

Scientific Report:
**Study of national plant
proteins with potential for
application in plant-based
products**



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The Good Food Institute (GFI) is an international non-profit organization working to transform the food production system. We operate in Brazil, the United States, India, Israel, European countries, and the Asia-Pacific region to build a world where alternative proteins are no longer alternatives. We are funded by philanthropy, and all of our work is provided free to society. We exist to make food systems better for the planet, people, and animals. To achieve this, we identify the most effective solutions, seek resources and talent, and empower partners throughout the food system to make alternative proteins more accessible. Reimagining how we obtain protein for human consumption is urgent and essential. Plant-based analogs to animal-derived products are one of the concrete alternatives to help Brazil transition to safe, fair, and sustainable agriculture. Side by side with sustainable animal-derived proteins, we can form a consistent response from our country and our agricultural economy to the new scenario,

in which different sources of protein for human consumption will coexist. This is an 'and' market, not an 'or' market: there is space and demand for everyone to act.

To identify the biggest challenges for the development of plant-based products analogous to animal products with the quality, price, and sensory characteristics sought by consumers, GFI Brazil conducted a survey with professionals from the ingredient and plant-based product processing industries. According to the survey 'Opportunities and Challenges in the Production of Plant-Based and Animal-Origin Products,' 84% of respondents said it was a priority to develop new sources of national plant proteins. Thus, to accelerate the development and application of new sources of plant proteins produced nationally, GFI Brazil commissioned a study focusing on National Plant Proteins. The study was conducted by researchers and professors from the Faculty

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List of abbreviations and acronyms

BSG

Brewer spent grain.

Conab

Brazilian National Supply Company.

CS

Chemical score.

DDG

Dried distillers grain.

DDGS

Dried distillers grain with solubles.

EA

Emulsifying activity.

EAI

Emulsifying activity index.

EC

Emulsifying capacity.

Embrapa

Brazilian Agricultural Research Corporation.

ES

Emulsion stability.

ESI

Emulsion stability index.

FAO

Food and Agriculture Organization.

FC

Foam capacity.

FS

Foam stability.

GHG

Greenhouse Gas.

IAC

Campinas Agronomic Institute.

IBGE

Brazilian Institute of Geography and Statistics.

MAPA

Ministry of Agriculture, Livestock and Food Supply.

MDIC

Ministry of Development, Industry and Foreign Trade.

PDCAAS

Protein digestibility-corrected amino acid score.

Taco

Brazilian Food Composition Table.

WDG

Wet distillers grains.

Introduction



By 2050, world food production needs to be 70% to 100% higher than today to serve the estimated population of 10 billion people (Tilman *et al.*, 2011; FAO, 2019). Thus, it is necessary to rethink the current food production system, since much of the cultivable land on Earth is used as pasture or to provide animal feed (FAO, 2020). The territorial expansion of agricultural activity should be analyzed with caution, as it results in serious short and long-term environmental implications, such as deforestation, loss of biodiversity, emission of greenhouse gases (GHG) and consumption of already scarce water resources (Tilman *et al.*, 2011).

The current food production system generates large, and sometimes unavoidable, by-products and waste: about 38% of waste is generated during food processing (Tassoni *et al.*, 2020). In addition, in a document presented in 2011, the United Nations Food and Agriculture Organization (FAO) claims that about 1.3 billion tons of food are lost or wasted per year along the entire production chain,

from production to consumption (FAO, 2011). In Latin America, fruits and vegetables are the major contributors to food waste, accounting for 62% of waste. Roots/tubers and cereals contribute 43% and 16% to these indices, respectively (FAO, 2011). In general, hulls/peels/skins, stems, seeds, bran and residues of shavings after the extraction of oil, starch, juice and sugars are considered plant residues, since these have no commercial value and have as their main destination the disposal in landfills (Ganesh *et al.*, 2022). In addition to waste, food processing chains result in numerous by-products, which end up being destined for animal feed, such as cakes obtained from oil extraction, or are marketed at a lower market value, such as grains broken down during the processing of legumes, such as beans.

Due to this context and the positive correlation between the consumption of proteins of plant origin and health, in addition to the growth of the

vegetarian, vegan and flexitarian public, plant proteins are increasingly gaining the preference of consumers and driving research aimed at exploring new ingredients (Aiking, 2011; Alves & Tavares, 2019; Boland *et al.*, 2013). Brazil is a strong candidate to take the lead in the plant-based market, since it is one of the most important food producers in the world, contributing to the food of about 1.5 billion people around the world and with more than 400 agricultural products (Embrapa, 2021a). The large extension of the country, which covers different regions and climates, enables the production of different agricultural products, whether native, such as peanuts and cassava, or of foreign origin, such as corn, soybeans and wheat. Brazil's significant agricultural diversity enables the exploration of alternative sources in relation to soybeans and peas, which are widely studied in the development of plant-based products. Regarding the food industry, Brazil is currently the second largest exporter of industrialized foods: it exports to 190 countries (ABIA, 2021), behind only the United States; however, nationally, the vast majority of plant proteins marketed and used in the development of alternative products in relation to animal proteins still come from imports. In a research carried out by GFI Brazil and published at the end of 2021, the development of national ingredients was pointed out as the main strategy for advancing the plant-based market in Brazil (Ambiel *et al.*, 2021). Developing national agricultural chains for exploring alternative protein sources and developing products processed from these sources can contribute not only to strengthening Brazil's agriculture, but also to dynamize the food industry and the capacity to export processed products of interest in the international market.

Thus, the objective of this report is to map plant sources grown in Brazil, which have the potential to be protein ingredients for the plant-based industry, as well as to identify the plant-based raw materials, residues and/or by-products of the industries with the best technological and economic performance for application in plant-based products. This study aims to provide technical contributions for new research and the applications of new ingredients by industries.



Peanut

CHAPTER 1

Research methodology



Data collection



This report presents 18 plant sources grown in Brazil: rice, potatoes, three cultivars of beans (black, cowpea and mung), chickpeas, peanut, sesame seed, corn, cassava, wheat, barley, oat, rye, canola, lentil, sorghum and sunflower. A search of the scientific literature was carried out in different databases—including Scopus, Science Direct and Web of Science—in order to collect data regarding the extraction methods, composition, functional properties, allergenicity, nutritional

value and sensory characteristics of these 18 plant sources. The production, import and export data presented in this report were obtained from official national sources, such as the Brazilian Institute of Geography and Statistics (IBGE), Brazilian National Supply Company (CONAB), Ministry of Development, Industry and Foreign Trade (MDIC), Ministry of Agriculture, Livestock and Food Supply (MAPA) and associations of pre-processing of the different raw materials.

Criteria for technical and economic evaluation of the proteins



This report addresses 18 different plant protein sources, including cereals (rice, oat, rye, barley, corn, sorghum, and wheat), cruciferous (canola), legumes (peanuts, black beans, cowpea beans, mung beans, chickpeas, and lentils), tuberous roots (cassava), seeds (sesame and sunflower), and tubers (potatoes). To create a qualitative comparison between proteins, a color scale was established that classifies the technical and economic criteria of proteins as excellent, good, medium, low and/or poor, as shown in Table 1.

For the “protein concentration” criterion, we considered the protein concentration present in the whole plant source (raw material) before processing. Regarding the technological properties, this study evaluated solubility, emulsifying capacity (EC), emulsifying activity (EA), emulsifying

activity index (EAI), emulsion stability (ES), emulsion stability index (ESI), foaming properties, foaming capacity (FC) and foam stability (FS) and gelling agents, when available. However, the lack of standardization in the techniques used in the literature to evaluate emulsifying, foaming and gelling properties of plant proteins makes it difficult to compare the data presented in the different studies. Thus, considering that poorly soluble proteins are related to inferior technological properties (Silva *et al.*, 2021), insoluble proteins were classified as “poor/bad” following the technological classification presented in Table 1. On the other hand, proteins that were very soluble or that showed good technological properties (emulsifying, foaming and/or gelling properties) even at low concentrations were classified as “excellent.”

Table 1. Color scale that classifies the technical and economic criteria of the plant sources evaluated in this study as excellent, good, medium, low or poor/bad

Color ADD	Label	Protein Concentration in the Plant Source	PDCAAS ¹	Allergenicity	Flavor and Aroma	Solubility	Price (R\$/kg of protein)	Brazilian Production in 2021 (ton)
	Excellent	> 30%	> 0,8	Rare, only in specific cases	Pleasant/ Imperceptible	Excellent (even at low concentrations)	< 15	> 1.000.000
	Good	20% - 30%	0,60 - 0,79	Low allergenic potential	Very acceptable	Good	15 - 30	100.000 - 1.000.000
	Medium	10% - 20%	0,40 - 0,59	Moderate allergenicity	Acceptable with aftertaste	Average	30 - 45	10.000 - 100.000
	Low	5% - 10%	0,2 - 0,39	High allergenicity (low risk)	Little acceptable	Little soluble	45 - 55	1.000 - 10.000
	Poor/bad	< 5%	< 0,20	High allergenicity (severe risk)	Unpleasant	Insoluble	> 55	< 1.000

¹ "Protein digestibility-corrected amino acid score" refers to the digestibility of the protein corrected by the limiting amino acid.

For comparison purposes, the price per kilo of protein was determined considering the national average price per kilo of the plant source and the amount of protein present in it. However, it is noted that, in most of the sources evaluated in

the present study, the raw materials of interest for obtaining protein would be residues or by-products, which are often discarded or destined for animal feed.



Color accessibility

This publication uses, in some graphics, an accessible captioning system called [ColorADD®](#). This is a universal and inclusive language system that allows people with color blindness to identify colors whenever they are a factor in identification, orientation or choice. By using symbols, this system can simulate the additive properties of color and can represent primary and secondary colors and shades.

CHAPTER 2

Analysis of the results



Canola

Brazilian production of the plant sources evaluated in this study

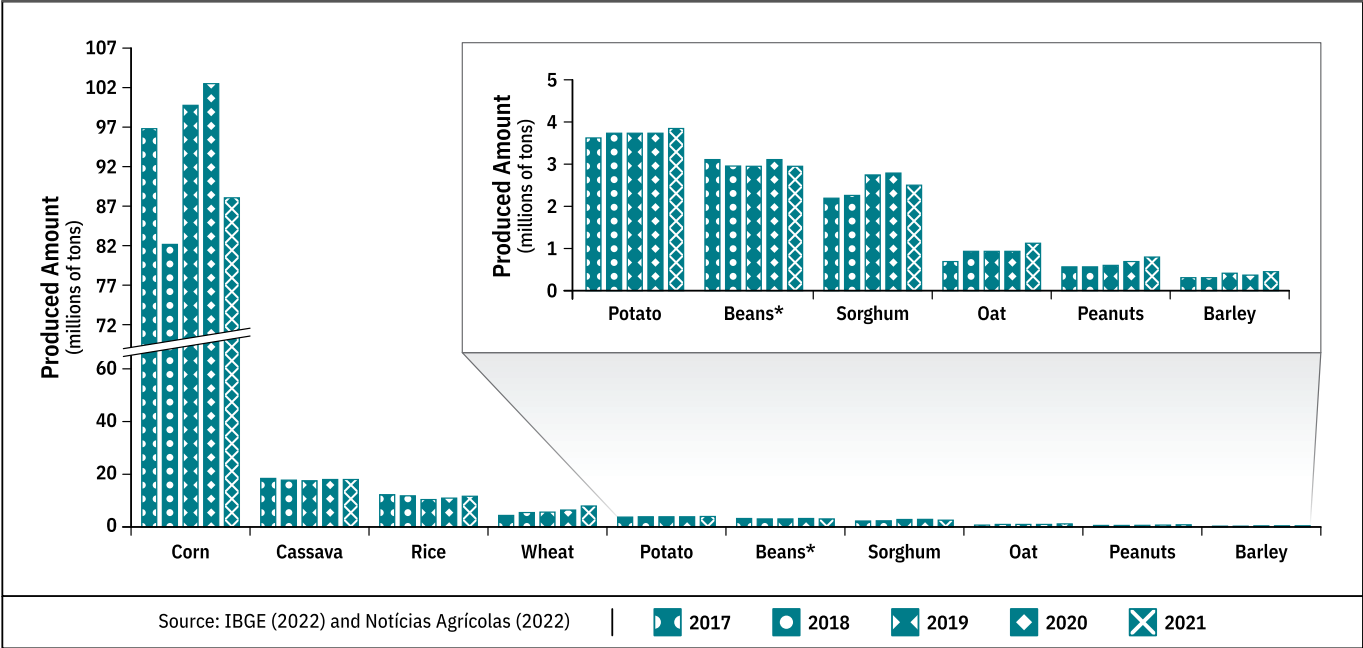


Rye

The Brazilian production of the evaluated products showed little fluctuation in the last five years, and growth trends, albeit discrete, were observed for oats, peanuts, barley and wheat (Appendix 1). Among the sources listed, corn is the product with

the highest production volume in Brazil (Figure 1). Nevertheless, Brazil also stands out in world corn production, being the third largest producer in the world, behind the United States and China (Embrapa, 2021a).

Figure 1. Ranking of the 10 plant-based raw materials most produced in Brazil among those evaluated in this study



* Corresponds to the total of all types of beans (pinto, black, black-eyed, kidney, white, etc.)

Evaluating the production of this plant by region, the Central-West region stands out. The state of Mato Grosso is responsible for more than 30% of national production, which, with the states of Paraná, Goiás and Mato Grosso do Sul, account for almost 70% of the total Brazilian corn production (Appendix 1).

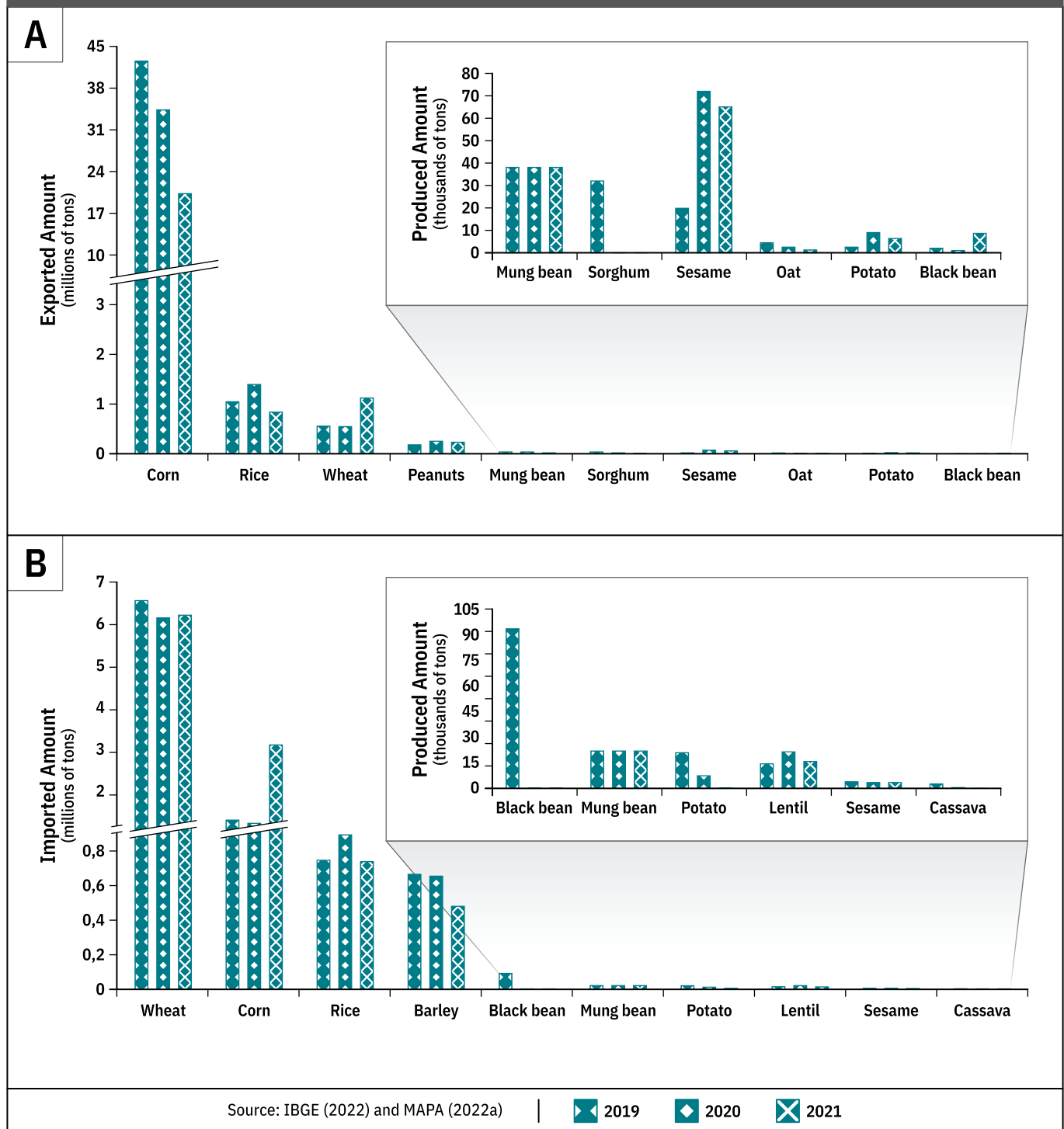
After corn, cassava is the second most produced plant crop in Brazil, followed by rice, wheat, potatoes, beans and sorghum (Figure 1). Brazil is among the world's 10 largest producers of rice and sorghum. Most of the national rice production is consumed nationally, and the South region is responsible for more than 80% of the national production of this cereal (Appendix 1). Sorghum, on the other hand, is still mostly intended for animal feed, being produced mainly in the states of Goiás and Minas Gerais. Globally, Brazil also stands out for its bean production, being among the three largest dry bean producers in the world (FAO, 2020). Among the bean cultivars evaluated with a focus on obtaining proteins, mung beans are the most mentioned in the literature (Scopus database); however, their production is still incipient in Brazil. Nationally, carioca beans are the most cultivated and consumed, but the cowpea and black cultivars stand out for their importance in the Northeast region and in the states of Rio de Janeiro and Rio Grande do Sul, respectively.

Chickpeas and lentils are legumes that stand out for their high protein content, originating from the Middle East and Southwest Asia, respectively. In Brazil, their production is not yet significant, and no official data are found. Some national cultivars were developed by Embrapa and by Campinas Agronomic Institute (IAC) (Artiaga *et al.*, 2015), and are legumes of interest for planting in the Brazilian Central-West region, as a planting option for the winter because it does not require large amounts of water.

Sesame is still not widely cultivated in Brazil, but production has increased in recent years, with the potential to be cultivated as a second crop (Embrapa, 2021a). Despite the small production in Brazil, rye and canola are possibilities for winter production (Embrapa, 2021b; EBC, 2018), being grown in cold countries and with mild temperatures, respectively.

Despite being a major producer of agricultural products with relevant exports, Brazil is still an importer of several raw materials. Figure 2 shows the 10 raw materials, among those evaluated in this study, most imported and exported in recent years. Although wheat is the fourth product with the highest national production among those evaluated in this study, Brazil is still very dependent on wheat imports, since it is the basis of numerous bakery products consumed in the daily lives of Brazilians. Other products, such as rice and corn, also appear in the list of products with significant imports and exports. Imports of these raw materials can be related to several factors, such as seasonality, crop of agricultural products, price in the international market and increase in exports.

Figure 2. Ranking of the 10 raw materials A) exported and B) imported, among those evaluated in this study



Properties of interest of plant sources to meet the market of plant-based products



In plant matrices, proteins perform storage or protective functions, and these functions are distinct from those performed in animal matrices. Thus, it is normal to observe differences in composition between animal and plant protein sources. Carbohydrates are the most abundant constituents in plant sources, however, among the plant species evaluated in this study, most have good protein levels in their source, such as canola, barley, sesame and sunflower, in addition to legumes, such as peanuts, chickpeas, different types of beans and lentils (Appendix 2).

From the perspective of the development of plant-based products, it is interesting to determine the levels of the various plant protein sources (legumes, cereals, tuberous roots, cruciferous, tubers and seeds) in terms of nutritional quality, allergenicity, techno-functionality and sensory contribution so replacements can be carried out without significant loss in product quality. Figure 3 presents a summary of the main characteristics to be considered in plant-based proteins. The plant sources evaluated in this study were classified based on the criteria presented in Table 1, and the complete data can be found in Appendices 2 and 3 of this document.

Figure 3. Summary of the properties of interest of plant sources to meet the plant-based market

Plant Source	Protein in the plant source ¹	PDCAAS ²	Allergenicity ²	Flavor and Aroma ²	Solubility ²	Price (R\$/kg of protein) ³	National Production in 2021 (ton) ⁴
Peanut							
Rice							
Oat							
Potato							
Canola							
Rye							
Barley							
Black bean							
Black-eyed bean							
Mung bean							
Chickpea							
Sesame							
Sunflower							
Lentil							
Cassava		*	*				
Corn							
Sorghum							
Wheat							
Color labels: Excellent Good Median Low Bad							

* Information not found

¹ Corresponds to the amount of protein (g/100g) present in the plant source. Source: NEPA (2004) and Philippi (2002) (Appendix 2).

² Source: Diverse scientific articles (Appendix 2).

³ Source: Conab [s.d].

⁴ Source: IBGE (2022).

Vegetable consumption is increasingly encouraged by physicians and dietary experts, since studies prove the efficiency of a plant-based diet in controlling metabolic syndrome (clinical expression of insulin resistance), decreasing hypertension, dyslipidemia, obesity and glycemic control (Hoffman & Falvo, 2004; Micha *et al.*, 2010; Rizzo *et al.*, 2011; Adeva-Andany *et al.*, 2022).

Despite these benefits, there are some factors that impact the replacement of animal proteins with plant-derived proteins, such as nutritional quality and allergenicity. In addition, replacing animal proteins with plant proteins in food formulations is challenging due to factors such as sensory aspects (taste, aroma and bitterness) and technological properties.

Next, we highlight the main factors related to nutritional, allergenicity, sensory aspects and technological properties, such as emulsifying and gelling properties, which are to be considered when using plant proteins in human food.



Nutritional quality



For the protein present in a food to be considered of high quality, it must be present in significant quantities and have the essential amino acids in the quantities necessary for the human organism, be fully or mostly digestible/absorbable and not present undesirable compounds, such as anti-nutritional factors. Chemical score (CS) is the measure of the amino acid composition of a protein compared to the composition of a reference standard amino acid. The *Protein Digestibility-Corrected Amino Acid Score* (PDCAAS) is related to the composition of the amino acids present in a protein source corrected for protein digestibility.

The amino acid composition of plant sources varies greatly according to cultivar, place of cultivation, soil, time of year and type of processing. Among the sources listed, potato proteins stand out for having a high PDCAAS value (Appendix 2), since they have an amino acid profile considered adequate, including lysine, methionine, threonine and tryptophan, in addition to having good digestibility (84%), being compared to egg and milk proteins (Hussain *et al.*, 2021). Canola also stands out for its PDCAAS value, having more than 400 mg of essential amino acids per gram of protein, in addition to containing a large amount of sulfurized amino acids (Chmielewska *et al.*, 2021).

In general, legumes are considered good sources of protein, being rich in lysine, while cereals are deficient in lysine, but rich in sulfur amino acids, being complementary when evaluating the nutritional composition. However, these sources have a limitation, when they are evaluated in relation to PDCAAS, mainly related to their low digestibility. The structural conformation of legume proteins, as well as the presence of protease inhibitors, can limit the action of digestive enzymes. In addition, legumes often have other compounds classified as antinutritional, such as phytates, saponins and/or polyphenols, which decrease and/or prevent nutrient absorption (Bessada *et al.*, 2019). It is worth noting that it is possible to reduce or even eliminate these antinutritional compounds with proper processing, such as hulling/peeling/skinning, immersion in water, hydrothermal treatments, germination and fermentation (Patterson *et al.*, 2017; Kumar *et al.*, 2022). Similarly, in cereals, the presence of antinutritional compounds, such as tannins, the amino acid sequence, and the secondary and tertiary structure of proteins are related to their low digestibility (Day, 2013).



Allergenicity



Often, plant-based products are used as alternatives by consumers that are allergic to animal-based products. However, with the increased consumption of protein products that are alternatives to animal-based products, allergies to different plant sources have been reported. Storage proteins, such as vicilins and legumins, present in oilseeds, and prolamins, present in legumes and cereals, are a large part of proteins related to allergenicity (Breiteneder & Radauer, 2004; Maruyama, 2021). Proteins linked to the lipid fraction of some plants, as well as peptides related to plant defense mechanisms, are also reported to be allergenic (Maruyama, 2021). Among the sources evaluated in the present study, peanuts,

black and mung beans, chickpeas, sesame seeds, lentils, corn and wheat were reported to have established allergenicity, that is, the amino acids (or their sequence) that can trigger allergic responses have been mapped (Appendix 2). Although the allergenicity of other sources, such as canola and rye, is not well defined, the similarity of protein fractions with those of other sources, such as mustard and wheat, respectively, indicates that caution should be taken in the consumption of these sources. A low allergenic potential was reported for rice, oats, potatoes, sunflower and sorghum, and some specific cases of allergy were identified (Appendix 2).

Sensory aspects



The taste of protein ingredients depends on the source, extraction methods and various treatments in the course of obtaining them. The taste given by these ingredients depends on the product to which it is added and the formulation, since they can interact with the other ingredients of the formulation. In addition, it should be considered that the sensory information reported in the literature for proteins from different plant sources depends on the sensory panel used: trained or untrained tasters (Clapperton & Piggott, 1979; Fiorentini *et al.*, 2020; Losó *et al.*, 2012). In general, for legumes, the “*beany*” flavor is frequently described (Chang *et al.*, 2019; Xu *et al.*, 2019; Yang *et al.*, 2021). This term refers to the bean-like

flavor and is mainly related to the degradation of lipids and/or amino acids (Murat *et al.*, 2013). For cereals, astringency and bitterness are frequently reported (Huang & Zayas, 1991; Holtekjølén *et al.*, 2008; Kaleda *et al.*, 2021), being related to the presence of phenolics. In this category, the highlights are rice, which was reported as having a pleasant (Nadathur & Carolan, 2016), but granular taste (Hu *et al.*, 2019), and sorghum, with a neutral taste (Pereira *et al.*, 2017). Among the sources evaluated, cowpea, sesame, sunflower and wheat have flavor without or with less impact of sensory characteristics considered unpleasant (Appendix 2).

Technological properties



In addition to the nutritional aspect, proteins can meet various technological purposes, the most common being the function of conferring texture and/or stabilizing food formulations. The molecular basis for the functionality of proteins is related to their structure, conformation and their ability to interact with other ingredients. Due to the presence of hydrophilic and hydrophobic groups in the same molecule, proteins are able to retain water in their structure, form foams, gels and can bind to fat. This may give excellent interfacial and emulsifying properties to proteins (Lam & Nickerson, 2013). In addition, proteins can be used as foaming agents, films and as thickening agents, contributing to the quality and sensory attributes of food (Miquelim *et al.*, 2010; Joshi *et al.*, 2012; Evangelho *et al.*, 2017, Guerrero *et al.*, 2010).

Regarding the technological functional properties, proteins obtained from plant sources usually present lower solubility when compared to proteins of animal origin (Silva *et al.*, 2021). However, protein conformation is quite variable and depends not only on protein source or factors intrinsic to the molecule (such as amino acid composition and sequence, secondary and tertiary structures, presence of sulfide bonds and others), but is deeply affected by extraction and recovery methods and environmental factors, such as medium pH, heat treatment (Coelho & Salas-Mellado, 2018). Alkaline extraction is the simplest process for extracting protein from plant sources, in addition to having low cost and being highly scalable. In this process, to obtain high extraction yields, high pH values (> 10) and temperature values (> 70°C)

are often used, which can impair the technological properties of the proteins obtained (Silva *et al.*, 2021). Accordingly, processes with less degradation in technological properties, such as dry fractionation, ultrasound-assisted extraction and the use of enzymes, have been sought (Pojić *et al.*, 2018). The use of enzymes to obtain protein concentrates is very attractive because it uses mild process conditions (pH 7-8, > 50 °C) without generating highly acidic or alkaline solutions, thus having a low polluting effect. In addition, it is possible to achieve high levels of separation and purity through this technique (Yu *et al.*, 2020).

In general, extraction processes have a direct influence on the secondary and tertiary structures of proteins. Particularly, in processes using proteases, even the amino acid sequence (primary structure) can be affected. Thus, it is very difficult to reach a conclusion about the functionality of plant proteins without considering the process used to obtain them. In addition, the lack of standardization of the methodologies used to quantify the technofunctional properties of plant proteins makes it difficult to compare the results obtained in different studies. In order to minimize these divergences, Embrapa Agroindústria de Alimentos prepared a guide for the technological and functional characterization of protein ingredients for the market of plant-based products (Silva *et al.*, 2022).

Considering the importance of knowing the technological properties of proteins from different plant sources for use as ingredients in plant-based formulations, the results of this research showed that, in general, proteins obtained from cereals have better emulsifying properties than those obtained from legumes (Appendix 3), while most studies involving gelling properties of plant

proteins evaluate legume proteins. Commercially, pea protein, a legume rich in globulins, is widely used in the plant-based industry in sausage and hamburger formulations, mainly due to its gelling properties; thus, it is believed that proteins from other legumes may have similar characteristics. In turn, proteins from cereals, such as corn and sorghum, have significant fractions of prolamines (protein fractions with a strong hydrophobic character), being applied in sauces and “vegetable milks” due to their emulsifying properties.



CHAPTER 3

Residues and by-products from plant sources destined to obtain plant proteins



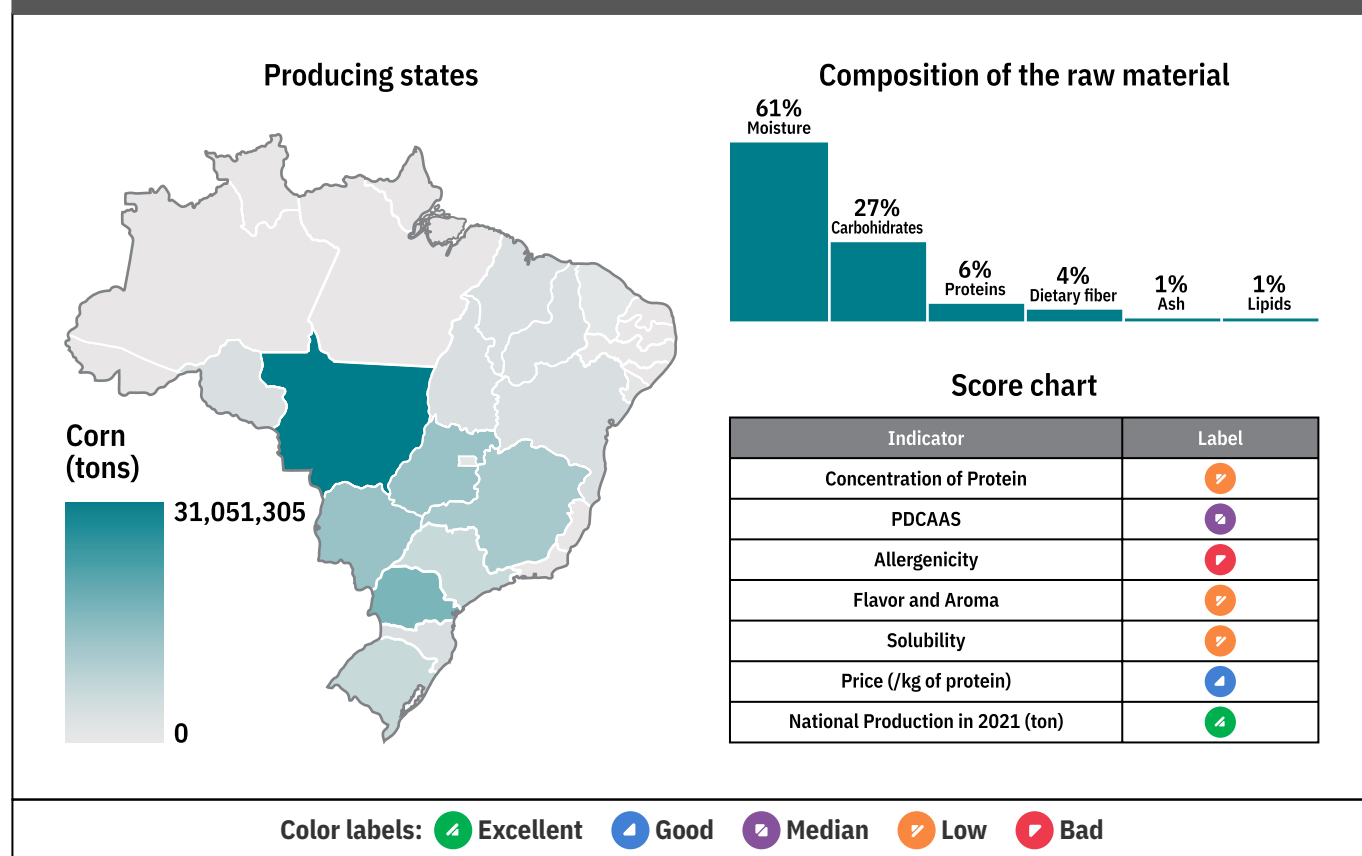
Corn

Proteins are valuable constituents of plant raw materials that are currently still underutilized, and are often destined for animal feed after the processing of various foods, such as to obtain oil, flour and starch (Appendix 4). A variety of proteins can be obtained from the residues or by-products of food processing, optimizing the use of resources and contributing to a more sustainable agriculture (Waglay *et al.*, 2014; Colantuono, 2018; Los *et al.*, 2020), as in the case of oilseeds—peanuts, canola, sesame, sunflower, etc.—which, after removal

of the oil, result in cakes with a protein content ranging between 37% and 63% (on a dry basis) and of starch production industries that use plant sources, such as corn, cassava and potatoes.

Corn should be noted because it is the crop with the highest production volume in Brazil, being produced in all regions of the country, but predominantly in the states of the Central-West region, such as Mato Grosso, Goiás and Mato Grosso do Sul (Figure 4).

Figure 4. General information on corn as a source of plant protein: composition of the raw material, national production and classification according to the color criteria shown in Table 1



Corn can be processed by dry or wet methods to obtain different products such as flours (cornmeal) (Figure 5), oil (Figure 6) and ethanol (Figure 7). In addition to the processes presented, it is also

possible to produce other products, such as corn starch, obtained through the wet milling process, after the separation of gluten, while flakes can be obtained from the hydration of the flour.

Figure 5. Frequent products and residues from dry corn processing to obtain flours and derivatives

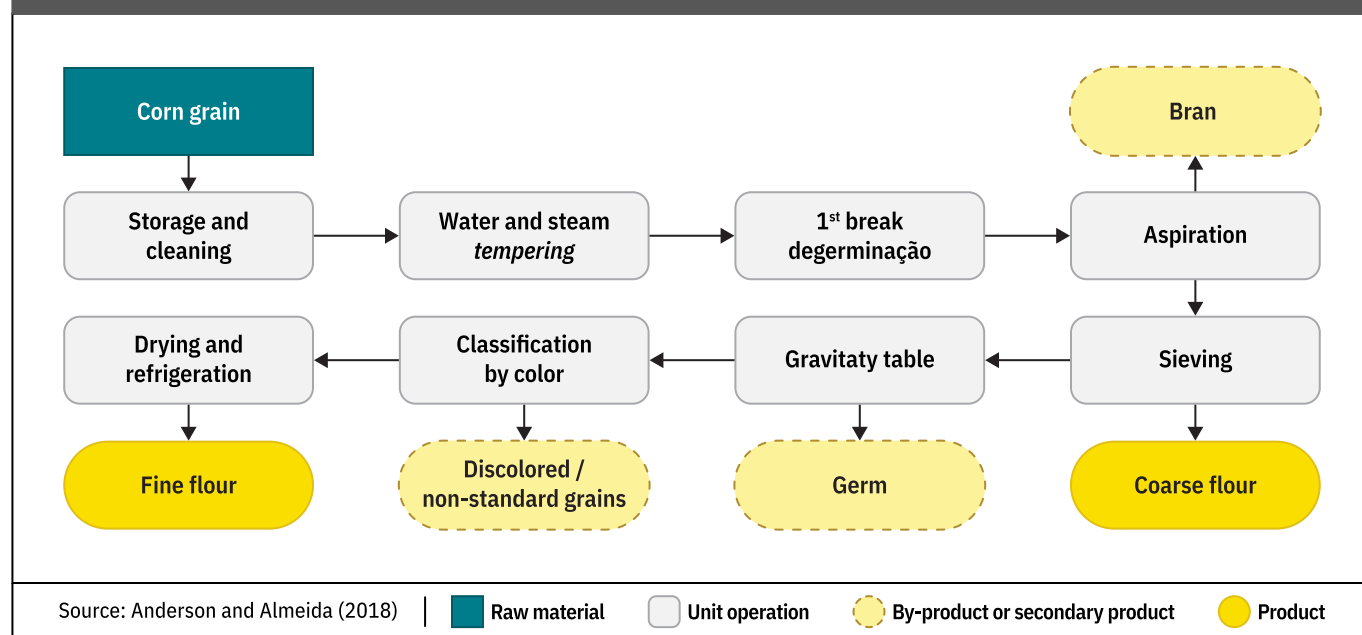
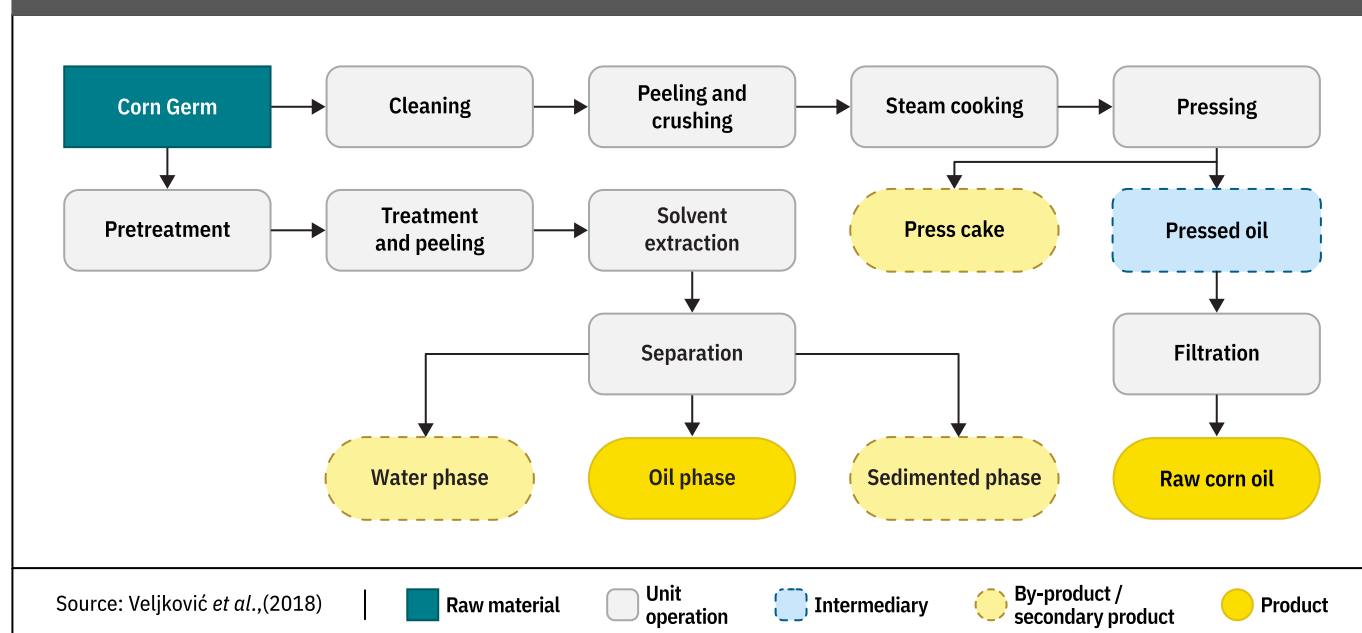
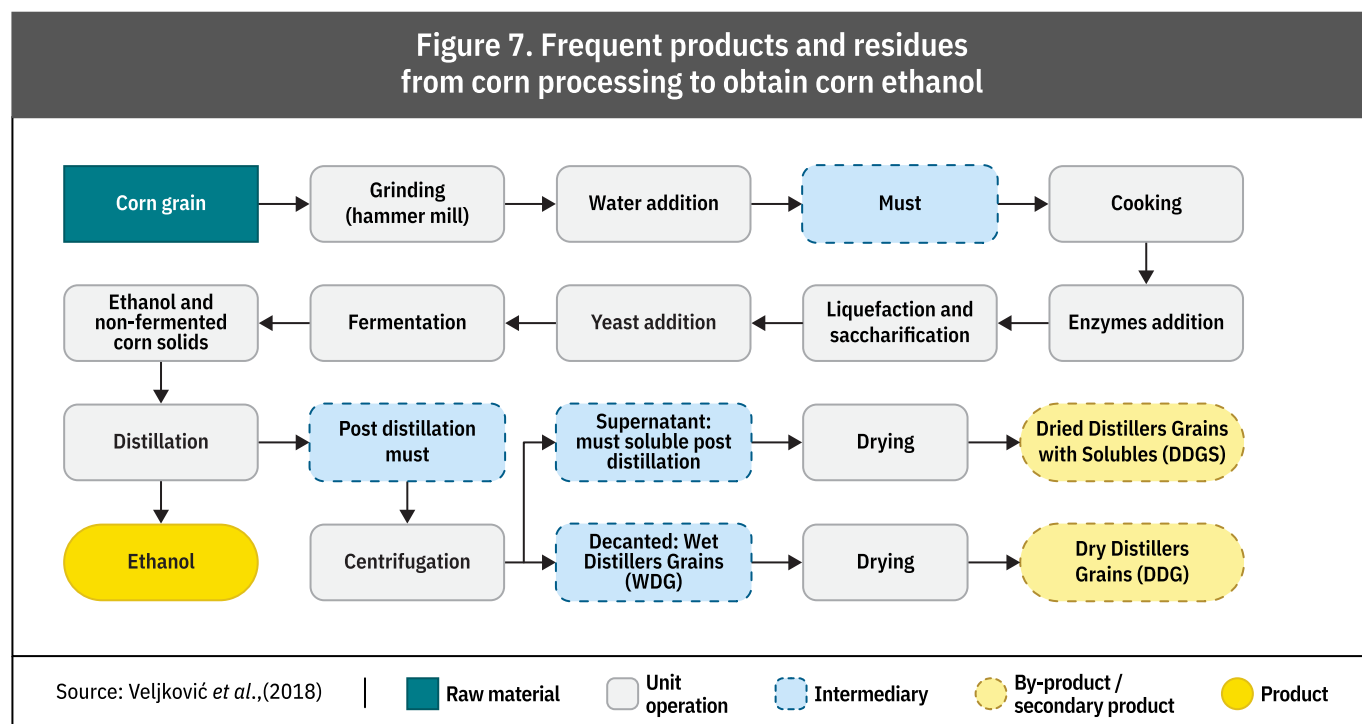


Figure 6. Frequent products and residues from corn processing to obtain corn oil



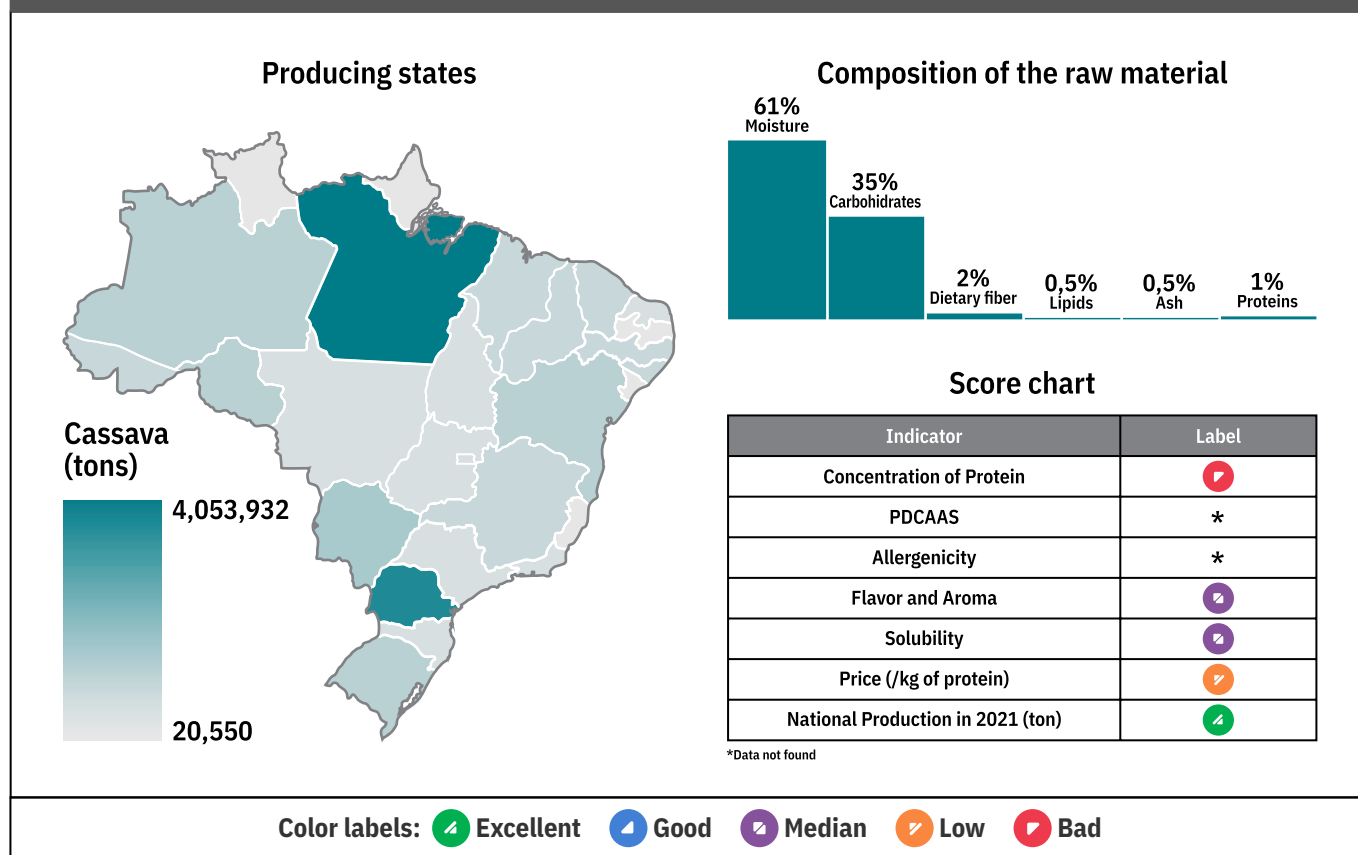
In the domestic market, corn has been destined for the production of ethanol (Figure 7), representing, in 2022, 13% of the total national biofuel (REDAÇÃO AGRISHOW, 2022). In the previous year, this amount was equal to 8% (MAPA, 2021), indicating a rising market. This process generates, as a by-product, *Dried Distillers Grains* (DDG), which is the dry corn grain after the fermentation and distillation process, which, according to producers, has 26% to 30% protein.



Corn proteins contain a considerable amount of zeins, prolamin-type proteins with good interfacial properties that have been the subject of studies related to coating for foods, such as cheeses, and to the production of biodegradable plastic (Fontes, 2022), mainly due to their low solubility in water and due to resistance to bacterial attack. Given their inherent hydrophobicity and biodegradability, zein nanoparticles have been successfully applied as carriers for controlled release of hydrophobic drugs and as biomaterial for the development of colloidal release systems. Regarding plant-based products, corn proteins can be used as taste-masking agents (Flozein Products, 2021).

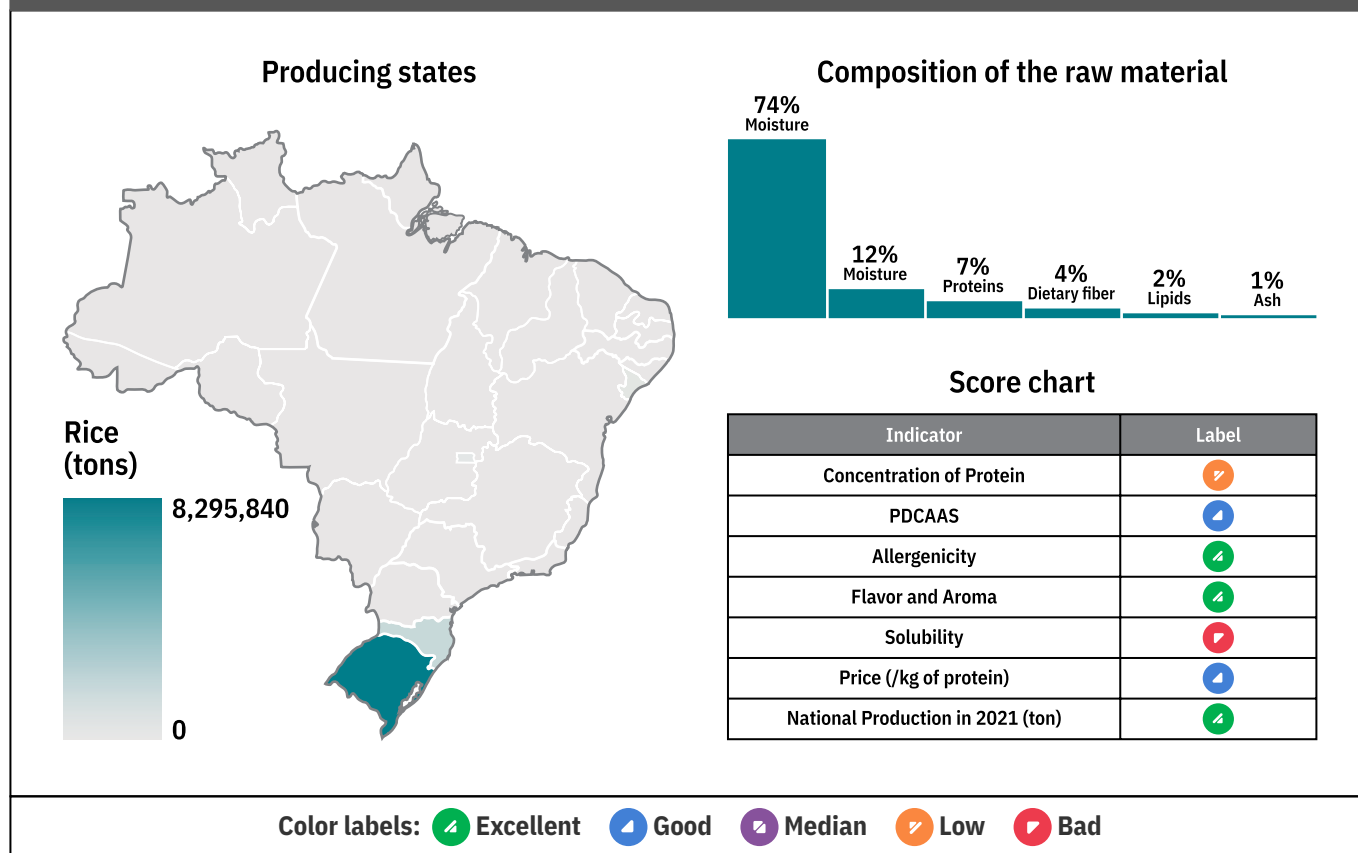
Another raw material that should be noted because it is produced in all regions of Brazil is cassava (Figure 8). Although there is little protein concentration in this root (~ 1%), the great availability of this raw material makes it an interesting source of protein for human consumption.

Figure 8. General information on cassava as a source of plant protein: composition of the raw material, national production and classification according to the color criteria shown in Table 1



The main products obtained from cassava are: flours, starch and tapioca powder. Similarly to proteins obtained from the coagulation and precipitation of the protein present in “potato juice” (by-product of starch production) (Pęksa & Miedzianka, 2021), it is believed that cassava root protein may have industrial potential for adding value as the by-product of the cassava starch industry. In addition, planting cassava results in a large amount of leaves, peels and stems. During cassava cultivation, approximately 10 tons of dried leaves are produced per hectare, which are commonly left in the field, with no industrial destination (Oresegun *et al.*, 2016). These leaves have great potential as a protein source, since they have more than 20% dry basis of protein.

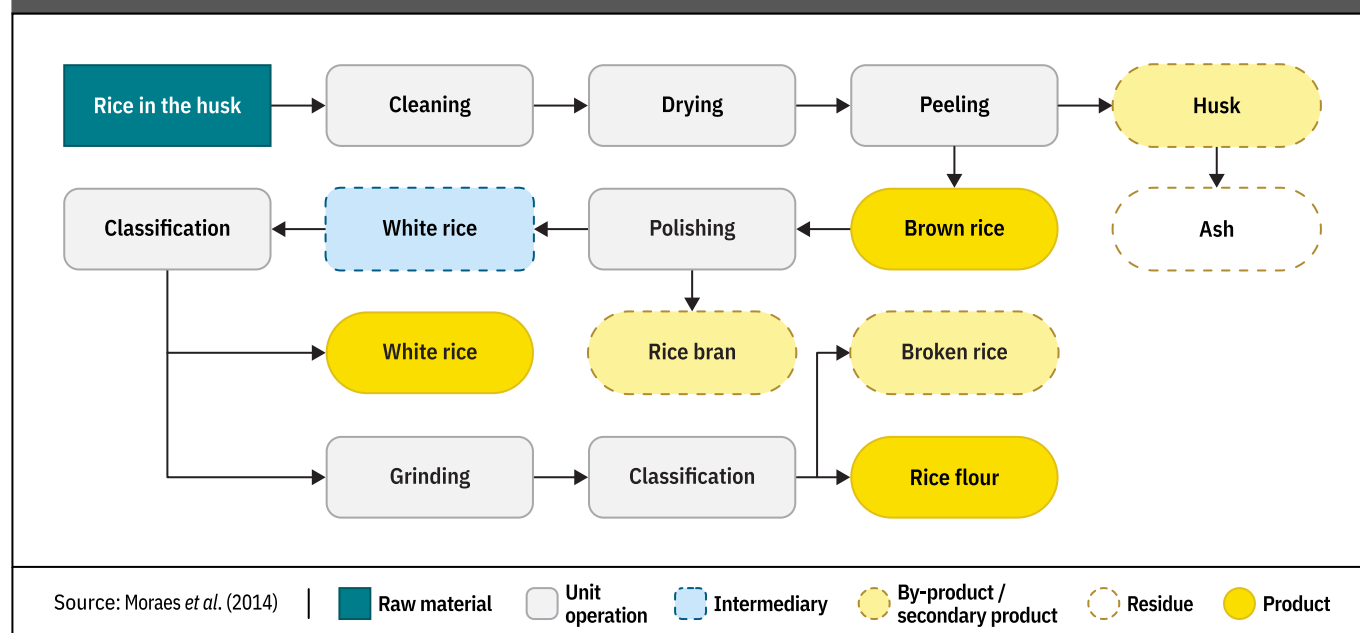
Figure 9. General information on rice as a source of plant protein: composition of the raw material, national production and classification according to the color criteria shown in Table 1



Rice is widely consumed throughout the Brazilian territory because it is a staple ingredient of Brazilian cuisine. However, rice production is concentrated in the South region, with the state of Rio Grande do Sul accounting for more than 80% of national production (Figure 9).

Rice is consumed mainly in the form of white or parboiled grains, in which the hull is removed and the grain is polished (Fabian & Ju, 2011). The main by-product of rice processing is broken grains that are destined for the production of white rice flour for gluten-free formulations, while its main residue is rice bran, consisting mainly of rice hulls and dust from the polishing of the grain (Figure 10).

Figure 10. Products and residues from rice processing



Rice bran is rich in protein, lipids, dietary fiber, vitamins and minerals. The composition of rice bran is 15%-20% lipids, 34%-52% carbohydrates, 7%-11% fiber, 6%-10% ash, 8%-12% moisture and 11%-16% highly nutritious proteins (Amagliani *et al.*, 2017). The recovery of rice bran proteins to obtain protein concentrates is highly recommended, since their proteins have great potential to be used as functional food ingredients and nutritional supplements (Fabian & Ju, 2011). Since it is hypoallergenic, it is a suitable ingredient for infant food formulations and gluten-restricted diets (Phimolsiripol *et al.*, Schoenlechner, 2012; Hirano *et al.*, 2016). In addition, rice proteins have been reported to have high antioxidant capacity (Gomes & Kurosawa, 2020).

Efforts are made to promote this ingredient. However, more and more companies focus on the extraction and commercial availability of rice bran protein concentrates (Amagliani *et al.*, 2017; Silva *et al.*, 2021). The major challenge in the application

of rice bran proteins is the low solubility and strong aggregation and/or extensive cross-linking of disulfide bonds (Ju *et al.*, 2001). In addition, rice bran has a high content of phytates (1.7%) and fibers (12%) (Juliano, 1985), components that can bind strongly to proteins, making it difficult to obtain proteins with high purity.

Potato is also a vegetable widely produced in Brazil. This raw material is the fifth most produced variety in Brazil with crops in the South, Southeast, Northeast and Central-West states (Figure 11), totaling a national production volume of more than 3.7 million tons.

Potatoes are marketed fresh or industrialized in different forms, such as chips, ready-to-fry preparations, puree formulations and as ingredients in recipes. Figure 12 shows one of the main potato processing methods for starch production. The recovery of proteins from potato water is of particular interest, as the production of starch from 1000 kg of potato releases 5m³ to 12m³ of wastewater, which contains 30% to 41% protein on a dry base (Waglay *et al.*, 2014).

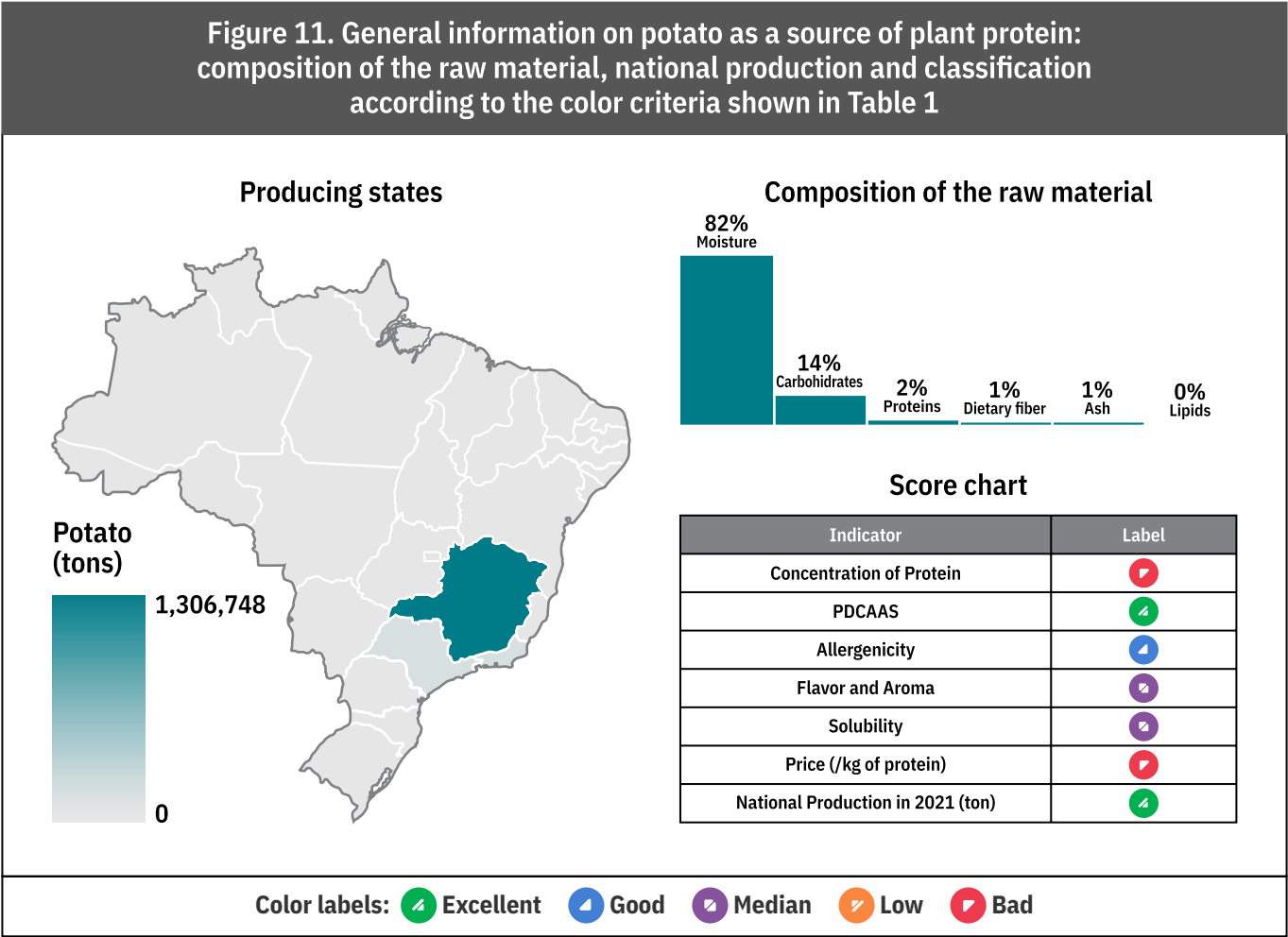
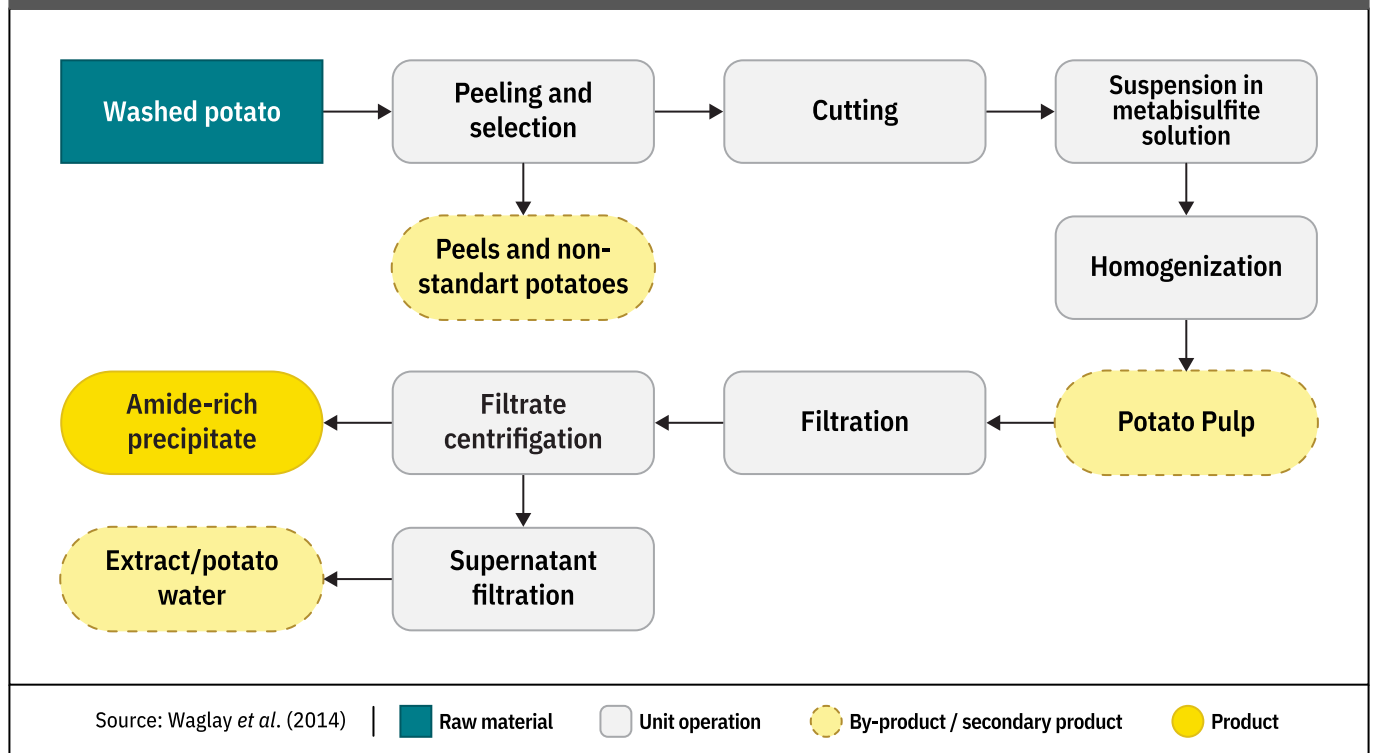


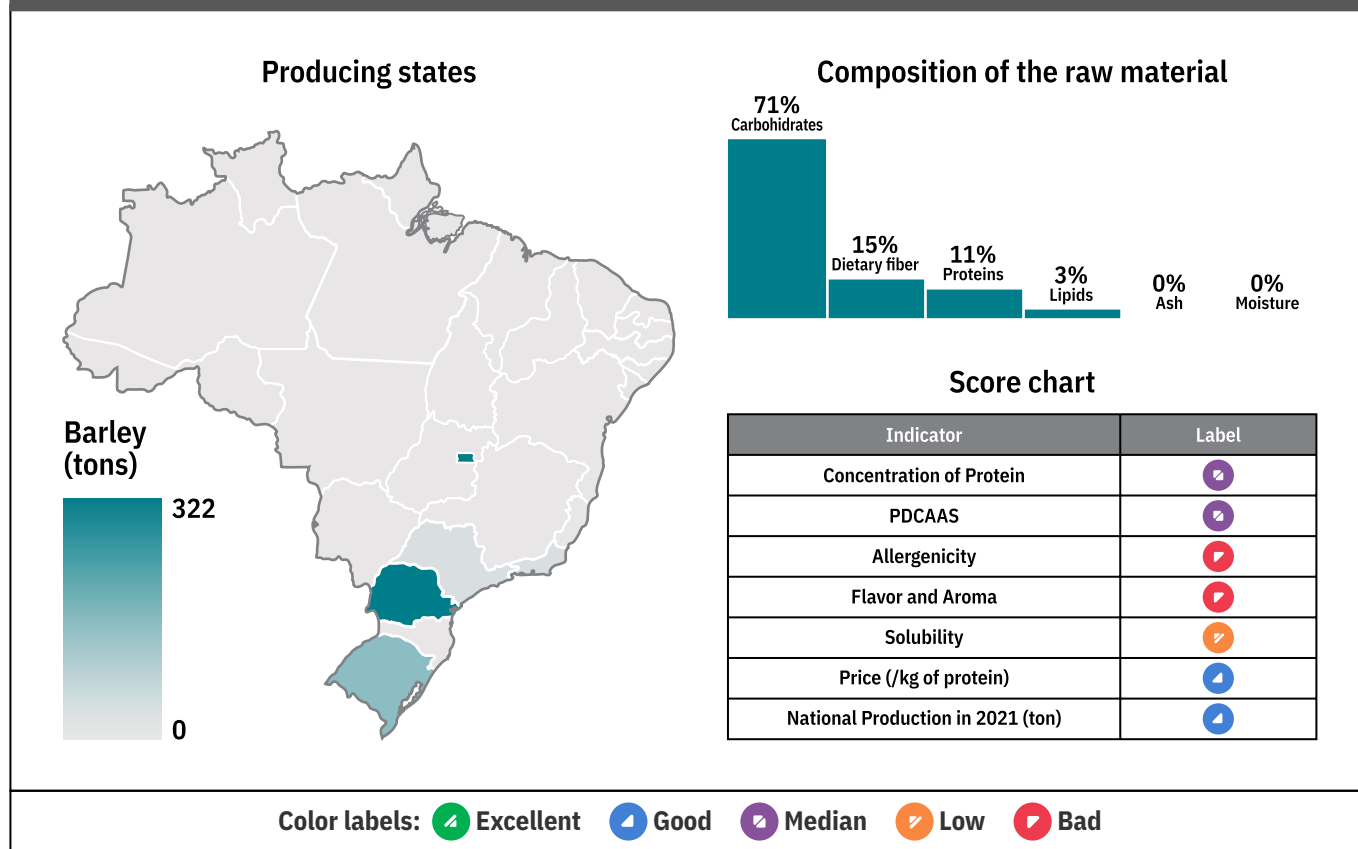
Figure 12. Products and residues from potato processing to obtain potato starch



Compared to proteins from other plant sources, potato proteins are considered to be of excellent nutritional quality, since they contain a high proportion of lysine, an essential amino acid that is often lacking in plant foods (Peksa *et al.*, 2009) and, like rice bran, they have low or no reported allergenicity (Majamaa *et al.*, 2001), being a particularly interesting ingredient for foods intended for children and athletes. Potato proteins are commonly composed of two major fractions, patatin (up to 40% of the total weight of protein), whose molecular mass ranges from 39 kDa to 45 kDa and protease inhibitors (~ 50%), whose molecular weight ranges from 4 kDa to 25 kDa, in addition to other high molecular weight proteins (~ 10%) (Bártová & Bártá, 2009).

Industrially, potato extract protein recovery is accomplished by a combination of thermal coagulation and acid precipitation (Cheng *et al.*, 2010, Miedzianka *et al.*, 2012). Although thermal/acid precipitation results in a high protein recovery yield, it often leads to complete loss of protein functionality, which limits its application in human food (Cheng *et al.*, 2010, Miedzianka *et al.*, 2012).

Figure 13. General information on barley as a source of plant protein: composition of the raw material, national production and classification according to the color criteria shown in Table 1

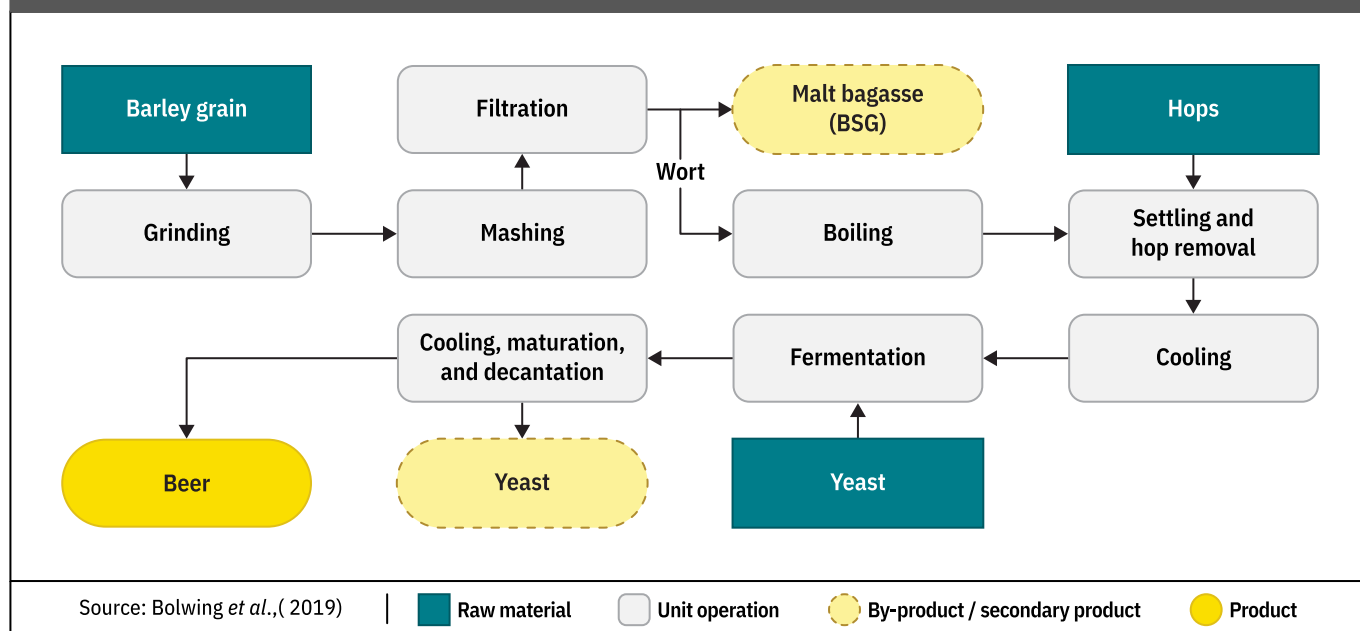


In Brazil, the South and Southeast are the only regions that produce barley, with the state of Paraná being the leading producer (Figure 13). More than 450 tons were produced in 2021 (Figure 1). Such amount, in addition to the volume imported by Brazil (third place in volume of imports) (Figure 2), show the importance of this plant species among the raw materials studied in this report.

The high demand for barley is due to the fact that Brazil is a major beer producer (the 3rd in the world) and barley is the main raw material for the production of national beer (Embrapa, [s.d]). Brewer's spent grain (BSG) is the most abundant by-product generated in the beer production

process, with a generation on dry bases of 15 kg-20 kg per 100 l of beer produced (Wen *et al.*, 2019) (Figure 14). Data from the latest FAO report indicated that more than 3.7 billion tons of BSG were produced worldwide last year, while, in Europe, annual BSG production was estimated at ~ 1 billion tons (FAOSTAT, 2022). BSG is a fiber-rich material and its protein content ranges from 11% to 30% on a dry base (Wen *et al.*, 2019).

Figure 14. Products and residues from beer processing



Studies have shown that BSG proteins and hydrolysates have promising potential as functional food ingredients for health (Cermeño *et al.*, 2019; Vieira *et al.*, 2017). However, there are still few studies dedicated to the technological properties of the protein fraction of this raw material. It is believed that the difficulty in separating the protein part from the fibrous part of BSG and the low solubility of its fractions are the main challenges.



CHAPTER 4

Conclusions and prospects



Sorghum



Brazil has a wide variety of plant sources with potential for use as raw materials to obtain proteins. Corn, cassava, rice and potatoes should be highlighted, as these varieties are the most significant for the national economy due to the volume produced. Obtaining proteins from by-products and residues from the production chain of other products—such as ethanol, oil, starch and/or flour—contributes to a more sustainable system with less impact on the environment.

The deliberate use of plant-based proteins to replace animal proteins is still a challenge to be overcome mainly due to the taste, nutritional quality and technological properties of these proteins. Methods for extraction and combinations of different sources enable overcoming the challenges of the plant-based market.

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Appendices



Appendix 1 - Production volume of plant-based raw materials (ton) per federative unit (FU) of Brazil between 2019 and 2021

FU	Year	Peanut	Rice	Oats	Potato	Canola	Rye	Barley	Beans¹	*Black beans	*Cowpea beans	Sesame seeds	Sunflower	Cassava	Corn	Sorghum	Wheat
AC	2019	122	4,540	0	0	0	0	0	3,025	**	**	0	0	628,422	75,412	0	0
	2020	120	4,626	0	0	0	0	0	2,941	0	3,800	0	0	586,202	79,067	0	0
	2021	122	4,473	0	0	0	0	0	2,855	0	3,600	0	0	532,059	105,885	0	0
AL	2019	5,316	20,177	0	0	0	0	0	9,802	**	**	0	0	384,152	54,122	1,155	0
	2020	5,276	16,072	0	25	0	0	0	10,735	0	3,800	0	0	532,553	61,097	912	0
	2021	5,138	24,436	0	47	0	0	0	13,403	0	3,900	0	0	508,652	81,642	912	0
AP	2019	0	820	0	0	0	0	0	805	**	**	0	0	108,530	1,138	0	0
	2020	0	835	0	0	0	0	0	812	0	0	0	0	112,244	1,150	0	0
	2021	0	815	0	0	0	0	0	3,820	0	0	0	0	113,506	1,186	0	0
AM	2019	0	1,100	0	0	0	0	0	1,432	**	**	0	0	876,452	6,683	0	0
	2020	0	1,680	0	0	0	0	0	1,508	0	2,500	0	0	890,124	6,824	0	0
	2021	0	1,147	0	0	0	0	0	1,169	0	2,500	0	0	720,488	6,363	0	0
BA	2019	5,918	471	0	300,257	0	0	0	179,570	**	**	0	0	648,444	1,886,858	106,056	14,600
	2020	5,297	744	0	390,789	0	0	0	194,060	0	82,500	0	0	706,887	2,646,955	127,809	17,000
	2021	5,026	732	0	393,914	0	0	0	164,055	0	100,200	0	0	766,772	2,450,153	123,788	17,737
CE	2019	428	15,877	0	0	0	0	0	110,067	**	**	0	0	642,188	423,601	1,120	0
	2020	602	16,394	0	0	0	0	0	124,746	0	110,800	0	0	641,142	633,317	3,500	0
	2021	531	19,362	0	0	0	0	0	110,981	0	118,900	0	0	560,249	414,411	10,440	0
DF	2019	0	0	0	4,250	0	0	315	33,609	**	**	0	1,680	20,550	499,800	24,000	6,870
	2020	0	0	0	4,250	0	0	315	46,484	2,700	100	0	1,680	20,550	486,138	48,000	11,400
	2021	0	0	0	4,250	0	0	315	27,240	2,000	100	0	1,050	20,550	324,000	35,100	10,500
ES	2019	3	353	0	5,746	0	0	0	9,766	**	**	0	0	118,470	37,533	0	0
	2020	3	345	0	5,937	0	0	0	9,421	0	0	0	0	127,529	39,422	0	0
	2021	3	371	0	7,118	0	0	0	9,920	0	0	0	0	126,760	41,670	0	0
GO	2019	0	165,383	0	218,084	0	0	0	341,045	**	**	2,000	44,477	184,776	11,979,032	1,110,706	67,953
	2020	0	144,419	0	183,104	0	0	0	351,454	0	15,600	2,000	38,320	182,254	11,838,775	1,173,014	110,884
	2021	0	124,510	0	177,618	0	0	0	340,325	0	15,900	1,500	36,661	180,820	10,750,433	1,140,088	84,035
MA	2019	189	155,552	0	0	0	0	0	31,047	**	**	0	0	464,148	1,803,512	21,882	0
	2020	260	154,856	0	0	0	0	0	27,260	0	26,900	0	0	434,344	2,177,432	20,274	0
	2021	256	168,014	0	0	0	0	0	26,707	0	37,600	0	0	440,241	2,267,556	21,081	0
MT	2019	1,993	444,634	0	0	0	0	0	278,957	**	**	94,000	75,706	287,237	31,504,274	136,840	0
	2020	2,718	378,442	0	0	0	0	0	335,345	0	156,300	55,000	30,296	270,376	33,650,671	162,006	0
	2021	2,040	392,293	0	0	0	0	0	355,501	0	170,200	58,400	17,264	258,812	31,051,305	141,198	288
MS	2019	6,135	53,825	38,582	0	0	0	0	31,323	**	**	0	35	807,343	9,963,206	35,102	43,120
	2020	7,875	51,298	46,510	0	0	0	0	34,262	0	0	0	0	906,533	10,696,608	32,144	73,198
	2021	8,171	67,115	20,847	0	0	0	0	12,957	0	0	0	0	997,672	5,418,082	72,195	21,470
MG	2019	7,314	7,438	16,600	1,199,571	0	0	2,095	531,604	**	**	0	4,073	525,053	7,468,417	843,932	242,367
	2020	12,429	7,936	4,247	1,267,243	0	0	0	553,065	12,100	9,300	0	2,569	518,141	7,689,309	862,632	296,770
	2021	21,290	8,302	3,164	1,306,748	0	0	0	536,826	12,800	9,300	0	3,544	547,267	6,788,836	565,017	207,262

FU	Year	Peanut	Rice	Oats	Potato	Canola	Rye	Barley	Beans¹	*Black beans	*Cowpea beans	Sesame seeds	Sunflower	Cassava	Corn	Sorghum	Wheat
PA	2019	106	94,508	0	0	0	0	0	20,883	**	**	0	0	3,711,214	827,720	39,771	0
	2020	104	112,470	0	0	0	0	0	19,891	0	18,100	0	0	3,813,369	893,065	47,056	0
	2021	104	113,734	0	0	0	0	0	20,759	0	18,300	0	0	4,053,932	1,122,835	54,785	0
PB	2019	368	2,073	0	15	0	0	0	21,143	**	**	0	0	143,990	39,414	0	0
	2020	696	2,772	0	158	0	0	0	36,103	600	19,300	0	0	141,910	77,585	0	0
	2021	284	3,651	0	0	0	0	0	21,366	1,200	26,700	0	0	131,811	48,172	0	0
PR	2019	5,029	138,446	191,861	763,181	1,000	7,098	247,733	635,728	**	**	0	14	3,270,654	16,519,549	370	2,408,810
	2020	5,603	150,967	192,962	744,147	1,000	6,974	278,661	624,587	336,400	0	0	40	3,474,295	15,786,934	7,207	3,130,147
	2021	4,857	152,888	211,891	769,378	1,500	7,130	321,516	631,295	373,800	0	0	77	3,404,917	10,528,860	4,723	3,231,985
PE	2019	93	3,022	0	0	0	0	0	47,498	**	**	0	0	400,096	30,489	2,264	0
	2020	73	3,622	0	0	0	0	0	64,684	11,200	32,100	0	0	433,938	92,173	2,441	0
	2021	78	5,772	0	0	0	0	0	71,155	9,400	41,000	0	0	421,311	66,731	49	0
PI	2019	50	78,444	0	0	0	0	0	78,642	**	**	0	0	365,109	1,835,613	62,810	0
	2020	52	103,759	0	0	0	0	0	82,984	0	59,400	0	0	444,433	2,199,753	37,836	0
	2021	31	97,188	0	0	0	0	0	53,280	0	79,000	0	0	405,718	2,145,035	21,261	0
RJ	2019	0	627	0	303	0	0	0	2,838	**	**	0	0	216,496	7,967	0	0
	2020	0	114	0	306	0	0	0	2,036	1,400	0	0	0	151,558	7,866	0	0
	2021	0	785	0	198	0	0	0	1,144	1,200	0	0	0	158,860	8,557	0	0
RN	2019	0	3,053	0	0	0	0	0	26,390	**	**	0	0	219,150	30,687	298	0
	2020	0	3,154	0	0	0	0	0	23,996	0	17,800	0	0	211,288	31,252	298	0
	2021	0	3,241	0	0	0	0	0	10,396	0	19,500	0	0	230,030	13,345	221	0
RS	2019	3,723	7,172,101	642,211	452,332	42,000	3,222	140,694	91,774	**	**	0	3,813	886,955	5,735,186	11,789	2,287,720
	2020	2,791	7,753,663	609,277	363,873	60,000	2,289	93,057	81,146	54,100	0	0	4,079	788,415	4,211,208	6,815	2,104,160
	2021	2,996	8,295,840	803,552	510,858	74,100	3,437	110,929	89,767	58,800	0	0	4,618	842,953	4,389,617	6,263	3,547,866
RO	2019	194	123,940	0	0	0	0	0	16,036	**	**	0	0	521,258	1,004,717	0	0
	2020	169	112,848	0	0	0	0	0	15,660	0	0	0	0	519,582	1,036,905	0	0
	2021	164	114,942	0	0	0	0	0	11,968	0	0	0	0	842,953	1,355,590	0	0
RR	2019	0	97,655	0	0	0	0	0	970	**	**	0	0	82,792	50,390	0	0
	2020	0	71,054	0	0	0	0	0	1,072	0	2,100	0	0	85,520	79,128	0	0
	2021	0	83,830	0	0	0	0	0	1,514	0	2,900	0	0	58,210	114,159	0	0
SC	2019	129	1,062,159	29,335	117,483	0	0	2,388	108,976	**	**	0	0	348,412	2,767,609	0	146,039
	2020	85	1,215,651	24,635	107,647	0	0	1,080	98,713	57,200	0	0	0	336,523	2,701,935	0	171,727
	2021	15	1,195,939	26,891	101,125	0	0	1,935	98,334	76,900	0	0	0	341,727	2,006,694	0	317,969
SP	2019	541,158	57,374	11,692	650,522	0	0	12,390	264,859	**	**	0	3,160	1,358,067	4,800,956	227,037	373,336
	2020	603,697	59,038	20,646	700,300	0	0	14,033	256,170	0	0	0	2,696	1,504,057	4,503,594	183,235	432,674
	2021	741,538	58,791	20,728	582,210	0	0	18,132	229,660	0	0	0	1,640	1,456,284	3,709,901	233,679	435,413
SE	2019	1,339	28,159	0	0	0	0	0	3,836	**	**	0	0	147,465	687,221	0	0
	2020	1,506	34,153	0	0	0	0	0	3,776	0	0	0	0	136,438	904,506	0	0
	2021	1,519	51,227	0	0	0	0	0	1,828	0	0	0	0	135,067	741,765	0	0
TO	2019	966	636,908	0	0	0	0	0	26,450	**	**	0	0	225,763	1,085,843	47,113	0
	2020	1,504	690,099	0	0	0	0	0	32,379	0	64,700	0	0	234,915	1,430,951	54,075	0
	2021	62	671,205	0	0	0	0	0	52,539	0	67,300	20,300	0	265,699	1,509,160	75,972	0

Caption: ¹ Corresponds to the total types of beans produced nationally (carioca, black, cowpea, jalo, white, etc.)

*Black beans/Cowpea beans: refer to the 2020/21 and 2021/ crops

** no data were found on the 2019/20 crop

Appendix 2 - Centesimal composition (g/100 g), protein quality (PDCAAS), and allergenicity of plant-based raw materials

	COMPOSITION (%)						PDCAAS			ALLERGENICITY
	Moisture	Carbohydrate	Fibers	Lipids	Ash	Proteins	Score	Ingredient	Evaluation	
Peanut*	6.4	20.3	8.0	43.9	2.2	27.2	0.56	NS	Chemical score x digestibility (FAO)	Well established, with high severity. Of 32 proteins from this source, 17 of them are known to be allergenic. Allergy common in the US and UK. 100 mcg is enough to trigger adverse reactions. Proteins resistant to chemical and thermal denaturations, as well as to digestion.
							0.70	NS	NS	
							0.82	NS	Experimental model	
Rice*	12.2	77.5	4.8	1.9	1.2	7.3	0.45 – 0.47	Raw rice	In vitro	Lower allergenic potential. Of 131 proteins identified in the endosperm, only 9 have sequences similar to the allergens already catalogued. Improbable clinical relevance. Specific cases: occupational allergy, inhalation of flour in confectionery, asthma and rhinitis. Main allergens: protease and α-amylase inhibitors.
							0.63	Endosperm protein	Experimental model	
							0.90	Bran protein	Experimental model	
Oats*	9.1	66.6	9.1	8.5	1.8	13.9	0.45 – 0.51	NS	Experimental model	Lower allergenic potential. Rare incidence, limited to childhood. One reason is the use of topical creams in young children with atopic dermatitis. Main allergenic: avenin, a prolamine fraction that also causes celiac disease. Specific cases: occupational allergy, inhalation of flour in confectionery, asthma and rhinitis. Main allergens: protease and α-amylase inhibitors.
							0.51	Raw flour	IVPD X AAS (In vitro)	
							0.57	Cooked protein concentrate	IVPD X AAS (In vitro)	
							0.60	Protein concentrate	IVPD X AAS (In vitro)	
Potato*	82.9	14.7	1.2	-	0.6	1.8	0.93	Protein isolate	NS	Lower allergenic potential. Rare incidence, being classified as GRAS and non-allergenic by the FDA (2016). Main allergenic: patatin (mild allergy). Heat treatment reduces allergenicity. Not classified as allergenic for labeling. Less allergenic than egg and milk.
							0.99	Juice protein concentrate	NS	
							1.05	Protein concentrate	NS	

Appendix 2 - Centesimal composition (g/100 g), protein quality (PDCAAS), and allergenicity of plant-based raw materials

	COMPOSITION (%)						PDCAAS			ALLERGENICITY
	Moisture	Carbohydrate	Fibers	Lipids	Ash	Proteins	Score	Ingredient	Evaluation	
Canola	6.0	10.2	10.0	49.0	2.8	22.0	0.86	Protein isolate	Children (1-2 years); TD (experimental model)	Known in hypersensitive individuals. Main allergenic: napin, belonging to the albumin 2S protein family - intrinsically allergenic. Resistant to digestion and heat treatment. The European Union and Canada recommend including canola in nutritional labeling as a potential allergenic. Cross-reaction with mustard.
							0.87	CanolaPROTM	Children (2 – 5 years)	
							1.00	Protein hydrolysate	Children (1-2 years); TD (experimental model)	
							1.04	IsolexxTM	Children (2 – 5 years)	
Rye*	10.8	73.3	15.5	1.8	1.7	12.5	0.59	Protein extract	TD (experimental model)	Proteins with high similarity to wheat, but with less allergenic potential. Possible moderate to high IgE response in patients sensitive to food allergies. Presence of gluten – non-immunological reactions such as celiac disease. Specific cases: occupational allergy.
Barley	10.5	69.3	4.2	2.7	2.8	11.3	0.44	Pre-peeled flour	IVPD X AAS (In vitro)	Rare incidence, except for celiac disease. May be relevant in children and adolescents - relatively severe clinical symptoms. Mean age of patients with clinical characterization of allergy is 1 year, motivated by early exposure. Type of processing influences allergy. Presence of gluten – non-immunological reactions such as celiac disease.
							0.59	Protein extract	TD (experimental model)	
Black bean*	14.9	58.8	21.8	1.2	3.8	21.3	0.53	Cooked beans	NS	Established. There are 8 allergenic components of size between 16 and 78 kDa, 6 of which are resistant to heat treatment (roasting). Main allergen: 28 kDa protein. Vicillin is a potential food allergen with cross-reactive characteristics (lentil and lima bean). Causes respiratory allergy in sensitive patients.
Cowpea beans*	12.7	61.2	23.6	2.4	3.5	20.2	0.80	Raw beans	NS	Lower allergenic potential.
Mung beans	3.9	65.7	NC	1.0	2.7	26.8	0.52	Protein isolated from cooked beans	Experimental model	Established. Proteins with sequences similar to soybean, lentil, pea and lupin allergens: There are 4 allergenic proteins, with Vig r2 (52 kDa, pI 5.7) and Vig r3 (50 kDa, pI 5.8) being the two main ones.
							0.56	Protein isolated from raw beans	Experimental model	
							0.59	Cooked bean protein	WHO/FAO Standard Score	
							0.64	Uncooked bean protein	WHO/FAO Standard Score	
Chickpeas*	12.3	57.9	12.4	5.4	3.2	21.2	0.52	Cooked chickpeas	NS	Well established. Cross-reactions with lentils and peas. Incidence in India, Spain, Europe, Asia, the Mediterranean and some regions of the West motivated by high consumption.
							0.59	Chickpea flour	NS	
							0.65	Chickpea flour	IVPD X AAS (In vitro)	
							0.71	Protein	NS	
Sesame seeds*	3.9	21.6	11.9	50.4	2.9	21.2	0.44	Seed protein (alkaline extraction)	IVPD	Low prevalence, but potentially severe (anaphylaxis). Main allergens: Oleosins (17 and 15 kDa). Incidence in the USA, Canada, the Middle East (major cause of anaphylaxis) and Israel (3rd most common food allergy).
							0.55	Oil extraction by-product (cooked, 32 % ptn)	IVPDCAAS (in vitro)	
							0.71	Oil extraction by-product (uncooked, 32 % ptn)	IVPDCAAS (in vitro)	
							0.80	Protein concentrate from defatted cake (alkaline extraction)	Mathematical model	
Sunflower	NC	18.8	6.1	49.6	NC	22.8	0.59	Protein isolate	NS	Rare, but reported. Specific cases: occupational allergy (bird feeders).
							0.60	Concentrated flour	NS	
							0.63	Flour	NS	
Lentils*	11.5	62.0	16.9	0.8	2.6	23.2	0.47	Oven green lentil	Experimental model	Well established and one of the most reported in the Mediterranean, motivated by high consumption. Incidence also in Asian countries. Cross-reaction with lentils in children. Main allergenic: Len c1.
							0.53	Cooked green lentil	Experimental model	
							0.57	Extruded green lentil	Experimental model	
							0.63	Cooked whole green lentil	NS	
Cassava*	61.8	36.2	1.9	0.3	0.6	1.1	-	-	-	-
Corn*	63.5	28.6	3.9	0.6	0.7	6.6	0.40	Flours (10 – 70 % ptn)	NS	Established. There is cross-reaction with wheat and barley, as well as pollen. Lipid transfer protein (9 kDa) is the main allergen and trypsin inhibitor (16 kDa) is the minor. Allergen of 50 kDa (γ-zein, possibly) is resistant to pepsin/trypsin and stable to heat, presenting a sequence similar to the wheat glutenin epitope.
							0.42	Gluten flour	NS	
							0.46	Protein	NS	
							0.52	NS	NS	
Sorghum	NC	67.4	14.2	3.2	1.7	13.5	0.22	Sorghum (wet cooking)	NS	Lower allergenic potential, being relatively safe for celiacs and those allergic to wheat-like proteins. Possible cross-reaction with pollen grains. Main allergens: polcalcin, Sor h 1 and Sor h 13
							0.22 – 0.58	Flours (various varieties)	NS	
							0.33 – 0.46	Heated flours 105°C/30 min (3 varieties)	NS	
Wheat	NC	72.6	11.6	1.9	NC	13.7	0.42	Wheat (hard, winter)	NS	Occupational allergy: inhalation of wheat flour in confectionery (asthma and rhinitis). Protease and alpha-amylase inhibitors are the main allergens. Presence of gluten – non-immunological reactions such as celiac disease. Risk of anaphylaxis in case of physical exercise after ingestion of gluten-rich foods.
							0.43	-	NS	
							0.51	-	From 6 months of age	

Caption: NC: not counted; peanut: grain, raw; rice: whole grain, raw; oats: flakes, raw; potato: inglesa, raw; canola: seed; rye: flour, whole grain; barley: whole grain flour; black beans: raw; cowpea (Embrapa = fradinho): raw; mung beans (Embrapa = moyashi): grain, powder; chickpeas: raw; sesame: seed; sunflower: seed; lentil: raw; cassava: raw; corn: raw; sorghum: grain; wheat: flour, whole grain. NS: Not Specified

* Composition data obtained from TACO Table (2004), which includes dietary fiber content in total carbohydrates.

TD: true digestibility; IVPD: in vitro protein digestibility; AAS: amino acid score; IVPDCAAS: in vitro Protein digestibility-corrected amino acid score.

Source: Nepa (2004); Philippi (2002); Aider (2011); Aly (2021); Ge (2021); Queiroz (2015); López (2016); Ciftci *et al* (2022); Sousa *et al* (2011); Pedó *et al* (1990); Han *et al* (2015); Sanchez-Velazquez *et al* (2021); Kleba *et al* (2018); Hussain *et al* (2021); Jiménez-Muñoz *et al* (2021); Chmielewska *et al* (2021); Fleddermann *et al* (2013); Ertl *et al* (2016); Bai *et al* (2018); Nosworthy *et al* (2017); Anyango *et al* (2011); Pape (2016); Turck *et al* (2021); Tavano *et al* (2016); Day (2013); Di (2022); Sà (2022); Escamilla-Silva *et al* (2003); Alexandrino (2017); Nosworthy (2018); Taylor *et al* (2017); Baladrán-Quintana *et al* (2019); Day *et al* (2022); Toomer (2018); Sicherer *et al* (2003); Lehmann *et al* (2006); Mondoulet *et al* (2006); Hirano *et al* (2016); Makinen *et al* (2017); Majamaa *et al* (2001); Wanasundara *et al* (2016); Zimmermann *et al* (2021); Ruiz Segura *et al* (2020); National Library of Medicine (2006); Lee *et al* (2020); Shakoore *et al* (2016); Kumari *et al* (2012); Kumari *et al* (2005); Gupta *et al* (2021); Kaseira *et al* (2011); Carbonaro *et al* (2015); Misra *et al* (2011); Hildebrand *et al* (2021); Dadon *et al* (2014); Honjoja *et al* (2021); Leduc *et al* (2006); Adatia *et al* (2017); Dreskin *et al* (2021); Lavine *et al* (2015); Zitouni *et al* (2000); Pascual *et al* (1999); Soyak Aytekin *et al* (2022); López-Torrejón *et al* (2003); Lee *et al* (2005); Pasini *et al* (2002); Bokka *et al* (2019); Battais *et al* (2003); Moraes *et al* (2012);.

Appendix 3 - Technofunctional Properties of Plant Proteins.

	Protein extraction				Solubility			Emulsifying				Foaming				Gelling			Sensory
	Mtd.	Ext.	Precip.	Ingred.	Mtd.	pH	Res.	Mtd.	pH	A/C	S	Mtd.	pH	C	S	Prod.	Cond. +	MGC	
Peanut	pH	8.5	4.5	Isolate 96.6%	PCS	7	~80%	TBD	6.0 / 8.0	0.25 / 0.25 (OD)	19.2 / 20.4 min	Mixture	7.4	50 and 35% (0 and 60 min)	-	-	-	-	Protein hydrolysate: bitter, umami, salty and full-bodied.
	pH	9.0	4.5	Isolate 85.9%	PCS	7	79.4%		-	-	-	-	-	-	-	14%; 90°C; 1h	pH 10	-	
Rice	pH	10	4.5	89.3% ptn	SPEC.	-	~18%	TBD	7	15 m ² /g	25 min	-	-	-	-	-	-	-	Endosperm protein 80.5%; unpleasant, granular and lumpy; Pleasant, nutty. ^{79,80}
	pH	Alkaline	4.5	88.9% ptn	PCS	7	~7%	TBD	7	1.8 m ² /g	50%	-	-	-	-	-	-	-	
	pH	9	4.5	80.5% ptn	-	-	-	-	-	-	-	Mixture	7	~ 90%	~ 70%	-	-	-	
	pH	9	4.5	74.3% ptn	-	-	-	-	-	-	-	Mixture	7	~ 540%	~ 58%	-	-	-	
	Salt	7	(NH4)2SO4	-	-	-	-	-	-	-	-	-	-	-	-	10%; 95°C; 30 min	-	-	
	CAR	6 (native)	-	Flour 24.4%	-	-	-	-	-	-	-	-	-	-	-	14%; 95°C; 1h30min	-	-	
Oats	pH	9.5	4.5	Isolate 90.1%	PCS	7	60.8%	TBD	7	40 m ² /g	30 min	-	-	-	-	-	-	-	Application in meat analogue: bitter taste, aftertaste, and cereal flavor directly proportional to the concentration.
	pH	9.2	Not precip.	Isolate 82.6%	PCS	7	73.3%	-	-	-	-	DFA	7	136 mm/min			-	-	
	pH	9.2	5.0	Isolate 89.6%	-	-	-	-	-	-	-	-				15%, 110°C; 30 min	pH 3; pH 7	-	
Potato	pH/UF	5.7 - 6.0 (native)	4	Concentrate 85.8%	PCS	7	~70 %	TBD	7	25 m ² /g	20 min	Mixture	7	140%	80 and 80% (30 and 60 min)	-	-	-	Potato juice protein obtained by hot coagulation: bitter, baked, salty and "off flavor"; by ultrafiltration: soft, from raw potato.
	-	-	-	Ptn. Insoluble	-	-	-	-	-	-	-	-	-	-	-	15%; 85°C; 30 min	-	-	
Canola	pH	Max. solubility	Min. solubility	Isolate	PCS	6 / 8	28 / 45%	VL	6 / 8	30.9 / 37.8%	23.1 / 37.9%	Mixture	6 / 8	190.3 / 235%	64.3 / 60% (30 min)	2-20%; 100°C;1h; pH 2 - 10	pH 6 / 8	12 / 14 %	Applications in sausage: 1) addition of 2% rapeseed protein (58.8% ptn) resulted in better flavor and aroma compared to soy concentrate and isolate. 2) addition of 2.0 and 4.5% resulted in astringent/bitter and oily/straw flavor, respectively. ^{51,55,86,87}
Rye	HA	-	-	Secalin 91 %	PCS	7	~7%	TBD	7	77.9 m ² /g	21.0 min	Mixture	7	54%	39% (60 min)	-	-	-	Aplication in bread: 1) aftertaste.
	OS	Phenol	Acet ammonium-methanol	Protein	-	-	-	-	-	-	-	-	-	-	-	(un) Freezing	pH 12.0 / 8, 10 and 13 mg/mL	-	
Barley	pH	11	4.5	Concentrate 70%	-	-	-	-	-	-	-	Mixture	5 / 8	87 / 70%	80 / 70% (30 min)	-	-	-	Applications in bread: 1) bitterness directly proportional to the concentration added; 2) intense flavor and odor ("off"), in addition to bitterness dependent on the barley cultivar.
	pH	11.2	5.4	Concentrate 76.5%	PCS	6 / 8 (0.1%)	10 / 30%	HL	6 / 8	0 / 45%	0 / 41%	-	-	-	-	-	-	-	
	pH	-	-	-	-	-	-	-	-	-	-	-	-	-	-	14%; pH ≥ 8; heating	95°C / 40 min	-	
Black beans	pH	9	4.5	"Isolate" 64.9%	PCS	7	78.2%	-	-	-	-	Mixture	-	272%	36% (30 min)	-	-	-	Applications in bread: acceptance reduced as the addition of black bean flour (21.1% ptn) increased, acceptable up to 10% replacement in relation to wheat; Application of pregelatinized flours (broken grains) in baked snacks and instant gluten-free pasta (penne type): acceptance index > 60%; Extruded: acceptable, with undesirable aftertaste; "beany and green".
	pH	10	4.5	Isolate 82.1%	PCS	7	~66%	TBD	7	10 m ² /g	~ 250 min	Mixture	7	60%	75 / 55% (0 e 30 min)	2-20%; 100°C; 1h ; pH 3 - 9	pH 7	10%	
Cowpea beans	pH	9	4.5	Isolate 87.7%	PCS	7	75%	TBD	7	8.9 m ² /g	37.5 min	Mixture	7	93%	88 / 70% (30 and 60 min)	-	-	-	Application in bread and cake: isolate 90% when added at a concentration of 2.0% in bread and 3.5% in cake: general sensory acceptability > 7 – acceptable replacement.
	pH	8	4.5	Isolate 92.4%	PCS	7	91.5%	-	-	-	-	-	-	-	-	6-16 mg/m; 100°C; 30 min	pH 7	12%	
Mung beans	pH	9	4.5	Isolate 86.2%	PCS	6 / 8	15 / 100 mg/mL	TBD	-	23.6 m ² /g	72.6 min	-	-	-	-	8-20%; 10°C; 1h	-	12%	Application in yogurt: addition of concentrate 80.7% at 3% concentration: evaluation of volatile compounds – unpleasant taste of beans ("beany").
	pH	9 and 11	4.0	Isolate 81.5%	PCS	-	-	HL	-	63.2%	62.7%	Mixture	-	89.7%	78 / 70 % (30 and 60 min)	2-20%; 100°C;1h	pH 7	12%	
Chickpeas	pH	8.3	5	Isolate	-	-	-	TBD	7	150 m ² /g	-	-	-	-	-	-	-	-	Chickpea flour: "Beany".
	pH	9.5	4.5	Isolate 84%	-	-	61.1%	HL	-	70.3%	87.3%	Mixture	-	50%	76%	-	-	-	
	pH	9	4.5	Protein 73.6%	PCS	7	~55%	TBD	7	5.7 m ² /g	~20 min	SC	7	R5 = 35%; Gi = 85%; FE = 105%	2-20%; 100°C;1	-	14%		

	Protein extraction				Solubility			Emulsifying				Foaming				Gelling			Sensory
	Mtd.	Ext.	Precip.	Ingred.	Mtd.	pH	Res.	Mtd.	pH	A/C	S	Mtd.	pH	C	S	Prod.	Cond. +	MGC	
Sesame seeds	pH/salt	7.5	4.5	Isolate 100%	PCS	7	~18%	TBD	7	16.8 m ² /g	17.4 min	SC	7.0	FE = 537%; FS = 88%			-	-	Application in bread: addition of 87% isolate in concentration of up to 7.6% in wheat flour bread resulted in flavor and overall quality significantly equal to that of wheat bread – acceptable replacement.
	pH	9.0	5	Protein	PCS	7	~3%	HL	-	~66%	~68%	Mixture	-	22%	~75 / 65% (30 and 60 min)	-	-	-	
	pH	11	4.5	Isolate 90.5%	PCS	9 - 12	~45% (pH 9)	HL	9 -12	~9%	~60%	Mixture	9 -12	15% (pH9)	40 / 30% (30 and 60 min)	2-20%; 100°C;1h; pH 9 - 12	9 / 12	25 / 14%	
Sunflower	pH	9	4.5	Isolate	PCS	6 / 8	~50 / 65%	TBD	6 / 8	25 / 35 m ² /g	25 / 31 min	Mixture	6 / 8	23 / 28%	7 / 1% (20 min)	-	-	-	Application in bread: concentrate (75%) extracted from the cake was added at concentrations of 5, 10 and 20% to gluten-free bread (rice flour + corn starch) – acceptable replacement in terms of appearance, color, odor, texture, flavor, aftertaste, global acceptance and acceptance index.
	pH	10	4.5	Isolate 78.4%	PCS	7	40%	HL	7	48%	45.4%	Mixture	7	34.9%	61 / 51 (30 and 60 min)	2-20%; 100°C;1h	-	16%	
Lentil	pH	9	4.5	Protein 79.1%	PCS	7.0	~60%	TBD	7	5 m ² /g	17 min	SC	7	R5 = 42%; Gi = 102%; FE = 79%		2-20%; 100°C;1h	-	12%	Flours, concentrates and isolates: "beany, green and grassy".
Corn	Zein				PCS	7	~10%	TBD	7	87 m ² /g	30 min	Mixture	7	10%	50% (30 min)	-	-	-	Corn gluten meal (by-product of corn starch production): undesirable; Protein isolate from corn germ obtained by alkaline extraction and ethanol precipitation VS protein flours from corn germ washed with acid, heat treated and washed with ethanol: isolate is more acidic, less bitter and more astringent.
	Zein				-	-	-	-	-	-	-	-	-	-	-	10-20%; 35:65 without heating.	37°C	15 – 20%	
Sorghum	Ungerminated sorghum flour 11 % protein				-	-	-	-	-	-	-	-	-	-	-	100°C; 8 min	-	8%	Application in bread: 1) sorghum flour containing 9.5% ptn – 100% sorghum bread had better taste (neutral) and odor; 2) 9.5% ptn red and 11.6% ptn white sorghum flour – flat bread with a concentration of 30, 40 and 50% of the sorghum flour had a flavor and general acceptability greater than or equal to wheat bread; Application in cookies: 7.8% ptn sorghum flour - cookies with 58.3 and 66.6% sorghum flour (+ rice flour/corn starch) had sensory attributes of residual bitterness, sweetness, sandiness, chocolate flavor and global quality equivalent to commercial cookie; Sorghum flour - neutral and mild flavor; Some cultivars have a bitter and astringent flavor (presence of tannins).
	pH	8	4.8	Protein	PCS	6 / 8	15 / 60%	TBD	7.0	0.567 (OD)	1 min	-	-	-	-	-	-	-	
	HA	-	5.0	Kafrin	-	-	-	HL	-	~35%	~35%	Mixture	-	2.7%	0%	-	-	-	
Wheat	pH	11	4.5	Protein	-	-	-	TBD	-	22 m ² /g	~65%	Mixture	-	~32%	~89% (20 min)	-	-	-	Application in pasta: addition of gluten at a concentration of 1, 3 and 5% to frozen cooked pasta – we observed no modification of flavor compared to no addition of gluten and small improvement of palatability with 3% addition; Application in bread: process of micronization (jet milling) of whole wheat flour reduced aftertaste.
	-	-	-	Gluten 75.1%	PCS	7	4.6% (ISN)	TBD	-	0.18 m ² /g	~82%	Mixture	-	~8 cm ³	~78% (10 min)	-	-	-	
	-	-	-	Soluble protein 76%	-	-	-	-	-	-	-	-	-	-	-	120°C; 10 min; 2 atm	-	19%	

Caption: CP: chickpeas; Method (Mtd.); Extraction (Ext.); Precipitation (Precip.); Result (Res.); Protein (ptn); Emulsifying/foaming activity or capacity (A/C); Emulsifying/foaming stability (S); Production (Prod.); Favorable condition (Cond. +); Minimum gelling concentration (MGC); Application (Appl.); PCS: protein content of the supernatant; TBD: turbidimetric; HL/VL: height/volume of the emulsified layer; OS: organic solvent; HA: hydroalcoholic; UF: ultrafiltration; CAR: classification by air; DFA: dynamic foam analyzer; SC: sparging chamber.

Source: Ge (2021); Wanasundara (2016); Su (2011); Hu (2019a); Nadathur et al (2016); Kaleda et al (2021); Zwijnenberg et al (2002); Duzgun et al (2020); Zhao et al (2022); Mansour et al (1996); Guo et al (2010); Yoshie-Stark et al (2006); Horstswald et al (2009); Robles-Ramirez et al (2020); Holtekjolen et al (2008); Bento et al (2021); Mariscal-Moreno et al (2021); Simons et al (2015); Campbell et al (2016); Yang et al (2021); Xu et al (2019); El-Adawy (1997); Zorzi et al (2020); Chang et al (2019); Wu et al (2006); Wu et al (2003); Huang et al (1991); Pereira et al (2017); Ferreira et al (2009); Pereira Filho et al (2015); Queiroz et al (2011); Yousif et al (2012); Wang et al (2020); Protonotariou et al (2020) Wu et al (2009); Li et al (2020); Liu et al (2022); Hu et al (2019b); Zhang et al (2022); Wei et al (2022); Yuno-Ohta et al (1994); Kortekangas et al (2020); Zhong et al (2018); Guhmann et al (2018); Nieto et al (2016); Zhang et al (2017); Flores-Jimenez et al (2019); Qazanfarzadeh et al (2021); Lim et al (2013); Houdé et al (2018); Wang et al (2013); Bilgi et al (2004); Ferreira et al (2018); Shevikani et al (2015); Peyrano et al (2016); Brishiti et al (2020); Brishiti et al (2017); Zhang et al (2009); Mesfin et al (2021); Boye et al (2010); Achouri et al (2012); Di et al (2022); Sharma et al (2016); Dabbour et al (2019); Malik et al (2017); He et al (2021); Gagliardi et al (2020); Singt et al (2017); Babiker et al (1998); Georget et al (2016); Tian et al (2022); Liu et al (2021); Confort et al (2003).

Appendix 4 - Frequent products and residues from the processing of plant sources.

	Transformation or Step	Residues and/or By-products	Main products	% m/m residues (relative to m _{input})	Protein content of the residues	Destination of residues
Peanut	Extraction with solvent	Hull, skin and bran	Flour and oil	Hull: 30 % Skin: 3 %	Bran (deffatted peanut meal): up to 50% ptn	D, F, HF
Rice	Skinning and polishing	Hull, bran and broken grains	Brown rice, white rice and flour	Hull: 20% Bran: 10% Broken grains: 16 %	Bran: 14- 18% (db)	D, F, HF
Oats	Skinning and polishing	Hull and oat powder	Flake and flour	Hull: up to 30%	Hull: 2-4% Oat powder: traces	F
Potato	Various processings	Skin, pulp, potato juice and hash	Starch	Skin: ~10% or 12.0 – 27.6 % db Pulp: 14.2 – 17.0 % db Potato juice: 5 – 12m ³ per ton of potato	Skin: 2 – 10 % (wb) Pulp: 4.9 – 6.1 % (db) Potato juice: 2.5% Hash: 10.5 – 11 % (db)	F, HF
Canola	Extraction with expeller, extraction with solvent	Cake from expeller and bran from solvent	Oil	Variable	Bran: 35 - 40 % (db)	F, HF
Rye	Grinding	Bran	Flour	-	Bran 18.2% (db); 14 – 18%	D, F, HF
Barley	i. Malting ii. Others	i. Brewer's spent grain (BSG) ii. Hull	Beer	BSG: 31 %	BSG: 16-27% (db)	D, F, HF
Sesame seeds	Extraction with expeller	Cake (from which flour can be obtained by grinding)	Oil and Flour obtained from the cake	-	Cake: 39.7%	D, F, HF
Sunflower	i. Extraction with expeller -> extraction with solvent OR ii. Pressing -> Solvent	i. Cake from expeller and bran from solvent; ii. Hull and bran (from which "flour" can be obtained)	Oil and "Flour"	2. Bran: 40% 2. Hull: 20-30 %	ii. Defatted and hulled bran: 50 %	F, HF
Cassava	Various processings	Leaves, stem and peel; i. Wastewater (manipueira) ii. Fibrous bagasse	i. Flour ii. Starch (or sweet tapioca powder)	Peel: 2 – 5 %	Bagasse: 2,3 %; Peel: ; 4,5%, 6,9%	F, HF
Corn	Grindings: i. Dry ii. Wet iii. Wet grinding for biorefineries	Bleached grits, bran, and germ, gluten, liquor, fiber, steepwater solubles (SS), corn gluten meal (CGM), and corn gluten feed (CGF)	i. Flour and grits ii,iii. Starch	Bran: 12 %; Germ: 7.5 %; Gluten: 5.6 %	CGM: 60 – 71%;CGF: 18 – 21 % (db) Bran: 10 – 13 % Germ: 12 – 21 %; Gluten: 19 – 24 % Liquor: 40 – 50 %	D, F, HF
Sorghum	i. Decortication ii. Leaf and panicle removal iii. Stalk pressing	Lignocellulosic materials: i. Bran ii. Stalks and panicles (clusters)	i, ii. Flour; iii. Juice/syrup (from sweet sorghum stalks)	Bran: 7.9%	Bran: 10,2-10,4% Panicles: 7.95 and 9.64 % (db) Stalks: 4.86 and 12.5% (db)	F, HF
Wheat	Grinding	Bran and germ; gluten;	Flour	Bran and germ: 23-27%	Bran: 15– 22 % wb; Germ: 26 – 35 % wb	D, F, HF

Caption: D: disposal, F: feed, HF: human food; wb: wet base; db: dry base.

The letters (i, ii, and iii) represent the different steps of processing and relate the products and by-products generated from these steps.

*Information obtained from specialist in the areas of cereals and oilseeds.

Sources: Pereira Filho *et al* (2015); Esteves (2000); Carrão-Panizzi *et al* (1994); Sorita (2021); Baier (1996); Goes *et al* (2013); Decker *et al* (2014); Nutrient (2001); Fernandes [s.d]; Fernandes *et al* (2008); Mussatto *et al* (2006); Queiroga *et al* (2017); Carvalho *et al* (2005); Dourado *et al* (2019); Matte *et al* (2021); Ramos *et al* (2000); Paes (2006); abimILHO (2015); Strazzi [s.d]; Zhang *et al* (2021); Machado *et al* (2014); Da Silva *et al* (2004); Demarchi (1993); Rosenfelder (2013); Dapčević-hadnadev *et al* (2018).

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