Project 2: Week 4
20. April, 2005
Synthesis and Characterization of a Amyloid-β Fibril-forming Peptide.

A. Today (20. April, 2005)
1. Discuss the theory of chromatography
2. Observations from the lyophilization of the peptide samples
   a. Notes from last week:
      The last step in the cleavage procedure was to take up the peptide pellets in 5 mL water after evaporating away the ether.
      i. Not all of the pellets dissolved. The Aβ(25-35)wt seemed less soluble than the Aβ(25-35)rev.
      ii. Pelleted the undissolved peptide and took up the pellets in a small volume (~ 50-100 μL of 1,1,1,3,3,3 hexafluoroisopropanol (HFIP); both peptides dissolved in HFIP.
      iii. Added the dissolved pellet back to the supernatant. The solution became turbid; the turbidity looked like it was not due to a precipitate, but rather to droplets of an immiscible liquid.
      iv. Froze the two peptide samples in liquid N₂ and placed on the lyophilizer.
   v. Masses for lyophilized products:

<table>
<thead>
<tr>
<th>Peptide</th>
<th>Sequence</th>
<th>Mass of tube</th>
<th>Mass of tube + peptide</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aβ(25-35)wt</td>
<td>GSNKGAIIGLM</td>
<td>6.8852 g</td>
<td>6.9735 g</td>
</tr>
<tr>
<td>Aβ(25-35)rev</td>
<td>MLGIIAGKNSG</td>
<td>6.9142 g</td>
<td>6.9750 g</td>
</tr>
</tbody>
</table>

3. Determine the yield and % yield for the two peptide samples
4. From the literature, find out what you can about the effect that halogenated alcohols have on peptide structure, and in particular, HFIP.
5. In groups, carry out HPLC analyses of the two peptides.
6. When not engaged with the isolation procedure, do the following:
   a. From the literature, find out what you can about the effect that halogenated alcohols have on peptide structure, and in particular, HFIP.
   b. Take a look at Keeler’s NMR tutorial, which can be found on the class website under “Manuals”.

B. For Wednesday (27. April, 2005)
1. Write a description of what a 1D $^1$H-NMR spectrum of the amino acids alanine and leucine should look like.