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ICP Waters Report 113/2012

Biological intercalibration: Invertebrates 1612

International Cooperative Programme on Assessment and Monitoring Effects of Air Pollution on Rivers and Lakes

Convention on Long-Range Transboundary Air Pollution

Norwegian Institute for Water Research
– an institute in the Environmental Research Alliance of Norway

REPORT

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Abstract

The 16th intercalibration of invertebrates in the ICP Waters programme had contribution from three laboratories. The laboratories identified a high portion of the individuals in the test samples, usually > 90% of the total number of species. Few faults were recorded on genus level. The mean Quality assurance index was > 90% for all participating laboratories, indicating excellent taxonomic work.

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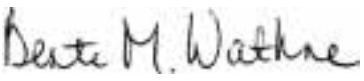
1. Interkalibrering	1. Intercalibration
2. Invertebrater	2. Invertebrates
3. Akvatisk fauna	3. Aquatic fauna
4. Overvåking	4. Monitoring



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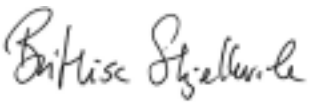

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CONVENTION ON LONG-RANGE TRANSBOUNDARY AIR POLLUTION

INTERNATIONAL COOPERATIVE PROGRAMME ON ASSESSMENT AND MONITORING EFFECTS OF AIR POLLUTION ON RIVERS AND LAKES

Biological intercalibration: Invertebrates 1612

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Preface

The international cooperative programme on assessment and monitoring of air pollution on rivers and lakes (ICP Waters) was established under the Executive Body of the UNECE Convention on Long-range Transboundary Air Pollution (LRTAP) in July 1985. Since then ICP Waters has been an important contributor to document the effects of implementing the Protocols under the Convention. Numerous assessments, workshops, reports and publications covering the effects of long-range transported air pollution have been published over the years.

The ICP Waters Programme Centre is hosted by the Norwegian Institute for Water Research (NIVA), while the Norwegian Climate and Pollution Agency (Klif) leads the programme. A programme subcentre is established at Uni Research, University of Bergen. The Programme Centre's work is supported financially by Klif and from the UNECE LRTAP Trust Fund.

The main aim of the ICP Waters Programme is to assess, on a regional basis, the degree and geographical extent of the impact of atmospheric pollution, in particular acidification, on surface waters. More than 20 countries in Europe and North America participate in the programme on a regular basis.

The Programme objective is to establish and maintain an international network of surface water monitoring sites and promote international harmonisation of monitoring practices. A tool in this work is the inter-laboratory quality assurance tests. The bias between analyses carried out by the individual participants of the Programme has to be identified and controlled. The tests will also be a valuable tool in improving the taxonomic skill of the participating laboratories.

We here report the results from the 16th intercalibration on invertebrate fauna.

Bergen, November 2012

Arne Fjellheim
ICP Waters Programme Subcentre

Contents

Summary

The 16th intercalibration of invertebrates in the ICP Waters programme had contribution from three laboratories. The biological intercalibration is important for harmonising biological material/databases and will be of high value in programmes where community analyses is in focus or where the ecological status should be stated, like EU Water Framework Directive. The biological intercalibration under the ICP Waters programme is a unique test, as it operates on a species level.

The laboratories generally identified a high portion of the total number of species in the test samples, usually > 90% of the total number of species. Few faults were recorded on genus level. The mean Quality assurance index was > 90% for all participating laboratories, indicating excellent taxonomic work. None of the participants did misidentifications that could result in a wrong acidity index, based on the Raddum score (Raddum et al., 1988).

1. Introduction

The purpose of the biological intercalibration is to evaluate the quality of the taxonomic work on the biological material delivered to the Programme centre. The quality can influence on the evaluation of the samples, which is based on the species and their tolerance (Raddum *et al.* 1988, Fjellheim and Raddum 1990, Raddum 1999). The control is therefore important for evaluation of the significance of trends in biotic indexes both for a specific site/watershed, as well as for comparisons of trends between different regions and countries. The material is also used for multivariate statistical analysis (Larsen *et al.* 1996, Skjelkvåle *et al.* 2000, Halvorsen *et al.* 2002, Velle, 2013). The results of this type of data treatment are especially sensitive to the quality of the species identification. The biological intercalibration focuses on the taxonomic skills of the participants and is a tool for improving the quality of work at the different laboratories as well as harmonisation of the biological database.

The methods for intercalibration of biological material were outlined in 1991 at the 7th ICP Waters Task Force meeting in Galway, Ireland. The different countries/laboratories have to know, first of all, their home fauna. Since the fauna in different geographical regions vary, it is necessary to prepare specific samples for each participating laboratory, based on their home fauna. It is a problem for the exercise of the intercalibration that it is not possible to use standardised samples for all participants. To solve this problem, each laboratory send identified samples of invertebrates from their own monitoring sites to the Programme centre. The Programme centre will additionally add species known to be present in the region of the specific laboratory. Based on this, each laboratory receives individual test samples composed of species representing their own monitoring region.

The taxonomic skill of the different participants is measured by using a quality assurance index, see Raddum (2005). This index evaluates the skill of identifying the species as well as the genus. It also takes into account the effort of identifying all specimens in the sample. The highest index score is 100, while a value of 80 is set as the limit of good taxonomic work.

2. Methods

2.1 Preparation of test-samples

Samples of identified invertebrates were received from all participating laboratories. These samples were used to compose test samples, with the addition of specimens from earlier exercises and from own stocks. The geographical distribution of species is checked by the use of the Fauna Europaea Web Service 2012 (http://www.faunaeur.org). This is a database of the scientific names and distribution of multicellular European land and fresh-water animals (see example in Figure 1).

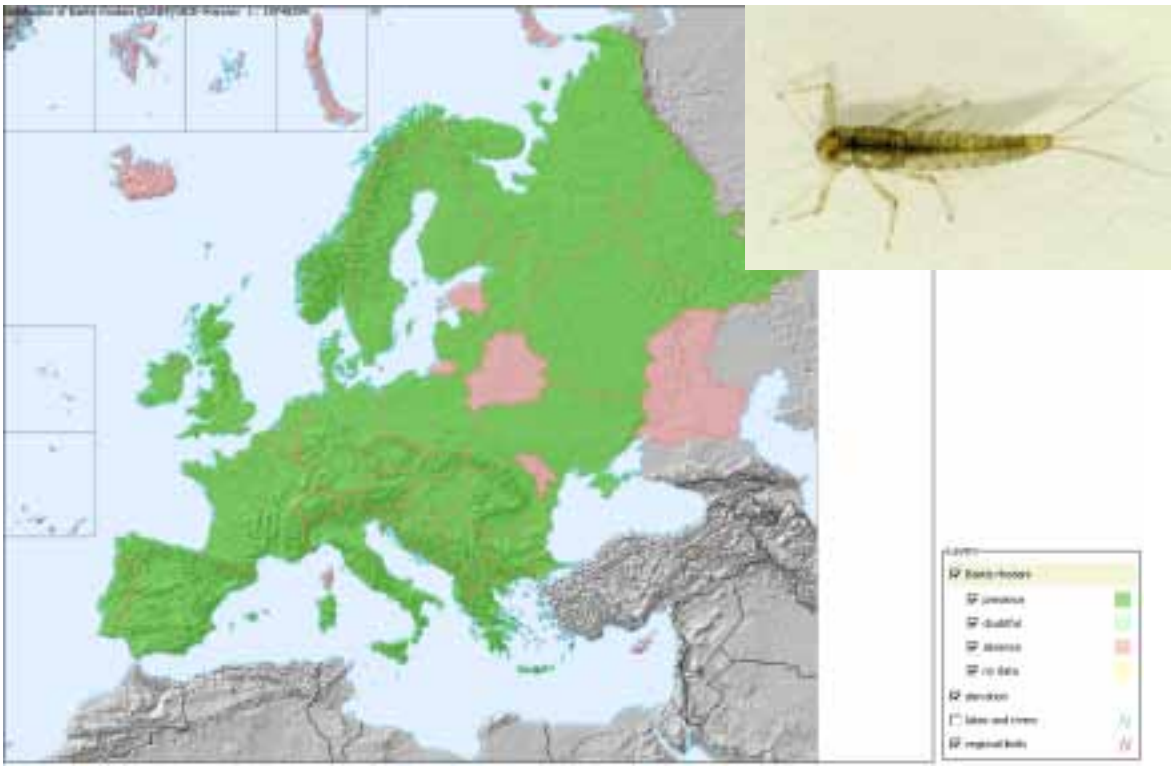

Figure 1. *Geographical distribution of the mayfly* Baetis rhodani *in Europe. This is an example of a widely distributed freshwater species. The species is recorded in all countries participating in the ICP Waters intercalibration with the exception of Estonia. Map after Fauna Europaea Web Service, http://www.faunaeur.org, Photo: Arne Fjellheim*

2.2 Identification

To minimise possible faults, the following procedure have been used in preparing the test samples:

- The participating country has first identified the source material for the test samples. Two of us have verified the identification of the species/taxa as far as possible without damaging the individuals.

– The content of the two test samples for each laboratory, with respect to species and numbers, is listed in a table. Two persons control that the correct number and species is placed in the test samples according to the list.

2.3 Damages of the material

The quality of the test material may be reduced during handling and shipping. Taxonomically important parts of the body, as gills, legs, cerci, mouthparts etc., can be lost or destroyed in actions connected with identification, sample composition and transportation. Contamination of larvae may also occur during these processes as well as during the identification work at the participating laboratories. All mentioned possibilities for faults could influence on the results of the identifications and disturb the results in a negative way.

2.4 Evaluation

The results of the tests are sent to the laboratories for eventual comments before publishing the report. In this way we can remove taxonomical biases, for example misidentified or destroyed test material. In cases of disagreement, material may be sent back to the programme subcentre for control. This procedure may act educational for both parts.

For calculation of faults (in percent), we must take into account possible destructions of the material as mentioned above. Further, a wrong identification of a species is one fault even if the sample contains many individuals of the species. We encourage the participants to give comments on matters that may impede the identification. For example may misidentification of species, in cases where important taxonomic characters have been destroyed, be neglected if this is pointed out by the participants.

We have discriminated between "short coming" identification, probably due to damaged material, and virtual fault (wrong species – or genus name). Due to this, some subjective evaluations of the results have to be made. The percent of faults is therefore not always the exact calculated percent of faults, but can be a modified value where some "expert judgement" is taken into account.

It is also of interest to know how many individuals that have been identified of the total number in the sample. This is named *% identified*. A low percent means that many individuals are not identified and will consequently reduce the value of the taxonomic work.

Available material for making test samples varies. The number of individuals and number of species delivered will therefore differ. Normally each laboratory gets between 50 and 100 individual species in the two samples. Samples with low diversity will be easier to handle than samples with high diversity, see Appendix tables. This should also be kept in mind when the results are evaluated. On the other hand, small samples should be avoided as only a few misidentifications could result in a low score.

We have calculated the quality assurance index, Qi, for important groups of invertebrates as well as the mean index for each participant. The Qi integrates the separate levels of the identifications as follows:

Qi = % correct species/10 * % correct genus/10 * % identified individuals/100

Qi will be a number between 0 and 100. 100 is the highest score that can be obtained. A score ≥80 is regarded as good taxonomical work.

2.5 Test of the subcentre

The ICP Waters subcentre in Bergen is tested each second year with the help from Sweden. The Swedish University of Agricultural Sciences in Uppsala prepares and evaluates the test of the subcentre. Methodology and implementation is otherwise identical to the other tests.

3. Results and discussion

Three laboratories participated in the intercalibration of invertebrates in 2012 (Appendix A). The content of species in the test samples delivered – and the results of the identification by the different laboratories are shown in Appendix Tables 1 – 3.

3.1 Mayflies

The identification of mayflies (Ephemeroptera) was generally very good (Figure 2, Appendix Table 1-3). Laboratory 1 and 2 identified the mayflies without faults. The results from laboratory 3 were acceptable. The Qi was calculated to 100, 100 and 82 for laboratories 1, 2, and 3 respectively. This indicates high quality of work.

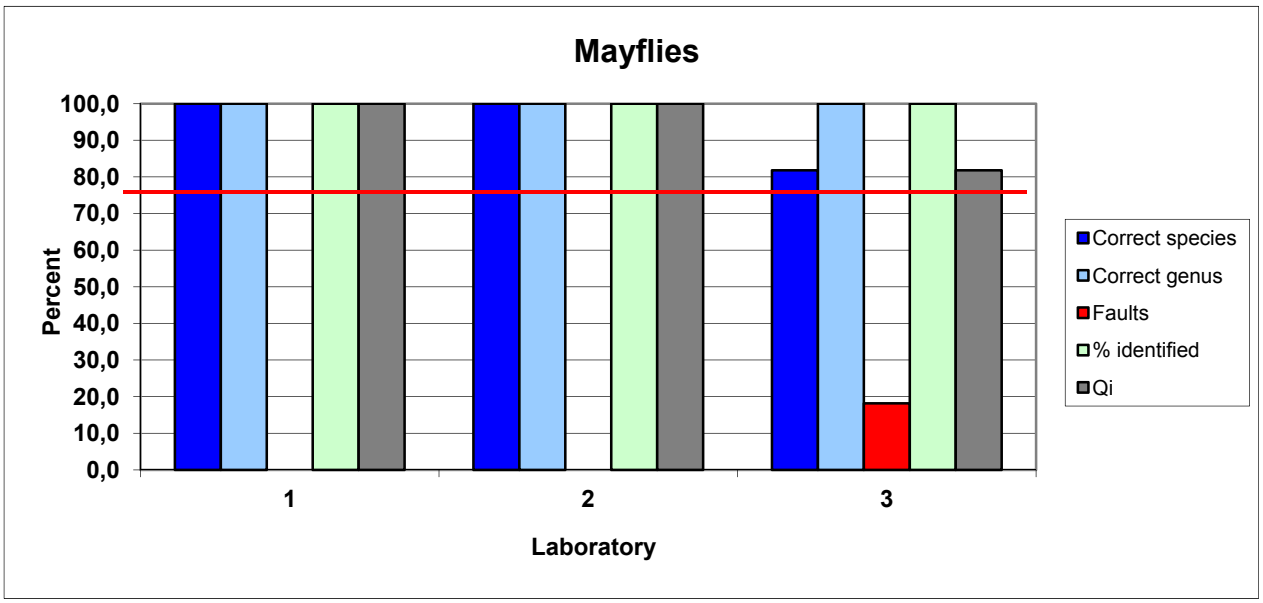

Figure 2. *Results of the identification of mayflies.*

3.2 Stoneflies

The identification of the stoneflies is presented in Figure 3 and Appendix tables 1 – 3. The results are regarded as very good, and show a good taxonomical knowledge of the group. The Qi was calculated to 88, 95 and 94 for laboratories 1, 2 and 3, respectively, well above the limit of acceptance.

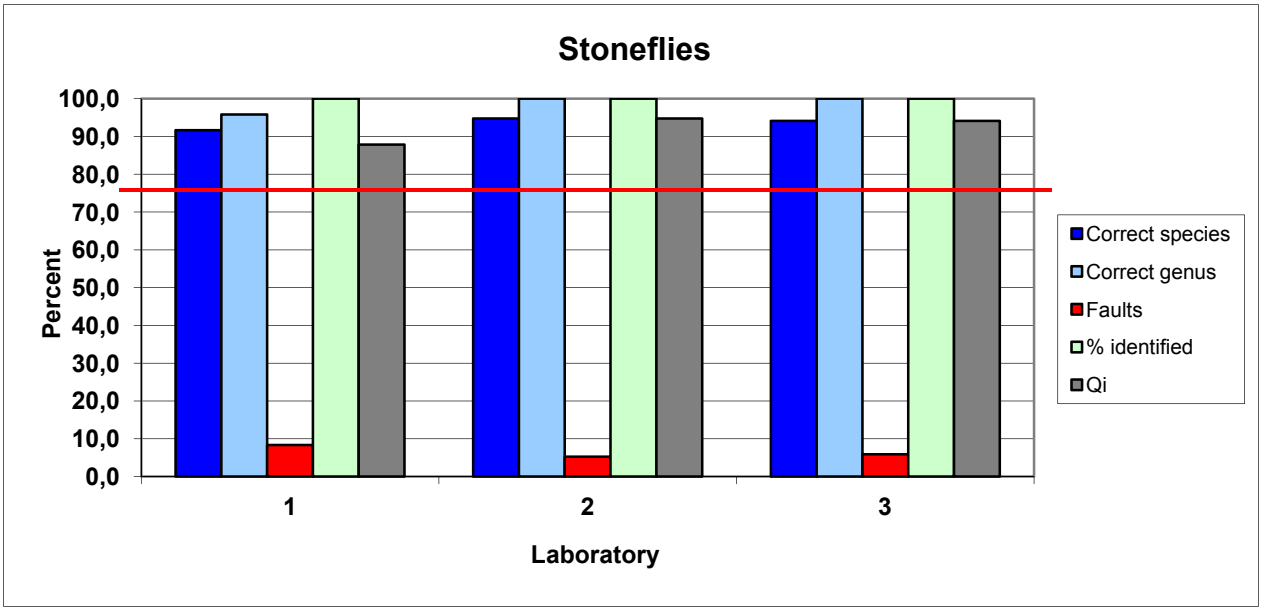

Figure 3. *Results of the identification of stoneflies.*

3.3 Caddisflies

The identification of caddisflies (Trichoptera) is presented in Figure 4 and Appendix tables 1 – 3. The quality of the identification was very good for all laboratories, Qi values being 94, 95 and 96, for participants 1, 2 and 3, respectively.

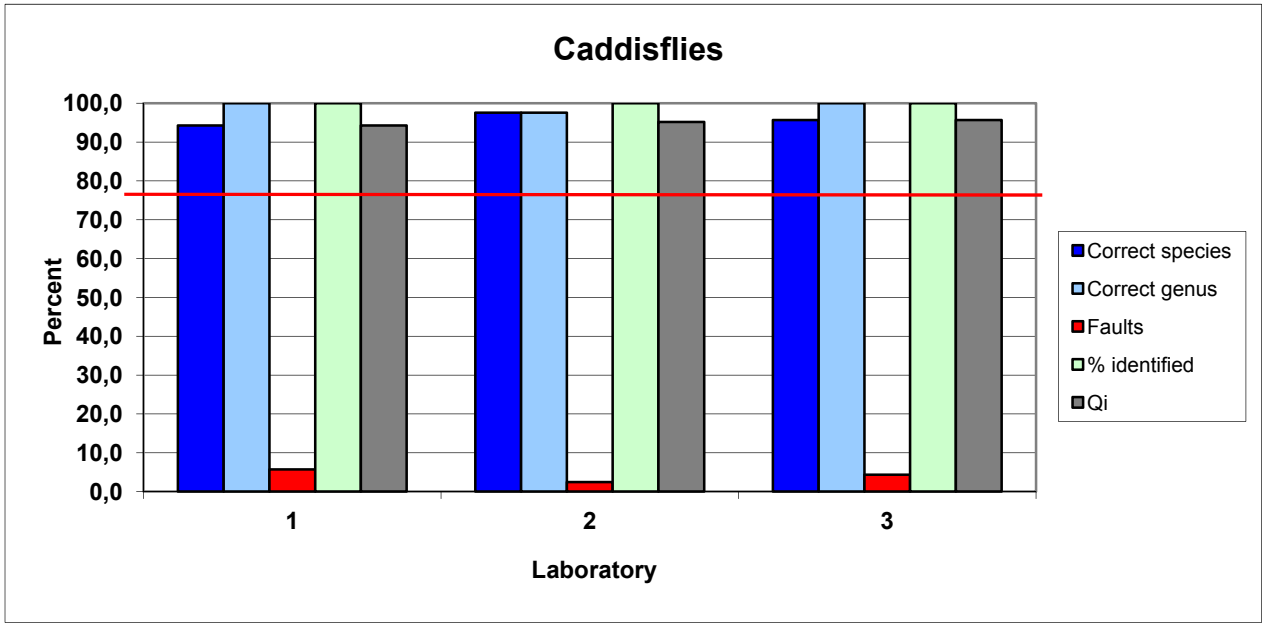

Figure 4. *Results of the identification of caddisflies.*

3.4 Other groups

In this intercalibration we have included water beetles (Coleoptera), larger crustaceans (Malacostraca), leeches (Hirudinea), molluscs (Gastropoda), alder-flies (Megaloptera), Diptera etc. Both larvae and imagines have been included for some of the groups. Leeches, molluscs and larger crustaceans are sensitive to acid water and important for the evaluation of acidification. The tolerance of the invertebrates among Coleoptera, Megaloptera, Diptera etc. is little known, but generally they are regarded as tolerant to acidic water and consequently have low importance for evaluation of acidity indices. However, all species will be important for invertebrate community analysis. Figure 5 and Appendix tables 1 – 3 shows the results of the identification of these groups. The identifications made by laboratory 2 were perfect with no faults. The quality of laboratory 1 and 3 was also very good. The latter identified the species correctly but failed to identify one individ of Diptera. The Qi score was 96, 100 and 97, for participants 1, 2 and 3, respectively. This indicates excellent work.

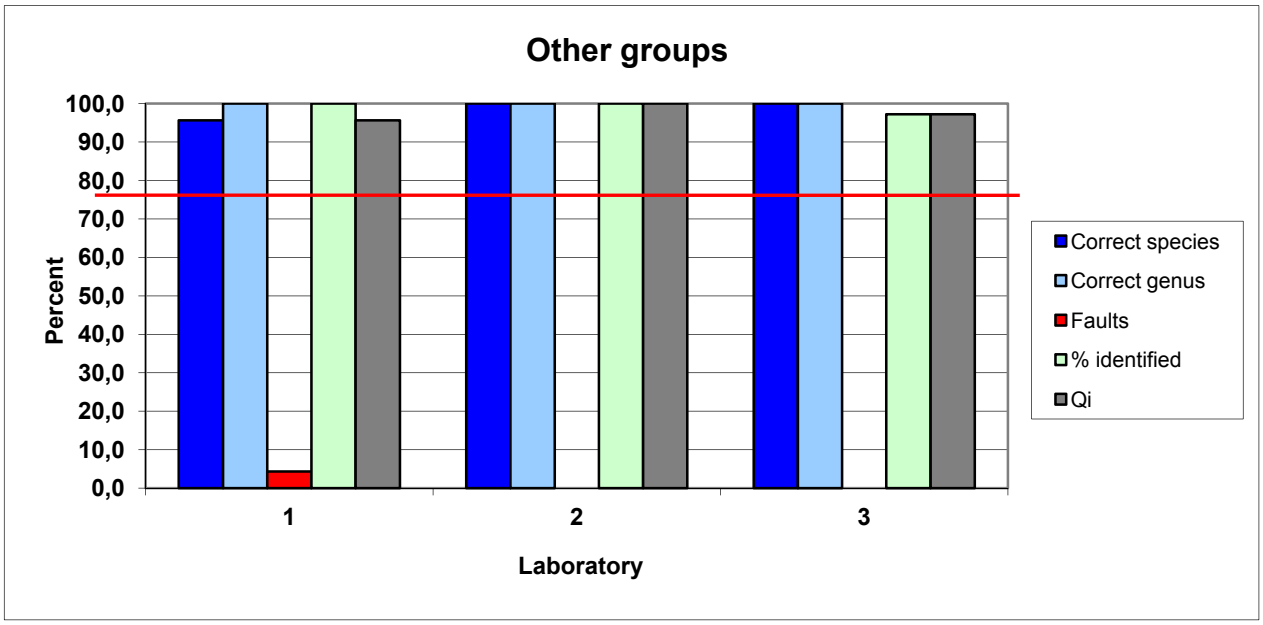

Figure 5. *Results of the identification of miscellaneous groups*

3.5 Total number of species in the sample

There were generally low discrepancy between the number of individuals put into the samples and the reported number of larvae. A total of 295 individual specimens were sent to the different laboratories. Of these 99.7 percent were reported back to the programme sub-centre.

4. Evaluation/conclusion

The laboratories generally identified a high portion of the total number of species in the test samples. Shortcoming identification was low and indicated good taxonomic skills by the participants. The mean skill of identifying species, genus and Qi score per laboratory is shown in Figure 6. Laboratory 1 to 3 got a mean Qi score of 94, 97 and 92 respectively. All tests were characterized as excellent taxonomic work. The biological intercalibration is important for harmonising biological material/databases and will be of high value in programmes where community analyses is in focus or where the ecological status should be stated.

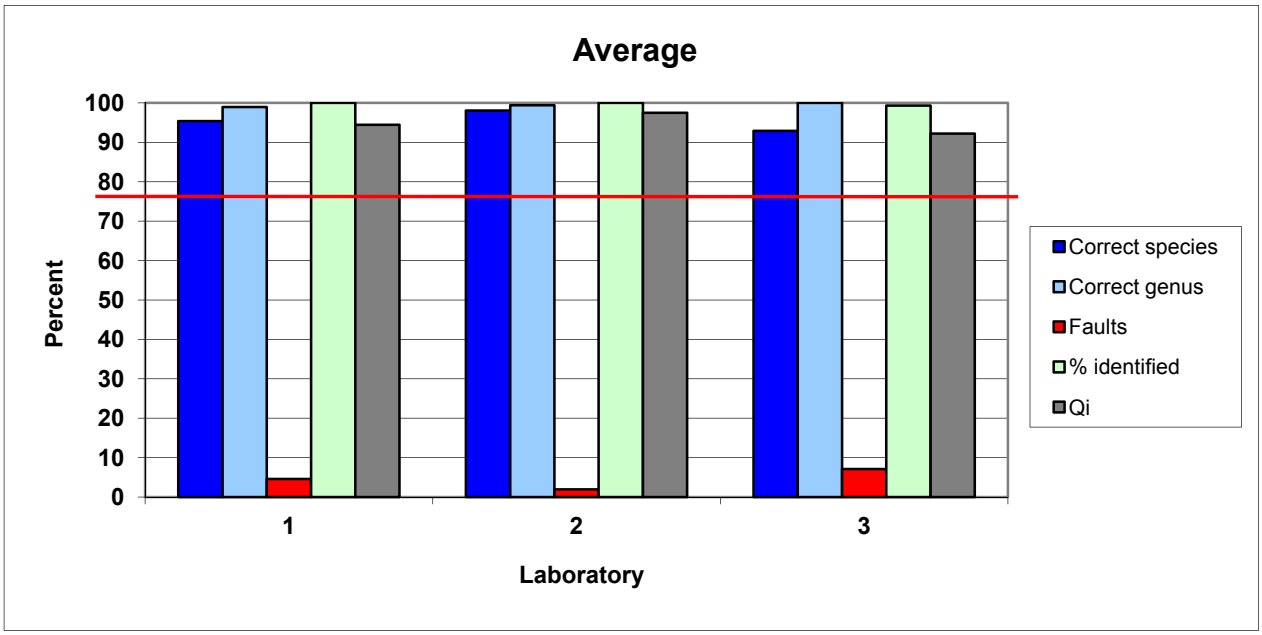

Figure 6. *Mean skill in percent of identifying species and genus and mean Qi for each laboratory.*

None of the participants did misidentifications that could result in a wrong acidity index, based on the Raddum score (Raddum et al., 1988).

The biological intercalibration under the ICP Waters programme was the first regular test aiming to test taxonomic skills of identifying benthic invertebrates. Today, similar tests are run by the the North American Benthological Society (http://www.nabstcp.com) and by the Natural History museum, London (Identification Qualifications – IdQ test). The invertebrate groups covered in the latter test are those used in the BMWP water quality score system (Armitage et al., 1983) and include groups used for monitoring freshwater environments under the EU water framework directive (Schartau et al. 2008).

5. References

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Appendix A. Responsible laboratories

Each participating laboratory is identified by a number, which is identical with table number. Laboratories participating in the intercalibration of invertebrates in 2011 and their code numbers are:

1. Swedish University of Agricultural Sciences, Dept. of Environmental Assessment, P.O. Box 7050, S-75007 Uppsala, **Sweden.** Responsible taxonomists: Lars Erikson and Magda-Lena Wiklund
2. Uni Research AS, P.O.box 7810, N-5020 Bergen, Norway. Responsible taxonomists: Torunn Landås and Arne Johannessen
3. Institute for Environmental Studies, Faculty of Science, Charles University, Prague, **Czech Republic.** Responsible taxonomist: Dr. Evzen Stuzlik

Appendix table 1. Identified species/genus in sample 1 and 2 by Laboratory 1

	Sample 1		Sample 2	
Taxa:	**Delivered**	**Identified**	**Delivered**	**Identified**
Ephemeroptera				
Baetis rhodani	1	1	1	1
Caenis horaria	1	1	1	1
Caenis luctuosa	1	1	1	1
Ephemera danica			1	1
Ephemera vulgata	1	1		
Heptagenia dalecarlica			1	1
Heptagenia sulphurea			1	1
Leptophlebia marginata	1	1		
Nigrobaetis digitatus	1	1	1	1
Nigrobaetis niger	1	1	1	1
Seratella ignita	1	1		
Plecoptera				
Amphinemura borealis	1	1	1	1
Amphinemura sulcicollis				1
Amphinemura standfussi			1	
Arcynopteryx compacta	1	1		
Capnia bifrons	1	1	1	1
Diura nanseni	1	1	1	1
Isoperla grammatica	1	1	1	1
Leuctra nigra	1	1	1	1
Nemoura avicularis	1	1	1	1
Nemoura cinerea	1	1	1	1
Nemoura sp.		1		1
Nemurella pictetii	1		1	
Protonemura meyeri	1	1	1	1
Siphlonurus burmeisteri	1	1	1	1
Taeniopteryx nebulosa	1	1	1	1
Trichoptera				
Agapetus ochripes	1		1	1
Agapetus sp.		1		
Arctopsyche ladogensis			1	1
Ceraclea annulicornis	1	1		
Ceratopsyche silfveni	1	1	1	1
Cheumatopsyche lepida			1	1
Chimarra marginata	1	1	1	1
Cyrnus flavidus	1	1	1	1
Cyrnus trimaculatus			1	1
Glyphotaelius pellucidus	1	1		
Hydropsyche angustipennis			1	2
Hydropsyche pellucidula			1	1
Hydropsyche siltalai	1	1	1	
Ironoquia dubia			1	1
Lepidostoma hirtum	1	1	1	1
Micrasema gelidum	1	1		
Molannodes tinctus			1	1
Neureclipsis bimaculata	1	1	1	1

	Sample 1		Sample 2	
Taxa:	**Delivered**	**Identified**	**Delivered**	**Identified**
Philopotamus montanus	1	1	1	1
Polycentropus flavomaculatus			1	
Polycentropus irroratus			1	2
Rhyacophila nubila	1	1		
Ryacophila fasciata			1	1
Sericostoma personatum			1	1
Setodes argentipunctellus	1	1		
Tinodes waeneri	1	1	1	1
Wormaldia subnigra			1	1
Megaloptera				
Sialis lutaria	1	2	1	1
Sialis sordida	1			
Malacostraca				
Asellus aquaticus	1	1		
Gammarus pulex			1	1
Hirudinea				
Glossophonia complanata	1	1		
Helobdella stagnalis			1	1
Gastropoda				
Acroloxus lacustris	1	1	1	1
Bithynia tentaculata			1	1
Gyraulus acronicus	1	1	1	1
Potamopyrgus antipodarum			1	1
Corixidae				
Callicorixa praeusta	1	1		
Aphelocheirus aestivalis	1	1		
Coleoptera				
Elmis aenea			1	1
Elodes sp.	1	1	1	1
Hydraena gracilis	1	1	1	1
Limnius volckmari	1	1	1	1
Orectochilus villosus			1	1

Appendix table 2. Identified species/genus in sample 1 and 2 by Laboratory 2

	Sample 1		Sample 2	
Taxa:	**Delivered**	**Identified**	**Delivered**	**Identified**
Ephemeroptera				
Alainites muticus	1	1	1	1
Baetis rhodani	1	1	1	1
Caenis horaria			2	2
Caenis luctuosa	2	2		
Cloeon dipterum/inscriptum			1	1
Ephemerella aurivilli	1	1	1	1
Heptagenia dalecarlica	1	1	2	2
Heptagenia sulphurea	2	2		
Leptophlebia marginata			1	1
Leptophlebia vespertina	2	2	1	1
Nigrobaetis niger			1	1
Plecoptera				
Amphinemura borealis	2	2		
Amphinemura standfussi				1
Amphinemura sulcicollis			2	1
Brachyptera risi	1	1	1	1
Capnia atra			1	1
Diura nanseni	1	1	1	1
Isoperla grammatica	1	1	1	1
Leuctra fusca	1	1		
Nemoura avicularis	1	1		
Nemurella pictetii			1	1
Protonemura meyeri	1	1	1	1
Siphonoperla burmeisteri	1	1		
Taeniopteryx nebulosa	1	1	1	1
Trichoptera				
Agapetus ochripes	1	1		
Agrypnia obsoleta			1	1
Athripsodes aterimus	1	1		
Athripsodes cinereus	1	1	1	1
Ceratopsyche silfvenii			1	1
Chimarra marginata	1	1		
Cyrnus insolutus			1	1
Cyrnus trimaculatus	1	1		
Ecclisopteryx dalecarlica	1	1		
Ecnomus tenellus			1	1
Holocentropus dubius	2	2		
Hydropsyche angustipennis	1	1		
Hydropsyche pellucidula			1	1
Hydropsyche siltalai	1	1	1	1
Lepidostoma hirtum	1	1	1	1
Molanna angustata			1	1
Molannodes tinctus			1	1
Mystacides azurea	1	1		
Mystacides longicornis			1	1
Nemotaulius punctatolineatus			1	1

	Sample 1		Sample 2	
Taxa:	**Delivered**	**Identified**	**Delivered**	**Identified**
Neureclipsis bimaculata	1	1	1	1
Oecetis testacea			1	1
Philopotamus montanus	1	1		
Phryganea bipunctata	1	1		
Cyrnus flavidus			1	
Plectrocnemia conspersa				1
Polycentropus flavomaculatus	1	1	1	1
Potamophylax cingulatus			1	1
Potamophylax latipennis	1	1		
Rhyacophila fasciata	2	2		
Rhyacophila nubila			2	2
Sericostoma personatum	1	1		
Trianodes bicolor	1	1		
Wormaldia subnigra			1	1
Hirudinea:				
Erpobdella octoculata	1	1		
Helobdella stagnalis			1	1
Diptera:				
Antocha vitripennis			1	1
Dicranota sp.	1	1		
Coleoptera:				
Elmis aenea	2	2	2	2
Limnius volckmari	2	2		
Olimnius tuberculatus			2	2
Orectochilus villosus			1	1
Megaloptera:				
Sialis fuliginosa			1	1
Sialis lutaria	1	1		
Gastropoda:				
Hippeutis complanata			1	1
Radix balthica			1	1
Valvata piscinalis			1	1
Odonata:				
Erythromma najas	1	1		
Pyrrhosoma nymphula			1	1

Appendix table 3. Identified species/genus in sample 1 and 2 by Laboratory 3

	Sample 1		Sample 2	
Taxa:	**Delivered**	**Identified**	**Delivered**	**Identified**
Ephemeroptera				
Baetis buceratus	1			
Baetis vernus		1		
Baetis fuscatus			1	
Baetis rhodani				1
Caenis horaria			1	1
Caenis luctuosa	1	1	1	1
Caenis macrura	1	1		
Caenis robusta			1	1
Ecdynurus dispar	1		1	1
Ecdynurus cf. *dispar*				1
Ecdynurus aurantiacus		1		
Ecdyonurus torrentis			1	
Epeorus sylvicola	1	1		
Ephemera danica	1	1	1	1
Ephemera vulgata	1	1	1	1
Ephemerella mucronata	1	1	1	1
Habrophlebia fusca			1	
Habrophlebia lauta				1
Leptophlebia marginata	1			
Leptophlebia vespertina		1	1	
Leptophlebia sp.				1
Seratella ignita			1	1
Siphlonurus armatus	1	1		
Plecoptera				
Amphinemura borealis	1	1	1	1
Diura bicaudata	1	1	1	1
Isoperla oxylepsis	1	1	1	1
Leuctra niger			1	1
Leuctra rauscheri			1	
Leuctra handlirschi				1
Nemoura avicularis	1	1		
Nemoura cinerea	1	1	1	1
Nemurella pictetii	1	1		
Protonemura auberti			1	1
Protonemura montana	1	1	1	
Protonemura sp. juv				1
Taeniopteryx nebulosa	1	1	1	1
Trichoptera				
Anabolia sp.			1	1
Beraeodes minutus	1	1		
Brachycentrus subnubilus	1	1	1	1
Chaetopteryx villosa	1	1		
Cyrnus flavidus			1	2
Drusus annulatus	1	1		
Holocentropus dubius	1	1		
Hydropsyche sp.			1	1

	Sample 1		Sample 2	
Taxa:	**Delivered**	**Identified**	**Delivered**	**Identified**
Hydropsyche pellucidula	1	1	1	1
Leptocerus tineiformis			1	
Leptocerus interruptus				1
Limnephilus coenosus			1	1
Molannodes tinctus	1	1		
Mystacides azurea	1	1	1	1
Oligotricha striata			1	1
Phryganea bipunctata	1	1		
Polycentrupus flavomaculatus	1	1	1	1
Pseudopsilopteryx zimmeri			1	1
Ptilocolepus granulatus	1	1		
Gastropoda				
Ancylus fluviatilis	1	1		
Teodoxus fluviatilis	1	1	1	1
Odonata				
Aeshna cyanea			1	1
Libellula quadrimaculata	1	1		
Coleoptera				
Deronectes latus	1	1		
Elmis aenea			1	1
Hydroporus palustris			1	1
Nebrioporus assimilis	1	1	1	1
Megaloptera				
Sialis fuliginosa	1	1		
Sialis lutaria			1	1
Malacostraca				
Asellus aquaticus	1	1	1	1
Gammarus fossarum	1	1		
Gammarus lacustris			1	1
Heteroptera				
Aphelocheirus aestivalis			1	1
Glaenocorisa propinquua	1	1		
Notonecta glauca	1	1		
Sigara fossarum			1	1
Sigara nigrolineata			1	1
Sigara semistriata	1	1	1	1
Velia caprai	1	1		
Diptera				
Atherix ibis	1	1	1	1
Chaoborus cf.obscuripes			1	1
Chelifera sp.	1		1	1
Dicranota sp.	1	1		
Wiedemannia sp.			1	1
Hirudinea				
Erpobdella octoculata			1	1
Erpobdella vilnensis	1	1	1	1
Bivalvia				
Pisidium casertanum			1	1

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