Biodiesel Co-product Utilization: Glycerol-derived Films for use in Commercial Applications

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ABSTRACT

Glycerol is the co-product made when biodiesel, a renewable fuel made from fats and oils, is produced. As much as 10 % of the overall biodiesel product mixture is glycerol. In order to use more glycerol, it has been polymerized with other chemicals to form gels that can be converted to films when exposed to elevated temperatures in a furnace. The resultant films were studied for their ability to absorb solvents and the effects of the solvent on the physical, mechanical and elastic properties of the film were evaluated. Polymer films (bioplastics) made from glycerol can absorb various solvents in response to the size, shape and polarity of the solvent. These types of polymers are important to develop for use in biodegradable food and beverage containers, filters, drug delivery systems, soil conservation, water remediation, and timed-release applications.

Keywords: biodiesel, glycerol, polyesters, bioplastics, antimicrobial

I. INTRODUCTION

Developing new outlets for glycerol would have a significant impact on the economics of biodiesel production if value-added products made from glycerol can be identified. Glycerol is the major co-product produced from the process used to make biodiesel. Before the introduction of biodiesel-derived glycerol, the glycerol market was already saturated with uses in the food industry and in many pharmaceutical, chemical, and personal care applications. Therefore, increased production of biodiesel created a need to find new uses for glycerol. The renewable fuels market survives largely off of tax incentives and legislative mandates. However, adding value to the co-product streams of biofuel production will provide revenue for the industry. Production and marketing of such new high-value products would decrease cost of biodiesel production and provide new products to improve the environment and human health. For several years, our research team has studied the synthesis of gels and films made from glycerol and characterized their physical, thermal and mechanical properties using a variety of analytical tools [1].

Researchers at the United States Department of Agriculture (USDA) developed materials from the hydrolysis of starch-acrylonitrile co-polymers that absorbed greater than 400 times its weight in water and later became known as "Super Slurper" [2]. Today, we continue similar work by studying the absorption of various solvents into the matrix of poly(glutaric acid-glycerol) films [3, 4]. In our previous studies, various changes to the physical and chemical environment (i.e. temperature, solvent pH, solvent polarity index (SPI)) affected the absorption of solvents into the film. During these studies, we learned that the glycerol derived polymer films swell more in polar aprotic solvents like dimethyl sulfoxide (DMSO) and tetrahydrofuran (THF) than in polar protic solvents like methanol and water.

The absorbtion studies showed a clear correllation among polarity, molecular size and shape, and solvent absorption into poly(glutaric acid-glycerol) films. These films were shown to absorb up to 219.8% of their weight in solvent [5]. The absorption properties of the films also respond to external stimuli such as changes to pH, temperature, and polarity. Alternatively, these polymer films can be used in a variety of applications. Preliminary studies have also shown that these polymer films can be used in antimicrobial food packaging technologies or as conductive polymers.

However, challenges in their production and use still exist. Specifically, improving their mechanical properties (i.e. % elongation and tensile strength) and decreasing the underside tackiness to the side of the films that are not directly exposed to heat during production. This under-side tackiness is observed in a small percentage of film samples. Minimal assistance from industrial partners could overcome these small obstacles. Conversely, the adhesion properties of these polymers can also be optimized to form biobased tackifiers and adhesive tapes.

II. MATERIALS AND METHODS

Synthesis of Polymer Gels and Films

Polymer films were made by first synthesizing polyester gels from glycerol and glutaric acid at 135 ^{o}C as previously described [1] in a 2:1, 1:1, and 1:2 molar ratio of glutaric

acid and glycerol in a 250mL round bottom flask (RBF). Experiments were performed using an IKA stir plate equipped with a 250mL heating block and thermocouple. The mixture was reacted with stirring for 1h and then reacted further for 2h under vacuum. The polyester gels were characterized, without further purification, by NMR for degrees of branching and by GC and FTIR to quantify unreacted starting materials.

15g of the polyester gels consisting of a 2:1 molar ratio of glutaric acid:glycerol were transferred to individual aluminum weighing dishes and cured in a furnace at 150°C for 12 h. This film serves at the control. The polymers were characterized by ATR-IR and NMR (^{1}H and ^{13}C) as previously described.

Absorption and Erosion Measurements

A. Procedure and Calculations. The samples were bored from the aluminum pans into circles that were 1 inch in diameter. The samples were weighed (W_d) and immersed into 15 mL of solvent in capped, 2 oz jars. The samples were removed from the jars after 10 hours of incubation at room temperature, blotted with a Kimwipe (Kimberly-Clark, Marietta, GA), and weighed (W_s) .

Weight changes due to solvent absorption was determined by the following equation,

$$\% \text{ Weight Change} = (W_s - W_d) / W_d * 100 \quad (1)$$

where W_s and W_d represent the weight of the swelled and dry films, respectively.

B. Erosion. Erosion is defined as the physical depletion of a material, and is in contrast to degradation which involves chemical bond cleavage. Erosion of the films is calculated from the weight of the film before and after the film has been absorbed and desorbed twice and calculated by the following equation,

$$\% \text{ Erosion} = (W_d - W_{de}) / W_{de} * 100 \quad (2)$$

where W_{de} represents the weight of the desorbed films.

C. Polymer Resorption. To evaluate the ability to reuse these materials, the polymer films were allowed to desorb by gravity filtration and solvent evaporation in fume hoods until the polymer films returned to their original weight. If erosion was high, the film would break into pieces and the pieces would be recovered as efficiently as possible[4]. In such cases, the weight of the desorbed film could be less than the original weight of the film. The desorbed polymer films were then re-submerged into solvent and evaluated for solvent absorption as previously described using Equation 1. The amount of solvent resorbed was expressed relative to the weight of the eroded materials rather than the original weight of the film.

Nisin film preparation

Film were produced by mixing polyester gels with 10, 20, and 30 mg of nisin/mL of gel. The mixture transferred to individual aluminum weighing dishes and cured in a furnace at 150°C for 12 h. Film with no added nisin was used as the control.

Bacterial inhibition testing

L. monocytogenes Scott A 724, *E. coli* O157:H7 Oklahoma, and *S*. Enteritidis (ATCC 13076) were obtained from the culture collection of the U.S. Dept. of Agriculture, Agricultural Research Service, Eastern Regional Research Center. Prior to inoculum preparation, *E. coli* O157:H7 and *S*. Enteritidis were grown in tryptic soy broth (TSB: Remel Inc., Lenexa, Kans., U.S.A.), and *L. monocytogenes* was grown aerobically at 37 ◦C for 16 to 18 h in brain heart infusion broth (BHIB: Difco Laboratory, Detroit,Mich., U.S.A.).

Bacterial inhibition by antimicrobial films was evaluated using an agar diffusion method and a liquid incubation method as described by Appendini and Hotchkiss [6].

In the agar diffusion test, each film sample (1.4 × 1.4 cm) was placed on the surface of a BHI agar plate overlaid with the seeded semi-soft BHI agar (0.5% [w/v] agar). The seed density of overlay was approximately 106 CFU/mL of *L. monocytogenes*. The agar plateswere incubated at 37 ◦C for 24 h. Diameters of inhibition zone around film specimen were used to determine antimicrobial activity of each film sample.

For the liquid incubation test, 6 pieces of films (total surface area of approximate 24 cm2) were placed in a glass bottle with 50 mL liquid medium (BHIB, orange juice, or liquid egg white), creating a ratio of 2.08 mL of liquid/cm2 of exposed polymer surface. Orange juice and pasteurized liquid egg white (LEW) were purchased from a local store; these contained no preservatives as stated on their labels.

The orange juice was autoclaved (121 ◦C for 25 min) before use. The pH of liquid egg white was 8.46 and the pH of orange juice was 3.78. Themedium in the bottle was inoculated with 1mL of an overnight culture of selected strains and shaken at 24 or 4 ◦C at 150 rpm. The final cell populations were approximately 107 CFU/mL for orange juice and 104 CFU/mL for all other tests. One milliliter of the inoculated medium was sampled at each sampling time. Specimens were serially diluted with sterile phosphate buffer (Hardy Diagnostics, Santa Maria, Calif., U.S.A.), then pour plated onto BHI agar. Plates were incubated at 37 ◦C for 24 h. Inoculated medium without a film served as a control. The nisin diffusing from films to liquid media during incubation was equivalent to 200 IU/mL of liquid medium.

III. RESULTS AND DISCUSSION

Synthesis

Polymers made by the esterification of glycerol and glutaric acid were prepared by modifying our published synthetic protocol [3]. These polymers were soluble in polar organic

solvents but would not dissolve in water or nonpolar solvents. The resultant films made with a 2:1 (glutaric acid:glycerol) molar ratio, lost an average of 7.6 ±0.4% of their initial weight, presumably in the form of water as the by-product of esterification. The final products were clear, flexible, solid materials with a yellow hue. The pre-cured gels and cured films previously made under similar conditions have been extensively characterized by GC [7,1], TGA [1], FTIR [1] and NMR [8] (^{1}H and ^{13}C).

Absorption Studies

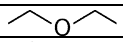
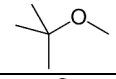
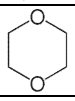
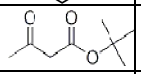
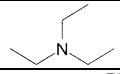
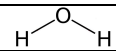
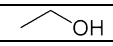
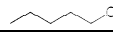
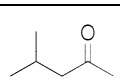
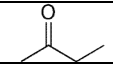
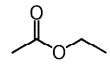
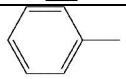
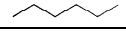
Previously, only seven polar and non-polar solvents were used to investigate absorption into poly(glutaric acid-glycerol) films[4]. In this study, the number of solvents was increased to twenty-five constituting a diverse group that would allow the investigation of solvents by pKa, SPI, functional group classes, and molecular size and structure. Previous studies suggested that when aprotic polar solvents have similar pKa's (±3 units), the solvent with the highest polarity index would absorb the most solvent [4]. The expanded group of solvents revealed that this correlation remained true when considering a very narrow pKa range (Table I). There was no apparent absorption trend that could uniformly characterize the entire group of solvents. However, there were subsets of data that were informative.

When the solvent polarity was ≤ 4.0, the absorption was less than or equal to 5.3 ±1.5%. There were no apparent similarities among these solvents to explain this observation as they vary randomly in molecular composition, molecular structure, pKa, dielectric constant, and protic and aprotic character. The only low absorbing solvent with a SPI > 4.0 is methyl isobutyl ketone (SPI = 4.2) which is absorbed into the film at 3.7 (+2.5)% of the film weight. However, there are high and low absorbers at SPI 4.2 and SPI 4.3 which indicates that the absorption into films with polarities above an SPI of 4.0 is intrinsically controlled by phenomena other than polarity.

When the solvents were divided into functional group types, it was clear that two factors influence solvent absorption: polarity and molecular size and shape. As the polarity of the solvents increases, the absorption into the films also increases. Consequently, an increase in polarity is accompanied by a decrease in chain length of the molecules. Therefore, it appears that the ability of molecules to enter into the matrix of the film could depend on a combination of the solvent polarity and size and shape of the solvent molecules. Methanol (SPI = 6.6) swells the polymer film by 26.5 ±2.1% of its original weight and is the highest absorbing mono-alcohol. The ability of alcohols to swell the polymer films decreases as the aliphatic chains grow. The films did not swell at all in n-pentanol (SPI = 3.6).

When comparing acetic acid to its methyl and ethyl ester derivatives (methyl and ethyl acetate, respectively), it is clear that solvent polarity and solvent absorption into the poly(glutaric acid-glycerol) film are directly related while molecular size and solvent absorption

Table I. Absorption of solvents into poly(glutaric acid-glycerol) films listed by increasing pKa of the solvent.

Solvent	Structure	pKa	SPI	Absorbance (%)
diethyl ether		-3.5	2.9	2.6 (±0.7)
methyl t-butyl ether		-2.0	2.5	0.0 (±0.0)
1,4-dioxane		2.1	4.8	163.8 (±0.3)
acetic acid		4.75	6.2	131.1 (±13.1)
pyridine		5.2	5.3	200.4 (±3.5)
t-butyl acetoacetate		10.0	n/a	1.7 (±0.4)
triethylamine		10.7	1.8	5.3 (±1.5)
ethylene glycol		14.2	6.9	72.6 (±6.5)
chloroform		15.5	4.4	168.9 (±12.9)
Water		15.8	9.0	4.7 (±1.2)
methanol		15.5	6.6	26.5 (±2.1)
ethanol		15.9	5.2	18.6 (±0.4)
isopropanol		16.5	4.3	10.0 (±2.0)
n-butanol		16.1	4.0	2.4 (±1.2)
n-pentanol		16.8	3.6	0.0 (±0.0)
Methyl isobutyl ketone		19.6	4.2	3.7 (±2.5)
acetone		19.7	5.4	77.7(±2.8)
2-butanone		20	4.7	71.0 (±7.3)
ethyl acetate		21	4.3	46.3 (±5.3)
methyl acetate		25	4.4	73.4 (±3.5)
acetonitrile		28.9	6.2	57.8 (±9.3)
dimethyl sulfoxide		35	6.5	186.0 (±11.4)
THF		38	4.2	106.4 (±9.0)
toluene		38	2.4	2.6 (±0.8)
Hexane		60	0.0	0.0 (±0.0)

are inversely related. As the carboxylic acid hydrogen is replaced with methyl (SPI = 4.4) and ethyl groups (SPI = 4.3), the absorption decreases with increased chain length and decreased polarity.

The trend continued with the ketone series. Acetone, the simplest ketone, was absorbed into the matrix of the polymer film and swelled it by 77.7 (±2.8)% of its original weight. The ketones became less polar by adding alkyl groups to acetone (SPI = 5.4) to form 2-butanone (SPI = 4.7) and methyl isobutyl ketone (SPI = 4.2).

The ether series contains both cyclic and aliphatic ethers (Table VI) and these solvents also swell poly(glutaric acid-glycerol) films in amounts directly proportional to their SPI values. The cyclic ethers, tetrahydrofuran (SPI = 4.2) and 1,4-dioxane (SPI = 4.8), swelled the polymers to 106.4 (±9.0) and 163.8 (±0.3)% of their weight, respectively. It is reasonable to assume that more 1,4-dioxane is absorbed by the films than tetrahydrofuran because of the additional oxygen atom available for hydrogen bonding. The cyclic ethers have higher SPI values, are more compact, and their oxygen atoms are more exposed and available for hydrogen bonding than their aliphatic counterparts.

None of the solvents from the alcohol or ketone groups were among the high absorbers and only 6 of the 25 solvents studied were high absorbers. From the molecules discussed so far, acetic acid and the cyclic esters (THF and 1,4-dioxane), were designated as three of the six high absorbers. The other three high absorbing solvents were chloroform, pyridine, and dimethyl sulfoxide (DMSO) which swelled the films by 168.9 (+12.9), 200.4 (+3.5)%, and 203.0 (+8.9)%, respectively. While these molecules are either small or heterocylic, there was no apparent connection among the solvents to explain their affinity to the matrix of the film.

Resorption and Erosion Studies

Resorption studies were performed on the polymer films that had been recovered after solvent desorption. Generally, complete solvent desorption was achieved within two hours of removing the film from the solvent. Erosion was low for all of the low absorbing solvents with SPI ≤ 4.0, with all of them except one (MTBE) showing no erosion at all [5].

Bacterial inhibition testing

Histograms (Figure I) were created to show the response of microbial growth in the presence of nisin-infused films. The data shows that the polyester films do not inherently inhibit microbial growth. However, at concentrations at or above 10 mg nisin/mL gel, microbial growth is slightly reduced after 24 hours and continues to decrease over the following 24 hours.

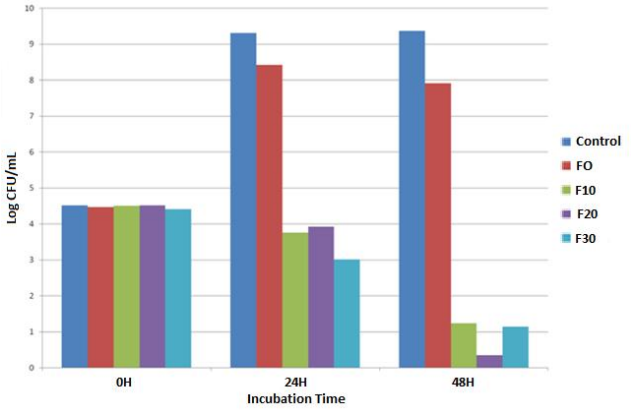


Figure 1. Effect of Films on Growth of L. innocua in TSB at 22°C.

IV. CONCLUSIONS

This research shows that there is a correlation among polarity, molecular size and shape, and solvent absorption into poly(glutaric acid-glycerol) films. These films were shown to absorb, on average, 0.0% to 219.8% solvent with less than 22% erosion after two-10 hour incubations. It also shows that the original hypothesis that suggested a correlation existed among pKa, polarity, and absorption, observed with the smaller data set, could not be used to reliably predict solvent absorption into poly(glutaric acid-glycerol) films. While the underlying reason for solvent absorption is still unclear, it was shown that among functional group classes that polarity and molecular size and shape can be used to accurately predict which solvents should be more readily absorbed into poly(glutaric acid-glycerol) films.

Microbial growth was retared around nisin-infused films at the lowest concentration studied (10mg nisin/mL gel).

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