

PRODUCT INFORMATION SHEET

Almond, Brazil Nuts, Cashew, Coconut, Hazelnut, Macadamia, Pecan, Pine Nuts, Pistachio, Walnut (4 Samples qualitative - Detection of "Action Levels" included)

PT originated REFERENCE MATERIAL (RM)

Description of the RM

Name / Material of related PT	DLA ptALS1 (2025) Allergen Screening I - 4 Samples qualitative: Almond, Brazil Nuts, Cashew, Coconut, Hazelnut, Macadamia, Pecan, Pine Nuts, Pistachio, Walnut - Samples 1-4
Identifier / LOT	RM 2025-ALS1 Set
Expiry date	June 2029
Matrix	Allergens in simple matrix (see tables)
Physical description	Powder
Packaging / weight	Sealed foile poche / 20 g
Storage conditions	2-10°C (dry and dark)
Name and address manufacturer	DLA - Proficiency Tests GmbH Hauptstr. 80, 23845 Oering, Germany Tel. ++49-(0)4532-9183358, Fax. ++49(0)4102-9944976 eMail. pt@dla-lvu.de, website. www.dla-lvu.de

Assigned values and uncertainties of 2/3 and 1/6 "action levels":

The "action level" in mg/kg is calculated from the allergen protein reference dose per 100 g food portion (based on FAO/WHO 2022 and Vital 4.0 2024 (for more details see final report)). The contents of the allergenic items in the PT-samples 1-4 were gravimetrically adjusted to 2/3 and 1/6 of the "action levels" (s. Tab. 2).

Each assigned value, here the spiked allergen-contents, is afflicted with a standard uncertainty. As uncertainties the following factors were considered: gravimetric weighing, protein contents, homogeneity and stability of allergen protein.

All uncertainties were expressed in the form of their standard deviations and then added as variances. The square root from the sum of the total variances results in the combined uncertainty "Uc". Multiplied with the coverage factor k=2 the extended uncertainties of the assigned values " $U(X_{pt})$ " are obtained (for more details see final report).

For all parameters the extended combined uncertainties (k=2) of the "2/3 action levels" and "1/6 action levels" are in the range of $U(X_{pt}) = \pm 5-10 \%$.

Interpretation of 2/3 and 1/6 Action Level Detection

A positive detection of the 2/3 action level enables a reliable decision as to whether a measurement result is below the action level, even if the measurement uncertainty of the analytical method is about $\pm 50\%$.

The positive detection of the 1/6 action level can demonstrate the suitability of the applied method at lower values close to the LOQ or LOD.

Composition of Material

Ingredients	Amounts
Rice flour, organic	74 - 76 %
Maltodextrin	24 - 26 %
Allergen-Premixes	0,05 - 0,42 %
Ingredients: Maltodextrin (88% - 97%), Silicon dioxide (1% - 2%) and Allergens (2,2% - 10% each)	

Added allergenic ingredients 2/3 and 1/6 values of the "action levels" are indicated as actually spiked total food item and calculated protein contents in mg/kg

Ingredients *	"Action Level"	Sample 1	Sample 2	Sample 3	Sample 4
Protein % (Kf = Kjeldahl factor) **	Vital 4.0	[mg/kg]	[mg/kg]	[mg/kg]	[mg/kg]
Almond: roasted, milled Protein 21,1% (Kf 5,18) (mixture 23 products from USA, Europe, Australia, Western Asia)	Protein 10 mg/kg	2/3 Level Food 31,2 Protein 6,58	negative	negative	1/6 Level Food 7,88 Protein 1,66
Brazil Nut: milled Protein 12,9% (Kf 5,46) (1 product)	Protein 10 mg/kg	negative	2/3 Level Food 51,2 Protein 6,60	1/6 Level Food 12,9 Protein 1,66	negative
Cashew: roasted, milled Protein 18,5% (Kf 5,30) (mixture 9 products from Asia and unknown origin)	Protein 10 mg/kg	2/3 Level Food 35,7 Protein 6,60	negative	1/6 Level Food 9,05 Protein 1,67	negative
Coconut: flours Protein 15,9% (Kf 5,30) (mixture 6 products from Asia and unknown origin)	Protein 10 mg/kg #	2/3 Level Food 40,9 Protein 6,50	1/6 Level Food 10,8 Protein 1,71	negative	negative
Hazelnut: roasted, milled Protein 14,1% (Kf 5,30) (mixture 10 products from 5 countries, Europe)	Protein 30 mg/kg	negative	negative	1/6 Level Food 35,8 Protein 5,05	2/3 Level Food 141 Protein 19,9
Macadamia: roasted, milled Protein 8,28% (Kf 5,30) (mixture 7 products from Kenia and Australia)	Protein 10 mg/kg	negative	1/6 Level Food 20,2 Protein 1,67	2/3 Level Food 80,3 Protein 6,65	negative
Pecan: raw, milled Protein 9,86% (Kf 5,30) (mixture 8 products from USA, Mexico and unknown)	Protein 10 mg/kg	2/3 Level Food 67,2 Protein 6,63	negative	negative	1/6 Level Food 16,8 Protein 1,66
Pine Nuts: roasted, milled Protein 25,1% (Kf 6,25) (mixture 6 products from Pakistan, Russia, Italy, Spain and unknown)	Protein 10 mg/kg	2/3 Level Food 26,3 Protein 6,60	negative	1/6 Level Food 6,66 Protein 1,67	negative
Pistachio: roasted, milled Protein 21,1% (Kf 5,30) (mixture (10 products from USA, Iran, Turkey, Greece, Italy, Spain)	Protein 10 mg/kg	negative	1/6 Level Food 7,80 Protein 1,66	negative	2/3 Level Food 31,2 Protein 6,58
Walnut: roasted, milled Protein 15,8% (Kf 5,30) (mixture 11 products from North America, South America, Europe and unknown)	Protein 10 mg/kg	negative	2/3 Level Food 41,9 Protein 6,62	1/6 Level Food 10,5 Protein 1,66	negative

* Allergen contents of „food item“ as indicated in the column of ingredients according gravimetric mixing

**Protein contents according to laboratory analysis (total nitrogen, Kjeldahl)

No action level available for coconut (value based on values of tree nuts)

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

The detectability or absence of the allergens was tested by DLA using lateral flow assays. The results are in agreement with the spiking of the PT samples 1-4.

Minimum Sample Size / Homogeneity

A minimum sample size of 5 g is recommended for sample preparation.

The homogeneity tests were performed with sample sizes of 5 g (Microtracer analysis).

The mixture homogeneity was examined 10-fold by microtracer analysis. Homogeneity was sufficient with a probability of $\geq 25\%$ (excellent mixture) and a HorRat value in the range of acceptance (0,3 to 1,3) (for more details see final report of PT).

Reference Values

All reference values were derived from the related proficiency test (PT) round (for more details see final report).

Reference Values qualitative (Consensus Values*)

Methods	Sample 1	Sample 2	Sample 3	Sample 4
Almond	2/3 Level			1/6 Level
Almond ELISA / LFD	positive	negative	negative	positive
Almond PCR	positive	negative	negative	none**
Brazil Nut		2/3 Level	1/6 Level	
Brazil Nut ELISA / LFD	negative	positive	positive	negative
Peanut PCR	negative	positive	positive	negative
Cashew	2/3 Level		1/6 Level	
Cashew ELISA / LFD	positive	negative	positive	negative***
Cashew PCR	positive	negative	positive	negative
Coconut	2/3 Level	1/6 Level		
Coconut ELISA / LFD	positive	positive	negative	negative
Celery PCR	positive	positive	negative	negative
Hazelnut			1/6 Level	2/3 Level
Hazelnut ELISA / LFD	negative	none**	positive	positive
Hazelnut PCR	negative	negative	positive	positive
Macadamia		1/6 Level	2/3 Level	
Macadamia ELISA / LFD	negative	positive	positive	negative
Macadamia PCR	negative	positive	positive	negative
Pecan	2/3 Level			1/6 Level
Pecan ELISA / LFD	positive	positive****	positive****	positive
Pecan PCR	positive	negative	negative	positive
Pine Nuts	2/3 Level		1/6 Level	
Pine Nuts ELISA / LFD	positive	negative	positive	negative
Pine Nuts PCR	positive	negative	positive	negative
Pistachio		1/6 Level		2/3 Level
Pistachio ELISA / LFD	none**	positive	none**	positive
Pistachio PCR	negative	positive	negative	positive
Walnut		2/3 Level	1/6 Level	
Walnut ELISA / LFD	none**	positive	positive	negative
Walnut PCR	negative	positive	positive	negative

* Consensus values $\geq 75\%$ positive or negative results of PT-participants

** Due to the number of $< 75\%$ concordant results, no consensus value could be set

*** Several participants obtained a positive result for sample 4 which contains pistachio (without addition of cashew).

**** For samples 2 and 3 without addition of pecan also positive consensus values were obtained. Several participants indicated cross-reactivity to walnut which is contained in samples 2 and 3.

Note: Some unexpected positive results can be due to possible cross-reactivities of the applied methods (for more details see final report). Possible cross-reactivities should be documented in the manufacturer's test kit information.

Intended Use

- for laboratory use only, no food!
- validation of analytical methods
- control sample for quality control charts
- test material for inter-laboratory comparisons

Storage Instructions and Handling Information

The material should be stored as stated on page 1. Before use of the material should be warmed up to room temperature. The content should be mixed thoroughly prior to subsampling (see minimum sample size above). The material should be weighed immediately after opening of the foil poche/bottle and closed again afterwards in order to avoid uptake of moisture. Frequent freezing and thawing should be avoided as this can lead to changes in the properties of the material. The expiry date of this RM is based on the current knowledge and applies only for proper storage conditions in the originally closed foil poche/bottle.

Safety Precautions

The usual laboratory safety precautions should be applied when working with the RM considering the type of ingredients, parameters and its concentration.

Data Source

The material was analyzed by 31 laboratories within the related proficiency test (PT). Different analytical methods have been used.

Various

The user must determine the suitability of this RM for its particular purpose. DLA makes no warranty of any kind, expressed or implied, other than its products meet all quality control standards set by DLA. We do not guarantee this product can be used for a special application.



approved (PT-Coordinator)	updated
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