White-Paper: Thermogels Additive effects and dissolution speed  
August 28, 2017

Introduction
Polymers which have the correct architecture including poly(ethylene glycol) and hydrophobic polyester blocks at the right molecular weights have the capability to dissolve in low-temperature water and then turn into gels as the water is heated. This thermogelation property is related to the selective dehydration of the PEG chain at higher temperatures which cause the polymer micelles to bridge together forming the gel. This process involves hydrogen bonding and water structure within the solution and, as such, it can be affected by additives.

A functionality issue for thermogels is the excessive time required for their dissolution (typically days). This is due to the fact that, by definition, they are on the edge of being water soluble. Naturally, increasing the PEG block size can improve their water solubility but then such polymers are no longer thermogels but simply water soluble within the meaningful temperature range.

Purpose
In this research, we explore strategies to improve thermogel dissolution time as well as explore the effects of various additives on dissolution speed and gelation. We focus our efforts on known thermogel PLCL-PEG-PLCL at a constant solution concentration of 20% w/v water.

Materials and Methods
PLCL-PEG-PLCL (~1700-1500-1700 Da, 60:40 CL:LA) PolyVivo AK109 (Lot#60301ELH-A)  
PEG 400Da (Fluka)  
Dichloromethane (Malinckrodt)  
Mannitol (Mannogen EZ Spray dried)  
N-methylpyrrolidinone (NMP, Aldrich)  
Dimethylsulfoxide (DMSO, Aldrich)  
mPEG-P(DL)La (2000-2200 Da) PolyVivo AK009 (Lot# 70207STR-C)

Methods
Aliquots of known thermogel PLCL-PEG-PLCL (AK109, (200-300 mg)) were put into 5 ml vials containing 12 mm magnetic stir-bars along with the additives indicated in Table 1. These were then prepared as shown in Table 1.

Table 1. PLCL-PEG-PLCL aliquot preparations

<table>
<thead>
<tr>
<th>Formulation</th>
<th>PLCL-PEG-PLCL (mg)</th>
<th>Additive (mg)</th>
<th>Treatment/Preparation</th>
</tr>
</thead>
<tbody>
<tr>
<td>SGT170726A</td>
<td>238.2</td>
<td>None (control)</td>
<td>None</td>
</tr>
<tr>
<td>SGT170726B</td>
<td>300.7</td>
<td>Mannitol (309.9 mg)</td>
<td>Stirred under argon at 40 °C to mix and chopped up using metal spatula</td>
</tr>
<tr>
<td>SGT170726C</td>
<td>213.1</td>
<td>NMP (213 µL)</td>
<td>Stirred to mix ambient</td>
</tr>
<tr>
<td>SGT170726D</td>
<td>209.4</td>
<td>DMSO (209 µL)</td>
<td>Freeze dried in HarvestRight freeze-dryer</td>
</tr>
</tbody>
</table>
Additionally, a sample of 8.3 mg of PLCL-PEG-PLCL was prepared in a DSC pan and analyzed using a TA instruments Q2000 modulated DSC. Under 50 ml/min argon flush, the sample was chilled to -80 °C and then heated with modulation of ± 1 °C/min with a ramp of 3 °C/min up to 200 °C.

To each of these, a volume of water equivalent to 5 times the mass was added to each vial to generate a 20% w/v solution of the PLCL-PEG-PLCL (regardless of additive mass). The solutions were then stirred/shaken at room temperature and observed for dissolution. Upon dissolution, the samples were placed in a static 37 °C incubator (Quincy Labs, Inc. Model 12-140)

### Results/Discussion

#### Dissolution/Gelation

Table 2. Below summarizes the dissolution and gelation results. Figure 1. Shows the dry-state condition of the prototypes.

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Dry Appearance</th>
<th>Dissolution</th>
<th>Gelation</th>
</tr>
</thead>
<tbody>
<tr>
<td>SGT170721A</td>
<td>Viscous gel</td>
<td>Solution after 2 days of agitation at room temperature</td>
<td>Solution at room temperature, Gelled at 37°C</td>
</tr>
<tr>
<td>SGT170721B</td>
<td>White clumpy powder</td>
<td>Gel after 2 days of agitation at room temperature</td>
<td>Gelled at room temperature, solid precipitate at 37°C</td>
</tr>
<tr>
<td>SGT170721C</td>
<td>Solution</td>
<td>Solution after 3 minutes of agitation at room temperature</td>
<td>No gelation up to 37°C</td>
</tr>
<tr>
<td>SGT170721D</td>
<td>Viscous gel</td>
<td>Solution after 2 days of agitation at room temperature</td>
<td>Solution at room temperature, Gelled at 37°C</td>
</tr>
<tr>
<td>SGT170721E</td>
<td>Viscous gel with yellow layer</td>
<td>Sample observed to be contaminated with oil, dissolution determined to be &gt; 2 hours</td>
<td>Not tested</td>
</tr>
<tr>
<td>SGT170721F</td>
<td>Solution</td>
<td>Gel after 3 minutes of agitation at room temperature</td>
<td>Gelled at room temperature, solid precipitate at 37°C</td>
</tr>
<tr>
<td>SGT170726A</td>
<td>Viscous gel</td>
<td>Solution after 1 day of agitation at room temperature</td>
<td>No gelation up to 37°C</td>
</tr>
<tr>
<td>SGT170726B</td>
<td>Viscous gel</td>
<td>Solution after 1 day of agitation at room temperature</td>
<td>No gelation up to 37°C</td>
</tr>
<tr>
<td>SGT170726C</td>
<td>Viscous gel</td>
<td>Dissolution between 1 - 4 days of agitation at room temperature</td>
<td>Gel at room temperature, liquefied at 37°C</td>
</tr>
</tbody>
</table>
Figure 1. Dry Form (A) 170721* series (B) 170726* series

**Crystalline form**
One manner to improve dissolution speed is to convert a material into a porous form with improved surface area for water access. Earlier attempts at simply freeze-drying the PLCL-PEG-PLCL from water in a conventional freeze-drier (HarvestRight) from water failed to generate a porous foam as the gel collapsed on itself. Additionally, freeze-drying from DMSO solvent (SGT170726D) failed to generate a porous gel as the .

In this research, the polymer was dissolved in dichloromethane (prototype SGT170726E) and deep vacuum dried (Welch Duoseal) with cooling in a dry-ice/acetone bath. **Figure 2** shows images from this.

Figure 2. (A) deep vacuum drying in dry-ice/acetone bath, (B) crystalline-foam PLCL-PEG-PLCL freshly pulled from dry-ice/acetone bath

The gel, however, did not maintain this form. Storing the gel overnight in the freezer at -20 °C it collapsed back down into the typical, liquid-gel form.

To explore this further, DSC was performed. **Figure 2**. Shows the results from this.
As determined by DSC, the glass transition for this material is around –47.45°C, which puts it well below the temperature of conventional freezer storage. Outright melting occurs at 12.59°C indicating that warming the solid polymer to room temperature will liquefy it into a gel.

**Additives**
As for additives for additives, the results indicate the following trends:

**NMP**
NMP was applied as it is a biocompatible solvent. This solvent added at a 1:1 ratio with the polymer did leave a material which was readily water soluble but the solvent itself then interfered with gelation. This may still be applied in situations where it is expected that the system will leach the solvent away leaving only the polymer.

**Mannitol**
Mannitol is often used as an anti-caking agent for parental formulations. It actually provides for the capability to render the thermogel as a porous solid with high surface area simply by soaking up the gel and providing mechanical support. Despite this, it does not improve dissolution speed and acts to lower the gel-temperature likely by encouraging hydrogen bonding.

**mPEG-P(DL)La (2000-2200Da)**
The polymer selected for this application is known, by itself, to be fully water soluble with no notable thermogelation. It is also naturally a porous solid. The addition of this material did not enhance the dissolution rate of the thermogel and acted to prevent gelation at 37°C.

**PEG 400**
Predissolving the thermogel in PEG 400Da solvent acted to improve solubilization in water. Care must be taken, however, to not use too much PEG 400Da as it also acts to lower the gel temperature at high concentrations. PEG 400Da seems most effective when an equivalent volume that is 50% the mass of the thermogel is added (e.g. 0.5 ml PEG 400/1 g of thermogel) for the PLCL-PEG-PLCL AK109 type thermogel. This is suitable to improve dissolution speed to a few hours while not drastically lowering the gel point.