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A New Approach to Environmentally Friendly Protein Plastics and Foams

María Cecilia Basso, a Antonio Pizzi, a,b, * and Luc Delmotte c

New formaldehyde-free and isocyanate-free bioplastics and biofoams were prepared by reacting ovoalbumin and dimethyl carbonate (DMC) at a moderate temperature. Analysis by $^{13}$C NMR revealed a reaction between dimethyl carbonate and the amino and/or hydroxyl groups of the side chain of amino acids. The densities were between 0.6 and 1.1 g/cm$^3$ for the obtained plastics. Thermal and mechanical resistances peaked at 175 °C and 7.7 MPa, respectively. The Brinell hardness was 2. The prepared foam exhibited a density of 0.1 g/cm$^3$ and an open cell structure. Impregnation with hexamethylene diamine (DAH) allowed for the preparation of materials with elastic mechanical behavior through the reaction of DAH and DMC. The new plastics and foams derived from ovoalbumin protein were markedly more environmentally friendly.

Keywords: Bioplastics; Biofoams; Protein; Dimethyl carbonate; Hexamethylene diamine; Aldehyde-free; Isocyanate-free; NMR analysis

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INTRODUCTION

Currently, there is a growing demand for natural products to be used in industrial applications because of environmental concerns and the depletion of non-renewable resources. In the framework of sustainable development, renewable biomaterials can provide an attractive alternative to petroleum-based products (Kaplan 1998; Gardziella et al. 2000; Frattini 2008; Raqueza et al. 2010; Ronda et al. 2013).

Among bio-based raw materials, proteins are an interesting alternative because of their ubiquity of supply, abundance in nature, and high functionality (Raqueza et al. 2010). Some technologies have been used to cross-link proteins effectively: for example, thermostables networks have been obtained by reaction of protein skeletal amido-groups, and also some amino acid amino groups of casein can react with formaldehyde or other aldehydes (Krische and Spitteler 1900; Ralston 2008). Cottonseed protein has been used to yield bioplastics by hot-press molding; in such a process methylene bridges are formed mainly by cross-linking with formaldehyde, whereas imine covalent bonds can be formed by reacting with glyoxal and glutaraldehyde (Yue et al. 2011; Yue et al. 2014).

This study describes a novel, simple, and non-conventional method for obtaining new bioplastics and biofoams from industrial egg white albumin (ovoalbumin) without any addition of either aldehydes or isocyanates. These products were prepared using dimethyl carbonate (DMC), which is an important carbonylating and methylating reagent used in various industrial fields, such as medicine, pesticides, composites, materials, flavoring agents, and electronic chemicals (Kongpanna et al. 2015). This compound
presents special environmental advantages (Tundo 2001; Thébault et al. 2015), such as the ability to cross-link at a moderate temperature and under normal atmospheric pressure. Usually, dimethyl carbonate has been used in the past to produce diverse materials by reaction with synthetic compounds and more recently with hydrolysable and condensed tannins (Thébault et al. 2014, 2015). However, proteins have not been used before as a raw material for such type of reaction.

In this context, the cross-linking reaction of ovoalbumin protein with DMC is a new reaction and approach for the development of environmentally friendly plastics and their derivatives. Several formulations are presented and the prepared biomaterials are characterized according to their main properties and chemical compositions.

EXPERIMENTAL

Materials

Albumin powder (chicken egg white), pentane, lead nitrate (Pb(NO₃)₂), and diamino hexane (DAH) were supplied by Acros Organics (Geel, Belgium). Dimethyl carbonate (DMC) and 1,5,7-triazabicyclo[4.4.0]dec-5-ene (TBD) were purchased from Sigma-Adrich (Steinheim, Germany). Glycerol was purchased from VWR International (Leuven, Belgium).

Plastics and Foam Preparation

The new biomaterials were prepared according to the mixture of components shown in Table 1 at 22 ± 2 °C.

**Table 1.** Composition of Bioplastics (A, D, E, and P) and Biofoam (F) Derived from Albumin and DMC

<table>
<thead>
<tr>
<th>Sample</th>
<th>A</th>
<th>D</th>
<th>E</th>
<th>P</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water (%)</td>
<td>47.4</td>
<td>47.4</td>
<td>35.9</td>
<td>35.9</td>
<td>33.5</td>
</tr>
<tr>
<td>Albumin (%)</td>
<td>24.5</td>
<td>24.5</td>
<td>42.6</td>
<td>42.5</td>
<td>39.5</td>
</tr>
<tr>
<td>DMC (%)</td>
<td>28.1</td>
<td>28.1</td>
<td>21.3</td>
<td>21.2</td>
<td>19.5</td>
</tr>
<tr>
<td>TBD (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.2</td>
</tr>
<tr>
<td>Pentane (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>7.5</td>
</tr>
<tr>
<td>Pb(NO₃)₂ (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.4</td>
</tr>
<tr>
<td>Apparent density (g/cm³)</td>
<td>0.6</td>
<td>1.1</td>
<td>1.0</td>
<td>1.1</td>
<td>0.1</td>
</tr>
<tr>
<td>Compression strength at 20% strain (MPa)</td>
<td>3.1</td>
<td>0.4</td>
<td>4.2</td>
<td>7.7</td>
<td>0.4</td>
</tr>
<tr>
<td>Brinell Hardness</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>2</td>
</tr>
<tr>
<td>Thermal resistance (°C)</td>
<td>110</td>
<td>175</td>
<td>175</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Albumin powder was mixed with water and mechanically stirred until homogeneity was reached. Samples E and P contained a catalyst, TBD and Pb(NO₃)₂ respectively, and for sample F a blowing agent (pentane) was incorporated. Finally, DMC was added and mixed, and the obtained samples were put in a ventilated oven preheated at 80 ± 2 °C for 12 h. Sample D was kept in the oven for 1 h and then it was impregnated.
with DAH (70% water solution) at 22 ± 2 °C for 24 h. Lastly, it was rinsed under water in order to remove excess DAH that has not been fixed.

**Plastics and Foam Characterization**

Cylindrical blocks of the prepared materials with the dimensions of 3 cm x 1.5 cm (D x L) were weighed to measure their bulk density. The cellular morphology of sample F biofoam was observed using scanning electron microscopy (SEM; TM3000, Hitachi, Japan). Thermal resistance was determined by progressive heating in a glycerine bath, and water resistance was measured by immersion of the samples in boiling water for 2 h. The mechanical resistance to compression was determined with an Instron 4467 Universal Testing Machine (USA) at a load rate of 2.0 mm min⁻¹. Brinell hardness tests were performed on the basis of NF B 51–126 (2007) using an Instron 4467 machine and a 10 mm diameter ball. No significant differences in the results were noted within the specimens after four repetition materials were tested for each case.

A bio-based plastic sample (sample A) was ground finely for nuclear magnetic resonance (NMR) analysis. The plastic powder was analyzed by solid state Cross Polarization-Magic Angle Spinning (CP-MAS) ¹³C NMR (Bruker, USA). Spectra were obtained using a Bruker AVANCE II 400 MHz spectrometer at a frequency of 100.6 MHz and at sample spin of 12 kHz. A recycling delay of 1 s depended on the ¹H spin lattice relaxation times (t₁), which were estimated with the inversion-recovery pulse sequence and a contact time of 1.0 ms. The number of transients were approximately 15,000, and the decoupling field was 78 kHz. Chemical shifts were determined relative to the reference, tetramethyl silane (TMS). The spectra were accurate to 1.0 ppm. The spectra were run with the suppression of spinning side bands. Furthermore, the solid product obtained from the reaction between 7.0 g of DAH (70% aq. solution) and 8.0 g of DMC (80 °C) was dried and then analyzed by CP-MAS ¹³C NMR.

**RESULTS AND DISCUSSION**

All of the mixtures presented in Table 1 yielded materials based on ovoalbumin and DMC, which is a non-toxic compound (Tundo 2001). The materials were completely aldehyde-free and isocyanate-free, therefore providing greater environmental acceptability over conventional plastics and foams, and even several other bio-based plastics and biofoams (Tondi and Pizzi 2009; Yue et al. 2011; Li et al. 2012a,b; Martinez de Yuso et al. 2014; Gama et al. 2015; Oliveira et al. 2015).

Comparative tests (not reported in Table 1) were conducted by preparing solid control samples without DMC. In this case, the materials obtained were very brittle and weak, showing the determinant role of DMC on cross-linking.

Formulations A, E, and P yielded rigid plastics. The macroscopic aspect of these samples varied greatly depending on the specific composition of the materials, as can be observed in Fig. 1. Moreover, their densities rose from 0.6 to 1.1 g/cm³ when the ovoalbumin proportion (% w/w) was increased (Table 1) and a catalyst was used. The thermal resistance property presented a similar trend: Sample A underwent degradation at 110 °C, while samples E and P just began to degrade at temperatures greater than 175 °C.
Figure 2 presents the compression strength curves for the prepared bio-based plastics. Samples P, A, and E exhibited the typically brittle thermosetting material behavior. The former, which includes Pb(NO$_3$)$_2$, exhibited a higher mechanical resistance (Table 1), which was consistent with a greater density. The value of Brinell hardness for this sample was 2.0, which is similar to polyethylene (Richardson and Lokensgard 2004). Pb(NO$_3$)$_2$ has been recognized as an efficient catalyst for the methoxycarbonylation reaction of aliphatic amines with dimethyl carbonate (Baba et al. 2002). However, other catalysts that are less dangerous for health and environment, such as AlCl$_3$, FeCl$_3$, or sodium acetate, could be used (García Deleon et al. 2002; Sun et al. 2010).

A rigid foam (sample F) can be obtained from the composition of sample E (Table 1) when a blowing agent, i.e., pentane, is added. Foaming occurs when the temperature exceeds the boiling point (36 °C), allowing for the expansion of the mixture simultaneously with the cross-linking reaction between the functional groups of ovoalbumin and DMC. This cellular material, namely sample F, was much lighter than the sample E plastic, exhibiting densities of 0.1 and 1 g/cm$^3$, respectively. The macroscopic aspect of both samples is shown comparatively in Fig. 3.

An SEM image of sample F (Fig. 4) revealed that this solid had an alveolar structure formed by broken and disordered cells. Thus, the sample F presented a mainly open porosity structure like other bio-based foams already reported (Basso et al. 2014; Lacoste et. al 2015 a,b). This property is highly suitable for several applications, including acoustic insulation or for floral and hydroponic foams (Landrock 1995; Lacoste et al. 2015b). The stress-strain curve obtained from the compression tests of this material (Fig. 5, curve F) indicate that the structure formed was fundamentally rigid. As for several tannin/furanic foams (Celzard et al. 2010), the stress-strain curve always showed three typical phases: linear elastic, collapse, and densification.
Fig. 3. Macroscopic examination of sample E (plastic) and sample F (foam)

Fig. 4. SEM image of sample F foam (X50 mag.)

Fig. 5. Stress-strain curves for the compression of sample F and sample D
Nonetheless, the high number of hydrophilic group of proteins would make it possible to forecast that adhesion to a wood surface would occur. However, the present data were not sufficient to be certain of it.

All of the prepared material samples (A, D, E, P, and F) were insoluble in boiling water. However, the materials appeared to experience some swelling during the boiling water test. Consequently, $^{13}$C NMR analysis was used to investigate this behavior. The CP-MAS $^{13}$C NMR spectrum of the dried reaction product of ovoalbumin with DMC (sample A; Fig. 6a) was compared with the spectrum of the albumin alone (Fig. 6b).

Fig. 6. A CP-MAS$^{13}$C NMR comparison of: a) albumin + DMC (sample A) and b) albumin.
The albumin + DMC spectrum (Fig. 6a) exhibited only one indication that DMC reacted with the albumin protein, namely shown by the peak at 157.47. This peak corresponds to the exact shift calculated for the DMC having eliminated a molecule of methanol to react with an amino group or a –OH group of the albumin protein. In the spectrum of albumin alone (Fig. 6b) there is a peak at 157.04. It is a small but significant difference. In albumin, the relative abundance of amino acids from greatest to least was: 1. leucine, 2. alanine + glutamic acid + aspartic acid, 3. lysine + phenylalanine, and 4. tyrosine + methionine. Thus, the shift at 157.47 ppm shows the reaction that occurred principally with the amino group of the side chain of lysine and with the –OH group (perhaps) of the side chain of glutamic acid. Furthermore, no reactions were observed with either the amide or C=O groups of the skeletal peptide chain. It is possible that cross-linkages were present, however, were not abundant, which would explain why the plastics exhibited swelling in hot water immersion.

In regards to the peaks in both the albumin and albumin + DMC spectra, these belonged to the albumin protein and are as follows: 173 ppm was assigned to C=O of peptide bonds and some –COOH of aspartic and glutamic acids; 60 ppm and 54 ppm were assigned to –N-C-C=O of the carbon connecting one peptide group to another peptide group (e.g., C5 of leucine); 40 ppm (C4 of leucine); 24 ppm (C3 of leucine); 20 ppm (C1 and C2 of leucine); 16 ppm (C-C of alanine).

Environmentally friendly protein plastics and foams can be obtained from the reaction between DMC and proteins containing a high proportion of amino acids having -NH2 or -OH groups, and perhaps –SH groups in their side chain. For example, the proteins from legumes and some vegetables, such as white beans, kidney beans, green peas, lentils, lima beans, soybeans, wing beans, peas (green), potatoes, cassavas, and yams present a high content of lysine, an amino acid with a –NH2 group on its side chain. (Young and Pellet 1994).

Impregnation of sample A with a diamine (hexamethylene diamine) (DAH) solution before total hardening lead to obtain a plastic showing an elastic mechanical behavior (sample D, Fig. 5). This was a result of the reaction between DAH and DMC having already partially reacted with albumin.

The quantitative MAS 13C NMR spectrum of the dried reaction product of DAH with DMC is shown in Fig. 7. The peaks at 156 to 158 ppm correspond to the C=O stretching of DMC and the peak at 52 ppm correspond to the –O-CH3 stretching of DMC. The other peaks represent the amine groups. The peaks at 43 and 41 ppm, 32 ppm, and 26 ppm correspond to the C1 and C6, the C2 and C5, and the C3 and C4 of the di-amine, respectively. The most interesting peak was at 40 ppm, which belonged to the C1 and C6 of the di-amine. This represents where the DMC reacted with the amino group of the amine, thus the C of the amine vicinal to the N of the amine reacted with DMC to form the following link type:
Fig. 7. Quantitative MAS $^{13}$C NMR of the reaction between dimethyl carbonate and hexamethylene diamine

The development of a network by the generation of urethane bridges allows for a better distribution of stress, thus improving the elastic properties of the materials. The intrinsic length of the urethane bridges lengthens the chains, rendering greater flexibility and elasticity to the network formed (Fig. 5, curve D). The same reaction between DMC having partially reacted with tannins and DAH has been described previously by Thébault et al. (2014, 2015). Even though the plasticizing effect is mainly due to the formation of urethane bridges, some residual water can also contribute to plasticisation with the temporary role of an external plasticizer.

CONCLUSIONS

1. Novel, rigid plastics of high mechanical and thermal resistance were developed from ovoalbumin and DMC.
2. $^{13}$C NMR analysis revealed that the reaction occurred mainly between DMC and the amino group of the side chains of the lysine and the –OH group of the side chain of glutamic acid and not with the peptide skeletal groups of the protein.

3. Flexible plastics were prepared by impregnation and reaction with DAH.

4. Expanded materials, *i.e.*, foams, can be obtained by including a blowing agent (*e.g*., pentane) in their composition.

5. Novel plastics and foams that are derived from proteins, not composed of aldehydes or isocyanates, yield materials that are markedly more environmentally friendly.

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**REFERENCES CITED**


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