

Molecular Biology Lab Presentations
Spring 2010

As part of the Molecular Biology course you will be asked to make an oral presentation to the class on an interesting lab topic/technique that we cannot do in our laboratory. You can choose from the topics below. Or if you prefer, you may present on a topic other than one listed below if you have an interest in it. In some instances, some of the topics below overlap or require some background in other areas.

Some Possible Topics for Molecular Lab Presentations

- Cloning of DNA (recombinant DNA technology)
- Cell culture (usually referred to as tissue culture)
- DNA sequencing/Genome projects
- Genome analysis (*Genome/Transcriptome/Proteome/Interactome*)
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- Measuring gene expression
 i.e. Microarray analysis (gene expression, protein expression, genotyping, reverse transcriptase (real-time) q PCR)
- Molecular methods of associating/mapping specific genes with disease (defects in these genes lead to specific diseases) Similar to DNA fingerprinting
- Making of transgenic organisms
- Cloning of organisms
- Protein structure prediction
- Methods for analyzing/altering gene expression or introducing mutations
- Chromatin analysis (accessibility, histone modifications)
- Gene therapy approaches
- RNA interference
- Micro RNA analysis

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A

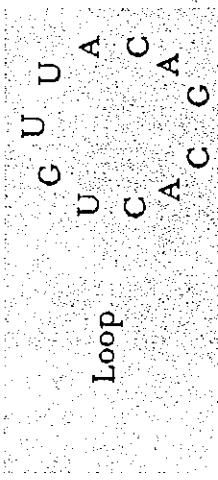
DNA: Inverted
 Coding strand NNNN-TAGCGGCCCATC-NNNNNNNNN-GATGGCCCGCTA-TTTTTT
 Template strand NNNN-ATCGCCCGTAG-NNNNNNNNN-CTACCCCGCAT-AAAAAA
 (N = any base)

TRANSCRIPTION

Repeats

Messenger RNA NNNN-UAGCGGCCCAUC-NNNNNNNNN-GAUGGCCCGCUA-UUUUUUUU

B



Loop

Stem:
(base pairing of inverted repeats)

Terminator



Messenger RNA ... AUUA- UUUUUUUU ... 3' end

Some common named upstream elements in RNAP II promoters

Element	consensus	TF that binds
GC Box	TGCGGCGAATGCC	SP-1 Protein
CAAT Box	GGNCAATCT	CAAT-box complex CBFs
Ap1 box	GCCNNGGC	AP-1
Octamer	ATGCAAAT	Oct-1 Oct-2

***In this short list, the first 3 are considered specific transcription factors (vs. the general TF2A, TF2B etc.) However they are not really specific to any one cell type of gene class. (They could be found in virtually any gene transcribed by RNAP II)

Octamer/Oct-1/Oct-2 are more specific. The octamer sequence is only found in promoters of genes whose products are active in white blood cells. For example your antibody genes have Octamer elements in the promoters.

Topics List
Molecular Biology
Exam3

03/29/2010
Lectures 17-25

Chapter 11 DNA Replication

E. coli

- Origin (OriC) and the proteins that initiate
DnaA, B, C
- RNA primer
- Semi-discontinuous replication
 - Okasaki experiment/pulse-chase method
 - Orientation of replication bubble
 - Role of Pol I in lagging strand synthesis
 - how do each of pol I enzymatic activities fit in?
 - Review animation

Eukaryotes

- DNA pols
- Genome differences
 - Special considerations—how do eukaryotes replicate DNA in a timely manner?
 - Linear chromosome replication—potential for shortening
 - How overcome? /
 - Telomerase—Nobel prize 2009
 - Role in cell aging/cancer

In vitro replication

- PCR
- Sequencing
- Disruption as a disease treatment
- Nucleotide analogs (especially dideoxynucleotides)

Chapter 14 Transcription

- Major similarities/differences from replication
- Mechanism (templated polymerization of NTPs)
- Mechanism (experimental evidence)
- Transcription unit (gene) designation
- Basic structure of a gene
- Cis and trans elements necessary for transcription

E. coli

- Basic gene structure
- One RNA pol
 - Sigma (s) and core
- Stages
 - Initiation
 - Elongation
 - Termination (sequence/structure/intrinsic/rho-dependent)
 - Polycistronic mRNA
 - Antitermination by phage

Eukaryotes

Basic differences of genes/ gene structure (vs. prokaryotes)

3 polymerases

details of (RNAPol I, III, and II) genes/transcription

Gene structure (reg elements/ Promoters/upstream elements/distant elements/enhancers)

TFs (general, common, gene (cell-type) specific)

General role of each type of TF

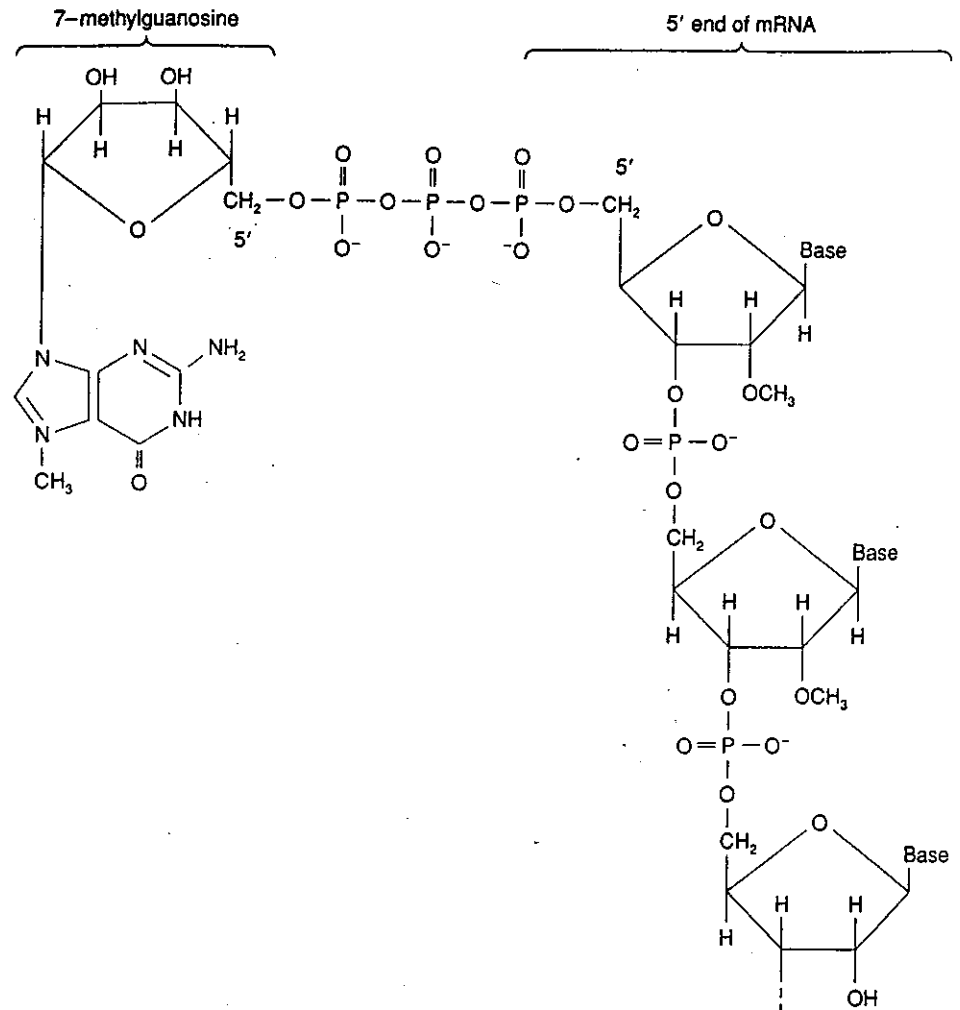


Figure 9-17 Structure of the cap at the 5' end of eukaryotic mRNA. Note the unusual 5' to 5' linkage. All caps contain 7-methylguanylate (shown in red) attached by a triphosphate linkage to the ribose of the 5' end of the mRNA. This cap was encountered earlier (Figure 8-10). The 2' hydroxyl group of the first mRNA ribose is also methylated while the second sugar is not always methylated.

instead of the usual 5' to 3' linkage. (Figure 9-17). The cap may protect the mRNA from exonucleases, but its main role is to promote and stimulate efficient translation. The 5' untranslated region at the cap end of the eukaryotic mRNA is folded into a secondary structure held together by base-pairing. This secondary structure inhibits efficient translation. Specific proteins, called cap-binding proteins, bind to the cap and unwind the secondary structure of the 5' end of the mRNA. This facilitates efficient translation. These proteins are also involved in guiding the 40S eukaryotic ribosomal subunit to the cap, and thus to the 5' end of the mRNA. Chapters 8 and 13 contain further information on mRNA modification.