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**Alternative Sources of Plant Protein in Tropics
and Subtropics: The Case Study of Bolivia**

BACHELOR'S THESIS

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Declaration

I hereby declare that I have done this thesis entitled Alternative sources of plant protein in the Bolivia region independently, all texts in this thesis are original, and all the sources have been quoted and acknowledged by means of complete references and according to Citation rules of the FTA.

In Prague date

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Štěpán Diewock

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Abstract

This work focused on plants as alternative sources of protein in the tropics and subtropics, specifically in the Bolivian region. The primary objective of the work was to review the available publications and scientific studies dealing with this topic and then to create a comprehensive review. The first part of the thesis informed about protein, specifically its functions, importance to the human body and the reason why it is referred to as the most important component of the diet. It also addressed the question of whether protein of plant origin can be described as a sufficient source of protein. Plant proteins were compared with animal proteins in terms of the amino acids and other substances contained, digestibility, impact on human health and the environmental impact of their production. In the second part of the work was created a summary table of a total of 52 different plant species with information on the basic botanical systematics, edible parts, processing methods and most importantly protein content. These 52 plant species were identified as potentially high-quality sources of plant proteins in Bolivia. They belonged to a total of 24 different families, of which the Fabaceae family was the most abundant with 14 representatives (26.9%). Of the 52 plant species, 4 suitable species were selected based on their solid protein content, good adaptation to the natural conditions prevailing in Bolivia and the potential in expanding their cultivation. Selected species were quinoa (*Chenopodium quinoa* Willd.), Andean lupin (*Lupinus mutabilis* Sweet), mountain peanut (*Plukenetia volubilis* L.), pequi (*Caryocar Brasiliense* Cambess.). These selected species were described in more detail from the following aspects: botanical systematics, habitat, botanical description, nutritional values, and their uses.

Key words: protein, plant, crop, underutilized, nutritional value, chemical composition, Bolivia.

Abstrakt

Tato práce se zabývala rostlinami jako alternativními zdroji bílkovin v tropech a subtropích, konkrétně v oblasti Bolívie. Primárním cílem práce bylo prozkoumat dostupné publikace a odborné studie zabývající se tímto tématem a následně z vytipovaných rostlin, které představovaly kvalitní zdroj bílkovin, vytvořit ucelený přehled. První část práce se zabývala bílkovinami, konkrétně jejich funkcemi, významem pro lidské tělo a důvodem, proč jsou označovány jako nejdůležitější složka stravy. Dále se zde řešila otázka, zda mohou být bílkoviny rostlinného původu označovány jako dostatečně kvalitní zdroj bílkovin. Rostlinné bílkoviny byly porovnány s živočišnými bílkovinami z hlediska obsažených aminokyselin a dalších látek, stravitelnosti, vlivu na lidské zdraví a ovlivňováním životního prostředí jejich produkcí. V druhé části práce byla vytvořena přehledná tabulka s informacemi o základní botanické systematice, jedlých částech, způsobu zpracování, a především obsahu bílkovin celkem 52 různých druhů rostlin, které byly identifikovány jako potenciálně velmi kvalitní zdroj rostlinných bílkovin v Bolívii. Vybraných 52 druhů rostlin patřilo celkem do 24 různých čeledí, z nichž nejpočetněji zastoupená byla čeleď Fabaceae se 14 zástupci (26,9 %). Z 52 druhů rostlin byly vybrány na základě kvalitního obsahu bílkovin, dobré adaptaci na přírodní podmínky panující v Bolívii a potenciálu v rozšíření jejich pěstování 4 vhodné druhy. Vybrané druhy byly quinoa (*Chenopodium quinoa* Willd.), Andean lupin (*Lupinus mutabilis* Sweet), mountain peanut (*Plukenetia volubilis* L.) a pequi (*Caryocar Brasiliense* Cambess.). Následně byly tyto druhy podrobněji popsány z následujících hledisek: botanické systematiky, biotope, botanického popisu, nutričních hodnot a jejich využití.

Klíčová slova: bílkovina, rostlina, plodina, nedostatečně využívané, výživová hodnota, chemické složení, Bolívie

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List of the abbreviations used in the thesis

AA = Amino acids

EAA = Essential amino acids

NEAA = Nonessential amino acids

PAH = Phenylalanine hydroxylase

NAD⁺ = Nicotinamide adenine dinucleotide

ASNS = Asparagine synthetase

CNS = Central nervous system

PEM = Protein-energy malnutrition

WPC = Whey protein concentrate

WPI = Whey protein isolate

PDCAAS = Protein digestibility corrected amino acid score

FAO = Food and Agriculture Organization

WHO = World Health Organization

DIAAS = Digestible indispensable amino acid score

IAA = Indispensable amino acids

NUS = Neglected and underutilised species

1. Introduction

Continuing population growth is one of the biggest global problems the world is facing today. The population is increasing rapidly mainly in the economically weaker countries, where are already relatively large numbers of food-insecure people. This fact raises a few crucial questions about the future, how to ensure sustainable food production for the mankind with least impact on the environment possible. In 2020, up to 2.37 billion people worldwide had no access to sufficient food and between 720 – 811 million people suffered from hunger (FAO et al. 2021).

Protein is the most important macronutrient and has several vital functions in the human body. In general, an adult person needs to take in at least 0.8 g of protein in the diet per 1 kg of weight for proper body growth and maintenance of functions (Bilsborough & Mann 2006). Protein ingested from the diet can be of dual origin, animal, or plant (Frej 2020). Animal protein is of higher quality because it contains the full range of essential amino acids needed for proper body functioning. Plant protein alone does not contain all essential amino acids, but with the right combination of plant sources, a complete intake can be achieved (Kaur et al. 2022). Plant proteins are also healthier and compared to animal proteins they minimize the risk of cancer, which can occur with excessive meat consumption (Perraud et al. 2022).

Animal protein production also has a negative impact on the environment. Combined with the increasing number of people on Earth, there are serious concerns about how to ensure sufficient production of high-quality food with sufficient protein in the future. From this perspective, plant proteins could be the solution (Wu et al. 2014). One of the other factors, why plant protein production is increasingly coming to the fore, is the price. Plant proteins are cheaper than animal proteins, which is a very important criterion, especially in economically weak countries where people simply do not have enough money (Day 2013). Because of the lack of protein in the diet in these countries, there are large numbers of people suffering from hunger and general malnutrition (Cheng et al. 2019). One such country is Bolivia, which is the focus of this thesis. Specifically, whether there is a sufficient diversity of possible sources of plant protein to constitute a diet of sufficient quality.

2. Aims of the Thesis

The main objective of this bachelor's thesis was to review the available literature on important alternative sources of plant protein in tropics and subtropics specifically in the region of Bolivia and their traditionally used processing methods.

3. Methodology

The bachelor thesis was written in the form of a literature review. The necessary information and data used in this thesis originate principally from scientific articles freely available in scientific databases. Among the most frequently used databases were ScienceDirect, Google scholar, Web of Science or Ebsco. Some of the information and sources found came from the library of the Czech University of Life Sciences Prague and the library of the Faculty of Science of Charles University. The selection of suitable publications was based on the selected keywords: protein, plant, crop, underutilized, nutritional value, chemical composition, Bolivia. Plants were selected based on protein content at least approaching 10 g per 100 g. Suitable species described in more detail were selected based on their high protein content, good adaptation to the natural conditions of Bolivia, and the great potential for increasing their cultivation.

4. Literature Review

4.1. Proteins

Proteins are ubiquitous. We find them in the structures of all living organisms from bacteria and viruses to mammals including humans (Whitford 2005). We classify them as very diverse and very numerous folders of the biomolecules. More than 50 % of the dry weight of cells consists of proteins. It follows that proteins play a major role in the formation of the structures of individual cells as well as their function (Garret & Grisham 2016).

The first person to describe proteins was Gerardus Johannes Mulder towards the end of the first half of the 19th century. He cooperated with Jöns Jakob Berzelius in his studies, who created the word protein. It's a combination of the Latin word *primarius* and the Greek god *proteus* (Whitford 2005).

4.1.1. Protein structure

Proteins are relative complex polymers that are made up of twenty different amino acids, but not every protein contains each of the twenty amino acids. In total protein contains up to thousands of individual amino acids, which are connected to each other by already substituted amide bonds. The structure and function of individual proteins is based on the sequence in which the amino acids are linked together by these amide bonds (Damodaran et al. 2007). Protein molecules are divided by their structural organization into four levels. Primary, secondary, tertiary, and quaternary structure (Garret & Grisham 2016).

Overall proteins are composed of a total of six elements namely carbon, hydrogen, oxygen, nitrogen, sulphur, phosphorus. The elements are sorted according to their amount contained in proteins from most to least represented (Whitford 2005).

4.1.1.1. Primary structure

The primary structure of a protein is a linear sequence in which individual amino acids are connected by amide bonds. Amide bonds are more commonly known as peptide bonds (Damodaran et al. 2007). The amino acid sequence is determined genetically by

the variation in the order of the four nucleotide bases (adenine, guanine, cytosine, thymine) in the DNA molecule. Using the triple combinations that made up these nucleotides, one of the twenty amino acids is encoded for proteosynthesis (Holeček 2006).

4.1.1.2. Secondary structure

The secondary structure is based and builds on the primary structure. The hydrogen bonds of adjacent amino acids interact with each other allowing the peptide chain to arrange into characteristic structures (Garret & Grisham 2016). We distinguished two structures, α -helix, or β -sheet structure. The β -sheet structure can be also described as folded segments (Whitford 2005). The α -helix represents the most common structure found in proteins. The individual parts of the helix are stabilized by hydrogen bonding. A total of thirteen atoms of the N-H group are found in this structure. Each N-H group is hydrogen bonded to the relevant C=O group (Damodaran et al. 2007). The β -sheet structure is also a helical layout, but it's a very elongated and extended conformation compared to the α -helix (Whitford 2005). Overall, it's a more stable structure formed by the interaction of hydrogen bonds between two beta strands of the same molecule (Damodaran et al. 2007).

4.1.1.3. Tertiary structure

The tertiary structure is a compact three-dimensional shape that originated from the linear structure and is formed by folded polypeptide chains (Garret & Grisham 2016). Compared to the secondary structure, there is a major difference in the amino acid sequence. The secondary structure represents the designation of two adjacent amino acids, the tertiary structure in contrast includes amino acids with much longer reach. These amino acids from different type of secondary structure interact with each other in the already complete protein structure (Nelson & Cox 2004). To ensure the stability of the structure there are covalent disulphide bridges, hydrogen bonds, salt bridges and hydrophobic reactions (Baynes & Dominczak 2019). We distinguish fibrillar or globular structure according to the shape and properties of the protein. If hydrogen bonds connect different polypeptide chains, it is a fibrillar structure. However, if hydrogen bonds connect parts of the same chain, it is a globular structure (Whitford 2005).

4.1.1.4. Quaternary structure

The quaternary structure is found in proteins that contain two or more polypeptide chains. Each of these polypeptide chains is referred to as a subunit (monomer) of the protein (Garret & Grisham 2016). Between the chains take place the same interactions as in the tertiary structure. Specifically, hydrogen bonds, disulphide bonds, hydrophobic interactions (Whitford 2005). The resulting proteins are classified according to the number of subunits they are composed from. We distinguish dimers, trimers, tetramers, etc. (Damodaran et al. 2007). Haemoglobin is an example of protein with a quaternary structure. This protein found in the blood is tetramer formed by four subunits. It is composed of two alpha subunits and two beta subunits (Bailey 2019).

The quaternary structure of a protein can be also different in nature and divided into homogeneous and heterogeneous. Homogeneous contains two or more identical subunits. Heterogeneous, on the other hand, is composed of two or more distinct subunits. Homogeneous proteins include β -lactoglobulin, which contains identical subunits. The above-mentioned haemoglobin is one of the heterogeneous proteins (Damodaran et al. 2007).

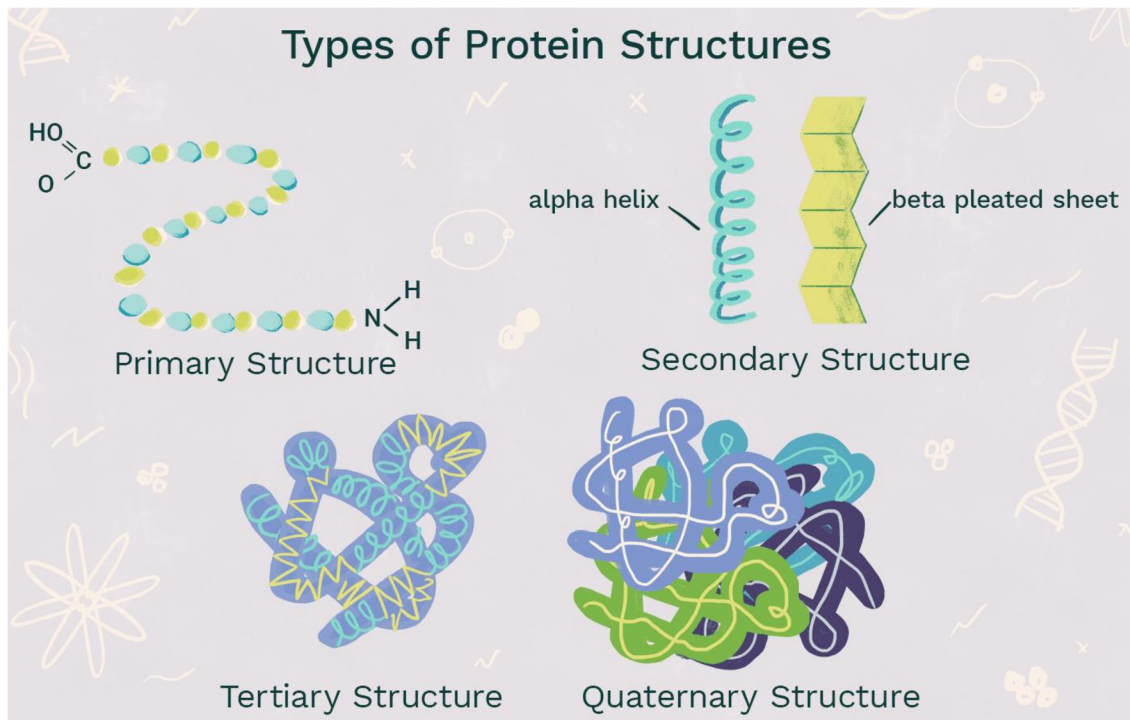


Figure 1. Types of protein structures (Bailey 2019)

4.1.2. Amino acids

Amino acids (AA) are organic compounds consisting of an amino group and acids. Enough AA in diet is essential for the health, growth, development, reproduction, and overall survival of organisms (Wu et al. 2013). There are hundreds of AA in living organisms, which are present in both free and bound forms. Proteins contain only bound AA, of which there are twenty types, namely: alanine, arginine, asparagine, aspartate, cysteine, glutamine, glutamate, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine (Barett 1985). These AA are known as L- α -amino acids and represent the basic structural units of proteins.

Each of these AA is composed of more parts. The basic component is central α -carbon atom to which is covalently binds a hydrogen atom (-H), a basic amino group (-NH₂), a carboxyl group (-COOH) and a side chain group (-R) (Baynes & Dominczak 2019). The basic shape is the same for all. The differences between them form a side chain whose structure influences their properties and nature. Specifically, variability in chemical structure, size, electrical charge, water solubility (Nelson & Cox 2004). In terms of water solubility, we divide them into two groups. The first group are hydrophilic (or polar), which react with water. The opposite is hydrophobic (or non-polar), which do not react with water (Fellows 2009).

Based on whether the human body is able synthesize AA alone and on growth or nitrogen balance AA are classified as nutritionally essential (indispensable) or non-essential (dispensable) (Lopez & Mohiuddin 2020).

4.1.2.1. Essential amino acids

The term essential amino acids (EAA) refer to amino acids that the human body is unable to synthesize in sufficient quantities to maintain the health, growth and development of the body and must be supplied in sufficient quantities in the diet (Hou Y et al. 2015). EAA are vital AA. They have a major influence on the use of other AA for protein synthesis in cells and tissues, especially in skeletal muscles. Insufficient intake can lead to extensive metabolic and psychological problems and, more importantly, to the overall degradation of the organism (Hou & Wu 2018). EAA overall include nine

proteinogenic AA, namely: histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, valine (Reeds 2000).

Histidine

It is essential for protein synthesis and is needed as a precursor to the dipeptide carnosine, which is an important buffer and antioxidant in muscle and brain. Histidine is the second least represented AA in overall body protein. According to WHO/FAO, the required amount is 10 mg per kilogram of body weight (M. Brosnan & J. Brosnan 2020).

Isoleucine

Isoleucine significantly increases the uptake of glucose into the cell and especially its conversion into energy. It speeds up muscle recovery after activity and promotes muscle growth (Patel 2022).

Leucine

Leucine is widely used in the field of dietary supplements. It is added to whey protein as it represents the most powerful activator of muscle protein synthesis of all AA. More specifically, it is the component responsible for stimulating postprandial muscle protein synthesis and must be present in sufficient quantities to optimize the effect of the postprandial response.

The importance of leucine also lies in its insulinotropic properties, which can be used as a resource to stimulate endogenous insulin release. This improves postprandial glycaemic control in patients with type 2 diabetes. A greater insulin response stimulates glucose uptake in the patient's blood and reduces the postprandial degree of hyperglycaemia (van Loon 2012).

Lysine

It is an indispensable component for the human body. First, it helps to produce collagen which is the most abundant protein in the body and forms connective tissue that makes up bones, ligaments, cartilage and is also needed to repair broken bones. In addition to the formation of collagen, it also enables the formation of the vital substance carnitine. Carnitine is the main stimulus in the production of energy from fatty acids and

is concentrated mainly in skeletal and cardiac muscle tissues, which are energetically dependent on fatty acids (Renee 2018).

Methionine

Methionine has a leading role in the initiation of protein synthesis. Regularly forms the first AA in protein. It is part of antioxidant systems and serves as a precursor for cysteine and cystine. For the human body it also has a great value as a supplier of sulphur (Romanet et al. 2020).

Phenylalanine

Phenylalanine mediates two important functions for the human body. Firstly, it is involved in protein synthesis, but also undergoes a chemical reaction in which it is converted into the NEAA tyrosine. The conversion occurs by oxidation of phenylalanine, which is catalysed by PAH. The resulting tyrosine is further dismantled into products that are involved in the citric acid cycle (Flydal & Martinez 2013).

Threonine

Threonine is one of the basic components of protein and is involved in protein synthesis. The catabolic reactions of threonine produce several products very important for metabolism. Namely, glycine, acetyl CoA and pyruvate (Kidd & Kerr 1996).

Tryptophan

Tryptophan has a huge impact on the overall health of the organism. It is one of the basic building blocks for protein synthesis in humans and animals. Tryptophan has an important function in the liver, where part of it is unfolded through metabolic mechanism called the kynurenine pathway into the molecules the body needs. In the nervous system and intestine, it is an essential substrate for the synthesis of serotonin and in the pineal gland it mediates the synthesis of melatonin. Tryptophan is also needed when dietary of niacin is deficient. It is necessary for the synthesis of the essential cellular cofactor NAD⁺ (Moffett & Namboodiri 2003).

Valine

Valine is an AA of hydrophobic nature. This property is used in industry to produce pharmaceutical products or in cosmetics for skin care. The use of valine also

includes supplementation of dietary products in human and animal nutrition, where it has become an important feed ingredient (Oldiges et al. 2014).

4.1.2.2. Nonessential amino acids

NEAAs include AAs that the body can synthesize in sufficient quantities for growth, development, and overall health of the organism. They are perceived to be dispensable in the diet and the amount of synthesis depends on a sufficient supply of EAAs and glucose (Hou & Wu 2017). They have a crucial effect on the functioning of the organism as they play an important role in many processes such as the regulation of gene expression, cell signalling, antioxidant reactions, neurotransmission, and immunity (Wu et al. 2013). A total of eleven proteinogenic AAs is classified as NEAAs, namely: alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, proline, serine, tyrosine (Hou & Wu 2017).

Alanine

Alanine is involved in protein formation or can be used to synthesize other AAs. It is a component of almost all proteins and peptides in the body. In addition, alanine transports ammonia from muscles to the liver and forms so-called alanine cycle, in which it is involved in gluconeogenesis (Kohlmeier 2003).

Arginine

Arginine is one of the most versatile AA as it is involved in the formation of many important substances. It is a precursor for the synthesis of proteins, but also for synthesis of urea, nitric oxide, polyamines, proline, glutamate, creatine, and agmatine. Furthermore, arginine is significantly involved in improving sexual function, immunity, and overall health (Morris 2006).

Asparagine

Asparagine is the product of asparagine synthetase (ASNS), which converts aspartate with glutamine to asparagine and glutamate in ATP-dependent reaction. ASNS is found in almost all organs but is very sensitive to cellular stress. When ASNS is deficient or mutated, neurological disorders can occur. The importance of ASNS lies in

the treatment of acute lymphoblastic leukaemia, which occurs mainly in children (Lomelino et al. 2017).

Aspartic acid

Aspartic acid occurs in the body in two forms: L-aspartic acid and D-aspartic acid. L-aspartic acid is involved in protein synthesis and in the formation of immune-promoting antibodies. D-aspartic acid is not involved in protein synthesis. It is found in the pituitary gland and testes, where it is involved in the synthesis and regulation of testosterone and luteinizing hormone (Zoppi 2021).

Cysteine

Cysteine is a sulphur-containing AA. In the human body, it is used to produce the antioxidant glutathione and to stabilize the structure of proteins. Stabilization is achieved by oxidation of cysteine, which forms disulphide bonds that strengthen the tertiary and quaternary structure of proteins. Cysteine is a component of β -keratin, which is found in skin, hair, and nails. The number of disulphide bonds affects the hardness of keratin (Burns et al. 2016).

Glutamic acid

Glutamic acid forms proteins and its ionic form called glutamate has an important function in the central nervous system (CNS). It is the main excitatory transmitter in the brain and mediates synaptic transmission of excitations (Garattini 2000).

Glutamine

Glutamine is the most abundant AA in the blood, the most abundant free AA in the body and is essential for many processes. It is constantly produced from skeletal muscle and serves as an essential component for the cultivation of human cells. Also, it is an important precursor for the synthesis of other metabolites such as purines, pyrimidines, and glutathione. Glutamine is also one of the most effective substances for gluconeogenesis and, finally, has a strong influence on cell volume regulation and immune system function (Roth 2008).

Glycine

Glycine is the most important NEAA and represents 11,5 % of the total AA content in the body. It is synthesized mainly from serine by the activity of the liver and kidneys. Glycine has an important role in metabolism and nutrition and is especially used for protein synthesis. It forms the structure of two important extracellular proteins, elastin, and collagen. In the CNS, glycine has a neurotransmitter function and controls food intake, behaviour, and homeostasis. The regulation of immune function is also controlled by glycine and therefore a more severe absence of glycine in the body can lead to immune response failure, poor growth, and other adverse health effects (Razak et al. 2017).

Proline

Proline is unique among proteinogenic AAs in its cyclic shape. This regular cyclic structure influences the secondary structure of proteins and specifically imparts stiffness to them. In addition to this property, proline is involved in the maintenance of cellular homeostasis, development, and acclimatization to stress in plants (Alvarez et al. 2022).

Serine

Serine is an important AA for protein synthesis and operates as a precursor for several important essential compounds such as sphingomyelin. In an addition, the proper functioning of the organism is dependent on serine, as it plays a crucial role in the structure and function of the nervous system. Insufficient synthesis of serine leads to a group of serious diseases of the organism with significant neurological disabilities occur. Especially in children in early infancy severe developmental delays has been observed (El-Hattab 2016).

Tyrosine

Tyrosine belongs to NEAA, but it is dependent on a sufficient supply of EAA phenylalanine, from which is subsequently formed. Inadequate intake of phenylalanine results in insufficient synthesis of tyrosine and needs to be adequately supplemented from the diet. Tyrosine is found in the highest amount in the milk protein casein, as well as in long-ripened wines. The primary function of tyrosine is in protein synthesis and it also a precursor of a few important neurotransmitters, such as dopamine, which is involved in mood regulation (Kapalka 2010).

4.1.3. Types of protein

According to their structure in the organism, proteins are divided into two groups: simple proteins and conjugated proteins (Holeček 2006).

4.1.3.1. Simple proteins

This group includes proteins that are composed only of substances of protein nature i.e., amino acids. Most of the simple proteins, nevertheless, also contain a small amount of carbohydrate, but the proportion of protein is highly predominant and for that reason we still include them into this group (Blanco & Blanco 2022). We distinguish two types of simple proteins. Globular (spherical) and fibrillar (fibrous) proteins (Holeček 2006).

Globular proteins

Globular proteins are spherical in shape and represent the most abundant group of proteins in nature. A key characteristic is very good solubility in aqueous solutions. The compact three-dimensional structure is stabilized by interactions between the amino acid side chains. The diversity of the structure translates into a multitude of functions in the organism. They are involved in many important processes such as: transport, catalysis, regulation, immunity, and cell signalling (Shen 2019). Examples of globular proteins are albumins, globulins, histones (Blanco & Blanco 2022).

Fibrillar proteins

Fibrillar proteins, also called scleroproteins, are a very important group of proteins found only in animal tissues (Blanco & Blanco 2022). The structure is formed by polypeptide chains located along the same axis, forming characteristic long filaments. This arrangement gives them great physical strength and prevents them from dissolving in water. Strength is an essential property that makes fibrous proteins an important structural component. The most notable representatives are keratin, collagen, and elastin.

Keratin is mostly contained in hair, nails, horns, hooves, and wool. Collagen is the most important connective tissue in animals and is a part of tendons, cartilage, bones, teeth, skin, and blood vessels (Shen 2019). Elastin is a component of connective tissue and forms very elastic fibres (Blanco & Blanco 2022).

4.1.3.2. Compound proteins

Compound proteins are a set of proteins containing a substance of non-protein nature in their structure. The part containing the protein chain is called the apoprotein, the part containing the non-protein substance is called the prosthetic group. According to the type of the prosthetic group, we classify compound proteins into nucleoproteins, chromoproteins, glycoproteins, phosphoproteins, lipoproteins, and metalloproteins (Blanco & Blanco 2022).

4.1.4. Protein function in the human body

Proteins, along with carbohydrates and fats, are vital macromolecules that form an integral part of the diet. They are involved in the structure and function of almost every cell in the body and adequate supply is essential for growth, development, and survival of the organism (Lieberman & Peet 2017). The primary function of proteins is to create structures, as they are the only building blocks in the diet and cannot be replaced by other substances (Blanco & Blanco 2022). They form structural elements in connective tissues and maintain cell shape. Other vital functions include transport of oxygen (haemoglobin, myoglobin) and other substances in the blood, muscle contraction (actin, myosin, troponin), control of cell division and gene expression, protection against diseases (immunoglobulins). Certain proteins have important catalytic, control, and regulatory functions (enzymes, hormones, receptors) (A-Level Biology 2022). They also have a storage function and in extreme cases can be used as source of energy for the organism if other necessary substances are not available (Hoffman & Falvo 2004).

The recommended protein intake depends on many factors such as age, gender, physical activity, pregnancy, and associated lactation (Wu 2016). Protein should make up 10-35 % of daily caloric intake. Any lower or higher intake has a negative effect on the body (Wempen 2022). In general, it is stated that for an adult person to maintain growth and all body functions, the minimum daily protein intake is 0.8 g per 1 kg of weight (Bilsborough & Mann 2006).

Higher protein intake is required in children for optimal growth and body development, in elderly, where muscle mass decreases and body functions weaken, and in pregnant women, where sufficient protein intake is needed for proper foetal development. Increased protein intake is also necessary after childbirth, when a woman

needs adequate amount of protein for lactation (Millward 2012). Muscle-building athletes must also include increased amount of protein in the diet to maximize the effect of training. The recommended amount of daily protein intake for athletes ranges from 1.6-2.2 g per kilogram of body weight (Gunnars 2020).

4.1.4.1. Protein deficiency

Inadequate protein intake is a global problem, occurring mainly in developing countries, especially among young children and pregnant women. These two groups have increased protein requirements that cannot be met under local conditions and are therefore the most affected by protein-energy malnutrition (PEM). Protein deficiency causes impaired growth, damaged tissue, reduced number of synapses which is associated with delayed mental development. At the same time, the body is more susceptible to various types of parasitic and infectious diseases that children are usually unable to resist (Batool et al. 2015). Overall, approximately 13 million children die annually in developing countries because of malnutrition (Briassoulis et al. 2001).

PEM represents a widespread biological disorder that occurs in the human body because of insufficient nutritional intake. The individual suffers from an energy deficiency due to which protein must be used as an energy source. As a result, there is no protein left to support growth, the immune system or for tissue repair (Singh et al. 2017). The two main diseases of PEM are kwashiorkor and marasmus (Sengupta et al. 2019). Kwashiorkor is the result of a poor diet based on enough calories derived mainly from carbohydrates, but at the same time insufficient protein intake. It causes the typical bilateral pitting pedal oedema and ascites. Marasmus is a serious disease resulting from overall caloric insufficiency. A person suffering from marasmus experiences a visible reduction in growth, loss of muscle mass, adipose tissue, and may cause the development of pitting oedema (Titi-Lartey & Gupta 2022).

4.1.4.2. Protein surplus

While people in developing countries struggle with a lack of protein in the diet, the exact opposite problem occurs in developed countries. People have access to a diet based plenty of calories and that often leads to the intake of more protein than the body can properly process (Cheng et al. 2019). Higher protein intake than the recommended

amount of 10-35 % of total daily energy intake is desirable only for athletes, pregnant and lactating women, and people performing physically demanding work. In these cases, the body can process the obtained protein properly and a safe daily intake is 2-3.5 g per kilogram of weight (Huizen 2018). In other cases, the body is not able to use the acquired protein in the right way. Instead, it builds up fat stores from them, and the rest of the unnecessary AA are excreted. Excessive protein intake does not affect health in the short-term, but long-term intake of excessive amounts of protein can lead to an increased risk of serious health problems (Cronkleton 2020).

Inability of the body to use excessive amounts of protein efficiently poses a danger to the liver, kidneys, and bones. High amounts of protein cause the production of large amounts of acids. This process puts a lot of pressure on the kidneys, which must increase their activity due to the higher acidity. At the same time, a buffer of active bone resorption begins to be released from the skeleton. There is a high loss of calcium and the impossibility of its reabsorption. Excessive bone loss is a consequence, and the combination of these defects leads to the excretion of large amounts of calcium in the urine (hypercalciuria). In combination with low fluid intake, hypercalciuria causes the formation of kidney stones. As a result of a high-protein diet, a lot of saturated fat and cholesterol enters the body, and this can lead to an increased risk of coronary artery disease or even cancer (Delimaris 2013).

4.1.5. Sources of protein

Protein is the most important macronutrient, and it is a component of almost all foods (Frej 2020). The protein content of each food ingredient is not the same but varies depending on its origin (Hayes 2020). According to the origin of protein in the food spectrum, we divide them into animal proteins and plant proteins (Frej 2020). Nowadays, protein supplements are becoming a relatively abundant source of protein. This industrial protein is made from whey and is popular and used source of protein especially among athletes (Hoffman & Falvo 2004).

4.1.5.1. Animal protein

In general, proteins of animal origin have a higher nutritional value. They are referred to as complete proteins because they supply the body with all the essential amino

acids needed. Compared to proteins of plant origin, they are also more highly valued because of their higher digestibility in the gastro-intestinal tract (Kaur et al. 2022). Animal proteins can be found in all types of meat, eggs, milk, and dairy products such as cheese, yoghurt, and curd (Frej 2020). Meat is a very important component of the diet, as it is the primary source of water, fat, and essential micronutrients in addition to protein. It is a prominent source of iron, zinc, selenium, vitamins B6 and B12, vitamin D and provides significant amounts of omega-3 fatty acids (Ferguson 2010).

Proteins of animal origin are still more in demand worldwide than proteins of plant origin, even though they are less sustainable from an ecological point of view (Langyan et al. 2022). Animals have been a staple food for mankind for thousands of years and to ensure sufficient animal production are currently used extensive and intensive farming systems. However, the world's population is constantly increasing and the demand for animal products is growing in direct proportion, which brings with it problems in terms of sustainability and negative environmental impact. Livestock production consumes huge amounts of water, can contribute to environmental pollution (groundwater contamination), and generates greenhouse gas emissions which contribute to global warming. The use of modern technologies and efforts to make livestock production more efficient will minimise negative environmental impacts and increase sustainability (Wu et al. 2014).

Due to the unclear sustainability of animal protein, other possible sources of protein are being sought. In addition to plant-based proteins, insects are coming to the fore as a very high quality but relatively unusual source of protein. Insect proteins have a high nutritional value and represents a potentially rich source of protein, minerals, vitamins, and fatty acids (Gravel & Doyen 2020). Some species are consumed in the larval stage, others as insect exoskeletons. Insects are also processed in the food industry and added to various foods (Kaur et al. 2022). Compared to animal protein, insects are slightly less digestible, but at the same time more digestible than plant proteins. They are more environmentally friendly as they release less greenhouse gases. Insect farming is relatively easy, inexpensive, and due to their high-quality protein content and excellent overall nutritional value, represents one of the best solutions to provide sufficient quality nutrition for an ever-growing population (Gravel & Doyen 2020).

4.1.5.2. Plant protein

Plants are the second traditional source of protein for the human population. In addition to protein, they also provide the body with other macronutrients as well as large amounts of micronutrients, mainly unsaturated fats, vitamins, antioxidants, and fibre (Sokolowski et al. 2020). Plant proteins are increasingly coming to the fore as they are perceived as a very healthy and sustainable source of protein. In contrast to animal protein, plants are not complete proteins as they are usually deficient in one or two essential amino acids. Therefore, they do not have as high a nutritional rating as animal proteins and are considered as a source of protein with low nutritional value. The deficient amino acid is lysine in cereals and methionine with cysteine in legumes. Despite their low nutritional value, crops can be a sufficient source of all the amino acids needed by the human body, if they are consumed in the right combination (Kaur et al. 2022).

The low nutritional value of plant proteins is also caused by their lower level of digestibility compared to animal proteins. Digestibility expresses the amount of protein successfully absorbed as a proportion of the total protein consumed. It depends on the structure of the protein and the presence of digestion-inhibiting compounds. These compounds are referred to as antinutritional factors and include phytates, tannins, trypsin inhibitors, and lectins. Digestibility of plant proteins can be improved by removing antinutritional factors, which can be achieved by sufficient cooking, baking, drying, fermentation, etc (Sá et al. 2020).

Plant proteins are cheaper than animal proteins, but their consumption is still relatively limited. A large proportion of plant protein is used as animal feed and thus for the subsequent production of animal protein. This method is relatively inefficient, as only a fraction of the plant protein is ultimately converted into animal protein for human consumption. To ensure a sustainable diet, it is important to include more plant-based foods (Day 2013). This will lead to a reduction of the negative impact on the environment and to a possible improvement in the health of the population (Alves & Tavares 2019). Including more plant proteins in the food spectrum significantly reduces the risk of many serious diseases that can affect the human body through excessive consumption of animal proteins, especially red meat (Perraud et al. 2022).

Plant proteins represent the most important source of protein in the human diet of all existing options. They account for more than 50% of protein consumption. Important sources of plant protein are cereals, legumes, pseudocereals, nuts and almonds, seeds. Globally, the most consumed crops supplying sufficient protein intake are cereals and legumes. Cereals, namely wheat, barley, rice, and maize, have been staple food for humans for many millennia. For example, rice is one of the most consumed cereals worldwide. Legumes have excellent nutritional values and are considered as the best option in plant-based protein intake. These include beans, lentils, soybean, chickpea, and peas. Chickpea products are a highly used and high-quality source of protein (Langyan et al. 2022).

Soybeans is a very important crop that has been included in the crop rotation in countries around the world. Soya have been a staple of the Asian diet for several thousand years. During the 20th century it was introduced to other parts of the world and since then it becomes one of the largest sources of plant protein in the world. Soybeans contain the most protein of all cereals and legumes and have excellent overall nutritional values. In addition to protein, it is a rich source of many important food components, including edible oil. In its raw state, humans cannot take advantage of all its benefits and therefore it needs to be cooked before consumption. Soya is also used to produce many foods, especially tofu, that are part of the diet of a large part of the population (Nadathur et al. 2017).

4.1.5.3. Protein as a dietary supplement

The basis of protein supplements is whey, which is one of the two proteins found in milk. Milk contains 20% of it, the remaining 80% is casein, the other milk protein (Gangurde et al. 2011). Whey was used on a much smaller scale in the past than at present. It was used in medicine as a medicinal product, otherwise it was mainly considered as a waste product. The potential of whey was only discovered in the late 20th century through science and whey became an important nutritional component in many people's lives (Smithers 2008). It represents a supplementary source of protein in various nutritional applications such as infant feeding. Due to their content of large number of branched-chain amino acids, which strongly support muscle formation, whey protein is mostly used in the nutrition of athletes (Boland 2011).

Whey is produced as a by-product of the manufacture of dairy products, especially cheese. The use of rennet or organic acid coagulates the casein protein. As a result of this reaction, the casein curd separates from the milk and excess liquid, whey, is produced. Whey has a yellow colour, but this can change slightly depending on the origin of the milk. It can be made from any type of milk, but cow's milk is most used (Smithers 2008). Compared to casein, whey protein is more soluble, absorbs much more quickly, and has a higher quality rating. In comparison to other protein sources, whey protein performs best in terms of biological value. Depending on the method of production, different types of whey are produced of which three main forms are used: whey protein concentrate, whey protein isolate, and hydrolysed whey protein (Gangurde et al. 2011).

Whey protein concentrate (WPC) contains a certain amount of fat, carbohydrates, lactose, cholesterol, and bioactive substances compared to whey protein isolate (Hoffman & Falvo 2004). It is produced in several variants differing in protein concentration. For example, there are variants with 35% protein (WPC35), 50% protein (WPC50) or 80% protein (WPC80). Each of these variants has different characteristics and is used for different purposes. WPC35 is perfectly soluble and has a milky taste. It can be found manufacturing procedures in the production of dairy products or in infant formula. WPC80 has a lower carbohydrate profile than WPC35. It can bind water and is an important element for sports nutrition (Bacenetti et al. 2018). Forms of WPC with protein content of more than 70% are most used as a dietary supplement in the form of protein powder (Gangurde et al. 2011).

Whey protein isolate (WPI) represent the best possible source of protein available. The protein content is at least 90% and the isolates are also safe for people with lactose problems (Hoffman & Falvo 2004). During the manufacturing process, fat and lactose are significantly removed and a maximum of 1% is subsequently present in the products. Compared to WPC they are much faster absorbed and have a faster and better-quality effect (Gangurde et al. 2011).

Hydrolysed whey protein is partially pre-digested and hydrolysed form of whey protein. During hydrolysis, the peptide chains are split into peptides, which are then more easily absorbed. Hydrolysates have a bitter taste compared to WPC and WPI and are significantly more expensive (Gangurde et al. 2011).

4.1.6. Quality of protein in the diet

For the overall health of the body, it is necessary to consume a balanced diet with enough quality protein (Hayes 2020). At the same time, an adequate supply of micronutrients such as vitamins and minerals must be provided, without which the body cannot perform to its full potential (Frej 2020). The correct determination of the quality and protein content of each food ingredient is very important as it affects the subsequent economic value of the product (Hayes 2020). The assessment of protein quality is based on digestibility, essential amino acids content and amino acids availability. The subsequent evaluation of protein quality is performed using several methods such as protein efficiency ratio, biological value, net protein utilization, and protein digestibility corrected amino acid score (Hoffman & Falvo 2004).

Protein digestibility corrected amino acid score (PDCAAS) is an international method established in 1991 by the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) to detect and evaluate the quality of protein in the human diet (Millward et al. 2008). The PDCAAS method works based on two important steps. First, the structure of the amino acids in protein must be observed and the one amino acid that is present in the lowest amount is the observed protein must be identified. This amino acid is referred to as the limiting amino acid and determines the overall amino acid score, which determines the overall efficiency of the observed protein. The resulting limiting amino acid value is then compared to a reference formula that represents the minimum protein requirements for the human body to maintain tissue and growth. To obtain the correct total value, a second step must be included in the calculation, which is to consider the digestibility of the protein (Schaafsma 2005). After taking protein digestibility into account, we get the final chemical score. The human body is not able to use excessive amounts of substances, so chemical scores exceeding 100% are rounded up to 100%. On the other hand, if the chemical score does not reach the 100% thresholds, it means that the protein is not able to provide a sufficient supply of amino acids for the minimum required nutrition of the human body (Schaafsma 2012). In general, the quality of protein in terms of digestibility is higher for animal protein than for plant protein. Plant proteins, except for some soy protein isolates, have lower PDCAAS values than the 100% required to match the body's needs. The plant protein with the lowest PDCAAS value is wheat gluten, which achieves a final value of only 25%. The lower values of plant proteins

compared to animal proteins are due to the poorer digestibility or possibly the absence of a certain amino acid or amino acids (Berrazaga et al. 2019).

Digestible indispensable amino acid score (DIAAS) is a newer method proposed by the FAO as a replacement for the older PDCAAS method, which has been widely criticised. DIAAS is based on the observation of the actual amino acid digestibility, which is determined for each amino acid separately (Rutherford et al. 2015). For the final evaluation of protein quality by DIAAS, it is necessary to know not only the IAA (indispensable amino acids) content and profile, but also the protein content of the food source. Compared to the PDCAAS method, DIAAS is more accurate in revealing the nutritional value of dietary protein as well as its quality. Especially for plant protein sources, better accuracy of results is observed and in situations where it is not one specific protein but a mixture of different types of proteins (Wolfe et al. 2016).

4.2. Overview of Bolivia

Bolivia, officially called the Republic of Bolivia or Bolivian Plurinational State, is a landlocked country located in the western and central parts of South America. Bolivia borders a total of five countries, namely Peru to the northwest, Brazil to the northeast, Paraguay to the southeast, Argentina to the south, and Chile to the southwest (Arnade & McFarren 2023). Its total area of 1,098,581 km² makes it the twenty-eighth largest country in the world and fifth largest country in South America (Worldometer 2023). In 2022, Bolivia had a population of over twelve million inhabitants. An interesting fact may be that Bolivia has two capital cities, Sucre, and La Paz. Sucre is home to the Supreme Court, which makes it the constitutional capital. The institutions for executive and legislative power are in La Paz, making it the administrative capital (Arnade & McFarren 2023).

4.2.1. Geography

Bolivia is generally perceived as a high-mountain country, but in terms of topography its geo-relief is very diverse and can be divided into three regions: the Andean, the Sub-Andean, and the lowland (Morales 2010). The Andean region is in the western part of Bolivia and is composed of two dominant mountain ranges. Further west, near the border with Chile, is the Cordillera Occidental, in which is located Bolivia's

highest peak Sajama (6,542 m), the important Uyuni salt flat and many active volcanoes. Further east is the Eastern Cordillera, known as the Royal Cordillera in the vicinity of La Paz. Between these mountain ranges is the important Altiplano plateau, which extends into southern Peru and northern Argentina. On this plateau is located the highest navigable lake in the world, Lake Titicaca, whose area is divided between Bolivia and Peru, and another important lake, Lake Poopó (Arnade & McFarren 2023). The Altiplano region is historically a very important economic and cultural area for the Bolivians.

The Andean region transitions eastwards into the Sub-Andean region, which is a transitional area between the mountains and the lowlands. It is located along the slopes of the Andes and consists of numerous temperate valleys which, in the north, become semi-tropical valleys, known as Yungas (Morales 2010). The Sub-Andean region transitions into extensive lowlands. To the northeast and east of the Andes is a vast area called the Oriente, which is part of the Amazon basin and forms over two-thirds of Bolivia's total territory. The Oriente area is made up of swamps, savannahs, and tropical forest. In the southeast of the country, the Oriente region transitions into a fertile flat area called the Bolivian Chaco, which is part of the Gran Chaco (Arnade & McFarren 2023).



Figure 2. Physical map of Bolivia (Ezilon.com 2009)

4.2.2. Climate

Although Bolivia is situated in a tropical zone, the huge difference in altitude creates overall very diverse climatic conditions. From the warm equatorial plains in the east to the freezing conditions of the Andes in the west. Throughout the year there is a regular alternation between a rainy season lasting from December to March and a dry season lasting from April to November (Country Reports 2023). In the Altiplano, very cold winds blow year-round, which significantly affect life here. Daytime temperatures range between 7 and 16 °C, night-time temperatures fall below freezing and rainfall is relatively variable throughout the year. In contrast, the lowland Oriente has a tropical climate with temperatures around 25 °C or slightly higher and abundant rainfall throughout the year. The highest rainfall totals are monitored in the northern part, where annual rainfall ranges from 1,800 mm or more. Also in the Oriente area, strong cold winds called surazos occasionally blow in from the south, bringing with them sharp changes in temperature (Arnade & McFarren 2023).

4.2.3. Flora of Bolivia

Bolivia's flora is one of the most diverse in the world due to the natural conditions prevailing in the country. The partly encroaching Amazonian tropical rainforest, plains, valleys, and mountains are the source of great diversity of plants, some of which are very rare and grow endemically (Meneses et al. 2015). Interesting native species include the quishuara and klena trees that grow in the Altiplano. In the Yungas area we find the quinine tree, a native species from which quinine is produced. Other important species is the coca bush and the characteristic saúco tree, whose fruit is used to make medicinal syrups (Arnade & McFarren 2023). The vascular plants are relatively numerous with approximately 17,000 species described so far, of which 2,343 are endemic. Finally, it is important to note that current knowledge of Bolivia's flora is far from complete, as studies only began in the late 20th century (Meneses et al. 2015).

4.3. Neglected and underutilised species

The diets of people around the world are based on a very narrow range of plant species, of which three basic types dominate: wheat, rice, and maize. The human population continues to grow, which causes an increasing demand for more food to be

produced. However, in the future it will be a problem to ensure food security based mainly on the production of these three species, which ultimately do not even provide a sufficient supply of nutrients for the body, and even their production has its limits (Mayes et al. 2012). Continuous improvements in the production of staple foods have reduced the number of people on the planet suffering from hunger, but at the same time have worsened the nutritional diversity of the population. Poor diets are one of the main causes of malnutrition, which according to the FAO, affects nearly two billion people. Current food production systems also have a negative impact on the environment. They cause loss of biodiversity, pollution of ecosystems, contamination and overuse of water, and land degradation. A shift towards healthier farming is desirable for several reasons and neglected and underutilised species (NUS) appear to be the ideal solution to these serious problems (Hunter et al. 2019). NUS is the name given to indigenous plant species that are cultivated in certain areas and represent a hidden and under-utilised potential in terms of their nutritional quality. They form an important part of the food and culture of the people who have cultivated them for many centuries (Mayes et al. 2012).

In the world, approximately 7,000 food plant species were used at least to a small extent, which is still only a fraction of the edible species found in the nature. Around 30 plant species are used to meet 95% of current food demand, which is very few compared to the potential (Janick 1999). The decline in food diversity has been driven primarily by political and socio-economic reasons. It is linked to the beginning of the Green Revolution, which saw the transformation of agriculture into industrial farming systems focused on monoculture crops that are highly profitable and can be grown on a large scale. This move began to deepen the displacement of NUS, which however have a higher nutritional profile than the species prevalent in the market. They contain more macronutrients and micronutrients that are necessary for the proper functioning of the human body (Hunter et al. 2019).

Interest in NUS is steadily increasing precisely because of efforts to change current farming practices in the face of uncertain prospects for sustainability. NUS are a resource of increasing diversity in agriculture, improving land use and avoiding land degradation. At the same time, the improvement of NUS cultivation techniques represents the creation of a job opportunity and economic income for the poor and therefore the alleviation of their difficult living situation, as NUS are mostly grown in economically

very poor areas. There is need to start exploiting the full potential of these crops. The aim is not to introduce NUS cultivation directly into new areas, but rather to devise workable ways of maximising their potential and use in existing areas. There is a need to make NUS a sustainable and nutritious food source to meet the dietary needs of the population and avoid malnutrition problems in these areas (Janick 1999).

Bolivia is one of the most biologically rich and diverse of plant species in the world despite the adverse climatic conditions of the Andes, which occupy a large part of the country. The Bolivian region is also one of the main centres of origin of important cultivated plants grown worldwide. The Andean region has specific conditions that do not allow the cultivation of crops such as wheat, rice, or maize. The present frost, drought, wind, poor and saline soils are stressors that only native species can accept. The people of Bolivia and Peru have always had a diet based mainly on plant-based foods, and diversity in the consumption of different types of crops has been very important to them. Tubers, legumes, cereals, and fruits are the main crops used. The Andean region is otherwise very poor, as the inhabitants grow crops for their own consumption and sell only a tiny fraction of the total production. The cultivation of these crops stems from a long tradition and is a part of the culture of the local people. These crops provide an extremely high-quality diet, rich in macronutrients and micronutrients, and are a way of improving the nutrition of the world's population (Padulosi et al. 2014).

4.3.1. Selected plant species in Bolivia

In total, data on 52 different plant species were collected in Table 1. These selected plants represented representatives from a total of 24 different families, of which Fabaceae was the most abundant family with 14 plant species (26.9%). The second most represented family was Amaranthaceae with 5 representatives (9.6%). This was followed by Arecaceae, Euphorbiaceae, Cucurbitaceae, Asteraceae, and Malvaceae all with 3 representatives (6.3%). The remaining families had a maximum of 2 representatives.

The most common source of protein were seeds, which also had the highest protein content. Other sources of protein were leaves, roots, fruit, tubers, or flowers. In general, for the higher protein content, the plants need to be dried or processed into flour. However, the protein values in the dried parts of the plants or in the flour were not included in the table.

Table 1. Selected plant species in Bolivia

	Scientific name	English name	Local name	Family	Edible parts	Protein g/100g	Use	References
1)	<i>Amaranthus caudatus</i> L.	Amaranth Love-lies-bleeding	Millmi Coimi	Amaranthaceae	grain leaves	12 – 16	cooked flour	1
2)	<i>Amaranthus hypochondriacus</i> L.	Prince's-feather	Quintonil Quelite	Amaranthaceae	grain seeds leaves	13 – 19	roasted flour	2, 52
3)	<i>Anacardium occidentale</i> L.	Cashew nut	Acajú	Anacardiaceae	nuts	21	fresh roasted powder	2, 20
4)	<i>Arachis hypogaea</i> L.	Peanut Groundnut	Cacahuete	Fabaceae	seeds	22 – 30	fresh cooked roasted	4, 40
5)	<i>Arracacia xanthorrhiza</i> Bancr.	Arracacha White carrot	Arracacha	Apiaceae	roots leaves	9	cooked fried roasted	2
6)	<i>Astrocaryum aculeatum</i> G.Mey.	Tucuma of Amazonas	Tucuma	Arecaceae	fruit pulp	9	fresh	1, 14
7)	<i>Bactris gasipaes</i> Kunth.	Peach palm	Tembé Palma de Castilla	Arecaceae	fruit pulp	5 – 9	cooked flour	1
8)	<i>Bertholletia excelsa</i> Bonpl.	Brazil nut	Castaña de Brazil	Lecythidaceae	nuts	12 – 17	fresh flour	1, 2
9)	<i>Bixa orellana</i> L.	Achiote tree Annatto Lipstick tree	Achiote Annatto	Bixaceae	seeds	13.70 – 14.80	flour	2, 32
10)	<i>Cajanus cajan</i> (L.) Huth	Pigeon pea No-eye pea	Guandul	Fabaceae	seeds	20 – 22	dried cooked toasted flour	46

(continued)

Table 1. Selected plant species in Bolivia (continued)

	Scientific name	English name	Local name	Family	Edible parts	Protein g/100g	Use	References
11)	<i>Canavalia ensiformis</i> (L.) D.C.	Jack bean	Chichasaro lima bean	Fabaceae	seeds	23 – 34	cooked flour	2, 23
12)	<i>Canna edulis</i> L.	Achira	Achira	Cannaceae	tubers roots	12	dried cooked flour	2, 36
13)	<i>Caryocar Brasiliense</i> Cambess.	Pequi Souari nut	Piquiá	Caryocaraceae	seeds pulp leaves	25.27	fresh cooked roasted flour	4, 38
14)	<i>Caryodendron orinocense</i> H.Karst.	Cacay nut Orinoconut Tacay	Castanhola	Euphorbiaceae	nuts	20	fresh toasted flour	1, 3, 19
15)	<i>Cucurbita ficifolia</i> Bouché	Black-seed squash Malabar ground Big leaf squash	Lacoyote Chilacayote	Cucurbitaceae	seeds fruit flowers	25.20 – 37	cooked roasted flour	2, 35
16)	<i>Cucurbita maxima</i> Duchesne	Squash	Zapallito	Cucurbitaceae	seeds fruit flowers	39.25	cooked roasted	2, 34
17)	<i>Erythrina edulis</i> Triana ex Micheli	Basul	Pajuro	Fabaceae	seeds	18 – 25	dried cooked flour	2, 24
18)	<i>Erythroxylum coca</i> Lam.	Coca	Coca	Erythroxylaceae	leaves	20.28 ± 1.65	raw dried flour	4, 37
19)	<i>Glycine max</i> (L.) Merr.	Soybean	Haba de soja	Fabaceae	seeds	39.40 – 44.40	cooked flour	44
20)	<i>Helianthus annuus</i> L.	Sunflower	Inti wayta	Asteraceae	seeds	33.85 ± 0.88	fresh roasted	2, 27

(continued)

Table 1. Selected plant species in Bolivia (continued)

	Scientific name	English name	Local name	Family	Edible parts	Protein g/100g	Use	References
21)	<i>Hymenaea courbaril</i> L.	Courbaril	Jatobá	Fabaceae	seeds pulp	11.50 ± 0.19	fresh dried flour	1, 2, 11
22)	<i>Chenopodium pallidicaule</i> Aellen	Canihua Kaniwa	Cañahua Qañiwa	Amaranthaceae	seeds leaves	15 – 19	flour	1, 2
23)	<i>Chenopodium quinoa</i> Willd.	Quinoa	Kinuwa Jiura	Amaranthaceae	seeds leaves	14.40 – 19	cooked flour	1, 2, 6, 7
24)	<i>Inga edulis</i> Mart.	Ice-cream bean	Pacay	Fabaceae	seeds pulp	11	fresh dried cooked	51
25)	<i>Jatropha curcas</i> L.	Physic nut Poison nut Purging nut	Curcas	Euphorbiaceae	seeds	32.88	cooked roasted	3, 30
26)	<i>Lagenaria siceraria</i> (Molina) Standl	Bottle ground Calabash	Guacal Jicara	Cucurbitaceae	seeds leaves flowers	19.25 ± 1.01	cooked toasted	2, 33
27)	<i>Lecythis pisonis</i> Cambess.	Cream nut Paradise nut Monkey nut	Sapucaia	Lecythidaceae	nuts	18 – 29	fresh cooked flour	4, 39
28)	<i>Lepidium meyenii</i> Walp.	Maca Pepper grass Pepper weed	Maca	Brassicaceae	hypocotyl roots leaves	10 – 14	dried cooked roasted	2
29)	<i>Lupinus mutabilis</i> Sweet	Andean lupin	Tarwi Tauri Chuchus	Fabaceae	seeds	32 – 52.60	cooked dried flour	2, 53
30)	<i>Macadamia integrifolia</i> Maiden & Betche	Macadamia nut Bush nut Bauple nut	Macadamia	Proteaceae	nuts	7.80 – 9.20	fresh dried roasted	43

(continued)

Table 1. Selected plant species in Bolivia (continued)

	Scientific name	English name	Local name	Family	Edible parts	Protein g/100g	Use	References
31)	<i>Madia sativa</i> Molina	Coast tarweed	Tarweed	Asteraceae	seeds	28 – 31	fresh cooked	29
32)	<i>Mauritia flexuosa</i> L.f.	Moriche palm	Buriti	Arecaceae	seeds pulp	11	fresh cooked flour	1, 2
33)	<i>Mirabilis expansa</i> (Ruiz. & Pav) Standl.	Mauka	Mauka Chago	Nyctaginaceae	roots leaves	7 – 17	cooked	2
34)	<i>Oxalis tuberosa</i> Molina	Oca	Uqa	Oxalidaceae	tubers	8 – 10	dried cooked roasted flour	2, 8
35)	<i>Pachyrhizus ahipa</i> (Wedd.) Parodi	Andean yam bean	Ajipa	Fabaceae	seeds roots	25.20 – 31.40	fresh dried cooked flour	2, 12, 13
36)	<i>Pachyrhizus tuberosus</i> (Lam.) Spreng.	Amazonian yam bean	Jacatupe	Fabaceae	seeds leaves tubers	29 – 32	fresh cooked flour	2, 31
37)	<i>Persea americana</i> Mill.	Avocado Alligator pear	Palta	Lauraceae	seeds fruit	15.55 ± 0.36	fresh cooked soaked powder	42
38)	<i>Phaseolus lunatus</i> L.	Lima bean Butter bean	Haba lima	Fabaceae	seeds	15.93 ± 0.55	cooked flour	2, 25
39)	<i>Phaseolus vulgaris</i> L.	Common bean	Nuña	Fabaceae	seeds	16.50 – 25.20	cooked	2, 26
40)	<i>Plukenetia volubilis</i> L.	Mountain peanut Inca peanut	Sacha inchi	Euphorbiaceae	seeds leaves	25 – 30	roasted cooked dried	41, 54

(continued)

Table 1. Selected plant species in Bolivia (continued)

	Scientific name	English name	Local name	Family	Edible parts	Protein g/100g	Use	References
41)	<i>Psophocarpus tetragonolobus</i> (L.) D.C.	Winged bean Asparagus bean Goa bean	Frijol alado	Fabaceae	seeds tubers leaves flowers roots	32 – 37	fresh cooked	48
42)	<i>Salvia hispanica</i> L.	Chia	Chia	Lamiaceae	seeds	15 – 25	dried	2, 9
43)	<i>Smallanthus sonchifolius</i> (Poepp.) H. Rob.	Yacon	Yacón	Asteraceae	leaves tubers	17 – 21	fresh dried cooked	10
44)	<i>Spondias mombin</i> L.	Yellow mombin	Taperebé	Anacardiaceae	leaves seeds fruit	7.70 – 11	fresh roasted	2, 21, 22
45)	<i>Theobroma bicolor</i> Humb. & Bonpl.	Jaguar tree	Balamte Macambo	Malvaceae	seeds	24	toasted powder	1, 15
46)	<i>Theobroma cacao</i> L.	Cocoa	Kakawa	Malvaceae	seeds	10 – 15	fermented dried	1, 16
47)	<i>Theobroma grandiflorum</i> (Willd. ex Spreng.) K.Schum.	Capuacu	Capuacu	Malvaceae	seeds	8.80	flour	1, 17, 18
48)	<i>Tropaeolum tuberosum</i> Ruiz. & Pav.	Mashwa	Añu	Tropaeolaceae	tubers leaves flowers	6.90 – 15.70	cooked roasted	2, 28
49)	<i>Ullucus tuberosus</i> Caldas.	Ullucu Oca quina	Olluco Papalisa Lisa	Basellaceae	tubers leaves	7 – 14	fresh cooked	2
50)	<i>Vigna luteola</i> (Jacq.) Benth.	Cowpea Hairy cowpea	Bajuco marullero	Fabaceae	seeds flowers roots	22.20	cooked	45

(continued)

Table 1. Selected plant species in Bolivia (continued)

	Scientific name	English name	Local name	Family	Edible parts	Protein g/100g	Use	References
51)	<i>Xanthosoma sagittifolium</i> (L.) Schott	New cocoyam	Walusa	Araceae	tubers leaves	8.48 – 10.10	cooked flour	5, 47
52)	<i>Zea mays</i> L.	Maize Corn	Mahis	Poaceae	seeds	7 – 13	fresh cooked flour	49, 50

1. (Shanley & Medina 2011), 2. (Food and Agriculture Organization of the United Nations 1995), 3. (Heller 1996), 4. (Shepard et al. 2020), 5. (Barthlott & Winiger 1998), 6. (Pereira et al. 2019), 7. (Angeli et al. 2020), 8. (Holban & Grumezescu 2018), 9. (Mohd Ali et al. 2012), 10. (Lachman et al. 2003), 11. (Pereira Santos et al. 2020), 12. (Doperto et al. 2011), 13. (Leidi et al. 2003), 14. (Santos et al. 2018), 15. (Olander 2017), 16. (Bertazzo et al. 2011), 17. (Rogez et al. 2004), 18. (Salgado et al. 2011), 19. (Reckin 1983), 20. (Ogunwolu et al. 2009), 21. (Igwe et al. 2010), 22. (Esua et al. 2016), 23. (Akpapunam & Sefa-Dedeh 1997), 24. (Intiquilla et al. 2016), 25. (Palupi et al. 2022), 26. (Celmeli et al. 2018), 27. (Petraru et al. 2021), 28. (Campos et al. 2018), 29. (Schmeda-Hirschmann 1995), 30. (Abou-Arab & Abu-Salem 2010), 31. (Kisambira et al. 2015), 32. (Wurts & Torreblanca R. A. 1983), 33. (Hassan et al. 2008), 34. (Mythili Md & Kavitha Md 2017), 35. (Falfan-Cortés 2016), 36. (Benjamin Caballero 2003), 37. (Penny et al. 2009), 38. (Ascari et al. 2013), 39. (Teixeira et al. 2018), 40. (Toomer 2018), 41. (Sethuraman et al. 2020), 42. (Ejiofor et al. 2018), 43. (Borompichaichartkul et al. 2009), 44. (Sharma et al. 2014), 45. (Harouna et al. 2018), 46. (Solomon et al. 2017), 47. (Wada et al. 2019), 48. (Bassal et al. 2020), 49. (Zambrano et al. 2021), 50. (Cuevas-Rodríguez et al. 2004), 51. (Lim 2012), 52. (Sánchez-López et al. 2020), 53. (Carvajal-Larenas et al. 2016), 54. (Wang et al. 2018)

4.3.2. Selected plant species

A total of four species were selected, *Chenopodium quinoa* Willd., *Lupinus mutabilis* Sweet, *Plukenetia volubilis* L., and *Caryocar Brasiliense* Cambess. These selected species represent a quality source of protein and the natural conditions prevailing in Bolivia are very favourable to them. For these reasons, they were selected as suitable species that have significant potential in expanding their cultivation, which would gradually improve the situation of protein deficiency in the diet of the local population and have the potential to be sustainable crops and nourish the local population for many years to come. These selected species have been described in more detail in the following section.

4.3.2.1. *Chenopodium quinoa* Willd.



Figure 3. *Chenopodium quinoa* Willd. (Inkanat 2021)



Figure 4. *Chenopodium quinoa* Willd. seeds (Hofacker 2023)

Botanical name: *Chenopodium quinoa* Willd.

English name: Quinoa

Local name: Kinuwa, Jiura

Family: Amaranthaceae

Biotope: Quinoa is plant native to the Andean region, where it has been cultivated for centuries in Bolivia and Peru. It is a plant with very good adaptation and high resistance to abiotic stress, which allows it to be grown in a variety of conditions, including very adverse ones (Pereira et al. 2019).

Description: Quinoa is classified as a pseudocereal. It has a strong, woody stem bearing broad leaves. The plant grows up to at least 1.5 metres and matures between 90 and 125 days. It produces seeds which form in clusters on the panicle and are red, pink, orange, yellow, white, or black in colour (Oelke et al. 1992).

Nutritional value: Quinoa is a very useful crop for mankind as it has an excellent nutritional value. It contains large amounts of two important macronutrients, carbohydrates, and proteins, but also carotenoids, vitamins, minerals, and other important micronutrients. The protein content (**14.40 – 19%**) is of high quality as it includes sufficient lysine and methionine supply, which are shortages in cereals (Bhargava et al. 2006).

The seeds are processed in different ways. They can be cooked, added to soups or salads, milled into flour, and then baked into tortillas or pastries. Seeds can be also fermented to make a beer (Pereira et al. 2019). The whole plant can be used as green fodder for livestock. Interestingly, NASA has selected quinoa as a suitable food ingredient for astronauts (Bhargava et al. 2006).

4.3.2.2. *Lupinus mutabilis* Sweet



Figure 5. *Lupinus mutabilis* Sweet (CABI 2022)

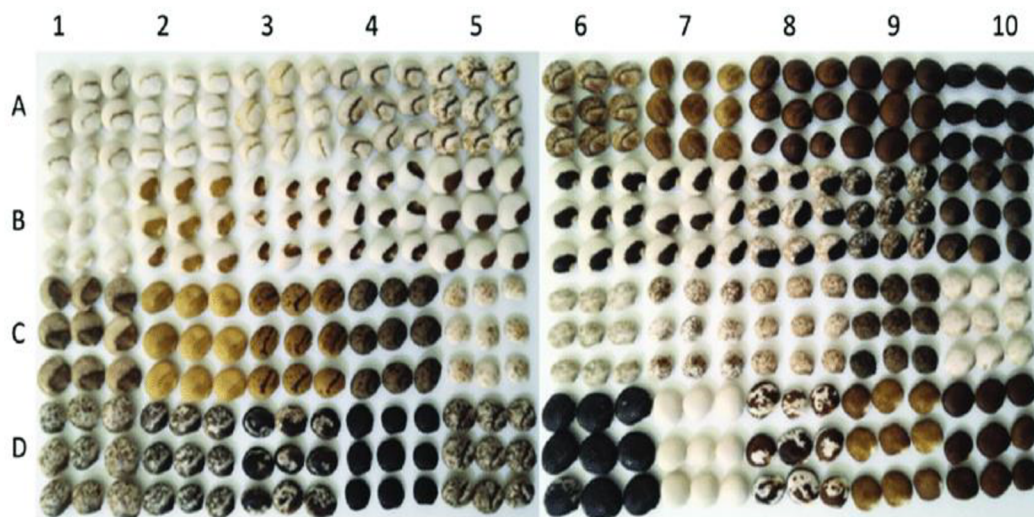


Figure 6. Variants of *Lupinus mutabilis* Sweet seed coat (Guilengue et al. 2022)

Scientific name: *Lupinus mutabilis* Sweet

English name: Andean lupin

Local name: Tarwi, Tauri, Chuchus

Family: Fabaceae

Biotope: Andean lupin is a plant capable of growing in a wide range of altitudes (1,500 – 3,800 m) and is found mainly in the Andean region. The crop is adapted to grow in the tropics, subtropics, and temperate zone (Gulisano et al. 2019).

Description: Tarwi is a legume and is grown for its highly nutritious seeds, which colour range from black to white. Depending on the genotype and the environment in which it is grown, it reaches a height of 0.40 – 2.50 m. The growing cycle is relatively long (240 – 300 days) and influenced by the amount of rainfall, which ranges from 350 – 800 mm per year. An amount close to the upper limit is ideal. Suitable are loamy-sandy soils with good nutrient balance and surface (Food and Agriculture Organization of the United Nations 1995).

Nutritional value: Seeds have a very high nutritional profile and achieve similar values to soybeans. In addition to their high protein content (**32 – 52.60 %**), they also contain a lot of oil. The amino acid content of lysine and cysteine is particularly high. The seeds contain a certain number of alkaloids, which can be got rid of by soaking the seeds overnight before cooking (Carvajal-Larenas et al. 2016).

The most common way of processing the seeds is to cook them and then eat them as part of traditional dishes. The seeds are also used to make flour, which is mainly used to bake bread but also other products. The flour has a higher protein content than cooked seeds and its use significantly improves the protein and caloric value of the resulting product (Food and Agriculture Organization of the United Nations 1995).

4.3.2.3. *Plukenetia volubilis* L.

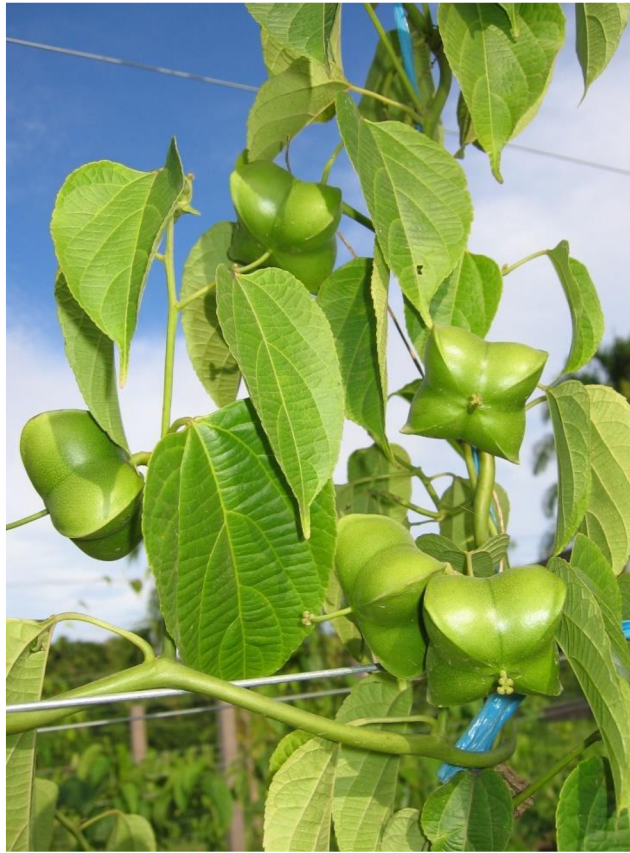


Figure 7. *Plukenetia volubilis* L. (NusHub 2006)



Figure 8. Fruit and seeds of *Plukenetia volubilis* L. (Shanantina 2020)

Scientific name: *Plukenetia volubilis* L.

English name: Inca peanut, Mountain peanut

Local name: Sacha inchi

Family: Euphorbiaceae

Biotope: Sacha inchi is a plant native to the Amazonian tropical rainforest. It is a tropical plant as it is ideally suited to temperatures between 25 – 30 °C. Plant is adapted to grow at altitudes between 200 – 1500 m and requires plenty of sunlight for proper growth and development (Wang et al. 2018).

Description: It is a perennial and climbing plant that reaches a height up to 2 m. It has hairy leaves that are approximately 10 – 12 cm long and 8 – 10 cm wide. The fruit is a star-shaped capsule, which is green at first but turns black-brown as it matures. Inside each asterisk are 4 – 6 edible oval seeds, dark brown in colour (Preedy et al. 2011).

Nutritional value: Ripe seeds are an excellent source of high-quality protein (25 – 30%), which contains many essential amino acids. In addition to protein, they are also high in lipids with high levels of omega-3, omega-6, omega-9 fatty acids. They are a source of the highly regarded edible oil, which is produced from them and used extensively in cooking (Kodahl 2020). The seeds also contain abundant amount of minerals, antioxidants, and vitamin E (Wang et al. 2018).

The seeds contain terpenoids, saponins, and flavonoids, which are harmful to the human body and need to be removed before consumption. The seeds are most often roasted but can also be cooked. The leaves of Sacha inchi are also edible, but like the seeds they contain substances that need to be removed before consumption. The leaves are either cooked or dried for a long time and then made into tea (Srichamnong et al. 2018).

4.3.2.4. *Caryocar Brasiliense* Cambess.



Figure 9. *Caryocar Brasiliense* Cambess. fruit (Costa 2012)

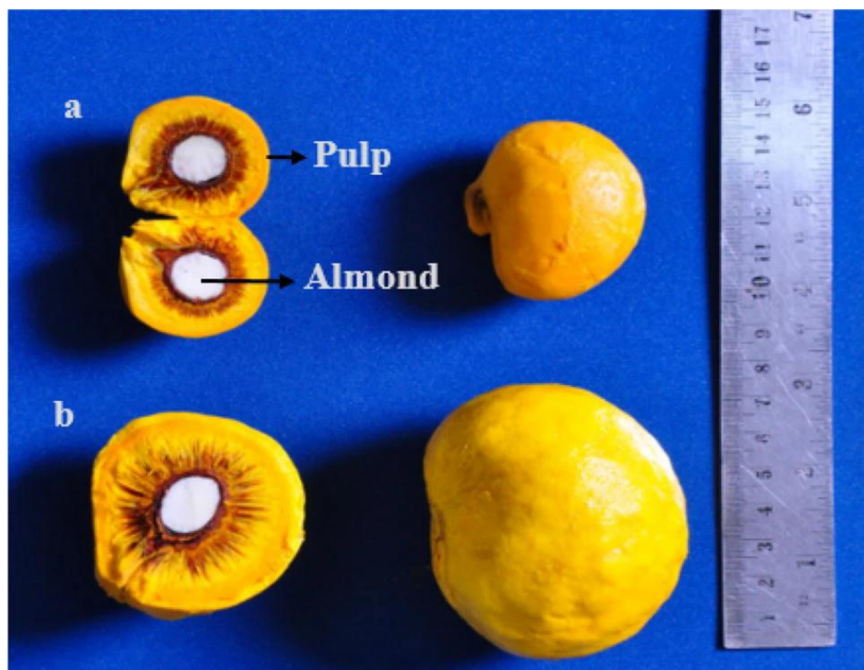


Figure 10. *Caryocar Brasiliense* Cambess. detail (dos Santos da Silva et al. 2020)

Scientific name: *Caryocar Brasiliense* Cambess.

English name: Pequi, Sounari nut

Local name: Piquiá

Family: Caryocaraceae

Biotope: Piquiá is a representative of subtropical and tropical mainly drier regions. It is adapted to grow in areas with nutrient-poor soils, mostly on plateaus and to a lesser extent in savannahs. For sufficient growth, it needs plenty of sunshine, 1000-1500 mm of rainfall per year and a dry season in which it flowers and fruits (Lorenzi 2002).

Description: *Caryocar brasiliense* Cambess. is a tree with a massive crown, growing to a height of 8 – 10 metres. It has a relatively thick trunk up to 40 cm in a diameter and a large hairy leaf. It is particularly useful for its fruits, which are seeds used for food and for oil production (Lorenzi 2002).

Nutritional value: Piquiá is a fruit of important economic importance. The most exploited part of the fruit is the pulp, which is an excellent reservoir of vitamins, proteins (25,27%) and lipids. It also contains natural antioxidants, namely carotenoid and phenolic compounds, which have a positive effect on the development and maintenance of human health, while reducing the risk of cancer, cerebrovascular and cardiovascular diseases (Paula et al. 2022).

The tasty and nutritious pulp and seeds are consumed. The pulp is eaten raw or as an ingredient in cooking. The traditional method is to prepare pequi with chicken and rice (Guedes et al. 2017). The seeds are covered with tiny spines that can injure the oral cavity when eaten (Lorenzi 2002). For this reason, the seeds are left to dry in the sun. After about two days of drying, the spines are scraped off and the seeds can be eaten. They are either extracted to make edible oil or roasted and then eaten (Guedes et al. 2017).

5. Conclusion

The aim of this thesis was to analyse the available literature on plants as a potential alternative source of proteins, specifically in the region of Bolivia. Currently, plants as a source of protein still do not receive the attention they deserve. However, they do offer a solution for economically weaker countries to provide people with a sufficient quality source of protein in their diet and alleviate malnutrition problems, which occur mainly in developing countries. Given the world's growing population and concerns about the sustainability of producing enough nutritious food in the future, they also represent a possible solution how to feed the world.

A total of 52 plant species belonging to 24 different families were analysed in this survey. The most represented family was the Fabaceae family with 14 representatives (26.9%), followed by the Amaranthaceae family with 5 representatives (9.6%), Arecaceae, Euphorbiaceae, Cucurbitaceae, Asteraceae, and Malvaceae families all with 3 representatives (6.3%). From these plants, 4 species were selected that contained high quality protein content and were also well adapted to the natural conditions of Bolivia, and their widespread cultivation had a great potential. More precisely, they were: *Chenopodium quinoa* Willd., *Lupinus mutabilis* Sweet, *Plukenetia volubilis* L., *Caryocar Brasiliense* Cambess. These species were described in more detail.

Seeds were the most common and highest quality source of protein. Other quality sources of protein were leaves, roots, and tubers. Fruits or flowers were the less nutritious parts, but still represent a useful and important component of many people's diets. The edible parts are eaten raw, cooked, or dried and ground into flour. The flour is then used to bake bread and many other products.

Proteins of animal origin will always be considered more nutritionally valuable and complete. However, their production puts a negative burden and impact on the environment and is not sustainable for the future. This is the moment, where plant proteins come into focus, as they represent a quality and healthier alternative to ensure adequate protein nutrition for the population now and for the future. With a lower environmental impact, with a great potential for expanding the cultivation of the species and overall, their production, in improving the bad situation of malnutrition in developing countries and, above all, sustainability.

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