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9th International Conference on Food Digestion, 19-21 May 2026, Gdansk (Poland)

Behind the scenes of INFOGEST Quant: insights into the step-by-step protocol for protein digestibility and DIAAS determination

Ana Blanco-Doval, **Lotti Egger**, Raquel Sousa, Cédric Brügger, Eliane Binz, André Brodkorb, Didier Dupont, Isidra Recio, and **Reto Portmann**
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nature protocols

https://doi.org/10.1038/s41596-025-01307-9

Protocol extension

Check for updates

INFOGEST Quant: standardized in vitro determination of digestibility and DIAAS of dietary proteins based on the INFOGEST static digestion model

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The INFOGEST Quant protocol

nature protocols

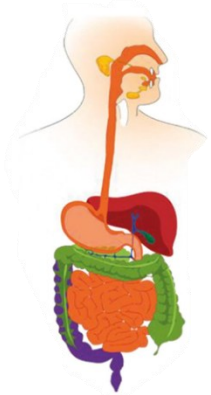
https://doi.org/10.1038/s41596-025-01307-9

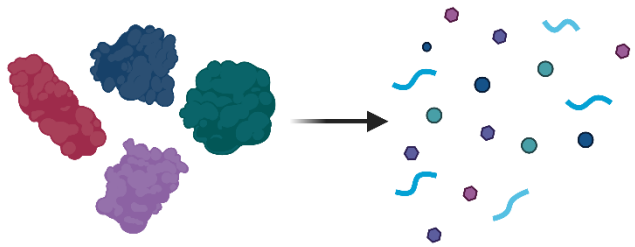
Protocol extension

Check for updates

INFOGEST Quant: standardized in vitro determination of digestibility and DIAAS of dietary proteins based on the INFOGEST static digestion model

Lotti Egger[1,5], Ana Blanco-Doval[1,5], Raquel Sousa[1], Cédric Brügger[1], Eliane Binz[1], André Brodkorb[2], Didier Dupont[3], Isidra Recio[4] & Reto Portmann[1,5]





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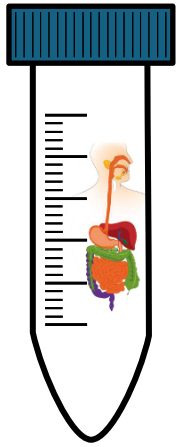
The INFOGEST Quant protocol – *overview*

~ INFOGEST 2.0

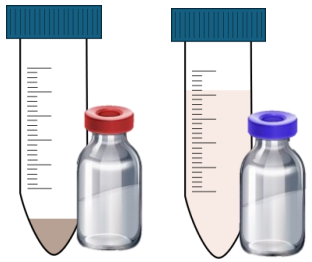


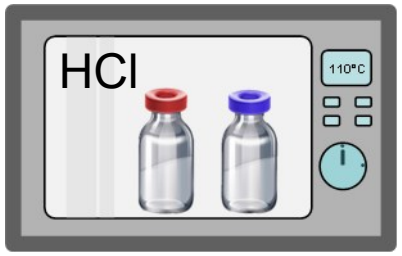
MeOH

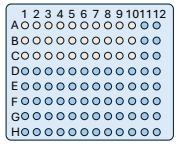
Digesta

Supernatant

Pellet







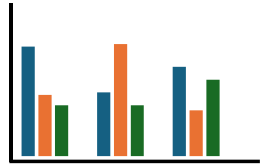
Total amino acids (*HPLC-UV*)

Total amino groups (*OPA method*)

Total nitrogen (*Kjeldahl*)

DIAAS

Proxy DIAAS



Total protein digestibility

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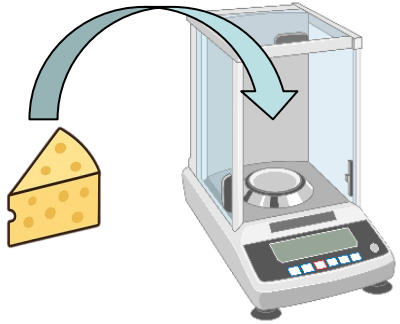
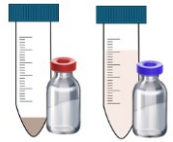
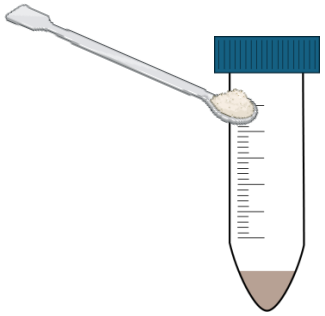
Critical points of the INFOGEST Quant method

Sample amount for IVD

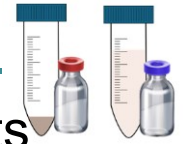


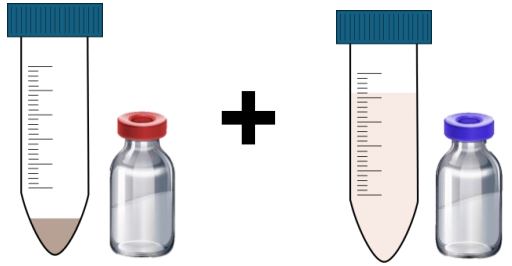
Pellet aliquots





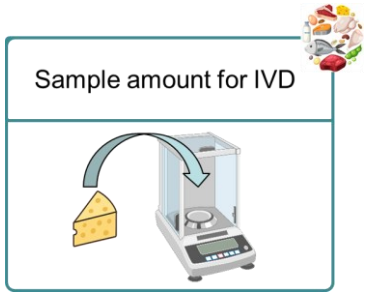
Analysis of supernatants and pellets



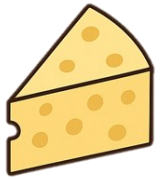


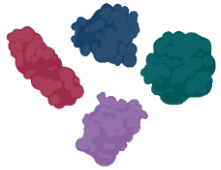
Critical points of the INFOGEST Quant method

Sample amount for IVD

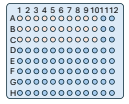


The amount of sample for digestion is adjusted to **40 mg of protein** in order to normalize the protein-to-enzyme ratio among experiments.

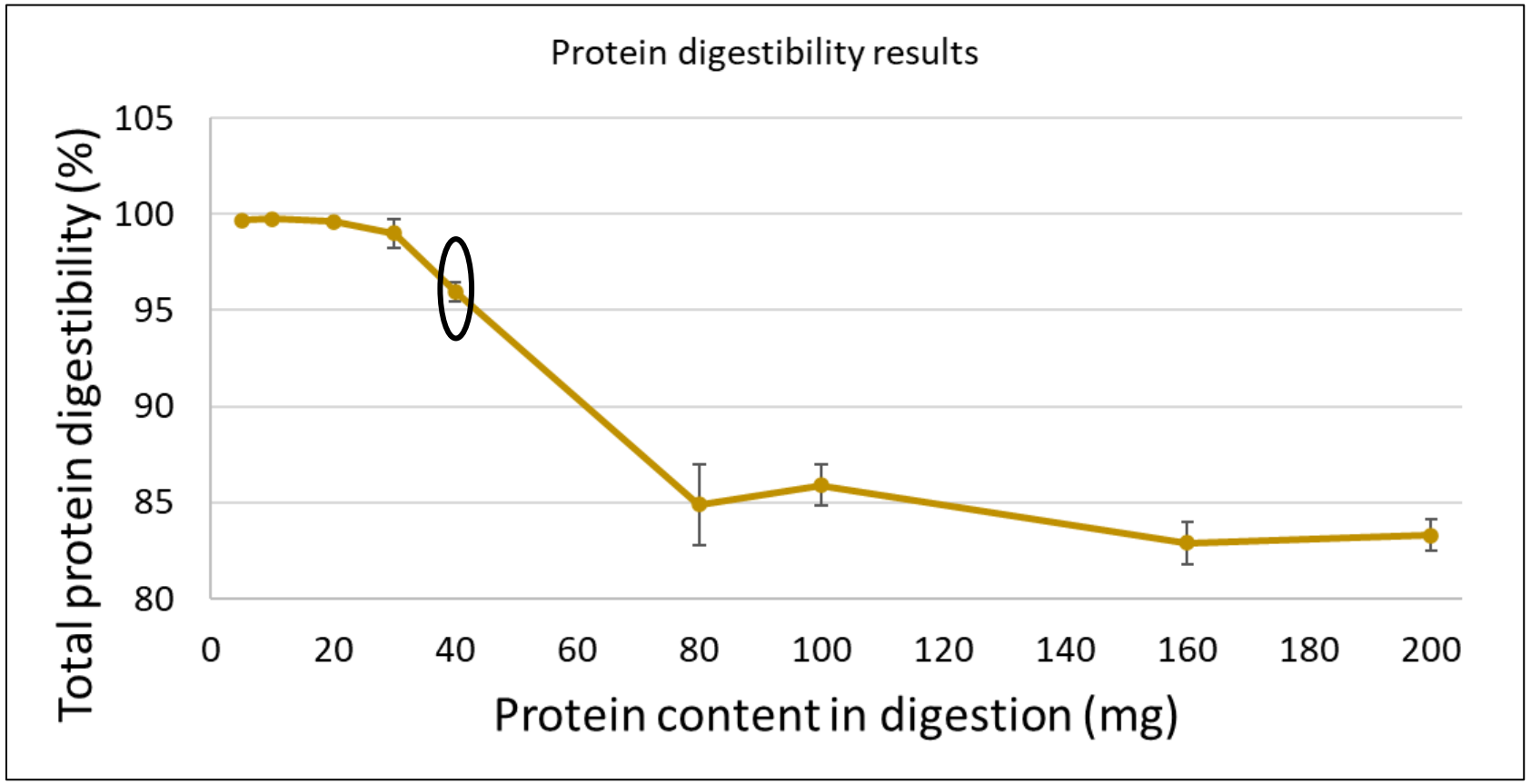




5 mg	**80** mg
10 mg	**100** mg
20 mg	**160** mg
30 mg	**200** mg
40 mg	



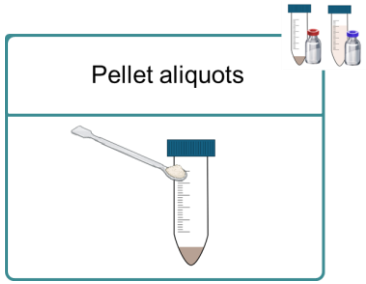
Total amino acids (*HPLC-UV*)



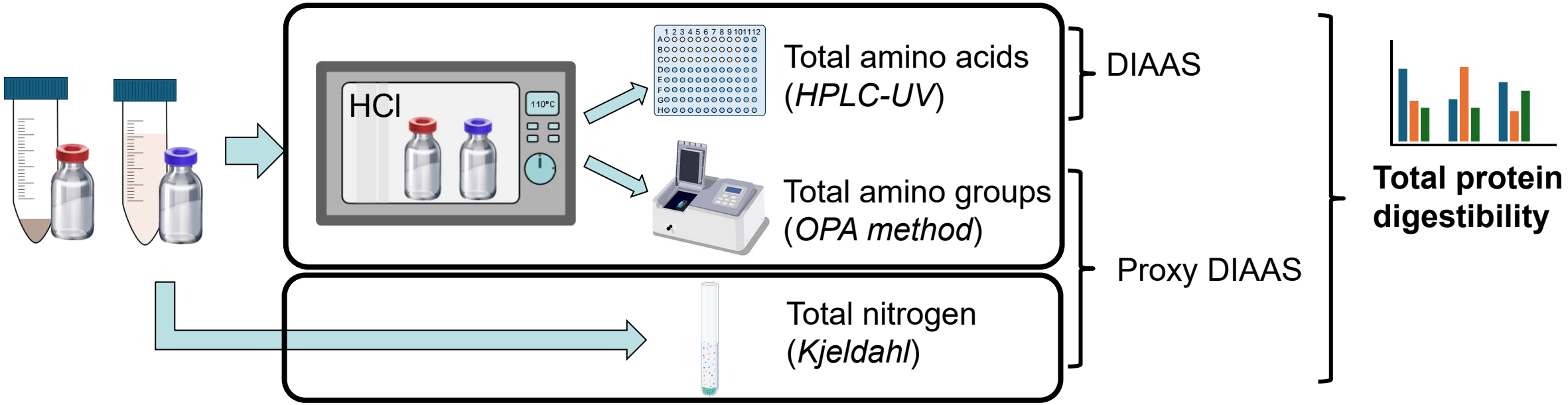
1 g food → 40 mg protein

Critical points of the INFOGEST Quant method

Pellet aliquots

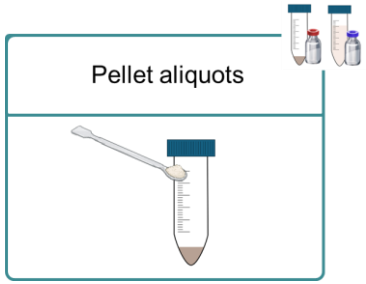


The **pellets** (non-absorbable fractions) **must not be aliquoted** for various analytical workflows; instead, the whole pellets are used for chemical analysis.



Critical points of the INFOGEST Quant method

Pellet aliquots



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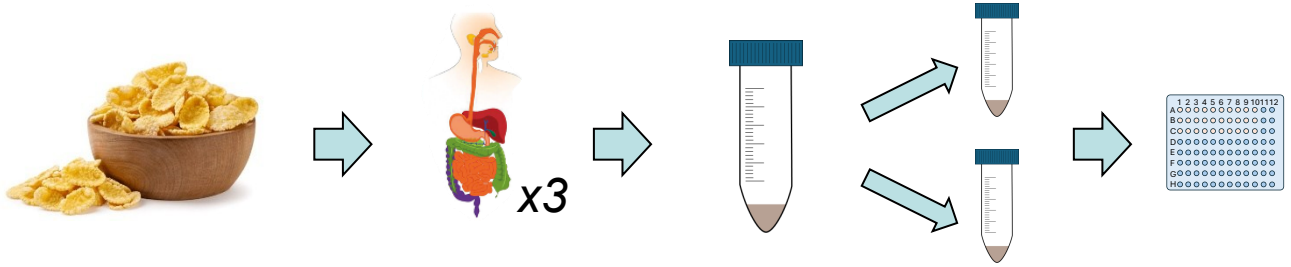
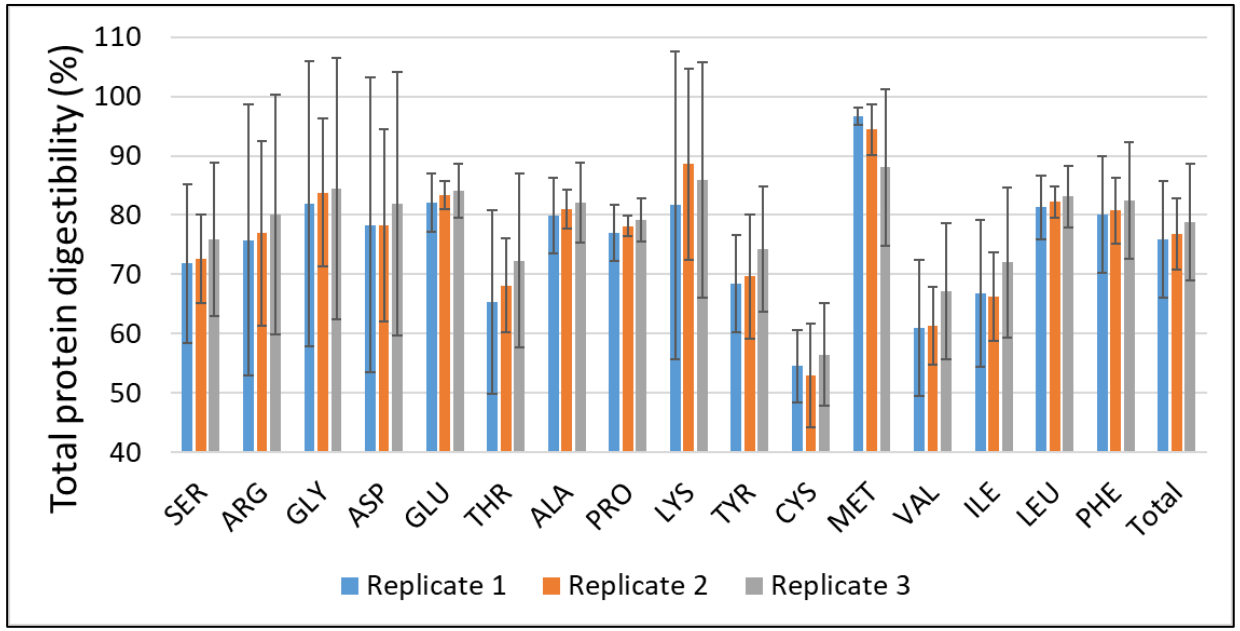
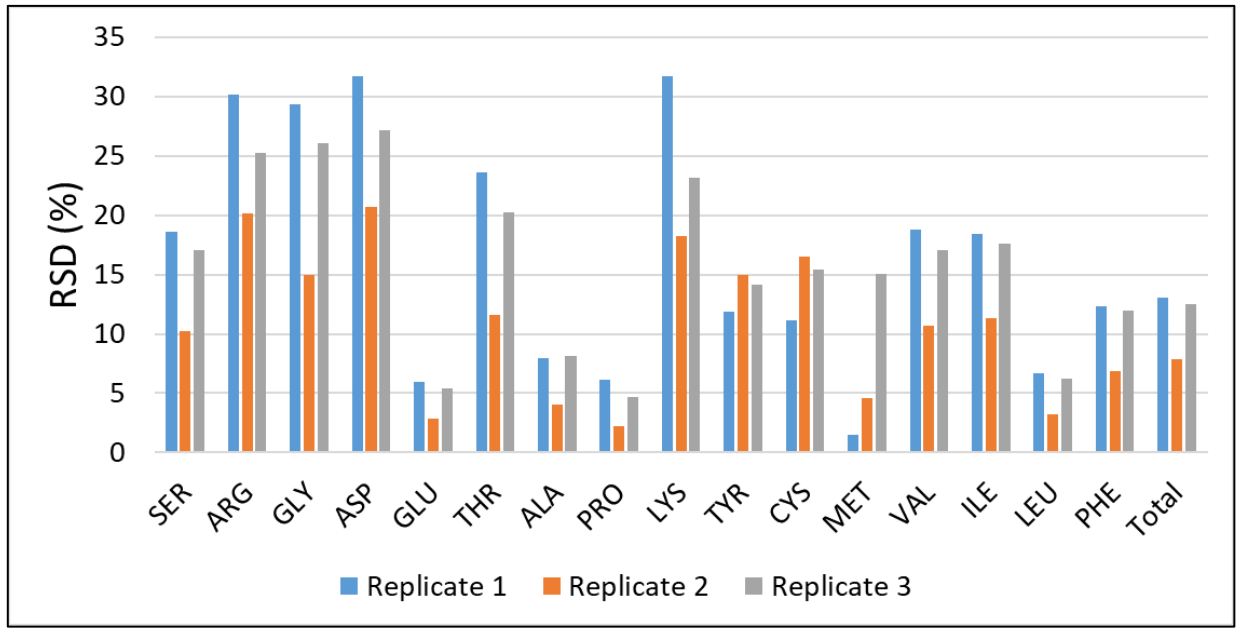


Total amino acids (*HPLC-UV*)

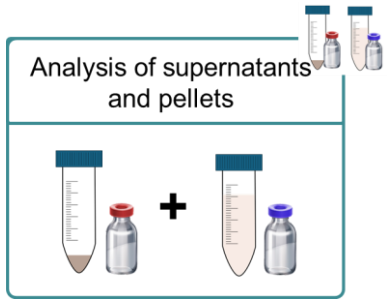




IMPORTANT NOTE: *The pellet is not homogeneous.*

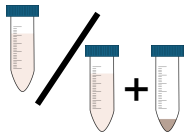
Critical points of the INFOGEST Quant method

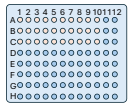
Analysis of supernatant and pellets



Calculations are done based on results in the **absorbable fraction** in relation to the **sum of the absorbable and non-absorbable fractions**.

$$In\ vitro\ digestibility\ [\%] = \frac{(FS - CS)}{(FS - CS) + \max(0; FP - CP)} \times 100 =$$

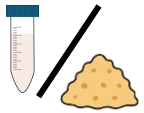
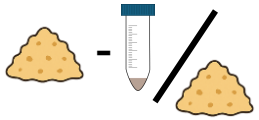
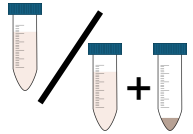




Input

Supernatant

Pellet

	Milk (pasteurised)	**Sheep milk**	**Quark**	**Quinoa** (cooked)	**Chia** (ground)	**Chia** (hydrated)
	83.3	84.3	93.1	58.8	33.1	0.2
	99.9	99.8	99.9	87.1	83.2	41.4
	99.9	99.8	99.9	82.0	66.3	0.4
Recovery (%)	83.4	84.5	93.2	71.7	49.8	58.9

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Critical points of the INFOGEST Quant method

Other points

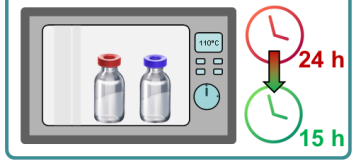
Enzyme blank

The enzyme blank tube to correct for enzyme autolysis must include a **protein-free substrate** (e.g. a protein-free cookie).

Sample size

Mastication is simulated by grinding the solid samples to a particle size of **2-3 mm** (based on *in vivo* data).

15 h acid hydrolysis



The **acid hydrolysis** of the digested samples lasts **15 h** instead of 24 h, as done in the food material.

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ICF Gdańsk 2026

Behind the scenes of INFOGEST Quant: insights into the step-by-step protocol for protein digestibility and DIAAS determination

Thank you for your attention

Biochemistry of milk and microorganisms

INFOGEST

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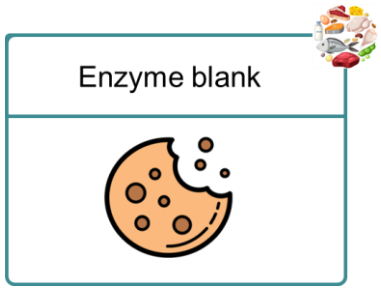
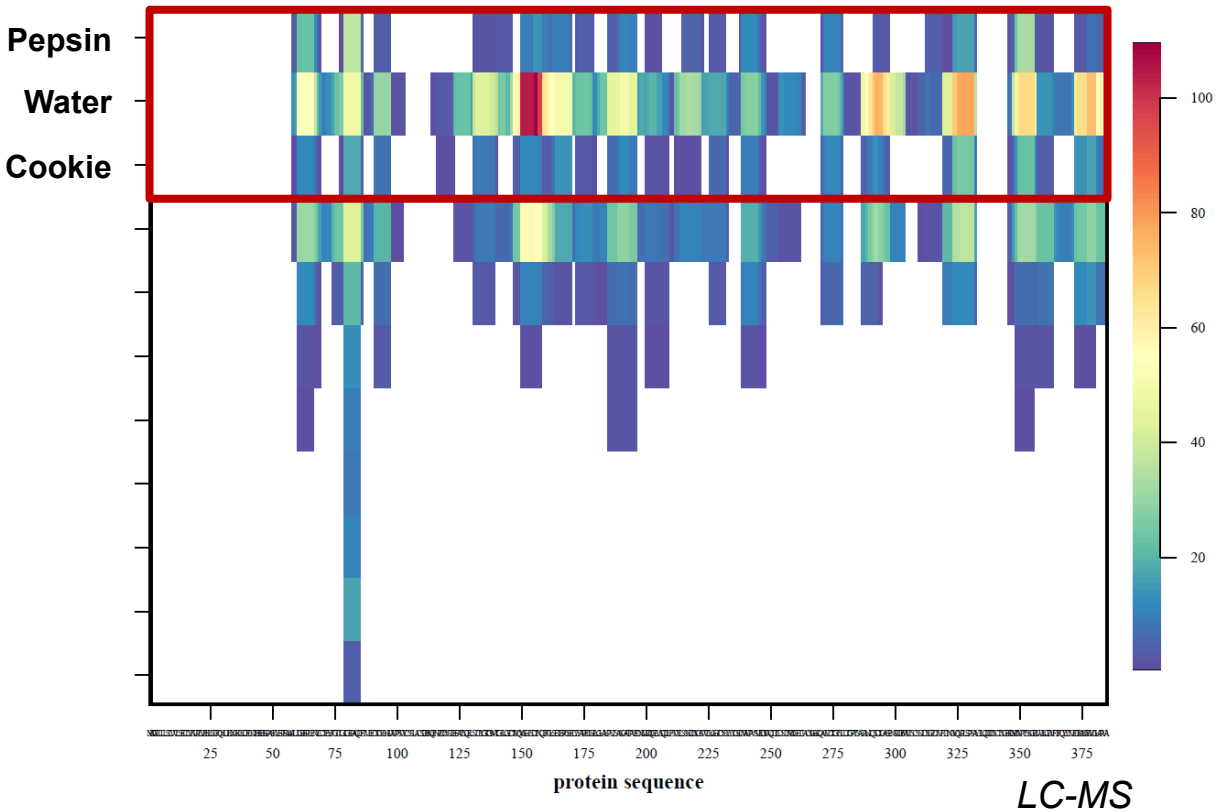
Critical points of the INFOGEST Quant method

Enzyme blank

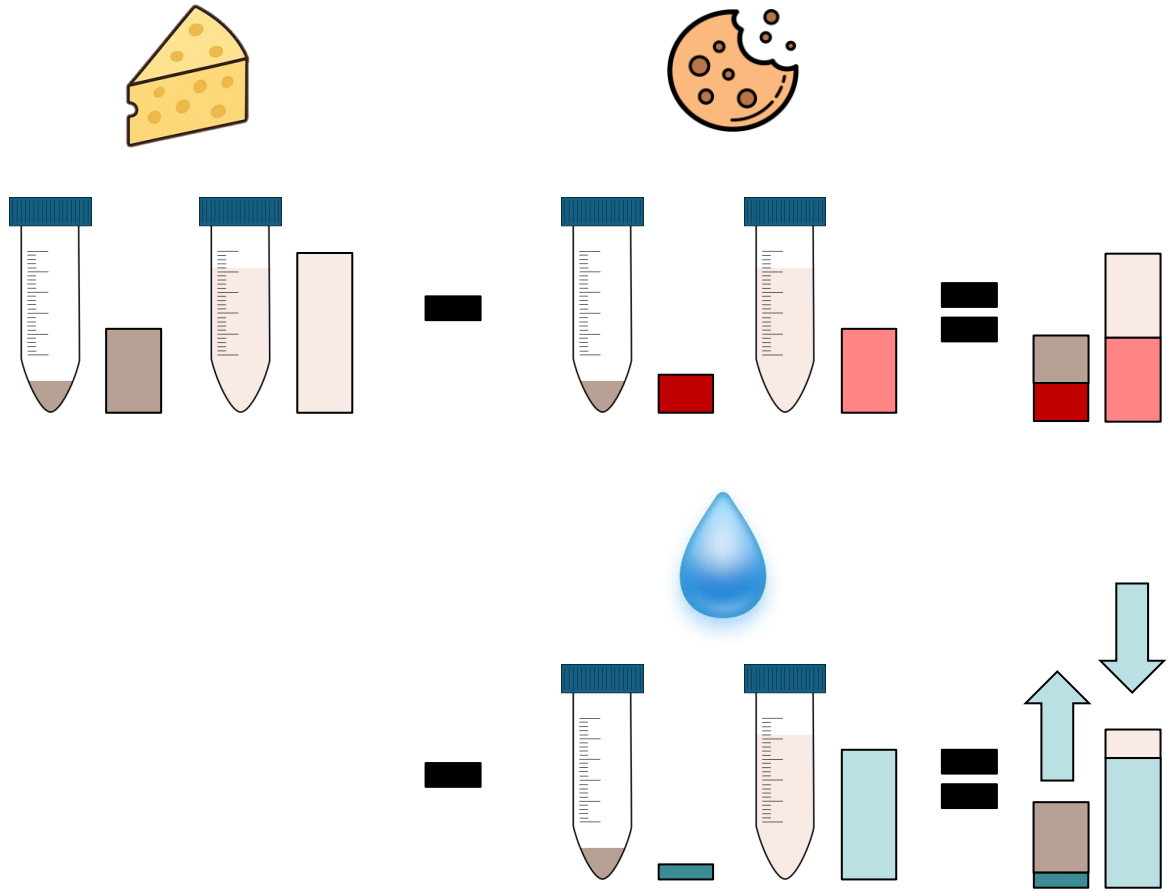


The enzyme blank tube to correct for enzyme autolysis must include a **protein-free substrate** (e.g. a protein-free cookie).

Pepsin – Gastric phase 120 min



LC-MS

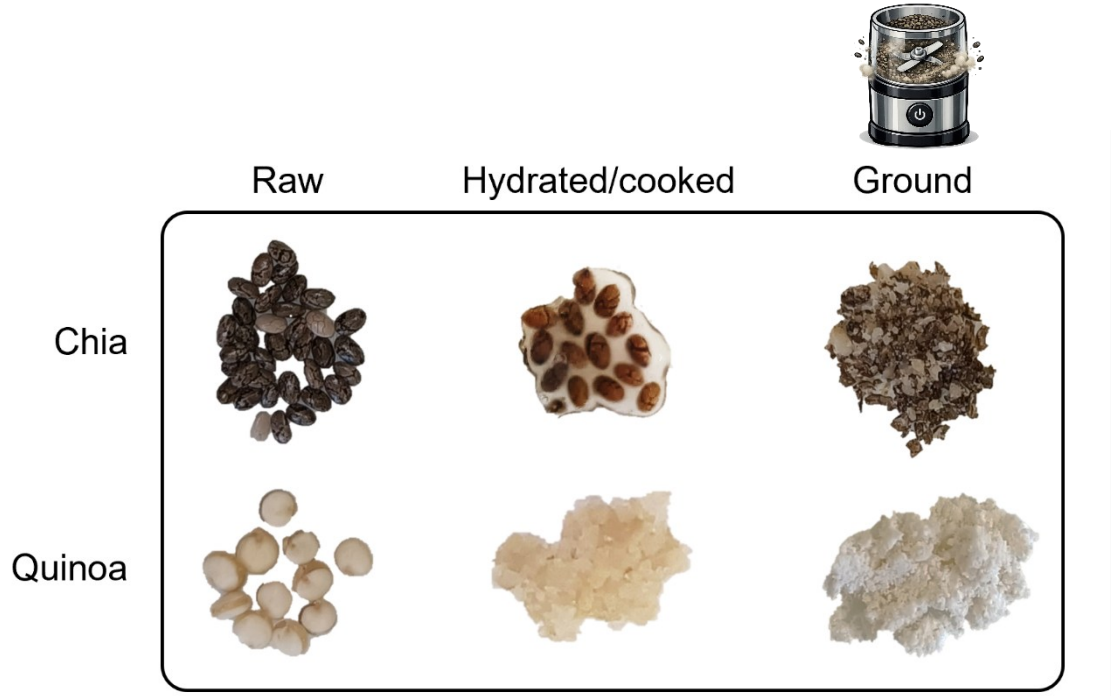


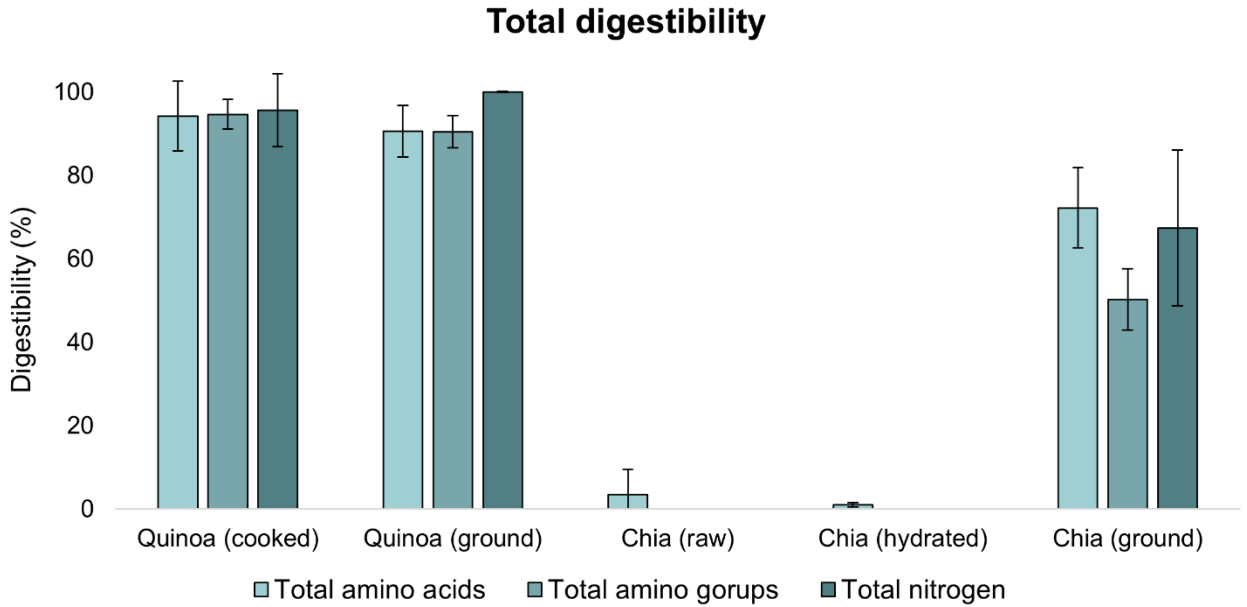
Underestimation of digestibility

Critical points of the INFOGEST Quant method

Sample size

Mastication is simulated by grinding the solid samples to a particle size of **2-3 mm** (based on *in vivo* data).

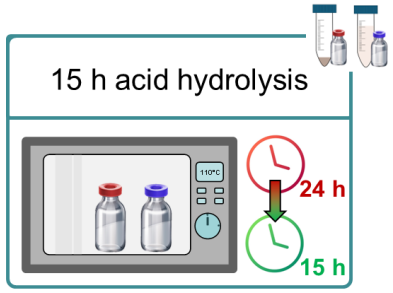




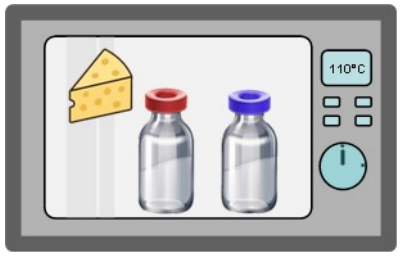
IMPORTANT NOTE: *Foods for digestion should be prepared as they are consumed, and not milled to a powder for homogeneity (if this is not the case in real life).*

Critical points of the INFOGEST Quant method

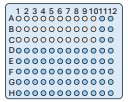
15 h acid hydrolysis



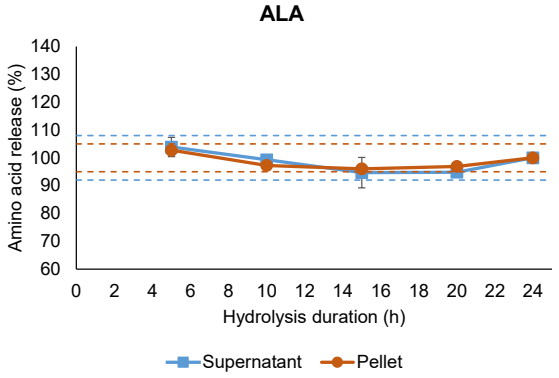
The **acid hydrolysis** of the digested samples lasts **15 h** instead of 24 h, as done in the food material.

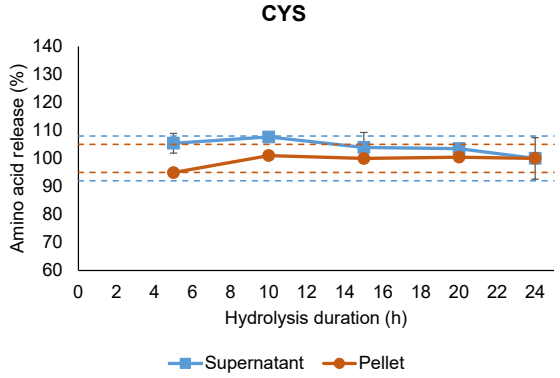


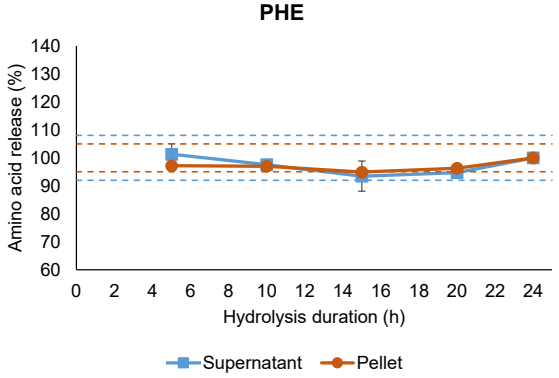
5 h
10 h
15 h
20 h
24 h

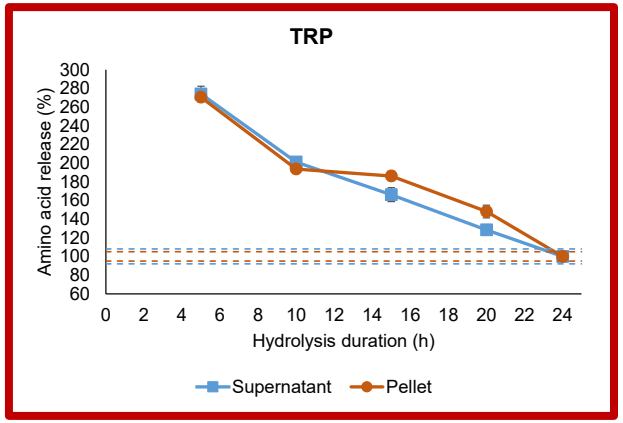


Total amino acids
(*HPLC-UV*)









**Results: % relative to 24 h (reference method).*