Supplementary Materials:

An In Situ Wet Chemical Methodology to Fabricate Hydroxyapatite based Nanostructured Biocomposites**

Prakash Hariram Kithva, Lisbeth Grøndahl, Rajendra Kumar, Darren Martin and Matt Trau*

On the determination of timing of polymer solution addition to the reaction mixture:

The pH of the reaction mixture (Ca(OH)$_2$ + H$_3$PO$_4$) is maintained as the acid is added due to the dissolution of non-dissociated Ca(OH)$_2$. The dissolution of Ca(OH)$_2$ is accompanied by the consumption of hydroxyl ions and calcium ions as a result of neutralization and ACP formation. However, the pH of the reaction mixture starts decreasing after the complete dissolution of Ca(OH)$_2$ (event 1) and the ACP starts transforming into crystalline HA (event 2) once the former reaches a critical concentration, C* (C* is about 10.5 mM at 30 °C and 12.6 mM at 95 °C and would be slightly greater than 12.6 mM at 98 °C)\textsuperscript{15}. Therefore, the polymer solution should be added to the reaction mixture before the ACP particles starts transforming into crystalline HA for maximum interaction between ACP and polymer and just after the pH of the reaction mixture starts to decrease from its initial value to avoid precipitation of, particularly, chitosan. The equation to determine the time at which the pH of the reaction mixture starts to decrease is given below\textsuperscript{15}.

\[
[Ca(OH)_2] = \left(\frac{k_{sp}}{4}\right)^{\frac{1}{3}} (V_i + fr \cdot t) + \frac{3}{2} C \cdot fr \cdot t 
\]

where, [Ca(OH)$_2$] is the total dissolved quantity of Ca(OH)$_2$, (mol), $k_{sp}$ is the solubility product of Ca(OH)$_2$, (the unit for the term $(k_{sp}/4)^{1/3}$ is mol/L), $V_i$ is the initial volume of suspension (L), $fr$ is the acid flow rate (4x10$^{-3}$ L/min), $t$ is the time (min), and $C$ is the concentration of H$_3$PO$_4$ (mol/L)\textsuperscript{15}.

The $k_{sp}$ of Ca(OH)$_2$ at 98°C is calculated to be 1.60x10$^{-6}$ using the vant Hoff’s equation (Eqn. 2\textsuperscript{S}).

The $k_{sp}$ of Ca(OH)$_2$ at 25°C is 6.6x10$^{-6}$ and the heat of dissociation, $\Delta H_r$ is −17.88 kJ/mol for the reaction, Ca(OH)$_2$ → Ca$^{2+}$ (aq) + 2OH$^-$ (aq)\textsuperscript{25}.

\[
\ln \frac{k_{sp}^{25}}{k_{sp}^{98}} = \frac{\Delta H_r}{R} \left( \frac{1}{298} - \frac{1}{T} \right) 
\]

where, $R$ is the universal gas constant (8.314 J/mol-K) and $T$ is the absolute temperature.

Therefore, in the suspension containing 15mmol Ca(OH)$_2$ in 225mL water at 98°C, about 1.66 mmol of Ca(OH)$_2$ dissociates into Ca$^{2+}$ and OH$^-$ ions. The pH of the reaction mixture starts to
decrease (event 1), in the present study, after approximately 50 minutes (or about 198 mL) of H₃PO₄ addition. The ACP concentration in the suspension would be 9.365 mmol/L (or 3.96 mmol in 423 mL) which is well below the critical concentration. Similarly, in a suspension of 15mmol Ca(OH)₂ in 150 mL water, the pH start decreasing after adding 143 mL acid and the ACP concentration would be 14.6 mmol/L (or 4.28 mmol in 293 mL). Figure 1S shows a schematic describing the effect of concentration of Ca(OH)₂ suspension on the events 1 and 2. The concentration of dissolved Ca(OH)₂ and hence the concentration of OH⁻ ions until the time t₁ can accurately be calculated using Eqn. 1S. After the time t₁, Eqn. 1S can not be used to calculate the concentration of OH⁻ ions as the consumption of OH⁻ ions is not only due to the neutralization by H⁺ ions, but also due to the formation of crystalline HA from ACP. However, the variation of pH with time after the time t₁ is drawn following the experimentally observed trend of the same IS. The increased rate of decrease of conductivity, as a result of consumption of large number of ions after the time t₂ is an indication of the start of ACP → HA transformation, via a combination of the following schemes.

\[
\begin{align*}
\text{Ca}_3(\text{PO}_4)_2 + 2\text{Ca}^{2+} + \text{PO}_4^{3-} + \text{OH}^- &\rightarrow \text{Ca}_5(\text{PO}_4)_3(\text{OH}) \\
3\text{Ca}_3(\text{PO}_4)_2 + \text{Ca}^{2+} + 2\text{OH}^- &\rightarrow 2\text{Ca}_5(\text{PO}_4)_3(\text{OH}) \\
5\text{Ca}_3(\text{PO}_4)_2 + 3\text{OH}^- &\rightarrow 3\text{Ca}_5(\text{PO}_4)_3(\text{OH}) + \text{PO}_4^{3-}
\end{align*}
\]

**Information related to TGA:**

The weight fraction of nHA in composite films is measured using Equation 1 given in the experimental section, whose derivation is given below. It is assumed that the weight loss in composite films is only from the polymers, which is true as HA does not degrade temperatures below 650°C.

\[
W_p + W_c = W_{cp}
\tag{3S}
\]

\[
W_{ash} + W_c = W_r
\tag{4S}
\]

-where, \(W_p\) is weight of polymer, \(W_{ash}\) is weight of residue in pure polymer films after heating, \(W_c\) is weight of nHA, \(W_{cp}\) is weight of composite and \(W_r\) is weight of residue in the composite. By subtracting Eqn. 4S from Eqn. 3S, we eliminate the weight of HA in both the composite and the residue. Thus,

\[
W_p - W_{ash} = W_{cp} - W_r \quad \text{or} \quad \frac{W_p - W_{ash}}{W_p} = \frac{W_{cp} - W_r}{W_p} \quad \text{or} \quad W_p = \frac{W_{cp} - W_r}{f} \tag{5S}
\]
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-where \( f = \left( W_p - W_{ash} \right)/W_p \) is essentially the weight loss in pure polymer film. Equation 5S estimates the weight of polymer in the composite. The final nHA content in the composite is given by Eqn. 6S.

\[
\frac{W_c}{W_{cp}} = \frac{W_{cp} - W_p}{W_{cp}}
\]  

(6S)

By substituting Eqn. 5S for \( W_p \) in Eqn. 6S, we get

\[
\frac{W_c}{W_{cp}} = 1 - \frac{W_{cp} - W_r}{fW_{cp}} = 1 - \frac{\% \text{ wt. loss in composite}}{\% \text{ wt. loss in polymer}}
\]

(7S)

Figure 1S. TGA thermograms of polymers and hybrid films.

Table 1S: Weight loss and weight fraction of HA measured via TGA thermograms of films.

<table>
<thead>
<tr>
<th>Material</th>
<th>Weight loss (%)</th>
<th>Estimated nHA content, (Wt. %)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HCHO treated chitosan</td>
<td>63.5</td>
<td>0</td>
</tr>
<tr>
<td>CS-60nHA</td>
<td>27.8</td>
<td>56.2</td>
</tr>
<tr>
<td>Sodium Alginate</td>
<td>64.5</td>
<td>0</td>
</tr>
<tr>
<td>SA-60nHA</td>
<td>28.9</td>
<td>55.2</td>
</tr>
</tbody>
</table>
Stress vs. Strain curves of polymer and composite films:

Figure 2S: Stress vs. Strain curves of (a) sodium alginate, (b) formaldehyde treated chitosan, (c) SA-60nHA and (d) CS-60nHA films. Note the changed x- and y- axes scale.

Reference