Evaluation Report
proficiency test

DLA ptAU07 (2020)

Ethanol in Marzipan

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Allgemeine Informationen zur Eignungsprüfung (EP)
General Information on the proficiency test (PT)

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EP-Koordinator *PT-Coordinator*	Dr. Matthias Besler-Scharf
Status des EP-Bericht *Status of PT-Report*	Abschlussbericht / Final report (23 November 2020) Gültig ist die jeweils letzte Version/Korrektur des Berichts. Sie ersetzt alle vorangegangenen Versionen. Only the latest version/correction of the report is valid. It replaces all preceding versions.
EP-Bericht Freigabe *PT-Report Authorization*	Dr. Matthias Besler-Scharf (Technischer Leiter / Technical Manager) *- gezeichnet / signed M. Besler-Scharf* Alexandra Scharf MSc. (QM-Beauftragte / Quality Manager) *- gezeichnet / signed A. Scharf* Datum / Date: 23 November 2020
Unteraufträge *Subcontractors*	Im Rahmen dieser Eignungsprüfung wurden nachstehende Leistungen im Unterauftrag vergeben: Keine As part of the present proficiency test the following services were subcontracted: none
Vertraulichkeit *Confidentiality*	Die Teilnehmerergebnisse sind im EP-Bericht in anonymisierter Form mit Auswertenummern benannt. Daten einzelner Teilnehmer werden ausschließlich nach vorheriger Zustimmung des Teilnehmers an Dritte weitergegeben. Participant result are named anonymously with evaluation numbers in the PT report. Data of individual participants will be passed on to third parties only with prior consent of the participant.

Contents

1. Introduction

The participation in proficiency testing schemes is an essential element of the quality-management-system of every laboratory testing food and feed, cosmetics and food contact materials. The implementation of proficiency tests enables the participating laboratories to prove their own analytical competence under realistic conditions. At the same time they receive valuable data regarding the verification and/or validation of the particular testing method [1, 5].

The purpose of DLA is to offer proficiency tests for selected parameters in concentrations with practical relevance.

Realisation and evaluation of the present proficiency test follows the technical requirements of DIN EN ISO/IEC 17043 (2010) and DIN ISO 13528:2009 / ISO 13528:2015 [2, 3].

2. Realisation

2.1 Test material

The test material is a common in commerce food from a pre-pack (marzipan praline with dark chocolate and marzipan filling) from a European supplier. According to the declaration, the product contains the ingredient ethanol.

The composition of the PT samples is shown in Table 1.

Table 1: Composition of DLA-Samples

PT-Sample
Marzipan (71%) with Dark Chocolate Coating   Ingredients: Almonds, sugar, cocoa mass, cocoa butter, whole milk powder, invert sugar syrup, emulsifier: soy lecithins, vanilla extract, alcohol

Note: The metrological traceability of temperature, mass and volume during production of the PT samples is ensured by DAkkS calibrated reference materials.

Afterwards two pralines (approximately 24 g) were sealed in vacuum bags. And the samples were chronologically numbered.

2.1.1 Homogeneity

The **homogeneity** of the PT samples was examined 6-fold by enzymic UV-test (mod. ASU §64 LFGB L 40.00-12). The repeatability standard deviation was 5,5% and thus within the range of repeatability standard deviations of the ASU method (s. 3.6.2), which are in the range of 1,9% to 10,8% [18]. The results of homogeneity analysis are given in the documentation.

The calculation of the **repeatability standard deviations S_r of the participants** was used as an indicator of homogeneity. It is 5,5% for the parameter ethanol. Thus it is similar to corresponding repeatability standard deviations of precision data of the standardized methods (e.g. ASU §64 LFGB L 40.00-12, s. 3.6.2) (see Table 2) [18].
The repeatability standard deviations of the participants' results are given in the statistic data (see 4.1).

Furthermore, the homogeneity was graphically characterized for information by the **trend line function of participants' results for chronological numbered single samples** (s. 5.2.2 Homogeneity).

In case the criterion for sufficient homogeneity of the test items is not fulfilled the impact on the target standard deviation will be verified. If necessary the evaluation of results will be done considering the standard uncertainty of the assigned value by z'-scores (s. 3.8 and 3.11) [3].

2.1.2 Stability

According to the manufacturer the shelf life is > 6 months for the product when stored at room temperature. Experience has shown that the storage stability or shelf life of the samples additionally sealed under vacuum in a gas-tight film bag is thus guaranteed during the investigation period under the specified storage conditions at a temperature of < 10°C.

2.2 Sample shipment and information to the test

Two portions of test material were sent to every participating laboratory in the 26th week of 2020. The testing method was optional. The tests should be finished at 4th September 2020 the latest.

With the cover letter along with the sample shipment the following information was given to participants:

The two portions contain identical samples with the parameter Ethanol in the matrix of marzipan with chocolate coating. The ethanol content must be determined in the marzipan portion. The analysis method is optional.

Note: please store the samples at 2-10 °C on arrival

Please note the attached information on the proficiency test.
(see documentation, section 5.3 Information on the PT)

2.3 Submission of results

The participants submitted their results in standard forms, which have been handed out with the samples (by email).
The finally calculated concentrations of the parameter as average of duplicate determinations of both numbered samples were used for the statistical evaluation. For the calculation of the repeatability- and reproducibility standard deviation the single values of the double determination were used.
Queried and documented were single results, recovery and the used testing methods.
In case participants submitted several results for the same parameter obtained by different methods these results were evaluated with the same evaluation number with a letter as a suffix and indication of the related method.

All 10 participants submitted their results in time.

3. Evaluation

3.1 Consensus value from participants (assigned value)

The robust mean of the submitted results was used as assigned value (X_{pt}) („consensus value from participants") providing a normal distribution. The calculation was done according to algorithm A as described in annex C of ISO 13528 [3]. If there are < 12 quantitative results and an increased difference between robust mean and median, the median may be used as the assigned value (criterion: Δ median - rob. mean > 0,3 σ_{pt}) [3].

The condition is that the majority of the participants' results show a normal distribution or are distributed unimodal and symmetrically. To this end, an examination of the distribution is carried out, inter alia, using the kernel density estimate [3, 12].

In case there are indications for sources of higher variability such as a bimodal distribution of results, a cause analysis is performed. Frequently different analytical methods may cause an anomaly in results' distribution. If this is the case, separate evaluations with own assigned values (X_{pti}) are made whenever possible.

The statistical evaluation is carried out for all the parameters for a minimum of 7 values are present, in justified cases, an evaluation may also be carried out from 5 results onwards.

The actual measurement results will be drafted. Individual results, which are outside the specified measurement range of the participating laboratory (for example with the result > 25 mg/kg or < 2,5 mg/kg) or the indicating "0" will not be considered for the statistic evaluation [3].

3.2 Robust standard deviation

For comparison to the target standard deviation σ_{pt} (standard deviation for proficiency assessment) a robust standard deviation (S*) was calculated. The calculation was done according to algorithm A as described in annex C of ISO 13528 [3].

3.3 Repeatability standard deviation

The repeatability standard deviation S_r is based on the laboratory´s standard deviation of (outlier free) individual participant results, each under repeatability conditions, that means analyses was performed on the same sample by the same operator using the same equipment in the same laboratory within a short time. It characterizes the mean deviation of the results within the laboratories [3] and is used by DLA as an indication of the homogeneity of the sample material.

In case single results from participants are available the calculation of the repeatability standard deviation S_r, also known as standard deviation within laboratories S_w, is performed by: [3, 4].

The relative repeatability standard deviation as a percentage of the mean value is indicated as coefficient of variation CV_r in the table of stat-

istical characteristics in the results section in case single results from participants are available.

3.4 Reproducibility standard deviation

The reproducibility standard deviation S_R represents a inter-laboratory estimate of the standard deviation for the determination of each parameter on the bases of (outlier free) individual participant results. It takes into account both the repeatability standard deviation S_r and the within-laboratory standard deviation S_s. Reproducibility standard deviations of PT´s may differ from reproducibility standard deviations of ring trials, because the participating laboratories of a PT generally use different internal conditions and methods for determining the measured values.

In the present evaluation, the specification of the reproducibility standard deviation, therefore, does not refer to a specific method, but characterizes approximately the comparability of results between the laboratories, assumed the effect of homogeneity and stability of the sample are negligible.

In case single results from participants are available the calculation of the reproducibility standard deviation S_R is performed by: [3, 4].

The relative reproducibility standard deviation CV_R in percent of the mean is given as variation coefficient in the statistical data of participant for each parameter. The significance of CV_R is further explained in section 3.9.

3.5 Exclusion of results and outliers

Before statistical evaluation obvious blunders, such as those with incorrect units, decimal point errors, too few significant digits (valid digits) or results for another proficiency test item can be removed from the data set [2]. Even if a result e.g. with a factor >10 deviates significantly from the mean and has an influence on the robust statistics, a result of the statistical evaluation can be excluded [3].

All results should be given at least with 2 significant digits. Specifying 3 significant digits is usually sufficient.

Results obtained by different analytical methods causing an increased variability and/or a bi- or multimodal distribution of results, are treated separately or could be excluded in case of too few numbers of results. For this results are checked by kernel density estimation [3, 12].

Results are tested for outliers by the use of robust statistics (algorithm A): If a value deviates from the robust mean by more than 3 times the robust standard deviation, it can be classified as an outlier (see above) [3]. Due to the use of robust statistics outliers are not excluded, provided that no other reasons are present [3]. Detected outliers are only mentioned in the results section, if they have been excluded from the statistical evaluation.

3.6 Target standard deviation (for proficiency assessment)

The target standard deviation of the assigned value σ_{pt} (= standard deviation for proficiency assessment) can be determined according to the following methods.

If an acceptable quotient S^*/σ_{pt} is present, the target standard deviation of the general model by Horwitz is preferably used for the proficiency assessment. It is usually suitable for evaluation of interlaboratory studies, where different methods are applied by the participants. On the other hand the target standard deviation from the evaluation of precision data of an precision experiment is derived from collaborative studies with specified analytical methods.

In cases where both above-mentioned models are not suitable, the target standard deviation is determined based on values by perception, see under 3.6.3.

For information, the z-scores of both models are given in the evaluation, if available.

For valuation of <u>ethanol</u> the target standard deviation of the evaluation by a precision experiment (s. 3.6.2) was applied in the present PT (German official method ASU §64 L 40.00-12).
Additionally the standard uncertainty was considered evaluating the results by z'-scores (s. 3.6.8). The reason for the relatively high statistical uncertainty could be the use of different methods (enzymatic methods, head-space, destillation, GC/MS, GC/FID).

In addition, the target standard deviation according to the general model of Horwitz (see 3.6.1) was given for information.

3.6.1 General model (Horwitz)

Based on statistical characteristics obtained in numerous PTs for different parameters and methods Horwitz has derived a general model for estimating the reproducibility standard deviation σ_R [6]. Later the model was modified by Thompson for certain concentration ranges [10]. The reproducibility standard deviation σ_R can be applied as the relative target standard deviation σ_{pt} in % of the assigned values and calculated according to the following equations [3]. For this the assigned value X_{pt} is used for the concentration c.

Equations	Range of concentrations	corresponds to
$\sigma_R = 0{,}22c$	$c < 1{,}2 \times 10^{-7}$	< 120 µg/kg
$\sigma_R = 0{,}02c^{0,8495}$	$1{,}2 \times 10^{-7} \leq c \leq 0{,}138$	≥ 120 µg/kg
$\sigma_R = 0{,}01c^{0,5}$	$c > 0{,}138$	$> 13{,}8$ g/100g

with c = mass content of analyte (as relative size, e.g. 1 mg/kg = 1 ppm = 10^{-6} kg/kg)

3.6.2 Value by precision experiment

Using the reproducibility standard deviation σ_R and the repeatability standard deviation σ_r of a precision experiment (collaborative trial or proficiency test) the target standard deviation σ_{pt} can be derived considering the number of replicate measurements m of participants in the present PT [3]:

$$\sigma_{pt} = \sqrt{\sigma_R^2 - \sigma_r^2 \left(m - 1 \, / \, m \right)}$$

The relative repeatability standard deviations (RSD_r) and relative reproducibility standard deviation (RSD_R) given in Table 2 were determined in ring tests using the indicated methods.
The resulting target standard deviations σ_{pt}, which were identified there, were used to evaluate the results and to provide additional information for the statistical data.

Table 2: Relative repeatability standard deviations (RSD_r) and relative reproducibility standard deviations (RSD_R) according to selected evaluations of tests for precision and the resulting target standard deviation σ_{pt} [18]

Parameter	Matrix	Mean	RSD_r	RSD_R	σ_{pt}	Method / Literature
Ethanol	Honey	0,00046	10,8%	13,2%	10,8%	Enzymatic [18]
Ethanol	Honey	0,0035	2,4%	5,8%	5,55%	Enzymatic [18]
Ethanol	Honey	0,0151	1,9%	7,8%	7,68%[1]	Enzymatic [18]

[1] used for evaluation or given for information (s. chapter 4)

3.6.3 Value by perception

The target standard deviation for proficiency assessment can be set at a value that corresponds to the level of performance that the coordinator would wish laboratories to be able to achieve [3].

For the present evaluation the target standard deviation according to 3.6.2 was regarded suitable.

Table 3 shows selected statistic data of participants results of present PT compared to PT results of previous years.

Tabelle 3: Characteristics of the present PT (on grey) in comparison to the previous PT from 2016 (SD = standard deviation, CV = coefficient of variation)

Parameter	Matrix	rob. Mean	rob. SD (S*)	rel. SD (VK$_{S*}$) [%]	Quotient S*/σ$_{pt}$	DLA Report
Ethanol	Marzipan	0,667	0,112	16,8	2,4[1]	DLA 39-2016
Ethanol	Marzipan	0,620[2]	0,134	21,6	1,9[1]	ptAU07 (2020)

[1] calculated by z'-Score
[2] median (applied for evaluation)

3.7 z-Score

To assess the results of the participants the z-score is used. It indicates about which multiple of the target standard deviation (σ_{pt}) the result (x_i) of the participant is deviating from the assigned value (X_{pt}) [3].
Participants' z-scores are derived from:

$$z_i = \frac{\left(x_i - x_{pt}\right)}{\sigma_{pt}}$$

The requirements for the analytical performance are generally considered as fulfilled if

$$-2 \leq z \leq 2 \ .$$

The valid z-Score for each parameter is indicated as z-Score (σ_{pt}). The value indicated as z-Score (Info) only obtains a informative character. The both z-Scores were calculated with the different target standard deviations in accordance with 3.6.

3.7.1 Warning and action signals

In accordance with the norm ISO 13528 it is recommended that a result that gives rise to a z-score above 3,0 or below −3,0, shall be considered to give an "action signal" [3]. Likewise, a z-score above 2,0 or below −2,0 shall be considered to give a "warning signal". A single "action signal", or "warning signal" in two successive PT-rounds, shall be taken as evidence that an anomaly has occurred which requires investigation.
An error or cause analysis can be carried out by checking the analysis process including understanding and implementation of the measurement by the staff, details of the measurement procedure, calibration of equipment and composition of reagents, transmission error or an error in the calculation, in the trueness and precision and use of reference material. If necessary, the problems must be addressed through appropriate corrective action [3].
In the figures of z-scores DLA gives the limits of warning and action

signals as yellow and red lines respectively. According to ISO 13528 the signals are valid only in case of a number of ≥ 10 results [3].

3.8 z'-Score

The z'-score can be used for the valuation of the results of the
participants, in cases the standard uncertainty has to be considered (s.
3.11). The z'-score represents the relation of the deviation of the
result (x_i) of the participant from the respective consensus value (X) to
the square root of quadrat sum of the target standard deviation (σ_{pt}) and
the standard uncertainty ($U_{(Xpt)}$) [3].

The calculation is performed by:

$$z_i' = \frac{x_i - x_{pt}}{\sqrt{\sigma_{pt}^2 + u_{(x_{pt})}^2}}$$

If carried out an evaluation of the results by means of z 'score, we have
defined below the expression in the denominator as a target standard
deviation σ_{pt}'.

The requirements for the analytical performance are generally considered
as fulfilled if

$$-2 \leq z' \leq 2 \, .$$

For warning and action signals see 3.7.1.

3.9 Reproducibility cofficient of variation (CV_R)

The variation coefficient (CV_R) of the reproducibility (= *relative
reproducibility standard deviation*) is calculated from the standard
deviation and the mean as follows [4, 13]:

$$CV_R = \frac{S_R * 100}{X}$$

In contrast to the standard deviation as a measure of the absolute varia-
bility the CV gives the relative variability within a data region. While
a low CV, e.g. <5-10% can be taken as evidence for a homogeneous set of
results, a CV of more than 50% indicates a "strong inhomogeneity of
statistical mass", so that the suitability for certain applications such
as the assessment of exceeded maximum levels or the performance evalu-
ation of the participating laboratories possibly can not be done [3].

3.10 Quotient S*/σ_{pt}

Following the HorRat-value the results of a proficiency-test (PT) can be considered convincing, if the quotient of robust standard deviation S* and target standard deviation σ_{pt} does not exceed the value of 2.
A value > 2 means an insufficient precision, i.e. the analytical method is too variable, or the variation between the test participants is higher than estimated. Thus the comparability of the results is not given [3].

3.11 Standard uncertainty of the assigned value

Every assigned value has a standard uncertainty that depends on the analytical method, differences between the analytical methods used, the test material, the number of participating laboratories (P) and on other factors. The standard uncertainty ($U_{(Xpt)}$) for this PT is calculated as follows [3]:

$$u_{(x_{pt})} = 1,25 \times \frac{s^*}{\sqrt{p}}$$

If $U_{(Xpt)} \leq 0,3$ σ_{pt} the standard uncertainty of the assigned value needs not to be included in the interpretation of the results of the PT [3]. Values exceeding 0,3 imply, that the target standard deviation could be too low with respect to the standard uncertainty of the assigned value.

The traceability of the assigned value is ensured on the basis of the consensus value as a robust mean of the participant results.

4. Results

All following tables are anonymized. With the delivering of the evaluation report the participants are informed about their individual evaluation number.

In the first table the characteristics are listed:

Statistic Data
Number of results
Number of outliers
Mean
Median
Robust mean(X_{pt})
Robust standard deviation (S*)
Number with m replicate measurements
Repeatability standard deviation (S_r)
Coefficient of Variation (CV_r)in %
Reproducibility standard deviation (S_R)
Coefficient of Variation (CV_R)in %
Target range:
Target standard deviation σ_{pt} or σ_{pt}'
Target standard deviation for information
lower limit of target range $(X_{pt} - 2\sigma_{pt})$ or $(X_{pt} - 2\sigma_{pt}')$ *
upper limit of target range $(X_{pt} + 2\sigma_{pt})$ or $(X_{pt} + 2\sigma_{pt}')$ *
Quotient S/σ_{pt} or S*/σ_{pt}'*
Standard uncertainty U(X_{pt})
Number of results in the target range
Percent in the target range

* Target range is calculated with z-score or z'-score

In the table below, the results of the participating laboratories are formatted in 3 valid digits**:

Auswerte-nummer	Parameter	Abweichung	z-Score	z-Score	Hinweis
Evaluation number	[Einheit / Unit]	Deviation	σ_{pt}	(Info)	Remark

** *In the documentation part, the results are given as they were transmitted by the participants.*

4.1 Ethanol in g/100g

Vergleichsuntersuchung / Proficiency Test

Statistic Data	
Number of results	11
Number of outliers	0
Mean	0,568
Robust Mean	0,592
Median (X_{pt})	**0,620**
Robust standard deviation (S*)	**0,134**
Number with 2 replicates	8
Repeatability SD (S_r)	0,0334
Repeatability (CV_r)	5,48%
Reproducibility SD (S_R)	0,150
Reproducibility (CV_R)	24,7%
Target range:	
Target standard deviation σ_{pt}'	**0,0694**
Target standard deviation (for Information)	0,0267
lower limit of target range	**0,481**
upper limit of target range	**0,759**
Quotient S*/σ_{pt}'	*1,9*
Standard uncertainty U(X_{pt})	*0,0505*
Results in the target range	*8*
Percent in the target range	*73%*

Comments:

The median was applied as the assigned value.

The target standard deviation was calculated using data from a precision experiment (ASU §64 L 40.00-12)(3.6.2). Additionally the target standard deviation according to the model of Horwitz (s. 3.6.1) was given for in-formation.

The distribution of results showed a slightly increased variability. The quotient S*/σ_{pt} was > 2,0. Therefore the evaluation was done by z'-scores considering the standard uncertainty. The quotient S*/σ_{pt}' was below 2,0 then. The robust standard deviation was in the range of previous PTs (see 3.6.3). The comparability of results is given.

The repeatability standard deviation was in the range of established values for the used determination methods (s. 3.6.2).

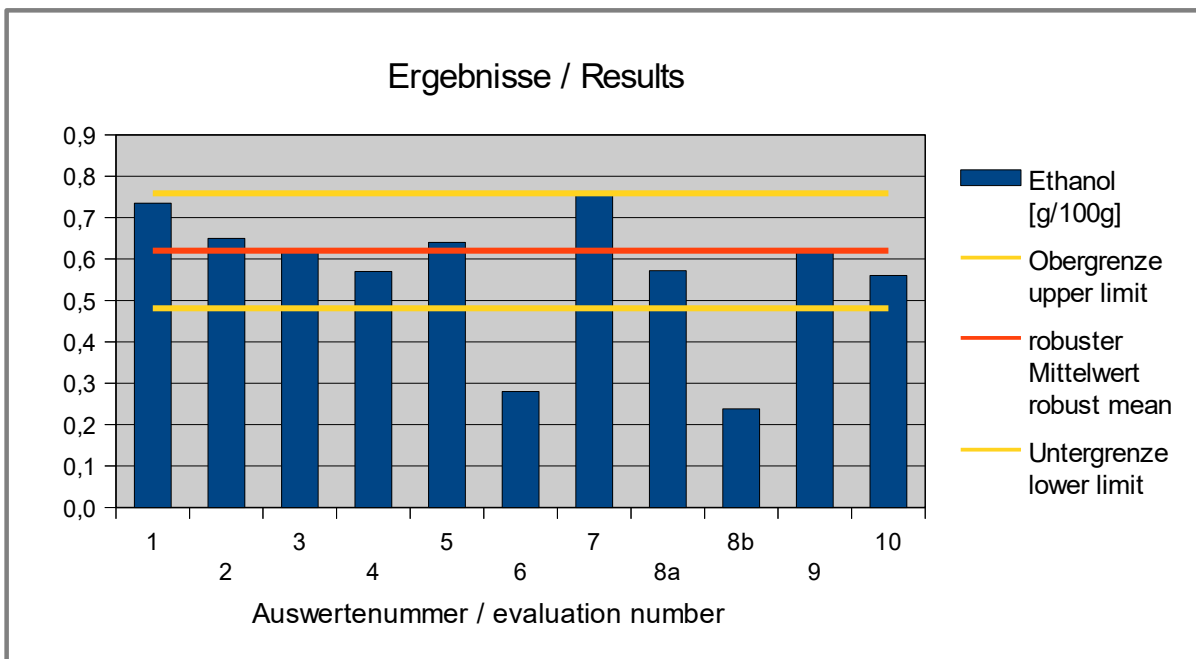*73% of results were in the target range.*

Abb. / Fig. 1: Ergebnisse **/** Results Ethanol

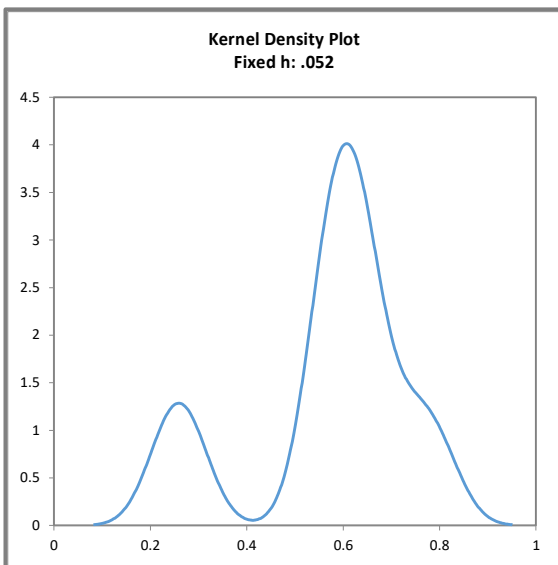**Abb. / Fig. 2:**
Kerndichte-Schätzung der Ergebnisse
(mit h = 0,75 x σ_{pt} von Xpt)

Kernel density plot of results
(with h = 0,75 x σ_{pt} of Xpt)

Comment:
The kernel density shows almost a symmetrical distribution of results with a slight shoulder at approx. 0,8 g/100g and a side peak at approx. 0,25 g/100g, due to two participants' results outside the target range.

Ergebnisse der teilnehmenden Institute:
Results of Participants:

Auswerte-nummer / Evaluation number	Ethanol [g/100g]	Abweichung [g/100g] / Deviation [g/100g]	z'-Score (σ_{pt})	z-Score (Info)	Hinweis / Remark
1	0,735	0,115	1,7	4,3	
2	0,650	0,030	0,43	1,1	
3	0,620	0,000	0,00	0,00	
4	0,570	−0,050	−0,72	−1,9	
5	0,640	0,020	0,29	0,75	
6	0,280	−0,340	−4,9	−13	
7	0,763 *	0,143	2,1	5,4	result without recovery rate
8a	0,572	−0,048	−0,69	−1,8	single result
8b	0,238	−0,382	−5,5	−14	single result
9	0,620	0,000	0,00	0,00	
10	0,560	−0,060	−0,86	−2,3	single result

* calculated by DLA
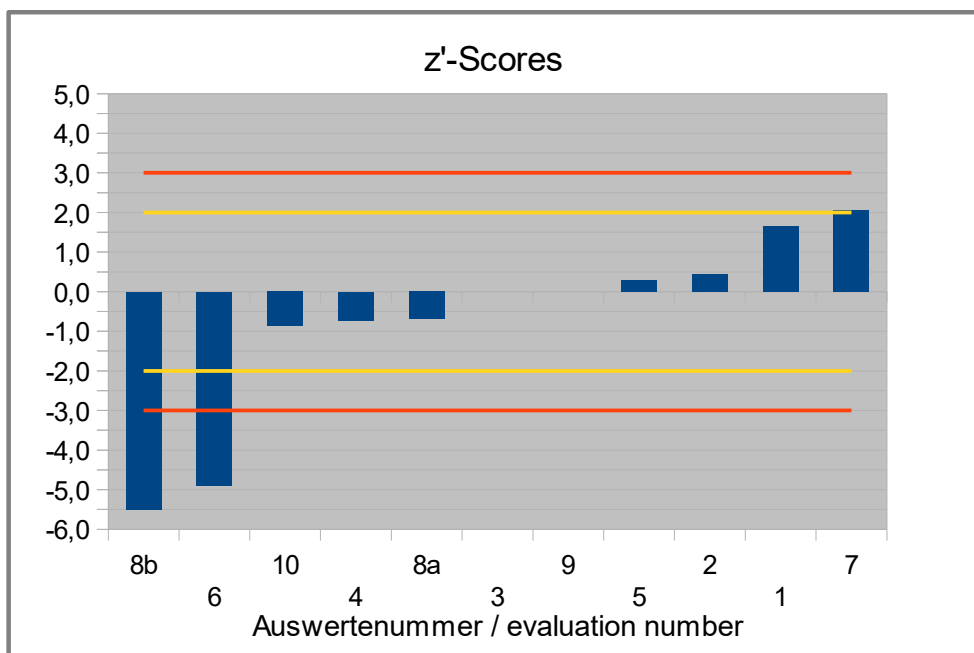
Abb. / Fig. 3:   z'-Scores Ethanol

5. Documentation

5.1 Details by the participants

Note: Information given in German were translated by DLA to the best of our knowledge (without guarantee of correctness).

5.1.1 Primary Data

Partici-pant	Unit	Sample I DLA No.	Sample II DLA No.	Date of analysis	Result (Mean)	Result I	Result II	Limit of quantification	Incl. Recove-ry rate	Recovery rate
				Day/Month					yes / no	in %
1	g/100g	9	37	07.07.20	0,735	0,731	0,738	0,05	no	-
2	g/100g	10	36	28.07. 29.07.	0,65	0,65	0,65	0,14	no	103
3	g/100g	2	44	27.08.20	0,62	0,61	0,62	0,009	no	
4	g/100g	21	25	01.09.20	0,57	0,63	0,53	0,01	no	
5	g/100g	16	30	01.07.20	0,64	0,65	0,62	0,001	no	-
6	g/100g	No. 8	No. 38	17.08.20	0,28	0,26	0,29	0,01	No	85
7	g/100g	15	31	14.08.20	0,795	0,77	0,82	0,1	yes	96
8	g/100g	20	26	26.08.20	0,405	0,572	0,238			
9	g/100g	18	28	04.09.20	0,62	0,59	0,65	0,01	no	
10	g/100g	24		01.07.20	0,56	0,56			No	N/A

5.1.2 Analytical Methods

| Partici-pant	Method description As in test report / norm / lite-rature	Sample preparation and proces-sing	Measuring me-thod	Calibration / Refe-rence material	Recovery rate with same matrix	Method Accredited ISO/IEC 17025	Further Remarks
					ja / nein	ja / nein	
1	ASU L 40.00-12 (Enzymatic Determination)	aqueous extr. and carrez precipitation, according to method no. 307.1 SLMB	Enzymatic Method			yes	
2	Enzymatic assay, r-biopharm AG 10 176 290 035, 2017-08 LOT: 45180000				no	yes	
3	yes	Samples extracted with water, precipitation and an aliquot taken for measurement.	GC- Headspace -Method	external calibration, no reference material	no	yes	
4	enzymatic assay	aqueous extraction, precipitation by carrez				yes	
5	in-house method	Headspace	GC-FID	3-Point-Calibration; REF-Material: ice tea	-	yes	-
6	ref. AOAC 2016.12		Headspace-GC-MS		Yes	No	
7	Internal Method MP.0797.R0.2020	Evaluation of amount of marzipan and complete homogenization	distillation and GC-FID determination	External calibration/ surrogate 1 pentanol/std ethanol	96	yes	
8						Yes	different results noted
9						yes	
10	Enzymatic assay				N/A	No	

5.2 Homogeneity

5.2.1 Homogeneity of PT-samples

Homogeneity of ethanol was tested by enzymatic assay (Megazyme K-ETOH / mod. according to ASU §64 L 40.00-12):

Independant Samples	g/100g
1	0,785
2	0,797
3	0,898
4	0,863
5	0,793
6	0,839

General Mean	0,829	
Repeatability standard deviation	0,045	5,5%

5.2.2 Trend line function of the participants results

By comparison of the increasing sample numbers and the measurement results of participants, the homogeneity of the chronological numbered PT items can be shown by the trend line for information:
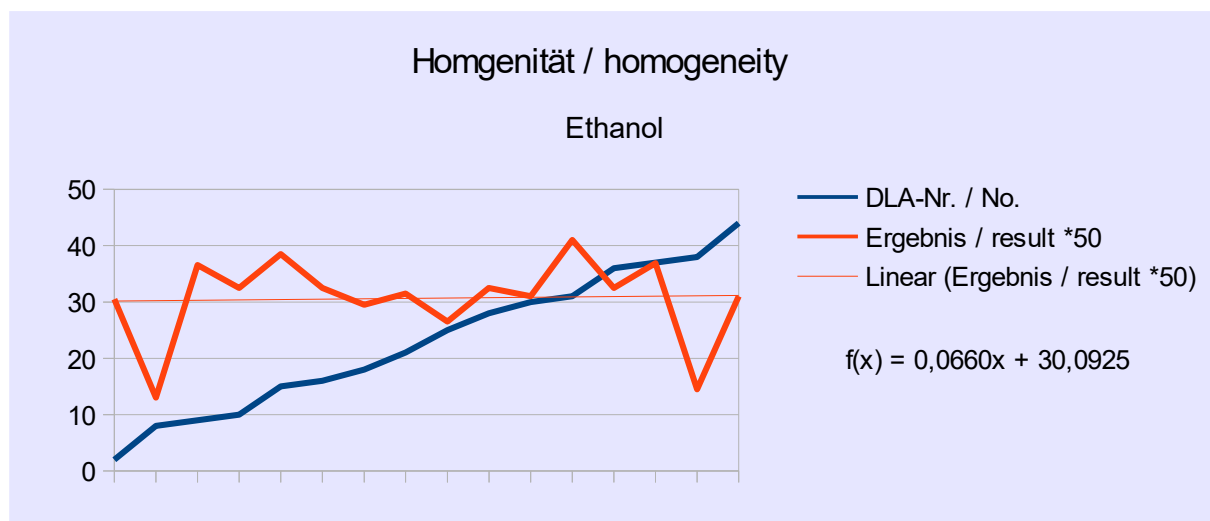
Abb./Fig. 4:
Trendfunktion Probennummern vs. Ergebnisse (1*50 dargestellt)
trend line function sample number vs. results (1*50 shown)

5.3 Information on the Proficiency Test (PT)

Before the PT the participants received the following information in the sample cover letter:

PT number	**DLA ptAU07 (2020)**
PT name	**Ethanol in Marzipan**
Sample matrix*	**Samples I + II:** Marzipan with chocolate coating / ingredients: Marzipan (71%) with chocolate coating / ingredients: almonds, sugar, invert sugar syrup, alcohol with 29% dark chocolate coating
Number of samples and sample amount	2 identical samples I + II: 20 g (2 pieces) each.
Storage	Samples I + II: cooled 2 - 10°C (PT period), frozen < -18°C (long term)
Intentional use	Laboratory use only (quality control samples)
Parameter	quantitative: **Ethanol in Marzipan-Portion**
Methods of analysis	Analytical methods are optional
Notes to analysis	The analysis of PT samples should be performed like a routine laboratory analysis. In general we recommend to homogenize a representative sample amount before analysis according to good laboratory practice, especially in case of low sample weights.
Result sheet	The results for sample I and II as well as the final results calculated as mean of the double determination (samples I and II) should be filled in the result submission file. The recovery rates, if carried out, has to be included in the calculation.
Units	g/100g
Number of significant digits	at least 2
Further information	For information please specify: – Date of analysis – DLA-sample-numbers (for sample I and II) – Limit of detection – Assignment incl. Recovery – Recovery with the same matrix – Method is accredited
Result submission	The result submission file should be sent by e-mail to: **pt@dla-lvu.de**
Last Deadline	**the latest September 04th 2020**
Evaluation report	The evaluation report is expected to be completed 6 weeks after deadline of result submission and sent as PDF file by e-mail.
Coordinator and contact person of PT	Matthias Besler-Scharf PhD / A.Scharf MSc.

* Control of mixture homogeneity and qualitative testings are carried out by DLA. Any testing of the content, homogeneity and stability of PT parameters is subcontracted by DLA.

6. Index of participant laboratories in alphabetical order

Teilnehmer / Participant	Ort / Town	Land /
		ITALY
		GREAT BRITAIN
		FRANCE
		Germany
		Germany
		Germany
		Germany
		Germany
		VIETNAM
		Germany

[Die Adressdaten der Teilnehmer wurden für die allgemeine Veröffentlichung des Auswerte-Berichts nicht angegeben.]

[The address data of the participants were deleted for publication of the evaluation report.]

7. Index of references

1. DIN EN ISO/IEC 17025:2005; Allgemeine Anforderungen an die Kompetenz von Prüf- und Kalibrierlaboratorien / General requirements for the competence of testing and calibration laboratories
2. DIN EN ISO/IEC 17043:2010; Konformitätsbewertung – Allgemeine Anforderungen an Eignungsprüfungen / Conformity assessment – General requirements for proficiency testing
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4. ASU §64 LFGB: Planung und statistische Auswertung von Ringversuchen zur Methoden-validierung / DIN ISO 5725 series part 1, 2 and 6 Accuracy (trueness and precision) of measurement methods and results
5. Verordnung / Regulation 882/2004/EU; Verordnung über über amtliche Kontrollen zur Überprüfung der Einhaltung des Lebensmittel- und Futtermittelrechts sowie der Bestimmungen über Tiergesundheit und Tierschutz / Regulation on official controls performed to ensure the verification of compliance with feed and food law, animal health and animal welfare rules
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15. MTSE SOP No. 010.01 (2014): Quantitative measurement of mixing uniformity and carry-over in powder mixtures with the rotary detector technique, MTSE Micro Tracers Services Europe GmbH
16. Homogeneity and stability of reference materials; Linsinger et al.; Accred Qual Assur, 6, 20-25 (2001)
17. AOAC Official Methods of Analysis: Guidelines for Standard Method Performance Requirements, Appendix F, p. 2, AOAC Int (2016)
18. ASU § 64 LFGB L 40.00-12 Untersuchung von Honig. Bestimmung des Gehaltes an Ethanol; enzymatisches Verfahren, nach DIN 10762; September 2006 [Determination of ethanol in honey – Enzymatic method]
19. ASU § 64 LFGB L 36.00-13 Bestimmung von Ethanol in Bier mit geringem Alkoholgehalt; Dezember 1992 [Determination of ethanol in beer with low alcohol content – Enzymatic method]