



## Review article

# Exploring Azolla as a sustainable feedstock for eco-friendly bioplastics: A review

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## ABSTRACT

In today's world, environmental concerns about plastic pollution of aquatic and terrestrial ecosystems are at the forefront of many conversations. However, a solution that is gaining momentum is bioplastics. Bioplastics come from sustainable biological sources such as plants, bio-waste, or microorganisms, rather than non-renewable fossil fuels like petroleum or natural gas. The properties of Azolla, including its growth in aquatic environments, high nutrient content, and ability to symbiotically fix nitrogen, make it an intriguing candidate for sustainable bioplastics feedstock. By analyzing the current state of research on bioplastics, this review aims to demonstrate the feasibility, challenges and environmental sustainability of this new environmentally friendly alternative to plastics. Thus, we contribute to the ongoing discourse on addressing plastic pollution and environmental degradation through innovative, sustainable materials. The research results show that the unique properties of Azolla such as rapid growth and nutritional content make it a strong contender for sustainable bioplastics raw materials. Azolla-based bioplastics can be helpful as an environmentally friendly alternative to conventional plastics. However, it is crucial to address challenges related to cultivation, processing, and economic feasibility for practical implementation. Azolla-based bioplastics are an opportunity to reduce the environmental impact of plastic waste and contribute to a more sustainable future.

## 1. Introduction

Plastics, once celebrated for their wide application and convenience, have left irreparable environmental effects. The proliferation of non-biodegradable conventional plastics has led to an alarming global crisis characterized by plastic pollution [1]. These synthetic materials, predominantly derived from non-renewable petrochemical sources, have been linked to the destruction of ecosystems, marine pollution, and other environmental problems. The need for new environmentally friendly substitutes for traditional plastics is growing in light of this pressing dilemma [2].

Bioplastics have been demonstrated to be a convincing answer to this problem. Bioplastics are substances derived from sustainable biological sources like plants, bio-waste, or microorganisms instead of relying on non-renewable fossil fuels such as petroleum or natural gas [3]. Compared to traditional plastics, their environmental friendliness is frequently acknowledged, though it can depend on how they are made and disposed of. The bioplastics are still in the early stages of development. Nevertheless, it is expected to

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experience substantial growth as awareness of pollution caused by traditional plastics continues to increase, pointing towards a promising future [4]. They are created to decompose and can be composted, providing an eco-friendly option to regular plastic made from petroleum-based plastic. Bioplastics are categorized into biobased and biodegradable groups. Bioplastics are manufactured from renewable biomass sources like starch, cellulose, proteins, or microbial polyesters [5]. Some examples are polylactic acid (PLA) and polyhydroxyalkanoates (PHA). Under certain conditions, microorganisms can decompose biodegradable plastics into natural substances such as carbon dioxide, water, and biomass [6]. Biodegradability can be attained in both plastics derived from natural sources and those derived from petroleum. Nevertheless, there has been inconsistency in the inconsistent usage of the term “bioplastic” highlighting the necessity of differentiating between biobased and biodegradable plastic [7]. The advancement of bioplastics is motivated by the goal of decreasing the use of petroleum, advancing the utilization of renewable materials, and tackling the increasing environmental issues linked to conventional plastics [8]. Bioplastics provide various benefits like decreased greenhouse gas emissions, the ability to decompose, decreased reduced reliance on fossil fuels, and the opportunity to make use of waste. Bioplastics made from plants have strong mechanical and chemical qualities, yet they pose a risk to the future due to competing with human food sources. Moreover, the techniques for extracting bioplastics from plants are intricate and essential [9]. Therefore, given the increasing worries regarding plant resources for bioplastics, it is imperative to find a more cost-effective and accessible alternative source of biodegradable plastic polymer for our sustainable future. Challenges in the bioplastics market growth include the requirement for consistent definitions, certification procedures, and recycling and composting facilities [10]. If these elements are missing, the possible advantages of bioplastics may not be fully experienced and they could lead to environmental problems like traditional plastics do. They could lead to environmental problems like traditional plastics. Current studies are focused on enhancing the characteristics and usability of bioplastics in order to increase their competitiveness when bioplastics’ characteristics and usability of bioplastics in order to increase their competitiveness compared to conventional plastics [11].

Macro algae, commonly referred to as algae, are valuable renewable biomass sources because of their rapid growth rates, low land and freshwater requirements, and ability to sequester carbon dioxide. Macro algae have attracted considerable attention as promising feedstocks for the production of bioplastics. However, water fern, *Azolla* stands out within macroalgae due to its unique properties [12]. *Azolla* is a member of the Salviniaceae family and is distinguished by dense mats of tiny, floating fronds that form on the surface of freshwater bodies such as lakes, ponds, and streams with a slow flow. Under ideal circumstances, this free-floating, heterosporous aquatic fern can double its biomass in days and shows excellent growth rates [13]. An image of an *Azolla* fern is shown in Fig. 1.

*Azolla* fern is known to be an environmental problem and is freely available. During the last few years, there have been several studies in literature verified the high potential of *Azolla* to be applied to biofuel [14], a source of food for humans [15] and animal consumption [16], biofertilizer [17], environmental bioremediation [18], greenhouse gas mitigation [19] and also in removing nitrogenous compounds from the water [20] and a weed and mosquito control agent [15]. It also has several applications as a biocontrol agent for improving water quality and wastewater treatment [21–24]. The unique growth form of *Azolla* and its adaptability to different waters make it a valuable resource. The nutrient profile of *Azolla* is particularly remarkable [25]. It is rich in essential nutrients, including proteins, vitamins, minerals, and amino acids. In particular, the high protein content of *Azolla* combined with its rapid growth makes it a compelling candidate for biomass production [26]. Its capacity to effectively store atmospheric carbon dioxide, which aligns well with lower greenhouse gas emissions, highlights its sustainability. In addition, their unique composition offers opportunities for developing biopolymers with remarkable properties. Apart from its nutritional benefits, *Azolla* has unique properties that make it an excellent candidate for the production of bioplastics. *Azolla* has high cellulose content, which is a critical component in the production of bioplastics. *Azolla* biomass contains cellulose that can be harvested and processed into bioplastics. *Azolla*’s quick growth rate guarantees a regular flow of raw materials for bioplastic synthesis, allowing for a more reliable and sustainable manufacturing process. Compared to traditional plant-based raw materials, *Azolla* cultivation for bioplastics can have a much smaller environmental impact because of its low water and area needs [27–29].

The massive biomass of *Azolla* waste produced is underutilized and often discarded or decomposes naturally. The extraction and processing of the biomass are the initial stages of using *Azolla* to produce bioplastics. *Azolla*’s propensity to blanket the surface of

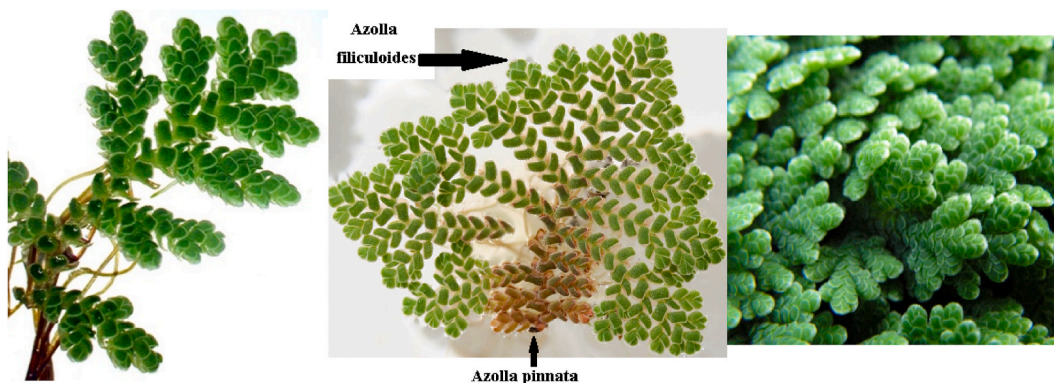


Fig. 1. *Azolla* fern.

bodies of water and its fast development make extraction and processing particularly difficult. The many techniques for gathering, desiccating, and preparing *Azolla* biomass for use in the synthesis of bioplastics are used. The selection of processing techniques may impact the quality and properties of the resulting bioplastics. Its complex organic components must be broken down into simpler biopolymers to convert *Azolla* biomass into bioplastics. Chemical and enzymatic conversion methods play an essential role in this conversion. Chemical processes such as hydrolysis and esterification are used to extract and modify the biopolymers in *Azolla* [30]. In addition, enzymatic methods utilize enzyme activity to facilitate the conversion of *Azolla* biomass into bioplastics precursors. By understanding the processes involved in converting *Azolla* biomass into bioplastics and characterizing the resulting materials, researchers and industry representatives can gain valuable insights into the potential of *Azolla* as a sustainable and environmentally friendly feedstock for the bioplastics industry. This knowledge is vital to utilize *Azolla*'s unique properties for a more environmentally friendly and sustainable approach to plastics production [24].

Recent advancements in using *Azolla* for bioplastic production focus on its potential as a sustainable feedstock, leveraging its rapid growth and high biomass yield [31]. Researchers are increasingly exploring using *Azolla* biomass as a raw material for bioplastics. Studies have shown that *Azolla* can be processed to extract polysaccharides, which can be used in bioplastic formulations [32]. This approach highlights *Azolla*'s potential to contribute to a circular economy by utilizing waste materials [33]. Also, recent research has investigated the fermentation of *Azolla*-derived substrates to produce PHAs, a type of bioplastic. This process involves using specific bacteria that can convert the organic matter in *Azolla* into PHA, which is biodegradable and can serve as an alternative to conventional plastics [34]. Innovations in creating bioplastic composites that incorporate *Azolla* fibres have emerged. These composites aim to enhance the mechanical properties of bioplastics while maintaining biodegradability [35]. The addition of *Azolla* not only improves strength but also reduces the environmental footprint of the materials. *Azolla*'s nutrient-rich profile is being utilized to enhance the growth of microorganisms in bioplastic production. By using *Azolla* as a substrate, researchers can optimize the fermentation processes, leading to higher yields of bioplastics [36]. Life cycle assessments (LCA) are increasingly being conducted to evaluate the environmental benefits of using *Azolla* for bioplastic production [37]. These studies often conclude that *Azolla* can significantly reduce greenhouse gas emissions compared to traditional plastic production methods. These advancements indicate a growing interest in *Azolla* as a sustainable alternative for bioplastic production, aligning with global efforts to reduce plastic waste and promote environmentally friendly materials [38].

In this review, *Azolla* was investigated being to be an innovative approach, a sustainable and environmentally friendly alternative to conventional plastics. The use of *Azolla*-based bioplastics offers numerous advantages over traditional plastics. First, *Azolla* is a renewable resource that proliferates and requires little water and fertilizer. This makes it an economically viable and sustainable alternative. In addition, *Azolla*-based bioplastics have a significantly lower carbon footprint than conventional plastics because they do not rely on fossil fuels for production. These bioplastics are non-toxic and can decompose safely within a few months, reducing environmental pollution risk. By focusing on these objectives, research into *Azolla* as a bioplastic feedstock aims to contribute to a circular economy and promote sustainable practices in material production [39–41].

## 2. Bioplastics: types and properties

Bioplastics are derived from plant or microbes sources. Bio-based plastics are biodegradable, can degrade and destroy when introduced in the environment, and break down naturally over time. Parkesine is the first man-made bioplastics (1862) derived from cellulose [42]. After these efforts regarding bioplastics, many studies were started to develop bioplastics because of awareness in society regarding the effect of plastic waste on the environment, such as the destruction of ecosystems, marine pollution, greenhouse gas emission and other environmental problems. The need for new environmentally friendly substitutes for traditional plastics is growing in light of this dilemma [2].

Biodegradable plastics have become a new way to escape the damage caused by the increasing rate of plastics use due to their potential biodegradability and harmlessness [43]. However, biodegradable plastics are still only a small part of the global plastics market and require more effort in research and commercialization. Bioplastics are good quality, and in terms of resistance to wear and tear, stability, lightweight, and strength; they are equal to conventional plastics, and therefore they are used for similar applications. Increasing the use of bioplastics is very useful and will reduce human dependence on fossil fuels and reduce the environmental risks associated with it. The additional benefits of bioplastics include eco-friendly, biodegradable, low greenhouse gas emission, energy efficiency, reclamation of byproducts, reduce carbon footprint, reduction in litter, greater water vapor permeability, clearer and more transparent, good surface quality and less probability of imparting different taste to the product when stored.

The field of bioplastics encompasses a variety of materials, each with its composition and properties. Every polymer is described as bioplastics that are either biobased (derived from microbes or renewable raw materials), biodegradable (breaks down naturally, under the right environmental conditions), or both. Thus, bioplastics can be classified into three categories [44–46].

- 1 **Fossil-based biodegradable plastics**, a category of plastics that decomposes naturally in the environment. Polyesters, such as polycaprolactone, polybutylene succinate, polybutylene adipate-co-terephthalate and polyglycolic acid, are in this category. The ester linkage in these plastics makes them unstable and suitable for biodegradation.
- 2 **Bio-based and non-biodegradable plastics**, such as polyethylene and polyvinyl chloride, are included in this category. These plastics are chemically similar to category 1. However, because they do not release additional carbon dioxide during incineration, they have a lower carbon footprint. Bio based plastics such as polyethylene (PE), polyethylene terephthalate (PET) and polymers, such as polyamides (PA) and polyurethanes (PUR) are in this category.

**3 Bio-based and Biodegradable Plastics** are derived from natural polymers like polysaccharides, proteins, from animal origins or lipids of plants. Plastics are made of microorganisms such as poly hydroxybutyrate are in this category. Compostable plastics, such as polylactic acid (PLA), polyhydroxyalkanoates (PHA), polybutylene succinate (PBS), and starch blends are included in this category.

The classification of four types of degradable plastics is shown in Fig. 2. Only fossil based non-biodegradable plastics are not included as bioplastics.

The mechanical characteristics of bioplastics include tensile strength, flexibility, and impact resistance [47]. Additives, manufacturing techniques, and polymer mix are some variables that affect these qualities. One of the most critical aspects of developing bioplastics is customizing their features to meet the demands of specific applications [48]. The thermal properties of bioplastics, such as their melting point and glass transition temperature, vary depending on the polymer type and intended use [8]. Understanding these properties is crucial for processing and application design. Some bioplastics may have lower heat resistance than conventional plastics. One of the main advantages of bioplastics is their biodegradability [49]. They can be broken down into harmless substances under the right conditions, reducing their environmental impact. However, the rate and extent of biodegradability can vary among different types of bioplastics and require specific disposal conditions.

Bioproduction and biodegradation of bioplastics are shown in Fig. 3. The degradation mechanism starts when microbes attaching to the surface of polymers followed by secretion of extracellular enzymes for hydrolysis attack, which induces depolymerization of high molecular weight polymers into simple monomers [50]. Biodegradation by microbes usually occurs under both aerobic and anaerobic conditions, but the aerobic process is more efficient in terms of energy gain. In aerobic digestion, microbes utilize oxygen and release water and carbon dioxide as end products. In anaerobic digestion, biodegradation of polymers occurs in the absence of oxygen (anaerobic and methanogenic microbes) and yields methane, carbon dioxide, water, and other metabolites as end products [51–53].

### 3. Azolla as a source of bioplastics

Petroleum and fossil fuels are usually used to produce conventional plastics. Our need for non-renewable fossil fuels multiplies the problem of conventional plastics. But bioplastics are often obtained from the fermentation or chemical synthesis of renewable resources such as sugarcane, corn and kawsa [54].

Making bioplastics from renewable resources reduces the adverse effects of conventional plastics on the environment. Therefore, the production of bioplastics is environment friendly due to the reduction of fossil fuel consumption. Growing plants to produce bioplastics can help reduce atmospheric carbon dioxide levels. This carbon reduction is beneficial for both the environment and bioplastics. However, the use of agricultural raw materials such as sugar, wheat, potatoes, corn and rice in the production process of bioplastics has the potential to negatively affect global food production.

Hence, there is the demand for economically efficient bioplastics feed stocks. Azolla fern is known to be an environmental problem and is freely available [55]. Azolla biomass has the potential to serve as a viable and environmentally sustainable alternative to

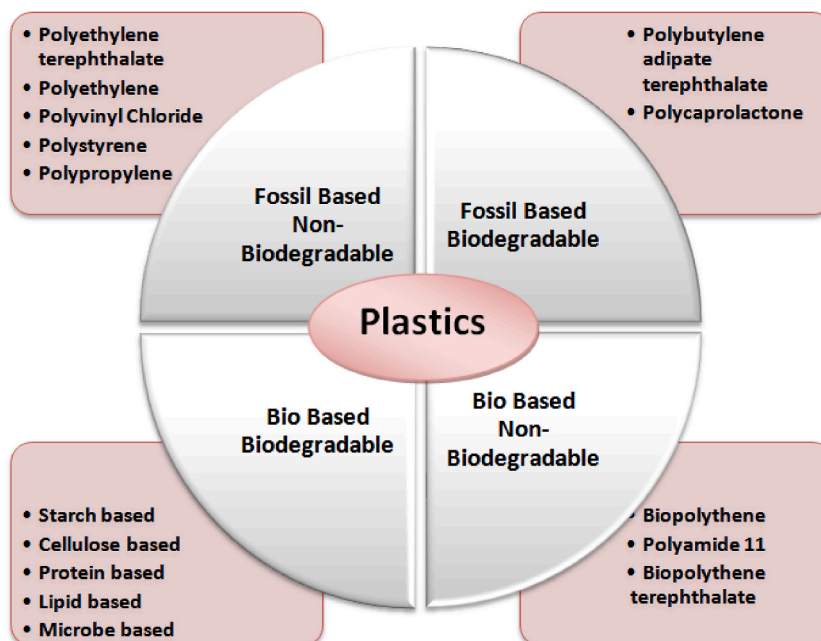


Fig. 2. Classification of four types of degradable plastics.

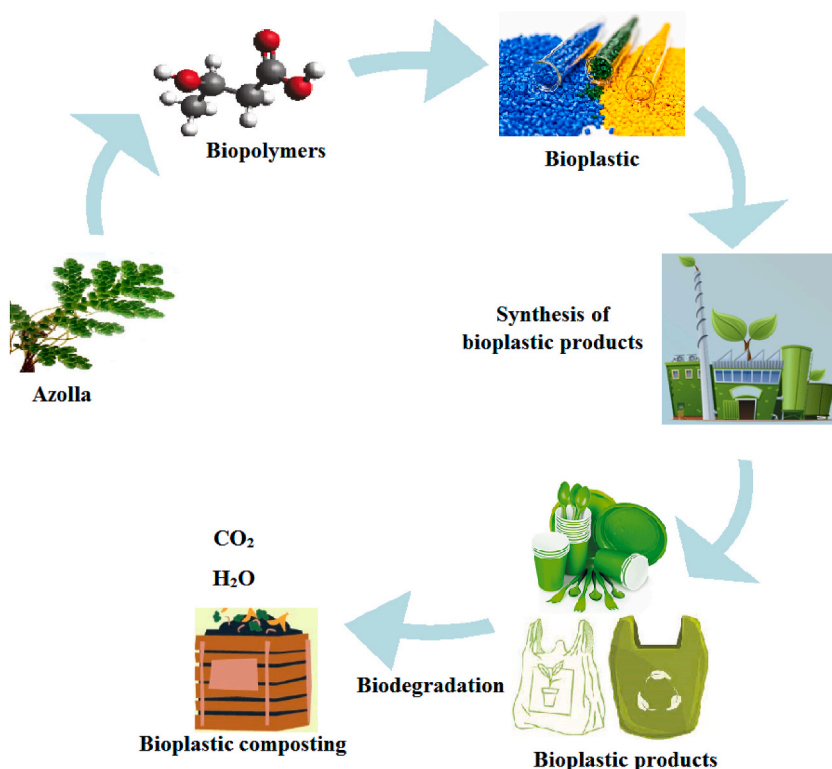


Fig. 3. Bioproduction and biodegradation of bioplastics.

traditional plastic, thereby offering economic advantages. Azolla presents numerous quantitative benefits for bioplastics production, making it an attractive alternative to conventional feedstocks.

Below are the benefits of using Azolla as a feedstock for bioplastics production:

**High Biomass Productivity:** Azolla is freely and can double its biomass in days and shows excellent growth rates. It can achieve an average productivity of 90.0–97.2 kg/ha per day DW (dry weight) when continuously harvested over extended periods. Azolla has a rapid growth rate, doubling its biomass in less than two days under optimal conditions. It does not compete with human food [56].

**Nutritional Composition:** Azolla is rich in essential nutrients including proteins, lipids, vitamins, cellulose, minerals and amino acids, which can act as an effective agent for producing polyhydroxybutyrate [57].

·**Proteins:** Azolla has high protein levels, making up 176–208 g/kg of dry weight. This is significantly higher in essential amino acids than soybean, making it a valuable protein source for bioplastics and animal feed. These proteins can potentially create protein-derived bioplastics such as zein, gluten, or casein [58].

·**Lipids:** Lipids in Azolla biomass range from 79 to 100 g/kg on a dry weight basis. These fats can create lipid-derived bioplastics like polyhydroxyalkanoates [59].

·**Phenolic substances:** Azolla also has 21–69 g/kg of (poly) phenols when measured on a dry weight scale. Although Azolla's inclusion rate in animal diets may be restricted due to high phenolic levels, these compounds could improve bioplastic characteristics [60].

By extracting and processing these biomolecules, Azolla can be a sustainable feedstock for producing a wide range of bioplastic products, including pellets, films, fibers, coatings, adhesives, and molded articles. The bioplastics derived from Azolla are entirely non-toxic and biodegradable, making them suitable for food packaging, agriculture, and consumer goods [61].

**Environmental Benefits:** Azolla's growth contributes to carbon capture; it sequesters large amounts of CO<sub>2</sub>, making it a sustainable option for reducing greenhouse gas emissions. Also, the symbiotic relationship with cyanobacteria allows Azolla to fix atmospheric nitrogen, reducing the need for synthetic fertilizers and enhancing soil health. It also has a high degradation speed [62].

**Economic Viability:** Azolla can be grown on small plots of land with low labour costs, making it economically viable for bioplastic production, especially in regions with limited arable land. Azolla can be integrated into biorefinery systems, producing bioplastics and biofuels, biofertilizers, and animal feed, maximizing resource efficiency and profitability. The quantitative values associated with Azolla highlight its potential as a sustainable and economically viable feedstock for bioplastics production. Its high biomass yield, rich nutritional content, environmental benefits, and integration capabilities make it a promising alternative for sustainable materials [63].



### 3.1. Various methods of producing bioplastics through *Azolla* biomass

There are different methods to produce bioplastics by *Azolla*, such as direct use of *Azolla* biomass, biorefinery or combination approaches. Chemical and enzymatic conversion methods play an essential role in utilizing *Azolla* as a feedstock for bioplastics production. These processes allow for the efficient extraction and conversion of *Azolla*'s biomolecules into useable forms for bioplastic synthesis [64].

**Cellulose and Hemicellulose Extraction:** *Azolla*'s cell walls are rich in cellulose and hemicellulose, which can be extracted through chemical or enzymatic hydrolysis. Acid hydrolysis using sulfuric acid or enzymatic hydrolysis using celluloses and hemicelluloses breaks down these polysaccharides into fermentable sugars that can be used for bioplastic production [65].

**Proteins Extraction:** *Azolla* contains significant amounts of proteins. These proteins can be extracted using chemical methods like alkaline extraction or enzymatic methods involving proteases. The extracted proteins can then produce protein-based bioplastics like zein, gluten, or casein [66].

**Lipid Extraction:** Lipids can be extracted using chemical solvents like hexane or supercritical fluid extraction. The extracted lipids can be used to produce lipid-based bioplastics such as polyhydroxyalkanoates [67].

**Polymer Synthesis:** The extracted biopolymers can be converted into bioplastics using conventional plastic manufacturing methods like extrusion, injection molding, or compression molding. Chemical modification or enzymatic treatment may be used to enhance the properties of the bioplastics [68].

These chemical and enzymatic conversion methods enable *Azolla*'s biomolecules to be efficiently extracted and processed into a wide range of bioplastic products. Integrating these methods into an *Azolla* biorefinery system enables the sustainable and economical production of bioplastics alongside other valuable products [54].

**Blending *Azolla* biomass with bioplastics:** PLA (Poly Lactic Acid) is a fully biodegradable plastic produced from plant sources such as sugar beet and corn, making them excellent candidates for blending with *Azolla* biomass, as they are biodegradable [69].

**Blending *Azolla* Biomass with Starch:** Starch that contains long-chain amylase and amylopectin has good film-forming properties and is widely used in several industrially based products such as bioplastics. Starch-based bioplastics are often affordable, biodegradable and renewable. Most importantly, bioplastics are starch blends are rapidly biodegradable with the increase of starch to *Azolla* biomass ratio [70].

***Azolla* biomass as a feed for PLA Production:** Although PLA is a biodegradable bioplastic, but because it is derived from plant resources such as corn, sugarcane, etc, etc, it competes with human food. So to solve this problem PLA can be produced by the fermentation of *Azolla* biomass with bacteria and then polymerization [71].

**Biorefinery Approach to Producing PHAs:** The bio-refining method allows the production of more products from *Azolla* biomass using less energy compared to fossil refineries. The biorefinery approach has a wide range of benefits, such as producing more products with low energy, cost-effectiveness, and minimal waste. The most usual biorefinery approach is *Azolla* biomass to use *Azolla* biomass as feedstock for production products containing ethanol, biodiesel and Polyhydroxybutyrate (PHB) formation. *Azolla* biomass produces a wide range of products such as starch, proteins, vitamins, lipids, biopolymers, etc. These products can be used using biorefining method as raw materials for production such as bioplastics, biofuels, biofertilizers, etc [72].

**Hydrolysis of *Azolla* biomass for PHA production:** Hydrolysis of *Azolla* biomass for PHA production can be used for bioplastics production. The researchers showed when recombinant *E. coli* is grown *Azolla* biomass, PHB biopolymer production can be achieved.

Significant research has provided evidence about the prospective utilization of *Azolla*, in producing fundamental constituents of biodegradable polymers, such as PHB. Overall, the development of bioplastics from *Azolla* biomass has the potential to revolutionize the plastic industry while addressing the environmental concerns associated with traditional plastic [73].

There are some essential technical factors when utilizing *Azolla* for bioplastic production. Regarding the growth and collection of products, it is worth mentioning that *Azolla* can be grown in ponds, lakes, or bioreactors. The fast growth rate allows for high yields, as the biomass doubles in less than 48 h. Biomass can be collected using basic methods such as skimming or filtration [74]. Then materials in the downstream must be processed. Following the harvest, the *Azolla* biomass goes through multiple processing stages to extract the required biopolymers. This can involve drying, grinding, chemical extracting, enzymatic breaking down, fermenting, and creating polymers [75]. Super critical fluid extraction, advanced methods, also have potential for utilization [76]. *Azolla*'s cell walls contain substantial levels of cellulose and hemicellulose. Such polysaccharides may be extracted for the production of bioplastics via methods such as acid hydrolysis, enzymatic hydrolysis, or chemical derivatization. *Azolla* also has significant amounts of proteins and lipids. These biomolecules are utilized in the creation of bioplastics by means such as protein-based plastics (e.g. zein, gluten, casein) or lipid-based plastics (e.g. polyhydroxyalkanoates) [77].

Through traditional plastic production techniques such as extrusion, injection molding, or compression molding, the biopolymers obtained from *Azolla* can be transformed into bioplastics. The addition of substances like plasticizers or cross-linking agents can further enhance the qualities of these bioplastics. By incorporating *Azolla* into a larger biorefinery system, we cannot only produce bioplastics but also other essential products such as biofuels, animal feed, and biofertilizers, making *Azolla* a key player in the bioplastics production process.

Overall, *Azolla* shows potential as a viable source for creating bioplastics due to its high biomass production, cellulose and hemicellulose levels, and ease of cultivation. Incorporating bioplastics manufacturing into an *Azolla* biorefinery can improve the operation's overall sustainability and economic feasibility [57].

### 3.2. Expanding *Azolla* cultivation for bioplastics manufacturing

*Azolla* is an excellent candidate for bioplastic production because of its abundant biomass and favorable chemical makeup. To facilitate the production of bioplastics on a large scale, *Azolla* cultivation can be expanded through the utilization of the following methods:

**Improving the Growth Conditions of *Azolla*:** Optimizing nutrient availability, light, temperature, and CO<sub>2</sub> levels are crucial factors for maximizing *Azolla* biomass production. Studies have indicated that increased CO<sub>2</sub> levels at 800 ppm can increase *Azolla* biomass by 36–47 % while maintaining protein content [78].

**Creating *Azolla* strains with high productivity:** Genetic modification and selective breeding can develop *Azolla* varieties with increased growth rates, biomass yields, and desired chemical compositions for making bioplastics [34].

**Using Continuous Harvesting Techniques:** *Azolla* can be constantly gathered from cultivation ponds or bioreactors, allowing for consistent high yields for extended periods. During a study, *Azolla* cultures were harvested non-stop for more than 100 days, producing an average of 90–97 kg dry weight per hectare per day [79].

**Combining *Azolla* Farming with Biorefineries:** *Azolla* farming can be incorporated into a biorefinery setup that generates biofuels, animal feed, biofertilizers, and other valuable items. This allows for effective use of all *Azolla* parts and enhances the overall financial aspect of the system [80].

**Improving the Efficiency of Post-Production Processes:** Methods such as enzymatic hydrolysis, fermentation, and polymer synthesis can be fine-tuned to effectively extract and transform *Azolla*'s cellulose, hemicellulose, proteins, and lipids into bioplastics [81].

By utilizing these tactics to expand the cultivation and processing of *Azolla*, it becomes possible to manufacture bioplastics in large quantities using this sustainable, high-yield material source. Incorporating *Azolla* biorefineries into a circular economy framework optimizes the utilization of this adaptable water plant [82].

### 3.3. Industries utilizing *Azolla* for bioplastics

#### 3.3.1. Sectors employing *Azolla* for biodegradable plastic production

In recent years, *Azolla*, a small water fern, has been noticed for its possible application in bioplastics. Bioplastics, made from sources like vegetable fats, oils, and starches, present a sustainable option instead of conventional petroleum-based plastics. Several sectors have started to investigate the utilization of *Azolla* in the production of bioplastics [83].

**Farming and cultivation of crops and livestock:** *Azolla* is under investigation for its potential use as a feedstock in the agricultural sector for producing bioplastics. Its fast growth rate and capacity to fix nitrogen make it an appealing choice for sustainable biomass production. Scientists are investigating ways to collect and convert *Azolla* into materials that can be used to make bioplastic [35].

**Wrapping materials:** The packaging sector is engaging in research on the utilization of *Azolla*-derived bioplastics for a range of purposes, including food containers, single-use utensils, and packaging products. These bioplastics provide biodegradability and lower environmental effects than traditional plastics [78,79,84].

**Table 1**

Comparison *Azolla*-based bioplastics with some other common bioplastics.

Feature/Characteristic	<i>Azolla</i> -based Bioplastics	PLA (Polylactic Acid)	PHA (Polyhydroxyalkanoates)	Starch-based Bioplastics
<b>Raw Material Source</b>	<i>Azolla</i> biomass (cellulose, hemicellulose, proteins and lipids)	Corn starch and sugarcane	Microbial fermentation of sugars	Corn starch, potatoes and tapioca
<b>Biodegradability</b>	High (within months)- entirely non-toxic and biodegradable, breaking down into nutrients for soil	Moderate (can take years) compostable under controlled conditions, but may not degrade well in natural environments.	High (within months) is readily biodegradable in both aerobic and anaerobic environments	High (within months)
<b>Production Process</b>	Biorefinery, fermentation, chemical conversion	Fermentation, polymerization	Microbial fermentation	Extrusion, gelatinization
<b>Mechanical Properties</b>	Moderate to high strength	Good tensile strength	Excellent flexibility and toughness	Variable, often lower strength
<b>Thermal Stability</b>	Moderate	Suitable (up to 60 °C)	Excellent (up to 180 °C)	Lower (degrades at high temps)
<b>Cost of Production</b>	Potentially low (utilizes waste)	Moderate to high- production requires more complex and costly fermentation processes	High (due to microbial processes)- production requires more complex and costly fermentation processes	Low to moderate
<b>Environmental Impact</b>	Low (sustainable, fast-growing)	Moderate (depends on agriculture)	Low (renewable resources)	Moderate (depends on crop use)
<b>Applications</b>	<i>Azolla</i> bioplastics can be used to make a wide range of products like packaging films, fibers, coatings, adhesives, and molded articles	PLA is commonly used for packaging, disposable tableware, and 3D printing filaments	PHA has applications in medical devices, personal care products, and packaging	Food packaging, disposable items

**Car-related industries:** The automotive sector is investigating the integration of Azolla-derived bioplastics in car parts like interior trims, door panels, and dashboard elements. These bioplastics offer a more eco-friendly option than conventional plastics, without compromising on traditional plastics and without compromising strength and durability [34,85].

**Electronic devices:** The electronics sector is exploring the potential of utilizing bioplastics derived from Azolla for making casings, housings, and various components in electronic devices. These bioplastics have the potential to be more biodegradable and have a more negligible minor environmental impact [86].

**Building the structure:** The construction sector is investigating the utilization of Azolla-derived bioplastics in construction components like insulation, panels, and coatings. These biodegradable plastics offer a sustainable option compared to traditional petroleum-based materials with similar performance qualities [32].

With continuing research and development, more sectors are expected to start using Azolla-based bioplastics as a sustainable and eco-friendly option instead of conventional plastics [64].

#### 4. Comparing Azolla-based bioplastics with other bioplastics

Here is a comparison table that compares azolla-based bioplastics with some other common bioplastics and assesses its viability. In summary, azolla-based bioplastics compare favorably with other bioplastics regarding biodegradability, scalability, cost-effectiveness, and environmental benefits. Its unique properties and ability to be integrated into a biorefinery system make it a promising feedstock for sustainable bioplastics production. With further research and development, azolla has the potential to become a viable alternative to conventional plastics and other bioplastics [78,84] (see Table 1).

#### 5. Challenges and limitations

Azolla is found in different parts of the world, especially in tropical and temperate regions. It grows in freshwater environments such as ponds, lakes, low-flow rivers and rice fields. Azolla species are well adapted to survive in diverse climatic conditions, including humid and arid regions [18]. Azolla's unique characteristics and ecological importance make it an attractive subject for study. This section examines the limitations in growing and harvesting Azolla, the technological hurdles in converting Azolla into bioplastics, the compatibility with existing bioplastics production processes, and the economic feasibility and scalability of this innovative approach [87].

The widespread use of Azolla as a source of bioplastics has limitations because its consumption is very high. In addition, Azolla biomass is highly perishable and cannot be stored for long. Therefore, all these applications of Azolla require the production of its fresh biomass in bulk. Azolla is cheap to grow and can be propagated in natural waters to produce biomass. Biomass productivity depends on the doubling time and relative growth rate and for its high nitrogen fixation potential [88]. Although Azolla is a fast-growing plant, several challenges are associated with its cultivation and harvesting. These include the need for a controlled aquatic environment, susceptibility to contaminants, and vulnerability to climatic fluctuations [21]. Ensuring consistent, large-scale cultivation of Azolla as a raw material for bioplastics requires careful attention to these factors and the development of sustainable cultivation methods. The conversion of Azolla biomass into bioplastics involves complex chemical and enzymatic processes [50]. The availability of efficient and

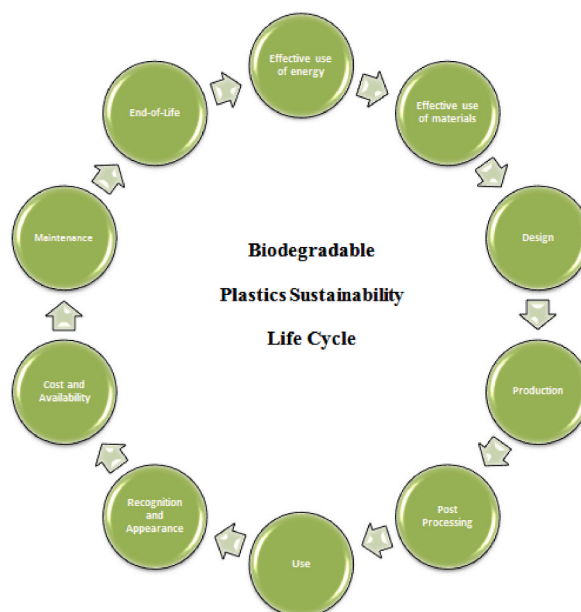


Fig. 4. Proposed sustainability life cycle for biodegradable plastics.



cost-effective technologies for biomass conversion is a crucial challenge. Researchers need to optimize these processes to maximize the yield and quality of bioplastics while minimizing the environmental impact [35].

The production of bioplastics generally uses established manufacturing processes and infrastructures that were developed for conventional plastics. The integration of Azolla-based bioplastics into existing production systems poses challenges in terms of compatibility, including equipment and processing parameters. Adapting these systems to Azolla-based feedstocks requires research and development efforts. The economic viability of Azolla-based bioplastics is a significant consideration. Although Azolla is abundant and rapidly growing, the costs associated with cultivation, harvesting, and conversion must be carefully evaluated [89].

Furthermore, achieving scalability to meet the demands of large-scale bioplastics production presents economic and logistical challenges. These challenges and limitations underscore the complexity of developing Azolla-based bioplastics as a sustainable alternative to traditional plastics. Addressing these issues is essential for realizing the full potential of Azolla as a renewable feedstock for bioplastics and ensuring its competitiveness in the market. Overcoming these obstacles requires interdisciplinary collaboration, technological innovation, and a commitment to sustainable practices in agriculture and manufacturing [73].

## 6. Environmental and sustainability aspects

In this section, we explore the Azolla-based bioplastics' environmental and sustainability aspects and focus on critical dimensions such as conducting a life cycle analysis (LCA) of Azolla-based bioplastics, assessing their carbon footprint, green house gas emissions, evaluating their biodegradability and environmental impact. The use of biotechnology and the usage of alternative feedstock have both been statistically shown to support the sustainability [35,90]. Proposed sustainability life cycle for biodegradable plastics is shown in Fig. 4.

Life cycle analysis (LCA) is a quantitative method to evaluate potential effects (positive and negative) of products on the environment during a product's life cycle. LCA provides a comprehensive assessment of the environmental impact of a product from its inception to disposal [37]. Two types of studies were applied to study LCA of bioplastics.

- 1 Cradle-to-gate (upstream product process), which includes raw materials from Products, extraction, and manufacturing.
- 2 Cradle to grave (downstream product process), which includes end-of-life assesses the impact of recycling or biodegradation process/final removal of the product at the end, followed by analyzing the impact of bioplastics on the environment [51].

Evaluating Azolla-based bioplastics through LCA helps us understand the sustainability of these materials throughout their entire life cycle, encompassing cultivation, processing, manufacturing, use, and end-of-life scenarios. This analysis considers factors such as resource use, energy consumption, toxicity, greenhouse gas emissions, global warming, recycling and biodegradation to assess Azolla-based bioplastics' ecological, Environmental and sustainability aspects [91].

Based on LCA studies of bioplastics, Azolla-based bioplastics eliminate toxic production processes and reduce greenhouse gas emissions, and finally, it reduces the negative effects of bioplastics on the environment. Assessing Azolla-based bioplastics carbon footprint and greenhouse gas emissions is essential for determining their contribution to mitigating climate change. The cultivation and processing of Azolla, as well as the manufacturing of bioplastics, generate emissions [52]. However, Azolla's capacity to sequester atmospheric carbon dioxide offset some of these emissions.

important to investigate the effects of the end of the bioplastics life cycle on the environment. Investigating the effects of the end of the bioplastics life cycle on the environment is very important. Life cycle analysis studies showed that disposal of bioplastics by burial or incineration is inappropriate. One of the most important advantages of bioplastics is their potential for biodegradation, which can reduce the environmental harm associated with persistent plastic waste [53]. Assessing the biodegradability of Azolla-based bioplastics is crucial for understanding their behavior in natural environments and composting facilities.

By analyzing these environmental and sustainability aspects, we gain insights into how Azolla-based bioplastics align with the broader goals of reducing plastic pollution and environmental degradation. This assessment contributes to the ongoing discourse on sustainable materials and provides valuable information for policymakers, industries, and consumers seeking eco-friendly alternatives to traditional plastics. More time is needed to conduct more comprehensive research to overcome the existing barriers to the production of bioplastics. This eco-friendly approach will reduce our dependence on fossil fuel-based polymers [16].

## 7. Future prospects

This section explores the dynamic and evolving landscape of Azolla-based bioplastics. It highlights the path forward in realizing their potential as a green alternative by considering innovative approaches to enhance their properties, potential synergies with the circular economy and waste reduction strategies, and their integration into the existing bioplastics industry and market trends [92].

The future of Azolla-based bioplastics holds exciting possibilities for enhancing their properties. Researchers are continually exploring innovative approaches, such as genetic modification of Azolla strains, to tailor the biopolymer composition and improve the bioplastics' biodegradable characteristics, processability, advancements in manufacturing technologies, expansion of applicability, establishment of standards, reduction in the cost of production and mechanical properties such as heat and shock-resistance [93,94]. Advances in nanotechnology and polymer processing techniques may also unlock new avenues for refining the material properties of Azolla-based bioplastics [21].

Azolla-based bioplastics can play a vital role in promoting circular economy principles. Manufacturers can extend the life cycle of Azolla-based bioplastics by designing products for reusability and recycling. Moreover, Azolla cultivation can be integrated into

wastewater treatment processes, simultaneously cleaning water bodies and providing a sustainable feedstock for bioplastics [95]. For Azolla-based bioplastics to make a significant impact, their seamless integration into the existing bioplastics industry and their alignment with market trends are essential. This involves collaboration with existing manufacturers and compliance with bioplastics regulatory standards. Additionally, understanding market dynamics, consumer preferences, and the growing demand for eco-friendly alternatives is vital for positioning Azolla-based bioplastics as a competitive and sustainable choice in the global marketplace [14].

As the development of Azolla-based bioplastics continues to evolve, prospects are encouraging. Innovations in materials science, coupled with a commitment to sustainability, hold the potential to position Azolla-based bioplastics as a compelling solution to address the environmental challenges posed by conventional plastics [96].

## 8. Conclusion

This review provides a comprehensive overview of Azolla-based bioplastics, offering insights into their viability and implications for the broader bioplastics and sustainability landscape. Our exploration of Azolla-based bioplastics has revealed several key findings. Azolla, an aquatic fern, exhibits impressive growth rates and high nutritional content, making it a compelling source of bioplastics feedstock. The processes involved in extracting and processing Azolla biomass and chemical and enzymatic conversion methods have been examined in detail. We have discussed the challenges related to Azolla cultivation, technological hurdles in conversion, compatibility with existing manufacturing processes, and the economic feasibility of scaling up production. The review also assessed Azolla-based bioplastics environmental and sustainability aspects, including their life cycle analysis, carbon footprint, and biodegradability. Finally, we explored the prospects of Azolla-based bioplastics, emphasizing innovative approaches, synergies with circular economy principles, and integration into the existing bioplastics industry and market trends. These findings revealed that Azolla-based bioplastics hold significant promise as an eco-friendly alternative to conventional plastics. Their unique properties, rapid growth, and nutritional content make them a strong contender for sustainable bioplastics feedstock. However, challenges related to cultivation, processing, and economic feasibility must be addressed for practical implementation. Azolla-based bioplastics present an opportunity to reduce the environmental impact of plastic waste and contribute to a more sustainable future.

Additionally, integrating Azolla-based bioplastics into circular economy and waste reduction strategies aligns with the global push for sustainable resource management. As a result, bio-plastics production based on Azolla creates a suitable and sustainable solution to deal with the existing environmental challenges with traditional plastics. Their development and integration into the bioplastics industry represent a positive step toward a more sustainable and eco-friendly future. Further research and collaboration will be instrumental in realizing the full potential of Azolla-based bioplastics and driving progress in the broader field of sustainability and materials science.

Future research on Azolla can focus on optimizing its cultivation techniques and investigating its potential applications in various fields. Investigating the best management practices for sustainable cultivation of Azolla, including nutrient optimization and disease control, can contribute to its efficient and widespread use as bioplastics feedstock. Furthermore, exploring future research and the prospect of new applications such as Azolla-based bioplastics, pharmaceutical compounds, and of biotechnological developments can open up new opportunities for using Azolla.

Bioplastics production and sales are developing quickly, and world-famous brands have become interested in investing in this field. Scientific research is needed to reduce the problems associated with current bioplastics production technology and eliminate our dependence on conventional fossil-based polymers.

## CRedit authorship contribution statement

**Reyhaneh Kouchakinejad:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Zahra Lotfi:** Writing – review & editing, Validation, Methodology, Investigation, Conceptualization. **Abooli Golzary:** Writing – review & editing, Validation, Supervision, Project administration, Methodology, Conceptualization.

## Availability of data and materials

The data that support the findings of this paper are available from the corresponding author upon reasonable request.

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