



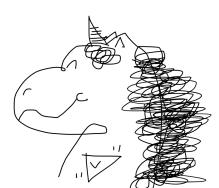
# Aroma Development and Volatile Composition in Mango (*Mangifera indica L.*)

A Theoretical Report on the Biochemical Pathways, Ripening Physiology, and Quality Determinants in mango.

**Lucas Daniel Paz Zuleta, TZS159**  
MSc students at the University of Copenhagen

Last compiled: 20-10-2025

[Link to Git repository](#)



## Preface

This written report has been prepared as part of the course NPLK14014U - Fruit and Berry Crop Physiology and Quality at the University of Copenhagen. The course explores the physiological and biochemical factors determining fruit development, yield, and quality in relation to fresh consumption and processing. Emphasis is placed on how genetic, environmental, and management factors influence internal and external fruit quality, including the formation of secondary metabolites and sensory attributes such as aroma.

The present project is a theoretical study on the development and composition of aroma in mango, *Mangifera indica L.*, The focus is on the biochemical formation of volatile organic compounds (VOCs) and their relationship to fruit ripening and sensory quality. The report integrates physiological and biochemical knowledge to describe how preharvest conditions, cultivar differences, and post-harvest handling affect aroma expression and the perception of fruit quality.

The assignment was carried out by Lucas Daniel Paz Zuleta (TZS159), MSc student at the University of Copenhagen, as an individual written report fulfilling the course requirements for NPLK14014U - Fruit and Berry Crop Physiology and Quality.

## Summary

### Signatures

Copenhagen, 20-10-2025



Lucas Daniel Paz Zuleta (TZS159)

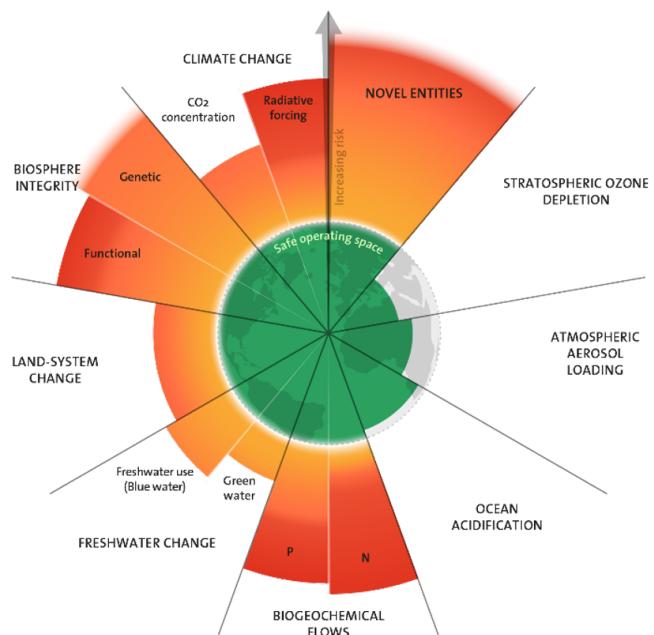
20-10-2025

# Contents

1	Introduction . . . . .	1
1.1	Aim and Objectives . . . . .	1
1.2	Market Trends & Target Consumer Group . . . . .	1
1.2.1	Existing Market . . . . .	2
2	Product Concept . . . . .	3
2.1	Macronutrients in Hemp Seedssc . . . . .	4
2.1.1	Protein . . . . .	5
2.1.2	Fatty Acids . . . . .	5
2.1.3	Carbohydrates . . . . .	5
2.2	Micronutrients: Vitamins and Minerals . . . . .	6
2.3	Potential Side Streams . . . . .	6
2.4	Dietary Pattern of the Chosen Consumer Group and Product Fit . . . . .	6
3	Formulation and Raw Materials . . . . .	7
4	Processing and Manufacturing . . . . .	8
4.1	Effects on Processing . . . . .	10
5	Final Nutritional Profile of the Bar . . . . .	11
5.1	Nutritional Composition Overview . . . . .	11
5.2	Macronutrient Composition . . . . .	11
5.2.1	Protein Content and Quality . . . . .	12
5.2.2	Dietary Fibre Content . . . . .	13
5.2.3	Fatty Acid Profile . . . . .	15
5.3	Comparison with Market Products - ROO'bar . . . . .	16
5.3.1	Macronutrients Comparison . . . . .	16
5.3.2	Protein Quality Comparison . . . . .	18
5.3.3	Dietary Fibre Comparison . . . . .	18
5.3.4	Fatty Acid Profile Comparison . . . . .	19
6	Environmental Impact - sustainability Perspective . . . . .	19
7	Conclusion . . . . .	20

# 1 Introduction

With an increasing population and an estimate of almost 10 billion by 2050. The future protein demand is projected to increase as a correlated factor to the population increase [1]. Proportionally, Makkar et al. 2014 predicts an increase in animal consumption of 60-70%. The increasing demand for animal protein risks further extending planetary boundaries and resulting in conflicts related to sustainability. The planetary boundaries framework by the Potsdam Institute for Climate Impact Research defines “a safe operating space for humanity”. It maps out nine Earth system processes critical for maintaining the planet’s stability and resilience, processes which are all affected by the Anthropocene (Figure 1.1) [2].



**Figure 1.1:** Schematic of the nine Planetary Boundaries [2].

Thus, humanity stands in front of a challenge in sustaining the supply of animal proteins. Meanwhile, the EAT-Lancet commission declares numerous arguments for a protein shift towards plant-based alternatives. Where human health and environment are two pillar arguments of the diet recommendations (Willett et al., 2019).

## 1.1 Aim and Objectives

The scope of this report narrows down to a nutrient-rich product with low water impact and low CO<sub>2</sub> footprint. The formulation targets two of the nine planetary boundaries, freshwater change and CO<sub>2</sub>-concentration. This report aims to present a hemp protein bar, which served as an alternative to animal-source protein bars, which could help in mitigating the transgression of these planetary boundaries.

## 1.2 Market Trends & Target Consumer Group

With animal sourced protein risking straining the planets’ resources, a shift in the traditional diet of the Nordic countries has been studied. On the very subject, Geirsdóttir et al. 2023 provided a thorough scoping-review on Nordic Nutrition Recommendations. They concluded that a shift towards a more plant-based protein diet

would benefit both health and the environment. Hence, given the need for a shift, the demand for plant-based protein is expected to increase, while the European meat consumption is expected to decline, the consumption still exceeds that of the respective countries' national recommendations (typically 300-500 g/week) [3]. The Smart Protein Project collects data which helps understanding the status and attitude towards a plant-based diet in various European Countries. Among many surveys, it is stated that "Plant-based sweets, meat alternatives, and milk substitutes emerge as the most sought-after categories for expanding plant-based options." This accounts for 27% of the cohorts "express for desire" of such product. Among the top 6 drivers for choosing these products, "health" and "environmentally friendly" are mentioned (45% and 21% respectively) [prenote 4, postnote].

Looking at market predictions and current trends, the global growth of plant-based protein supplements is currently outpacing the growth of that animal source. Which yet again correlates with the future need for sustainable plant-based protein [5]. The consumption of such is seen in either ready-to-eat products or supplements as concentrates or isolates. It is shown that nutritious / functional protein bars are an emerging market as in 2023, it was worth 0,92 billion \$ in Europe. Offering a convenient, ready-to-eat alternative to reach fitness goals. The main consumers of such products are found to be Millennials and Generation Z, with enthusiasm over high protein bars [6]. Hence, targeting younger consumers is of interest. Furthermore, a plant-based protein bar would target any consumer interested in reducing animal consumption, complementing their daily protein intake, and supplementing regular meals.

### 1.2.1 Existing Market

As development of plant-based fitness supplements and ready to eat products are increasingly popular, a wide range of products are available. Below is a showcase of an Estonian and Finnish producer, which specialize in hemp-based products and/or raw material. A showcase of a current hemp bar product is also presented.

#### Nordic Hemp, Estonia

Specializes in organic industrial hemp growing and processing of raw material. Production of 6000 ha / year [7].

- Sorting
- Dehulling
- Processing
- Protein isolation

#### Impolan Kasvitila, Finland

Impola plant farm is a family-owned company in its fourth generation [8]. They grow and produce product for end-consumers.

- Pet and Feed products
- Pressing
- Dehulling
- Hemp chocolate
- Hemp muesli
- Hemp meal



**Figure 1.2:** Hemp protein bar by Nordic Hemp. Selection of products from Impolan Kasvitila, Finland.

### ROO'bar by Smart Organic, Bulgaria

Roobar is the flagship brand by Smart Organic AD. They are the largest producer of “minimalistic plant-based bars.” They focus on “... 4-5 ingredients... organic, vegan, raw, and gluten-free”. The production is estimated to 1 million bars per month, and they are accessible in around 50 countries [9].

- Broad range of different bar products
- Owner of a wide range of ready-to-eat brands

**Table 1.1:** Ingredients: Dates, almonds, hemp protein (18%). Nutrient declaration per 100 g.

Energy	1582 kJ / 377 kcal
Fat	11 g
- Fatty acids	1.9 g
Carbohydrates	49 g
- Sugars	33 g
- Dietary fibre	11 g
Protein	14 g
Salt	0 g



**Figure 1.3:** ROO'bar hemp protein bar

## 2 Product Concept

It is estimated that only 35% of the globally produced plant protein is consumed by humans. Meanwhile the current market offers a wide range of plant-based proteins available which are well fitted for human consumption. Common choices include formulations with oat, wheat, hemp, soy and pea to name a few [10]. The Cannabis sativa L., a Cannabaceae known as “hemp” is of increasing interest because it is highlighted as an environmentally friendly and economically high-potential crop. Its history as a source for medicine, fiber and food dates back 6000 years, and the cultivation of hemp with close to no levels of tetrahydrocannabinol (THC), the psychoactive compound found in cannabis, has increased to use in foods since strains with a THC content below 0.3% was approved in EU in 1996. Among its many advantages are fast growth and low dependency on pesticides, which benefits biodiversity and healthy soils. Furthermore, its seeds provide a valuable source of nutrients [11]. Globally,

the trend of increasing interest for hemp seeds is clear, as the production increased from 2,718 tons to 5,449 tons between 2015 and 2020.

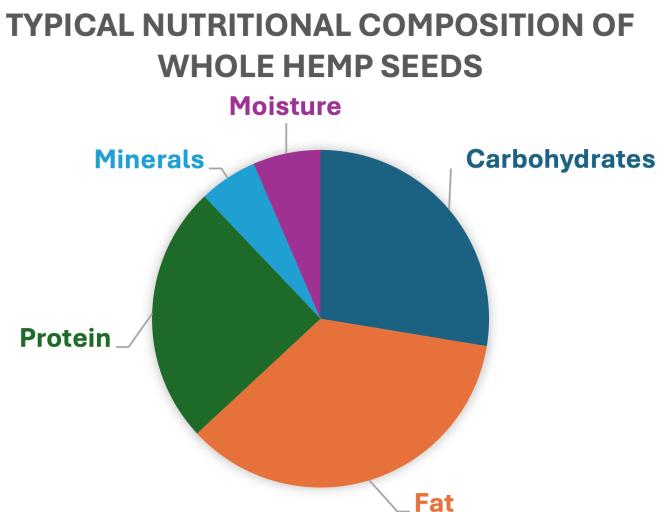
Thus, developing a convenient hemp-based bar would harness the many benefits of the not-so-novel ingredient that is hemp seed. Below is a mock up of the Hemp Protein Bar (Figure 1.4).



**Figure 1.4:** AI-generated illustration of a medieval marketplace. Generated using DALL-E 3 (OpenAI, 2025) with the prompt: "Mock up of a hemp bar picture half coved in chocolate".

## 2.1 Macronutrients in Hemp Seeds

Hemp seeds typically contain around 20-30% protein, 25%-35% lipids, 20-30% carbohydrates and 4-7% of ash (Figure 1.5). It has a balanced composition with particularly low starch content [12].



**Figure 1.5:** Composition diagram of whole hemp seeds

### 2.1.1 Protein

Hemp seeds are an excellent source of high-quality protein, typically containing 20-30% protein, with dehulled hemp seeds having even higher protein contents, ranging from 30% to 38% [11]. The two main proteins in hemp seeds are albumin (33%) and edistin (65%), which have very similar structures to plasma proteins which helps the digestibility in humans. Hemp seeds provide a complete amino acid profile, containing all nine essential amino acids required by humans, making them comparable to other high-quality proteins like egg white and soybeans [11].

The protein is also particularly rich in arginine, glutamic acid, and aspartic acid, where arginine is especially valuable as it is a precursor to nitric oxide, which enhances blood flow and helps maintain normal blood pressure, contributing to cardiovascular health [11]. Hemp proteins are highly digestible, with dehulled hemp seeds demonstrating superior protein digestibility (83.5-92.1%) [12]. This is partly attributed to the absence of protease inhibitors in hemp seeds [13].

A study has analysed the macronutrient composition and protein quality of 30 hemp seed products from Western Canada, including whole seeds, dehulled seeds, and hemp seed meal. Crude protein, fat, and amino acid profiles were determined, and protein quality was assessed using the Protein Digestibility-Corrected Amino Acid Score (PDCAAS) method, based on a rat bioassay and FAO/WHO amino acid requirements for young children. Average protein content ranged from 24.0% in whole seeds to 40.7% in hemp seed meal. Protein digestibility was 84-98%, with protein digestibility-corrected amino acid score (PDCAAS) values of 46-66%, highest in dehulled seeds. The protein digestibility-corrected amino acid score of dehulled hemp seed is comparable to lentils and is about half that of casein, and almost twice that of almonds [11].

### 2.1.2 Fatty Acids

Hemp seed oil is primarily composed of polyunsaturated fatty acids, PUFA (over 80%) including fatty acids like essential linoleic acid (omega-6) and alpha-linolenic acid (omega-3) [11]. Dehulled hempseeds have a healthy balance of omega-6 to omega-3 polyunsaturated fatty acids (2.5:1) [11].

Unsaturated fatty acids help protect against cardiovascular disease, obesity, diabetes, and inflammation. EFSA recommends an optimal omega-6/omega-3 ratio of 3:1 to 5:1. Hemp seed oil typically shows a ratio of 2.5-3.5:1, which is a desirable range linked to lower chronic disease risk [12].

### 2.1.3 Carbohydrates

About 98% of the carbohydrates in hemp seeds are dietary fiber, mainly insoluble dietary fibers (80%), such as cellulose, lignin and hemicellulose. Dietary fiber resists enzymatic digestion in the small intestine and undergoes partial or complete fermentation in the large intestine. The remainder of the carbohydrates in hemp seeds is starch. Therefore, hemp seeds are considered a low-starch food and an excellent source of dietary fiber. [12]

The dietary fibers from hemp seeds are associated with positive effects on the digestive tract support by acting as prebiotics. The fermentation of fibers in the colon generates short-chain fatty acids that have beneficial roles in the body. [13]. In the Western countries the consumption of dietary fiber is lower than the recommended intake, making hemp seeds an attractive ingredient to meet the recommended daily intake of dietary. However, processing might affect the amounts of dietary fibers in hemp seeds.

## 2.2 Micronutrients: Vitamins and Minerals

Hemp seeds are rich in vitamins and minerals. Just 50 mg of hemp seed can supply at least half of the recommended daily allowance of copper, magnesium, and zinc, and exceed the recommended daily allowance of the vitamins A, D, and E [11]. Hemp seeds also contain other micronutrients such as phosphorus, potassium, calcium, sodium, iron, and manganese [11]. Hemp seed oil contains fat-soluble vitamins, in particular vitamin E (tocopherols) and vitamin A which respectively has an antioxidant role and is beneficial for skin integrity and Vitamin D is important for bone health and the immune system [12]

Besides the macro- and micronutrients, secondary metabolites, such as terpenes, phytosterols and flavonoids, constitute essential components of the defence response of the hemp plant to biotic and abiotic stresses. However, the composition of these secondary metabolites can be influenced by cultivation conditions, providing a distinct fingerprint of different production regions. It is suggested that these compounds contribute with antioxidative, antimicrobial, and anti-inflammatory properties in the human body. Phytosterols for instance, are not synthesized in humans, but can when ingested from plants, reduce cholesterol levels in the human body by changing the cholesterol solubility in the intestine [14].

## 2.3 Potential Side Streams

The industrial hemp plant has a versatile plant body which consists of seeds, leaves, stem, and flowers with several application opportunities depending on the part of the plant. Particularly the stem is a valuable source to produce hemp fiber which can be used for rope, building materials, paper, or textiles. The seeds, dehulled or whole, can be utilized as a food source. The hemp flower can be used to produce cosmetic and pharmaceutical products, including essential oils. When looking further into hemp as a natural source to bast fiber a life cycle assessment reveals that hemp performs better than glass fiber by weight and compared to cotton, hemp requires less water and pesticides to grow although hemp fiber is known to be coarser and stiffer than cotton which has a softer appearance [15].

When processing the stem to hemp fiber a by-product of shives is made. It can be used to produce hemp concrete which is a bio-composite and carbon-negative alternative to concrete for construction and insulation [16]. These useful side streams make hemp even more attractive to use as an ingredient in foods.

## 2.4 Dietary Pattern of the Chosen Consumer Group and Product Fit

Our hemp seed bar is uniquely positioned to seamlessly integrate into several contemporary dietary patterns, directly addressing the needs and preferences of our diverse target consumer groups within the Millennial and Generation Z age group.

For vegetarians and vegans dehulled hemp seeds are an excellent source of high-quality, plant-based protein. The seeds naturally contain all nine essential amino acids required by humans, offering a complete protein profile that can be challenging to obtain from other plant sources [11]. The protein in dehulled hemp seeds also boasts superior digestibility (83.5%-92.1%) compared to whole hemp seeds and hemp meal, making its nutrients more accessible. They also provide a healthy balance of omega-6 to omega-3 polyunsaturated fatty acids (typically 2.5:1 to 3.5:1), which is desirable for overall human nutrition. [12] Hemp protein and flour can serve as an alternative for soy ingredients [17].

A health-conscious person or athlete would also use it for its excellent source of protein, especially because of

the high value protein composition with the nine essential amino acids. Furthermore, it scores high in terms of digestibility which makes our bar effective for muscle recovery and satiety [11]. Many athletes need to have control on their calorie needs. Our bar could therefore be an easy boost of calories or be used as a substitute for a snack or meal. The bar is rich in healthy fats, including the beneficial omega-6 to omega-3 ratio, which is important for cardiovascular health and may help prevent chronic diseases. [12]

Our bar is also a good source of essential minerals like copper, magnesium, and zinc, and provide vitamins A, D, and E, which also talks to the health-conscious person [11].

For individuals with dietary restrictions and/or allergies our bar is also a great option, as our bar is naturally gluten-free, making it a safe nutritious option for people with celiac disease or gluten sensitivities [11]. Hemp proteins are generally considered to have low allergenicity compared to common proteins like soy, dairy, or wheat, broadening its appeal for those with various food allergies [11, 12, 13].

For environmentally aware consumers our hemp-based product directly supports environmental sustainability due to it having a low environmental impact, actively contributing to improved soil health, water quality, have carbon-negative crops and requiring no or little pesticide use. (Chapter 1 hemp book & hemp nutritional value PDF & hemp seed bioactivity)

The pre-packaged protein bar format offers convenience and time efficiency, requiring no preparation or cleanup for the people on-the-go. It delivers the balanced nutrition derived from dehulled hemp seeds in an easily consumable form, perfectly fitting the needs of busy individuals seeking healthy and convenient dietary options.

### 3 Formulation and Raw Materials

The formulation of the hemp seed bar is an important factor, as it regulates the final nutritional composition and determines the parameters for the upstream and downstream processing steps. Using consistent suppliers and high-quality raw materials are key factors in maintaining a predictable, consistent production. Production as such would decrease faults, which reduces waste and ensures safe a safe high-end product to the final consumer.

Given the lack of sweetness of hemp seeds, the bar is formulated with naturally sweet components such as dried dates and agave sirup. Along with a caramelly flavor, important cohesiveness of the bar is achieved. The dates are dried which reduces water content and thus increases sugar concentration and intensifies flavor. Minimum processing other than dehydration and pitting ensures valuable components such as micronutrients and fibers are included.

To complement the protein value (which lacks in lysine), potato protein isolate is used. During its production, it undergoes heavy processing which concentrates the protein content while reducing other macronutrients, minerals and vitamins[18].

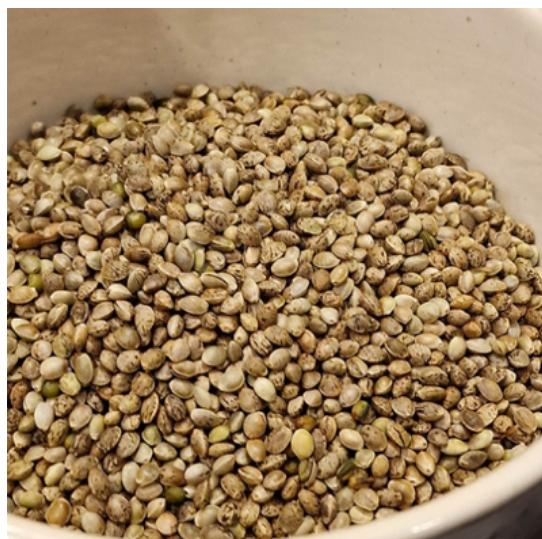
Rolled oats are used as a structural component. It is chosen as it is a gluten free, cheap, readily available raw material known to consumers. It undergoes dehulling, rolling and steaming, which maintains the high fiber content while inactivating lipase enzymes to prevent oxidation and extend shelf life [19]. Ground flax seeds are also included to account for the loss of dietary fibers in Hemp. The flax seeds are grounded to inhibit toxic effects of cyanogenic glycosides [20].

Preprocessing of hemp seeds aims to partially remove the hull/husk (Figure 1.6). The macronutrient profile of the

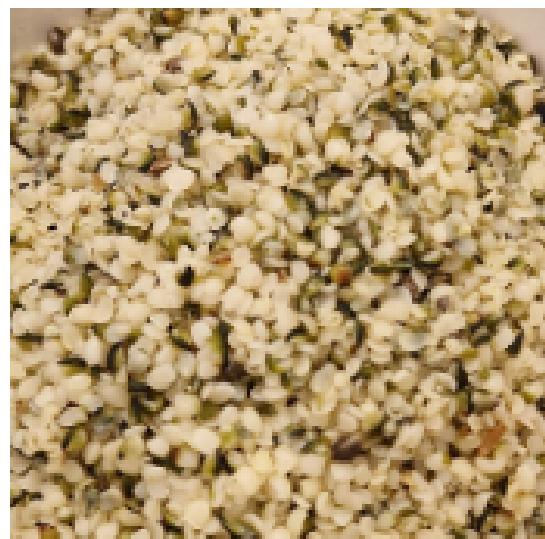
seed is significantly affected by the processing method. Although the formulation does not involve hemp protein isolate, HPI, many studies on this subject point at the effects of preprocessing.

For example, a study by House et al., 2010 evaluated HPI derived from whole seeds, hemp seed meal, and solely hulls. The study could show that the amino acid composition is different among the three processed raw materials. In the works by Shen et al., 2020 the HPI of dehulled and whole seeds were comprehensively investigated in terms of aromatic components, colour, and protein characterization. Dehulled seeds would increase the extraction yield by 21.52 % and protein recovery yield (46.90%) of the HPI. Naturally, the HPI with whole seeds would contain increased amounts of lipids and carbohydrates. The preprocessing also had a profound impact on colour, where whole seeds generated a darker coloured HPI. This matter is further explained in the section “4.1 Effects on Processing”

Vanilla extract, cocoa and sea salt are used to characterize the aroma of the bar. Potentially masking some of the earthy off-flavours from the other ingredients.



(a) Whole hemp seeds



(b) Dehulled hemp seeds

**Figure 1.6:** Whole and dehulled hemp seeds (Svensk Hampaindustri, 2025).

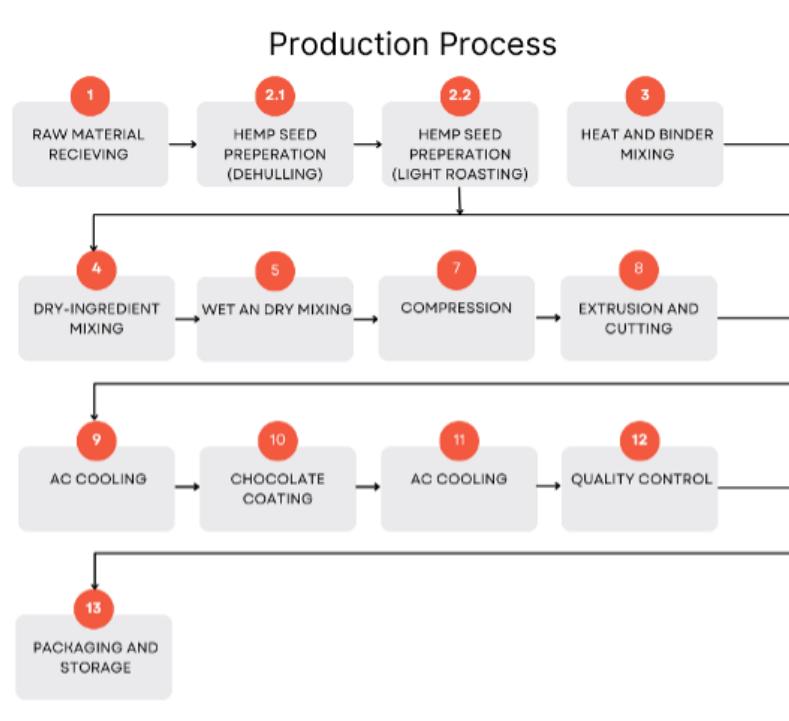
## 4 Processing and Manufacturing

The production process of the protein bar can be seen on Figure 1.7.

*Sourcing and preparing* raw materials - all ingredients are inspected for quality and stored under appropriate conditions to maintain freshness. All ingredients are weighed and portioned accurately according to the recipe formulation, ensuring batch-to-batch uniformity. This ensures that only safe, high-quality, and properly measured ingredients enter the mixing stage, laying the foundation for a consistent protein bar in terms of flavor, texture, and nutritional profile.

*Dehulling hemp seed* - Increases absorption of proteins and nutrients.

*Light roasting* - of the dehulled hemp seed (75°C-80°C) - improves digestibility, and reduces anti-nutritional factors and lower microbial load.



**Figure 1.7:** A flowchart with a visualization of the production process of the hemp protein bar

*Mixing* to homogeneous mixture - evenly distributed throughout the mixture providing a consistent texture and flavor in every bar. Time and temperature in mixing are controlled to ensure that every batch is consistent.

*Compression* - shaping the mixture into a form that is suitable for further processing. Using large presses ensures a uniform sheet, which helps create consistency in texture and flavor throughout the bar.

*Extrusion and cutting* - A combination of extrusion and cutting technologies are used to shape the mixtures into bars. The extrusion process forces the mixture through a die, after which a knife cuts the long bars into the desired length.

*AC cooling* - is used to ensure that the bar maintains the desired temperature throughout the process, as mixing and pressure from extrusion can increase the temperature of the product.

*Chocolate coating* - for flavour.

*AC cooling* - to solidify the chocolate.

*Quality control* - Both automated sensors and visual inspection for defects or foreign objects. Taking samples to ensure that our product meets quality standards, as well as for example checking that the product contains enough protein to be claimed as a product with a high protein content.

*Packaging* - wrap each bar individually and in a material that keeps the protein bar safe and to ensure that the bar does not undergo oxygenation and to preserve freshness. This process also includes labelling and coding to ensure traceability and compliance with regulatory requirements.

## 4.1 Effects on Processing

Hemp seeds contain several antinutritional, such as phytic acid, tannins and saponins. The presence of tannins and saponins can reduce the bioavailability of nutrients and disrupt both metabolism and digestive functions [11]. The presence of phytic acid can lead to mineral deficiencies (e.g. iron, zinc and calcium), as it can inhibit the absorption of these [11]. Polyphenols, of which tannins are a part, are often found in the shell of hemp seeds [12]. The same applies to phytic acid. Studies have shown that there is significantly more phytic acid present in whole hemp seeds 3.5 g/100g compared to 2.1 g/100g in hulled hemp seeds. That is a reduction of 40% [11].

According to House et al. (2010), whole hemp seeds have a protein digestibility of approximately 84-86% and a protein digestibility-corrected amino acid score (PDCAAS) value of 49-53%, while dehulled hemp seeds reach a digestibility of 91-97% and a PDCAAS value of 63-66%. This improvement is primarily due to the fact that the hull contains a large part of the fiber fraction of the seed, which inhibits digestibility, so when the hull is removed, the fiber content is significantly reduced, making the protein more available and thus improving the overall protein assessment (PDCAAS) [21]. Therefore, by using a majority of dehulled hemp seeds instead of whole hemp seeds, we reduce the content of antinutrients and increase the nutritional quality of the protein.

Plant proteins naturally contain antinutritional factors such as trypsin inhibitors, glucosinolates, phenols and phytates, together with a high content of dietary fiber, which can negatively affect protein and amino acid digestibility and bioavailability. Heat processing can effectively help to remove or reduce these compounds, leading to higher protein digestibility. However, high heat treatment can have negative effects as it can affect the chemical transformations of amino acids. Some amino acids, such as lysine, can be chemically transformed and become unavailable during heat treatment or other severe processes, leading to problems such as the formation of Maillard reaction products. This underlines the need for appropriate processing conditions to avoid such [22].

**Table 1.2:** Protein digestibility-corrected amino acid scores of hemp protein sources in comparison to other food proteins [21].

Protein source	PDCAAS (%)
Casein	100
Egg white	100
Beef	92
Soy protein isolate	92
Chickpeas (canned)	71
Pea flour	69
Kidney beans (canned)	68
Dehulled hemp seed	61
Pinto beans (canned)	57
Rolled oats	57
Lentils (canned)	52
Hemp seed	51
Hemp seed meal	48
Whole wheat	40
Almond	23

At the same time, a study has shown that hemp protein isolates improved proteolysis when heated at 75 °C and 80 °C, but already at 90 °C it caused reduced proteolysis. [13] Therefore, we have chosen to lightly roast our dehulled hemp seeds to achieve the highest possible protein absorption, but with an eye not to reach too high a temperature, as we want to avoid a greater loss of lysine and reduced proteolysis.

## 5 Final Nutritional Profile of the Bar

### 5.1 Nutritional Composition Overview

In this section of the report, the macronutrient composition of the hemp seed protein bar is examined, namely protein, dietary fibre, and fatty acids. The final nutritional profile was estimated based on the formulation and calculated contributions of each ingredient. Table 1.3 shows the magnitude of each ingredient in the bar, where the highlighted cells represent the top contributors that will be examined in greater detail. This section further outlines the amino acid and fatty acid spectrum, evaluates potential nutrition and health claims considering EU regulations and EFSA opinions, and compares the bar's profile to existing market products.

**Table 1.3:** The table indicates the 10 ingredients that the product is made of, and the respective values for three of the most notable macronutrients and moisture content. The top five contributor for each of the factors is highlighted in green, ranging from dark to light-green from highest to lowest value corresponding to the amount of the macronutrient in the bar.

No.	Ingredient	Per bar [g]	Protein [g/100g]	Dietary fibre [g/100g]	Fat [g/100g]	Moisture [g/100g]	Carbohydrates
1	Dates (pitted and dried)	16.50	2.00	6.40	0.50	18.40	nd
	Contribution		0.33	1.06	0.08	3.04	nd
2	Rolled Oats	10.00	12.90	9.90	6.90	10.00	nd
	Contribution		1.29	0.99	0.69	1.00	nd
3	Potato Protein Isolate	8.50	93.20	0.20	0.10	7.00	nd
	Contribution		7.92	0.02	0.01	0.60	nd
4	Dehulled Hemp Seeds	6.50	25.93	4.58	52.07	5.50	nd
	Contribution		1.69	0.30	3.38	0.36	nd
5	Agave Syrup	5.00	0.11	0.00	0.87	18.93	nd
	Contribution		0.01	0.00	0.04	0.95	nd
6	Whole Hemp Seeds	3.50	20.45	37.68	31.03	5.86	nd
	Contribution		0.72	1.32	1.09	0.21	nd
8	Cocoa Powder (Unsweetened)	2.50	16.50	24.00	21.90	5.10	nd
	Contribution		0.41	0.60	0.55	0.13	nd
9	Vanilla Extract	0.50	0.00	0.00	0.00	0.00	nd
	Contribution		0.00	0.00	0.00	0.00	nd
10	Sea Salt	0.50	0.00	0.00	0.00	0.00	nd
	Contribution		0.00	0.00	0.00	0.00	nd
Total in bar		53.50	12.36	4.28	5.84	6.27	24.75
% in bar			23.11	8.00	10.92	11.72	46.26
Energy Content [kcal/100g]			49.45	8.56	52.58	0.00	99.00
Total kcal in bar		209.58					
[kcal/100g]		391.74					

### 5.2 Macronutrient Composition

As shown in Table 1.3, the hemp seed protein bar delivers 209.6 kcal per 53.5 g bar, with a macronutrient profile characterised by 12.36 g protein, 4.28 g dietary fibre, 5.84 g fat, 6.27 g moisture, and 24.75 g carbohydrates. This balance between protein, fibre, and healthy fats underlines the bar's potential as a nutrient-dense snack.

The overall macronutrient profile was calculated using data from various sources to determine the contribution of each ingredient. For each raw material, published nutrient values were combined with the amount used per bar (g) to estimate the contribution of the respective ingredient. The calculations followed Equation 5.1.

$$\text{Contribution [g]} = \frac{\text{Nutrient content [g/100g]} \times \text{Ingredient weight [g]}}{100} \quad (1.1)$$

This approach was applied for protein, dietary fibre, fat and moisture. Carbohydrates were excluded from direct analysis, thus their content was derived by calculating the difference, as outlined in Equation 5.2.

$$\text{Carbohydrates [g]} = \text{Total weight [g]} - (\text{Protein} + \text{Fat} + \text{Dietary fibre} + \text{Moisture}) \quad (1.2)$$

The energy content of the bar was subsequently estimated while adhering to the conversions factors defined in regulation (EU) no. 1169/2011 (protein 4 kcal/g, carbohydrates 4 kcal/g, fibre 2 kcal/g) [23]. The energy content for moisture was assumed to contribute with 0 kcal/g, thus neglected in the calculations.

Based on published nutrient values and the calculations described, the bar (53.5 g) provides 209.6 kcal, equivalent to 391.7 kcal/100 g. Its macronutrient composition - 23.1% protein, 8.0% dietary fibre, and 10.9% fat - qualifies the product for the nutrition claims, “High protein” and “High fibre.” These claims comply with the conditions defined in the Annex of Regulation (EC) No 1924/2006, which governs nutrition and health claims across the European Union [24].

### 5.2.1 Protein Content and Quality

Protein intake is essential for muscle protein synthesis, but the source and type of protein substantially influences its digestibility and utilisation. Since humans cannot synthesise essential amino acids, these must be obtained through diet. Consequently, the overall protein composition, and particularly the essential amino acids profile, is a key to determine protein quality [25].

The hemp seed protein bar provides a total of 12.36 g protein per bar. As shown in Table 1.3, the main contributors for protein are potato protein isolate, rolled oats and dehulled hemp seeds. Dehulling is a processing step which has a significant effect on protein quality. It improves digestibility and reduces antinutritional factors, while also elevating the protein fraction of the hemp seed [11]. To further characterise the protein profile, Table 1.4 presents an in-depth analysis of the protein composition, with respect to each ingredient’s amino acid composition. The data was compiled from multiple sources, yet the methodologies used for amino acid determination has been similar across the studies [25, 26, 27, 28].

The amino acid profile derived from the three main protein contributors includes all nine essential amino acids, confirming the bar as a complete protein source for the consumer. In each of the three main protein contributors, Leucine is the most abundant essential amino acid. Overall, Leucine makes up 5.32% of the total amino acids contributed by the three ingredients. The combined amount of essential amino acids, derived from these contributors, amounts to 2568.83mg, of which Leucine alone constitutes 22.54%.

For both rolled oats, and dehulled hemp seeds, the lowest contribution stems from Tryptophan. General consensus, regarding the hemp seed amino acid profile identifies Lysine as the limiting essential amino acid. However, published values vary considerably depending on the analytical methods used, cultivation conditions and cultivar [11].

Conversely, Tryptophan was not identified in published data for potato protein isolate, suggesting a scarce amounts present. For this ingredient, Methionine and Histidine was reported as the limiting essential amino acids, only contributing with 1300 mg/100g, which is substantially lower than the levels of the other essential amino acids.

The hemp seed protein bar exhibits an overall balanced essential amino acid profile, with only Tryptophan contributing under 1% of the total amino acids, largely due to the absence of reported values in the potato protein isolate. The combination of the three main protein contributors therefore provides an amino acid profile, that can support several potential health benefits. In dehulled hemp seeds, Leucine, Phenylalanine, and Valine are three most abundant essential amino acids. Of these, only Valine is the only one not shared by the top three essential amino acid contributors in potato protein isolate.

Valine is one of the branched-chain amino acids (BCAAs), together with Isoleucine and leucine [29]. Elevated levels of Valine have been associated with various proposed benefits including improved weight gain, weight gain ratio, enhanced intestinal morphology, strengthened immune responses and increased bone density and strength [30]. As an essential amino acid, sufficient dietary intake is important, since valine is directly associated with protein synthesis and functions as a glucogenic amino acid within energy metabolism [30]. Although, valine itself has not been authorised any specific health claims under EU law, protein as a whole is recognised by EFSA to contribute to the maintenance and growth of muscle mass [31].

The combination of potato protein isolate, rolled oats, and dehulled hemp seeds enhances the amino acid profile, positioning the hemp protein bar as a sustainable and nutritionally valuable alternative to other conventional protein bars on the market. In addition, the presence of bioactive amino acids supports the bar's functional value, making hemp a promising ingredient, as a source for high-quality plant protein.

### 5.2.2 Dietary Fibre Content

Dietary fibres are carbohydrate polymers that cannot be absorbed in the human small intestine. These polymers, which contains three or more monomer units, have shown positive potential health benefits with significant prospect of improving carbohydrate metabolism and reducing cholesterol levels [32, 33]. In addition, certain dietary fibre fractions act as prebiotics, and has shown physiological beneficial effects by supporting colonic fermentation and short-chain-fatty acids [33, 34].

The hemp seed protein bar provides a total of 4.28 g dietary fibre per bar. This corresponds to 8 g/100g which exceeds the conditions specified in the Annex of Regulation (EX) No. 1924/2006, for allowing a product to be labelled as "high fibre".

As shown in Table 1.3, the main dietary fibre contributors of the hemp seed protein bar are whole hemp seeds, dates (pitted), and rolled oats, who each contributes with 1.32 g, 1.06 g, and 0.99 g, respectively. Processing also influences the dietary fibre profile of the ingredients. In oats, kilning has been used to inactivate lipase enzymes [35], drying dates concentrates their fibre fractions while using whole hemp seeds (un-hulled) preserves their full dietary fibre content. Together, these three ingredients provide the majority of the dietary fibre in the product. When compared with the relative proportions in the product (Table 1.5), it can be noted, that whole hemp seeds, despite representing only 6.54% of the bar's weight, contributes the largest share of the total dietary fibre. Conversely, dates and rolled oats, which consist of 30.84% and 18.69% of the bar, respectively, provide less fibre relative to their much larger share of the formulation.

Table 1.5 contains several blank entries, reflecting that not all ingredients contribute to each of the listed dietary fibre type, and that published data remain scarce, particularly for whole hemp seeds. Nevertheless, it is noteworthy that whole hemp seeds consistently display higher values g/100g for their respective fibre fractions. This highlights the role for whole hemp seeds as the most fibre-dense ingredient and the main contributor to the label "high fibre".

**Table 1.4:** Amino acid composition of the three main protein-contributing ingredients in the hemp seed protein bar (rolled oats, potato protein isolate, and dehulled hemp seeds). The light orange rows indicate essential amino acids (EAAs). Within each amino acid column, the green shading represents relative contribution, ranging from light green (third highest contributor) to dark green (highest contributor). Red cells highlight the lowest contributing ingredient for that specific amino acid.

Ingredient	Rolled Oats	Potato Protein Isolate	Dehulled Hemp Seeds	Total in bar [mg/bar]	protein % in bar
Protein per bar [g]	1.29	7.92	1.69	10.9	
Ile [mg/100g]	552	3100	1290	274.45	2.52
Leu [mg/100g]	994	6700	2140	579.67	5.32
Lys [mg/100g]	575	4800	1260	408.91	3.75
Met [mg/100g]	221	1300	940	121.68	1.12
Cys [mg/100g]	376	300	650	39.57	0.36
Phe [mg/100g]	729	4200	1430	366.23	3.36
Tyr [mg/100g]	420	3800	1280	328.03	3.01
Thr [mg/100g]	464	4100	1270	352.19	3.23
Trp [mg/100g]	179		380	8.71	0.08
Val [mg/100g]	818	3700	1780	333.67	3.06
Arg [mg/100g]	884	3300	4550	349.52	3.21
His [mg/100g]	309	1300	970	123.32	1.13
Ala [mg/100g]	685	3300	1520	295.88	2.72
Asn [mg/100g]	1150			14.84	0.14
Glu [mg/100g]	2920	7100	3740	663.17	6.09
Gly [mg/100g]	751	3200	1610	290.33	2.66
Pro [mg/100g]	729	3300	1620	298.14	2.74
Ser [mg/100g]	751	3400	1700	307.69	2.82
total EAA [mg/bar]	62.45	2313.22	193.16	187.31	1.72

The three main ingredients providing dietary fibres, contribute with a diverse palette of fibres. Whole hemp seeds mainly contribute with insoluble fibres such as cellulose, lignin, and hemicellulose. Rolled oats supply cellulose +  $\beta$ -glucan, lignin, and arabinoxylan, while dates provide these fraction as well as pectin. These fibres differ in solubility and fermentability, thus contributing to a complementary total dietary fibre profile of the hemp seed protein bar [36].

The data used for calculating the fibre fractions for both dates and rolled oats were obtained from published sources given as g/100g dry weight, and g/kg dry weight, respectively. In order to express these values on an as-is basis g/100g, the first step was to make a moisture content correction for the respective ingredients. These conversions were carried out according to Equation 3 and Equation 4, respectively.

$$x_{\text{as-is}} \left[ \frac{\text{g}}{100\text{ g}} \right] = x_{\text{DW}} \left[ \frac{\text{g}}{100\text{ g}} \right] \cdot (1 - x_{\text{moisture}}) \quad (1.3)$$

And

$$x_{\text{as-is}} \left[ \frac{\text{g}}{100\text{ g}} \right] = \frac{x_{\text{DW}} \left[ \frac{\text{g}}{100\text{ g}} \right]}{10} \cdot (1 - x_{\text{moisture}}) \quad (1.4)$$

The values for the dietary fibre fractions for whole hemp seeds were given in % of dry weight, so the calculation had to follow Equation 5.

$$x_{\text{as-is}} \left[ \frac{\text{g}}{100\text{ g}} \right] = (x_{\text{DW}} \left[ \frac{\text{g}}{100\text{ g}} \right] \cdot (1 - x_{\text{moisture}})) \cdot x_{\text{DFfraction}} \quad (1.5)$$

These conversions (Equation 3-5) ensured that all reported values, despite differences in study and unit expression, were standardised to a consistent unit. This enabled a direct comparison between the fibre fractions contributed by the three ingredients.

### 5.2.3 Fatty Acid Profile

The hemp seed protein bar has a total fat content of 5.84 g. The main contributors to this fat content are dehulled hemp seeds, whole hemp seeds, and rolled oats, which contribute with 3.38 g, 1.09 g, and 0.69 g, respectively, as shown in Table 1.3. Processing steps also affect the lipid quality.

Processing steps also affect the lipid quality. Dehulling the hemp seeds increases the fat fraction by removing the hull mass. For a deeper insight into the specific fatty acid profile of the hemp protein bar, Table 1.6 was constructed to illustrate the distribution of fatty acid fractions from the top three ingredients.

It can be seen in Table 1.6, that the fatty acid profile is dominated by the polyunsaturated fatty acids (PUFAs). Notably, linoleic acid (LA, C18:2 n-6) and  $\alpha$ -linoleic acid (ALA, C18:3 n-3) constitutes 69.47% of the total fatty acids quantified in the bar. These two essential fatty acids, LA (an omega-6) and ALA (an omega-3), cannot be synthesised by the human body and must therefore be obtained through diet. The abundance in the fatty acid profile highlights the nutritional value of the hemp protein bar. Previous studies have reported that LA and ALA from hemp seeds may the nervous system, supporting the health of blood vessels and to protect against cardiovascular diseases [14].

Omega-6 tends to have pro-inflammatory pathways, whereas omega-3 supports anti-inflammatory responses. Therefore, maintaining an appropriate balance between these fatty acids is imperative [14]. Although EFSA does not prescribe a fixed omega-6 to omega-3 ratio, its Adequate Intake levels for LA (4% of energy) and ALA

(0.5% of energy) imply a target ratio of approximately 3:1. The hemp protein bar provides 1.022 g of omega-6 and 0.430 g of omega-3, yielding a ratio of 2.38:1, close to, but slightly below the recommended guidelines [37].

**Table 1.5:** Contribution of the main dietary fibre sources (dates, rolled oats, and whole hemp seeds) to the hemp seed protein bar, expressed as total dietary fibre per bar and distribution of fibre fractions. Coloured cells indicate relative contribution, with light green representing lowest top three value and dark green representing the highest of the top three. The red coloured cells indicate the lowest value for each ingredient.

Ingredient	DF per bar [g]	Fructans [g/100g]	Cellulose + b-glucan [g/100g]	Arabino-xylan [g/100g]	Galacto-mannan [g/100g]	Lignin [g/100g]	Pectin [g/100g]	Hemi-cellulose [g/100g]
Dates (pitted)	1.06	0.23	1.26	1.17	0.60	2.59	0.91	
Rolled Oats	0.99		3.98	2.53		1.81		
Whole Hemp Seeds <sup>1</sup>	1.32		16.95			11.30		7.54
Total in bar [g/bar]	3.36	0.00	0.28	0.04	0.01	0.19	0.01	0.10

As stated, EFSA has set the Adequate Intake levels for LA and ALA to 4E% and 0.5E%, respectively. Based on a diet of 2000 kcal, this corresponds to approximately 8.9 g/day of LA and 1.1 g/day of ALA.

The hemp protein bar will provide roughly 11.5% and 39% of the daily requirements of LA and ALA, respectively. Although the substantial contribution to the daily fatty acid intake, these levels do not meet the conditions set by EU for nutrition and health claims. ALA falls below the *geq* 0.3 g/100kcal threshold for the claim “source of omega-3 fatty acids” [38], and LA remains below the  $\geq$  1.5 g/100kcal threshold required for the claim “contributes to the maintenance of normal blood cholesterol levels” [35].

Saturated fatty acids (SFAs) in the hemp protein bar sum to 0.277g per bar, or 13.27% of the total fatty acid profile. Palmitic acid (C16:0) is the predominant fraction, contributing with 0.199 g per bar. Monounsaturated fatty acids (MUFA) account for 0.360 g of the hemp bar, representing 17.24% of the 2.09 g total fatty acids. Table 1.6 illustrates that oleic acid (C18:1, n-9) is the dominant MUFA, consistently ranking among the three most abundant fatty acid fractions in dehulled hemp seeds, whole hemp seeds, and rolled oats.

## 5.3 Comparison with Market Products - ROO’bar

### 5.3.1 Macronutrients Comparison

A comparison of the macronutrients between the project hemp protein bar and ROO’bar hemp protein bar can be seen in Table 1.7. The project bar is slightly more energy dense than the ROO’bar, with an energy surplus of 14.7 kcal/100g. The higher energy levels primarily stem from protein and carbohydrates, which are present in 9.1 g and 5.3 g greater amounts, respectively. The total fat content is slightly lower in the project hemp protein bar, yet the fat profile appears of a higher quality when reflected by the fraction of fatty acids as the project bar has a surplus of 2.0 g/100g. Although the carbohydrate fraction is higher in the project hemp bar, the dietary fibre content is

**Table 1.6:** Contribution of the main dietary fibre sources (dates, rolled oats, and whole hemp seeds) to the hemp seed protein bar, expressed as total dietary fibre per bar and distribution of fibre fractions. Coloured cells indicate relative contribution, with light green representing lowest top three value and dark green representing the highest of the top three. The red coloured cells indicate the lowest value for each ingredient.

No.	Ingredient	Fat per bar [g]	C14:0 [g/100g]	C16:0 [g/100g]	C18:0 [g/100g]	C24:0 [g/100g]	C16:1, n-7 [g/100g]	C18:1, n-9 [g/100g]	C20:1, n-11 [g/100g]	C18:2, n-6 [g/100g]	C18:3, n-3 [g/100g]	Total FA [g/bar]
1	Dehulled Hemp Seeds	3.38	0.035	4.913	1.876	0.098	0.076	8.734		24.016	11.229	1.725
2	Whole Hemp Seeds	1.09		2.385	0.805			4.232		17.511	4.497	0.320
3	Rolled Oats	0.69	0.005	1.036	0.069	0.077	0.014	2.315	0.055	2.719	0.090	0.044
Total in bar [g/bar]		5.16	0.001	0.199	0.073	0.004	0.003	0.358	0.000	1.022	0.430	2.089
% of FA in fat			0.02	3.86	1.41	0.07	0.05	6.93	0.01	19.80	8.32	

lower, indicating that the carbohydrate quality is comparatively less favourable.

Macronutrient comparison between the ROO'bar hemp protein bar and the project's hemp protein bar, expressed per 100g. The difference column represents (project bar - ROO'bar). Values > 0 are green, indicating a higher content, whereas values < 0 are red, indicating a lower content.

**Table 1.7:** Contribution of the main dietary fibre sources (dates, rolled oats, and whole hemp seeds) to the hemp seed protein bar, expressed as total dietary fibre per bar and distribution of fibre fractions. Coloured cells indicate relative contribution, with light green representing lowest top three value and dark green representing the highest of the top three. The red coloured cells indicate the lowest value for each ingredient.

Component	ROO'bar	Unit 100g	Hemp Protein Bar	Unit 100g	Difference
Energy	377	kcal	391.7	kcal	14.7
Fat	11	g	10.9	g	-0.1
Fatty acids	1.9	g	3.9	g	2.0
Carbohydrates	49	g	54.3	g	5.3
Dietary Fibers	11	g	8.0	g	-3.0
Protein	14	g	23.1	g	9.1
Salt	0	g	0.9	g	0.9

### 5.3.2 Protein Quality Comparison

The macronutrient fraction with the largest difference is that of protein. The project hemp protein bar consists of 23.1 g/100g product, compared to the ROO'bar which only consist of 14 g/100g product. This results in a surplus of 9.1 g/100g product in the project bar. This surplus primarily stem from the potato protein isolate in the project bar. In addition, to the potato protein isolate, the project bar also includes dehulled hemp seeds to the formulation, which adds great amounts of high-quality protein. These amounts of protein qualify the project bar for the nutritional claim "high protein" under regulation (EC) No 1924/2006, while the ROO'bar remains just above the threshold for "source of protein".

### 5.3.3 Dietary Fibre Comparison

The content of dietary fibre in the ROO'bar is greater than that of the project bar. The ROO'bar consist of 11 g/100g dietary fibre, whereas the project bar only consists of 8 g/100g. Both values are above the criteria for the nutritional claim for "high fibre" when expressed in g/100g, although neither of the protein bars reach the required concentration when expressed in g/100kcal. The lower content of fibre in the project bar reflects the dehulling processing step, where the fibre rich hull is removed. In contrast, the ROO'bar formulation relies on hemp protein isolate which is richer in dietary fibres than potato protein isolate. Consequently, while the project bar provides a higher fraction of carbohydrates, the ROO'bar has a comparatively higher quality of carbohydrate profile with respect to dietary fibre.

### 5.3.4 Fatty Acid Profile Comparison

Although the total fat content in the two bars is nearly identical with the projects hemp protein bar containing 10.9 g/100g and the ROO'bar 11.0 g/100g, the fatty acid profile differs substantially. The fatty acid fraction of the project bar contains 3.9 g/100g, more than twice the amount in the ROO'bar that contains 1.9 g/100g. Table 1.6 shows that this difference is attributed to the high levels of PUFAs in both whole hemp seeds and dehulled hemp, particularly LA and ALA. The resulting ratio between these fatty acids in the project bar, are approximately 2.4 1, which is close to the EFSA's recommended range for balance between omega-3 and omega-6 fatty acids. Neither of the products reach the threshold for authorised EU health claims, but the higher levels of PUFA in the project bar indicates a superior quality lipid profile.

## 6 Environmental Impact - sustainability Perspective

Looking at environmental impacts of the hemp seed bar, it is interesting to investigate the impacts from the hemp seeds, as dehulled and whole hemp seeds contribute to a large proportion of the ingredients (Table 1.3). As a crop, hemp is considered carbon-negative because it absorbs more carbon from the air during growth than it yields during its production. E.g. one ton of harvested hemp stem absorbs approximately 1.6 tons of CO<sub>2</sub>, equating to 9-13 tons of CO<sub>2</sub> absorption per hectare [11, 12]. To compare environmental impacts from different food products life cycle assessment methods are often used. Life cycle assessment methodologies have some limitations due to the methodological choices. It is important to be aware of both what is included and excluded from the assessment e.g. what source of power the calculation is made from. However, International Organization for Standardization (ISO) standard exists for life cycle assessments [39]. Though it is important to be aware that life cycle assessments may fail to provide sufficient guidance on environmental, and nutrition impacts that users should be capturing when comparing the overall sustainability and health impacts of different food products [40].

Depending on the genotype and agronomic practices of the hemp seed production, results from a lifecycle assessment revealed that the carbon footprints for most genotypes are below 0.675 kg CO<sub>2</sub> eq for 1 kg produced hemp seeds and for the genotype with the lowest carbon footprint it is reported to be 0.161 kg CO<sub>2</sub> eq for 1 kg produced hemp seeds. The environmental impacts of hemp seed production were analysed with a focus on the agronomic practices such as changing genotype, plant density, and N fertilization in a Mediterranean environment in central Italy. Though it is noted that the lowest carbon footprint is comparable with other crops, e.g., 0.186 kg CO<sub>2</sub> eq for soybeans [41].

Moreover, the cultivation of hemp positively affects soil health and water quality, and it supports wildlife by providing habitat and food [11]. Hemp has deep tap roots that contribute to improved soil structure and carbon storage [42]. The deep root system also allows for efficient water uptake, contributing to a lower water footprint compared to many other crops. This makes it a more resource-efficient choice for food production [42]. Furthermore, hemp crops can remediate organic contaminants and heavy metals from the soil, making them suitable for use on land unsuitable for other food crops. Expanding hemp grain production not only supports environmental sustainability in agriculture but can also deliver substantial social benefits, particularly for low-income groups, rural communities, and other socially or economically disadvantaged populations [11].

In this report the focus is on a hemp seed bar and not on whole diets, which gives rise to the possibility of overlooking the interplay between the amount of nutrients in the studied food product, and the amount of nutrients from other foods that an individual has eaten. For example, the hemp seed bar may be a main contributor to total vitamin E intake of a person who consumes few other foods that contain vitamin E. In contrast, for a person

who consumes many other foods that contain vitamin E, the vitamin E content in the hemp seed bar may only represent a small proportion of their total vitamin E intake. When summarizing a nutritional value of a food product, e.g. based on its nutrient density, the fact that the analyzed food product is part of a whole diet is ignored. To complicate it even more the ingestion of sufficient quantities of nutrients is no guarantee of nutrition because the nutrients need to be bioavailable to provide nutrition and bioavailable nutrients starts with bioaccessible nutrients. To really make life cycle assessment studies useful, the nutrient aspect of the food product is an important aspect to consider. It can either be assessed in parallel with the environmental impacts or by using methods that integrate the two dimensions such as nutritional life cycle assessment where reference nutrient intake values are incorporated [40].

When looking into existing literature, it is found that data lack on nutritional life cycle assessments of hemp seeds, but the existing data on life cycle assessments of hemp seeds suggest that hemp seeds have low carbon footprints and studies of the nutritional value of hemp seeds indicate that hemp seeds are good sources to multiple nutrients that are easily digestible in humans as mentioned previously [43, 41].

To calculate the carbon footprint of a food product, a database such as CarbonCloud can be a useful tool [44]. It is a database built on models that follow the ISO 14067, which is the international standard to assess carbon footprint [40]. The carbon footprints in the CarbonCloud database are divided into 5 categories: Agriculture, transport, processing, packaging and storage. Currently, there is no calculations of a hemp seed bar, as presented in this report. But when looking at excising calculations on products such as an energy bar, the biggest contribution to the carbon footprint is agriculture which covers all the inputs from biological processes such as the production and use of fertilizer and pesticides, as well as the energy use for machinery and refinement processes on the farm. As the second largest contribution processing is found and then packaging. Most probably the hemp seed bar would have a similar distribution in a carbon footprint calculation and careful and optimised agriculture practices for all the ingredients are very important to produce a product with a low carbon footprint which make studies such as Campiglia et al. (2020) highly relevant, because many parameters can be adjusted in the agronomic practices.

The specific ingredients also have a huge impact on the carbon footprint. Dried egg white or whey protein is a common ingredient in protein and energy bars on the market today [45, 46]. When looking at the carbon footprint of producing 1 kg whey powder with 60% protein content the carbon footprint is 19,21 kg CO<sub>2</sub>e and the carbon footprint of producing 1 kg whey powder with just 10-15% protein content the carbon footprint is 4,14 kg CO<sub>2</sub>e. On the other hand the carbon footprint of producing 1 kg dried egg white with 94,5% dry matter is varying between 31,06 kg CO<sub>2</sub>e and 49,79 kg CO<sub>2</sub>e depending on where it is produced. As described in this report an energy bar containing hemp seeds is also a good source of protein. According to data from the database CarbonCloud, 1 kg of 50% hemp protein has a carbon footprint between 0,72 CO<sub>2</sub>e and 1,15 CO<sub>2</sub>e depending on where it is produced, which is lower than the animal-based protein sources and a reason why switching from animal-based to plant-based ingredients can have a big impact on the final carbon footprint of a food product [44].

## 7 Conclusion

In conclusion, the projects hemp seed protein bar offers the consumer a nutritionally rich and environmental friendly protein bar. The bar's formulation, which includes dehulled hemp seeds, whole hemp seeds, and potato protein isolate, results in a well balanced macronutrient profile with high levels of protein, dietary fibre, and healthy fats. The processing methods employed, such as dehulling and light roasting of hemp seeds, enhance the bioavailability of nutrients while reducing antinutritional factors.

The projects hemp protein bar's amino acid profile confirms it as a complete protein source, with all nine essential amino acids present. This complete protein profile supports muscle maintenance and growth. The dietary fibre content stems primarily from whole hemp seeds, dates, and rolled oats. The levels of dietary fibre were not adequate for EU health claims, but may contribute to digestive health and overall well-being. The fatty acid profile is dominated by polyunsaturated fatty acids, particularly linoleic and  $\alpha$ -linolenic acids, which are essential for cardiovascular health.

When compared to existing market products, such as the ROO'bar hemp protein bar, the project bar demonstrates superior protein content and a more favourable fatty acid profile, although it has a slightly lower dietary fibre content. The environmental impact of hemp cultivation further enhances the bar's appeal, as hemp is a carbon-negative crop that supports soil health and biodiversity.

The hemp protein bar demonstrates a strong potential for an environmental friendly product, which offers a low carbon footprint, since hemp requires significantly less water during cultivation compared to many conventional crops. Hemp seeds serve as a sustainable ingredient, particularly in terms of freshwater needs and  $CO_2$  emissions. The development of this theoretical hemp protein bar highlights its potential as a healthy and sustainable alternative to animal-based protein bars. However, further research into the nutritional life cycle assessment of hemp seeds is essential to fully understand the balance between their environmental impact and nutritional benefits.

Overall, the hemp seed protein bar shows promise as an addition to the market focused on health-conscious consumers seeking nutritious and sustainable food options. Future research could focus on further optimizing the formulation to reach the threshold for macronutrient levels for EU health claims. Furthermore, a comprehensive LCA of the environmental benefits of hemp-based products can be conducted.

# Bibliography

- [1] Maeve Henchion et al. “Future Protein Supply and Demand: Strategies and Factors Influencing a Sustainable Equilibrium”. In: *Foods* 6.7 (2017). ISSN: 2304-8158. DOI: [10.3390/foods6070053](https://doi.org/10.3390/foods6070053). URL: <https://www.mdpi.com/2304-8158/6/7/53>.
- [2] Katherine Richardson et al. “Earth Beyond Six of Nine Planetary Boundaries”. In: *Science Advances* 9 (2023), eadh2458. DOI: [10.1126/sciadv.adh2458](https://doi.org/10.1126/sciadv.adh2458).
- [3] OECD and FAO. *OECD–FAO Agricultural Outlook 2023–2032*. Paris: OECD Publishing, 2023. DOI: [10.1787/08801ab7-en](https://doi.org/10.1787/08801ab7-en).
- [4] *Evolving Appetites: An In-Depth Look at European Attitudes Towards Plant-Based Eating*. ProVeg International. 2023. URL: <https://proveg.org/report/evolving-appetites-an-in-depth-look-at-european-attitudes-towards-plant-based-eating/> (visited on 08/14/2025).
- [5] *Plant-based Protein Supplements Market Size*. Market.us. 2024. URL: <https://market.us/report/plant-based-protein-supplements-market/> (visited on 08/15/2025).
- [6] *High-Protein Bars Market*. PW Consulting Health Care Research Center. Oct. 9, 2024. URL: <https://pmarketresearch.com/hc/high-protein-bars-market/> (visited on 08/15/2025).
- [7] *Nordic Hemp (official site)*. Nordic Hemp. URL: <https://nordichemp.ee/> (visited on 08/22/2025).
- [8] *Front page*. Impolan Kasvitila. URL: <https://www.impolankasvitila.fi/en/front-page> (visited on 08/22/2025).
- [9] *ROO'bar*. Smart Organic. URL: <https://smartorganic.com/de/roobar/> (visited on 08/22/2025).
- [10] Stefan H. M. Gorissen et al. “Protein content and amino acid composition of commercially available plant-based protein isolates”. In: *Amino Acids* 50.12 (2018). PMCID: PMC6245118, pp. 1685–1695. DOI: [10.1007/s00726-018-2640-5](https://doi.org/10.1007/s00726-018-2640-5). pmid: [30167963](https://pubmed.ncbi.nlm.nih.gov/30167963/).
- [11] Milica Pojic and Brijesh K Tiwari. *Industrial Hemp: Food and Nutraceutical Applications*. Academic Press, 2022.
- [12] Lidia Montero et al. “Hemp seeds: Nutritional value, associated bioactivities and the potential food applications in the Colombian context”. In: *Frontiers in nutrition* 9 (2023), p. 1039180.
- [13] RE Aluko. “Hemp seed (*Cannabis sativa L.*) proteins: composition, structure, enzymatic modification, and functional or bioactive properties”. In: *Sustainable protein sources*. Elsevier, 2017, pp. 121–132.
- [14] Virginia Tănase Apetroaei et al. “Hemp seeds (*Cannabis sativa L.*) as a valuable source of natural ingredients for functional foods—A review”. In: *Molecules* 29.9 (2024), p. 2097.
- [15] Gurpreet Kaur and Rina Kander. “The Sustainability of Industrial Hemp: A Literature Review of Its Economic, Environmental, and Social Sustainability”. In: *Sustainability* 15 (2023), p. 6457. DOI: [10.3390/su15086457](https://doi.org/10.3390/su15086457).

- [16] Hiroki Yano and Wen Fu. "Hemp: A Sustainable Plant with High Industrial Value in Food Processing". In: *Foods* 12 (2023), p. 651. DOI: [10.3390/foods12030651](https://doi.org/10.3390/foods12030651).
- [17] Peiyi Shen et al. "The impact of hempseed dehulling on chemical composition, structure properties and aromatic profile of hemp protein isolate". In: *Food Hydrocolloids* 106 (2020), p. 105889. DOI: [10.1016/j.foodhyd.2020.105889](https://doi.org/10.1016/j.foodhyd.2020.105889).
- [18] Gerrit A Van Koningsveld et al. "Effects of pH and heat treatments on the structure and solubility of potato proteins in different preparations". In: *Journal of Agricultural and Food Chemistry* 49.10 (2001), pp. 4889–4897.
- [19] Sofia Ekelund et al. "Oat Kilning and Its Effects on Liquid Oat-Base Production". In: *Foods* 13.24 (2024), p. 4083. DOI: [10.3390/foods13244083](https://doi.org/10.3390/foods13244083).
- [20] Wioleta Nowak and Magdalena Jeziorek. "The Role of Flaxseed in Improving Human Health". In: *Healthcare (Basel)* 11.3 (2023). PMCID: PMC9914786, p. 395. DOI: [10.3390/healthcare11030395](https://doi.org/10.3390/healthcare11030395). pmid: 36766971.
- [21] James D House, Jason Neufeld, and Gero Leson. "Evaluating the quality of protein from hemp seed (*Cannabis sativa L.*) products through the use of the protein digestibility-corrected amino acid score method". In: *Journal of agricultural and food chemistry* 58.22 (2010), pp. 11801–11807.
- [22] FAO Expert Consultation on Dietary Protein Quality Evaluation in Human Nutrition. *Dietary Protein Quality Evaluation in Human Nutrition: Report of an FAO Expert Consultation*. Report 92. Expert consultation held in Auckland, New Zealand, 31 March–2 April 2011. Rome: Food and Agriculture Organization of the United Nations (FAO), 2013. 64 pp.
- [23] European Parliament and Council of the European Union. *Regulation (EU) No 1169/2011 of the European Parliament and of the Council of 25 October 2011 on the provision of food information to consumers, amending Regulations (EC) No 1924/2006 and (EC) No 1925/2006 of the European Parliament and of the Council, and repealing Commission Directive 87/250/EEC, Council Directive 90/496/EEC, Commission Directive 1999/10/EC, Directive 2000/13/EC of the European Parliament and of the Council, Commission Directives 2002/67/EC and 2008/5/EC and Commission Regulation (EC) No 608/2004*. Official Journal of the European Union, L 304, pp. 18–63. 2011. URL: <https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX:32011R1169>.
- [24] European Parliament and Council of the European Union. *Regulation (EC) No 1924/2006 of the European Parliament and of the Council of 20 December 2006 on nutrition and health claims made on foods*. Official Journal of the European Union, L 404, pp. 9–25. 2006. URL: <https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=celex:32006R1924>.
- [25] Stefan HM Gorissen et al. "Protein content and amino acid composition of commercially available plant-based protein isolates". In: *Amino acids* 50.12 (2018), pp. 1685–1695.
- [26] DTU Fødevareinstituttet. *Fødevare ID: 59 — Havregryn, uspec. (Oats, rolled, average values)*. <https://frida.fooddata.dk/food/59>. Frida fooddata.dk, National Food Institute, Technical University of Denmark. 2025. (Visited on 08/29/2025).
- [27] Anna PęksA and Joanna MiedziAnkA. "Amino Acid Composition of Enzymatic ally Hydrolysed Potato Protein Preparations." In: *Czech Journal of Food Sciences* 32.3 (2014).
- [28] Hee-Young Hwang, Van-Long Truong, and Woo-Sik Jeong. "A comparative study of nutrient compositions, phytochemicals, and antioxidant activities of two dehulled hemp seed cultivars harvested from different regions in the Republic of Korea". In: *Food Bioscience* 64 (2025), p. 105752.

- [29] Shakshi Sharma et al. “L-VALINE TREATMENT IMPROVES CELLULAR MITOCHONDRIAL FUNCTION”. In: *Innovation in Aging* 7.Suppl 1 (2023), p. 933.
- [30] R Abbas. “Valine in Broiler Diets: A Review”. In: *International Journal of Scientific Research* 3.4 (2024), pp. 11–40.
- [31] European Commission. *Commission Regulation (EU) No 432/2012 of 16 May 2012 establishing a list of permitted health claims made on foods, other than those referring to the reduction of disease risk and to children's development and health*. Official Journal of the European Union, L 136, pp. 1–40. 2012. URL: <https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX:32012R0432>.
- [32] Margarita S Dodevska et al. “Characterisation of dietary fibre components in cereals and legumes used in Serbian diet”. In: *Food Chemistry* 141.3 (2013), pp. 1624–1629.
- [33] Knud Erik Bach Knudsen, Bent Borg Jensen, and Inge Hansen. “Digestion of polysaccharides and other major components in the small and large intestine of pigs fed on diets consisting of oat fractions rich in β-D-glucan”. In: *British journal of nutrition* 70.2 (1993), pp. 537–556.
- [34] Ricardo Ramos-Sánchez et al. “Hemp seed-based foods and processing by-products are sustainable rich sources of nutrients and plant metabolites supporting dietary biodiversity, health, and nutritional needs”. In: *Foods* 14.5 (2025), p. 875.
- [35] Sofie Ekelund et al. “Oat Kilning and Its Effects on Liquid Oat-Base Production”. In: *Foods* 13.24 (2024), p. 4083.
- [36] Abdessalem Mrabet et al. “Date palm fruits as a potential source of functional dietary fiber: A review”. In: *Food Science and Technology Research* 25.1 (2019), pp. 1–10.
- [37] Nutrition EFSA Panel on Dietetic Products and Allergies (NDA). “Scientific opinion on dietary reference values for fats, including saturated fatty acids, polyunsaturated fatty acids, monounsaturated fatty acids, trans fatty acids, and cholesterol”. In: *Efsa Journal* 8.3 (2010), p. 1461.
- [38] European Commission. *Commission Regulation (EU) No 116/2010 of 9 February 2010 amending Regulation (EC) No 1924/2006 of the European Parliament and of the Council with regard to the list of nutrition claims*. Official Journal of the European Union, L 37, pp. 16–18. 2010. URL: <https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX:32010R0116>.
- [39] Anders Bjørn et al. “Goal Definition”. In: *Life Cycle Assessment*. Springer International Publishing, 2017, pp. 67–74. DOI: [10.1007/978-3-319-56475-3\\_7](https://doi.org/10.1007/978-3-319-56475-3_7).
- [40] S. McLaren et al. *Integration of Environment and Nutrition in Life Cycle Assessment of Food Items: Opportunities and Challenges*. Rome: Food and Agriculture Organization of the United Nations (FAO), 2021.
- [41] Enio Campiglia et al. “Hemp Seed Production: Environmental Impacts of *Cannabis sativa* L. Agronomic Practices by Life Cycle Assessment (LCA) and Carbon Footprint Methodologies”. In: *Sustainability* 12.16 (2020), p. 6570. DOI: [10.3390/su12166570](https://doi.org/10.3390/su12166570).
- [42] Rachel A Burton et al. “Industrial hemp seed: From the field to value-added food ingredients”. In: *Journal of Cannabis Research* 4.1 (2022), p. 45.
- [43] Violeta Tănase Apetroaei et al. “Hemp Seeds (*Cannabis sativa* L.) as a Valuable Source of Natural Ingredients for Functional Foods—A Review”. In: *Molecules* 29 (2024), p. 2097. DOI: [10.3390/molecules29092097](https://doi.org/10.3390/molecules29092097).
- [44] *ClimateHub: Benchmark\_Store\_Shelf*. CarbonCloud. URL: [https://apps.carboncloud.com/climatehub/organizations/Benchmark\\_Store\\_Shelf](https://apps.carboncloud.com/climatehub/organizations/Benchmark_Store_Shelf) (visited on 08/26/2025).

- [45] *Protein Bar – Hazelnuts & Cocoa – 12 stk.* The Protein Kitchen. URL: <https://theproteininkitchen.dk/shop/protein-bar-hazelnut-dates-egg-whites/> (visited on 08/29/2025).
- [46] *Blueberry Muffin Protein Bars.* Quest Nutrition. URL: <https://www.questnutrition.com/products/blueberry-muffin-protein-bar> (visited on 08/29/2025).