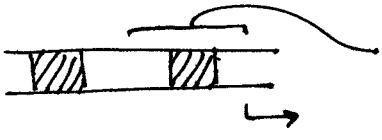
BE.440

22 September 2004

Essigmann

Last Day: Transcription (Front End)

a) promoter architecture
b) how pol. finds promoter
c) elongation of transcript
d) termination
e) specialized endings (euk. cells) { capping, splicing, poly A }
f) translation

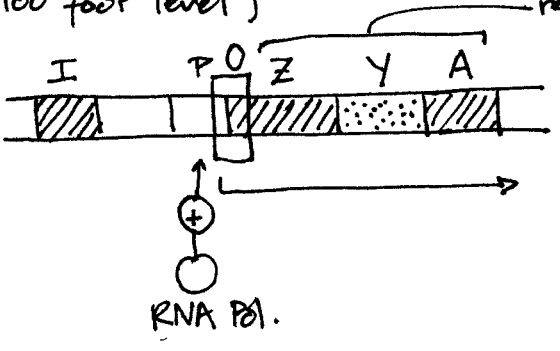


emphasized fact that these segments cause 10^3 X variation in constitutive expression.

Effectors: other way to regulate. Good for responding to changes in the environment.

Paradigm of ⊕ and ⊖ Regulation: Lac operon

(100 foot level)



Lac operon → β-gal

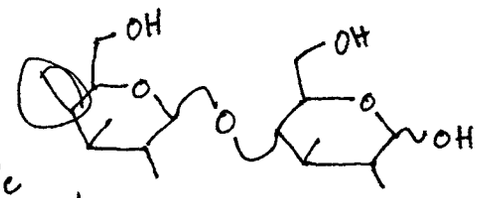
lactose

Gal ⟷ Glc → Glycol → TCA → En. → Growth

Gal epimerase

Chemistry:

Lactose
Gal (β1→+)Glc



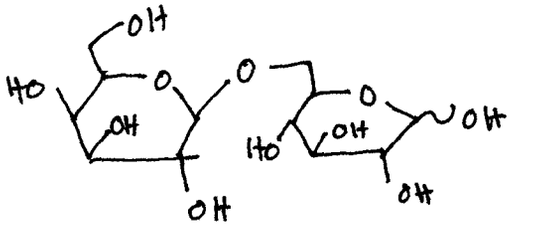
β-gal → Gal + Glc

epim.

β-gal

(tiny amount) → allolactose Gal (β1→6)Glc

for later



non metabolizable
potent inactivator
repressor

Consider Two Scenarios:

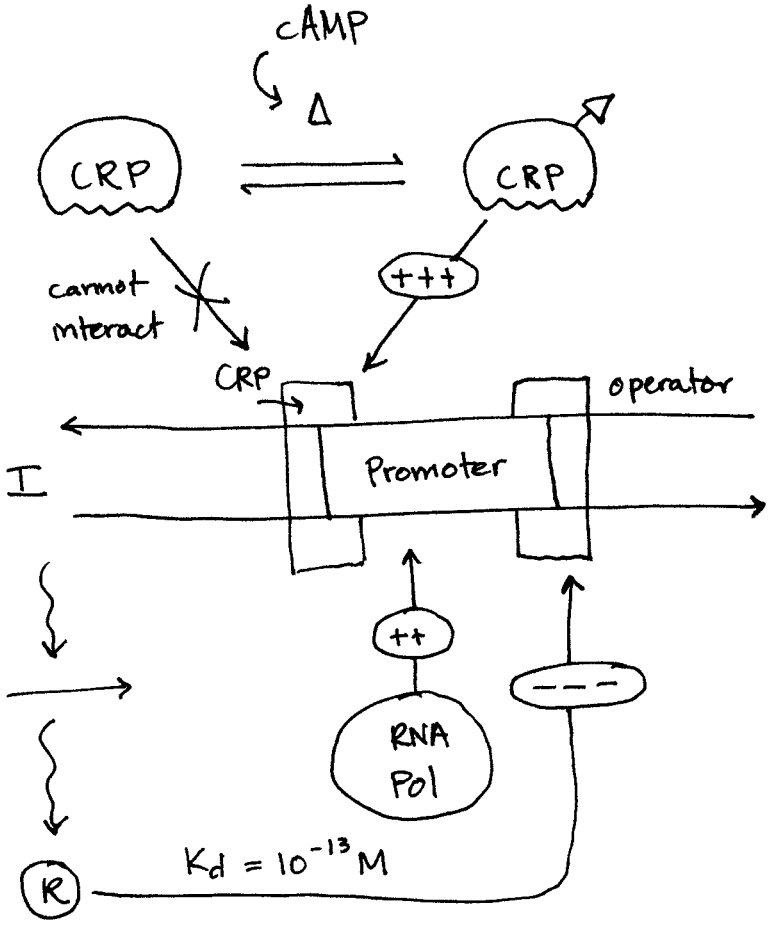
1 lactose → E. coli

- genes for lac utilization (βgal, Lac A, Lac Y) turned on

2 lac + Glc → E. coli

- cell uses all Glc
- then turns on genes for Lac utilization
- ↑ Glc → inhibits adenyl cyclase
  ⟹ ↓ cAMP

Promoter / operator region (100 mm level)



(3) Positive Regulation — binding of CRP = needed to ensure that cell will use glc first if available

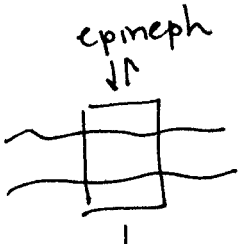
(1) Draw at interface of 2 long B' bonds

(2) Negative Regulation

• = allolact.

R• } cannot bind DNA

Note:



activation
adenyl cyclone → [↑] cAMP
(glucose)n
glucose

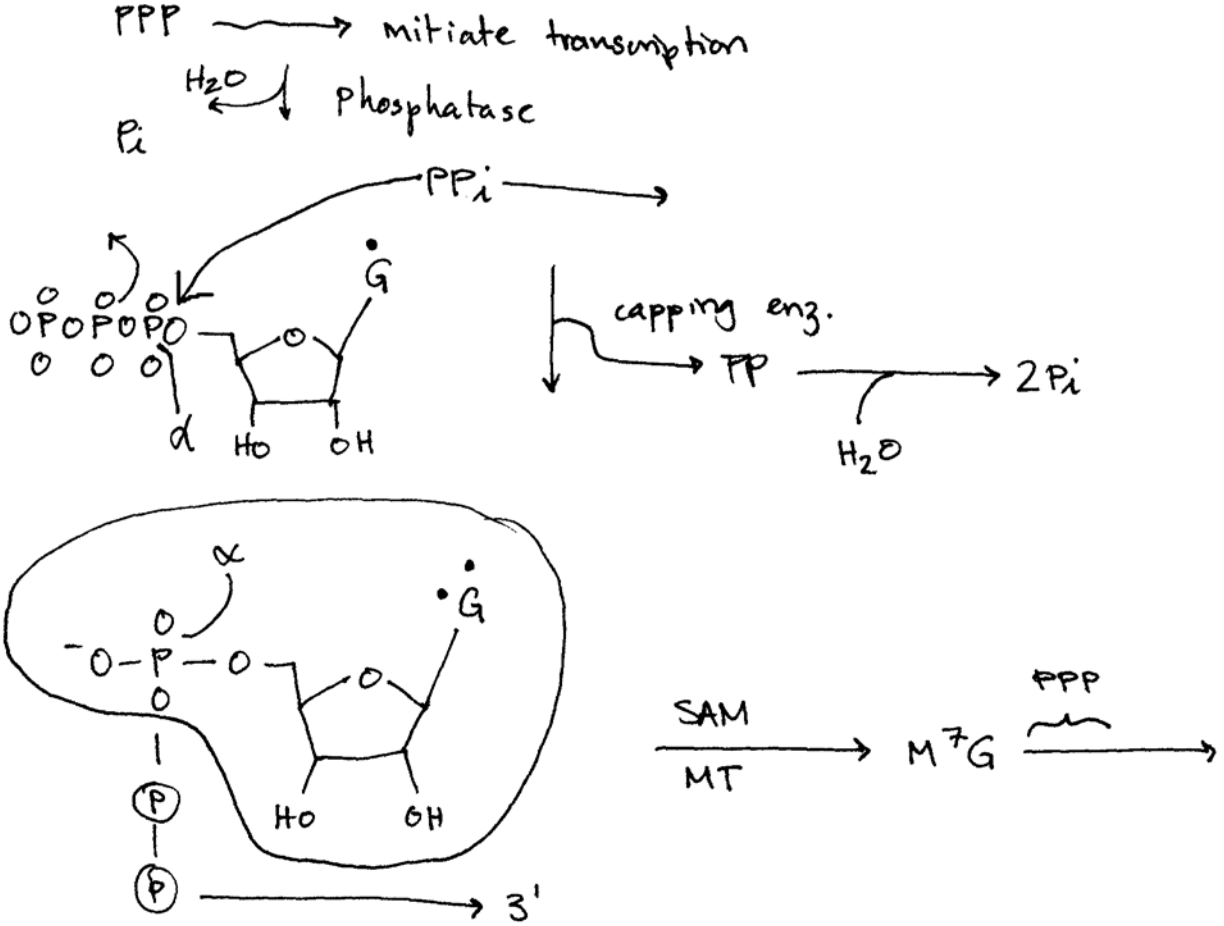
Cell is doing same thing here, preserving polymeric glucose if

[↓] cAMP [↑] glucose

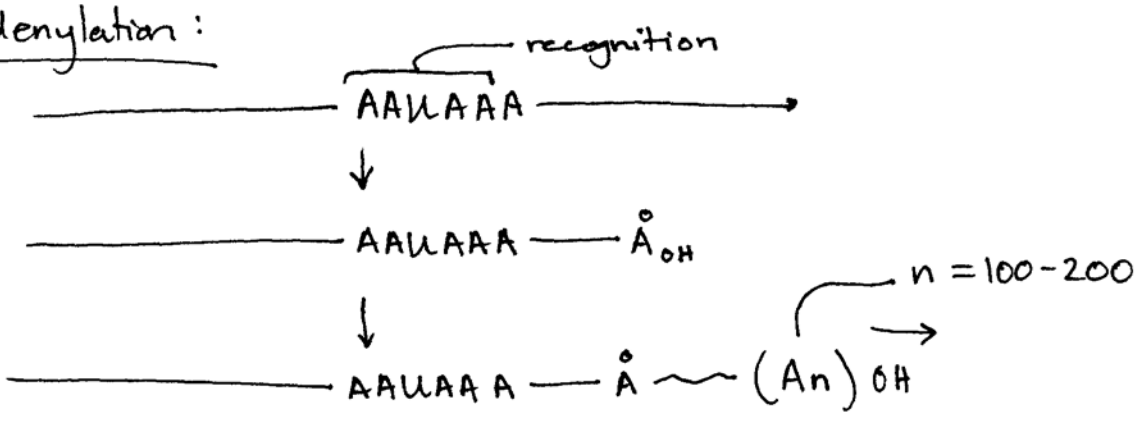
Optional Endings for Transcription (eukaryotes)

1. 5' PPP is "capped" by 7 MeG (from SAM)
2. 3' end polyAdenylated
3. Introns spliced out as lariat structures

1. Capping:

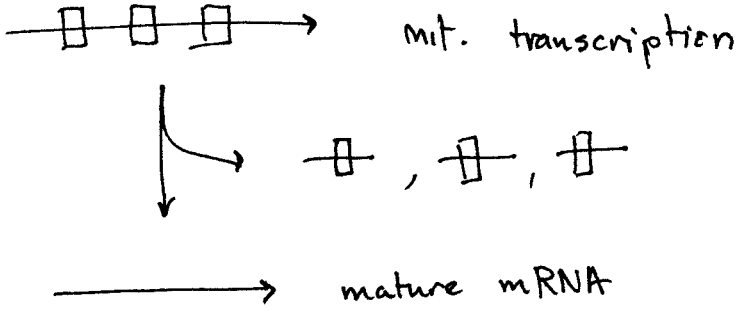


2. PolyAdenylation:

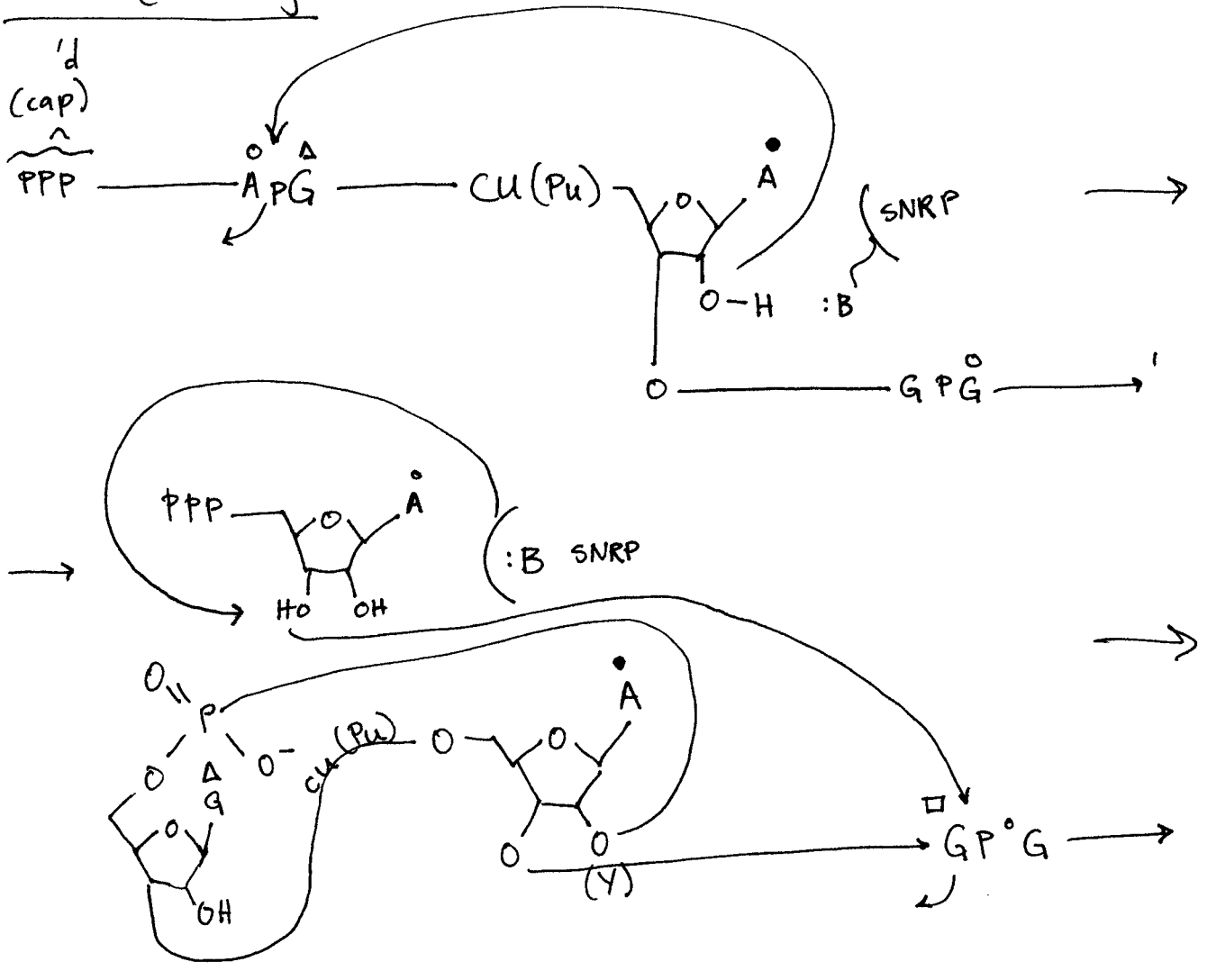


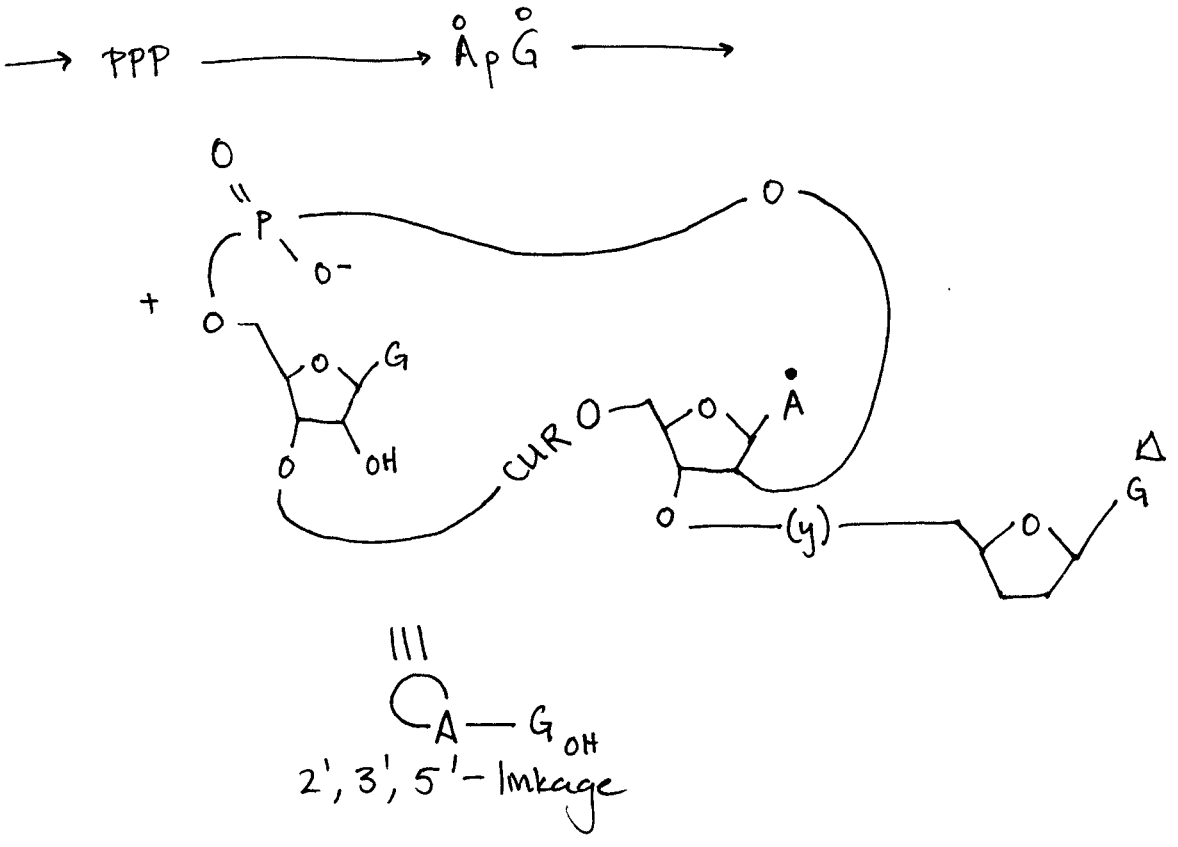
3. Splicing : occurs on SNRPs (spliceosome)

overview :



details (chemistry) :

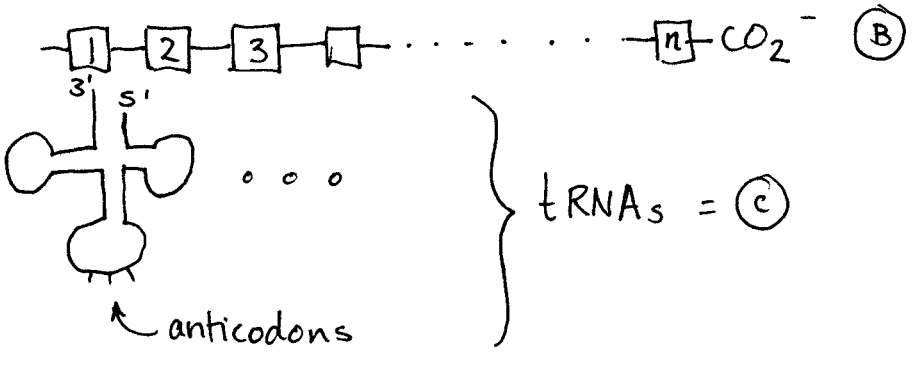


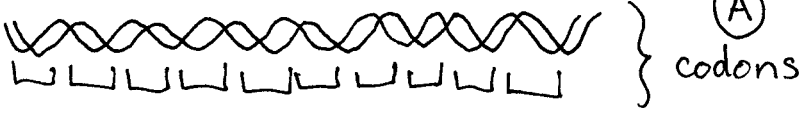


Translation. mRNA-div'd PS (opposed to non-ribosomal PS)

PROBLEM: How does lin. code in DNA (A) get "translated" into lin. segments of amino acids in protein? (B)

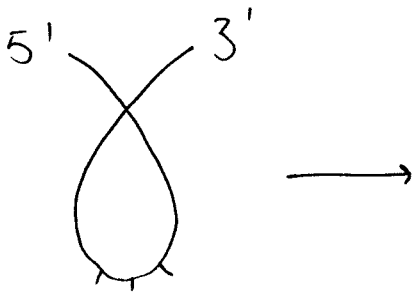
ANSWER: Translation, (C) using tRNA as the adaptor or connector.





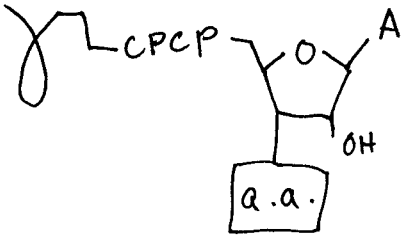
POINTS:

1. DNA 5' → 3'
2. transcription 5' → 3'
3. translation H → C
   by recoding mRNA 5' → 3'



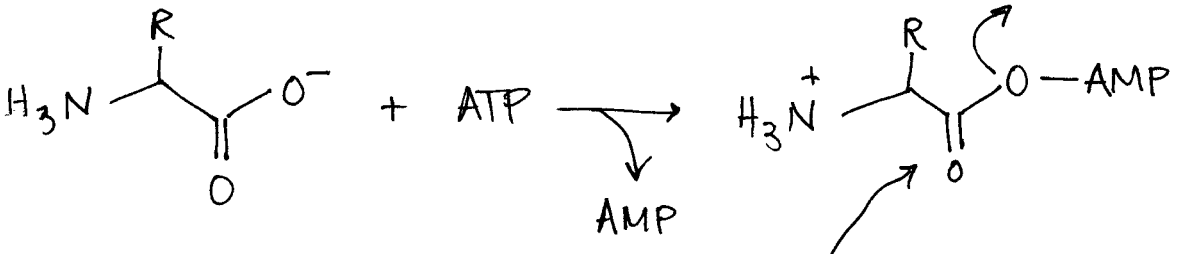
1. Attachment of amino acids to tRNA

tRNA Synthetases

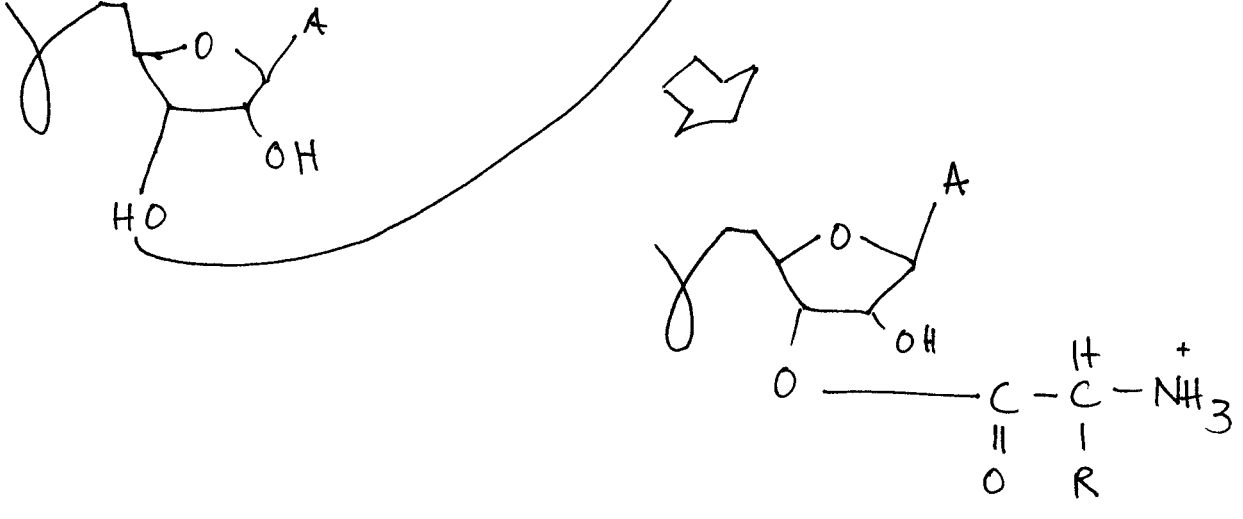


tRNA synthetases (2 steps)

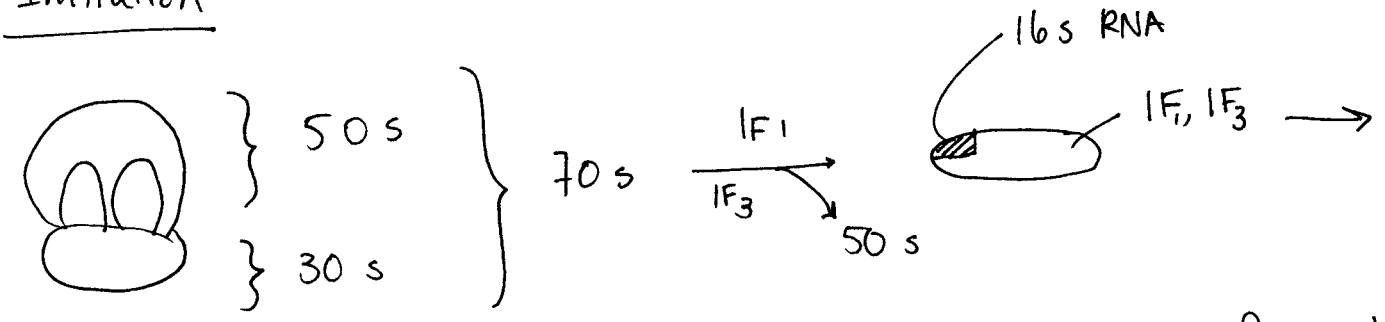
1. amino acid

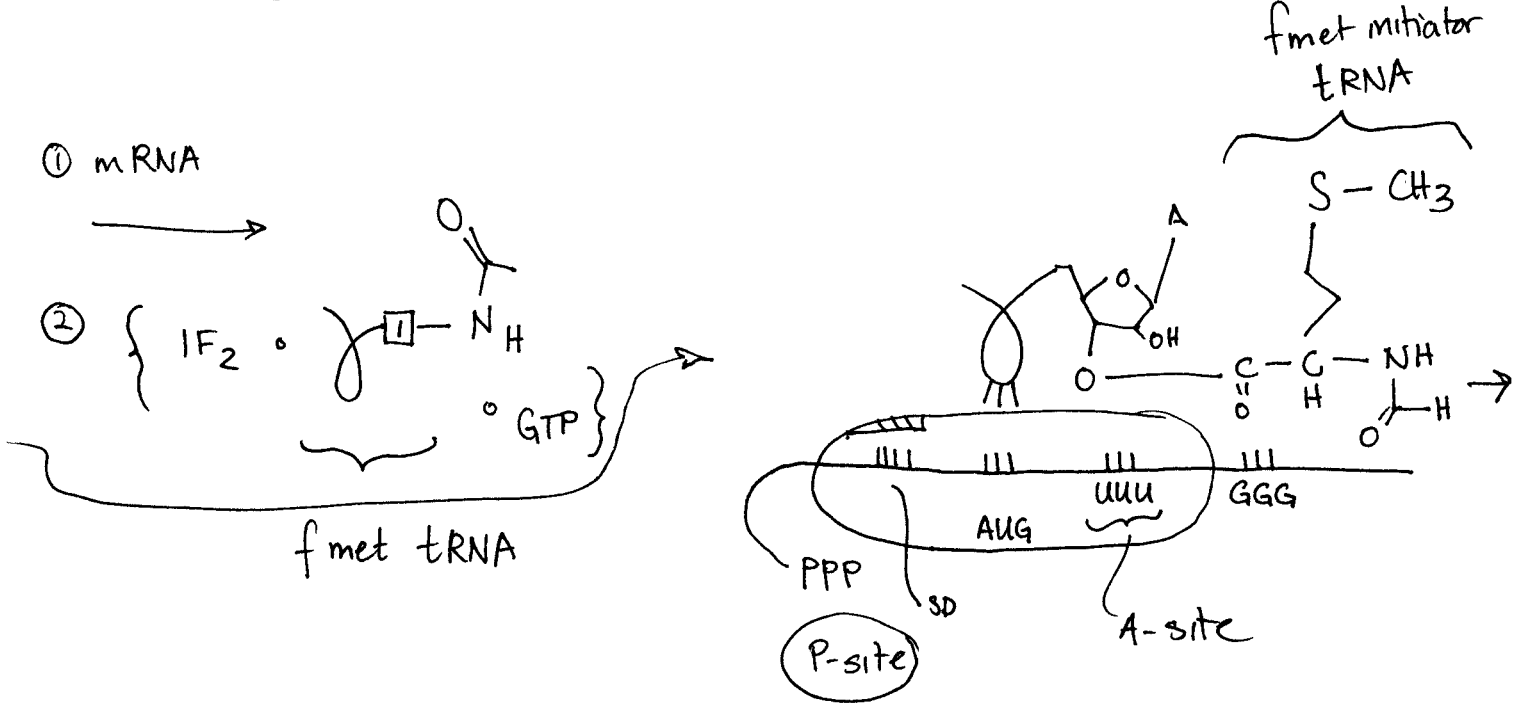


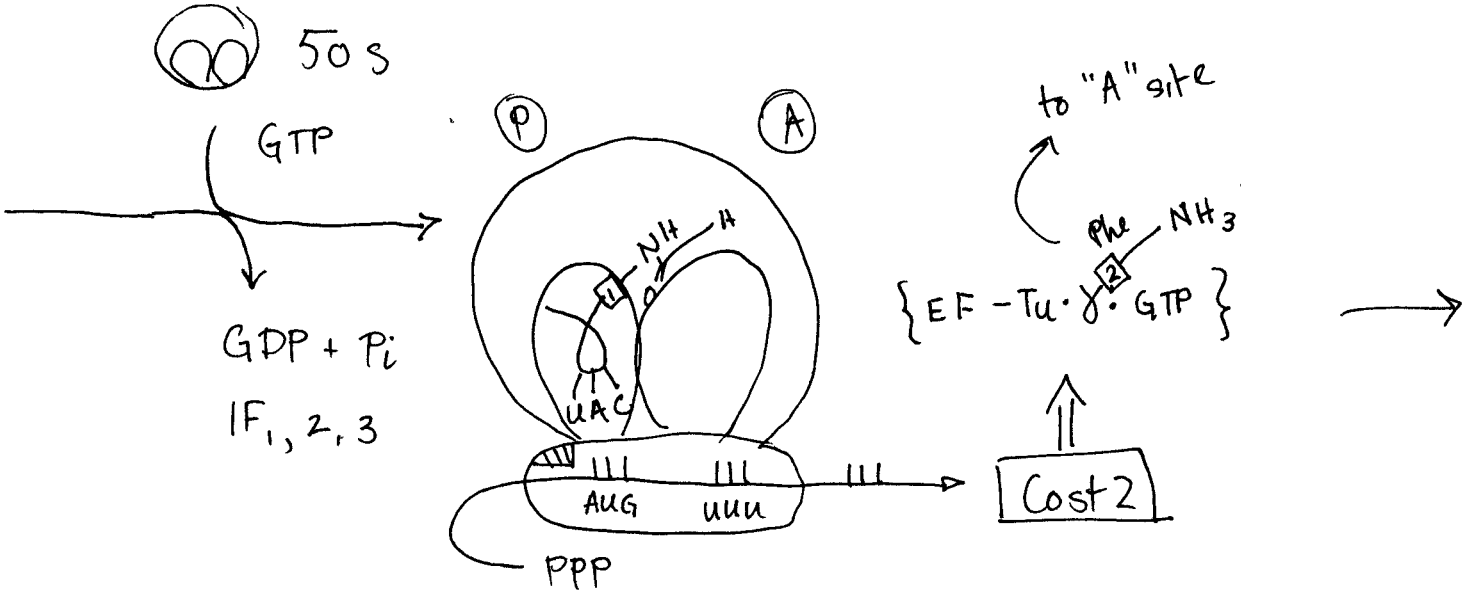
2. charging

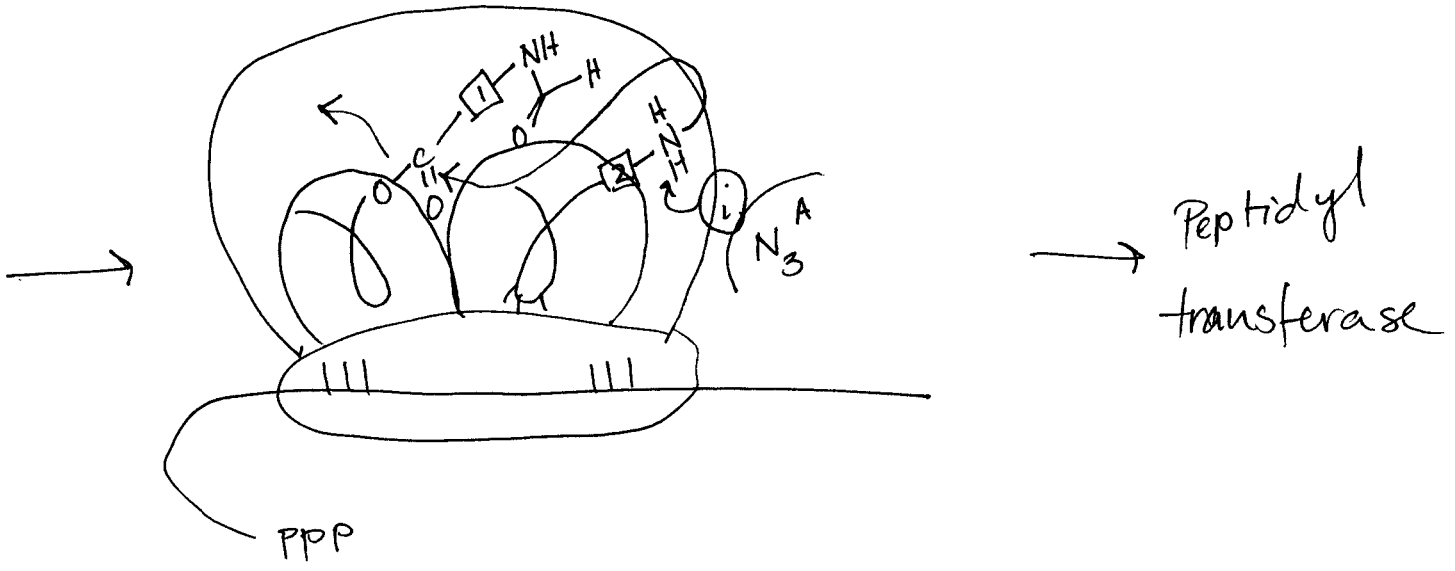


2. Initiation








NH
H
1
C
O
O
O
H
N
H
2
i
A
N_3
PPP
Peptidyl
transferase