



**HAL**  
open science

## Challenging in Rats the Use of $^{13}\text{C}$ Spirulina as Reference Protein for the Dual Isotope Method to Determine Amino Acid Bioavailability (P08-061-19)

Romain Tessier, Nadezda Khodorova, Juliane Calvez, Daniel Tomé, Claire C. Gaudichon

### ► To cite this version:

Romain Tessier, Nadezda Khodorova, Juliane Calvez, Daniel Tomé, Claire C. Gaudichon. Challenging in Rats the Use of  $^{13}\text{C}$  Spirulina as Reference Protein for the Dual Isotope Method to Determine Amino Acid Bioavailability (P08-061-19). *Nutrition* 2019, Jun 2019, Baltimore, United States. *Current Developments in Nutrition*, 3 (Supplement\_1), 2019, 10.1093/cdn/nzz044.P08-061-19 . hal-03296220

**HAL Id: hal-03296220**

**<https://hal.inrae.fr/hal-03296220>**

Submitted on 28 Jul 2021

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution 4.0 International License

### Challenging in Rats the Use of $^{13}\text{C}$ Spirulina as Reference Protein for the Dual Isotope Method to Determine Amino Acid Bioavailability (P08-061-19)

Romain Tessier,<sup>1</sup> Nadezda Khodorova,<sup>2</sup> Juliane Calvez,<sup>1</sup> Daniel Tomé,<sup>2</sup> and Claire Gaudichon<sup>2</sup>

<sup>1</sup>INRA-AgroParisTech; and <sup>2</sup>AgroParisTech-INRA

**Objectives:** In order to establish DIAAS in humans, the FAO recommended to develop a new method to measure indispensable amino acid (IAA) digestibility. This method uses two isotopic labeling, one for the protein to test and one for a reference protein. Spirulina was chosen as the  $^{13}\text{C}$  reference protein due to its commercial availability and affordability. However, the real digestibility of spirulina protein has not been measured *in vivo*. This work aims to assess the digestibility of spirulina and its repeatability in different meal tests in rats.

**Methods:** 23 Wistar male rats were fed a test meal containing 0.5 g of  $^{15}\text{N}$  protein from either spirulina ( $n = 7$ ), sunflower ( $n = 8$ ) or goat milk isolate ( $n = 8$ ) and 10 mg of  $^{13}\text{C}$  labeled spirulina. Rats were euthanized 6 h after the meal and their digestive luminal contents (stomach, small intestine, ileum, caecum, colon) were collected. Protein digestibility was determined for the test and the reference proteins by measuring  $^{15}\text{N}$  and  $^{13}\text{C}$  enrichments in the digesta by EA-IRMS. Caecal IAA digestibility of  $^{13}\text{C}$  spirulina was determined by measuring the quantity of AA in the

caecum by UPLC and the  $^{13}\text{C}$  enrichment in AA by GC-C-IRMS. Group effects were tested using one way ANOVA and differences between groups using Bonferroni test.

**Results:** Six hours after ingestion, most of the dietary  $^{15}\text{N}$  and  $^{13}\text{C}$  were found in the caecum and colon. But there at least twice more  $^{15}\text{N}$  nitrogen in the caecum and colon in the spirulina group than in the two other groups. Therefore, spirulina protein digestibility ( $86.0 \pm 0.7\%$ ) was lower ( $P < 0.001$ ) than sunflower ( $95.1 \pm 0.5\%$ ) and goat milk digestibility ( $97.2 \pm 0.2\%$ ).  $^{13}\text{C}$  spirulina digestibility tended to be different ( $P = 0.06$ ) when mixed to spirulina ( $90.6 \pm 0.6\%$ ), sunflower ( $88.8 \pm 0.5\%$ ) or goat milk ( $89.0 \pm 0.5\%$ ) isolates. The caecal IAA digestibility of  $^{13}\text{C}$  spirulina was lower in the spirulina group than in sunflower and goat milk groups for every IAA tested, and the mean was  $91.6 \pm 0.2\%$  for sunflower,  $91.4 \pm 0.4\%$  for goat milk and  $85.4 \pm 0.6\%$  for spirulina.

**Conclusions:** Spirulina protein is of lower digestibility than other animal or plant proteins. Protein and amino digestibility of a tracer dose of  $^{13}\text{C}$  spirulina appears to vary depending on the protein component of the meal. These results question the use of spirulina as a reference protein for the dual isotope method.

**Funding Sources:** French Research National Agency (ANR), SOFIPROTEOL.

**Supporting Tables, Images and/or Graphs**

	Val	Leu	Ile	Thr	Phe	Val	Met	IAA Mean
Sunflower	91.5±0.4 a	91.5±0.4 a	91.0±0.3 a	91.8±0.4 a	89.8±0.5 a	91.7±0.4 a	93.4±0.3 a	91.5±0.2 a
Goat milk	91.0±0.8 a	91.0±0.9 a	91.4±0.8 a	91.7±0.9 a	88.7±1.1 a	92.4±0.8 a	93.3±0.7 a	91.4±0.4 a
Spirulina	84.8±1.5 b	83.7±1.6 b	84.3±1.5 b	88.4±1.1 b	80.4±2.0 b	88.1±1.1 b	88.9±1.5 b	85.4±0.6 b
Group effect	<0.001	<0.001	<0.001	0.01	<0.001	<0.01	<0.001	<0.001