Genetics 632, Course Outline:
The purpose of Genetics 632 is to provide historical, basic and up to date information about the flow and regulation of genetic information (from DNA to RNA). It also aims to provide the newest information about relevant technology. Focus is on the biology and regulation of transcription factors, epigenetics, chromatin, and regulation and biology of RNA processes (microRNAs, RNA processing/splicing, etc.).

Lectures will be in 321 MacNider (except on Feb. 27, when the lecture will be in G10 Bondurant)

Instructors:

Baldwin and Strahl	January 9 through March 2 (Martin Luther King Holiday is January 16, no class)
Marzluff:	Begins March 12 (after Spring Break) (Good Friday Holiday is April 6)
	Classes End April 25.

Discussion Sections: Wed. or Friday (2:00 to 3:00 PM). The Wed session will be in 1074 Bondurant, and the Friday session will be in 4074 Bondurant.

Teaching assistants: Stephen McDaniel and Joe Durand

1.Your responsibilities:

a. Read papers (usually two or three with a possible review article) and do your very best to understand them before class. We will try to introduce the papers to some degree in the preceding lecture, pointing out techniques, etc. Lectures will encompass: (1) the concepts and conclusions of the papers, (2) limited discussion of some of the experimentation based on your in-class questions, and (3) topics other than the papers. It is important that you understand the major experiments (for some papers we will assign only part of the experiments). Please be prepared to ask about an experiment or procedure that you don't understand (either in class or in the discussion section).

Papers generally will be available on the class website (sakai.unc.edu). Lecture notes will also be posted on the website. You need to have ONYEN/password.

It is important to try to understand the variety of experimental techniques that we will cover (for example, luciferase assays, lacZ assays, in vitro transcription, DNase I footprinting, 2-hybrid screens, ChIP assays, etc.). Some of these techniques will be covered in the Discussion section.

PLEASE BRING THE PAPERS THAT WE ARE DISCUSSING TO CLASS!

b. Answer assigned questions and deposit in class dropbox. These will be graded by TAs. Questions and answers will be posted on the website.

c. Class discussion: we will ask questions in class about the reading, or homework questions.

d. Attend discussion section. This 1 hr period is designed for you to discuss techniques, methodologies, etc. from the assigned reading. Also, you may discuss assigned questions from the lectures. Additionally, for some weeks, we will cover a paper in the discussion section. This is also a good place to have questions about the lectures or about papers answered by the TAs (or passed on to me). One question on the final exam will be derived from the "discussion experience".

e. Do well on the first and final exams. The first exam (January 25) will be relatively short (covering the first 7 lectures). The “final exam” will be a take home exam (given out the last day of this overall section, March 2). For the final exam, we expect you to go farther than just understanding the papers and the lectures. You will need to integrate concepts and mechanisms. The final exam will cover all of the transcription material (i.e., all of the Baldwin and Strahl lectures).

f. Have fun and learn something. . . .

Today:

1. Goals of Genetics 632 and the section on Transcription.
   - gain historical perspectives on transcription research
   - gain knowledge and familiarity with methods and terminology
   - understand structural aspects of transcription factors (DNA binding domains, transactivation domains, dimerization domains, etc.).
   - learn similarities and differences between prokaryotic and eukaryotic mechanisms
   - appreciate the new findings on chromatin regulation and control of transcription
   - recognize that many diseases are controlled by "dysregulated" transcription

2. Discuss historical approaches to understanding the flow of genetic information. Question at the time: what molecular material encodes genetic information and how is this code expressed?

   - (1944) Avery, MacLeod, and McCarty: could "transform" the phenotype of rough, small, non-pathogenic bacteria (pneumococcus) to smooth, large and pathogenic through exposure to DNA from pathogenic strain. DNaseI removed activity. Suggests DNA contains genetic information.
   - (1951) Hershey and Chase: exposed growing cultures of T2 phage to either ^{35}S-methionine or ^{32}P, then used these "labeled" phage to infect bacteria. They found the ^{32}P associated with the bacteria and not the ^{35}S. Since P is largely associated with DNA (which was known), they concluded that DNA comprised the "genetic" information required for maintenance of the phage lineage.
   - (1950) Erwin Chargaff: published that A=T and G=C in DNA.
   - (1953) Watson and Crick: published a model of double-stranded DNA (using Chargaff's rule and X-ray crystallographic data "obtained" from Rosalind Franklin).
   - (1960) Brenner, Jacob, Meselson, and Spiegelman: showed that during phage infection of bacteria that an RNA component (complementary to phage DNA) was found on the bacterial ribosomes. Ribosomes were thought to be the site of protein synthesis. Thus phage DNA yields phage RNA which associates with ribosomes, which presumably "translates" the RNA information into protein.
   - Subsequent work on the triplet code: programming cell-free extracts (containing ribosomes) with different RNAs showed that the genetic code is a triplet-code of bases found in the RNA.

2. The bacterial (E. coli) genome:
   - circular genome (single origin of replication)
   - protein coding genes - approx. 4000
   - ribosomal/tRNA operons: 16S - tRNA - 23S – 5S - distal tRNAs
   - non-coding/small RNAs: approximately 500 (some intergenic, some anti-sense within a gene)

3. Preview of Wed. (Jan. 11) lecture:
   - (1960) Jacob and Monod: model for the regulation of expression of the lac operon in bacteria.
     - Addition of lactose to bacteria leads to a 10,000 fold induction of β-galactosidase
     - Other gene products were also induced
     - These genes mapped together on the bacterial chromosome (proposed the operon)
     - Another gene locus (i), which maps at a different locus, appears to control the operon expression
     - Different mutations in the I gene product have different phenotypes.
     - Techniques: mostly genetics, lacZ assays
   - Discussion of bacterial RNA polymerase and preview of Burgess et al paper.
     - Bacterial RNA polymerase was known to be comprised of multiple subunits.
     - This paper identified the sigma subunit of RNA polymerase as being important for promoter recognition.
     - Techniques: gel electrophoresis, "transcription assays"
   - Discussion of promoters and preview of Ross et al paper.
     - This paper analyzes a strong bacterial promoter.

- They show that RNA polymerase subunit α contacts the promoter and functions to enhance transcription.
- Techniques: in vitro transcription, DNaseI footprinting

4. Reading assignments for Wed. (see Blackboard 1/11/12 for the pdfs of these papers):

- **F. Jacob and J. Monod**. "On the regulation of gene activity". Published in Cold Spring Harbor Laboratory Symposium, 1961. A review of the lac operon. Read the text associated with Figs. 1-4 and Tables 1-5 (approximately the first half of the review). A scanned paper is on the 1/12/11 Blackboard file.

- **R.R. Burgess et al**. "Factor stimulating transcription by RNA polymerase." Nature 221, p. 43-46. A study describing the identification of sigma factor. Read text for Figs. 1-4 and Tables 1-3. A scanned paper is on the 1/12/11 Blackboard file.

- **W. Ross et al**. "A third recognition element in bacterial promoters: DNA binding by the α subunit of RNA polymerase". Science 262, 1407-1413. A paper describing a regulatory element in certain promoters and the involvement of polymerase α in contacting that site. Paper is on the 1/12/11 Blackboard file.

5. Questions due for Wed. Jan. 11 – see Sakai file for that day.

Baldwin and Strahl lectures: (321 MacNider except Feb. 27 ---which will be G10 Bondurant)
Discussion: Wed and Fri. 2:00 – 3:00, Wed (1074 Bondurant); Fri (4074 Bondurant)

January 9	Introduction and Historical Perspectives on Regulation of Gene Expression (Baldwin)
January 11	Lac Operon, Sigma Factor, and Introduction to promoters (Baldwin)
January 13	cAMP and CAP, Lac Operon Revisited. Introduction to Phage Lambda (Baldwin)
January 16	**Martin Luther King Memorial Day (no class)**
January 18	Phage Lambda Gene Regulation, Trp Operon/Attenuation, and Bacterial Enhancers (Baldwin)
January 20	Basic Eukaryotic Gene Transcription (Strahl)
January 23	Role of the C-terminal Domain of RNA pol II; Elongation cycle (Strahl)
January 25	**MINI EXAM** (covering the first six lectures) Enhancers, locus control regions, silencing elements, insulators and gene organization. Introduction to transcription factor structure/function (Baldwin)
January 27	Transcription factors and genomic imprinting (Baldwin)
January 30	DNA Methylation and Genomic Silencing (Strahl)
February 1	Chromatin and Associated Mechanisms of Transcription (Strahl)

February 3 Introduction to Histone Modifications: Acetylation as a Paradigm (Strahl)

February 6 Histone Code Hypothesis and Histone Cross-Talk (Strahl)

February 8 Role of Histone Methylation in Heterochromatin Formation and Gene Expression (Strahl)

February 10 Histone Methylation in Transcription Elongation (Strahl)

February 13 ATP-Dependent Chromatin Remodeling (Strahl)

February 15 Variant Histones and their Roles in Chromatin Function (Strahl)

February 17 Signaling to Chromatin Factors via Post-Translational Modifications (Strahl)

February 20 Inducible transcription factors I: HIF and NF-κB (Baldwin)

February 22 Inducible transcription factors II: STATs, SMADs, and p53 (Baldwin)

February 24 p53 (Baldwin)

February 27 Transcription and cancer (Baldwin) [**G10 Bondurant**]

February 29 More cancer mechanisms and HIV Transcription (Baldwin)

March 2 Steroid receptors (Baldwin) (**TAKE HOME EXAM – due back March 9**)

Section 3 – Taught by Bill Marzluff

Dr. Marzluff
EMAIL: marzluff@med.unc.edu
Office 208 Fordham Hall 2-2140
For appointments call Delphine Bull 2-8920

RNA Processing and Postranscriptional regulation: From the chromosome to the ribosome

There will be weekly section meetings starting the week of Mar. 19, where you will discuss additional papers relevant to one of the current lecture topics. The last two sections will have presentations by students on assigned papers related to posttranscriptional regulation. We have two special lectures (list in bold) scheduled this semester which are relevant to this course and you are encouraged to attend them.

Mon. Mar. 12 1. RIBOZYMES: tRNA processing: RNase P: an RNA enzyme

Wed. Mar. 14 2. rRNA processing: role of snoRNAs in methylation of rRNA

Wed. Mar. 14 3 P.M., Nobel symposium lecture by Dr. Tom Cech

Fri. Mar. 16 3. Capping and Polyadenylation

Mon. Mar. 19 4. Gene organization: hnRNPs and snRNPs: splicing

SECTION MEETING 1 ENZYMES IN rRNA PROCESSING

Wed. Mar. 21 5. Exon definition: coupling of splicing and polyadenylation

Fri. Mar. 23 6. Alternative splicing I: Drosophila sex determination

Mon. Mar. 26 7. Alternative splicing II: coupling splicing and transcription

SECTION MEETING 2: ALTERNATIVE SPLICING AND DISEASE: SMA and SMN

Wed. Mar. 28 8. Histone mRNA processing and regulation

Fri. Mar. 30. 9. Regulation of transcription elongation: P-TEFb and HIV-Tat

Mon. Apr . 2 10. Translation regulation: cytoplasmic polyadenylation

SECTION MEETING 3: TRANSLATIONAL REGULATION IN XENOPUS DEVELOPMENT

Wed. Apr. 4 11. Mechanism of mRNA Degradation

Fri. Apr. 6 HOLIDAY

Mon. Apr. 9 12. Exon junction complexes and NMD

SECTION MEETING 4: Student talks: Alternative splicing regulation of neural development

Wed. Apr. 11 13. siRNAs: structure/function of “Slicer”

Fri. Apr. 13 14. Application of siRNAs in mammalian cells

Mon. Apr. 16 15. Micro RNA Mechanism and role of miRNAs in cancer

Mon. April 16 2:P.M. Special seminar Dr. Phil Sharp

SECTION MEETING 5: Student talks: Regulation of miRNA function by lncRNAs

Wed. Apr. 18 16. Regulation of heart development by miRNAs

Fri. Apr. 20 17. PAR-CLIP: Identification of RNP binding sites

Mon. Apr. 23 18. LncRNAs: role in regulating mRNA degradation

Wed. Apr. 25 19. Lnc RNAs: are they really non-coding: translation profiling

EXAMS WILL BE AVAILABLE ON LINE ON SUNDAY APRIL 29 AT NOON DUE AT 11:59 P.M. ON WEDNESDAY MAY 2.