

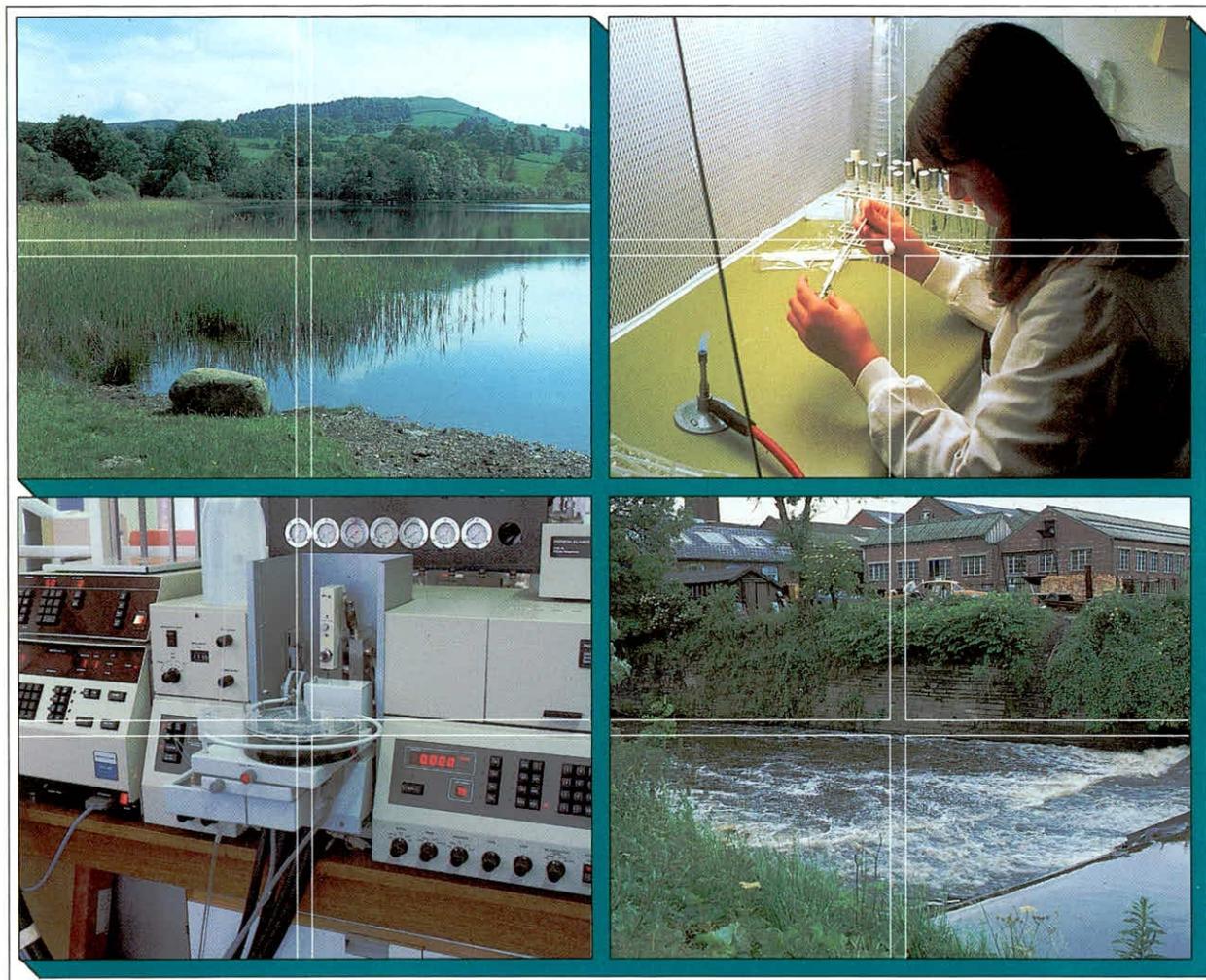
An audit of performance in the analysis of biological samples in 1995 Tweed River Purification Board

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Project Leader:
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1. INTRODUCTION

The 1995 General Quality Assessment (GQA) Survey included the sampling of aquatic macro-invertebrates for biological assessment of river quality throughout the United Kingdom. In England and Wales the survey was undertaken by the National Rivers Authority (NRA), the River Purification Boards (RPBs) sampled in Scotland, the Industrial Research and Technology Unit (IRTU) undertook the work in Northern Ireland and the Government Laboratory covered the Isle of Man (IOM).

The majority of sites surveyed were sampled in two seasons, spring and autumn. Standard collection procedures were used and the sampling strategy was compatible with RIVPACS (River InVertebrate Prediction And Classification System), a computer model developed by the Institute of Freshwater Ecology (IFE).

Samples were sorted for the families of macro-invertebrates included in the Biological Monitoring Working Party (BMWP) system. Taxa present were recorded on site data sheets. Although attempts had been made to standardise sample processing and recording techniques, these did vary somewhat from region to region.

In order to undertake this massive programme of fieldwork and sample processing, a large number of staff, many of whom were relatively inexperienced, were employed by the surveying agencies. In view of the number of staff involved and the variability of sample processing techniques, it was recognised that a quality assurance exercise was necessary to minimise and quantify errors. Each laboratory appointed at least one experienced analyst to act as an internal analytical quality control (AQC) checker. For most agencies, these checkers re-sorted about 10% of the laboratory's samples, those samples chosen for re-sorting being selected at random. In addition, IFE was contracted to undertake an independent, external audit of the quality of the laboratory analysis of biological samples for each NRA region, each RPB, IRTU and IOM. This commission was consistent with the audit performed by IFE for the National River Quality Survey in 1990 and for the routine biological monitoring of river sites each year between 1991 and 1994. This audit was originally intended as a measure of the quality of the AQC analyses and is termed the main audit or AQC audit. The data collected for the 1995 GQA Survey was not adjusted for errors identified by either of the quality assurance procedures. Therefore the NRA contracted IFE to subject their samples to a further audit of the primary analysis (the primary audit) to provide an independent assessment of the quality of the Survey data .

This report presents the results of the 6 samples audited for Tweed RPB. For samples that have been subjected to an internal AQC check, the results measure the performance of the AQC analyst rather than that of the primary analyst.

2. SAMPLE SELECTION

Samples for audit were selected internally by each of the agencies being monitored. The number of samples selected for audit varied between the different agencies and the biologists processing these samples had no prior knowledge of which samples were to be audited. Some agencies only sent to IFE samples that had been processed twice. Others adopted a random selection process, whereby some samples had been analysed just once and some had been re-sorted. The manner of sample selection, which biologists would be monitored and the number of audit samples from each season, were left to the discretion of the agency, within the limits of the total number of samples that IFE was contracted to audit.

3. SAMPLE PROCESSING

The normal protocol for NRA, RPB, IRTU and IOM biologists was to sort their samples within the laboratory and to select examples of each scoring taxon within the BMWP system. In most cases, the invertebrates were placed in a vial of preservative (4% formaldehyde solution or 70% industrial alcohol) and the BMWP taxa were listed on a data sheet. The vial of animals and the sorted material were then returned to the sample container and preservative added. Thus, each sample available to IFE for audit should have included:

- i) a data sheet containing a list of the BMWP families found in the sample.
- ii) a vial containing representatives from each family.
- iii) the preserved sample.

When these three elements were present, the sequence of operations at IFE was as follows:

- a) The remainder of the sample was sorted, without reference to the data sheet or to the vial of animals, and the BMWP families identified.
- b) The families contained within the vial were identified.
- c) A comparison was made between the listing of families and those found in the sample by IFE.
- d) A comparison was made between the listing of families and those identified from the vial by IFE.
- e) "Losses" or "gains" from the original listing of families were noted. In the case of "gains", each additional family was identified, where possible, to species level, in order to clarify any specific repetitive errors. Single representatives of a "gained" taxon were noted as such.
- f) An error code, selected from a list on the result sheet, was assigned by the IFE auditor for each "loss" or "gain".

Occasionally a sample did not include a vial containing representative examples of the families listed on the data sheet, while some arrived with the vial damaged in transit such that the representative specimens were no longer separated. For these samples, only operations a), c), e) and f) above were appropriate.

Several directives were issued to IFE relating to the treatment of BMWP taxa. Every taxon recorded on the data sheet must be supported by a voucher specimen of that family in the vial (or, for very large specimens, left in the sample). The only exceptions to this rule were the native crayfish, *Austropotamobius pallipes*, the medicinal leech, *Hirudo medicinalis* and the pearl mussel, *Margaritifera margaritifera* (which does not belong to a BMWP family), all of which are protected species. Where possible, IFE gave the benefit of doubt to the analyst in cases of the "loss" of Planariidae, specimens of which have been known to disintegrate in preservative. Animals deemed to have been dead at the time of sampling, cast insect skins, pupal exuviae and empty mollusc shells were to be excluded from the listing of families present. Isolated posterior ends of "living" specimens were not acceptable as records of a taxon. In these cases, thorax plus abdomen was deemed acceptable but abdomen only was deemed unacceptable. Terrestrial representatives of BMWP scoring families were also to be excluded from the audit. For this reason, Clambidae, Chrysomelidae and Curculionidae, which appear in the BMWP list, were excluded for the purposes of the audit since most representatives of these families are, at best, only semi-aquatic. Trichopteran pupae, although not routinely identified by many biologists, were to be included in the listing of families.

4. REPORTING

The results of each sample audit were recorded on a standard report form and sent to the Regional Biologist. Copies of these report forms are presented in the Appendix. For audit samples where a vial of animals was included, the comparison between the listing of families and the taxa found in the vial by IFE was shown in the section of the report form headed "VIAL". Discrepancies could be due to carelessness, misidentifications or errors in completing the data sheet listing the families present. Families not on the listing but found by IFE in the remainder of the sample were entered in the section of the report form headed "SAMPLE" under "Additional BMWP taxa found by IFE". Taxa recorded here represent families missed by the analyst(s) on sorting the sample. When the families listed as "losses" in the first section of the report form were compared with the full list of families recorded in the sample by IFE, some apparent losses from the vial were offset by the presence of those families in the remainder of the sample. These taxa were therefore listed both as "losses" from the vial and as "gains" from the sample and were neither a net loss nor a net gain. In these cases, the families were marked with an asterisk in both boxes. Such errors are noted as "omissions".

Species identifications, state of development (eg adult or larval coleopterans) and the presence of a single representative of a family within the remainder of the sample were recorded in the centre section of the report form under "species name".

IFE was asked to interpret each error to provide a possible cause. An error code, selected from a list of options at the foot of each result sheet, was entered against each taxon in the column headed "Presumed cause of error".

For those samples in which the vial of animals was damaged or missing, the "VIAL" sections of the report form were not applicable (N/a). Families not on the list but present in the sample were entered in the section under "SAMPLE" : "Additional taxa" as before. Families recorded on the list but not found by IFE were indicated in the section above this. If the vial of animals was retained by the sorter, entries in this box could include the sole representative of a family which was removed, a family seen at the site which escaped or was released (without mention being made on the data sheet), inaccurate identification, the wrong family box being ticked on the data sheet or the family being present in the sample but missed by IFE.

The final section of the result sheet summarises the audit, giving details of the numbers of "losses", "gains" and "omissions", together with the net effects on BMWP score and the number of scoring taxa.

5. RESULTS

The results of the audit for Tweed RPB are summarised Table 1. Table 2 displays the statistics of these regional audit results centered around the target of acceptability of no more than two missed taxa per sample. These data are presented for each analyst and for the RPB as a whole. Table 3 compares these results for each RPB laboratory. Table 4 presents data for Tweed RPB for the net effects of the audit on the BMWP score and number of taxa. This table is again based on the target of no more than two missed taxa per sample. The figure of 13 for an acceptable underestimate of BMWP score is based on twice the average score of all taxa in the BMWP listing (excluding Clambidae, Chrysomelidae and Curculionidae, which are excluded from the audit). This average score is 6.57. Table 5 compares this BMWP data for each RPB laboratory. Table 6 lists the taxa missed in sorting by Tweed RPB's analysts in the 1995 audit and Table 7 lists all such taxa for the entire 1995 audit (Primary and AQC Audits for NRA regions and Main Audit for other organisations) for the whole of the UK.

6. ACKNOWLEDGEMENTS

Grateful thanks to John Murray-Bligh of NRA Thames Region, who provided an invaluable service in the development and implementation of improved methodology and in providing helpful advice throughout.

Table 1. The 6 samples audited for Tweed RPB

River	Site	Analyst	Losses	Gains	Omissions
SPRING					
Yarrow Water	Craig Douglas	DC	0	0	0
Tweed	Lowood Bridge	DC	0	1	0
Slitrig Water	Hummelknows Bridge	JWC	0	1	0
AUTUMN					
Whiteadder Water	Playhaugh Bridge	DC	0	0	0
Tweed	Dawyck	DC	0	0	1
Ettrick Water	Brockhoperig	JWC	1	1	0

Table 2. Statistics of 1995 audit results for Tweed RPB

Analyst	n	Mean gains	Standard error	No.samples >2 gains	% samples >2 gains	Highest no. gains	Mean errors (l+g+o)	Standard error
DC	4	0.50	0.29	0	0.00	1	0.50	0.29
JWC	2	1.00	0.00	0	0.00	1	1.50	0.50
Tweed RPB	6	0.67	0.21	0	0.00	1	1.00	0.26

Table 3. Summary of errors found in 1995 audit for each RPB

RPB	n	Mean gains	Standard error	No.samples >2 gains	% samples >2 gains	Highest no. gains	Mean errors (l+g+o)	Standard error
Clyde	42	1.00	0.16	5	11.90	3	1.21	0.18
Forth	40	1.75	0.29	11	27.50	8	2.03	0.33
Highland	12	2.42	0.58	4	33.33	6	2.75	0.55
North East	34	2.06	0.31	11	32.35	6	2.21	0.32
Solway	7	1.00	0.38	0	0.00	2	1.00	0.38
Tay	10	1.20	0.29	1	10.00	3	1.40	0.31
Tweed	6	0.67	0.21	0	0.00	1	1.00	0.26
All RPBs	151	1.55	0.13	32	21.19	8	1.77	0.14

Table 4. Net effects of the Audit on BMWP score and number of scoring taxa

Analyst	n	Mean net effect on BMWP score	% of samples underestimated by score >13	Maximum underestimate of BMWP score	Mean net effect on no. of taxa	% of samples underestimated by >2 taxa	Maximum underestimate of no. of taxa
DC	4	3.00	0.00	7	0.50	0.00	1
JWC	2	0.50	0.00	5	0.50	0.00	1
Tweed RPB	6	2.17	0.00	7	0.50	0.00	1

Table 5. Summary of errors found in 1995 audit for each RPB

RPB	n	Mean net effect on BMWP score	% of samples underestimated by score >13	Maximum underestimate of BMWP score	Mean net effect on no. of taxa	% of samples underestimated by >2 taxa	Maximum underestimate of no. of taxa
Clyde	42	6.50	16.67	27	0.83	11.90	3
Forth	40	10.55	27.50	63	1.57	22.50	8
Highland	12	14.83	50.00	47	2.17	25.00	6
North East	34	12.76	38.24	41	2.00	32.35	6
Solway	7	7.14	28.57	20	1.00	0.00	2
Tay	10	6.70	20.00	15	1.10	10.00	3
Tweed	6	2.17	0.00	7	0.50	0.00	1
All RPBs	151	9.52	27.15	63	1.41	19.21	8

Table 6. Taxa missed by Tweed RPB's analysts

Family	n	% of Tweed RPB's missed taxa in audit	% of missed taxa in audit for all RPBs
Simuliidae	1	25.00	3.14
Caenidae	1	25.00	4.04
Gyrinidae	1	25.00	0.90
Hydroptilidae	1	25.00	3.14

Table 7. Missed taxa for all samples in 1995 audit

Family	n	% of all missed taxa in 1995 audit
Hydrophilidae (incl. Hydraenidae)	68	5.97
Hydroptilidae	59	5.18
Sphaeriidae	52	4.57
Hydrobiidae (incl. Bithyniidae)	50	4.39
Planariidae (incl. Dugesiidae)	46	4.04
Caenidae	40	3.51
Elmidae	39	3.42
Leptoceridae	39	3.42
Psychomyiidae (incl. Ecnomidae)	39	3.42
Lymnaeidae	33	2.90
Simuliidae	32	2.81
Nemouridae	31	2.72
Limnephilidae	30	2.63
Planorbidae	29	2.55
Haliplidae	28	2.46
Tipulidae	25	2.19
Baetidae	23	2.02
Glossiphoniidae	22	1.93
Goeridae	22	1.93
Leptophlebiidae	22	1.93
Dytiscidae (incl. Noteridae)	21	1.84
Ephemerellidae	20	1.76
Valvatidae	20	1.76
Hydropsychidae	18	1.58
Ancylidae (incl. Acroloxidae)	16	1.40
Asellidae	16	1.40
Leuctridae	16	1.40
Piscicolidae	16	1.40
Rhyacophilidae (incl. Glossosomatidae)	16	1.40
Scirtidae	16	1.40
Sericostomatidae	15	1.32
Gyrinidae	14	1.23
Erpobdellidae	13	1.14
Lepidostomatidae	13	1.14
Polycentropodidae	13	1.14
Chloroperlidae	11	0.97
Odontoceridae	11	0.97
Dendrocoelidae	10	0.88
Heptageniidae	10	0.88

Table 7. (cont.)

Family	n	% of all missed taxa in 1995 audit
Gammaridae (incl. Crangonyctidae)	9	0.79
Taeniopterygidae	9	0.79
Hydrometridae	8	0.70
Oligochaeta	8	0.70
Physidae	7	0.61
Chironomidae	6	0.53
Coenagriidae	6	0.53
Perlidae	6	0.53
Brachycentridae	5	0.44
Calopterygidae	5	0.44
Perlodidae	5	0.44
Sialidae	5	0.44
Beraeidae	4	0.35
Corixidae	4	0.35
Corophiidae	4	0.35
Ephemeridae	4	0.35
Unionidae	4	0.35
Capniidae	3	0.26
Dryopidae	3	0.26
Gerridae	3	0.26
Libellulidae	3	0.26
Siphonuridae	3	0.26
Aphelocheiridae	2	0.18
Neritidae	2	0.18
Platycnemididae	2	0.18
Aeshnidae	1	0.09
Cordulegasteridae	1	0.09
Notonectidae	1	0.09
Philopotamidae	1	0.09
Viviparidae	1	0.09

APPENDIX

Results of individual sample audits

EXTERNAL AUDIT OF BIOLOGICAL SAMPLES

REGION: Tweed RPB

LABORATORY: Galashiels

DATE: 20.4.95

WATER-COURSE: Yarrow Water

PRIMARY ANALYST: DC

AQC ANALYST:

SITE: Craig Douglas

CODE: 95B0293

SORT/AQC METHOD: Not known

RESULTS OF MAIN AUDIT

Family name	Species name (where appropriate)	Presumed cause of error (see footnotes)
-------------	-------------------------------------	---

VIAL

BMWP taxa not found by IFE

None

Additional BMWP taxa found by IFE

None

SAMPLE

BMWP taxa not found by IFE (For samples where vial is broken or absent)

N/a

Additional BMWP taxa found by IFE

Simuliidae

Simulium argyreatum 1 only

9

SUMMARY OF AUDIT

LOSSES 0

GAINS 1

OMISSIONS: 0

NET EFFECTS:

ON BMWP SCORE 5

ON NO. OF TAXA 1

1 No representative of family in vial

2 Alternative terrestrial specimen in vial

3 Posterior end only in vial

4 Empty shell or case or cast skin in vial

5 Specimen dead at time of sampling

6 Taxon in vial but not recorded

7 Mis-identification

8 Typographical error - wrong box ticked

9 Taxon missed in sorting

10 Unexplained error

11 Taxon added in internal AQC

12 Recorded taxon that was rejected by AQC analyst

Omission (*) = Recorded, not in vial but found by IFE in sample (no net loss or gain)

EXTERNAL AUDIT OF BIOLOGICAL SAMPLES

REGION: Tweed RPB

LABORATORY: Galashiels

DATE: 3.5.95

WATER-
COURSE: Tweed

PRIMARY
ANALYST: DC

AQC
ANALYST:

SITE: Lowood Bridge

CODE: 95B0369

SORT/AQC
METHOD: Not known

RESULTS OF MAIN AUDIT

Family name	Species name (where appropriate)	Presumed cause of error (see footnotes)
-------------	-------------------------------------	---

VIAL

BMWP taxa not found by IFE

None

Additional BMWP taxa found by IFE

None

SAMPLE

BMWP taxa not found by IFE (For samples where vial is broken or absent)

N/a

Additional BMWP taxa found by IFE

Caenidae

Caenis rivulorum 1 only

9

SUMMARY OF AUDIT

LOSSES 0

GAINS 1

OMISSIONS: 0

NET EFFECTS:

ON BMWP SCORE 7

ON NO. OF TAXA 1

1 No representative of family in vial

2 Alternative terrestrial specimen in vial

3 Posterior end only in vial

4 Empty shell or case or cast skin in vial

5 Specimen dead at time of sampling

6 Taxon in vial but not recorded

7 Mis-identification

8 Typographical error - wrong box ticked

9 Taxon missed in sorting

10 Unexplained error

11 Taxon added in internal AQC

12 Recorded taxon that was rejected by AQC analyst

Omission (*) = Recorded, not in vial but found by IFE in sample (no net loss or gain)

EXTERNAL AUDIT OF BIOLOGICAL SAMPLES

REGION: Tweed RPB

LABORATORY: Galashiels

DATE: 24.4.95

WATER-
COURSE: Slitrig Water

PRIMARY
ANALYST: JWC

AQC
ANALYST:

SITE: Hummelknows Bridge

CODE: 95B0324

SORT/AQC
METHOD: Not known

RESULTS OF MAIN AUDIT

Family name	Species name (where appropriate)	Presumed cause of error (see footnotes)
-------------	-------------------------------------	---

VIAL

BMWP taxa not found by IFE

None

Additional BMWP taxa found by IFE

None

SAMPLE

BMWP taxa not found by IFE (For samples where vial is broken or absent)

N/a

Additional BMWP taxa found by IFE

Gyrinidae	Orectochilus villosus (larva) 1 only	9
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SUMMARY OF AUDIT

LOSSES 0

GAINS 1

OMISSIONS: 0

NET EFFECTS:

ON BMWP SCORE 5
ON NO. OF TAXA 1

- 1 No representative of family in vial
- 2 Alternative terrestrial specimen in vial
- 3 Posterior end only in vial
- 4 Empty shell or case or cast skin in vial

- 5 Specimen dead at time of sampling
- 6 Taxon in vial but not recorded
- 7 Mis-identification
- 8 Typographical error - wrong box ticked

- 9 Taxon missed in sorting
- 10 Unexplained error
- 11 Taxon added in internal AQC
- 12 Recorded taxon that was rejected by AQC analyst

Omission (*) = Recorded, not in vial but found by IFE in sample (no net loss or gain)

EXTERNAL AUDIT OF BIOLOGICAL SAMPLES

REGION: Tweed RPB

LABORATORY: Galashiels

DATE: 30.10.95

WATER-

PRIMARY

AQC

COURSE: Whiteadder Water

ANALYST: DC

ANALYST:

SITE: Playhaugh Bridge

CODE: 95B1123

SORT/AQC

METHOD: Preserved

RESULTS OF MAIN AUDIT

Family name	Species name (where appropriate)	Presumed cause of error (see footnotes)
-------------	-------------------------------------	---

VIAL

BMWP taxa not found by IFE

None

Additional BMWP taxa found by IFE

None

SAMPLE

BMWP taxa not found by IFE (For samples where vial is broken or absent)

N/a

Additional BMWP taxa found by IFE

None

SUMMARY OF AUDIT

LOSSES 0

GAINS 0

OMISSIONS: 0

NET EFFECTS:

ON BMWP SCORE 0

ON NO. OF TAXA 0

1 No representative of family in vial

5 Specimen dead at time of sampling

9 Taxon missed in sorting

2 Alternative terrestrial specimen in vial

6 Taxon in vial but not recorded

10 Unexplained error

3 Posterior end only in vial

7 Mis-identification

11 Taxon added in internal AQC

4 Empty shell or case or cast skin in vial

8 Typographical error - wrong box ticked

12 Recorded taxon that was rejected by AQC analyst

Omission (*) = Recorded, not in vial but found by IFE in sample (no net loss or gain)

EXTERNAL AUDIT OF BIOLOGICAL SAMPLES

REGION: Tweed RPB

LABORATORY: Galashiels

DATE: 9.11.95

WATER-COURSE: Tweed

PRIMARY ANALYST: DC

AQC ANALYST:

SITE: Dawyck

CODE: 95B1204

SORT/AQC METHOD: Preserved

RESULTS OF MAIN AUDIT

Family name	Species name (where appropriate)	Presumed cause of error (see footnotes)
-------------	-------------------------------------	---

VIAL

BMWP taxa not found by IFE

Lepidostomatidae *

1

Additional BMWP taxa found by IFE

None

SAMPLE

BMWP taxa not found by IFE (For samples where vial is broken or absent)

N/a

Additional BMWP taxa found by IFE

Lepidostomatidae *

Lepidostoma hirtum

1

SUMMARY OF AUDIT

LOSSES 0

GAINS 0

OMISSIONS: 1

NET EFFECTS:

ON BMWP SCORE 0

ON NO. OF TAXA 0

1 No representative of family in vial

2 Alternative terrestrial specimen in vial

3 Posterior end only in vial

4 Empty shell or case or cast skin in vial

5 Specimen dead at time of sampling

6 Taxon in vial but not recorded

7 Mis-identification

8 Typographical error - wrong box ticked

9 Taxon missed in sorting

10 Unexplained error

11 Taxon added in internal AQC

12 Recorded taxon that was rejected by AQC analyst

Omission (*) = Recorded, not in vial but found by IFE in sample (no net loss or gain)

EXTERNAL AUDIT OF BIOLOGICAL SAMPLES

REGION: Tweed RPB

LABORATORY: Galashiels

DATE: 21.9.95

WATER-COURSE: Ettrick Water

PRIMARY ANALYST: JWC

AQC ANALYST:

SITE: Brockhoperig

CODE: 95B0964

SORT/AQC METHOD: Preserved

RESULTS OF MAIN AUDIT

Family name	Species name (where appropriate)	Presumed cause of error (see footnotes)
-------------	-------------------------------------	---

VIAL

BMWP taxa not found by IFE

Lepidostomatidae

1

Additional BMWP taxa found by IFE

None

SAMPLE

BMWP taxa not found by IFE

(For samples where vial is broken or absent)

N/a

Additional BMWP taxa found by IFE

Hydroptilidae

Hydroptila sp. 1 only

9

SUMMARY OF AUDIT

LOSSES 1

GAINS 1

OMISSIONS: 0

NET EFFECTS:

ON BMWP SCORE -4

ON NO. OF TAXA 0

1 No representative of family in vial

2 Alternative terrestrial specimen in vial

3 Posterior end only in vial

4 Empty shell or case or cast skin in vial

5 Specimen dead at time of sampling

6 Taxon in vial but not recorded

7 Mis-identification

8 Typographical error - wrong box ticked

9 Taxon missed in sorting

10 Unexplained error

11 Taxon added in internal AQC

12 Recorded taxon that was rejected by AQC analyst

Omission (*) = Recorded, not in vial but found by IFE in sample (no net loss or gain)