clinical outcomes from well-conducted randomized controlled trials, the identification of a suitable experimental approach that could be adopted by different investigators could allow the requisite much larger numbers of volunteers to be studied and might settle the debate if agreement could be reached on a suitable method. It is quite clear to me that the IAAO method could not serve such a purpose.

The author had no conflicts of interest.

D Joe Millward

Department of Nutritional Sciences
School of Biosciences and Medicine
Faculty of Health and Medical Sciences
University of Surrey
Guildford GU2 7XH
United Kingdom
E-mail: d.millward@surrey.ac.uk

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Reply to DJ Millward

Dear Sir:

With Millward's letter, he continues to criticize the merits of the indicator amino acid oxidation (IAAO) method to assess human protein requirements. The issues raised are old and repeatedly described and discussed in the literature, as well as carefully considered during a rigorous review of the article before acceptance for publication, and noted in Fukagawa's editorial published with the article (1).

Apparently, Millward's chief criticism of the IAAO method is that the intake of the indicator amino acid phenylalanine was inadequate at the higher intakes of protein and thus the breakpoint in the response was due to a deficiency of phenylalanine. He used a circular argument with regard to whether phenylalanine intake was adequate or deficient.

We have previously measured the phenylalanine requirement in the presence of an excess of all other amino acids (ie, protein), including tyrosine, and found it to be $13.6 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$ (population upper 95% CI) (2). During the present experiment, we provided phenylalanine at an intake of $30.5 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$, which is well in excess of the phenylalanine requirement in the presence of excess tyrosine ($40 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$) and other amino acids. Therefore, very clearly, phenylalanine would not be limiting at any intake of protein in the current experiment. In which case, as we discussed in the article, the breakpoint was due to the plateau in protein synthesis that occurred when the intake of protein was adequate. To argue that phenylalanine was deficient because it plateaued in oxidation is therefore incorrect, and ignores the extensive work done to show how the IAAO method works.

Two main principles of the IAAO method are 1) that the excess intake of phenylalanine is proportioned between protein synthesis and oxidation and 2) phenylalanine oxidation progressively declines with increasing intake of the limiting amino acid (or of total protein) from deficient to adequate and reaches a steady nadir (breakpoint) when a sufficient amount is consumed. Millward has suggested that the intake of phenylalanine, as the indicator, be allowed to change in proportion to the protein intake. If there was a change in the intake of phenylalanine along with protein intake, then the percentage of dose oxidized would not change with each increment of protein because the degree of excess of phenylalanine would be the same for every protein intake. This means that the percentage of dose oxidized would not vary or vary only very little with intakes between deficient and adequate, and it would increase thereafter because phenylalanine would be in excess. This is the same as using the direct oxidation approach, which has other well-recognized issues (3). This is also the key reason why the indicator amino acid must be controlled to the same intake in all treatments; otherwise, the slope of the response line may be due to changes in intake of the indicator amino acid rather than intake of the test protein or amino acid (3).

Millward repeatedly argues that the requirement of every indispensable amino acid varies directly with protein intake, the "adaptive metabolic demand." However, this theory is untested by direct experimentation. In contrast, there is no evidence currently available that the requirement for any amino acid, other than the single most limiting amino acid in the diet, varies with protein intake. This principle of the limiting amino acid defining the protein requirement is the principle whereby the amino acid score of a protein is derived. If one accepts that amino acid score is a valid concept, as Millward does (4), then "adaptive metabolic demand" cannot also be correct.

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Minghua Tang

Department of Nutrition Science
Purdue University
West Lafayette, IN

George P McCabe

Department of Statistics
Purdue University
West Lafayette, IN

*Rajavel Elango**Wayne W Campbell*

Department of Pediatrics
University of British Columbia
Vancouver
Canada

Department of Nutrition Science
Purdue University
700 West State Street
West Lafayette, IN 47907
E-mail: campbellw@purdue.edu

Paul B Pencharz

Department of Pediatrics and Nutrition Science
University of Toronto
Toronto
Canada

Ronald O Ball

Department of Agricultural, Food, and Nutrition Science
University of Alberta
Edmonton
Canada

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Erratum

Mason C, Xiao L, Imayama I, Duggan C, Wang C-Y, Korde L, McTiernan A. Vitamin D₃ supplementation during weight loss: a double-blind randomized controlled trial. *Am J Clin Nutr* 2014;99:1015–25.

The units for C-reactive protein (CRP) are incorrectly stated as “mg/mL” in the following places: the Results section of the Abstract on page 1015, the Results section of the text on page 1020, Table 2 on page 1020, Table 3 on page 1021, and Table 4 on page 1022. The correct unit is “mg/L.”

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Erratum

Gale C, Thomas EL, Jeffries S, Durighel G, Logan KM, Parkinson JRC, Uthaya S, Santhakumaran S, Bell JD, Modi N. Adiposity and hepatic lipid in healthy full-term, breastfed, and formula-fed human infants: a prospective short-term longitudinal cohort study. *Am J Clin Nutr* 2014;99:1034–40.

Because of a copyediting error, the abbreviation for magnetic resonance spectroscopy (MRS) incorrectly appears in the fourth paragraph of the Subjects and Methods section. The first sentence of the fourth paragraph [“Whole-body MRS images were acquired on a Phillips 1.5 Tesla system by using a T₁-weighted rapid-spin-echo sequence (repetition time of 500 ms, echo time of 17 ms, echo train length of 3) by using a Q body coil.”] should read as follows: “Whole body magnetic resonance images were acquired on a Phillips 1.5 Tesla system by using a T₁-weighted rapid-spin-echo sequence (repetition time of 500 ms, echo time of 17 ms, echo train length of 3) by using a Q body coil.” The fifth sentence of the fourth paragraph [“All MRS images were analyzed independently of the investigators and blind to participant identity and feeding group by VardisGroup (www.vardisgroup.com) by using an image segmentation program (SliceOmatic; Tomovision).”] should read as follows: “All magnetic resonance images were analyzed independently of the investigators and blind to participant identity and feeding group by VardisGroup (www.vardisgroup.com) by using an image segmentation program (SliceOmatic; Tomovision).”

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