

# Research Progress and Application of Mung Bean Protein in 3D Food Printing: Performance Regulation and Application Expansion

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## Abstract:

3D food printing technology has opened up a new manufacturing path for personalized food customization. Mung bean protein, with its high nutritional value and excellent functional properties, has broad application prospects in this field. This paper reviews the extraction process and modification techniques of mung bean protein, focusing on the intrinsic connection between its key properties and 3D printing performance. It also explores the impact of pretreatment methods, printing parameter settings, and additive usage on the final 3D printing effect. By comparing mung bean protein with other legume proteins through evidence-based data, the paper demonstrates its application potential in areas such as meat product analogues, antioxidant functional snacks, and infant complementary foods. At the same time, it points out the current research deficiencies, such as the destruction of protein secondary structure due to high-temperature printing, the disruption of gel networks by freeze-thaw cycles, low sensory acceptance, and immature large-scale production control. Finally, it looks forward to future development directions, providing theoretical support for the scientific research and “tailored nutrition” of plant protein-based 3D printed foods.

**Keywords:** Mung bean protein; 3D food printing; Performance regulation; Rheological properties; Application of mung bean protein.

## 1. Introduction

With the upgrading of consumption and the diversification of health demands, the food industry is

transitioning from standardized production to personalized customization. The emergence of 3D food printing technology is a response to this trend. It not only enables the customization of complex-shaped

foods through precise structural design, meeting the personalized shape demands of special groups such as children, but also flexibly adjusts raw material components based on individual nutritional data, providing 'tailored nutrition' for each person and offering a new technological platform for the development of functional foods and special diets. Moreover, its advantages in reducing food waste and promoting the intelligence of the food industry have further accelerated its application penetration in the food field. Additionally, the core of this technology lies in the printing materials, which must have good extrudability and formability. The size and quality, as key factors for evaluating the printing effect, are significantly influenced by the entire process including pretreatment, printing implementation, and post-treatment [1].

Among the numerous potential printing raw materials, plant proteins have become a research hotspot due to their high nutritional value and strong sustainability of resources. Among them, mung bean protein, with its unique composition and functional characteristics, has gradually entered the research field. It is rich in lysine, has a high digestibility, and exhibits excellent gel properties and biological functions such as antioxidation. Compared with pea and broad bean proteins, after appropriate modification, mung bean protein shows superior elasticity, shear thinning properties, and stability during frying, which precisely meet the core requirements of 3D food printing for raw materials. It becomes an ideal raw material for 3D-printed meat substitutes [1].

However, current research mainly focuses on the evaluation of the printing performance of a single protein, lacking comprehensive comparisons between mung bean protein and other bean proteins. Moreover, the exploration of the regulatory mechanism for 3D printing performance is also somewhat insufficient. Therefore, this paper intends to integrate the latest research achievements to reveal the core mechanism of 3D printing of mung bean protein and its application prospects, providing more theoretical reference basis for its clinical application.

## 2. Application Foundation of Green Bean Protein

### 2.1 Physical and Chemical Properties

Mung bean protein mainly contains 7S, 8S and 11S globulins. Among them, 8S globulin accounts for 89.0% and contains 16–60 kDa subunits and specific N-terminal amino acid sequences. It also has 68% amino acid sequence homology with soybean  $\beta$ -protein and  $\alpha$ -granule protein. This structural similarity provides a molecular basis for

the regulation of its functional characteristics [2]. The protein secondary structure contains  $\alpha$ -helix,  $\beta$ -sheet, etc. The maximum fluorescence emission is 393 nm; it is an irregular spherical shape, with a particle size of 6.75–31.28  $\mu\text{m}$ , and the denaturation temperature is 84.6°C, with lower thermal stability than soybean and chickpea proteins [3]. The solubility shows a „U-shaped“ pH response. The isoelectric point is at pH 5.0, and it reaches 80% - 100% at pH 3.0 or 7.0. It should be noted that there is a pH-dependent difference in the solubility of 8S and 11S globulins: at pH 4.0, the solubility of 11S (20%) is higher than that of 8S (5%), and the trend reverses when the pH drops to 2.0; in terms of ionic strength compatibility, 8S can be dissolved in 0.1 - 0.5 M NaCl, while 11S can only be dissolved in 0.35 M NaCl, and the difference stems from the different surface charge density and molecular conformation of the two [2]. Additionally, by combining carboxymethyl cellulose and other polysaccharides, hydrogen bonds can be formed to construct protein-polysaccharide complexes, which can increase the solubility from 1.69% to 43.62% near the isoelectric point, solving the problem of protein precipitation [2].

The minimum gel concentration is 12%. At concentrations of 15%–20%, it exhibits viscoelasticity and shear thinning behavior, which is suitable for 3D printing requirements [3]. The water retention is 1.7–2.2 g/g, and the oil retention is 6.1–6.7 g/g. The water retention is positively correlated with the content of 8S ( $R^2 = 0.87$ ). Because the surface of 8S exposes more hydrophilic amino acid residues, the oil retention is related to the proportion of hydrophobic regions. In neutral conditions, the oil retention is significantly better than that of soy protein [2]. The foaming capacity in a neutral environment is 62.50% and the stability is 95.20% [3].

### 2.2 Nutritional Characteristics

Mung bean protein contains 18 kinds of amino acids. The lysine content is 3.49–7.74 g/100g protein, which is higher than that of rice, wheat and other grains, and can make up for the deficiency of lysine in grains; the limiting amino acids are methionine and cysteine, and glutamic acid and aspartic acid are the main non-essential amino acids [3]. From the comparison of essential amino acids, the leucine content of the isolated mung bean protein is higher than that of pea protein and soy protein. Except that the sulfur-containing amino acids are slightly lower than those of soy protein, all the essential amino acids can meet the daily requirements of children and adults aged 1–18 [2]. The digestion and absorption performance of mung bean protein is outstanding. The corrected amino acid score of its digestibility reaches 93, which is higher than that of red

beans and millet, etc. After heating or enzymatic hydrolysis, the in vitro true protein digestibility can be increased to 99.26%, approaching the level of animal protein [3]. Protease inhibitors and phytic acid in mung beans, which are anti-nutritional factors, can affect digestibility. Through specific processes, they can be eliminated: boiling for 30 minutes reduces the activity of protease inhibitors by 75%, and germination for 48 hours combined with boiling for 20 minutes can completely eliminate them; extrusion treatment reduces phytic acid from 1.2 mg/g to 0.3 mg/g, reducing interference to digestion. Moreover, after simulated gastrointestinal digestion, the soluble nitrogen ratio reaches 64.14%. This in vitro data has a correlation with the in vivo DIAAS value (93) with  $R^2 = 0.92$ , verifying the high digestibility [2].

Meanwhile, the soybean protein has abundant biological activity. The <3kDa peptide segments obtained through Alcalase enzymatic hydrolysis have a DPPH scavenging rate of 95.3%, and the  $Fe^{2+}$  chelation activity  $IC_{50}$  is 14.84  $\mu$ g/mL. They can also eliminate reactive oxygen species and enhance antioxidant enzyme activity [3]. It contains 27kDa  $\alpha$ -amylase inhibitor and angiotensin-converting enzyme inhibitory peptides, which can regulate the intestinal flora [3]; after being fermented by *Rhizopus microsporus*, the hydrolysis degree can reach 18.3%, generating more

active peptides, enhancing antioxidant and antibacterial functions, providing a direction for the development of functional foods [2].

### 3. Extraction, Modification and Pre-treatment of Mung Bean Protein

#### 3.1 Extraction Method

The extraction of mung bean protein mainly adopts two methods: wet method and dry method. In the wet method, it includes alkali dissolution and acid precipitation methods as well as salt dissolution method. Among them, the alkali dissolution and acid precipitation method can obtain highly pure separated protein, but this method requires a large amount of water and is prone to cause wastewater pollution. The salt dissolution method can better retain the functional components of the protein, although its purity is relatively lower. The dry method extraction, such as air classification method, has the advantages of low energy consumption and environmental friendliness, but the purity of the extracted protein is relatively lower and is suitable for food systems with low requirements for purity [4]. As shown in Table 1 for details.

**Table 1. Extraction Methods of Mung Bean Protein**

Extraction Method	Core Characteristics	Advantages	Disadvantages	Applicable Scenarios
Wet Method - Alkali-Soluble Acid-Precipitation Method	Separating proteins through alkali dissolution and acid precipitation steps	High-purity isolated proteins can be obtained	High water consumption, prone to wastewater pollution	3D printing systems with high requirements for protein purity
Wet Method - Salt-Soluble Method	Dissolving proteins using salt solutions	Better retention of protein functional components	Relatively low protein purity	Printing scenarios requiring retention of protein functional activity
Dry Method - Air Classification Method	Extracting proteins by means of air classification technology	Low energy consumption, environmentally friendly	Relatively low protein purity	Food systems with low requirements for protein purity

#### 3.2 Modification Technology

For physical modification, ultrasonic treatment can disrupt the aggregation of protein macromolecules through the cavitation effect, reducing the particle size from 818nm to 172nm, significantly improving the solubility and emulsifying stability index of mung bean protein, while enhancing the intermolecular hydrophobic interaction and hydrogen bond binding, increasing the storage modulus by more than 25%, and meeting the requirements of 3D printing for material viscoelasticity; while high-pressure treatment can change the structure by disrupting protein

hydrophobic and electrostatic interactions, optimizing the density of the gel network, but excessive pressure can lead to excessive protein aggregation, deteriorating the rheological properties, and thus is not conducive to the stability and support of the printed structure after printing [3]. In addition, non-thermal physical modification techniques can achieve precise control of the function of mung bean protein: cold plasma treatment can increase the hydrophobicity of the mung bean protein surface by 35%, while reducing the allergen content by 40%, and will not damage the secondary structure of the protein, effectively reducing the risk of food allergy during printing and masking

some bean odors; ultra-high pressure homogenization can further reduce the particle size of mung bean protein from 818nm to 150nm, increasing the storage modulus to 1200Pa, reducing the shear thinning behavior index  $n$  to 0.16, more closely meeting the core requirements of 3D printing for „easy extrusion and stable shaping“, especially suitable for precise replication of complex shapes [5].

In chemical modification, glycosylation can increase the proportion of  $\beta$ -folding structure in mung bean protein by 17.5%, enhance the shear thinning behavior, reduce the flow behavior index from 0.22 to 0.18, and lower the extrusion resistance, meeting the core requirements of „easy extrusion and stable forming“ during printing. Phosphorylation modification can introduce anionic phosphate groups, increasing the water-holding capacity by 35.9%, reducing interlayer cracking caused by water loss during printing, but excessive phosphorylation will lead to excessive cross-linking of the protein, resulting in a 15% or more decrease in foaming stability and affecting the uniformity of the printing material [5].

In biological modification, the cross-linking catalyzed by transglutaminase can enhance the network strength of protein gels by forming interpeptide bonds. When the addition amount is 4U/g, it can significantly improve the printing accuracy to  $\pm 0.1$ mm and reduce inter-layer offset; however, when the enzyme dosage exceeds 8U/g, it will cause excessive polymerization of protein molecules, resulting in a rough surface structure, and increase the brittleness of the gel. During the printing process, it is prone to nozzle blockage or fracture after molding, which is not conducive to the precise replication of complex shapes [6]. Additionally, the „chemical-biological composite modification“ strategy can further optimize the printing adaptability and functional characteristics of mung bean protein: first, through the Maillard reaction, the proportion of  $\beta$ -folding structure of mung bean protein is increased by 17.5%, enhancing the shear thinning ability of the system; then, using *Lactobacillus plantarum* fermentation for 72 hours, the protein hydrolysis degree reaches 18.3%, generating more antioxidant active peptides with a molecular weight of  $< 3$ kDa. Ultimately, not only does the DPPH free radical scavenging rate increase from 35% to 82%, but the room temperature collapse rate of the printed samples can be reduced from 12.5% to 2.1%, while the fermentation process can degrade the precursor substances of bean odor, further improving the sensory acceptance of the product [4].

### 3.3 Preprocessing Method

In the physical pre-treatment stage, multiple techniques can optimize the 3D printing compatibility by regulating

the structure and properties of mung bean protein and the composite system. Among them, microwave treatment can increase the starch gelatinization degree from 42% to 89% in the composite system, significantly reducing the viscosity of the system, reducing the shear resistance during extrusion, and enhancing the interaction between protein and starch, thereby improving the uniformity of the printing material. Thermal treatment can promote the unfolding of protein molecules and form an ordered gel network, increasing the storage modulus by more than 40%. The collapse rate of the printed sample placed at room temperature for 2 hours is only 3.2%, which is much lower than that of the untreated group (12.5%). In addition, ultrasonic pretreatment can disrupt the protein aggregation state, reduce the particle size from 750nm to 210nm, enhance the fluidity of the system, increase the interlayer fusion of printing by 18%, and further optimize the molding effect [7].

Additive regulation is a key means to improve printing performance. Besides the basic compounding scheme, different additives can specifically address specific problems in printing. When compounding carboxymethyl konjac glucomannan, its molecular chain can form hydrogen bonds and hydrophobic interactions with mung bean protein, increasing the gel strength from 120g/cm<sup>2</sup> to 280g/cm<sup>2</sup>, while the ratio of elastic modulus to storage modulus remains below 0.25, ensuring that the system is dominated by elasticity. The supportability of the complex structure after printing is enhanced, with the deformation rate being lower than 5%; adding xanthan gum can lock moisture by constructing a three-dimensional network structure, increasing the adhesive force of the system by 35%, and reducing the inter-layer fracture rate from 11% to 2% after printing. It is particularly suitable for multi-layer complex shape printing [8]. At the same time, compounding 0.3% sodium alginate can enhance the pseudoplasticity and thixotropy of the system, reducing the flow behavior index to 0.15, and quickly restoring the structural stability after extrusion, reducing the „stringing“ phenomenon after nozzle discharge, and further improving the printing accuracy [8].

## 4. Key Performance and Regulatory Mechanism of 3D Printing of Soybean Protein

### 4.1 Key Performance and Printing Compatibility

Mung bean protein exhibits key performance that is highly compatible with 3D printing at a concentration ranging



from 15% to 20%. It has excellent viscoelasticity, with the storage modulus being much greater than the loss modulus, presenting typical solid-like behavior, providing a basic support for the printing structure; and it has a significant shear thinning behavior, with the flow behavior index being less than 1, which can reduce viscosity during extrusion and quickly restore the structure after molding, facilitating precise molding [4].

In terms of gel and print compatibility, at this concentration, mung bean protein can form a continuous and dense gel network, featuring both good cohesion and molding stability, it can maintain a uniform ink state before printing and quickly fix the three-dimensional configuration after extrusion, reducing the risk of interlayer collapse [4]. After being modified by transglutaminase, the gel strength and network regularity are further improved. This not only significantly increases the yield stress, enhancing the anti-collapse ability, but also optimizes the water retention property to reduce deformation caused by water migration during printing. At the same time, the dense gel structure can improve the precision and integrity of the printed products [9].

## 4.2 Factors Affecting Printing Performance and Optimization

Among the formula parameters, the ratio of protein to water directly affects the printing smoothness and shape retention effect. When the ratio is 1:1, the viscosity of the system is too high, making it impossible to extrude through the nozzle; when the ratio increases to 1:3, the viscosity drops sharply, and after printing, the structure is prone to collapse; while when the ratio is 1:2.5, all three protein-based inks present the optimal state, with the flow behavior index ranging from 0.120 to 0.171. This enables smooth extrusion and maintains a clear outline after printing, with a design retention rate exceeding 90% [4].

On the printing parameters, the nozzle diameter, speed and layer height need to be precisely matched to ensure the optimal printing accuracy. Experiments have shown that when the nozzle diameter is set to 1.5mm, the printing speed is controlled at 14mm/s, the layer height is adapted to the nozzle diameter in a 1:1 ratio, combined with a 60% filling density and a linear filling pattern, the size deviation of the printed sample can be controlled within  $\pm 0.5$ mm, with a smooth surface without inter-layer gaps. This is significantly superior to other parameter combinations [1].

The post-treatment adaptability of the soybean protein-based samples is excellent. After being fried, the shape shrinkage rate of these samples is low, and the shells are uniformly formed. Specifically, for the soybean

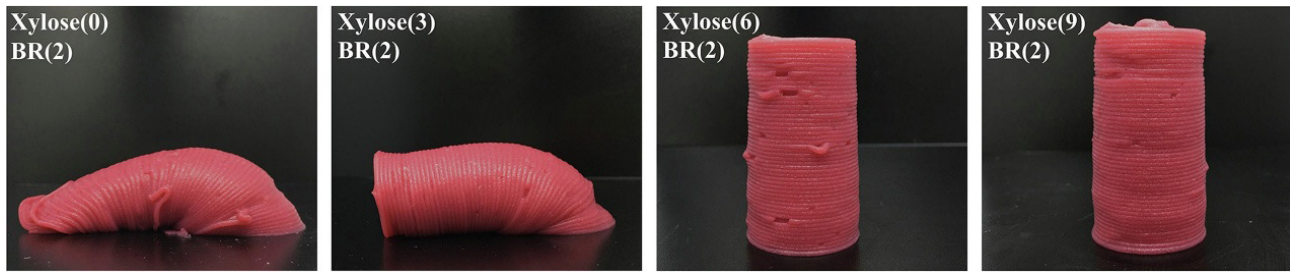
protein-based samples prepared with a protein-to-water ratio of 1:2.5, after being frozen at  $-18^{\circ}\text{C}$  for 12 hours and then fried in soybean oil at  $160^{\circ}\text{C}$  for 3 minutes, the height shrinkage rate is only 2.5%, which is much lower than that of pea protein-based and broad bean protein-based samples; at the same time, the uniformity of the shell thickness reaches 92%, with no local over-frying or damage, and can effectively preserve the integrity of the cubic structure during printing [4].

## 5. Application Scenarios and Case Analysis

### 5.1 Analogous Products for Meat Products

As consumers' demands for sustainable diets and health increase, plant-based protein-based meat substitutes have become a research hotspot in the food industry. The core requirement lies in precisely regulating the properties of raw materials and processing techniques to replicate the fiber structure, texture, and sensory characteristics of animal meat [10]. Mung bean protein, due to its excellent gelation properties and modification potential, demonstrates significant advantages in this field. By optimizing its concentration and adding transglutaminase, a dense and directional fibrous network structure can be constructed. The simulated meat printed out shows, through texture profile analysis, a hardness of 7.23N and a chewing force of 1.76N, which is highly similar to the texture parameters of pork tenderloin. At the same time, its storage modulus can reach 122562Pa, ensuring that the structure remains stable and does not collapse after heating [4].

To further enhance the sensory authenticity, 0.5% natural beet red pigment was added to the mung bean protein-based printing system, which could simulate the red-brown color change process of meat when heated: when not heated, the system presented a bright red color due to the uniform dispersion of the pigment. After being fried at  $160^{\circ}\text{C}$  for 3 minutes, a Maillard reaction occurred between the pigment and the protein, and the color turned to a natural brownish color, consistent with the thermal color change pattern of real meat [4]. Compared with the soybean protein-based simulated meat, the mung bean protein system has a higher oil retention capacity, and the oil distribution after frying is more uniform, avoiding a greasy surface, and its corrected amino acid score for digestion is better than that of soybean protein, with better nutritional and taste compatibility [10]. This technical approach provides a feasible solution for the industrial production of highly realistic plant-based meat. As shown in Figure 1 for details.



**Fig. 1** Printing behavior of raw colorant-containing meat analogs depending on the amount of xylose added; BR: beetred Unit:%.

## 5.2 Antioxidant Functional Snacks

Mung bean protein is a natural precursor of antioxidant active substances. The amino acid residues such as tyrosine and histidine contained in its molecular structure can be converted into active peptides with specific sequences after enzymatic hydrolysis, making it the core raw material for antioxidant functional snacks. Using the alkaline dissolution and acid precipitation method to extract high-purity mung bean protein as the raw material, and after enzymatic treatment with Alcalase and others, the active peptide components with a molecular weight of less than 3 kDa are obtained. These components not only retain the natural nutrition of mung bean protein but also significantly enhance biological activity. They can exert antioxidant effects by eliminating free radicals and regulating the oxidative stress pathway [3].

After combining the active peptide with xanthan gum and other components, the system exhibits a shear-thinning characteristic with a flow behavior index  $n < 1$  and a viscoelasticity where the storage modulus is greater than the loss modulus, which meets the requirements of „easy extrusion and stable shaping“ for extrusion-based printing. By adjusting parameters such as nozzle diameter and printing speed, various structural customizations such as cartoon shapes and thin and crispy sheets can be achieved [7].

After printing and undergoing post-treatment such as low-temperature baking, it still retains over 85% of its antioxidant activity and has no beany flavor or a crispy texture; relying on 3D printing technology to achieve the combination of „individualized shape + precise function“, it meets the demands of consumers for healthy snacks [3,7].

## 5.3 Infant and Toddler Supplementary Food

Mung bean protein is an ideal base material for infant complementary foods. Its amino acid composition is balanced, and its lysine content is significantly higher than that of cereal proteins. It can form nutritional complementation with cereal-based complementary foods, and

its molecular structure is simple and the digestion and absorption rate is high, which is suitable for the delicate digestive system of infants [3]. Compared with soybean protein, mung bean protein has no bean odor and has a higher sensory acceptance. At the same time, it has good gel properties and shear thinning behavior. After physical modification, the energy storage modulus and yield stress can meet the core requirements of 3D printing for the extrudability and formability of raw materials, providing a basis for the structural customization of complementary foods [3,4,7].

## 6. Existing Issues and Future Prospects

### 6.1 Key Challenges

Currently, the practical application of mung bean protein in 3D food printing technology faces multiple challenges: Printing in high-temperature environments can easily cause the secondary structure of mung bean protein to be disrupted, affecting the product quality; The stability during the post-processing stage is poor. The freezing-thawing cycle will cause the structure to be damaged due to ice crystal compression, and post-heating processing is prone to causing excessive protein aggregation, resulting in a harder texture [4,9]. In addition, the low sensory acceptance of consumers for the product has become a major obstacle in market promotion: Firstly, after 3D printing and post-processing of mung bean protein, it is prone to form a rough texture due to protein aggregation, and the natural beany smell has not been effectively masked, resulting in a gap from the delicate taste and flavor of traditional snacks; Secondly, during the printing process, water gels and other auxiliary materials added to ensure the shape stability may cause the product to have a sticky sensation or an unpleasant smell, further reducing the sensory experience [3,7].

### 6.2 Future Outlook

Regarding the core challenges in 3D food printing of

mung bean protein, future research can break through from three aspects: raw material modification, process optimization, and application expansion, to promote industrialization: First, develop „physical+biological“ combined modification technology, such as ultrasonic pretreatment combined with Alcalase enzymatic hydrolysis, to regulate the molecular weight of active peptides, while retaining antioxidant activity and improving thermal stability; through phosphorylation and glycosylation modification, enhance protein water-holding capacity and reduce the structural damage rate during freezing-thawing cycles; Second, in the pre-treatment stage, use microwave to cooperate with starch gelatinization to improve the rheological properties of the protein system; in the printing stage, based on the „size stability window“ theory, match the nozzle diameter, printing speed and yield stress parameters; post-processing develops low-temperature baking and vacuum freeze-drying processes to avoid texture hardening caused by excessive protein aggregation; Third, use lactic acid bacteria fermentation to degrade the precursor substances of bean off-flavors, optimize the ratio of xanthan gum and other auxiliary materials, balance the formability and taste; expand to special food fields, such as custom infant complementary food and dietary plans for people with swallowing difficulties, relying on 3D printing to achieve „form - nutrition“ dual customization.

## 7. Conclusion

Mung bean protein, with its outstanding performance and good printing adaptability, has demonstrated unique advantages in the field of 3D food printing, especially in the development of meat product simulators and functional foods (such as antioxidant foods, infant complementary foods, etc.), where it has broad application prospects.

By optimizing the extraction and modification processes, adjusting the printing parameters and post-processing procedures, the performance stability and product quality can be further enhanced. However, at present, green bean protein still faces multiple bottlenecks in the industrialization process of 3D printing: High-temperature printing and post-processing heating are prone to causing the disruption of protein secondary structure, resulting in hardening of texture; the ice crystal effect during the freezing-thawing cycle will damage the integrity of the gel network; the natural bean odor and the stickiness of the additives restrict the sensory acceptance; meanwhile, the control of raw material uniformity and cost management in large-scale production have not yet formed a mature solution.

In the future, efforts should be made to break through the technical bottlenecks of large-scale production, solve key problems such as high-temperature denaturation and struc-

tural damage caused by freezing and thawing, and provide new paths for the high-value utilization of plant proteins. At the same time, efforts should be made to enhance the research on industrial standardization of mung bean protein, promote its large-scale application in personalized nutrition and special needs foods, and achieve precise nutrition supply tailored to each individual.

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