

**Summer Review Course (CGS 650) - Laboratory**  
**July 18 - August 12, 2011**  
**QE 119 - 12:30 to 5 PM**

**Instructors:**

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**Course description:** This course has been designed to provide the students with a hands-on experience on different molecular and cell biology laboratory techniques currently used in basic science research.

**Course objectives:** At the end of this course the students will be able to perform the following techniques:

- 1) Eukaryotic cell tissue culture
- 2) Eukaryotic cell transfection
- 3) Immunohistochemistry/Fluorescence microscopy
- 4) Protein analysis by Western Blot
- 5) Immunoprecipitation
- 6) Real-Time PCR

**Tentative Schedule**

<b>July</b>	
18	Orientation Laboratory Safety
19	Tissue culture MCF7 cells: <ul style="list-style-type: none"> <li>• Count cells</li> <li>• Seed flasks</li> </ul>
20	Transfection (control and GFP) Start serum starvation
21	Reading assignment (cells starving)
22	Estradiol treat cells (calculations and dilutions) Harvest RNA and store in Trizol Visualize GFP transfected cells
25	Make RNA, DNase treat
26	Take OD readings Make cDNA
27	RT-PCR reactions
28	RT-PCR data analysis Protein standard curves
29	Tissue culture – seed macrophages
<b>August</b>	
1	Harvest macrophages Make protein lysates Take protein readings Set up IP reactions (CD44)
2	Wash IPs, run gels

	Set up Western blots
3	Block blots Put in primary antibody (CD44)
4	Put blots in secondary antibody Develop blots Wash blots Add a different primary (moesin)
5	Add secondary Wash blots Develop again – compare results
8	Immunohistochemistry (IHC) <ul style="list-style-type: none"> <li>• Blocking</li> <li>• Primary antibody</li> </ul>
9	IHC Finish <ul style="list-style-type: none"> <li>• Secondary antibody</li> <li>• Mounting, visualization</li> </ul>
10	**free day**
11	Confocal demonstration
12	1) Lab. Clean up 2) Return coats 3) Survey