Results of Proficiency Test Preservatives in Toothpaste (Triclosan) November 2020

Organized by: Institute for Interlaboratory Studies
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1 INTRODUCTION

Triclosan is a disinfectant. In addition, it is also used as a preservative in personal care products. The concentration of triclosan when used as a preservative is lower than when used as a disinfectant. Triclosan can occur both as a disinfectant or as a preservative in textiles, building materials, kitchen utensils, personal care products such as hand soap, bathroom accessories, cleaning products, pesticides, plastics, medical implants and office supplies.
In 2020 on request of a number of laboratories, the Institute for Interlaboratory Studies has started a new proficiency test of the determination of Triclosan in Toothpaste.
In this interlaboratory study 7 laboratories in 5 different countries registered for participation. See appendix 3 for the number of participants per country. In this report the results of this proficiency test are presented and discussed. This report is also electronically available through the iis website www.iisnl.com.

2 SET UP

The Institute for Interlaboratory Studies (iis) in Spijkenisse, the Netherlands, was the organizer of this proficiency test (PT). Sample analyses for fit-for-use and homogeneity testing were subcontracted to an ISO/IEC17025 accredited laboratory. It was decided to send one sample Toothpaste of approximately 10 grams and labelled #20725 which was artificially fortified on Triclosan. The participants were requested to report rounded and unrounded test results. The unrounded test results were preferably used for statistical evaluation.

2.1 QUALITY SYSTEM

The Institute for Interlaboratory Studies (iis) in Spijkenisse, the Netherlands, has implemented a quality system based on ISO/IEC17043:2010. This ensures strict adherence to protocols for sample preparation and statistical evaluation and 100% confidentiality of participant's data. Feedback from the participants on the reported data is encouraged and customer's satisfaction is measured on regular basis by sending out questionnaires.

2.2 PROTOCOL

The protocol followed in the organization of this proficiency test was the one as described for proficiency testing in the report 'iis Interlaboratory Studies: Protocol for the Organisation, Statistics and Evaluation' of June 2018 (iis-protocol, version 3.5). This protocol is electronically available through the iis website www.iisnl.com, from the FAQ page.

2.3 CONFIDENTIALITY STATEMENT

All data presented 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

A regular toothpaste was purchased from a local supermarket and was artificially fortified with Triclosan. After homogenization 22 PE bottles of 10mL were filled with approximately 10 grams of Toothpaste and labelled #20725.
The homogeneity of the subsamples was checked by determination of Triclosan on five stratified randomly selected subsamples using an in-house test method.

	Triclosan in mg/kg
sample #20725-1	1483
sample #20725-2	1398
sample #20725-3	1400
sample #20725-4	1478
sample #20725-5	1402

Table 1: homogeneity test results of subsamples #20725

From the above test results the relative standard deviation (RSD) was calculated and compared with 0.3 times the relative standard deviation of this proficiency test.

	Triclosan
RSD (observed)	3.1%
Reference method	iis20H06
0.3*RSD iis20H06	3.3%

Table 2: evaluation of the relative standard deviation of subsamples #20725

This is the first proficiency test for Triclosan in Toothpaste organized by iis and it is expected that most laboratories use an in-house method. Therefore, the homogeneity could not be determined by comparing the variation to a known reproducibility. However, the relative standard deviation (RSD) of the homogeneity tests was determined to be sufficiently small (RSD<5%) to be sure that the batch is homogeneous.

After the evaluation of the PT test results the RSD of the PT was checked to see if the RSD obtained from the homogeneity test results is indeed lower than the variation between the test results as reported by the participants. The relative standard deviation of subsamples is smaller than 0.3 times the relative standard deviation of this proficiency test. This underpins the decision that homogeneity of the subsamples was good.

To each of the participants 1 sample labelled #20725 was sent on November 4, 2020.

2.5 ANALYZES

The participants were requested to determine the concentration of Triclosan applying the analytical procedure that is routinely used in the laboratory. It was also requested to report if the laboratory was accredited for this determination and to report the sample used for intake.

It was explicitly requested to treat the sample as if it was a routine sample and to report the test results using the indicated units on the report form and not to round the test results, but to report as much significant figures as possible. It was also requested not to report “less than” test results, which are above the detection limit, because such test results cannot be used for meaningful statistical evaluation.

To get comparable test results a detailed report form and a letter of instructions are prepared. On the report form the reporting units are given as well as the reference test methods (when applicable) that will be used during the evaluation. The detailed report form and the letter of instructions are both made available on the data entry portal www.kpmd.co.uk/sgs-iis-cts/. The participating laboratories are also requested to confirm the sample receipt on this data entry portal. The letter of instructions can also be downloaded from the iis website www.iisnl.com.

3 RESULTS

During five weeks after sample dispatch, the test results of the individual laboratories were gathered via the data entry portal www.kpmd.co.uk/sgs-iis-cts/. The reported test results are tabulated per determination in appendix 1 of this report. The laboratories are presented by their code numbers.

Directly after the deadline, a reminder was sent to those laboratories that had not reported test results at that moment. Shortly after the deadline, the available test results were screened for suspect data. A test 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 reported test results (no reanalyzes). Additional or corrected test results are used for data analysis and original test results are placed under 'Remarks' in the test result tables in appendix 1. Test results that came in after the deadline were not taken into account in this screening for suspect data and thus these participants were not requested for checks.

3.1 STATISTICS

The protocol followed in the organization of this proficiency test was the one as described for proficiency testing in the report ‘iis Interlaboratory Studies: Protocol for the Organisation, Statistics and Evaluation’ of June 2018 (iis-protocol, version 3.5).

For the statistical evaluation, the *unrounded* (when available) figures were used instead of the rounded test results. Test results reported as ‘<…’ or ‘>…’ were not used in the statistical evaluation.

First, the normality of the distribution of the various data sets per determination was checked by means of the Lilliefors-test a variant of the Kolmogorov-Smirnov test and by the calculation of skewness and kurtosis. Evaluation of the three normality indicators in combination with the visual evaluation of the graphic Kernel density plot, lead to judgement of the normality being either ‘unknown’, ‘OK’, ‘suspect’ or ‘not OK’. After removal of outliers, this check was repeated. If a dataset does not have a normal distribution, the (results of the) statistical evaluation should be used with due care.

According to ISO5725 the original test results per determination were submitted to Dixon's, Grubbs' or Rosner's outlier tests. Outliers are marked by D(0.01) for the Dixon's test, by G(0.01) or DG(0.01) for the Grubbs' test and by R(0.01) for the Rosner's test. Stragglers are marked by D(0.05) for the Dixon's test, by G(0.05) or DG(0.05) for the Grubbs' test and by R(0.05) for the Rosner's test. Both outliers and stragglers were not included in the calculations of averages and standard deviations.

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. In this PT the criterion of ISO13528 paragraph 9.2.1 was met for all evaluated tests, therefore the uncertainty of all assigned values may be negligible and need not be included in the PT report.

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

3.2 GRAPHICS

In order to visualize 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 test results are plotted. The corresponding laboratory numbers are on 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 reference test method. 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. The Kernel Density Graph is a method for producing a smooth density approximation to a set of data that avoids some problems associated with histograms. Also, a normal Gauss curve was projected over the Kernel Density Graph for reference.

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 variation of this interlaboratory study.
The target standard deviation was calculated from the literature reproducibility by division with 2.8. In case no literature reproducibility was available, other target values were used. In some cases, a reproducibility based on former iis proficiency tests could be used.

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 whether the reported test result is fit-for-use.

The z-scores were calculated according to:

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

The $z_{(target)}$ scores are listed in the test result tables in appendix 1.
Absolute values for z<2 are very common and absolute values for z>3 are very rare.
The usual interpretation of z-scores is as follows:

	|z|	< 1	good
1 <	|z|	< 2	satisfactory
2 <	|z|	< 3	questionable
3 <	|z|		unsatisfactory

4 EVALUATION

During the execution of this proficiency test no problems were encountered with the dispatch of the samples. One participant reported after the final reporting date. Finally, 7 laboratories reported 7 numerical test results. No outlying test results were observed. In proficiency studies outlier percentages of 3% - 7.5% are quite normal.

4.1 EVALUATION PER COMPONENT

In this section, the test results are discussed per component.
The test methods which were used by the various laboratories were taken into account for explaining the observed differences when possible and applicable. These test methods are also in the tables in appendix 1 together with the original data. The abbreviations used in these tables are explained in appendix 4.

Unfortunately, a suitable reference test method, providing the precision data, is not available for the determinations, therefore the calculated reproducibilities were compared against the reproducibility estimated from the Horwitz equation.

Triclosan (CAS no. 3380-34-5): The determination of this component at a concentration level of 1200 mg/kg may be problematic. No statistical outliers were observed. However, the calculated reproducibility is not in agreement with the calculated reproducibility estimated from the Horwitz equation.

4.2 PERFORMANCE EVALUATION FOR THE GROUP OF LABORATORIES

A comparison has been made between the estimated target reproducibility and the reproducibility as found for the group of participating laboratories. The number of significant test results, the average, the calculated reproducibility (2.8 * standard deviation) and the target reproducibility derived from the reference method (in casu Horwitz Equation) are compared in the next table.

Component	unit	n	average	2.8 * sd	R (target)
Triclosan	mg/kg	7	1214	376	187

Table 3: reproducibility of tests on sample #20725

From the table above it can be concluded that without statistical calculations the group of participating laboratories do have some difficulties with the analysis of Triclosan when compared with the target reproducibility. See also the discussion in paragraph 4.1.

4.3 OVERVIEW OF THE PROFICIENCY TEST OF NOVEMBER 2020

The evolution of the uncertainty expressed as relative standard deviation for Triclosan in Toothpaste as observed in this proficiency scheme is listed in table 4.

Year	Component	Observed RSD%	Target RSD%	Concentration mg/kg
2020	Triclosan	11%	5%	1214

Table 4: uncertainty in % for Triclosan

4.4 EVALUATION ANALYTICAL DETAILS

For this PT also some analytical details were requested and are listed in appendix 2.
Based on the answers given by the participants the following can be summarized:
- Four out of seven reporting participants mentioned that they are accredited for the determination of Triclosan in toothpaste.
- Three out of seven reporting participants used around 0.5 grams and two others reported to have use 1 grams.

Because the amount of analytical details and participating laboratories is small, no conclusions could be drawn from these analytical details.

5 DISCUSSION

All reporting participants were able to detect Triclosan in sample #20725.
The concentration of Triclosan in sample of #20725 was below the the limit of rejection of 0.3%M/M.

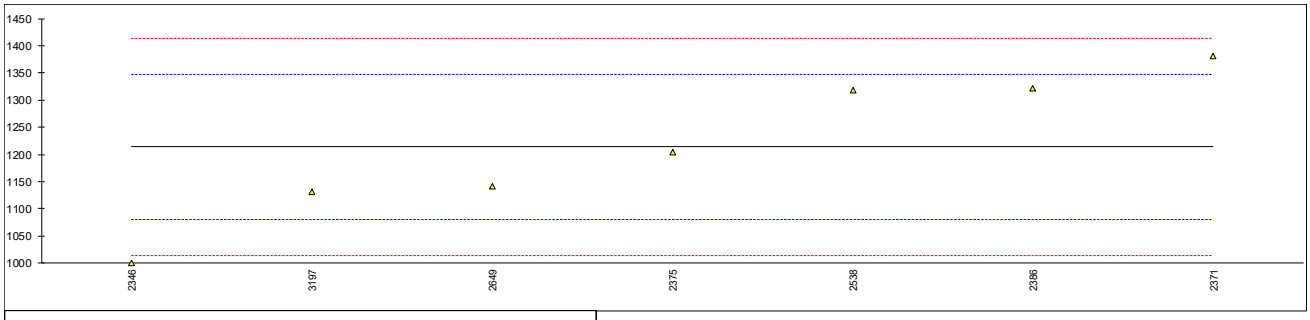
6 CONCLUSION

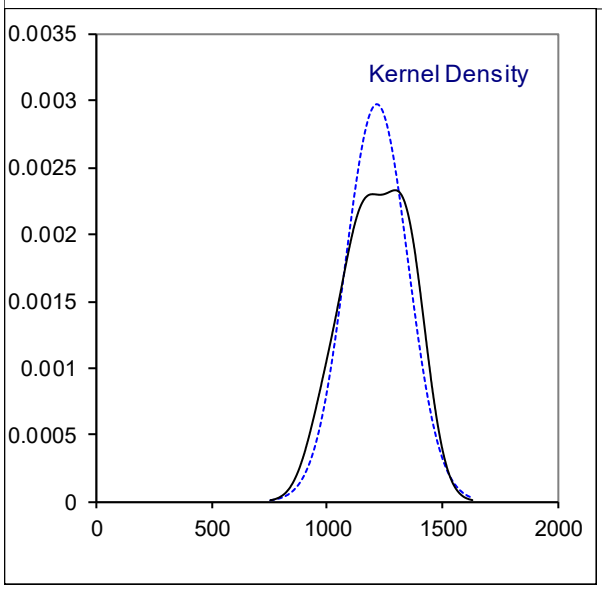
Each laboratory should evaluate its performance in this study and make decisions about necessary corrective actions. Therefore, participation on a regular basis in this scheme could be helpful to improve the performance and the quality of the analytical results.

APPENDIX 1

Determination of Triclosan (5-Chloro-2-(2,4-Dichlorophenoxy)phenol) CAS No. 3380-34-5 in sample #20725; results in mg/kg

lab	method	value	mark	z(targ)	remarks
2346	In house	1000		-3.21	
2371	In house	1380.988		2.50	
2375	In house	1205	C	-0.14	First reported 1650
2386	In house	1321.9		1.62	
2538	In house	1318		1.56	
2649	In house	1142	C	-1.08	First reported 1781.54
3197	In house	1131	C	-1.25	First reported 1900
	normality	unknown			
	n	7			
	outliers	0			
	mean (n)	1214.13			
	st.dev. (n)	134.346	RSD = 11%		
	R(calc.)	376.17			
	st.dev.(Horwitz)	66.705			
	R(Horwitz)	186.77			





APPENDIX 2

Analytical details

lab	ISO/IEC17025 accredited	sample intake (g)
2346	---	-
2371	Yes	1 g
2375	No	-
2386	No	1 g
2538	Yes	ca. 0,5 g
2649	Yes	0.5 g
3197	Yes	0,5 g

APPENDIX 3

Number of participants per country

1 lab in BANGLADESH
2 labs in GERMANY
1 lab in HONG KONG
1 lab in TAIWAN
2 labs in TURKEY

APPENDIX 4

Abbreviations

C	= final test result after checking of first reported suspect test 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
R(0.01)	= outlier in Rosner's outlier test
R(0.05)	= straggler in Rosner's outlier test
ex	= test result excluded from statistical evaluation
n.a.	= not applicable
n.e.	= not evaluated
n.d.	= not detected

Literature

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