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Applications and Challenges of Protein Quality Assessment in Food

Yang Cong [1, 2, 3, 4], Ruiming Zhang [1, 2, 3, 4]

[1] Shanghai Le Bonta Wellness Co., Ltd; Shanghai, China
[2] Standard Investment (China) Co., Ltd, Taicang, Jiangsu, China
[3] Jiangsu Hua Sun Health Technology Co., Ltd; Taicang, Jiangsu, China
[4] Standard Foods (China) Co., Ltd, Taicang, Jiangsu, China

Abstract: As a key nutrient to maintain normal physiological functions of the human body, protein plays an increasingly important role in people's pursuit of a healthy life. With the growing demand for healthy dietary standards, the evaluation of protein quality in food has become particularly critical, providing a scientific basis for the innovation and development of healthy foods. Given the diversity of protein sources, fluctuations in content, and the complexity of amino acid composition, it is crucial to choose appropriate evaluation methods to accurately analyze the protein quality in food. This article deeply explores the protein quality evaluation methods widely used internationally, with a particular focus on the two evaluation systems of digestible amino acid score (DIAAS) and protein digestibility corrected amino acid score (PDCAAS). This article analyzes in detail the application scope, advantages, and challenges that may be encountered in practical applications of these two methods, aiming to provide a solid theoretical support for the development of high-quality protein foods.

Keywords: Protein Quality; PDCAAS; DIAAS.

1. Introduction

When evaluating the nutritional value of food, we should not only pay attention to the quantity of each nutrient provided, but also pay attention to its quality. Especially for protein, the significant difference in its amino acid composition and digestion efficiency makes the quality evaluation of dietary protein particularly critical. Protein is not only one of the three major energy sources for the human body, but also the main source of nitrogen and essential amino acids (EAAs). These amino acids are essential for synthesizing new proteins, promoting tissue growth and maintaining body functions [1]. Evaluating the quality of protein not only helps us understand its nutritional value in food, but also plays an important role in guiding consumers to eat a reasonable diet and preventing malnutrition.

Protein comes from a wide range of sources, and proteins from different sources have significant differences in amino acid composition and digestion efficiency [4]. Only proteins with appropriate amino acid composition and high digestibility can meet the needs of the human body. The ability of different proteins to meet these needs varies significantly and is usually quantitatively evaluated through protein quality indicators. Animal-based proteins are generally considered to be high-quality proteins because of their comprehensive amino acid types, high content and easy absorption; while most plant-based proteins are considered to be of lower quality than animal proteins because the proportion of amino acids they contain is very different from the needs of the human body and their absorption rate is not high [15]. However, accurate assessment of protein quality requires comprehensive consideration of multiple factors, including amino acid composition, human body's demand for protein, protein digestibility, measurement methods and animal models. A comprehensive evaluation method should be both scientific and reasonable and cost-effective. In addition, protein utilization takes a relatively short time, so the quality of protein consumed over a period of time is extremely critical. Unlike many micronutrients and energy, the human body does not store protein or EAAs in the liver, tissues or bones. Studies have shown [2] that amino acid levels in the blood rise after a meal, but return to baseline levels a few hours later. During this period, the absorbed amino acids are metabolized by the body and used to synthesize proteins and other substances, while the unmetabolized amino acids are oxidized [3]. Therefore, in order to reduce the nutritional waste of amino acids, especially to reduce the metabolic pressure of protein on specific populations, it is crucial to scientifically evaluate protein quality.

Given the dietary changes aimed at human health, the importance of protein quality evaluation is obvious. However, it is not easy to translate this concept into practical application. Existing protein quality evaluation methods can be used to rank proteins, but applying them to food is more complicated because protein is not only a single nutrient, but also carries multiple essential nutrients, namely 9 essential amino acids plus dietary nitrogen. Therefore, when evaluating protein quality from a food perspective, factors such as data reliability, the needs of different populations for essential nutrients, and food processing and preparation need to be considered. In this article, we will review the concept of protein quality, explore protein digestion and amino acid absorption, and introduce different methods used to evaluate protein quality, with a particular focus on the application of PDCAAS and DIAAS measurement data and the challenges they face.

2. Protein Quality

2.1. Definition of Protein Quality

Protein is a macromolecular organic compound composed of amino acid sequences, which plays a key role in the development, maintenance and various physiological functions of the human body [1]. For adult individuals in my country, the recommended daily intake (RNI) is 0.98 grams of protein per kilogram of body weight for men and 0.97 grams of protein per kilogram of body weight for women[3]. Amino acids are not only the basis of protein, but also participate in a variety of physiological activities such as energy conversion, biosynthesis, immune response and muscle function, and are essential for physical health and life processes. Amino acids are divided into essential amino acids (EAA), conditionally essential amino acids and non-essential amino acids (NAA). EAA cannot be synthesized by the human body and must be ingested through diet. With age, the body's demand for EAA changes, with the highest demand in infancy (0.5 to 1 year old) and then gradually decreasing. This shows that in the early stages of life, EAA is essential for growth and development, while in maturity, the demand shifts more to maintaining body functions[5].

The quality of protein is usually based on its ability to fully meet the human body's EAA requirements. Common criteria include its amino acid composition, the EAA requirements of a specific population, and the digestibility of EAA. First, these criteria take into account the digestibility of protein, that is, the proportion of ingested protein that is broken down into absorbable amino acids, dipeptides, and tripeptides in the body. There are significant differences in the digestibility of protein from different food sources. Some foods contain sufficient digestible EAA to meet the human body's needs, while others may be insufficient in the supply of one or more EAA. Second, when evaluating protein quality, reference is made to an idealized reference protein model that is based on perfect digestibility and contains ideal levels of all essential amino acids. This reference model is adjusted according to the needs of different age groups [10].

2.2. Human Digestion of Protein and Absorption of Amino Acids

The premise for the human body to utilize the amino acids in food protein is that the protein must first be broken down into free amino acids and smaller peptide chains, such as dipeptides and tripeptides, which can then be absorbed by the blood[1]. This digestive process is a complex series of steps, starting with the physical destruction of food when chewing in the mouth. The food then enters the stomach, where it mixes with acidic gastric juice, and pepsin in the gastric juice begins to perform a preliminary decomposition of the protein to form polypeptides. However, at this point, protein digestion is not complete and not enough absorbable amino acids and peptides have been produced. Subsequently, the food enters the small intestine at the rate of gastric emptying. Under the combined action of enzymes such as trypsin, chymotrypsin, and carboxypeptidase A in the small intestine and enzymes in the brush border of the small intestine, proteins and polypeptides are further broken down into amino acids, dipeptides, tripeptides, and oligopeptides. Compared with pepsin, enzymes in the small intestine digest proteins more thoroughly because most of the protein decomposition process actually occurs in the small intestine[6].

The terminal small intestine, especially the terminal ileum, is the main site for the absorption of amino acids and peptides, where they are almost completely absorbed [7]. However, amino acids and peptides that are not absorbed in the terminal small intestine will enter the large intestine. Although some parts of the large intestine also contain amino acid transporters, there is currently no evidence that the large intestine can significantly absorb these amino acids [8]. Even if there is some absorption in the large intestine, its contribution to the overall amino acid absorption level is minimal. In addition, amino acids and peptides that are not absorbed in the small intestine and undigested proteins may be further broken down and fermented by the microbiota in the large intestine [9]. At the same time, colonocytes have the ability to synthesize and metabolize amino acids, which may enter the body through the blood rather than the digestive tract. Since protein digestion and absorption mainly occur in the small intestine, and microbial protein formation in the large intestine is different, when evaluating protein digestibility, samples collected from different locations may lead to differences in measurement results, which is particularly important when considering methods for determining protein quality.

3. Protein Quality Measurement Methods

Since the development of food protein nutritional evaluation technology, researchers have developed a variety of methods to measure different sources and types of protein [1]. These technologies are based on different scientific principles and quantify the quality of protein through specific parameters and formulas. Specifically, these methods can be in vivo or in vitro. In vivo methods may involve humans or animals as experimental subjects. In vivo experiments include methods that rely on amino acid digestibility using different sample collection points, and some methods based on animal growth conditions. These methods evaluate the quality of protein by measuring the weight gain of animals under a specific protein diet and comparing it with a standard protein. In vitro evaluation methods measure protein quality in vitro by simulating the digestive environment in the human body, which will be discussed later in this chapter.

3.1. Evaluation Methods Based on Weight Change

Protein is essential for the growth and tissue maintenance of organisms. Based on the contribution of protein to weight change, the quality of protein can be evaluated by measuring the ratio of the weight gain of experimental subjects over a certain period of time to the amount of protein intake. This method includes the protein efficiency ratio (PER) and the net protein retention ratio (NPR), which are usually compared with a standard protein (such as casein, with a PER of 2.5) [1]. The advantage of these methods is that they are simple to operate. The quality of protein is evaluated by feeding experiments on growing rodents over a period of time, collecting data, and measuring their weight gain at the end of the experiment. However, these methods have limitations. For example, rodents may have a higher digestibility of certain sulfur-containing amino acids than humans, which may lead to an overestimation of the evaluation results [11]. In addition, these methods do not take into account other nutrients in the experimental diet that may affect weight gain.

3.2. Evaluation Methods Based on Nitrogen Balance

Since protein is the main source of nitrogen in organisms and the nitrogen metabolism process is relatively independent, protein intake and utilization efficiency can be indirectly evaluated by monitoring the nitrogen balance state. Nitrogen balance evaluation methods include biological value (BV), net protein utilization (NPU) and true digestibility (TD). In these methods, rodents are fed with a diet containing the test protein, while a control group is fed with a nitrogen-free diet. The quality of the protein can be calculated by measuring the basal nitrogen loss of the control group and the nitrogen consumption and excretion of the test group [12]. The basic assumption of these methods is that the ingested nitrogen is either utilized by the organism or excreted. However, these methods do not fully consider the delayed effect of nitrogen excretion and the contribution of metabolism to nitrogen excretion [12].

3.3. Evaluation Method Based on Amino Acid Score

As mentioned above, the key to protein quality lies in whether it can fully meet the human body's needs for essential amino acids. The ideal measurement method should be based on the composition of amino acids and their digestibility in the human body.

3.3.1. Evaluation Mechanism of Amino Acid Scoring Method

The amino acid scoring method (AAS) can scientifically evaluate the differences in amino acid composition and content of proteins from different food sources and human needs by comparing the ratio of EAA in the test food with the reference protein (usually eggs) or the human body requirement standard [13]. The advantages of this method are its simplicity and economy, as it does not require in vivo digestion tests. The implementation of AAS is to use chemical analysis technology to quantitatively determine the content of each amino acid in food. By adjusting experimental conditions such as mobile phase and ion exchange column, the concentration of different amino acids can be accurately measured.

However, one limitation of AAS is that it does not fully consider the degree of absorption and utilization of amino acids in the human body. To make up for this deficiency, researchers introduced the digestibility of food protein on the basis of AAS and developed two more advanced evaluation methods, PDCAAS and DIAAS [13,14]. These two methods more comprehensively evaluate the bioavailability of protein by comparing the amount of digestible EAA in food with the human body requirement standard, thereby more accurately predicting the potential of food protein in meeting human needs. The calculation methods, evaluation principles, advantages and limitations of the PDCAAS and DIAAS methods can be seen in Table 1. Next, their applications and challenges will be discussed in detail.

Table 1. Comparison of three protein quality evaluation methods based on amino acid composition and digestibility

Method	Calculations	Measurement Principle	Biological Evaluation	Advantages	Applications	Limitations
AAS	AAS = (Amount of First Limiting Amino Acid in Test Protein / Amount of First Limiting Amino Acid in Reference Pattern) × 100	Compare the similarity of the amino acid ratio of the tested food protein to the reference protein amino acid pattern or the human amino acid score pattern	no	Easy to operate, economical	Preliminary exploration of high-quality protein sources, research on protein complementarity, and development of high-quality protein foods	do not consider the digestibility of food protein
PDCAAS	PDCAAS = AAS × Faecal digestibility TP%	Ration of EAAlim in test protein compared to reference protein corrected for faecal protein digestibility	yes	Relatively accurate, widely used, and more comprehensive data	Evaluate protein quality under different production processes and processing methods	Fecal digestion rate is not as accurate as ileal digestion rate. There is a difference between protein digestibility and amino acid digestibility. Truncation ignores the nutritional differences of some high-quality proteins.
DIAAS	DIAAS = (mg of digestible dietary indispensable amino acid in 1 g of the dietary protein) / (mg of the same dietary indispensable amino acid in 1 g of the reference protein)	Ratio of EAAlim in test protein compared to reference protein corrected for ileal amino acid digestibility	yes	accurate	Comparing or arranging the quality of proteins from different sources, especially high-quality proteins. to evaluate the complementarity of mixed protein sources	High cost. Measurement methods are not uniform. Database of ileal amino acid digestibility is not comprehensive.

3.3.2. Protein Digestibility Corrected Amino Acid Score (PDCAAS)

PDCAAS (Protein Digestibility Corrected Amino Acid Score) was proposed by FAO/WHO in 1989 [13]. This method comprehensively considers the concentration and digestibility of EAA in food. By comparing with the reference model of different age groups, the score of each EAA is determined, and the lowest score (i.e. the score of the first limiting amino acid) reflects the overall quality of the tested protein. The PDCAAS score directly reflects the ability of food protein to meet the human body's EAA needs. It is currently the most widely recognized method for evaluating food protein quality and guides product development and production.

First, the evaluation results of protein quality by PDCAAS can be applied to people of all ages, which is conducive to the development of high-protein quality products for different target groups. According to the latest recommendations of FAO [14], considering the differences in the needs of essential amino acids in different age groups, a new scoring model is proposed for four age groups: infants, preschool children (1-2 years old), older children and adolescents (4-18 years old) and adults (> 18 years old).

Second, the data reliability of PDCAAS has also been verified. The use of standardized amino acid analysis methods during the determination process can provide good intra-laboratory repeatability and good inter-laboratory reproducibility for most amino acids [13]. However, a study comparing the PDCAAS values of four different types of soy raw materials, such as isolated soy protein (SPI) and soy protein concentrate (SPC), in two different laboratories [16] found that there were differences in the results measured between different laboratories. The error of the analytical method may be an important factor in the difference in PDCAAS, which can be alleviated by applying the amino acid nitrogen recovery correction factor.

In addition to measuring the quality of proteins from different sources, PDCAAS can also measure the impact of different production processes and processing methods on protein quality. Nosworthy et al. used the PDCAAS method to evaluate the impact of processing on protein quality, depending on the selected crops and varieties [17], and found that heat processing had the greatest impact on soybeans compared with wheat and oats. Similarly, another study found that optimizing the production process, such as the application of cell disruption technology, can also help improve the extraction rate and digestibility of microalgae protein, thereby improving its PDCAAS score [18].

PDCAAS can be used on product packaging to convey product protein quality information to consumers because of its scientific and feasible characteristics. Enacted in the Nutrition Labeling and Education Act (NLEA) of the United States in 1990, PDCAAS complies with the requirements of the Food and Drug Administration (US-FDA) labeling regulations for declaring protein content [26].

However, the PDCAAS method also has some limitations. First, PDCAAS is based on fecal digestibility rather than ileal digestibility, which may not accurately reflect the absorption of amino acids in the small intestine. This can easily lead to an overestimation of protein digestibility when evaluating low-quality plant proteins. Nitrayová et al. [19] found that the calculated PDCAAS values of poor-quality protein sources such as rye and barley are at risk of being overestimated compared to rice. Secondly, PDCAAS calculates digestibility based on protein, ignoring the differences in digestibility between different amino acids. This makes it impossible to accurately reflect the difference between the digestibility of limiting amino acids and the overall digestibility of protein in some cases. For example, the true digestibility of methionine, cysteine and tryptophan in soybeans, peas and lentils is significantly lower than its protein digestibility [20], thus affecting the accuracy of the evaluation results. In addition, the PDCAAS score that exceeds 1 will be ignored, which limits its ability to distinguish different high-quality protein sources [32]. PDCAAS believes that protein that exceeds human needs will be metabolized and has no additional nutritional value, which eliminates the nutritional differences between high-protein foods such as soy protein isolate and whey protein.

3.3.3. Digestible Essential Amino Acid Score (DIAAS)

DIAAS (Digestible Essential Amino Acid Score) is similar to PDCAAS in terms of calculation principle, but differs in some key aspects. The DIAAS assessment is based on digestibility at the ileum level rather than at the fecal level, which reduces the impact of non-food protein metabolism in the large intestine on the assessment results. In addition, DIAAS measures digestibility at the level of individual amino acids rather than in units of protein, which allows the assessment to more accurately reflect the differences in digestibility of different amino acids. Another notable feature of DIAAS is that its score has no upper limit and can quantify the quality of protein that exceeds the human body's requirements.

When applying DIAAS for assessment, three different reference models were used according to the needs of people of different age groups, including an infant model based on the amino acid composition of breast milk, a model based on children aged 0.5 to 1 year and 3 to 10 years [14], which are suitable for infants aged 0-6 months, children aged 0.5-3 years and people over 3 years old, respectively.

Application studies of DIAAS have shown that it can effectively distinguish and rank the quality of different protein sources. For example, a study[21] compared the DIAAS values of whey protein, milk protein, soy, pea and wheat proteins and found that the DIAAS values of dairy products were generally higher than those of plant proteins, which is consistent with the PDCAAS evaluation results, but DIAAS provides a more detailed distinction. The results of PDCAAS and DIAAS methods for measuring the quality of common proteins can be seen in Table 2. In addition, the study found that DIAAS can be used in clinical applications that require more accurate measurement of protein quality[30] to select parenteral and/or enteral feeding regimens containing high-quality protein (rich in branched-chain amino acid leucine).

The DIAAS scoring system does not overemphasize the excess of certain EAAs when evaluating protein quality, which makes it effective in identifying those EAAs that are lacking in a specific protein source, thereby guiding the improvement of the overall protein quality of mixed protein foods. Studies have shown [22] that cereal proteins with low lysine but high methionine (Met) and cysteine (Cys) can complement legume proteins with high lysine but low Met and Cys, significantly improving the DIAAS score and improving the overall protein quality. A study measuring ileal digestibility in sows found [31] that the combination of milk and breakfast cereals can increase the DIAAS to nearly or even greater than 100, and the measured value is not significantly different from the DIAAS value predicted based on the individual ingredients.

Table 2. PDCAAS and DIAAS of common food protein sources

Protein Source	PDCAAS	DIAAS	Complete	Limiting AAs
Egg	1.00	1.22	yes	Methionine/cysteine
Whey protein isolate	1.00	1.25	yes	Histidine
Whey protein concentrate	1.00	1.33	yes	Histidine
Soy protein isolate	1.00	0.98	yes	Methionine /cysteine
Pea protein concentrate	0.86	0.73	no	Methionine /cysteine
Oat	0.57	0.68	no	Lysine
Wheat	0.63	0.53	no	Lysine
Sunflower seed	0.37	/	no	Lysine

*PDCAAS and DIAAS were calculated using the recommended amino acid scoring pattern for older child, adolescent, and adult.

The degree of protein complementarity is affected by the ratio of various protein sources. By adjusting the mixing ratio of different proteins, the quality and bioavailability of the overall protein can be maximized. Table 3 lists the DIAAS values of whey protein concentrate (WPC), whey protein isolate (WPI), soy protein isolate (SPI) and pea protein (PPC) at different mixing ratios. The combination ratio of different protein sources has a significant effect on the final protein quality. As shown in Table 3, mixing with soy protein isolate and pea protein in different ratios improves the DIAAS score of whey protein isolate. This concept of protein complementarity is of great significance for food formulation design.

Table 3. Different protein combinations alter DIAAS

Protein Mixture	Ratio	DIAAS
SPI/PPC	50/50	0.89
SPI/WPI	50/50	1.30
SPI/WPC	50/50	1.28
SPI/PPC/WPI	25/25/50	1.32
SPI/PPC/WPC	25/25/50	1.29

*DIAAS were calculated using the recommended amino acid scoring pattern for older child, adolescent, and adult.

**Composition and True ileal IAA digestibility of WPI, WPC, PPC and SPI were from Mathai J K, Liu Y, Stein H H. (2017).

However, DIAAS assessment also faces some challenges. First, the process of determining ileal digestibility is limited because it usually requires surgical means, including stoma at the end of the ileum or ileo-rectal anastomosis to collect samples [23], which may greatly limit the amount of data collected, and the establishment and application of the DIAAS database are also limited by this. Second, in terms of animal model selection, the digestive tract structure and physiological function of growing pigs are more similar to those of adults, making them an ideal model for evaluating protein digestibility [25], followed by growing rats [24]. However, in actual application, due to differences in the purpose of measurement and the diversity of animal model selection, inconsistent evaluation results may occur. Therefore, selecting appropriate animal models and developing standardized digestibility measurement methods are crucial to establishing a comprehensive and unified ileal amino acid digestibility database. It is worth noting that although DIAAS can provide more accurate protein quality information, its measurement process requires expensive technical equipment to calculate the digestibility of amino acids. This cost factor may limit the application of the DIAAS method in a wider range of fields [14].

3.4. In Vitro Evaluation Methods

Researchers simulate the digestive environment in the human body in the laboratory and measure protein quality by simulating static or dynamic digestion processes. Static models focus on the static reactions of chemicals, while dynamic models attempt to reproduce the gradual involvement of peristalsis and enzymes in the human body.

The in vitro protein digestibility (IVPD) method evaluates protein digestibility by comparing the original protein content in the food with the total protein content after digestion. On this basis, the in vitro protein digestibility corrected amino acid score (IVPDCAAS) method further corrects the first limiting amino acid to more accurately evaluate the amino acid availability compared with the reference protein. Nosworthy [27] used the ICPDCAAS method to measure buckwheat flour and bean flour mixed in different proportions, and also tested the quality of the real mixed protein through animal experiments. It was found that the in vitro experimental values were very close to the scores of the in vivo experiments. This suggests that the PDCAAS method can be used in practical applications to develop the best theoretical mixture before testing.

Raquel Sousa's study[28] evaluated a variety of protein sources, including whey protein (WPI), corn protein, collagen, black beans, pigeon peas, All-Bran®, and peanuts, using the INFOGEST in vitro digestion protocol. The results showed that the total protein digestibility and individual amino acid digestibility determined in vitro were highly consistent with the in vivo data, with minimal average differences. The correlation between the in vitro DIAAS values and the in vivo true ileal digestibility DIAAS values was very high, indicating that the in vitro method has a high accuracy in predicting in vivo digestibility.

In vitro analysis provides a low-cost and easy-to-operate means of predicting in vivo digestibility, but it is still unable to fully simulate the complexity of the in vivo digestive process[29]. In vitro models may not be able to fully reproduce the effects of gastric acid pH changes, intestinal motility, and microbial flora. Therefore, the in vitro evaluation results need to be further verified by in vivo experiments.

Overall, although in vitro methods provide convenience for the initial evaluation of protein digestibility and can be used for rapid screening, in vivo studies, especially data based on ileal digestibility, remain the gold standard for evaluating protein quality. Future studies should continue to optimize in vitro assessment techniques, improve their predictive accuracy, and perform valid comparisons with in vivo data.

4. Conclusion

Protein is an essential nutrient for the human body. Its source mainly depends on daily diet. It plays a vital role in maintaining good health and balanced nutrition. For many years, the Food and Agriculture Organization of the United Nations (FAO) and the World Health Organization (WHO) as well as researchers around the world have been committed to studying and improving the evaluation methods of protein quality. At present, a variety of evaluation methods have been developed, including nitrogen balance method, amino acid composition analysis, weight monitoring, protein digestibility determination, amino acid utilization evaluation and in vitro simulated digestion. These methods have their own characteristics, both advantages and limitations. Among them, the protein digestibility corrected amino acid score (PDCAAS) and the digestible essential amino acid score (DIAAS) are more scientific in theory and are widely recognized by food researchers and product development personnel at home and abroad. PDCAAS is considered to be a highly feasible and reliable evaluation method in the food industry due to its accuracy of evaluation results and relatively convenient operation procedures. It is used to improve production processes and processing methods, improve the quality of high-protein products for specific populations, and product nutrition labeling. The DIAAS method scores by measuring the digestibility of protein in the ileum, providing a more scientific and accurate assessment. It is particularly suitable for accurately measuring the quality of protein from different sources and predicting the nutritional value of different protein combinations.

In summary, in the process of developing high-protein quality health products, both PDCAAS and DIAAS methods are scientific and authoritative quality assessment methods, and can play an important role in evaluating production processes, product formulas, and nutritional claims. On a theoretical basis, if the focus is on the accuracy of the test results, the DIAAS scoring results are based on ileal digestibility and are a more accurate method. In practical applications, if it is necessary to comprehensively consider the difficulty and accuracy of sample collection, and to ensure the stability and consistency of research results when costs need to be controlled, PDCAAS has a low cost, rich application scenarios, and relatively reliable test results. It can be used as the preferred method for the food industry to develop high-protein quality products at low cost.

References

[1] Yang Yuexin, Ge Keyou. Encyclopedia of Chinese Nutrition Science[M]. People's Medical Publishing House: 2019.

[2] Atherton, P.J.; Etheridge, T.; Watt, P.W.; Wilkinson, D.; Selby, A.; Rankin, D.; Smith, K.; Rennie, M.J. Muscle full effect after oral protein: Time-dependent concordance and discordance between human muscle protein synthesis and mTORC1 signaling. Am. J. Clin. Nutr. 2010, 92, 1080–1088

[3] Witard, O.C.; Jackman, S.R.; Breen, L.; Smith, K.; Selby, A.; Tipton, K.D. Myofibrillar muscle protein synthesis rates subsequent to a meal in response to increasing doses of whey protein at rest and after resistance exercise. Am. J. Clin. Nutr. 2014, 99, 86–95.

[4] Herreman, L.; Nommensen, P.; Pennings, B.; Laus, M.C. Comprehensive overview of the quality of plant- And animal-sourced proteins based on the digestible indispensable amino acid score. Food Sci. Nutr. 2020, 8, 5379–5391.

[5] Millward, D.J. Identifying recommended dietary allowances for protein and amino acids: A critique of the 2007 WHO/FAO/UNU report. Br. J. Nutr. 2012, 108, s3–s21.

[6] Heda, R.; Toro, F.; Tombazzi, C.R. Physiology, Pepsin; StatPearls Publishing: Treasure Island, FL, USA, 2021.

[7] Jahan-Mihan, A.; Luhovyy, B.L.; Khoury, D.; Harvey Anderson, G. Dietary proteins as determinants of metabolic and physiologic functions of the gastrointestinal tract. Nutrients 2011, 3, 574–603.

[8] van der Wielen, N.; de Vries, S.; Gerrits, W.; Jannink, K.; Moughan, P.; Mensink, M.; Hendriks, W. Presence of unabsorbed free amino acids at the end of the small intestine of humans and pigs: Potential implications for amino acid bioavailability. Curr. Dev. Nutr. 2021, 5, 530.

[9] van der Wielen, N.; Moughan, P.J.; Mensink, M. Amino acid absorption in the large intestine of humans and porcine models. J. Nutr. 2017, 147, 1493–1498.

[10] Min L I, Wang Z L, Gou L Y, et al. Evaluation of the protein requirement in Chinese young adults using the indicator amino acid oxidation technique[J]. Biomedical and Environmental Sciences, 2013, 26(8): 655-662.

[11] Deglaire, A.; Moughan, P.J. Animal models for determining amino acid digestibility in humans--A review. Br. J. Nutr. 2012, 108, S273–S281.

[12] Boye, J.; Wijesinha-Bettoni, R.; Burlingame, B. Protein quality evaluation twenty years after the introduction of the protein digestibility corrected amino acid score method. Br. J. Nutr. 2012, 108, S183–S211.

[13] FAO/WHO Protein Quality Evaluation. Report of the Joint FAO/WHO Expert Consultation; Food & Agriculture Org.: Rome, Italy, 1991;

[14] Leser S. The 2013 FAO report on dietary protein quality evaluation in human nutrition: Recommendations and implications. Nutrition Bulletin. 2013;38(4):421-428.

[15] Chinese Nutrition Society. Dietary Nutrient Reference Intakes for Chinese Residents (2023 Edition) [M]. People's Medical Publishing House; 2023.

[16] HUGHES, G. J., RYAN, D. J., MUKHERJEA, R. & SCHASTEEN, C. S. 2011. Protein Digestibility-Corrected Amino Acid Scores (PDCAAS) for Soy Protein Isolates and Concentrate: Criteria for Evaluation. Journal of Agricultural and Food Chemistry, 59, 12707-12712.

[17] Nosworthy M G, Hernandez-Alvarez A J, Franczyk A J, et al. Effect of cooking on the in vitro and in vivo protein quality of soy, oat and wheat varieties[J]. Cereal Chemistry, 2023, 100(2): 460-472.

[18] WANG Y, TIBBETTS S M, BERRUE F, et al. A rat study to evaluate the protein quality of three green microalgal species and the impact of mechanical cell wall disruption[J]. Foods, 2020, 9(11): 1531.

[19] Nitrayová, S.; Brestenský, M.; Patráš, P. Comparison of two methods of protein quality evaluation in rice, rye and barley as food protein sources in human nutrition. Potravin. Slovak J. Food Sci. 2018, 12, 762–766.

[20] Sarwar G. The protein digestibility–corrected amino acid score method overestimates quality of proteins containing antinutritional factors and of poorly digestible proteins supplemented with limiting amino acids in rats[J]. The Journal of nutrition, 1997, 127(5): 758-764.

[21] Mathai J K, Liu Y, Stein H H. Values for digestible indispensable amino acid scores (DIAAS) for some dairy and plant proteins may better describe protein quality than values calculated using the concept for protein digestibility-corrected amino acid scores (PDCAAS)[J]. British Journal of Nutrition, 2017, 117(4): 490-499.

[22] Herreman L, Nommensen P, Pennings B, et al. Comprehensive overview of the quality of plant-And animal-sourced proteins based on the digestible indispensable amino acid score[J]. Food science & nutrition, 2020, 8(10): 5379-5391.

[23] Trommelen, J.; Tomé, D.; van Loon, L.J.C. Gut amino acid absorption in humans: Concepts and relevance for postprandial metabolism. Clin. Nutr. Open Sci. 2021, 36, 43–55.

[24] Ertl, P.; Knaus, W.; Zollitsch, W. An approach to including protein quality when assessing the net contribution of livestock to human food supply. Animal 2016, 10, 1883–1889.

[25] Deglaire, A.; Moughan, P.J. Animal models for determining amino acid digestibility in humans--A review. Br. J. Nutr. 2012, 108, S273–S281.

[26] FDATitle 21 of the United States Code of Federal Regulations, Part 101.9(c)(7)(ii). http://www. accessdata.fda. gov/scripts/ cdrh/ cfdocs/cfcfr/CFRSearch.cfm (accessed 12/6/2010).

[27] Nosworthy, M.G.; Franczyk, A.; Zimoch-Korzycka, A.; Appah, P.; Utioh, A.; Neufeld, J.; House, J.D. Impact of processing on the protein quality of pinto bean (Phaseolus vulgaris) and buckwheat (Fagopyrum esculentum Moench) flours and blends, as determined by in vitro and in vivo methodologies. J. Agric. Food Chem. 2017, 65, 3919–3925.

[28] Sousa R, Recio I, Heimo D, et al. In vitro digestibility of dietary proteins and in vitro DIAAS analytical workflow based on the INFOGEST static protocol and its validation with in vivo data[J]. Food Chemistry, 2023, 404: 134720.

[29] Bohn, T.; Carriere, F.; Day, L.; Deglaire, A.; Egger, L.; Freitas, D.; Golding, M.; Le Feunteun, S.; Macierzanka, A.; Menard, O.; et al. Correlation between in vitro and in vivo data on food digestion. What can we predict with static in vitro digestion models? Crit. Rev. Food Sci. Nutr. 2018, 58, 2239–2261.

[30] Paddon-Jones D, Coss-Bu JA, Morris CR, Phillips SM, Wernerman J. Variation in Protein Origin and Utilization: Research and Clinical Application. Nutrition in Clinical Practice. 2017;32(1_suppl):48S57S. doi:https://doi.org/ 10. 1177 / 0884533617691244.

[31] Fanelli NS, Bailey HM, Guardiola LV, Stein HH. Values for Digestible Indispensable Amino Acid Score (DIAAS) Determined in Pigs Are Greater for Milk Than for Breakfast Cereals, but DIAAS Values for Individual Ingredients Are Additive in Combined Meals. The Journal of Nutrition. 2021;151(3):540-547. doi: https://doi.org/10.1093/jn/nxaa398.

[32] Huang S, Wang LM, Sivendiran T, Bohrer BM. Review: Amino acid concentration of high protein food products and an overview of the current methods used to determine protein quality. Critical Reviews in Food Science and Nutrition. 2017;58(15):2673-2678. doi:https://doi.org/10.1080/ 10408 398. 2017. 1396202.