ICES COOPERATIVE RESEARCH REPORT

RAPPORT DES RECHERCHES COLLECTIVES

NO. 207

REPORT ON THE RESULTS OF THE ICES/IOC/OSPARCOM INTERCOMPARISON PROGRAMME ON THE ANALYSIS OF CHLOROBIPHENYLS IN MARINE MEDIA-STEP 2

and

THE INTERCOMPARISON PROGRAMME ON THE ANALYSIS OF PAHs IN MARINE MEDIA-STAGE 1

https://doi.org/10.17895/ices.pub.7985

ISBN 978-87-7482-686-6

ISSN 2707-7144

International Council for the Exploration of the Sea
Palægade 2-4, DK-1261 Copenhagen K
DENMARK

January 1995

TABLE OF CONTENTS

Section	**Page**

RESULTS OF THE INTERCOMPARISON PROGRAMME ON THE ANALYSIS OF PAHs IN MARINE MEDIA–STAGE 1

REPORT ON THE RESULTS OF THE ICES/IOC/OSPARCOM INTERCOMPARISON PROGRAMME ON THE ANALYSIS OF CHLOROBIPHENYL CONGENERS IN MARINE MEDIA - STEP 2

J. de Boer[1], L. Reutergårdh[2], J. van der Meer[3], J.A. Calder[4]

1) Netherlands Institute for Fisheries Research (RIVO)
P.O. Box 68
1970 AB IJmuiden, The Netherlands

2) Swedish Environmental Protection Agency
Special Analytical Laboratory
S-17185 Solna, Sweden

3) Netherlands Institute for Sea Research
P.O. Box 59
1790 AB Den Burg
Texel, The Netherlands

4) NOAA, Office of Oceanic and Atmospheric Research
1335 East-West Highway, SSMC1
Silver Spring, MD 20910, USA

SUMMARY

This report gives an account of the second step of the ICES/IOC/OSPARCOM Intercomparison Programme on the Analysis of Chlorobiphenyls (CBs) in Marine Media. Results were received from 58 laboratories in sixteen countries. In this exercise, CB Nos. 28, 31, 52, 101, 105, 118, 138, 153, 156 and 180 were analysed in an unknown CB solution, a cleaned seal blubber extract, and a cleaned sediment extract. An extra test, which included the analysis of an unknown CB solution vs. a supplied known solution, was carried out by laboratories that produced outlying results in the first exercise and laboratories who participated for the first time in this second exercise.

Standard deviations for reproducibility of 1.16 - 1.17 for the standard solution, 1.20 - 1.33 for the cleaned seal blubber extract, and 1.31 - 1.56 for the cleaned sediment extract were found for all laboratories without outliers for CBs 52, 101, 118, 138, 153 and 180. The results for CBs 28, 31, 105 and 156 showed larger standard deviations.

The two major difficulties identified for participants in this exercise were the preparation of calibration solutions and the chromatographic separation. It is suggested that in any further exercise results will only be accepted when based on calibration solutions prepared from solids of known purity and analysed with two columns with different stationary phases and minimum lengths of 50 m and maximum internal diameters of 0.25 mm.

The third step of this exercise should be undertaken only after the laboratories have been given time to revise their methods and install the necessary gas chromatographic columns. A repeat exercise using an unknown CB solution is suggested, along with analysis of an uncleaned seal blubber extract. The common quality level for the analysis of sediment extracts is considered to be far from what is required. Therefore, it is suggested to wait with further action until improvement is obtained with the standard solution and the uncleaned seal blubber extract.

To five laboratories, advice was given not to participate in further steps of this exercise, because their calibration procedures and chromatographic methods first need a drastic revision. Eleven laboratories that did not produce any results, even three months after the deadline, should be excluded from further participation until the present learning exercise is concluded.

1 INTRODUCTION

In 1988 the International Council for the Exploration of the Sea (ICES), the Intergovernmental Oceanographic Commission (IOC), and the Oslo and Paris Commissions (OSPARCOM) initiated a stepwise intercomparison programme to improve the analysis of chlorinated biphenyls in marine media.

The objectives of this exercise were:

1) to determine the variation in the results of the analyses of chlorobiphenyl congeners among participating laboratories;

2) to identify sources of error which cause this variation; and

3) to reduce the variation in the results by means of a learning process through a series of step-by-step intercomparison exercises.

The exercises were to be conducted in four steps:

1) analysis of standard solutions;

2) check of participants' own ability to prepare standards and to analyse cleaned extracts;

3) analysis of uncleaned extracts; and

4) analysis of samples of marine organisms and sediments.

The first step of this study was completed in 1990 (de Boer *et al.*, 1992). In the results of this first step, the variation coefficients were 10-13% among the majority of the participants which were adequate to permit the organization of the second step. In addition to this second step, an extra exercise was designed for laboratories who produced outlying results during the first step. Although the agreement for the programme was that participants could only commence participation at the beginning of the programme and not during subsequent steps, owing to the many requests from interested laboratories to join this study after the first step, a later group of institutes was permitted to join the exercise owing to their laboratory activities in marine monitoring programmes. These laboratories were requested to demonstrate their ability in analysing chlorobiphenyls (CBs) in the same extra exercise that was designed for the group of outliers from the first step. In this report, the results of both exercises are given.

The aim of the second step was to check the ability of participants to prepare their own CB solutions and to compare the results of the participants' analyses of a cleaned seal blubber extract and/or a cleaned marine sediment extract.

The extra test was, in fact, a short version of the first step of this study. Participants were asked to optimize their instruments, to prepare linearity graphs of the electron capture detector, and to analyse an unknown CB solution against a supplied calibration solution.

The exercise was coordinated by J. de Boer (Netherlands Institute for Fisheries Research, IJmuiden), who was assisted by J.A. Calder (NOAA, Washington) for the evaluation of the sediment data and by L. Reutergårdh (Swedish EPA, Solna) for the evaluation of the seal blubber data. The statistical evaluation of all data was performed by J. van der Meer (Netherlands Institute for Sea Research, Texel) on behalf of the ICES Working Group on Statistical Aspects of Trend Monitoring.

2 PARTICIPANTS

The names and addresses of all participants in this second step are given in Table 1. The laboratories marked with an asterisk participated in the extra exercise, repeating the first step (Nos. <75) or as a new participant (Nos. >75). The West Vancouver Laboratory from Canada (No. 8), the Instituto de Ciencias del Mar y Limnología from Mexico (No. 76) and the Institute of Marine Environmental Protection from China (No. 77) decided not to participate further after the first step. The National Environmental Research Institute (No. 9) and the National Agency of Environmental Protection (No. 10), both from Denmark, merged into one institute, the National Environmental Research Institute, which participated in this second step under No. 10.

The Laboratoire Municipal de Bordeaux from France (No. 17), the Alfred Wegener Institüt für Polar und Meeresforschung from Germany (No. 26), the Institut für Küsten- und Binnenfischerei from Germany (No. 30), and the Instituto Hydrografico from Portugal (No. 44) withdrew from this exercise after having received the ampoules for the second step. The Fisheries Research Centre from Ireland (No. 34), the Woods Hole Oceanographic Institution from the USA (No. 64), the Institute of Applied Geophysics from the (then) USSR (No. 74), the Southern Californian Coastal Water Research Project from the USA (No. 82) and the SMHI Oceanographic Laboratory from Sweden (No. 86) received samples but did not return results.

Results from 55 laboratories were used in the statistical evaluation. Results from the Rijksstation voor Zeevisserij, Ostend, Belgium, the Free University, Brussels, Belgium, and the Icelandic Fisheries Research Laboratories, Reykjavik, Iceland, were received, but were too late to be included in the statistical evaluation. All laboratories analysed the unknown CB solution A, 49 laboratories analysed the seal blubber extract B and 46 laboratories analysed the sediment extract C.

3 MATERIALS AND METHODS

The guidelines for the conduct of this second step are attached to this report as Annex A. A small change from the CB solution used in the first step was made, namely, CB189, 2,3,4,5,3',4',5'-heptachlorobiphenyl, was replaced by CB156, 2,3,4,5,3',4'-hexachlorobiphenyl. The latter CB is of greater toxicological importance. The following CBs were used for the unknown solution A in this exercise:

CB28	-	2,4,4' - trichlorobiphenyl
CB31	-	2,5,4' - trichlorobiphenyl
CB52	-	2,5,2',5' - tetrachlorobiphenyl
CB101	-	2,4,5,2',5' - pentachlorobiphenyl
CB105	-	2,3,4,3',4' - pentachlorobiphenyl
CB118	-	2,4,5,3',4' - pentachlorobiphenyl
CB138	-	2,3,4,2',4',5' - hexachlorobiphenyl
CB153	-	2,4,5,2',4',5' - hexachlorobiphenyl
CB156	-	2,3,4,5,3',4' - hexachlorobiphenyl
CB180	-	2,3,4,5,2',4',5' - heptachlorobiphenyl

In this report CB138 is actually the sum of CB138, CB163 and a third compound (Larsen and Riego, 1990; de Boer and Dao, 1991), known to coelute in environmental samples.

The standard solution A, the internal standard solution D, and the blank E were prepared and ampouled at RIVO. Standards of CBs 28, 52, 101, 105, 118, 138, 153 and 180 were obtained from the Community Bureau of Reference Materials (BCR), Brussels. Except for CB105 these CBs were all certified standards with a purity of >99%. The purity of CB105 was 99%. CBs 31 and 156 were obtained from Promochem, Wesel, Germany. The minimum purity of these two CBs was given as 98%. The two internal standards tetrachloronaphthalene (TCN) and octachloronaphthalene (OCN) were obtained from Promochem, Wesel, Germany.

Iso-octane, nanograde quality, was used as a solvent in all solutions. Before flame sealing, the ampoules were chilled in liquid nitrogen to prevent the formation of carbon particles during flame sealing.

The seal blubber extract, solution B, was prepared by the Swedish Environmental Protection Agency, Solna, Sweden. The seal sample was a common seal (*Phoca vitulina*), one-year-old male, weight 40 kg, length 128 cm, drowned in a fishing net on 5 June 1990 off western Iceland and was supplied by the Icelandic Fisheries Laboratories. The fat content of the blubber was 91.4%. The fat was dissolved in n-hexane at a concentration of 100 mg/ml and treated with H_2SO_4 (1:2). The hexane was extracted, iso-octane was added, and this solution was concentrated 5 to 1 under nitrogen at 50°C. Ampoules were filled with 2.5 ml of this solution, which corresponded to 1250 mg fat.

The sediment extract, solution C, was prepared by the Rijkswaterstaat, Tidal Waters Division, Groningen, The Netherlands. The sediment sample was taken from the Wadden Sea and consisted of a fine material with a particle size of <63 μm (100%). The material was dried for 48 hours at 35°C. After grinding and homogenizing, the material was extracted with 25% acetone in hexane. This extract was concentrated on a Kuderna Danish apparatus in portions of 20 ml, corresponding to 10 g of sediment, down to 5 ml and subsequently under nitrogen to 1 ml. These extracts were transferred to a column of 4 g desulphurizing agent and then to a 2 g silica column. After elution with 20 ml hexane, 2 ml iso-octane was added and the extracts were concentrated to 1 ml. These 50 solutions of 1 ml each were transferred to a 1 litre measuring flask and dissolved in iso-octane. Each ampoule was filled with 4 ml from this 1 litre solution, corresponding to 2 g of sediment. The ampoules were filled at RIVO in IJmuiden.

The weight of each ampoule was written on the ampoule. Participants were requested to check these weights upon receipt of the ampoules and report any change in weight, so that spare ampoules could be sent immediately if necessary.

Participants were requested to prepare their own CB standard and to analyse this standard with the solutions A, B, C and E.

Participants were further requested to use two gas chromatographic columns with different stationary phases and to inject the solutions three times on each column. The best estimate for each CB (in general, the lowest value) was to be reported together with an indication of on which column this value was produced. The choice of using peak heights instead of peak areas was based on the experience gained in the first step. Table 2 lists the different GC columns and conditions which have been used. A number of suggestions were given for the optimization of the instruments. Participants were requested to optimize their instruments according to the guidelines for the first step. Emphasis was placed on checking the linearity of the electron capture detector, the use of appropriate concentrations or dilution of standards and samples, and the use of a multi-level calibration where necessary. All ampoules were dispatched to the participants during the last week of August 1990.

3.1 Extra Test

The guidelines for the extra test are attached to this report as Annex B. The solutions for this test were prepared and ampouled at RIVO. This extra test was a brief repetition of the first step. The following samples were supplied: 1 ampoule X with the five CBs 28, 31, 118, 153 and 180 in known concentrations, 1 ampoule Y with the same CBs in unknown concentrations, 1 ampoule D with the internal standards TCN and OCN, and 1 ampoule E with only iso-octane as the blank. Participants were requested to analyse the Y-solution, using the X-solution as a standard. CB149 was added to the Y-solution to make the analysis of CB118 more realistic in comparison with environmental samples. Linearity graphs were required to be constructed for CB28 and CB153. Two columns were to be used with different stationary phases and both results were to be reported. Only peak heights were used. An overview of the columns and conditions used is given in Table 3.

3.2 Homogeneity Test

A number of ampoules were tested for homogeneity at RIVO. The results are shown in Table 4. The number of ampoules of each type of sample used was different due to the limited availability of material. The stock solutions together with the ampoules (except B) were analysed to check for any systematic errors between the CB concentrations in the stock solutions and those in the ampoules.

The results show coefficients of variation below 10%, which is an acceptable analytical error. A few exceptions above 10% can be attributed to analytical difficulties, namely, difficult separation and low concentrations, e.g., CBs 28/31 in B and CB105 and CB156 in B and C. No systematic deviations were observed between the CB concentrations in the stock solutions and the mean CB concentrations in the ampoules.

The cleanliness of the ampoules was checked by filling four ampoules with 5 ml iso-octane, shaking, 9-fold concentration of the iso-octane, and testing for the presence of CBs. The results are given in Table 4g. Only for CBs 28, 101 and 180 were detectable amounts found in all ampoules and for most other CBs in one ampoule at the level of 0.5-1 pg/μl. The influence of these levels on the final results is negligible.

Table 5 shows the target values of the CBs in the ampoules A and Y, as they were prepared by weighing.

4 RESULTS

Results from 55 laboratories were returned in time to be used for the statistical evaluation. Results were accepted until 8 January 1991. A few laboratories returned their results later than this date (see Section 4.3). With 65 sets of ampoules sent out, this means 85% participation. For the extra test 12 laboratories out of 18 returned results, which is a return rate of 67%. All data delivered for this second step of the exercise are given in Tables 6, 7 and 8. The data delivered for the extra test are shown in Table 9.

4.1 Remarks from Participants

Many participants confirmed that they had carried out a recent linearity test of their ECDs. Some participants observed instability of OCN when using polar columns or retention gaps. Also, some participants reported impurities present in the D-solution.

Lab. No. 7 Used only one GC column (DB-5). A more polar column is not routinely used in this laboratory. A PCB-standard solution from the National Research Council of Canada (NRC) was used instead of its own weighed standard of CB crystals. A linear calibration curve was used with repeated injections of a single standard instead of a high and a low standard.

Lab. No. 10 Reported a fast deterioration of the DB-1701 column, in contrast to what was observed during normal routine analysis. Nevertheless, some results from this column were used. Both columns were installed in one injector with a 1 m 530 μm pre-column and a glass T-split. The total injection volume was 2 μl.

Lab. No. 12 Injected 3 μl by mistake for all samples. Columns with diameters <0.32 mm were not available. Therefore, CBs 149 and 118 and CBs 28 and 31 could not be separated. CB185 (2,3,4,5,6,2',5'-heptachlorobiphenyl) was used as an internal standard. The NRC standard solution was used instead of solid standards, which were not available.

Lab. No. 13 Reported high bleeding of the DB-17 columns. Therefore, only DB-5 results were reported.

Lab. No. 14 Injected all samples manually. Two impurity peaks were found in the OCN/TCN solution.

Lab. No. 16 Used only one GC column (CP-Sil 8).

Lab. No. 36 Reported that both columns were attached to one injector, which is a normal practice in this laboratory.

Lab. No. 37 Used two columns attached to one injector. Glasswool was inserted in the injector to improve the response of the CBs. CB143 (2,3,4,5,2',6'-hexachlorobiphenyl) was used as an internal standard.

Lab. No. 40 Reported that CB31 was not available. The CB31 concentration of ampoule X was used as a standard.

Lab. No. 47 Used a PTV injector.

Lab. No. 49 Used CB198 (2,3,4,5,6,2',3',5'-octachlorobiphenyl) as an internal standard in A and B. For ampoule C no internal standard was used. A 50 m x 0.33 mm x 1 μm HP-5 column was preferred instead of a 50 m x 0.20 mm x 0.33 μm HP-5 column.

Lab. No. 50 Reported that two columns were connected to one injector via a 3m retention gap. Coelution of CB101 and CB90 (2,3,5,2',4'-pentachlorobiphenyl) and of CB28 and CB50 (2,4,6,2'-tetrachlorobiphenyl) were reported on both columns. Also CB156 was not fully separated on either column.

Lab. No. 52 Used DCBE 16 (dichlorobenzyl C_{16} alkyl ether) as an internal standard.

Lab. No. 53 Used a VG Trio-1 bench-top GC/MS for this exercise; negative chemical ionization with iso-butane as reagent gas was used. ECD results were reported later.

Lab. No. 54 Reported that the results of these two congeners were probably slightly high due to partial separation of CB28 and CB31.

Lab. No. 63 Used a split injection for solutions A and B and a splitless injection for solution C. All preparations were done on a weight-to-weight basis. The density of iso-octane (0.691 g/ml at 22°C) was used to convert to a weight-to-volume basis. Next to TCN and OCN, CB103 (2,4,6,2',5'-pentachlorobiphenyl) and CB198 (2,3,4,5,6,2',3',5'-octachlorobiphenyl) were used as internal standards. Only peak areas were used for calculation.

Lab. No. 66 Used only one GC column (CP-Sil 8) which is a normal practice in this laboratory. The peak heights of the internal standards TCN and OCN in the participant's own standard had to be corrected due to coelution with some CBs. CB88 (2,3,4,6,2'-pentachlorobiphenyl) is normally used as an internal standard. The results based on calculation without the use of an internal standard were preferred because of the observed variation in results based on internal standard calculation.

Lab. No. 69 Could not reach the recommended optimum gas velocity for helium with the 0.15 mm i.d. column because the maximum manometer pressure was restricted to 250 kPa. Nevertheless, with a run-time of 120 minutes the best separation for CBs was obtained.

Lab. No. 70 Used only one GC column (30 m DB-5). A standard solution of the National Institute of Standards and Technology (NIST) was used, except for CBs 28, 31 and 156, for which solid standards were available. Results for solution C could not be reported due to losses during concentration. An additional GC/MS analysis of solutions A and B was completed.

Lab. No. 72 Used only one GC column (50 m CP-Sil 8 CB). This laboratory suggested the use of CB88 (2,3,4,6,2'-pentachlorobiphenyl) as an internal standard in the future, although they used TCN and OCN for this exercise.

Lab. No. 78 Could not analyse solution C due to sensitivity problems.

Lab. No. 79 Did not use internal standards, because of evaporation losses.

Lab. No. 80 Did not use internal standards, because the retention time of OCN was too long. Only one GC column was used (50 m SE-54). Only indicative values for solution C were given, which were not used for the statistical evaluation.

Lab. No. 81 Used a VG Auto Spec high resolution GC/MS for this exercise. Because of optimization problems, the results for the extra test were suspected of not being as good as possible. The polar column was not available for the extra test.

Lab. No. 83 Used commercial CB standard solutions of Dr Ehrenstorfer, Germany.

Lab. No. 85 Reported some differences in results based on peak heights and peak areas. Only the results based on peak heights were used for this exercise.

Lab. No. 88 Could not separate CBs 28 and 31.

4.2 Remarks from the Coordinators

Lab. No. 2 Some negative peaks were observed in the CP-Sil 19 chromatograms.

Lab. No. 6 The use of longer columns or a reduction in the internal diameter is advised. No separation was obtained for CBs 28 and 31 on either column. Coelution of CB52 with other compounds was observed in solution C.

Lab. No. 7 Only one GC column was used. Standard solutions were used instead of solid standards, which may have led to deviations from the target values in solution A.

Lab. No. 10 This laboratory produced very acceptable chromatograms for all three solutions.

Lab. No. 11 Many negative peaks were observed in the B and C solutions on the SE-54 column. Insufficient separation of CBs 28 and 31.

Lab. No. 12 This laboratory used 0.32 mm i.d. columns, which caused insufficient separation. In combination with the commercial standard solutions, this led to poor results, especially for the Y solution. CB149 was identified in the Y solution, but the concentration given was not correct. Tailing peaks were observed on both columns.

Lab. No. 13 Only one 30 m x 0.32 mm i.d. column was used, which caused insufficient separation. Tailing peaks were observed, together with a high bleeding and a high background noise. Several peaks were observed in the blank. The chromatographic system did not appear to be very well equilibrated.

Lab. No. 14 The column length of 60 m only partly compensated for the loss of separation power of the 0.32 mm i.d. DB-5 columns. Several peaks were present in the blank.

Lab. No. 16 Only one 25 m x 0.32 mm i.d. CP-Sil 8 column was used. The restricted separation caused deviating results.

Lab. No. 18 Although the CP-Sil 8 column has the appropriate dimensions, the results were rather poor especially for the B and C solutions. This could result from a combination of errors in the preparation of the standard and chromatographic conditions that are not optimum. Several negative peaks were observed.

Lab. No. 21 This laboratory produced acceptable results.

Lab. No. 22 Presumably an error was made in the preparation of the CB180 standard. Insufficient sensitivity was obtained for solution C. No values were reported for CBs 31 and 156.

Lab. No. 23 Only one GC column (CP-Sil 8 CB) was used. Some tailing was observed in the chromatograms.

Lab. No. 24 Insufficient separation was obtained with the two columns which were used, particularly for the separation of CB118 and CB149 in the Y solution, which resulted in too high a value for CB118. A large solvent peak was present in the chromatograms of the DB-5 column. CBs 52, 101, 105, 138 and 156 were not analysed, which makes the contribution of this laboratory rather limited.

Lab. No. 25 Negative peaks were observed in the OV1-701 chromatograms.

Lab. No. 27 A high background with a negative drift was observed for the DB-1 column. The separation of CBs 28 and 31 was insufficient, which may have caused the deviating result for CB31 in A. The chromatograms indicated that poorer results might be expected than were finally reported. Lengthening of the columns or reduction of the internal diameter is advised.

Lab. No. 28 Tailing peaks were sometimes found in the DB-5 chromatograms. Errors in the preparation of the standard must have been made for CB31 and CB156. A high background was observed in chromatograms of both columns.

Lab. No. 29 The baseline was not always drawn correctly, which might have influenced the peak heights. Negative peaks were observed. The results for B were biased to the high side.

Lab. No. 33 The two 30 m x 0.25 mm i.d. GC columns did not separate CBs 28 and 31. In solution C, more coelution may have occurred. Not enough sensitivity was obtained to measure most CBs in C. Some negative peaks were observed.

Lab. No. 35 This laboratory produced acceptable results. A few negative peaks were observed which were not in conflict with the quantification.

Lab. No. 36 Very acceptable results were obtained for A and B, but not enough sensitivity was obtained for the analysis of solution C. Tailing of the OCN peak was observed.

Lab. No. 37 Very acceptable results were obtained for all three solutions. Some negative peaks were present in the B chromatograms.

Lab. No. 39 Although the chromatograms looked good and for the A solution acceptable results were found except for CB52, the results for most CBs in B and C seem to be about 50% too high. A calculation error could have been made.

Lab. No. 40 With the columns used it should be possible to obtain a better resolution. Some CBs in A were on the high side (weighing errors?) and also results for B and C were high. The results for the extra test were acceptable.

Lab. No. 43 Very acceptable results were obtained for all three solutions.

Lab. No. 45 Very acceptable results were obtained. Only the results for CB101 in B and for CBs 138 and 153 in C are on the high side. Some leading peaks were observed on the DB-5 column.

Lab. No. 46 Very acceptable results were reported for A and B.

Lab. No. 47 For most CBs errors must have been made in the preparation of standards, which is presumably the basic reason for the deviating results. Also separation needs improvement.

Lab. No. 48 The separation on the two columns could be improved. Presumably weighing errors for CBs 153 and 156 have been made. Only a few results in B and C were acceptable. The baseline correction was not appropriate in the B and C chromatograms.

Lab. No. 49 No chromatograms were returned. Although very acceptable results were obtained for the unknown solution A, the results for B and C were rather poor. The limited chromatographic performance, using only one 0.32 mm i.d. column, may be the reason.

Lab. No. 50 Presumably a weighing error for CB105 has been made. A few negative peaks were observed, which were not in conflict with the quantification.

Lab. No. 51 Although the chromatograms looked good, all results for the A solution were on the low side. This laboratory reported afterwards a correction of 20% for their A results, due to a calculation error.

Lab. No. 52 This laboratory initially returned results expressed in ng/g. Unfortunately, this was only discovered after the statistical evaluation had been conducted. Tables 6-8 now contain the correct results, which are very acceptable. Dividing these data by 0.694 gives the results which were used for the statistical evaluation.

Lab. No. 53 This laboratory produced GC/MS results. The results were rather poor for A as well as for C. Only one GC column was used. See also Section 4.3.

Lab. No. 54 Weighing errors for CB52 and CB101 may have been made. Many deviating results were found for B and C. Improvement of the chromatographic separation is suggested.

Lab. No. 57 Although 60 m columns compensated somewhat for the loss in resolution due to the 0.32 mm i.d., results for B and C still suffered due to insufficient separation. Some parts of the C chromatograms dropped under the baseline.

Lab. No. 58 Lengthening the columns or some reduction of the internal diameter could improve the results.

Lab. No. 59 No DB-1701 chromatograms were returned. Lengthening of the column or reduction of the internal diameter could improve the results.

Lab. No. 62 A commercial standard solution was used as CB standard. This is likely to be the cause of the rather deviating results. The data set based on Aroclor contained no better data. Some parts of the A and B chromatograms on the DB-17 column dropped under the baseline.

Lab. No. 63 This laboratory produced very acceptable results. It is a pity that peak areas were used for calculation, although for this exercise it was agreed to use peak heights. Some negative peaks were observed in all chromatograms.

Lab. No. 66 This laboratory used only one GC column. Values uncorrected for the internal standard were used for this exercise, as suggested by the laboratory. Acceptable data were obtained for A, but several deviations were found for B and C.

Lab. No. 67 Considerable noise and bleeding were observed in the chromatograms. Only one GC column was used with nitrogen as a carrier gas. Separation of CBs 118 and 149 and CBs 28 and 31 could not be obtained. The results for the extra test were insufficient. For A, B and C, several deviating results were obtained. Chromatographic conditions must be greatly improved.

Lab. No. 68 Acceptable results were obtained, although some improvement of the separation is recommended.

Lab. No. 69 It should be possible to obtain better results with the CP-Sil 8 column. Some bleeding was observed and the restricted separation obtained may have caused deviating values for B.

Lab. No. 70 Only one GC column was used. It is advised to use a longer column or to reduce the internal diameter to obtain a better separation. Some deviating results in A may indicate an imprecise preparation of standards.

Lab. No. 72 Only one GC column was used. Very acceptable results were found, except for CB180 in B and C and CB156 in C.

Lab. No. 73 Although the chromatography looks good, high results were found for CBs 101, 118, 138 and 180 in B.

Lab. No. 78 For A, B and Y the results produced were too low. Imprecise standard preparation and too low sensitivity (all chromatograms showed rather small peaks) could be reasons for this performance.

Lab. No. 79 This laboratory only had difficulties with the C solution, probably due to a restricted sensitivity.

Lab. No. 80 Only one GC column was used. No internal standards were used. Nitrogen was used as a carrier gas. All results for A and B were too high, probably influenced by an imprecise weighing of standards. The indicative values for C showed several deviations.

Lab. No. 81 Only GC/MS data were delivered. The values were marginally high for several CBs in A, B and C. This may be due to calibration difficulties.

Lab. No. 83 This laboratory used a commercial standard solution and was the exception in producing reliable results by this method. Some slightly leading peaks were observed.

Lab. No. 85 Acceptable results were obtained for A and Y, except for CB31 in A, but the results for B and C were often high. The results of the second column for Y showed too much variation. With the columns used it should be possible to improve the separation. A high background noise was observed.

Lab. No. 87 Only one GC column was used. The internal diameter of 0.32 mm is wide to obtain a sufficient separation between the CBs. For example, CBs 28 and 31 could not be separated. The results of the uncleaned B extract were used for this exercise. For both the cleaned and the uncleaned extracts, deviating results were found. The results for C were rather high, which might be due to a calculation error.

Lab. No. 88 A high bleeding was observed in the chromatograms. The results for Y were very poor. Only a few results for A, B and C were acceptable. Further optimization of the chromatographic conditions is advised.

4.3 Late Results

Lab. No. 1 All results were biased to the high side. The concentrations found in the blank were sometimes higher than those in the C solution. Results were reported from a

RSL 300 and a SE-54 column, but no choice was made between the results. The fast temperature program had a negative influence on the resolution of both columns.

Lab. No. 3 The results from this laboratory must be considered as outlying over the whole range of CBs for solutions A, B and Y. A total reconsideration of calibration procedures, chromatographic conditions and optimization is strongly recommended.

Lab. No. 53 A set of additional results produced with GC/ECD was returned after a first set using GC/MS. Although for A some results were now acceptable, the overall results must be considered as insufficient. Chromatograms showed tailing peaks and insufficient sensitivity was obtained for the analysis of solution C.

Lab. No. 84 This laboratory used GC/MS with chemical ionization for the detection of the CBs. The results were very acceptable for A and Y. Several CBs in the B solution were too high.

5 STATISTICAL EVALUATION

The statistical evaluation was partly based on international standard ISO 5725 for interlaboratory tests (ISO, 1986). According to this standard, the repeatability value r is the value below which the ratio of two single test results (maximum/minimum) obtained with the same method on identical test material, under the same conditions (same operator, same apparatus, same laboratory and a short interval of time) may be expected to lie with a probability of 95%. The reproducibility value R is the value below which the ratio of two single test results obtained with the same method on identical test material, under different conditions (different operators, different apparatus, different laboratories and/or different time) may be expected to lie with a probability of 95%. Because the error in this exercise appeared to show a relative character, different from the ISO standard, a model with a multiplicative error structure was used. After log-transformation and back transformation, the model provided standard deviations for the repeatability S(r) and the reproducibility S(R) which must be applied as a factor instead of using them as coefficients of variation. For small S(r)s and S(R)s, the values $S(r) - 1$ and $S(R) - 1$ may be roughly compared with the values of the coefficients of variation CV(r) and CV(R).

Tables 10–13 show the results of r, R, S(r) and S(R) for this exercise. Recall that the relations between r and S(r) and R and S(R) are, respectively, $2.8 \log S(r) = \log r$ and $2.8 \log S(R) = \log R$. In addition to these parameters, the coefficient of interclass correlation (cic) was determined. This coefficient gives the ratio between the variation due to the bias (accuracy) and the sum of this variation plus the variation due to precision. A high cic (close to 1) means that the major part of the variation has been caused by the bias.

Principal component analyses were performed for A, B, C and Y to indicate the outlying laboratories. Figures 6, 7 and 8 reflect the results of these principal component analyses for solutions A, B and C. Table 14 shows the outlying laboratories which were determined by this principal component analysis. The main criterion which has been applied in this principal component analysis is the deviation from the target values (A, Y) or the mean values (B, C).

6 DISCUSSION

With 58 participating laboratories, this second step of the Intercomparison Programme on the Analysis of CBs in Marine Media has, like the first step, resulted in valuable information being obtained on the performance of laboratories dealing with the analysis of CBs in the marine environment. Unfortunately, nine laboratories did not return results. Although there might be several reasons for not returning results, it must be emphasized again that these laboratories failed to appreciate the large amount of money and effort which has been expended in the organization of these exercises.

As a logical continuation of the first step, the participants were asked to prepare their own calibration solutions and to analyse a cleaned seal blubber extract and/or a cleaned sediment extract. An extra test was requested to be performed by the laboratories that had been outliers in the first step. The results will be discussed for each solution.

6.1 Unknown CB Solution

The S(R)s varied between 1.25 and 1.35 for the unknown solution A for 53 laboratories for CBs 52, 101, 118, 138, 153 and 180. Excluding the outliers, the S(R)s were reduced to 1.16–1.17; and for a selected group of 39 laboratories, which produced full data sets for all CBs, S(R)s for all CBs were between 1.16–1.33 (Table 10). The cic showed that the main variation was due to the bias between the laboratories. The precision of the individual laboratories was generally not a problem. However, the precision obtained in this exercise may not be a realistic estimate of the laboratory precision. In this exercise three injections per solution were requested, which were allowed to take place within a short period of time. To estimate the long-term precision of a laboratory, several injections should be made with periods of, for example, one week in between. An increase in the precision may be expected then, which might go together with a reduction in the bias.

CBs 31, 105, 118 and 156 showed the highest S(R)s, namely, 1.26–1.33 in the selected group. The mean results are within 10% of the target values, except for CB28 (13%) and CB52 (16%). A chromatogram of the A solution is shown in Figure 1.

A comparison with the results of the first step shows that the S(R)s have increased from 1.11–1.13 to 1.16–1.33 for the results of the selected group of laboratories. This means that, in addition to the error of the final chromatographic determination, there is an error associated with the preparation of standards. It is essential that this calibration error be reduced since it will affect all further results.

There are a number of sources of error in the preparation of calibration solutions. Firstly, place calibrants must be solid materials of known purity >95%, preferably certified standards. Commercially available standard solutions should not be used, because often deviations from the given concentrations occur. Weighing these materials is not simple. At least 5 mg should be weighed directly into a measuring flask. No weighing papers or glasses should be used. The balance must be able to weigh to at least 0.01 mg. Care should be taken to maintain a balanced temperature in the weighing room. Electrostatic problems should not hinder the weighing. Before weighing the standards, the variation in the display of the balance should be measured for some time. Two standards must be prepared independently of each other. These standards should be checked against a previously prepared solution. Solvents should be weighed as well. Iso-octane or heptane should be used. Standard solutions must be kept in a refrigerator and the weight must be checked regularly. Solvent losses via evaporation must be corrected. Preferably the stock solutions should be stored in ampoules. When stored in measuring flasks or bottles, standard solutions should not be stored longer than one year.

The Coordinators recommend that this part of the exercise be repeated during the next step. They also recommend that an unknown CB solution be supplied during any further steps for a continuous control of the calibration. Furthermore, it was suggested that participants using commercial standard solutions be excluded from further participation in this programme.

6.2 Seal Blubber Extract

Chromatograms of the seal blubber extract are shown in Figures 2 and 3. The S(R)s varied between 1.23 and 1.47 for the analysis of the seal blubber extract B by 45 laboratories. Excluding the outliers, the S(R)s were reduced to 1.20–1.33 (Table 11). Again, the main variation was due to the bias. The highest S(R) was obtained for CB52, namely, 1.33, and the lowest for CB118, namely, 1.20. Statistical results for CB28 and CB31 are not given because too few results were delivered for these CBs. For CBs 28 and 31, a large variation was found, possibly due to the presence of two additional components which coeluted with CBs 28 and 31 on an SE-54 column. This was detected with multidimensional gas chromatography at RIVO (Figure 5). In addition, these peaks were very small in relation to the other CB peaks.

The errors made in the calibration, of course, also had an effect on the B results. However, in comparison with sample A, the difference in the S(R)s still gives rise to some optimism. Obviously, there are difficulties in analysing CBs 105 and 156, with S(R)s of 1.38 and 1.62, respectively, for a selected group of 35 laboratories. From the viewpoint of toxicity this is disquieting, since CBs 105 and 156 may be of importance in the estimation of the toxicological impact of PCBs (Hong and Bush, 1990; Safe, 1987). Insufficient chromatographic separation is the main reason for this poor performance. On SE-54 columns, CB105 elutes close to CB132 and CB153, and CB156 coelutes with CB171 and close to CB202. Columns with diameters of 0.32 mm give insufficient resolution for a good separation of these CBs. Column lengths of 25 m are also insufficient. A step forward in the analysis of an uncleaned seal blubber extract may be made only when results obtained using columns with minimum lengths of 50 m and internal diameters of 0.25 mm are reported.

6.3 Sediment Extract

The results for the sediment extract C show S(R)s between 1.69 (CB118) and 3.06 (CB52) for 33 laboratories. Excluding the outliers, the S(R)s were reduced to between 1.31 (CB153) and 1.56 (CB52) (Table 12). No statistical data are given for CBs 28, 31, 105 and 156 because too few complete data sets were available and the quality of the remaining data sets was very poor. The analysis of these CBs must be judged to be impossible for the majority of the participants in this exercise. For the remaining six CBs high S(R)s were also found. A chromatogram of the sediment extract is shown in Figure 4. Sensitivity was a major problem for most laboratories. However, this extract was prepared from a common Wadden Sea sediment and should certainly not be regarded as uncontaminated. Many sediment samples from the Joint Monitoring Programme will contain considerably lower CB concentrations. If more material had been used for one extract, there might have been fewer problems. On the other hand, for this exercise only a cleaned extract was analysed. Higher S(R)s may be expected when undertaking an analysis of a real sediment sample. For an acceptable analysis of CBs in North Sea sediments, the Coordinators consider that the sensitivity required for this sediment is a minimum condition.

In addition to the sensitivity problems, poor chromatographic separation resulted in poor performance. As was the case for solution B, the separation conditions were often far from optimum. Negative peaks were regularly observed, indicating the presence of electron-donating compounds in the chromatographic system. Peaks present in the blank chromatograms also suggested imprecise results. An S(R) of 1.43 corresponds to a reproducibility R of 2.75. This means that, in joint studies of CB levels, the differences between two values will be within a factor of 2.75, with a probability of 95%; 95% of all results will be found in an area with extremes which differ by a factor of 4.2 from each other. With this variation, identification of any trend is impossible.

The Coordinators suggest that the next step focus on improvement in the calibration and progress in the analysis of CBs in seal blubber. Only when those exercises show sufficient improvement can the analysis of a sediment extract be considered again.

6.4 Extra Test

Table 13 shows the summary of the results obtained for the analysis of an unknown CB solution. S(R)s between 1.18 (CB28) and 2.80 (CB118) were obtained for a group of nine laboratories (without outliers). Thirteen laboratories participated in this exercise. The separation of CB118 from CB149 was difficult for most laboratories, although in most cases the separation was obtained on at least one column. The results from laboratories 3, 12, 67 and 88 are insufficient. A total reconsideration of calibration procedures, chromatographic conditions, and optimization is recommended for these laboratories.

6.5 Qualification of Laboratories

Based on the results of the principal component analyses and on their chromatographic performance, the participating laboratories could be classified according to the quality level of their results. For each of the solutions, the Coordinators have identified three groups of laboratories, that can be specified by the following qualifications:

Group 1: All results were within 20% of the target or mean values, with the exception of a maximum of one result per solution. Acceptable chromatographic performance and calibration.

Group 2: Several deviating results. Several deficiencies in calibration procedure or the chromatographic system.

Group 3: Poor chromatography and/or difficulties with calibration or statistical outlier.

Applying these criteria resulted in the following division:

Unknown solution A

Group 1: Lab. Nos. 2, 10, 11, 21, 27, 36, 37, 39, 43, 45, 46, 49, 50, 52, 58, 59, 63, 68, 72, 79, 83, 84 (total: 22).

Group 2: Lab. Nos. 1, 6, 7, 12, 13, 14, 23, 24, 25, 28, 29, 33, 35, 40, 48, 51, 54, 57, 66, 69, 70, 73, 81, 85, 87, 88 (total: 26).

Group 3: Lab. Nos. 3, 16, 18, 22, 47, 53, 62, 67, 78, 80 (total: 10).

Seal blubber extract B

Group 1: Lab. Nos. 10, 21, 35, 36, 37, 43, 45, 46, 50, 51, 52, 63, 68, 70, 72, 79 (total: 16).

Group 2: Lab. Nos. 1, 2, 11, 13, 14, 25, 27, 28, 29, 39, 40, 48, 49, 59, 62, 66, 69, 73, 81, 84, 85, 88 (total: 22).

Group 3: Lab. Nos. 3, 6, 7, 12, 16, 18, 22, 47, 54, 57, 67, 78, 80, 87 (total: 14).

Sediment extract C

Group 1: Lab. Nos. 10, 35, 37, 43, 63, 83, 84 (total: 7).

Group 2: Lab. Nos. 2, 11, 14, 21, 22, 24, 27, 28, 36, 39, 45, 50, 51, 52, 57, 58, 59, 62, 66, 72, 80, 81, 85 (total: 23).

Group 3: Lab. Nos. 1, 6, 7, 12, 13, 16, 18, 23, 33, 40, 47, 48, 49, 53, 54, 67, 79, 87, 88 (total: 19).

This overview shows that only seven laboratories, Nos. 10, 37, 43, 46, 63, 68 and 83, have produced fully acceptable results for the requested analyses. Only four of these laboratories, Nos. 10, 37, 43 and 63, analysed all three solutions. The poor results for the sediment extract are reflected by the fact that only seven laboratories are in Group 1, while 19 laboratories are in Group 3.

Laboratories classed in Group 1 for one or more solutions are advised to maintain the present quality level and to try to improve where possible. Especially the quality of the analysis of CBs 105 and 156, which are of toxicological importance, needs improvement even for the best laboratories. Laboratories classed in Group 2 for one or more solutions are advised to note the deficiencies which appear from this exercise, to install the appropriate chromatographic columns, to reconsider their calibration procedures, and to follow further the suggestions given in this report under Section 4.2. Group 3 laboratories are advised to reconsider totally their calibration procedures, chromatographic conditions, and optimization of their instruments.

7 CONCLUSIONS AND RECOMMENDATIONS

a) The second step of the ICES/IOC/OSPARCOM Intercomparison Programme on CB Analysis has led to between-laboratory standard deviations of 1.16-1.17 for all laboratories except outliers for CBs 52, 101, 118, 138, 153 and 180 for the analysis of an unknown CB solution. Including CBs 28, 31, 105 and 156, between-laboratory standard deviations of 1.16–1.33 were obtained for a selected group of 39 laboratories. These results must be considered as insufficient with respect to the final objective of this exercise, which is a reduction of the variation in the results of CB analyses.

b) For the analysis of a cleaned seal blubber extract, between-laboratory standard deviations of 1.20 to 1.33 were obtained for all laboratories except outliers for CBs 52, 101, 118, 138, 153 and 180. Considering the standard deviations obtained for the standard solution and which are included in the results of the analysis of the seal blubber extract, there may be some optimism about further progress in the analysis of CBs in seal blubber. However, the quality of the analysis of CBs 28, 31, 105 and 156 is still insufficient.

c) The analysis of a cleaned sediment extract has resulted in between-laboratory standard deviations of 1.31 to 1.56 for all laboratories except outliers for CBs 52, 101, 118, 138, 153 and 180. For the time being, programmes requiring analysis of CBs in sediments by several laboratories must be judged as impracticable.

d) The preparation of reliable calibration solutions has been identified in this exercise as one of the major problems. It is strongly recommended that laboratories use solid calibrants as the basis for their calibration solutions. Participants who insist on using commercial standard solutions should demonstrate the quality of these solutions by checking them against weighed solid standards.

e) Insufficient chromatographic separation, which is especially shown by the poor results for CBs 28, 31, 105 and 156, is another reason for the disappointing results. It is suggested that in further steps of this programme results will only be accepted which are obtained with gas chromatographic columns with minimum lengths of 50 m and maximum internal diameters of 0.25 mm. Reduction of the internal diameter to 0.20 mm or less is strongly recommended. In addition, the Coordinators suggest that the results of laboratories who insist on producing results based on only one column no longer be accepted.

f) Some laboratories have still produced results without the use of internal standards. The Coordinators suggest that these results no longer be accepted. During further steps in this programme, the choice of the internal standards will be left to the participants.

g) Based on earlier agreements during the design of this exercise, it is advised that the following laboratories will be excluded from further participation: Lab. Nos. 17, 26, 30, 34, 44, 64, 74, 82 and 86, because they withdrew during this second step or could not produce any results, even within five months after the deadline.

h) It is advised that laboratories take notice of the recommendations in this report and as soon as possible take the necessary measures to improve their performance. It is suggested that a period of about five months be given to the participants to revise their methods where necessary, install new columns, etc., and practice their revised methods.

i) In order to obtain a realistic estimate of the long-term precision of the laboratories, the Coordinators suggest that participants analyse a certified reference material fish oil six times with intervals of about one week between analyses. The Coordinators suggest that this exercise be planned for November and December 1991.

j) Depending on the results of this exercise, a next step may be planned for 1992 in which an unknown CB solution may be analysed together with a cleaned and an uncleaned seal blubber extract and a cleaned and an uncleaned sediment extract.

ACKNOWLEDGEMENTS

The Coordinators would like to thank all participants for their kind cooperation and many useful comments. Dr G. Andunsson of the Icelandic Fisheries Laboratories is acknowledged for the care and transport of the seal blubber sample and Mrs U. Eriksson of the Swedish Environmental Protection Agency is acknowledged for the preparation and ampouling of the seal blubber extracts. Mr F. Smedes and Mr J. Hermans of Rijkswaterstaat, Tidal Waters Division, The Netherlands are acknowledged for the preparation of the sediment extracts. The help of Mrs Q.T. Dao, Mr P.G. Wester and Mr W. Koedijk of the Netherlands Institute for Fisheries Research with the ampouling of the extracts is much appreciated. The comments of Dr D.E. Wells of the Marine Laboratory in Aberdeen were a valuable help in designing this exercise and, last but not least, Ms M. Sørensen of the ICES Secretariat is acknowledged for her kind administrative assistance.

REFERENCES

de Boer, J., and Dao, Q.T. 1991. Analysis of seven chlorobiphenyl congeners by multidimensional gas chromatography. J. High Resolut. Chromatogr., 14: 593–596.

de Boer, J., Duinker, J.C., Calder, J.A., and van der Meer, J. 1992. Report on the Results of the ICES/IOC/OSPARCOM Intercomparison Exercise on the Analysis of Chlorobiphenyl Congeners in Marine Media – step 1. ICES Cooperative Research Report No. 183, pp. 1-56.

Hong, C.S., and Bush, B. 1990. Determination of mono- and non-*ortho* coplanar PCBs in fish. Chemosphere, 21(1,2): 173–181.

International Organization for Standardization (ISO) 1986. Precision of test methods – Determination of repeatability and reproducibility for a standard test method by interlaboratory tests. 2nd edition. ISO 5725.

Larsen, B., and Riego, J. 1990. Interference from 2,3,5,6,3',4'-hexachlorobiphenyl (CB163) in the determination of 2,3,4,2',4',5'-hexachlorobiphenyl (CB138) in environmental and technical samples. Intern. J. Environ. Anal. Chem., 40: 59–68.

Safe, S. 1987. Determination of 2,3,7,8-TCDD toxic equivalent factors (TEFs): support for the use of the *in vitro* AHH induction assay. Chemosphere, 16(4): 791–802.

Table 1 : Participants.

LAB. NO.	CODE	INI-TIALS	NAME	INSTITUTE	ADDRESS	CITY	COUNTRY
1	RVZB	P.	Roose	Rijksstation voor Zeevisserij	Ankerstraat 1	B-8400 Oostende	Belgium
2	IHEB	E.	de Wulf	Instituut voor Hygiene en Epidemiologie	Juliette Wytmanstraat 14	B-1050 Brussels	Belgium
3 *	VUBB	C.	Joiris	Free University, Laboratory for Ecotoxicology	Pleinlaan 2	B-1050 Brussels	Belgium
6	BIOC	R.F.	Addison	Bedford Institute of Oceanography	P.O. Box 1006	Dartmouth, N.S. B2Y 4A2	Canada
7		B.	Grift	Freshwater Institute	501 University Crescent	Winnipeg, Manitoba R3T 2N6	Canada
10	SCSS	E.	Storr-Hansen	National Environmental Research Institute	Mørkhøj Bygade 26 H	DK-2860 Søborg	Denmark
11		K.	Erkomaa	National Board of Waters and the Environment	Hakunimaantie 4 - 6	SF-00430 Helsinki	Finland
12*	IMRF	H.	Haahti	Finnish Inst. of Marine Research	P.O. Box 33	SF-00931 Helsinki	Finland
		E.L.	Poutanen				
13	IPLF	J.C.	L'Hopitault	Service des Eaux de L'Institut Pasteur de Lille	B.P. 245	59019 Lille Cedex	France
14	LMRF	A.	Franco	Laboratoire Municipal et Region de Rouen	29, Rue Bourg l'Abbe	76000 Rouen	France
16		J.	Tronczynski	IFREMER Centre de Nantes	B.P. 1049	44037 Nantes Cedex 01	France
		M.	Lucon				
18		T.	Gaultier	Laboratoire Municipal du Harve	5, Rue Raymond Guènot	76600 Le Havre	France
21*	LABF	J.	Dussauze	Laboratoire Municipal de Brest (H.S.R.)	16, Rue Alexandre Ribot	29287 Brest	France
22	ICBF	A.	Abarnou	IFREMER Centre de Brest	B.P. 70	29280 Plouzané	France
23		A.	Medail	Laboratoire Municipal de Toulon (H.S.R.)	6, Avenue Francois Cuzin	83000 Toulon	France
24*		M.	Chapat	Institut Bouisson Bertrand (N.E.H.)	778, Rue de la Croix-Verte	34090 Montpellier	France
		H.	Mion				
25	BFRG	E.	Hüschenbeth	Bundesforschunganstalt für Fisherei	Wüstland 2	D-2000 Hamburg 55	Germany
				Labor für Radioökologie der Gewässer			
27	BFGG	H.	Bergmann	Bundesanstalt für Gewässerkunde	Postfach 309	D-5400 Koblenz	Germany
		B.	Schubert				
28*	DHIG	H.	Gaul	Bundesamt für Seeschiffart und Hydrographie	Wüstland 2	D-2000 Hamburg 55	Germany
				Labor Sulldorf			
29	VUCG	R.	Kruse	Staatliches Veterinäruntersuchungsamt	Schleusenstrasse	D-2190 Cuxhaven-F	Germany
				für Fische und Fischwaren			
33	LWKG	T.	Petenati	Landesamt für Wasserhaushalt und Küsten,	Saarbrückenstrasse 38	D-2300 Kiel 1	Germany
				Schleswig-Holstein			
35	RIVO	Q.T.	Dao	Netherlands Institute for Fishery Investigations	P.O. Box 68	1970 AB IJmuiden	The Netherlands
36	IVPT	J.B.	Luten	CIVO - TNO, Instituut voor Visserij Produkten	P.O. Box 183	1970 AD IJmuiden	The Netherlands
37	DGWN	W.P.	Cofino	Rijkswaterstaat Dienst Getijdewateren	Nijverheidsstraat 2	2288 BB Rijswijk	The Netherlands
		F.	Smedes				
		J.	Hermans				
39	SIIF	K.	Martinsen	Center for Industrial Research	P.O. Box 124, Blindern	0314 Oslo 3	Norway
		A.L.	Kvernheim				
40*	NIVA	R.G.	Lichtenthaler	Norwegian Institute for Water Research	P.O. Box 69	Korsvoll 0808 Oslo 8	Norway
		L.	Berglind				
43	IMRN	J.	Klungsøyr	Institute of Marine Research	P.O. Box 1870	N5024 Bergen-Nordnes	Norway
		S.	Wilhelmsen				
45	INIP	O.	Castro	Instituto National de Investiçãcao das Pescas	Avenida de Brasilia	1400 Lisbon	Portugal
		A.M.	Ferreira				

LAB. NO.	CODE	INI-TIALS	NAME	INSTITUTE	ADDRESS	CITY	COUNTRY
46		P.	Viana	Derecção-Geral Qualidade do Ambiente	Av. Alm Gago Coutinho 30-2	1000 Lisbon	Portugal
		J.	Matos				
47	IEOV	J.	Fumega	Instituto Español de Oceanografia	Cabo Estay - Canido Apartado 1552	36280 - Vigo	Spain
		A.	Gonzalez-Quyano Mosteiro	Centro Costero de Vigo			
48		J.	Albaigés	Centro de Investigacion y Desarrollo	Jordi Girona, 18-26	08034 Barcelona	Spain
49		P.	Azpeitia	Laboratorio de Contaminacion Toxicologia CONTOX	Cronos 8	28037 Madrid	Spain
50	NSLS	L.	Reutergårdh	Swedish Environmental Protection Agency	Englundavägen 5	S-17185 Solna	Sweden
		U.	Eriksson	Special Analytical Laboratory			
51	BLUK	C.R.	Allchin	MAFF, Fisheries Laboratory	Remembrance Avenue	Burnham-on-Crouch Essex CMO 8HA	UK
52	ALUK	D.E.	Wells	DAFS, Marine Laboratory	P.O. Box 101	Aberdeen, AB9 8DB	UK
		A.	Kelly				
53	CRUK	J.P.	Dawson	Clyde River Purification Board, Rivers House	Murray Road, East Kilbride	Glasgow, G75 0LA	UK
54	FRUK	I.M.	Ridgway	Forth River Purification Board	Avenue North, Riccarton	Edinburgh EH14 4AP	UK
57		D.A.	Kurtz	Department of Entomology, 122 Pesticide Research Lab.	Penn State University	University Park, PA 16802	USA
58		C.S.	Peven	Battelle Ocean Sciences	397 Washington Street	Duxbury, MA 02332	USA
59		J.L.	Sericano	GERG Texas A&M University	10 South Graham Road	College Station, TX 77845	USA
		T.L.	Wade				
62		C.	Younghans-Haug	UCSC-CDFG, Trace Organics Facility	100 Shaffer Road	St. Cruz, CA 95060	USA
63		M.M.	Schantz	NIST, Chemistry Building 222	Route 270, Quince Orchard Road	Gaithersburg, MD 20899	USA
		S.A.	Wise				
		W.E.	May				
66		M.Th.J.	Hillebrand	Netherlands Institute for Sea Research	P.O. Box 59	1790 AB Den Burg, Texel	The Netherlands
		J.P.	Boon				
		K.	Booij				
67*		F.	Benijts	Laboratorium ECCA	Klaartestraat 24	B-9710 Gent-Zwijnaarde	Belgium
68		A.E.	v.d. Zande	Rijksinstituut voor Natuurbeheer (RIN)	P.O. box 9201	6800 HB Arnhem	The Netherlands
69	VETN	A.	Polder	National Veterinary Institute	P.O. Box 8146, Dep.	0033 Oslo 1	Norway
		J.U.	Skaare	Department of Pharmacology & Toxicology			
70		D.F.	Gadbois	National Marine Fisheries Service	30 Emerson Avenue	Gloucester MA 01930	USA
				Northeast Fisheries Center			
72		J.	Nieuwenhuize	Delta Institute for Hydrobiological Research	Vierstraat 28	4401 EA Yerseke	The Netherlands
73		Ö.	Andersson	Swedish National Food Administration	Box 622,	S-75126 Uppsala	Sweden
78*		K.	Himberg	Technical Research Centre of Finland (VTT)	P.O. Box 203	SF-02151 Espoo	Finland
		E.	Sippola	Food Research Laboratory			
79*		H.	Büther	Staatliches Amt Für Wasser- und Abfallwirtschaft	Postfach 8440	D-4400 Münster	Germany
80*		J.W.	Readman	International Atomic Energy Agency	19 Avenue des Castellans	Fontvieille, MC 98000 Monaco	Monaco
		J.P.	Villeneuve	Marine Environmental Studies Laboratory			
81*		M.	Oehme	Norwegian Institute for Air Research	P.O. Box 64	N-2001 Lillestrøm	Norway
		M.	Schlabach				
83*		G.	Jonsäll	Nat. Laboratory for Agricultural Chemistry	Box 7004	S-750 07 Uppsala	Sweden
84*		S.	Einarsson	Icelandic Fisheries Laboratories	P.O. Box 1390	121 Reykjavik	Iceland
85*		K.	Olafsdottir	University of Iceland	Armuli 30	108 Reykjavik	Iceland
87		J.C.	Duinker	Institut für Meereskunde	Düsternbrookerweg 20	D-2300 Kiel 1	Germany
88*		N.	Kluyev	Inst. of Evolutionary Morphology and Ecology of Animals	Leninsky av. 33	Moscow 117071	USSR
				Laboratory of Analytical Ecotoxicology			

Table 2. Gaschromatographic columns and conditions, 2nd step.

LAB. NO.	STATIONARY PHASE	COLUMN LENGTH (m)	INT. DIAMETER (mm)	FILM THICKNESS (µm)	CARRIER GAS	LIN. GAS VELOCITY (cm/sec.)	INJ. TECHNIQUE
2	CP-Sil 8	50	0.25	0.26	He	36	on column
2	CP -Sil 19	25	0.25	0.20	He	33	on column
6	DB-5	30	0.25	0.25	He	38	splitless
6	SPB-20	30	0.25	0.25	He	32	splitless
7	DB-5	60	0.25	0.25	H2	33	splitless
10	DB-5	60	0.25	0.11	He	25	splitless
10	DB-1701	60	0.25	0.15	He	25	splitless
11	SE-54	50	0.2	0.25	He	17	splitless
11	NB-1701	50	0.2	0.25	He	35	splitless
12	SE-54	50	0.32	0.15	He	35	splitless
12	OV-1701	25	0.32	0.15	He	36	splitless
13	DB-5	30	0.32	0.25	He		on column
13	DB-17	30	0.25	0.25	He/H2		on column
14	DB-5	60	0.317	0.25	H2	30	splitless
14	DB-1701	60	0.26	0.15	H2	27	splitless
16	CP-Sil 8 CB	25	0.32	0.13	He	28	direct
18	CP-Sil 8 CB	50	0.25	0.28	H2	21.3	on column
18	OV-1	25	0.32	0.10	H2	11	on column
21	BP-5	50	0.22	0.25	H2	40	on column
21	CP-Sil 19 CB	50	0.25	0.20	H2	30	on column
22	CP-Sil 8	50	0.25	0.12	H2	50	on column
22	CP-Sil 19						on column
23	CP-Sil 8 CB	50	0.25	0.24			solid inj.
24	DB-5	30	0.32	0.25	N2	17.8	on column
24	SPB-608	30	0.25	0.25	N2	24.1	on column
25	SE-54	50	0.25	0.27	He	39	splitless
25	OV-1701	50	0.25	0.23	He	42	splitless
27	DB-1	30	0.25	0.25	He	22	splitless
27	DB-1701	30	0.25	0.25	He	24	splitless
28	DB-5	60	0.25	0.25	He	17	on column
28	DB-1701	60	0.25	0.25	He	17	on column
29	DB-5	60	0.25	0.25	H2	30	splitless
29	RTX-20	60	0.25	0.25	H2	30	splitless
33	SPB-608	30	0.25	0.25	He	25	splitless
33	RTX-5 *	30	0.25	0.25	He	27	splitless
35	CP-Sil 8 CB	50	0.15	0.30	H2	28	splitless
35	CP-Sil 19 CB	60	0.15	0.20	H2	37	splitless
36	DB-1701	30	0.32	0.25	He	36	splitless
36	CP-Sil 8 CB	50	0.25	0.13	He	22	splitless
37	SE-54	50	0.15	0.20	H2	38	splitless
37	CP-Sil 19 CB	50	0.15	0.20	H2	37	splitless
39	SPB-5	60	0.25	0.25	H2	43	splitless
39	CP-Sil 19 CB	50	0.25	0.21	H2	41	splitless
40	MeSigum	50	0.2	0.50	H2	43	splitless
40	SPB-5	60	0.25	0.25	H2	35	splitless
43	SE-54	50	0.2	0.11	H2	39	splitless
43	SP-2330	60	0.25	0.20	H2	36	splitless
45	DB-5	60	0.254	0.25	He	22	splitless
45	DB-1701	60	0.256	0.25	He	22	splitless
46	SE-54	50	0.25	0.22	H2	40	splitless
46	OV-1701	60	0.25	0.22	H2	40	splitless
47	CP-Sil 8 CB	25	0.15	0.4	He	30	splitless PTV
47	OV-101	50	0.25	0.25	He	35	splitless PTV
48	SE-54	50	0.25	0.25	He	25	splitless
48	DB-17	30	0.252	0.25	He	32	splitless
49	HP-5	50	0.33	1.05	He		splitless
49	RSL-300	25	0.25	0.20	He		splitless
50	SE-30	50	0.2	0.33	He	24.6	splitless
50	SE-54	50	0.2	0.33	He	21.9	splitless
51	HP-5	50	0.2	0.11	H2	45	on column
51	CP-Sil 19 CB	50	0.25	0.20	H2	42	on column
52	CP-Sil 8 CB	50	0.25	0.25	H2	35	on column
52	CP-Sil 19 CB	50	0.25	0.20	H2	35	on column

LAB. NO.	STATIONARY PHASE	COLUMN LENGTH (m)	INT. DIAMETER (mm)	FILM THICKNESS (µm)	CARRIER GAS	LIN. GAS VELOCITY (cm/sec.)	INJ. TECHNIQUE
53	DB-5	45	0.25	0.25	He	25	on column
54	DB-5	30	0.249	0.25	He	25	on column
54	DB-1701	30	0.25	0.25	He	25	on column
57	SPB-5	60	0.32	0.25	H2	32	direct
57	RTX-35	60	0.32	0.25	H2	32	direct
58	DB-5	30	0.25	0.25	H2	19.1	splitless
58	DB-17	30	0.25	0.25	H2	19.1	splitless
59	DB-5	30	0.25	0.25	He	32	splitless
59	DB-1701	30	0.25	0.25	He	40	splitless
62	DB-17	30	0.25	0.25	He	36	splitless
62	DB-5	30	0.25	0.25	He	36	splitless
63	DB-5	60	0.25	0.25	He	30	splitless
63	DB-1701	40	0.25	0.25	He	30	splitless
66	CP-Sil 8 CB	50	0.25	0.12	He	29	splitless
67	SE-54	25	0.25	0.25	N2		splitless
68	DB-5	30	0.25	0.25	He	28	on column
68	DB-5	30	0.25	0.25	He	28	solid inj.
69	CP-Sil 8 CB	50	0.15	0.20	He	10	splitless
69	SP-2250	30	0.25	0.20	He	40	splitless
70	DB-5	30	0.25	0.25	He	28.7	splitless
72	CP Sil-8-CB	50	0.25	0.12	He	30	splitless
73	SE-54	50	0.20	0.33	He	18	splitless
73	RTX-1701(OV-1701)	60	0.25	0.25	He	20	splitless
78	SE-51	25	0.20	0.25	He	25	splitless
79	SE-54	50	0.25	0.50	H2	38.5	splitless
79	OV-1701	50	0.25	0.32	H2	38.5	splitless
80	SE-54	50	0.25	0.17	N2	15	splitless
81	HP Ultra 2	25	0.20	0.11	He	31	splitless
81	RTX-1701	30	0.25	0.10	He	35	splitless
83	DB-5	60	0.25	0.25	He	25.6	splitless
83	DB-1701	60	0.25	0.25	He	24.5	splitless
85	HP Ultra 2	50	0.20	0.33	He	19.7	splitless
85	HP Ultra 1	25	0.20	0.33	He	28	splitless
87	SE-54	50	0.32	0.25	H2		on column
88	HP-5	25	0.32	0.52		30	splitless
88	SP-2250	30	0.25	0.25		30	splitless

Table 3. Gaschromatographic columns and conditions, extra test.

LAB. NO.	STATIONARY PHASE	COLUMN LENGTH (m)	INT. DIAMETER (mm)	FILM THICKNESS (µm)	CARRIER GAS	LIN. GAS VELOCITY (cm/sec.)	INJ. TECHNIQUE
12	SE-54	50	0.32	0.15	He	55	splitless
12	OV-1701	25	0.32	0.15	He	36	splitless
21	BP-5	50	0.22	0.25	H2	40	on column
21	CP-Sil 19 CB	50	0.25	0.20	H2	30	on column
24	DB-5	30	0.32	0.25	N2	17.8	on column
24	SPB-608	30	0.25	0.25	N2	24.1	on column
28	DB-5	60	0.25	0.25	He	17	on column
28	DB-1701	60	0.25	0.25	He	17	on column
40	MeSigum	50	0.20	0.50	H2	43	splitless
40	SPB-5	60	0.25	0.25	H2	35	splitless
67	SE-54	25	0.25	0.25	N2		splitless
78	SE-51	25	0.20	0.25	He	25	splitless
78	DB-23	30	0.25	0.25	He	25	splitless
79	SE-54	50	0.25	0.50	H2	38.5	splitless
79	OV-1701	50	0.25	0.32	H2	38.5	splitless
81	HP Ultra 2	25	0.20	0.11	He	31	splitless
81	RTX-1701	30	0.25	0.10	He	35	splitless
83	DB-5	60	0.25	0.25	He	25.6	splitless
83	DB-1701	60	0.25	0.25	He	24.5	splitless
85	HP Ultra 2	50	0.20	0.33	He	19.7	splitless
85	HP Ultra 1	25	0.20	0.33	He	28	splitless
88	HP-5	25	0.32	0.52		30	splitless
88	SP-2250	30	0.25	0.25		30	splitless

Table 4. Homogeneity test of the ampoules, concentrations expressed in pg/μl

a) Ampoule A:

CB	A1	A2	A_{stock}	mean	s.d.	s.d.%
28	44	44	45	44	0.58	1.3
31	44	45	44	44	0.58	1.3
52	51	50	52	51	1.0	2.0
101	44	46	48	46	2.0	4.3
105	38	39	40	39	1.0	2.6
118	49	48	49	49	0.58	1.2
138	68	66	66	67	1.2	1.8
153	70	73	71	71	1.5	2.1
156	37	35	37	36	1.2	3.3
180	32	32	33	32	0.58	1.8

b) Ampoule B:

CB	B1	B2	B3	mean	s.d.	s.d.%
28	11	11	13	12	1.2	10
31	4.9	5.9	7.4	6.1	1.3	21
52	23	24	26	24	1.5	6.3
101	52	55	59	55	3.5	6.4
105	13	14	16	14	1.5	11
118	30	34	37	34	3.5	10
138	136	140	155	144	10	6.9
153	243	247	261	250	9	3.6
156	7.0	7.8	9.1	8.0	1.1	14
180	34	36	37	36	1.5	4.2

c) Ampoule C:

CB	C1	C2	C3	C4	C5	C_{stock}	mean	s.d.	s.d.%
28	1.2	1.1	1.1	1.1	1.1	1.1	1.1	0.04	3.6
31	0.86	0.73	0.81	0.78	0.76	0.82	0.79	0.05	6.3
52	0.73	0.59	0.60	0.63	0.61	0.60	0.63	0.05	8.3
101	1.4	1.3	1.2	1.2	1.2	1.2	1.3	0.08	6.2
105	0.43	0.37	0.34	0.32	0.35	0.33	0.36	0.04	11
118	1.5	1.3	1.3	1.2	1.1	1.2	1.3	0.14	11
138	2.0	1.7	1.7	1.6	1.7	1.8	1.8	0.14	7.8
153	2.1	1.9	1.9	1.8	1.8	1.9	1.9	0.11	5.8
156		0.27	0.24	0.19	0.19	0.20	0.22	0.04	16
180	0.9	0.8	0.8	0.7	0.7	0.8	0.78	0.08	10

d) Ampoule D:

	D1	D_{stock}
TCN	2.1	2.1
OCN	3.8	3.8

e) Ampoule X:

CB	X1	X2	X3	X4	X_{stock}	mean	s.d.	s.d.%
28	900	940	940	940	900	924	22	2.3
31	900	940	920	960	900	924	26	2.8
118	720	700	760	720	700	720	24	3.3
153	760	780	780	800	780	780	14	1.8
180	700	660	680	700	660	680	20	2.9

f) Ampoule Y:

CB	Y1	Y2	Y3	Y4	Y_{stock}	mean	s.d.	s.d.%
28	46	42	42	43	46	44	2.0	4.5
31	46	44	44	43	44	44	1.1	2.5
118	41	40	43	41	44	42	1.6	3.8
153	74	73	76	71	75	74	1.9	2.6
180	38	41	40	39	42	40	1.6	4.0

g) Blank values ampoules/iso-octane

CB	1	2	3	4
28	0.45	0.32	0.21	0.22
31	<0.13	<0.11	<0.12	<0.12
52	<0.21	<0.18	<0.19	<0.20
101	1.25	0.73	0.76	0.65
105	0.19	<0.07	<0.07	<0.08
118	0.64	<0.11	<0.12	<0.12
138	0.43	<0.07	<0.08	<0.06
153	0.44	<0.08	<0.08	<0.08
156	0.41	<0.08	<0.03	<0.03
180	0.86	0.49	<0.41	0.36

Table 5. Target values of CB in ampoules A and Y (pg/μl)

CB	A	Y
28	40.5	40.5
31	40	40
52	43.5	
101	56	
105	41.7	
118	56	48
138	82	
149		75.2
153	80	80
156	41.3	
180	40	48

Table 6. RESULTS OF THE ANALYSIS OF AMPOULE A (STANDARD SOLUTION).

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
2	1	-	-	47	51	43	54	83	79	52	45
2	2	45	42	47	52	42	58	84	79	52	43
2	3	43	41	46	52	42	55	85	79	50	43
6	1	-	-	76.6	69.4	52.7	65.6	94	85.5	39.7	49.7
6	2	-	-	62.7	58.2	52.5	65.4	91.7	85.3	42.5	48.3
6	3	-	-	71.1	66.9	53.2	66.9	96.4	88.4	40.9	50.7
7	1	76.3	56.41	52.64	44	54.45	50.87	58.42	53.2	43.48	32.86
7	2	87.83	60.17	57.47	43.26	61.21	58.96	66.95	58.22	45.09	33.88
7	3	83.93	57.11	54.89	44.85	57.62	51.25	65.58	60.47	42.87	35.17
10	1	44.2	46.3	51.8	55	37.1	55.2	82.4	78.6	27.1	38.3
10	2	42.8	45.5	49.3	52.1	40.1	57.8	80.9	77	29.6	37.7
10	3	41.1	44.3	49.6	53.2	45.2	62.1	88.3	81.4	36.7	43.7
11	1	42.8	44.4	49.4	50.4	41.4	57	82.4	78.4	40.5	42.5
11	2	43.1	44.4	49.1	50.6	41.6	57.4	82.5	78.6	40.4	42.4
11	3	42.8	44.3	49.3	50.7	41	57.7	83.3	80.2	40.2	42.3
12	1	-	-	54.7	56.3	56.8	66.8	81.7	65.1	48	45.7
12	2	-	-	56.5	57.1	56.6	66.8	82.1	65.6	47.7	45.7
12	3	-	-	55.6	56.9	54.7	65.2	81.3	65.1	46.9	45.5
13	1	61	-	53	48	-	55	74	76	-	41
13	2	41	-	38	44	-	46	63	61	-	34
13	3	56	-	53	53	-	60	75	72	-	37
14	1	48.5	48	51.3	57	45	66.8	91.2	90	43	48.5
14	2	46.5	47.5	51	61.2	43.8	62.5	93	91.2	43.1	47.5
14	3	51	51	52.5	56.9	45	66.2	93	90	44	48.1
16	1	101.3	-	53	67.8	-	87.5	107.2	88.4	-	130.1
16	2	65.7	-	46.8	62.7	-	93.5	117.7	79.3	-	178.7
16	3	101.5	-	53.9	68.6	-	84.3	121.6	109.4	-	157.4
18	1	41.8	40.4	44.1	43.4	77.3	12.7	52.8	65.4	28.6	25.9
18	2	37.4	44.2	40.4	38.5	72.9	21.6	69.7	57.3	31.5	25.6
18	3	40.6	47.9	45.1	48.9	76.6	23.2	76.5	56.6	37.3	32.4
21	1	39.7	43.5	43.5	47.9	31	56.6	81.9	76.3	30.9	42.1
21	2	39.5	42.5	44.8	47.8	30.9	56	81.8	77.4	30.8	41.5
21	3	39.5	42.4	44.2	50.2	31.4	57.6	82.5	77.1	32.1	41.7
22	1	-	-	53.3	50.1	40.7	54.1	80.7	73.9	-	80.6
22	2	48.1	-	52.9	48.2	36.6	51.2	80.4	74.3	-	84.7
22	3	49.2	-	51.7	45.5	34	47.3	78	67.8	-	79.4
23	1	34.6	27.2	42.4	43.2	34.3	54.3	68.7	66	39.1	43.6
23	2	31.8	32.2	38.4	50	49.2	66.7	76.9	71.1	47.3	50.7
23	3	32.1	30.5	38.1	44.9	46.5	64.1	79.7	72.9	44.8	49.9
24	1	37.3	36.7	-	-	-	56.3	-	78.3	-	35.5
24	2	38.9	38.7	-	-	-	54.2	-	79.4	-	42.1
24	3	39.3	39.1	-	-	-	53.2	-	78.8	-	41.9
25	1	42.4	43.6	58.3	50.9	38	43	87.7	73.7	20.4	40
25	2	50.6	45.6	68.6	58	47	50.1	89.5	91	25.5	45.3
25	3	35.9	42.8	49.4	53.3	43.3	43.3	70.5	74.3	20.8	42.5
27	1	49.7	18.6	48.8	51.8	44	54	80.7	73.7	40.3	40.6
27	2	49.7	19.5	47.5	51.4	43.7	57.6	85.7	71.3	40.4	43.6
27	3	53.3	19.2	51.8	54.9	43.8	61.3	90	77.1	39.8	46.5
28	1	38.6	23.5	61.3	52	46.4	55.5	75.7	70.7	22.8	41.2
28	2	38.4	23.9	51.3	51.1	45.4	55.7	78	73.4	21.4	42.7
28	3	38.4	24	51.4	52.3	46	54	74.4	71.1	19.1	39.7
29	1	48.4	49.8	54.7	59.7	44.2	64.8	84.1	85.6	41.4	38.9
29	2	48	50.2	54.2	60.5	42.3	62.8	80.7	89	41.4	41.7
29	3	45.7	48.5	53	57.4	42.6	61.6	79.8	84.3	41.7	39.5
33	1	-	-	43.9	47.6	38.9	37.4	82.3	70.9	30.6	32.6
33	2	-	-	44	44.9	40.6	37.7	82.7	74.3	32.3	33.3
33	3	-	-	44.1	44.1	37.3	35.8	78.4	65.6	30.3	31.5
35	1	44	44	51	44	38	49	68	70	37	32
35	2	40	39	45	41	41	47	66	77	40	35
35	3	43	43	50	42	40	45	65	76	41	34
36	1	45.5	43.9	46.1	53.8	41.5	62.8	84.4	80.9	40.1	41.1
36	2	44.8	43.9	47	52.2	40.1	61.4	81	81.4	39.8	41.2
36	3	43.9	45.3	44.5	51.3	39.8	61.3	82.4	80.5	39	41.1
37	1	43	44.4	48.6	48.8	40.2	53.7	76.4	75.5	-	40
37	2	45	46.8	49.9	50.6	41.4	54.8	77.9	79.1	-	40.5
37	3	44.5	46.2	49.3	50.2	42.8	55.6	78.9	79.8	-	41.3
39	1	51	47	55	61	47	59	100	86	41	43
39	2	54	50	62	65	49	60	98	83	41	42
39	3	54	50	62	66	50	63	98	94	44	47
40	1	48	36	55	58	60	80	130	113	50	56
40	2	50	35	54	56	48	63	92	78	40	44
40	3	50	35	52	57	51	67	100	86	43	48
43	1	44.1	36.7	47.8	51	46.7	56.7	83.6	70.9	36.3	40
43	2	41.9	34.4	44.3	47.7	46.2	55.9	83.5	67	36.1	39.3
43	3	42.1	33.8	43.3	46.2	45.9	53.9	83	65.6	35.3	38.9

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
45	1	46.3	41.1	51.1	54.6	41.4	64.8	85.2	86.1	36.1	39.4
45	2	44.1	41.1	47.8	55.7	43.9	67	94.3	99.4	38.3	43.2
45	3	43.4	40.2	45.6	51.2	43.9	63.7	82.5	82.8	38.3	39.4
46	1	41.2	32.8	45.2	58.2	41.9	63.9	100	90.1	42	43.3
46	2	43.4	34.3	46.9	56.6	41.2	62.9	97.4	90.3	40	41
46	3	41.7	34	46.4	56.6	38.2	59.4	94.3	85.5	40.2	41.3
47	1	36.9	17.9	89.1	59.3	78.5	64.7	95.6	94.5	60.1	48.9
47	2	38.3	18.2	89.1	59.3	73	69.7	92	92.2	56.3	46.2
47	3	39.6	18.2	86.2	60.3	74.9	66.7	95.7	94.8	57.6	48.3
48	1	42.8	51.7	51.6	47.7	40.2	65.8	81	60.3	27.7	36.5
48	2	41.8	52.9	51.8	49.1	42.5	62.3	79	58.8	26.3	33.5
48	3	36.4	45.5	47.4	44.5	32.6	66.6	82.9	62.2	19.6	33.1
49	1	42	47	51	52	48	59	85	84	43	42
49	2	41	43	47	52	38	51	77	74	38	38
49	3	43	45	53	51	45	50	81	83	35	41
50	1	47.6	46.5	48.2	50.3	18.7	62.6	94.8	73.6	39.5	43.5
50	2	45.9	45.6	46.3	50.5	18.7	63.5	95.1	74.3	41.7	43.2
50	3	48.1	47.6	51.2	53.8	18.3	61.7	98.8	77.8	39.1	43.8
51	1	36	33	38	35	29	45	63	54	33	33
51	2	37	32	37	34	29	45	61	55	34	32
51	3	36	32	36	34	30	45	61	55	32	32
52	1	43.96	44.54	49.63	54.44	43.92	60.44	91.09	83.61	40.58	40.54
52	2	43.83	43.26	45.72	55.48	43.27	64.47	91.77	85.2	41.08	40.04
52	3	44.23	44.28	47.63	55.41	61.01	61.42	91.12	81.89	42.94	40.69
53	1	60.6	-	39.3	36.4	-	93.9	78	62.4	-	34.4
53	2	59.2	-	34.3	39.8	-	99.6	78	60.4	-	35.4
53	3	62.7	-	36.4	36.8	-	94.4	79	62.8	-	35.8
54	1	93	-	50.8	61.9	-	63.2	91.9	70.4	-	40.6
54	2	96.4	-	52.6	63.4	-	65.6	92.9	73.7	-	41.6
54	3	99.4	-	55	66.4	-	69	99.3	79.5	-	44.6
57	1	44.7	50.6	52.6	55.1	42.2	58.7	91.9	77.3	41.6	42.1
57	2	43.3	50.2	54	59.6	41	58.9	97.8	80.4	40.1	41.1
57	3	43.9	45.7	48.8	50.2	40.1	55.8	95.6	80.1	40.4	36.6
58	1	39.2	40.9	52.8	54.6	40.4	62.1	86.9	86.1	37.3	42.6
58	2	38.9	40.7	52.9	53.9	39.5	60.1	87.9	83.5	39.2	43.9
58	3	38.6	40.5	52.2	53.3	39	60.4	84.1	83.2	38.1	43.9
59	1	46.9	48.7	53.3	58.7	44.7	61.8	91.1	82.3	42.1	44.6
59	2	49.4	46.7	52.8	58.8	43.2	62.1	92.1	85.2	41.4	44.5
59	3	43	50.9	51.3	57.2	44.6	60	90.8	79.3	40.8	43.4
62	1	46.1	59.6	66.6	69.1	48	69.2	104	98.5	54.9	51
62	2	46	58.9	66.5	68.2	46.7	69.2	105	103	53.1	49.7
62	3	46.2	59	66.1	68.7	47.3	69.8	107	101	53.7	51.2
63	1	42.8	44.5	42.9	53.3	42.6	59.6	69.1	77.9	36.7	38.4
63	2	43.2	44.5	42.5	53.5	42.3	58.6	68.9	77.6	36.2	38.6
63	3	42.9	44.8	42.5	53.8	41.9	56.7	68	76.5	35.9	38.3
66	1	46.5	47.7	51.1	54.1	45.2	61	83.1	72.5	38.7	29.7
66	2	46.9	47	51	54.2	43.9	61.6	85.1	74.1	39.1	29.9
66	3	46.9	47.4	49.2	52.9	46	63	84.2	71.2	41.9	31.5
67	1	-	-	61.1	72.2	42.7	74.2	90	90.8	81.6	55.8
67	2	-	-	54.2	56.6	40.6	60.6	79.3	74	71.6	54.7
67	3	-	-	50.3	47.2	37.8	50.7	73.9	68.9	66.7	48.8
68	1	47.7	38	43	48.1	36.5	57.6	74.5	77.6	44	44.8
68	2	49.9	39	44.3	48.3	38.1	58	74.6	76.7	44.2	43.7
68	3	48.2	39	42.7	46.3	37.7	59.3	73.2	75.9	46.3	43.2
69	1	56.73	47.89	44.27	51.65	43.28	51.87	91	79.68	55.63	52.49
69	2	58.21	49.17	50.23	55.06	41.87	50.66	89.08	86.17	37.72	45.48
69	3	56.86	49.09	47.63	54.61	41.42	51.83	90.46	87.85	43.69	48.39
70	1	46.94	47.26	38.85	46.47	72.73	67.06	101.01	78.93	58.77	54.51
70	2	47.36	47.74	38.39	45.4	73.39	65.5	97.97	73.84	60.66	54.78
70	3	48.28	48.83	38.45	46.29	75.09	66.68	98.61	75.99	60.83	54.96
72	1	44.6	47.8	50.5	53.6	41.6	57.3	83.7	75.7	38.9	35.3
72	2	43.2	47.5	52.9	55.1	40.7	57.3	80.6	75	44.9	38.3
72	3	43.8	47.1	51.2	53.6	43.1	59.9	84.9	75.6	35	37.2
73	1	53.2	55.5	56.2	63.2	35.1	66.3	89.2	79.1	35.1	43.3
73	2	52.9	54.7	57.4	62.2	37.4	66.4	95.2	85.5	37	45.5
73	3	53.2	54.2	57.3	62.2	37.8	66.2	95.4	85	36.9	45.8
78	1	12	14	10	13	14	19	21	24	-	14
78	2	12	17	15	18	16	18	24	26	-	16
78	3	8	20	13	16	16	19	28	23	-	13
79	1	43.4	47.9	37.5	43.7	37.9	56.8	72.8	69.9	34.2	38.4
79	2	38.7	45.5	39.2	54.9	47.1	55.1	84.7	75.3	40.5	42.5
79	3	39.7	49.6	43.8	59.1	48.6	56	84.5	74	39.6	42.2
80	1	56.19	76.23	65.19	56.79	66.03	72.42	89.25	86.7	53.52	56.5
80	2	77.25	81.18	74.43	65.22	64.8	83.22	103.95	93.6	55.35	54.3
80	3	78.9	81.36	65.52	69.54	60.99	81.78	104.04	96.27	57.96	58.7

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
81	1	58.5	42.1	52	59.3	50.2	65	90.7	95.2	42.1	46.3
81	2	50.9	44.9	49.1	53.3	44.5	57.9	93.2	92.6	47.8	50.3
81	3	48.4	56.8	58.7	62.4	43.2	60.9	98.9	107.3	42.7	51.7
83	1	43.4	48.3	53.8	56.2	42.5	62.3	88.6	81.8	41.5	47.5
83	2	44.2	48.8	51.9	55.7	43	63.2	84.8	78.5	44.2	46.9
83	3	43.8	48	53.3	56.1	44.1	62.7	92.3	85.9	42.7	48
85	1	41.8	55.4	42.4	54.9	33.8	63.1	80.3	83.1	34.3	43.1
85	2	41.8	53.4	43.2	48	31.7	70.1	83.2	80	36.3	42.5
85	3	43.8	55.2	53.8	54.6	31.5	68.6	79.5	70.6	37.8	37.6
87	1	-	-	31	39	29	37	76	60	27	36
87	2	-	-	36	46	34	43	84	71	30	36
87	3	-	-	37	47	36	44	94	76	33	40
88	1	-	-	56.4	49.1	51.3	89	16.8	64.3	-	44.5
88	2	-	-	42.3	41.6	51.2	84.6	-	65.6	-	48.3
88	3	-	-	56.9	51.2	50.3	86.2	-	62.6	25.5	45.8

Table 7. RESULTS OF THE ANALYSIS OF AMPOULE B (SEAL BLUBBER EXTRACT).

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
2	1	-	-	22	67	26	-	190	280	-	-
2	2	2.4	9.9	23	68	26	44	190	280	10	46
2	3	2.9	11	23	70	26	44	190	270	9.5	46
6	1	-	-	46.8	96.2	-	59.3	226.7	292.8	7.32	63.8
6	2	-	39.7	51.3	85	18.5	57.6	220.7	272.4	8.58	60.4
6	3	-	30.3	54.7	98.6	17.2	65.5	240.6	300.2	9.13	68.9
7	1	7.46	33.76	24.88	48.07	17.34	35.06	92.98	118.62	7.67	23.31
7	2	8.35	37.5	25.27	49.49	16.27	37.02	108.45	123.41	7.93	25.93
7	3	7.87	36.28	23.58	50.15	8.54	27.83	98.13	134.86	9.24	28.47
10	1	3.2	-	22.7	58.1	12.4	40.3	151	211.2	4.6	38.3
10	2	3.3	-	22.8	55.4	11.6	36.5	132.8	193.2	4.1	31.1
10	3	3.1	-	21.7	56.2	12.2	38.2	138.6	198.1	4.5	34.8
11	1	-	-	30.3	74.7	20.2	54	199	329	10.2	55.2
11	2	-	-	29.9	77.4	20.1	54	202	329	9.4	58.9
11	3	-	-	29.8	78.1	20.3	53	205	325	9.3	58.9
12	1	-	-	38.1	82.8	27	55.8	170.1	202	11.8	54.5
12	2	-	-	34.6	77.2	26.6	54.9	171.8	199.1	11.9	55.2
12	3	-	-	36.9	80.7	27	56	173.1	196.8	11.7	54.9
13	1	10	-	20	60	-	45	161	165	-	45
13	2	8	-	16	59	-	42	139	161	-	47
13	3	10	-	21	56	-	43	146	157	-	45
14	1	17.5	0.8	20	70	17.5	49.5	212.5	266	6.4	50
14	2	16.9	0.8	18.5	70.4	18.1	47.5	194	267.5	6.6	50
14	3	17.5	1	18.1	74.3	18.8	47.8	195	288	6.7	55
16	1	5.8	-	22	336.8	-	54.6	247.9	296.6	-	162.9
16	2	3.2	-	17.1	277.6	-	47.1	234.9	373.1	-	183.9
16	3	6.1	-	9.4	313.9	-	51.7	215.5	309.6	-	188.2
18	1	-	27.7	59.9	72.5	51.8	20.2	197	220	9.5	50.1
18	2	-	22.4	50.1	103.4	58.2	28.8	208	248	10	43.7
18	3	-	24.6	55.5	81.5	48.7	25	197	311	9.2	48.7
21	1	3.5	1.85	20.67	71.9	14.3	46.5	205	276	5.83	52
21	2	3.55	1.95	4.75	70.5	13.8	45.1	207	284	5.62	51.1
21	3	3.57	1.86	20.8	68.5	14	43.4	207	287	5.77	51.8
22	1	-	-	25.8	64.8	19.5	43.4	177.8	231.8	-	98.8
22	2	-	-	22.4	63.8	6.4	40.4	202.6	196.2	-	105
22	3	-	-	17.4	62.8	7.6	39.2	199.2	218.8	-	97.6
25	1	3.1	-	24.9	73.8	17.6	41.2	198.3	327.1	5.9	47.3
25	2	4.5	-	37.7	67.1	21.1	40.2	183.7	317.8	4.5	46.8
25	3	2.9	-	28	58	14.4	32.8	180	264.6	5	44.5
27	1	4.8	-	25.7	68.1	19.1	43.7	196	216	5	46.7
27	2	5.7	-	25.3	67.5	18.9	43.7	206	220	5.4	47.2
27	3	5.7	-	26.9	69.8	18.9	44	198	222	4.7	48.5
28	1	3	-	30	98.6	16.6	42.4	160	255.7	3.9	49
28	2	3	-	29	99.1	13.6	40.3	148.5	240.9	3.7	48.3
28	3	3	-	31	92	14.8	40.8	162.2	249.6	3.7	52.6
29	1	3.19	2.44	34.2	74.9	21.1	46.1	178.5	319	11.5	60.6
29	2	3.21	2.43	33.3	76.9	23.8	46.6	201.8	351.7	12.2	63.6
29	3	3.97	2.61	35.5	80	22.1	48.2	190.9	329.5	13.3	62
35	1	5.5	1.4	26	58	16	41	137	244	8.3	39
35	2	5.6	1.2	22	58	15	32	127	291	7.9	44
35	3	5.2	1.4	24	63	15	34	128	278	9.1	44
36	1	4.44	-	21.5	68	12.6	42	194	265	5.64	49.8
36	2	4.4	-	21.4	71	13.1	41.9	176	243	5.8	50.9
36	3	4.47	-	21.9	70.6	13.1	43.8	182	240	5.68	49.8
37	1	3.2	0.6	23.8	65.1	15	39.4	157	280	-	47.5
37	2	3.3	0.7	24.7	65.4	14.6	39.7	160	285	-	48.7
37	3	3.3	0.7	24.2	65.9	14.9	40.5	163	287	-	47.8
39	1	14	1.6	40	86	19	47	200	270	4.6	52
39	2	17	1.8	31	93	20	59	210	280	7.5	52
39	3	17	2	32	93	20	47	210	280	7.9	59
40	1	-	-	34	92	22	53	219	282	11	54
40	2	-	-	35	95	22	55	228	319	10	60
40	3	-	-	34	96	24	63	249	333	11	66
43	1	3.5	1.7	23.3	72	17.8	42.3	190	260.1	6.2	45.6
43	2	3.2	1.4	19.4	68.5	16.6	38.8	177.4	241.2	5.1	41.6
43	3	2.3	1.2	19.9	68.1	18.6	39.7	180.2	247.5	5.4	42.4
45	1	6.8	-	23.3	77	17.1	45.9	154.9	263.6	5.6	39.2
45	2	8.4	-	24.4	74	17.1	44.4	162.2	276.9	7.1	44.1
45	3	9	-	24.4	74	14.9	42.6	138.6	243.6	5.6	35.9
46	1	2.89	-	24.3	59.1	15.8	33.6	186	264	6.79	39.5
46	2	3.07	-	25.2	61.6	14.9	35.8	188	266	5.09	36.1
46	3	2.88	-	24.3	59.3	14.3	35.3	179	271	7.12	37.4
47	1	11.9	-	43.4	69.8	16.4	54.2	133.3	161.8	9.6	52
47	2	11	-	41.6	71.1	15.5	50.1	132.2	165.8	8.9	51.1
47	3	11	-	39.9	68.3	15.4	50.2	129.8	159	7.9	47.4

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
48	1	7.2	-	17.6	37.4	12.7	50.2	160.8	125.2	4.8	34.7
48	2	4.3	-	16.6	37.8	10.5	44	140.7	111.5	3.8	27.3
48	3	5.1	-	16.9	36.7	10.2	50.1	149.4	121.3	3.7	29.2
49	1	6	-	25	67	28	28	197	265	11	49
49	2	6	-	28	75	27	33	211	304	16	53
49	3	5	-	28	69	27	32	215	315	15	49
50	1	4	0.7	24.7	67.1	7.9	48.4	193.2	239	-	49.8
50	2	4.2	0.9	22.9	65.3	7.6	45.7	184.1	228.6	-	47.8
50	3	4	0.8	24.2	68.5	7.6	47.1	190.6	241.6	-	49.8
51	1	-	-	29	73	16	50	174	235	9	48
51	2	-	-	28	70	15	48	167	227	8	46
51	3	-	-	-	-	-	-	-	-	-	-
52	1	-	-	23.62	56.89	13.98	38.1	195.48	283.18	6.7	44.84
52	2	-	-	22.35	58.23	14.49	37.84	189.37	278.5	6.82	43.26
52	3	-	-	21.47	59.81	14.9	39.34	182.99	258.6	7.02	44.89
54	1	8.7	-	29.2	88.7	-	48	228.8	256.4	-	50.5
54	2	9	-	27	90.1	-	47.6	221	252.2	-	49.5
54	3	9.3	-	29.3	89.3	-	49.2	234.6	261.5	-	50.7
57	1	6.53	-	23.1	52.6	14.1	31	76	110	4.38	13.7
57	2	7.17	-	24.2	51	12.6	29.6	77.7	117.6	4.23	12.7
57	3	7.1	-	23.7	49.6	11.5	27.9	79.6	109.1	3	10.5
59	1	2.6	0.5	23.6	60.2	16.3	40.1	207.5	244.6	8.6	49.6
59	2	2.8	0.8	22.4	59.8	15.8	38.7	208.4	246	7.4	48.4
59	3	2.4	0.7	21.5	62.8	15.7	39.6	217.8	248.6	7.6	48.8
62	1	3.1	-	32.9	83.7	12.9	56.2	144	283	8.51	50.6
62	2	3	-	34.2	83.6	12.3	55.7	137	292	8.16	48.1
62	3	3.1	-	34.1	85.5	12.2	58.3	136	288	7.94	47.3
63	1	17.1	5.61	21.5	89	12.7	44.9	142	269	5.51	47.1
63	2	16.6	5.69	21.3	88.1	12.9	43.6	144	260	5.3	46
63	3	17	5.79	23.9	88.2	13.1	44	147	267	5.53	47.3
66	1	4.9	-	30.6	82.9	24.3	53	207.6	259.5	11.1	40.8
66	2	4.6	-	30.5	83	21.5	51.1	204.7	263.3	10.7	40.3
66	3	4.3	-	30	80.5	24.8	56.2	206.3	247.2	11.7	41.8
67	1	-	-	45.4	51.4	-	35.8	105	105	8.81	32.6
67	2	-	-	30	45	-	32.4	94.2	116	7.2	25.8
67	3	-	-	32.2	47.6	-	46.6	102	100	9.32	29
68	1	4.5	-	20.7	52	16.3	40.5	183.9	261.8	7.7	44
68	2	5	-	21.1	52.4	16.8	41.1	175.6	264.3	8	44.5
68	3	4.7	-	22.1	49.5	15.3	37.2	171.4	264.8	7.5	42
69	1	22.79	-	27.99	80.97	19.74	48.88	218.26	288.45	9.08	59.21
69	2	25.55	-	31.38	83.05	20.11	48.33	230.85	296.65	18.07	62.63
69	3	28.21	-	27.93	82.29	23.99	48.2	215.94	287.15	19.21	61.48
70	1	1.93	-	19.92	52.38	26.96	40.95	187.01	207.42	5.69	50.01
70	2	2.25	-	19.43	50.02	32.75	39.94	172.21	185.43	6.27	48.16
70	3	2.13	-	19.49	50.6	30.63	41.27	184.63	197.49	6.71	50.01
72	1	-	-	28.7	71.8	19.2	42.5	186.2	275.1	-	52.75
72	2	-	-	28.1	71.5	19.1	43.2	174.5	254.6	-	48.7
72	3	-	-	29.6	73.3	20.9	45.6	189.2	255.9	-	53.1
73	1	3.7	-	28	80	16	57.9	218	295	9.58	59.2
73	2	3.73	-	28.2	85.3	15.5	55.5	218	298	9.11	58.2
73	3	3.67	-	27.8	77.6	15.8	55.3	215	294	9.54	60
78	1	-9	-	-	10	-	14	38	50	-	17
78	2	-9	-	-	12	-	11	35	41	-	13
78	3	-9	-	-	12	-	16	48	51	-	16
79	1	5.47	4.37	14.6	51.3	13.5	32.1	104	152	6.4	33
79	2	3.51	1.32	20.8	64.9	15.9	40.9	140	206	7.05	39.5
79	3	3.88	1.46	20.9	70.7	17.6	49.7	147	210	6.84	44.4
80	1	11.4	-	29	76.9	25.7	52	230.4	303.9	36	92.7
80	2	15.3	-	31.3	84.5	27	51.4	221.3	305.4	53.1	97.7
80	3	9.1	-	28.7	85.3	15.8	44.4	215.8	309.7	49.9	106.1
81	1	3.8	0.31	24.9	76.3	19.1	48.6	196.5	266.7	5.3	47.1
81	2	3.7	0.48	25	76.6	19.6	44.4	209.5	272	6.1	50.3
81	3	3.6	0.4	26.2	81.5	18.6	48.6	198.4	277.4	6.9	51
85	1	4.95	-	25	72.6	18.2	57.3	238.8	350.3	6.97	55.6
85	2	1.7	-	26.8	77.7	12.8	64.1	256.2	395.3	9.12	59.2
85	3	2.54	-	24.7	79.9	17.4	60.5	225.4	305.3	7.19	56.2
87	1	7	6	30	71	6	59	321	315	11	111
87	2	-	-	-	-	-	-	-	-	-	-
87	3	-	-	-	-	-	-	-	-	-	-
88	1	-	-	18.8	51.2	-	62	169	264.4	25.5	43.8
88	2	-	-	18	50.4	-	63.8	178.4	264.3	26.9	42.1
88	3	-	-	23.9	50.6	-	66.4	191	278.6	36.6	48

Table 8. RESULTS OF THE ANALYSIS OF AMPOULE C (SEDIMENT EXTRACT).

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
2	1	1.4	1	0.5	1.2	0.8	1.4	2.3	2.4	0.2	1.1
2	2	1.4	1	0.6	1.3	0.9	1.3	2.4	2.6	0.2	1.1
2	3	1.4	1	0.6	1.3	0.9	1.5	2.4	2.6	0.3	1
6	1	-	-	1.07	1.74	-	1.89	2.74	2.47	0.44	1.23
6	2	-	-	1.06	1.6	-	1.79	2.73	2.34	0.37	1.21
6	3	-	-	1.06	1.76	-	1.96	2.91	2.52	0.47	1.35
7	1	2.71	-	-	1.42	1.79	2	2.38	2.07	-	0.98
7	2	3	-	-	1.53	-9	2.05	2.39	2.22	-	1.02
7	3	2.54	-	-	1.46	1.24	2.12	2.53	2.27	-	1.08
10	1	1.19	1.03	0.59	1.15	0.56	1.33	1.73	1.97	0.14	0.76
10	2	1.15	1	0.56	1.06	-9	1.5	1.65	2.1	0.18	0.8
10	3	1.16	0.98	0.54	1.09	0.46	1.44	1.77	1.96	0.16	0.85
11	1	1.49	1.02	0.88	1.28	0.64	1.73	2.41	2.74	-	1.2
11	2	1.49	1.01	0.93	1.16	0.65	1.61	2.41	2.63	-	1.24
11	3	1.4	0.99	0.84	1.15	0.62	1.61	2.37	2.59	-	1.24
12	1	-	-	2.16	3.09	-	2.9	3.39	3.41	-	1.41
12	2	-	-	1.7	2.71	-	2.78	3.39	3.31	-	1.47
12	3	-	-	1.57	2.58	-	2.71	3.3	3.28	-	1.47
13	1	3	-	-	4	-	3	5	4	-	2
13	2	2	-	1	2	-	2	4	4	-	2
13	3	-	-	-	-	-	-	-	-	-	-
14	1	1.33	0.87	0.8	1.41	0.64	1.7	2.49	2.23	0.22	1.1
14	2	1.34	0.88	0.7	1.38	0.61	1.6	2.36	2.18	0.23	1.08
14	3	1.38	0.9	0.6	1.41	0.58	1.51	2.46	2.3	0.21	1.08
16	1	1.7	-	21	1.6	-	2.6	3	4.7	-	3.5
16	2	0.4	-	19.6	0.7	-	1.7	3.7	1.3	-	4.6
16	3	2	-	20	1.3	-	1.9	2.8	4.2	-	3.4
18	1	1.45	1.27	10.7	1.28	1.27	2.5	2.5	2.15	0.3	1.3
18	2	1.68	1.31	11	1.4	1.25	1.4	2.7	2.39	0.3	1
18	3	0.91	0.85	10.6	1.75	0.9	0.8	1.5	2.7	0.2	1
21	1	1.26	0.99	0.78	1.29	0.43	1.5	2.38	2.35	0.19	1.06
21	2	1.25	0.98	0.76	1.3	0.43	1.58	2.36	2.32	0.19	1.06
21	3	1.22	0.97	0.76	1.28	0.44	1.53	2.35	2.38	0.17	1.05
22	1	2.5	-	0.5	1.1	-	1.1	2	3.3	-	1
22	2	2.7	-	0.4	1.4	0.5	1.1	2.3	2.9	-	1.6
22	3	3.9	-	0.6	1.4	0.4	1.4	2.8	2.9	-	1.9
23	1	2.5	2.3	0.8	3	2.9	2.9	3	2.4	2.2	3
23	2	1.2	0.8	0.8	3.5	1.6	3	3	4.3	0.8	1.6
23	3	1.8	1.6	2.4	5	2.6	5.5	4	5.6	-	2.7
24	1	1.3	1	-	-	-	2.2	-	2.2	-	1.2
24	2	1.3	1	-	-	-	2.2	-	2.3	-	1.2
24	3	1.3	1	-	-	-	2	-	2.4	-	1.2
27	1	1.2	0.4	0.5	0.9	0.4	1.2	2.3	1.7	-	0.6
27	2	1	0.3	0.5	1	0.4	1.2	2.7	1.9	-	0.8
27	3	1.2	0.4	0.5	1	0.4	1.2	2.5	1.8	-	0.7
28	1	1.2	-	1.2	1.6	0.7	1.8	2.8	3.5	0.16	1.34
28	2	1.3	-	1	1.8	0.65	1.9	2.9	3.7	0.16	1.7
28	3	1.4	-	1.1	1.8	0.64	1.9	3	3.8	0.17	2
33	1	-	-	2.5	-	-	-	-	-	-	-
33	2	-	-	3.7	-	-	-	-	-	-	-
33	3	-	-	2.4	-	-	-	-	-	-	-
35	1	1.2	0.86	0.73	1.3	0.43	1.5	2	2.1	0.45	0.9
35	2	1.4	0.9	0.8	1.5	0.59	1.8	2.4	2.3	0.64	1.1
35	3	1.3	0.94	0.78	1.4	0.62	1.8	2.2	2.3	0.67	1.1
36	1	-	-	-	-	-	1.6	2.71	2.52	-	1.18
36	2	-	-	-	-	-	1.57	2.84	2.62	-	1.21
36	3	-	-	-	-	-	1.5	2.58	2.54	-	1.12
37	1	1.4	1	0.7	1.2	0.5	1.4	1.9	2.5	-	1.1
37	2	1.4	1	0.7	1.2	0.5	1.4	1.9	2.5	-	1.1
37	3	1.3	1	0.7	1.2	0.5	1.4	1.9	2.6	-	1
39	1	-	-	-	-	-	-	-	-	-	-
39	2	1.8	1.1	1	2	0.72	1.7	3	2.9	0.15	1.1
39	3	1.8	1.1	1	2	0.75	1.8	3.1	3.1	0.22	1.3
40	1	1.2	0.5	1.4	2.6	1.4	2	2.7	2.1	0.4	0.9
40	2	1.2	0.4	1.4	2.5	1.4	2.1	2.8	2.3	0.4	1
40	3	1.2	0.5	1.4	2.5	1.6	2.3	3.2	2.6	0.4	1.1
43	1	1.3	0.8	0.6	1.3	0.6	1.3	2.1	1.9	0.2	0.8
43	2	1.2	0.8	0.6	1.3	0.7	1.4	2.1	1.9	0.2	0.8
43	3	1.3	0.8	0.6	1.3	0.9	1.4	2.1	1.9	0.2	0.8
45	1	1.2	0.7	0.6	1.3	0.7	1.7	2.1	2.4	0.3	1.2
45	2	1.1	0.6	0.6	1.2	0.8	1.6	2.4	2.8	0.3	1.2
45	3	1.2	0.6	0.6	1.5	0.7	1.6	2.6	3	0.3	1.4
47	1	0.63	0.13	0.58	0.97	0.58	1.01	1.36	1.27	0.12	0.55
47	2	0.67	0.15	0.58	0.82	0.61	0.97	1.31	1.32	0.12	0.55
47	3	0.66	0.13	0.65	0.87	0.6	0.94	1.31	1.28	0.16	0.51

LAB. NO.	INJ. NO.	CB28	CB31	CB52	CB101	CB105	CB118	CB138	CB153	CB156	CB180
48	1	1.2	1.3	0.8	1.2	1.3	0.6	0.8	1.3	0.3	0.7
48	2	1	0.6	0.6	1.2	0.9	0.9	0.9	1.3	0.2	0.7
48	3	1	0.6	0.6	1.3	0.9	0.7	0.6	1.2	0.2	0.6
49	1	2	0.6	0.6	1.3	2.2	2.5	3	4.3	0.8	0.9
49	2	1.3	0.8	0.6	1	1.7	2.4	3.6	4.1	1.1	0.9
49	3	1.2	0.7	0.5	1.1	1.9	2.4	3.5	4	1	0.9
50	1	1.4	1	0.8	1.5	-	1.5	2.5	2.3	-	1.1
50	2	1.4	1.1	0.7	1.4	-	1.6	2.6	2.3	-	1.1
50	3	1.4	1	0.8	1.5	-	1.7	2.6	2.4	-	1.2
51	1	1.1	0.7	0.6	1.1	-	1.4	2.1	2	0.2	0.9
51	2	1.1	0.7	0.6	1	-	-	-	-	0.2	-
51	3	1	0.7	-	-	-	-	-	-	0.2	-
52	1	1.04	-	-	1.08	-	1.47	2.45	2.31	-	1
52	2	1	-	-	0.97	-	1.43	2.37	2.3	-	0.96
52	3	1	-	-	-	-	1.35	2.14	2.03	-	0.88
53	1	3.3	-	2.2	0.7	-	2.8	5.1	2.5	-	7.2
53	2	3.2	-	2	0.7	-	2.8	5.5	2.1	-	6.1
53	3	3.2	-	2	0.8	-	3	5.5	2.2	-	6.3
54	1	3.07	-	0.96	1.95	-	1.92	3.09	2.5	-	1.05
54	2	3.03	-	0.95	1.95	-	1.92	3.18	2.53	-	1.05
54	3	3.06	-	0.88	2	-	2.12	3.47	2.72	-	1.12
57	1	1.38	-	-	1.26	0.81	1.63	1.68	1.71	-	0.54
57	2	1.47	-	-	1.53	0.77	1.76	1.52	1.49	-	0.42
57	3	1.55	-	-	1.31	0.82	1.74	1.56	1.5	-	0.41
58	1	1.06	1.21	0.74	1.43	0.54	1.84	2.31	2.63	0.19	1.11
58	2	1.03	1.2	0.73	1.37	0.56	1.74	2.37	2.64	0.19	1.06
58	3	0.96	1.14	0.71	1.38	0.57	1.74	2.32	2.67	0.2	1.08
59	1	1.12	0.81	0.45	1.04	0.5	1.44	2.46	3.03	0.22	1.02
59	2	1.01	1.01	0.47	0.98	0.45	1.42	2.52	2.99	0.18	0.99
59	3	0.98	0.87	0.48	0.9	0.47	1.37	2.33	2.82	0.19	0.93
62	1	1.28	1.11	0.69	1.22	0.49	1.55	1.85	2.52	0.51	0.96
62	2	1.26	1.09	0.73	1.21	0.5	1.59	1.79	2.62	0.5	0.95
62	3	1.3	1.11	0.68	1.2	0.51	1.62	1.81	2.47	0.52	0.95
63	1	1.33	0.88	0.64	1.04	1.24	1.43	2.15	2.2	0.14	1
63	2	1.31	0.85	0.56	1.11	1.29	1.39	2.19	2.21	0.17	1
63	3	1.37	0.84	0.59	1.06	0.32	1.33	2.17	2.24	0.15	0.99
66	1	1.7	1.2	0.9	2	1.5	2.3	3.4	3.2	-	1.1
66	2	1.7	1.3	-	2	1.7	2.5	3.7	3.5	-	1.2
66	3	1.5	1.1	0.8	1.7	1.2	2	3	2.7	-	0.9
67	1	-	-	3.1	2.7	-	2.7	3.5	3.2	0.2	1.9
67	2	-	-	3.1	2	-	2.6	3.1	2.9	0.2	2.2
67	3	-	-	3.6	4	-	4	5.5	3.3	1	2.2
72	1	1.11	0.9	0.64	1.19	0.81	1.42	2.25	2.18	0.39	1.25
72	2	0.97	0.8	0.61	1.08	0.72	1.26	1.98	1.82	0.25	-
72	3	1.11	0.9	0.67	1.21	0.88	1.44	2.22	2.11	0.44	1.6
79	1	1.2	1.25	-	1.59	1.56	1.78	3.5	4.52	-	1.48
79	2	2.24	2.42	-	2.53	1.4	2.16	3.46	4.18	-	2.17
79	3	0.75	1.31	-	1.88	1.28	2.33	3.03	4.17	-	1.6
81	1	1.63	0.75	0.7	1.23	0.46	1.36	3.43	3.07	0.18	1.15
81	2	1.57	0.93	0.69	1.3	0.51	1.49	3.41	2.99	0.21	1.19
81	3	1.66	0.78	0.7	1.47	0.48	1.51	3.72	3.53	0.27	1.25
83	1	0.664	1.25	0.562	0.98	0.373	1.16	1.88	2.01	0.108	1.03
83	2	0.658	1.22	0.565	1.111	0.43	1.17	2.16	2.4	0.134	1.1
83	3	0.675	1.17	0.579	1.15	0.449	1.16	1.88	2.06	0.157	1.08
85	1	1.22	0.88	0.42	1.01	0.8	2.4	1.79	1.66	0.295	1.66
85	2	1.67	1.27	0.81	1.69	1.45	4.05	2.99	2.67	0.625	1.67
85	3	1.3	1.04	0.93	1.22	1.11	2.97	2.4	2.3	0.614	1.14
87	1	7	33	66	11	8	14	57	48	2	18
87	2	8	31	64	14	7	16	54	46	2	17
87	3	8	35	66	15	8	15	54	49	2	16
88	1	-	-	4.3	-	-	-	1.8	1.4	-	5.7
88	2	-	-	5.3	-	-	-	1.4	1.3	-	5.1
88	3	-	-	5.6	-	-	-	1.3	1.2	-	7.2

Table 9. RESULTS OF THE ANALYSIS OF AMPOULE Y.

LAB. NO.	INJ. NO.	COLUMN	CB28	CB31	CB118	CB153	CB180
12	1	1	-	-	335.8	193.8	132.8
12	2	1	-	-	356.5	205.1	130.9
12	3	1	-	-	350	207.4	135.9
12	1	2	-	-	128.4	202.5	120.3
12	2	2	-	-	124.2	203.6	119.1
12	3	2	-	-	124.2	203.6	119.1
21	1	1	40	43.7	49	74.5	49.5
21	2	1	39.1	42.5	50	77.4	49
21	3	1	40.1	43.9	50.7	78.9	50.7
21	1	2	-	-	50.3	78.4	51.1
21	2	2	-	-	50.3	78.4	51.7
21	3	2	-	-	50.4	78.1	51.6
24	1	1	39.3	39.1	92.5	75.2	40.6
24	2	1	39	38.6	93.1	79.9	41.4
24	3	1	39.6	38.7	89.7	81.1	42.5
24	1	2	38.4	40.2	76.6	73.9	44.1
24	2	2	40	40.5	79.1	75.7	50.3
24	3	2	38.8	40	67.1	73.8	45.8
28	1	1	66.1	65.9	77.7	87	69
28	2	1	57.2	50.4	107.5	96	63.5
28	3	1	52.4	54.6	79.6	100.1	61.1
28	1	2	48.7	49	58.3	86.2	56.9
28	2	2	49.3	50.3	58.2	88.4	58.2
28	3	2	49.7	50.3	57.4	87.5	57.4
40	1	1	45	45	68	85	56
40	2	1	47	45	70	87	57.5
40	3	1	42.5	41.5	63	85	56
40	1	2	44	45	54.5	83.5	54
40	2	2	45	44.5	55	85	54
40	3	2	45	45	55	84.5	53.5
67	1	1	-	-	94.6	83.6	55.6
67	2	1	-	-	88.3	78.1	49.1
67	3	1	-	-	88.3	71.5	46
67	1	2	-	-	-	-	-
67	2	2	-	-	-	-	-
67	3	2	-	-	-	-	-
78	1	1	16	14	34	28	18
78	2	1	15	14	34	27	16
78	3	1	20	9	35	28	17
78	1	2	15	16	26	16	13
78	2	2	16	17	24	17	17
78	3	2	16	13	26	14	17
79	1	1	40.2	46.5	51	82.4	52.9
79	2	1	40.5	46.6	49.2	75.4	52
79	3	1	42	55.4	52.3	82.3	54.4
79	1	2	-	-	48.7	78.3	49.8
79	2	2	-	-	49.4	75.9	50.1
79	3	2	-	-	50.7	80	51.5
81	1	1	38.1	43.4	55.3	87.4	43.6
81	2	1	29.2	33.8	46.8	73.4	48.5
81	3	1	35.7	35.7	46.2	68	48.1
81	1	2	-	-	-	-	-
81	2	2	-	-	-	-	-
81	3	2	-	-	-	-	-
83	1	1	38.8	35.8	-	77.4	48.7
83	2	1	37.9	37.3	-	67.2	42.2
83	3	1	33.6	34	-	66.4	40.8
83	1	2	38	39.2	50.6	83.8	53
83	2	2	39.1	37.4	51.7	86.8	51.4
83	3	2	36.9	34.9	45.6	73.4	45.6
85	1	1	40.2	42	42.8	70.7	46.2
85	2	1	39.6	40.8	47.6	66.8	46
85	3	1	38	34.4	41.2	70	42
85	1	2	59	97.1	202.6	55.4	57.2
85	2	2	40.8	27.6	176.6	74.4	52
85	3	2	-	-	-	-	-
88	1	1	-	-	11.5	8.6	9.6
88	2	1	-	-	13	8.2	-
88	3	1	-	-	12.5	8.1	-
88	1	2	-	-	12.3	10.8	10.2
88	2	2	-	-	12.6	9.1	10.5
88	3	2	-	-	13.4	9.6	9.4

Table 10. Summary of results for standard solution A

a) All results (53 laboratories)

CB	mean	r	R	Sr	SR	% deviation from target values	cic
52	48.7	1.21	2.08	1.07	1.30	+12	0.93
101	51.5	1.19	1.90	1.07	1.26	- 8	0.92
118	57.7	1.22	2.24	1.07	1.33	+ 3	0.94
138	82.8	1.19	1.90	1.06	1.26	+ 1	0.93
153	76.0	1.19	1.86	1.07	1.25	- 5	0.92
180	42.8	1.17	2.30	1.06	1.35	+ 7	0.96

b) Results without outliers (46 laboratories)

CB	mean	r	R	Sr	SR	% deviation from target values	cic
52	50.0	1.19	1.56	1.07	1.17	+15	0.84
101	51.5	1.18	1.51	1.06	1.16	- 5	0.84
118	59.4	1.17	1.56	1.06	1.17	+ 6	0.87
138	85.2	1.16	1.50	1.06	1.16	+ 4	0.86
153	78.4	1.19	1.51	1.06	1.16	- 2	0.83
180	42.4	1.14	1.52	1.05	1.16	+ 6	0.90

c) Results of selected group (39 laboratories)

CB	mean	r	R	Sr	SR	% deviation from target values	cic
28	45.8	1.17	1.65	1.06	1.19	+13	0.90
31	43.2	1.13	2.23	1.04	1.33	+ 8	0.98
52	50.5	1.17	1.62	1.06	1.19	+16	0.90
101	53.2	1.15	1.52	1.05	1.16	- 5	0.89
105	43.8	1.22	2.10	1.07	1.30	+ 5	0.93
118	58.8	1.22	1.93	1.07	1.26	+ 5	0.91
138	85.3	1.18	1.55	1.06	1.17	+ 4	0.86
153	79.2	1.17	1.56	1.06	1.17	- 1	0.88
156	39.2	1.23	1.93	1.08	1.27	- 5	0.90
180	42.0	1.15	1.58	1.05	1.18	+ 5	0.90

Table 11. Summary of results for seal blubber extract B

a) All results (45 laboratories)

CB	mean	r	R	Sr	SR	cic
52	26.0	1.59	2.43	1.18	1.37	0.73
101	71.7	1.16	2.38	1.06	1.36	0.97
118	44.5	1.24	1.81	1.08	1.23	0.87
138	177.1	1.16	2.05	1.05	1.29	0.96
153	242.9	1.20	2.33	1.07	1.35	0.95
180	48.9	1.19	2.92	1.06	1.47	0.97

b) Results without outliers (40 laboratories)

CB	mean	r	R	Sr	SR	cic
52	26.3	1.55	2.23	1.17	1.33	0.70
101	70.8	1.14	1.72	1.05	1.21	0.94
118	45.5	1.23	1.68	1.08	1.20	0.84
138	179.4	1.16	1.91	1.05	1.26	0.95
153	250.7	1.17	2.10	1.06	1.30	0.95
180	48.7	1.17	1.90	1.06	1.26	0.94

c) Selected results (35 laboratories)

CB	mean	r	R	Sr	SR	cic
52	26.2	1.57	2.35	1.17	1.36	0.72
101	70.2	1.17	1.84	1.06	1.24	0.93
105	17.85	1.37	2.47	1.12	1.38	0.88
118	43.8	1.24	1.84	1.08	1.24	0.87
138	176.4	1.16	2.07	1.05	1.30	0.96
153	246.6	1.20	2.28	1.07	1.34	0.95
156	7.7	1.44	3.87	1.14	1.62	0.93
180	46.6	1.20	2.54	1.07	1.40	0.96

Table 12. Summary of results for sediment extract C

a) All results (33 laboratories)

CB	mean	r	R	Sr	SR	cic
52	1.08	1.53	22.81	1.17	3.06	0.98
101	1.48	1.47	4.46	1.15	1.71	0.93
118	1.80	1.53	4.38	1.16	1.69	0.92
138	2.68	1.39	6.09	1.12	1.91	0.97
153	2.71	1.57	5.22	1.17	1.80	0.93
180	1.31	1.40	6.48	1.13	1.95	0.97

b) Results without outliers (28 laboratories)

CB	mean	r	R	Sr	SR	cic
52	0.78	1.58	3.47	1.18	1.56	0.86
101	1.43	1.37	2.74	1.12	1.43	0.90
118	1.63	1.38	2.75	1.12	1.43	0.90
138	2.34	1.34	2.53	1.11	1.39	0.90
153	2.43	1.35	2.15	1.11	1.31	0.85
180	1.11	1.40	2.53	1.13	1.39	0.87

Table 13. Summary of results for the extra test, solution Y

a) All results (12 laboratories)

CB	r	R	Sr	SR	cic
28*	1.27	2.90	1.09	1.46	0.95
31*	1.72	3.77	1.21	1.61	0.83
118	1.19	8.77	1.07	2.17	0.99
153	1.20	10.39	1.07	2.31	0.99
180	1.18	5.41	1.06	1.83	0.99

* 9 laboratories

b) Results without outliers (9 laboratories)

CB	r	R	Sr	SR	cic
28	1.25	1.58	1.08	1.18	0.76
31	1.71	1.82	1.21	1.24	0.20
118	1.22	2.80	1.07	2.80	0.96
153	1.21	1.35	1.07	1.35	0.60
180	1.16	1.41	1.05	1.41	0.81

Table 14. Outliers determined after a principal component analysis

	A			B			C			Y		
outliers based on deviation from target value (A,Y) or mean (B, C)	16	18	22	6	7	16	12	16	18	12	67	78
	47	51	53	18	21	47	23	47	48	88		
	62	78	80	48	57	67	53	67				
laboratories of which results were insufficient to be used for statistical treatment	24	88		2	51	78	7	13	24			
				87			33	36	39			
							51	52	57			
							66	72	79			
							88					

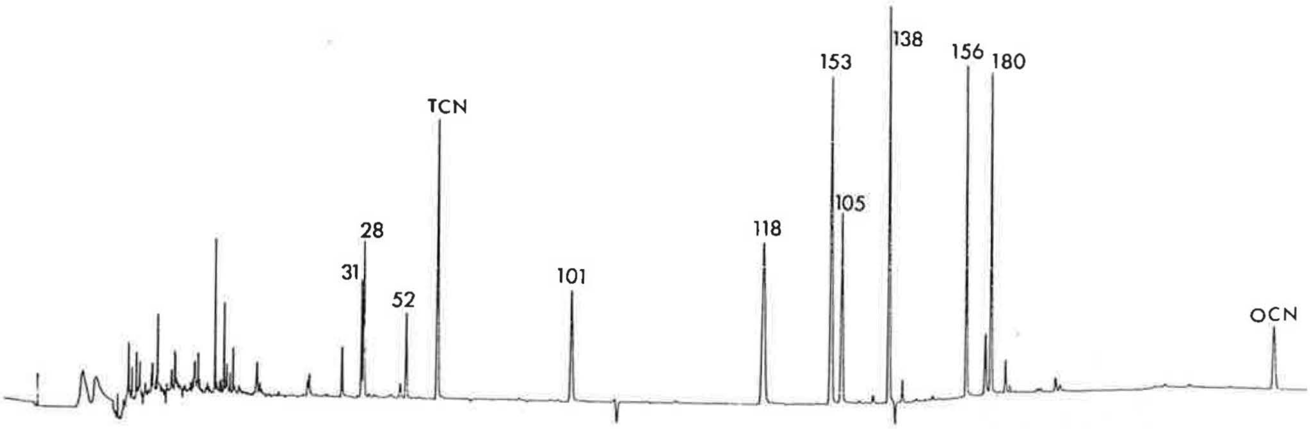


Figure 1 Chromatogram of the standard CB solution on a 50m x 0.15 mm CP-Sil 8 column.

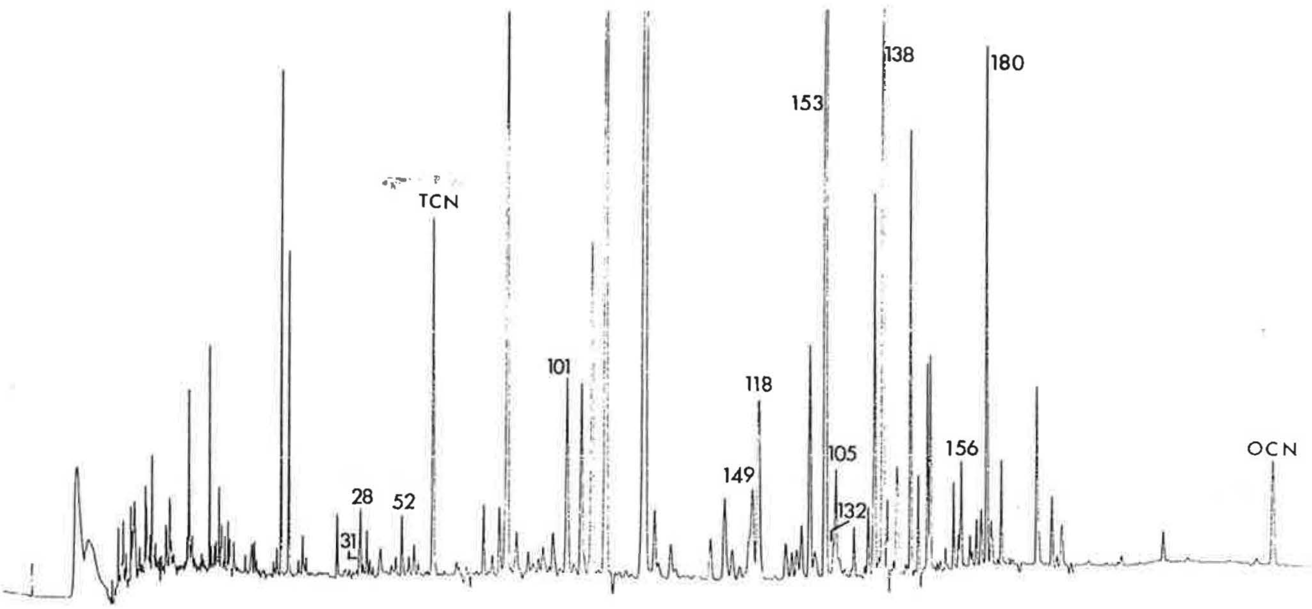


Figure 2 Chromatogram of the seal blubber extract on a 50m x 0.15 mm CP-Sil 8 column.

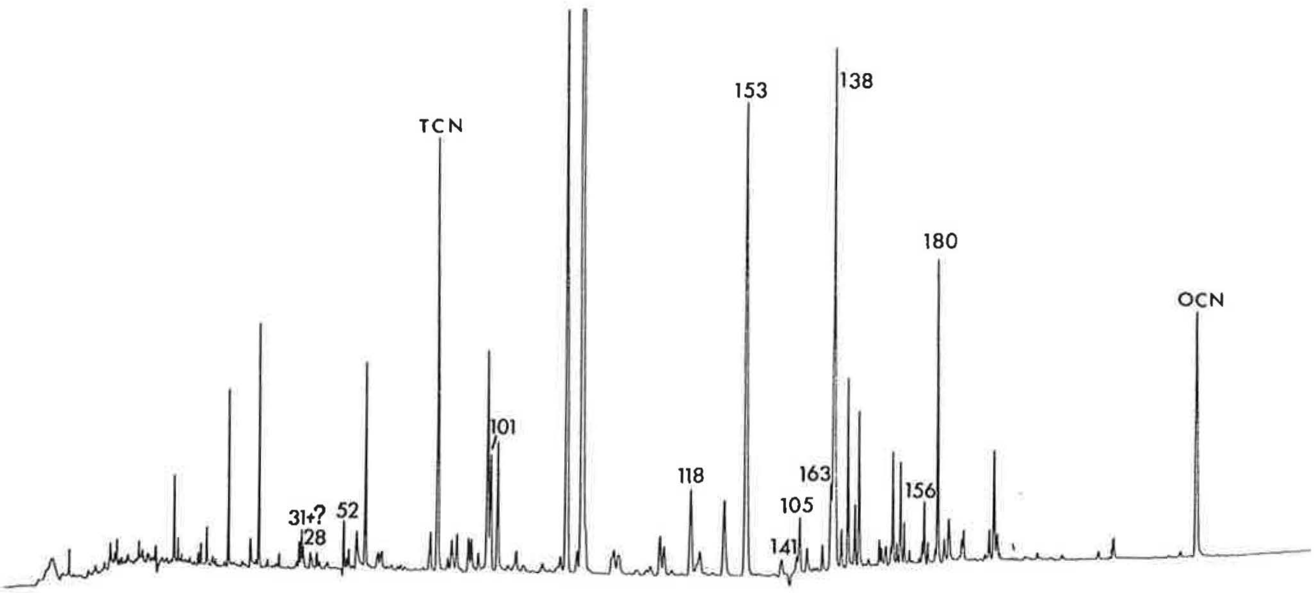


Figure 3 Chromatogram of the seal blubber extract on a 60m x 0.15 mm CP-Sil 19 column.

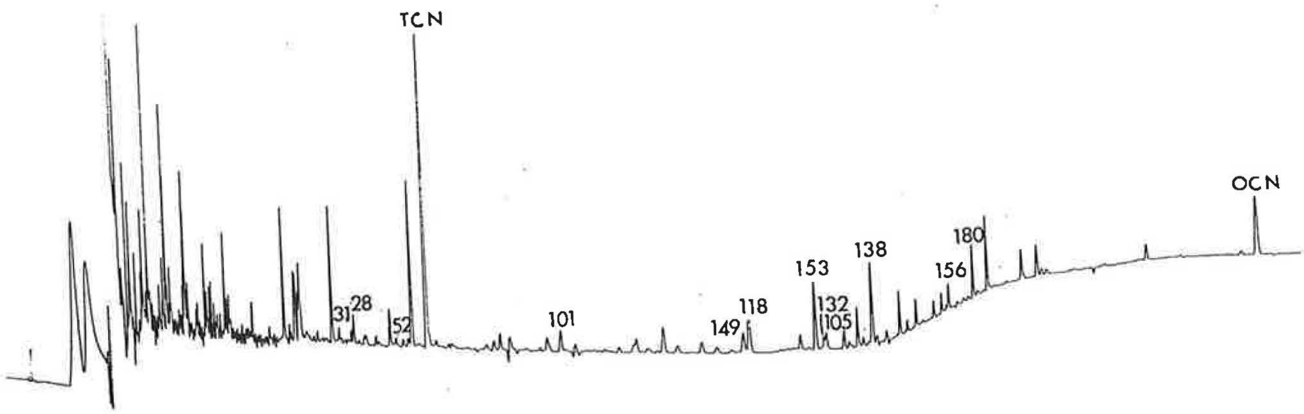


Figure 4 Chromatogram of the sediment extract on a 50m x 0.15 mm CP-Sil 8 column.

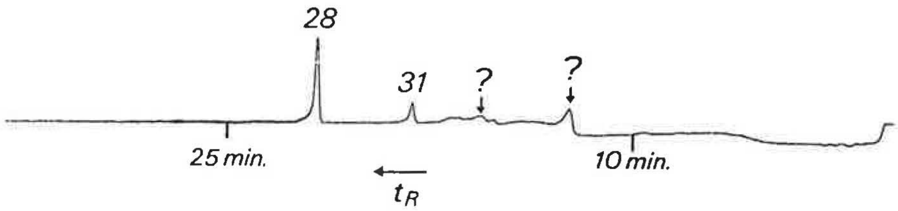


Figure 5 Heart cut of the CB 31/28 cluster from the seal blubber extract on a SB-Smectic column.

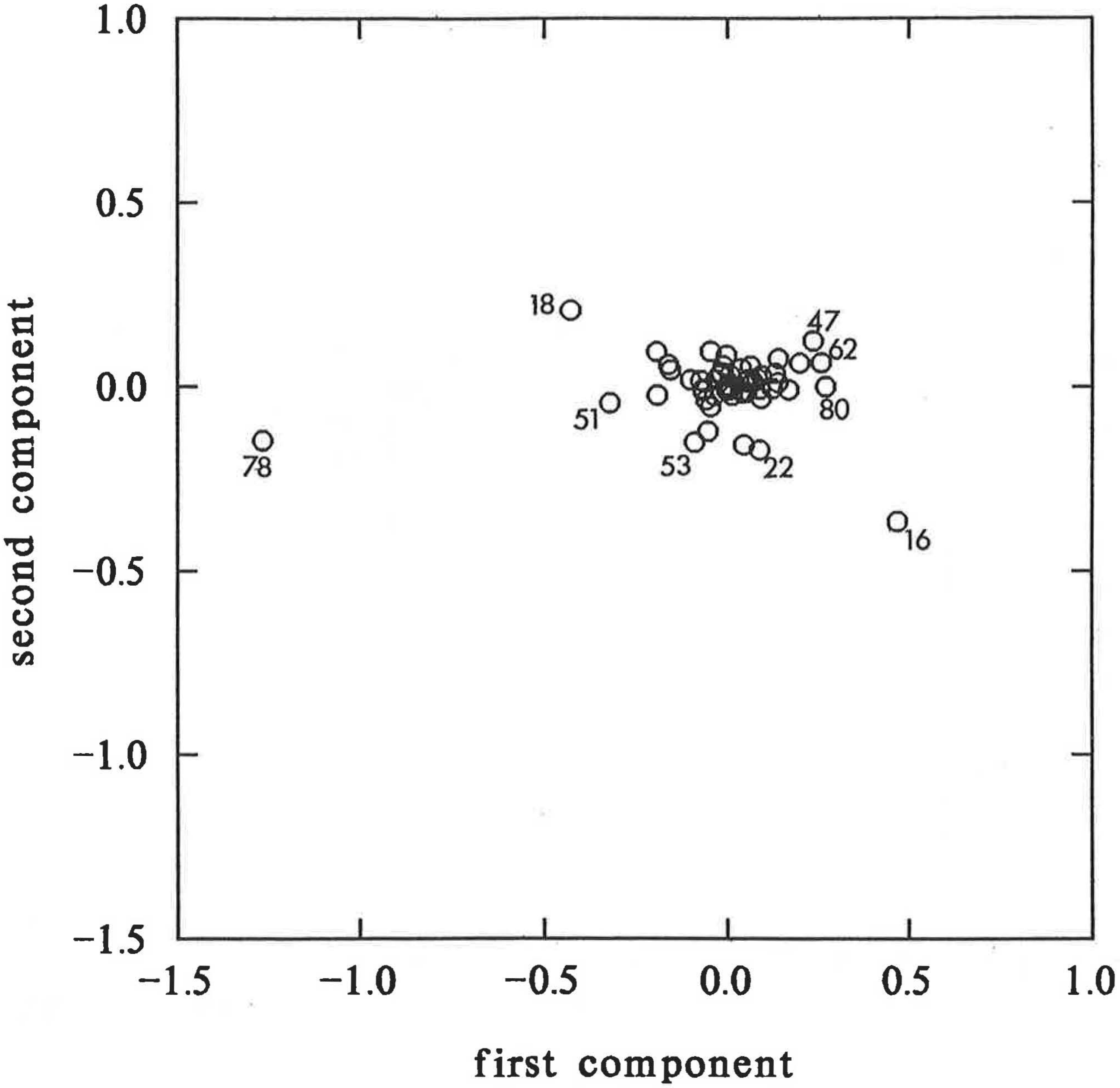


Figure 6 Principal component analysis of the unknown CB solution A

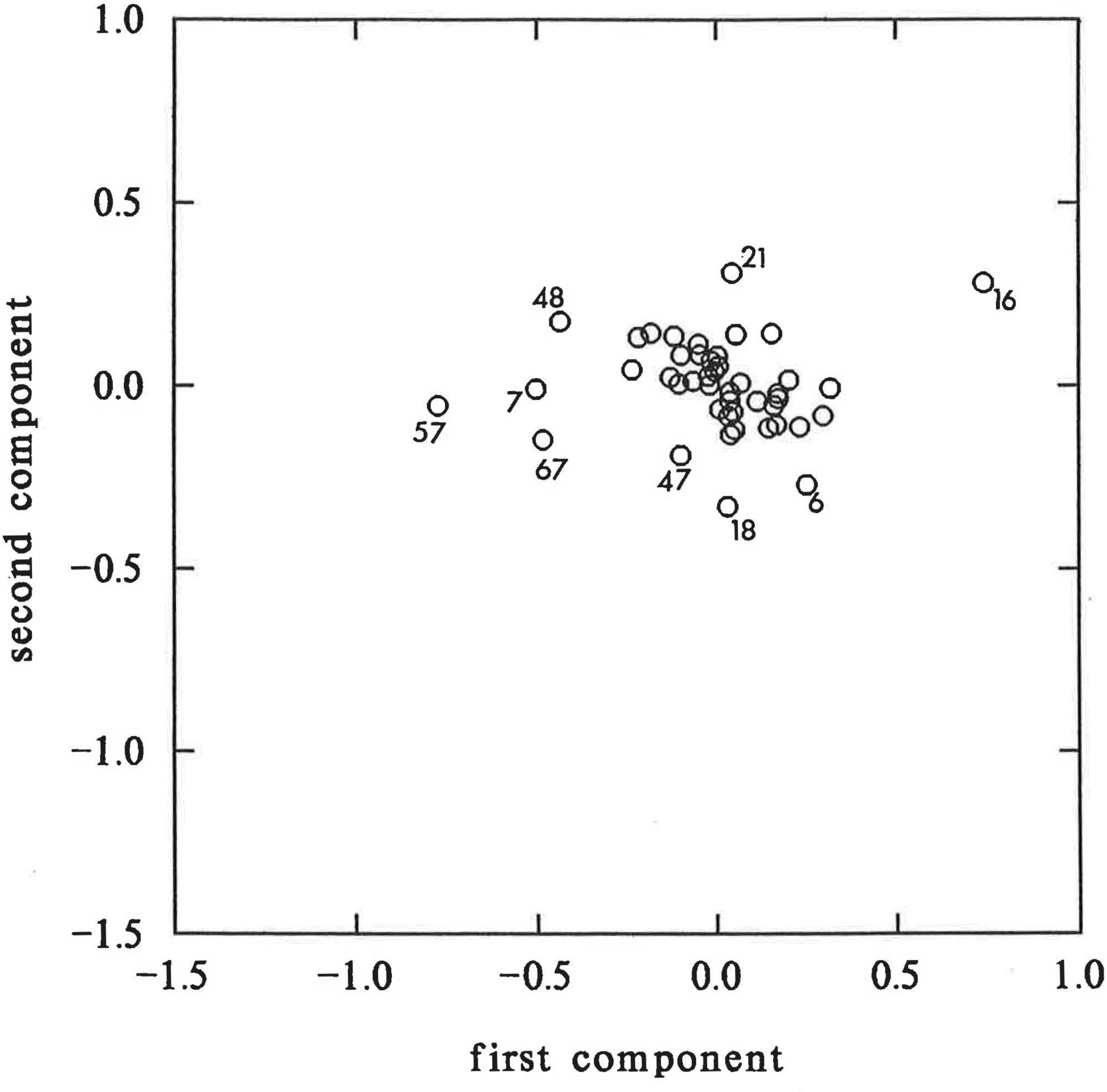


Figure 7 Principal component analysis of the seal blubber extract B

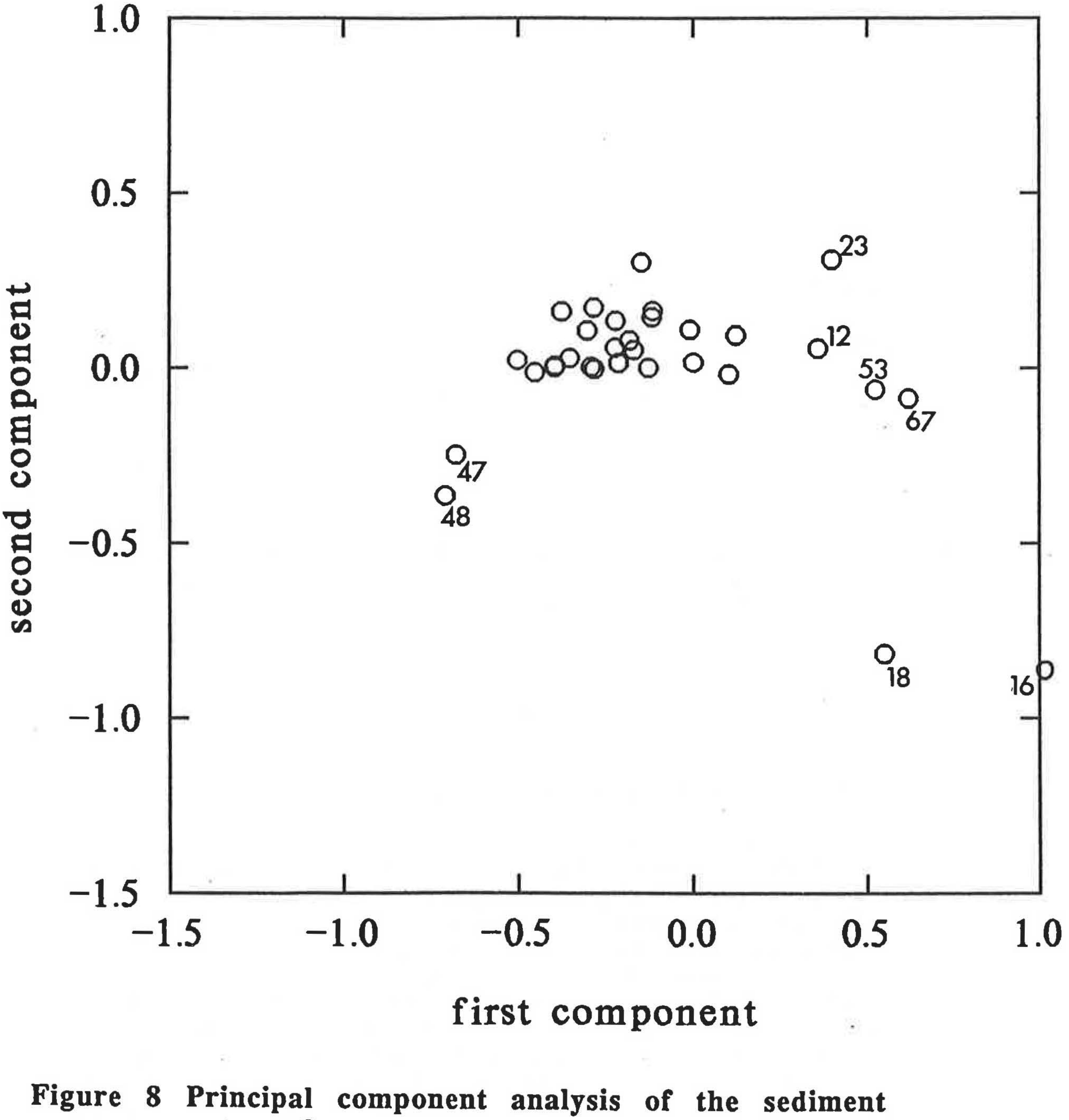


Figure 8 Principal component analysis of the sediment extract C

Annex A

GUIDE-LINES FOR THE ICES/IOC/OSPARCOM INTERCOMPARISON EXERCISE ON THE ANALYSIS OF CHLOROBIPHENYL CONGENERS IN MARINE MEDIA - 2ND STEP.

Dear participant,

Please find enclosed the following ampoules to be used for the 2nd step of the ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CBs in marine media.

1 Ampoule A:	This ampoule contains 10 CBs (no.s: 28, 31, 52, 101, 105, 118, 138, 153, 156 and 180) dissolved in about 5 ml iso-octane.
1 Ampoule B:	This ampoule contains a cleaned seal blubber extract in iso-octane.
1 Ampoule C:	This ampoule contains a cleaned sediment extract in iso-octane.
1 Ampoule D:	This ampoule contains two internal standards tetrachloronaphtalene, concentration: 2 µg/ml and octachloronaphtalene, concentration: 4 µg/ml, both dissolved in about 5 ml iso-octane.
1 Ampoule E:	This ampoule contains 5 ml iso-octane and serves as the blank.

The total weights are written on the ampoules A-D.
You will find only 4 ampoules if you had requested to analyse only seal blubber or sediment. The ampoules which should be analysed are solutions A, E and B or C. Stock solutions of individual chlorobiphenyls are > 98% purity by ECD chromatograms. However, the used CBs were not certified standards. Therefore we do not recommend the use of these standard for reference purposes.

1. Please weigh all ampoules upon receipt and report the condition of the ampoules as soon as possible, as received (annex 1).

2. For this exercise the following advice is given:

   a Use 2 capillary GC columns of different polarity. One column should be a 5% phenyl 95% dimethylsiloxane column (SE-54, CP-Sil8, DB-5, etc.). It is advised to use a more polar stationary phase for the second column, e.g. OV-17, CP-Sil 19, DB-17. The first step of this exercise confirmed that the internal diameter of the capillary columns was not critical for the analysis of standard solutions, but for the analysis of real samples the use of narrow bore columns is essential. Therefore we strongly recommend the use of columns with internal diameters of 0.25 mm or less. The analysis of the cleaned extracts on wide bore columns (> 0.25 mm) will definitely give poor separation of closely eluting CBs. It is advised also to use column lengths of at least 50 m, although the length is less critical than the internal diameter. A film thickness of 0.2-0.4 µm is also advised.

   b Hydrogen should be used as the first choice of carrier gas, but if not available, helium is acceptable.

   c The GC conditions should be optimised according to the advice given in the guide lines for the first step of this exercise. This optimization covers gas flow, injector and detector temperatures, oven temperature program, splitter

closing time in case of splitless injection, initial oven temperature of the on-column injection.
After the first step of this exercise it appeared that different participants were not able to calculate the linear gas velocity. This is done for example by an injection of dichloromethane vapour in the split mode or on column. The length of the column divided by the time between the moment of injection and the appearance of the dichloromethane peak in the chromatogram gives the linear gas velocity in cm/s. The optimum flow for the carrier gas should be set at 30-45 cm/s for helium and 25-30 cm/s for hydrogen.
Identify the linear range of your electron capture detector. If it is not possible to work within the linear range, than use a multi level calibration in the concentration range of the CBs in the extracts.
Please note the comments in paragraph 4.2 of the report on the first step of this exercise. Although a number of laboratories were not qualified as outliers, the quality of their analyses can still be improved at different points. Check all materials for contamination. In the first step many high blank values were reported. This may be due to contaminated syringes, autosamplers, autosampler vials, septa, injector liners, solvents, glassware, etc.

d Inject a fixed volume for all standards, samples and blanks. This volume should be 1 µl or less, and if possible, automatically injected.

e It is strongly recommended to use a balance for the preparation of dilutions. Iso-octane (2,2,4-trimethylpentane) is recommended as a solvent for all dilutions.

3. Complete Annex 2.

4. Prepare your own CB-standard. Weigh all the solvents necessary for dilution. It is recommended to prepare twice a CB-standard to check your own weighing. Do not use commercial CB-standard <u>solutions</u>. The concentrations are not reliable.

5. Prepare test chromatograms of the solutions A, B, and C. Decide upon concentration or dilution of these solutions and your own standard. Try to work in the linear range of your detector. Always use at least 2 different dilutions of your standard. It might be necessary to use 3 or 4 different dilutions of the standard when the total amount of injected compound cannot be brought into the linear range of the detector. Add the internal standard to all solutions, including the blank. Concentrate or dilute the blank in the same way as the samples. Inject all solutions on both columns. The solutions A, B and C and the standards should be injected three times on each column, so e.g. according to the next scheme (for both columns):
E, Standard 1, A, B, C, Standard 2, Standard 1, A, B, C, Standard 2, Standard 1, A, B, C, Standard 2.
If necessary a third and/or a fourth standard must also be injected three times.
It might be necessary to use different attenuation settings for the analyses of the different samples. Also for the seal extract and the sediment extract, the necessary standard dilutions may be different.

Measure the peak heights of the 10 CBs and the internal standards and indicate them on the chromatograms. Calculate the concentrations of the 10 CBs in the columns A, B, C and E and complete the annexes 3 and 4. Report 3 results and indicate on which column they were measured. If you have 6 equal values from both columns, select the values from one column. If one set of values differs from the other, choose the correct set of values, based on chromatographic performance. In general the lowest values will be the most reliable ones. Also indicate which internal standards have been used for calculation.In general TCN is advised to use for the first half of the chromatogram and OCN for the second half. However, on some columns there may be an interference of CBs with one of the internal standards. We leave it to your choice to decide which internal standard is the best to use.

6 Return all completed annexes <u>and chromatograms</u> **<u>before 1 December 1990.</u>**

Laboratories coded 1-55 are requested to return their results to:

J. de Boer
RIVO
P.O.Box 68
1970 AB IJMUIDEN
The Netherlands
(TEL. 31-2550 64736, FAX: 31-2550 64644)

Laboratories coded 56-90 are requested to return their results to:

J.A. Calder
NOAA, National Ocean Service
Office of Ocean Services, N/OS
Universal Building South, room 615
1825 Connecticut Ave, NW
Washington DC, 20235
USA
(TEL: 1-202 673 3803, FAX: 1-202 673 3850)

Your laboratory code is: ....

We thank you for your willing co-operation and wish you much success with your analysis.

J. Calder
J. de Boer

vdW.

ANNEX 1

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media, 2nd step.

Receipt/confirmation letter.

I acknowledge the receipt of a set ampoules for the ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media, 2nd step.
The ampoules have been received in good condition .... yes / no

Damaged: ampoules no.: ....., ..., ..., ..., ...

Loss of weight: ampoules no.: ....., ..., ..., ..., ...

I request for new ampoules coded: ....., ..., ..., ..., ...

Date: Signature

Name participant:

Name and address Institute:

Return this annex to:

J. de Boer
RIVO
P.O. Box 68
1970 AB IJmuiden
The Netherlands

ANNEX 2

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media, 2nd step.

GC conditions

COLUMN A

Apparatus (type)	:	....
EC-detector (type)	:	....
Injection volume	:	.... µl
Used injection technique	:	....
Splitter closing time	:	.... min.
Detector temp.	:	.... °C
Injector temp.	:	.... °C
Recorder range	:	.... mV
Chartspeed	:	...... mm/min.
Carrier gas	:	....
Flow carrier gas	:	...... ml/min.
Detector purge gas	:	....
Detector purge flow	:	...... ml/min
Septum purge flow	:	...... ml/min.
Split ratio	:	....
Stationary phase	:	....
Material: glass / fused silica		
Length	:	.... m.
Int. diameter	:	.... mm.
Film thickness	:	.... µm.
Chemical bonded: yes / no		
Temperature program:		

Initial temp.: ..... °C (..... min.)
1st rate: .....°C to ..... °C
Isothermal: ..... min. ..... °C
2nd rate: ..... °C/min. to ..... °C
Isothermal: ..... min. .....°C
3rd rate: ..... °C/min. to ..... °C
Isothermal: ..... min. ..... °C
Lineair gas velocity : ...... cm/s

COLUMN B

Apparatus (type)	:	....
EC-detector (type)	:	....
Injection volume	:	.... µl
Used injection technique	:	....
Splitter closing time	:	.... min.
Detector temp.	:	.... °C
Injector temp.	:	.... °C
Recorder range	:	.... mV
Chartspeed	:	...... mm/min.
Carrier gas	:	....
Flow carrier gas	:	...... ml/min.
Detector purge gas	:	....
Detector purge flow	:	...... ml/min.
Septum purge flow	:	...... ml/min.
Split ratio	:	....
Stationary phase	:	....
Material: glass / fused silica		
Length	:	.... m.
Int. diameter	:	.... .mm.
Film thickness	:	.... µm.
Chemical bonded: yes / no		
Temperature program:		

Initial temp.: .....°C (..... min.)
1st rate: ..... °C to ..... °C
Isothermal: ..... min. ..... °C
2nd rate: ..... °C/min. to ..... °C
Isothermal: ..... min. ..... °C
3rd rate: ..... °C/min. to ..... °C
Isothermal: ..... min. ..... °C
Lineair gas velocity : ...... cm/s

Laboratory code: ....

ANNEX 3

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CBs in marine media, 2nd step.

Results of the analysis of ampoule A (standard solution).

		Concentrations of CBs in pg/µl				
CB	1st inj.:	2nd inj.:	3rd inj.	mean	column	int.st.
28						
31						
52						
101						
105						
118						
138						
153						
156						
180						

Results of the analysis of ampoule B (seal blubber extract)

		Concentrations of CBs in pg/µl				
CB	1st inj.:	2nd inj.:	3rd inj.	mean	column	int.st.
28						
31						
52						
101						
105						
118						
138						
153						
156						
180						

Please complete all columns.

ANNEX 4

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CBs in marine media, 2nd step.

Results of the analysis of ampoule C (sediment extract)

		Concentrations of CBs in pg/µl				
CB	1st inj.:	2nd inj.:	3rd inj.	mean	column	int.st.
28						
31						
52						
101						
105						
118						
138						
153						
156						
180						

Results of the analysis of ampoule E (blank)

		Concentrations of CBs in pg/µl				
CB	1st inj.:	2nd inj.:	3rd inj.	mean	column	int.st.
28						
31						
52						
101						
105						
118						
138						
153						
156						
180						

Please complete all columns.

Annex B

ICES/IOC/OSPARCOM INTERCOMPARISON ON THE ANALYSIS OF CB'S IN MARINE MEDIA - GUIDE LINES FOR EXTRA TEST ON THE QUANTIFICATION OF CB'S IN STANDARD SOLUTIONS.

Dear participant:

Please find enclosed the following ampoules to be used for an extra test on the quantification of chlorobiphenyls in standard solutions.

1 Ampoule X : This ampoule contains 5 CB's dissolved in about 5 ml iso-octane in the following concentrations:

CB No.	Concentration (ng/ml)
28	800
31	810
118	800
153	800
180	800

1 Ampoule Y : This ampoule contains the same 5 CB's dissolved in about 5 ml iso-octane in an unknown concentration. One or two extra CB's with an unknown identity may be added.

1 Ampoule D : This ampoule contains the internal standards octachloronaphtalene (OCN), concentration: 4 µg/ml and tetrachloronaphtalene (TCN), concentration: 2 µg/ml, dissolved in 5 ml iso-octane.

1 Ampoule E : This ampoule contains 5 ml iso-octane and serves as the blank.

The total weights are written on the ampoules X, Y and D. Stock solutions of individual chlorobiphenyls were >98% purity by ECD chromatograms. However, these standard solutions should not be used as reference standards for quantitative purposes!

1. Please weigh all ampoules upon receipt and report the condition of the ampoules as soon as possible as received (annex 1a).
2. For this exercise the following advice is given:
   a) Use 2 capillary GC columns of different polarity. One of these columns should be a SE-54 or SE-54 like column (CP-Sil 8, DB-5, etc.) (5% phenyl 95% dimethylsiloxane). The relative retention times of the 5 CB's in ampoule X on a SE-54 column are: (according to M.D. Mullin et al., 1984: High resolution PCB-analysis: Synthesis and chromatographic properties of all 209 PCB-congeners, Environ. Sci. Technol. 18, 6, 468-476).

	tr.rel. to octachloronaphtalene
CB 28	0.4031
CB 31	0.4024
CB118	0.6693
CB153	0.7036
CB180	0.8362

b) The capillary columns which you use for this exercise should have minimum lengths of 25 m (preferably, however, 50 m) and internal diameters of 0.25 mm or less. We emphasize to use these dimensions.

c) Hydrogen should be used as the first choice of carrier gas; but if not available, helium is acceptable.

d) The optimum linear gas velocity for the carrier gas should be set at:

hydrogen	: 30 - 45 cm/s
helium	: 25 - 30 cm/s

e) Inject a fixed volume for all samples, standards and blanks. This volume should be not more than 1 μl, if possible, automatically injected.

f) When using the splitless injection technique, first select the optimum injection temperature and optimum splitter closing time. To find the optimum injection temperature a test can be performed in which e.g. 5 times a solution of CB118 (or a lower chlorinated CB) and CB180 (concentration of both about 80 ng/ml) is injected at different injector temperatures. The highest ratio of CB180/CB118 will correspond with the optimum injector temperature. This optimum injector temperature will probably be around 270°C. To avoid discrimination effects it is necessary to optimize the splitter closing time. A test can be performed in which e.g. 5 times a solution of CB118 and CB180 is injected at different splitter closing times. The point at which the ratio of CB180/CB118 will not more increase with a lengthening of the splitter closing time will correspond with the optimum splitter closing time. The optimum splitter closing time is very much depending of the construction of the injector. In all cases the minimum splitter closing time must be kept at 1 minute. To optimize the injector temperature together with the splitter closing time, one might use a simplex procedure (Ref.: Anal. Chim. Acta 46 (1969) 193-206, Anal. Chemistry 45, 3(1973), 278-283). After selection of the optimum injector temperature and optimum splitter closing time the temperature program of the oven can be varied to obtain the best resolution for all CB's. The optimum initial oven temperature should be around 90°C.

g) When using the on-column technique, first select the optimum temperature program of the oven and the optimum initial temperature. Due to the variety in on-column injectors, a detailed optimization procedure cannot be given. Because often more parameters are important, the simplex procedure for optimization is strongly advised.

h) A balance should be used for the preparation of dilutions.

i) 2,2,4-Trimethylpentane (iso-octane) is strongly advised to be used as a solvent for all dilutions. Complete annex 2 a) for the optimum GC conditions.

3. Identify the linear range of your detector. Graphs as shown in annex 3 a) must be constructed for the CB's 28 and 153. For those who have not carried out this test before, instructions can be found in the report on the first step of this CB-intercomparison exercise - annex B: guide-lines, paragraph 3. The linearity test may be performed with your own standards or with dilutions of ampoule X. However, the quantity of X is limited, so be carefull when preparing dilutions.

4. Inject sample Y to prepare a test chromatogram. Select two dilutions of solution X (X1 and X2), bracketing the CB-concentrations in Y. If necessary, Y may be diluted or concentrated. Add an amount of the internal standard solution. One of the two internal standards may be used to your choice. Inject all standards, samples and blanks according to the next scheme:

day 1 : E, X1, X2, Y (column A)
day 2 : E, X1, X2, Y (column A)
day 3 : E, X1, X2, Y (column A)
day 4 : E, X1, X2, Y (column B)
day 5 : E, X1, X2, Y (column B)
day 6 : E, X1, X2, Y (column B)

Measure the peak heights of the 5 CB's and the internal standard in the standards, sample and blank. Indicate them in the chromatogram. Calculate the concentrations of the 5 CB's in the unknown sample and in the blank and complete the annex 4 a).

5. Return all completed annexes, graphs **and chromatograms before 1 December, 1990**, to:

J. de Boer
RIVO
P.O. Box 68
1970 AB IJMUIDEN.
The Netherlands
(tel. 31-255064736, facs: 31-255064644).

We thank you for your willing co-operation and wish you success with your analysis.

The Co-ordinators:

J. Calder.
J. de Boer.

/ct

ANNEX 1a

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media, extra test.

Receipt/Confirmation letter.

I acknowledge the receipt of a set ampoules for the ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media (extra test). The ampoules have been received in good condition....................yes / no.

Damaged	: ampoule no.:	.....,	.....,	.....,	......
Loss of weight	: ampoule no.:	.....,	.....,	.....,	.....
I request for new ampoules coded:		.....,	.....,	.....,	.....

Date:.............................   Signature:.............................

Name participant:

Name Institute:

Return this annex to:
Netherlands Institute for Fishery Investigations
Attn.: J. de Boer
P.O. Box 6
1970 AB IJmuiden
The Netherlands

ANNEX 2a

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media, extra test.

GC conditions

<u>COLUMN A</u>

Apparatus (type) : ....
EC-detector (type) : ....
Injection Volume : .... µl
Used inj. techn. : ....
Split. clos. time : .... min.
Detector temp. : .... °C
Injector temp. : .... °C
Recorder range : .... mV
Chart speed : .... mm/min.
Carrier gas : ....
Flow carrier gas : .... ml/min.
Detector purge gas : ....
Detector purge flow : .... ml/min.
Septum purge flow : .... ml/min.
Split ratio : ....
Stationary phase : ....
Material: glass/fused silica
length : .... m
int. diameter : .... mm
film thickness : .... µm
chemical bonded : .... yes/no
temperature program
initial temp. ....°C (..............min.)
lst rate: ....°C to .... °C
isothermal: ....min .... °C
2nd rate: ....°C/min. to .... °C
isothermal: ....min. .... °C
3rd rate: ....°C/min. to .... °C
isothermal: ....min. .... °C
lineair gas velocity: ....cm/sec.

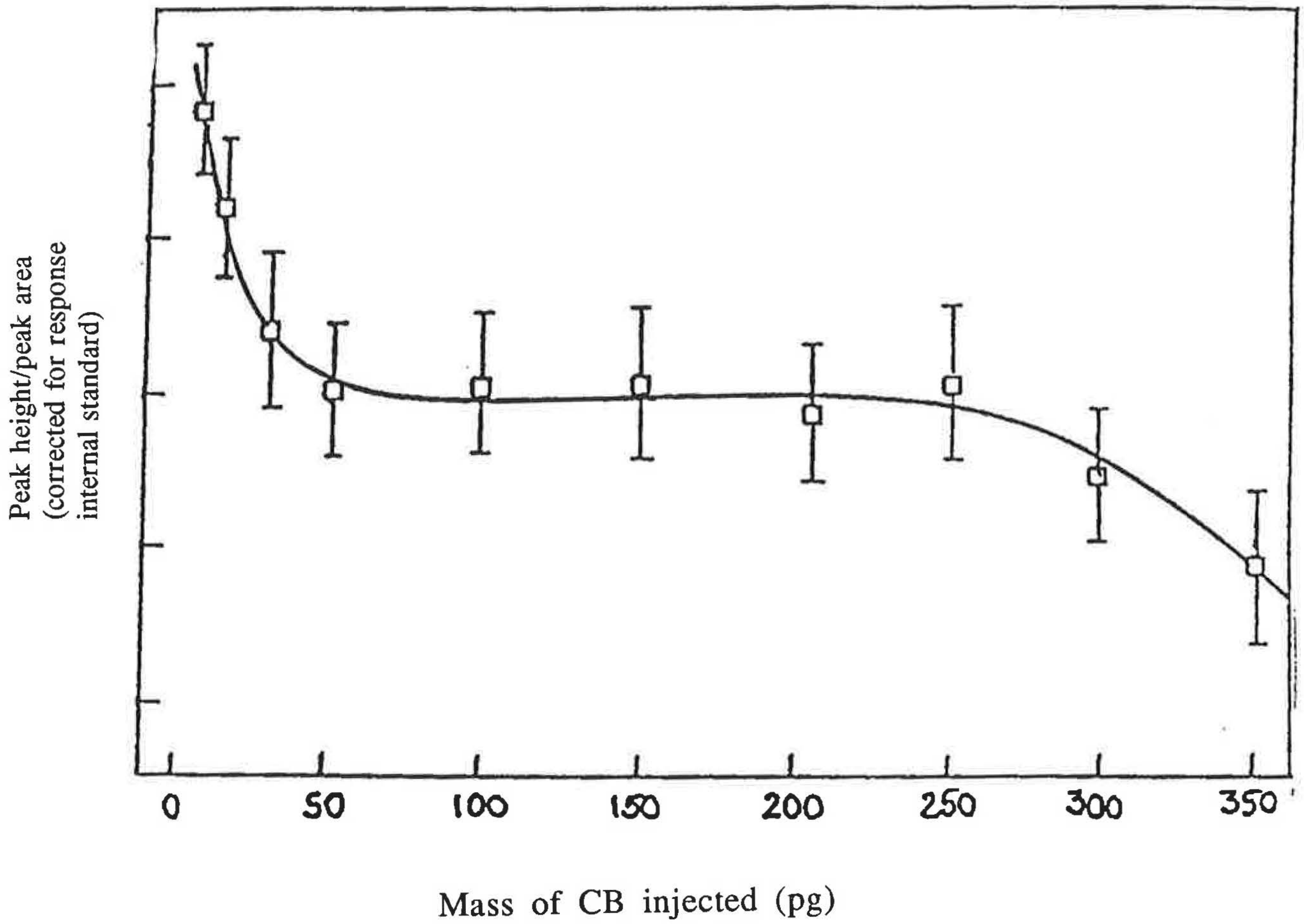
<u>COLUMN B</u>

Apparatus (type) : ....
EC-detector (type) : ....
Injection Volume : .... µl
Used inj. techn. : ....
Split. clos. time : .... min.
Detector temp. : .... °C
Injector temp. : .... °C
Recorder range : .... mV
Chart speed : .... mm/min.
Carrier gas : ....
Flow carrier gas : .... ml/min.
Detector purge gas : ....
Detector purge flow : .... ml/min.
Septum purge flow : .... ml/min.
Split ratio : ....
Stationary phase : ....
Material: glass/fused silica
length : .... m
int. diameter : .... mm
film thickness : .... µm
chemical bonded : .... yes/no
temperature program
initial temp. ....°C (..............min.)
lst rate: ....°C to .... °C
isothermal: ....min .... °C
2nd rate: ....°C/min. to .... °C
isothermal: ....min. .... °C
3rd rate: ....°C/min. to .... °C
isothermal: ....min. .... °C
lineair gas velocity: ....cm/sec.

Laboratory: ....

annex 3a

Example of a linearity response curve



ANNEX 4A

ICES/IOC/OSPARCOM intercomparison exercise on the analysis of CB's in marine media, extra test.

Results of the analysis of ampoule Y.

Concentrations of CB's in pg/µl								
	Column 1				Column 2			
CB	day 1	day 2	day 3	int. st.	day 1	day 2	day 3	int. st.
28								
31								
118								
153								
180								

Results of the analysis of ampoule E.

Concentrations of CB's in pg/µl								
	Column 1				Column 2			
CB	day 1	day 2	day 3	int. st.	day 1	day 2	day 3	int. st.
28								
31								
118								
153								
180								

Laboratory:...

Used dilutions of standard solution X: ....and ....

/ct

REPORT ON THE RESULTS OF THE INTERCOMPARISON PROGRAMME ON THE ANALYSIS OF PAHs IN MARINE MEDIA - STAGE 1

R.J. Law* and M.D. Nicholson**

Ministry of Agriculture, Fisheries and Food
Directorate of Fisheries Research

* Fisheries Laboratory
Remembrance Avenue
Burnham-on-Crouch
Essex CMO 8HA, United Kingdom

** Fisheries Laboratory
Pakefield Road
Lowestoft
Suffolk NR33 OHT, United Kingdom

SUMMARY

This report gives an account of stage 1 of the Intercomparison Programme on the Analysis of PAHs in Marine Media, which is the fourth round of the ICES hydrocarbon intercomparison programme. This exercise concerns the determination of specific polycyclic aromatic hydrocarbons (PAHs), and the first stage comprised two phases. Results were received from 14 of 19 laboratories in phase 1, and 17 of 18 laboratories in phase 2. The techniques used in the participating laboratories were capillary gas chromatography (GC) with flame-ionization detection (FID) and mass spectrometric (MS) detection, high-performance liquid chromatography (HPLC) with ultra-violet absorption and fluorescence detection, and a low-temperature (Shpol'skii) fluorescence technique.

1 INTRODUCTION

Three previous intercomparison exercises on analyses of hydrocarbons conducted under the auspices of ICES (Law and Portmann, 1982; Farrington *et al.*, 1986; Uthe *et al.*, 1986) have demonstrated that, whilst good comparability can be obtained for total hydrocarbon determinations using techniques such as ultra-violet fluorescence (UVF) spectrometry, there is a serious lack of comparability between measurements of specific hydrocarbon concentrations in different laboratories. This applies to both aliphatic and aromatic hydrocarbons, and to all sample types and analytical methods employed. Similar problems have been encountered in other such investigations (Grahl-Nielsen *et al.*, 1978; Hilpert *et al.*, 1978; MacLeod *et al.*, 1982).

These difficulties were discussed at the meeting of the ICES Marine Chemistry Working Group held in Helsinki, Finland in 1986. It was proposed at that meeting that a programme of intercomparison be undertaken with the intention of identifying the sources of errors, and reducing the errors themselves, thereby improving the general level of analytical comparability. This intercomparison programme was approved at the 1986 ICES Statutory Meeting (C.Res. 1986/2:16). A meeting of analysts interested in participating in such a programme was held at ICES Headquarters in Copenhagen, Denmark, in February 1987, at which the outline of the first stage of the programme was agreed.

2 BACKGROUND

Although improvements in analytical comparability are desirable for all matrices (water, sediment and biota), attention was to be focused initially on the determination of aromatic hydrocarbon concentrations in biota. This recognizes that in many cases improvements in procedures for these analyses would be directly applicable to the other sample types. In the absence of any coordinated monitoring effort within ICES, there were no obvious target compounds, so a primary list of aromatic hydrocarbons was compiled. These compounds were chosen primarily on the basis of their analytical and chromatographic behaviour, although in the absence of evidence for the inclusion of other compounds they *could* be used for monitoring and investigative purposes. This list consisted of seventeen polycyclic aromatic hydrocarbons (PAHs) selected from those PAHs fulfilling three basic criteria:

1) compounds with 3 to 6 fused rings;

2) containing only carbon and hydrogen (i.e., no heterocyclics); and

3) not an alkyl-substituted PAH.

The primary list comprised:

Fluorene
Phenanthrene*
Anthracene
Fluoranthene*
Pyrene*
Benz[*a*]anthracene*
Chrysene*
Benzo[*b*]fluoranthene
Benzo[*j*]fluoranthene
Benzo[*k*]fluoranthene*
Benzo[*a*]pyrene*
Benzo[*e*]pyrene*
Perylene
Benzo[*ghi*]perylene*
Indeno[1,2,3-*cd*]pyrene*
Dibenz[*a,c*]anthracene
Dibenz[*a,h*]anthracene

From this list, a subset of ten compounds was selected for use in the first stage of the intercomparison programme, and these compounds are indicated above by means of an asterisk. Although benzo[*k*]fluoranthene was initially selected for use in this exercise, benzo[*b*]-fluoranthene was ultimately actually used, for logistic reasons. All ten compounds may be analysed by both gas chromatographic (GC) and high-performance liquid chromatographic (HPLC) techniques. The first stage of the programme was intended to check instrument calibration by the analysis of standard solutions to be distributed by the coordinator. In order that the same solutions could be utilized for both GC and HPLC analyses, they were to be prepared in acetonitrile.

3 MATERIALS AND METHODS

3.1 Acetonitrile

Preliminary work had been performed at the coordinator's laboratory in Burnham-on-Crouch to confirm acetonitrile as a suitable solvent for GC analysis; its use as an HPLC solvent is routine. The tests were carried out using a Hewlett-Packard 5890a gas chromatograph with an HP 7673a autosampler. Injections were made in the on-column mode at 60 °C, via a fused silica retention gap (0.5 m x 530 μm internal diameter) onto an analytical column of 0.32 mm internal diameter (25 m, 5% phenylmethylsilicone stationary phase). Under these conditions, any of the following solvents could be used with acceptable results: pentane, hexane, dichloromethane or acetonitrile. The acetonitrile used in the preparation of the standard solutions for both phase 1 and phase 2 was of HPLC grade, and was supplied by Rathburn Chemicals, Walkerburn, Scotland.

3.2 Hexane

In phase 2 of the exercise solutions for GC analysis were circulated in hexane, which was of glass-distilled grade. This was also supplied by Rathburn Chemicals.

3.3 PAH Standards

These were supplied as follows:

Compound	CAS Registry No.	Purity	Supplier*
Phenanthrene	85-01-8	99.5%	Aldrich
Fluoranthene	206-44-0	99%	Koch-Light
Pyrene	129-00-0	>99%	Aldrich
Benz[*a*]anthracene	56-55-3	99%	Aldrich
Chrysene	218-01-9	98%	Aldrich
Benzo[*b*]fluoranthene	205-99-2	99%	Aldrich
Benzo[*a*]pyrene	50-32-8	98%	Aldrich
Benzo[*e*]pyrene	192-97-2	99%	Aldrich
Benzo[*ghi*]perylene	191-24-2	98%	Aldrich
Indeno[1,2,3-*cd*]pyrene	193-39-5	99%	BCR

*Aldrich Chemicals, Gillingham, Dorset SP8 4JL, UK.
Koch-Light Ltd., Haverhill, Suffolk CB9 8PU, UK.
EC Community Bureau of Reference, Brussels, Belgium.

3.4 Preparation of Standard Solutions

Standard solutions were prepared by sequential weighing of each of the ten pure PAHs into a single volumetric flask (for each solution), using an electronic balance fitted with a ^{210}Po static eliminator disc and capable of weighing in grams to four decimal places. Precautions were taken to prevent spillage and inhalation of the pure materials, and the balance and surrounding bench were thoroughly cleaned afterwards as a further precaution. Dissolution of the PAHs was achieved by ultrasonication rather than by shaking and inversion of the flask, and the solutions were made "up to volume" by weight rather than by volume, so as to avoid problems resulting from changes in solvent volume with variations in ambient temperature.

As flame-sealable ampoules could not be obtained in time for use in phase 1, the two standard solutions (designated STD1 and STD2, being respectively the solutions of declared and unknown concentrations) and an aliquot of the acetonitrile used in their preparation were transferred to separate 6 ml Hypo-vials (Pierce and Warriner, Chester, England) and sealed with colour-coded crimp seal caps fitted with PTFE liners. Each vial was weighed after sealing and the weight recorded on a return slip enclosed with the samples. For phase 2, separate sets of standards were prepared for GC and liquid chromatographic (LC) analyses, in hexane and acetonitrile, respectively. In this case the two standards (designated G1 and G2 [GC analysis] and H1 and H2 [LC analysis]) and an aliquot of the relevant solvent were transferred to 2 ml glass ampoules (Jencons (scientific) Ltd., Leighton Buzzard, England) and the samples frozen with liquid nitrogen prior to sealing with a flame. The ampoules were labelled, then weighed, and the weights were recorded as before.

3.5 Distribution of Samples

The vials or ampoules were packed in small polythene freezer boxes inside padded envelopes for protection, and distributed by post. Phase 1 samples were distributed in November 1988 with a deadline for the return of results at the end of February 1989; phase 2 samples were distributed in October 1989 with a deadline of 1 January 1990. In practice, results were accepted until May 1989 and 9 February 1990, respectively.

After phase 2 of the exercise was completed, two additional laboratories expressed interest in joining the intercomparison exercise, and one of the laboratories involved in both phases 1 and 2 requested further samples in order to rectify problems identified in the exercise. Accordingly, a further set of samples was prepared as for phase 2. These samples, designated as stage 1 (phase 3) of the exercise, were distributed in July 1990. One set of results was received, from laboratory No. 16 on 1 March 1991. A further set of samples was sent out in April 1991.

4 RESULTS

4.1 Stage 1 (Phase 1)

For phase 1 of the first stage of the exercise, samples were distributed to 19 laboratories, and results were received from 14, a return rate of 73%. The most common comment received from participants was of a noticeable and continuing weight loss from the sample vials on storage. As no losses of whole liquid were observed during a trial period within this laboratory during which filled vials were suspended upside-down, this suggests that the crimp-seal caps of at least some of the Hypo-vials were not vapour tight. If the loss of solvent vapour were significant, a bias could have been introduced in the concentrations reported, although as it could proceed at a similar rate in both standard solutions this may have resulted only in an increase in the scatter of results.

The results obtained during phase 1 are given in Annex 1. From these it can be seen that the overall means reported for the ten PAHs determined vary from 97% to 110% of the nominal concentrations in the unknown solution (STD2), with eight means falling within the range 97% to 101%. In addition, one laboratory (No. 13) carried out analysis of an NBS (U.S. National Bureau of Standards, now National Institute for Standards and Testing, NIST) reference PAH solution [SRM1647] after calibration with STD1, and reported slight discrepancies in the concentration values obtained only for pyrene and benzo[*b*]fluoranthene. This suggests that losses of acetonitrile from the vials were not a major source of error.

Although the overall means corresponded well with the nominal concentrations, the range of results reported was rather wide. This occurred despite the fact that the more widespread use of autosamplers has resulted in very low relative standard deviations (RSDs) being reported in many cases, often <3% of the mean value.

A number of laboratories also reported problems with GC analysis of acetonitrile solutions. One laboratory (No. 3) transferred the supplied solutions into benzene, with consequent losses of the lower boiling components phenanthrene, fluoranthene and pyrene.

4.2 Stage 1 (Phase 2)

Samples were distributed to 18 laboratories and results were received from 17, a return rate of 94%. The remaining laboratory cited instrumental failure as the reason for non-participation in the exercise. Fifteen sets of results were submitted for standard solution G2 (in hexane), seven of which were analysed by GC-FID, seven by GC/MS, and one by low-temperature (Shpol'skii) fluorescence. Eight sets of results were returned for standard solution H2 (in acetonitrile), five of which were analysed using HPLC with fluorescence detection and three with detection by UV absorption. The operating conditions used for GC and LC analyses are given in Tables 1 and 2, and the results submitted for stage 2 are given in Annex 2. Examples of the chromatograms and spectra obtained by the various techniques employed are given in Figures 1 to 4. It is apparent that all the techniques provide sufficient resolution and specificity for the analysis of a ten-compound PAH standard solution, but this may not be the case when more complex mixtures including alkylated PAHs, lipids, etc., are analysed in later stages of the intercomparison exercise.

Despite the improved sealing of ampoules and the optimization of solvents used for each method, the results generally showed greater variability than those obtained for phase 1. Laboratory 16 reported very low and variable results for all their GC/MS analyses, associated with an instrumental problem,. Results obtained by laboratories 4 and 8 (by GC/MS) were consistently somewhat high, and those of laboratory 12 were consistently low. In the latter case, this was caused by falling MS sensitivity when the samples were run, but no explanation has been found in the former cases. Laboratory 14 reported anomalously high values for indeno[123-*cd*]pyrene by GC-FID, and both HPLC-UV and HPLC-UVF yielded low results for benzo[*ghi*]perylene. Analysis of unknown solution H2 by GC-FID has confirmed the nominal concentration as correct, and so the nominal concentration for the circulated standard solution H1 must be wrong.

One interesting aspect of phase 2 was the inclusion of results obtained by a novel low-temperature fluorescence method, using the hexane standards prepared for GC analysis. Hexane is not an optimal solvent for the determination of the larger PAHs using this method, and before analysis the hexane must be replaced or, if the analyte concentrations are sufficiently high, heavily diluted with octane. Results obtained for pyrene, benz[*a*]anthracene, benzo[*a*]pyrene, and indeno[1,2,3-*cd*]pyrene (Annex 2) were in good agreement with the nominal values, whereas results obtained for benzo[*b*]fluoranthene, benzo[*e*]pyrene, chrysene and benzo[*ghi*]perylene were much poorer. The first two of these compounds suffer from spectral interference and are difficult to quantify, although as future work will be conducted at a higher optical resolution this should prove to be less of a problem from now on. The large discrepancies seen for chrysene and benzo[*ghi*]perylene were harder to explain, as these compounds show a strong quasilinear fluorescence and do not suffer from spectral overlap. Further work on solutions G1 and G2 was carried out at the Free University during the summer of 1990, and a second set of results was submitted in August (see Table 3). Interference from vibrational lines within the spectra of pyrene and the internal standard pyrene-d_{10} on the determination of benzo[*b*]fluoranthene and benzo[*e*]pyrene were eliminated by working at a higher spectral resolution (0.1 nm) and by adding the internal standard at a lower concentration. The earlier difficulties with the determination of chrysene and benzo-[*ghi*]perylene were also discovered to be due to storing the solutions in a freezer at −20 °C, and allowing solution G1 insufficient time to warm up. As the concentrations of some PAHs were close to the solubility limit, it was felt that freezing out and/or adhesion of these compounds to the glass of the ampoule must have caused the concentrations within G1 as made up to be lower than expected. On the second occasion, the ampoules were stored at 4 °C, and before opening they were put in an ultrasonic bath for several minutes and allowed to reach room temperature. After addition of the internal standard, the solutions were diluted 1000 times with octane prior to analysis. The results for the eight determinands analysed were in excellent agreement with the nominal values; analysis of phenanthrene and fluoranthene was not attempted.

4.3 Remarks from Participants

4.3.1 Phase 1

Lab. No. 2: Reported evaporation of solvent from the sample vials.

Lab. No. 3: Reported extremely poor performance with acetonitrile on a DB-1 column using cold on-column injection. The solvent was replaced by benzene prior to analysis.

Lab. No. 8: Reported problems believed to be due to the acetonitrile solvent in GC/MS analysis: 1) with leaks in the transfer line, and 2) with septa for the autosampler vials.

Lab. No. 11: Reported a loss of weight of around 1% in the solutions on receipt. Acetonitrile presented no problems when splitless injection was used, but caused peak splitting with cold on-column injection.

Lab. No. 13: Analysis of SRM1647 revealed slight discrepancies for the nominal concentrations of pyrene and benzo[*b*]fluoranthene in STD1. Seals appeared tight and intact, but weight losses of 0.3–0.4 g were noted for each solution.

Lab. No. 14: Reported problems of peak splitting, tailing and column deterioration when using acetonitrile with on-column injection. A major deviation from linearity was noted for phenanthrene at low concentration. A greater degree of variability was observed for GC/MS than for GC analysis. It was recommended that all future work to be undertaken in this exercise should use only GC-FID.

4.3.2 Phase 2

Lab. No. 2: Reported a head crash on their GC/MS data system which disabled the instrument for some time and limited the data to two replicates.

Lab. No. 3: Reported the hexane supplied to be of excellent quality.

Lab. No. 10: Reported that analyses were conducted using only UV-absorbance detection as their fluorescence detector was broken.

Lab. No. 11: GC/MS analysis was delayed due to instrument problems.

Lab. No. 13: Reported a problem with non-linear response for their fluorescence detector. Some dark coloured particles were observed in solutions H1 and H2.

Lab. No. 14: Had problems with their gas chromatograph. Observed greater variability in the concentrations determined for the standard solution using peak height rather than peak area as the basis for calculation. The differences were significant at the 99% level for all 10 PAHs. Recommended the use of peak area for future determinations.

No participant reported loss of weight in the ampoules used in phase 2 of the exercise.

5 STATISTICAL TREATMENT OF DATA

5.1 Summaries

Summaries of the results from phase 1 are given in Tables 4a–d. Laboratory numbers with decimal values of 1 and 2 indicate that results using two different methods have been submitted or that two separate submissions have been made using the same method. These tables contain, for each hydrocarbon determined at each laboratory, the number of replicates, and the averages, biases and precisions, respectively. Tables 5a–d give the corresponding results from phase 2.

Bias is calculated as

$$\mathbf{Bias_{ij} = 100\ (x_{ij} - c_{ij})\ /\ c_{ij}\ \%}$$

and precision as

$$\mathbf{Precision_{ij} = 100\ s_{ij}\ /\ c_{ij}\ \%}$$

where x_{ij} and s_{ij} are the average concentration and the standard deviation for the i'th hydrocarbon in the j'th laboratory, and c_{ij} is the corresponding true concentration. In the absence of any coordinated monitoring effort within ICES, or within most of the laboratories involved in the exercise, it is difficult to assign target values for bias and precision, but as an aid to interpretation, biases in excess of 20% have been highlighted using bold type in Tables 4c and 5c, and precision values exceeding 10% have been highlighted using bold type in Tables 4d and 5d.

Most laboratories exhibited a large bias for benzo-[*ghi*]perylene in phase 1. To a lesser extent, benzo[*b*]-fluoranthene tended to be biased in phase 2.

Figures 5 to 10 present the statistical information derived from the results of the exercise in a pictorial manner. A number of different approaches have been adopted, in order to study different aspects of the data received and to draw useful information out of the mass of results. In each case, the figures are preceded by a guide page, which explains the way in which the subsequent figure can be interpreted.

In Figure 5 precision and bias are plotted against each other, for each of the ten determinands in each phase of the exercise. To aid in the visual interpretation, the same scales are used in all plots, and dotted lines on the plots indicate the 10% precision and 20% bias targets. In most cases, the majority of laboratories are clustered within this area of the plots. The values of bias and precision tend to apply to all hydrocarbons measured within a laboratory. This can be seen in, e.g., Figure 6a, a visual correlation matrix of the biases observed in phase 1. The top row shows the biases for the first hydrocarbon plotted against those of the second, third, etc. The second row shows the biases for the second hydrocarbon plotted against those of the third, fourth, etc., and so on. Figure 6b gives a similar display for the precisions from phase 1. Figures 7a and 7b give the corresponding displays for the data from phase 2. The association between hydrocarbons is stronger in phase 2 than in phase 1.

5.2 Statistical Analyses

5.2.1 Variability between laboratories: Principal Component Analysis

To examine the interrelationships between the biases and precisions obtained for each hydrocarbon within laboratories, Principal Component Analyses were made on the covariance matrices of biases and precisions for phases 1 and 2. The coefficients of the first and second components for the data from phase 1 were as follows:

Total SD Hydrocarbon #	Bias (28%) Compl	Comp2	Precision (14%) Compl	Comp2
1	0.25	0.30	0.20	0.49
2	0.01	0.36	0.21	0.08
3	0.22	0.32	0.11	-0.07
4	0.15	0.36	0.28	-0.16
5	0.10	0.12	0.20	-0.02
6	0.23	0.21	0.21	-0.14
7	0.26	0.13	0.33	-0.25
8	0.77	-0.57	0.66	-0.25
9	0.26	0.32	0.27	-0.16
10	0.29	0.20	0.35	0.74
(% variation)	(49%)	(23%)	(78%)	(13%)

For the phase 2 data, the coefficients of the first and second components were as follows:

Total SD Hydrocarbon #	Bias (50%) Compl	Comp2	Precision (12%) Compl	Comp2
1	0.33	0.28	0.33	0.35
2	0.25	0.09	0.29	-0.11
3	0.25	0.02	0.32	-0.31
4	0.34	-0.61	0.26	0.82
5	0.37	-0.09	0.19	-0.15
6	0.30	-0.05	0.31	-0.13
7	0.21	0.10	0.27	-0.05
8	0.37	0.26	0.28	0.03
9	0.35	0.50	0.44	-0.19
10	0.34	-0.45	0.40	-0.12
(% variation)	(78%)	(12%)	(84%)	(5%)

The amount of variability explained by the first component tends to be high, except for the biases observed in phase 1. The coefficients in the first components are positive and, especially for the phase 2 data, are close to the value $(0.1)^{½}$, which they would have if the covariances of biases or precisions were all equal. The second components are contrasts between various subgroups of hydrocarbons.

The total standard deviation is larger for the phase 2 biases than for those in phase 1. Thus, there is more between-laboratory variability during phase 2, and this variability consists of bias or poor precision acting on all hydrocarbons. This is at odds with the expectation that better agreement would be obtained in phase 2 than in phase 1, as the solvent was optimized for each method.

These results can be interpreted further from the plots of the scores from the first two principal components, standardized to have unit variance, shown for phase 1 biases and precisions in Figures 8a and 8b, respectively. Figures 9a and 9b show the corresponding plots for phase 2. The figures are presented as biplots, and laboratory numbers are given a suffix .0, .1 or .2 for sole data set, first, or second data set, respectively. The scores for the biases in phase 2 show one laboratory (No. 16) separated from the rest. Similarly, the plots for the precisions show a few laboratories detached from the main group of laboratories.

5.2.2 Variability between methods: Canonical Discriminant Analysis

Figures 10a and 10b, for phases 1 and 2 respectively, show the results of an analysis carried out in order to determine whether biases can be explained by the method of analysis. Laboratories are shown plotted on the first two canonical axes, chosen so that the ratio of the variability between methods to the variability between laboratories within methods is greatest. Both plots show some clustering of laboratories sharing the same method, but only for the phase 2 data was the difference between methods significant. Arithmetic differences between the two standards (GC and HPLC) were removed prior to the statistical analysis.

The canonical coefficients were as follows:

Hydrocarbon	Phase 1		Phase 2	
	Canl	Can2	Can1	Can2
1	-35	-8	9	-10
2	-12	14	0	39
3	-20	14	-3	-25
4	80	-17	22	24
5	51	-13	-10	-45
6	32	-4	0	-17
7	-32	-2	2	2
8	10	3	-10	22
9	-21	7	-15	12
10	-16	12	7	4

which gave the following means for each method:

Method	Phase 1		Phase 2	
	Canl	Can2	Canl	Can2
GC-FID	2.7	-0.3	-2.3	1.8
GC-MS	-1.9	1.2	-3.0	-1.3
HPLC-UV	-0.0	0.1	6.4	-1.3
HPLC-UVF	-0.9	1.8	6.3	0.4

6 CONCLUSIONS AND FUTURE PLANS

Compared with the wide variations in concentrations reported for analyses of specific aromatic hydrocarbons in previous ICES exercises (Law and Portmann, 1982; Farrington *et al.*, 1986; Uthe *et al.*, 1986), these data represent a considerable improvement. It must be borne in mind, however, that in stage 1 of this exercise only standard solutions were analysed. Considerable care is still needed in the calibration of instruments and, by analogy with the current chlorobiphenyl exercise (de Boer *et al.*, 1994), in the preparation of standard solutions. Laboratories can clearly produce very precise data in the short term; in the longer term the variability will presumably be greater. All participating laboratories, with the exception of laboratory number 16, were adjudged capable of proceeding to stage 2 of the exercise. It has already been mentioned that this laboratory was well aware of its problems, and remedial action has been take to correct them. This has involved modifications to equipment and to their operating procedures, and a one-week training visit by an analyst to the coordinator's laboratory. A further set of results has now been submitted from laboratory 16 under phase 3 of the exercise and, if these results are deemed acceptable, this laboratory will also be allowed to pass on to stage 2. These results were not presented here as phase 3 results were awaited from two other laboratories (see Section 7).

The proposed intercomparison programme agreed at MCWG in 1987 (see Section 2) was designed to improve comparability for data obtained on all matrices routinely analysed (water, sediment, and biota). Initially it was agreed to concentrate on the determination of PAHs in biota, and at the meeting of analysts held in 1990 it was agreed that the most appropriate species to use would be *Mytilus edulis* (blue mussel). However, it was also agreed that the first matrix to be studied should be a sediment because:

1) a sediment sample is more readily stabilized;

2) the North Sea Task Force have included the determination of PAHs in sediments in their monitoring requirements; and

3) the subsequent stages of this exercise would be easier to prepare and conduct.

Stage 2 of the exercise was planned to involve the analysis of a cleaned-up sediment extract, and of both distributed and the laboratories' own standard solutions. It was originally intended to run this stage during 1990, and to have a preliminary report available early in 1991. This proved impractical because of staffing problems in the coordinator's laboratory, and the timetable was changed to run the exercise later in 1991. For various reasons, however, this second stage of the exercise was not conducted.

It had been agreed that, if the results for stage 2 were acceptable, stage 3 would involve the analysis of a raw sediment extract, and stage 4 analysis of a fresh sediment sample. In stage 2 and subsequent stages of the exercise, laboratories would also be asked to submit analytical quality control data summarizing analyses of certified and/or laboratory reference materials undertaken during the period of the intercomparison programme. This would allow a more meaningful estimate of long-term variability (within laboratory) to be made, and meaningful targets to be set for future exercises and monitoring/survey programmes. In addition, the use of internal standards would be encouraged, but because of the use of multiple analytical techniques it would not be possible for the coordinator to include internal standards in the solutions distributed and each analyst would have to add his own favoured compounds.

7 ENVIRONMENTAL PAHs

It would be prudent at this point to make some statements about the present scope of this exercise in relation to the environmental occurrence of PAHs and their sources. Leaving aside diagenetic processes occurring in shallow sediments which generate specific PAHs such as perylene, environmental PAHs arise from two major anthropogenic sources. These are fossil fuel combustion and oil. In certain localized areas, PAHs have also been generated during industrial processes, and these tend to be similar in composition to those arising from combustion. PAH assemblages arising from combustion of fossil fuels contain only parent, non-alkylated PAHs, whereas in oils a major fraction of the PAHs can occur as alkylated PAHs. In consequence, chromatograms of the aromatic fractions of samples contaminated by oil and petroleum products are normally much more complicated than those for samples where combustion is the main source. This has implications for the analytical techniques that can be used for analysis, as many compounds may coelute and less specific methods may exhibit insufficient resolution to resolve adequately such complex mixtures. Thus GC-FID, even with capillary columns, is inadequate for these separations, whereas GC-MS can resolve such complex mixtures by the use of mass chromatography or multiple-ion detection methods. HPLC is normally a lower resolution technique than capillary GC; microbore technology can yield similar resolution but at the expense of very long (>24hr) run times, and this is not currently used routinely in monitoring laboratories. When combined with a relatively non-specific detection technique such as UV-absorption, problems may be expected. HPLC-UVF and LC-MS would be expected to yield more specificity, but would still be limited in capability by the overall HPLC resolution available.

Whilst this exercise is targeted primarily at combustion PAHs, these problems are minimized, especially as only a subset of the PAHs encountered in the environment are being analysed. Lower resolution techniques may still, in addition, experience some difficulties with co-extractives from real samples. Results generated from this exercise should not, however, be extrapolated to predict performance when oil-contaminated samples are being analysed for specific hydrocarbons.

8 REFERENCES

de Boer, J., Reutergårdh, L., van der Meer, J., and Calder, J.A. 1994. Report on the Results of the ICES/IOC/OSPARCOM Intercomparison Programme on the Analysis of Chlorobiphenyl Congeners in Marine Media - Step 2. This volume.

Farrington, J.W., David, A.C., Livramento, J.B., Clifford, C.H., Frew, N.M., and Knap, A. 1986. ICES/IOC Intercomparison Exercise on the Determination of Petroleum Hydrocarbons in Biological Tissues (mussel homogenate) - ICES/2/HC/BT. ICES Cooperative Research Report No. 141, pp. 1–75.

Grahl-Nielsen, O., Law, R., Palmork, K., Portmann, J.E., and Wilhelmsen, S. 1978. Anglo-Norwegian oil programme - Intercalibration of analytical methods. ICES, Doc. C.M.1978/E:22.

Hilpert, L.R., May, W.E., Wise, S.A., Chesler, S.N., and Hertz, H.S. 1978. Interlaboratory comparison of determinations of trace level petroleum hydrocarbons in marine sediment. Analytical Chemistry, 50:458–463.

Law, R.J., and Portmann, J.E. 1982. Report on the First ICES Intercomparison Exercise on Petroleum Hydrocarbon Analyses in Marine Samples. ICES Cooperative Research Report No. 117. 55 pp.

MacLeod, W.D. jr, Prohaska, P.G., Gennero, D.D., and Brown, D.W. 1982. Interlaboratory comparisons of selected trace hydrocarbons from marine sediments. Analytical Chemistry, 54:386–392.

Uthe, J.F., Musial, C.J., and Sirota, G.R. 1986. Report on the Intercomparative Study 03/HC/BT on the Determination of Polycyclic Aromatic Hydrocarbons in Biological Tissue. ICES Cooperative Research Report No. 141, pp. 76–85.

LIST OF PARTICIPANTS

Lab. No.	Analyst and Laboratory
1	Dr L. Tuinstra RIKILT Wageningen, Netherlands
2	Dr M. Ehrhardt Institute for Marine Research Kiel, Federal Republic of Germany
3	Dr E. Levy Bedford Institute of Oceanography Dartmouth, Canada
4	Dr P. Mackie MAFF, Torry Research Station Aberdeen, UK
5	Mrs G. Riekwel-Booy TNO/CIVO Institutes IJmuiden, Netherlands
6	Dr E.-L. Poutanen Institute for Marine Research Helsinki, Finland
7	Dr F. Smedes Rijkswaterstaat Haren, Netherlands
8	Dr N. Theobald German Hydrographic Institute Hamburg, Federal Republic of Germany
9	Dr A. Contente-Mota Environmental Quality Directorate Lisbon, Portugal
10	Prof U. Kirso Academy of Sciences Tallinn, Estonian SSR
11	Dr J. Klungsøyr Institute for Marine Research Bergen, Norway
12	Mr R. Law MAFF Fisheries Laboratory Burnham-on-Crouch, UK
13	Dr M. Quilliam NRC Atlantic Laboratory Halifax, Canada
14	Dr J. Robson DAFS Marine Laboratory Aberdeen, UK
15	Mr C. Phinney University of Massachusetts Boston, USA
16	Dr J. Biscaya Hydrographic Institute Lisbon, Portugal
17	Dr F. Ariese Free University Amsterdam, Netherlands
*	Dr P. Roose Rijksstation voor Zeevisserij Ostend, Belgium
*	Dr J. Tronczynski IFREMER Nantes, France

* These two laboratories have been supplied with Stage 1 samples for analysis prior to participation in Stage 2.

Table 1. GC and GC/MS operating conditions.

Lab. No.	Apparatus	Stationary Phase	Length	ID (mm)	Carrier	Temperature Programme	Injector
2	HP 5993C GC/MS	CP-Sil-8	50m	0.23	Helium	40 (1) @20-200, @3-260,@2-300C	Splitless
3	HP 5890 GC	DB-1	15m	0.25	Helium	60 (0.5) @8-300C	OCI
4	HP 5970 GC/MS	HP-1	25m	0.2	Helium	60 @4-280C	OCI
8	HP 5970B GC/MS	HP-1	25m	0.2	Helium	45 (2) @5-230 (2) @5-300C	Splitless
9	PE 8500 GC	SE-54	30m	0.22	Hydrogen	70 (0.8) @30-140 @3-260C	Splitless
9	PE 8500 GC/MS	SE-54	30m	0.22	Helium	70 (1) @10-190 (2) @10-260C	Splitless
11	HP 5880 GC	SE-54	50m	0.32	Hydrogen	60 (1) @15-150 @4-270C	Splitless
11	HP 5987A GC/MS	SE-54	40m	0.32	Helium	60 (1) @15-150 @5-280C	OCI
12	HP5890 GC	HP-5	25m	0.32	Hydrogen	60 (1) @5-320C	OCI
12	Incos50 GC/MS	HP-5	25m	0.32	Helium	60 (1) @5-300C	OCI
14	Carlo Erba 4160	DB-1	50m	0.32	Hydrogen	65 (1) @5-300C	OCI
15	Varian 6000 GC	DB-5	30m	0.25	Helium	50 (3) @6-290C	Splitless
16	HP 5985 GC/MS	HP-5	25m	0.2	Helium	40 (2) @15-300C	Splitless

OCI : On-column injection.

Temperature Programme : eg. 45 (1) @5-300C = Injection temperature 45°C held for 1 minute, raised at 5°C per minute to 300°C.

Table 2. HPLC operating conditions.

Lab. No.	Instrument	Column Type	Size (mm)	Eluent	Detectors
1	Waters 590	Vydac 201 TP B5	200 x 0.3	acetonitrile/water 85/15%	UV & UVF
5	Maxima 820	ChromSpher PAH	100 x 3.0	acetonitrile/water gradient	UVF
6	Waters 510	Waters radial PAK 5-PAH 10		acetonitrile/water gradient	UV
7	HP 1090	Vydac TP B5	250 x 4.6	methanol/water gradient	2 x UVF
10	Knauer	Perkin Elmer HCODS	250 x 2.6	methanol/water 95/5%	UV
13	HP 1090	Vydac 201 TP 54		acetonitrile/water gradient	Diode Array & UVF

Table 3. Supplementary results obtained by low-temperature fluorescence. (ng / µl).

Compound	Concentration in solution G2						Range	Mean	RSD (%)	Nominal Value
Pyrene	71.4	71.8	71.4	68.4	69.2	68.8	68.4-71.8	70.2	2	68
Benz[a]anthracene	48.3	47.8	46.9	47.6	45.7	45.7	45.7-48.3	47	2.1	47
Chrysene	46.4	46.6	41.2	42.3	46.4	46.8	41.2-46.8	45	5.1	45
Benzo[b]fluoranthene	27.8	25.2	26.2	24.9	27.2	25.2	24.9-27.8	26.1	4.2	26
Benzo[e]pyrene	80	85	92	89	80	83	80-92	85	5.2	78
Benzo[a]pyrene	119	121	119	113	119	111	111-121	117	3.4	115
Benzo[ghi]perylene	79.4	76.4	74.5	79.4	74.2	74.2	74.2-79.4	76.4	3	75
Indeno[123-cd]pyrene	59	58	56	59	47	49	47-59	55	8.8	53

Table 4a. Numbers of replicates Phase 1

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	6	6	6	6	6	6	6	6	6	6
	9.0	6	6	6	6	6	6	6	6	6	6
	11.2	6	6	6	6	6	6	6	6	6	6
	12.2	6	6	6	6	6	6	6	6	6	6
	14.2	6	6	6	6	6	6	6	6	6	6
gc-ms	2.0	5	6	5	6	5	5	5	6		
	4.0	6	6	6	6	6	6	6	6	6	6
	8.0	6	6	6	6	6	6	6	6	6	6
	11.1	6	6	6	6	6	6	6	6	6	6
	12.1	6	6	6	6	6	6	6	6	6	6
	14.1	6	6	6	6	6	6	6	6	6	6
hplc-uv	6.0	6	6	6	6	6	6	6	6	6	6
	10.2	5	5		5	5	5	5	5	5	5
	13.2	6	6	6	6	6	6	6	6	6	6
hplc-uvf	1.0	6	6	6	6	6	6	6	6	6	6
	5.0	6	6	6	6	6	6	6	6	6	6
	7.1	6	6	6	6	6	6	6	6	6	6
	7.2				6	6		6			
	10.0	1			7	7		7	7	4	7
	13.0	1	6	6	6	6	6	6	6	6	6

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 4b. Average of individual determinations of PAHs (in ng/μ1). Phase 1

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	29.2	22.7	74.7	19.8	37.5	48.3	30.7	6.4	45.2	10.3
	9.0	37.9	25.4	76.6	23.2	37.5	51.2	37.7	8.6	44.1	12.6
	11.2	41.0	25.9	82.5	22.1	38.4	52.7	31.9	7.9	46.9	11.7
	12.2	41.9	27.1	84.8	22.7	40.0	60.9	33.7	7.7	49.6	11.9
	14.2	44.1	27.3	86.6	24.0	41.5	54.8	36.2	7.7	50.5	11.8
gc-fid mean	.	38.8	25.7	81.0	22.4	39.0	53.6	34.0	7.7	47.3	11.7
gc-ms	2.0	43.8	20.2	88.2	23.2	38.8	55.2	26.9	4.1	.	.
	4.0	36.3	24.8	73.0	20.7	37.0	48.2	31.8	7.0	44.4	10.9
	8.0	42.0	26.2	89.5	23.6	38.2	47.9	33.0	6.2	44.8	11.0
	11.1	40.6	24.2	83.3	19.4	39.4	46.3	31.3	8.0	35.8	9.3
	12.1	41.2	29.2	77.9	23.2	38.3	49.6	34.9	6.7	49.8	10.7
	14.1	43.2	22.9	83.7	22.7	40.5	56.6	40.0	9.5	55.5	11.9
gc-ms mean	.	41.2	24.6	82.6	22.1	38.7	50.6	33.0	6.9	46.0	10.7
hplc-uv	6.0	40.1	25.6	80.2	21.3	37.8	55.0	32.0	6.9	46.9	11.1
	10.2	39.1	21.7	.	19.9	37.9	49.7	31.0	6.1	43.4	13.2
	13.2	41.1	26.3	84.4	22.6	39.5	53.7	33.7	8.1	49.3	12.1
hplc-uv mean	.	40.1	24.5	82.3	21.3	38.4	52.8	32.2	7.1	46.5	12.1
hplc-uvf	1.0	38.9	26.6	81.8	22.5	37.9	49.6	33.1	6.5	47.5	10.7
	5.0	40.5	24.9	86.9	21.6	38.2	51.8	33.5	10.7	48.7	11.7
	7.1	43.0	27.5	107.8	23.7	41.8	54.5	35.0	8.0	50.8	12.2
	7.2	.	.	.	23.8	42.0	.	35.0	.	.	.
	10.1	.	.	34.3	22.2	.	53.0	33.6	8.8	44.4	9.2
	13.1	38.0	26.4	78.9	22.3	38.9	52.3	34.0	7.7	48.3	13.3
hplc-uvf mean	.	40.1	26.3	77.9	22.7	39.8	52.2	34.0	8.3	47.9	11.4
overall mean	.	40.1	25.3	80.8	22.2	39.0	52.2	33.4	7.5	47.0	11.4
Reference Concentration		41.2	26	84	21.2	40	52.4	33.2	6.8	48.8	10.8

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 4c. Biases (%). Phase 1

METHOD	LAB	B1	B2	B3	B4	B5	B6	B7	B8	B9	B10
gc-fid	3.0	**-29.2**	-12.7	-11.1	-6.8	-6.3	-7.9	-7.7	-6.1	-7.4	-4.3
	9.0	-8.1	-2.5	-8.8	9.5	-6.2	-2.3	13.5	**27.0**	-9.6	16.5
	11.2	-0.4	-0.3	-1.8	4.2	-4.0	0.6	-4.0	16.7	-3.9	8.0
	12.2	1.6	4.0	0.9	7.2	-0.1	16.2	1.5	13.0	1.7	9.9
	14.2	7.1	4.9	3.1	13.1	3.8	4.5	9.0	13.0	3.6	9.3
gc-fid mean	.	-5.8	-1.3	-3.6	5.5	-2.6	2.2	2.5	12.7	-3.1	7.9
gc-ms	2.0	6.3	**-22.3**	5.0	9.4	-3.0	5.3	-18.9	**-40.2**	.	.
	4.0	-12.0	-4.5	-13.1	-2.5	-7.4	-8.1	-4.1	2.2	-9.1	1.2
	8.0	1.9	0.6	6.6	11.5	-4.6	-8.6	-0.8	-8.6	-8.3	2.2
	11.1	-1.6	-7.1	-0.9	-8.5	-1.5	-11.6	-5.8	17.9	**-26.6**	-14.4
	12.1		12.4	-7.3	9.2	-4.2	-5.3	5.2	-1.5	1.9	-1.2
	14.1	4.8	-11.9	-0.3	7.1	1.2	7.9	**20.4**	**40.0**	13.7	9.7
gc-ms mean	.	-0.1	-5.5	-1.7	4.4	-3.3	-3.4	-0.6	1.6	-5.7	-0.5
hplc-uv	6.0	-2.6	-1.7	-4.5	0.5	-5.6	4.9	-3.8	1.5	-4.0	2.8
	10.2	-5.0	-16.6	.	-6.2	-5.2	-5.1	-6.6	-10.0	-11.1	**22.2**
	13.2	-0.2	1.1	0.5	6.8	-1.2	2.5	1.5	19.6	1.0	11.9
hplc-uv mean	.	-2.6	-5.8	-2.0	0.3	-4.0	0.8	-3.0	3.7	-4.7	12.3
hplc-uvf	1.0	-5.5	2.2	-2.6	6.0	-5.2	-5.3	-0.3	-3.9	-2.6	-0.6
	5.0	-1.7	-4.1	3.4	1.9	-4.5	-1.2	0.9	**56.9**	-0.2	8.5
	7.1	4.3	5.8	**28.4**	11.9	4.5	4.0	5.5	17.6	4.0	13.0
	7.2	.	.	.	12.3	5.1	.	5.4	.	.	.
	10.1	.	.	**-59.1**	4.6	.	1.1	1.1	**28.7**	-9.0	-14.6
	13.1	-7.8	1.4	-6.0	5.0	-2.8	-0.2	2.5	13.2	-1.0	**23.3**
hplc-uvf mean	.	-2.7	1.3	-7.2	6.9	-0.6	-0.3	2.5	**22.5**	-1.8	5.9
overall mean	.	-2.7	-2.9	-3.8	4.8	-2.5	-0.4	0.7	10.4	-3.7	5.7

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 4d. Precisions (%).

Phase 1

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	0.8	0.9	1.1	2.1	2.3	1.9	1.8	1.1	1.9	2.1
	9.0	**12.6**	**13.1**	6.8	**12.5**	9.4	**10.0**	**15.0**	**31.9**	**11.0**	**20.3**
	11.2	1.4	1.9	1.5	2.4	2.2	2.6	2.3	3.0	2.2	2.8
	12.2	1.0	2.7	1.3	1.4	1.3	1.2	1.0	1.7	0.8	3.2
	14.2	1.6	2.4	2.0	3.2	2.2	2.0	7.1	3.1	2.1	4.3
gc-fid mean	.	3.5	4.2	2.5	4.3	3.5	3.6	5.4	8.2	3.6	6.5
gc-ms	2.0	**12.7**	8.2	9.8	**15.1**	7.3	8.6	6.7	7.7	.	.
	4.0	1.7	2.7	1.2	2.1	1.6	1.0	1.8	2.8	0.8	1.9
	8.0	3.5	5.9	6.6	5.1	5.1	5.3	5.3	4.4	4.6	5.0
	11.1	8.1	2.9	1.2	5.5	7.5	2.6	2.5	2.2	6.7	7.2
	12.1	1.5	1.9	1.1	3.8	2.5	2.3	3.2	4.4	2.3	2.3
	14.1	2.1	2.8	3.3	**12.5**	8.2	8.0	**13.1**	**20.2**	**13.6**	9.1
gc-ms mean	.	4.9	4.1	3.8	7.4	5.4	4.6	5.4	6.9	5.6	5.1
hplc-uv	6.0	1.2	1.4	1.4	1.9	1.1	0.8	2.4	2.6	1.2	1.0
	10.2	4.5	6.4	.	7.7	6.9	5.8	4.3	**20.0**	1.6	**12.0**
	13.2	0.4	1.4	0.6	0.5	0.4	0.8	0.8	1.2	1.9	4.6
hplc-uv mean	.	2.0	3.1	1.0	3.3	2.8	2.5	2.5	7.9	1.6	5.9
hplc-uvf	1.0	3.8	1.4	3.1	2.1	3.1	3.0	2.6	2.4	2.4	1.5
	5.0	2.2	4.1	5.0	3.5	3.4	3.7	4.1	3.7	4.0	3.6
	7.1	0.8	1.0	1.4	1.4	1.0	0.5	0.5	7.4	1.0	4.5
	7.2	.	.	.	2.1	1.0	.	0.9	.	.	.
	10.1	.	.	4.4	7.7	.	9.8	6.4	4.4	6.6	**13.3**
	13.1	9.6	2.1	0.7	1.5	2.4	0.5	0.5	0.9	1.4	**18.2**
hplc-uvf mean	.	4.1	2.2	2.9	3.0	2.2	3.5	2.5	3.8	3.1	8.2
overall mean	.	3.9	3.5	2.9	4.7	3.6	3.7	4.1	6.6	3.7	6.5

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 5a. Numbers of replicates

Phase 2

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	6	6	6	6	6	6	6	6	6	6
	9.2	6	6	6	6	6	6	6	6	6	6
	11.2	6	6	6	6	6	6	6	6	6	6
	12.2	6	6	6	6	6	5	5	6	5	5
	14.1	6	6	6	6	6	6	6	6	6	6
	14.2	6	6	6	6	6	6	6	6	6	6
gc-ms	2.0	2	2	2	2	2	2	2	2	2	2
	4.0	6	6	6	6	6	6	6	6	6	6
	8.0	6	6	6	6	6	6	6	6	6	6
	9.1	6	6	6	6	6	6	6	6	6	6
	11.1	6	6	6	6	6	6	6	6	6	6
	12.1	6	6	6	6	6	6	6	6	6	6
	15.0	6	6	6	6	6	6	6	6	6	6
	16.0	12	12	12	12	12	7	7	7	7	7
uvf	17.0			5	6	6	6	6	6	6	6
hplc-uv	6.0	6	6	6	6	6	6	6	6	6	6
	10.0	6	6		6	6	6	6	6	6	6
	13.2	6	6	6	6	6	6	6	6	6	6
hplc-uvf	1.0	6	6	6	6	6	6	6	6	6	6
	5.0	6	6	6	6	6	6	6	6	6	6
	7.1	6	6	6	6	6	6	6	6	6	6
	7.2				6	6		6			
	13.1	6	6	6	6	6	6	6	6	6	6

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 5b. Average of individual determinations of PAHs (in ng/μl). Phase 2

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	46.0	52.9	70.7	43.6	45.8	73.1	106.5	28.2	70.7	49.4
	9.2	46.2	53.0	71.3	45.9	50.0	79.5	123.8	30.3	78.7	54.6
	11.2	43.3	51.1	68.3	39.5	44.3	63.9	94.2	26.2	62.4	44.2
	12.2	45.2	49.2	66.1	42.6	46.5	73.8	111.4	28.1	69.9	49.5
	14.1	56.1	54.8	71.3	47.0	50.7	80.9	122.7	30.4	82.5	51.8
	14.2	42.9	50.4	70.2	52.8	51.4	80.7	119.2	26.8	91.0	59.5
gc-fid mean	.	46.6	51.9	69.6	45.2	48.1	75.3	113.0	28.3	75.9	51.5
gc-ms	2.0	39.5	45.8	61.4	37.1	40.0	68.2	114.5	22.5	66.6	30.0
	4.0	57.8	61.4	83.7	53.2	61.2	87.9	131.3	32.9	95.5	59.3
	8.0	50.2	54.9	73.4	50.2	54.7	79.8	123.2	30.3	78.5	55.3
	9.1	39.9	41.7	69.5	48.0	48.4	78.6	99.0	29.8	77.1	57.3
	11.1	45.2	47.7	65.1	40.2	46.1	74.3	108.0	28.4	75.0	51.5
	12.1	37.3	46.7	62.1	38.9	42.0	68.6	111.2	22.0	58.2	39.9
	15.0	43.8	47.3	63.6	40.3	45.9	67.6	99.6	27.9	66.8	48.2
	16.0	24.1	30.3	43.4	17.4	21.9	35.7	74.5	12.6	38.1	23.4
gc-ms mean	.	42.2	47.0	65.3	40.6	45.0	70.1	107.7	25.8	69.5	45.6
Reference Concentration		45.0	51.0	68.0	47.0	45.0	78.0	115.0	26.0	75.0	53.0
uvf	17.0	.	.	63.6	44.2	101.4	55.1	119.8	42.7	117.7	52.2
uvf mean	.	.	.	63.6	44.2	101.4	55.1	119.8	42.7	117.7	52.2
hplc-uv	6.0	145.0	95.3	67.0	19.3	34.5	71.3	164.0	56.3	52.3	37.3
	10.0	130.0	88.0	.	17.0	27.8	67.3	169.2	51.0	49.3	34.7
	13.2	120.3	81.0	53.7	17.7	33.1	63.3	159.5	48.1	45.7	32.6
hplc-uv mean	.	131.8	88.1	60.4	18.0	31.8	67.3	164.2	51.8	49.1	34.9
hplc-uvf	1.0	140.3	87.0	58.7	16.7	30.0	67.3	163.0	51.3	46.3	32.0
	5.0	121.7	78.4	50.5	15.6	28.3	59.8	149.0	43.5	46.4	36.0
	7.1	138.0	91.3	59.5	19.3	34.7	70.3	174.7	54.1	49.3	37.2
	7.2	.	.	.	19.2	35.1	.	177.2	.	.	.
	13.1	102.5	84.3	54.5	17.4	32.2	64.7	165.5	48.0	45.0	34.1
hplc-uvf mean	.	125.6	85.2	55.8	17.6	32.1	65.5	165.9	49.2	46.7	34.8
Reference Concentration		142.0	92.0	59.0	16.0	31.0	71.0	176.0	55.0	64.0	34.0

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 5c. Biases (%).

Phase 2

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	2.1	3.8	3.9	-7.2	1.7	-6.3	-7.4	8.5	-5.7	-6.9
	9.2	2.7	3.9	4.8	-2.4	11.1	1.9	7.7	16.6	4.9	3.1
	11.2	-3.9	0.2	0.4	-16.0	-1.7	-18.0	-18.1	0.6	-16.9	-16.6
	12.2	0.4	-3.5	-2.8	-9.3	3.3	-5.4	-3.1	7.9	-6.8	-6.5
	14.1	**24.6**	7.5	4.8	0.1	12.6	3.7	6.7	17.0	10.0	-2.2
	14.2	**-4.8**	-1.2	3.2	12.3	14.3	3.5	3.6	3.1	**21.4**	12.2
gc-fid mean		3.5	1.8	2.4	-3.8	6.9	-3.4	-1.8	9.0	1.2	-2.8
gc-ms	2.0	-12.2	-10.2	-9.7	**-21.1**	-11.1	-12.6	-0.4	-13.5	-11.2	**-43.4**
	4.0	**28.3**	**20.5**	**23.1**	13.2	**36.1**	12.7	14.2	**26.7**	**27.4**	11.9
	8.0	11.5	7.7	7.9	6.7	**21.6**	2.2	7.1	16.4	4.6	4.3
	9.1	-11.4	-18.3	2.2	2.0	7.6	0.8	-14.0	14.7	2.8	8.1
	11.1	0.4	-6.5	-4.3	-14.4	2.4	-4.7	-6.1	9.0		-2.9
	12.1	-17.1	-8.5	-8.6	-17.3	-6.7	-12.0	-3.3	-15.3	**-22.4**	-9.2
	15.0	-2.7	-7.3	-6.5	-14.3	2.0	-13.4	-13.4	7.3	-11.0	-9.2
	16.0	**-46.5**	**-40.7**	**-36.1**	**-63.0**	**-51.3**	**-54.2**	**-35.2**	**-51.5**	**-49.2**	**-55.8**
gc-ms mean		-6.2	-7.9	-4.0	-13.5		-10.1	-6.4	-0.8	-7.4	-13.9
uvf	17.0			-6.5	-5.9	**125.4**	**-29.4**	4.2	**64.4**	**56.9**	-1.4
uvf mean				-6.5	-5.9	**125.4**	**-29.4**	4.2	64.4	**56.9**	-1.4
hplc-uv	6.0	2.1	3.6	13.6	**20.8**	11.3	0.5	-6.8	2.4	-18.2	9.8
	10.0	-8.5	-4.3		6.3	-10.2	-5.2	-3.9	-7.3	**-22.9**	2.0
	13.2	-15.3	-12.0	-9.0	10.6	6.6	-10.9	-9.4	-12.6	**-28.6**	-4.2
hplc-uv mean		-7.2	-4.2	2.3	12.6	2.6	-5.2	-6.7	-5.8	**-23.2**	2.5
hplc-uvf	1.0	-1.2	-5.4	-0.6	4.2	-3.2	-5.2	-7.4	-6.7	**-27.6**	-5.9
	5.0	-14.3	-14.8	-14.4	-2.3	-8.7	-15.8	-15.3	**-21.0**	**-27.5**	5.7
	7.1	-2.8	-0.8	0.9	**20.8**	11.9	-1.0	-0.8	-1.6	**-23.0**	9.5
	7.2				19.8	13.3		0.7			
	13.1	**-27.8**	-8.4	-7.7	8.9	3.9	-8.9	-6.0	-12.8	**-29.7**	0.3
hplc-uvf mean		-11.5	-7.3	-5.4	10.3	3.4	-7.7	-5.8	-10.5	**-27.0**	2.4
overall mean		-4.6	-4.5	-1.7	-2.1	8.3	-8.1	-4.6	2.4	-7.8	-5.1

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Table 5d. Precisions (%).

Phase 2

METHOD	LAB	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
gc-fid	3.0	2.4	3.4	3.4	2.5	2.7	3.2	2.1	3.1	2.7	2.8
	9.2	4.9	4.7	3.4	4.1	4.4	4.9	5.4	4.4	5.3	5.0
	11.2	5.0	1.1	1.3	4.0	4.6	5.8	6.0	6.3	6.8	6.2
	12.2	0.9	1.9	1.4	1.7	2.1	0.7	0.8	5.1	1.2	2.1
	14.1	0.6	2.3	2.0	1.5	1.8	1.4	1.6	1.6	1.4	1.5
	14.2	14.0	9.8	8.9	16.9	6.2	9.7	10.1	11.0	16.9	15.6
gc-fid mean	.	4.6	3.9	3.4	5.1	3.6	4.3	4.3	5.3	5.7	5.5
gc-ms	2.0	2.2	4.7	5.0	0.3	4.4	3.8	6.8	4.4	14.0	9.3
	4.0	1.0	0.7	0.8	0.8	1.9	1.6	1.7	0.5	0.8	1.6
	8.0	1.2	1.1	1.0	0.9	0.8	1.6	1.9	1.6	1.0	1.0
	9.1	7.5	8.3	15.4	7.5	10.6	11.4	6.2	13.1	13.2	15.0
	11.1	1.6	1.3	0.5	2.6	2.9	5.3	3.6	4.0	4.5	5.1
	12.1	3.9	3.0	2.9	2.2	4.1	5.5	5.2	6.0	5.0	7.2
	15.0	3.3	3.4	3.5	3.5	4.1	3.6	3.7	4.3	4.0	4.0
	16.0	12.7	13.5	10.6	4.4	5.2	12.3	11.9	7.8	11.2	10.6
gc-ms mean	.	4.2	4.5	5.0	2.8	4.2	5.6	5.1	5.2	6.7	6.7
uvf	17.0	.	.	1.5	2.3	11.8	3.7	3.0	13.1	6.7	5.8
uvf mean	.	.	.	1.5	2.3	11.8	3.7	3.0	13.1	6.7	5.8
hplc-uv	6.0	0.9	0.9	2.1	3.2	2.7	1.9	2.1	2.2	0.8	1.5
	10.0	0.4	1.8	.		1.3	0.7	1.0	2.0	0.8	4.4
	13.2	0.4	0.6	0.5	0.6	0.6	0.6	0.5	0.5	0.5	1.9
hplc-uv mean	.	0.6	1.1	1.3	1.3	1.5	1.1	1.2	1.6	0.7	2.6
hplc-uvf	1.0	3.0	2.7	4.1	3.2	4.1	1.5	2.2	3.0	1.3	0.0
	5.0	1.4	1.2	2.5	2.9	1.3	1.9	2.1	4.3	2.9	3.0
	7.1	1.8	1.4	1.8	2.3	3.2	1.8	2.0	3.4	1.6	4.8
	7.2	.	.	.	2.4	2.6	.	2.7	.	.	.
	13.1	0.7	0.1	0.5	0.3	0.3	0.3	0.3	0.4	0.5	2.1
hplc-uvf mean	.	1.7	1.4	2.2	2.2	2.3	1.4	1.9	2.8	1.6	2.5
overall mean	.	3.3	3.2	3.6	3.1	3.6	3.8	3.6	4.6	4.7	5.0

D1=Phenathrene D2=Fluoranthene D3=Pyrene D4=Benz[a]anthracene D5=Chrysene
D6=Benzo[e]pyrene D7=Benzo[a]pyrene D8=Benzo[b]fluoranthene D9=Benzo[ghi]perylene D10=Indeno[123-cd]pyrene

Figure 1. GC-FID chromatogram of standard G2 obtained in laboratory 3.

Peaks in elution order are: phenanthrene, fluoranthene, pyrene, benz[*a*]anthracene, chrysene, benzo[*b*]fluoranthene, benzo[*e*]pyrene, benzo[*a*]pyrene, perylene, indeno[123-*cd*]pyrene, benzo[*ghi*]perylene.

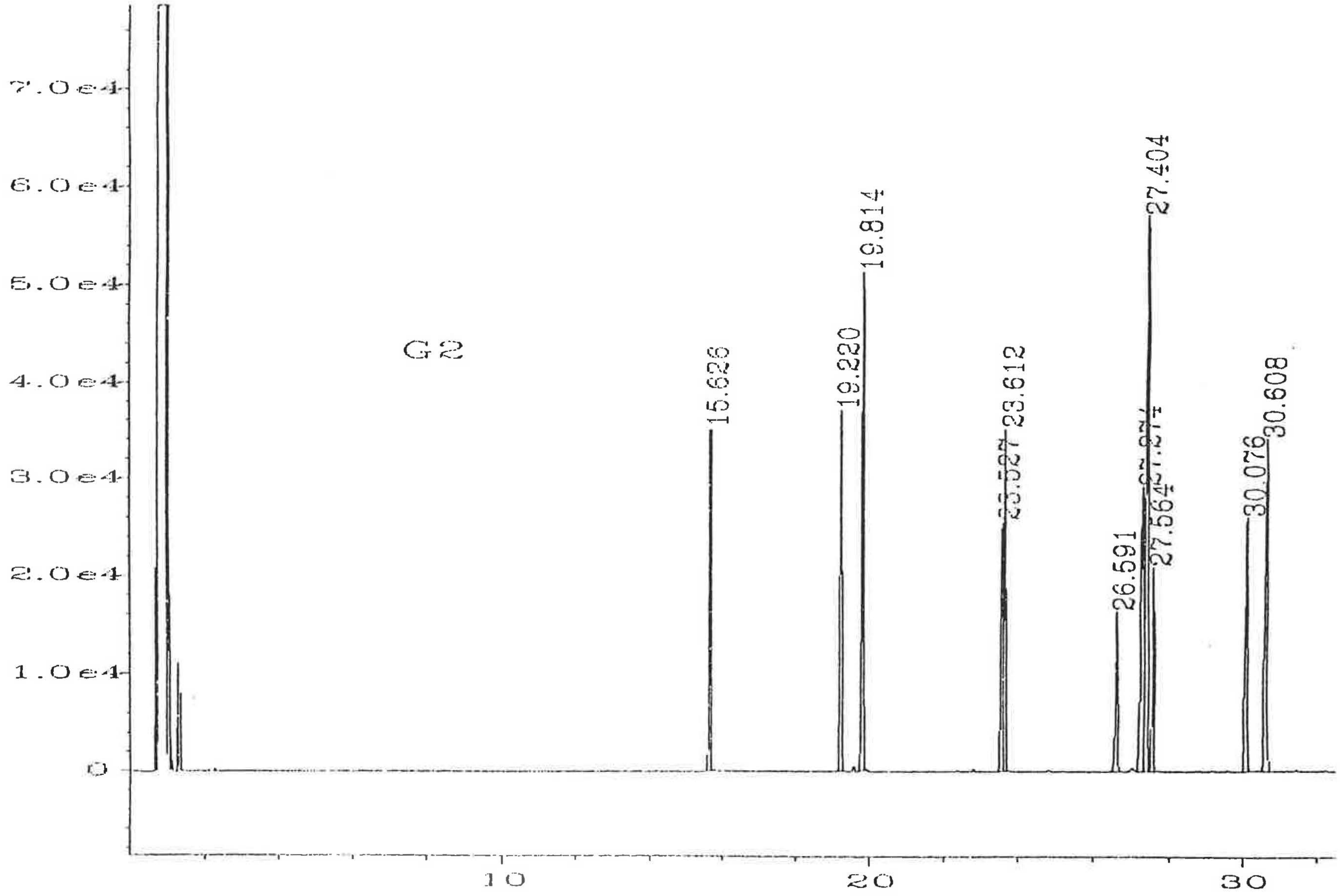


Figure 2. Total ion chromatogram (summation of MID signals) of standard G2 by laboratory 4. Peaks as for Figure 1.

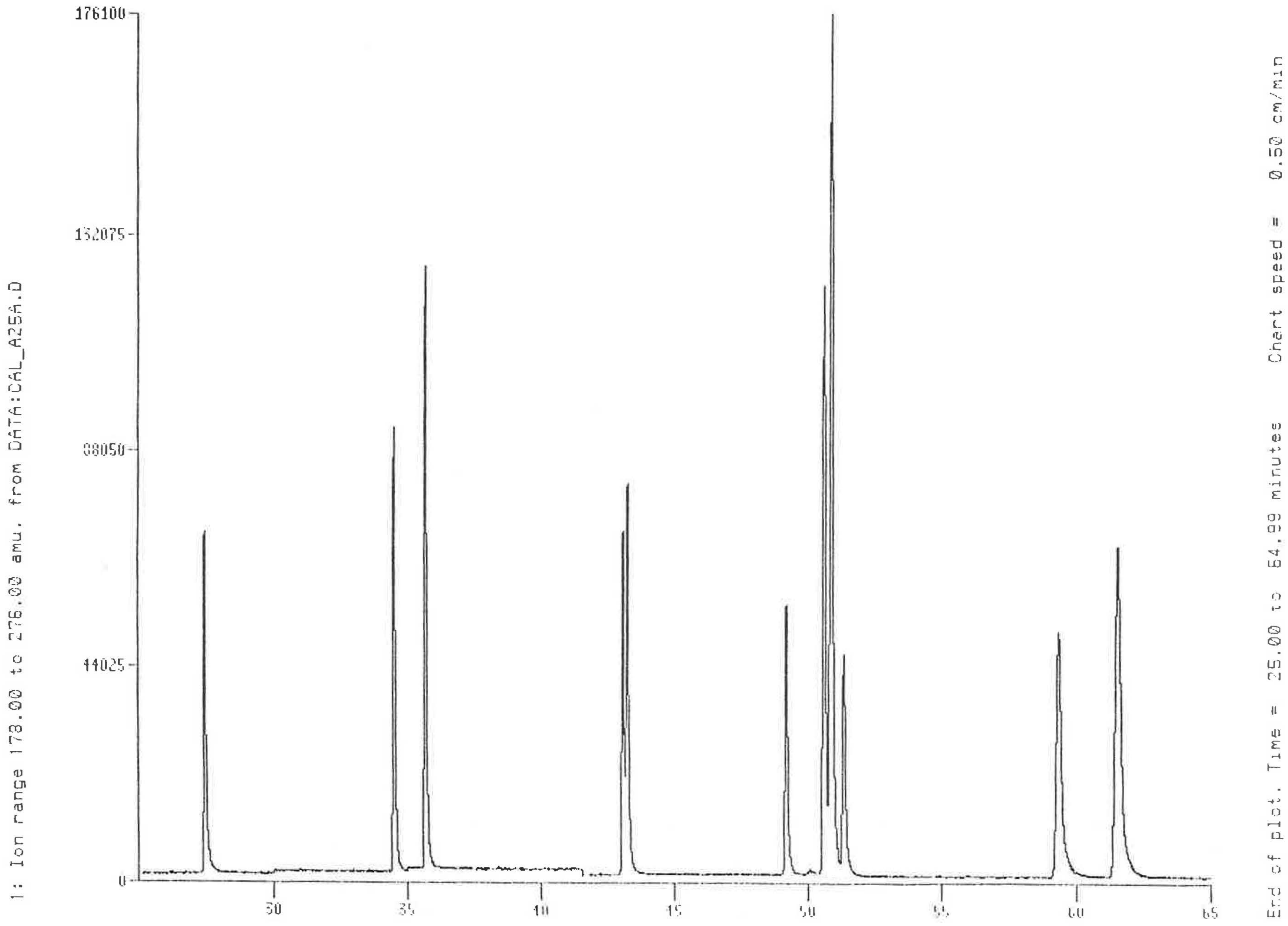
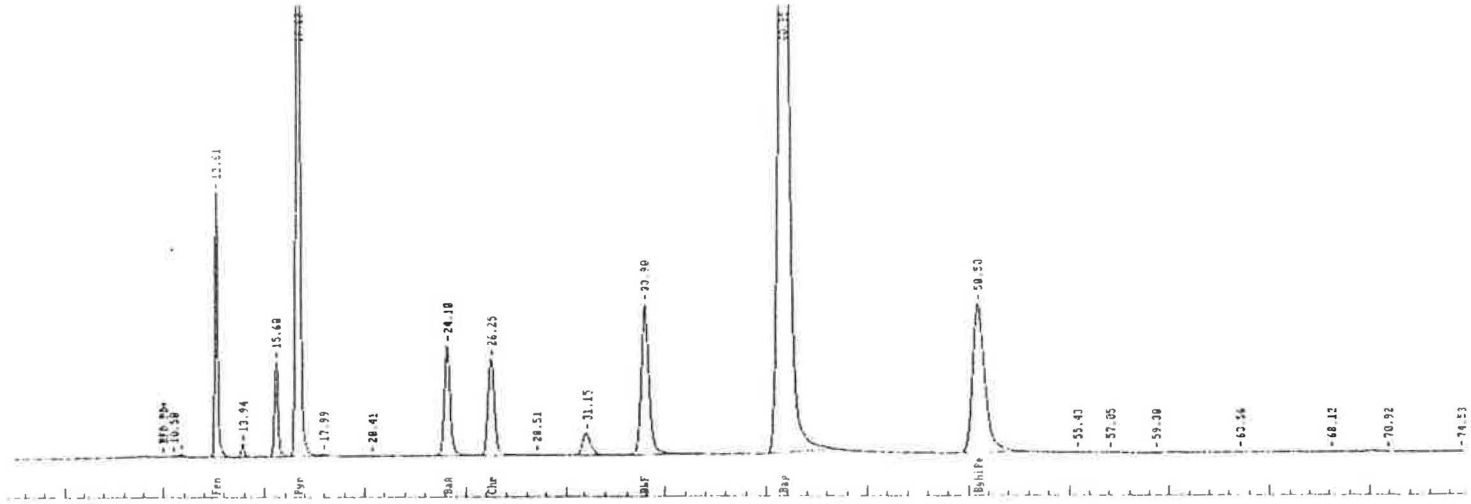


Figure 3 HPLC chromatograms from laboratory 7 of standard H2 obtained using two fluorescence detectors connected in series, each with its own programme of 2 and 6 wavelength pairs. Peaks in elution order are:
(a) phenanthrene, pyrene, benz[*a*]anthracene, chrysene, benzo[*b*]fluoranthene, benzo[*a*]pyrene and benzo[*ghi*]perylene
(b) fluoranthene, benz[*a*]anthracene, chrysene, benzo[*e*]pyrene, benzo[*a*]pyrene and indeno[123-*cd*]pyrene

(a)



(b)

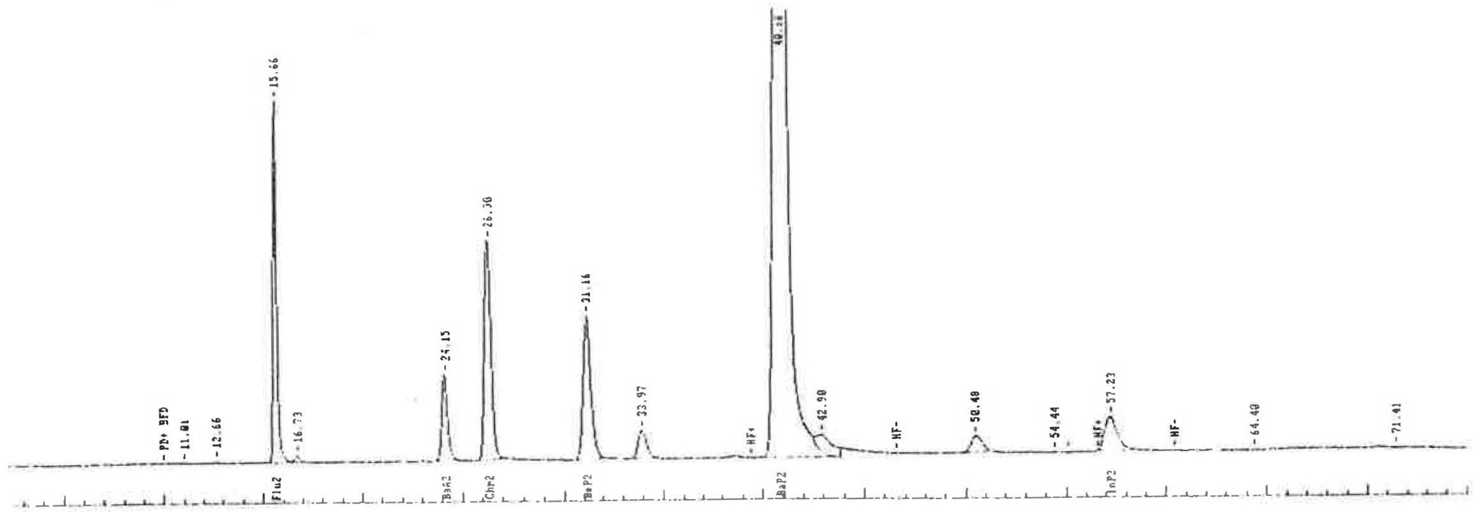
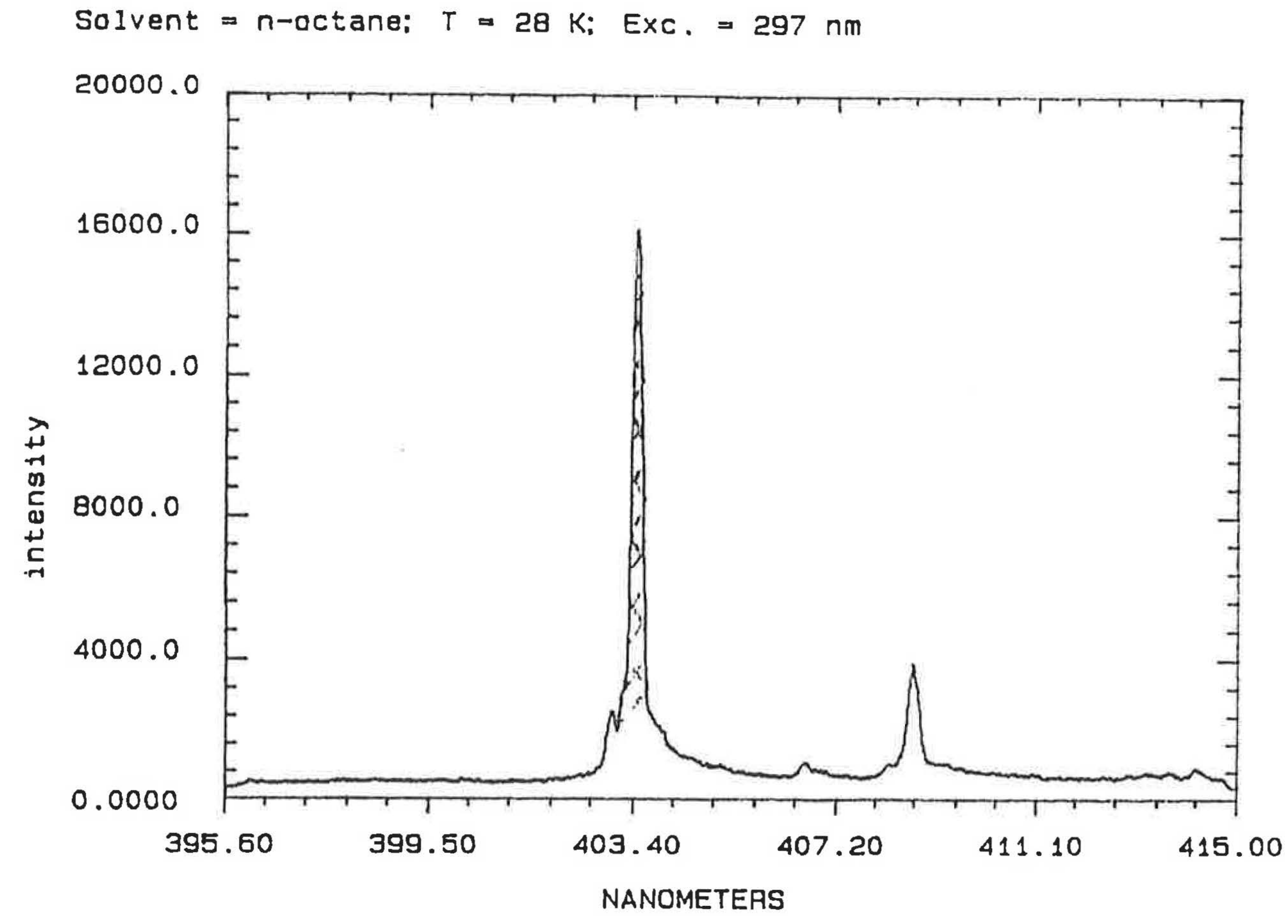
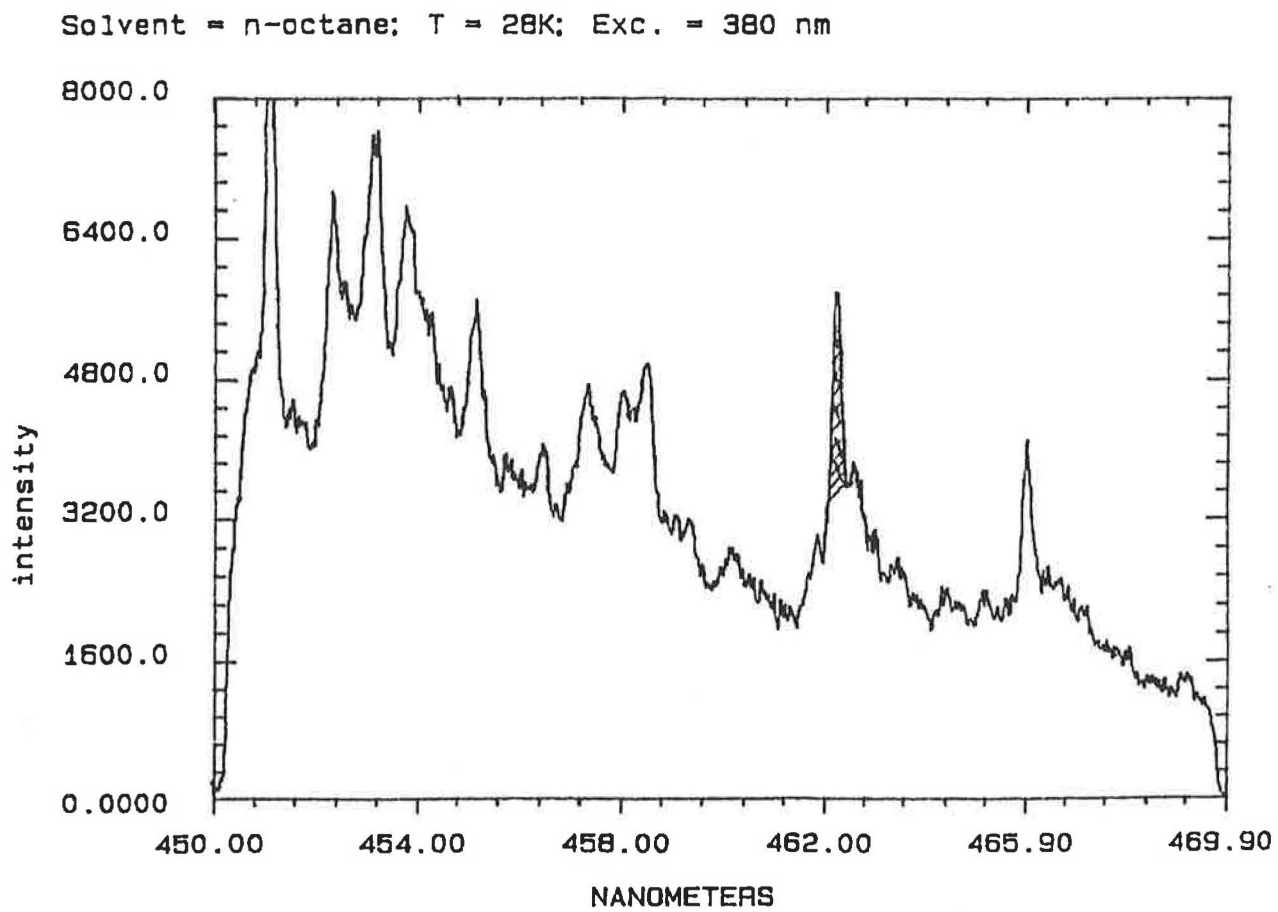


Figure 4. Examples of spectra obtained from standard G2 by laboratory 17 using the low temperature (Shpol'skii) fluorescence technique.
(a) benzo[*a*]pyrene (b) indeno[123-*cd*]pyrene

(a)

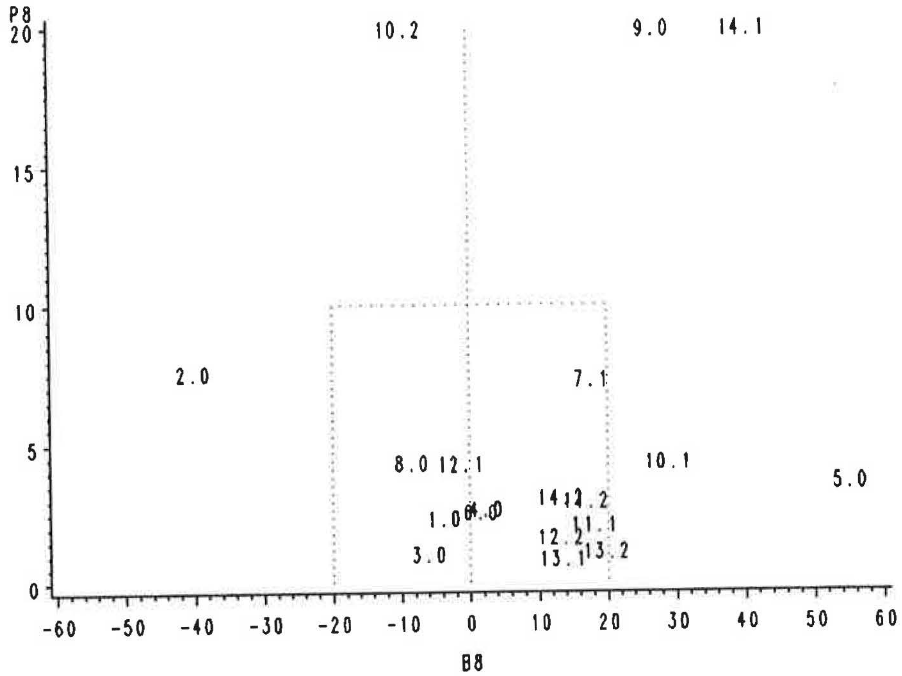


(b)



Guide to Figures 5ab

Precision versus Bias



Commentary

The objective of Figure 5 is to show which results are unsatisfactory, and whether this is because they are too biased (i.e. systematically too high or too low) or lack precision (i.e. repeated measurements of the same value are too different).

Above, we see precision (P) plotted against bias (B) for each laboratory. The example above is for Determinand 8, hence P8 Vs. B8. The lab number is used as the plotting symbol with extension 0 if the lab submitted only one set of results. Where there are two sets they are distinguished by the extension 1 or 2.

For reference a box has been drawn to enclose laboratories with bias less than 20% and precision better 10%.

Biases above 55% have been plotted at 55% (lab 5.0). Precisions above 20% have been limited to 20% (labs 10.2, 9.0, 14.1).

These plots makes it easy to see in which way (bias or precision) laboratory performance could be improved and for which determinands. For example in Phase 1, the results for lab 9.0 were generally unbiased but has poor precision for determinands 1, 2, 4, 7, 8, 9 and 10.

Figure 5a Phase 1

Precision V Bias

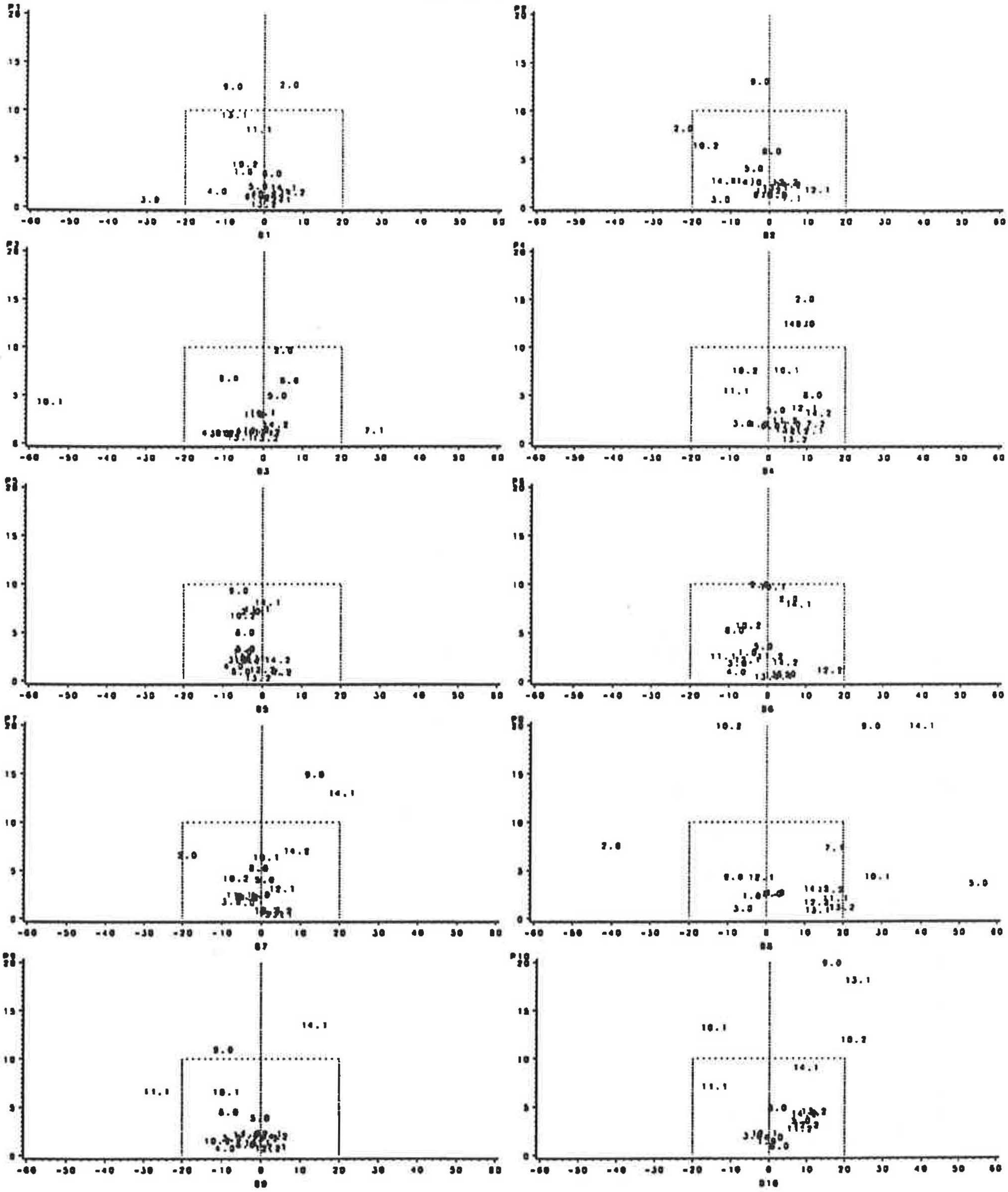
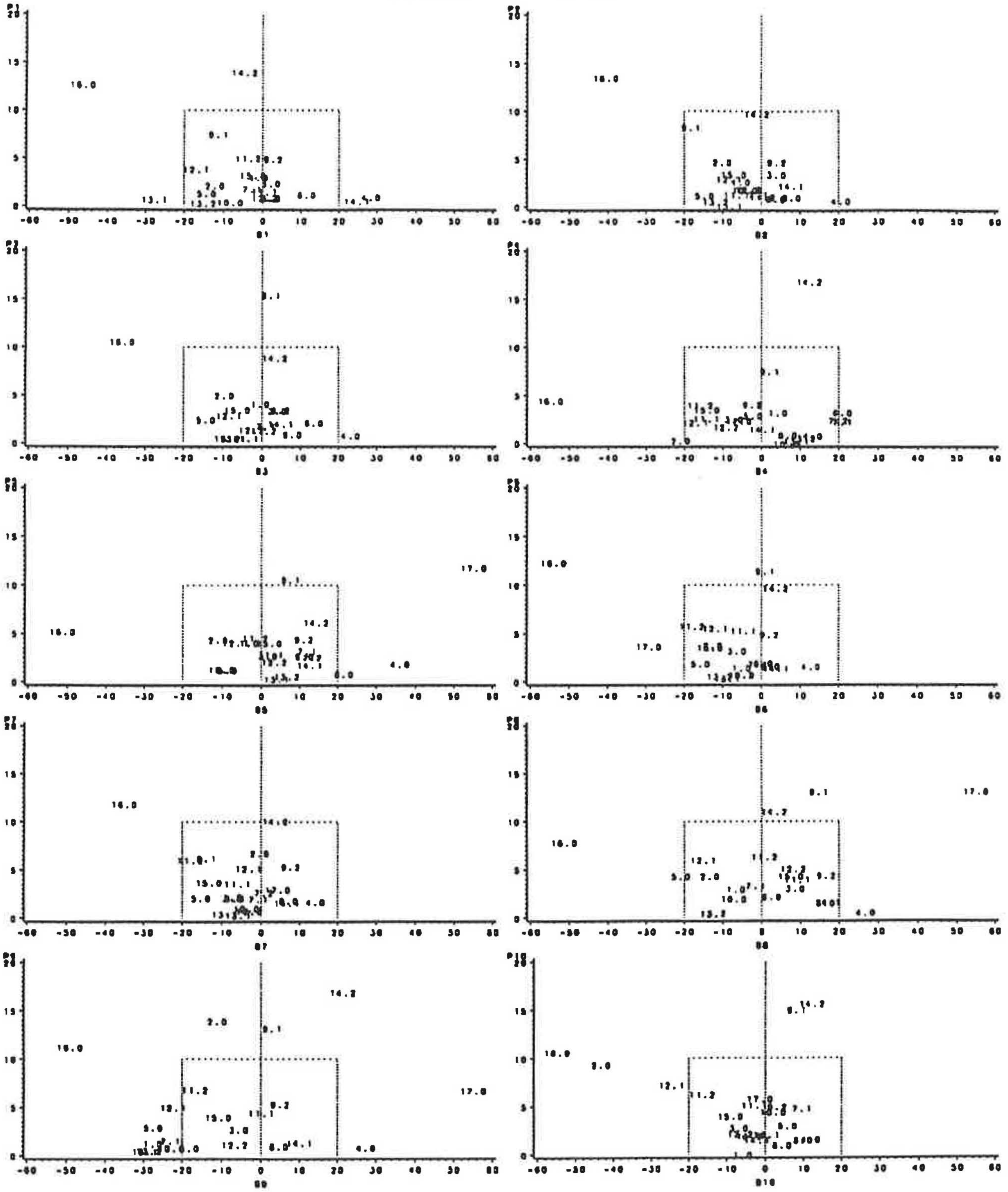


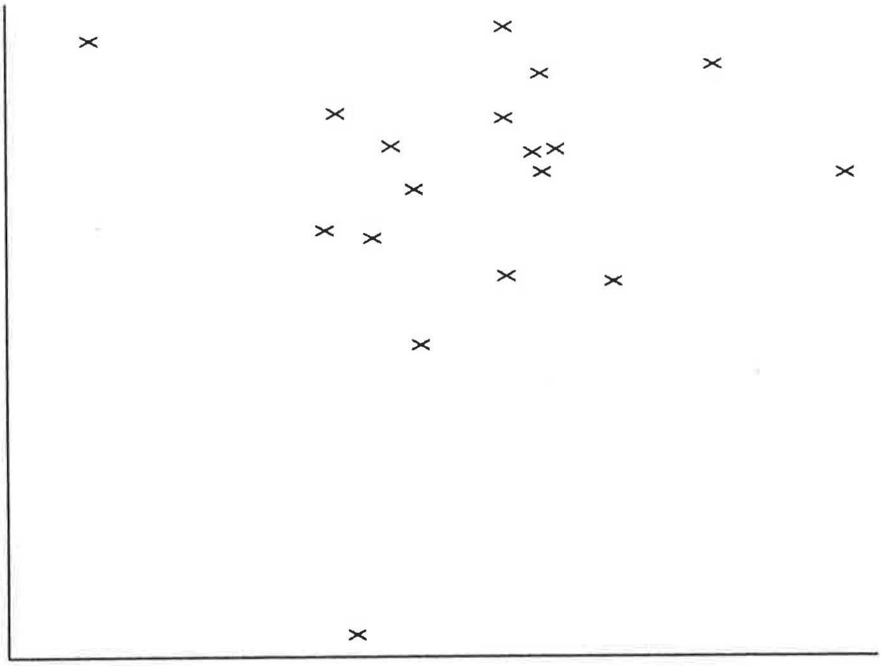
Figure 5b Phase 2

Precision V Bias



Guide to Figures 6ab 7ab

All pairwise Plots



Commentary

The objective of the series of plots in Figures 6 and 7 is to reveal any patterns in bias or precision either within a determinand or between determinands.

Figures 6a, b show an array of the laboratory biases for each determinand plotted against those for all other determinands. These plots can be thought of as a visual correlation matrix. The first row corresponds to determinand 1 plotted successively against determinands 2,3, ...10. The second row is determinand 2 plotted against determinands 3, 4, ... 10, and so on.

If there is no bias, the points within each graph will lie in an elliptical cluster with axes parallel to the x-y axes. If there is bias, the ellipse will be tilted, upwards if the bias is in the same direction, downwards if in opposite directions.

Outlying points which are distant from the ellipse could suggest an intermittent error.

However, the effectiveness of these graphs comes from looking across the full array of pairwise plots. For example, an outlying point can often be tracked across several plots. This may reveal a problem in only one determinand if the displacement occurs on only one axis, or a general problem in the laboratory if displacement occurs on several axes.

The example above shows the biases for determinand 1 plotted against the biases for determinand 2. Points seem to lie in an upwardly titled ellipse, with possibly two outliers.

Figures 7a and 7b show similar plots for the laboratory precisions.

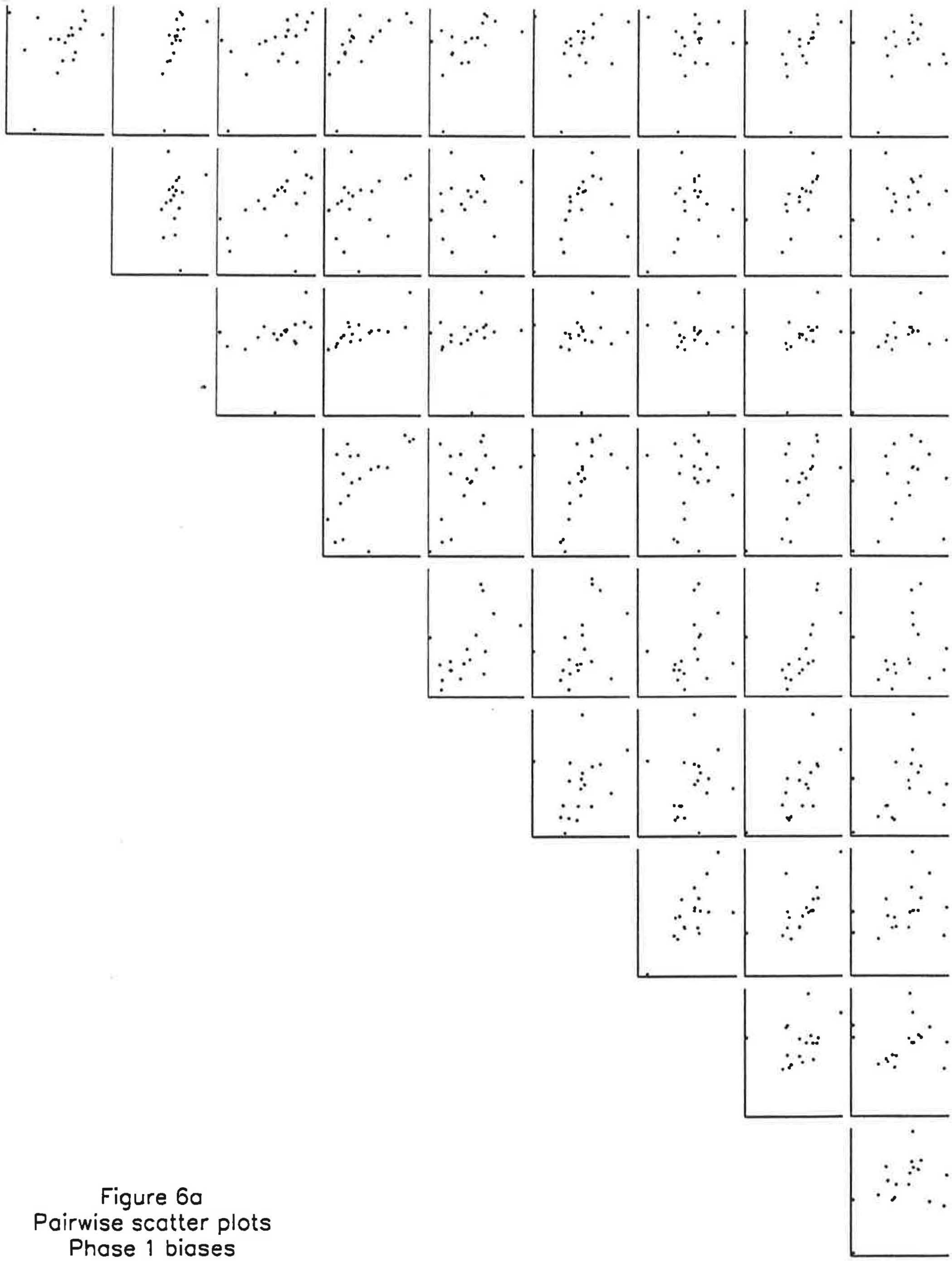


Figure 6a
Pairwise scatter plots
Phase 1 biases

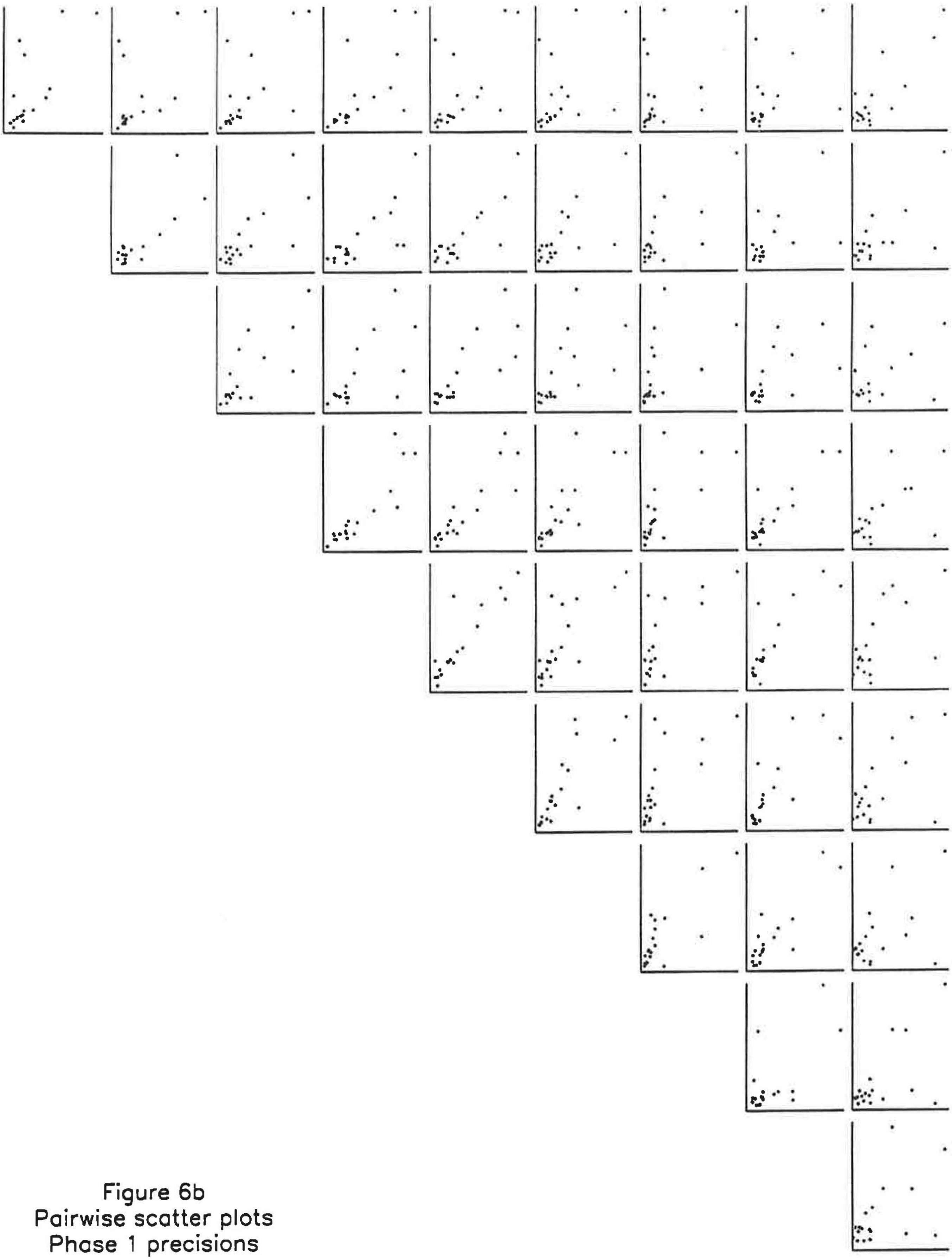


Figure 6b
Pairwise scatter plots
Phase 1 precisions

Figure 7a
Pairwise scatter plots
Phase 2 biases

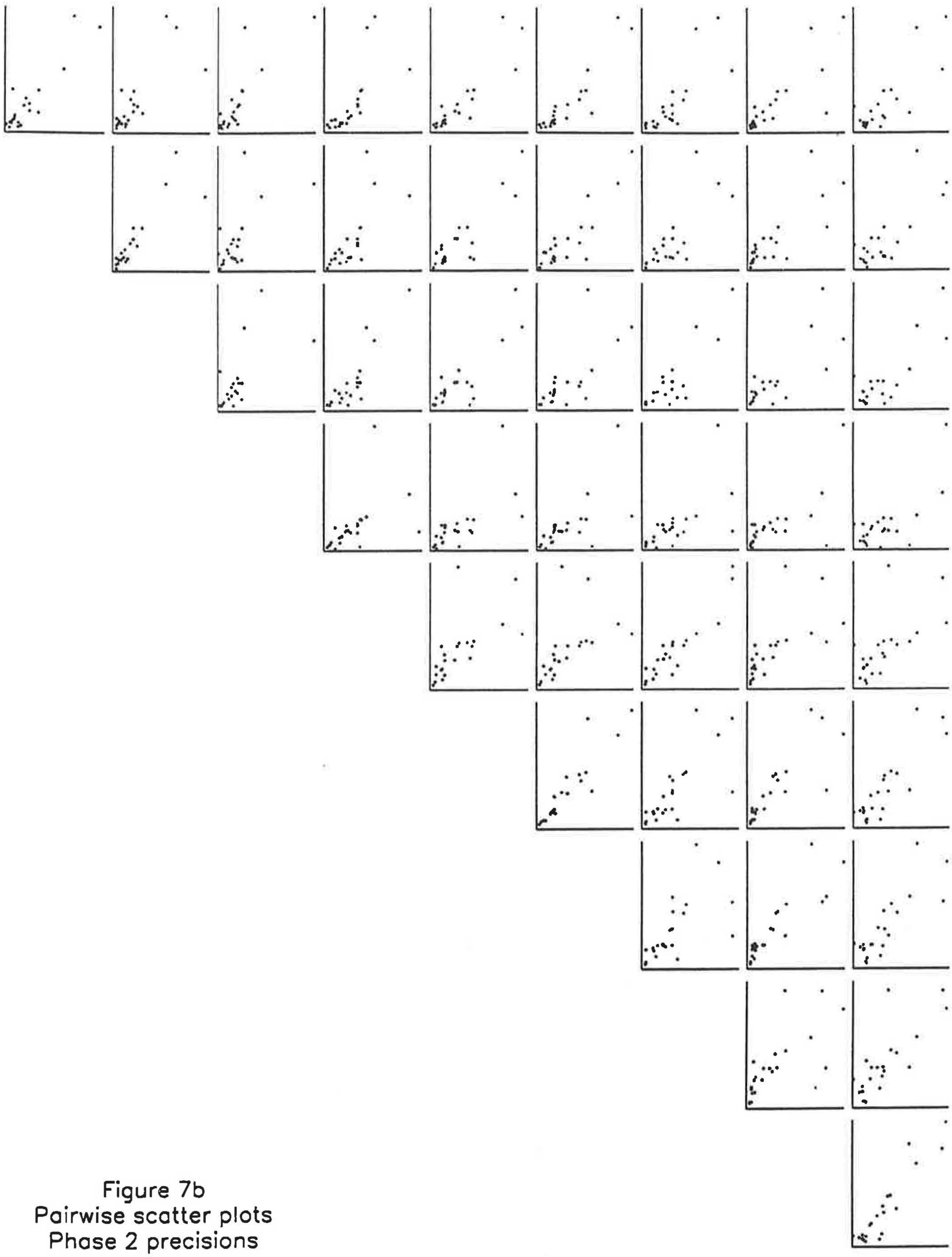
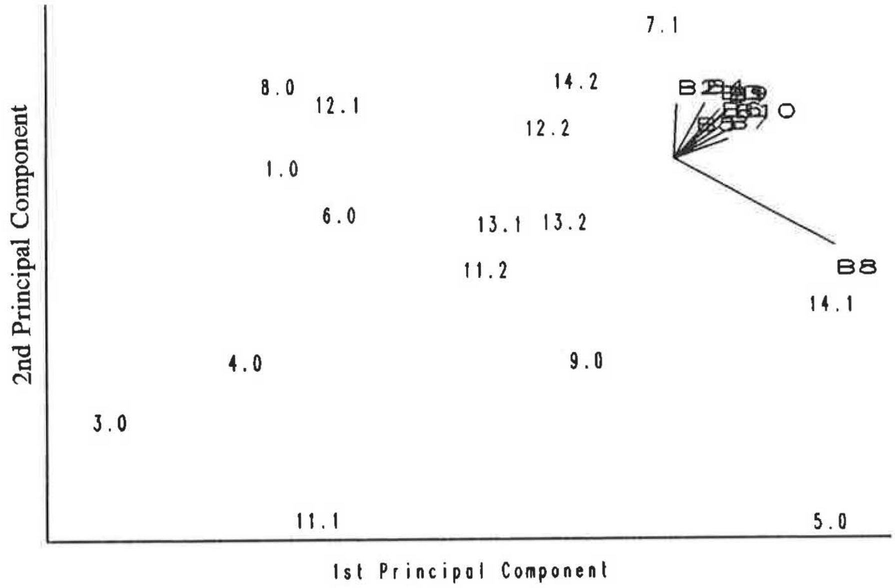


Figure 7b
Pairwise scatter plots
Phase 2 precisions

Guide to Figures 8ab 9ab

Principal Component Biplots



Commentary

Figure 8 plots the first two principal components obtained from a principal component analysis of the laboratory biases. The purpose here is to try and simplify the information plotted in Figure 6 by viewing the variability in the biases for all ten determinands in only one scatter plot.

The first principal component is obtained by constructing the weighted average of the biases giving the largest possible variance.

The second component is the weighted average which has the second greatest variance and is uncorrelated with the first.

The weight that each determinand has in the first and second components is indicated by the lines. There is one line for each determinand. The angle and size of a line indicates the magnitude of the contribution from a determinand to each component.

The lab number is used as the plotting symbol with an extension of zero when the lab submitted only one set of results. Where there were two sets they are distinguished by the extension 1 or 2.

Here we see that all lines point to the right, suggesting that the first component is a measure of overall average bias. The bias for determinand 8 makes a proportionally greater contribution to both components, and the second component seems to be a contrast between determinand 8 and the rest.

This can be useful for interpreting the positions of individual labs. For example, laboratory 5.0 has a large bias for determinand 8. This can also be seen in Figure 5a and the Guide to Figure 5a.

In Figure 9 the same thing is done for the precisions.

Principal Component Scores

Figure 8a
Phase 1 Bios

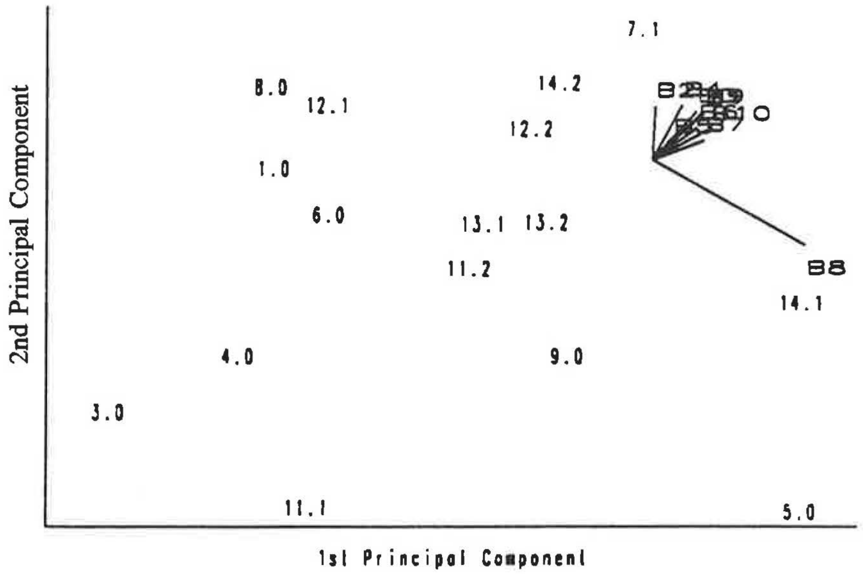
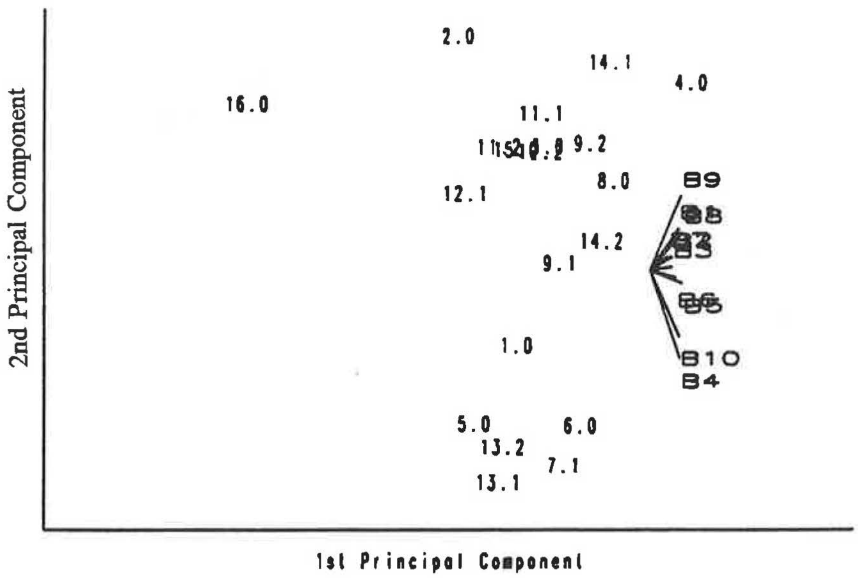


Figure 8b
Phase 2 Bios



Principal Component Scores

Figure 9a
Phase 1 Precision

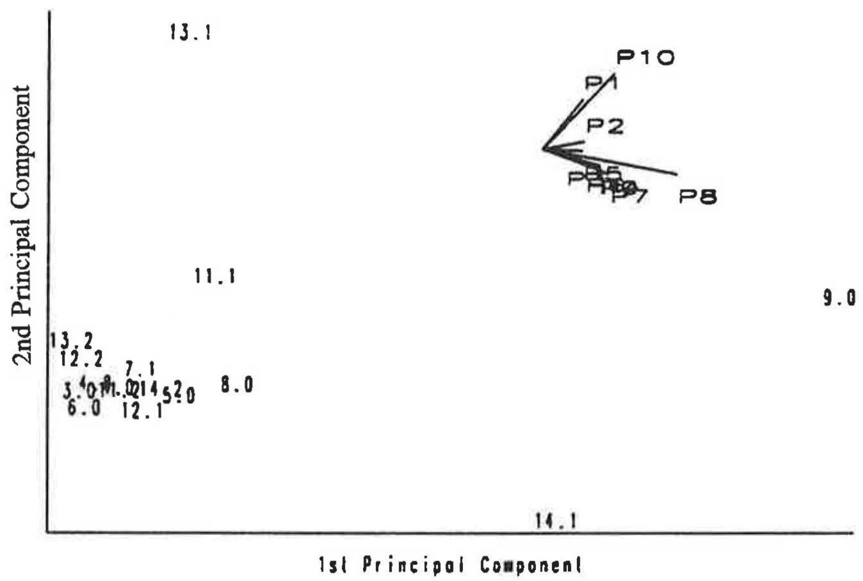
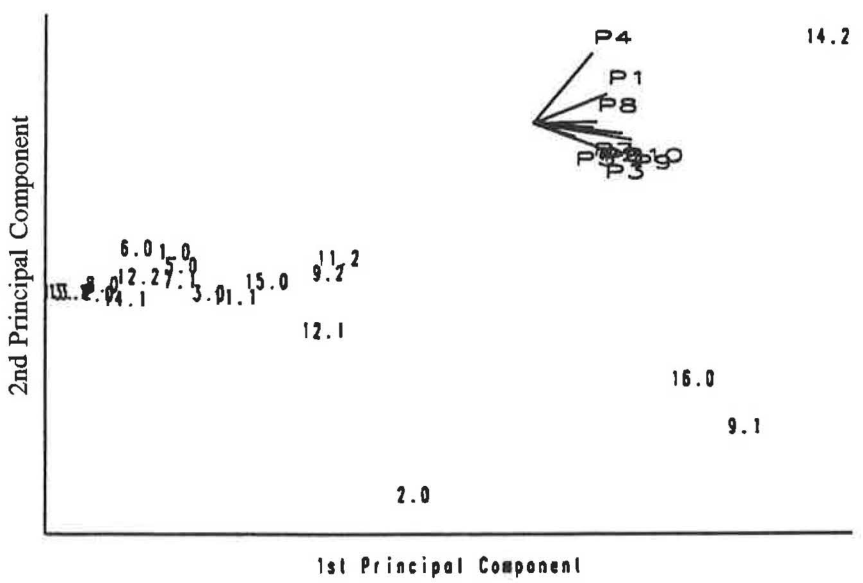


Figure 9b
Phase 2 Precision

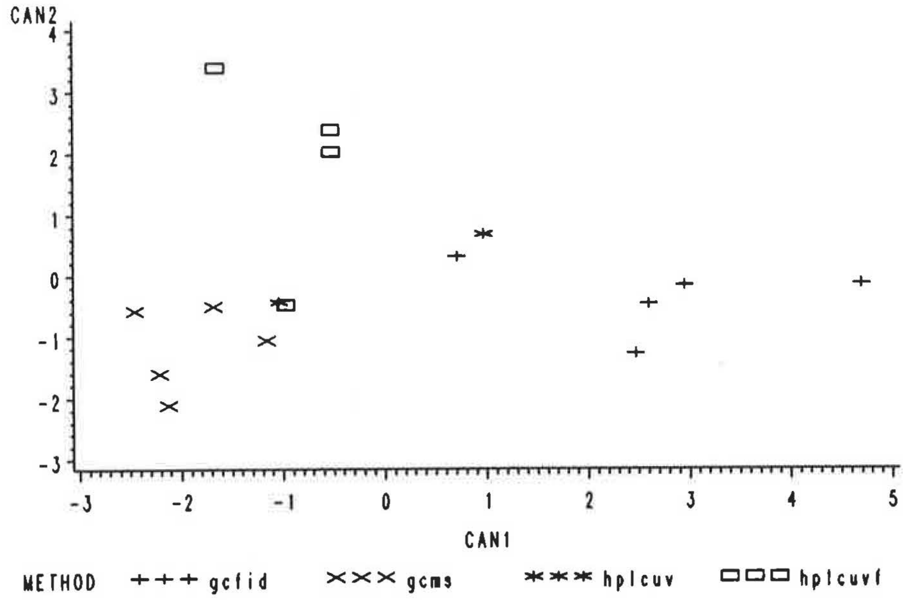


Guide to Figure 10ab

Canonical Discriminant Scores

Figure 10a

Phase 1 Bias



Commentary

Figure 10 gives a graphical summary of a canonical discriminant analysis of the biases. The objective of this analysis is to see if laboratories using the same method have similar biases, different from those of laboratories using other methods.

Effectively, this is an analysis of variance between analytical methods carried out on the biases of all ten determinands simultaneously.

The first canonical variable is obtained from the biases of the 10 determinands by constructing the weighted average which gives the largest variance between methods.

The second canonical variable is the weighted average which has the second largest variance between methods, and is uncorrelated with the first.

Laboratories are plotted with different symbols for each method.

For the Phase 1 results, there is visual evidence of some difference between methods, particularly along axis 1. The points for GCFID lie to the right and separate from the other methods. However, this difference was not significantly great.

Canonical Discriminant Scores

Figure 10a

Phase 1 Bias

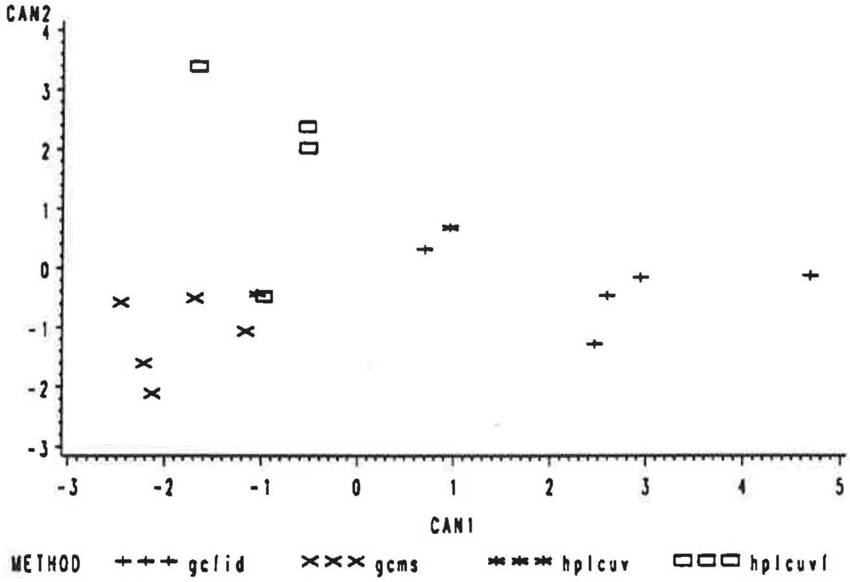
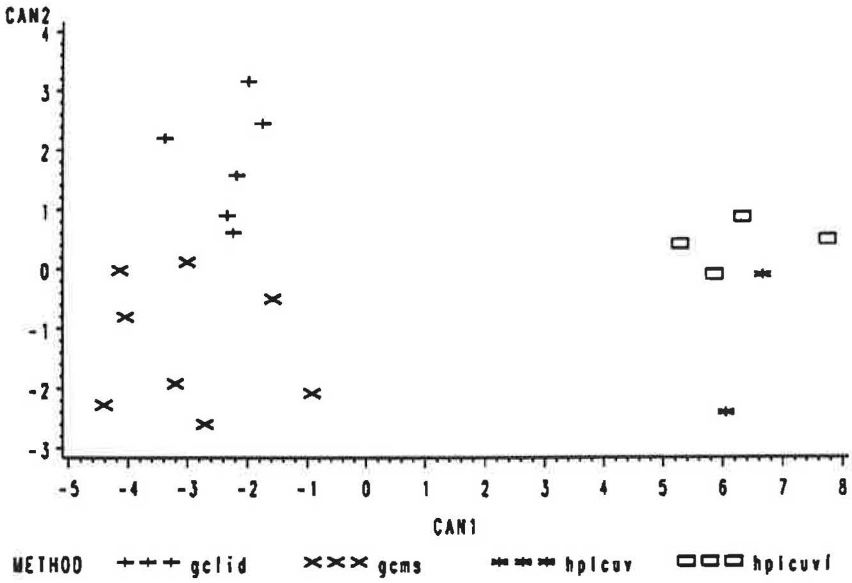


Figure 10b

Phase 2 Bias



Annex 1. Results submitted during Stage 1 (phase 1) of the exercise.
Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

LAB NO.	1						2						3					
COMPOUND																		
Phenanthrene	36.3	38.3	39.1	39.1	40.0	40.8	40.6	36.1	46.9	47.2	48.2		28.7	29.7	29.2	29.2	28.9	29.3
Fluoranthene	26.3	26.7	26.7	26.7	26.0	27.0	23.2	20.6	17.9	20.5	17.6	21.4	22.7	23.0	22.8	22.7	22.7	22.3
Pyrene	78.9	78.9	84.7	84.7	81.4	82.1	92.0	90.2	74.5	88.2	96.1		75.1	75.1	75.0	74.7	75.5	72.9
Benz[a]anthracene	22.6	21.7	23.0	22.6	22.3	22.6	19.4	21.9	20.4	24.2	27.9	25.3	19.7	19.9	20.2	20.3	19.3	19.2
Chrysene	36.6	36.6	37.9	38.1	38.4	39.9	41.8	41.3	36.9	34.9	39.0		37.6	37.0	38.5	38.3	37.4	36.0
Benzo[e]pyrene	47.0	49.1	50.0	50.0	49.7	51.9	54.0	60.9	51.8	50.5	58.8		48.6	48.7	48.3	49.7	47.5	46.8
Benzo[a]pyrene	34.3	32.3	32.3	33.2	32.6	33.9		28.2	28.3	28.4	23.2	26.6	30.8	30.9	30.6	31.6	30.1	29.9
Benzo[b]fluoranthene	6.8	6.3	6.5	6.5	6.5	6.6	4.6	4.4	4.0	3.4	3.5	4.5	6.5	6.4	6.4	6.4	6.3	6.3
Benzo[ghi]perylene	47.1	45.6	47.6	48.1	47.6	49.1							45.3	45.6	44.8	46.7	44.5	44.1
Indeno[123-cd]pyrene	10.8	10.4	10.8	10.8	10.8	10.8							10.3	10.4	10.4	10.7	10.1	10.1
METHOD	HPLC/UVF						GC/MS						GC/FID					

[Annex 1 : continued]

Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

	4						5						6					
Phenanthrene	37.7	36.0	35.9	35.9	36.0	36.1	41.8	41.4	40.5	39.9	40.1	39.4	39.3	40.0	40.6	40.1	40.6	40.2
Fluoranthene	25.7	24.2	23.8	25.1	25.2	25.0	26.3	26.2	24.6	24.0	24.7	23.8	24.9	25.6	25.9	25.5	25.9	25.5
Pyrene	74.7	72.2	72.6	72.3	73.6	72.4	89.9	88.0	86.1	82.4	92.8	82.0	77.9	80.4	81.2	80.7	80.7	80.2
Benz[a]anthracene	21.5	20.2	20.5	20.6	20.5	20.7	22.9	21.9	21.2	21.5	21.4	20.7	20.8	21.2	21.4	21.3	22.0	21.1
Chrysene	38.3	36.5	36.6	37.0	37.0	36.8	40.5	39.3	37.3	37.2	37.5	37.5	37.6	37.3	38.0	37.5	38.5	37.7
Benzo[e]pyrene	49.1	47.5	48.3	48.1	47.9	48.0	55.5	52.0	50.6	50.6	51.4	50.5	54.6	54.9	55.0	54.8	55.8	54.7
Benzo[a]pyrene	32.9	31.2	31.5	31.7	32.0	31.7	35.8	34.4	32.2	32.6	32.8	33.1	31.0	32.9	32.0	32.5	32.3	31.0
Benzo[b]fluoranthene	7.2	7.0	6.7	7.1	6.8	6.9	10.6	10.6	10.5	10.4	11.1	10.8	6.8	6.8	7.2	6.7	7.0	6.9
Benzo[ghi]perylene	45.0	43.9	44.1	44.2	44.7	44.2	51.4	50.6	46.5	47.4	47.6	48.6	45.8	46.8	47.5	47.0	47.3	46.8
Indeno[123-cd]pyrene	11.3	10.7	10.9	10.9	11.0	10.8	12.2	11.2	11.6	11.4	11.8	12.1	10.9	11.1	11.2	11.2	11.1	11.1
METHOD	GC/MS						HPLC/UVF						HPLC/UV					

[Annex 1 : continued]

Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

	7						7						8					
Phenanthrene	43.3	42.5	42.6	43.1	43.0	43.3							44.4	42.8	41.4	42.0	40.3	41.0
Fluoranthene	27.4	27.3	27.5	27.9	27.7	27.2							27.6	23.8	26.6	27.6	24.8	26.5
Pyrene	106	107	108	109	109	108							94.8	82.0	91.0	95.1	83.6	90.6
Benz[a]anthracene	23.6	23.2	24.0	23.9	23.9	23.7	23.6	23.1	23.8	24.4	23.9	24.1	24.3	21.8	24.2	24.5	22.8	24.2
Chrysene	41.8	41.4	41.7	42.0	42.5	41.4	41.7	41.7	42.1	42.5	42.5	41.7	39.3	34.7	39.4	39.7	36.6	39.3
Benzo[e]pyrene	54.6	54.1	54.5	54.8	54.3	54.8							47.5	43.2	48.8	50.4	46.7	50.8
Benzo[a]pyrene	34.9	35.0	35.0	35.3	35.1	34.9	34.9	34.6	34.8	35.4	35.1	35.2	32.8	29.8	33.9	34.1	32.4	34.7
Benzo[b]fluoranthene	8.5	8.2	7.7	8.6	7.4	7.6							6.2	5.7	6.3	6.5	6.1	6.5
Benzo[ghi]perylene	50.1	50.4	50.8	51.3	50.7	51.3							44.3	40.7	45.9	46.1	44.5	47.0
Indeno[123-cd]pyrene	11.7	11.7	12.3	12.0	12.7	12.8							11.0	10.0	11.5	11.3	11.1	11.3
METHOD	HPLC/UVF						HPLC/UVF*						GC/MS					

* Different set of detection wavelengths

[Annex 1 : continued]

Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

	9						10							10				
Phenanthrene	45.2	38.8	29.4	37.3	40.1	36.3								36.4	40.0	40.6	38.0	40.6
Fluoranthene	30.0	22.4	21.6	26.3	28.4	23.4								21.0	22.0	23.6	19.2	22.6
Pyrene	78.7	70.3	71.7	81.2	84.5	73.0	37.0	29.0	33.2	36.0	36.4	30.0	38.6					
Benz[a]anthracene	26.2	22.6	21.4	25.5	24.4	19.2	21.6	21.6	19.2	22.0	23.6	23.2	24.0	21.6	21.6	19.4	18.4	18.4
Chrysene	41.8	34.9	37.5	40.5	38.9	31.6								36.8	42.6	37.4	35.4	37.4
Benzo[e]pyrene	53.6	51.6	52.1	54.3	54.9	40.8	48.0	49.6	46.0	55.4	60.0	56.0	56.0	50.8	45.0	49.4	53.4	50.0
Benzo[a]pyrene	39.5	41.4	35.6	41.6	39.5	28.5	37.6	35.0	34.0	32.0	32.0	32.4	32.0	30.0	32.0	30.0	30.0	33.0
Benzo[b]fluoranthene	8.0	12.1	9.8	8.0	8.3	5.6	8.6	9.0	8.4	9.0				4.0	6.6	5.6	7.4	7.0
Benzo[ghi]perylene	46.5	40.5	43.5	49.9	48.6	35.6	50.0	42.0	40.8	46.0	44.6	41.6	46.0	43.6	43.6	43.6	44.0	42.0
Indeno[123-cd]pyrene	14.1	10.8	12.3	14.9	14.1	9.3	8.0	8.0	7.2	10.4	10.0	10.6	10.4	11.2	13.2	13.2	14.8	13.6
METHOD	GC/FID						HPLC/UVF							HPLC/UV				

[Annex 1 : continued]

Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

	11						11						12					
Phenanthrene	45.0	40.2	41.2	34.7	40.9	41.3	41.7	40.6	40.6	41.4	40.4	41.5	40.2	40.7	41.5	41.5	41.9	41.3
Fluoranthene	24.8	24.4	24.7	23.6	24.5	22.9	26.5	25.9	25.4	26.1	25.3	26.3	28.7	29.0	29.3	29.4	30.1	28.9
Pyrene	82.0	83.9	83.2	84.7	83.4	82.5	84.1	82.2	81.2	83.0	80.9	83.4	76.5	77.4	78.1	77.7	79.3	78.3
Benz[a]anthracene	20.4	19.6	19.1	21.0	18.0	18.3	22.8	22.3	21.5	22.1	21.5	22.4	23.0	23.0	23.6	22.9	24.4	22.0
Chrysene	40.7	39.7	37.6	44.6	37.5	36.3	39.6	38.8	37.5	38.3	37.3	38.9	37.7	37.7	38.4	38.3	40.2	37.6
Benzo[e]pyrene	46.0	47.4	47.6	43.8	46.4	46.7	54.7	53.1	51.4	52.4	51.1	53.7	49.5	49.4	50.5	50.2	50.6	47.4
Benzo[a]pyrene	31.0	31.8	30.5	31.1	32.7	30.6	32.7	32.1	31.0	31.6	31.1	32.7	35.1	34.8	35.6	35.6	35.6	32.9
Benzo[b]fluoranthene	8.0	8.2	8.1	7.8	8.1	7.9	8.2	8.0	7.7	7.9	7.7	8.1	6.7	6.8	7.0	7.0	6.4	6.3
Benzo[ghi]perylene	37.8	41.5	35.1	34.1	32.9	33.5	47.6	47.1	45.5	46.5	46.1	48.5	49.6	49.7	50.7	51.4	48.6	48.5
Indeno[123-cd]pyrene	9.7	10.6	9.0	9.0	8.5	8.7	11.9	11.6	11.3	11.7	11.4	12.1	10.7	10.6	10.9	11.0	10.4	10.4
METHOD	GC/MS						GC/FID						GC/MS					

[Annex 1 : continued]

Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

	12						13						13					
Phenanthrene	41.1	41.8	41.8	42.2	42.0	42.3	42.7	41.0	40.1	37.2	34.0	32.9	41.0	41.0	40.9	41.2	41.2	41.3
Fluoranthene	26.2	28.3	26.7	27.2	27.1	26.8	27.0	26.6	26.8	26.4	25.6	25.8	26.1	26.0	27.0	26.2	26.2	26.2
Pyrene	83.3	84.7	84.0	85.8	86.1	84.7	78.1	78.7	78.7	79.0	79.3	79.8	84.0	83.7	85.1	84.2	84.8	84.5
Benz[a]anthracene	22.3	22.7	22.6	23.1	23.0	22.7	22.6	22.6	22.0	22.3	22.3	21.8	22.6	22.6	22.5	22.6	22.7	22.8
Chrysene	39.2	40.0	39.7	40.6	40.4	39.8	40.2	39.6	38.0	39.3	38.0	38.2	39.4	39.6	39.3	39.6	39.5	39.7
Benzo[e]pyrene	59.9	61.0	60.4	61.6	61.4	60.9	52.5	52.1	51.9	52.6	52.2	52.4	53.6	53.5	53.1	53.9	54.0	54.2
Benzo[a]pyrene	33.2	33.7	33.5	34.1	33.9	33.8	33.8	34.0	34.0	34.1	33.9	34.3	33.4	33.7	33.5	33.7	33.6	34.2
Benzo[b]fluoranthene	7.6	7.6	7.6	7.7	7.9	7.7	7.8	7.7	7.7	7.7	7.6	7.7	8.2	8.2	8.2	8.0	8.1	8.1
Benzo[ghi]perylene	48.9	49.6	49.8	49.8	50.1	49.6	47.6	47.7	47.9	48.7	49.0	49.1	48.8	48.8	48.6	49.2	49.2	51.1
Indeno[123-cd]pyrene	11.6	11.6	11.8	12.0	12.5	11.7	14.0	12.3	9.9	13.8	15.5	14.4	11.8	11.7	11.8	11.9	12.3	13.0
METHOD	GC/FID						HPLC/UVF						HPLC/UV					

[Annex 1 : continued]

Concentration of PAH in unknown solution STD2 (ng μl^{-1}).

	14						14						Nominal Concentrations
Phenanthrene	44.3	43.8	43.1	42.7	43.4	41.8	44.6	44.8	42.9	44.2	44.0	44.2	41.2
Fluoranthene	22.1	23.0	23.9	23.1	23.3	22.0	28.0	26.3	26.7	27.6	27.5	27.5	26.0
Pyrene	83.8	88.0	85.3	82.2	83.2	79.9	88.7	84.5	85.0	88.0	85.8	87.4	84.0
Benz[a]anthracene	19.6	25.3	22.5	26.4	21.8	20.6	23.9	25.0	23.0	24.4	23.9	23.6	21.2
Chrysene	37.9	45.1	38.7	44.2	39.2	37.8	41.9	41.1	41.1	43.1	40.5	41.4	40.0
Benzo[e]pyrene	52.8	62.9	54.1	60.7	55.1	53.7	55.4	55.8	55.4	54.1	53.0	55.0	52.4
Benzo[a]pyrene	34.2	45.6	41.0	42.2	41.5	35.4	34.4	35.5	36.6	35.9	40.6	34.1	33.2
Benzo[b]fluoranthene	8.9	11.6	9.8	9.5	9.9	7.4	7.8	7.7	7.3	7.9	7.6	7.8	6.8
Benzo[ghi]perylene	50.7	61.9	60.7	57.9	57.1	44.5	51.9	50.4	49.5	49.4	50.5	51.5	48.8
Indeno[123-cd]pyrene	10.4	12.9	12.7	11.6	12.4	11.1	12.5	11.7	11.3	11.9	12.1	11.3	10.8
METHOD	GC/MS						GC/FID						

Annex 2. Results submitted during Stage 1 (phase 2) of the exercise.
Concentration of PAH in unknown solution H2 (ng μl^{-1}).

LAB NO.	1						5						6					
COMPOUND																		
Phenanthrene	132	142	142	140	144	142	120	119	124	121	123	123	144	144	144	145	147	146
Fluoranthene	82.0	88.0	88.0	88.0	88.0	88.0	77.2	80.5	77.9	78.7	77.8	78.4	95.0	95.0	94.0	96.0	96.0	96.0
Pyrene	54.0	60.0	58.0	60.0	60.0	60.0	48.2	49.3	51.6	51.1	50.8	52.0	67.0	66.0	66.0	66.0	68.0	69.0
Benz[a]anthracene	16.0	17.0	17.0	16.0	17.0	17.0	14.8	15.9	15.7	15.6	15.6	16.2	19.0	19.0	19.0	19.0	20.0	20.0
Chrysene	28.0	30.0	32.0	30.0	30.0	30.0	27.6	28.3	28.7	28.6	28.2	28.4	33.0	35.0	34.0	35.0	35.0	35.0
Benzo[e]pyrene	66.0	68.0	68.0	66.0	68.0	68.0	57.8	61.9	59.7	59.3	60.2	59.8	70.0	71.0	71.0	73.0	70.0	73.0
Benzo[a]pyrene	156	166	166	162	166	162	142	150	148	152	150	152	163	162	161	161	170	167
Benzo[b]fluoranthene	48.0	52.0	52.0	52.0	52.0	52.0	40.6	45.7	42.0	41.7	46.5	44.3	56.0	57.0	55.0	57.0	55.0	58.0
Benzo[ghi]perylene	46.0	46.0	48.0	46.0	46.0	46.0	43.2	45.2	47.7	46.8	47.5	48.1	52.0	52.0	52.0	52.0	53.0	53.0
Indeno[123-cd]pyrene	32.0	32.0	32.0	32.0	32.0	32.0	35.1	35.9	37.2	37.0	35.9	34.6	37.0	37.0	37.0	38.0	37.0	38.0
METHOD	HPLC/UVF						HPLC/UVF						HPLC/UV					

[Annex 2 : continued]

Concentration of PAH in unknown solution H2 (ng μl^{-1}).

	7						7						10					
Phenanthrene	141	140	138	136	139	134							131	130	130	130	130	129
Fluoranthene	91.9	92.8	92.0	89.4	91.2	90.2							91.0	88.0	88.0	88.0	86.0	87.0
Pyrene	61.0	60.6	59.2	59.0	59.1	58.2												
Benz[a]anthracene	19.5	19.7	19.7	19.2	18.8	19.1	19.2	19.4	19.2	18.9	18.6	19.7	17.0	17.0	17.0	17.0	17.0	17.0
Chrysene	34.6	34.7	33.8	34.8	36.5	33.7	34.8	34.4	34.6	35.4	36.6	35.0	28.0	28.0	28.0	28.0	27.0	28.0
Benzo[e]pyrene	71.2	71.5	70.6	68.8	68.5	71.0							68.0	67.0	67.0	68.0	67.0	67.0
Benzo[a]pyrene	176	178	173	171	171	179	178	179	176	172	173	185	171	170	169	171	167	167
Benzo[b]fluoranthene	55.9	55.8	54.4	52.2	51.5	54.8							53.0	51.0	51.0	51.0	50.0	50.0
Benzo[ghi]perylene	49.3	50.1	49.0	47.8	48.7	50.6							50.0	49.0	49.0	50.0	49.0	49.0
Indeno[123-cd]pyrene	36.3	35.0	36.4	37.6	39.1	39.0							36.0	36.0	34.0	33.0	33.0	36.0
METHOD	HPLC/UVF						HPLC/UVF*						HPLC/UV					

*See Phase 1

[Annex 2 : continued]

Concentration of PAH in unknown solution H2 (ng μl^{-1}).

	13						13						Nominal Concentration (solution H2)
Phenanthrene	104	103	103	102	102	101	120	120	120	120	121	121	142
Fluoranthene	84.3	84.1	84.3	84.4	84.4	84.4	80.5	80.5	80.7	81.0	81.5	81.8	92
Pyrene	54.1	54.2	54.4	54.5	54.8	54.9	53.4	53.4	53.6	53.7	53.9	54.2	59
Benz[a]anthracene	17.4	17.4	17.4	17.4	17.4	17.5	17.6	17.6	17.7	17.7	17.8	17.8	16
Chrysene	32.2	32.1	32.1	32.3	32.3	32.3	32.8	32.9	33.0	33.1	33.2	33.3	31
Benzo[e]pyrene	64.6	64.4	64.5	64.6	64.9	65.0	62.8	63.0	63.0	63.3	63.6	63.9	71
Benzo[a]pyrene	165	165	165	166	166	166	159	159	159	159	160	161	176
Benzo[b]fluoranthene	47.8	47.8	47.8	48.0	48.2	48.3	47.8	47.8	48.0	48.0	48.3	48.5	55
Benzo[ghi]perylene	44.5	44.8	45.0	45.1	45.2	45.3	45.4	45.4	45.6	45.6	45.9	46.3	64
Indeno[123-cd]pyrene	32.7	34.3	34.2	34.2	34.6	34.6	32.2	32.2	32.3	32.4	32.5	33.9	34
METHOD	HPLC/UVF						HPLC/UV						

[Annex 2 : continued]

Concentration of PAH in unknown solution G2 (ng μl^{-1}).

	2		3						4					
Phenanthrene	38.8	40.2	44.8	46.9	46.4	47.3	44.7	45.7	57.5	58.2	57.9	58.1	57.8	57.0
Fluoranthene	44.1	47.5	50.7	53.1	51.5	55.6	52.7	54.0	61.4	61.6	61.4	62.0	61.2	61.0
Pyrene	59.0	63.8	67.9	70.8	68.8	74.4	70.3	71.8	83.4	83.0	84.3	83.7	84.5	83.5
Benz[a]anthracene	37.0	37.2	42.9	44.8	42.8	45.5	42.8	43.0	53.1	53.1	53.2	52.7	53.9	53.3
Chrysene	38.6	41.4	45.0	46.6	44.9	47.9	45.1	45.0	61.3	60.7	60.4	60.9	61.3	62.8
Benzo[e]pyrene	66.1	70.3	72.6	75.8	71.5	76.6	71.3	70.7	86.7	86.4	88.5	87.7	89.6	88.7
Benzo[a]pyrene	109	120	106	109	105	110	105	104	128	132	134	131	132	131
Benzo[b]fluoranthene	21.7	23.3	28.1	29.0	27.6	29.4	27.6	27.6	32.7	33.1	33.0	32.9	32.9	33.0
Benzo[ghi]perylene	59.2	74.0	70.6	72.7	69.6	73.7	69.3	68.5	95.1	96.4	95.2	95.7	96.0	94.7
Indeno[123-cd]pyrene	26.5	33.5	49.2	50.8	48.5	51.5	48.4	47.8	58.4	59.6	60.8	58.7	59.0	59.3
METHOD	GC/MS		GC/FID						GC/MS					

[Annex 2 : continued]

Concentration of PAH in unknown solution G2 (ng μl^{-1}).

	8						9						9					
Phenanthrene	50.9	50.0	49.8	49.5	50.8	50.0	42.6	35.2	42.8	42.8	39.1	36.7	47.8	46.3	44.4	49.2	46.5	43.1
Fluoranthene	55.3	54.5	54.3	54.6	55.7	55.2	41.4	41.0	35.8	40.6	42.3	49.0	56.3	50.7	50.8	54.5	54.5	51.1
Pyrene	73.5	73.0	73.0	72.8	74.6	73.3	59.9	57.9	77.9	81.9	62.7	76.8	71.4	69.2	69.5	74.9	73.1	69.4
Benz[a]anthracene	50.2	50.1	50.5	50.7	49.5	49.9	44.4	44.4	51.6	46.5	48.2	52.6	44.6	44.4	45.7	48.0	48.4	44.0
Chrysene	54.6	54.9	55.2	55.0	54.3	54.3	45.2	53.6	47.9	41.5	48.5	53.7	49.0	48.5	49.8	52.0	52.8	47.8
Benzo[e]pyrene	79.6	78.4	79.7	79.6	82.1	79.1	65.8	71.6	88.9	76.2	83.8	85.3	75.2	76.5	79.5	82.4	85.4	78.0
Benzo[a]pyrene	122	121	123	124	127	122	88.0	93.7	106	98.0	102	106	116	120	123	129	133	122
Benzo[b]fluoranthene	30.1	29.8	30.3	30.3	31.0	30.1	25.1	27.2	34.0	28.6	32.2	31.9	29.2	29.8	30.2	31.4	32.0	29.3
Benzo[ghi]perylene	78.4	78.1	78.2	78.8	79.8	77.6	64.7	69.5	91.6	72.8	81.9	82.3	74.7	74.2	80.4	79.3	85.1	78.5
Indeno[123-cd]pyrene	55.0	55.0	55.2	55.9	56.0	54.7	46.9	51.2	68.5	54.3	62.5	60.5	52.7	51.9	54.7	56.2	59.0	53.3
METHOD	GC/MS						GC/MS						GC/FID					

[Annex 2 : continued]

Concentration of PAH in unknown solution G2 (ng μl^{-1}).

	11						11						12					
Phenanthrene	45.2	44.4	44.3	46.1	45.3	45.7	44.1	46.5	45.0	41.2	41.1	41.7	40.4	37.9	36.7	35.3	36.3	37.3
Fluoranthene	46.9	47.5	47.2	47.5	48.4	48.6	51.6	51.3	50.3	51.6	50.5	51.2	48.6	47.6	45.0	44.8	46.7	47.4
Pyrene	65.2	65.6	65.3	64.8	64.9	64.6	69.3	67.8	67.2	69.4	67.6	68.3	64.6	62.8	61.4	59.1	61.2	63.7
Benz[a]anthracene	38.9	41.9	40.8	39.9	41.0	38.9	38.5	41.6	42.2	38.4	38.1	38.2	39.8	38.2	37.4	39.3	40.1	38.5
Chrysene	44.6	47.5	46.6	46.1	47.2	44.4	43.7	46.5	47.2	42.7	42.6	42.8	44.7	43.2	41.9	41.8	39.4	40.8
Benzo[e]pyrene	74.1	77.9	73.8	79.1	73.7	67.3	60.9	68.8	70.5	60.2	61.0	62.2	75.5	70.9	64.8	64.4	70.1	66.1
Benzo[a]pyrene	106	113	108	110	110	101	89.1	102	104	88.4	89.8	91.9	119	114	106	103	110	115
Benzo[b]fluoranthene	28.3	28.8	28.5	29.9	27.8	26.8	26.0	27.7	28.6	24.6	24.8	25.2	24.8	22.6	21.6	20.2	21.4	21.6
Benzo[ghi]perylene	73.9	78.1	74.9	79.6	73.1	70.5	58.1	68.5	69.2	58.3	59.3	60.7	61.7	57.4	61.6	60.0	51.9	56.6
Indeno[123-cd]pyrene	51.9	51.3	50.5	55.9	51.8	47.5	41.8	48.1	48.7	41.3	42.2	43.1	44.5	39.6	44.2	37.6	38.8	34.7
METHOD	GC/MS						GC/FID						GC/MS					

[Annex 2 : continued]

Concentration of PAH in unknown solution G2 (ng μl^{-1}).

	12						14*						14+					
Phenanthrene	45.1	45.0	45.5	45.8	45.1	44.6	55.9	56.4	55.9	55.9	56.4	55.9	40.7	51.3	47.5	38.4	34.1	45.1
Fluoranthene	48.7	50.2	48.6	50.1	49.9	47.9	54.0	57.0	54.3	54.4	54.0	55.2	50.3	57.4	54.5	49.9	44.4	45.7
Pyrene	66.0	66.9	65.6	65.9	67.4	64.7	70.0	73.9	71.4	70.8	70.4	71.1	63.9	73.9	79.5	69.0	63.8	71.1
Benz[a]anthracene	42.8	43.2	43.4	42.4	42.8	41.1	46.1	48.1	46.8	46.5	47.3	47.4	51.0	66.4	57.2	50.7	45.0	46.4
Chrysene	46.6	47.2	47.3	46.3	46.8	44.7	50.4	51.8	50.4	49.5	50.6	51.3	49.3	54.8	54.3	52.4	48.4	49.3
Benzo[e]pyrene	73.7	74.2	74.6	73.2	73.4		80.7	82.4	80.4	79.1	81.2	81.3	81.9	90.9	74.5	69.7	84.9	82.4
Benzo[a]pyrene	111	112	112	112	110		123	125	121	120	124	123	122	132	128	118	98.9	116
Benzo[b]fluoranthene	26.8	29.2	29.4	27.7	29.0	26.3	30.4	31.2	30.1	30.0	30.4	30.4	26.2	28.5	29.3	22.8	24.2	29.8
Benzo[ghi]perylene	70.0	70.3	70.3	70.6	68.4		82.6	84.0	82.0	80.9	82.7	82.9	102	110	84.5	90.9	77.1	81.7
Indeno[123-cd]pyrene	50.1	50.4	50.4	49.0	47.8		51.7	53.0	51.7	50.7	52.5	51.4	63.2	69.7	67.2	53.5	50.1	53.0
METHOD	GC/FID						GC/FID						GC/FID					

* By peak area. + By peak height.

[Annex 2 : continued]

Concentration of PAH in unknown solution G2 (ng μl^{-1}).

	15						16											
Phenanthrene	44.0	41.7	44.0	45.2	45.4	42.5	31.0	34.4	28.6	21.7	25.1	16.5	28.2	20.6	19.9	15.7	22.3	24.7
Fluoranthene	47.7	44.7	47.6	49.2	48.8	45.7	14.2	28.6	26.2	35.1	28.7	36.6	22.7	35.9	33.3	34.6	37.9	29.4
Pyrene	64.4	60.1	63.9	66.1	65.6	61.4	39.5	54.9	50.1	44.2	45.2	34.6	53.3	38.1	39.6	31.8	42.0	47.8
Benz[a]anthracene	40.5	38.0	40.5	42.4	41.5	38.8	14.2	21.0	19.5	18.2	17.1	19.6	18.0	15.6	15.4	15.7	15.7	18.5
Chrysene	46.2	43.4	45.7	48.2	47.5	44.3	17.2	24.2	22.9	25.6	21.2	24.3	23.0	21.0	19.6	21.0	20.0	22.8
Benzo[e]pyrene	68.2	64.0	66.8	70.9	70.4	65.2	55.1	31.4	25.8	28.1	36.7	37.1	35.8					
Benzo[a]pyrene	100	94.2	98.6	105	104	96.0	98.8	69.0	59.2	60.6	73.6	79.7	80.7					
Benzo[b]fluoranthene	28.1	26.4	27.6	29.3	29.0	27.0	16.5	12.0	10.6	10.4	12.9	13.3	12.6					
Benzo[ghi]perylene	66.6	62.8	66.2	70.1	70.4	64.6	54.4	36.9	28.5	30.4	37.6	38.4	40.7					
Indeno[123-cd]pyrene	48.1	45.4	47.7	50.6	50.6	46.5	33.4	22.6	15.9	18.1	24.7	24.0	25.2					
METHOD	GC/MS						GC/MS											

[Annex 2 : continued]

Concentration of PAH in unknown solution G2 (ng μl^{-1}).

	17						NOMINAL CONCENTRATIONS OF STANDARDS	
							G2*	H2+
Phenanthrene							45	142
Fluoranthene							51	92
Pyrene	63.5	64.2	62.3	63.3	63.0	65.2	68	59
Benz[a]anthracene	44.8	44.1	44.7	45.6	42.4	43.7	47	16
Chrysene	108	106	98.9	95.5	96.1	104	45	31
Benzo[e]pyrene	57.0	50.9	53.0	55.6	59.0	55.0	78	71
Benzo[a]pyrene	121	114	119	119	122	124	115	176
Benzo[b]fluoranthene	47.2	42.1	44.7	39.0	44.7	38.7	26	55
Benzo[ghi]perylene	111	116	116	125	122	116	75	64
Indeno[123-cd]pyrene	52.5	56.5	54.4	48.4	49.2	52.4	53	34
METHOD	UVF							

* Used for: GC/FID gas chromatography/flame ionisation detector
GC/MS gas chromatography/mass spectrometry
UVF low temperature fluoresence

+ Used for: HPLC/UVF high performance liquid chromatography/fluorescence detector
HPLC/UV HPLC/UV absorption detector