Review Article

Antinutrients in mungbean and strategy for reduction: An overview

Abstract

Mung bean (Vigna radiata L. Wilczek) is a highly nutritious pulse, particularly popular in Asian countries and traditionally used in medicine. It is a rich source of protein, dietary fiber, essential vitamins, minerals, and amino acids, including calcium, magnesium, iron, phosphorus, and potassium. Trypsin inhibitors, tannins, phytic acid, saponins, and polyphenols are some of the antinutritional substances found in mung beans that can lower their nutritional value by preventing the digestion of proteins and carbohydrates, causing problems with the liver and intestines, and binding nutrients. These antinutrients can have both beneficial and detrimental impacts on human health, and they are essential to plant physiology for defense and seed storage, among other functions.Several processing techniques, including soaking, autoclaving, cooking, sprouting, roasting, and dehulling, can be employed to lessen their effects; each technique efficiently reduces a particular type of antinutrient. Moreover, mung bean varieties with reduced amounts of antinutritional substances may be developed using breeding techniques like selection, backcrossing, and mutation breeding, which would improve the beans' total nutritional value.

Keywords: Mungbean, Antinutrients, Processing, phytate, Trypsin, polyphenols

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Introduction

Legumes are a broad category of crops that belong to flowering plants and yield seeds in pods that are frequently processed for use in food and feed. With about 19500 species and 750 genera, legumes are the third-largest family of flowering plants (Abbas and Ahmad, 2018). They are an important source of dietary protein and play a big role in the diets of the impoverished in underdeveloped and emerging nations where it is generally more expensive to get animal protein. In addition, compared to other high-protein food kinds, legumes have minimal environmental impact (Afam *et al*., 2016).

The mung bean (*Vigna radiata* L. Wildzek) is a popular pulse that is consumed all throughout the world, but especially in Asian nations, where it has long been used in traditional medicine (Hou *et al*., 2019). India is the world's leading producer of mungbean, which is cultivated in practically every state. Mungbean production on 33.37 lakh hectares was 17.5 lakh tons in Kharif 2022–2023. It is grown both alone and in combination with other crops, including as maize, sorghum, cotton, and minor millets (http://www.agricoop.nic.in).

Like other members of the legume family, mung beans are composed of 63% carbohydrates, 16% dietary fiber, 1% fat, and 24% protein (Huang *et al.*, 2013). While its various food productslike cake, sprouts, noodles, and soupsevolved in oriental nations like China, the Philippines, and Thailand, its savoury foods, sweets, snacks, and dhal (thick stews made from dehulled and split grains) have evolved and gained popularity in the Indian subcontinent (Dahiya *et al*. 2013). In addition, mung beans are a highly well-liked Asian dish that have significant advantages over other legumes, including the ability to cleanse, reduce inflammation, fight tumor growth, lower cholesterol, and diuretic effects (Hu, 2003).

There has never been an attempt to scientifically validate the safety and quality of food produced using conventional processing techniques. Different processing techniques, such as soaking, dehulling, cooking, germination, and roasting, can be used to reduce the antinutrients; however, research is still needed to determine how beneficial these techniques are in compared (Luo& Xie, 2013). The health concerns associated with mung bean consumption may be significantly decreased with the documentation of processing techniques that effectively remove the antinutrients found in mung beans. Therefore, it is more than justified to try to enhance the nutritional qualities of mung beans with home remedies to lower antinutritional elements.Hou*et al.*,2019). Thus current review provides a knowledge about different processing techniques to be followed for elimination of antinutrients from mungbean.

Health benefits of mung bean

Green gram seeds are used medicinally to treat obesity, fever, and other conditions. In the Ayurvedic medical system, it is beneficial for skin conditions, heat-related illnesses, and weakness. In India, green gram flour is used to make herbal soap. A common ingredient in Asian cooking, green gram sprouts are high in vitamins and minerals. According to recent studies, green gram starch is a good source of slowly digesting carbohydrates for those with diabetes. It alters lipid and glucose metabolism favourably and causes a blood glycaemic response in humans (Randhir *et al*., 2004).

It has been observed that the green gram helps to moisturize the skin and regulate unsettled stomach. According to Tang *et al*. (2014), the main components of green gram's anti-melanogenesis, antioxidant, antimicrobial, anti-hypertensive, anti-inflammatory, immunomodulatory, and antitumor properties are believed to be its high levels of proteins, amino acids, oligosaccharides, and polyphenols. These components also play a role in the regulation of lipid metabolism.

Structure of mung bean cotyledon

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Mature seeds have three essential components: the embryo, cotyledons, and seed coat. 7–15% of the total seed mass is made up of the seed coat. Approximately 85% of the seed mass is made up of cotyledons, with the remaining 1-4% coming from the embryo, as seen in Fig.1. The exterior structures of the seed are the testa, hilum, micropyle, and raphe. The outside portion of the seed, known as the testa (smooth or rough), almost completely covers the seed surface. Where the seed was attached to the stalk, the hilum is an oval scar seen on the seed coat.The micropyle is a tiny hole near the hilum in the seed coat. An edge on the hilum that faces the micropyle is called the raphe. The embryonic structure is the surplus material that remains after the seed coat is removed from the grain. There are two cotyledons and a brief pivot above and below them that make up the early-stage structure. The seed coat provides a flimsy assurance, but the two cotyledons are not genuinely attached to one another other than at the pivot. The seed is hence unusually susceptible to breaking (Patel *et al*., 2016).

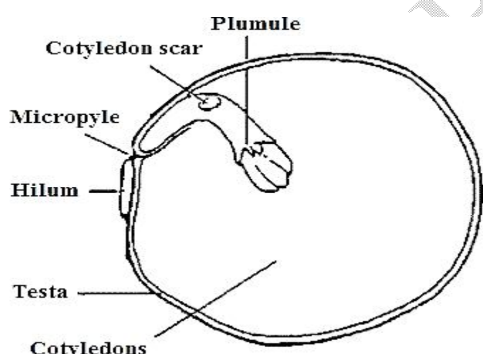

Fig.1 : Structure of mung bean cotyledon (Patel *et al.*, 2016).

What are the antinutrients?

Chemicals known as antinutrients hinder the body's ability to grow and function normally when they are present in food, whether it comes from humans or animals. anti-nutritional factors (ANFs) or non-beneficial compounds that can affect human and animal growth as well as reduce their nutrient intake, absorption, and utilization. These include phytic acid, saponins, alkaloids, certain oligosaccharides, protease inhibitors, glucosinolates, tannins, and cyanogenic glycosides (Ali*et al.*,2022; Dey*et al.*, 2022). Antinutreints are known to alter the absorption of nutrients such as vitamins, minerals, and proteins in addition to inhibiting enzyme activities. There are a plethora of studies that have proven the negative impact of ANFs on nutrient bioavailability in different living organisms (Ali*et al.*,2022; Dey*et al.*, 2022). However, the deleterious effects of ANFs on nutrient metabolism vary according to age,

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species, concentration of ANFs, processing, and interactions with other nutrients. According to Diouf *et al.* (2019), anti-nutritional agents are primarily found in foods and feed material made from grain legumes and pulses, as well as in pulses themselves.

Beneficial effects of antinutrients in plant

There can be no doubt that the so called "anti-nutrients" of legumes have a biological function. They are certainly important in the physiology of seedlings as N or C storage compounds and to facilitate nutrient uptake and rhizosphere establishment (Lal*et al.*, 2017).

Table 1: The major role of anti-nutrients in consumption and plant growth regulation.

Sr. no.	Anti-nutrient	Effects on consumption	Role in plant growth	References
1.	Phytic acid	1. Animal nutrition inhibitor. 2. Decreases the level of blood glucose.	Micronutrient chelation and phosphorus storage for growth and development.	Gupta *et al.*, 2015; Dilworth *et al.*, 2005;
2.	Saponins	1. By causing damage to red blood cells, induce vomiting and diarrhea. 2. Impacts the gastrointestinal membranes' ability to absorb nutrients. 3. Detrimental effect on the feed efficiency and chick development.	Resistance against diseasesinvegetables.	Akande *et al.*, 2010; Cárdenas *et al.*, 2015
3.	Trypsin inhibitor	Protease inhibitors reduce the activityof some enzymes duringingestion.	Confer biotic stress tolerance and act as biopesticides.	Battelino *et al.*, 2019; Amaral *et al.*, 2022; Ribeiro *etal.* 2015
4.	Tannins	1. Damage the intestines and inhibit digestion enzymes. 2. Have been linked to decreased protein digestibility, growth rate, feed efficiency, and feed intake.	Act againstpathogenic bacteria,have antibacterial actions, and are antioxidants.	Akande *et al.*, 2010;

Anti- nutritional profiling of mungbean

Antinutritional substances found in mung beans include tannins, phytic acids,

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polyphenols, trypsin inhibitors, and saponins. Sivakumaran *et al.* (2017), Dahiya *et al.* (2015), Singh *et al.* (2015), Abeykoon *et al.,* 2021).

1.Tannins

The seed coat of most legumes contains significant levels of tannins, which are polyphenolic chemicals. It interferes with iron absorption by forming an irreversible bond with iron. Moreover, they bind proteins, decreasing their availability, and interfere with the absorption of B vitamins. The ancient word "tannin" refers to a customary invention. The method of turning raw animal hides or skins into sturdy, non-putrescible leathers by applying plant extracts from different plant sections was referred to in the logical writing as tanning. Plant polyphenolic component tannin, which has a molecular weight ranging from 500 to over 3000, is an astringent, bitter substance that either binds or precipitates proteins and other organic molecules, such as amino acids and alkaloids (Gemede & Ratta, 2014).

Molecular formula: $C_{76}H_{52}O_{46}$

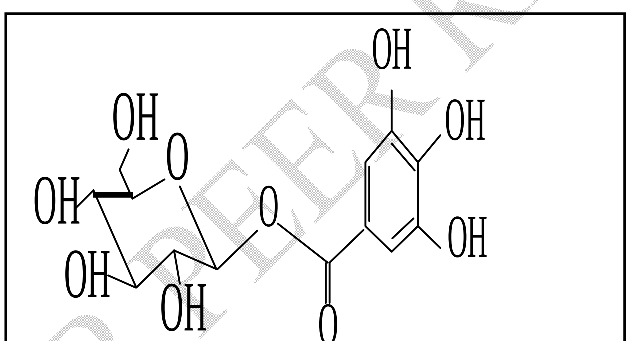

Fig.2 : Structure of hydrolyzed tannin (Diouf *et al.* 2019)

2.Phytate phosphorus

Foods strong in fiber, such as legumes and cereals, also have high levels of phytate or phytic acid. In the fully grown seeds of both monocot and dicot plants, phytate, or myo-inositol 1,2,3,4,5,6-hexakis (dihydrogen phosphate), is a major form of phosphorous storage. It typically accounts for more than 80% of the soluble myo-inositol phosphate in seeds and about 75% of the total phosphorous (Dorsch *et al*., 2003). Plant seed components make up the majority of animal and human feeds. Because humans lack phytases, seed phytic acid is typically inaccessible to monogastric animals like humans and is expelled as manure (Gupta *et al.,*2015).Water quality problems and eutrophication are caused by the excretion of undigested phytic acid in fertilizer (Yang*et al*., 2008). A phosphatase called phytotasehydrolyzes phytate to produce free orthophosphate and inositol (Zeller*et al*., 2015). It's available in a lot of seeds. Cereals contain a large amount

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of phosphorus in the form of phytate. Iron, zinc, calcium, and magnesium are bound by phytotate, which prevents them from being absorbed (Aneta and Dasha, 2019). Phytic acid (IP6) and phytin (phytate salts) are the two main types of phytates that are encountered. Phytates are chelating agents; their primary mechanism of antinutritional action is the binding of metals (Ca+2, Mg+2, Fe+2, Zn+2, and Cu+2), which results in inadequate absorption (Nissar*et al*., 2017).

Furthermore, protein consumption is negatively impacted by the creation of protein-phytate complexes (Wang & Guo, 2021). Moreover, phytates affect the activities of trypsin, pepsin, and amylase (Khan & Ghosh, 2013).Researchers have reported that the estimated values of phytate content in mungbean are as follows: 57.62 mg/100g (Oburbaga and Anyika, 2010), 63.2 mg/100g (Ravindran *et al*., 1976), 58 mg/100g (Mubarak, 2005), 66.47 mg/100g to 69.24 mg/100g (Kakati *et al*., 2010), 44.80 mg/100g (Dahiya *et al*., 2015). According to Srinivasan *et al*. (2007), phytic acid can be viewed as an anti-nutritional substance from a nutritional standpoint, yet it gives grains protection from the bruchid beetle (*Callosobruchus maculates*) when they are being stored. Wide variations in reported phytic acid levels in mung beans were discovered by Sompong *et al*. (2009). These variations could be caused by genetic variations, since some reports indicate that the phytate content of mung beans is inherited on a genetic basis. Upendra Pokharel (2021) investigated the impact of processing techniques on antinutritional factors found in mung beans. The findings showed that the raw mung bean's phytate content was 626.54 mg/100 g, which is comparable to the 622 mg/100 g found by Singh *et al*. (2015). However, this value is lower than the range of 727 to 940 mg/100 g found by Bindu *et al*. (2017). According to Zafar *et al*. (2023), phytic acid levels in mung bean cultivars ranged from 0.45 to 1.2 percent, with a mean value of 0.69 percent. **Molecular formula: $C_6H_{18}O_{24}P_6$**

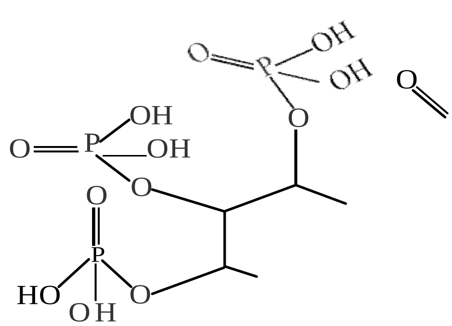

Fig.3: Structureofphyticacid
Source: Aneta and Dasha (2019)

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3.Polyphenols

All higher plants contain phenolic compounds, which are widely distributed bioactive secondary metabolites mainly produced by the pentose phosphate, phenylpropanoid, and shikimic acid pathways (Balasundram*et al*., 2006). They can range from simple molecules to extremely complex polymers and share a structural characteristic of having one or more hydroxyl groups directly attached to the aromatic ring. Based on the number of phenolic hydroxyl groups connected and the structural components that link benzene rings, phenolic compounds are subdivided into subgroups of phenolic acids, flavonoids, tannins, and stilbenes (Singh *et al*., 2016). It is estimated that about 8,000 phenolic chemicals found in flora have been identified and extracted (Ouchemoukh*et al*., 2017). Foods' sensory qualities are influenced by phenolic compounds, and tannins are mainly responsible for theastringency of food sources (Landete, 2012).

4.Saponins

Saponins are naturally occurring substances that are found in every cell of legume plants. A complex and chemically diverse category of chemicals, saponins derive their name from their ability to produce stable, soap-like foams in aqueous solutions (Arunasalam*et al*., 2004). Saponins are glycosidic, amphiphilic, and heat-stable substances that are typically found in a wide range of plant foods. They consist of one or more oligosaccharide moieties bonded to an aglycone that is either steroidal or triterpenoid. These compounds have amazing foaming and emulsifying qualities because of the strong hydrophilia of the sugar chains and the excessive hydrophobicity of the aglycone (Timilsenae*et al*.,2023).

They are structurally made up of a lipid-soluble aglycone that is coupled to one or more water-soluble sugar residues of various kinds and quantities of sugars that are found in many different plants. This aglycone can contain either a sterol or a triterpene group. The types, quantities, and content of the steroid ring determine the architectures of saponin found in various plant diets (Rao and Sung, 1995). The figure depicts the structure of soyasaponin III, which is found in mung beans.

Molecular formula: $C_{58}H_{94}O_{27}$

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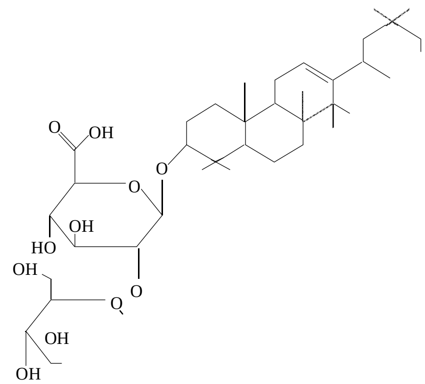

Fig.4: Structure of soyasaponin III present in mung bean
Source: Shi *et al.* (2004)

5.Trypsin inhibitor

A protein known as a trypsin inhibitor (TI) function as a kind of serine protease inhibitor (serpin) by regulating the activation and synergistic reactions of other proteins, thereby reducing the biological activity of trypsin. Based on molecular weight, legume TIs are categorized into two families: Bowman-Birk (BBTIs) with molecular weights of about 8 kDa and Kunitz (KTIs) with molecular weights of about 20 kDa. While mung beans, cowpeas, lentils, etc. exclusively contain trypsin inhibitors from the BBTI family, soybeans have inhibitors from both groups. According to Vanderven et al. (2005), KTI has two disulphide bonds while BBTI has seven. Trypsin is an enzyme that breaks down a variety of proteins, mostly during digestion in humans and other animals like young ruminants and monogastric animals. When trypsin inhibitor is consumed, it acts as an irreversible and competitive substrate (Silverman *et al.*, 2001). It competes with proteins to bind to trypsin and therefore renders it unavailable to bind with proteins for the digestion

Furthermore, chymotrypsin function is partially interfered with by trypsin inhibitors (Vagadia*et al.*, 2017). Trypsin inhibitors lower the effectiveness of proteins, which means that the body of the consumer cannot fully and efficiently utilize the protein (Klomklao*et al.*, 2011).

Strategies for reduction of Anti-nutrients from mung bean

Strategies for reducing anti-nutrients in mung beans include traditional processing methods such as soaking, sprouting, fermentation, and cooking. Each of these methods has been shown to significantly lower the levels of various anti-nutrients, thereby improving the digestibility and nutritional quality of mung beans(Samtiya*et al.*2020).

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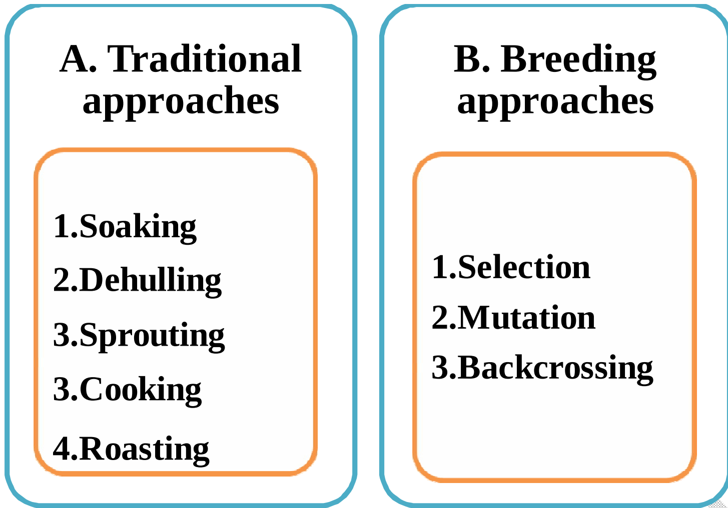

Fig.5 Different approaches for reduction of anti-nutrients from mung bean

A. Traditional approaches

1. Soaking

According to Siddiq and Uebersax (2012), soaking causes water to permeate the protein fraction and starch granules, which desaturates the protein and makes the texture of the beans mushy. Since phytate dissolves in water, soaking beans in water for the entire night significantly reduced the amount of phytate present. After soaking for 12 hours, mung beans' phytic acid content decreased by 18%. After soaking, mung beans' levels of polyphenols decreased by 23% whereas those of the trypsin inhibitor decreased by 7% (Grewal and Jood, 2006). Mung beans that are soaked for 24 hours have a 28–35% different tannin content (Kakati *et al*., 2010).

2. Cooking

Cooking often inactivates volatile molecules and heat-sensitive antinutritive factors including chymotrypsin and trypsin inhibitors. Tannin content in a variety of pulses, including kidney, mung, lentil, and cowpea, may have decreased after boiling as a result of tannins binding to proteins and other organic materials (Kaur *et al*., 2020; Kumar *et al.*, 2021). Cooking destroys polyphenols in addition to tannins (Yasmin *et al*., 2008). Pre-soaked cooked mung beans had a 15% and 20% decrease in phytic acid and tannin, respectively. Tannins and phytic acid are reduced in cooked mung beans that haven't been soaked by 25% and 30%, respectively (Singh *et al*.2015).

3. Autoclaving

Trypsin inhibitor and hemagglutinin were completely eliminated from mung beans using autoclaving (Mubarak, 2005). Phytic acid and tannin have been reduced by 34% and

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44%, respectively, in presoaked autoclaved mung bean samples (Singh *et al.*, 2015). Phytic acid and tannin have been reduced by 32% and 40%, respectively, in unsoaked autoclaved mung bean samples (Singh *et al.*, 2015).

4.Roasting

Roasting is done to enhance the product's sensory attributes and inactivate harmful enzymes, which enhances the product's nutritional value and storage capacity (Rackis*et al.*, 1986). According to Singh *et al*. (2015), roasting mung beans can reduce their phytic acid, tannin, trypsin inhibitor, and polyphenols by up to 30%, 17%, 92%, and 17%, respectively.

5.Dehulling

One of the important post-harvest processes for improving the palatability of food grains is dehulling, or the removal of the seed coat from pulses. However, dietary fiber and minerals are lost as a result. The embryo and sticky layer that are present between the hull and the cotyledons are also removed by dehulling (Kumar *et al*., 2021). Dehulling decreased condensed tannin and polyphenol concentration while increasing protein content. (Egounlety and Aworh 2003; Alonso *et al*., 2000).

6. Sprouting

During sprouting, the enzymatic system of the seed gets activated. It has been recognized as one of the best processing techniques for raising the nutritional content of pulses and improving the nutrients' digestibility, especially protein and carbs (Kumar *et al*., 2021). This is the most efficient way to lessen the phytic acid in legumes. During germination, phyttic acid is broken down, increasing the availability of inorganic phosphorus (Virginia *et al*., 2012). Tannin, phytic acid, and trypsin inhibitor are all decreased by 67%, 31%, and 23%, respectively, upon mung bean sprouting (Mubarak, 2005).

Physical processing	**Comments**	**Results**	**References**
Soaking	Deeping the seeds in water with or without anyadditives.	Reducestheoligosaccharideslikera□nose and stachyose.	(Egounlety and Aworh 2003)
Autoclaving	Heating at high temperature121°C under pressure.	Reduction in trypsininhibitors and tannins.	(Abbas, and Ahmad, 2018)(Kumar *et al*., 2021)

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Cooking	Ordinarydomestic cookingby using pressure cooker.	Reduction of tanninscontent after cooking due to the binding of tannins withproteins and other organic substances during cooking.	(Kaur *et al.*, 2020) ,(Kumar *et al.*, 2021)
Sprouting	Soaking the seed in ethanolfor one minute. (forsterilization) followed by keeping it in moist cotton until germination appear.	Reduces antinutritional factors such as trypsin inhibitors, phytic acid stachyose and raffinose.	(Mubarak 2005).
Roasting	Dry heating about 120°C-250°C.	Removes of trypsin inhibitor	(Khattab *et al.*, 2009)

Table 2 Overview of different approaches for reduction of anti-nutrients from mung bean

B. Breeding approaches

Selective breeding techniques and hybridization are the main components of traditional breeding practices used to reduce antinutrients in mung beans. For decades, farmers have employed these methods to enhance various aspects of crops, such as their nutritional value (Duraiswamy*et al.,*2023). Here's a thorough explanation of how these techniques can be used to lower the antinutrient content of mung beans:

1. Selection

The process of selection includes discovering the plants with desirable traits, keeping them, and getting rid of the ones that have unfavorable traits. It was also shown that screening pulse germplasm for reduced enzyme inhibitors was an effective method of locating possible donors with reduced inhibitors (Ahmar*et al*., 2020). Orf and Hymowitz (1979), identified two zero Kunitz inhibitor lines in soybeans, PI 157-440 and PI 196-168.

2. Backcrossing and Mutation breeding

A backcross occurs when an organism combines with either of its parents. Plant mutation breeding, also known as variant breeding, is an approach for creating novel crop varieties that involves causing spontaneous genetic diversity in plants using chemical or physical methods(Ahmar*et al*., 2020). A technique backcrossing is used to improve or introduce a certain trait in a plant while keeping other desired traits from the original parent. Using this method to add a feature like lower antinutrient levels to well-adapted or high-yielding cultivars is especially helpful (Hospital, 2005).In order to add variety to a plant population, mutation breeding entails causing genetic mutations. If the pathways for

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antinutrient synthesis are affected by these mutations, it may result in lower amounts of antinutrients. By carefully applying these methods, breeders can develop mung bean varieties with reduced antinutrient levels, enhancing the nutritional quality of the crop while maintaining desirable agronomic characteristics (Wani*et al*., 2014). The two primary approaches for reducing anti-nutritional characteristics in crops are backcrossing and mutation breeding (Wilcox et al., 2000; Yuan *et al*., 2009). Using both spontaneous and induced mutations in important crops, phytic acid has been successfully reduced (Pramitha *et al*., 2021).

Future prospects

Large amount of mung bean is imported from India as it has the highest production in the world Nowadays, production rate is also increasing day by day mainly, and hence consumption is also increasing due to its beneficial effect in the human body. Present reviewprominentlydeals with antinutrients in mung bean and effect of various processing methods to reduce those antinutrients. Hence, review might help in the establishment of the effective and optimized way for the use of green gram in household level and industrial levels.

Conclusions

This review highlights the key antinutritional components in mung beans, such as trypsin inhibitors, tannins, and phytic acid, which can reduce nutrient absorption and cause health issues. To mitigate these effects, processing methods like dehulling, pressure cooking, and sprouting are most effective, preserving nutrients while reducing antinutrients. Additionally, breeding techniques like selection, backcrossing, and mutation breeding show promise in eliminating harmful compounds

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