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PHArmed: A Biological Process for PHA Production from Apple Waste Residues

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Abstract

PHArmed is an innovative Portuguese company responsible for producing PHA in its rawest form, using renewable carbon sources from apple residues. With the increasing environmental urge to find alternatives to regular plastic and an existing expensive way of producing PHA, PHArmed solves both issues with a biodegradable and cheaper product, with biocompatible properties that make it viable for both medical and pharmaceutical industries. Using apple waste from other companies reduces production costs and helps solve the issue regarding waste treatment. After a complete definition of the manufacturing process, the industrial plant would be installed in Pombal, near two large apple waste-producing companies, with a work team of 15 operators. An economic assessment was performed, and an initial 11.8 M€ investment was estimated for the first year. The project was shown to be economically viable and capable of generating a financial surplus, reaching a break-even point after six years.

1. Introduction

As the global population and living standards rise, food production is growing to meet these demands, making waste generation an increasingly alarming issue. In 2021, an estimated total of above 58 million tons of food waste were generated at EU level (Eurostat 2023). An effective management and valorization plan of this waste, in all its different processing steps, is crucial to meet the global efforts to fight the large amounts of domestic and industrial organic waste that are produced worldwide and incorrectly disposed of (Ojha, Bußler et al. 2020). The identification of potentially high-value raw materials within this waste motivates the search for new applications of their properties of interest in different areas, such as the energy or food sectors (De la Peña-Armada and Mateos-Aparicio 2022). Most studies report fruits and grains as the most popular feedstocks for this type of research (Zuin and Ramin 2018).

According to EuropeanBioplastics (2023), global bioplastics production is a growing sector, with its capacity set around 2.18 million tons in 2023 and expected to increase to 7.43 by the next five years. The transformation of the residues of vegetable and fruit processing into bio-based polymeric films is attractive due to their valuable composition, typically with a high content in cellulose/ lignin, its low cost of acquisition and processing and the versatility and biocompatibility of the product (Li, Zhou et al. 2022). Orange waste (Bátori 2019) or banana peels (Ramadhan and Handayani 2020) are, among others, some of the already reported raw materials for the small-scale production of bioplastics.

Apple residues or by-products are one of the most produced agri-food waste types (Ramadhan and Handayani 2020), popularly used for animal feed, organic fertilizing, and pectin extraction (Lobo and Dorta 2019). Within the Portuguese scenario, data from SUMOL+COMPAL and Indumape, two of the biggest juice industries in the country and main apple residues producers, approximates that around 9,600 tons of apple residues are generated annually, consisting of everything from the peel to seeds, pulp, and stalks, which represent 25-30% of the dry mass of the fruit, as estimated in the literature (Lyu et al. 2020). Its potential as a raw material for biofuels and biopolymers has been recently under investigation. Besides showing a high composition of phenolic compounds, which give apple pomace antioxidant, antimicrobial, and anti-inflammatory properties of interest to cosmetic and pharmaceutical industries, its rich content in fermentable sugars provides a feedstock to produce added microbial products (Borrero-de Acuña et al. 2014).

This article suggests an alternative method for the production of polyhydroxyalkanoates (PHA), which have proven to be a promising alternative for the future of plastics. It explores bioplastic production from a renewable source, namely apple fruit waste.

The production process follows the feast-and-famine process, a PHA storage process proposed by Dionisi et al. 2005, consisting of three anaerobic and aerobic sequential steps. Acidogenic fermentation transforms bio-waste into a mixture of Volatile Fatty Acids (VFAs), such as acetic or butyric acid, which are fed into a Sequencing Batch Reactor (SBR) and taken as the carbon source for PHA production by bacteria. Under periods of famine, the absence of substrate leads to a decrease in cell growth potential. If fed with excess carbon, the accumulation of PHA inside bacteria becomes the dominant phenomenon (Huang et al. 2018).

Through this method, a differentiated product can be introduced in the market, aiming to replace synthetic plastics and mitigate the problem of fossil resource over-exploitation. The higher carbon-to-nitrogen ratio of apple waste is an advantageous condition for bacterial

PHA production, which can create a bio-versatile polymer by using a carbon feedstock that is cheaper and independent of fossil resources (Liu, Kumar et al. 2021). The development of a Portuguese company with sustainable PHA production can be a potentially high-profitable business and give a major contribution to a circular economy. By selling biocompatible, non-toxic and quickly degradable bioplastics, PHArmed could be expected to thrive in the medical and healthcare business (Ray and Kalia 2017), as PHAs are suitable to produce surgical materials and other medical devices and are a promising polymer in the tissue engineering field (Bonartsev et al. 2019).

This study elaborates an analysis of the different stages of the design of a chemical product, following the four-step approach (Needs, Ideas, Selection, and Manufacturing) after the characterization of the raw material of interest. It aims to develop a process diagram for PHA production from apple waste and perform an in-depth study of the technical and economic viability of the overall process.

2. Raw Material

2.1 *Production and waste generation*

According to data from the Food and Agriculture Organization of the United Nations, worldwide apple production in 2021 was approximately equal to 93,000 tons (FAO , 2023). Within the Portuguese scenario, apples are the most produced fruit in the fresh fruit category. This fruit represents about 58% of the total registered production in 2019 and about 31.8% of the country's agricultural surface destined to plant all fresh fruits (Agrogarante 2019). It should be noted that apple production is concentrated in the south of the country and in the north, specifically in the regions of Ribatejo and Oeste and in Trás-os-Montes, respectively.

Apple is a promising crop for these regions due to its good climate adaptability and market needs, both for domestic consumption and export. Table 1 shows that, from 2018 to 2020, exports prevailed in comparison to national consumption. Figure S1 of Supporting Information shows detailed data on what concerns national apple production.

Year	Exports (ton)	Imports (ton)
2018	64,000	55,000
2019	71,000	51,000
2020	66,000	41,000

Table 1: National imports and exports from 2018 to 2020 (Agrogarante 2019).

According to CONFAGRI, the National Confederation of Agricultural Cooperatives and Agricultural Credit of Portugal, apple production reached 368.2 ktons in 2021, which was considered the second most productive harvest in the last 35 years (CONFAGRI 2022). However, a decrease in apple productivity of around 20% is expected due to scarce precipitation and high temperatures (CONFAGRI 2022).

The data that was used to design the production process concerns the production of apple residues from SUMOL+COMPAL and Indumape, two fruit juice production industries located in Pombal. Thus, it was considered that the retrieved quantitative data on apple residues was referred to the prevalent apple species in that region, namely Fuji, Golden Delicious, and Jonagored.

2.2 *Characterization*

Apple pomace consists of a heterogeneous mixture composed mainly of peel and pulp (95%) and a small percentage of stems and seeds (2-4%). It has several nutrients, in which the high concentration of phytochemicals and carbohydrates, as well as small amounts of proteins, vitamins, and minerals, stand out. More specifically, carbohydrates come from insoluble sugars such as cellulose (127.9 g/kg, in dry weight (DW) basis), hemicellulose (7.2 to 43.6 g/kg DW), and lignin (15.3 to 23.5 g/kg DW), with simple sugars such as glucose, fructose and galactose (**Table 2**). These sugars are used as sources of carbon to produce products in the industry, and therefore, apple pomace proved to be a good and cheaper source of carbon for the production of bioplastics (Lyu et al. 2020).

	Composition (%)
Fructose	21.85
Glucose	10.55
Sucrose	4.39

Table 2: Sugars composition in apple pomace.

In addition to these components, apple pomace also shows, although to a lesser extent, some minerals such as P (0.070-0.076%), Ca (0.06-0.10%), Mg (0.02-0.36%), and Fe (31.8-38.3 mg/kg DW) (Lyu et al. 2020).

3. Chemical Product Design

For the creation of the product, a strategy was developed in terms of marketing. The main objectives were based on developing a different product that would be able to stand out in the market, targeting a specific niche of customers, and trying to find space in the market where there is an opportunity for profit and growth. The four-step strategy of Cussler EL, Moggridge GD. 2011 for the design of a chemical product was followed.

3.1 *Needs and Ideas*

In the initial step, the raw material's properties and the active compounds were analyzed, and different ideas came of how they could be valued and turned into a product. Table 3 summarizes some of these envisioned, along with the market and product needs associated.

Product	Market Needs	Product Needs
Apple Cider	Low market need	Similar in price to regular cider
Shampoo	Innovative products for various hair-related issues; Growing popularity of leave-in treatments and masks for hair protection and nourishment	Low price; Nice smell; Provide protection (antioxidant properties); and cleansing
Coating film for food protection	The food packaging industry is fast-growing; Need for sustainable and recycled plastics; Reduce plastic waste	Inhibition of pathogens growth; Prevent transfer of odors and flavors; Barrier to O_2 and moisture
Wood Protecting Resin	Market increase; Need for non-toxic and biodegradable resins; High demand for applications like landscaping; fire protection.	Non-toxic and biodegradable natural substances; Antifungal and antibacterial
PHA	Increasing demand for biodegradable materials in the packaging, food, and pharmaceutical industries; Growing of the medical polymers market (replacing conventional glass, ceramics, etc.)	Biocompatible; Biodegradable
Biogas	Replaces traditional fossil fuels (FF), reduces the emissions of greenhouse gases, nitrous oxide, etc.	-

Table 3: Market and Product needs for products envisioned.

An analysis of Table 3 guides the decision-making process and allows a more detailed insight into what are the potential opportunities for innovation and the market gaps that need to be filled, through the development of differentiated and unique products.

3.2 *Selection*

To choose between the six different products envisioned, a matrix analysis was developed to deepen the evaluation of the products. The authors have graded each product within different criteria, from 1 to 10, according to the significance and importance of certain factors on the success of the product. The products were evaluated according to:

- Production Price: compares the production price of the product considered to the average production cost of a similar product already availed in the market.
- Innovation: refers to the novelty of the idea and its possible differentiation factors compared to others in the same market field.
- Available information: regards the amount of data and information that can be gathered for each product to understand the feasibility of its production.
- Market size: identifies the size and number of groups of potential buyers for the product.
- Longevity: considers the capability of the product to prevail among others in the market for long periods.
- Market Need: relates to the functional needs or desires of the targeted market.

Table 4 summarizes this analysis and presents the different scores assigned to each product idea.

Product	Production Price: 20%	Innovation: 20%	Available info: 10%	Market size: 20%	Longevity: 10%	Market Need: 20%	Total
PHA	5	7	6	6	6	7	6.2
Coating Film	3	9	3	6	6	7	5.9
Shampoo	6	1	9	9	3	2	4.8
Apple cider	3	4	4	7	3	2	3.9
biogas	2	7	6	6	5	8	5.7
Wood protecting resin	5	7	3	6	6	7	5.6

Table 4: Evaluation table regarding product ideas.

A deeper explanation of how the final selection was performed can be found in Section 4.1 of the Supporting Information. However, Table 4 easily leads to the conclusion that PHA and coating film were the two main options, according to the total weight of the defined criteria. More specifically, PHA arises as a highly innovative product, sharing a large market size and need due to its versatility in end use and relatively low production price, while still possessing more available information when compared to the coating film.

4. Manufacture

4.1 Description of production process

The raw material required for the process is obtained from two large Portuguese companies, SUMOL+COMPAL and Indumape, both close to one another and to the chosen location for the industrial plant. This way, while boosting the local economy, companies' waste is easily handled and transported to the site at low costs. Despite being limited to the supply of these two companies, the approximate amount of 10,000 annual tons of apple residues is significantly enough to meet the requirements of the designed industrial plant.

Given that, in Portugal, apples are not considered a seasonal fruit, the waste collection from these two fruit processing industries would be done every month and stored in two tanks of 350 m^3 capacity each. Because the apple pomace will undergo a process of fermentation, a pretreatment step before the reaction is recommended, to increase the yield of the overall process, as well as to avoid the deterioration of the material over time. This can be done through heating, mechanical, chemical or enzymatic processes (Cheng, Yue et al. 2020), that reduce the cellulose and lignin content (Zytner, Kumar et al. 2023) and help to release the fermentable sugars (Peinemann and Pleissner 2020). However, the cost of this step was not considered nor detailed in the process flowsheet, due to the need for a more in-depth study of the different possible routes for the pretreatment of the material.

The following process was developed following data from a large-scale designed plant for PHA production from wastewater, from a report by Nazeer Khan et al. (2021). Despite not using the same raw materials and having different purposes of component valorization, the overall methodology present in this report was followed by the authors of this work, because the design of process diagrams for PHA production is not yet significantly reported in the literature.

In the first step of the process, an estimated amount of 26 tons/day of apple waste is mixed with an equal part (in volume) of water and pumped into a set of 5 acidogenic fermenters, each with 100m^3 of capacity and sludge retention time of 7 days. The fermentation process is carried out at a relatively low temperature of 35 °C, and the outlet stream is sent to a centrifuge, where solids are separated from a solution with a VFAs concentration of about 77 wt.%. This VFA-rich stream is then separated into two different reactors. In these reactors, a mixed-microbial culture is used to increase the success of the selection step and to enhance the PHA accumulation performance.

The first split stream is fed into a fully aerobic SBR, operating about 24 hours a day, using cycles of 12 hours with alternating feast and famine phases. The VFAs stream enters the reactor, along with water and excess biomass, allowing for the consumption of the carbon source and biomass growth. After the feeding and the reaction, the settling phase is followed by an effluent withdrawal to keep the biomass concentration inside the reactors between 1.5-2.0 kg/m^3 (Montiel Jarillo et al. 2021).

The remaining solution of the second split is bypassed to an accumulation batch reactor and is mixed with the outlet stream of the SBR, as seen in **Error! Reference source not found.**, presented in the next section. The accumulation reactor is operated through a pulse-feeding strategy, and it is continuously stirred and aerated (Campanari et al. 2017).

Following the accumulation step, the PHA-rich biomass goes through a centrifuge, and a polymer content of 30 wt.% in the cell dry mass is reached. This dried biomass is then sent to a flue gas dryer, with a working temperature of 100 °C, in which around 5 tons of dried PHA-rich biomass are produced per day.

In the extraction process, this previously dried biomass is fed into an extraction reactor. Using dimethyl carbonate (DMC) at a temperature of 90 °C, with a solvent-to-biomass mass ratio of 19, the polymer is extracted with a process yield of 50%. This PHA-rich solution is filtered, and the waste biomass is discarded, as it is mainly composed of cellular residues and other components from which the PHA was extracted. Due to not having an interestingly high PHA- content, this biomass is forwarded to a sludge digester, but the possibility to use it as a biofertilizer or even biogas production could be explored. On the other hand, the PHA-rich biomass is sent to an evaporator, in which a percentage of 99.5% of the solvent is expected to be recovered and reused in the next cycle.

Error! Reference source not found. sketches this entire process flowsheet diagram (PFD) and presents the flow rates associated with each equipment.

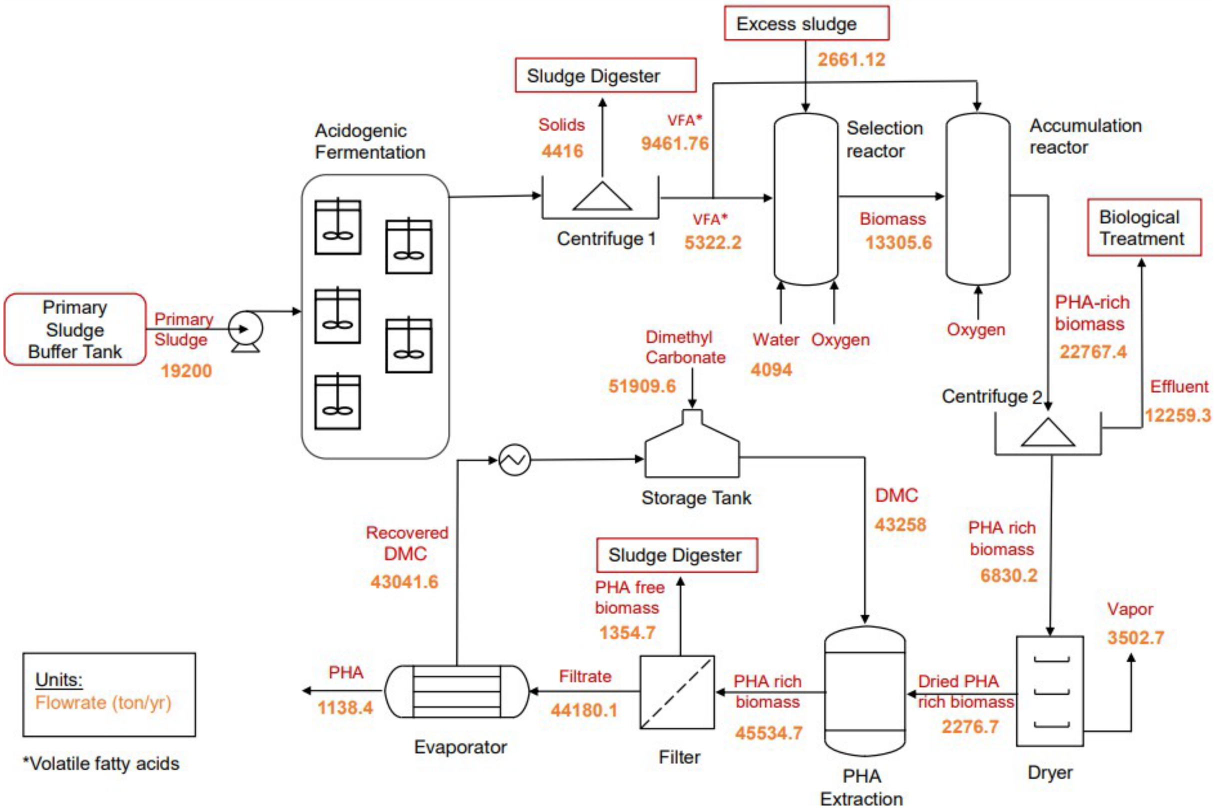

Figure 1: Flowsheet for the industrial production of PHA with the respective flowrates (ton/yr).

The daily amount of 3.12 tons of produced PHA will be stored in appropriate storage tanks and will be sold in 25 kg and 50 kg bags.

4.2 Product Specifications

PHA comes from the family of bacteria synthesized bio polyesters, presenting numerous advantages, such as biodegradability, biocompatibility, and flexible strengths (Colombo et al. 2017). It can be synthesized by different and naturally occurring bacterial cultures, such as *Ralstonia eutropha, Pseudomonas spp.*, etc. (Verlinden, Hill et al. 2007, Yang, Liu et al. 2007). The final product is produced in its rawest phase, allowing it to achieve a higher number of these characteristics and therefore reach a broader market. When compared to other bioplastics, the high carbon (approx. 50%) and low nitrogen content (<2%) in apple pomace are both favorable factors for PHA production (Liu, Kumar et al. 2021). Besides, the use of this raw material as a carbon source in anaerobic fermentation allows for a sharp decrease in the production price, which is usually one of the main drawbacks for the commercialization of PHAs and industrialization of this technology (Surendran, Lakshmanan et al. 2020). This biopolymer can be used as a substitute for regular plastic, packaging materials (including films, boxes, coatings, fibers, and foams), or even as a biofuel. However, it shows the biggest potential for medical applications, such as implants or biocompatible medical devices, and for use in the pharmaceutical industry, as drug delivery carriers (Vu et al. 2021). Since no specific study was done on the final product, information from other researchers was used to describe the physical and chemical characteristics of this polymer. Endothermic sample peaks determine the PHA's melting temperature in a range between 133 °C and 145 °C. Other extracted samples and the existing commercial product show similar crystallization temperatures, rounding 100 °C. The molecules are sufficiently mobile at crystallization temperatures to reorganize into orderly structures, preventing brittleness. Because typical plastics do not need to be exposed to high temperatures, the usage of PHA might be an alternative for general-purpose applications (Vu et al. 2021).

From a commercial point of view, the melting and crystallization temperatures are essential factors in determining product quality and its end use. From previously analyzed samples, crystallinity degree and melting enthalpy have also been studied, as shown in Table 5.

	T_m melting temp. (°C)	T_C Crystallization temp. (°C)	Crystallinity degree (%)	Melting Enthalpy (J/g)
PHA	133-145	100	12.81	18.71

Table 5: Obtained PHA Properties (Vu et al. 2021).

As it still is a considerably new product, the degradation, storage, and shelf-life conditions are hard to determine, since they depend on the environmental factors, intrinsic properties of the PHA and the chosen production method. Meereboer, Misra et al. (2020) reported that the degradation of PHA in aquatic environments at approximately 5 °C is negligible. On the other hand, some studies show that PHAs are degradable even at a maximum temperature of 60 °C, with moisture levels above 50% (Rudnik 2013).

With the essential goal of mass production and financial success for the proposed company, it is not only necessary to understand the properties of the proposed product but also to compare it with rival products in the market. The comparison is made between the produced PHA from food waste with a commercial and more expensive PHA, that utilizes another source other than food residue and regular petroleum-based plastic (Vu et al. 2021). This comparison is shown in Table 6.

	T_m melting temp. (°C)	Crystallinity degree (%)	Time to complete degradation	Production Price (€/kg)
Produced PHA	133-145	12.81	7 weeks	1.25-2.5
Commercial PHA	148.89-161.62	74.09	-	2.5-5
Regular plastic	100-200	10-80	Up to 450 years	1

Table 6: Property comparison between produced products. (Vu et al. 2021)

Regarding melting temperatures, PHA produced from food waste has a similar value to commercial PHA, both within the normal regular temperature margin found in common plastic. However, produced PHA shows a far shorter biodegradation period when compared to regular plastic. This creates a clear advantage, not only as it reduces the cost of the degradation itself, but also indicates that it is a sustainable product, fit for what the current times, government, and laws demand. The Portuguese Decree-Law No. 78/2021 of 24 of September of 2022 stipulated the goal of "reducing the impact of certain plastic products on the environment and amending the rules concerning plastic products at points of sale (…)", mostly due to their long degradation period (DRE 2021).

It is also easily verifiable that regular plastic is inexpensively produced, only costing 1 €/kg. Price production is a problem for commercial PHA derived from non-renewable carbon sources, reaching almost five times the value of regular plastic. The usage of food waste as this source drastically reduces the production price to half the value, making it a much more compatible end-use product, especially due to the advantages it presents when compared to other polymers.

5. Economic analysis

To evaluate the economic viability of a project, an economic analysis is a crucial step in optimizing the process and assessing its future performance. This analysis is carried out after the manufacturing process is defined and all the units and their capacities are known.

5.1. Gantt Diagram

Gantt diagrams are used to organize and structure projects. Events and milestones are arranged on a timeline and an overview is maintained in the case of dependencies and overlaps. To organize the timeline of the product PHArmed, six significant events (Marketing, Investments, Research and Development, Engineering, Production, and Sales) were selected. Furthermore, four stages of the Chemical process design (Needs, Ideas, Selection, and Manufacturing) were chosen to monitor the project's progress. The Gantt diagram of Figure 2 presents on the timeline for the first eight years of the company. Dark blue bars show cost and labor-intensive phases, while light blue bars indicate periods of medium economic effort.

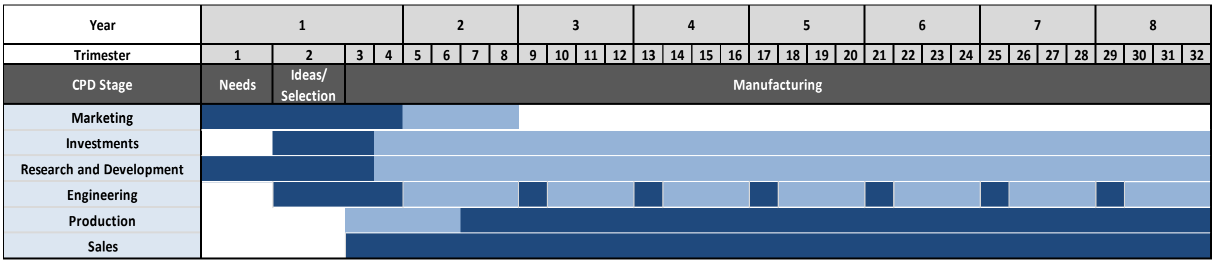

Figure 2: Gantt Diagram for the PHArmed project.

Since the product will be based on a Business-to-Business (B2B) strategy, it is important to approach other companies as early as possible to introduce the company and the product and connect to potential customers. When the product is finalized, marketing becomes crucial to generate demand and orders for the product. After establishing the product, Marketing becomes less important in a B2B market.

Investments are highest when acquiring production capacity and equipment after finalizing product specifications. The need for research and development is highest during product development. Further development in later phases of the project and optimization of the process can lead to higher yields and profits. The Engineering department assists in the development of the product and is responsible for maintenance and repairs between production cycles. Production and sales are linked and reach a steady level after a start-up phase. After reaching the target production volume, the process flow is only interrupted by maintenance and repairs. Most of the costs are spent on the purchase of raw materials and basic materials.

5.2. Capital Investment and Total Product Cost

The capital expenses associated with the purchase of the unit's equipment are summarized in Table 7. This data was based on already reported data, and it was scaled down according to the corrected flow rates handled by the process in question (Nazeer Khan et al. 2021). All values were updated by the CEPCI value of 2022.

Equipment	Capital Cost (€)
Fermentation reactors	972,850
Centrifuge 1	247,156
Selection reactor	277,600
Accumulation reactor	277,600
Centrifuge 2	247,156
Dryer	124,104
Extraction reactor	52,854
Filter	358
Evaporator	2,681
Waste Storage Tank	28,000
Water Storage Tank	2,000

Table 7: Cost of each unit of equipment.

An additional 30% was added to the total value to account for non-dimensioned equipment (such as valves, pumps, PHA, and solvent storage tanks). Table S1 of Supporting Information elaborates, in detail, the fixed-capital investment (FCI) for the plant, considering direct and indirect costs and working capital of 15% of the FCI.

Capital is needed to purchase the equipment and start the production process, which can be borrowed from a bank in the form of a loan. The investments costs are estimated at 6.05 million euros, including the equipment shown in Table 7 and the installation and transport costs of the components to the production site. As no profit is made in the first two trimesters, further costs are added to the loan. The solvent, the bacteria, and the research costs for these trimesters are also included in the loan. The research costs are assumed to be an additional 10% of the personnel costs in the first year and 5% in the following years to optimize the process.

In total, a loan of 6.75 million euros is needed. Based on the current interest rate situation, an annual interest rate of 5% and a repayment of 1% can be assumed.

Concerning the operational expenses and the utilities required for the process, electricity and steam involve the greatest financial amount. Considering a unit price of 0.24 €/kWh and the energy requirement for each piece of equipment (Table S2 of Supporting Information), a total of 94,192€ is expected to be spent per trimester (GlobalPetrolPrices 2022). Regarding steam consumption, over 14 tons per day are consumed in the drying and evaporator units, accounting for approximately 31,705 € every three months. A total of 6,947 € is allocated to costs with natural gas (for heating of the acidogenic fermenters and the drying step) and cooling water (for solvent cooling).

As per expenses with raw materials, the large requirement of solvent per ton of dried PHA turns it into an important financial factor. With a unit price of 1,000 €/ton, the initial investment cost for DMC purchase would be 142,218 € (for the first trimester), ensuring enough solvent for a day of work by a safety margin of 20%. Considering a 99.5% recovery of this component per evaporation cycle, an inferior value of 53,331 € would be spent in the subsequent trimesters to compensate for the loss.

Lastly, in terms of personnel costs, the total annual cost payable to employees is 362,365 €, as detailed in Table S3 of Supporting Information. The fixed monthly salary per individual depends on the type of function performed. All employees are entitled to a food allowance of 5.20 € per day of work (DN. 2022), and social security contributions are considered at the updated rate of 23.75 € (CGD 2015).

5.3. Location

The chosen business location is one of the most crucial factors when contemplating any industrial installation project, regarding profit and business growth. In the case of a PHA production industrial company, a close location to the raw material suppliers should be chosen to reduce transport costs.

It is also crucial to verify that the chosen location has a good amount of qualified labor. Safety margins at the human and environmental levels closest to the facility should also be considered, as some of the resources used may be environmentally unfriendly and require handling and direct human contact (Sfredo et al. 2006). To respond to the aforementioned parameters, the industrial facility is strategically located on the outskirts of the city of Pombal, in location A, as shown in Figure 3, due to its proximity to the main apple waste producers in the region: Indumape, at location B, and SUMOL+COMPAL, at location C.

Figure 3: Suggested location for the industrial plant (A) and nearby industries (B and C). From: ©2024 Airbus, CNES/ Airbus, IGP/DGRF, Maxar Technologies, 200m. (Google 2024).

After considering potential locations in terms of price and all other factors previously discussed, location A, with an average area of 8,000 m^2, was chosen for the implementation of the company's facilities.

Due to the very short distance between this location and the two companies providing the raw material needed (2 to 5 km), transportation would be easy, especially as apple waste does not have any safety precautions regarding its transportation. With an annual available amount of 9,600 tons of waste, a monthly transportation of 800 tons from companies B and C to the selected location would be the best option. The transportation would be done by 100 m^3 capacity trucks, able to carry up to 25 tons of load capacity, which would require around 30 to 35 trips per month. This way, a total investment of 240,000 € would be made, equivalent to the price of 6 trucks of 25 tons (estimated at 40,000 € each). From 2020, these big-capacity trucks from TIR are allowed in Portugal, facilitating this transportation. Gas prices were also taken into consideration for the economic analysis.

The chosen location has great proximity to major highways only 3 km away from A1, which connects the two largest cities in Portugal, facilitating the distribution of the product and the other needed resources. In addition to minimizing transport costs, this location was chosen due to easy access to labor while still maintaining a safe distance from the nearest city.

The analysis of all the points mentioned led to the conclusion that the site offered was the one that offered the greatest advantages concerning the installation needs.

5.4. Marketing strategy

When developing a marketing strategy, it is fundamental to consider the markets in which PHArmed will be launched, and so a strategy to attract as many people as possible and generate high sales must be defined. Therefore, initial market research is essential in the early stages of the company.

Unlike most products, the PHA would not be produced towards a specific end goal but rather sold in a raw state to reach a broader market. This way, most companies can transform it to meet their objectives. This strategy was thought to be a B2B model, decreasing the need for a marketing strategy that relies on intensive publicity campaigns. It becomes crucial that the marketing strategy influences the businesses to know whether the proposed raw material will be able to satisfy their needs for the desired final product.

To do so, company representatives would be required to present and promote PHArmed to other companies. Besides, free samples should be first provided to those who are interested, to encourage experimentation and build a long-lasting, trustworthy relationship. These representative employees would choose promising regions where individual interested companies would be located (most likely Porto, Lisbon, and the south of Portugal).

While marketing should follow the development of several areas, such as technology innovation and new means of information and communication, there is not a significant dependence of PHArmed on social media, as it does not aim to establish direct connections with final consumers (Silva 2018). Therefore, advertisements will not be a direct necessity for the promotion of the produced product. The biggest expenses associated with marketing would be the company's representative of sales (ca. 1150 €/month) and eventual small product losses due to handling of small samples. To promote the differentiation of the product to other businesses and to sell it as a complete substitute to all other polymers, the biodegradable and biocompatible properties of the PHA produced are crucial points of focus, to justify the relatively higher prices of this product when compared to other already available PHA options.

5.5. Economic Feasibility

The sales price level of PHA polymers is still much higher than traditional petroleum-based plastics, having a reported value, in 2016, of 5€/kg of bio-based PHA (Van den Oever, Molenveld et al. 2017). This is mainly driven by the production price associated with the carbon source (Khatami, Perez-Zabaleta et al. 2021). However, as the general awareness towards more sustainable plastic alternatives grows and considering the estimated production price of for 1.25-2.5 €/kg of the PHA plastic suggested by the authors, a sales price of 3.75 €/kg produced PHA can be considered, making it economically competitive with others while still offering significant revenue to the company.

Considering this value, the price of sale for 25 kg and 50 kg bags of PHA is defined as 93.75 € and 187.5 €, respectively. With an estimated annual production of 1139 tons of this product, the company can expect an annual revenue of 4.27 M€.

A financial forecast was performed to estimate when the breakeven of the company would be achieved. To do so, all incomes, production, and operational costs were identified to assess the evolution of NET and PV cash flows during the first years. An annual increase of 2% in production costs was considered.

Figure 4 shows the trend for accumulated costs and accumulated profits, which points to a payback time of a little over 22 trimesters for the investment.

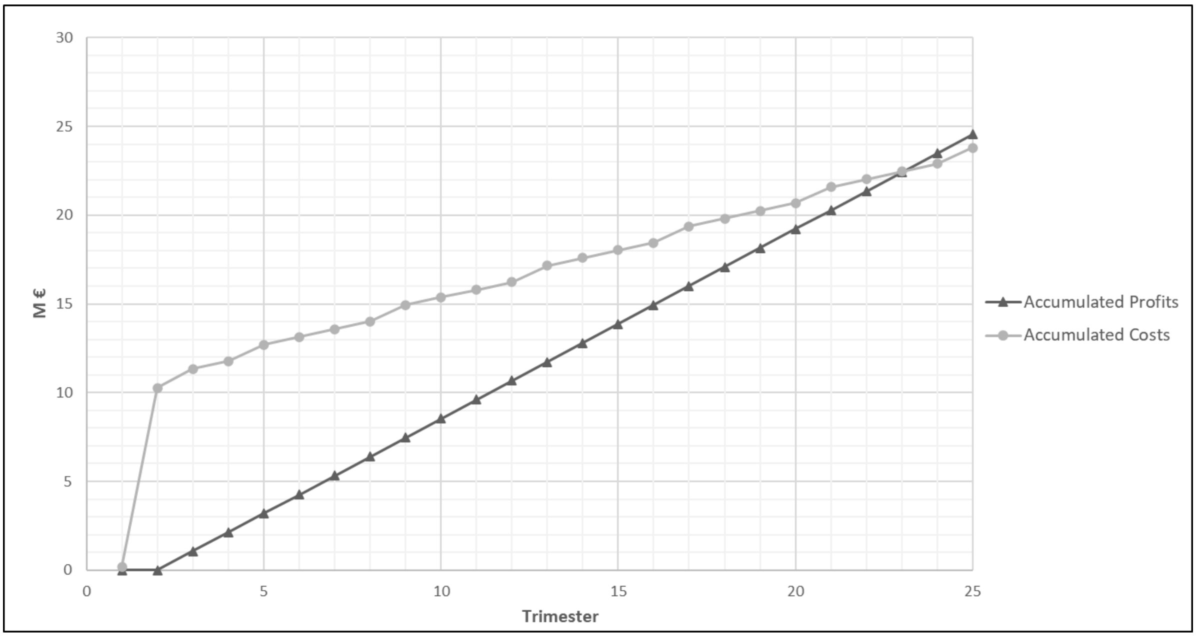

Figure 4: Accumulated profits and costs on a trimestral basis.

An Internal Rate of Return (IRR) of 28% was calculated, far exceeding the percentage of 12% considered for the cost of capital. The net present value of the project was also estimated to be 544 M€. All these financial indicators point to the profitability of the project, which appears to generate a significant sales volume, able to support the initial capital and operational expenses, within a relatively short period.

6. Conclusions

In conclusion, PHArmed offers an innovative approach to PHA production from apple waste, bringing an environmentally sustainable alternative to the bioplastics sector, while addressing the urgent need of residues valorization.

PHArmed combines the biocompatible and biodegradable properties of PHA with a sustainable production process, presenting the final polymer in its rawest, non-modulated form, which not only differentiates it from its competitors but also enables PHArmed to sell its product to industries in various sectors (although targeted to medical and pharmaceutical areas).

This project offers a competitive value for the production cost when compared to direct competitors, mainly due to the cheaper feedstock, making it possible to sell the polymer at an attractive price of 3.75 €/kg and still achieve a break-even point by the 22nd trimester of the project. The entire technical and economic analysis performed by the authors leads to conclude that PHArmed can be a lucrative investment opportunity, capable of generating a financial surplus and address a viable and sustainable solution to the global challenge of plastic pollution and waste management.

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Appendix

SUPPORTING INFORMATION

2.1 Production and waste generation

Figure S1 reports Portuguese apple production from the year 2012 to 2020. As seen, it fluctuates between 16 to 25 tons per hectare, with the available data averaging a production of 20 tons/ha (Agrogarante 2019).

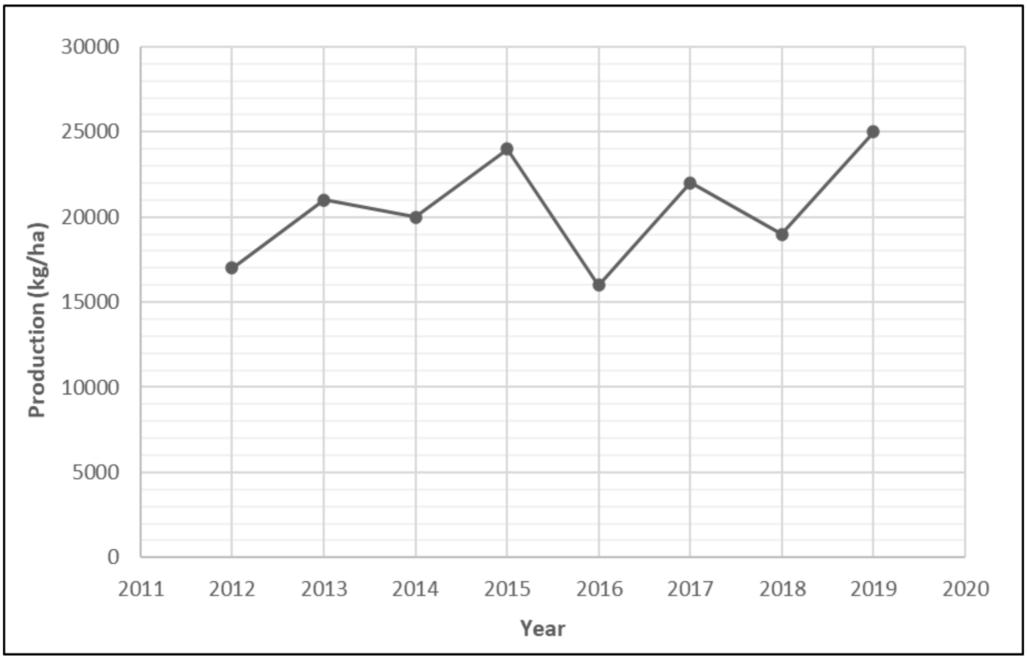

Figure S1: Portuguese Apple Production from 2012 to 2019.

4.1 Needs and Ideas

As the suggested raw material is apple waste, a cosmetic of natural origin could be an attractive product to develop. The idea of creating a natural shampoo was to take advantage of the antioxidant properties of apple polyphenols and provide protection to the scalp. However, market needs for this type of product are relatively low, due to very high competition in the area, which would hinder a product differentiation strategy.

Regarding the production of apple cider from waste, the idea would be to reuse apple pulp and fruit waste to create an innovative food product (or in this case, a beverage) with added value. Even though nowadays, due to health-conscious reasons, the fruit-based drinks market is growing, which gives space to the growth of improved craft ciders, the product would not be innovative enough to be economically feasible, not prevailing in the highly competitive market.

The wood-protecting resin idea was thought to work as a coat protection for wood surfaces, offering resistance against wear and corrosion. Even though the construction sector accounts for 30% of the consumption of epoxy resins (EpoxyEurope 2018) and the interior designers and building market have both size and need for the product, there was limited information available in this field, making it a difficult product to develop.

5.2 Capital Investment and Total Product Cost

Capital Investment

Table S1 lists all item ponderations for the calculation of the total capital investment needed. Working capital of 15% was considered for an initial start-up of the plant (Kobe 1958).

	% FCI	M€
Direct Costs		
Equipment Purchase	30	2.938
Equipment Transportation	25	0.734
Equipment Installation	9	0.881
Control	6	0.588
Electric Equipment	5	0.490
Services	8	0.783
Indirect Costs		
Engineering and Supervision	5	0.490
Construction Costs	4	0.392
Contractor Costs	3	0.294
Contingencies	5	0.490
Land		0.047
Total Fixed-Capital Investment (FCI)	100	8.126
Working Capital (WC)	15	1.434
Total Capital Investment		**9.560**

Table S1: Total Capital Investment breakdown cost.

Electricity Use

A breakdown of the electricity requirement of each unit is presented in Table S2. A unit cost of 0.24 €/kWh was used to estimate the total daily spending of each equipment.

Equipment	Electricity Requirement (kWh/m^3 sludge)	Total electricity spent (kWh/day)
Fermentation reactors	96.9	3920.93
Centrifuge 1	1.88	76.07
Selection reactor	2.51	28.15
Accumulation reactor	2.40	115.16
Centrifuge 2	1.88	90.21
Dryer	0.16	1.54
Extraction reactor	0.01	1.25

Table S2: Electricity requirement of the plant.

Labor Costs

In terms of costs allocated to the company's workers (Table S3), two different teams were defined. As the unit runs 24 hours a day, two operators and a head chief of production are needed for night shifts. For these workers, an increase of 25% is implemented to the basic salary (14 months).

	N° of workers	Salary (€)	Food Allowance	Financial Charges (€)	Total cost/year (€)	Total cost/ trimester (€)
Operators	3	42,000.00	343.00	9,975.00	57,123.00	14,280.75
Operators working the night shift	2	35,000.00	2,288.00	8,312.50	46,744.50	11,686.13
Head chiefs of production	2	42,000.00	2,288.00	9,975.00	55,407.00	13,851.75
Cleaning staff	1	10,500.00	1,144.00	2,493.75	14,709.75	3,677.44
Quality control	2	33,600.00	2,288.00	7,980.00	45,012.00	11,253.00
Sales	2	31,220.00	2,288.00	7,414.75	42,066.75	10,516.69
Engineers	3	77,700.00	3,432.00	18,453.75	101,301.75	25,325.44
				Total €	362,364.75	90,591.19

Table S3: Labor costs associated with the plant.