

# Plasticization of Poly-L-lactide for Tissue Engineering

Matthieu Cuénoud, Pierre-Etienne Bourban, Christopher J. G. Plummer, Jan-Anders E. Månson

Laboratoire de Technologie des Composites et Polymères (LTC), Ecole Polytechnique Fédérale de Lausanne (EPFL), Station 12, Lausanne CH-1015, Switzerland

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**ABSTRACT:** Plasticization of medical grade poly-L-lactide (PLLA) by addition of polyethylene glycol (PEG) with various molar masses has been evaluated as means of producing low stiffness matrices for bioresorbable scaffolds for soft-tissue engineering applications. As reported previously, the  $T_g$  of injection molded specimens of the PLLA/PEG blends decreased strongly with PEG content, so that at PEG contents of 15 and 25 wt % it became significantly lower than normal human body temperature, implying an essentially rubber-like mechanical response *in vivo*. The degree of crystallinity of the moldings also increased strongly with PEG content, reaching a maximum of about 60 wt % at 25 wt % PEG. Moreover, after the immersion in phosphate-buffered saline for 5 days in 37°C to simulate

conditions *in vivo*, the moldings with the highest PEG contents showed increased water uptake and, for relatively low molar mass PEG, significant mass loss, associated with phase separation and leaching of the PEG. Blends with relatively low PEG contents also showed large increases in their degree of crystallinity. The implications of these changes for the *in vivo* performance of the blends and their potential for development as matrices for bioresorbable scaffolds are discussed in the light of results from a series of PLLA/PEG copolymers. © 2011 Wiley Periodicals, Inc. *J Appl Polym Sci* 121: 2078–2088, 2011

**Key words:** polylactide; polyethylene glycol; plasticization; ageing; biomaterials

## INTRODUCTION

Synthetic biodegradable polyesters are widely used in tissue engineering as a matrix for bioresorbable scaffolds and templates for tissue regeneration.<sup>1</sup> The use of bioresorbable scaffolds limits the number of required surgical operations and the scaffold degrades over time without leaving foreign substances in the body, thus reducing the risk of inflammation.<sup>2</sup> Synthetic thermoplastic polylactides (PLA) are considered to be among the most promising biodegradable polyesters owing to their mechanical property profile, ease of processing, biocompatibility, and biodegradability.<sup>3</sup> Certain PLA grades are FDA approved and have been used in surgery as suture materials and for load bearing applications such as orthopedic fixation devices.<sup>4</sup>

PLA degrades by hydrolysis of the ester bond without the need for enzyme catalysis, but degradation rates are generally low and it also has the disadvantage of a relatively low ductility.<sup>5</sup> PLA has therefore been modified by manipulation of its stereochemistry and processing conditions, copoly-

merization and blending, with the aim of tailoring its mechanical properties.<sup>5,6</sup> The most widely used approach to improving the ductility of PLA is to blend it with plasticizers such as poly(ethylene glycol) (PEG),<sup>5</sup> which is an FDA approved water-soluble semicrystalline thermoplastic.<sup>7,8</sup> Blends of PEG and PLA with high and low degrees of stereoregularity are reported to be miscible for PEG contents of up to 20 and 30 wt %, respectively,<sup>9,10</sup> and Hu et al.<sup>9</sup> observed a decrease in room temperature tensile modulus by two orders of magnitude on addition of 30 wt % PEG to PLA, for example. However, even under ambient conditions, such blends may show a tendency to phase separate over time,<sup>9</sup> which raises the question of their stability *in vivo* applications, i.e., in an aqueous environment at a temperature of approximately 37°C.

Porous biocomposite scaffolds based on PLA have already been shown to be promising for bone tissue engineering, based on investigations of their mechanical properties<sup>11</sup> and *in vivo* response.<sup>12</sup> Porous neat PLA scaffolds show a compressive elastic modulus of between 180 and 10 MPa in the porosity range 78–92%, and mechanical properties comparable to those of native bone may be obtained in the presence of fillers<sup>11</sup> and biodegradable phosphate glass fibers.<sup>13</sup> However, it is also of interest to produce scaffolds with a reduced elastic modulus, to replace softer tissues such as cartilage. The elastic modulus of a foam is generally expected to

Correspondence to: J.-A. E. Månson (jan-anders.manson@epfl.ch).

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be linearly proportional to the elastic modulus of the cell walls at constant porosity.<sup>14</sup> The present preliminary study has therefore been aimed at assessing the extent to which plasticization of PLA by PEG is suitable for this purpose, and hence whether the mechanical properties of cartilage may potentially be reproduced in porous PLA/PEG scaffolds *in vivo*. Polymer blending was the method of choice because the varying the blend composition provides a straightforward means of tailoring the mechanical response, which is of particular interest for the development of scaffolds with property gradients, suitable for attachment to a rigid (bone) substrate, for example. The same FDA approved poly-L-lactide (PLLA) was used as in the previous studies of bone replacement scaffolds referred to above, combined with PEG with different molar masses because the degree of plasticization is known to depend on the molar mass of the plasticizer.<sup>15</sup> The molar mass of the PEG was nevertheless limited to below 40,000 g/mol to avoid eventual problems with PEG excretion from the body.<sup>8</sup> Specimens of the various blends were prepared by melt extrusion, followed by injection molding, along with specimens of pure PLLA and a series of commercial PLLA-PEG copolymers. To evaluate their mechanical response to implantation in the human body, the properties of the as-injected specimens were then compared with those measured after immersion in phosphate-buffered saline (PBS) at 37°C for 1 week, which is the time required for the water content to reach saturation under these conditions.<sup>16</sup>

## MATERIALS AND METHODS

### Materials

Neat bioresorbable PLLA (PLLA1) with an estimated molar mass of 146,000 g/mol<sup>6</sup> and two triblock copolymers containing 4 wt % PEG, PLLA-PEG-PLLA (Copo1) and PLLA-co-poly-D-lactide (PDLA)-PEG-PLLA-co-PDLA (Copo2) with intrinsic viscosity of 1.6, 2.1, and 1.8 dL/g, respectively, were provided by Boehringer Ingelheim Germany in the form of pellets. Three grades of poly(ethylene glycol) (PEG), in the form of flakes, were added to the PLLA: PEG1500 (weight average molar mass  $M_w = 1500$  g/mol, Fluka), PEG6000 ( $M_w = 6000$  g/mol, Aldrich), and PEG35000 ( $M_w = 35,000$  g/mol, Aldrich). The PLA-based polymers and the PEG were dried overnight under vacuum at 75 and 35°C respectively, to limit degradation by hydrolysis during processing.

### Processing

PLLA1, Copo1, and Copo2 were melt-extruded using a micro-compounder with twin conical coro-

tating screws and a capacity of 5 cm<sup>3</sup> (Micro 5 Compounder, DSM, Netherlands). Compounding was carried out under nitrogen flow with a screw rotation speed of 125 rpm, a temperature of 200°C, and a residence time of 3 min. The polymer melt was then injected at 8 bars into a mold maintained at a temperature of 40°C to produce dumbbell-shaped tensile test specimens following the ASTM D638 protocol (thickness: 2 mm, gauge width: 4 mm, and gauge length: 25 mm). PLLA1/PEG blends with PEG contents of 5, 15, and 25 wt % were processed under the same conditions. All the materials were stored dry in a freezer at -30°C to avoid excessive ageing. The specimens referred to as "dry" were transferred to a desiccator at room temperature prior to testing. The specimens referred to as "wet" were immersed in a phosphate buffered saline solution (PBS) for 5 days at 37°C. After removal from the PBS and cooling to room temperature, the surfaces of the specimens were wiped with a towel prior to testing to remove any excess water. The PLLA was assumed not to be severely degraded after this treatment, following Grizzi et al.,<sup>17</sup> who have shown that 2 mm thick plates of PLA undergo insignificant degradation after 5 weeks in PBS at 37°C.

### Tensile tests

Mechanical tests were performed at ambient temperature using a tensile test machine (UTS, Test Systeme, Germany) with a cross-head speed of 3 mm/min. The strain was measured with an extensometer over the full gauge length of 25 mm. The tensile modulus,  $E$ , was determined according to ASTM D638 from the initial linear elastic region of the stress-strain curve in the strain range  $\epsilon_1 = 0.0005$  and  $\epsilon_2 = 0.0025$ . The yield stress,  $\sigma_y$ , and the yield strain,  $\epsilon_y$ , defined as the point at which the stress reached its maximum value, and the elongation at break,  $\epsilon_{max}$ , were also determined. Three to five specimens of each material were tested.

### Differential scanning calorimetry

Samples of approximately 10 mg in mass were cut from the ends of the tensile test specimens. The glass transition temperature,  $T_g$ , crystallization temperature,  $T_c$ , and melting temperature,  $T_m$ , were determined by differential scanning calorimetry (DSC Q100, TA instrument, United States), to a precision of about  $\pm 2^\circ\text{C}$ .  $T_g$  was taken to correspond to the mid-point of the associated step-like increase in heat flow rate observed during temperature scans. The scanning conditions were -60 to 200°C (first heating), 200 to -60°C (first cooling), and -60 to 200°C (second heating) at a rate of 10 K/min under nitrogen, with an isothermal step of 1 min between the

scans.  $X_c$ , the degree of crystallinity of the PLLA was then calculated from

$$100(\Delta H_m - \Delta H_c)/[\Delta H_{100\%}(1 - W_{\text{PEG}})],$$

where  $\Delta H_m$  is the measured enthalpy of fusion corresponding to the PLLA melting peak,  $\Delta H_c$  is the measured enthalpy of crystallization,  $\Delta H_{100\%}$  is the enthalpy of crystalline PLLA, taken to be 91 J/g,<sup>18</sup> and  $W_{\text{PEG}}$  is the weight fraction of PEG.  $\Delta H_{100\%}$  for PEG was taken to be 197 J/g.<sup>19</sup>

### Thermogravimetric analysis

Thermogravimetric analysis (TGA) was carried out on a TGA/SDTA 851e (Mettler-Toledo, Switzerland) using 30 mg samples cut from the dry and wet specimens. The specimens were heated under air-flow of 30 mL/min from 30 to 200°C at a rate of 5°C/min. The weight changes were considered to be due uniquely to water release from the specimens given that the degradation temperatures of both PEG and PLLA are over 200°C under these conditions. The water uptake was defined as

$$100(W_{20^\circ\text{C}} - W_{190^\circ\text{C}})/W_{20^\circ\text{C}},$$

where  $W_{20^\circ\text{C}}$  and  $W_{190^\circ\text{C}}$  are weights measured at 20°C and 190°C, respectively. Each specimen was also weighed using a balance (XS204 DeltaRange, Mettler Toledo, Switzerland) before and after 5 days in PBS, i.e. in the dry and wet states. The percentage weight change was in this case defined as

$$100(W_{\text{Dry}} - W_{\text{Wet}})/W_{\text{Dry}}.$$

## RESULTS AND DISCUSSION

In the dry state, all the moldings were observed to be transparent. The surfaces of PLLA/25 wt % PEG were somewhat greasy, particularly in the case of PEG1500, which suggested that some segregation of PEG had taken place. In the wet state, PLLA, Copo1, Copo2, PLLA/15 wt % PEG1500, PLLA/15 wt % PEG6000, and all the blends containing 5 wt % PEG remained optically transparent. However, PLLA/15 wt % PEG35000, PLLA/25 wt % PEG1500, and PLLA/25 wt % PEG6000 became opaque, with pronounced blistering at their surfaces, and PLLA/25 wt % PEG35000 became white and highly swollen, consistent with previous work.<sup>20</sup> The surfaces of PLLA/25 wt % PEG were no longer greasy after immersion in PBS.

### Water absorption

Figure 1 shows the results from thermogravimetry for the water uptake of the moldings. In the dry state, the

water content remained well below 1% in each case, ranging from 0.13 wt % for the neat PLLA to 0.87 wt % for PLLA/15 wt % PEG35000. After 5 days in PBS, however, the water uptake strongly increased at high PEG contents, reaching about 17.9 wt % for PLLA/25 wt % PEG35000 [Fig. 1(c)]. PLLA/15 wt % PEG35000 and PLLA/25 wt % PEG35000 also absorbed significantly more water than the corresponding PLLA/PEG1500 and PLLA/PEG6000 blends. Finally, the water uptakes of Copo1 and Copo2 given in Table I were comparable with that of PLLA/5 wt % PEG (which varied little with the molar mass of the PEG), consistent with the correlation between water uptake and PEG content in the blends.

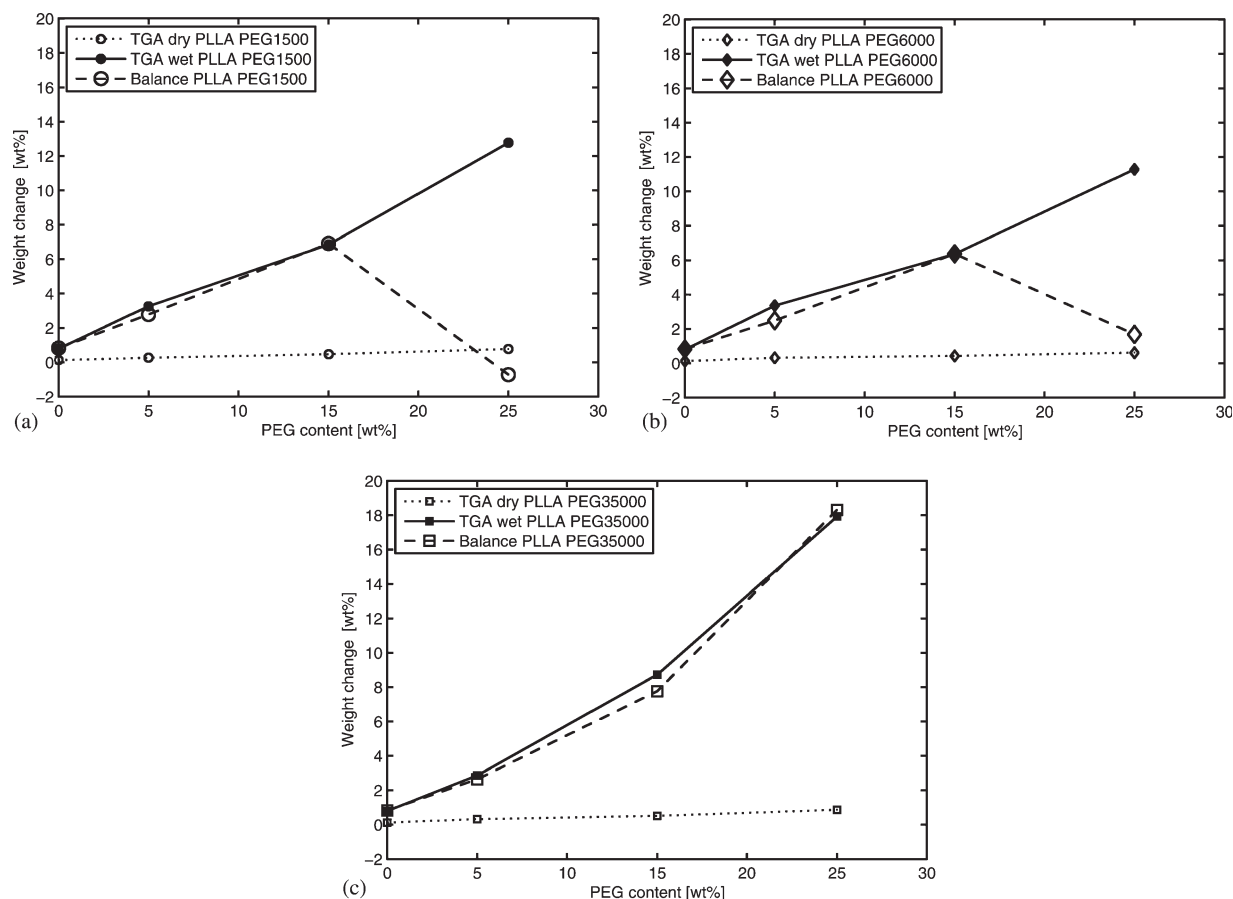
One might expect thermogravimetric measurements of the water content and direct measurements of the weight change after immersion in water (wet) to give similar results in the absence of any degradation or dissolution of the PLLA and PEG. This was the case for PLLA1, Copo1, Copo2, PLLA/5 wt % PEG, and PLLA/15 wt % PEG, as may be seen from Figure 1. On the other hand, for PLLA/25 wt % PEG1500 and PLLA/25 wt % PEG6000, weight changes of -0.71 and 1.69 wt % respectively, were observed, whereas thermogravimetry indicated water contents of 12.77 and 11.29 wt %. These differences were interpreted as corresponding to leaching of part of the PEG during immersion in PBS, as reported elsewhere,<sup>20</sup> which was presumably enhanced in the present case by the increased molecular mobility of the lower molar mass PEG. The opacity of the specimens containing high PEG contents in the wet state suggested that mass loss was preceded by phase separation in the presence of the PBS.

### Thermal properties

#### Dry state

Figure 2 shows DSC traces of the first heating scans from PLLA1, Copo1, and Copo2 in the dry state. PLLA1 showed a  $T_g$  of 58°C, a cold-crystallization peak at 96°C,  $T_m$  of 182°C, and an initial degree of crystallinity of 5.3%. Copo1 showed a  $T_g$  of 55°C, a less marked cold-crystallization peak than PLLA,  $T_m$  of 178°C and an initial crystallinity of 9.7%, whereas Copo2 showed a  $T_g$  of 49°C and was fully amorphous. A small exotherm in the temperature range immediately below the main melting peaks of PLLA1 and Copo1 indicated a transformation from the less stable  $\alpha'$  crystalline phase of PLLA to the  $\alpha$  phase during the scans.<sup>21</sup> In specimens cooled from the melt at 10 K/min,  $T_g$  was -45, -42, -47°C;  $T_m$  was 49, 64, and 68°C; and the degree of crystallinity was 83.9, 89.3, and 80.9% for PEG1500, PEG6000, and PEG35000, respectively.

The blends showed transition temperatures intermediate between those of the PEG and the neat



**Figure 1** TGA measurements of the water uptake for specimens in the dry and wet states, along with the overall weight differences between the dry and the wet states as a function of PEG content.

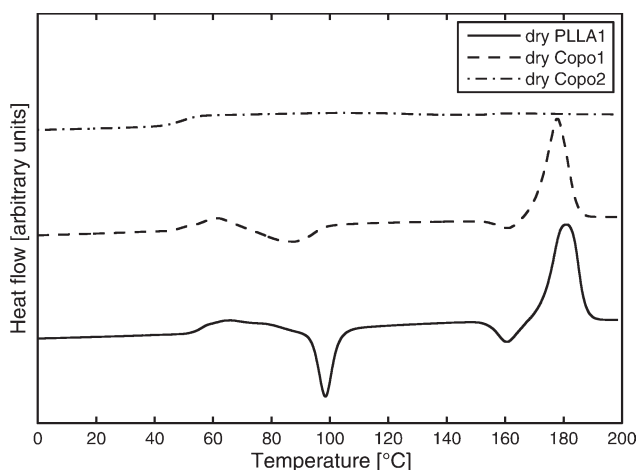
PLLA in the dry state. Figure 3 shows DSC traces of the first heating scans from PLLA/PEG6000. Regardless of their molar mass, the blends containing 5 and 15 wt % of PEG showed a single  $T_g$  suggesting complete miscibility, and  $T_g$  decreased with PEG content, falling below normal body temperature at a PEG content of 15 wt % as shown in Figure 4, consistent with previous work.<sup>9,15,22-24</sup> An attempt was made to fit the  $T_g$  data with the Fox equation<sup>9</sup>:

$$\frac{1}{T_g} = \frac{w_1}{T_{g1}} + \frac{w_2}{T_{g2}} \quad (1)$$

**TABLE I**  
Results from Thermogravimetric and Thermal Measurements on Copo1 and Copo2

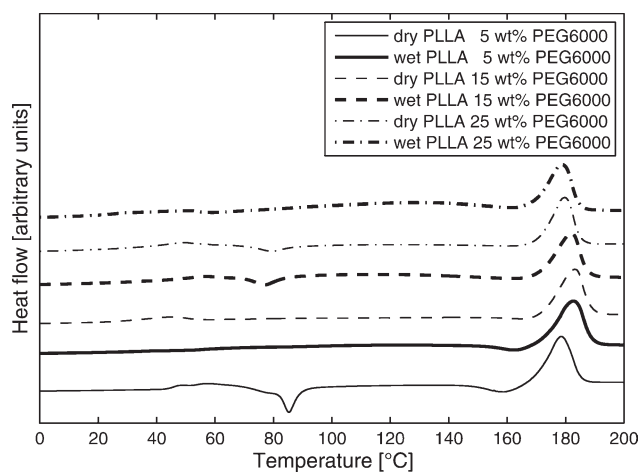
	Weight change TGA (wt %)		Weight change balance (wt %)	Degree of crystallinity (%)	
	Dry	Wet	Wet	Dry	Wet
Copo1	0.16	1.94	1.6	10.9	11.5
Copo2	0.22	3.5	2	—	—

where  $w$  is the weight fraction and the subscripts 1 and 2 refer to the blend constituents. Predictions from Eq. (1), taking  $T_g$  for PLLA to be 58°C and varying  $T_g$  for PEG in the range -40 to -66 are also shown in Figure 4. Although Eq. (1) was broadly consistent with the  $T_g$  of PLLA/15 wt % PEG, it



**Figure 2** DSC first heating thermograms of PLLA1, Copo1, and Copo2 in the dry state.



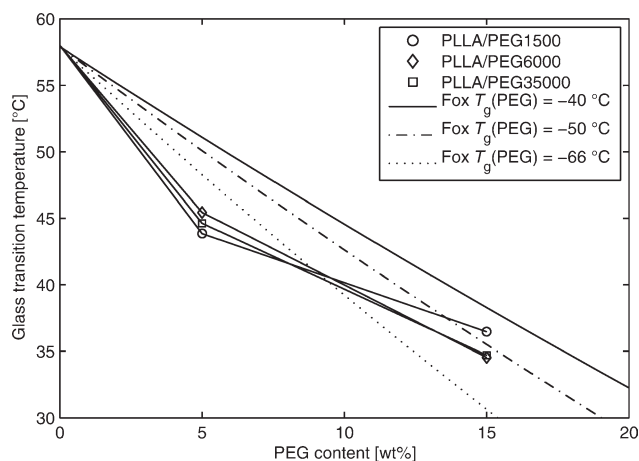


**Figure 3** DSC first heating thermograms of PLLA/PEG6000 with different PEG contents in the dry state.

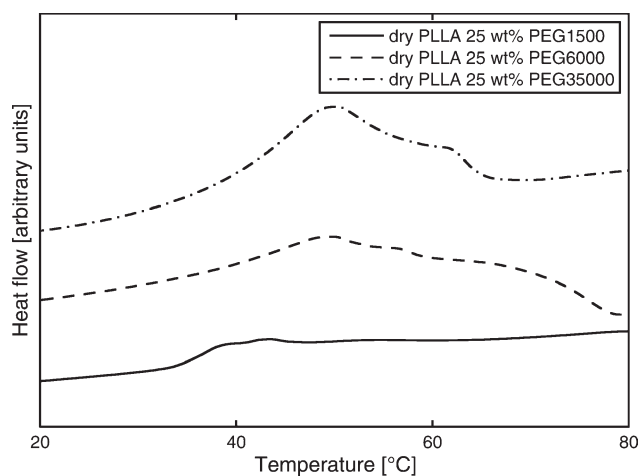
overestimated that of PLLA/5 wt % PEG regardless of the molar mass of the PEG.

Figure 5 shows thermograms from the first heating scans of the blends containing 25 wt % PEG. PLLA/25 wt % PEG1500 showed a  $T_g$  of 36°C, i.e., much greater than one would expect on the basis of Figure 4, followed by a small endothermic peak which was associated with melting of the PEG. In the blends containing 25% wt % PEG6000 and PEG35000, however, the  $T_g$  were less clearly defined owing to overlap of the PLLA glass transition and the PEG melting peaks centered at 48 and 50°C, respectively. The areas under the PEG melting peaks also increased with molar mass. This confirmed these blends to contain two distinct phases and also indicated the extent of phase separation to increase with the molar mass of the PEG.<sup>9,15</sup>

No clear dependence of the PLLA melting behavior on the PEG content of the blends and the molar



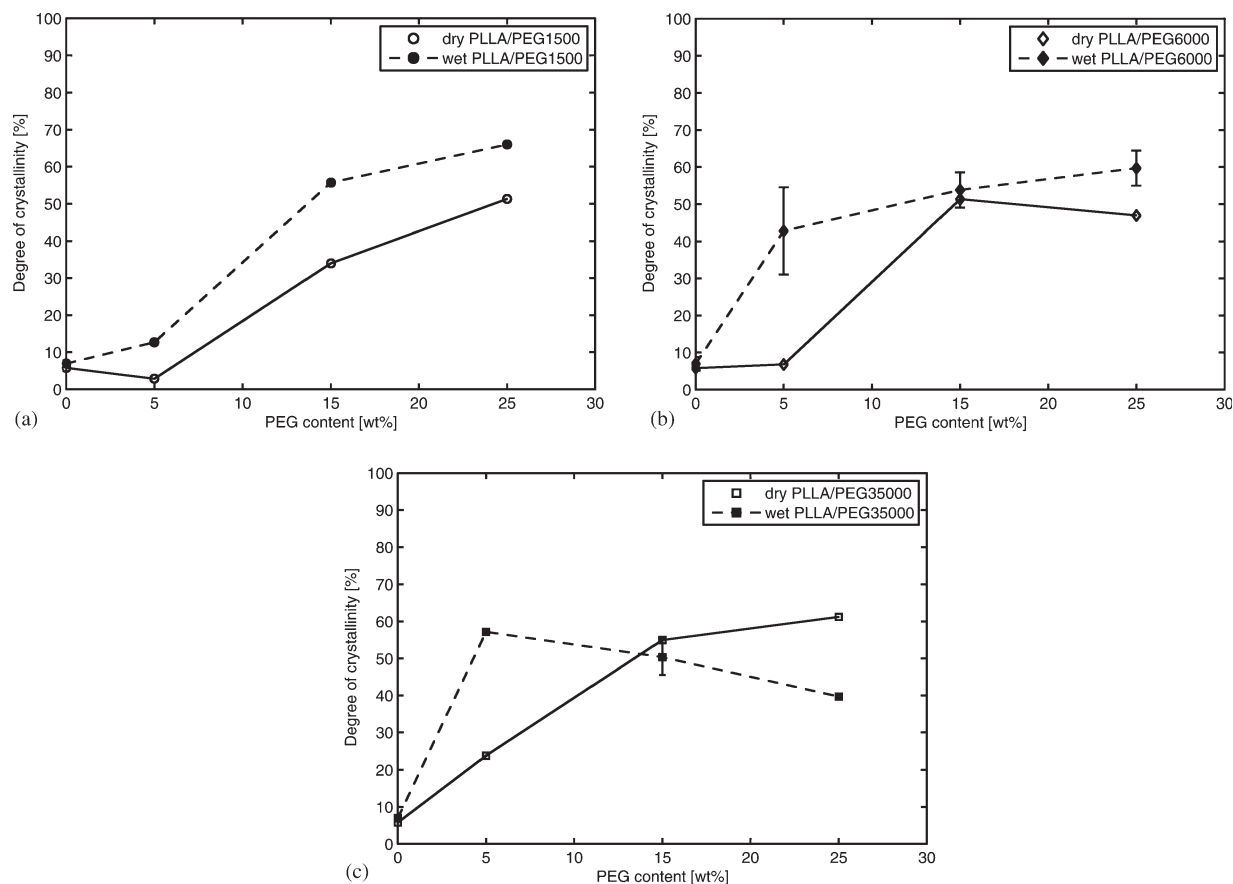
**Figure 4**  $T_g$  of the PLLA/PEG blends in the dry state as a function of the PEG content along with predictions from the Fox equation.



**Figure 5** DSC Glass transitions of PLLA 25 wt % PEG blends.

mass of the PEG was observed by DSC.  $T_m$  remained in the same range<sup>22,25</sup> as that of PLLA1 (182°C), with the exception of the blends with PEG1500, which exhibited significantly reduced  $T_m$  of about 174°C for 15 and 25 wt % PEG. Similar results have been obtained elsewhere for PLA/PEG400 blends,<sup>24</sup> which suggests  $T_m$  to decrease in the presence of low molar mass PEG. As with PLLA1 and Copo1, a small endothermic peak associated with the  $\alpha'$  to  $\alpha$  transformation was observed immediately below the main melting peak for PLLA/5 wt % PEG, but not at higher PEG contents.

As reported elsewhere,<sup>22,25</sup> the apparent initial degree of crystallinity of the dry blends increased strongly with PEG content and molar mass, as shown in Figure 6, reaching up to 60 wt % in the blends containing 25 wt % PEG. These increased degrees of crystallinity were associated with a strong decrease in the magnitude of the cold crystallization peaks observed in the first DSC heating scans (cf. Fig. 3). It was therefore inferred that plasticization in the presence of PEG led to significantly increased crystallization rates during processing. Thus, the crystallization temperature of the PLLA during the first cooling scan also increased systematically with the PEG content and decreasing PEG molar mass, as shown in Figure 7, as reported by Hu et al.<sup>25</sup> and Li et al.<sup>26</sup> Even so, for the cooling rate of 10 K/min used for the DSC measurements a PEG content of 5 wt % was sufficient to ensure a final degree of crystallinity of about 60 wt %, which compares with about 20 wt % for the PLLA1 after cooling. The apparent inconsistencies between the observed  $T_g$  in the dry specimens and Eq. (1), may be at least in part due to the significantly lower degrees of crystallinity in PLLA/5 wt % PEG than at higher PEG contents, confinement in the presence of crystalline lamellae generally tending to limit the mobility of the amorphous phase, as will be discussed further in



**Figure 6** Degree of crystallinity of the different moldings in the dry and wet states.

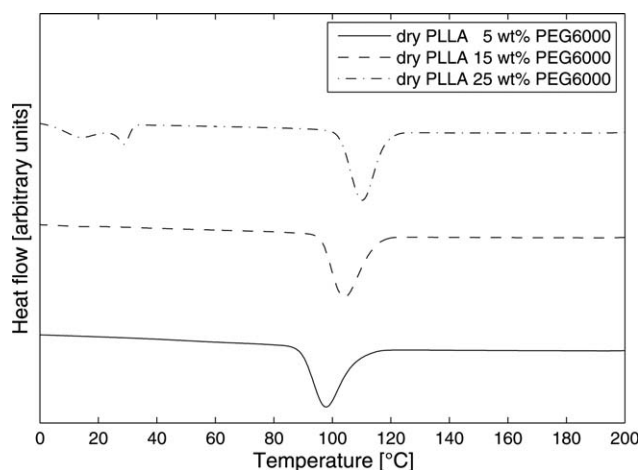
what follows in the context of the  $T_g$  data for the same specimens in the wet state.

Exothermic crystallization peaks of PEG were also observed for the blends containing 25 wt % PEG in the temperature range 10–30°C (as shown for example in Fig. 7 for PLLA/25 wt % PEG6000), again reflecting the phase separation inferred from the first heating scans, as well as additional transitions in the temperature range –35 to –20°C (–30, –24, and –21°C for PEG1500, PEG6000, and PEG35000, respectively), which were assumed to be associated with the glass transition of the PEG. These values were somewhat higher than the  $T_g$  for the corresponding pure polymers, and the fact that the lowest  $T_g$  was observed for PEG1500 suggested a higher degree of phase purity in this case.

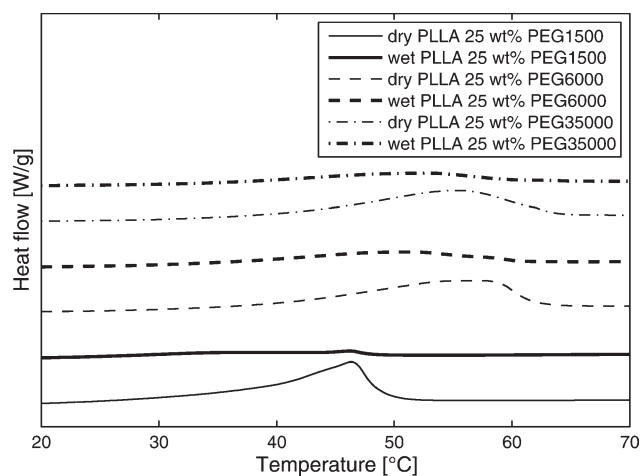
Crystallization of the PEG was also observed in the second heating scans for the blends containing 25 wt % PEG, which showed melting peaks at 46, 55, and 56°C for PEG1500, PEG6000, and PEG35000, respectively, i.e., a somewhat lower temperatures than for the pure PEG as shown in Figure 8. The melting point depression was nevertheless reduced for PEG1500 compared with the other molar masses, again suggesting a higher degree of phase purity.

#### Wet state

In the wet state, the  $T_g$  of PLLA1 (58°C) was similar to that in the dry state, whereas those of Copo1 and Copo2 (53 and 48°C, respectively) were somewhat lower than those of the corresponding polymers in the dry state. The glass transitions were associated



**Figure 7** DSC first cooling thermograms showing crystallization in PLLA/PEG6000 in the dry state.

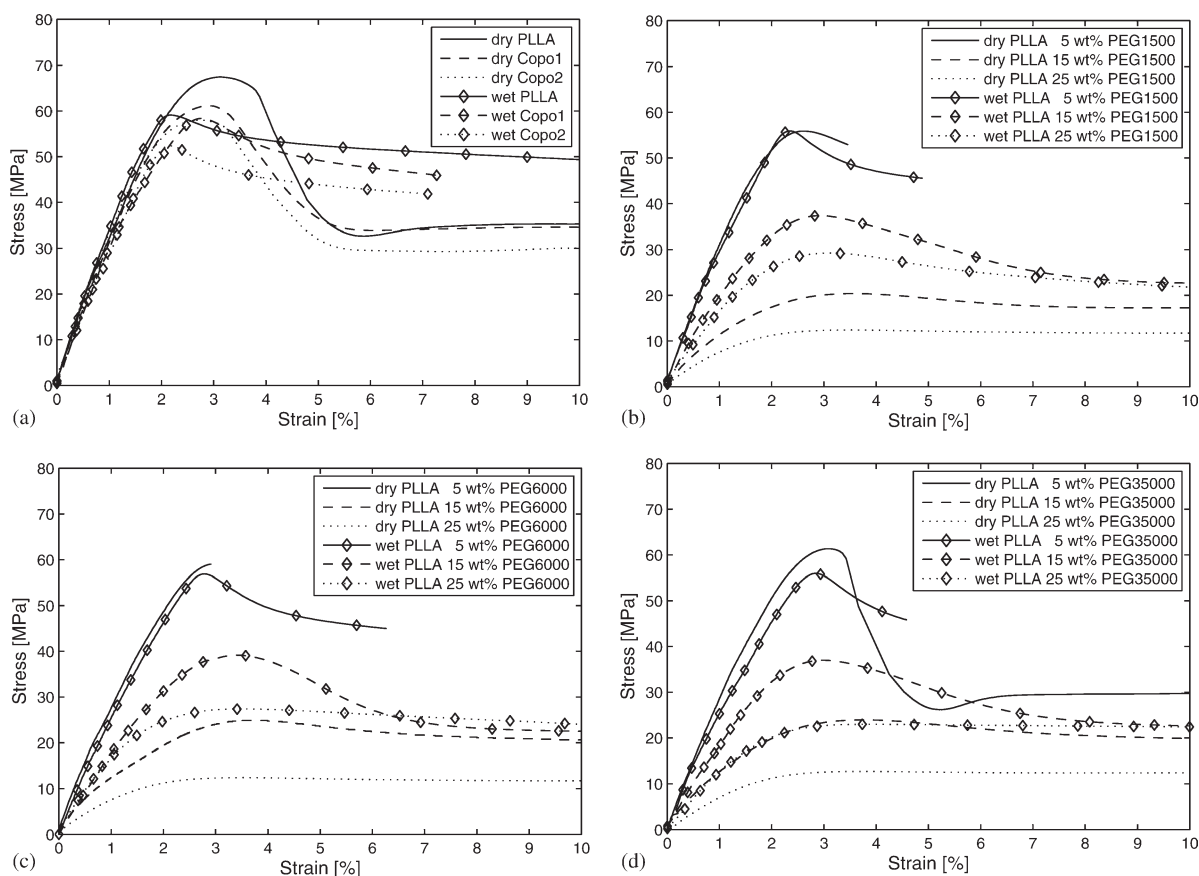


**Figure 8** DSC second heating thermograms in the vicinity of the PLLA glass transition in dry and wet PLLA/25 wt % PEG moldings.

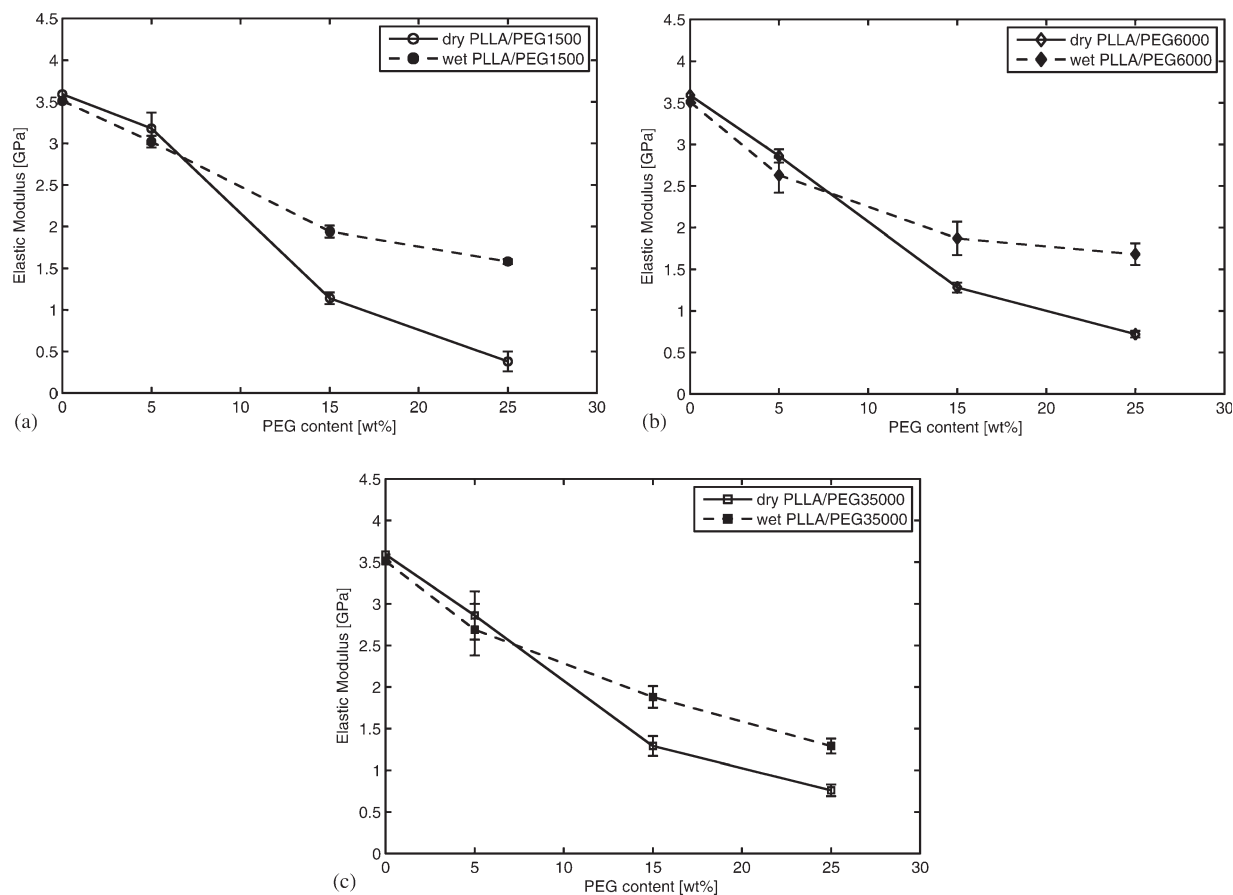
with marked endothermic peaks suggesting significant physical ageing, reflecting the fact that  $T_g$  remained well above 37°C. The degrees of crystallinity of PLLA1 and Copo1 also increased slightly in the wet state (from 5.4 to 6.5 wt % for PLLA1 and from 10.9 to 11.5 wt % for Copo1).

For PLLA/5 wt % PEG,  $T_g$  in the wet state was generally higher than in the dry state (48 and 52°C in the wet state, compared with 44 and 45°C in the dry state for PLLA/PEG1500 and PLLA/PEG6000). This may be attributed to relatively large increases in crystallinity during immersion in the PBS (see below), which are expected to reduce the mobility of the amorphous phase. On the other hand, the  $T_g$  was lower in the wet state than in the dry state at 15 wt % PEG (34 and 29°C compared with 36 and 34°C for PLLA/PEG1500 and PLLA/PEG6000, respectively), indicating plasticization effects to dominate.

Figure 6 shows the degree of PLLA crystallinity determined from first heating DSC scans on specimens in the wet state. As with PLLA1 and Copo1, the degrees of crystallinity were consistently higher than in the dry state, although they tended to reach a plateau at about 60 wt %, so that for PEG35000, for which values of 60 wt % were obtained with as little as 5 wt % PEG, there was no further increase at higher PEG contents (and indeed, the apparent degree of crystallinity decreased somewhat at the highest PEG contents in this case, although this is thought to be an experimental artifact arising from increased baseline curvature owing to water loss during the scans).



**Figure 9** Stress–strain curves for the different moldings in the dry and wet states.



**Figure 10** Elastic modulus,  $E$ , as function of PEG content in the dry and wet states.

These results confirm the PEG<sup>25</sup> and the related water uptake to favor crystallization of the PLLA by increasing its mobility, even where the nominal  $T_g$  in the dry and the wet states were greater than 37°C, as in the case of PLLA/5 wt % PEG6000 and PLLA/5 wt % PEG35000. On the other hand, these data provide no information on the state of the PLLA/5 wt % PEG blends in the PBS prior to the observed increases crystallization, for which  $T_g$  is expected to be significantly lower than the values given for the wet specimens.

The PEG melting peaks observed in the second heating scans were less marked than for the specimens initially in the dry state, particular for the lower PEG molar mass as shown in Figure 8. This is consistent with a loss in PEG owing to dissolution by the PBS, as suggested by the changes in specimen weight (Fig. 1) and hence with the results of Sheth et al.,<sup>22</sup> who also reported dissolution of PEG from PLA/PEG with  $T_g$  less than 37°C on immersion in PBS at 37°C.

## Mechanical properties

### Dry state

The stress–strain behavior of the various materials is summarized in Figure 9, and the trends in  $E$  and  $\sigma_y$

are shown in Figures 10 and 11, and in Table II. In the dry state,  $\sigma_y$  of PLLA1 was 67 MPa and the elongation at break was of the order of 140%. Copo1 and Copo2 showed similar stress–strain curves with similar  $\epsilon_{\max}$  to PLLA1 but lower  $\sigma_y$  (62 and 59 MPa, respectively). All of these materials deformed by localized necking, and the necked regions generally became opaque. Necking was also observed for PLLA/5 wt % PEG and PLLA/15 wt % PEG in the dry state, regardless of the molar mass of the PEG, but not for PLLA/25 wt % PEG, which deformed homogeneously.

As seen from Figure 10,  $E$  in the dry state was strongly influenced by the PEG content for all the PEG molar masses considered, that of PLLA/PEG1500 decreasing from around 3.6 GPa (PLLA1) to 1.3 GPa and 0.7 GPa on addition of 15 and 25 wt % PEG, respectively. The  $\sigma_y$  for PLLA/5 wt % PEG were comparable with those of Copo1 and Copo2, although PLLA/5 wt % PEG1500 and PLLA/5 wt % PEG6000 showed even lower elongations at break than the copolymers. At higher PEG contents, however,  $\epsilon_{\max}$  increased to more than 100% in the blends, and the  $\sigma_y$  decreased systematically with molar mass, consistent with previous work.<sup>9,15,22,24</sup> These trends were associated with various failure mechanisms: PLLA/15 and 25 wt % PEG yielded by



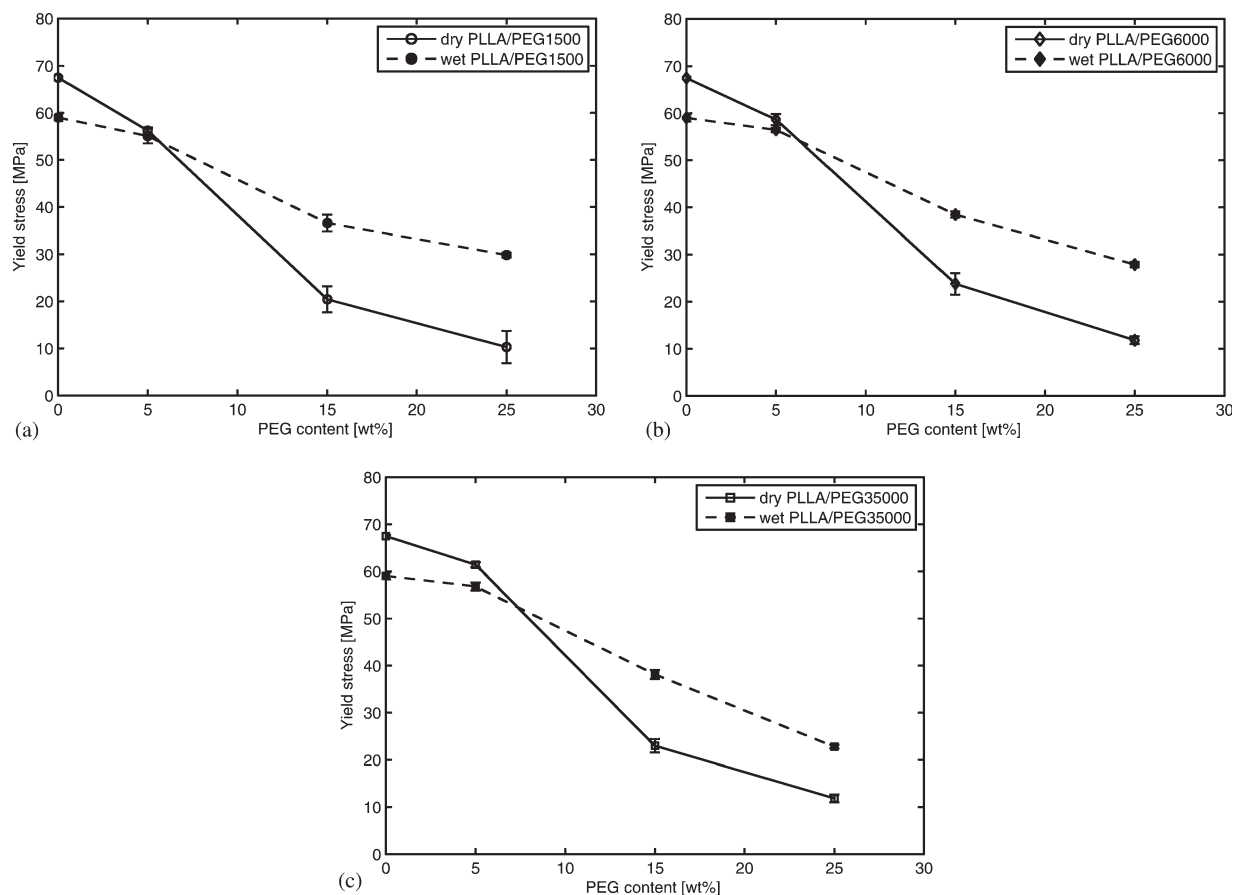


Figure 11 Yield stress,  $\sigma_y$ , as function of the PEG content in the dry and wet states.

TABLE II  
Mechanical Properties of the Copolymers and Blends in the Dry and Wet States

	Elastic modulus (GPa)		Yield stress (MPa)		Yield strain (%)		Elongation at break (%)	
	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet
PLLA1	3.59 ± 0.27	3.51 ± 0.16	67.4 ± 0.3	59 ± 0.7	2.9 ± 0.2	2.1 ± 0.1	139 ± 15.2	14.6 ± 0.5
Copo1	3.45 ± 0.28	2.99 ± 0.21	62.3 ± 1.3	59 ± 0.6	2.8 ± 0.1	2.8 ± 0.2	121.6 ± 40.1	7.3 ± 0.4
Copo2	3.6 ± 0.4	3.11 ± 0.18	59 ± 1.1	51.5 ± 0.8	2.7 ± 0.3	2.2 ± 0.1	181.6 ± 7.4	10.3 ± 3.8
PLLA 5 wt % PEG1500	3.18 ± 0.19	3.02 ± 0.07	56.2 ± 0.7	55.1 ± 1.5	2.6 ± 0.1	2.3 ± 0.1	3.3 ± 0.2	6.9 ± 1.7
PLLA 15 wt % PEG1500	1.14 ± 0.07	1.94 ± 0.07	20.4 ± 2.8	36.6 ± 1.8	3.7 ± 0.1	2.9 ± 0.2	249 ± 22.7	176.7 ± 72.1
PLLA 25 wt % PEG1500	0.38 ± 0.12	1.58 ± 0.03	10.3 ± 3.4	29.8 ± 0.5	N/A	3.2 ± 0.1	197 ± 26	42.5 ± 20.3
PLLA 5 wt % PEG6000	2.86 ± 0.08	2.63 ± 0.21	58.6 ± 1.8	56.5 ± 0.5	3 ± 0.1	2.7 ± 0.2	3.3 ± 0.3	5.2 ± 1
PLLA 15 wt % PEG6000	1.28 ± 0.06	1.87 ± 0.2	23.8 ± 2.3	38.5 ± 0.7	3.6 ± 0.2	3.3 ± 0.1	241.7 ± 10.7	207.7 ± 25
PLLA 25 wt % PEG6000	0.72 ± 0.04	1.68 ± 0.13	11.8 ± 0.8	27.9 ± 0.6	N/A	3.4 ± 0.2	176.9 ± 10	139 ± 12.05
PLLA 5 wt % PEG35000	2.86 ± 0.29	2.69 ± 0.31	61.4 ± 0.5	56.8 ± 0.8	3.2 ± 0.2	2.9 ± 0.2	134.5 ± 86.2	4.2 ± 0.4
PLLA 15 wt % PEG35000	1.29 ± 0.12	1.88 ± 0.13	23 ± 1.4	38.1 ± 1	3.8 ± 0.2	3.1 ± 0.2	233.1 ± 2.2	140.5 ± 32.4
PLLA 25 wt % PEG35000	0.76 ± 0.07	1.29 ± 0.09	11.8 ± 0.8	22.8 ± 0.4	N/A	3.8 ± 0.5	236.7 ± 4.4	97.4 ± 61.4

homogenous necking, PLLA/5 wt % PEG1500 and PLLA/5 wt % PEG6000 showed extensive crazing and brittle fracture while PLLA/5 wt % PEG35000 underwent localized necking.

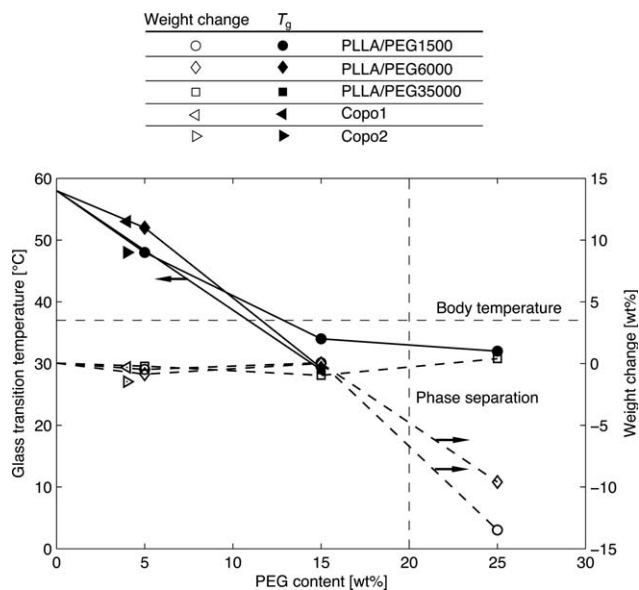
#### Wet state

In the wet state, PLLA1, Copo1, and Copo2 showed lower  $E$  and lower  $\sigma_y$  than in the dry state, and homogenous deformation up to failure, which occurred at around 7% strain for the copolymers. PLLA/5 wt % PEG showed further decreases in  $E$  and  $\sigma_y$  compared to those observed for the specimens in the dry state, particularly at the highest PEG molecular weights. On the other hand, PLLA/15 wt % PEG and PLLA/25 wt % PEG showed higher  $E$  and  $\sigma_y$  than observed for the specimens in the dry state. The corresponding failure modes were crazing in PLLA/5 wt % PEG, localized necking in PLLA/15 wt % PEG and homogenous deformation in PLLA/25 wt % PEG.

#### Overall assessment of the different materials

Figure 12 summarizes the data for  $T_g$  (in specimens for which a well-defined  $T_g$  could be observed) and the difference between the overall weight change and the water uptake (Fig. 1) in the various moldings after 5 days at 37°C in PBS, which is assumed to reflect any weight loss from the PLLA/PEG matrix. The moldings may be separated into two categories: those whose  $T_g$  remained above 37°C and those whose  $T_g$  was lower than 37°C, and which may therefore be assumed to show a rubbery response under these conditions, consistent with the overall aim of this work as set out in the introduction, namely the identification of materials with potential for the replacement of soft tissue. At the same time however, the excessive weight loss from the matrix of moldings containing 25 wt % PEG1500 and PEG6000 suggested these materials to be particularly unstable during immersion in PBS with respect both to phase separation and to leaching of the PEG, as reflected by the relatively high  $T_g$  for PLA/25 wt % PEG1500, and DSC also indicated substantial phase separation in PLLA/25 wt % PEG35000. Moreover, plasticization of the PLLA matrix by the PEG coupled with the effects of water absorption resulted in substantial increases in crystallinity in the blends with low PEG contents, which showed relatively low degrees of crystallinity in the dry state.

The composition of the blends was reflected by their mechanical response at room temperature. Thus,  $E$  in both the dry and the wet states could be reduced by increasing the PEG content, reflecting the corresponding changes in  $T_g$ , although the presence of short PEG chains may also be detrimental to the me-



**Figure 12** Summary of  $T_g$  in the wet state (solid symbols) and the matrix weight change defined as the difference between the overall weight change and the water uptake after immersion in the PBS, as a function of the initial PEG content for the various moldings.

chanical properties at large deformations. Thus, Copo1 and Copo2, which contained 4 wt % of PEG, were significantly more ductile than the PLLA/5 wt % PEG1500 and PLLA/5 wt % PEG6000, although comparable decreases in  $E$  were observed in each case. Nevertheless, at higher concentrations of PEG,  $\sigma_y$  decreased substantially owing to the increased molecular mobility, so that ductility was recovered, even for the lowest PEG molar masses. Depending on the PEG molar mass, the decreases in  $E$  with PEG content in the blends were more limited in the wet state than in the dry state, despite the significant water uptake, which one would expect to lead to increased plasticization of the PLLA, as observed for PLLA1, Copo1, Copo2, and PLLA/5 wt % PEG (although this was not reflected by large changes in  $T_g$ ). These results underline the importance of phase separation, crystallization of the PEG (as evidenced by the DSC scans), and leaching of the PEG at high PEG contents during immersion in PBS at 37°C, all of which may contribute to the observed increases in  $E$ . These effects were generally reduced for the highest molar mass PEG, presumably owing to its reduced mobility.

Given that large decreases in stiffness *in vivo* will generally require  $T_g$  to be below 37°C, PLLA/15 wt % PEG35000 ( $T_g$  in the wet state of approximately 34°C) was considered to be the candidate material with the most potential for use in soft scaffolds owing to its relatively good stability in PBS at 37°C. However, the relatively high degrees of crystallinity of such blends, particularly after relatively short exposures to PBS at 37°C, are expected to limit the range of moduli and

deformability that may be achieved above  $T_g$ , despite their relatively minor influence on  $E$  in the glassy state. It would therefore be of interest to investigate the replacement of PLLA by a PLA with a reduced tendency to crystallize, which may also contribute to the thermodynamic stability of the blends.<sup>9,10</sup>

## CONCLUSIONS

Consistent with previous work, the  $T_g$  of injection molded specimens of a bioresorbable medical grade PLLA was found to decrease strongly with PEG content, falling below normal body temperature at PEG contents of 15 and 25 wt %. At the same time, however, the degree of crystallinity of the PLLA also increased strongly with PEG content, reaching up to 60 wt % for 25 wt % PEG. Moreover, after immersion in PBS for 5 days at 37°C, to simulate conditions *in vivo*, the blends with the highest PEG contents also showed increased water uptake, and significant mass loss, assumed to be due to phase separation and leaching of the PEG, while the blends with the lower PEG contents showed relatively large increases in their degree of crystallinity. The maximum decreases in the room temperature elastic moduli were limited to about 60% under these conditions, an effect that was attributed both to phase separation and crystallization of the PEG, and to leaching of the PEG at high PEG contents. Of the blends investigated, PLLA/15 wt % PEG35000 was considered to be the most interesting candidate for use in soft scaffolds owing to its relatively good stability in PBS at 37°C, which may be imputed in turn to the relatively high molar mass of the PEG, and its  $T_g$  of approximately 34°C, implying the amorphous content to be in the rubbery state *in vivo*. However, while they clearly contribute to mechanical stability, the relatively high degrees of crystallinity of such blends, particularly after relatively short exposures to PBS at 37°C, are expected to limit the range of moduli and deformability that may be achieved above  $T_g$ . It would be of interest therefore to investigate the use of PLA with a reduced degree of crystallinity as a matrix. Moreover, copolymerization rather than blending with the PEG results in improved stability during ageing, but although the available materials showed decreases in  $T_g$  consistent with those observed in the blends for a given PEG content, the maximum available PEG content was too low to reduce  $T_g$  to below 37°C. It is therefore envisaged to prepare PLA-PEG copolymers with higher PEG contents (typically 20 wt %) for further assessment.

The emphasis will nevertheless remain on the use of blends, since these retain several key advantages, including relatively straightforward specimen preparation and versatility with regard to fine tuning of the final properties. The next step will be to prepare foamed specimens from PLLA/PEG in the appropri-

ate composition range and investigate their mechanical response and biocompatibility *in vitro* under much longer term exposure to simulate body conditions, where not only leaching of the PEG, but also bioresorption of the PLLA will begin to play a significant role in the evolution of their properties. It will also be important to investigate eventual modifications to the short- and long-term *in vitro* response arising from the sterilization procedures required for biocompatibility and *in vivo* testing.

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