European Commission

JRC VALIDATED METHODS, REFERENCE METHODS AND MEASUREMENTS REPORT

EURL-FCM-02-2016 Proficiency Test Report

Temperature control during migration test and quantification of migrated FCM No 500 by article filling

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2018

EUR 29121 EN

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JRC109543

PDF ISBN 978-92-79-79934-1 ISSN 1831-9424 doi:10.2760/940236

Luxembourg: Publications Office of the European Union, 2018

How to cite: E. Tsochatzis, J.F. Alberto Lopes, P. Robouch and E.J. Hoekstra; "EURL-FCM-02-2016 Proficiency Test Report: Temperature control during migration and quantification of migrated FCM No 500 by article filling", EUR 29121 EN, European Commission, Ispra, 2018, ISBN 978-92-79-79934-1, doi:10.2760/940236, JRC109543

European Union Reference Laboratory
for Food Contact Materials

EURL-FCM-02-2016
Proficiency test report

Temperature control during migration test and determination of migrated FCM No 500 by article filling

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268-PT Accredited by the
Belgian Accreditation Body (BELAC)

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Abstract

The European Union Reference Laboratory for Food Contact Materials (EURL-FCM) organised a proficiency test (PT) to assess the analytical capabilities of the EU National Reference Laboratories (NRLs) and Official Control Laboratories (OCLs) on the "temperature control" during migration tests by article filling at 70 °C for 2 h in food simulant D1 (ethanol 50 %, v/v), with the subsequent determination of the specific migration of 2,5-bis(5-tert-butyl-2-benzoxazolyl)thiophene (cf. FCM No 500 in EU Reg. 10/2011).

A total of 41 laboratories (25 NRLs and 16 OCLs) from 26 different countries reported results for the "temperature control" exercise. Thirty two laboratories (24 NRLs and 8 OCLs) submitted results for the "FCM No 500 quantification".

For "temperature control", participants were requested to implement the learnings of the previous PTs when designing their experimental set up. Some general instructions were provided by the EURL-FCM. The test specimens consisted of a set of five 0.3 L polypropylene cups. Laboratories were requested to monitor the temperature of the food simulant inside one of the test specimens during the contact phase. Participants were requested to provide details of their operating procedure and to report their temperature readouts. The results show that 59 % of the 41 laboratories were able to maintain the temperature of the food simulant inside the tolerance range (70 °C ± 2 °C) during the entire migration contact time. This is a significant improvement compared to the results obtained in 2015. Another evaluation approach was investigated checking whether the mean temperature of the entire experiment falls in the range of 68-72 °C; 85 % of the laboratories performed successfully.

For the "FCM quantification", the EURL-FCM confirmed the test items to be adequately homogeneous and stable. Laboratories had to develop an appropriate analytical method and to provide details. The assigned values of the migration of FCM No 500 were calculated as the robust mean and robust standard deviation of the results reported by the participants. The laboratory performance was expressed in terms of z and zeta scores in accordance with ISO 13528:2015. The outcome of this exercise was satisfactory for 80 % of the laboratories (24 out of 30). The remaining six laboratories had unsatisfactory z scores higher than 3. Although their quantification performance at 5 µg kg^{-1} level was unsatisfactory, this does not imply that their methods of analysis may be not be suitable for the determination at the legal specific migration limit (SML) of 600 µg kg^{-1}.

Laboratories were requested to estimate and report their measurement uncertainties. Half of them reported reasonable estimates. The EURL-FCM will organise a dedicated training to share best practices on the topic.

The experimental information collected through the questionnaire further confirms the critical factors affecting the migration experiment: (1) covering/sealing the test specimen to reduce solvent losses, (2) reducing the filling time, (3) using calibrated thermometers/dataloggers and (4) avoiding pre-heating of the test specimen prior to the migration experiment.

List of abbreviations

D1	Food simulant: 50 % v/v ethanol in water
DG SANTE	Directorate General for Health and Food Safety
EURL-FCM	European Union Reference Laboratory for Food Contact Materials
FCM	Food Contact Material
FCM No 500	2,5-bis(5-tert-butyl-2-benzoxazolyl)-thiophene
HPLC-DAD	High Pressure Liquid Chromatography with Diode Array Detector
HPLC-FLD	High Pressure Liquid Chromatography with Fluorescence Detector
HPLC-UV	High Pressure Liquid Chromatography with UltraViolet detector
JRC	Joint Research Centre
LC-MS/MS	Liquid Chromatography with tandem Mass Spectrometer detector
LC-qTOF-MS	Liquid Chromatography with Quadrupole Time-Of-Flight Mass Spectrometer detector
LOQ	Limit of Quantification
NRL	National Reference Laboratory
OCL	Official Control Laboratory
PCA	Principal Component Analysis
PP	Polypropylene
PT	Proficiency Test
RSD	Relative Standard Deviation (in %)
SM	Specific Migration
SML	Specific Migration Limit

1. Introduction

The European Union Reference Laboratory for Food Contact Materials (EURL-FCM), hosted by the Joint Research Centre (JRC), organised the proficiency test (PT) **EURL-FCM-02-2016** to (i) monitor the temperature during the migration test, and (ii) to quantify the migration of FCM No 500 by article filling.

Temperature among other parameters determines to a great extent the migration of a substance from a FCM to food. Therefore, it should be controlled and monitored accurately during migration tests. Laboratories observed that the required contact temperature is seldomly reached within an acceptable time nor maintained in the required range when the migration test is performed at elevated temperatures for short contact times.

During the PT organised by the EURL-FCM in 2015 (ILC01-2015 [1]) laboratories performed migration tests by article filling at 70 °C, maintaining the required temperature within the **tolerance range of ± 2 °C** specified in the EN 13130-1:2004 documentary standard [2]. Results showed that only 30 % of the 53 migration experiments (performed by the 45 laboratories) were successfully carried out maintaining the temperature of the food simulant within the tolerance range during the entire contact time.

The EURL-FCM decided to organise a follow-up exercise using the same test specimens and applying the same migration conditions, to compare the results obtained in the frame of the two PTs. In addition, this PT included the **quantification of "FCM No 500"** (2,5-bis(5-tert-butyl-2-benzoxazolyl)-thiophene, an additive included in the positive list of Regulation (EU) No 10/2011 [3]), in food simulant D1 (50 % v/v ethanol in water) after migration testing by article filling at 70 °C for 2 h.

This PT was agreed with the Directorate General for Health and Food Safety (DG SANTE) as part of the EURL-FCM annual work programme 2016-2017. The PT was open to National Reference Laboratories (NRLs) and to Official Control Laboratories (OCLs) willing to participate.

This report summarises the outcome of the PT.

2. Scope

As stated in Regulation (EC) No 882/2004 [4] one of the core duties of EURLs is to organise interlaboratory comparisons for the benefit of NRLs and OCLs.

The following objectives were set for this PT:

I. To assess the ability of the laboratories to develop and perform a migration test protocol by article filling using food simulant D1 (50% v/v ethanol in water) for 2 h at 70 °C inside a thermostatic oven/incubator respecting the required tolerance range (± 2 °C).

II. To compare the obtained results of the temperature control measurements with the results of ILC 01-2015 in order to assess potential improvements in laboratory performance.

III. To evaluate the capacity of the laboratories to determine the specific migration (SM) of FCM No 500 in food simulant D1 after the migration experiment by article filling at 70 °C for 2 h.

IV. To identify any correlation between the migration protocols developed by the participants (cf. sample pre-heating, simulant pre-heating, oven's temperature, etc.) and the quantification of the SM.

V. To derive recommendations for laboratories (i) for temperature control, and (ii) for the quantification of the specific migration by article filling.

The reported results were assessed following the administrative and logistic procedures of the JRC Unit in charge of the EURL-FCM, which is accredited for the organisation of PTs according to ISO 17043:2010 [5].

3. Set up of the exercise

3.1 Time frame

The organisation of the EURL-FCM-02-2016 exercise was agreed upon by the NRL network at the plenary meeting held in June 2016. Invitation letters were sent (via e-mail) to all NRLs of the network on December 12, 2016 (Annex 1). NRLs were advised to contact other interested OCLs. The registration deadline was set to December 15, 2016. Samples were sent in early January 2017. The dispatch was monitored by the PT coordinator using the messenger's parcel tracking system on the internet. The deadline for reporting of results was set to March 01, 2017.

3.2 Confidentiality

The procedures used for the organisation of PTs are accredited according to ISO 17043:2010 [5] and guarantee that the identity of the participants and the information provided by them is treated as confidential. However, the lab codes of the NRLs that have been appointed in line with Regulation (EC) No 882/2004 may be disclosed to DG SANTE upon request for the purpose of an assessment of their (long-term) performance.

3.3 Distribution

Each participant received:

- One test specimen consisting of 5 polypropylene cups (0.3 L);
- 200 mg of the pure analytical calibration standard of 2,5-bis(5-tert-butyl-2-benzoxazolyl)-thiophene (Sigma Aldrich, ≥ 99.5 % purity);
- The "Confirmation" form (Annex 2);
- The "Shipping and Instructions" form (Annex 3);
- The "Sample Acknowledgment Receipt" form to be sent back to the JRC after receipt of the test item (Annex 4);
- Instructions for "Compilation of Results" (Annex 5);
- The "Results Reporting" form (Annex 6); and
- The "Questionnaire" form (Annex 7).

3.4 Instructions to participants

Detailed instructions for the compilation of the results were given to the participants in the letter that accompanied the samples (see Annex 5), regarding the temperature control of the migration experiment and the determination of FCM No 500. Some of the instructions were requests that had to be followed, some others were only considerations/suggestions based on the two EURL-FCM methods (with or without oven ventilation), meant to assist in the development of the oven protocol for the migration experiment. Laboratories were free to adapt the provided information; they were asked to describe the implemented protocol in the "Questionnaire" form (see Annex 7).

Participants were asked to perform a SM test by article filling with food simulant D1 (i.e. ethanol 50 % v/v) for 2 h at 70 °C ± 2 °C inside a thermostatic oven or incubator. Four test specimens (cups) were to be filled with 300 mL of D1, and then introduced in an oven/incubator.

The temperature of the food simulant inside the fourth cup (the one filled last and/or placed into the thermostatic oven/incubator last) had to be monitored. Participants were instructed to use only a **calibrated thermometer/data logger** for the temperature measurements. A total of 33 readouts were to be reported using the "Results reporting form" (Annex 6).

The first three cups were to be used for the quantification of migrated FCM No 500 applying an appropriate analytical method developed by each laboratory.

Participants were also requested to report the associated expanded measured uncertainty together with the coverage factor and to describe the approach used for the estimation of the measurement uncertainty.

A dedicated questionnaire was used to gather additional information related to measurements and laboratories (Annex 7).

The laboratory codes were given randomly and communicated to the participants (Annex 8).

4. Test specimen

4.1 Preparation

As in ILC01-2015, the tests specimens consisted of five polypropylene cups (0.3 L) meant to be used with high alcoholic content beverages. Packs of five randomly selected cups were wrapped in aluminium foil and provided to the participants (as shown on the cover page of this report).

For the determination of the migration of FCM No 500, amber vials with approximately 200 mg of the pure analytical standard 2,5-bis(5-tert-butyl-2-benzoxazolyl)-thiophene (Sigma Aldrich, ≥ 99.5% purity) were prepared and sent together with the cups.

4.2 Homogeneity and stability

The homogeneity and stability studies for the migration of FCM No 500 from the test specimens were performed by the EURL-FCM, according to the requirements set in ISO 13528:2015 [6].

Fifteen randomly selected test specimens of the sample were analysed for FCM No 500 in duplicate, applying the single-laboratory validated HPLC-FLD analytical method developed by the EURL-FCM. The EURL-FCM evaluated homogeneity according to the IUPAC International Harmonized Protocol F-test [7] and checked for significant and adequate heterogeneity based on ISO 13528:2015 [6].

Similarly for stability, three test specimens of the sample were analysed for FCM No 500 in duplicate at the end of the PT (10 weeks later) by the EURL-FCM, using the HPLC-FLD method mentioned above. The EURL-FCM evaluated the stability according to the IUPAC International Harmonized Protocol F-test [7] and the expanded criteria of ISO 13528:2015 [6].

The statistical treatment of data was performed by the EURL-FCM using the ProLab Software [8].

The homogeneity results and their statistical evaluation are presented in Annex 9. The test material showed sufficient homogeneity. Similarly, the test material showed sufficient stability and no significant trend was observed for the tested samples.

5. Assigned values and corresponding uncertainties

5.1 Assigned values

The assigned value (x_{pt}) for the mass fraction of FCM No 500 released during the migration experiment was derived as consensus value from results reported by the participants applying Algorithm A (ISO 13528:2015 – Annex C, [6]).

5.2 Associated uncertainties

Robust statistics (cf. Algorithm-A, ISO 13528:2015, [6]) was used to derive the uncertainty associated with the assigned value $u(x_{pt})$ from the results reported by the participants as follows:

$$u(x_{pt}) = 1.25 \frac{s^*}{\sqrt{n}} \qquad \text{Eq. 1}$$

Where "s*" is the robust standard deviation, and n is the number of reporting participants.

The factor 1.25 represents the ratio of the standard deviation of the median to the standard deviation of the arithmetic mean, for large samples p>10. This factor is recommended because proficiency testing results are typically not strictly normally distributed and contain unknown proportions of results from different distributions. The factor of 1.25 is considered to be a conservative estimate.

5.3 Standard deviation for proficiency assessment

The value of target standard deviation (σ_{pt}) determines the limits of satisfactory performance in a PT. It should be set as a value that reflects best practice for the investigated analysis. The standard deviation of the reproducibility found in collaborative trials among experts/competent laboratories is generally considered as an appropriate indicator of the best agreement that can be obtained between laboratories.

In the absence of appropriate collaborative trial data on this migration test and being aware of specific difficulties associated to the reproducibility of the release of target analytes from FCMs, the **robust reproducibility standard deviation** (s*) derived from the reported results was set as σ_{pt}.

6. Evaluation of results

6.1 Scores and evaluation criteria

The individual laboratory performance was expressed in terms of z and ζ scores according to ISO 13528:2015 [6]:

$$z = \frac{x_i - x_{pt}}{\sigma_{pt}} \qquad \text{Eq. 2}$$

$$\zeta = \frac{x_i - x_{pt}}{\sqrt{u^2(x_i) + u^2(x_{pt})}} \qquad \text{Eq. 3}$$

Where: x_i is the measurement result reported by a participant;

$u(x_i)$	is the standard measurement uncertainty reported by a participant;
x_{pt}	is the assigned value;
$u(x_{pt})$	is the standard measurement uncertainty of the assigned value;
σ_{pt}	is the standard deviation for proficiency test assessment.

The interpretation of the z and ζ performance scores is done according ISO 13528:2015 [6]:

|score| ≤ 2	satisfactory performance	(green in Annexes 11)
2 < |score| < 3	questionable performance	(yellow in Annexes 11)
|score| ≥ 3	unsatisfactory performance	(red in Annexes 11)

Two laboratories reported "less than" values. These truncated values were not used in the statistical treatment of data, and no evaluation of these results was done (no scores were provided).

The z scores compare the participant's deviation from the assigned value with the standard deviation for proficiency test assessment (σ_{pt}) used as common quality criterion.

The ζ scores state whether the laboratory's result agrees with the assigned value within the respective uncertainty. The denominator is the combined uncertainty of the assigned value $u(x_{pt})$ and the measurement uncertainty as stated by the laboratory $u(x_i)$. The ζ score includes all parts of a measurement result, namely the expected value (assigned value), its measurement uncertainty in the unit of the result as well as the uncertainty of the reported values. An unsatisfactory ζ score can either be caused by an inappropriate estimation of the concentration, or of its measurement uncertainty, or both.

The standard measurement uncertainty of the laboratory $u(x_i)$ was obtained by dividing the reported expanded measurement uncertainty by the reported coverage factor, k. When no uncertainty was reported, it was set to zero ($u(x_i) = 0$). When k was not specified, the reported expanded measurement uncertainty was considered as the half-width of a rectangular distribution; $u(x_i)$ was then calculated by dividing this half-width by $\sqrt{3}$, as recommended by Eurachem [9].

Uncertainty estimation is not trivial, therefore an additional assessment was provided to each laboratory reporting measurement uncertainty, indicating how reasonable their measurement uncertainty estimation was.

The standard measurement uncertainty from the laboratory $u(x_i)$ is most likely to fall in a range between a minimum and a maximum allowed uncertainty (case a": $u_{min} \leq u_i \leq u_{max}$). u_{min} is set to the standard uncertainties of the assigned values $u(x_{pt})$. It is unlikely that a laboratory carrying out the analysis on a routine basis would determine the measurand with a smaller measurement uncertainty than the expert laboratories chosen to establish the assigned value. u_{max} is set to the standard deviation accepted for the PT assessment (σ_{pt}). Consequently, case "a" becomes: $u(x_{pt}) \leq u(x_i) \leq \sigma_{pt}$.

If $u(x_i)$ is smaller than $u(x_{pt})$ (case "b") the laboratory may have underestimated its measurement uncertainty. Such a statement has to be taken with care as each laboratory reported only measurement uncertainty, whereas the measurement uncertainty associated with the assigned value also includes contributions for homogeneity and stability of the test item. If those are large, measurement uncertainties smaller than $u(x_{pt})$ are possible and plausible.

If $u(x_i)$ is larger than σ_{pt} (case "c") the laboratory may have overestimated its measurement uncertainty. An evaluation of this statement can be made when looking at the difference between the reported value and the assigned value: if the difference is smaller than the expanded uncertainty $U(x_{pt})$ then overestimation is likely. If the difference is larger but x_i agrees with x_{pt} within their respective expanded measurement uncertainties, then the measurement uncertainty is properly assessed resulting in a satisfactory performance expressed as a ζ score, though the corresponding performance, expressed as a z score, may be questionable or unsatisfactory.

It should be pointed out that "u_{max}" is a normative criterion when set by legislation.

6.2 Laboratory results and scorings

Forty five laboratories from 25 EU Member States registered to the "temperature control" exercise; 25 NRLs and 16 OCLs (91 %) reported results. Similarly, 32 laboratories registered to the "FCM No 500 quantification" exercise; 24 NRLs and 8 OCLs (94 %) reported results (two of which were "less than values").

6.2.1 Temperature control

Annex 10 presents the mean temperature and the corresponding standard deviation derived from the 33 readouts performed by the laboratories during migration test by article filling.

Three types of profiles have been identified (Figure 1):

a. the majority of the (or all) data points are below the **target temperature range** set by EN 13130-1:2004 (70 ± 2 °C);
b. all the data points are included in the above mentioned temperature range;
c. several data points are above the temperature range.

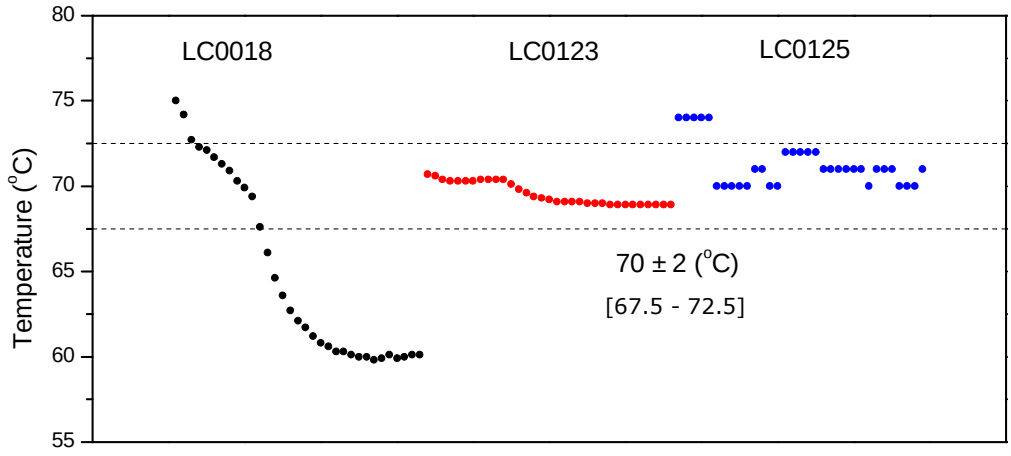

Figure 1: Examples of temperature profiles submitted by three participants (LC0###)

The first evaluation approach consisted in counting the number of temperature readouts included in the target temperature range (70 ± 2 °C), and compare it to the total number of temperatures to be reported (33, as requested in Annex 6).

Annex 10 shows that only 24 out of 41 laboratories (**59 %**) strictly complied with the requirements set by the CEN standard, thus **passing the test** and obtaining a score of 100 %.

Two laboratories (LC0002 and LC0003) reported all their temperatures below 68 °C, resulting in a score of 0 %, while the remaining 15 laboratories obtained scores ranging from 42 % to 94 %. Laboratories having obtained low scores may consider re-evaluating their oven protocol.

In the second evaluation approach the test is considered successful when the average of all reported temperature readouts is included in the target temperature range. Figure 2 and Annex 10 show that **35 laboratories (85 %) passed this test**. The remaining laboratories reported averages ranging from 64.6 to 67.3 °C.

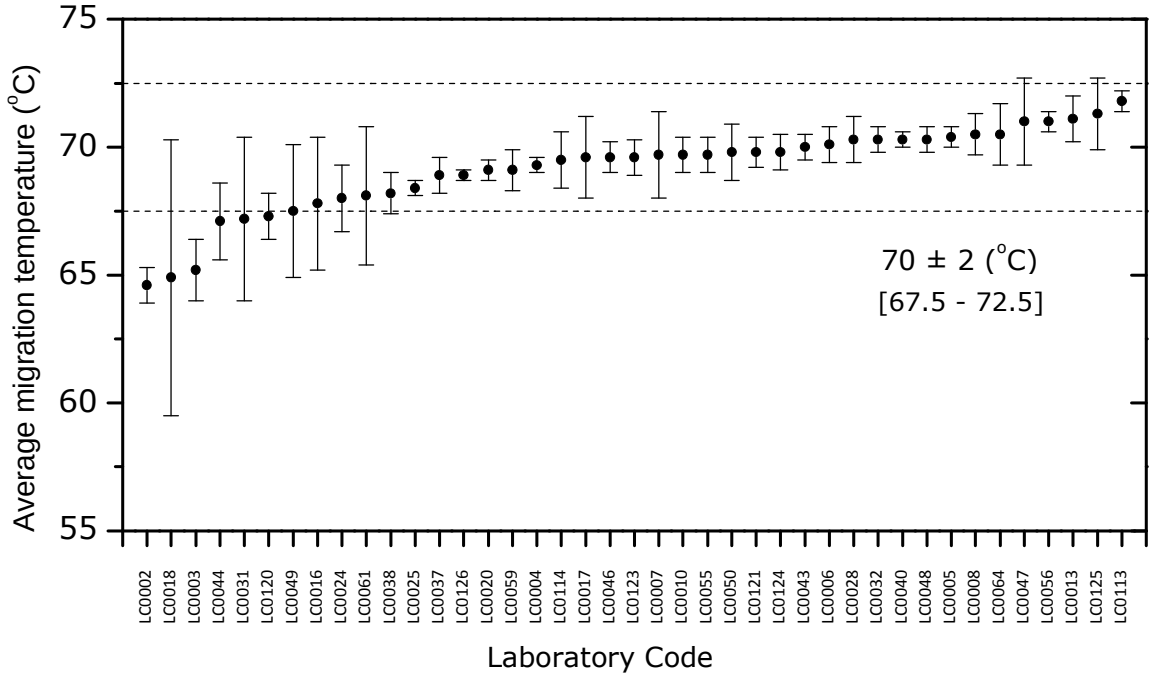

Figure 2: Average temperatures (and standard deviations) reported by the laboratories

6.2.2 FCM No 500 quantification

For the SM of FCM No 500 after migration by article filling with food simulant D1 at 70 °C for 2 h, a total of 32 laboratories participated (24 NRLs and 8 OCLs) and submitted their results. However, two laboratories could not quantify FCM No 500, as the limit of quantification (LOQ) of their analytical method was higher than the substance's concentration, though sufficient for analysis based on its respective SML [3, 10]. The results submitted by the participants, along with information regarding the results SM of FCM are available in Annex 11.

The included Kernel density plot was obtained using the software available from the Statistical Subcommittee of the Analytical Methods Committee of the UK Royal Society of Chemistry [11].

The assigned value of the mass fraction of "migrated FCM No 500 in D1" was obtained as a consensus value from participant results. The laboratory performance was assessed using the *z* and ζ scores.

Figure 3 present the laboratory performances for the mass fraction investigated assessed by the *z* and ζ scores. **80 % of the participants having reported results performed satisfactorily according to the *z score*** (67 % according to ζ score).

Two major analytical techniques were applied, based on (i) high pressure liquid chromatography (HPLC) with diode array, fluorescent or UV detectors, or (ii) liquid or gas chromatography coupled to mass spectrometers. No direct correlation could be identified between poor performance and the analytical technique used.

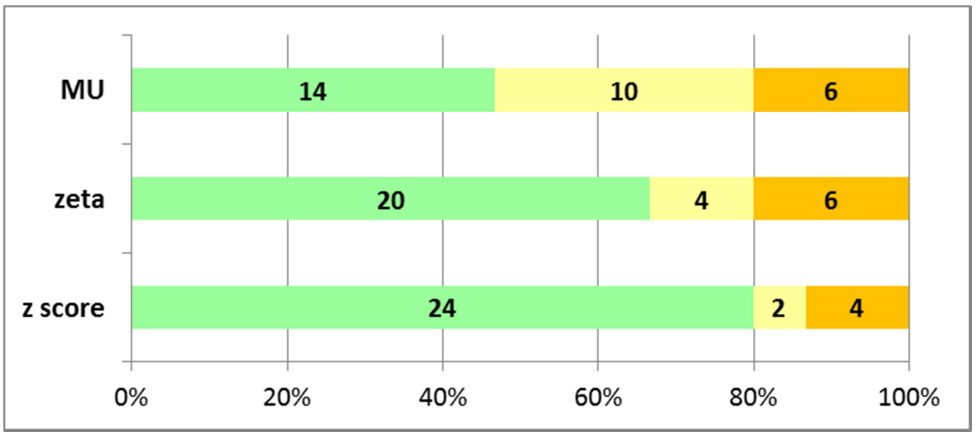

Figure 3: Overview of laboratory performance according to *z* and *ζ* scores, for FCM No 500 in D1, and measurement uncertainties (MU).
- Corresponding number of laboratories included in the graph.
- Satisfactory, questionable and unsatisfactory performance, in green, yellow and orange, respectively.
- For MU: Case "a" (green): $u(x_{pt}) \leq u(x_i) \leq \sigma_{pt}$; Case "b" (yellow): $u(x_i) < u(x_{pt})$; Case "c" (orange): $u(x_i) > \sigma_{pt}$

6.2.3 Measurement uncertainties

Laboratories that have to comply with ISO 17025:2005 [12] - the standard for the competence of testing and calibration laboratories - are supposed to provide realistic measurement uncertainties. For this PT the EURL-FCM requested laboratories to estimate and report their measurement uncertainty.

Figure 3 presents the evaluation of the reported measurement uncertainties. **Half of the participants reported realistic standard uncertainty** ranging from 0.4 to 1.7 mg kg^{-1} (case "a": $u(x_{pt}) \leq u(x_i) \leq \sigma_{pt}$). Some participants reported likely overestimated standard uncertainties above 2 mg kg^{-1}, while others reported underestimated uncertainties around 0.2 mg kg^{-1}. Four laboratories did not report any measurement uncertainty.

Note: x_{pt} = 5.63 mg kg^{-1}

6.3 Additional information extracted from the questionnaire

The questionnaire was answered by all participants giving valuable information on the laboratories, their way of working and their analytical methods.

The questionnaire related to two topics:

I. Migration conditions and temperature measurements;
II. Analytical methodologies.

Regarding the migration conditions, the following critical parameters were identified from the previous PTs (ILC 02-2016 and ILC 01-2015):

1. Pre-heating FCM specimen before the migration experiment;
2. Filling time of the FCM specimen with the selected food simulant;
3. Covering of the FCM specimen during the migration experiment;
4. Food simulant loss during the migration experiment;
5. Type of thermometer/datalogger used for temperature control during the migration experiment.

All these parameters may have potentially affected the results of the migration experiment, and subsequently the analytical determination of FCM No 500.

Figure 4 shows the percentage of laboratories having performed a **pre-heating step** of the FCM specimen prior to the migration experiment. Almost half of the laboratories decided to pre-heat the specimens, even when no further migration experiment was planned. For the laboratories that performed both exercises ("temperature control" and "FCM No 500 quantification"), the percentage of pre-heated/non pre-heated is almost the same.

The **pre-heating temperature** varied considerable among laboratories. These conditions are presented in Figure 5, together with the percentages of laboratories that used them. Eighteen out of the 30 laboratories pre-heated the test specimen. 22 % of them did it at a temperature below or close to the one used for the migration experiment (70 °C ± 2 °C); 61 % at a temperature close or slightly higher; and the remaining 17 % at significantly higher temperatures.

Figure 6 presents the **filling time** of the test specimen with food simulant just before the migration experiment. 83 % of the participants are able to fill up the test specimen in less than a minute. According to the EURL-FCM half a minute is the ideal filling time, requiring some training. 49 % of the laboratories reached already this target. A small filling time of the test specimen is crucial for the success of the migration experiment, as it may limit potential temperature drops at the beginning of the migration experiments.

The **covering of the test specimen** during the migration experiment is another factor affecting the migration experiment, as it is closely related to the overall food simulant loss. Figure 7 summarises the various types of coverings used by the participants. The majority of the laboratories used either aluminium foil or clock glasses to cover the test specimens during the migration experiment. Even though no correlation could be found between the type of covering and the volume of food simulant loss, the EURL-FCM observed that good results were obtained when the clock glass was used with the concave side turned down, allowing the food simulant to condense on the surface of the glass and drip back into the beaker. The loss of the food simulant is further decreased when ensuring a tighter seal (e.g. a silicone covered-metal o-ring). Due care should then be taken to avoid any contamination of the investigated food simulant.

The majority of the participants (66 %) reported **food simulant losses** up to 10 mL (Figure 8), which represents 3 % of the total food simulant volume used (300 mL). Although the influence of different volumes of food simulant losses on the performance of the migration experiment are not clearly established, it is recommended to reduce these losses as much as possible. As mentioned earlier, the use of a glass clock and a pressure on top may reduce the loss to to less than 3 mL (or 1 % of the initial volume).

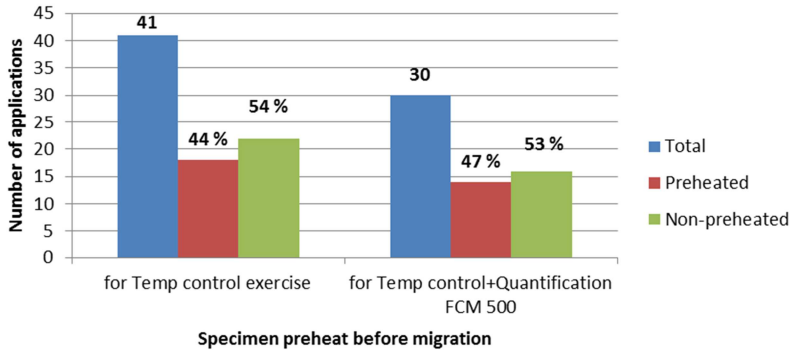

Figure 4.
Test specimen pre-heated or not before the test?

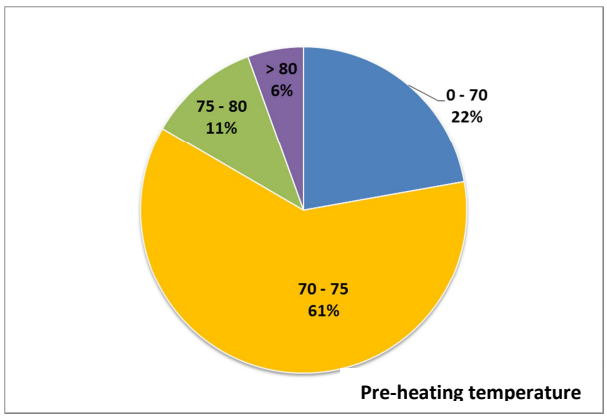

Figure 5.
Pre-heating temperatures used by 18 participants (out of 30)

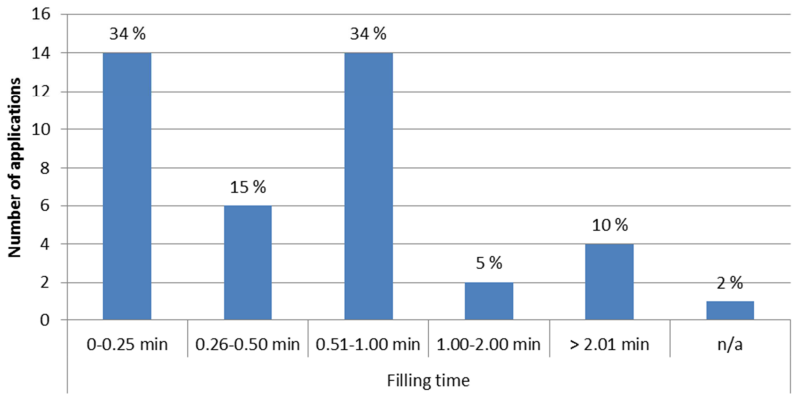

Figure 6.
Reported filling times

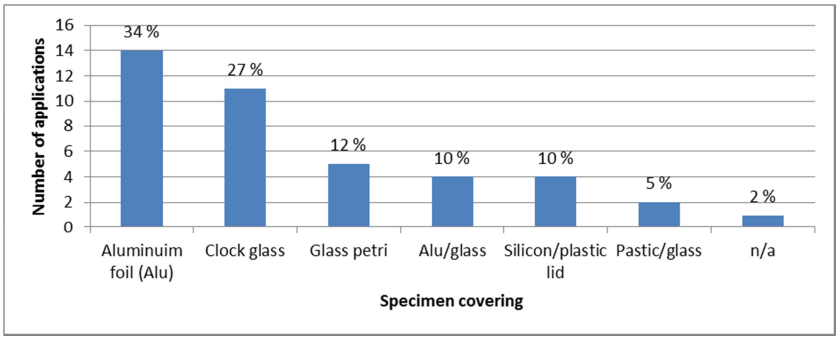

Figure 7.
Different test specimen coverings used during migration

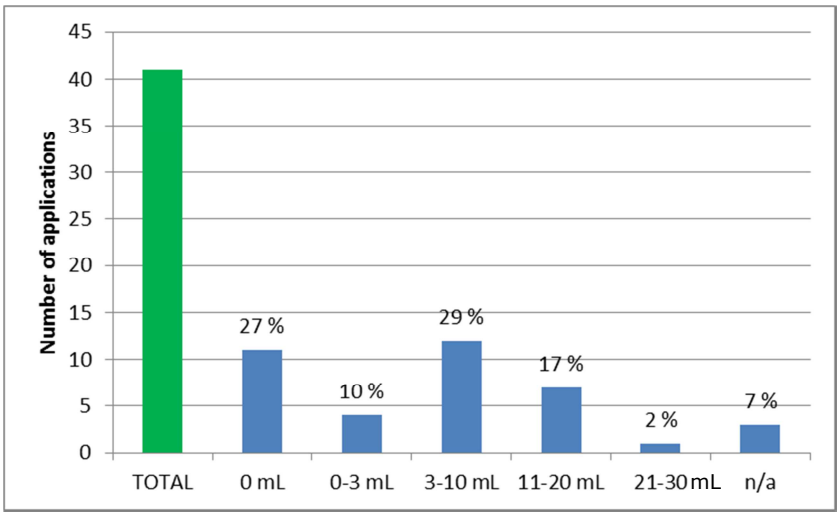

Figure 8.
Reported food simulant losses (mL) during migration

Despite the formal request by the EURL-FCM to use **calibrated thermometers or dataloggers**, a significant number of laboratories used non-calibrated equipment. Figure 9 presents the distribution of 41 laboratories having performed only the "temperature control" test (blue bars) or having also quantified the released FCM No 500 in D1 (red bars). It is regrettable to notice that 27 % of the participants continue to use non-calibrated equipment. This will be closely monitored by the EURL-FCM.

As mentioned earlier, the majority of the laboratories used HPLC methods coupled with FLD (53 %) or UV (13 %), followed mass spectrometry techniques (LC-MS/MS, 13 %; GC-MS, 9 %; LC-MS, 3 %; and UPLC-qTOF-MS 3 %) – Figure 10.

The vast majority of the laboratories (72 %) injected the food simulant D1 directly into the chromatographic system (Figure 11). Only few laboratories performed liquid-liquid extraction (LLE, 6 %) or evaporation/reconstitution (3 %). However, six laboratories did not provide any information (19 %).

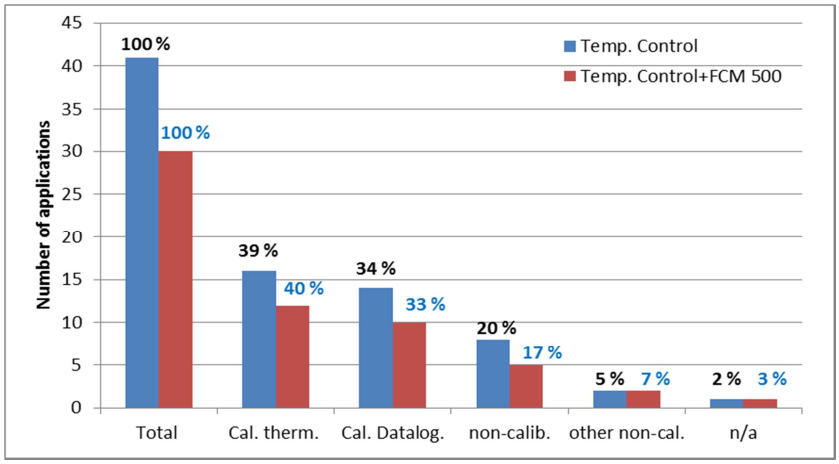

Figure 9.
Type of thermometer/data loggers used by the participants

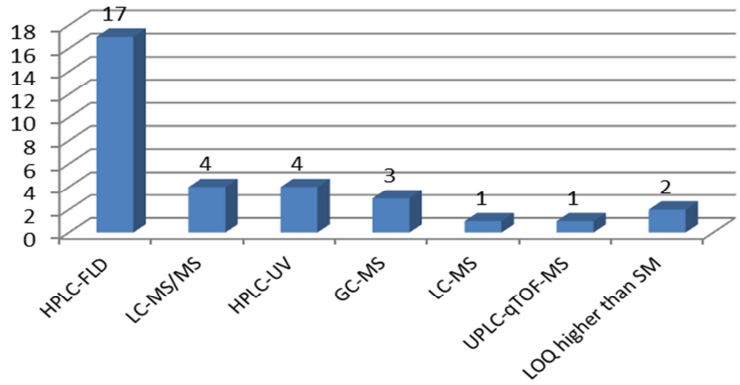

Figure 10.
Analytical instrumentation used for the quantification of FCM No 500 by the participating laboratories

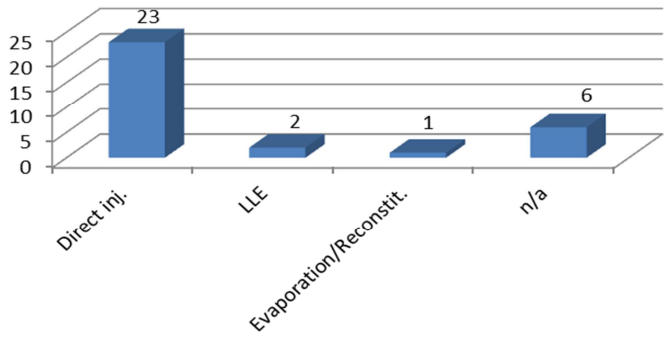

Figure 12.
Sample preparation used for the quantification of FCM 500 as reported by the participating laboratories

6.4 Origin of main variance of results

In order to identify and rank the parameter(s) that influence(s) the most the migration of target substances from the food contact material to the food simulant solutions, a statistical treatment of the reported results coupled with the experimental information provided in the questionnaire was performed using Principal Components Analysis (PCA).

PCA is a procedure for the identification of a smaller number of uncorrelated variables (called "principal components") from a large set of parameters, and explaining the maximum amount of the observed variance with the fewest number of principal components [13].

Figure 13 presents the PCA and cluster analysis of the participant's results (z scores, mean migration temperature, test specimen preheating temperature and time, food simulant loss, test specimen filling time). Figure 14 shows a similar analysis performed with the data reported by laboratories having used calibrated thermometers/dataloggers only. The first principal component is mainly driven by the mean temperature of the food simulant and the z score, while the second principle component is mainly driven by the test specimen pre-heating temperature. That is the reason why both figures show a clear differentiation between laboratories who pre-heated the test specimens and those who didn't. These two identified clusters become even more evidenced when rejecting results of laboratories that did not use calibrated thermometers/dataloggers (Figure 14).

Taking into consideration the material and the experiment used in this PT, it can be confirmed that **pre-heating of test specimens is not recommended**, as it may lead to unforeseen changes in the migration of target substances, especially if these values are close to SMLs.

6.5 General recommendations on the migration protocol

Based on the results of the present PT, the following recommendations can be drawn regarding the migration protocol at 70 °C for 2 h:

1. Use of calibrated thermometers or data loggers is compulsory;
2. Preheating the food contact material before migration should be avoided;
3. Food simulant losses should be minimised using proper covering/seal of the cup during the migration experiment.
4. Filling up the specimen with the food simulant should be done within half a minute to reduce temperature drops of the food simulant in the oven.

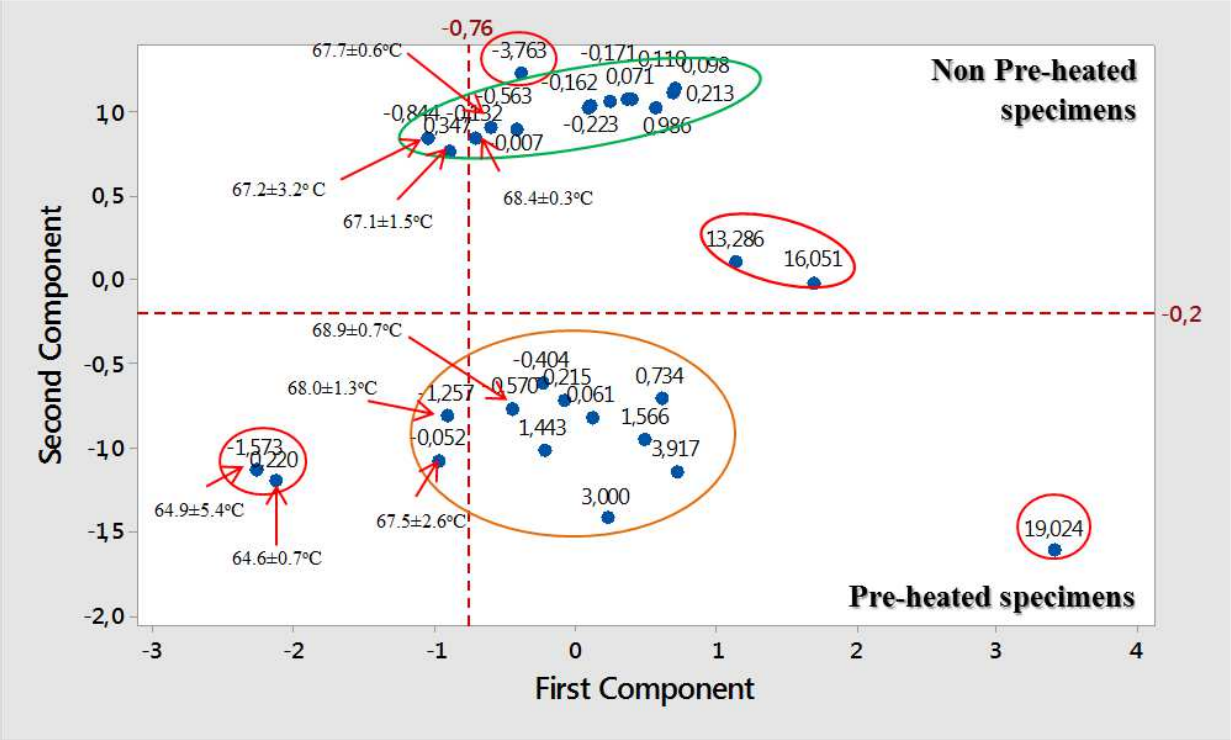

Figure 13. First versus second component after PCA analysis of z-scores and influencing parameters reported by all the laboratories.

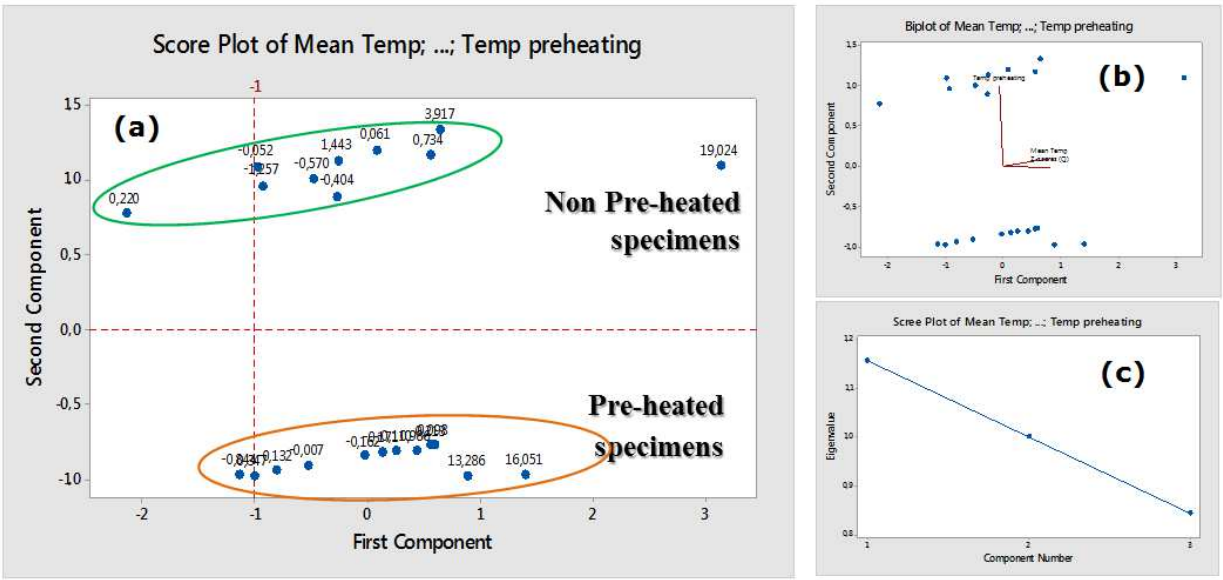

Figure 14 Principal component analysis of the laboratories that used calibrated thermometers and dataloggers: (a) Score plot, (b) biplot; and (c) scree plot

7. Conclusion

The proficiency test EURL-FCM-02-2016 was organised to assess the analytical capabilities of the EU NRLs and OCLs on the temperature control during migration tests by article filling at 70 °C for 2 h in food simulant D1 (ethanol 50 %, v/v), with the subsequent determination of the specific migration of 2,5-bis(5-tert-butyl-2-benzoxazolyl)thiophene (cf. FCM No 500 in EU Reg. 10/2011).

Results reported by the participants were evaluated based on the total mean migration temperature. A significant improvement was observed, with 85 % of the participating laboratories succeeding in having a mean temperature in the range of 70 ± 2 °C (compared to 28 % of the laboratories observed in 2015).

While 80 % of the participants having reported results for the determination of mass fraction of migrated FCM No 500 in food simulant D1 performed satisfactorily (according to the z score), only half of the participants estimated realistic measurement uncertainties. As a follow up the EURL-FCM will organise a dedicated training on the "estimation of the measurement uncertainty ".

The experimental information collected through the questionnaire further confirmed the critical factor affecting the migration experiment: (1) covering/sealing the test specimen to reduce solvent losses, (2) reducing the filling time, (3) using calibrated thermometers/dataloggers and (4) avoiding pre-heating of the test specimen prior to the migration experiment.

Acknowledgements

The EURL-FCM wishes to acknowledge the laboratories having participated to this PT (listed hereafter).

National Reference Laboratories

Austria	Austrian Agency for Health and Food Safety (AGES) Institut für Lebensmittelsicherheit, Wien
Belgium	Institute of Public Health, ISSP-LP, Bruxelles
Bulgaria	National Centre of Public Health & Analysis , Sofia
Croatia	Croatian Institute of Public Health, Zagreb
Cyprus	State General Laboratory, Nicosia
Czech Republic	National Institute of Public Health, Praha 10
Denmark	Danish Veterinary & Food Administration, Laboratory Århus, Lystrup
Denmark	Technical University of Denmark, National Food Institute, Analytical Food Chemistry, Søborg
Estonia	Health Board, Central Chemistry Laboratory, Tallinn
Finland	Finnish Customs Laboratory, Espoo
France	LNE (Laboratoire National de Metrologie et d'Essais), Trappes
France	SCL Laboratoire de Bordeaux-Pessac, Pessac
Germany	Bundesinstitut für Risikobewertung (BfR) (Federal Institute for Risk Assessment), Berlin
Greece	General Chemical State Laboratory, Laboratory of Articles and Materials in Contact with Foodstuffs, Athens
Hungary	National Food Chain Safety Office, Food and Feed Safety Directorate, Food Toxicological NRL, Budapest
Ireland	Public Analyst's Laboratory, Dublin
Italy	Istituto Superiore di Sanità, Rep. Esposizione e rischio da materiali, Roma
Lithuania	National Public Health Surveillance Laboratory, Laboratory of Chemistry, Vilnius
Luxembourg	Laboratoire National de Santé, Division de Contrôle Alimentaires, Luxembourg
Poland	National Institute of Public Health - National Institute of Hygiene, Warsaw
Portugal	Escola Superior de Biotecnologia (ESB), Universidade Católica Portuguesa, Packaging Department, Porto
Slovakia	Regional Public Health Authority Poprad (RUVZ), Poprad
Slovenia	National Laboratory of Health, Environment and Food, Ljubljana
Spain	Centro Nacional de Alimentación, Agencia Espanola de Seguridad, Alimentaria y Nutrición (AESAN), Majadahonda-Madrid
Sweden	National Food Agency, Uppsala
The Netherlands	Food and Consumer Product Safety Authority (VWA), Groningen
United Kingdom	The Food and Environment Research Agency, York

Official Control Laboratories

Belgium	Belgisch Verpakkingsinstituut, (BVI), Zellik
Belgium	Celabor, Chaineux
Belgium	Federaal Laboratorium voor de Voedselveiligheid (FLVVG), Gentbrugge
Belgium	Laboratoire Fédéral pour la Sécurité Alimentaire Liège (LFSAL), Wandre
Belgium	SGS Belgium N.V. - Division IAC, Antwerpen
Czech Republic	Zdravotni ustav se sidlem v Ostrave, Centrum Hygienických Laboratoři, Ostrava
Czech Republic	Zdravotni ustav se sidlem v usti n/labem, Centrum Hygienických Laboratoři, Hradec Kralove
Germany	Bayerisches Landesamt für Gesundheit und Lebensmittelsicherheit R 5 Bedarfsgegenstände, Erlangen
Germany	Chemisches und Veterinäruntersuchungsamt Fellbach Abt. Bedarfsgegenstände, Fellbach
Germany	Chemisches und Veterinäruntersuchungsamt Münsterland-Emscher-Lippe (CVUA-MEL), Bedarfsgegenstände, Münster
Germany	Dezernatsleiter Bedarfsgegenstände, kosmetische Mittel, Rückstände, Kontaminanten, Ladesamt für Verbraucheschutz Sachsen-Anhalt, Halle
Germany	Landesuntersuchungsanstalt für das Gesundheits- und Veterinärwesen Sachsen, Dresden
Germany	LAVES -Institut für Bedarfsgegenstände Lüneburg, Lüneburg
Germany	Thüringer Landesamt für Verbraucherschutz, Bad Langensalza
Italy	Istituto Zooprofilattico Sperimentale LER, Bologna
Spain	Centro de Salud Pública, Laboratorio de Salud Pública, Alicante
Spain	Conselleria De Sanidad Universal y Salud Pública, Laboratorio de Salud Pública de Valencia, Valencia
Spain	Ministerio de Economía industria y Competitividad, Centro Analitico de Inspeccion y Control De Calidad de Comercio Exterior, Madrid

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Annex 1: Invitation letter

Ref. Ares(2016)6915991 - 12/12/2016

EUROPEAN COMMISSION
DIRECTORATE-GENERAL
JOINT RESEARCH CENTRE
Directorate F - Health, Consumers and Reference Materials
Food & Feed Compliance

EURL
European Union Reference Laboratory
for Food Contact Materials

Geel, 12 December 2016
JRC.F.5/EH/mt/ARES(2016) 16-027

INVITATION LETTER

Inter-laboratory comparison exercise on temperature control during migration tests and migration of FCM substance No 500 by article filling

ILC02-2016

Dear Madam, dear Sir,

On behalf of the European Union Reference Laboratory (EURL) for Food Contact Materials (EURL-FCM), I invite you to participate in an inter-laboratory comparison (ILC) exercise on temperature control during migration tests by article filling combined with using your temperature-control protocol for the determination of the specific migration of FCM substance No 500.

Studies by National Reference Laboratories (NRLs) and the EURL-FCM indicated that the required contact temperature when performing migration tests by article filling is often reached only after an (unacceptably) long period of time or even not at all. As the contact temperature affects migration results, it may also affect the decision about compliance or non-compliance of samples. In the previous ILC01-2015 it appeared that the temperature during a 2 h at 70°C migration test could not maintained at the required temperature of 70±2°C during the entire 2 h contact period by a substantial amount of NRLs.

Therefore this ILC is a follow-up of the previous ILC01-2015 and aims at showing your improvement controlling the temperature in the food simulant at the contact surface with the filled article. You are asked to carry out a migration test by article filling of the provided polypropylene cups (volume 0.3 l) with food simulant D1 (ethanol 50%, v/v) for 2 h at 70°C to the best of your knowledge. You need to monitor the temperature, preferably by using a data logger, of the food simulant inside one of the test specimens during the contact period using a calibrated thermometer.

In this ILC you are also requested to use your temperature control protocol for performing a migration test in triplicate for the determination of the specific migration of FCM substance No 500 (2,5-bis(5-tert-butyl-2-benzoxazolyl)thiophene). Besides the specific migration, please report the associated expanded measurement uncertainty, the coverage factor and the approach followed for uncertainty calculation.

Via E. Fermi 2749, T.P. 260, I-21027 Ispra (VA) - Ispra. Telephone: (39-0332) 785319. https://ec.europa.eu/jrc/
E-mail: eddo.hoekstra@ec.europa.eu

Please answer our questionnaire to the best of your knowledge, because the EURL-FCM will use all information available to perform a root-cause-analysis. The results will be used to formulate further recommendations for performing migration tests by article filling at elevated temperatures for short contact times.

I remind you that it is your duty as NRL to participate in the ILCs organised by the EURL-FCM and that the work programme is decided with your agreement. There is no charge for participation. Feel free to involve your local official control laboratories.

Please confirm your participation until the 15th of December 2016 by sending back the completed participation form to Emmanouil TSOCHATZIS (Emmanouil.TSOCHATZIS@ec.europa.eu) or João Alberto Lopes (Joao-Filipe.ALBERTO-LOPES@ec.europa.eu). Once we have received your confirmation of participation, we will send a sample kit to you. The shipment of the sample kits is foreseen until the 17th of December 2016. You will find detailed instructions concerning the requested results in the sample kits. The deadline for submission of results is 1st March 2017. For further information, please contact Emmanouil TSOCHATZIS (phone: +39-0332789548) or João Alberto Lopes (phone: +39-0332789782).

Sincerely yours,

Eddo Hoekstra
Operating Manager, European Union Reference Laboratory for Food Contact Materials

Enclosure: 1

Cc: H. Emons (JRC), B. Schupp (SANTE)

Annex 2: Participation form

Ref. Ares(2016)6915991 - 12/12/2016

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CONFIRMATION OF PARTICIPATION

ILC 02 2016: Temperature Control (article filling) and specific migration- Follow-up 01-2015

To participate in the exercise, complete the form and return it until **16th December 2016** by e-mail to Emmanouil.TSOCHATZIS@ec.europa.eu or Joao-Filipe.ALBERTO-LOPES@ec.europa.eu

DETAILS OF THE INTERLABORATORY COMPARISON EXERCISE	
ILC code	ILC 02 2016
ILC Title	Temperature control and specific migration
Year	2016
Sample type	Food packaging material
Parameters for determination	Perform migration with temperature control and quantification of FCM substance No 500
Sample quantity	• 5 plastic PP cups (0.3 L) • ±100 mg FCM substance No 500
Packaging	padded cardboard box
Shipment conditions	no special precautions
Sample dispatch	
Deadline for results	

PARTICIPATING INSTITUTION	
Organisation	
Laboratory	
CONTACT INFORMATION	
Contact person	
Address for sample dispatch	
Telephone	
Fax	
e-mail	

Annex 3: Shipping kit and Instruction form

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Directorate F –Health, Consumers and Reference Materials
F.5 Unit – Food and Feed Compliance

European Union Reference Laboratory for Food Contact Materials

SHIPPING KIT and INSTRUCTION FORM

ILC02-2016: Temperature Control (article filling) and specific migration- Follow-up ILC01-2015

Material/samples sent:
• 5 polypropylene plastic cups (volume: 0.3 L, colour: orange)
• Vial containing 2,5-bis(5-tert-butyl-2-benzoxazolyl) thiophene (CAS: 70128-64-5) analytical standard

Documents sent (by e-mail and/or as print copy):
• JRC.I.1.Form.FIT-EURL.02 ver.1_Shipping kit and Instruction Form • JRC.I.1.Form.FIT-EURL.03 ver.1_Sample Receipt Acknowledgement • JRC.I.1.Form.FIT-EURL.04 ver.1_Instructions for compilation of results • JRC.I.1.Form.FIT-EURL.05 ver.1_Laboratory code • JRC.I.1.Form.FIT-EURL.06 ver.1_Results Reporting Form • JRC.I.1.Form.FIT-EURL.07 ver.1_Questionnaire Form

Instructions:
1. All samples should be stored at room temperature. The analytical standard (2,5-bis(5-tert-butyl-2-benzoxazolyl)thiophene (CAS: 70128-64-5)) must be stored in refrigerator (4°C).
2. Closing date: 1st of March
3. Other instructions for compilation of results, have been sent (see Instructions for compilation of results)

Sincerely yours

Eddo Hoekstra
Operating Manager, European Union Reference Laboratory for Food Contact Materials

European Commission
DG Joint Research Centre
Directorate F. Health, Consumers and Reference Materials
Food and Feed Compliance Unit
Via E. Fermi 2749, T.P. 260
I-21027 Ispra (VA), Italy

Annex 4: Sample acknowledgement receipt

EUROPEAN COMMISSION
DIRECTORATE GENERAL JRC
JOINT RESEARCH CENTRE
Directorate F –Health, Consumers and Reference Materials
F.5 Unit – Food and Feed Compliance

European Union Reference Laboratory for Food Contact Materials

To participate in the exercise, complete the form and return by e-mail to Emmanouil.TSOCHATZIS@ec.europa.eu or Joao-Filipe.ALBERTO-LOPES@ec.europa.eu **within 14 days** after the sample receipt

SAMPLE RECEIPT ACKNOWLEDGEMENT FORM

ILC 02 2016: Temperature Control (article filling) and specific migration-Follow-up 01-2015

LABORATORY NAME:	
LABORATORY CODE:	
SAMPLE CODE:	-
DATE OF RECEIPT:	
STATE OF SAMPLE:	

COMMENTS:

Date Name/Signature

<Sample Receipt Acknowledgement Form> *Page 1 of 1*

Annex 5: Instructions for compilation of results

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INSTRUCTIONS FOR COMPILATION OF RESULTS

ILC02 2016: Temperature Control (article filling) and specific migration- Follow-up of ILC01-2015

Perform a specific migration test by article filling with **food simulant D1** (i.e. ethanol 50% (v/v)) for **2 h at 70±2°C** inside a thermostatic oven or incubator. Fill four of the provided test specimens at least 0.5 cm from the top, put them into the oven and monitor the **temperature of the food simulant inside the fourth test specimen**, i.e. the one filled last and/or placed last into the thermostatic oven/incubator. Use only a **calibrated thermometer** to carry out the temperature measurements. The first three samples need to be analysed for migration of **FCM substance No 500** (2,5-bis(5-tert-butyl-2-benzoxazolyl)thiophene).

Read out the temperature values before and during the migration experiment based on the "**Results Reporting Form.doc**". For the control of the food simulant temperature, use only a calibrated thermometer or data logger as to verify that the food simulant temperature is at the desired temperature. In case a calibrated oven is not available, please also record the temperature of the air in the oven with a calibrated thermometer or data logger near the articles as to verify that the oven is operating in the desired temperature.

It is suggested first to establish the migration protocol and later perform the migration experiment together with the temperature monitoring. Based on the results of the previous ILC 01-2015 as well as from the EURLs' optimized in-house migration protocols in 70 ± 2°C for 2 h, some general considerations for two constant oven temperature programs are provided in Appendix I.

In the present ILC, there is also a requirement to quantify FCM substance No 500 (2,5-bis(5-tert-butyl-2-benzoxazolyl)thiophene). Besides the specific migration results, please report the associated expanded measurement uncertainty, the coverage factor and the approach followed for uncertainty calculation.

It is up to you whether and, if so, how (i.e. where, for how long, at which temperature/up to which temperature) to preheat the test specimens, the food simulant and the thermostatic oven/incubator in which the migration test is carried out afterwards. It is also up to you how/where to fill the test specimens and whether/how you cover the test specimens during the migration test. **You should perform the migration test to the best of your knowledge**, following eventually present standard operating procedures that are in place in your laboratory and trying to make sure that the food simulant reaches the desired test temperature of (70 ± 2)°C after placing the filled test specimens inside the thermostatic oven.

Report your results in the provided Word file "Results Reporting Form.doc" or fill in the print copy. Please also fill in the **questionnaire** and provide as much details as possible on the procedure that you have applied to perform the migration test. Please send your results and the completed questionnaire back by email to Emmanouil Tsochatzis (Emmanouil.TSOCHATZIS@ec.europa.eu, phone: +39 0332 78 9548) and João Alberto Lopes (Joao-Filipe.ALBERTO-LOPES@ec.europa.eu, phone: +390332789782) **1st March 2017**.

For further information, please contact Emmanouil Tsochatzis or João Alberto Lopes.

Sincerely yours.

Eddo Hoekstra
Operating Manager, European Union Reference Laboratory for Food Contact Materials

European Commission
DG Joint Research Centre
Directorate F. Health, Consumers and Reference Materials
Food and Feed Compliance Unit
Via E. Fermi 2749, T.P. 260
I-21027 Ispra (VA), Italy

Appendix I

General considerations
Use a calibrated thermometer/data logger (if possible) for food simulant D1 and oven temperature control.

A. Constant temperature with max fan operation

- Preheat the food simulant D1 at a temperature higher than the oven migration temperature in such way that it maintains the correct contact temperature in the filled test specimen.
- Fill the provided test specimens with 300 mL of preheated food simulant D1 outside the oven.
- If possible, cover the test specimen with a clock glass and put a small weight on top of it. This step is aiming at limiting the loss of food simulant due to evaporation.
- Put the filled test specimens into the thermostatic oven and monitor the **temperature of the food simulant inside the last test specimen** (i.e. the one filled last and/or placed into the thermostatic oven/incubator last).
- Use small intervals between the samples insert to the oven (e.g. 2 min).
- The time inserting the test specimens into the oven should be as short as possible, minimizing the temperature drop (5-10 s maximum) and equilibrating faster to the correct temperature conditions.

B. Constant temperature with no ventilation

- Preheat food simulant D1 at a temperature higher than the oven migration temperature.
- Fill the provided test specimens with 300 mL of preheated Food Simulant D1 outside the oven.
- If possible, cover the test specimen with a clock glass and put a small weight on top of it. This step is aiming at limiting the loss of food simulant due to evaporation.
- Put the filled test specimens into the thermostatic oven and monitor the temperature of the food simulant inside the last test specimen (i.e. the one filled last and/or placed into the thermostatic oven/incubator last).
- The time inserting one test specimen into the oven should be as short as possible, minimizing the temperature drop (5-10 s maximum) and equilibrating faster to the correct temperature conditions.
- Use longer time intervals between inserting test specimens into the oven (20-30 min) since the oven needs to equilibrate to the correct temperature conditions.

NOTE
The two procedures work for our oven but you need to verify whether these conditions are working for your oven.

Annex 6: Results reporting form

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F.5 Unit – Food and Feed Compliance

European Union Reference Laboratory for Food Contact Materials

Use this form to submit your results by entering data in the space provided and return it by **1st March 2017** (deadline) by e-mail to Emmanouil.Tsochatzis@ec.europa.eu or Joao-Filipe.ALBERTO-LOPES@ec.europa.eu

RESULTS REPORTING FORM – ILC 02-2016 Temperature Control (article filling) and specific migration- Follow-up 01-2015

LABORATORY CODE	

TEMPERATURE CONTROL

Temperature of the preheated food simulant just before article filling: ..... °C

Food simulant and thermostatic oven/incubator temperature monitored during the contact:

t [min]	T_{oven} [°C]	$T_{oven, display}$ [°C]	$T_{simulant}$ [°C]	Remarks
0				
1				
2				
3				
4				
5				
6				
7				
8				
9				
10				
15				
20				
25				
30				
35				
40				
45				
50				
55				
60				
65				
70				
75				
80				
85				
90				
95				
100				
105				
110				
115				
120				

where: t — exposure time; timer is started immediately after the 2nd test specimen is placed into the thermostatic oven/incubator
T_{oven} — temperature of the thermostatic oven/incubator, measured with a calibrated thermometer
$T_{oven, display}$ — temperature value shown at the display of the oven/incubator itself
$T_{simulant}$ — temperature of the food simulant inside the 3rd test specimen, measured with a calibrated thermometer

SPECIFIC MIGRATION

Perform the migration, by filling the provided polypropylene cups (volume 0.3 L) with food simulant D1 (ethanol 50%, v/v) for 2 h at 70°C.

Perform three replicates and report:
1. Three independent concentrations values for FCM substance No 500;
2. The concentration results would be based on the density of food simulant D1;
3. Report also the mean of your three measurement results with associated expanded uncertainty, the coverage factor and the approach followed for uncertainty calculation.

SAMPLE CODE	ANALYTE	REPLICATE 1* (µg/kg)	REPLICATE 2* (µg/kg)	REPLICATE 3* (µg/kg)	AVERAGE (µg/kg)	Expanded uncertainty (U)
	FCM 500					
Coverage factor "k"						
Approach used for uncertainty calculation						

*Two (2) number of decimals e.g. 0.01

PLACE AND DATE	LABORATORY MANAGER	SIGNATURE

Annex 7: Questionnaire form

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DIRECTORATE GENERAL JRC
JOINT RESEARCH CENTRE
Directorate F –Health, Consumers and Reference Materials
F.5 Unit – Food and Feed Compliance

EURL European Union Reference Laboratory for Food Contact Materials

QUESTIONNAIRE FORM

ILC02-2016: Temperature Control (article filling) and specific migration – Follow-up of ILC01-2015

To participate in the exercise, complete the form and return it until **1st March 2017** by e-mail to Emmanouil.TSOCHATZIS@ec.europa.eu or Joao-Filipe.ALBERTO-LOPES@ec.europa.eu

LABORATORY CODE:	

Please select your answer and fill in the required fields

A. MIGRATION CONDITION

PART I. Preheating of simulant, thermostatic oven and test specimen

1. Food simulant preheated portions (please include the volume; mL)

1. Single portion 2. Multiple portion 3. Other (please specify) (……mL)

2. Type of glassware used for preheating the food simulant

1. Glass Schott bottle 2. Erlenmeyer flask 3. Other (please specify)

3. Food simulant preheating:

1. Inside a thermostatic oven 2. Inside the same thermostatic oven 3. On a hot plate

4. Water bath 5. Other (please specify)

4. Food simulant preheating temperature (please fill also the temperature)

Constant oven temperature programs

1. $T_{simulant} < T_{oven}$ 2. $T_{simulant}=T_{oven}$ 3. $T_{simulant} > T_{oven}$ (……°C)

Gradient oven temperature programs

1. $T_{simulant} < T_{oven, min}$ 2. $T_{simulant}=T_{oven\ range}$ 3. $T_{simulant} > T_{oven, max}$ (……°C)

5. Time (t, min) of food simulant preheating

…… min

6. Verification of the desired food simulant temperature by:
1. Calibrated thermometer 2. Calibrated data logger 3. Non calibrated thermometer* 4. Other (please specify) * *In case of non-calibrated thermometer, please provide technical specifications (the one we supplied to you is not calibrated!)*: ---------- ----------
7. Way of measuring the desired food simulant temperature
1. Inside the food simulant portion 2. Inside a separate food simulant portion 3. Other (please specify) ……
8. Did you immerse the probe in the food simulant portion during the entire preheating phase OR did you immerse it at the end of the preheating phase?
9. Did you cover the food simulant portion which contained the probe and, if so, how did you do it?
10. Please insert here a picture of the experimental setting of preheating of the food simulant, if available.

11. Please provide details on the used thermostatic ovens/incubators

Model: ……
Dimensions: ……
Volume: ……
Electric power: ……
Temperature range: ……
Fan available: ……
Fan operation: ……
Level of fan operation: ……
Intervallic air exchange: ……

12. Temperature and time of thermostatic oven/incubator preheating

Time: …… min
Temperature: …… °C

13. Verification/control of preheated oven/incubator temperature?

1. YES 2. NO

If yes: 1. Calibrated thermometer 2. Calibrated data logger
3. Non calibrated thermometer* 4. Other (please specify)

* *In case of non-calibrated thermometer, please provide technical specifications (the one we supplied to you is not calibrated!):*

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11. Control of preheated oven/incubator temperature homogeneity?

1. YES 2. NO

If yes: 1. Calibrated thermometer 2. Calibrated data logger
3. Non calibrated thermometer* 4. Other (please specify)

* *In case of non-calibrated thermometer, please provide technical specifications (the one we supplied to you is not calibrated!):*

---

12. If you replied yes on question 11, how did you do it (e.g. several probes placed in different spots inside the thermostatic oven/incubator - close to the bottom/ceiling/side walls, in the centre,...)?

13. In case your thermostatic oven/incubator is equipped with a fan to enable air circulation inside, did you turn it on during the preheating phase? If so, at which level (low/medium/high)? Did you notice effects on the homogeneity of the temperature distribution inside the thermostatic oven/incubator?
1. YES 2. NO 1. Low 2. Medium 3. High *Notices/effect*: .....
14. In case your thermostatic oven/incubator provides the possibility for an automated intervallic exchange of air, did you enable it during the preheating phase? If so, at which frequency? Did you notice effects on the temperature constancy inside the oven/incubator?
1. YES 2. NO *If yes, please give details*: .....
15. Please add any other comments on the preheating of the thermostatic oven/incubator

PART II. Filling procedure

16. Test specimen preheating before filling
1. YES* 2. NO * *If yes:* ..... °C for ..... min (*time of preheating*) in ..... (*place of preheat; e.g. oven, …*)
17. Place of test specimen filling
1. Inside the oven 2. Outside the oven (lab/work bench)* * *If outside, please specify the distance:* ..... m from the thermostatic oven
18. Test specimen filling surface (either inside or outside)
1. Metal 2. Ceramic (insulated) 3. Ceramic (non-insulated) 4. Wood 5. Plastic 6. Other (please specify)

19. Test specimen insert into the oven
1. All at once 2. Sequential with time intervals* 3. Other (please specify) * *Intervals duration*: …… min
20. Filling time per test specimen (approximately, either inside or outside the oven)
…… min
21. In case of filling the specimen inside the oven, please provide the time that the ovens' door was opened
…… min
22. Test specimen covering
1. Aluminum foil 2. Clock glass 3. Petri dish 4. Silicon lid 5. Plastic lid 6. (Other please specify)
23. External insulation of the test specimen?
1. YES 2. NO (*If yes specify*:.....)
24. Number of persons involved in filling process
1. One 2. Two 3. Three 4. Other (please specify)
25. Please insert here a picture of the experimental setting for filling the test specimens, if available.

25. Please add any other comments regarding the filling procedure

PART III. Contact phase

26. Type of thermometer/data logger used
1. Calibrated thermometer 2. Calibrated data logger 3. Non calibrated thermometer* 4. Other (please specify) * *In case of non-calibrated thermometer, please provide technical specifications (the one we supplied to you is not calibrated!)*: ---------- ----------
27. Temperature control of the food simulant during migration
1. In the filled test specimen (1st, 2nd, 3rd replicate) 2. On a separate mode (e.g. glass bottle)
28. Simulant losses (mL) during migration
…… mL
29. Thermostatic migration oven program (please specify)
1. Constant oven programs 2. Gradient oven programs Temperature program: …… …… ……
30. Oven temperature verification during migration
1. YES 2. NO 1. Calibrated thermometer 2. Calibrated data logger 3. Non calibrated thermometer* 4. Other (please specify) * *In case of non-calibrated thermometer, please provide technical specifications (the one we supplied to you is not calibrated!)*: ---------- ----------
31. Migration oven internal fan operation (operation level)
1. YES* 2. NO **If yes*: 1. Low 2. Medium 3. High

32. Migration oven internal intervallic air exchange operation (operation level)
1. YES* 2. NO *If yes*: 1. Low 2. Medium 3. High
33. Load of the oven during migration
1. Low 2. Medium 3. High

PART IV. GENERAL COMMENTS

B. ANALYTICAL METHOD DESCRIPTION

Is the method validated?
Is the method accredited?
Did you analyse the sample according to an official method? If YES, please specify

EXPERIMENTAL PART

Sample amount used for analysis (mL) :	
Extraction solution used :	
Extraction procedure (please specify all the conditions used):	

Did you apply any special treatment to the samples provided? If YES, please specify	
Which analytical technique did you use?	
Please provide your method details? (e.g. injection vol., temperature program, eluents, etc..., if applicable)	
Does your laboratory carry out this type of analysis (same matrix, analytes) on a routine basis?	
Did you encounter any problems with sample analysis? If YES, please specify	
Other Comments	

Annex 8: Laboratory code letter

EUROPEAN COMMISSION
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Directorate F –Health, Consumers and Reference Materials
F.5 Unit – Food and Feed Compliance

European Union Reference Laboratory for Food Contact Materials

LABORATORY CODE

ILC 02 2016: Temperature Control (article filling) and specific migration-Follow-up 01-2015

LABORATORY NAME:	
LABORATORY CODE:	

Sincerely yours,

Eddo Hoekstra
Operating Manager, European Union Reference Laboratory for Food Contact Materials

European Commission
DG Joint Research Centre
Directorate F. Health, Consumers and Reference Materials
Food and Feed Compliance Unit
Via E. Fermi 2749, T.P. 260
I-21027 Ispra (VA), Italy

< Laboratory code>

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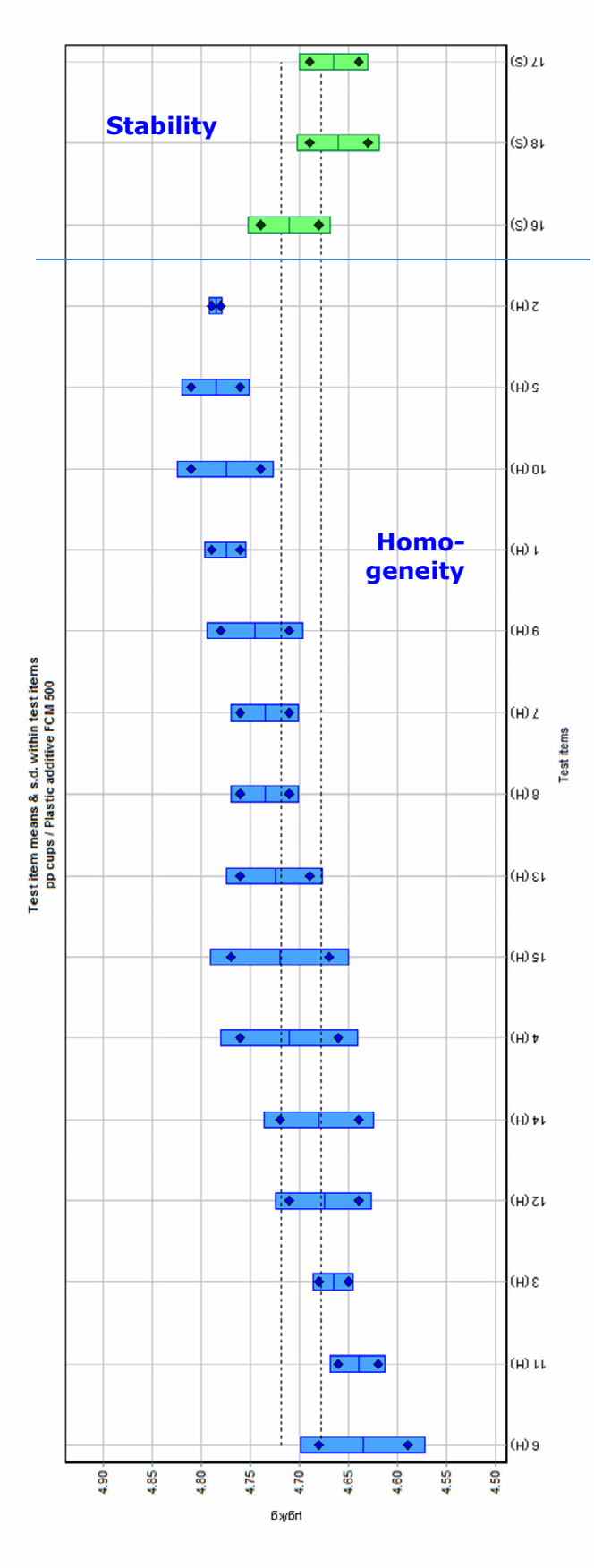
Annex 9: Homogeneity and stability



Analyte: **FCM No 500**

Mean (Homogeneity):	4.72 µg/kg
Mean (Stability):	4.68 µg/kg
RSD (Analytical):	1 %
RSD (Sample):	0.8 %
σ_{pt}:	31 %

[RSD: relative standard deviation]

<u>Homogeneity test</u> (*):

- ✓ adequate homogeneity: **Yes**
- ✓ significant heterogeneity: **No**
- ✓ F-test: **Passed**

<u>Stability test</u> (*):

- ✓ Stable: **Yes**
- ✓ T-test: **Passed**

(*) According to ISO 13528:2015

Annex 10: Temperature (T) results reported by participants

			Number of Temperature readouts			
Lab Code	**Mean T (°C)**	**Stdev (°C)**	**T < 67,5 °C**	**67,5 ≤ T ≤ 72,5 °C**	**T > 72,5 °C**	**Satisfactory**
LC0002	64.6	0.7	33	0	0	0%
LC0003	65.2	1.2	33	0	0	0%
LC0004	69.3	0.3	0	33	0	100%
LC0005	70.4	0.4	0	33	0	100%
LC0006	70.1	0.7	0	33	0	100%
LC0007	69.7	1.7	6	27	0	82%
LC0008	70.5	0.8	0	33	0	100%
LC0010	69.7	0.7	0	33	0	100%
LC0013	71.1	0.9	0	33	0	100%
LC0016	67.8	2.6	15	18	0	55%
LC0017	69.6	1.6	6	27	0	82%
LC0018	64.9	5.4	21	9	3	27%
LC0020	69.1	0.4	0	33	0	100%
LC0024	68.0	1.3	10	23	0	70%
LC0025	68.4	0.3	0	33	0	100%
LC0028	70.3	0.9	0	33	0	100%
LC0031	67.2	3.2	12	21	0	64%
LC0032	70.3	0.5	0	33	0	100%
LC0037	68.9	0.7	0	33	0	100%
LC0038	68.2	0.8	9	24	0	73%
LC0040	70.3	0.3	0	33	0	100%
LC0043	70.0	0.5	0	33	0	100%
LC0044	67.1	1.5	17	16	0	48%
LC0046	69.6	0.6	0	33	0	100%
LC0047	71.0	1.7	2	31	0	94%
LC0048	70.3	0.5	0	33	0	100%
LC0049	67.5	2.6	13	20	0	61%
LC0050	69.8	1.1	0	33	0	100%
LC0055	69.7	0.7	0	33	0	100%
LC0056	71.0	0.4	0	33	0	100%
LC0059	69.1	0.8	1	32	0	97%
LC0061	68.1	2.7	13	20	0	61%
LC0064	70.5	1.2	0	33	0	100%
LC0113	71.8	0.4	0	33	0	100%
LC0114	69.5	1.1	2	31	0	94%
LC0120	67.3	0.9	19	14	0	42%
LC0121	69.8	0.6	0	33	0	100%
LC0123	69.6	0.7	0	33	0	100%
LC0124	69.8	0.7	0	33	0	100%
LC0125	71.3	1.4	0	28	5	85%
LC0126	68.9	0.2	0	33	0	100%

* Satisfactory according to EN13130-1:2004: $67,5\ °C \leq \text{mean}(T) \leq 72,5\ °C$

Annex 11: Results for FCM No 500 after migration

Assigned values: x_{pt} = 5.63; $U(x_{pt})$ (k = 2.0) = 0.79; σ_{pt} = 1.72 (all values in µg kg^{-1})

	Lab Code	x_i	±	k	Technique	$u(x_i)$	z score	ζ score	MU
1	*LC0002	5.53	4.98	3	HPLC-FLD	1.66	-0.1	-0.1	a
2	LC0004	4.68	0.12	2	UPLC-FLD	0.06	-0.6	-2.4	b
3	LC0005	6.58	1.00	2	HPLC-FLD	0.50	0.6	1.5	a
4	LC0006	5.32	3.80	2	HPLC-FLD	1.90	-0.2	-0.2	c
5	LC0007	0.11	0.06	2	HPLC-FLD	0.03	-3.2	-14.0	b
6	LC0010	4.93	4.39	3.18	HPLC-DAD	1.38	-0.4	-0.5	a
7	LC0013	5.37	2.14	2	HPLC-UV	1.07	-0.2	-0.2	a
8	*LC0016	23.33	-	-	HPLC-FLD	0.00	10.3	45.1	b
9	LC0017	5.01	2.42	2	HPLC-FLD	1.21	-0.4	-0.5	a
10	*LC0018	3.09	0.90	2	HPLC-FLD	0.45	-1.5	-4.3	a
11	LC0024	3.53	5.28	2	HPLC-FLD	2.64	-1.2	-0.8	c
12	LC0025	5.22	-	-	HPLC-FLD	0.00	-0.2	-1.0	b
13	LC0028	5.08	0.40	2	LC-MS/MS	0.20	-0.3	-1.2	b
14	*LC0031	4.08	0.06	2	HPLC-FLD	0.03	-0.9	-3.9	b
15	LC0032	5.33	0.86	2	HPLC-FLD	0.43	-0.2	-0.5	a
16	LC0037	4.46	1.28	2	HPLC-FLD	0.64	-0.7	-1.6	a
17	LC0038	27.10	10.38	1.73	HPLC-UV	6.00	12.5	3.6	c
18	LC0043	5.00	0.40	2	LC-MS/MS	0.20	-0.4	-1.4	b
19	*LC0044	5.71	0.43	1.73	HPLC-FLD	0.25	0.0	0.2	b
20	LC0046	5.05	1.69	3.18	HPLC-FLD	0.53	-0.3	-0.9	a
21	LC0047	6.23	1.50	2	LC-MS	0.75	0.3	0.7	a
22	LC0048	10.57	12.04	2	LC-MS/MS	6.02	2.9	0.8	c
23	*LC0049	5.16	-	-	GC-MS	0.00	-0.3	-1.2	b
24	LC0050	7.20	8.24	2	HPLC-FLD	4.12	0.9	0.4	c
25	LC0055	4.94	1.44	2	LC-MS/MS	0.72	-0.4	-0.8	a
26	LC0056	5.52	0.88	2	HPLC-FLD	0.44	-0.1	-0.2	a
27	LC0061	4.47	-	-	-	0.00	-0.7	-3.0	b
28	LC0064	7.37	0.92	2	UPLC-qTOF-MS	0.46	1.0	2.9	a
29	LC0113	31.15	20.00	2	GC-MS	10.00	14.8	2.6	c
30	LC0114	10.43	2.08	2	HPLC-FLD	1.04	2.8	4.3	a

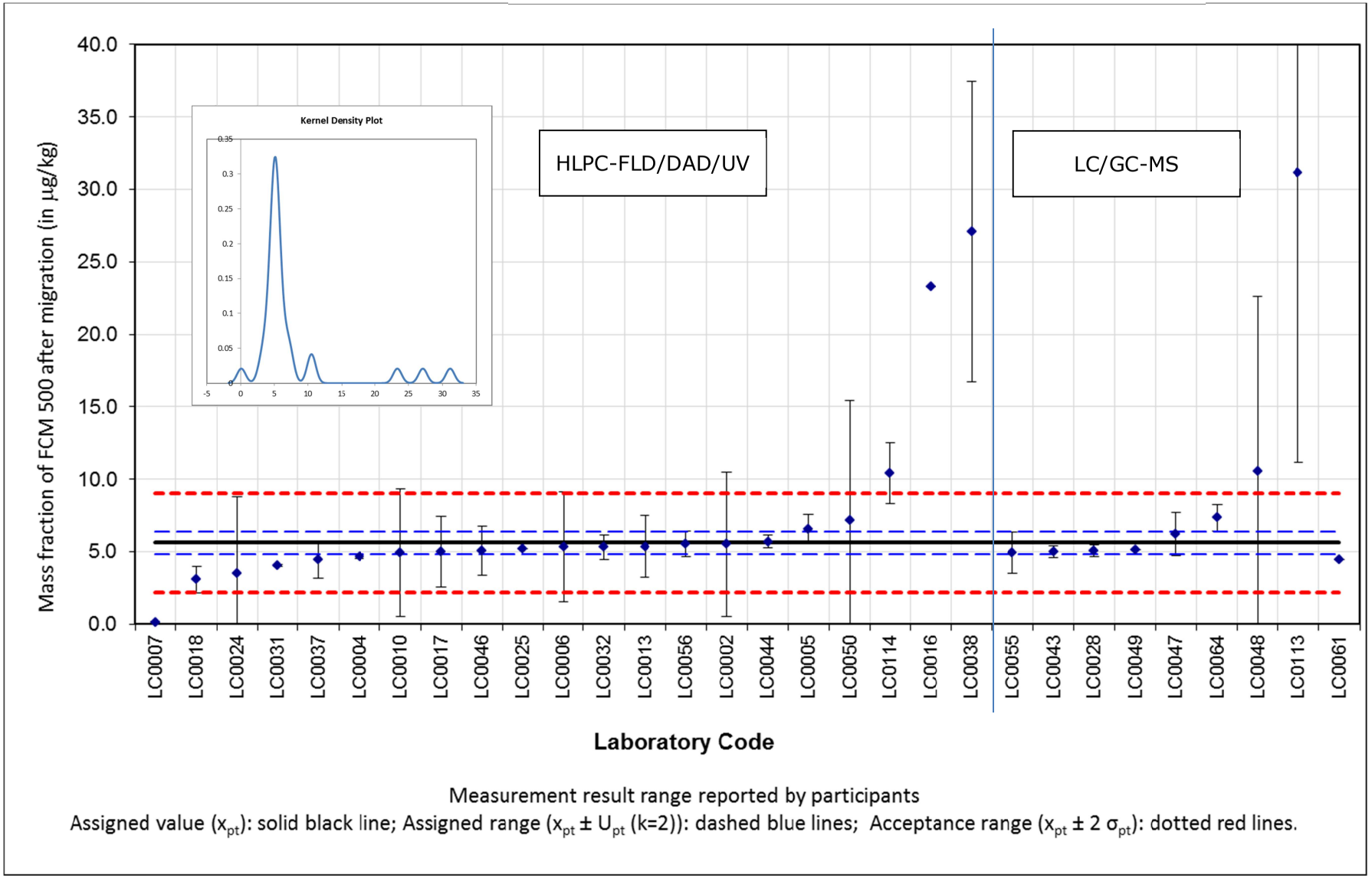
*: Mean T < 68 °C

Performance: satisfactory, questionable, unsatisfactory,

MU evaluation - a : $u_{min}(u(x_{pt})) \leq u_i \leq u_{max}(\sigma_{pt})$; b : $u_i < u(x_{pt})$; and c : $u_i > \sigma_{pt}$

EURL-FCM-02-2016: FCM No 500 in D1

x_{pt} = 5.63; U(x_{pt}) = 0.79 (k=2); σ_{pt} = 1.72 (µg kg^{-1})



Measurement result range reported by participants

Assigned value (x_{pt}): solid black line; Assigned range ($x_{pt} \pm U_{pt}$ (k=2)): dashed blue lines; Acceptance range ($x_{pt} \pm 2\ \sigma_{pt}$): dotted red lines.

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KJ-NA-29121-EN-N

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doi:10.2760/940236
ISBN 978-92-79-79934-1