DLA
Dienstleistung
Lebensmittel
Analytik GbR

**Evaluation Report**
proficiency test

**DLA 47/2016**

**Food Supplement I:**

# Vitamins A, D3, E, K1 and beta-Carotene

# in Multi Vitamin Capsule Powder

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# Inhalt / Content

# 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 mixture of two common in commerce food supplements "multi vitamin capsules" and lactose as bulking agent from European Suppliers. The first food supplement was crushed including the capsule shells, while the second food supplement was added without capsule shells. The materials were sieved, mixed and homogenized. Afterwards the samples were portioned to approximately 25 g into metallised PET film bags and chronologically numbered.

The composition (list of ingredients) and the amounts of vitamins were calculated according to the labelled values as given in table 1 and table 2 respectively.

Table 1: Composition of DLA-Samples

| **Multi vitamin capsules** |
|---|
| Ingredients including capsule shell (1. food supplement):<br>Dicalcium phosphate, magnesium oxide, gelatin, vitamin C, potassium chloride, niacin, magnesium stearate, vitamin E acetate, calcium D-pantothenate, ferrous sulfate, zinc oxide, vitamin B6 hydrochloride, copper sulfate, vitamin B2, vitamin B1 mononitrate, vitamin A acetate, folic acid, biotin, potassium iodide, chromium-III-chloride, sodium molybdate, sodium selenite, vitamin K1, vitamin D3, vitamin B12.<br><br>Ingredients without capsule shell (2. food supplement):<br>Bulking agent lactose, vitamin C, nicotinamide, vitamin E acetate, calcium D-pantothenate, vitamin B6, vitamin B2, vitamin B1, separating agent: magnesium stearate, silica, beta-carotene, biotin, folic acid, vitamin B12.<br><br>additional ingredient:<br>Lactose |

Table 2: Calculated amounts according to labelled values of vitamins

| Vitamin | Content per 100 g |
|---|---|
| Vitamin A | 23000 µg |
| Vitamin D3 | 87 µg |
| Vitamin E | 980 mg |
| Vitamin K1 | 870 µg |
| beta-Carotene | 39 mg |

*2.1.1 Homogeneity*

The **homogeneity of bottled numbered DLA-samples** was checked by 8fold determination of niacin by HPLC-UV. The repeatability standard deviation of 0,8 % is below the range of the repeatability standard deviations of method EN 15652:2009 for determination of niacin, which are in the range of 1,1% to 5,6% [21]. The results of the homogeneity test are given in the documentation.

The calculation of the **repeatability standard deviation $S_r$ of the participants** was also used as an indicator of homogeneity. It is < 5,0% (1,78% - 4,65%) for all analytes. Therefore these repeatability standard deviations are similar to precision data of the referring standardized methods (e.g. ASU §64 L 00.00-86, s. 3.6.2) (see Tab. 3) [16-20]. The repeatability standard deviations of the participants' results are given in the tables of statistic data (see 4.1 to 4.20).

Furthermore, the homogeneity was characterized by the **trend line function of participants' results for chronological bottled single samples**. The maximum deviations from the mean value of the trend lines for vitamin A and vitamin E were below 30% of the target standard deviations $\sigma_{pt'}$ and $\sigma_{pt}$, respectively (s. 5.2 homogeneity) and can therefore be regarded as low.

If the criteria for sufficient homogeneity of the test material are not fulfilled on a particular parameter, the impact on the target standard deviation is checked and optionally the evaluation of the results of the participants will be done using the z'-score considering the standard uncertainty of the assigned value (see 3.8 and 3.11) [3].

## 2.2 Sample shipment and information to the test

Two portions of test material were sent to every participating laboratory in the 24th week of 2016. The testing method was optional. The tests should be finished at 12th August 2016 the latest.

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

*The two identical samples are powdered multivitamin capsules including capsule shells (gelatin) containing Vitamins A, D3, E, K1 and beta-carotene. The recommendation is to take 3 g per day. Each sample bag contains 50 g. The samples contain vitamins in the form of approved food supplements nutrient compounds. The material was tested for homogeneity and is intended for laboratory use only. The methods for determination are optional (e.g. HPLC, ELISA).*
*In general we recommend to homogenize a representative sample amount before analysis according to good laboratory practice, especially in case of low sample weights.*

## 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.

From the 11 participants one participant submitted the results delayed in consultation with DLA. All other participants submitted the result 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].

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.

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 $\sigma_{pt}$ (standard deviation for proficiency assessment) a robust standard deviation ($S^x$) 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 statistical 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 as a percentage of the mean value is indicated as coefficient of variation $CV_R$ in the table of statistical characteristics in the results section in case single results from participants are available. Its meaning is explained in more detail in 3.9.

## 3.5 Exclusion of results and outliers

Before statistical evaluation obvious blunders, such as those with incorrect units, decimal point errors, and results for a another proficiency test item can be removed from the data set [2]. 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 identified as outliers by the use of robust statistics. If a value deviates from the robust mean by more than 3 times the robust standard deviation, it is classified as an outlier [3]. Detected outliers are stated for information only, when z-score are $< -2$ or $> 2$. Due to the use of robust statistics outliers are not excluded, provided that no other reasons are present [3].

## 3.6 Target standard deviation (for proficiency assessment)

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

If an acceptable quotient $S^*/\sigma_{pt}$ is present, the target standard deviation of the general model by Horwitz is preferably used for the proficiency assessment. It is usually suitable for for evaluation of interlaboratory studies, where different analytical 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.

***In the present PT for valuation of vitamin A, vitamin D3 and Vitamin K1 the target standard deviation according to the general model of Horwitz was applied (see 3.6.1) and for vitamin E and beta-carotene the target standard deviation according to precision experiments was applied (3.6.2). Wherein the results of beta-carotene were not evaluated by means of z-scores due to the number of < 7.***

### *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 $\sigma_R$ [6]. Later the model was modified by Thompson for certain concentration ranges [10]. The reproducibility standard deviation $\sigma_R$ can be applied as the relative target standard deviation $\sigma_{pt}$ 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 $\sigma_R$ and the repeatability standard deviation $\sigma_r$ of a precision experiment (collaborative trial or proficiency test) the target standard deviation $\sigma_{pt}$ 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 values given in Table 3 relative repeatability standard deviation ($RSD_r$) and relative reproducibility standard deviation ($RSD_R$) were determined in collaborative trials using the specified methods.
The in the table indicated resulting target standard deviations $\sigma_{pt}$ were used for evaluation of the results.
For information the target standard deviations according to Horwitz are given additionally.

Table 3: Relative repeatability standard deviations ($RSD_r$) and relative reproducibility standard deviations ($RSD_R$) from precision experiments and resulting target standard deviations $\sigma_{pt}$ [16-20]

| **Parameter** | **Matrix** | **Mean values** | **$RSD_r$** | **$RSD_R$** | **$\sigma_{pt}$** | **Method / Literature** |
|---|---|---|---|---|---|---|
| Vitamin A | milk powder | 653 µg/100 g | 2,1% | 3,4% | 3,06%[1] | HPLC [18] |
| Vitamin D3 | milk powder | 14,30 µg/100 g | 5,2% | 5,5% | 4,09% | HPLC [16] |
| | milk powder | 9,95 µg/100 g | 8,2% | 13,6% | 12,3%[1] | HPLC [16b] |
| | infant food, liquid | 1,38 µg/100 g | 5,9% | 12,1% | 11,4% | HPLC [16] |
| | infant food, powder | 10,1 µg/100 g | 2,4% | 7,1% | 6,89% | HPLC [16] |
| Vitamin E | oat powder | 0,279 mg/100g | 9,0% | 16,8% | 15,5% | HPLC [17] |
| | milk powder | 9,89 mg/100 g | 4,0% | 7,0% | 6,40% | HPLC [17] |
| | milk powder | 10,2 mg/100 g | 3,0% | 12,8% | 12,6%[1] | HPLC [17] |
| Vitamin K1 | 6 infant food (mean) | 77,37 µg/100 g | 4,47% | 5,91% | 4,99%[1] | HPLC [20] |
| β-Carotene | mixed vegetables | 18,05 mg/100g | 3,9% | 15% | 14,7%[1] | HPLC [19] |
| | pudding powder | 1,531 mg/100g | 5,6% | 9,3% | 8,42% | HPLC [19] |
| | vitamin drink | 2,248 mg/100g | 2,9% | 6,5% | 6,17% | HPLC [19] |

[1] *used in evaluation (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 3.6.1 and 3.6.2 were regarded suitable. Table 4 shows selected characteristics of participants results of the present PT in comparison to previous years.

## 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 ($\sigma_{pt}$) 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 z-score valid for the PT evaluation is designated z-score ($\sigma_{pt}$), while the value of z-score (Info) is for information only. The two z-scores are calculated using the different target standard deviations according to 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. For example a fault isolation or a root cause analysis through the examination of transmission error or an error in the calculation, in the trueness and precision must be performed and if necessary appropriate corrective measures should be applied [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].

Table 4: Characteristics of the present PT (on dark gray) in comparison to previous PTs since 2012 (SD = standard deviation, CV = coefficient of variation)

| Parameter | Matrix | robust Mean | rob. SD (S*) | rel. SD ($VK_{S*}$) [%] | Quotient $S^*/\sigma_{pt}$ | DLA-Report |
|---|---|---|---|---|---|---|
| Vitamin A | Multivit-amin-Tablets | 13700 µg/100g | 1800 µg/100g | 13,1% | 1,7 | DLA 32/2012 |
| Vitamin A | Multivit-amin-Powder | 690 µg/100g | 180 µg/100g | 26,1% | 2,2 | DLA 29/2014 |
| Vitamin A | Multivit-amin-Capsule Powder | 21900 µg/100g | 2870 µg/100g | 13,1% | 1,8 | DLA 47/2016 |
| Vitamin D3 | Multivit-amin-Tablets | 180 µg/100g | 23 µg/100g | 12,8% | 0,9 | DLA 32/2012 |
| Vitamin D3 | Multivit-amin-Powder | 28,6 µg/100g | 11,2 µg/100g | 39,2% | 2,0 | DLA 29/2014 |
| Vitamin D3 | Multivit-amin-Capsule Powder | 146 µg/100g | 10,3 µg/100g | 7,05% | 0,46 | DLA 47/2016 |
| Vitamin E | Multivit-amin-Tablets | 355 mg/100g | 71 mg/100g | 20,0% | 1,6 | DLA 32/2012 |
| Vitamin E | Multivit-amin-Powder | 92,7 mg/100g | 16,3 mg/100g | 17,6% | 1,4 | DLA 29/2014 |
| Vitamin E | Multivit-amin-Capsule Powder | 988 mg/100g | 211 mg/100g | 21,4% | 1,7 | DLA 47/2016 |
| Vitamin K1 | Multivit-amin-Tablets | 715 µg/100g | 113 µg/100g | 15,8% | 1,3 | DLA 32/2012 |
| Vitamin K1 | Multivit-amin-Capsule Powder | 933 µg/100g | 121 µg/100g | 13,0% | 1,1 | DLA 47/2016 |
| β-Carotene | Multivit-amin-Tablets | 2,54 mg/100g | 0,59 mg/100g | 23,2% | 2,4 | DLA 32/2012 |
| β-Carotene | Multivit-amin-Powder | 0,509 mg/100g | 0,160 mg/100g | 31,4% | 2,5 | DLA 29/2014 |
| β-Carotene | Multivit-amin-Capsule Powder | 32,2 mg/100g | 9,70 mg/100g | 30,1% | 2,0 | DLA 47/2016 |

## 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. 3.8). The z'-score represents the relation of the deviation of the result (x) of the participant from the respective consensus value (X) to the square root of quadrat sum of the target standard deviation ( $\hat{\sigma}$ ) and the standard uncertainty ($Ux_{pt}$) [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 $\sigma_{pt}$'.

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 coefficient of variation ($CV_R$)

The coefficient of variation ($CV_R$) 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 variability the $CV_K$ gives the relative variability within a data region. While a low $CV_R$, e.g. <5-10% can be taken as evidence for a homogeneous set of results, a $CV_R$ 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 values or the performance evaluation of the participants possibly can not be done [3].

## 3.10 Quotient $S^*/\sigma_{pt}$

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 $\sigma_{pt}$ 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

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

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

If $U(X_{pt}) \leq 0{,}3\ \sigma_{pt}$ the standard uncertainty of the consensus value needs not to be included in the interpretation of the results of the PT [3]. A clear exceeded the value of 0,3 is an indication that the target standard deviation was possibly set too low for the standard uncertainty of the assigned value.

The quotient $U(X_{pt})/\sigma_{pt}$ is reported in the characteristics of the test.

# 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 $\sigma_{pt}$ or $\sigma_{pt}$' |
| 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 ($X_{pt} + 2\sigma_{pt}$') * |
| Variation coefficient $V_K$ in % |
| *Quotient S*/$\sigma_{pt}$ or S*/$\sigma_{pt}$'* |
| *Standard uncertainty U($X_{pt}$)* |
| *Quotient U($X_{pt}$)/$\sigma_{pt}$ or U($X_{pt}$)/$\sigma_{pt}$'* |
| *Number of results in the target range* |
| *Percent in the target range* |

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

In the second table the individual results of the participating laboratories are listed:

| **Auswerte-nummer**<br>**Evaluation number** | **Parameter [Einheit / Unit]** | **Abweichung**<br>**Deviation** | **z-Score σpt** | **z-Score** (Info) | **Hinweis**<br>**Remark** |
|---|---|---|---|---|---|

## 4.1 Vitamin A (as Retinol in µg / 100 g)

**Vergleichsuntersuchung / Proficiency Test**

| **Statistic Data** | |
|---|---|
| *Number of results* | 10 |
| *Number of outliers* | 0 |
| Mean | 21900 |
| Median | 22400 |
| **Robust Mean (X)** | **21900** |
| **Robust standard deviation (S*)** | **2870** |
| *Number with 2 replicates* | 10 |
| Repeatability SD ($S_r$) | 1020 |
| Repeatability ($CV_r$) | 4,65% |
| Reproducibility SD ($S_R$) | 2640 |
| Reproducibility ($CV_R$) | 12,1% |
| *Target range:* | |
| **Target standard deviation $\sigma_{pt}$** | **1560** |
| Target standard deviation (for Information) | 669 |
| **lower limit of target range** | **18800** |
| **upper limit of target range** | **25000** |
| *Quotient $S^*/\sigma_{pt}$* | *1,8* |
| *Standard uncertainty $U_{(Xpt)}$* | *1130* |
| *Quotient $U_{(Xpt)}/\sigma_{pt}$* | *0,73* |
| *Results in the target range* | *8* |
| *Percent in the target range* | *80%* |

*Comments to the statistic data:*

The target standard deviation was calculated according to the general model of Horwitz.

The evaluation showed a normal variability of results. The quotient $S^*/\sigma_{pt}$ was below 2,0. The robust standard deviation is below the range of previous PTs (see 3.6.3). The comparability of results is given.

The repeatability and reproducibility standard deviations were higher than established values for the applied methods (see 3.6.2).

The quotient $U_{(Xpt)}/\sigma_{pt}$ was 0,73. Although it was not below 0,3 it is acceptable due to the other statistical data and the use of different analytical methods.

80% of results were in the target range.

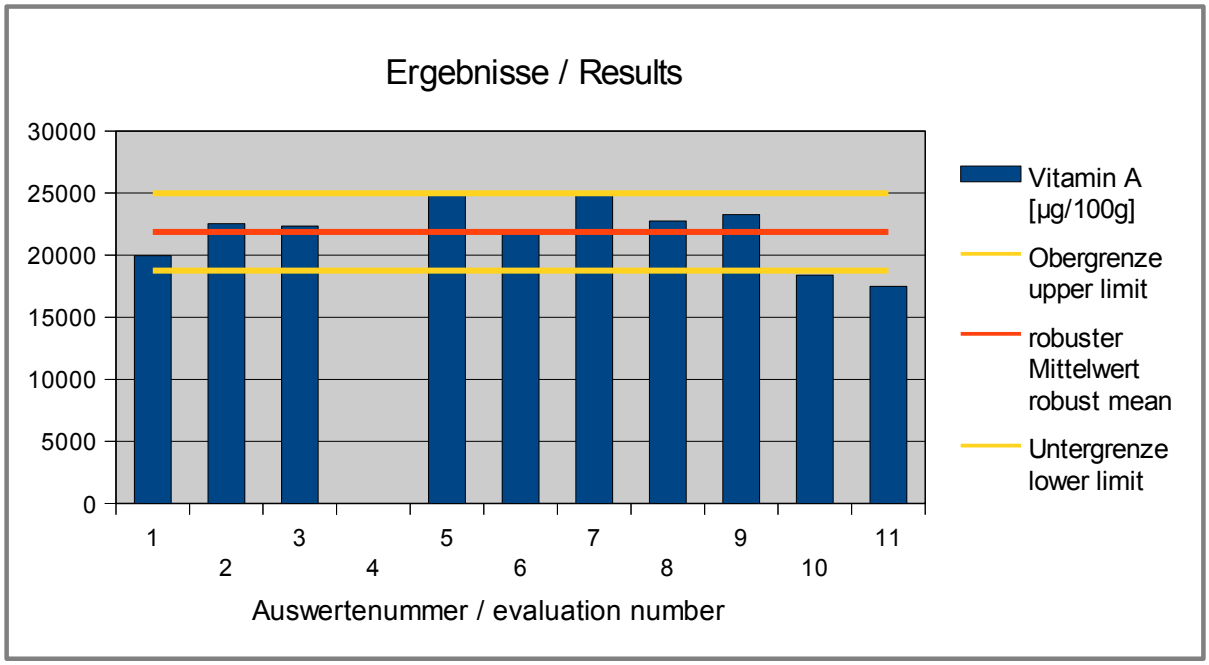


**Abb. 1:** Ergebnisse Vitamin A
**Fig. 1:** Results vitamin A

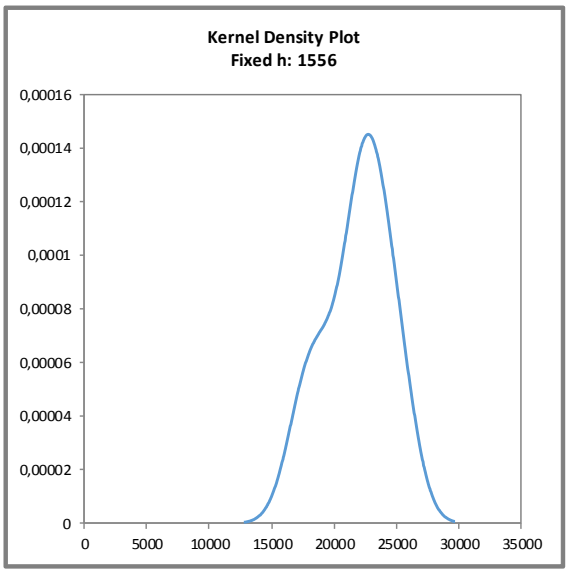


**Abb. 2:** Kerndichte-Schätzung der Ergebnisse für Vitamin A
(mit h = 1,0 x $\sigma_{pt}$ von $X_{pt}$)

**Fig. 2:** Kernel density plot of vitamin A (with h = 1,0 x $\sigma_{pt}$ von $X_{pt}$)

*Comments:*
The kernel density estimation shows almost a normal distribution with a slight shoulder at approximately 18000 µg/100g (s. fig. 2).

**Ergebnisse der Teilnehmer:**
**Results of Participants:**

| Auswerte-nummer<br>Evaluation number | Vitamin A [µg/100g] | Abweichung [µg/100g]<br>Deviation [µg/100g] | z-Score ($\sigma_{pt}$) | z-Score (Info) | Hinweis<br>Remark |
|---|---|---|---|---|---|
| 1 | 19950 | -1926 | -1,2 | -2,9 | |
| 2 | 22542 | 666 | 0,4 | 1,0 | |
| 3 | 22337 | 461 | 0,3 | 0,7 | |
| 4 | | | | | |
| 5 | 25044 | 3168 | 2,0 | 4,7 | |
| 6 | 21883 | 7 | 0,0 | 0,0 | |
| 7 | 25016 | 3140 | 2,0 | 4,7 | |
| 8 | 22750 | 874 | 0,6 | 1,3 | |
| 9 | 23270 | 1394 | 0,9 | 2,1 | |
| 10 | 18394 | -3482 | -2,2 | -5,2 | |
| 11 | 17496 | -4380 | -2,8 | -6,5 | |

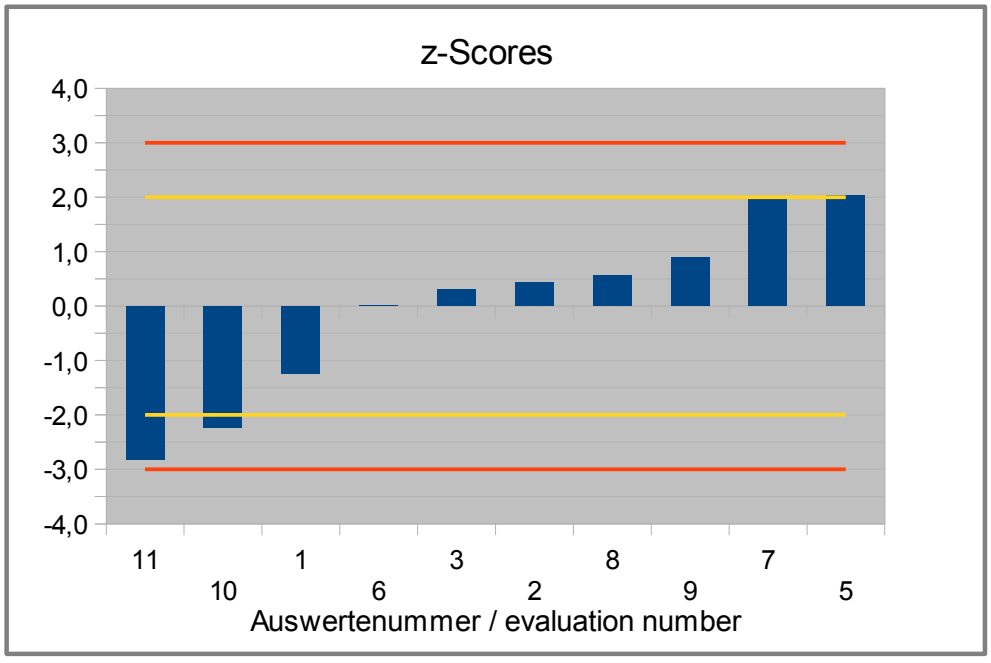


**Abb. 3:** Z-Scores Vitamin A
**Fig. 3:** Z-Scores vitamin A

## 4.2 Vitamin D3 (as Cholecalciferol in µg / 100 g)

**Vergleichsuntersuchung / Proficiency Test**

| **Statistic Data** | |
|---|---|
| *Number of results* | 8 |
| *Number of outliers* | 0 |
| Mean | 147 |
| Median | 146 |
| **Robust Mean (X)** | **146** |
| **Robust standard deviation (S*)** | **10,3** |
| *Number with 2 replicates* | 8 |
| Repeatability SD ($S_r$) | 6,36 |
| Repeatability ($CV_r$) | 4,33% |
| Reproducibility SD ($S_R$) | 15,3 |
| Reproducibility ($CV_R$) | 10,4% |
| *Target range:* | |
| **Target standard deviation $\sigma_{pt}$** | **22,1** |
| Target standard deviation (for Information) | 18,0 |
| **lower limit of target range** | **102** |
| **upper limit of target range** | **190** |
| *Quotient S*/$\sigma_{pt}$* | *0,46* |
| *Standard uncertainty $U_{(Xpt)}$* | *4,53* |
| *Quotient $U_{(Xpt)}/\sigma_{pt}$* | *0,21* |
| *Results in the target range* | *8* |
| *Percent in the target range* | *100%* |

*Comments to the statistic data:*

The target standard deviation was calculated according to the general model of Horwitz.

The evaluation showed a low variability of results. The quotient $S^*/\sigma_{pt}$ was clearly below 2,0. The robust standard deviation is below the range of previous PTs (see 3.6.3). The comparability of results is given.

The repeatability and reproducibility standard deviations were in the range of established values for the applied methods (see 3.6.2).

The quotient $U_{(Xpt)}/\sigma_{pt}$ of 0,21 was below 0,3.

All results were in the target range.

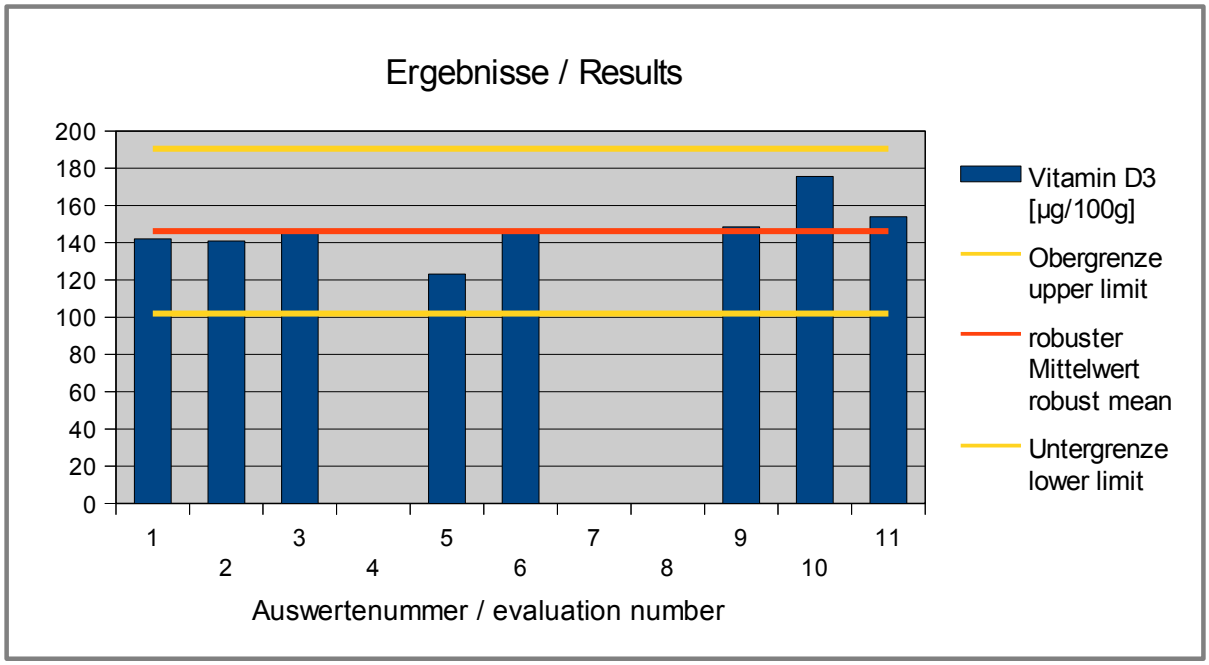


**Abb. 4:** Ergebnisse Vitamin D3
**Fig. 4:** Results vitamin D3

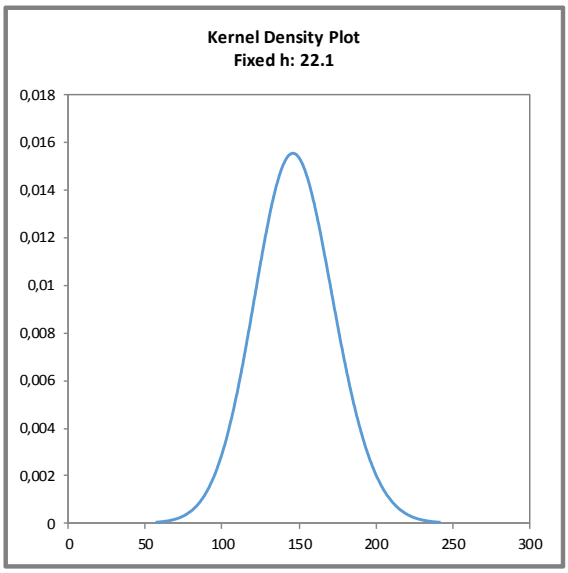


**Abb. 5:** Kerndichte-Schätzung der Ergebnisse für Vitamin D3
(mit h = 1,0 x $\sigma_{pt}$ von $X_{pt}$)

**Fig. 5:** Kernel density plot of vitamin D3 (with h = 1,0 x $\sigma_{pt}$ von $X_{pt}$)

*Comments:*
The kernel density estimation shows a normal distribution of results (s. fig. 5).

**Ergebnisse der Teilnehmer:**
**Results of Participants:**

| Auswerte-nummer<br>Evaluation number | Vitamin D3 [µg/100g] | Abweichung [µg/100g]<br>Deviation [µg/100g] | z-Score ($\sigma_{pt}$) | z-Score (Info) | Hinweis<br>Remark |
|---|---|---|---|---|---|
| 1 | 142 | -4,2 | -0,2 | -0,2 | |
| 2 | 141 | -5,3 | -0,2 | -0,3 | |
| 3 | 146 | -0,2 | 0,0 | 0,0 | |
| 4 | | | | | |
| 5 | 123 | -23,1 | -1,0 | -1,3 | |
| 6 | 146 | -0,2 | 0,0 | 0,0 | |
| 7 | | | | | |
| 8 | | | | | |
| 9 | 149 | 2,3 | 0,1 | 0,1 | |
| 10 | 176 | 29,3 | 1,3 | 1,6 | |
| 11 | 154 | 7,8 | 0,4 | 0,4 | |

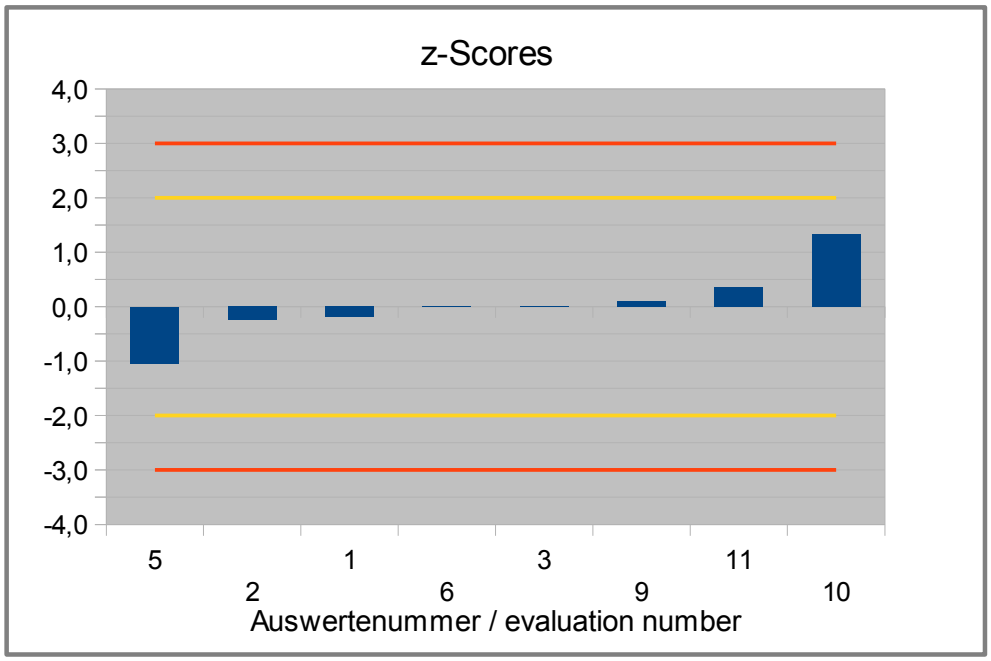


**Abb. 6:** Z-Scores Vitamin D3
**Fig. 6:** Z-Scores vitamin D3

## 4.3 Vitamin E (as D-alpha-Tocopherol in mg / 100 g)

### Vergleichsuntersuchung / Proficiency Test

| **Statistic Data** | |
|---|---|
| *Number of results* | 10 |
| *Number of outliers* | 0 |
| Mean | 988 |
| Median | 972 |
| **Robust Mean (X)** | **988** |
| **Robust standard deviation (S*)** | **211** |
| *Number with 2 replicates* | 10 |
| Repeatability SD ($S_r$) | 23,4 |
| Repeatability ($CV_r$) | 2,37% |
| Reproducibility SD ($S_R$) | 187 |
| Reproducibility ($CV_R$) | 18,9% |
| *Target range:* | |
| **Target standard deviation $\sigma_{pt}$** | **125** |
| Target standard deviation (for Information) | 39,6 |
| **lower limit of target range** | **738** |
| **upper limit of target range** | **1237** |
| *Quotient S*/$\sigma_{pt}$* | *1,7* |
| *Standard uncertainty $U_{(Xpt)}$* | *83,5* |
| *Quotient $U_{(Xpt)}/\sigma_{pt}$* | *0,67* |
| *Results in the target range* | *10* |
| *Percent in the target range* | *100%* |

*Comments to the statistic data:*

The target standard deviation was calculated according to the evaluation of a precision experiment (ASU / EN s. 3.6.2).

The evaluation showed an acceptable variability of results. The quotient $S^*/\sigma_{pt}$ was below 2,0. The robust standard deviation is below the range of previous PTs (see 3.6.3). The comparability of results is given.

The repeatability and reproducibility standard deviations were in the range of established values for the applied methods (see 3.6.2).

The quotient $U_{(Xpt)}/\sigma_{pt}$ was 0,67. Although it was not below 0,3 it is acceptable due to the other statistical data and the use of different analytical methods.

All results were in the target range.

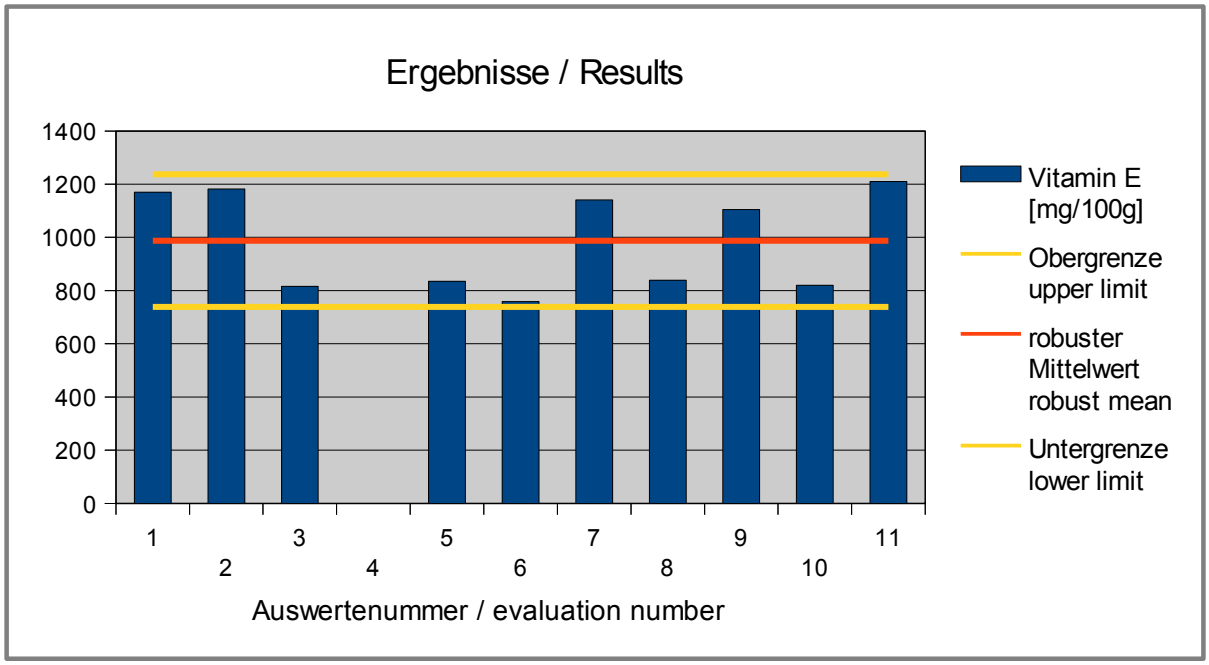


**Abb. 7:** Ergebnisse Vitamin E
**Fig. 7:** Results vitamin E

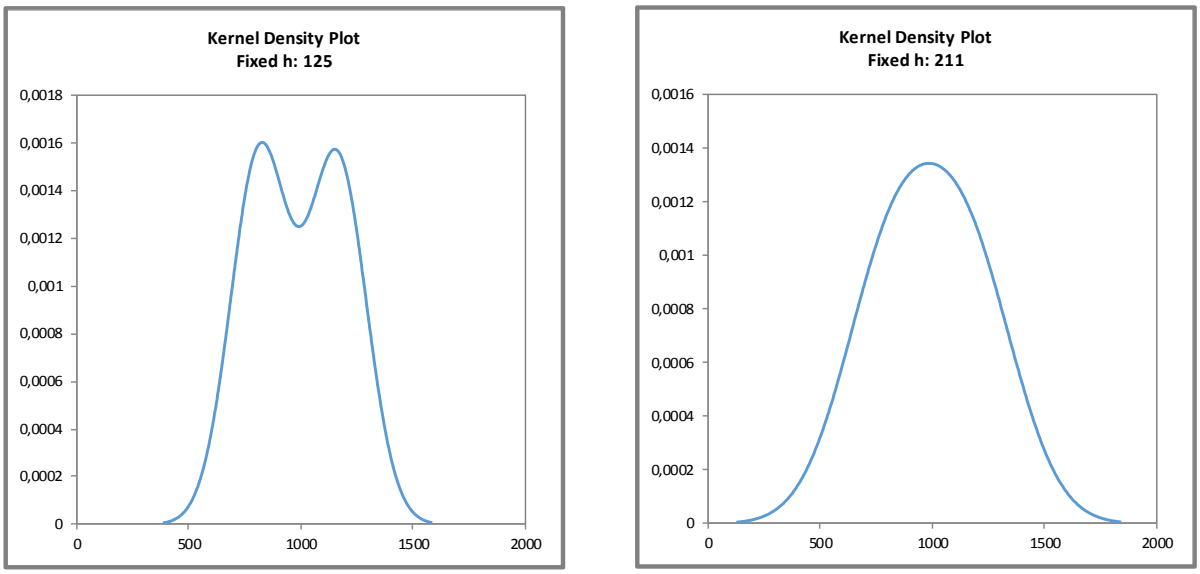


**Abb. 8:** Kerndichte-Schätzungen der Ergebnisse für Vitamin E
(links mit h = 1,0 x $\sigma_{pt}$ von $X_{pt}$ und rechts mit h = 1,0 x S* von $X_{pt}$)

**Fig. 8:** Kernel density plots of vitamin E (left with h = 1,0 x $\sigma_{pt}$ of $X_{pt}$ and right with h = 1,0 x S* of $X_{pt}$)

*Comments:*
The kernel density estimation shows two maximums when estimated against the target standard deviation. It is passing over into a normal distribution using the robust standard deviation as an estimator (s. fig. 8). The

information on the methods provided by the participants give no obvious indications of such grouping of results (s. documentation).

**Ergebnisse der Teilnehmer:**
**Results of Participants:**

| Auswerte-nummer<br>Evaluation number | Vitamin E [mg/100g] | Abweichung [mg/100g]<br>Deviation [mg/100g] | z-Score ($\sigma_{pt}$) | z-Score (Info) | Hinweis<br>Remark |
|---|---|---|---|---|---|
| 1 | 1170 | 182 | 1,5 | 4,6 | |
| 2 | 1182 | 194 | 1,6 | 4,9 | |
| 3 | 816 | -172 | -1,4 | -4,3 | |
| 4 | | | | | |
| 5 | 836 | -152 | -1,2 | -3,8 | |
| 6 | 759 | -229 | -1,8 | -5,8 | |
| 7 | 1142 | 154 | 1,2 | 3,9 | |
| 8 | 839 | -149 | -1,2 | -3,8 | |
| 9 | 1105 | 117 | 0,9 | 3,0 | |
| 10 | 820 | -168 | -1,3 | -4,2 | |
| 11 | 1210 | 222 | 1,8 | 5,6 | |

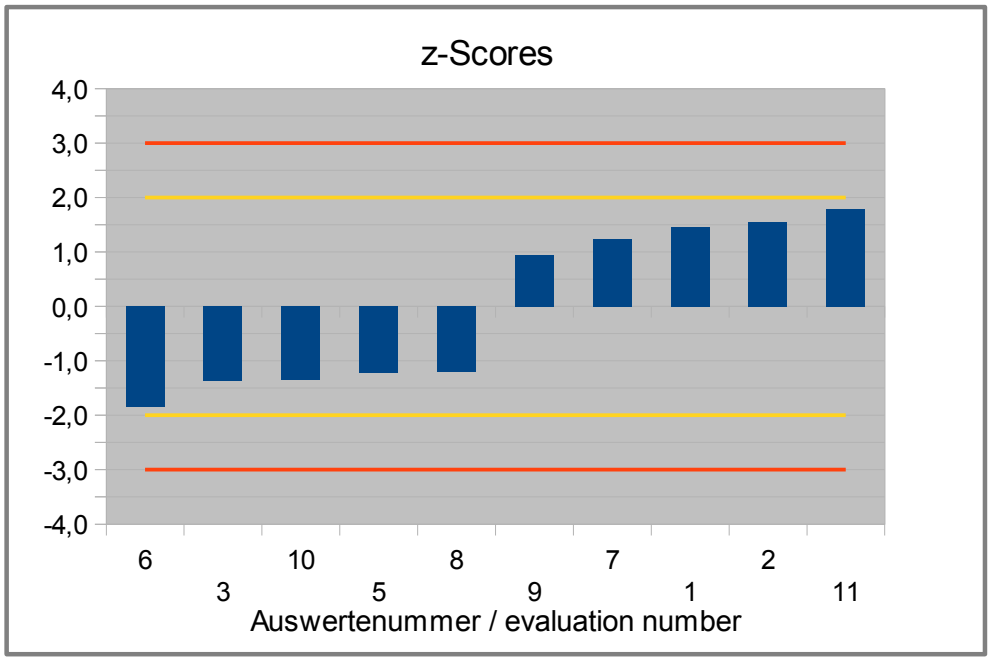


**Abb. 9:** Z-Scores Vitamin E
**Fig. 9:** Z-Scores vitamin E

## 4.4 Vitamin K1 (as Phylloquinone in µg / 100 g)

**Vergleichsuntersuchung / Proficiency Test**

| **Kenndaten** | |
|---|---|
| *Anzahl der Messergebnisse* | 7 |
| *Anzahl der Ausreißer* | 0 |
| Mittelwert | 933 |
| Median | 945 |
| **Robuster Mittelwert ($X_{pt}$)** | **933** |
| **Robuste Standardabweichung (S*)** | **121** |
| *Anzahl mit 2 Wiederholmessungen* | 7 |
| Wiederholstandardabweichung ($S_r$) | 16,6 |
| Variationskoeffizient ($VK_r$) | 1,78% |
| Vergleichsstandardabweichung ($S_R$) | 108 |
| Variationskoeffizient ($VK_R$) | 11,5% |
| *Zielkenndaten:* | |
| **Zielstandardabweichung $\sigma_{pt}$** | **107** |
| Zielstandardabweichung (zur Information) | 46,6 |
| **Untere Grenze des Zielbereichs** | **720** |
| **Obere Grenze des Zielbereichs** | **1150** |
| *Quotient S*/$\sigma_{pt}$* | *1,1* |
| *Standardunsicherheit $U_{(Xpt)}$* | *57,4* |
| *Quotient $U_{(Xpt)}/\sigma_{pt}$* | *0,54* |
| *Ergebnisse im Zielbereich* | 7 |
| *Prozent im Zielbereich* | *100%* |

*Comments to the statistic data:*

The target standard deviation was calculated according to the general model of Horwitz.

The evaluation showed a normal variability of results. The quotient $S^*/\sigma_{pt}$ was below 2,0. The robust standard deviation is below the range of previous PTs (see 3.6.3). The comparability of results is given.

The repeatability and reproducibility standard deviations were in the range of established values for the applied methods (see 3.6.2).

The quotient $U_{(Xpt)}/\sigma_{pt}$ was 0,54. Although it was not below 0,3 it is acceptable due to the other statistical data and the use of different analytical methods.

All results were in the target range.

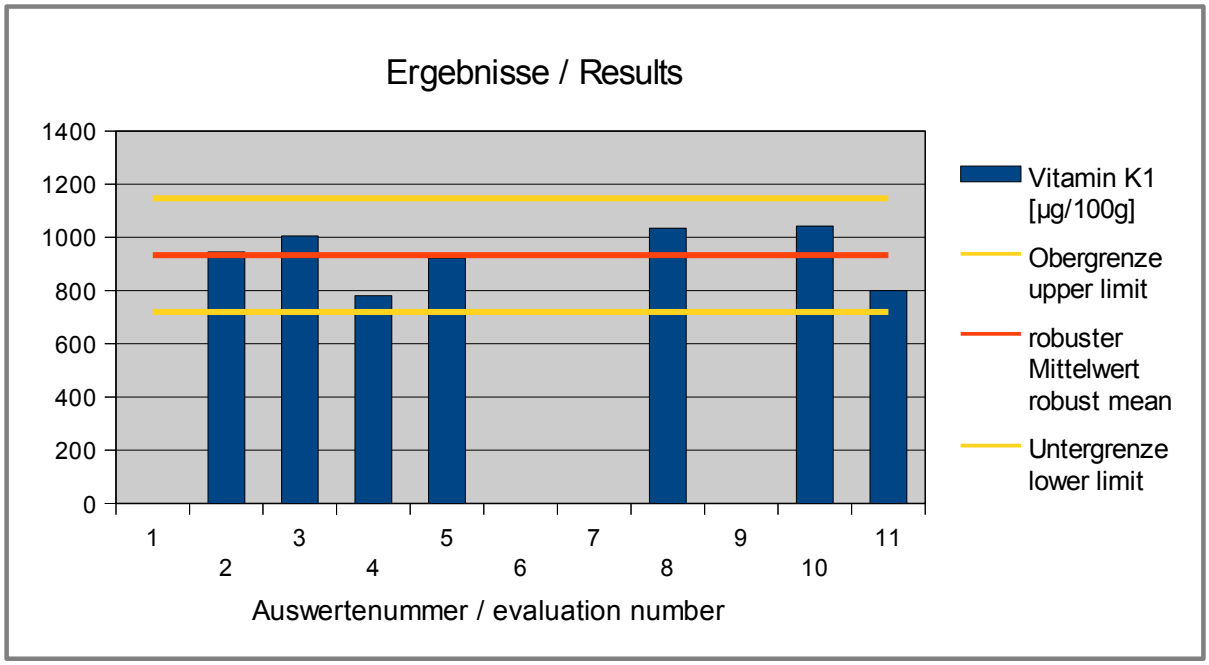


**Abb. 10:** Ergebnisse Vitamin K1
**Fig. 10:** Results vitamin K1

*Comments:*
The kernel density estimation was not calculated due to a number of < 8 results.

**Ergebnisse der Teilnehmer:**
**Results of Participants:**

| Auswerte-nummer<br>Evaluation number | Vitamin K1 [µg/100g] | Abweichung [µg/100g]<br>Deviation [µg/100g] | z-Score ($\sigma_{pt}$) | z-Score (Info) | Hinweis<br>Remark |
|---|---|---|---|---|---|
| 1 | | | | | |
| 2 | 945 | 12 | 0,1 | 0,3 | |
| 3 | 1005 | 72 | 0,7 | 1,5 | |
| 4 | 781 | -152 | -1,4 | -3,3 | |
| 5 | 922 | -11 | -0,1 | -0,2 | |
| 6 | | | | | |
| 7 | | | | | |
| 8 | 1035 | 102 | 1,0 | 2,2 | |
| 9 | | | | | |
| 10 | 1043 | 110 | 1,0 | 2,4 | |
| 11 | 800 | -133 | -1,2 | -2,9 | |

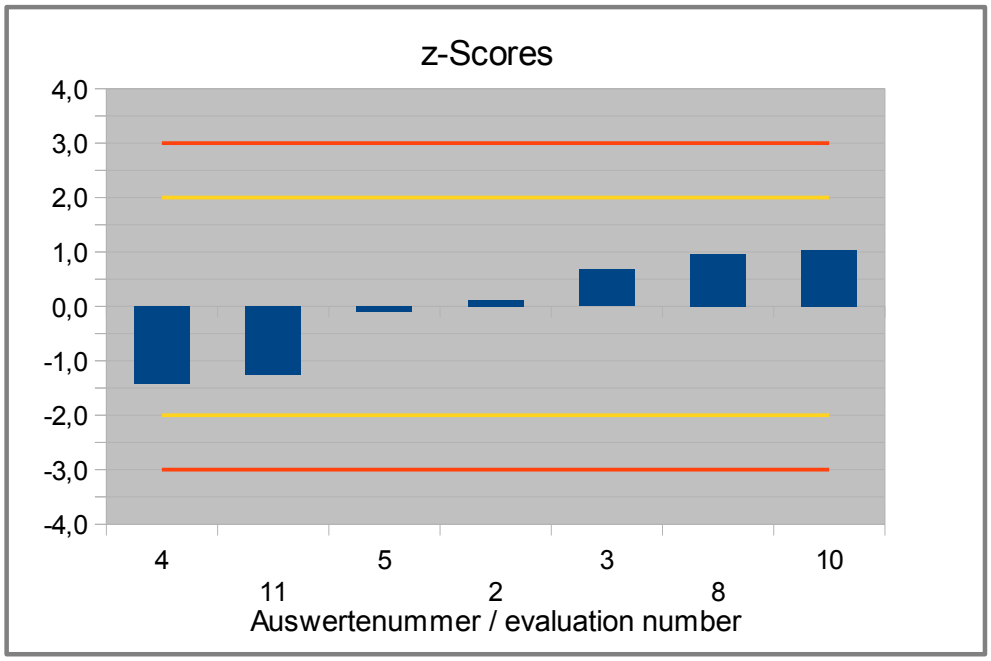


**Abb. 11:** Z-Scores Vitamin K1
**Fig. 11:** Z-Scores vitamin K1

## 4.5 beta-Carotene (in mg / 100 g)

**Vergleichsuntersuchung / Proficiency Test**

| **Statistic Data** | |
|---|---|
| *Number of results* | 6 |
| *Number of outliers* | 0 |
| Mean | 32,2 |
| Median | 33,3 |
| **Robust Mean (X)** | **32,2** |
| **Robust standard deviation (S*)** | **9,70** |
| *Number with 2 replicates* | 6 |
| Repeatability SD ($S_r$) | 0,98 |
| Repeatability ($CV_r$) | 3,04% |
| Reproducibility SD ($S_R$) | 8,58 |
| Reproducibility ($CV_R$) | 26,6% |
| *Target range:* | |
| **Target standard deviation $\sigma_{pt}$** | **4,75** |
| Target standard deviation (for Information) | 2,16 |
| **lower limit of target range** | **-** |
| **upper limit of target range** | **-** |
| *Quotient S*/$\sigma_{pt}$* | *2,0* |
| *Standard uncertainty $U_{(Xpt)}$* | *4,9* |
| *Quotient $U_{(Xpt)}/\sigma_{pt}$* | *1,0* |
| *Results in the target range* | - |
| *Percent in the target range* | - |

*Comments to the statistic data:*

A valuation of results using z-scores was not performed due to the small number of results of < 7. The following comments are for information only:

The target standard deviation was calculated according to the evaluation of a precision experiment (ASU / EN s. 3.6.2).

The evaluation showed an acceptable variability of results. The quotient $S^*/\sigma_{pt}$ was 2,0. The robust standard deviation is in the range of previous PTs (see 3.6.3). The repeatability and reproducibility standard deviations were in the range of established values for the applied methods (see 3.6.2). The quotient $U_{(Xpt)}/\sigma_{pt}$ of 1,0 was increased.

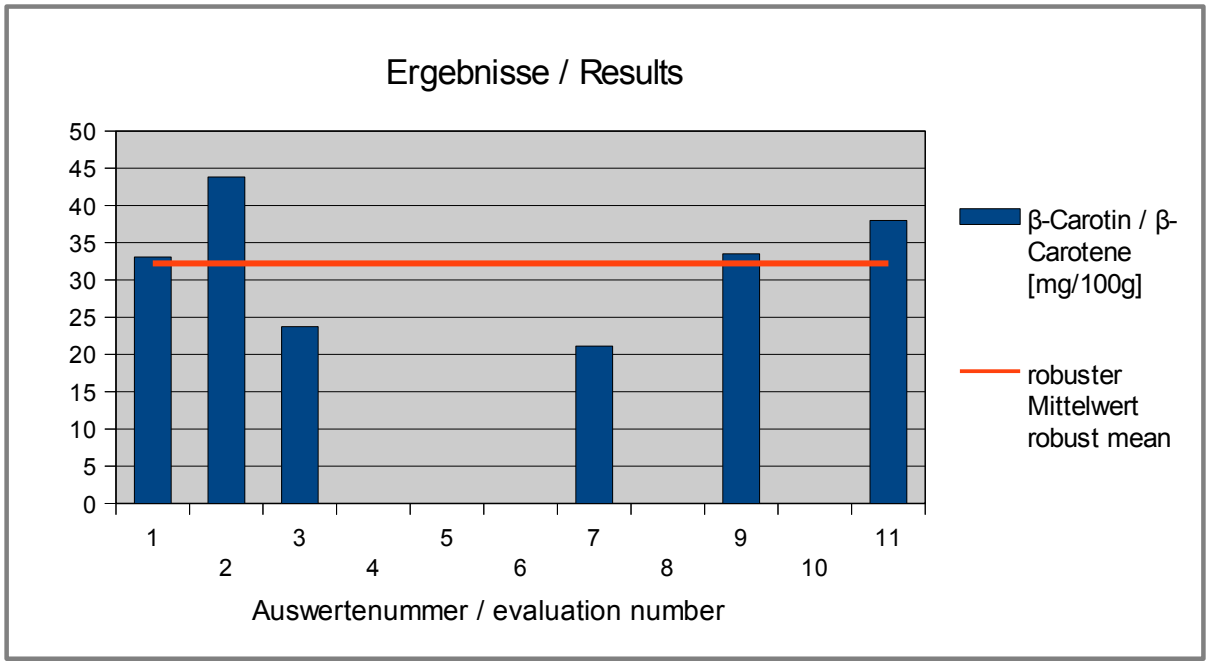


**Abb. 12:** Ergebnisse β-Carotin
**Fig. 12:** Results β-carotene

*Comments:*
The kernel density estimation was not calculated due to a number of < 8 results.

**Ergebnisse der Teilnehmer:**
**Results of Participants:**

| Auswerte-nummer<br>Evaluation number | β-Carotin / β-Carotene [mg/100g] | Abweichung [mg/100g]<br>Deviation [mg/100g] | z-Score ($\sigma_{pt}$) | z-Score (Info) | Hinweis<br>Remark |
|---|---|---|---|---|---|
| 1 | 33,1 | 0,9 | | | |
| 2 | 43,8 | 11,6 | | | |
| 3 | 23,7 | -8,5 | | | |
| 4 | | | | | |
| 5 | | | | | |
| 6 | | | | | |
| 7 | 21,1 | -11,1 | | | |
| 8 | | | | | |
| 9 | 33,5 | 1,3 | | | |
| 10 | | | | | |
| 11 | 38,0 | 5,8 | | | |

# 5. Documentation

## 5.1 Primary data

| Parameter | Teilnehmer | Einheit | Proben-Nr. A | Proben-Nr. B | Datum d. Analyse | Ergebnis (Mittel) | Ergebnis A | Ergebnis B | Bestim-mungsgren-ze | Inkl. WF | Wiederfin-dungsrate [%] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Unit** | **Sample No. A** | **Sample No. B** | **Date of analysis** | **Result (Mean)** | **Result A** | **Result B** | **Limit of de-termination** | **Incl. RR** | **Recovery rate [%]** |
| **Vitamin A** (als Retinol ohne Provitamine / as retinol without provitamins) | 1 | µg/100g | 19 | 55 | Jul 16 | 19950 | 20200 | 19700 | 4500 | no | n/a |
| | 2 | µg/100g | 4 | 16 | 16-08-08 | 22541,68 | 21736,63 | 23346,73 | 1,9 | no | 103,9 |
| | 3 | µg/100g | 29 | 46 | 16-07-19 | 22336,75 | 23154,75 | 21518,74 | 5 | no | |
| | 4 | µg/100g | 5 | 38 | n/a | | | | | | |
| | 5 | µg/100g | 7 | 36 | 16-06-30 | 25044,08 | 26647,12 | 23441,03 | 3090 | no | - |
| | 6 | µg/100g | 10 | 21 | 16-06-22 | 21883 | 22023 | 21743 | 9 | na | na |
| | 7 | µg/100g | 42 | 60 | Jul 16-21 | 25016,15 | 24491,66 | 25540,64 | 60 | yes | 102,47 |
| | 8 | µg/100g | 44 | 48 | 16-07-04 | 22750 | 22700 | 22800 | 3000 | no | - |
| | 9 | µg/100g | 13 | 25 | 16-07-06 | 23270 | 23470 | 23070 | | no | 97,8 |
| | 10 | µg/100g | 34 | 51 | | 18394,24 | 18190,3 | 18598,17 | | no | |
| | 11 | µg/100g | 27 | 53 | 16-08-08 | 17496 | 16579 | 18413 | N/A | NO | N/A |

| Parameter | Teilnehmer | Einheit | Proben-Nr. A | Proben-Nr. B | Datum d. Analyse | Ergebnis (Mittel) | Ergebnis A | Ergebnis B | Bestim-mungsgren-ze | Inkl. WF | Wiederfin-dungsrate [%] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Unit** | **Sample No. A** | **Sample No. B** | **Date of analysis** | **Result (Mean)** | **Result A** | **Result B** | **Limit of de-termination** | **Incl. RR** | **Recovery rate** [%] |
| **Vitamin D3** (als Cholecal-ciferol / as cholecalcife-rol) | 1 | µg/100g | 19 | 55 | Jul 16 | 142 | 136 | 147 | 75 | no | n/a |
| | 2 | µg/100g | 4 | 16 | 16-08-12 | 140,93 | 143,21 | 138,65 | 20 | no | 100,7 |
| | 3 | µg/100g | 29 | 46 | 16-07-19 | 146,07 | 148,93 | 143,2 | 0,01 | no | |
| | 4 | µg/100g | 5 | 38 | n/a | | | | | | |
| | 5 | µg/100g | 7 | 36 | 16-06-30 | 123,19 | 126,96 | 119,42 | 16 | no | - |
| | 6 | µg/100g | 10 | 21 | 16-07-13 | 146 | 140 | 152 | 10 | na | na |
| | 7 | µg/100g | 42 | 60 | | | | | | | |
| | 8 | µg/100g | 44 | 48 | 16-07-04 | < LOQ | < LOQ | < LOQ | 400 | no | - |
| | 9 | µg/100g | 13 | 25 | 16-07-15 | 148,5 | 142,9 | 154,1 | | | |
| | 10 | µg/100g | 34 | 51 | | 175,56 | 170,04 | 181,07 | | no | |
| | 11 | µg/100g | 27 | 53 | 16-08-09 | 154 | 156 | 151 | N/A | NO | N/A |

| Parameter | Teilnehmer | Einheit | Proben-Nr. A | Proben-Nr. B | Datum d. Analyse | Ergebnis (Mittel) | Ergebnis A | Ergebnis B | Bestim-mungsgren-ze | Inkl. WF | Wiederfin-dungsrate [%] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Unit** | **Sample No. A** | **Sample No. B** | **Date of analysis** | **Result (Mean)** | **Result A** | **Result B** | **Limit of de-termination** | **Incl. RR** | **Recovery rate** [%] |
| **Vitamin E** (als D-α-To-copherol / as d-α-tocophe-rol) | 1 | mg/100g | 19 | 55 | Jul 16 | 1170 | 1200 | 1140 | 1000 | no | n/a |
| | 2 | mg/100g | 4 | 16 | 16-07-26 | 1181,72 | 1185,85 | 1177,58 | 149 | no | 101,3 |
| | 3 | mg/100g | 29 | 46 | 16-07-19 | 816,22 | 817,22 | 815,21 | 0,5 | no | |
| | 4 | mg/100g | 5 | 38 | n/a | | | | | | |
| | 5 | mg/100g | 7 | 36 | 16-06-30 | 835,69 | 807,23 | 864,15 | 100 | no | - |
| | 6 | mg/100g | 10 | 21 | 16-06-22 | 759 | 784 | 734 | 215 | na | na |
| | 7 | mg/100g | 42 | 60 | Jul 16-21 | 1141,54 | 1144,89 | 1138,19 | 0,35 | yes | 94,21 |
| | 8 | mg/100g | 44 | 48 | 16-07-04 | 839 | 827 | 850 | 63 | no | - |
| | 9 | mg/100g | 13 | 25 | 16-07-06 | 1105 | 1112 | 1098 | | no | 99,2 |
| | 10 | mg/100g | 34 | 51 | | 820,31 | 826,63 | 813,99 | | no | |
| | 11 | mg/100g | 27 | 53 | 16-07-07 | 1210 | 1198 | 1222 | N/A | NO | N/A |

| Parameter | Teilnehmer | Einheit | Proben-Nr. A | Proben-Nr. B | Datum d. Analyse | Ergebnis (Mittel) | Ergebnis A | Ergebnis B | Bestim-mungsgren-ze | Inkl. WF | Wiederfin-dungsrate [%] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Unit** | **Sample No. A** | **Sample No. B** | **Date of analysis** | **Result (Mean)** | **Result A** | **Result B** | **Limit of de-termination** | **Incl. RR** | **Recovery rate** [%] |
| **Vitamin K1** (als Phylloqui-none / as phyl-loquinone) | 1 | µg/100g | 19 | 55 | n/a | n/a | n/a | n/a | n/a | n/a | n/a |
| | 2 | µg/100g | 4 | 16 | 16-07-28 | 945,16 | 934 | 956,32 | 250 | no | 91,8 |
| | 3 | µg/100g | 29 | 46 | 16-07-19 | 1005,22 | 1015,46 | 994,98 | 0,1 | no | |
| | 4 | µg/100g | 5 | 38 | 16-08-05 | 780,85 | 799,85 | 761,85 | 5.4 µg/100g | no | n/a |
| | 5 | µg/100g | 7 | 36 | 16-06-30 | 922,47 | 929,02 | 915,93 | 495 | no | - |
| | 6 | µg/100g | 10 | 21 | | | | | | | |
| | 7 | µg/100g | 42 | 60 | | | | | | | |
| | 8 | µg/100g | 44 | 48 | 16-07-04 | 1035 | 1020 | 1050 | 800 | no | - |
| | 9 | µg/100g | 13 | 25 | | | | | | | |
| | 10 | µg/100g | 34 | 51 | | 1042,89 | 1052,84 | 1032,94 | | no | |
| | 11 | µg/100g | 27 | 53 | 16-07-07 | 800 | 797 | 804 | N/A | NO | N/A |

| Parameter | Teilnehmer | Einheit | Proben-Nr. A | Proben-Nr. B | Datum d. Analyse | Ergebnis (Mittel) | Ergebnis A | Ergebnis B | Bestim-mungsgren-ze | Inkl. WF | Wiederfin-dungsrate [%] |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Unit** | **Sample No. A** | **Sample No. B** | **Date of analysis** | **Result (Mean)** | **Result A** | **Result B** | **Limit of de-termination** | **Incl. RR** | **Recovery rate** [%] |
| **β-Carotene** (als β-Carotin, ohne andere Provitamine / as β-carotene, without other provitamins) | 1 | mg/100g | 19 | 55 | Jul 16 | 33,1 | 33,3 | 32,9 | 0,01 | no | n/a |
| | 2 | mg/100g | 4 | 16 | 16-07-13 | 43,83 | 43,31 | 44,35 | 0,08 | no | 99,3 |
| | 3 | mg/100g | 29 | 46 | 16-07-20 | 23,73 | 22,67 | 24,78 | 0,01 | no | |
| | 4 | mg/100g | 5 | 38 | n/a | | | | | | |
| | 5 | mg/100g | 7 | 36 | - | - | - | - | - | - | - |
| | 6 | mg/100g | 10 | 21 | | | | | | | |
| | 7 | mg/100g | 42 | 60 | Jun 22-23 | 21,13 | 21,58 | 20,68 | 3 | yes | 127,73 |
| | 8 | mg/100g | 44 | 48 | | - | - | - | - | | |
| | 9 | mg/100g | 13 | 25 | 16-07-13 | 33,5 | 34 | 33 | | no | 98,5 |
| | 10 | mg/100g | 34 | 51 | | | | | | | |
| | 11 | mg/100g | 27 | 53 | 16-08-02 | 38 | 39 | 37 | N/A | NO | N/A |

## 5.2 Homogeneity

### 5.2.1 Homogeneity of bottled PT-samples

Homogeneity test of niacin by HPLC-UV:

| Independant samples | mg/100g |
|---|---|
| 1 | 16,2 |
| 2 | 16,3 |
| 3 | 16,3 |
| 4 | 16,1 |
| 5 | 16,3 |
| 6 | 16,0 |
| 7 | 16,3 |
| 8 | 16,4 |

| | | |
|---|---|---|
| Mean | 16,2 | |
| Repeatability Standard Deviation | 0,130 | 0,80% |

### 5.2.2 Comparison of sample numbers / test results and trend line

By comparison of the increasing sample numbers and the measurement results of participants, the homogeneity of the chronological bottled PT item can be characterized with the help of the trend line function:

| **Vitamin A** | without participants 5 + 11* | | | |
|---|---|---|---|---|
| Target standard deviation $\sigma_{pt}$ | 1556 | | | µg/100g |
| Sample numbers | 4 - 60 | | | |
| Total numbers of samples | 16 | | | |
| Slope | 27,0 | | | |
| Trend line range | 21815 | - | 22247 | µg/100g |
| Deviation trend line | 22031 | ± | 216 | µg/100g |
| **Percent of σpt** | 13,9 | % | | |

* lowest and highest results respectively

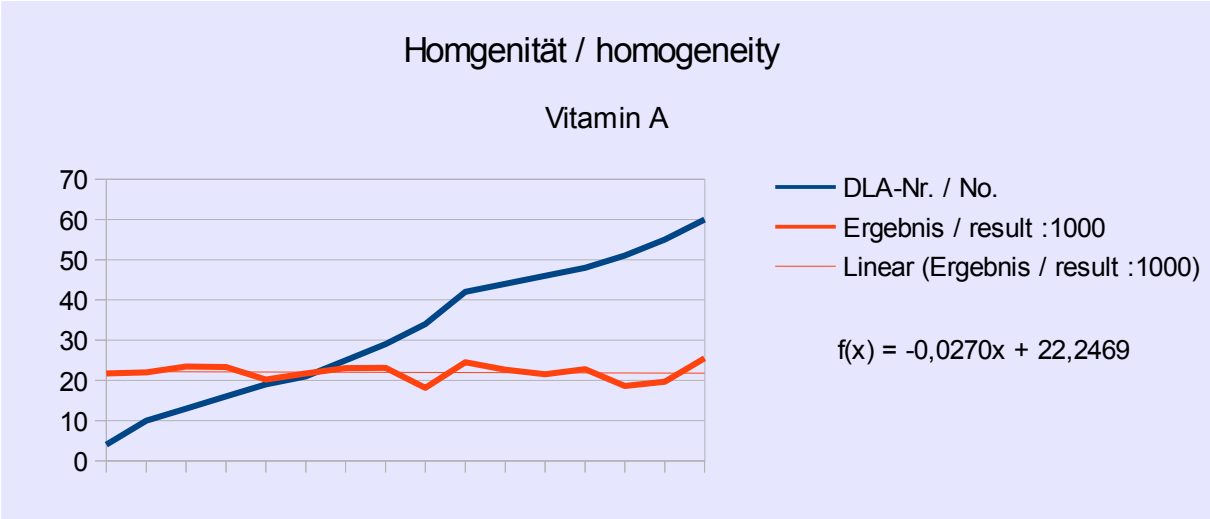


**Abb. 13:** Trendfunktion Probennummern / Vitamin A Ergebnisse (1/1000 dargestellt)
**Fig. 13:** trend line function sample number / vitamin A results (1/1000 shown)

| **Vitamin E** | | | | |
|---|---|---|---|---|
| Target standard deviation $\sigma_{pt}$ | 125 | | | mg/100g |
| Sample numbers | 4 - 60 | | | |
| Total numbers of samples | 20 | | | |
| Slope | 0,206 | | | |
| Trend line range | 986 | - | 990 | mg/100g |
| Deviation trend line | 988 | ± | 2,00 | mg/100g |
| **Percent of σpt** | 1,6 | % | | |

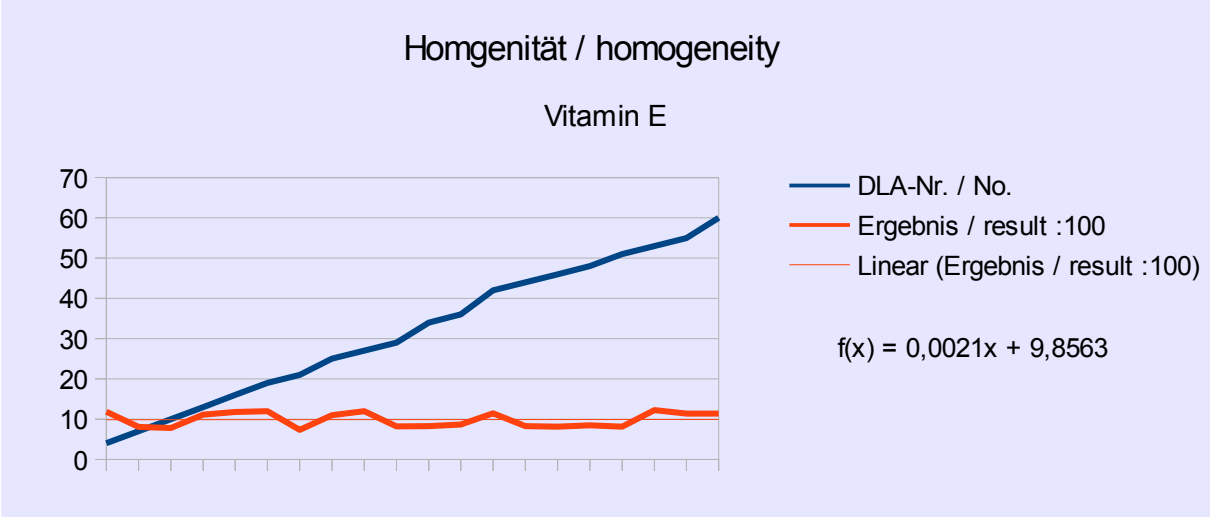


**Abb. 14:** Trendfunktion Probennummern / Vitamin E Ergebnisse (:100 dargestellt)
**Fig. 14:** trend line function sample number / Vitamin E results (:100 shown)

## 5.3 Analytical Methods

*Details by the participants*

| Parameter | Teilnehmer | Methodenbeschreibung | Wiederfindung mit gleicher Matrix | Methode akkreditiert | Sonstige Hinweise |
|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Method description** | **Recovery with same matrix** | **Method accredited** | **Further remarks** |
| **Vitamin A** (als Retinol ohne Provitamine / as retinol without provitamins) | 1 | HPLC | no | no | |
| | 2 | PV 2.020/002-03 | yes | yes | --- |
| | 3 | HPLC (SOP M840) | no | yes | |
| | 4 | | | | |
| | 5 | HPLC-DAD | no | no | - |
| | 6 | UPLC-DAD | n.a. | yes | |
| | 7 | ASU L 49.00-3, HPLC-DAD | yes | yes | |
| | 8 | HPLC-DAD | - | yes | |
| | 9 | Vitamin A and E; Determination in food and food supplements by HPLC according to ASU §64 method, modified saponification temperature | yes | yes | |
| | 10 | | no | yes | |
| | 11 | RP-HPLC | NO | YES | |

| Parameter | Teilnehmer | Methodenbeschreibung | Wiederfindung mit gleicher Matrix | Methode akkreditiert | Sonstige Hinweise |
|---|---|---|---|---|---|
| Analyte | Participant | Method description | Recovery with same matrix | Method accredited | Further remarks |
| **Vitamin D3** (als Cholecalciferol / as cholecalciferol) | 1 | HPLC | no | yes | |
| | 2 | PV 2.020/004-03 | yes | yes | --- |
| | 3 | LC-MS/MS (SOP M2885) | no | no | |
| | 4 | | | | |
| | 5 | HPLC-DAD | no | no | - |
| | 6 | UPLC-MS/MS | no | yes | |
| | 7 | | | | |
| | 8 | HPLC-DAD | - | yes | |
| | 9 | LC/MS/MS | | no | |
| | 10 | | no | yes | |
| | 11 | NP-HPLC | no | yes | |

| Parameter | Teilnehmer | Methodenbeschreibung | Wiederfindung mit gleicher Matrix | Methode akkreditiert | Sonstige Hinweise |
|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Method description** | **Recovery with same matrix** | **Method accredited** | **Further remarks** |
| **Vitamin E** (als D-α-Tocopherol / as d-α-tocopherol) | 1 | HPLC | no | yes | |
| | 2 | PV 2.020/003-03 | yes | yes | --- |
| | 3 | HPLC (SOP M840) | no | yes | |
| | 4 | | | | |
| | 5 | HPLC-DAD | no | no | - |
| | 6 | UPLC-DAD | no | yes | |
| | 7 | ASU L 49.00-5, HPLC-DAD | yes | yes | |
| | 8 | HPLC-DAD | - | yes | |
| | 9 | Vitamin A and E; Determination in food and food supplements by HPLC according to ASU §64 method, modified saponification temperature | yes | yes | |
| | 10 | | no | yes | |
| | 11 | RP-HPLC | no | yes | |

| Parameter | Teilnehmer | Methodenbeschreibung | Wiederfindung mit gleicher Matrix | Methode akkreditiert | Sonstige Hinweise |
|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Method description** | **Recovery with same matrix** | **Method accredited** | **Further remarks** |
| **Vitamin K1** (als Phylloquinone / as phylloquinone) | 1 | n/a | n/a | n/a | |
| | 2 | PV 2.019/019-01 | yes | yes | --- |
| | 3 | HPLC (SOP M2986) | no | yes | |
| | 4 | Based on BS EN14148:2003, after enzymatic removal of fat from the sample vitamin K1 is determined in an appropriate sample solution by HPLC separation coupled with post column reduction and subsequent fluorometric detection. Vitamin K1 isomers are quantified as a single unresolved peak with a C18 column | no | yes | method is accredited for infant formula, controls used was IF. Sample results were outside our working range, samples were diluted and re-injected. |
| | 5 | HPLC-DAD | no | no | - |
| | 6 | | | | |
| | 7 | | | | |
| | 8 | HPLC-DAD | - | yes | |
| | 9 | | | | |
| | 10 | | no | yes | |
| | 11 | RP-HPLC | no | no | |

| Parameter | Teilnehmer | Methodenbeschreibung | Wiederfindung mit gleicher Matrix | Methode akkreditiert | Sonstige Hinweise |
|---|---|---|---|---|---|
| **Analyte** | **Participant** | **Method description** | **Recovery with same matrix** | **Method accredited** | **Further remarks** |
| **β-Carotene** (als β-Carotin, ohne andere Provitamine / as β-carotene, without other provitamins) | 1 | HPLC | yes | yes | |
| | 2 | PV 2.019/012-03 | yes | yes | --- |
| | 3 | HPLC (SOP M932) | no | yes | |
| | 4 | | | | |
| | 5 | - | - | - | - |
| | 6 | | | | |
| | 7 | LAV 21.0055-02, HPLC-DAD | yes | yes | |
| | 8 | | | | |
| | 9 | Photometric determination of total carotinoides and beta-carotenes in food and food supplements | yes | yes | within june 13th and june 28th analysed values showed a decreasing tendency |
| | 10 | | | | |
| | 11 | RP-HPLC | no | yes | |

# 6. Index of participant laboratories in alphabetical order

| Teilnehmer / Participant | Ort / Town | Land / Country |
|---|---|---|
| | | UNITED KINGDOM |
| | | Germany |
| | | Germany |
| | | Germany |
| | | IRELAND |
| | | Germany |
| | | Germany |
| | | Germany |
| | | USA |
| | | SWEDEN |
| | | 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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17. ASU § 64 LFGB L 00.00-62 / DIN EN 12822:2014: Bestimmung von Vitamin E (α-, β-, γ- und δ-Tocopherol) in Lebensmitteln mittels HPLC / Foodstuffs. Determination of vitamin E by high performance liquid chromatography. Measurement of α-, β-, γ- and δ-tocopherol
18. ASU § 64 LFGB L 00.00-63/1 / DIN EN 12823-1:2014: Bestimmung von Vitamin A in Lebensmitteln mittels HPLC, Teil 1: Bestimmung von all-trans-Retinol und 13-cis-Retinol / Foodstuffs. Determination of vitamin A by high performance liquid chromatography. Measurement of all-E-retinol and 13-Z-retinol
19. ASU § 64 LFGB L 00.00-63/2 / DIN EN 12823-2:2000: Bestimmung von Vitamin A in Lebensmitteln mittels HPLC, Teil 2: Bestimmung von β-Carotin / Food-

stuffs. Determination of vitamin A by high performance liquid chromatography. Measurement of β-carotene
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