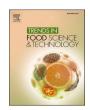
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Spirulina application in food packaging: Gaps of knowledge and future trends

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ARTICLEINFO

Keywords: Active packaging Bioactive compounds Smart packaging Natural indicators

ABSTRACT

Background: Conventional plastics have been widely applied in the food industry as packaging material because of their attractive characteristics, such as great mechanical resistance and moldability, as well as their highly cost-effective ratio. However, their use have negative impacts on the environment, including waste generation and pollution. Biodegradable and edible films produced from naturally available biomacromolecules have been seen as an alternative to synthetic polymers derived from petroleum in the development of food packaging. In this context, due to its rich composition and interesting bioactive properties, *Spirulina* has been considered a promising raw material for the production of multifunctional packaging materials.

Scope and approach: This article comprehensively reviews along it sections the state of the art in using Spirulina and phycocyanin to develop active and smart biodegradable and edible materials for application as food packaging, thus serving as a guide for researchers. Finally, the last section addresses recent trends and challenges related to the development of more environmentally friendly Spirulina-based packaging.

Key findings and conclusions: Spirulina has been gaining prominence in the food, pharmaceutical and cosmetic industries because of its rich composition and bioactive properties. Thus, studies have been conducted to analyze its potential use as a biopolymer and source of bioactive compounds in the development of active and smart packaging systems so as to extend and monitor the shelf life of packaged foods. Although the results are promising, few studies in the literature elucidate its use, which justifies the need to expand and encourage this line of research.

1. Introduction

When we think about food, we cannot only consider its use as an ingredient or its sensory and microbiological qualities; the production chains and the environmental impacts caused by food production, distribution, and consumption must also be taken into account. In this context, it is impossible to think about foodstuffs without turning our attention to packaging technologies. The growing concern of the population and researchers about the environmental impacts caused by the use of non-degradable materials has been noticeable. Due to the pandemic situation in which society currently lives, the demand for

packaged products has increased significantly over the past few years (Terra et al., 2021a). In 2018, the production of synthetic plastics almost crossed the 360 million tons mark, with only less than half of their consumption being recycled (Rai et al., 2021). Their profitability continued to grow in 2022, when the global packaged food market was valued at \$1.9 trillion, with an estimate to reach \$3.4 trillion by 2030 (Kan & Miller, 2022).

In addition to being a profitable trade worldwide, conventional food packaging is primarily intended to provide mechanical protection to the product and prevent possible microbiological and chemical contamination (Cheng et al., 2022; Flórez et al., 2022) during its transportation,

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storage, and sale (Braga et al., 2021; Oliveira-Filho et al., 2020; Zhang et al., 2022). However, most synthetic plastics derive from fossil fuel materials (Luo et al., 2021), such as polyethylene, polyethylene terephthalate, polyvinyl chloride, polypropylene, and polystyrene (Cheng et al., 2022), thus being non-biodegradable (Luo et al., 2018; Moghaddas Kia et al., 2018). This can cause significant environmental impacts (Chentir et al., 2019), especially those associated with pollution and waste production, due to the emission of greenhouse gases and the lack of circularity of plastic packaging, respectively, being mainly designated for single use and subsequent disposal (Kan & Miller, 2022).

An alternative to minimize these negative impacts is the use of biopolymers to produce biodegradable plastic packaging (Zhang et al., 2022). Although not all are entirely biodegradable, depending on their composition they can be easily decomposed by enzymatic action and microorganisms present in the soil in water, carbon dioxide, and biomass (Moghaddas Kia et al., 2018). Natural polymeric substrates such as proteins, polysaccharides and lipids from vegetables, animals, microorganisms, or food waste (Balti et al., 2017; Cheng et al., 2022; Onias et al., 2016) can be used alone or together to produce edible films and coatings.

The use of only one biopolymer implies films with delicate mechanical properties and susceptible to environmental conditions, e.g., luminosity, oxygen, and temperature (Askari et al., 2018). The combination of different materials, in turn, enhances the barrier against oxygen, carbon dioxide and water (Ferreira et al., 2021), and could therefore be a promising alternative to conventional plastics (Chentir et al., 2018, 2019), both ecologically and functionally.

The production of bioplastics uses natural resources to manufacture packaging that not only promotes the reduction of environmental impacts related to the disposal and accumulation of waste, but also contributes to the removal of pollutant gas emissions as it uses about 65% less energy when compared to the production of conventional polymers (Zhang et al., 2022). Additionally, bioplastics are usually low-cost materials (Moghaddas Kia et al., 2018). These biopolymers can be used to synthesize edible films and coatings with various convenient properties and functionalities, acting as semi-permeable barriers to gases and water vapor, and consequently extending the shelf life of the product (Ferreira et al., 2021) and promoting its quality and safety (Kuntzler et al., 2020).

Worldwide, the consumption of algae for cosmetic, food, medical (Smith et al., 2010), and biodiesel (Fabra et al., 2018) purposes has grown exponentially. This is because algae are extremely rich in essential human health compounds, such as proteins, vitamins, amino acids, and minerals from other edible plants (Ródenas de la Rocha et al., 2009).

Among the most promising substances is *Spirulina*, a type of microalga composed of 10–20% carbohydrate, 3–10% lipid, and 50–70% protein (Stejskal, Miranda, Martucci, Ruseckaite, Aubourg, et al., 2020), besides being rich in essential fatty acids, vitamins, and pigments, especially C-phycocyanin (C-PC) (Mosayebi et al., 2022). According to Ramji and Vishnuvarthanan (2022), due to its high phytonutrient value and the presence of phenolic compounds such as salicylic, chlorogenic, caffeic acids, and tocopherol, this microalga has immense potential to be applied as a medicine and nutritional additive.

The *Spirulina* species most used in dietary supplements are *S. maxima*, *S. platensis*, and *S. fusiformis* (Marles et al., 2011). In addition to the fact that the bioactive compounds of *Spirulina* can be directly applied to bioplastics, its biomass after extraction (with the addition of inorganic salts) becomes attractive for the production of bioplastics with improved physical properties (Zhang et al., 2022). A summary of the literature reports is presented in Table 1.

As already mentioned, food packaging can effectively maintain food quality and safety, providing protection against physical damage, light exposure, and possible microbiological and/or chemical contamination (Cheng et al., 2022). Therefore, it is necessary that these films have adequate mechanical and physicochemical properties. To improve them, some compounds can be incorporated into the film solution

Table 1Compilation of material and sources of ingredients in food packaging described in the literature

Material	Source	Properties	Reference
Chitosan	Spirulina	Barrier properties	Ramji and Vishnuvarthanan (2022)
Mucilage	Plantago seeds	Flexibility of the films	Tóth and Halász (2019)
Gum	Psyllium gum and modified starch	Reinforcement and stability the films networks	Askari et al. (2018)
Gelatin	Spirulina platensis	Antimicrobial effect	Stejskal, Miranda, Martucci, Ruseckaite, Aubourg, et al. (2020)
Gum	Zedo gum/ sodium caseinate and Spirulina platensis	Antioxidant effect	Moghaddas Kia et al. (2018)
Gum	Cashew gum polysaccharide	Antimicrobial effect	Moreira et al. (2020)
Phycocolloids	Red seaweed Porphyra columbina	Antioxidant effect	Cian et al. (2014)
Chitosan	Himanthalia elongata and Palmaria palmata	Antioxidant effect	Albertos et al. (2019)
Gelatin	Bovine gelatin- based films and C-PC	Antioxidant effect	Moreira et al. (2020)
Polycaprolactone (PCL) and poly (ethylene oxide) (PEO)	PCL/PEO and C-PC	pH Indicators	Terra et al. (2021a)
Polycaprolactone (PCL) and poly (ethylene oxide) (PEO)	PCL/PEO and curcumin or quercetin	pH Indicators	Terra et al. (2021b)
Protein isolate	Spirulina platensis	Antimicrobial effect	Stejskal, Miranda, Martucci, Ruseckaite, Barros-Velázquez, et al. (2020)
Chitosan	Spirulina platensis	Structural, physicochemical, mechanical, antimicrobial, and antioxidant	Luo et al. (2021)
Poly(lactic acid) (PLA) and Poly (ethylene oxide) (PEO)	PLA/PEO and Spirulina sp.	pH Indicators	Kuntzler et al. (2020)

matrix.

Studies on the use of *Spirulina* biomass (Cardoso et al., 2019; Fabra et al., 2018; Moghaddas Kia et al., 2018) and its blue extract (C-PC) (Balti et al., 2017; Chentir et al., 2019) have highlighted some positive effects on the properties of films based on these byproducts. The main characteristic of the addition of biomass in the preparation of food packaging is the increased film opacity, an interesting factor that enables its use in packaging as it provides a barrier to light. Although the incorporation of C-PC into gelatin-based films also demonstrated the same effect presented previously, there was a reduction in their mechanical properties (Chentir et al., 2019) probably due to inefficient interactions between the biopolymers, leading to brittle mechanical properties. In contrast, Balti et al. (2017) observed that the incorporation of the extract into chitosan-based films was responsible for increasing their mechanical properties, consequently enhancing their resistance.

Moreover, the need for alternative sources of biopolymers that do not

compete with food production to avoid insufficient demands for population supply has made the use of marine resources an interesting option. Because of the several advantages related to their cultivation, such as rapid reproduction and growth in a wide range of environments, their use would significantly increase the circular bioeconomy, given their reuse and return to the cycle (Fabra et al., 2018).

Therefore, the current work aims to present results found in the literature about the use of bioplastics, more specifically *Spirulina*, in food packaging through the generation and investigation of a bibliometric map of articles (Fig. 1), focusing on the reduction of some of their adverse effects on the environment, such as the use of wastewater as a source of nutrients (Cardoso et al., 2017) as well as those related to greenhouse gas emissions (Fabra et al., 2018).

To this end, an overview of the main applications of *Spirulina* in the development of edible and/or biodegradable films for food packaging is presented, highlighting their advantages and challenges as well as future trends.

2. Biopolymer-based packaging materials: an overview

Packaging is vital for containing and protecting food against physical, biological, and chemical contamination. The most used material in food packaging is plastic; however, it has generated severe environmental problems (Weber Macena et al., 2021). As a result, bio-based natural polymers or polymers synthesized with monomers from agricultural resources to develop biodegradable and/or edible materials has been proposed as an alternative to solve the plastic packaging problem (Gonçalves et al., 2021).

Biodegradable and/or edible films are produced from naturally available biomacromolecules, such as polysaccharides (e.g., starch, chitosan, pectin, cellulose, and its derivatives), proteins (e.g., zein, gelatin, collagen, and soy protein) and lipids (e.g., carnauba wax, beeswax, and palm oil), or combinations of these components (Kumar et al., 2022). Besides being sustainable, these films allow the transition from inert to biodegradable and multifunctional materials when several bioactive compounds, e.g., antimicrobial agents, antioxidants, indicators, flavors, and colors, are incorporated into the biopolymer matrix, expanding their functions (Motelica et al., 2020). Fig. 2 shows a schematic summary of the bio-based packaging materials studied in the

consulted literature.

Biopolymer packaging containing bioactive compounds can be classified as active or smart, depending on their characteristics and functions (Oliveira Filho et al., 2022). Active packaging is a system that releases substances with conservative properties (i.e., antioxidant and antimicrobial activity) and can interact with microorganisms and oxygen, delaying food quality deterioration caused by foodborne pathogens and the oxidation process (Oliveira Filho et al., 2022). In contrast, smart packaging allows the monitoring and verification of food quality and safety through smart devices (Zhang et al., 2021), such as indicators, sensors and data carriers (Zhang et al., 2022). These devices detect the presence of possible chemical, physical and biological changes (Terra et al., 2021a; 2021b), informing and alerting the consumer to the quality of the product before or after purchase (Kuntzler et al., 2020). Indicators can change color or color intensity according to the presence or absence of a given substance that causes an irreversible colorimetric reaction, thus providing qualitative information about the product (Oliveira Filho et al., 2022).

3. Applications of Spirulina and C-PC in food packaging

Numerous studies highlight the use of different compounds for food packaging, including chitosan (Ramji & Vishnuvarthanan, 2022), mucilage (Kang & Lipner, 2019; Tóth & Halász, 2019), gums (Askari et al., 2018; Zhang et al., 2022; Zibaei et al., 2021), microalgae and cyanobacteria (Larrosa et al., 2018), as well as natural compounds with antimicrobial (Stejskal et al., 2020a, 2020b) and antioxidant properties (Moghaddas Kia et al., 2018) that act as smart devices, detecting changes in food during storage (Moreira et al., 2020). All of these studies suggest positive and promising results about the use of these compounds to prepare edible films and coatings that extend the shelf life of products by reducing deterioration by microorganism action and prevent the oxidative process that occurs in foods.

Among the natural substrates that have been gaining visibility especially in western countries (Martelli, Alinovi, et al., 2020) because of their photosynthetic capacity, high growth rate, nutritional composition and bioactive properties are microalgae and cyanobacteria. Although these compounds are already established as food supplements on account of their nutritional composition (Lafarga et al., 2020),

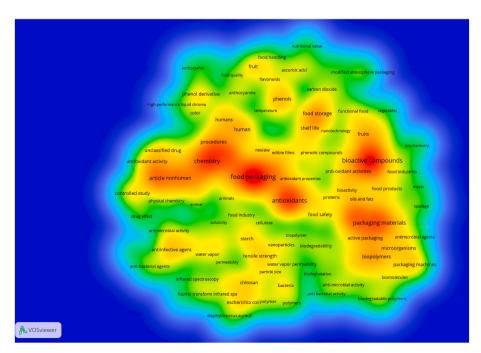


Fig. 1. Density visualization map resulting from the keywords applied in the search conducted in the present work using VosViewer software.

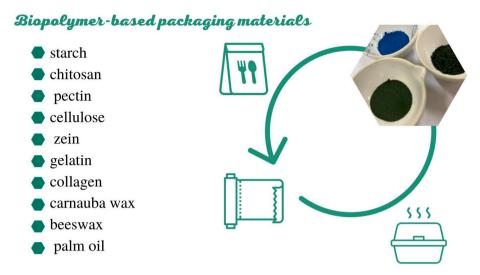


Fig. 2. Schematic summary of the biobased packaging material presented in the consulted literature.

several studies using distinct microalgae species, such as *Nannochloropsis gaditana*, *Scenedesmus* (Fabra et al., 2018), *Porphyra columbina* (Cian et al., 2014), *Chlorella* sp. (Medeiros Teodosio et al., 2021), *Spirulina platensis* (Ebrahimi & Rastegar, 2020), *Himanthalia elongata* and *Palmaria palmata* (Albertos et al., 2019), highlight their potential applicability in the production of edible films and coatings due to their thickening capability, provided by the polysaccharides and proteins synthesized by them (Ferreira et al., 2021).

The evaluation of the mechanical and physicochemical properties of films is relevant since these aspects are directly related to the protection that food packaging provides to the product. The study conducted by Fabra et al. (2018) aimed to evaluate the mechanical and physicochemical properties of films made with three different species of microalgae: Nannochloropsis, Spirulina, and Scenedesmus. After performing the tests, they verified that the microalga Nannochloropsis obtained the best results with regard to its mechanical and barrier properties (oxygen and water vapor permeability) when compared to the other microalgae.

Cian et al. (2014) also analyzed the same parameters as the previous study, together with the antioxidant activity of films made with phycobiliproteins (PF) and phycocolloids (PcF) of the microalga *Porphyra columbina*. The films were formulated with different PF:PcF ratios (0, 25, 50, 75 and 100% w/w). According to the authors, while the PcF films had excellent mechanical properties, the PF films showed high antioxidant capacity.

Besides protecting food in general, edible films and coatings can preserve fruits, vegetables, and meat products by delaying their ripening (in the case of the first two) and reducing lipid oxidation and microbial growth, for example. Several studies have focused on the application of these materials in fruits and vegetables in order to extend their postharvest shelf life using different biopolymers in their composition (Anjum et al., 2020; Dong & Wang, 2018; Kharchoufi et al., 2018; Kumar et al., 2021). For instance, Medeiros Teodosio et al. (2021) prepared edible coatings using Chlorella sp. at different concentrations (0.5, 1.0, 1.5, and 2.0%) in combination with pomegranate seed oil and studied their preservative effects on Spondias tuberosa over a 12-day storage period under refrigeration. The results showed that the coating with the highest concentration of Chlorella sp. was the one that most efficiently increased the shelf life of this fruit. Additionally, there was no mass loss, making it possible to obtain more turgid and firmer fruits while maintaining their greenish coloration for at least 10 days of storage.

On the other hand, Ebrahimi and Rastegar (2020) developed a coating based on guar gum enriched with *Spirulina platensis* extract and evaluated its effect on the mango fruit for three weeks, stored at room

temperature. During this period, the authors observed that the coating reduced the respiratory rate and mass loss of the fruit, making it firmer than the control, thus demonstrating the efficacy of the developed material in the preservative process.

Microalgae have also been applied in the packaging of meat products in recent years. Albertos et al. (2019) used the microalgae *Himanthalia elongata* and *Palmaria palmata* for the production of edible coatings and confirmed their positive and promising effects associated with their antioxidant and antimicrobial capacity on the shelf life prolongation of trout burgers. As reported by the authors, the use of edible films enriched with seaweeds in fish burgers controlled pH and water activity variations over storage and reduced microbial growth.

Shafiei and Mostaghim (2022) also prepared coatings using *Spirulina platensis* and *Chlorella vulgaris* to investigate the shelf life of calf filet. As reported, the application of 1% of *Spirulina platensis*, 2% of *Chlorella vulgaris*, 2% of chitosan, and 1% of natamycin was considered the best treatment, leading to an increase in all texture index parameters, except hardness. Moreover, the total microbial load was significantly reduced in all coated samples and the overall sensory acceptability scores were found to be significantly higher for the samples with microalgae addition than the control.

3.1. Physicochemical and barrier properties of Spirulina and C-PC-based materials

The incorporation of bioactive compounds into edible films for food packaging can positively or negatively affect the properties of these materials. The changes depend on the physical or chemical interactions between the biopolymers and the bioactive compounds, which end up affecting the structure and subsequently the functionality of the films. Therefore, it is crucial to investigate how these compounds interact with the biopolymer matrix and change the characteristics of the packaging material and packaged foods (Gonçalves et al., 2021). Many researchers have focused their studies on how the mechanical and barrier properties of biopolymer films are affected by the addition of *Spirulina*.

For application in food packaging, biopolymer films must maintain their integrity and withstand external stress, that is, they must have a good tensile strength (Taherimehr et al., 2021), which mainly depends on the structure of the polymer matrix, with ability to form strong molecular bonds between the polymer chains (Cruz et al., 2018). To be resistant, the films must have good mechanical properties, with high maximum strength (MF) and high strain at break (SB). Films with high MF and low SB are fragile, while those with high SB and low MF are weak (Moghaddas Kia et al., 2018). According to Ramji and

Vishnuvarthanan (2022), chitosan/montmorillonite-based nano-composite films incorporated with 0.5 g of *Spirulina* showed a high tensile strength of 46.3 MPa. In another study, Balti et al. (2017) observed that the addition of *Spirulina* extract improved the mechanical properties of crab chitosan-based films. The incorporation of *Spirulina* extract at a concentration of 20% increased the tensile strength of the films from 21.24 to 29.65 MPa and the strain at break from 26.13 to 34.29%.

Similar results were found in the study carried out by Luo et al. (2021), who developed chitosan films using different concentrations of *Arthrospira platensis* polysaccharide (APP) (0, 0.5% and 1% w/v). The authors identified a significant increase in the tensile strength and strain at break according to the concentration of APP. The enhanced mechanical properties of the film were attributed to strong molecular interactions between chitosan and APP. Fast-dissolving antioxidant nanofibers based on *Spirulina* and gelatin were obtained by Mosayebi et al. (2022) by electrospinning. The authors concluded that the increase in the concentration of *Spirulina* in solution mixtures resulted in films with low viscosity and electrical conductivity, in addition to an almost stable trend in their surface tension (29.25–32.19 mN/m).

The improved tensile strength of the films due to the incorporation of *Spirulina* probably occurs because of its solid intermolecular interaction with the polymer matrix, leading to a stronger interfacial adhesion and making the polymer chain more resistant to mechanical stress (Balti et al., 2017; Ramji & Vishnuvarthanan, 2022).

Barrier properties such as water vapor permeability, solubility, contact angle, and so on are the most studied properties in candidate food packaging materials because of their essential role during applications (Oliveira-Filho et al., 2020). Films and edible coatings with specific barrier properties depend directly on the type of food used and the intended application (Meritaine da Rocha et al., 2018). Certain food products are susceptible to reactions due to oxygen exposure and for this reason may be subject to some undesirable processes such as lipid rancidity, which justifies the development of films with good oxygen barrier properties. However, given the importance of the respiration process in fruits and vegetables, it is interesting that the film produced is permeable to gas exchange (Cruz et al., 2018).

Ramji and Vishnuvarthanan (2022) observed that the addition of *Spirulina* to chitosan/montmorillonite-based nanocomposite films increased their water solubility (10-13-14%) and reduced their water vapor permeability ($\sim 17-\sim 14-8~g/m^2/day$).

Terra et al. (2021a) synthesized pH indicators from electrospun fibers of C-PC, poly(ethylene oxide) (PEO) and polycaprolactone with high hydrophobicity (contact angle >90°) when compared to samples without C-PC. Regarding light barrier properties, Moghaddas Kia et al. (2018) and Benelhadj et al. (2016) produced edible films using *Spirulina*. According to them, the films containing this microalga were more opaque than those without its addition. The film opacity may be an exciting factor in fresh products sensitive to light. For this reason, further studies should be carried out in order to obtain films with enhanced structural and mechanical properties.

The barrier properties of films are essential when *Spirulina* is used as a sensor since the interaction of the polymer with the surrounding molecules will govern the speed of the sensor response according to its environment (Ramji & Vishnuvarthanan, 2022). Although studies on the effect of *Spirulina* on the mechanical and barrier properties of biopolymer films are still initial, the results are encouraging and have demonstrated a positive impact on the properties of biodegradable and edible food packaging materials.

3.2. Active packaging based on Spirulina and C-PC

As already established, food is susceptible to several environmental aspects, such as light, temperature, humidity and dust (Zhang et al., 2022), and mainly to oxidative processes and microbial degradation (Kuswandi & Jumina, 2020).

To minimize these negative impacts, food packaging materials have the primary function of protecting the product, promoting and maintaining its quality and safety (Cheng et al., 2022). As the leading causes associated with food spoilage originate from external and environmental factors, active packaging emerges as an efficient tool for this issue.

These packages are functionally active and interact directly with the food, releasing compounds with antimicrobial and/or antioxidant action, or absorbing undesirable substances that may be present there (Monção et al., 2022), thus prolonging its shelf life and maintaining its characteristics and quality (Flórez et al., 2022).

These materials can act as oxygen and ethylene scavengers, flavor/odor and moisture absorbers, ethanol emitters, preservative releasers, and temperature control systems (Kuswandi & Jumina, 2020). The most commonly evaluated packaging system is active packaging with antimicrobial and antioxidant action (Kontogianni et al., 2021; Martelli, Alinovi, et al., 2020; Mosayebi et al., 2022; Shafiei & Mostaghim, 2022).

As already mentioned, *Spirulina platensis* has gained visibility in the research area due to its rich nutritional composition and mainly its antioxidant and antimicrobial activity. The use of bioactive compounds present in this microalga can alter the permeability of the bacterial cell membrane, causing structural damage to both Gram-positive and Gramnegative bacteria (Alshuniaber et al., 2021), thus evidencing its high antimicrobial activity associated with the high amount of proteins and small peptides (Martelli, Alinovi, et al., 2020). In addition, these compounds also act as antioxidants, preventing lipid oxidation and the formation of free radicals that can cause undesirable sensory changes in foods and reduce their shelf life (Mosayebi et al., 2022).

Even though the development of studies on the application of *Spirulina platensis* and its extract in food active packaging has been growing in recent years, there is still a need for further research in this area that covers its different uses in food, seeking to better understand and comprehend its best application, the most appropriate methods of preparing films and/or nanoparticles, and the most effective concentrations of biomass and pigment to be used.

3.2.1. Antimicrobial activity

Since microbial activity is one of the main reasons for food spoilage, it is essential to develop food packaging materials with antimicrobial activity. In particular, the antimicrobial activity of *Spirulina* is entirely dependent on the alga species and the extraction protocol used (Oscar et al., 2017). Several bioactive compounds can be extracted from *Spirulina*, and the antimicrobial activity of the crude extract is possibly a result of the synergistic effect between the large amount of bioactive phenolic compounds and other compounds present in this alga, such as carotenoids, C-PC and chlorophyll (*a* and *b*) (Martelli et al., 2020). Thus, the use of *Spirulina* and C-PC as additives to food packaging becomes promising.

Tavakoli et al. (2021) conducted a dose-response study to investigate if the increase in the amount of *Spirulina platensis* extract would improve the antibacterial activity of films based on this cyanobacteria. The authors reported a significant effect of the increased amount of extract used, obtaining a different inhibitory zone with varying concentration ranges and a significant variation in the antibacterial potential of *Spirulina platensis* extract due to extraction solvent distinction.

Ramji and Vishnuvarthanan (2022) developed chitosan, clay, and *Spirulina* films with antimicrobial properties against *E. coli* and *S. aureus*. The excellent results were attributed to the interactions between the positive charges of chitosan molecules and the negative charges of the membranes of these bacteria, as well as the presence of chlorogenic acid in *Spirulina*, which was responsible for reducing the growth of these bacteria. In another work, Balti et al. (2017) proved that their chitosan films with *Spirulina* effectively eliminated *E. coli*, *S. aureus*, *P. aeruginosa*, *L. monocytogenes*, *S. typhimurium*, *B. subtilis*, and *B. cereus*. Unlike the previous example, these pure chitosan films did not show antimicrobial activity, which was attributed to the chlorogenic acid contained in *Spirulina*. Chlorogenic acid can bind and permeabilize the cell

membrane of microorganisms, causing cells to lose the ability to maintain membrane potential and leading to the formation of cytoplasmic macromolecules (Lou et al., 2011).

Chitosan/natamycin films containing *Spirulina platensis* and *Chlorella vulgaris* were used for refrigerated meat storage. As observed, the use of these microalgae decreased microbial proliferation, increasing the sensory acceptance of this food (Shafiei & Mostaghim, 2022). The encapsulation of *Spirulina* extract in chitosan nanoparticles proved to be efficient in reducing microbial counts in fish stored at low temperatures (Karimzadeh et al., 2023).

Gelatin-based films containing C-PC (bioactive extracted from *Spirulina*) showed antimicrobial activity against *M. luteus*, *E. coli*, *S. aureus*, and *Pseudomonas* sp. with C-PC concentrations above 6.25% (Chentir et al., 2019). This activity was associated with the presence of C-PC, which interacted with the membranes of these microorganisms (Safari et al., 2020). A remarkable antimicrobial effect was also obtained with gelatin and alginate/gelatin films containing *Spirulina* against *Enterobacteriaceae* when applied as packaging for fish storage (Stejskal et al., 2020a, 2020b). Edible films based on sorbitol and lysozyme were also successfully obtained with the addition of *Spirulina*, resulting in films with potential antimicrobial activity (Benelhadj et al., 2016).

3.2.2. Antioxidant activity

Recognized as safe food by the Food and Drug Administration (FDA) (Onias et al., 2016) for being non-toxic and non-carcinogenic (Chentir et al., 2018), *Spirulina* is the most well-known microalga worldwide (Larrosa et al., 2018). For this reason, the incorporation of *Spirulina* (and C-PC) into films and coatings becomes attractive, given its properties and potentially favorable results in food. Studies using *Spirulina* protein concentrate (Moreira et al., 2019; Mosayebi et al., 2022; Stejskal et al., 2020a, 2020b), biomass (Forghani et al., 2021; Kontogianni et al., 2021; Moghaddas Kia et al., 2018) and extract (C-PC) (Chentir et al., 2019; Karimzadeh et al., 2023; Moreira et al., 2019) have shown promising results regarding the antioxidant capacity of the microalga.

An analysis performed by Chentir et al. (2018) verified the feasibility of applying C-PC in bovine gelatin-based films using different concentrations of the dye (1.25%, 2.5%, 6.25%, and 12.5%). The authors observed that the incorporation of C-PC resulted in great antioxidant capacity. Karimzadeh et al. (2023) also applied Spirulina extract in chitosan nanoparticles and evaluated the increased shelf life of pike-perch filet during 14 days of refrigerated storage. According to the authors, fish filets coated with the microalga extract had low values of total volatile basic nitrogen (TVB-N) and thiobarbituric acid (TBA) on the last day of storage. This can be considered a positive result regarding food preservation. In another work, Moreira et al. (2019) used Spirulina protein concentrate and C-PC to produce PEO ultrafine fibers by electrospinning. Besides its high antioxidant potential, the authors confirmed that C-PC incorporated into the fibers was more stable.

Concerning *Spirulina* biomass, Kontogianni et al. (2021) evaluated the physicochemical and antioxidant properties of edible films based on whey protein combined with *Spirulina* biomass at different concentrations (0.5, 1, 2, 4, 6, and 8% w/w). The authors identified that the films containing 2% of biomass presented the highest percentage of radical scavenging effect. In another study, Moghaddas Kia et al. (2018) investigated the properties of biocomposite films based on Zedo gum and sodium caseinate in combination with *Spirulina* biomass at concentrations from 0 to 0.5%. The authors observed that the highest concentration resulted in films with significant antioxidant properties, corroborating the results obtained by Kontogianni et al. (2021).

Similar results were obtained using *Spirulina* protein concentrate (Moreira et al., 2019; Mosayebi et al., 2022; Stejskal et al., 2020a, 2020b), which demonstrates and reinforces the potential antioxidant characteristic of the microalga and its positive effects on food preservation.

Regarding the direct application of films or coatings on food, some researchers have discussed their use on peppers (Cardoso et al., 2017),

mango (Onias et al., 2016) and guava (Onias et al., 2018), obtaining promising results. As reported, there was a delay in mass loss and a prolongation of the shelf life of these types of food, evidencing the antioxidant capacity of the produced films. However, while the peppers and mango had their color, texture and firmness maintained, the films did not significantly influence the physical characteristics of the guava fruit

3.3. Smart packaging based on Spirulina and C-PC

The deterioration of foods such as meat, dairy products, and seafood is accompanied by changes in pH and the release of different volatile nitrogen compounds that can lead to sensory rejection and the formation of strange flavors in these products (Bekhit et al., 2021). These compounds are closely linked to changes in the food pH and its surrounding environment and can be detected using smart packaging containing pH-sensitive compounds (Fig. 3) (Zhao et al., 2022).

Alternatives to traditionally used synthetic pH indicators such as bromocresol purple and bromophenol blue have been explored. Synthetic indicators have potential toxicity that may pose a risk to the consumer's health, in addition to an inedible nature, which may alter the sensory properties of foods (Oliveira Filho et al., 2022). C-PC is a natural pigment sensitive to pH and its instability results in a color change (Patel et al., 2005). The color change of C-PC as a function of pH has makes this pigment attractive for application as a natural bioindicator in the development of intelligent packaging materials for food quality control (Moreira et al., 2018).

Three studies conducted by the same research group (Moreira et al., 2020; Terra et al., 2021a, 2021b) focused on the development of films with C-PC as a pH indicator for food quality control. The authors verified significant alterations in the film coloration according to pH changes. At alkaline pH above 8, the films tended to abruptly lose their blue and green tones, resulting in a color tending towards white. This behavior evidences their feasible application and potential use as a real-time smart device to transmit product information to the consumer.

Spirulina sp. contains in its composition some natural pigments that are sensitive to pH changes, such as β -carotene, tocopherols, phycoerythrin, and chlorophylls (Priyadarshani & Rath, 2012). Thus, the full use of the microalga biomass is promising for the development of colorimetric pH indicator films. A recent study carried out in 2020 reported the production of smart packaging through the incorporation of Spirulina biomass into nanofibers and evaluated the microalga efficiency as a colorimetric pH indicator. The colorimetric indicator containing 2% (w/v) biomass showed the best results, undergoing color changes between pH 5 and 7. As this range of pH values covers a wide variety of fresh foods, this material can be successfully used for monitoring food quality and safety (Kuntzler et al., 2020).

Although different studies have reported the application of both C-PC and *Spirulina* in the development of smart food packaging, the literature still needs to be more comprehensive regarding the use of these compounds. In this way, additional research in this area could address better strategies to improve the physical, structural, and mechanical properties of films and coatings since these are the main factors related to the challenges faced in their applicability and encourage the use of these natural substrates as a promising alternative to conventional plastics.

4. Biodegradability

Pollution caused by plastic materials for food packaging has become an increasingly severe problem over the years and thereby has received considerable attention in relation to environmental preservation. In this context, biodegradable packaging films have become a major trend in the packaging industry as they can replace traditional plastic packaging due to their degradability (Guo et al., 2022).

Biodegradable films have been developed through the combination



Fig. 3. Application of smart film in chicken quality monitoring. TVBN: Total Volatile Basic Nitrogen.

of different biopolymers and *Spirulina* (Balti et al., 2017; Cardoso et al., 2017; Ramji & Vishnuvarthanan, 2022; Zhang et al., 2020). Although some studies in the literature use biodegradable compounds, few investigate the biodegradability of films containing *Spirulina*. Ramji and Vishnuvarthanan (2022) evaluated the biodegradability of films based on chitosan/MMT K10/*Spirulina* by the soil burial method for 18 days. The results revealed that the biodegradability of the films measured in the form of mass loss increased steadily with increasing the days of burial in soil and the concentration of *Spirulina* used. In another work, Guru Moorthy et al. (2020) synthesized PVA films with *Spirulina* biomass and observed that the degradation of this composite increased as a function of the percentage of algae biomass added, evidencing that the biomass has a better efficiency when mixed with PVA than with corn starch.

5. Toxicity

Despite all the benefits *Spirulina* can provide, care must be taken concerning its origin and processing. In general, algae have a high capacity to accumulate different types of contaminants in their structure, which depends on environmental factors such as growing region, pH, light conditions, nutrient availability, and temperature (Sánchez-Rodríguez et al., 2001).

Due to the accelerated social, technological and industrial development, contaminations due to improper disposal of industrial effluents have become more frequent. These sources of contamination often contain highly toxic elements, e.g., arsenic (As), cadmium (Cd), mercury (Hg), lead (Pb) and antimony (Sb), which can be absorbed by algae through electrostatic or metabolic processes (Šesták et al., 1996). For this reason, specific legislation for their cultivation and quality control must be developed so that the final product arrives safely at the consumer's table.

Desideri et al. (2016) analyzed essential, trace, and toxic elements in 14 commercial samples of seaweeds, including *Spirulina*, and found amounts of As, Cd, and Pb of up to 67.6 mg/kg. In addition to the elements mentioned above, Al and Cu also appear as persistent contaminants in these algae (Mohy El Din, 2017; Rubio et al., 2021).

Another cause of severe contamination of *Spirulina* products can arise from its improper culture purity and the co-occurrence of potentially toxic cyanobacterial species such as *Microcystis aeruginosa* which can produce microcystins (Rzymski et al., 2015). Therefore, the final products made from *Spirulina* biomass can be potentially hazardous to human health when there is no solid quality control in their production and processing.

Nonetheless, the literature lacks studies about the toxicity of food packaging with the addition of *Spirulina*. As already mentioned, such toxicity is associated with the fact that its raw material can contain a series of emerging contaminants, e.g., heavy metals. In light of this, future works should take into account the source of its raw materials and their inherent toxicity.

6. Future trends for food packaging

Given the growing concern of society and researchers about the environmental impacts caused by conventional plastics, there has been an increasing search for new alternatives with minimal adverse effects to the environment to replace the use of these materials with natural and biodegradable ones that also increase the durability of foods and improve their quality.

Because of their broad applicability, low cost and biodegradability, biopolymers from natural sources are potential substitutes for non-degradable petroleum-based polymers (Ferreira et al., 2021). Their use in the food packaging area, especially in the preparation of active films, has been considered a field of intense development in recent years (Kumar et al., 2022). Their advantages include not only food protection as a result of the barrier formed against moisture loss, consequently delaying food deterioration, but also the prolongation of the shelf life of various food products due to the presence of active compounds in their composition, such as antioxidant and antimicrobial agents (Kumar et al., 2022; Rios et al., 2022).

Like other natural substances, *Spirulina platensis* extract (C-PC) has bioactive compounds that act as potential antioxidants. However, there are some limitations regarding its application precisely because it is sensitive to light, oxygen, heat, and humidity (Moreira et al., 2019).

The downstream processing of products from *Spirulina*, particularly C-PC, comprises several stages, including extraction and purification steps (Fratelli et al., 2021, 2022). In recent years, strategies have been developed to extract the bioactive compounds from *Spirulina* more sustainably, consuming less energy and avoiding the employment of toxic chemicals. Besides the challenges involved in these steps, maintaining the compounds in their stable form is also a largely reported issue (Giaconia et al., 2020).

The encapsulation method in micro and nanostructures can prevent the microalga degradation and increase its stability (Adjali et al., 2022) since it forms a barrier around the compound, thus providing protection against unfavorable conditions (Schmatz et al., 2020).

In this context, technological innovations such as nanotechnology emerge as an ally for the development of food packaging and are considered a trending topic for edible films. The use of nanomaterials, e. g., nanohydrogels, nanoparticles, nanofibers and nanoemulsions generally enhance the physical, mechanical and barrier properties of films, besides acting in delivery systems of pigments, enzymes and active compounds, being a good resource for obtaining new options of active and intelligent packaging (Trajkovska Petkoska et al., 2021) that help monitor the quality of the product and minimize its deterioration process (Rios et al., 2022). As a result, some studies have reported the use of *Spirulina* biomass and extract and evaluated its behavior and efficiency in food quality control as well as its active properties (Kuntzler et al., 2020; Moreira et al., 2018; Ramji & Vishnuvarthanan, 2022; Terra et al., 2021a, 2021b).

Although nanotechnology has several advantages, some challenges still need to be overcome. Problems related to the non-compatibility of the materials used to make the nanoparticles and their inefficient mixing can change their properties, increasing their fragility. In addition, there is some concern about the size of the nanoparticles. The smaller the particle, the greater the chances of interaction with human cells through the inhalation of these compounds, which can consequently cause respiratory and cardiac health problems, depending on the case. Therefore, further research is needed to minimize these adverse effects and help regulatory agencies oversee and regulate the dosage of compounds

(Javakumar et al., 2022).

Despite being an increasing and highly relevant area, there are still few studies in the literature that approach the use of *Spirulina* in the composition of food packaging and also as the main active compound for active and smart packaging. Given its potential in several sectors of the food industry, new studies should be conducted in order to analyze its interaction with other biopolymers and discover new findings that enable the improvement of the physicochemical, mechanical and optical properties of packaging materials so as to formulate quality films.

7. Conclusion

Several advancements in relation to food packaging technology have been made in the last years in multiple aspects. This includes the use of *Spirulina*, which has gained visibility in different western countries due to its composition and potential applicability in different areas of the food, pharmaceutical, and cosmetic industries. The incorporation of *Spirulina* biomass and high-value bioproducts such as C-PC into the formulation of food packaging has shown distinct and promising applications as colorimetric indicators and active packaging especially because of its active compounds, allowing real-time monitoring of food conditions and prolonging the shelf life of products, respectively.

Furthermore, the use of nanotechnology represents a strong trend in food packaging, given its advantages in terms of protection and preservation compared to traditional food packaging systems. Despite being a relevant area, few studies elucidate the use of *Spirulina* in isolation or in combination with other compounds, making further research in this area necessary, especially to encourage the use of this natural substrate as a promising alternative to conventional plastics.

The almost non-existent data about the toxicity of food packaging or even the solvents used for the solubilization of ingredients (polymers and salts) constitute a major literature gap. Therefore, it is highly desirable that new and practical packaging materials be explored and developed and that some studies be conducted to investigate their toxicity and biodegradability.

Declaration of competing interest

The authors declare that they have no conflicts of interest.

Data availability

No data was used for the research described in the article.

Acknowledgments

M.A. was supported by the Margarita Salas postdoctoral contract MGS/2021/21 (UP2021-021), financed by the European Union-Next Generation EU. The authors would also like to thank the São Paulo Research Foundation (FAPESP, grants nos. 2018/13408-1, 2020/06732-7, 2022/06293-9, and 2023/00857-0) and Higher Education Personnel Improvement Coordination (CAPES, grant no. 001) for funding this research.

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