situated in a zone directly or indirectly impacted by a nearby artificial structure upstream or downstream.
Lacking any instream structural modifications (weirs or dams) that affect the longitudinal and lateral connectivity, and natural movement of river bed, sediment load, water and biota (except for natural waterfalls).
Only very small artificial constructions with very minor local effects can be accepted
Water abstraction
REFCOND-Guidance
Levels of abstraction resulting in only very minor reductions in flow levels or lake level changes having no more than very minor effects on the quality elements.
Suggestion for GIG

<u>At the basin scale:</u>
No dams or water storage significantly altering the low flow regime; low flow alteration < 20% of the monthly minimum flow.
<u>At the reach scale:</u>
Only very minor reductions in flow level changes having no more than very minor effects on the quality elements.
No significant water abstraction in the reach. The cumulative effect of water regulation and abstraction at the basin and reach scales is < 20% of low flow discharge.
River flow regulation
REFCOND-Guidance
Levels of regulation resulting in only very minor reductions in flow levels or lake level changes having no more than very minor effects on the quality elements. Flow regulation that has the potential to recover to natural flow in near future.
Suggestion for GIG
<u>At the basin scale:</u>
No dams which significantly modify the natural hydrological flow regime (flow regulation) : e.g. suppression of frequent floods (<5 years) with anomalous development of vegetation in the channel, or low flow alteration (< to + or - 20% modification of the natural monthly minimum flow discharge).
The total storage capacity of the reservoirs in the catchment is < 5% of the mean annual discharge at the site.
No change of the natural (type specific) annual flow characteristics (seasonality of high and low flow)
<u>At the reach scale</u>
No by-passed section with residual flow (legal minimum discharge)
No significant hydropower peaking effect (ratio Q hydropeaking / Q baseflow < 2)
Absence of flow regulation (dam) on the reach itself.
Biological pressures
Introductions of alien species
REFCOND-Guidance
Introductions compatible with very minor impairment of the indigenous biota by introduction of fish, crustacea, mussels or any other kind of plants and animals.
No impairment by invasive plant or animal species.
Suggestion for GIG
NB: the issue is: to give a sound definition of “alien species” and “type-specific species” and to make clear if the one can shift into the other, and if so on what conditions. We consider this as an item that should be discussed and solved on a European level.
<u>Proposed definition of alien species</u> : non indigenous species recently introduced (i.e. during the XXth century) or in early stage of dissemination in the river reach, not known to present a risk of being invasive.

Proposed definition of invasive species : alien species in stage of active colonisation, which are quantitatively predominant in their respective community, and whose development significantly alter the composition and abundance of the type specific communities. These species, by direct or indirect effects, can induce a risk of extinction of indigenous biota, and alter the global ecosystem functioning.
At the site scale, no invasive species, but alien species which are not at the invasive stage are tolerated.
Fisheries and aquaculture
REFCOND-Guidance
Fishing operations should allow for the maintenance of the structure, productivity, function and diversity of the ecosystem (including habitat and associated dependent and ecologically related species) on which the fishery depends.
Stocking of non indigenous fish should not significantly affect the structure and functioning of the ecosystem.
No impact from fish farming.
Suggestion for GIG
No intensive (commercial) fishery.
Fisheries, fish management and/or aquaculture plants which have no significant impact on fish populations are tolerated, i.e. the type specific fish population is maintained (*for alien species, see line 184*)
Fishing or stocking of fish is limited, and must have no impact on the ecosystem functioning.
No or very limited direct pollution by aquaculture plants.
Biomanipulation
REFCOND-Guidance
No biomanipulation.
Suggestion for GIG
No biomanipulation.
Other pressures
Recreation uses
REFCOND-Guidance
No intensive use of reference sites for recreation purposes (no intensive camping, swimming, boating, etc.)
Suggestion for GIG
No nearby intensive recreational use at the site scale: No regular bathing activities or motor boating. Occasional recreational uses (such as camping, swimming, boating, etc.) should lead to no or very minor impairment of the ecosystem.

Chemical thresholds	Type	R-C1	R-C2	R-C3	R-C4	R-C5	R-C6
BOD_5 (mg/L)	mean	2.4	2.4	2	2.4	2.4	2.4
	90th perc.	3.6	3.6	2.75	3.6	3.6	3.6
Dissolved Oxygen Saturation (%)	mean	95-105	95-105	95-105	95-105	95-105	95-105
	10-90th perc.	85-115	90-110	90-110	85-115	85-115	85-115
$N\text{-}NH_4$ (mg/L)	mean	0.1	0.05	0.05	0.1	0.1	0.1
	90th perc.	0.25	0.12	0.12	0.25	0.25	0.25
$N\text{-}NO_3$ (mg/L)	mean	6	6	2	6	6	6
$P\text{-}PO_4$ (µg/L)	mean	40	30	20	40	40	40