Results of Proficiency Test
PFOA
September 2011

Organised by: Institute for Interlaboratory Studies
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CONTENTS

1 INTRODUCTION

Worldwide, many consumer products are produced that contain Teflon parts. In the production of Teflon perfluorooctanoic acid (PFOA) may be used.
PFOA persists indefinitely in the environment. It is a toxicant and carcinogen in animals.
In order to protect health and environment, the European Union promulgated on 27 December 2006 Directive 2006/122/EC, in which the placing on the market and the use of perfluorooctan sulfonates (C8F17SO2X, where X may be OH, being PFOA) is restricted: “Semi-finished products or articles, or parts thereof, if the concentration of PFOS is equal or greater than 0.1% by weight”.
Subsequent EC regulations on PFOS/PFOA are 552/2009 en 757/2010. The production and use is currently prohibited. Some exemptions are made.
On request of several participants, the Institute for Interlaboratory Studies decided to organise an interlaboratory study for the determination of PFOA in the 2011/2012 PT program.
In the interlaboratory study of September 2011, 36 laboratories from 14 different countries participated (See appendix 3). In this report, the results of the proficiency test are presented and discussed.

2 SET-UP

The Institute for Interlaboratory Studies (iis) in Spijkenisse, The Netherlands, was the organiser of this proficiency test. It was decided to send 2 different plastic samples (approximately 5 gram each), both positive (artificially fortified) on PFOA and labelled #11054 and #11055 respectively. Participants were also requested to report some details of the test method used.

2.1 QUALITY SYSTEM

The Institute for Interlaboratory Studies in Spijkenisse, the Netherlands, has implemented a quality system based on ISO guide 43, ILAC-G13:2007 and ISO/IEC 17043:2010. This ensures 100% confidentially of participant’s data. Also, customer’s satisfaction is measured on a regular basis by sending out questionnaires.

2.2 PROTOCOL

The protocol followed in the organisation was the one as described for proficiency testing in the report ‘iis Interlaboratory Studies: Protocol for the Organisation, Statistics and Evaluation’ of January 2010 (iis-protocol, version 3.2). This protocol can be downloaded from the iis website http://www.iisnl.com.

2.3 CONFIDENTIALITY STATEMENT

All data present in this report must be regarded as confidential and for use by the participating companies only. Disclosure of the information in this report is only allowed by means of the entire report. Use of the contents of this report for third parties is only allowed by written permission of the Institute for Interlaboratory Studies. Disclosure of the identity of one or more of the participating companies will be done only after receipt of a written agreement of the companies involved.

2.4 SAMPLES

Two different samples, both artificially fortified to be positive on PFOA (0.20 and 0.14% resp.), were selected. The first material (#11054) was a orange coloured plastic. The second material (#11055) was a purple coloured plastic. Both materials were divided over plastic bags, approx. 5 grams for each sample.
The homogeneity of the subsamples was checked by determination of PFOA content on 3 stratified randomly selected subsamples.

	PFOA in #11054	PFOA in #11055
Sample 1	1422 mg/kg	945 mg/kg
Sample 2	1396 mg/kg	987 mg/kg
Sample 3	1440 mg/kg	985 mg/kg

Table 1: results of the homogeneity test on the subsamples #11054 and #11055

From the above results of the homogeneity test, the relative between sample standard deviations RSD_r were calculated and compared with 0.3 times the relative proficiency target standard deviations RSD_R in agreement with the procedure of ISO 13528, Annex B2 in the next table:

	PFOA in #11054	PFOA in #11055
RSD_r (observed)	1.6%	2.4%
reference method	Horwitz	Horwitz
0.3 x RSD_R (reference method)	1.6%	1.7%

Table 2: relative repeatability standard deviations of PFOA contents of the subsamples #11054 and #11055

The calculated variation coefficient RSDr for #11054 is in good agreement and for #11055 almost in agreement with the estimated target, calculated using the Horwitz equation. Therefore, homogeneity of all subsamples was assumed.

To each of the participating laboratories one set of samples, (1* sample #11054 and 1* sample #11055) was sent on August 17, 2011.

2.5 ANALYSIS

The participants were requested to determine PFOA on both samples. It was explicitly requested to treat the samples as if it were routine samples and to report the analytical results using the indicated units on the report form and not to round the results, but report as much significant figures as possible. It was also requested not to report ‘less than’ results, which are above the detection limit, because such results can not be used for meaningful statistical calculations.
To get comparable results a detailed report form, on which the units were prescribed, was sent together with each set of samples. Also, a letter of instructions was added to the package.
The laboratories were requested to complete the report form with some details of the methods used.

3 RESULTS

During four weeks after sample despatch, the results of the individual laboratories were received. The original data are tabulated per sample in the appendix 1 of this report.
The laboratories are represented by the code numbers.

Directly after the deadline, a reminder fax was sent to those laboratories that did not report results at that moment.
Shortly after the deadline, the available results were screened for suspect data. A result was called suspect in case the Huber Elimination Rule (a robust outlier test) found it to be an outlier. The laboratories that produced these suspect data were asked to check the results. Additional or corrected results are used for the data analysis and the original results are placed under 'Remarks' in the result tables in appendix 1.

3.1 STATISTICS

The statistical calculations were performed as described in the procedures in the report ‘iis Interlaboratory Studies, Protocol for the Organisation, Statistics and Evaluation’ of January 2010 (iis-protocol, version 3.2).

First, the normality of the distribution of the various data sets per determination was checked by means of the Lilliefors-test. After removal of outliers this check was repeated.

In accordance to ISO 5725 (1986 and 1994) the original results per determination were submitted subsequently to Dixon and Grubbs outlier tests. Outliers are marked by D(0.01) for the Dixon test, by G(0.01) or DG(0.01) for the Grubbs test. Stragglers are marked by D(0.05) for the Dixon test, by G(0.05) or DG(0.05) for the Grubbs test. Both outliers and stragglers were not included in the calculations of averages and standard deviations.

Finally, the reproducibilities were calculated from the standard deviations by multiplying them with a factor of 2.8.

For each assigned value the uncertainty was determined in accordance with ISO13528. Subsequently the calculated uncertainty was evaluated against the respective requirement based on the target reproducibility in accordance with ISO13528. When the uncertainty passed the evaluation no remarks are made in the report. However, when the uncertainty failed the evaluation it is mentioned in the report and it will have significant consequences for the evaluation of the test results.

3.2 GRAPHICS

In order to visualise the data against the reproducibilities from literature, Gauss plots were made, using the sorted data for one determination (see appendix 1). On the Y-axis the reported analysis results are plotted. The corresponding laboratory numbers are under the X-axis.
The straight horizontal line presents the consensus value (a trimmed mean). The four striped lines, parallel to the consensus value line, are the +3s, +2s, -2s and -3s target reproducibility limits

of the selected standard. Outliers and other data, which were excluded from the calculations, are represented as a cross. Accepted data are represented as a triangle. Furthermore, Kernel Density Graphs were made. This is a method for producing a smooth density approximation to a set of data that avoids some problems associated with histograms (see appendix 4; nr.14 and 15).

3.3 z-scores

To evaluate the performance of the participating laboratories the z-scores were calculated. As it was decided to evaluate the performance of the participants in this proficiency test (PT) against the literature requirements, the z-scores were calculated using a target standard deviation. This results in an evaluation independent of the spread of this interlaboratory study.

The target standard deviation was calculated from the target reproducibility (preferably taken from a standardized test method) by division with 2.8.
The z-scores were calculated in accordance with:

$$z_{(target)} = (\text{result} - \text{average of PT}) / \text{target standard deviation}$$

The $z_{(target)}$ scores are listed in the result tables in appendix 1.

When a laboratory did use a test method with a reproducibility that is significantly different from the reproducibility of the reference test method used in this report, it is strongly advised to recalculate the z-score, while using the reproducibility of the actual test method used. This in order to evaluate the fit-for-useness of the reported test result. See also appendix 3; no. 16.

Absolute values for z<2 are very common and absolute values for z>3 are very rare. Therefore the usual interpretation of z-scores is as follows:

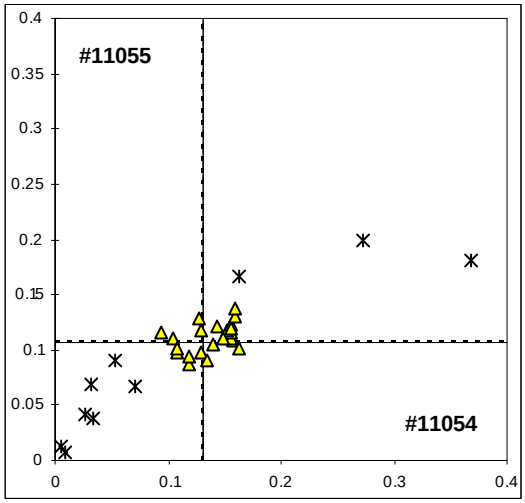
$\lvert z \rvert < 1$	good
$1 < \lvert z \rvert < 2$	satisfactory
$2 < \lvert z \rvert < 3$	questionable
$3 < \lvert z \rvert$	unsatisfactory

4 Evaluation

In this interlaboratory study, no problems were encountered with the dispatch of the samples.
Three participants reported test results after the final reporting date and three other participants did not report any test results at all.
Finally, 33 of the 36 participants submitted analysis results. The 33 reporting laboratories reported 66 numerical results. Observed were 13 outlying results, which is 16.5%. In proficiency studies, outlier percentages of 3% - 7.5% are quite normal.

A not-normal distribution was found for the reported PFOA test results of sample #11054.
Therefore this statistical evaluation should be used with due care.

From the reported test results it is clear that a significant number of laboratories have difficulties with this determination as they reported very low test results. During the preparation of the samples it was found that indeed some extra efforts may be needed to increase the recovery of PFOA from the matrix upto an acceptable level.
Therefore it is no surprise to find that the test results for both samples are strongly correlated as can be seen in the below Youden plot:



Youden plot of samples #11054 and #11055

A laboratory that reported a low test result for sample #11054, did also report a low test result for sample #11055.
In the dataset for sample #11055 the low test results were removed by the outlier detection method. However, in sample #11054 there are more low test results at close distance from each other and the Dixon's and Grubb's outlier test were unable to remove all these results. Therefore two test results were excluded manually based on the statistical evaluation on sample #11055. See also paragraph 5.

For the determination of PFOA, the CEN/TS 15968 method is considered to be the official EC test method. Regretfully this method does not yet mention reproducibility requirements. Therefore, the target requirements in this study were estimated using the Horwitz equation.

4.1 PERFORMANCE EVALUATION OF THE GROUP OF LABORATORIES

The calculated reproducibilities and the target reproducibilities derived from the literature standards, here Horwitz, are compared in the next table.

	unit	n	Average	2.8 * sd	R (lit)
PFOA in #11054	%M/M	24	0.139	0.059	0.021
PFOA in #11055	%M/M	27	0.109	0.057	0.017

Table 3: performance overview for samples #11054 and #11055

Without further statistical calculations, it can be concluded that there is not a good compliance of the group of participating laboratories with the target reproducibility.

4.2 EVALUATION

In this section the results are discussed per sample.

#11054: Severe analytical problems were observed at the relatively high concentration level of 0.14 - 0.15 %M/M in the evaluated material. Seven statistical outliers were detected and two test results were removed manually (see paragraph 4). The calculated reproducibility, after rejection of the 9 test results, is not in agreement with the target requirement estimated from the Horwitz equation.

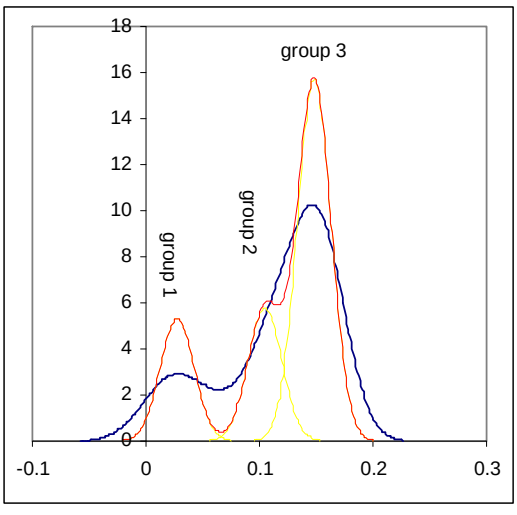
#11055: Analytical problems were observed at the concentration level of 0.11 %M/M in the evaluated material. Six statistical outliers were detected. The calculated reproducibility, after rejection of the statistical outliers, is not in agreement with the target requirement estimated from the Horwitz equation.

4.3 EVALUATION OF THE TEST METHODS USED

Most participants reported to have used an 'in house' test method. It is remarkable that only two participants reported to have used the CEN/TS 15968 method.
The reported details of the methods that were used by the participants are listed in appendix 2.

5 CONCLUSIONS

The final assigned value of 0.139 %M/M for sample #11054, even after removal of 9 test results, may still be rather low. The theoretical value, derived from the PFOA amount used during the preparation is 0.20%, which means that only 70% may have been recovered on average.
Also the total data set can be divided in 4 groups of test results (see also) below graph:

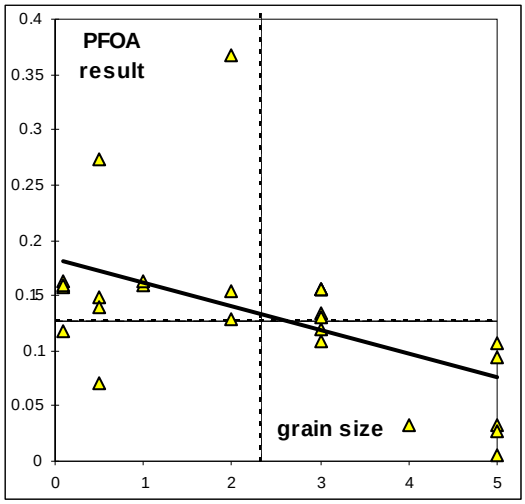


Division of results for #11054 into 3 groups

group 1: 6 test results with an average of 0.028 %M/M (= 14% recovery)
group 2: 7 test results with an average of 0.105 %M/M (= 53% recovery)
group 3: 18 test results with an average of 0.148 %M/M (= 74% recovery)
group 4: 2 very high test results (0.2733 and 0.3680 %M/M), not present in the above graph

From the reported test details it is clear that at least 4 laboratories (the other 2 did not report any details) from group 1 did not reduce the grain size of the samples.

When all reported grain sizes are plotted against the reported PFOA content, a significant correlation is observed, see below plot:



Correlation between grain size and reported PFOA result on sample #11054

The further the grain size was reduced, the higher the PFOA concentration reported. Obviously the extraction solvent is not able to reach the PFOA that is too deep inside the plastic matrix. Too few details were reported to see whether prolonged extraction does solve this problem. The two relative high test results that disturb the correlation and that also were statistical outliers, may be explained by possible contamination in the laboratory from used materials, e.g. ptfe tubing in analytical instruments, laboratory gloves, treated laboratory textiles, etc. See reference 17.

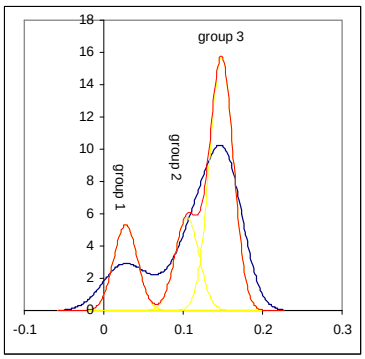
A number of participants used ultrasonification instead of Soxhlet extraction to release the components from the plastic matrix. These results were not significantly different from the other results, although several very low results are reported by these participants.

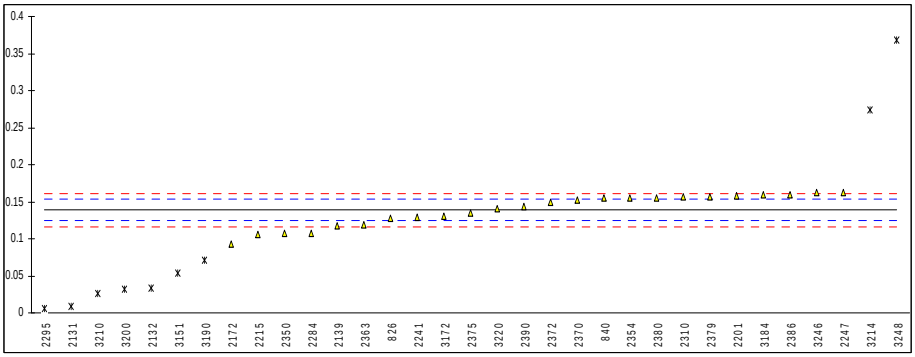
Each laboratory has to evaluate its performance in this study and make decisions about necessary corrective actions. Therefore, participation on a regular basis in this scheme could be helpful to improve the performance and the quality of the analytical results.

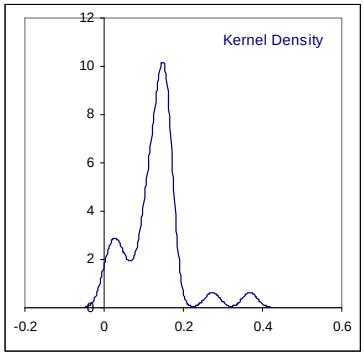
APPENDIX 1

Determination of PFOA on sample #11054; results in %M/M

lab	method	value	mark	z(targ)	remarks
826	EPA3540C/3550C	0.1278		-1.49	
840	in house	0.1546	C	2.09	first reported 0.2246
2115		-----		-----	
2129		-----		-----	
2131	in house	0.00827	ex	-17.47	see §4.1
2132	in house	0.0333	DG(0.05)	-14.12	
2139	CEN/TS15968	0.118		-2.80	
2172	in house	0.0933		-6.10	
2201	in house	0.1575		2.48	
2215	in house	0.1051		-4.53	
2241	in house	0.1285		-1.40	
2247	in house	0.1630	C	3.21	first reported 0.0397
2284	in house	0.1078		-4.16	
2295	in house	0.0054	ex	-17.85	see §4.1
2310	in house	0.1560		2.28	
2350	in house	0.10738	C	-4.22	first reported 0.21475
2354	in house	0.1554		2.20	
2363	EPA3550C/8321B	0.119		-2.67	
2370	INH-219-3	0.152		1.74	
2372	EPA3540C	0.1486		1.29	
2375	INH-219-3	0.13436		-0.61	
2379	in house	0.1561		2.29	
2380	in house	0.1556		2.22	
2386	DIN38414	0.160		2.81	
2390	in house	0.1432		0.57	
3151	in house	0.053064	DG(0.05)	-11.48	
3154		-----		-----	
3172	in house	0.13		-1.20	
3184	in house	0.1597		2.77	
3190	in house	0.071	DG(0.05)	-9.08	
3200	EPA3550C/8321B	0.0326	DG(0.05)	-14.22	
3210	CEN/TS15968	0.0265	G(0.05)	-15.03	
3214	in house	0.2733	D(0.05)	17.96	
3220	in house	0.14	C	0.14	first reported 0.02179
3246	in house	0.162		3.08	
3248	in house	0.3680	C,G(0.01)	30.62	first reported 0.0110
	normality	not OK			
	n	24			
	outliers	7			
	mean (n)	0.139	added 0.20%; recovery approx. 70%		
	st.dev. (n)	0.0212			
	R(calc.)	0.059			
	R(Horwitz)	0.021			

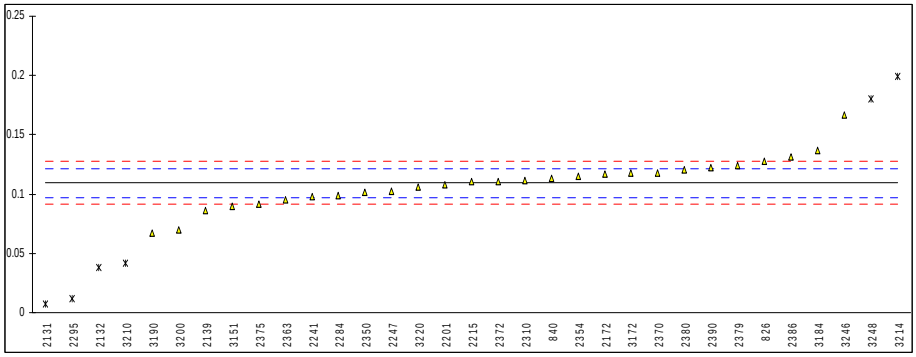


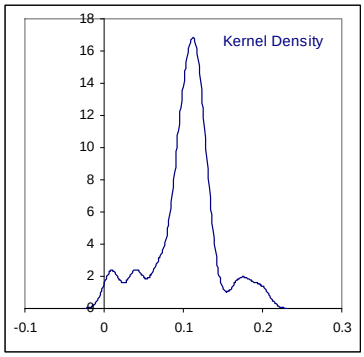




Determination of PFOA on sample #11055; results in %M/M

lab	method	value	mark	z(targ)	remarks
826	EPA3540C/3550C	0.1281		3.06	
840	in house	0.1131	C	0.60	first reported 0.1679
2115		-----		-----	
2129		-----		-----	
2131	in house	0.00715	DG(0.05)	-16.75	
2132	in house	0.0378	C,DG(0.05)	-11.73	first reported 0.0609
2139	CEN/TS15968	0.086		-3.83	
2172	in house	0.1167		1.19	
2201	in house	0.1080		-0.23	
2215	in house	0.1101		0.11	
2241	in house	0.0980		-1.87	
2247	in house	0.1020	C	-1.21	first reported 0.0364
2284	in house	0.0984		-1.80	
2295	in house	0.0118	DG(0.05)	-15.99	
2310	in house	0.1111		0.28	
2350	in house	0.10185	C	-1.24	first reported 0.2037
2354	in house	0.1150		0.92	
2363	EPA3550C/8321B	0.0947		-2.41	
2370	INH-219-3	0.118		1.41	
2372	EPA3540C	0.1106		0.20	
2375	INH-219-3	0.09136		-2.96	
2379	in house	0.1238		2.36	
2380	in house	0.1201		1.75	
2386	DIN38414	0.131		3.54	
2390	in house	0.1219		2.05	
3151	in house	0.089913		-3.19	
3154		-----		-----	
3172	in house	0.118		1.41	
3184	in house	0.1371		4.54	
3190	in house	0.067		-6.95	
3200	EPA3550C/8321B	0.0694		-6.55	
3210	CEN/TS15968	0.0421	DG(0.05)	-11.02	
3214	in house	0.1995	DG(0.05)	14.75	
3220	in house	0.1058	C	-0.59	first reported 0.035958
3246	in house	0.167		9.43	
3248	in house	0.1805	C,DG(0.05)	11.64	first reported 0.0162
	normality	OK			
	n	27			
	outliers	6			
	mean (n)	0.109	added 0.14%; recovery approx. 80%		
	st.dev. (n)	0.0205			
	R(calc.)	0.057			
	R(Horwitz)	0.017			





APPENDIX 2

Analytical details for samples #11054 and #11055

Lab	particle size used	Extraction method	Solvent(s) used	Detection	Corr	Other
826			dichloromethane/methanol	LC/MS/MS	N	
840	1mm X 2mm	Soxhlet extraction	dichloromethane/methanol	LC/MS	N	
2115						
2129		ultrasonic, 30 min, RT	dichloromethane	LC/MS/MS		
2131		ultrasonic	methanol & NH4-acetate	LC/MS/MS	Y	
2132	4mm x 4mm	ultrasonic	methanol	LC/MS/MS	N	
2139	powder	ultrasonic	methanol	LC/MS	N	
2172	5mm x 5mm	ultrasonic, 60°C, 2 hrs	methanol	LC/MS	N	ESTD
2201	grind to powder	ultrasonic	methanol	LC/MS/MS	N	
2215	1g	ultrasonic, 60°C	methanol	LC/MS	N	
2241	<2mm x 2mm	ultrasonic	methanol	UPLC/MS	N	ESTD
2247	cryomill powder	solvent extraction	methanol	LC/MS/MS	N	
2284	3mm x 3mm	ultrasonic	methanol	LC/MS	N	
2295	5mm	ultrasonic, RT	methanol	LC/MS/MS	Y	
2310	3mm X 3mm	solvent extraction	dichloromethane/methanol	LC/MS/MS	N	
2350	5mm x 5mm	Soxhlet extraction	dichloromethane/methanol	LC/MS	N	
2354	3 X 3mm	Soxhlet extraction	dichloromethane/methanol	LC/MS/MS	N	
2363	<3mm x 3mm	ultrasonic, 2 hrs, 70°C	methanol	ESTD	N	see sheet
2370	0.5g		dichloromethane/methanol	LC/MS	Y	
2372	<0.5mm	Soxhlet extraction	dichloromethane/methanol	LCMS	N	
2375	3mm x 3mm	solvent extraction	dichloromethane/methanol	LC	N	
2379	3mm x 3mm x 3mm	Soxhlet extraction	dichloromethane/methanol	LC/MS	N	
2380	0.50 – 0.52g	Soxhlet extraction	dichloromethane/methanol	LC/MS	N	
2386	<1mm	ultrasonic, 40°C, 120 min	methanol	LC/MS/MS	Y	rec 91-93%
2390	0.5g	Soxhlet extraction	dichloromethane/methanol	LC/MS	N	
3151		ultrasonic, 60°C, 120 min	methanol	LC/MS/Q-TOF		
3154				LC/MS/MS		
3172	3mm	ultrasonic	THF/ACN	LC/MS/QQQ	N	
3184	powder	ultrasonic, 70°C, 30 min	methanol	LC/MS/MS	N	
3190	grinded	ultrasonic	methanol	LC/MS/MS	N	
3200	5mm x 5mm	ultrasonic	methanol	UPLC/MS/MS	N	
3210	origin size		methanol	LC/MS/MS/QQQ	N	
3214	<500um	ultrasonic, 70°C, 2 hrs	methanol	LC/MS/MS	N	
3220	0.5 mm	solvent extraction	methanol	LC/MS	N	
3246	1mm	extract 1hr, 70°C	methanol	LC/MS	N	
3248	2mm x 2mm	solvent extraction	methanol	LC/MS	N	

APPENDIX 3

Number of participating laboratories per country

1 lab in	BANGLADESH
1 lab in	FRANCE
4 labs in	GERMANY
3 labs in	HONG KONG
3 labs in	INDIA
2 labs in	ITALY
3 labs in	KOREA
9 labs in	P.R. of CHINA
1 lab in	PAKISTAN
1 lab in	SWITZERLAND
3 labs in	TAIWAN R.O.C.
1 lab in	THAILAND
2 labs in	TURKEY
2 labs in	VIETNAM

APPENDIX 4

Abbreviations:

C = final result after checking of first reported suspect result
D(0.01) = outlier in Dixon's outlier test
D(0.05) = straggler in Dixon's outlier test
G(0.01) = outlier in Grubbs' outlier test
G(0.05) = straggler in Grubbs' outlier test
DG(0.01) = outlier in Double Grubbs' outlier test
DG(0.05) = straggler in Double Grubbs' outlier test
n.a. = not applicable

Literature:

1 iis Interlaboratory Studies, Protocol for the Organisation, Statistics & Evaluation, January 2010
2 EN 1122:2001, "Determination of Cadmium in plastics with the method of the wet decomposition".
3 ASTM D4004:98, "Determination of Metal Content by Flame Atomic Absorption (AAS) analysis"
4 ASTM E178-02
5 ASTM E1301-03
6 ISO 5725-86
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14 Analytical Methods Committee Technical Brief, No4 January 2001
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