PLANT SCIENCE TODAY
ISSN 2348-1900 (online)
Vol 9(4): 820–828
https://doi.org/10.14719/pst.1652

HORIZON
e-Publishing Group
HePG

REVIEW ARTICLE

Plant based proteins: Sustainable alternatives

Krishna Mistry[1], Sami Dilip Sardar[1], Hina Alim[1], Nimisha Patel[2], Mansee Thakur[3], Dilfuza Jabborova[4] & Ahmad Ali[1*]

[1]Department of Life Sciences, University of Mumbai, Vidyanagari, Santacruz (East), Mumbai 400 098, India
[2]Department of Life Sciences, J. C. Bose University of Science & Technology, YMCA, Faridabad 121 006, Haryana, India
[3]Department of Medical Biotechnology, MGMSBS, MGMIHS, Kamothe, Navi Mumbai, India
[4]Laboratory of Medicinal Plants Genetics and Biotechnology, Institute of Genetics and Plants Experimental Biology, Academy of Sciences of Uzbekistan, Tashkent, Uzbekistan

*Email: ahmadali@mu.ac.in

OPEN ACCESS

ARTICLE HISTORY
Received: 02 January 2022
Accepted: 04 August 2022

Available online
Version 1.0 : 15 September 2022
Version 2.0 : 01 October 2022

Additional information

Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

Reprints & permissions information is available at https://horizonepublishing.com/journals/index.php/PST/open_access_policy

Publisher's Note: Horizon e-Publishing Group remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Indexing: Plant Science Today, published by Horizon e-Publishing Group, is covered by Scopus, Web of Science, BIOSIS Previews, Clarivate Analytics, NAAS etc.
See https://horizonepublishing.com/journals/index.php/PST/indexing_abstracting

CITE THIS ARTICLE

Mistry K, Sardar S D, Alim H, Patel N, Thakur M, Jabborova D, Ali A. Plant based proteins: Sustainable alternatives. Plant Science Today. 2022; 9(4): 820–828. https://doi.org/10.14719/pst.1652

Abstract

Proteins can be procured from both plants and animals. Plant proteins are more preferable as the animal proteins may cause adverse health effects to human life. Proteins derived from plant sources are less expensive and hence are more cost effective. Quality of proteins relies on several factors and biological value is one such factor. Proteins with major essential amino acid are with high biological value. Every plant source is deficient in one or more essential amino acids so it is recommended to include multiple plant-based diets. Also proteins obtained from plant sources are less palatable so it is important to add flavor in order to make it more palatable. The quality and quantity of the proteins also depend on the techniques used for isolation and purification of proteins. Elucidation of the structure of proteins involves the use of techniques like nuclear magnetic resonance, X-ray crystallography and spectroscopy. Apart from the structural analysis the functioning of the protein could be determined by amino acid sequencing which could be performed using mass spectroscopy. Ultrasound assisted extraction, enzyme assisted protein extraction and electro activation method are few of the isolation and purification method which can be used in isolation and purification of these proteins. Owing to the vast availability of plant-based proteins it has various industrial applications like, plant based protein can be used as a dairy substitute, plant based meat analogue and its use as bioactive peptides which have been briefly discussed in the review.

Keywords

Biological value, essential amino acids, legumes, plant proteins, pseudocereals

Introduction

With the rise in population there is increase in demand of food supply. In such condition fulfilling the nutritional requirement not just involves the adequate calorie intake but also intake but also intake of proper macronutrient. Proteins are one of the important macronutrient, however the production of proteins on such largescale is of major concern. Proteins derived from animal source are considered to be of superior quality, but the resources required to sustain the continuous requirement is not adequate (1) In order to overcome this problem finding an alternative protein source as become necessary. Plant based proteins represent a promising solution as it could be cultivated easily at lower productivity cost and are also environmentally sustainable.

The quality of the protein depends on its constituent amino acids and the human body contain 20 amino acids which are further classified as essential and nonessential amino acids. Nonessential amino acids, are synthesized in our body and are not required essentially through the external diet; which includes alanine, arginine, aspartic acid, cysteine, glutamic acid, glycine, proline, serine, and tyrosine. On the other hand, essential amino acids are not made by our body and should be supplied through the external diet. This group includes histidine, leucine, isoleucine, valine, lysine, phenylalanine, methionine, threonine and tryptophan (2).

Table 1. Comparison between plant and animal source of protein

Animal protein	Plant protein	Reference
Less environmentally sustainable	Environmentally sustainable	(5)
Negative health impacts as it can cause various health hazards	Positive health impacts as it provide greater food safety	(6)
High cost due to which it is not affordable	Low cost easily affordable	(7)
Rich in all essential amino acids and provide complete protein	Deficient in essential amino acids as it provide incomplete protein	(6)
It consists of saturated fat and may contain harmful toxins and low in antioxidants	It consist of unsaturated fats and fiber also rich in various antioxidants and useful bioactive compounds and minerals	(8)

The nutritive value of protein depends on their amino acid content, bioavailability, purity, digestibility, antinutritional factor (substances that when present affects the availability of proteins by themselves or by some metabolic reactions) etc. Animal protein such as egg, meat, poultry, milk and fish contain complete and high quality proteins as they provide all the essential amino acids. However, they also include fats and cholesterol which are threat to human health and may cause heart disease, high blood pressure, stroke etc (3). Consuming more animal proteins are also associated with depletion of natural resources, damage to biodiversity, climate crisis, freshwater depletion etc (3). Therefore, there is shift towards plant-based proteins in recent times. However, these proteins are deficient in one or more essential amino acids.

Replacing plant proteins with animal proteins put forth various challenges. It is very important for protein content and quality should be intact while it is undergoing various processing. The cost of processing and raw material should be affordable. Plant based protein food should be tasteful flavors can be added to make it palatable. It should be easily digestible and available. The effects of anti-nutrients and allergens should be minimized (4).

Individuals who depend completely on plant proteins should have variation in their plant based diet i.e., they should have all types of plant diet which include legumes, cereals, seeds, nuts, variety of fruits and vegetables etc. to meet their protein demands. Plant proteins provide excellent source of protein with reduced fats and cholesterol.

As the world population is growing day by day and to provide safe and nutritious food to the current population without having any negative impact to the environment and maintain healthy ecosystem, it is important to replace traditional animal proteins and develop proteins with better digestibility and bioavailability. Plant proteins are inferior to animal proteins and are often recognized as incomplete. Table 1. Shows comparison between plant and animal sources of proteins. Therefore, protein products are developed with the help of advanced techniques by blending various plant proteins. While developing new formulations of protein, it is important to note that the protein should remain stable and active during transportation, storage and administration inside the human body, as these proteins are prone to degradation (3).

Biological value

There are various approaches to determine the quality of protein one such approach is to determine its biological value. Protein quality or its biological value relies on the amount of essential amino acids consumed through diet (9). Biological value of protein is defined on the basis of number of amino acids present in the particular protein sources that can be digested, absorbed and are utilized by the body to form new proteins (10).

Proteins of superior quality are those which have the most essential amino acid content in them i.e., they are with high biological value whereas proteins of inferior quality have low biological value since they lack one or more amino acids especially the essential amino acids (11). Table 1 represents a summary of biological value of important sources of proteins.

The biological value of protein is calculated by estimating the amount of nitrogen consumed and eliminated by the body as nitrogen is the main component of amino acids. Experimental animals with 2 dietary condition groups are considered; one by feeding the nitrogen free diet and the test sample are kept on protein diet. The amount of nitrogen lost in urine and fecal matter are calculated in both the groups.

The number of amino acids absorbed and retained by the body represents the biological value. Through the formula given below, the biological value of plant or animal sources can be calculated (12). Table 2 gives information about biological value of some plants and animal sources.

$$\text{Biological Value (BV)} = \frac{\text{reatained N}}{\text{absorbed N}} = \frac{[\text{N intake}]-[\Delta\text{ fecal N}]-(\Delta\text{ urinary N})}{[\text{N intake}]-(\Delta\text{ fecal N})} \quad \text{.......Eqn.1}$$

(where N = nitrogen content)

Table 2. Biological value of various protein sources (12)

Sources of Protein	Biological value
Egg	94
Fish	76
Beef	74
Sunflower seed	70
Casein	80
Oats	65
Rice	65
Peanuts	55
Soybeans	73
Wheat	65
Maize	59
Lentils	45

There are some other methods by which protein quality can be assessed such as net protein utilization (NPU) and protein efficiency rate (PER). Some researchers find NPU more suitable as it also considers the digestibility of the protein. In case of PER, it is assumed that all the protein are used for growth.

Sources of proteins

Animals, microorganisms and plants are the different sources of proteins. Major animal proteins include milk, egg, poultry, meat and fish which provide complete proteins with major essential amino acids. Egg proteins are divided among yolks and egg white. They also contains fats and cholesterol due to which it is considered as unhealthy and its consumption is declining over a past few decades especially for elderly people (13). Milk proteins are classified in to 2 major proteins i.e. whey and casein both are complete protein and also contain minerals like calcium and phosphorus (14). Meat proteins are associated with fatty acids mostly saturated fatty acids as well as cholesterol. There are many health-related problems associated with meat proteins. Heart disorders due to high cholesterol and fat, type-2 diabetes, bone health due to absorption of sulphur containing amino acids in animal proteins, high animal protein leads to more urinary excretion of calcium which affects bone density and leads to osteoporosis, high blood pressure etc (15). Fishes are rich source of proteins and also contains polyunsaturated fatty acids mainly omega-3 fatty acids; vitamins; minerals like calcium, zinc and iron. However, due to bioaccumulation of heavy metal like mercury, lead, nickel etc. fishes have become a matter of concern for human health (16).

Major plant protein sources

Legumes

Legumes belong to the family *Leguminosae*. They play an important role in human diet due to their rich protein content as well as certain minerals, vitamins and calories. Legumes are considered as poor man's meat as they are rich source of protein at low cost (17,18). Pulses which belong to the family of legumes are major contributors of protein in African and Asian diets. Some common pulses shown in Fig 1. include beans, peas, pigeon peas, chickpeas, peanuts (ground nuts), faba beans, soybeans, lentils, mung, kidney beans (also known as common or dry beans), cowpeas, and black gram (19).

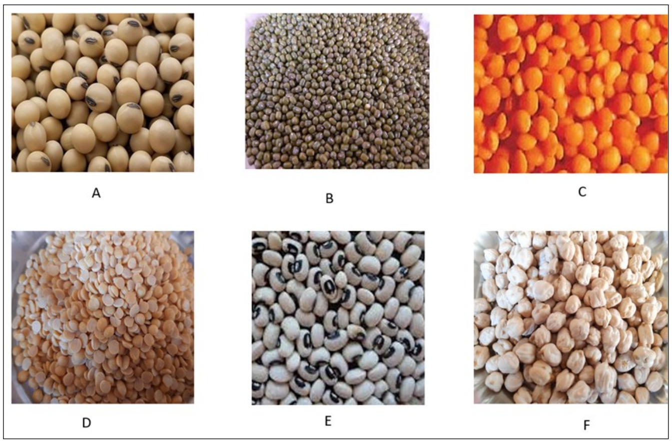

Fig. 1. Lentils and legumes: The given figure represents different sources of lentils and legumes. (**A**) Soy beans; (**B**) Green gram beans; (**C**) Red lentil; (**D**) Bengal gram; (**E**) Cow peas; (**F**) Chickpeas.

Soy proteins

Soy beans are legumes grown as pulses and for extracting oils. Soy beans contain approximately 34 to 37% of protein which is highest in comparison to other cereals and legumes. They also contain carbohydrates and dietary fibre. Human consumption needs a little more processing after which various soy products are manufactured, which includes soy milk for kids, soy flour, soy concentrates, tofu and soy isolates. Soy proteins include storage proteins like glycinin and β-conglycinin which contain most of the essential amino acids but low in sulfur containing amino acids like i.e., cysteine and methionine. Other minor protein includes lectins, lipooxygenase etc. but these proteins are removed as they may disturb the nutritional quality of proteins and also affects the taste (20).

Lentils

Lentils are the legumes belongs to the *Leguminosae* family. They are called "lentil" due to their lens shaped structure. Various types of lentils are available in market for consumption. Some of the very commonly used are green gram beans, red lentils, yellow pigeon pea, green and white peas, bengal gram, black gram etc. Lentils are rich in essential amino acids like phenylalanine, leucine, threonine, lysine, but contain low amount of sulfur containing amino acids like cysteine and methionine. Lentils also contain minerals like iron, potassium, phosphorus, zinc etc. They are also rich sources of vitamin B. Lentils are often consumed with cereals as they make complete protein source together (21).

Cereals

Cereals are edible seeds, which are often referred as grains and are belonging to grass family Gramineae. Cereals shown in Fig. 2. are staple food that provide nutrients like starch, proteins, vitamins and minerals. Wheat, barley, maize, rice, oat etc. are some of the major cereals.

Rice is the major staple food consumed in major

Fig. 2 Cereals and Pseudocereals: The given figure represents different cereals and Pseudocereals (**A**) Rice; (**B**) Wheat; (**C**) Amaranth; (**D**) Buckwheat; (**E**) Maize.

part of the world and especially in Asian countries. In market rice is available in 2 forms: brown rice and white rice. Various storage proteins are present in rice like albumin, globulin, gluteins and prolamins. Brown rice is comparatively more nutritious than white rice as it contains higher content of cereals, proteins minerals and vitamins. Aleuron layer and germ gets removed from brown rice due to polishing and provide us with white rice which contains low proteins, vitamins, minerals and large amount of fibre as compared with brown rice. The amino acid composition of prolamins include alanine, glutamic acid, arginine, hydrophobic amino acids like glutamine, valine and leucine but low sulphur containing amino acids and also deficit in lysine. Both gluteins and globulins are rich in sulphur containing amino acids with disulphide cross-linkage (22).

Another important cereal is wheat which is consumed in the form of chapati, bread, noodles, pasta etc (23). Gluten is the main storage protein present in wheat other proteins present are albumin, gliadins, globulin. Lysine is present in lower content but other sulphur containing amino acids are leucine, valine and isoleucine. L- glutamine is the readily available essential amino acid, which helps in immunity boost up and are considered important for athletes (24).

Pseudo cereals are gluten free and are recommended for patients with celiac disease and are used as food for infants. Pseudo cereals includes amaranth, buckwheat, quinoa. Pseudo cereals are rich in their protein content with high essential amino acids (25). Amaranth has the highest source of protein among all pseudo cereals i.e. approximately 13 to 16% whereas buckwheat contains 11 to 19 % and quinoa 12 to 14 %. Amaranth is rich in essential amino acids like methionine, arginine, cysteine, lysine, tryptophan as compared with cereals (26). Quinoa contain high amount of methionine, cysteine and lysine and are deficit in aromatic amino acids like tyrosine and phenylalanine. Buckwheat are nutritionally superior than cereals. It contains cereals like glutamic acid, arginine, lysine and aspartic acid but are limiting in amino acids like threonine, methionine, cysteine, phenylalanine and tyrosine (27). Buckwheat is unsuitable as staple food due to its low nutritional value but it can be have in combination with other sources to compensate the amino acids content (25).

Seeds and nuts as protein source

Peanuts

Peanuts are considered as highest source of plant based proteins. They also contain other nutrients like carbohydrates, fats, vitamins and minerals. Carbohydrates with high dietary fiber and monounsaturated and polyunsaturated fatty acids like omega fatty acids are present. Though peanuts contain all the major essential amino acids the other amino acids can get supplemented by including other plant based diets (28).

Flax seeds

Flax seeds are rich in proteins and oil content which consists approximately 73% of PUFA and 9% of saturated fatty acids. Flax seed protein includes amino acids like aspartate, glutamic acid, glutamine, asparagine whereas lysine, threonine and tyrosine are limiting amino acids of flax seeds (29).

Chia seeds

Chia seeds include approximately 25 to 41% carbs, 14 to 25% of protein, and 25 to 40% of fat. Chia seeds includes globulin as higher protein fraction and also contain aromatic and sulfur containing amino acids. Amino acids composition of chia protein includes aspartic acid, glutamic acid, threonine, histidine, leucine, isoleucine, lysine, valine, methionine, tryptophan, cysteine and phenylalanine. Amino acids present in chia seeds plays an important role in metabolic pathways i.e. plays an important role in hormonal regulation, immunity boost up, sulfur containing amino acids helps in maintaining structure of proteins, protection from cardiovascular disease (30).

Canola seeds

Canola seed occupies second rank in oil extraction after soybean. They are also rich in protein and provide a beneficial food source due to high levels of essential amino acids and sulfur based amino acids. All the essential amino acids are present but due to various processing method during oil extraction and increased temperature lysine contents gets reduced.

Various other seed and nuts rich in proteins include sunflower seeds, pumpkin seeds, cashews, almonds etc. Fig. 3. represents some of the seeds and nuts. Researchers are still looking for various plant-based protein sources

Fig. 3. Seeds and Nuts: The following are the seeds and nuts as source of plant based proteins (**A**) Peanuts; (**B**) Flax seeds; (**C**) Chia seeds; (**D**) Sunflower seeds; (**E**) Pumpkin seeds; (**F**) Almonds.

and their composition to meet the demand of growing world population. Processing methods performed to obtain plant based products also affects the quality of protein. Therefore, though plant protein contains majority of amino acids they are still deficient in one or more amino acid so it is recommended to include multiple plant based diets (31).

Protein alternatives as supplements

Animal based food products are considered to be the conventional source of proteins. Meat makes up a considerable portion of diet over the globe. However various health concerns have led to replacement of these animal based proteins with plant based proteins. Table 3. Given below mentions various plant protein based alternative along with the advantage and disadvantage.

Table 3. List of plant based protein alternatives

Product	Plant based alternative	Advantages	Disadvantages	Reference
Cheese	Protein from peanut, soy, vegetable fat	Good potential to lower the lipid profile Low cost of production	Amount of nutritional content may be low	(32)
Egg	Proteins from flax seed, pea and sunflower seed	Functional properties (emulsification, foaming) similar to egg Low fat content	Contains anti nutritional substance	(33)
Milk	Soy, almonds, coconut	Efficient for digestive disorder (lactose intolerance), Ideal as vegan source of milk	Less palatable if not flavoured	(34)
Microalgae	*Chlorella* sp. *Spirulina* sp.	Requires less land High protein content	Microalgae produced through genetic modification can have regulatory issues.	(35)

Tremendous research has been carried out in order to obtain plant based products which is palatable nutritious and most importantly has texture identical to meat. Consumers demand products that are sustainable, palatable, safe, nutritious, available and affordable.

Isolation and purification of plant based proteins

In order to understand the functionality of protein it is important to know the amino acid sequence and structure of particular protein. Methods like nuclear magnetic resonance, X-ray crystallography and spectroscopy are widely used for analysis of 3D structure of proteins. Apart from knowing the structure of protein it is also important to know the amino acid sequence. As mass spectroscopy relies on the separation of molecules on the basis of mass to charge ratio, it is considered as an ideal method for, peptide and protein sequencing, conformation analysis of protein and dynamics (36). Apart from amino acid sequencing is selected afterward taking into account. After understanding the characteristic properties such as structure and amino acid sequence isolation and purification from desired source can be performed.

Depending upon the physicochemical properties proteins could be isolated, quantitated and purified by various methods. Chemical, physical, biological properties of sources and type of proteins are considered for suitable isolation methods (36). While using these techniques various parameters like temperature, pH and type of solvent need to be carefully controlled (37). Proteins after isolation must be purified in order to separate them from non-protein part. For better purification of protein various techniques are applied such as ultrafiltration, dialysis, micellar precipitation techniques and isoelectric precipitation. The techniques mentioned are generally used for extraction of protein however while extraction combination of protein and non-protein part is obtained. In order to purify the obtained protein various techniques are available some of which are mentioned below (38).

Ultrasound assisted extraction is one of the methods use. This process has presented a considerable effect on the rate of different method in the food industry (39). The advantage of ultrasound assisted extraction has been reported earlier wherein they have shown that UAE in combination with micelles is effective in increasing the yield of wheat germ protein. Along with the increase in extraction, the overall time required was also reduced (40). In UAE milled soybean slurry used was subjected to ultrasound using laboratory probe for various time interval 0, 0.5, 1, 5 and 15 min. From this it was found that 1 min treatment gave approximately 10% improved yield of proteins. Further studies done by using Confocal laser scanning microscopy revealed the effect of ultrasound which showed the presence of undisrupted intact cells. From this it was found that the improved extraction of soya proteins was due to improved solubility. Thus, ultrasound assisted extraction is considered to be a reliable method because it gave improved yield in short time interval with lower energy consumption (40).

Solvent extraction is mostly used in initial step for separation and extraction of proteins. Various solvents could be employed for this purpose. Alcohol extraction method used for extraction of zein from maize (41). On commercial scale zein is extracted from corn gluten meal in small amount. 70% ethanol is used for extraction of zein which was then diluted using 40% ethanol and subjected to centrifugation. About 70-80 % protein was extracted. It also allowed removal of other components prior to dilution of extract (42).

Peanuts are found to have a wide industrial application as they have a superior content of oil and protein. But separation and isolation of oil and protein could be a tedious process. In order to separate the protein from the peanut seed enzyme assisted protein extraction technique was can be used. Through the use of various proteases this method has proved to give a considerable increase in yield of protein (36). Apart from enzyme assisted method various studies show the use of electro activation method wherein proteins from canola were isolated. Higher protein extractability with improved quality of protein was found through this technique. It can be observed that use of these advance techniques is supposed to be very efficient for isolation of proteins from plant sources.

Composition of plant proteins

Proteins are classified into various system due to their diverseness. Some are classified based upon biological and chemical properties and some on the basis of technological applications. Osbrone classification system is the oldest classification system based on proteins solubility in various solutions. Osbrone on the basis of solubility classified plant protein into four major fractions; albumin, prolamins, globulin, glutelins mentioned in Table 4. (43). Albumins are soluble in water and diluted salt solutions whereas globulins are soluble in salt solutions but insoluble in water. Prolamins are soluble in water alcohol mixture but insoluble if only water or alcohol is present and glutelins are insoluble in salt, alcohol or neutral water solutions but soluble in dilute acids or alkali solutions.

Table 4. Composition of plant proteins: major and minor fractions of plant proteins present in various sources

Components	2S Albumins	7-8S Globulins	11-12S Globulins	Prolamins	Glutelins
Major components	Legumes Cruciferae Compositae Castor beans Cotton seed Brazil nut	Legumes Cotton seed Palms cocoa	Legumes Compositae Cucurbitaceae Oats and rice Cruciferae Cannabis Brazil nut	cereals	wheat
Minor components		Cereals	French bean	Oats and rice	

Albumins

According to one report (44), albumins are storage proteins majorly present in legumes, oilseeds, and pulses rather than cereals. On ultra-centrifugation of various plant sources 3 fractions of albumin were isolated; 2S and 11S mainly present in all types of plant sources and 7S expect few sources present in all. Out of 3 fractions 2S is the major storage albumin protein in many sources. This globular small 2S albumin proteins are high in sulphur containing amino acids in various plant sources like legumes, lupines and peas though they occupy only 10 to 30% of total proteins.

Globulins

The major globulin content is present in legumes. They are present in both dicot, monocot and are also present in gymnosperms. As compared with albumin, globulins are deficient in sulphur based amino acids. The globulin fractions obtained after ultra-centrifugation is 7S and 11S. Some globulin proteins in various plant sources includes β-conglycinin (7S) and glycinin (11S) in soybean, β-conglutin (7S) and α- conglutin (11S) in lupins, leguminin (11S), convicilin (7S) and vicilin (7S) in peas, helianthnin (11S) in sunflower seeds, cucurbitin (11S) in *Cucurbita* spp. etc. (37).

Prolamins

Cereals are rich in prolamins except oats and rice (44). Prolamins were the first proteins studied scientifically and they were named gliadin but Osbrone proposed a new name i.e., prolamins as they yield high levels of proline and amides on hydrolysis. It also yields small amounts of arginine and histidine but lysine content is very little or totally absent. According to Osbrone there are four principle prolamins hordeins in barley, gliadins in wheat and rye, karfirins in sorghum and zein in maize. Prolamins present in oats are avenins.

Prolamins are grouped into four cereals categories. Triticeae which includes wheat, barley and rye and are further divided into three categories sulphur rich prolamins (S-rich) mainly includes amino acids proline, glutamine and sulphur containing amino acid cysteine with inter and intrachain disulphide bonds, High molecular weight prolamins (HMW) are rich in glycine and glutamine and sequence similar to S-rich prolamins and Sulphur poor amino acids. Prolamins in oats include avenins which occupies 10% of the seed protein; Prolamins in rice are divided into four class out of which class 1 to 3 are major prolamins and class 4 is minor prolamins but rich in sulphur amino acids methionine and cysteine (44). Prolamins in maize are zein which includes various fractions α, β, γ and δ. Sorghum includes karfirins are with disulfide bond with high cysteine residues which are further divided into subclasses α, β, γ and δ.

Glutelins

Glutens are mixture of two proteins i.e., prolamins and glutelins present in various grass varieties. Wheat contains gluten which are further categorized into two groups prolamins are called gliadin and glutelins are called glutenin. Gliadin are monomeric proteins and glutenins are polymeric proteins. Glutenins are with intrachain disulfide bonds present between two subunits i.e., HMW and LMW (i.e., high molecular weight and low molecular weight subunits) (44).

Application of plant based proteins

Plant based protein could be beneficial means as a protein source due to their composition, availability and other techniques available for isolating them. The availability of plants as source of proteins has now led to their use in various field. The application of plant-based protein is depicted in Fig. 4. Given below.

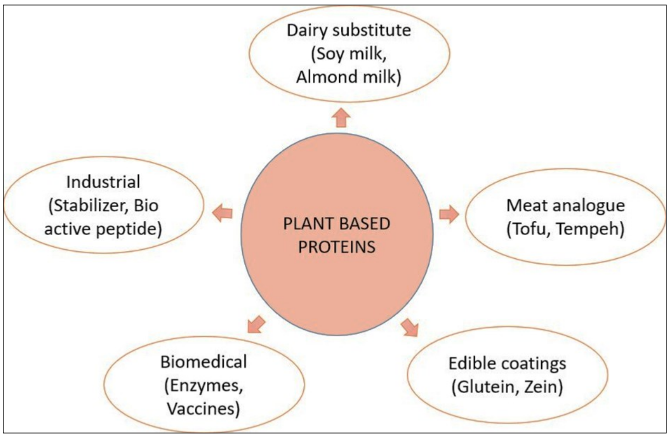

Fig. 4. Application of plant based proteins: Plant based protein could be beneficial means asa protein source.

Application of plant based protein in biomedical industry

Emergence of plant-based proteins has led to their commercialization and its production at industrial scale. The emergence of various molecular biology techniques has proved very useful in production of recombination proteins using plants. Monoclonal antibodies is one such products. As per one study, it has shown that the use of monoclonal antibodies produced from plants which has their use as immunotherapeutic agent for cancer (45). The production of monoclonal antibodies for various viral diseases such as rabies is possible using plant systems. Apart from monoclonal antibodies various other pharmaceutical products such as enzymes, hormones, vaccines etc. could be produced and considered to be of prime importance (46).

Neem leaf glycoprotein (NLGP) is an active ingredient present in neem leaf extract. NLGP enhance immune system in tumor bearing host by increasing amount of various immune cells like T helper cells, cytotoxic T cell, macrophage, monocyte, dendritic cell etc. (47). In cancer cells there are various suppressor like Tregs, myeloid derived suppressor cells (MDSCs) and tumor associated macrophages (TAMs) impairs the immune functions. Function of such suppressor cells is amplified in cancer cells (48). NLGP upregulates the immune system by downregulating the suppressor cells.

HIF1α regulates vascular endothelial growth factor (VEGF). VEGF is important for angiogenesis in cancer as it fulfills the nutrient and oxygen requirement for the cancer development and growth. NLGP reduces VEGF secretion by preventing the binding of HIF1α to VEGF. NLGP shows antimetastaic role by inhibiting VEGF by reducing angiogenesis (49).

Plant based protein as edible coatings

Plant based proteins are also used as edible films. Environmental concerns have led to replacement of coating material like plastic with edible plant protein based film. Plant protein based films provided an option of using a renewable resource. Various cereals, pulses could be used for production of edible films example soya, wheat etc. Gluten protein present in wheat is found to have high elastic properties which is used as a film or coating (50). Apart from gluten various other protein sources like zein, rice bran protein etc can also be used. Zein has proved to show excellent result as edible film due to its hydrophobic nature and higher amount of disulfate and hydrogen linkage.

Application of plant based protein as dairy substitute

Increased population and demand of food has led to consumption of plants as protein source which has various health effects (51). Plant based proteins could be used as an alternate source in disease and disorders Lactose intolerance is one of the increasing disorders which involves inability of an individual to digest milk due to lack of enzymes lactase (52) . Plant sources are thus used to obtain analogues of milk known as plant based non-dairy milk. Also, the milk obtained from plant source could be subjected to fortification with vitamins, minerals etc. Apart from lactose intolerance various diseases like chronic kidney disease could be cured by addition of plant-based proteins in diet (53). Including plant protein in various diet is nutritionally adequate and have pleiotropic effects which may be useful for chronic kidney disease patient (54).

Plant based protein as meat analogue

Plant-based meat alternatives (PBMA) have been used as a substitute to conventional animal based meat product. These meat analogues are derived through processing of different legumes, vegetable and fruits, examples of different plant based meat analogues are mentioned in the Fig. 4. (55). Vegetarianism and Veganism are the phenomenon which though sound similar but has a minor difference. Vegetarianisms is exclusion of animal meat (56). Whereas Veganism is referred to as complete exclusion of animal meat as well animal based products like milk. With the exclusion of animal and other non veg source of protein plant based protein is considered to be the most appropriate source of protein. A vegan diet is considered to have various health related benefits which is one of the reasons in global increase in trend of vegan diets.

Other industrial application of plant based proteins

The advances in food technology and search for renewable source of food material have led to the development of alteranative sources. Bioactive peptides are organic substances formed by amino acids joined by covalent bonds also known as amide or peptide bond. Studies perfomed by Samaei et al., depict the use of faba bean Faba bean based bioactive peptides in obtaining fortified fruit juice and other functional food. Apart from BAP plant based protein also as has good potential as stabilizers (57). Peanut protein Pea protein concentrate and soy protein isolate have shown resistance to lipid oxidation and reduced droplet flocculation.

Conclusion

Out of the four macromolecules protein is one of the important macromolecules which is building block of human body. The availability of protein is distributed in various sources out of which plant-based protein is one such source which has increase in demand. The review gives information about various sources of plant-based proteins with its composition. From the information mentioned about the composition of plant-based proteins it is clear that to get all the essential amino acids one protein source needs to be complimented with other. This is because one of the drawback of plant based protein over animal based protein source. The techniques used for purification discussed above are most widely used for extraction, however in order to improve the functionality of extracted protein advanced techniques are to be considered to be isolated. Some advance isolation techniques like ultrasound assisted extraction, enzyme assisted method and electro activation method provides efficient isolation of desired proteins however, these advanced techniques could be expensive. Thus, advance techniques which are more cost effective must be employed. The application of plant-based proteins in various aspects state the abundance availability of plant-based proteins. The usage of plant-based protein in various form should thus be considered as a replacement for animal and other sources which would help in overcoming the increase in demand of the protein in nutritional and other aspects. Thus, more research on plant based proteins need to be conducted.

Acknowledgements

The study was funded by the Research Society for the Study of Diabetes in India (RSSDI/HQ/Grants/2017/342).

Authors contributions

KM and SDS searched, collected and wrote the first draft. HA, and NP reviewed and edited the manuscript. MT and DJ critically reviewed and edited the same. AA analysed the manuscript, provided the regular assistance to revise and finalize it. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None.

References

1. Alcorta A, Porta A, Tárrega A, Alvarez MD, Vaquero MP. Foods for plant-based diets: Challenges and innovations. Foods. 2021 Feb 1;10 (2):293. https://doi.org/10.3390/foods10020293
2. Hoffman JR, Falvo MJ. Protein - Which is Best? J Sport Sci Med. 2004;3 (3):118-30.
3. Almeida AG, Franco Moreno YM, Mattar Carciofi BA. Plant proteins as high-quality nutritional source for human diet. Trends Food Sci Technol, 2020; 97:170-84. https://doi.org/10.1016/j.tifs.2020.01.011
4. Hertzler SR, Lieblein-Boff JC, Weiler M, Allgeier C. Plant proteins: Assessing their nutritional quality and effects on health and physical function. Nutrients. 2020; 12(12): 3704. https://doi.org/10.3390/nu12123704
5. Langyan S, Yadava P, Khan FN, Dar ZA, Singh R, Kumar A. Sustaining protein nutrition through plant-based foods. Frontiers in Nutrition. 2021;8. https://doi.org/10.3389/fnut.2021.772573
6. Fathi M, Mohebbi M, Razavi SM. Application of image analysis and artificial neural network to predict mass transfer kinetics and color changes of osmotically dehydrated kiwifruit. Food and Bioprocess Technology. 2011 Nov;4(8):1357-66. https://doi.org/10.1007/s11947-009-0222-y
7. Sabaté J, Soret S. Sustainability of plant-based diets: back to the future. The American journal of clinical nutrition. 2014 Jul 1;100 (suppl_1):476S-82S. https://doi.org/10.3945/ajcn.113.071522
8. Pojić M, Mišan A, Tiwari B. Eco-innovative technologies for extraction of proteins for human consumption from renewable protein sources of plant origin. Trends in Food Science and Technology. 2018 May 1;75:93-104. https://doi.org/10.1016/j.tifs.2018.03.010
9. Allison JB. Biological Evaluation of Proteins. Adv Protein Chem.1949; 5:155-200. https://doi.org/10.1016/S0271-5317(99)00117-7
10. Moore DR, Soeters PB. The biological value of protein. Nestle Nutr Inst Workshop Ser. 2015; 82:39-51. https://doi.org/10.1159/000382000
11. Friedman M. Nutritional value of proteins from different food sources. A review. J Agric Food Chem.1996;44(1):6-29. https://doi.org/10.1021/jf9400167
12. Brody T. Protein. Nutritional Biochemistry 2nd ed. University of California, Berkely, Academic Press.1998; 421-87. https://doi.org/10.1016/B978-012134836-6/50011-1
13. Abeyrathne ED, Lee HY, Ahn DU. Egg white proteins and their potential use in food processing or as nutraceutical and pharmaceutical agents-A review. Poult Sci. 2013; 92(12):3292-99. https://doi.org/10.3382/ps.2013-03391
14. Bar C, Mathis D, Neuhaus P, Dürr D, Bisig W, Egger L, Portmann R. Protein profile of dairy products: Simultaneous quantification of twenty bovine milk proteins. Int Dairy J. 2019; 97:167-75. https://doi.org/10.1016/j.idairyj.2019.01.001
15. Picota L, Bordenavea S, Didelota S, Fruitier-Arnaudina I, Sanniera F, Thorkelssonb G et al. Antiproliferative activity of fish protein hydrolysis on human breast cancer cell lines. Proc Bio. 2006; 41(5):1217-22. https://doi.org/10.1016/j.procbio.2005.11.024
16. Maurya PK, Malik DS, Yadav KK, Kumar A, Kumar S, Kamyab H. Bioaccumulation and potential sources of heavy metal contamination in fish species in river Ganga basin: Possible human health risks evaluation. Toxicol Rep. 2019;6:472-81. https://doi.org/10.1016/j.toxrep.2019.05.012
17. Jabborova DP, Enakiev YI, Davranov KD, Begmatov SA. Effect of co-inoculation with *Bradyrhizobium japonicum* and *Pseudomonas putida* on root morph-architecture traits, nodulation and growth of soybean in response to phosphorus supply under hydroponic conditions. Bulg J Agric Sci. 2018;1;24(6):1004-11.
18. Jabborova D, Wirth S, Kannepalli A, Narimanov A, Desouky S, Davranov K et al. Co-inoculation of rhizobacteria and biochar application improves growth and nutrientsin soybean and enriches soil nutrients and enzymes. Agronomy. 2020;10(8):1142. https://doi.org/10.3390/agronomy10081142
19. Singh U, Singh B. Tropical grain legumes as important human foods. Econ Bot. 1992; 46(3):310-21. https://doi.org/10.1007/BF02866630
20. Thrane M, Paulsen PV, Orcutt MW, Krieger TM. Soy Protein. Nadathur S et al. Sustainable Protein Sources 1st ed. Academic press. 2017;23-45. https://doi.org/10.1016/B978-0-12-802778-3.00002-0
21. Samaranayaka A. Lentil: Revival of poor man's meat. In: Nadathur S, Wanasundara JPD, Scanlin L. Sustainable protein sources 1st ed. Academic press. 2017;185-96. https://doi.org/10.1016/B978-0-12-802778-3.00011-1
22. Hoogenkamp H, Kumagai H, Wanasundara, JPD. Rice protein and rice protein products. In: Nadathur S, Wanasundara JPD, Scanlin L.

Sustainable protein sources 1st ed. Academic press. 2017; 47-65. https://doi.org/10.1016/B978-0-12-802778-3.00003-2

23. Jabborova D, Narimanov A, Enakiev Y, Kakhramon D. Effect of *Bacillus subtilis* 1 strain on the growth and development of wheat (*Triticum aestivum* L.) under saline condition. Bulg J Agric Sci. 2020;26(4):744-47.

24. Flambeau M, Redl A, Respondek F. Proteins from wheat. In: Nadathur S, Wanasundara JPD, Scanlin L. Sustainable Protein Sources 1st ed. Academic press. 2017;67-78. https://doi.org/10.1016/B978-0-12-802778-3.00004-4

25. Schoenlechner R, Siebenhandl S, Berghofer E. Pseudocereals. Gluten-free cereal products and beverages. 2008;149–*VI*. https://doi.org/10.1016/B978-012373739-7.50009-5

26. Orona-Tamayo D, Paredes-López O. Amaranth Part 1—Sustainable Crop for the 21st Century. In: Nadathur S, Wanasundara JPD, Scanlin L. Sustainable Protein Sources 1st ed. Academic press. 2017; 239-56. https://doi.org/10.1016/B978-0-12-802778-3.00015-9

27. Scanlin L, Lewis KA. Quinoa as a Sustainable Protein Source. Nadathur S, Wanasundara JPD, Scanlin L. Sustainable Protein Sources 1st ed. Academic press. 2017;223-38. https://doi.org/10.1016/B978-0-12-802778-3.00014-7

28. Arya SS, Salve AR, Chauhan S. Peanuts as functional food: A review. J Food Sci Technol. 2016; 53(1):31-41. https://doi.org/10.1007/s13197-015-2007-9

29. Marambe PWMLHK, Shand PJ, Wanasundara, JPD. An *in-vitro* investigation of selected biological activities of hydrolysed flaxseed *(Linum usitatissimum* L.) proteins. J Am Oil Chem Soc. 2008;85(12):1155-64. https://doi.org/10.1016/j.plaphy.2014.04.020

30. Orona-Tamayo D, Valverde ME, Paredes-López O. Chia-The New Golden Seed for the 21st Century. In: Nadathur S, Wanasundara JPD, Scanlin L. Sustainable Protein Sources 1st ed. Academic Press. 2017;265-81. https://doi.org/10.1016/B978-0-12-802778-3.00017-2

31. Wanasundara JPD, McIntosh TC, Perera SP, Withana-Gamage TS, Mitra P. Canola/rapeseed protein-functionality and nutrition. OCL. 2016; 23(4):D407. https://doi.org/10.1051/ocl/2016028

32. Bachmann, HP. Cheese Analogues: A review. Int Dairy J. 2001;11:505-15. (Cross Ref). https://doi.org/10.1016/S0958-6946(01)00073-5

33. Söderberg J. Functional properties of legume proteins compared to egg proteins and their potential as egg replacers in vegan food.

34. Sethi S, Tyagi SK, Anurag RK. Plant-based milk alternatives an emerging segment of functional beverages: A review. Journal of Food Science and Technology. 2016 Sep;53(9):3408-23. https://doi.org/10.1007/s13197-016-2328-3

35. Koyande AK, Chew KW, Rambabu K, Tao Y, Chu DT, Show PL. Microalgae: A potential alternative to health supplementation for humans. Food Science and Human Wellness. 2019 Mar 1;8(1):16-24. https://doi.org/10.1016/j.fshw.2019.03.001

36. Zhu Y, Fang Q. Analytical detection techniques for droplet microfluidics. Anal Chim Acta. 2013;787:24-35. https://doi.org/10.1016/j.aca.2013.04.064

37. Hadnadev M, Hadnadev TD, Pojic M, Saric B, Misan A, Jovanov T, Sakac M. Progress in vegetable proteins isolation techniques: A review. Food Res. 2017; 44(1):11-21. https://doi.org/10.5937/FFR1701011H

38. Moure A, Sineiro J, Domínguez H, Parajó JC. Functionality of oilseed protein products: a review. Food Res Int. 2006; 39(9):945-63. https://doi.org/10.1016/j.foodres.2006.07.002

39. Gendag E, Gorguc, A, Yilmaz FM. Recent advances in the recovery techniques in plant based proteins from agro industrial by products. Food Rev Int. 2020;1-22. https://doi.org/10.1080/87559129.2019.1709203

40. Chemat F, Huma Z, Khan, M. Applications of ultrasound in food technology: Processing, preservation and extraction. Ultrason Sonochem. 2011;18(2011):813-35. https://doi.org/10.1016/j.ultsonch.2010.11.023

41. Preece K, Hooshyar N, Krijgsman A, Frye P, Zuidam N. Intensified soy protein extraction by ultrasound. Chem Eng Process. 2016;113:94-101. https://doi.org/10.1016/j.cep.2016.09.003

42. Dickey L, Parris N, Craig JC, Kurantz MJ. Ethanolic extraction of zein from maize. Ind Crops Prod. 2000;13(2001):67-76. https://doi.org/10.1016/S0926-6690(00)00054-6

43. Takiguchi Y, Osada O, Uematsu M. Thermodynamic properties of {xC2H5OH+(1−x)H2O} in the temperature range from 320 K to 420 K at pressures up to 200 MPa. J Chem Thermodyn. 1996; 28(12):1375-85. https://doi.org/10.1006/jcht.1996.0120

44. Shewry P, Casey R. Seed Proteins. 1st ed. Springer publishing House. 1999;1-11. https://doi.org/10.1007/978-94-011-4431-5_1

45. Nessa M, Rahman M, Kabir, Y. Plant-produced monoclonal antibody as immunotherapy for cancer. Biomed Res Int. 2020;3038564. https://doi.org/10.1155/2020/3038564

46. Wadhave A, Jadhav A, Arsul, V. Plant proteins applications: A review. World J Pharm Sci. 2014;3(3):702-12.

47. Hao F, Kumar S, Yadav N, Chandra D. Neem components as potential agents for cancer prevention and treatment. Biochimica et Biophysica Acta (BBA)-Reviews on Cancer. 2014 Aug 1;1846(1):247-57. https://doi.org/10.1016/j.bbcan.2014.07.002

48. Bose A, Baral R. Neem leaf glycoprotein in cancer immunomodulation and immunotherapy. In: New Look to Phytomedicine. Academic Press. 2019 Jan 1;391-408. https://doi.org/10.1016/B978-0-12-814619-4.00016-1

49. Saha A, Nandi P, Dasgupta S, Bhuniya A, Ganguly N, Ghosh T et al. Neem leaf glycoprotein restrains VEGF production by direct modulation of HIF1α-linked upstream and downstream cascades. Frontiers in Oncology. 2020 Mar 6;10:260. https://doi.org/10.3389/fonc.2020.00260

50. Dangaran K, Tomasula PM, Qi P. Structure and function of protein -based edible films and coatings. In: Huber K, Embuscado M. (Editors) Edible Films and Coatings for Food Applications. Springer, New York, NY. 2009. https://doi.org/10.1007/978-0-387-92824-1_2

51. Mariotti F, Gardner C. Dietary protein and amino acids in vegetarian diets-A review. Nutrient. 2019;11(11):2661. https://doi.org/10.3390/nu11112661

52. Vanga SK, Raghavan V. How well do plant based alternatives fare nutritionally compared to cow's milk?. J Food Sci Technol. 2018;55(1):10-20. https://doi.org/10.1007/s13197-017-2915-y

53. Grant CA, Hicks AL. Comparative life cycle assessment of milk and plant-based alternatives. Environ Eng Sci. 2018;35(11):1235-47. https://doi.org/10.1089/ees.2018.0233

54. Paul AA, Kumar S, Kumar V, Sharma R. Milk analog: Plant based alternatives to conventional milk, production, potential and health concerns. Crit Rev Food Sci Nutr. 2019; 1-19. https://doi.org/10.1080/10408398.2019.1674243

55. Joshi VK, Kumar S. Meat analogues: Plant based alternatives to meat products-A review. International Journal of Food and Fermentation Technology. 2015;5(2):107-19. https://doi.org/10.5958/2277-9396.2016.00001.5

56. Dagnelie PC, Mariotti F. Vegetarian diets. Vegetarian and plant-based diets in health and disease prevention. 2017;3-10. https://doi.org/10.1016/B978-0-12-803968-7.00001-0

57. Samaei SP, Ghorbani M, Tagliazucchi D, Martini S, Gotti R, Themelis T et al. Functional, nutritional, antioxidant, sensory properties and comparative peptidomic profile of faba bean (*Vicia faba* L.) seed protein hydrolysates and fortified apple juice. Food Chemistry. 2020 Nov 15;330:127120. https://doi.org/10.1016/j.foodchem.2020.127120

§§§