Fabrication and Testing of Functional Food Packaging Materials

Project Report

Prepared by:

Verschuren Centre for Sustainability in Energy and the Environment

Project supported by:

Divert NS — Nothing Wasted

Introduction

One-third of the household wastes consist of food packaging materials. About 80% of which are single-use plastics, while only <11% of it gets recycled in Canada. Most of them end up in landfills and water bodies, polluting the ecosystem. Moreover, consumer awareness has created demand for sustainable and innovative food packaging solutions. Antimicrobial packaging (AP) is an innovative concept that can be defined as a mode of packaging in which the package can kill or inhibit the growth of microorganisms and thus extend the shelf life of perishable products and enhance the safety of the packaged products. There is myriad of studies on AP packaging; however, to date they have not been commercially viable due to multiple factors. Many of the antimicrobial agents used in AP packaging films are highly sensitive to high temperature and pressure during film production, which invariably results in significant structural and functional changes resulting in loss of the activity. For example, LLDPE film was co-extruded with grapefruit seed extract (GSE) at an extrusion temperature profile of 160-190 °C. The study reported that following extrusion, the GSE degraded and no antimicrobial activity was observed. Ramos, Jiménez, Peltzer, & Garrigós (2012) reported that only 25–44% weight of thymol and carvacrol were retained in PP film when subjected to temperature 190 °C for 18 min of hot press process. Panuwat Suppakul, Miltz, Sonneveld, & Bigger (2002) reported even higher loss of AM agent at about 96.7% weight after blown film extrusion process.

The Food and Agriculture Organization report shows that about one-third of all the food produced for human consumption is wasted at multiple stages in the supply chain. Although, Canada has a more highly advanced approach and technology for food waste redirection and treatment, it is still a major burden for the Canadian economy and it is estimated that wasted food costs the average Canadian household $1,100 per year. Furthermore, food waste prevention should be the first step in the waste management hierarchy, which has been challenging to achieve in developed and industrial nations. Microbial food spoilage contributes significantly to food waste generation, particularly in nutrient rich and perishable foods such as dairy and meat. The Verschuren Centre has embarked on a program with COPOL International Ltd. (COPOL) to develop packaging films that could potentially improve food safety and reduce food/packaging waste. COPOL manufactures Polypropylene (PP) films (mono- to multi-layers), for all applications including food packaging. COPOL is committed to continually investing in new technologies relevant to current market demands, while coping with evolving challenge to be environmentally responsible. The present project encompasses development of antimicrobial packaging with PP and chitin derivative(s), which is a thermally stable natural antimicrobial compound obtained from shellfish processing waste. Atlantic Canada occupies a major portion of shellfish landing, processing and export, resulting in large volumes of processing waste generation. Utilizing the shellfish processing waste further contributes to waste reduction, as this often ends up in landfill. It is anticipated that the PP-chitin packaging will be stable under the demanding plastic processing condition and retain its antimicrobial activity to inhibit the growth of microorganisms in food product and extend the shelf life of packaged food. AP packaging is in alignment with the "Food waste reduction" campaign (as mentioned in the website) by Divert NS, as the waste audit clearly points out that food makes up 11.5% of total materials going into Nova Scotia's landfills. The shelf life extension (of the packaged food) will also cut down the disposal of single-use plastic materials and extend their shelf life. As a result, there will be a significant reduction in the organics (from food waste) and plastics going into the landfill.

Proposed Solution- It was the goal of this project to create biopolymer infused polypropylene films that would provide equivalent or greater mechanical and barrier properties to existing polypropylene food films, but with enhanced antimicrobial properties, in order to extend shelf life and thereby reduce food wastage.

Experiments and Tests Performed

An extrudable biopolymer formulation (BPF) was identified and successfully developed by the VC in the early part of this project and used at varying weight percent incorporation in the polypropylene films to prepare BPF-PP films, in order to examine its impact on film structure and function. The goal being to add biopolymer within the film to retain or enhance film properties as it relates to use in food packaging. The prepared films were tested for mechanical and physicochemical properties as described below:

Wettability: The contact angle measurement was used to determine the wettability (hydrophobicity or hydrophilicity) of the films. This was measured using Drop Shape Analyzer (KRÜSS) by pendant drop method, where the contact angle between a drop of select liquid and the test surface is measured by the instrument software. In this study, contact angle at different points of the film surface was taken to determine the uniformity of the film as well.

Barrier Property: The gas barrier property or permeation test of the films was determined using the GAS-Transmission-Tester (Brugger), employing the pressure differential method. The test gas used in the present study was air. The films were cut to required size and adapters were used to fit the sample chamber.

Mechanical Properties: The BPF-PP films' mechanical properties including, tensile strength, elastic modulus and elongation at break, were tested using Universal Testing Machine (Shimadzu). Tensile strength is the measure of stress required to break the test material by stretching. Elongation at break, also known as facture strain, is the percentage increase in length before breaking. This indicates the capability of the test material to resist change in shape without crack formation/breaking. The elastic modulus is a measure of resistance of the test material/sample to elastic deformation.

Scanning Electron Microscopy: Visualization of the test films were carried out using SEM for observation of surface morphology and microstructure. The films were cut into discs of diameter ranging between 6 mm – 1 cm and used in the visualization under SEM at varying magnification ranging from 1 mm – to 2 µm. SEM images were taken at different locations of the same films.

Antimicrobial Test: The in-vitro antimicrobial efficacy of the packaging films prepared were tested against *Escherichia coli* using disc diffusion assay. The *E.coli* was sub-cultured on TSA plates for 24 hours at 37°C. The MHA plates were inoculated with *E.coli* at 0.5 McFarland standard number by streaking. The films were cut into discs of 1 cm diameter and placed on the inoculated MHA plates. The inoculated and treated plates were incubated at 37°C for 24 hours and the analyzed. Ampicilin discs of 10 µg were used as a positive control.

Film Packaging Test: Sample films produced with a range of inclusion of the BPF were sent to partners at NAIT (Northern Alberta Institute of technology) cheese lab for testing on cheese shelf life and packaging properties. Samples of two types of cheese were wrapped in specimen films and shelf life (up to 28days) determined at specific temperature and humidity by dissolution/homogentizing of cheese sample and plating to determine resultant microbial growth in samples. Additionally, samples were placed on film over inoculated media with E. Coli and Listeria in order to determine the bacteriostatic effect of the film on survival and growth of these gram negative and positive strains. To gain a comparative change in CFU for each film the net growth rates for each strain and time period were calculated.

Results

Wettability: The drop shape analysis was performed at different locations of the film. The contact angle measurement indicated that increasing incorporation of BPF does not significantly decrease the contact angle (indicating higher wettability). It was observed that doubling the incorporation of BPF (by weight percentage), decreased the average contact angle only by less than 0.5°. This is beneficial, as incremental incorporation of BPF will result in proportionate reduction in the quantity of synthetic polymers. However, the film thickness negatively affected the wettability of the films; i.e., thinner films (~40 μm) had lower contact angle indicating higher wettability compared to thicker films (~90 μm). Furthermore, films of ~40 μm thickness, lowest thickness among the films produced, resulted in non-uniform films, i.e., the thickness varied across the film surface. This could be attributed to the film thickness approaching the average particle size of the biopolymers used for incorporation. This resulted in melt voids in localized regions of the film caused by the biopolymer particles larger than the film thickness. Furthermore, thickness of the film also determines the exposure of the BPF on the surface, resulting in the negative relationship between the thickness and the wettability, as biopolymers are relatively more hygroscopic than PP. Generally, contact angles >90° is considered favourable as it indicates reduced interaction/less attraction with the test liquid. In this study, the BPF-PP films had contact angles in the range of 78 - 87°, and the contact angle increased as the film thickness increased, showing a directly proportional trend. This data is significant in contributing to ensuring films retain the appropriate moisture barrier properties with incorporation of biopolymer, particularly if the films produced comprise barrier layers, replacing non-recyclable components of plastics.

Barrier Property: Gas barrier property of the films prepared were tested at three different film thicknesses, ~40, ~60 and ~90 µm. The BPF-PP films had higher gas transmission rate and consequently higher permeability for air, compared to the control PP films (without BPF additive). This is contrary to the expectation and the literature reports, as biopolymers are known to reduce the gas permeability, thereby improving the barrier properties. While the results suggest otherwise, this could be primarily due to the melt voids causing localized regions of reduced thickness allowing for higher permeability, thereby skewing the overall performance. Although the average particle size of the biopolymers used in the film preparation were ~30 µm, few particles that are larger than or closer to the film thickness of 40 µm result in the lesions and voids. This was substantiated by the improvement in the barrier properties with increase in film thickness. Furthermore, the increase in weight percent incorporation of the BPF reduced the gas permeability compared to lower weight percent inclusion, but higher than the control PP films. The above showed promise that the BPF can improve the gas barrier properties, provided the film can be formed without melt voids and lesions, which can be achieved with smaller biopolymer particle size.

Mechanical Properties: The BPF-PP films' mechanical properties determined were the tensile strength, percent elongation at break and elastic modulus. The tensile strength of the control PP film was determined to be 27.6 MPa, comparable to the literature value. Inclusion of BPF into the films at lower weight percent did not reduce the tensile strength significantly (25.3 MPa). However, tripling the weight percent inclusion of BPF reduced the tensile strength. Although higher inclusion of BPF only reduced the tensile strength by <10 MPa, the elongation at break was drastically reduced at higher percent inclusion of BPF, suggesting these films can break relatively easier with less strain than films with lower BPF inclusion. This result indicates that inclusion of BPF at lower weight percent is possible without negatively impacting the mechanical properties, but inclusion beyond specific weight percent will result in significant reduction in specific mechanical properties, though this may also relate to film imperfections at the higher particle size used. Yet another challenge was the variation observed in the film with higher thickness, i.e., the standard deviation for films of thickness >60 µm, particularly for elongation at break values were significantly higher.

The elastic modulus of the control PP films and the BPF-PP films at different weight percent BPF inclusion were similar, indicating that the elasticity of the films are not impacted by the BPF incorporation.

SEM: The films were visualized under scanning electron microscope (SEM) to observe the surface morphology, distribution of the BPF in the films and the microstructure. Generally, SEM images showed that the BPF was evenly distributed throughout the film indicating that there was no phase separation and successful integration of BPF in PP films, as seen in Figure 1A and B. However, some melt voids and lesions were observed on certain regions of the BPF-PP films. The lesion can be observed in the Figure 1C in specific region of the BPF-PP film. This further resulted in non-uniform film thickness, which can also be seen in the Figure 1C. The lesions visualized through SEM, substantiates the results obtained from the mechanical and other properties test.

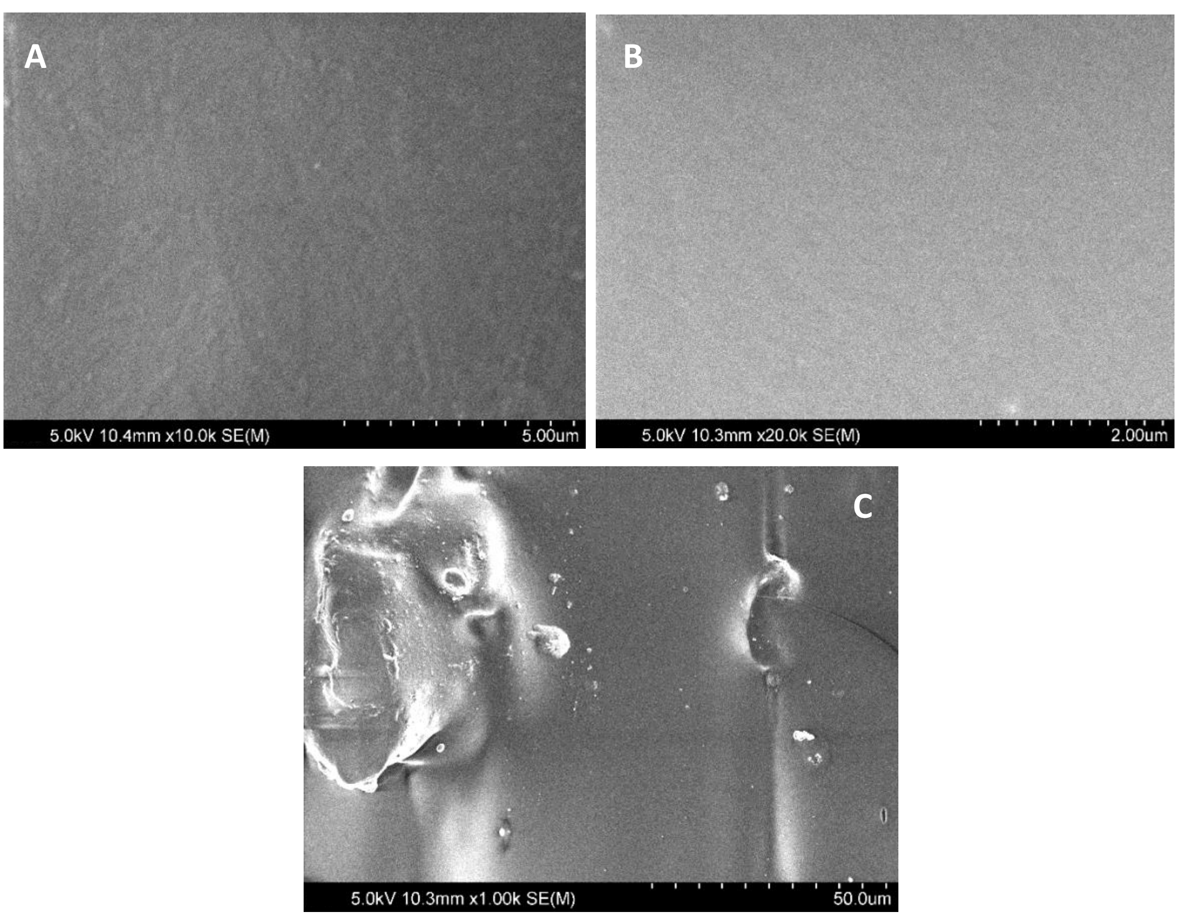


Figure 1: *SEM images of (A) Control PP films (B) BPF:PP films at lowest weight percent inclusion and (C) lesions observed in BPF:PP films at lowest weight percent inclusion*

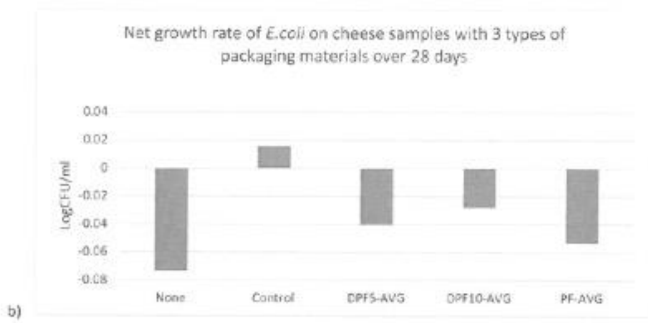
Antimicrobial Test

The preliminary in-vitro antimicrobial test was performed against *E.coli* to determine the efficacy of the BPF:PP films showed promise for on-contact prevention of microbial growth, however this is still not possibly to the degree we hope to achieve once higher inclusion thin film can be produced, as both the control PP films and BPF-PP films had on-contact reduction of microbial growth. This test was slightly challenging due to the mild coloration added by the inclusion of high levels of BPF compared to the control PP film, which are clearer. Quantitative analysis is essential to conclusively determine the antimicrobial property of BPF inclusion.

Data from NAIT cheese lab.

Growth rates for the two microbial strains on the cheese surface followed a similar and diminishing curve over the 28 day storage trial. The net growth rate evaluations for *E. coli* over 14 and 28 days show that the DPF5 and DPF10 packaging materials supported a negative growth rate over the first 14 days whereas the PF packaging material was similar to the control packaging. Findings on microbial growth with the various films suggest that the packaging materials with bioactive compounds present may be bacteriostatic as they may inhibit the growth or reproduction of bacteria (Figure 2 below, NAIT). Though the films reduced microbial growth rates, (Figure 3) they did not have a bactericidal effect on the E. coli and L. monocytogenes strains evaluated. In terms of their effectiveness against gram negative bacteria (i.e., E. coli) all three types of packaging materials (DPF5, DPF10 and PF) showed similar effectiveness. For gram positive bacteria (i.e., L. monocytogenes) the findings suggest that the PF type of packaging may be more effective.

Figure 2. Net growth rate reduction in packaged cheese with biopolymer addition in films.



Graphs 4: Growth rate of *L. monocytogenes* on cheese samples with different bioactive packaging materials.

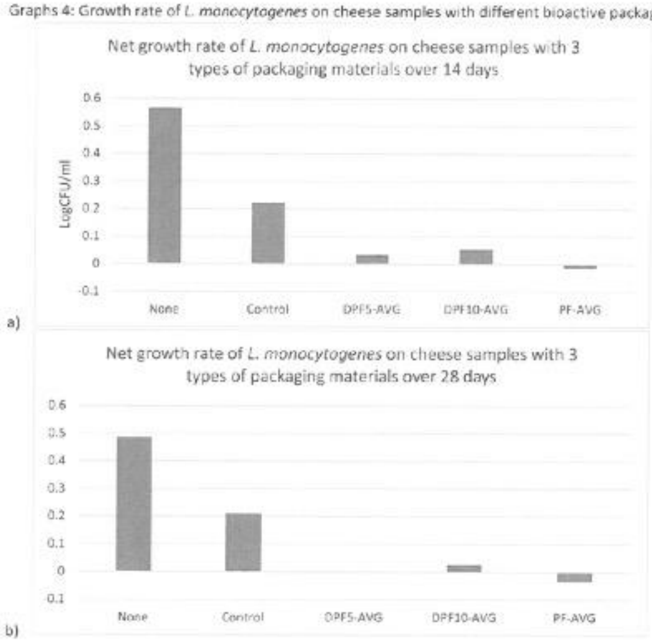
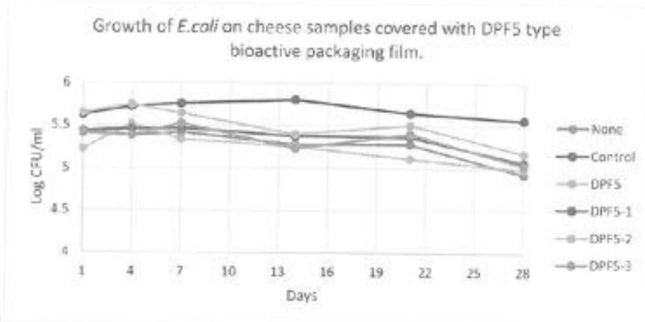
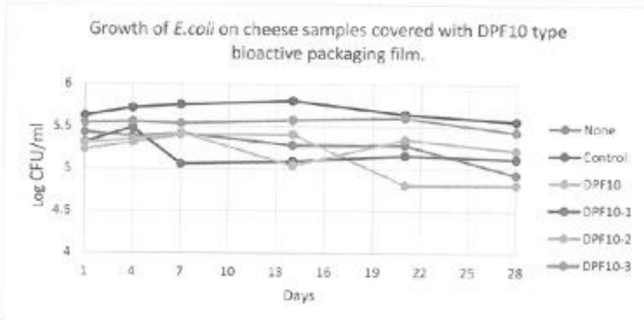


Figure 3. Reduction in microbial content of cheese stored at 4C in films with differing degree of biopolymer inclusion.



b)



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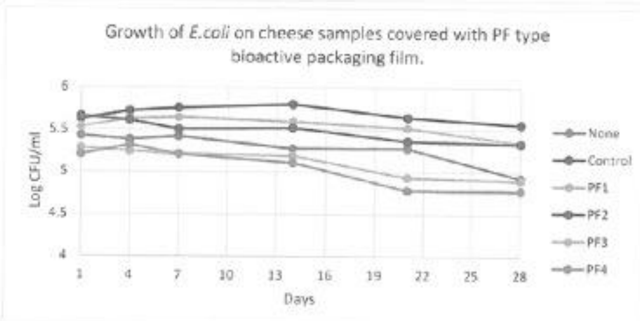

d)

Conclusive Remarks

The focus of the present project was to develop antimicrobial packaging films via extrusion, while simultaneously developing niche application for high-value compounds derived by biovalorization of marine by-products. The present study was successful in developing BPF-PP films without film failure or degradation. The mechanical properties at lower weight percent inclusion were not significantly impacted. Higher weight percent inclusion of the BPF, while having some positive impact such as reduced gas permeability, slightly negatively impacted mechanical properties. One of the major challenges encountered that caused this disruption was due to overlap of the particle size of the biopolymer and the film thickness, creating unevenness and potential particle voids in thin film. It is felt that this can be overcome with further optimization.

In terms of extending shelf life of cheese product, all treated films (BPF-PP) reduced the net growth rate of inoculated strains (E. Coli and Listeria) over a 28-day shelf life trial. Although these films had no direct bactericidal effect, the slightly improved film permeability (reduced gas barrier) did inhibit growth of microbes in storage conditions, thus showing good prospect for extending shelf life of packaged dairy products.

Future Directions

The outcomes for the present study are promising and show potential for incorporation of shell derived biopolymers in polypropylene films, though further optimization is required to achieve the ideal film properties for scaleup to commercial adaptation within our industry partners facility (COPOL). Particularly, to obtain uniform film thickness by achieving smaller particle size of the biopolymer, that is well within the desired film thickness, so as to prevent the melt voids and the resulting loss in mechanical properties. Moreover, the barrier properties are to be tested under different gases including oxygen and carbon dioxide to obtain further information on the barrier properties. The antimicrobial capacity to extend shelf life could be further elucidated at room temperature with different inclusion of a smaller particle size biopolymer providing for quantitative affirmation of the BPF-PP film efficacy in preventing and delaying the onset of microbial growth, and extending shelf life. Primarily, this study has also highlighted the need for developing key experimental viability approaches for testing and comparing antimicrobial efficacy as these had not been fully developed in the literature and film testing presents its own set of challenges when testing films/sheets or surfaces, such that a greater array of heat resistant biopolymers might be tested. The literature has a plethora of studies that evaluates compounds or specific formulations used to prepare films, though few in polypropylene, and there is very little to no information for methods of testing films or sheets. Hence the research team at VC is currently working to further develop a new approach suitable for testing packaging films for microbial inhibition capacity. Finally, expanding testing in real food systems is also in progress with NAIT and will provide indication of the new packaging material's efficacy as well as direct introduction of bioactive biopolymers in the cheese making process to extend the shelf life and ultimately reducing food waste.

On going discussions and feedback from the industry partner to strike a balance between the need for transparent/thin films and the particle size of the biopolymers has enabled the project to progress well and will be essential in the future in determining efforts to scale the BPF for testing at large scale extrusion. This study has enabled to establish proof-of-concept, develop prototype and test at bench scale. Moreover, the outcomes have highlighted specific aspects that needs to be further optimized at bench scale to ensure success. Not only will this create fully recyclable PP food films, but also achieve this through recovery of marine sourced biopolymer, creating a value stream and thereby reducing waste disposal in the seafood processing industry concurrently.

Acknowledgement of financial contribution

We gratefully acknowledge funds provided by Divert NS enabled the procurement of a film die/sheet former, which was essential in the preparation of the biopolymer films at the thickness and sheet dimensions relevant to our industry partners products and process.