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DIETARY SUPPLEMENTS AND FUNCTIONAL FOODS

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Review Based Book Chapter

PLANT BASED PROTEINS: OPTIONS, QUALITY AND EXTRACTION METHODS

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REVIEW BASED BOOK CHAPTER

PLANT BASED PROTEINS: OPTIONS, QUALITY AND EXTRACTION METHODS

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Abstract

Plant-based proteins are versatile, sustainable, and increasingly capable of meeting nutritional needs through careful selection and processing. As interest in sustainable and health-conscious diets continues to grow, plant-based proteins have become increasingly important in nutrition, food science, and agriculture. These proteins offer an alternative to animal-derived sources and are derived from a wide range of plants, each with unique benefits and limitations. Plant-based proteins are found in a variety of foods. Legumes, such as lentils, chickpeas, soybeans, and peas, are among the richest sources. Soy and pea proteins are particularly popular due to their relatively high protein content and favorable amino acid profiles. Whole grains like quinoa, brown rice, and oats also contribute valuable proteins; quinoa is notable as a complete protein, containing all nine essential amino acids. Nuts and seeds—including almonds, chia seeds, and pumpkin seeds—offer protein along with healthy fats. Additionally, some vegetables like spinach and broccoli contain small amounts of protein. With advancements in extraction technologies and a better understanding of protein blending, plant-based diets can now offer protein quality comparable to animal sources. As the demand for ethical and eco-friendly food options grows, the role of plant proteins will continue to expand in both consumer and industrial markets. Understanding the sources, quality, and extraction methods of plant-based proteins is crucial for optimizing their use in both human nutrition and food manufacturing.

Keywords

Plant Proteins, Proteins Sources, Protein Quality, Extraction Methods, Nutrition

1. Introduction

Global population is increasing rapidly and about to exceed 9 billion till 2050 [1]. Ensuring food security for such huge population is a global concern and it demands additional sources of nutrition particularly protein [2]. To meet the global dietary need

plant-based protein sources such as legumes, grains, Nuts etc. are gaining popularity, as they provide a higher percentage of protein (30-60%) with less environmental and sustainability concerns [3]. Plant based proteins are widely accepted as an alternative to animal protein due to their nutritional profile [4], therefore, plant-based protein industry is flourishing and expected to reach 27 billion dollars by 2030. As animal-based diets are considered a potential risk to environment and livestock, disturbing the biodiversity. So it is very important to shift towards plant based diet to protect the climate, animal welfare and meet the dietary needs of people as awareness is increased among public, food intake pattern are also influenced now people prefer sustainable and ethical food sources [5] especially, animal welfare has become a very sensitive topic and people consider themselves responsible for animal protection, so choice between animal and plant based food is influenced by religious beliefs, ethical concerns and cultural values, so plant-based proteins are reducing reliance on animal protein [6]. Now modern technology is employed to improve quality and function of plant protein to address the imposed challenges [7].

Plant proteins contain a very rich nutritional profile and recognized globally for their functional attributes. Along with protein they also supply fibers, vitamins and minerals which prevent chronic health issues [8]. Functional properties such as emulsification, solubility, gelling etc. are important characteristics of plant-based protein which influence their use in food processing [9]. This review explains the various sources and extraction methods that increase the functionality of plant proteins. Different techniques are used to isolate protein from plant sources, popular ones are enzyme assisted and microwave assisted techniques. These extraction methods affect the quality and quantity of protein [10]. It is believed that functional properties of plant-based proteins are less qualitative than animal-based proteins but many advance techniques have been discovered like enzymatic and chemical methods which enhance these characteristics [11]. Along with fulfillment of these dietary requirements, plant-based proteins are also considered sustainable alternative [12] as plant-based sources require less water and emit less green-house gases and put less harmful impact on the environment, where this industry gained a lot of importance, it still faces many

challenges like allergy to sources, less isolation techniques and less functional properties [13], so it is necessary to develop new techniques for isolation of protein and different processing methods which increase nutritional and functional attributes and also sync with environment [14]. Another main issue is less cultivation of plant sources like legumes; nuts etc. also only few plant crops are grown and there is a big room for diverse crops. For long term use of plants as protein sources, it is necessary to increase the production [15].

2. Types of Plant-based Protein

Plant based protein are categorized into: legumes (20-38 %), seeds (9-30%), cereals (6-15%), pseudo cereal (11-23%) and Nuts (18-38%) [16].

2.1. Legumes

Legumes include lentils, chickpeas, soybeans and peas. Legumes are known as a balanced food as they have high amino acid profile, minerals and vitamins [17]. Legumes are green and eco-friendly plant-based protein [18]. Legumes are believed to have high profile amino acids which make them a suitable alternative to animal proteins [19]. Legumes production is increasing due to its health promoting effects and environment friendly nature. They serve as good nutraceutical ingredient [20]. They prevent many chronic diseases like diabetes and cardiovascular diseases. Their eco-friendly nature made them useful for both consumer and cultivators [21]. Following are few most common legumes:

2.1.1. Soybean

Soybean (*Glycine max*) is a high protein containing legume which contain almost 35-50% protein [22]. It is used in many food products as an analogue to meat as it is rich in essential amino acids except sulfur containing Amino acids. Soybean is valued for its functional attributes which leads to disease prevention. Glycinin and B-conglycinine is the main storage protein of soybeans. Soybeans are rich in flavones and phytosterols which increase their nutritional and functional attributes and as a superior source of proteins [23]. Soybeans are used to mimic real meat due to its textural properties by using different techniques like moisture extrusion [24].

2.1.2. Lentils

Lentils (*Lens culinaris*) provide 25% protein and micronutrients like iron, zinc and folate [25]. Lentils are rich in functional properties like solubility and gelling characteristics which makes them useful for many food products [26]. They are also very eco-friendly and require less irrigation water and land than other protein sources. Lentils also show antioxidant characteristics due to presence of bioactive compounds like flavonoids and phenolics [27]. To compensate animal's protein, lentils are often combined with other legumes to serve as a complete amino acid profile. Lentils are considered affordable source of protein and used to treat malnutrition in under developed and developing countries [28].

2.1.3. Chick Peas

Chick peas (*Cicer arietenum*) are high protein source providing 18-26% proteins. Chick peas are highly nutritious plant which is rich in essential amino acids like lysine and with DIAAS (digestible indispensable Amino acids score) [29]. Chick peas are used in variety of foods and it is available in different forms like flour, concentrate etc. [30] essential amino acids like methionine and tryptophan's presence make them more qualitative. Quality of protein depends upon genes of chickpeas. Many bioactive peptides are also present which make them more beneficial and prevent diabetes by controlling glucose and insulin levels [31]. Hypoglycemic affect and low glycemic index are its well-known characteristics [32].

2.1.4. Peas

Peas (*Pisum sativum*) are a very good source of plant-based protein. They provide 24% protein and also rich in fiber and starch. The use of pea protein in food products not only satisfies consumer but also enhance economical-stability of farmers. R research reveals that functional properties of peas are limited; so many biological, physical and chemical methods are used to enhance its Functional properties [33]. Pea lacks sulfur containing Amino acids but consist of essential amino acids like threonine and lysine [34]. Pea contains probiotics, phytochemicals and fatty acids which make it valuable in food industry [35].

2.2. Cereals

Cereals are recognized for providing variety of nutrients including few essential amino acids [36]. Cereals are staple food in many parts of the world and eaten as a primary source of protein although animal proteins are more preferred but cereal gained a valuable position because of high nutritional content and availability of protein. Cereal possesses a limited biological activity because of absence of essential amino acids like Lysine which is why animal proteins are preferred over them, but their exceptional functional and nutritional qualities make them useful in every aspect, they are widely used in food industry, pharmaceutical industry and livestock [37]. Unfortunately, cereal grains are widely used in animal feed and are not consumed as a vital protein source by human. Currently 35% grains are used as animal feed while 41% is consumed by human. In order to ensure protein security in mankind, this trend should be changed [38]. Cereals are used as a staple food in many counties and they are largest grown crop worldwide so if cereals are used properly as a food, they are able to tackle food security issues and can satisfy this huge population's dietary requirement. Cereals are categorized as primary source of protein, they supply 10-15% protein which is obviously less than legumes but if used wisely in combination with other sources, they can be a good source of reliable calories [39]. Cereals possess a range of functional attributes which are highly liked by food industry because elasticity, solubility or moisture retention are the crucial Factor that affect the product manufacturing in an industry especially in baking industry [40]. It is believed that cereal protein has a bitter taste; so many pre-treatments are done to produce high quality product with a consumer accepted taste, however these proteins valued because of their antioxidant, antidiabetic and antihypertensive effects. Cereal proteins are classified into four classes based on storage characteristics that are Glutelin, albumin, globulin and prolamins. All of these offer different functional properties and nutritional qualities. Prolamins are alcohol soluble proteins and present as major storage Protein in all cereals but in Rice and Oat, Glutelin serve as major protein. Among all the cereals wheat has highest amount of amino acids while barley has lowest Amino acids ratio [41]. Oat is one of the most nutritious Cereals as it provides highest protein percentage as compared to other cereals 13.22%. Along with lysine presence oat is famous for its high amino acid profile.

While, glutamic acid is the most abundant amino acids among cereals [42]. In wheat prolamins are responsible for elasticity and viscosity which contributes to desirable dough formation. Prolamins are known as Glutelin and gliadin in wheat [43]. While in maize prolamins are known as zein and in Rice they are known as orzein and in oat prolamins are known as avenins [44]. In cereals like rice, maize and wheat etc., prolamins are present in endosperm portion while Globulin in aleurone portion and this is considered the main protein of cereal [45]. Cereals are not considered complete food and may cause deficiencies in the people completely relying on them for their nutritional needs particularly protein as they are deficit in lysine and tryptophan which are essential amino acids this limits their use as staple food [46]. Many countries including India, Pakistan, and Bangladesh etc. are using cereal as their main course which is point of concern as it can lead to malnutrition and inadequate protein intake [47]. Another drawback of cereal protein is their complex structure and composition of amino acids which slowdown the digestion and provides fewer nutrients because of presence of anti-nutritional factor [48]. Researchers and agriculturists are working on these limitations and improving the genes of cereal plants to ensure supply of balanced amino acids so that using them as staple food cannot be a risk anymore [49].

2.3. Pseudocereals

Pseudocereal are the edible cereals which belong to class dicotyledon and are highly appreciated because of their diverse nutritional benefits [50]. Pseudocereal are also well known because of their gluten free nature and distinct characteristic from true cereals. Pseudo cereal includes quinoa, buckwheat and amaranth [51]. Pseudocereal not only possess variety of nutrients but also consist of amazing functional properties which produce antioxidant effects and prevent chronic health diseases and promote health by the effects of phytochemicals such as polyphenols and saponins [52]. These are also center of attention because of complete protein profile and providing all essential amino acids along with fulfillment of dietary requirements of vitamins and minerals. They are also preferred due to providing no harm to the individual suffering from celiac disease [51]. Celiac disease is a common disease induced due to intake or gluten protein present in cereal like wheat, barley, rice etc. [53]. Pseudocereal are widely used in food industry particularly in baking and dairy due to their exceptional

techno-functional attributes. The amino acid profile of cereal is the most likeable factor about them, they possess superior nutritional characteristics particularly abundance of essential amino acids. Amaranth possesses highest amino acid content followed by buckwheat and quinoa [54]. Pseudocereals offer highly therapeutic effects along with nutritional attributes because of their dive diverse constituents such as peptides which are very helpful in maintaining good health by their antihypertensive and antioxidant effects [55]. People who are actively seeking plant-based proteins can consider pseudocereal as their primary choice because of their superior amino acid profile and their Gluten free property. They are also favorable for agriculturists because they don't require any extra treatment for growth and even grow in harsh environmental conditions which are why they are known as sustainable alternative source of proteins [56]. Pseudocereal are enriched with essential amino acids like Quinoa is considered more nutritious than traditional cereal because it contains Arginine 6.8%, lysine 6.2%, isoleucine 6.8%, aspartic acid 6.8% and glutamic acid 11.4% [57]. Quinoa is preferred amongst the pseudocereal because of presence of methionine. Pseudocereal are mostly used in the form of flour blended other ingredients and are widely used in baking for production of gluten free products without compromising protein nutrition and because of their exceptional techno-functional attributes such as water absorption and oil retention which are crucial for product development [58], moreover use of pseudocereal in baked good especially gluten free products can improve their quality with enhanced nutrition and desirable flavor and texture. They are widely used in sour dough fermentation where they provide antioxidant effects with characteristic texture [59], despite all these extraordinary attributes, pseudo cereal is under grown and they make just 1% of total production of cereals [60]. Their use is limited because of their undesirable taste and presence of few antinutritional compounds but this problem can be overcome by processing and pretreatment which could be helpful in enhancing palatability [61], for example sprouting is a very common treatment done to pseudo cereal for enhancement of nutritional characteristics and lowering anti nutritional effects of them, therefore it is important to grow Pseudocereal and increase their use for protein intake, it is unfortunate that cereal are fortified with micronutrients like

vitamin and minerals but still they make 80% of food consumption but pseudo cereal are ignored and undergrown [62].

2.4. Seeds

Seeds include sunflower seeds, canola, flax seeds, chia seeds, sesame seed, pumpkin seeds, hemp seeds etc. As, the demand for protein sources is increasing day by day and people are seeking for alternative sources while researchers are also conducting researches to get a sustainable protein source which could be alternate the animal protein, Seeds are emerged as potential protein source with a reliable nutrition profile and minimum impact on environment [63]. Seeds like rapeseed and lupin provide a large amount of protein content ranging from 30-60% and a variety of products are made from these oil seeds which is an indication of their potential to meet the dietary requirements of individual on daily basis and serve as an alternative of meat protein [64]. Seeds are rich in essential amino acids and also provide bioactive compounds which promote health and wellness [65]. Different seeds provide different range of protein like sunflower seed provide 25% and their meal extraction give 30-50% protein [66], while Hempseed and chia seeds are also significant source of protein and lipid and provide omega-6 and omega-3 fatty acids respectively and contribute to diverse nutritional intake and health benefits [67], while almond provide almost 16.3 g/100 of protein and peanut provides 25.8 g/100 of protein [68]. Oilseeds are preferred in low income and developing countries because they are cheaper source of protein and provide most of the essential amino acids which are required in diet such as methionine, phenylalanine etc. but there is a limiting amino acid lysine whose deficiency makes seed an incomplete source of protein which can lead to many serious functional problems in the body [69], overall they are a very likeable and cheap alternative to animal protein but deficiency of lysine make them dependent on other sources to compensate this limitation. Usually, seeds are combined with other plant-based sources which are rich in lysine for example legumes or cereal which collectively make an excellent source of protein with higher DAA value and a variety of amino acids also including essential amino acids [70].

2.5. Nuts

Nuts include Walnut, hazelnut, peanut, almond, pine nut, cashew, pecan etc. Nuts are very important part of diet as they promise to deliver 18-38% protein content which makes them very suitable to use in place of meat protein [71]. Nuts are famous for their diverse nutritional value and are significantly recognized worldwide as an important source of protein particularly pecan and walnuts are considered very nutritious and eaten on daily basis to contribute in dietary requirements in many parts of the world l, pecan is rich in gluten which is why liked by baking industry and it boasts a 83% of protein approximately with rich amino acids profile [72], similarly walnuts protein is extracted from its kernels and oil pressing wastes and utilized in many foods and taken as a protein source. It is believed to have a more balanced nutritional profile and serve as a major ingredient in many foods due to its exceptional functional characteristics [73]. Earlier Nuts were utilized as it is but with development in technology many extraction techniques have been introduced and now protein is extracted from nuts and used in different food products which is Increasing their demand at industrial level for addressing growing dietary needs [74]. Nuts are not only enriched with essential amino acids but also liked for their health promoting functions. They consist of many bioactive compounds, vitamins, minerals, dietary fibers; mono and polyunsaturated fatty acids and phytochemicals, all of them contribute to their anti-inflammatory and antioxidant effects [75]. It is also believed about nuts that their regular intake might be helpful in weight management by regulating appetite and satiety [76]. Regular use of nuts prevents risk of many chronic diseases like cancer, cardiovascular diseases and diabetes especially Type -2 diabetes. Epidemiological studies have been revealed that intake of nuts on regular basis can lead to many important health benefits like better glucose metabolism, improved lipid profile and better vascular health, which results in overall health betterment [77]. The Amino acid profile of nuts is very diverse which contributes in their significance, chestnut contain 13 amino acids and threonine is abundant in chest nut (54.09%) while Walnut contain 8 amino acids and valine is most abundant (69.43%) [78], but limiting Amino acids are also observed in nuts which is why it is recommended to eat a good combination of nuts so that their deficiency can be compensated, lysine is first limiting amino acid in walnut, cashew, Brazil nut, and pine

nut while methionine is limiting in almond and pecan is limited by tryptophan and peanut is limited by threonine [79].

3. Protein Quality of Plant-based Protein Sources

Protein quality refers to the characteristics of protein like its amino acids profile, bioavailability and digestibility. Plant based proteins are a good alternative to meat protein but their protein quality is not much appreciated because of lack of IAAs and poor digestion. It is mostly compensated by using them along with other protein sources but it can cause potential deficiency in under developed or developing countries where these plants are staple food and assumed to be the only source of protein in daily diet [80]. Modern research on these plants revealed that they consist of anti-nutritional factors which affect their overall nutrient profile and causes poor digestion and reduce bioavailability. Soaking as a pretreatment and cooking can reduce anti nutrients and lead to better absorption [81]. One of the major factor that contribute to lower protein quality is less essential amino acids as compared to animal meats particularly Sulphur containing amino acids which may not meet the dietary requirements especially in case of children [82], moreover Plant protein exhibit exceptional techno-functional attributes like gelling, foaming, thickening, emulsification, solubility etc. which enhance their application in food industry where emulsification is necessary for stabilizing oil and water mixture, soy and mung bean is enriched with emulsifying character due to their amphipathic nature. Solubility is also very likeable factor about plant proteins, they generally show good solubility but few sources are less soluble at neutral pH which negatively impacts the protein quality [83]. Similarly gelling is a very important function of protein and it plays an important role to mimic the animal meats texture, few plant protein form a stable Gell that sounds as animal protein. Foaming is important for aeration in products and enhance lightness while water and oil retention ability is very crucial to maintain a suitable moisture level and preserving color and flavor [84]. Overall plant proteins possess fine functional attributes but these can be further enhanced with modern techniques and research to make them more suitable for innovative formulations [85].

Table 1. Essential amino acids of plant-based proteins sources

Plant-based protein sources		Essential Amino Acids									References
		Histidine	Isoleucine	Leucine	Lysine	Methionine	Phenylalanine	Threonine	Tryptophan	Valine	
Legumes	Soybean	2.5-3.5	4.4-5.8	7.3-7.9	4.7-5.8	1.1-1.4	3.9-4.9	3.3-3.6	0.3-1.0	4.5-5.3	[86]
	Lentils	2.9-3.7	5.5-5.6	8.4-8.5	4.6-4.8	0.6-1.3	3.9-4.0	2.6-2.9	0.4-0.6	5.6-5.9	
	Chick-pea	2.5-2.7	4.1-4.3	6.0-7.5	3.5-6.1	1.1	3.8-6.0	2.1-3.1	0.6	3.9-4.4	
	Peas	2.4-3.5	4.6-6.3	8.0-9.0	5.0-7.0	0.8-1.2	4.0-5.2	3.4-3.8	0.6	4.9-6.2	
Cereals	Wheat	0.25	0.51	0.82	0.36	0.19	0.66	0.37	0.195	0.57	[42]
	Rice	0.16	0.34	0.66	0.23	0.21	0.42	0.27	0.097	0.39	
	Oat	0.33	0.45	0.94	0.44	0.28	0.62	0.42	0.110	0.61	
	Barley	0.23	0.31	0.72	0.33	0.16	0.52	0.33	0.113	0.44	
	Maize	0.22	0.30	0.92	0.29	0.24	0.46	0.35	0.044	0.43	
	Rye	0.24	0.35	0.67	0.34	0.15	0.46	0.12	0.088	0.45	
Pseudo cereals	Quinoa	2.89	3.57	5.96	5.43	3.63 MC	6.10 PT	2.99	1.18	4.21	[87]
	Amaranth	2.86	4.28	6.46	5.49	3.07 MC	6.40 PT	4.10	1.13	4.95	
	Buckwheat	2.34	3.77	6.30	5.09	3.04 MC	5.84 PT	3.83	1.45	5.14	
Seeds	Canola seed	3.40	4.54	7.67	5.83	1.85	4.05	5.24	2.91	5.53	[88]

	Sunflower seed	0.8	0.88	1.33	0.94	0.43	0.98	0.86	-	0.99	[89]
	Flax seed	0.56	1.12	1.35	0.97	0.44	0.91	0.87	-	1	
	Chia seed	0.67	0.61	1.06	1.15	0.5	0.94	1.06	-	0.76	
	Sesame seed	0.98	1.09	1.99	0.90	0.72	1.35	1.22	-	1.28	
	Pumpkin seed	0.94	2.55	3.2	1.15	0.89	1.51	1.06	-	1.4	
	Hemp seed	1.06	1.49	1.96	1.64	1.01	2.03	1.12	-	1.89	
Nuts	Walnut	2.43	4.00	7.76	2.71	2.14	4.63	3.00	0.55	4.61	[90]
	Hazelnut	2.65	3.69	7.40	2.93	1.90	4.54	2.95	0.98	4.66	
	Peanut	2.54	3.45	7.03	3.88	1.31	5.83	2.21	0.73	3.95	
	Almond	2.97	3.79	7.19	3.06	0.81	5.46	2.60	0.70	4.41	
	Pine nut	2.23	3.65	7.30	3.54	2.93	3.59	2.43	0.84	4.52	
	Cashew	2.68	4.15	8.00	4.59	2.27	4.83	3.22	1.31	5.65	
	Pecan	2.80	4.08	7.51	3.17	2.52	5.09	2.90	0.47	4.72	

- Essential amino acids of legumes, pseudo cereals and nuts are expressed as g/100 g proteins
- Essential amino acids of cereals and seeds (except canola seed) are expressed as g/100 g of raw samples
- Essential amino acids of canola seeds are expressed as canola protein hydrolysates
- In pseudo cereals MC stands for "Methionine + Cysteine" and PT stands for "Phenylalanine + Cysteine"

4. Extraction Methods for Protein

Extraction methods are used to extract protein from its source and use it in variety of products to meet innovation. Earlier many traditional methods were used to extract protein like acid – base extraction and heat assisted techniques but with advancement of technology many innovative techniques have been introduced which are equally helpful in protein extraction in less time, less energy consumption and causing less or no damage to protein structure or quality. These methods have replaced the traditional methods because of their eco-friendly nature and efficiency [91]. Advance techniques are improving the food processing methods and enhancing Extraction rate and protein yield which is why they are more preferred [92]. Extraction methods are divided into two categories on the basis of use of water: one is wet extraction and other is dry fraction, while based on technology extraction methods are divided into conventional methods (acid-base extraction, alkaline extraction) and emerging or advance methods (enzyme assisted, ultrasound assisted I, microwave assisted and high pressure assisted method) [93]. It is very important and crucial decision to select the extraction methods from all above-mentioned techniques because extraction methods affect the functional properties of proteins like solubility, emulsification, foaming, gelling etc. with appropriate selection of extraction method these properties are enhanced and protein quality is improved and lead to more diverse food applications [94].

In wet extraction chemicals and water are used as solvent which solubilize the proteins in them and extract them from solution This method was use in old times but due to excessive and energy consumption it is least preferred nowadays in industry [95].

In dry fractionation air classification and tribo- electrostatic separation is used to separate the protein from its source. It is a mechanical process and rate of extraction of protein is influenced by milling speed and separator's setting. This method is considered better then wet extraction because of environmental-friendly nature and optimized yield and purity of protein [96].

Among advance methods, enzyme assisted technique is an innovative technique in which enzymes are used to break down cell wall and causes release of bioactive

compounds and protein which are isolated and used in various products. Enzyme causes release of protein with higher quality and enhanced functional attributes [97]. Enzyme assisted extract also use solvent like conventional methods but it used water which ensures a sustainable process without causing any environmental hazards [98].

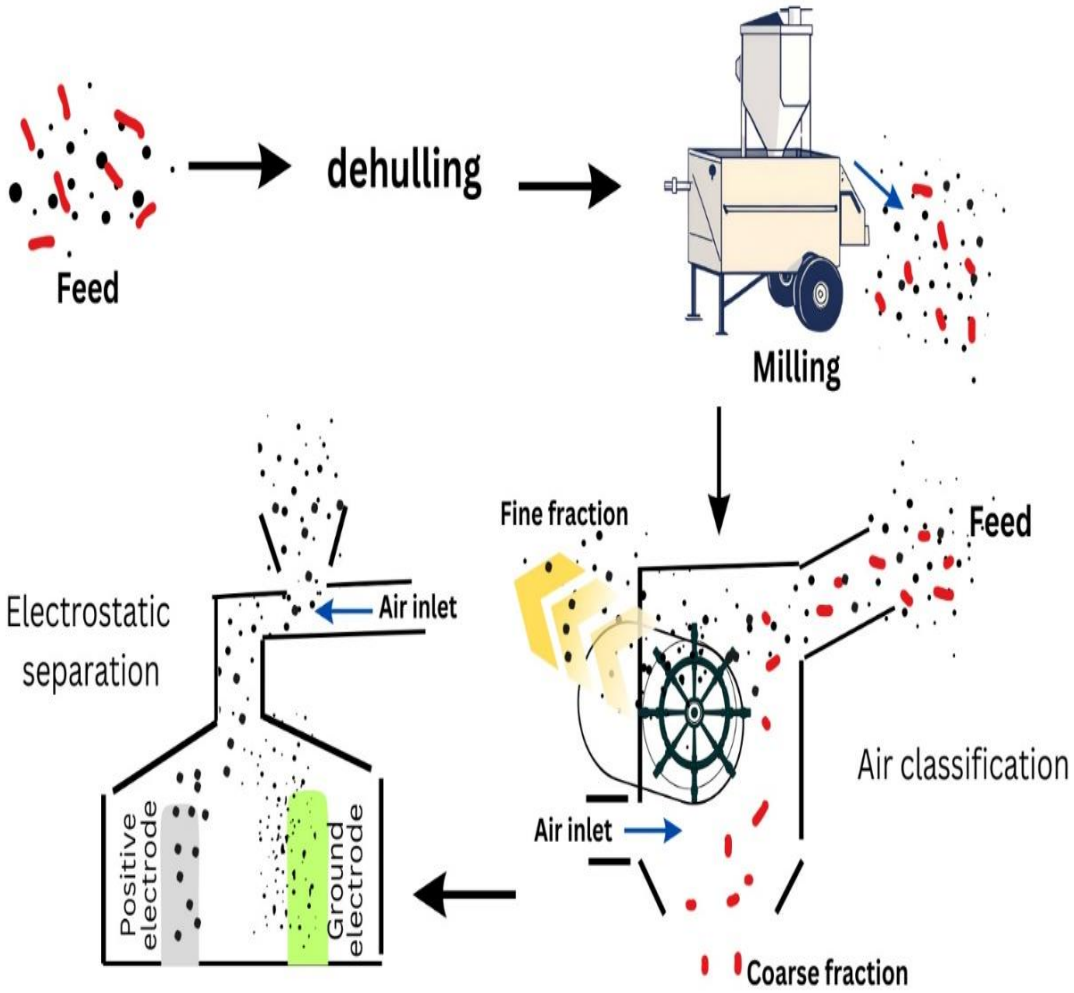

Figure1. Dry fractionation

Ultrasound assisted technique is another innovative method of extraction which uses high frequency acoustic Forces to rupture cell wall and causes transfer of protein to solvent. Usually acoustic streaming, shear, micro jet and shock waves are used which causes cavitation and facilitate extraction. This method is considered very efficient and widely used in developed countries [99].

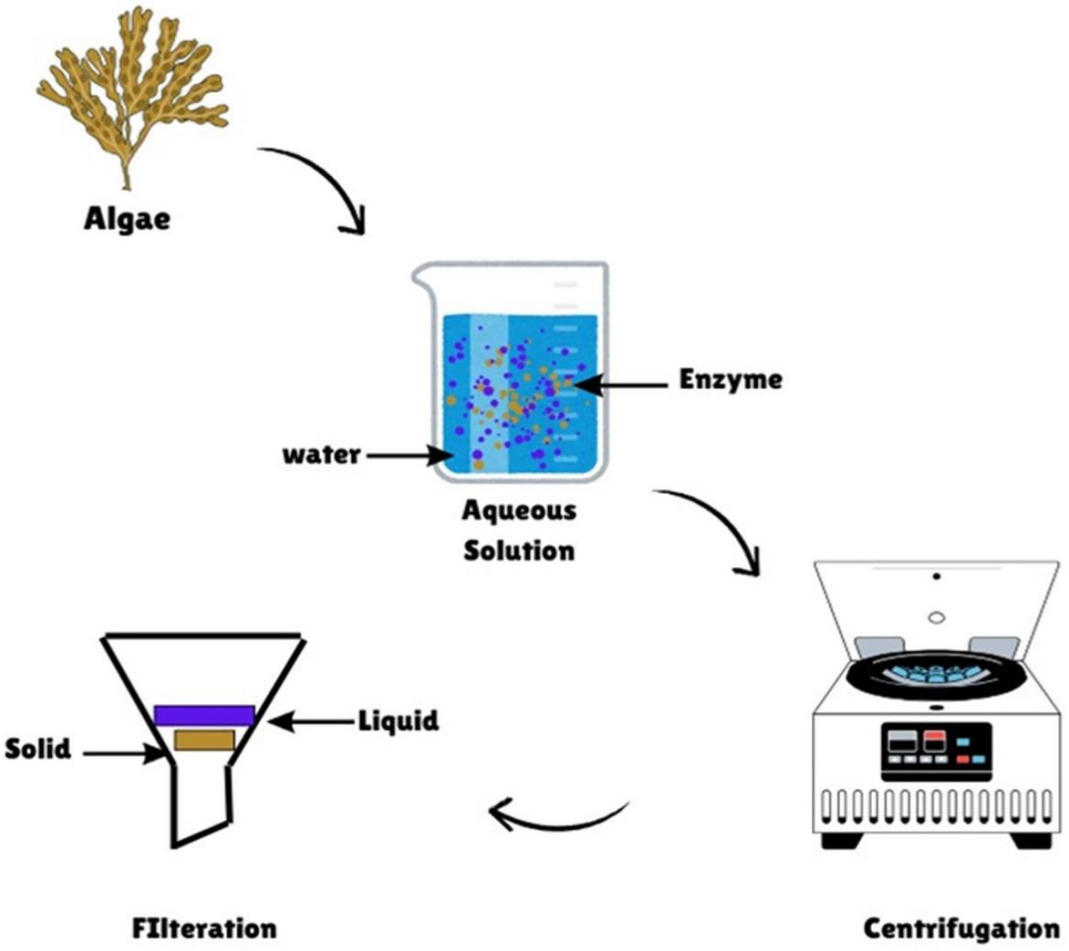

Figure2. Enzyme assisted technique

Microwave assisted technique involves use of microwave energy to heat the solvent which causes release of protein from plants effectively. Hot solvent comes in direct contact with the plant biomass and extract protein from matrix. Microwave technique is widely used in food industry to extract important constituents from the source for example phytochemicals like flavonoids and phenolics etc. are effectively extracted by this method. Microwave technique increases the efficiency of process such as it is found that extraction rate of protein in fermented brewer's grain is increased two folds by using microwave assisted method [100]. Furthermore, functional properties of

proteins are also enhanced along with other advantages like reduced time consumption, less solvent consumption and eco-friendly nature [101].

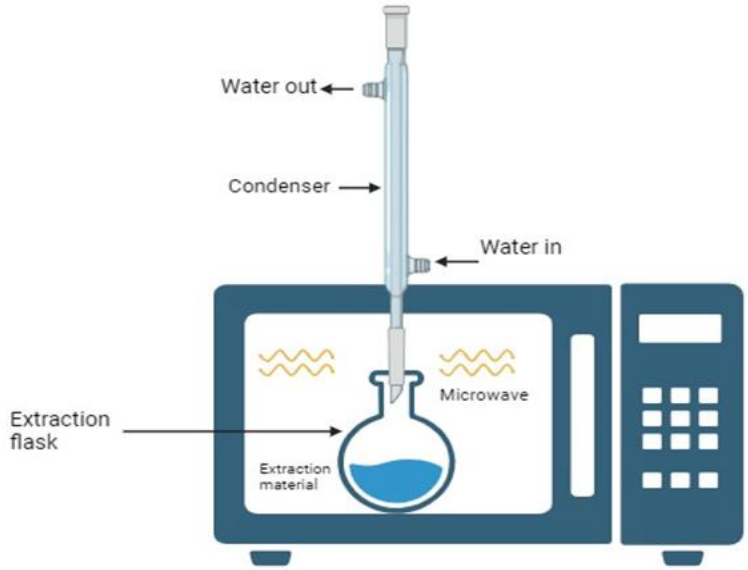

Figure 3: Microwave assisted technique

High pressure assisted method is an innovative and emerging technique used for extraction of protein from plant sources with enhanced protein quality and functional characteristics. In High pressure technique plant is suspended in a solution where high pressure (100-600mpa) is applied with a controlled pH (ideally 7.1) to extract its protein in a prescribed time (approximately 24 hours) this results in extraction of a pure and high-quality protein yield [102].

5. Environmental Impacts

Plant based proteins are very appreciated for their environmental-friendly nature. They play a significant role in conserving natural sources. Studies revealed that plant sources use relatively less land, water and energy as compared to animal protein and reduce any negative impact on environment but land use can vary depending upon crop type and cultivation practices [103] animal agriculture is a significant source of green-house gases. Plant based proteins sources are very helpful to reduce these greenhouse gases emission and protecting the environment. A study in Norway revealed that

replacing red meat with a protein source crop can reduce 1.07-1.76 million tons of carbon dioxide which makes 10-16% of food production emission [104], so it is believed that shifting from animal to plant sources for diet can be helpful in reducing climate change and sustainable environment. Another major concern is loss in biodiversity as using livestock for dietary purposes is a major factor of biodiversity loss and also lead to harmful gases emission, therefore shifting to plant sources can be a potential solution to mitigate this problem, even a minor decrease in meat intake can save a good number of animals contributing to maintain bio-diversity [105]. Current good habits and culture is putting a lot of pressure on ecosystem, as dietary choices and biodiversity are interconnected so it is important to be careful while choose your meal. Researchers have given three strategies to get out of this problem and ensure sustainable environment, first one is improvement in food production, second is reduction in food wastes and third is dietary change. It is understandable that animal diets put more negative impact on climate as compared to plant diet [106]. According to a survey livestock is 18% cause of greenhouse gases emission in which beef contributes 41% and milk 20%, so FAO suggest an ecological bases diet which is minimal processed and locally available [107].

6. Challenges Faced by Plant Protein Sources

Alternate protein sources are gaining popularity but still facing many challenges to make their place in market. One basic reason is that protein alternatives are failed to match the organoleptic properties of original meat because plant protein are globular and they are difficult to extract because mostly nutrients are present in storage organs like seeds and tubers for which a proper extraction technique is required, moreover to Mimic the emulsifying and gelling character of meat a very large amount of plant protein is required which exceed the affordability [108]. Another major challenge is to create the meat flavor in plant proteins because undesirable taste is developed in plant protein such as beany, bitter and astringency. Flavor present in plant protein is very unstable and modified on exposure to heat and pressure [109]. Creating meat like texture is also a difficult challenge for alternative proteins because plant release volatile compounds due to oxidation in polyunsaturated fatty acids which results in odor, off flavor and undesirable texture [110]. One of the most concern challenges is allergen

city or food allergies. Food allergy refers to adverse human health affects arising from an abnormal immunological response following exposure to a certain food. It is reported that plant-based foods are major cause of allergy in adults [111]. Allergy because of food is categorized into two types: first one is primary food allergy A, it is allergy to a plant origin food like soy or peanut etc., while second one is Secondary food allergy B, it is associated to pollen of a plant tree. It is reported that only wheat is associated with 20 kinds of allergies in which 15 are entirely food allergies [112]. Toxicological effects of plant proteins also limit their function because they may have anti nutritional factors, phytoestrogen and few oligosaccharides which decrease the bioavailability of protein and lower protein quality [113]. Plants protein industry is struggling to scale up their production level to meet the global dietary requirements. It is because of limitations in techno-functional attributes of plant proteins which are unable to match the animal proteins such as low digestibility, low solubility and poor gelling ability as compared to animal protein. Cultivation of plant protein sources is also limited because of genetic potential, phenotyping difficulty and environmental impacts from nitrogen fertilizer [114]. Many other factors like land availability, less consumer acceptance, agrochemicals requirements, and limited research on plant proteins contributing to slow development of this industry [115].

7. Opportunities

Plant based protein industry is rapidly growing and experiencing significant opportunities as the development in technology made it easy for protein extraction from plants and utilize it in various products. Many advanced methods like enzymatic extraction, high pressure processing, fermentation are modern techniques which can fasten and improve availability of protein, additionally, many techniques are developed to improve functional properties of plant proteins to compete with animal proteins like 3D extrusion and ultrasound are most advanced methods which have opened a path toward development of this industry [116]. Furthermore, bioactive peptides identification is improved which is leading to exceptional market potential in this field because their nutritional value is enhanced, but as the industry is growing, it is important to address regulatory challenges and ensure a sustainable growth and provision of protein to meet the demand along with better nutrition and functional

properties [117], moreover consumer awareness should be ensured and proper education should be provided along with marketing strategies to catch market and overcome social barrier and accept plants diet as an alternative protein, moreover, key formulation techniques are used to modify physically chemically and biologically by amyloid, fibrillations and dual methods to enhance nutritional, functional and organoleptic properties [118].

Conclusion

The exploration of plant-based protein sources from above literature highlighted that they are a significant and suitable alternative to animal protein. The key findings underscore that plant-based protein are enriched in essential amino acids though often required a strategic combination to compensate any deficiency. Furthermore, plant-based protein sources are also providing other nutrients like vitamins, minerals, dietary fibers and they are significantly lower in saturated fat and cholesterol as compared to animal protein. Technological advancement is improving functional properties of proteins and enhancing their flavor and texture to complete animal protein and appeal consumer who are more attracted towards meat. Growing global population, increasing protein demand, increasing consumer awareness, ethical concerns and search for a sustainable and eco-friendly source of protein is driving us to world of plant-based protein. Innovation in processing techniques like fermentation and extrusion etc. are making them more versatile and palatable. However, there is still a gap in research and need for further investigation. A novel and nutritious formulation is needed to be developed by in-depth research of nutrients present in plants. Assessment of the long-term effects of consuming high level of plant-based protein particularly in vulnerable population (children and elders) is needed. Addressing these research gaps will pave the way for development of even more nutritious, delicious and sustainable plant-based protein products ultimately contributing to heathier and secure future.

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Conflicts of interest

The authors do not have conflict of interest.

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