Measuring Protein's Punch: DIAAS is the New PDCAAS

The FAO's call for a more accurate protein scoring than PDCAAS has led to the development of the Proteos project, which uses pigs to predict amino acid uptake in humans.

by Paul J. Moughan

Amid the current focus on world food security and the importance of dietary protein to health, there has been a resurgence of interest in the subject of dietary protein quality. But how should a protein's potency to supply amino acids be described?

Dietary Protein Quality

Over recent years, there have been significant advances in how amino acid digestibility (disappearance from the digestive tract) and availability (uptake of the amino acid in a structural form utilizable for protein synthesis) are determined. One of the greatest challenges is to develop a scoring system where a single value can describe a protein's relative quality as a source of amino acids for the body. There is a great variety of food proteins that differ in their amino acid compositions, the structures of their proteins and how these proteins interact with other food constituents. Proteins such as milk, meat, eggs and well-processed soy have well-balanced amino acids that are highly digestible and bioavailable, while other proteins such as some cereals, beans and animal proteins such as collagen have less well-balanced amino acid patterns and are less well digested. Of course, people generally eat diets rather than single foods, and there is thus the opportunity to mix different foods together to obtain high-quality dietary intakes. There is also an opportunity to co-process vegetable- and animal-based proteins to generate new food proteins with optimal amino acid balances and new physicochemical functionalities. Scores can be calculated for both single foods and mixed-food diets to ascertain the overall protein quality. In the past, the globally recommended scoring method was the protein digestibility corrected amino acid score (PDCAAS), but this approach has shortcomings.

Realizing the limitations of PDCAAS, the Food and Agriculture Organization of the United Nations (FAO) Expert Consultation, which published its findings in 2013, has recommended a new score to replace PDCAAS, called DIAAS - the digestible indispensable amino acid score. In fact, the primary finding of the Consultation was that there should be an emphasis, when describing foods and diets, on individual amino acids. Each amino acid should be treated as an individual nutrient. However, where a single score is needed, DIAAS is recommended. DIAAS is based on the true "ileal" digestibility of each dietary indispensable amino acid. Ileal digestibility is that which occurs at the end of the small intestine (Figure 1). "True" digestibility denotes that the flow of amino acids at the ileum (end of the small intestine) has been corrected for the flow of basal endogenous amino acids, that are of the body rather than dietary origin.

The growing pig is recommended as the preferred animal model for the adult human, for determining ileal amino acid digestibility (Figure 2). For processed foods, lysine availability (digestible reactive lysine) needs to replace conventional measures of lysine digestibility. There were minor recommended changes to the reference amino acid patterns (i.e. amino acid requirements). A further major technical change that was recommended is that in the case of food ingredients (not sole foods or diets) the practice of truncating scores to 1.0 should be disbanded. The removal of the truncation step can be significant (Table 1).

Table 1. DIAAS values showing the effect of truncation of the score.

	Milk Protein Concentration	Whey Protein Isolate	Whey Protein Concentrate	Red Meat
Non-truncated	1.31	1.25	1.10	1.10
Truncated	1.0	1.0	1.0	1.0

The non-truncated values give information concerning the power of the protein to supply complementary amino acids, in formulating mixtures of proteins.

Are DIAAS values greatly different from PDCAAS?

DIAAS values are meaningfully different from PDCAAS. In particular, PDCAAS tends to overestimate the value of lower quality proteins.

If a single score is needed to describe a protein's relative quality as a supply of amino acids, then DIAAS is the preferred procedure. It must be noted, however, that encapsulating information into a single score, means that information is necessarily lost. Consider, the whey protein isolate in Table 2. The protein is most limiting in histidine (DIAAS = 1.09), but all of the dietary essential amino acids are supplied excess to requirement, and the protein is a very powerful supplier of tryptophan and the branched-chain amino acids.

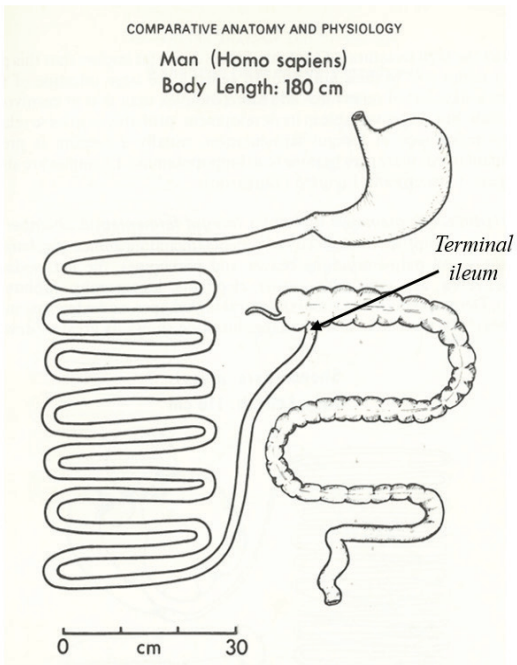
This latter information is lost in the score and is why the FAO Consultation emphasized describing and using the true ileal digestible contents of each amino acid individually. Nevertheless, a single score to describe a food's overall protein quality ranking is required, especially for purposes of trade in proteins and for substantiating food claims around protein. DIAAS is the most accurate scoring method for such purposes.

The practical implementation of DIAAS to date, however, has been hampered by a lack of published DIAAS values for foods in a form as consumed by humans. Not surprisingly, much work is underway globally to generate true ileal amino acid digestibility values and DIAAS values for human foods. The uptake and implementation of DIAAS will provide an important change in the accuracy of predicting dietary protein quality. This is of great consequence to many people in the developing world, and to any person trying to maintain lean body mass and particularly where food intake may be low (e.g., the elderly, dieters). It will be crucial for us to learn to use dietary protein more efficiently in light of high predictions for the world's human population.

Global Need for Protein

Dietary protein and amino acids promote lean body mass, normal

body weight and support optimal organ function. Amino acids also have specific crucial metabolic roles (e.g., leucine and muscle protein synthesis; tryptophan as a precursor for serotonin in the brain). Dietary protein and specific amino acids play important roles in body weight loss and the maintenance of body weight, satiety, and the attenuation of muscle loss that accompanies aging. Amino acids and overall dietary protein intake affect immune system performance, and especially when challenged by sub-clinical levels of infection. There is also much evidence to suggest that the achievement of peak performance in resistance training and high-intensity exercise may require higher-protein intakes than the currently advised recommended daily intakes. There may be a specific role for the branched-chain amino acids and leucine in particular. It is hardly surprising then that there has been an upsurge in the manufacture and marketing of high-protein foods and supplements.



Ref: Wrong OM, Edmonds CJ and Chadwick VS (1981) Comparative anatomy and physiology In: The Large Intestine, p 5, MTP Press Ltd, England.

Table 2. DIAAS Ratios for a Whey Protein

Amino acid isolate	Ratio 1
Threonine	1.80
Met + Cys	2.29
Valine	1.21
Isoleucine	2.22
Leucine	2.57
Tyr + Phe	1.71
Histidine	1.09
Tryptophan	3.35
Lysine	2.51

The Proteos Project

The Proteos project was established by a consortium of the world's food sectors in response to a call from FAO (FAO, 2013) to develop comprehensive true ileal amino acid digestibility and DIAAS data for foods. The project is overseen on behalf of the food sector funders, by the Global Dairy Platform (GDP), headquartered in Chicago. There is also a consortium of research providers including, Wageningen University and Research, the Netherlands; AgroParisTech, France; the Riddet Institute, Massey University, New Zealand, and the University of Illinois, USA.

This large, truly international research project has several aims. To date, a standardized research protocol has been developed, which will shortly be published in the scientific literature. Intra- and inter-variability in true ileal amino acid digestibility using the cannulated pig model have shown to be acceptably low. Ten diverse proteins have been given to growing pigs and adult humans under controlled conditions, and as expected, early observations show close agreement between the species.

The next step will be the generation of a statistical regression relationship between adult humans and the growing pig, to allow the relatively small correction to be made for digestibility between the species. The final and lasting contribution of Proteos will be the generation of a comprehensive data set for the amino acid digestibility and DIAAS values of the world's foods. It is important to note that the application of DIAAS employs "predicted human" amino acid digestibility values, not pig values, and publication of the equation to allow the generation of such predicted values is not too far away.

DIAAS Gaining Speed

In the six-year period since the publication of the FAO Expert Consultation report, there has been much scientific and technical activity around the new DIAAS method. A quick search of the literature using Scopus, with DIAAS as the keyword, gives 35 scientific papers with more than half of these published in the last two years. New DIAAS values have been published for some 90 human foods. Alongside the Proteos work, a considerable dataset of true ileal amino acid digestibility and DIAAS values for human foods is accumulating rapidly. What is often not recognized is that in animal nutrition, the more accurate true ileal amino acid digestibility system has been used in practice for the past 30 years.

This means that there is a very comprehensive dataset of pig true ileal amino acid digestibility data even for human foods, which are sometimes fed to pigs. Once Proteos releases a robust pig/human prediction equation, all of these data become available, meaning the generation of a global foods database, almost overnight.

Some of the key protein foods commonly consumed in Asia and Africa will be under-represented (though very little rat fecal crude protein digestibility data currently exist for such foods regardless), but work is already underway in China to provide DIAAS values for Asian foods. It is imperative to fund investigations into the foods of Africa, for which very few data on any type of amino acid digestibility currently exist.

The Way Forward

True ileal amino acid digestibility coefficients have been shown empirically to be accurate estimates of amino acid absorption. They have also been shown to be sensitive indicators of damage to proteins consequent upon processing and storage. Moreover, and because they include relevant corrections for endogenous (of body origin) amino acids at the end of the small intes-

tine, they reflect the effects of most of the plant antinutritional factors (ANFs). None of these claims can be made for the outmoded fecal crude protein digestibility measure. It is time to move forward.

Not surprisingly, recent work in the US by Dr. Mark Manary and colleagues has underscored the importance of poor protein quality, and especially the value of DIAAS, with high statistical correlations found with incidences of stunting in children. Similar findings with DIAAS have been reported by Dr. Anura Kurpad's group in Bangalore, India, providing further support for the usefulness in practice of DIAAS.

Work being conducted between the Punjab Agricultural University of India (PAU) and the Riddet Institute, New Zealand graphically illustrates the practical usefulness of DIAAS to unearth dietary amino acid deficiencies, hitherto not apparent. If we wish to use the world's protein sources efficiently and to describe the available amino acid levels as accurately as possible, then a shift to using the FAO recommended DIAAS should be made in all jurisdictions.

Accuracy Is the Agenda

In North America, questions have been raised with reference to regulatory frameworks, of unintended outcomes of using systems such as DIAAS, such as promoting the consumption of animal-based foods to the detriment of plant-based foods, ostensibly associated with greater environmental sustainability and health issues. However, DIAAS and similar scoring methods are purely about describing amino acid supply relative to requirements, and in this context, DIAAS is simply the most accurate approach.

If other attributes of foods need to be signaled to consumers then other indices (e.g., fiber content, carbon emissions, etc.) should be used, but the implementation of a valuable index to describe protein quality itself should not be compromised. The authors, while recognizing the technical superiority of DIAAS, not unreasonably, query the practicality of its implementation in the face of limited data on DIAAS. In response to this, however, once the interspecies prediction equation becomes available, a DIAAS dataset will be able to be generated immediately which will be more comprehensive than that of any existing rat fecal CP digestibility dataset.

Consideration for Animal Welfare C.P.F. Marinangeli and J.D. House question the ethics of using animal assays in the context of regulatory frameworks in a 2017 article published in Nutrition Reviews. The authors make the excellent suggestion of developing and using in vitro methods of protein and amino acid digestibility.

Unfortunately no such *in vitro* assay has been published, which has been adequately externally validated. Research to develop a validated robust *in vitro* amino acid digestibility assay should be of high priority. A great opportunity will soon exist to relate such an assay to the comprehensive *in vivo* database on true ileal amino acid digestibility, thus allowing routine prediction of accurate and relevant *in vivo* values.

Several Codex Committees have recently acknowledged that protein quality should be assessed by the most up-to-date methodology, DIAAS, but also acknowledge that currently, insufficient data are available to permit implementation of DIAAS into regulatory standards. The 2013 FAO report did give some examples of how DIAAS could be used for regulatory purposes, but it is critical to underline that these were merely exemplars and not a recommendation.

Careful consideration needs to be given as to how protein contents and DIAAS values should be used to allow product protein claims, and indeed in relation to this, the suggestion of Marinangeli and House of introducing the use of fixed, conservative, ileal digestibility coefficients for different food types has merit. Whether pragmatic, conservative coefficients or *in vitro* coefficients are introduced, they must ultimately relate back to a robust, accurate baseline.▼

Paul J. Moughan is a distinguished Professor and Fellow Laureate of the Riddet Institute, Massey University, New Zealand and is the Science Leader of the international Proteos project.