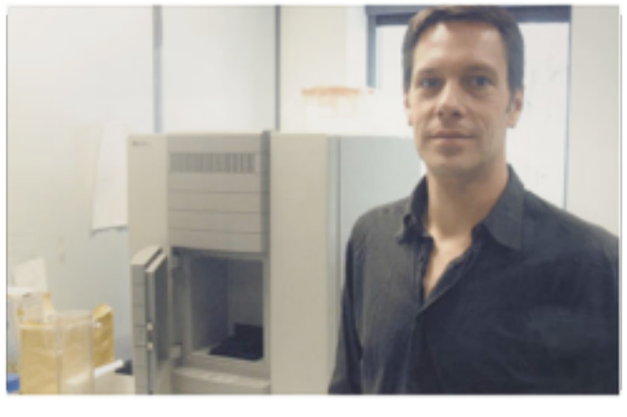
Epigenetics, aging and symmetry

or why DNA is not a program...

Jean Krivine (CNRS & Université Paris VII)

ENS - Winter school
ER06 2011

Joint work



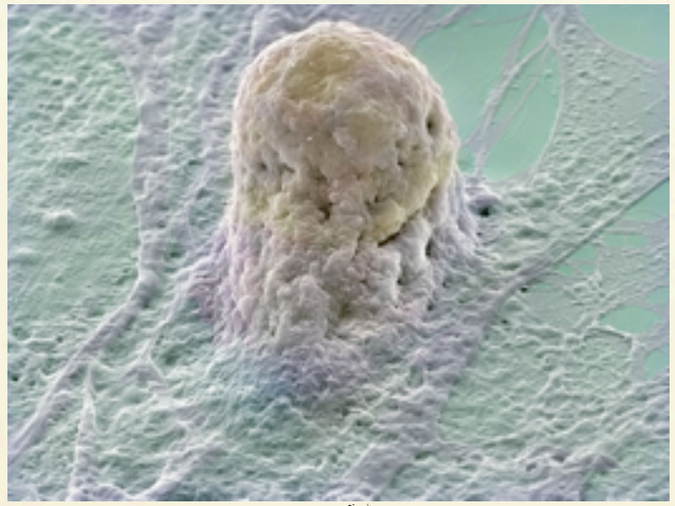
Arndt Benecke

Nicolas Tchitchek

IHÉS
Institut des Hautes Études Scientifiques

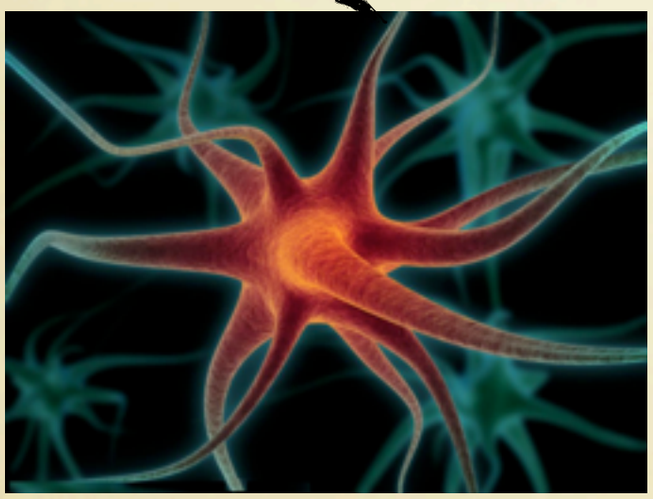
Paris
Centre de recherche
Cardiovasculaire
PARCC
H E G P

Cell differentiation



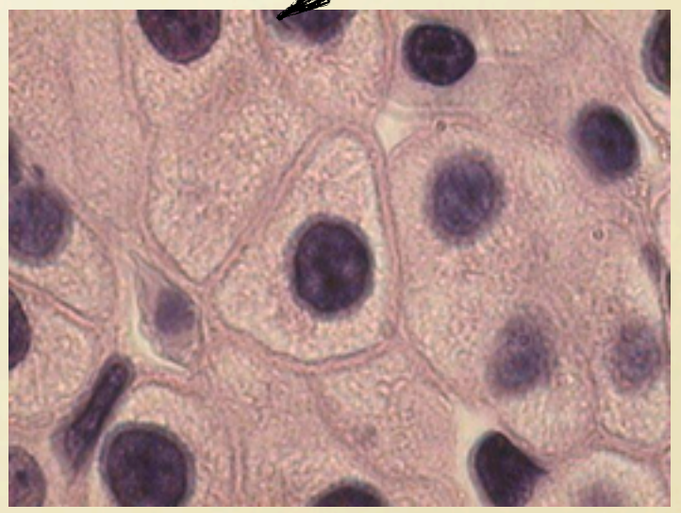
A stem cell ...

Neurons

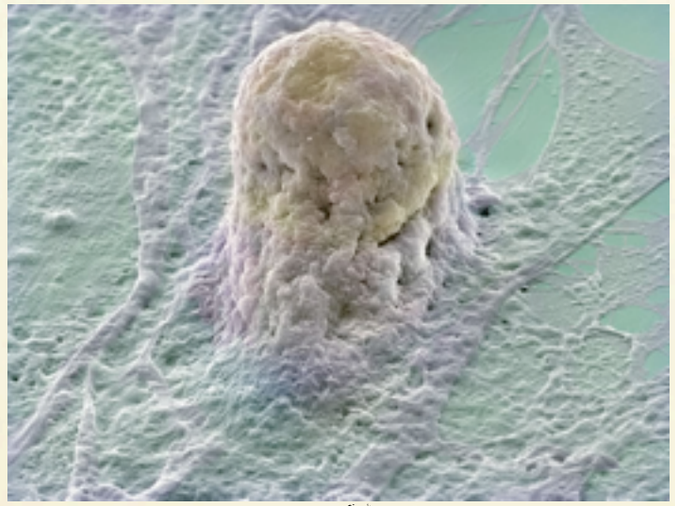


Red blood cells

Liver



Cell differentiation



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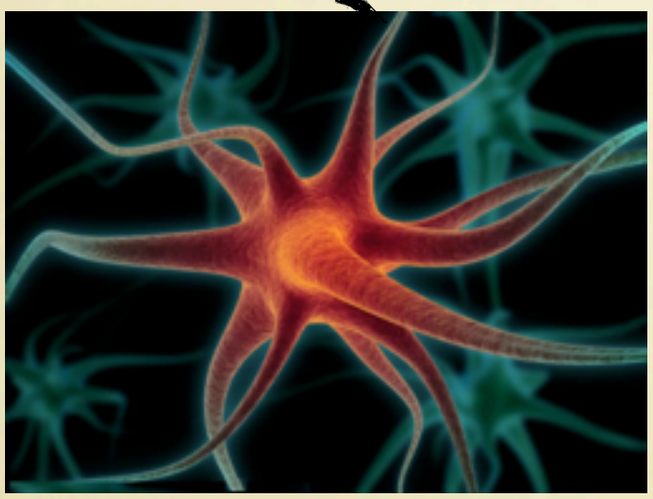
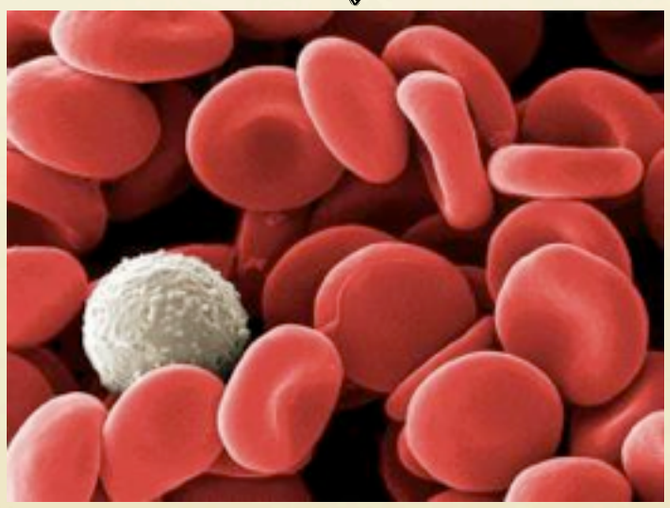
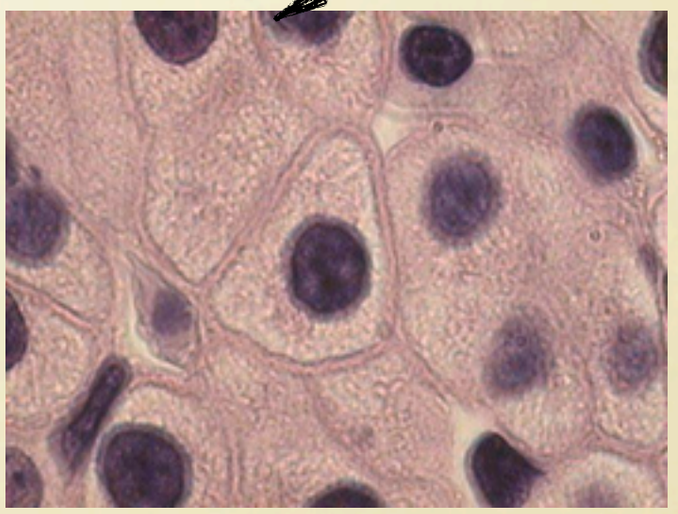


To keep thinking...

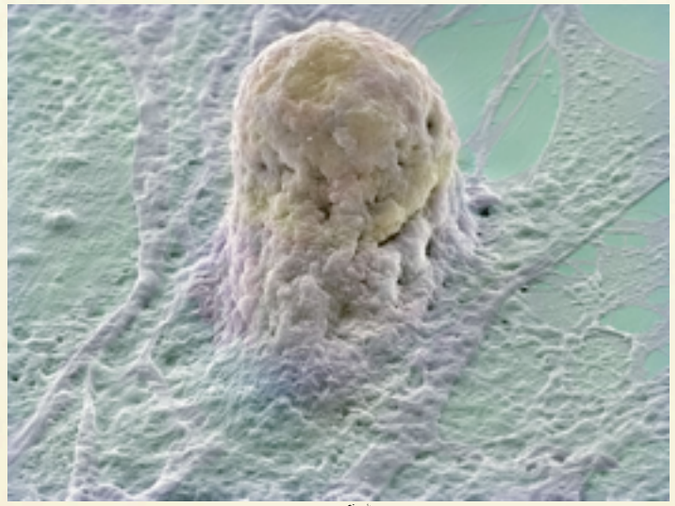
Red blood cells



Liver

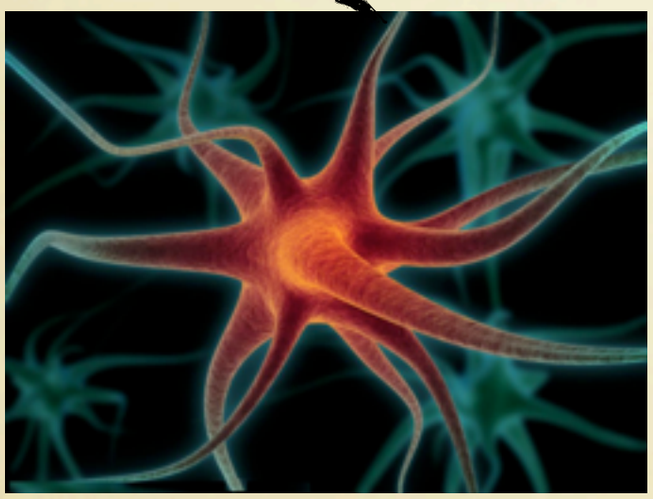


Cell differentiation



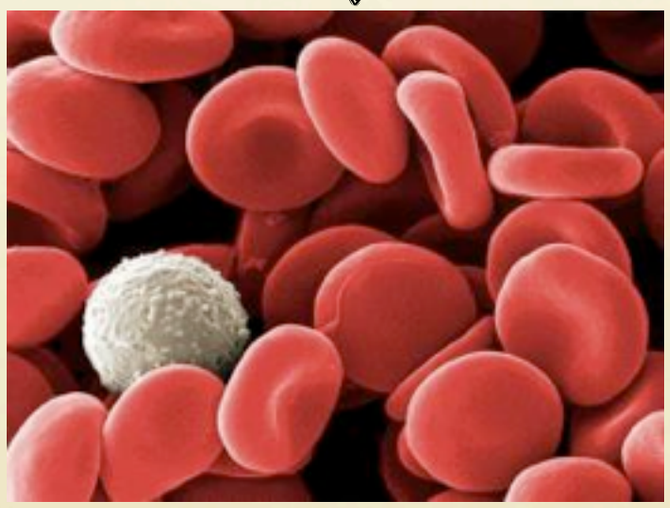
A stem cell ...

Neurons



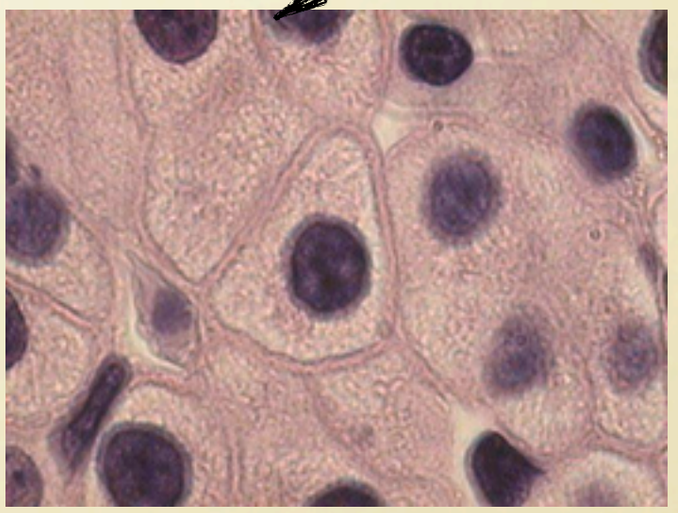
To keep thinking...

Red blood cells

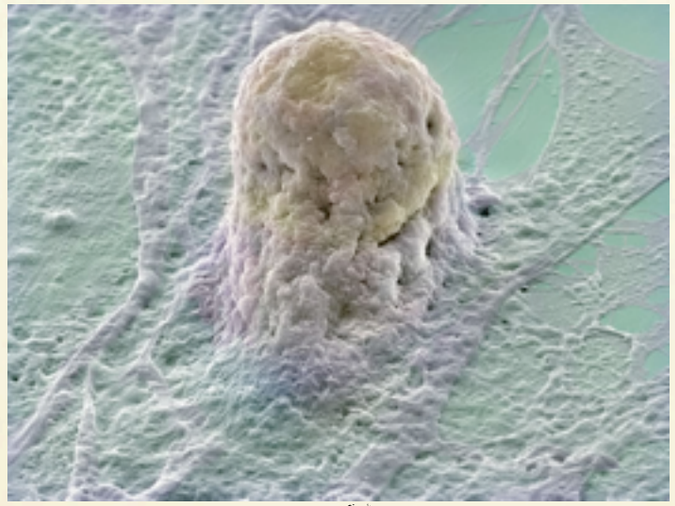


To keep breathing...

Liver

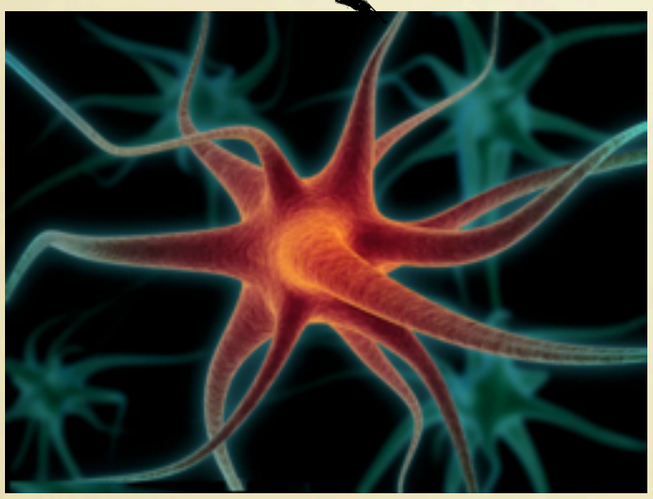


Cell differentiation



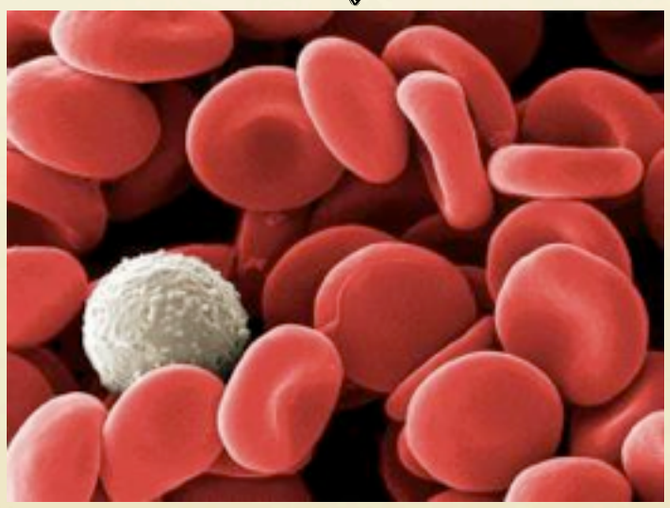
A stem cell ...

Neurons



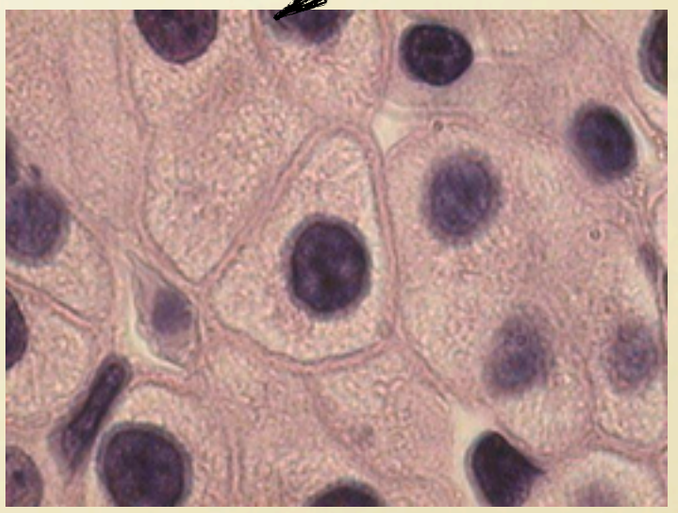
To keep thinking...

Red blood cells



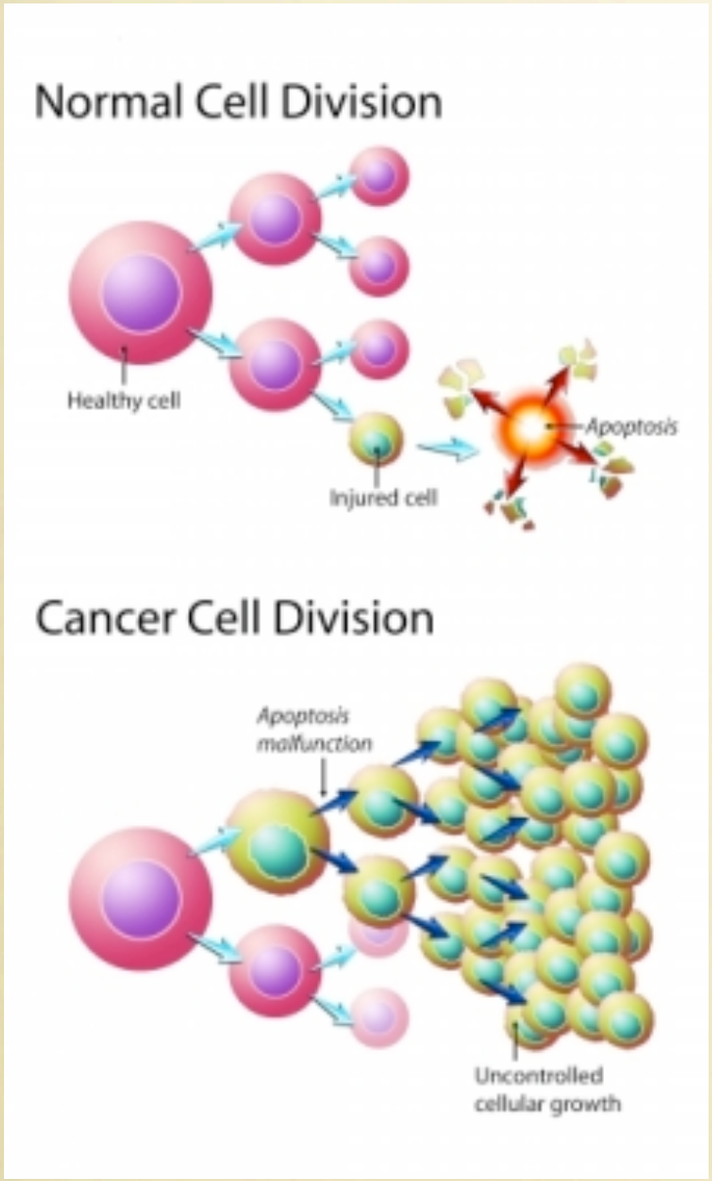
To keep breathing...

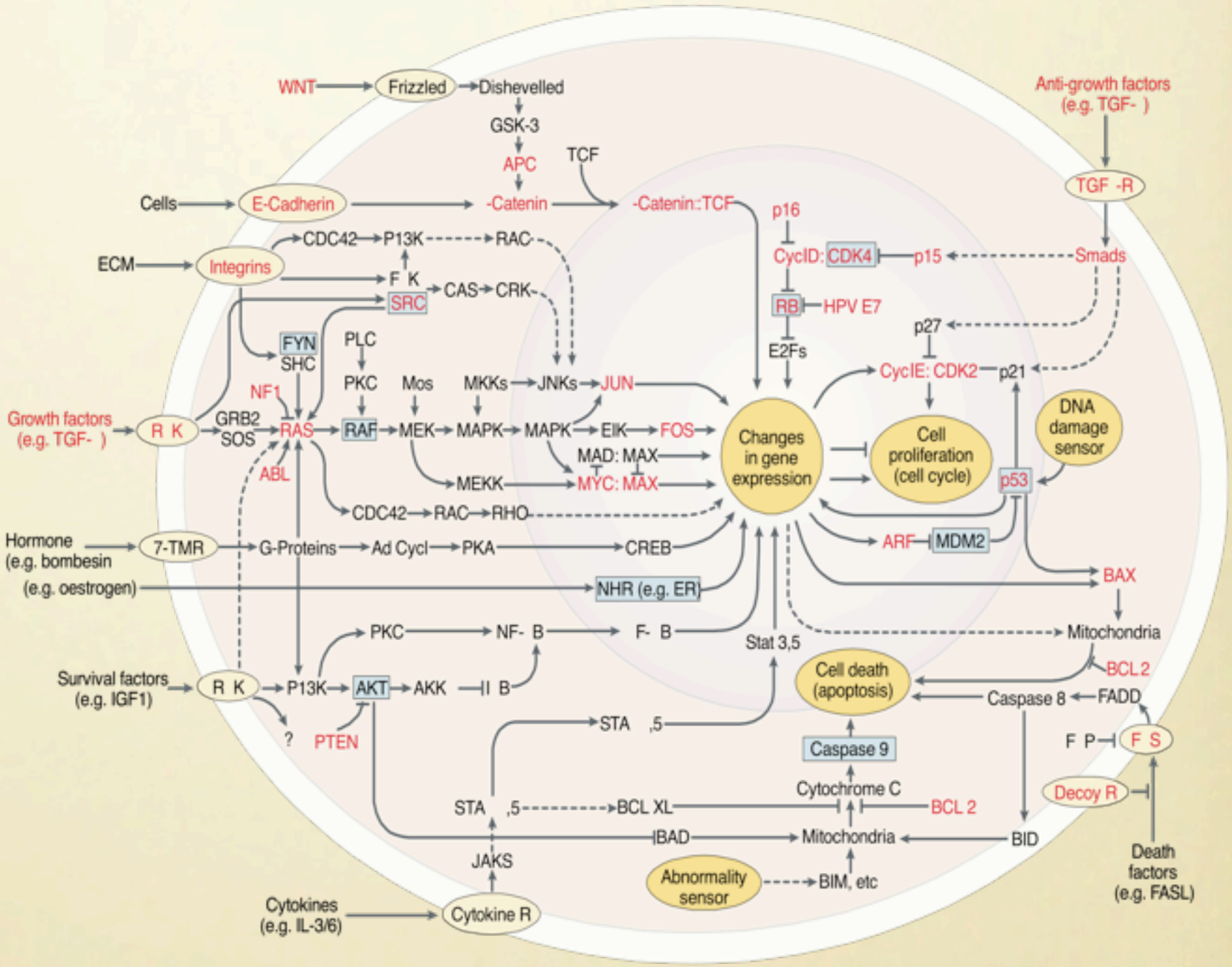
Liver



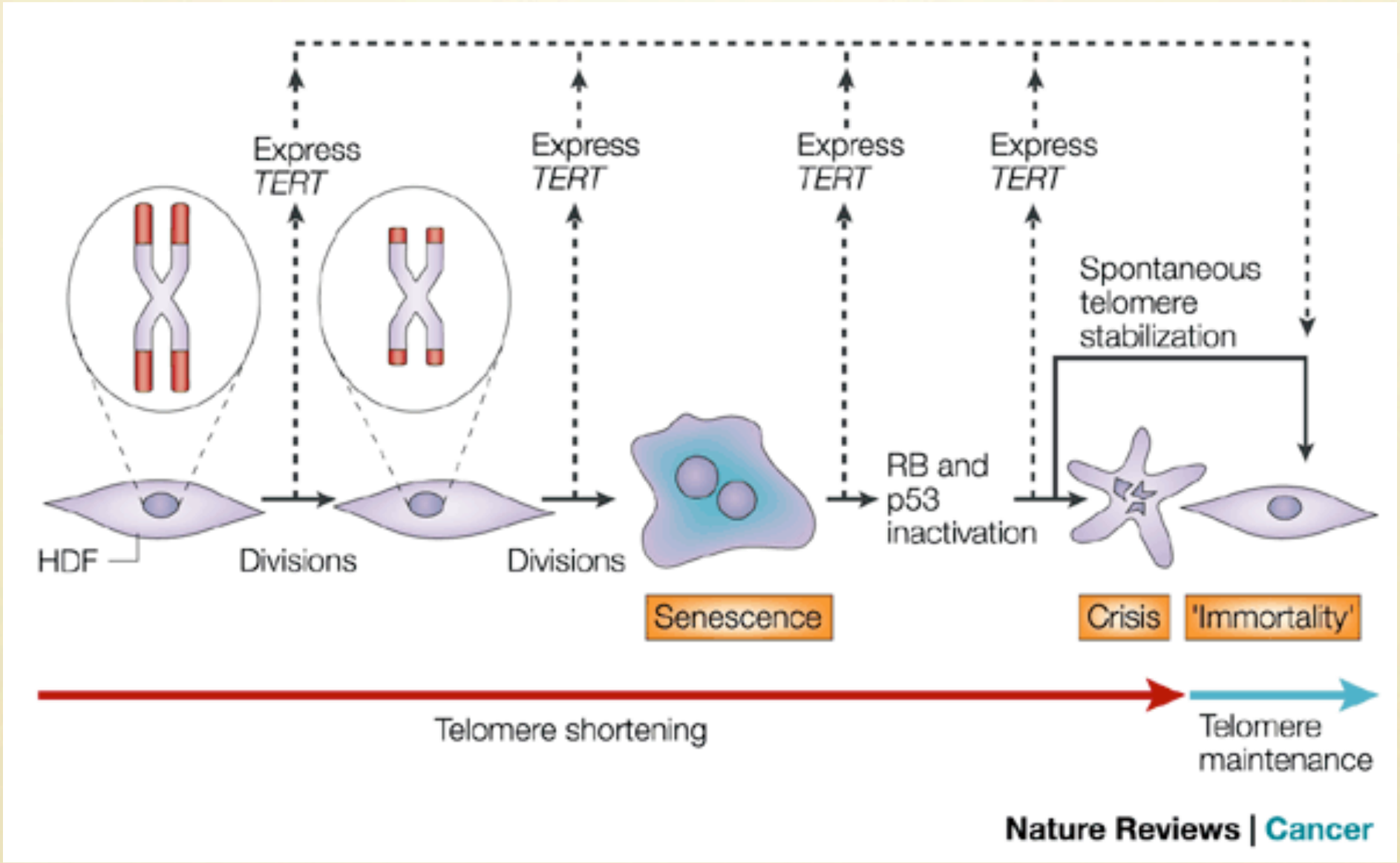
To keep drinking...

Cellular division

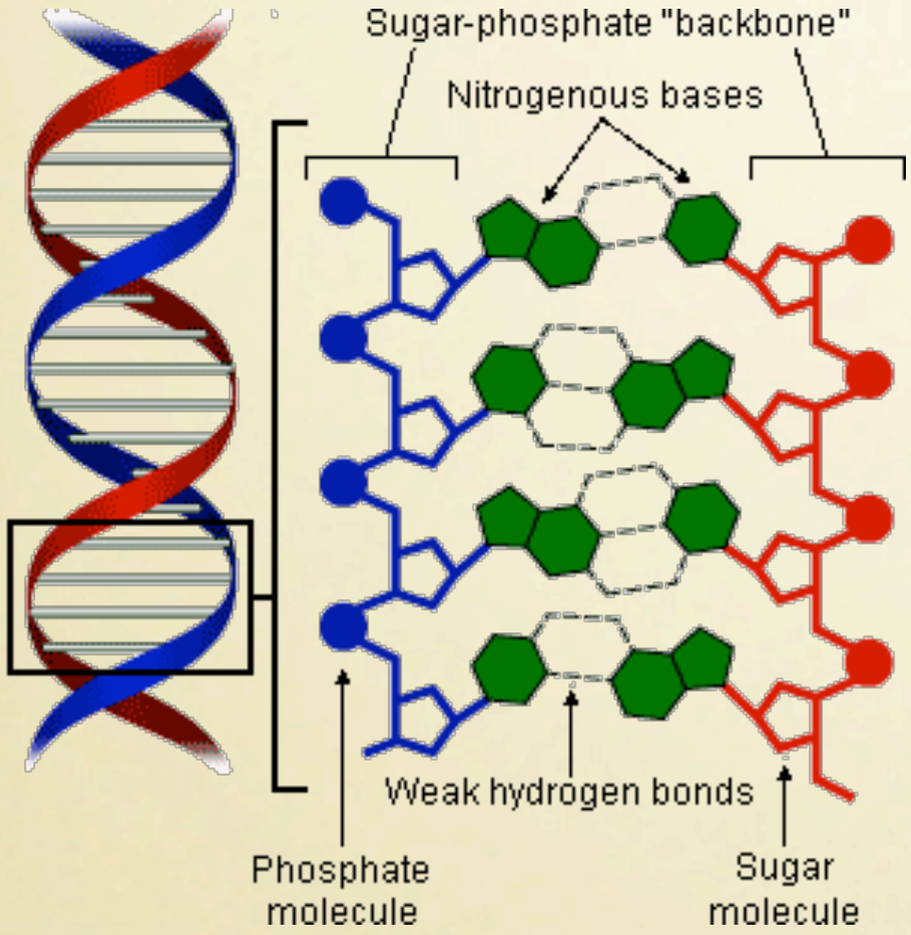




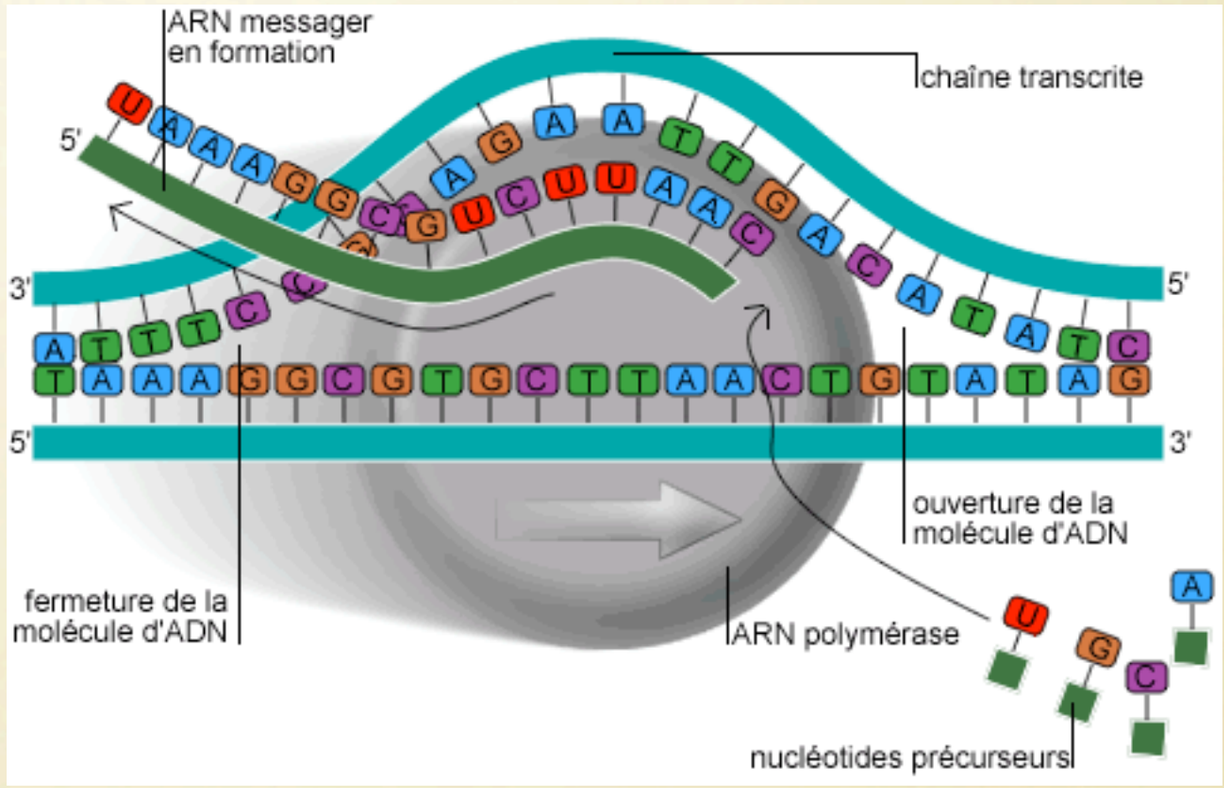
Senescence


Express
TERT
Express
TERT
Express
TERT
Express
TERT
Spontaneous
telomere
stabilization
RB and
p53
inactivation
HDF
Divisions
Divisions
Senescence
Crisis
'Immortality'
Telomere shortening
Telomere
maintenance
Nature Reviews | Cancer

Basics: DNA structure



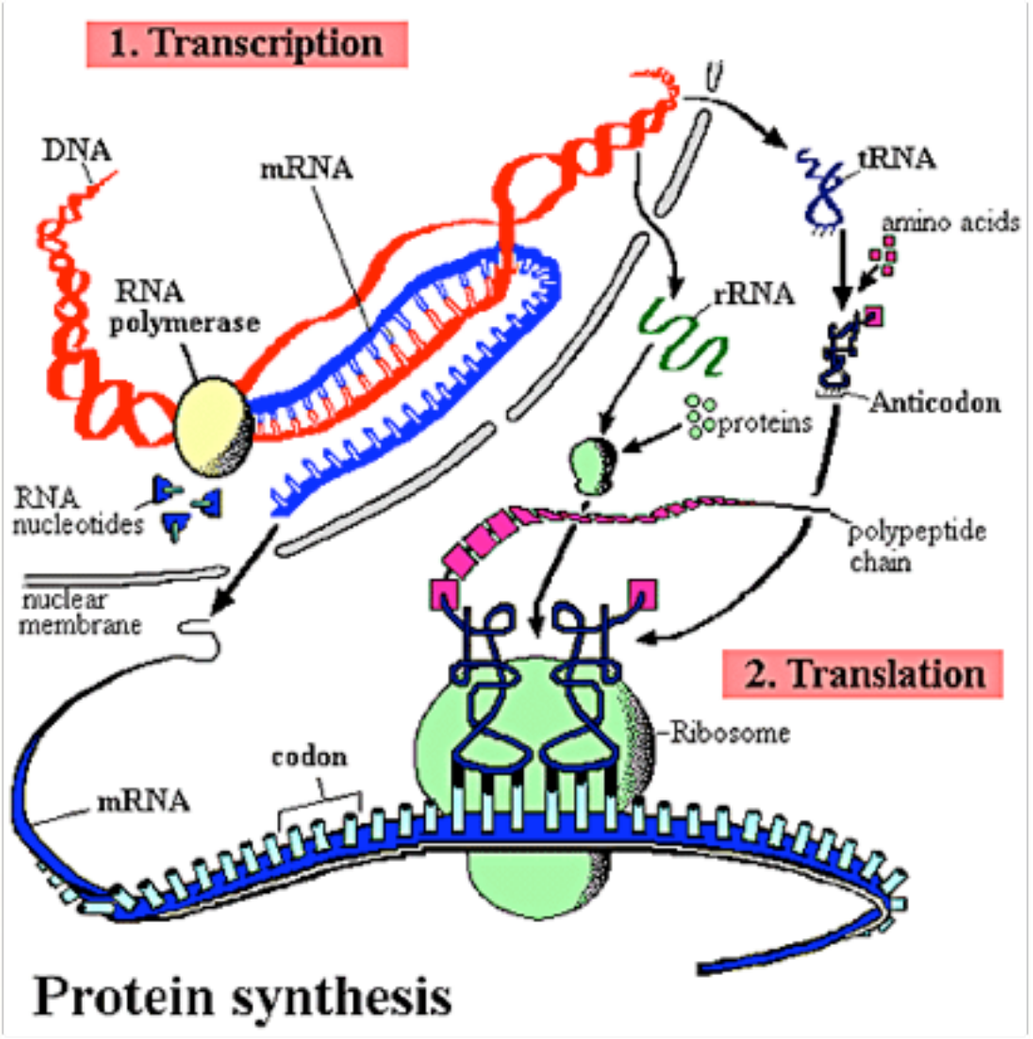
transcribed vs. non transcribed sequences…



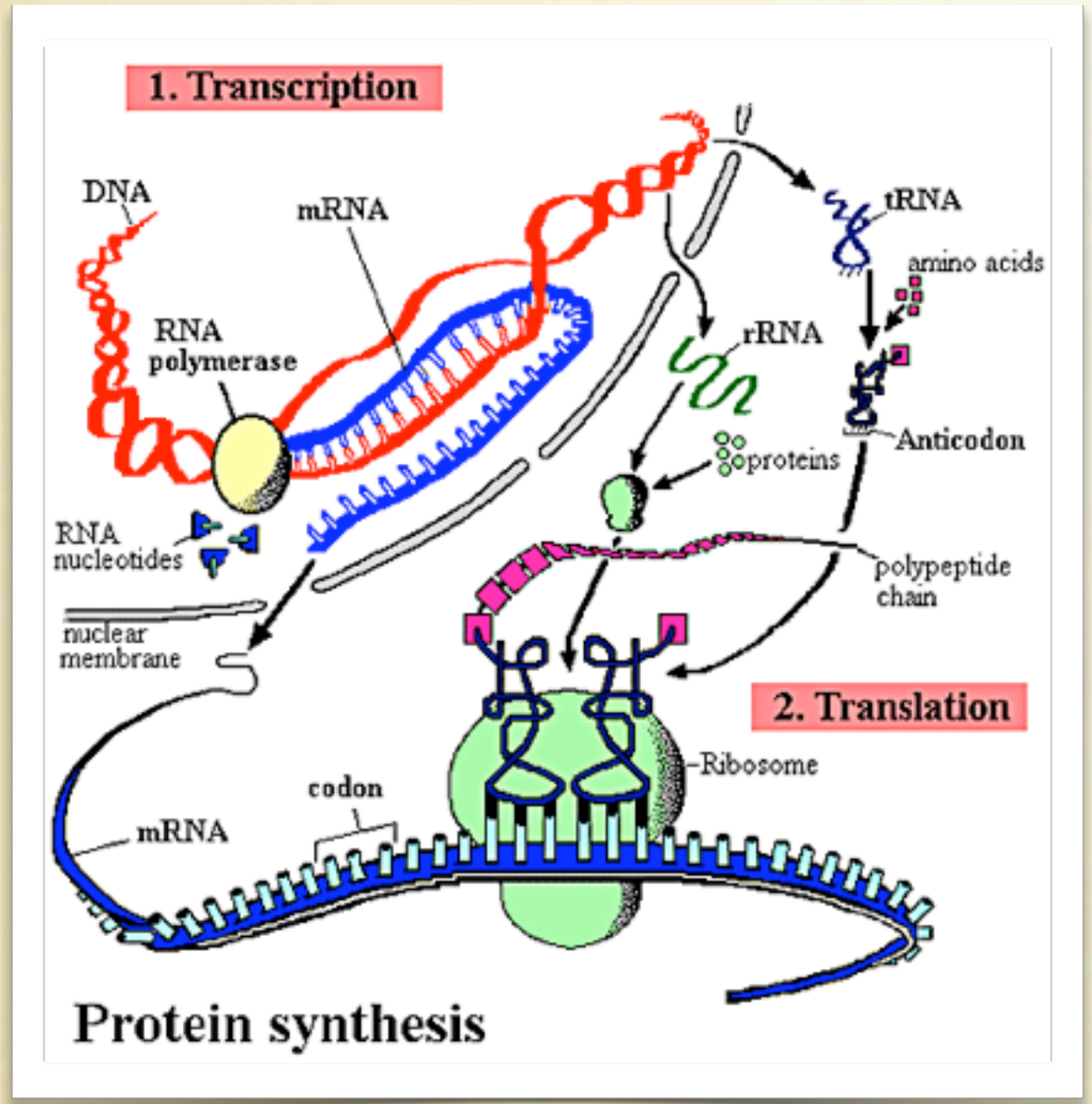
Coding vs. non coding RNA…

Proteins

Proteins


1. Transcription
DNA
mRNA
RNA polymerase
RNA nucleotides
nuclear membrane
tRNA
amino acids
rRNA
proteins
Anticodon
polypeptide chain
2. Translation
Ribosome
codon
mRNA
Protein synthesis

Proteins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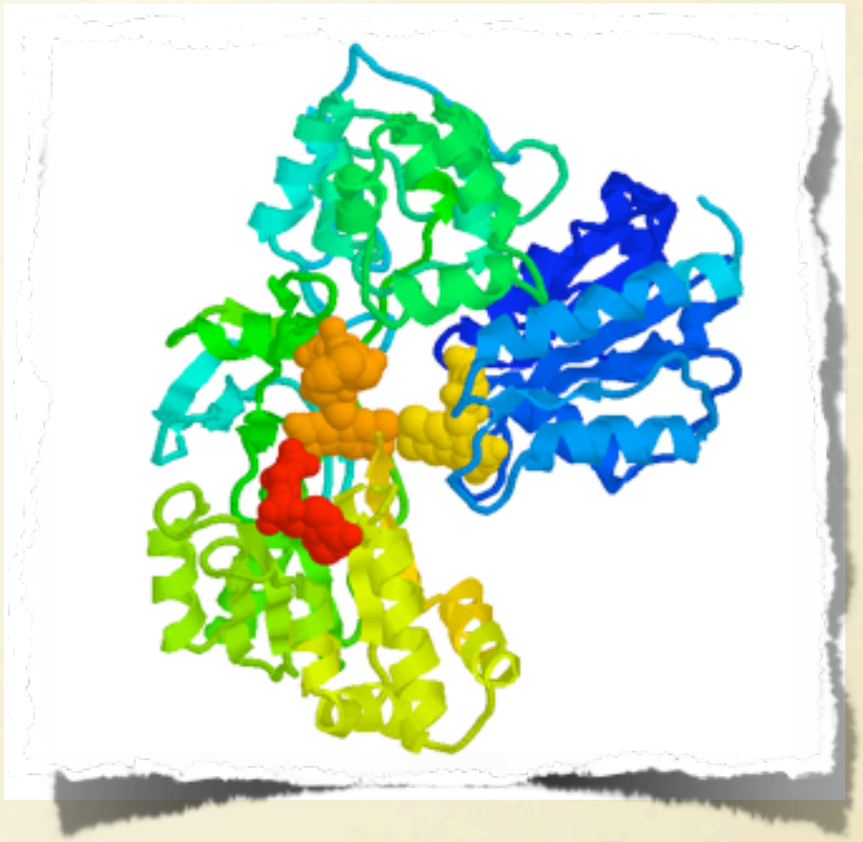
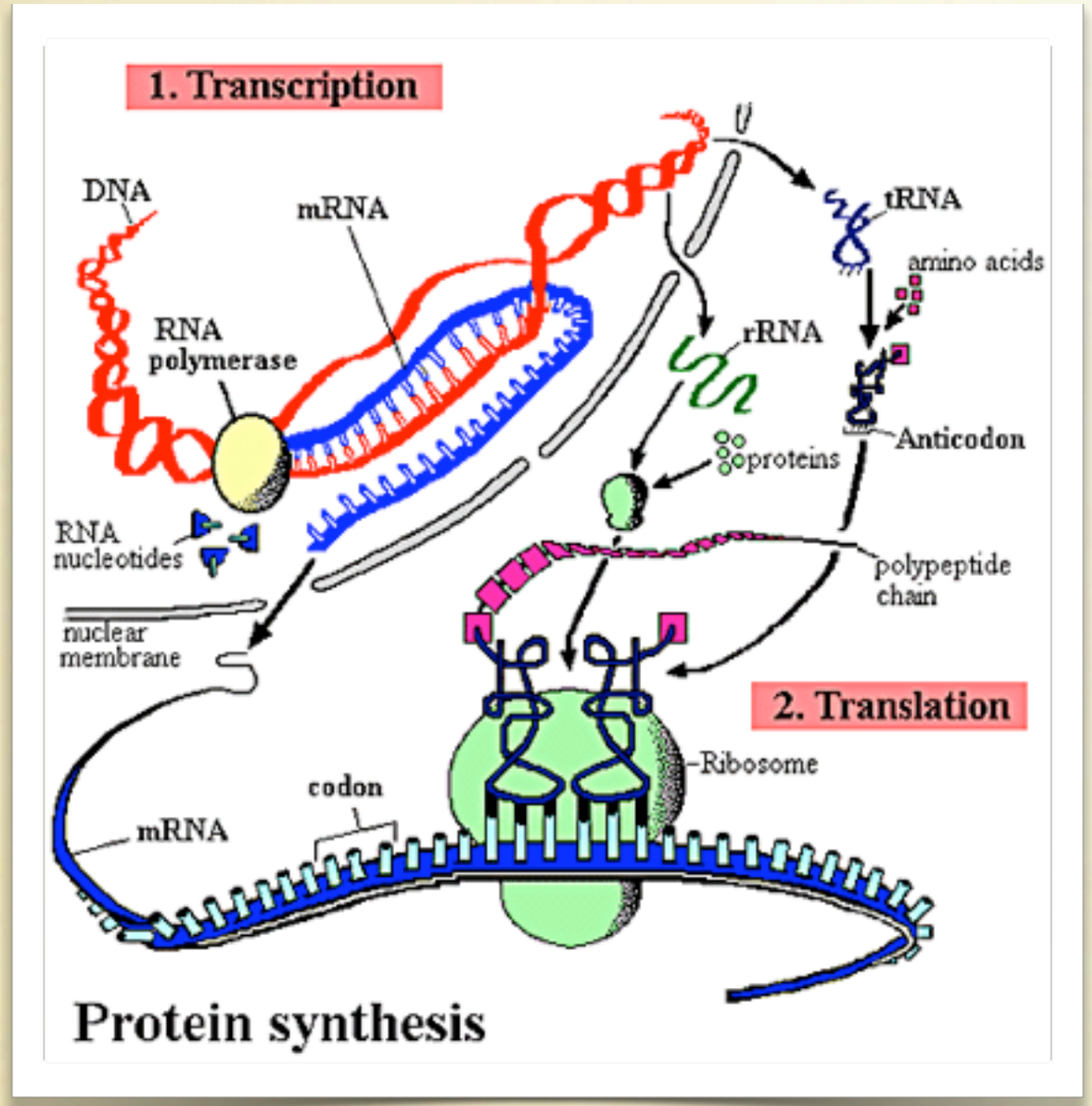


Proteins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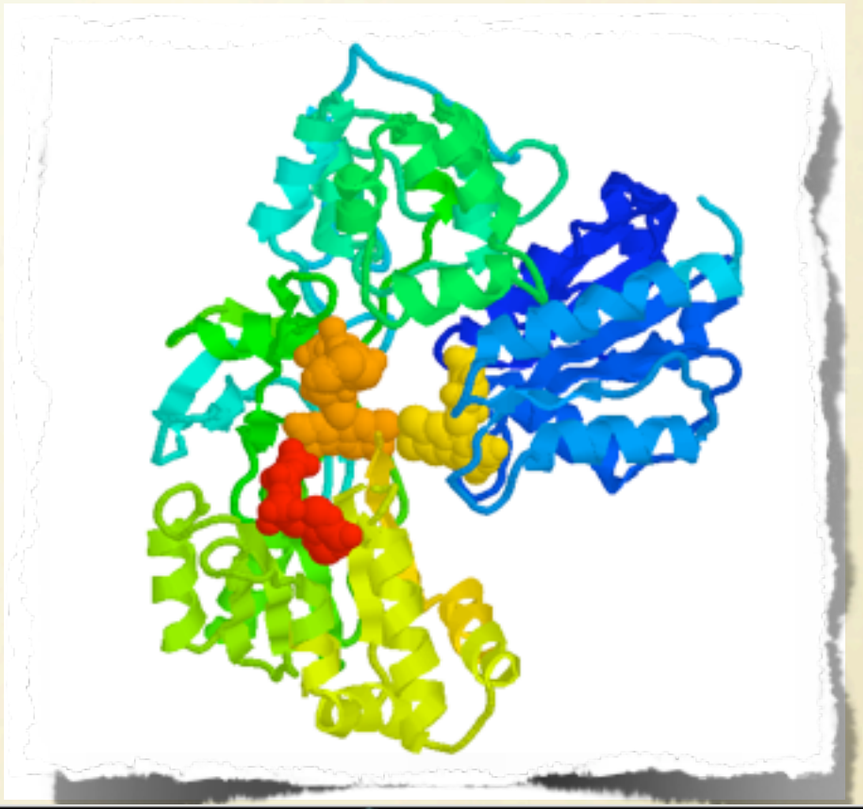
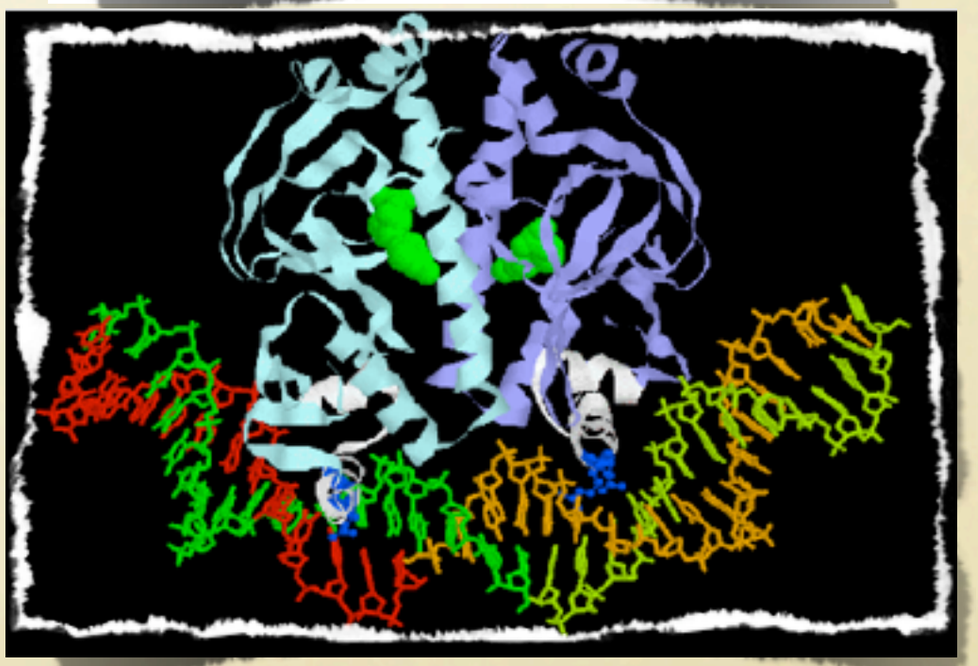
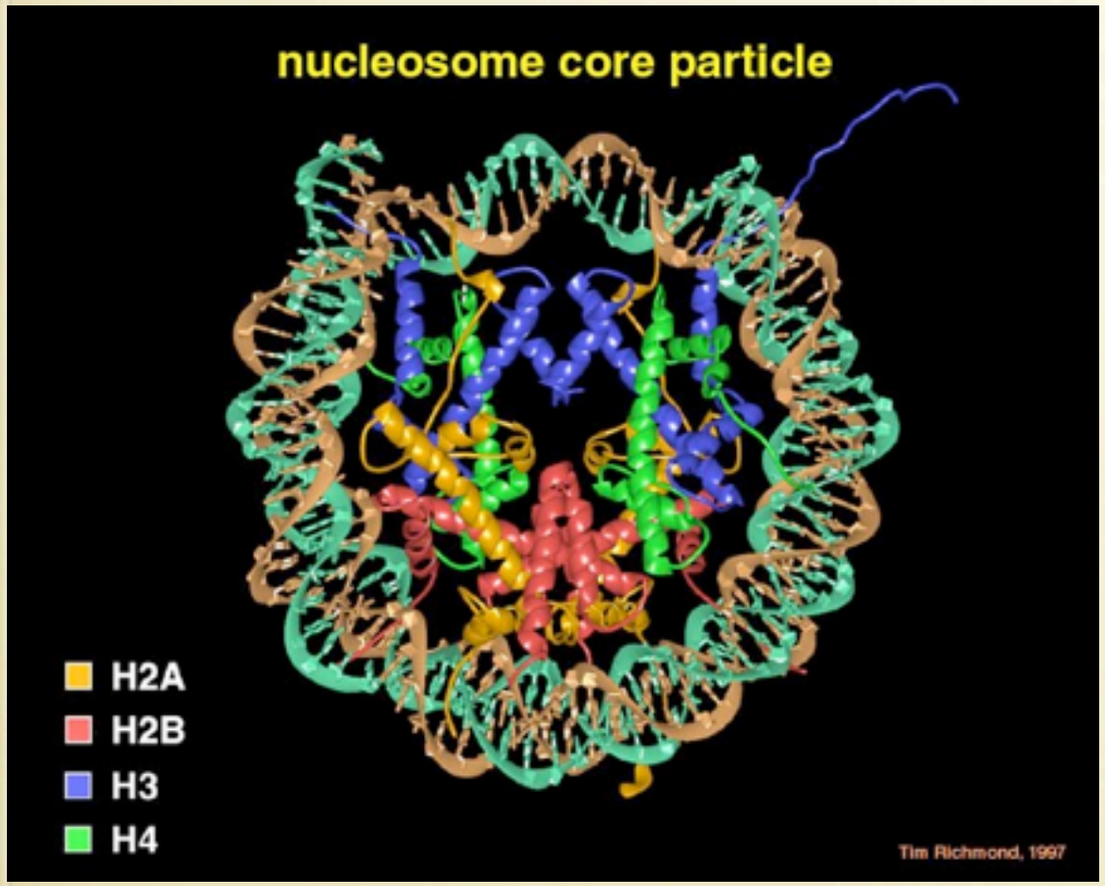
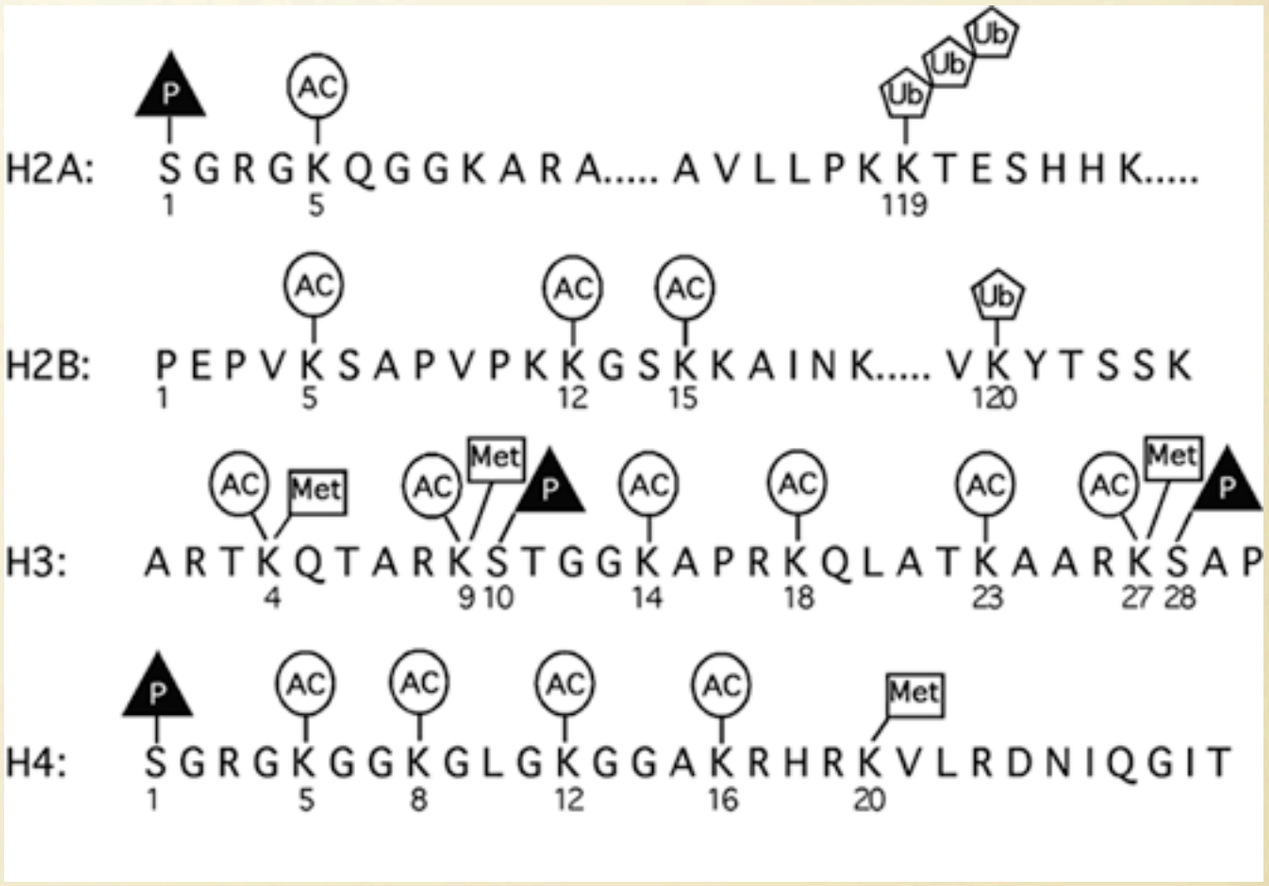


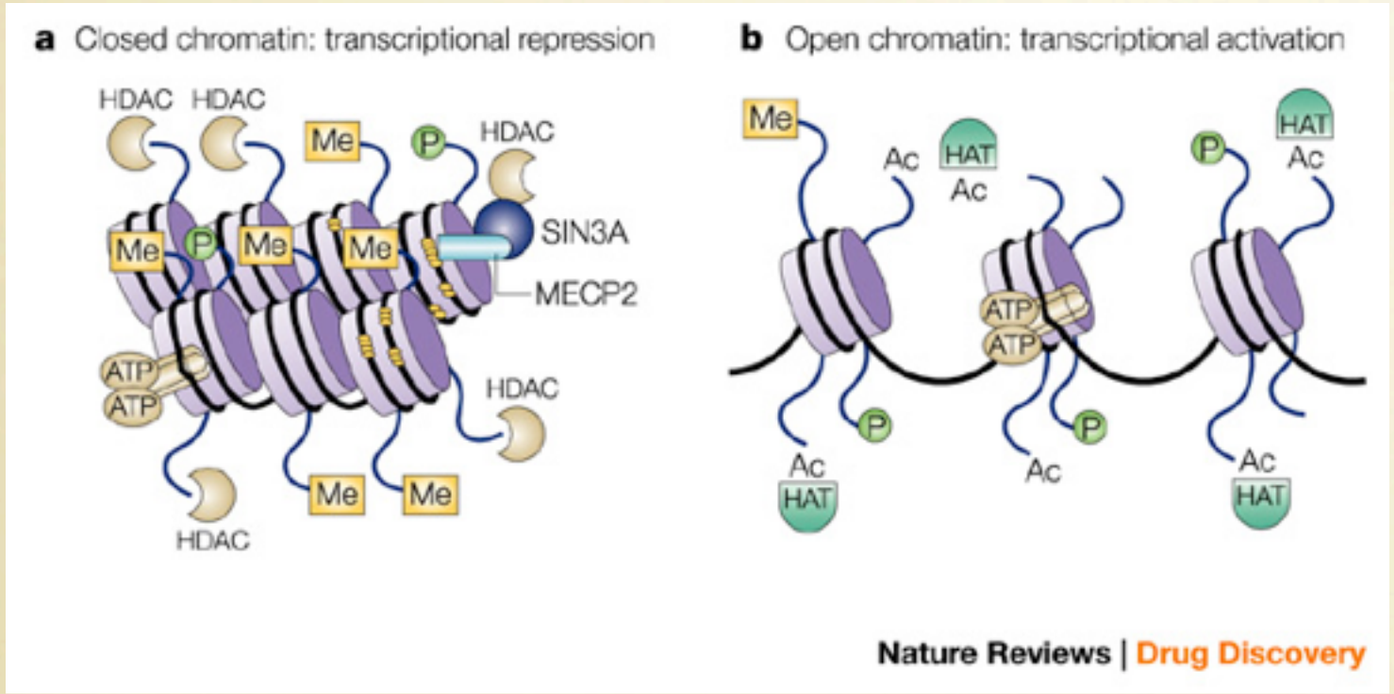


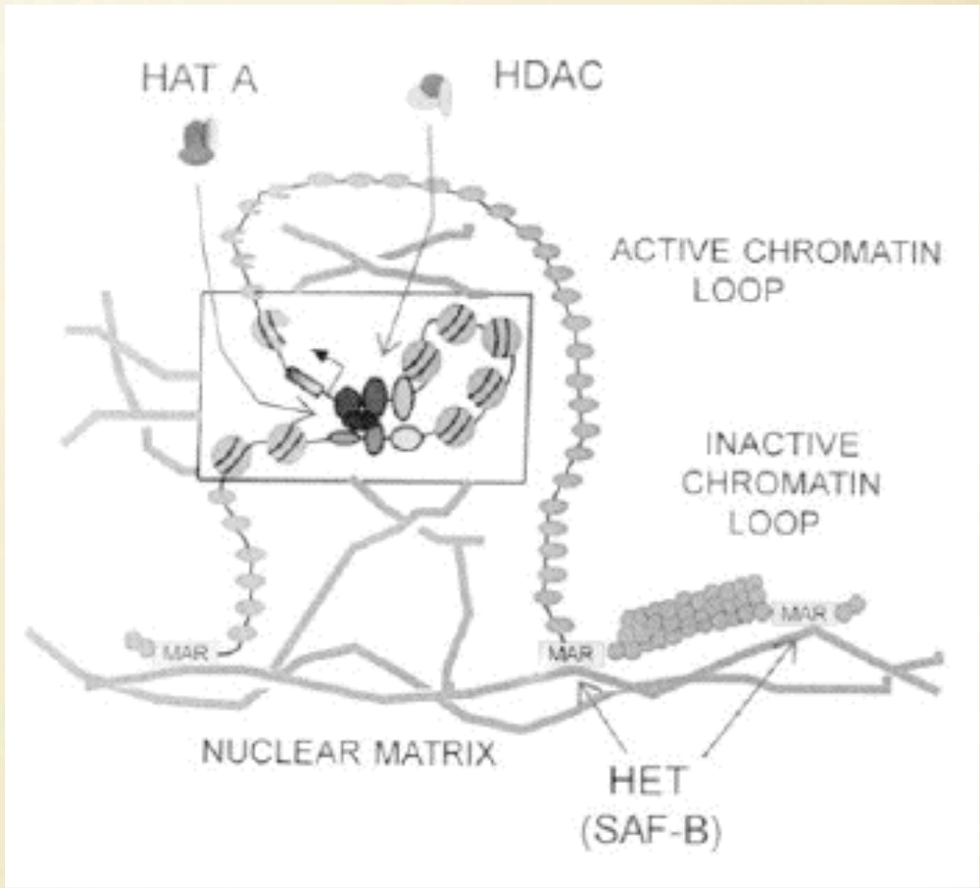
Nucleosomes





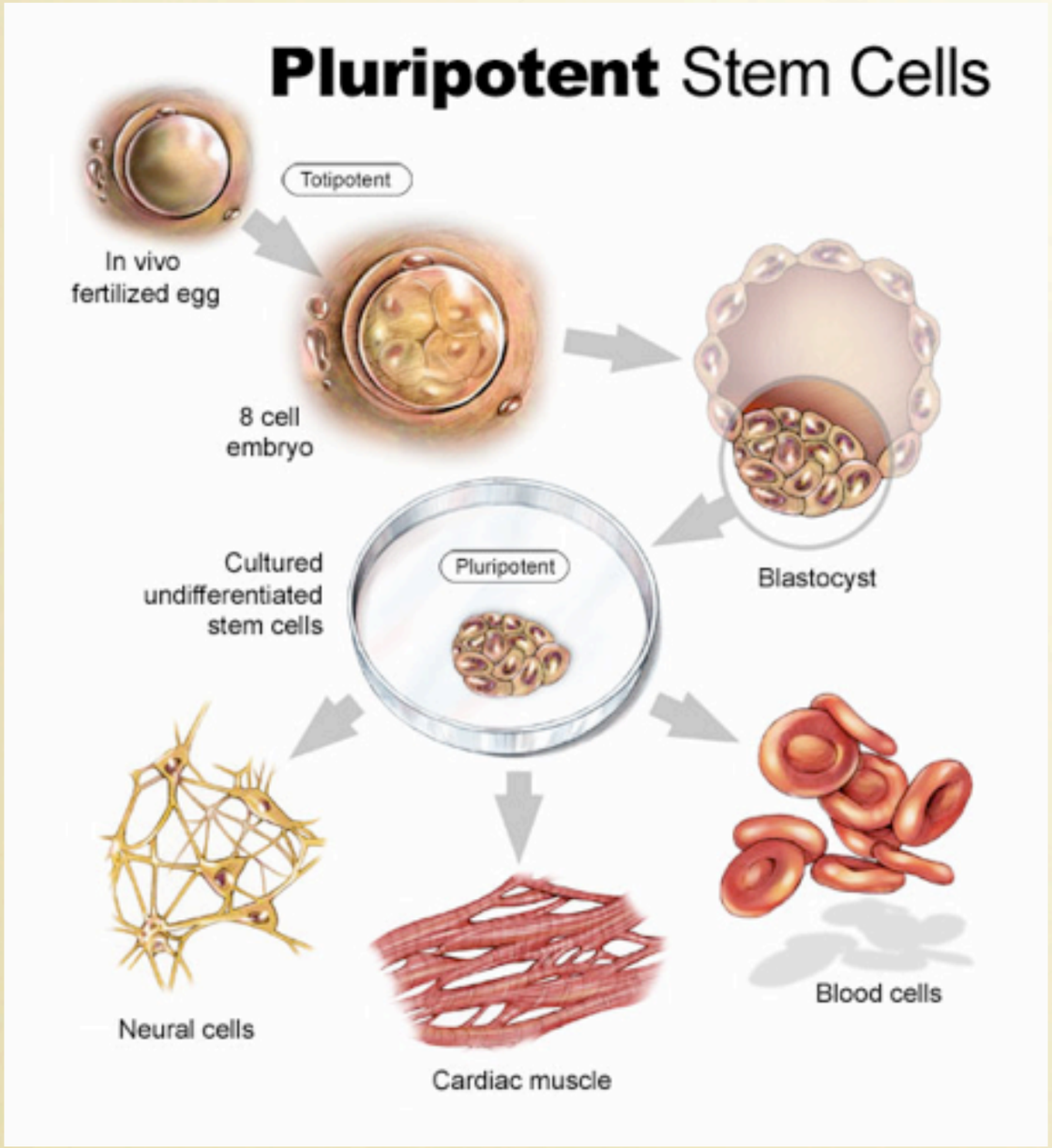
Chromatin structure

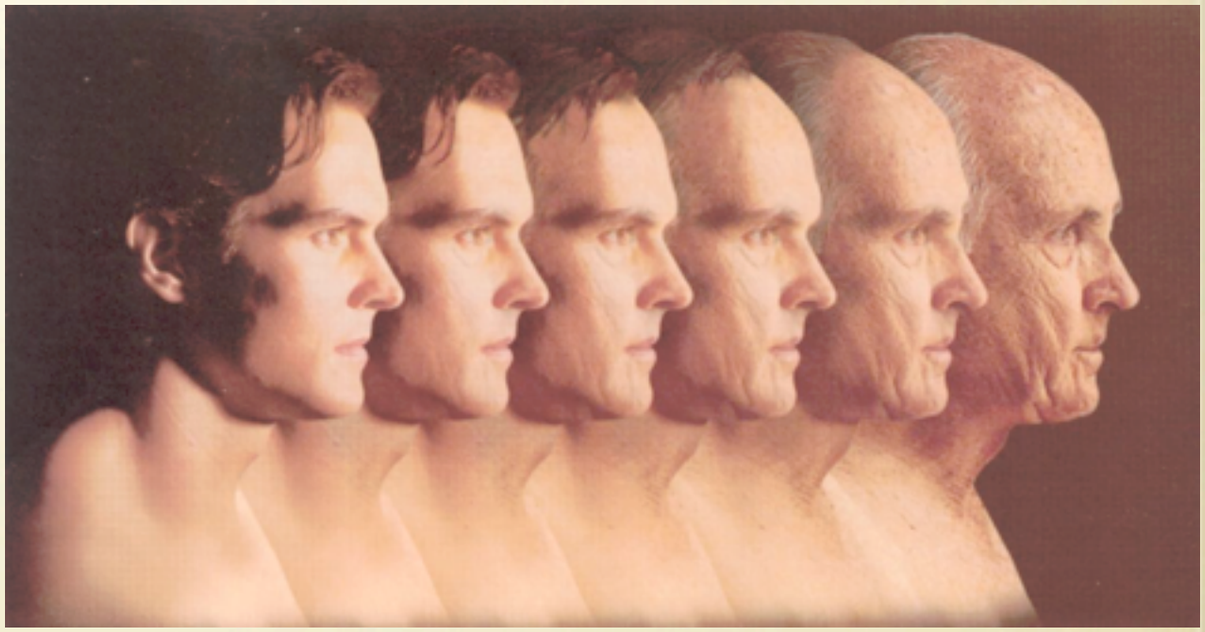




Aging...

Some continuity?





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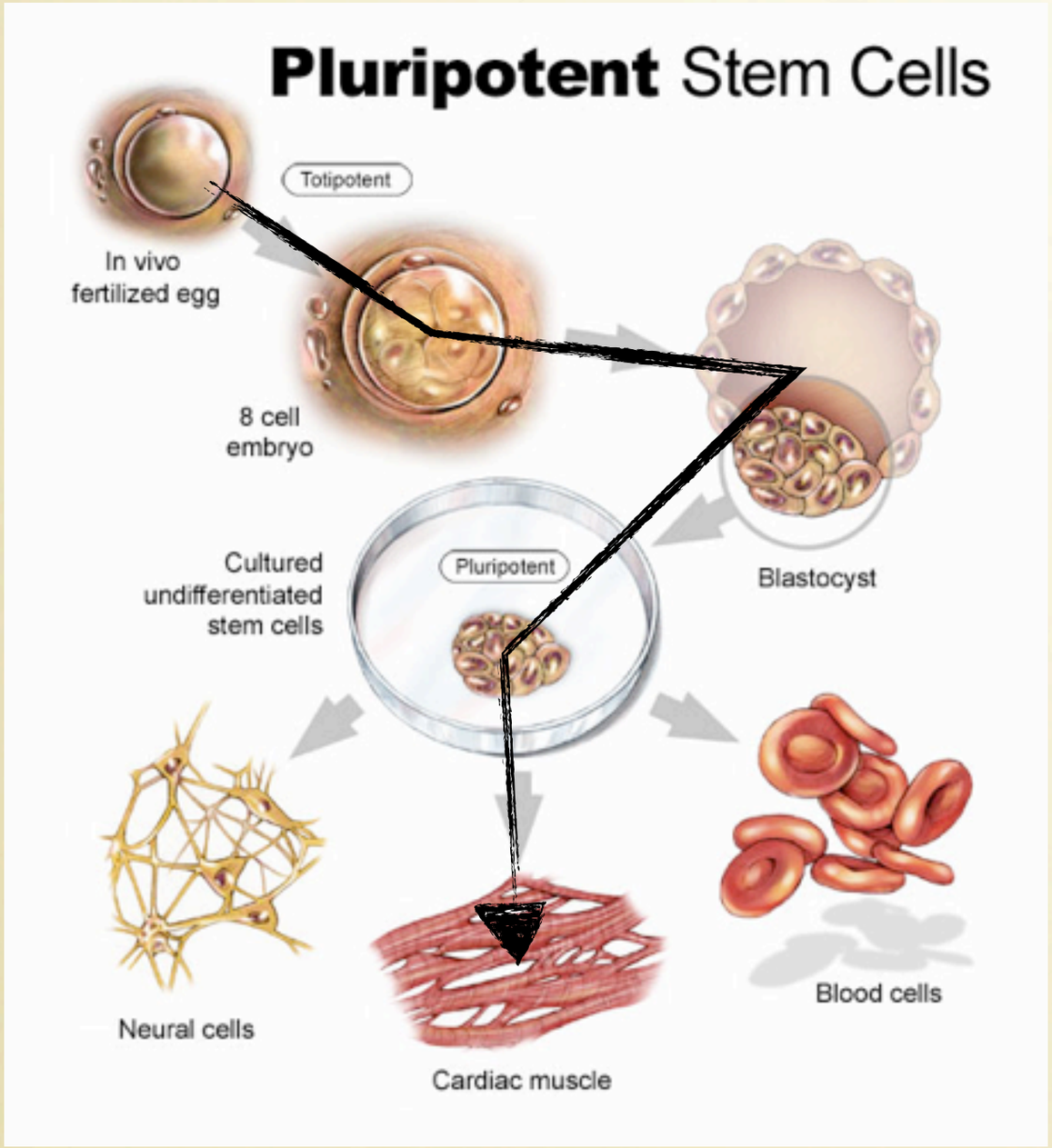
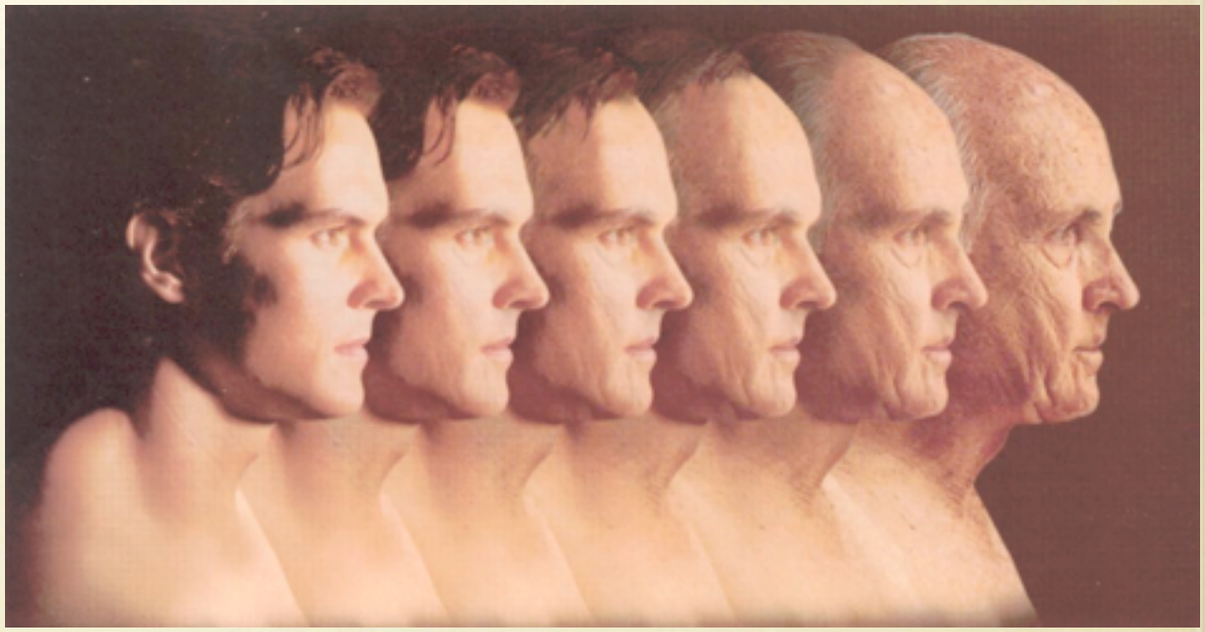




Non standard unifying view

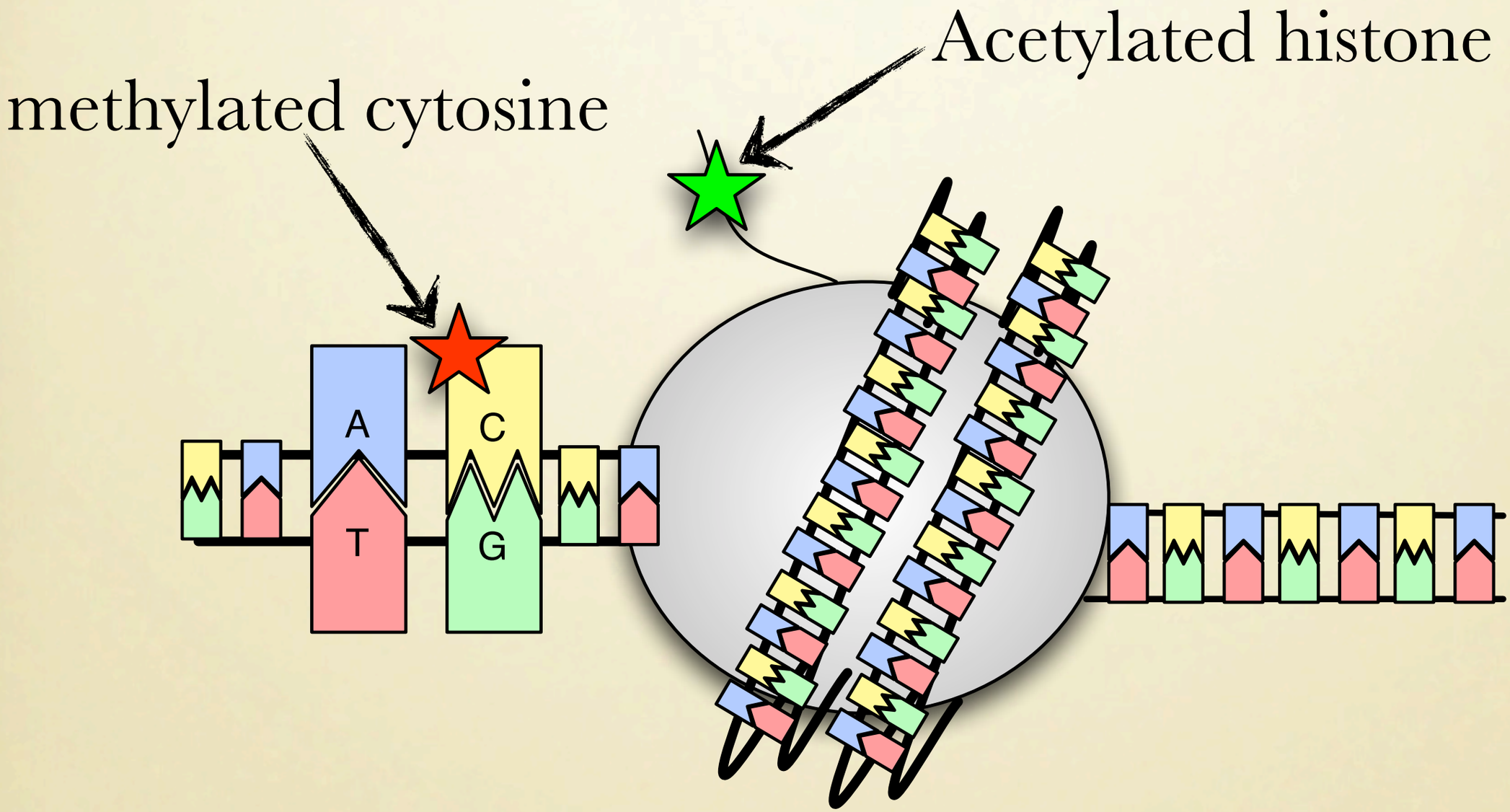
- Assume a set of cell phenotypes $\Phi = \{\varphi_1, \ldots, \varphi_n\}$
- A cell is Ψ-potent if it may eventually beget $\Psi \subseteq \Phi$ different types of differentiated cells
- E-sc are Φ-potent cells, H-sc are $\{\psi_1, \ldots, \psi_{11}\}$-potent
- Differentiated cells with phenotype ψ are $\{\psi\}$-potent
- Senescent cells (aging) are $\emptyset$-potent

SPECS

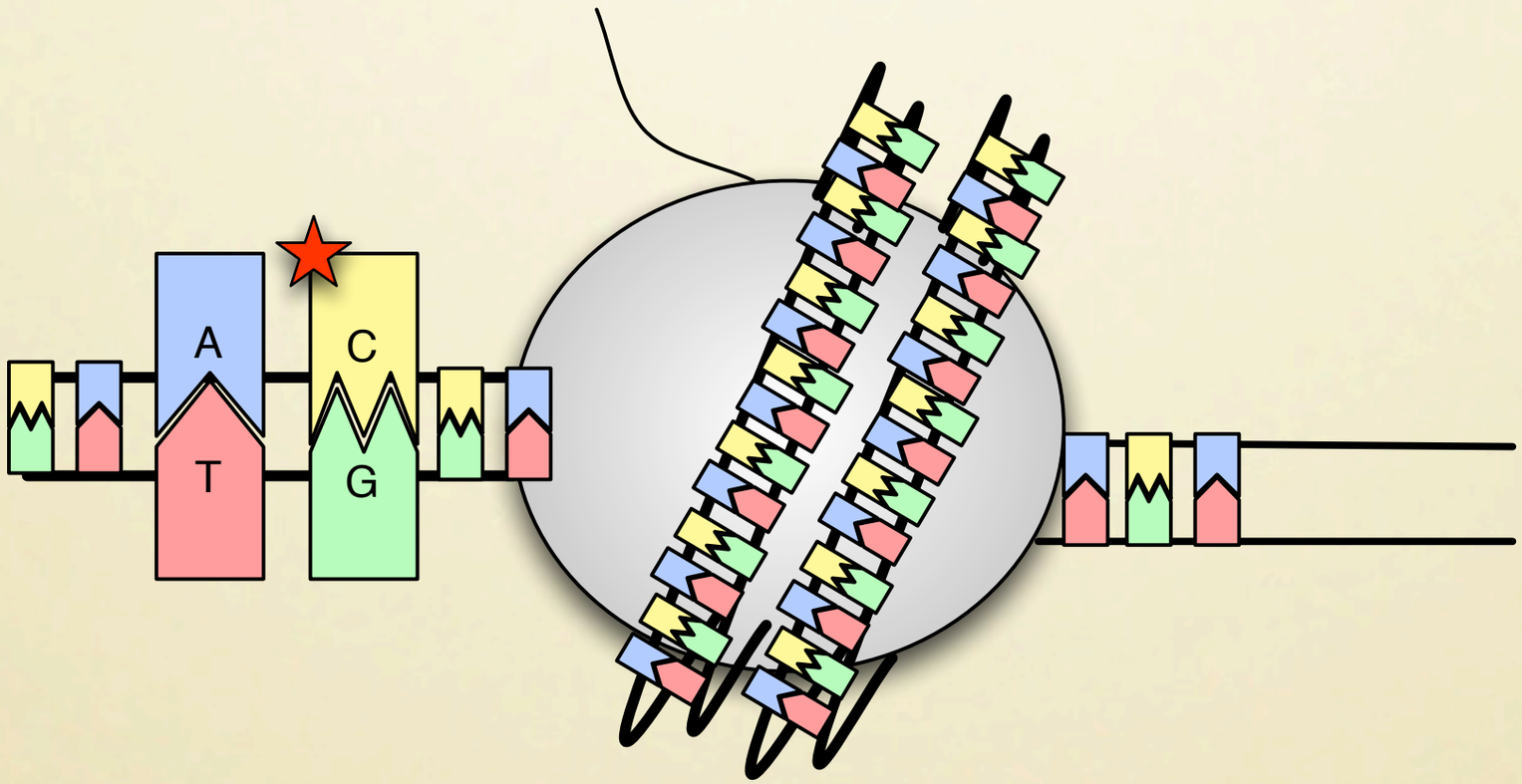
- Dissipative (potency is monotonically decreasing)
- Genome preserving (each daughter cell has a copy of the mother cell's genome)
- Branching (potency graph is a directed *tree*)

IF NOT ON THE GENES, WHERE IS THE INFORMATION?

Reconfiguring chromatin



Tug o' war



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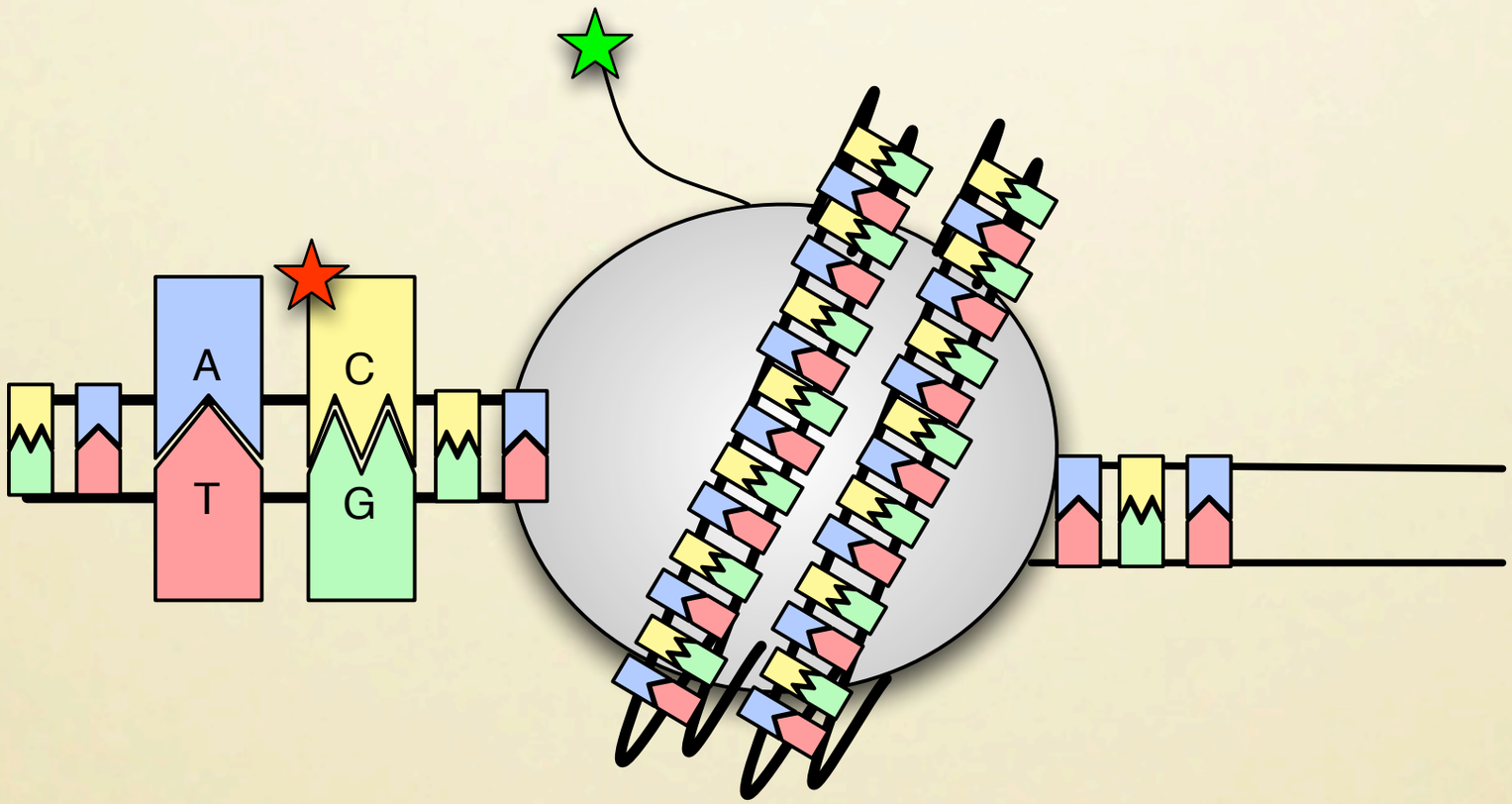

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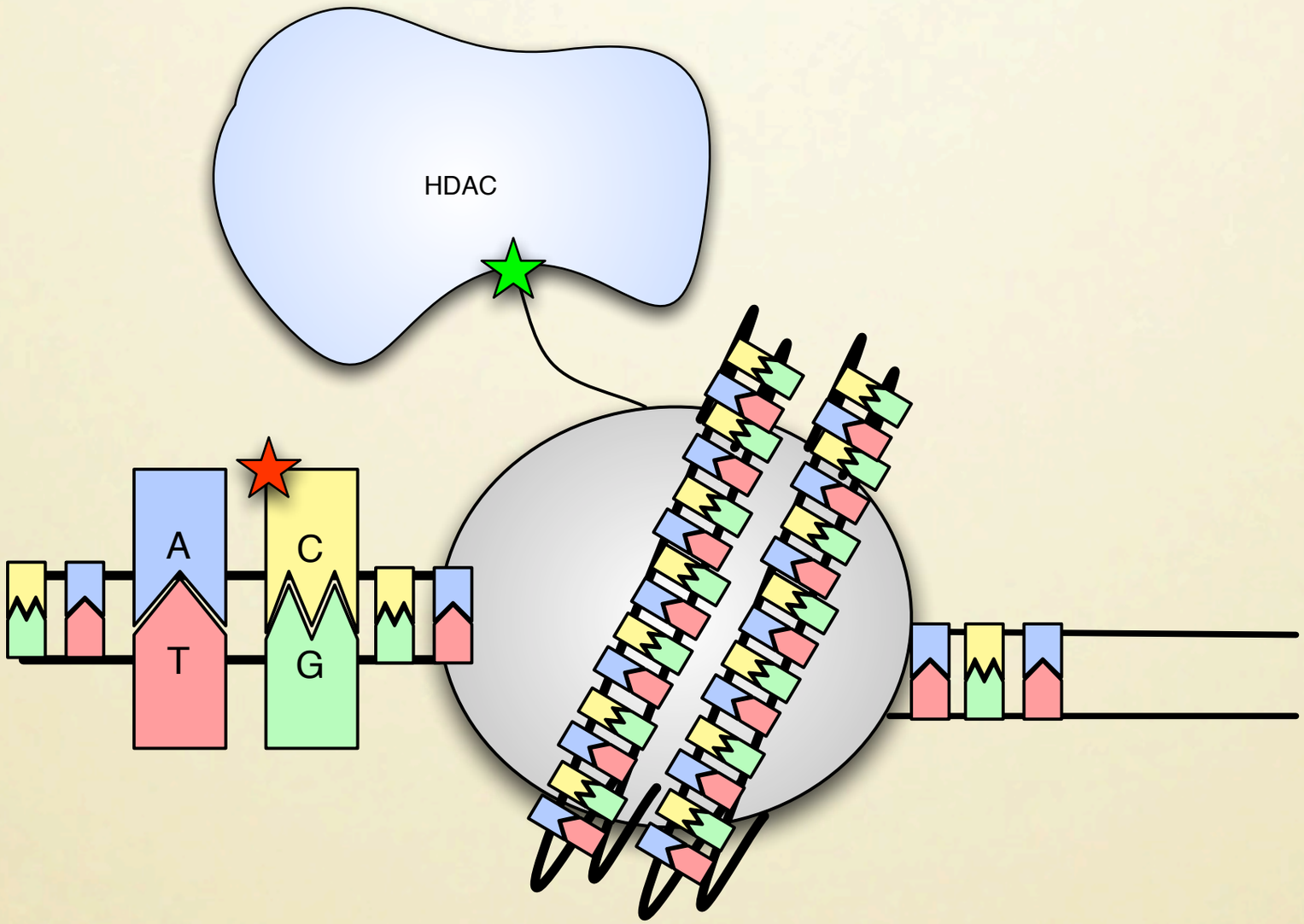

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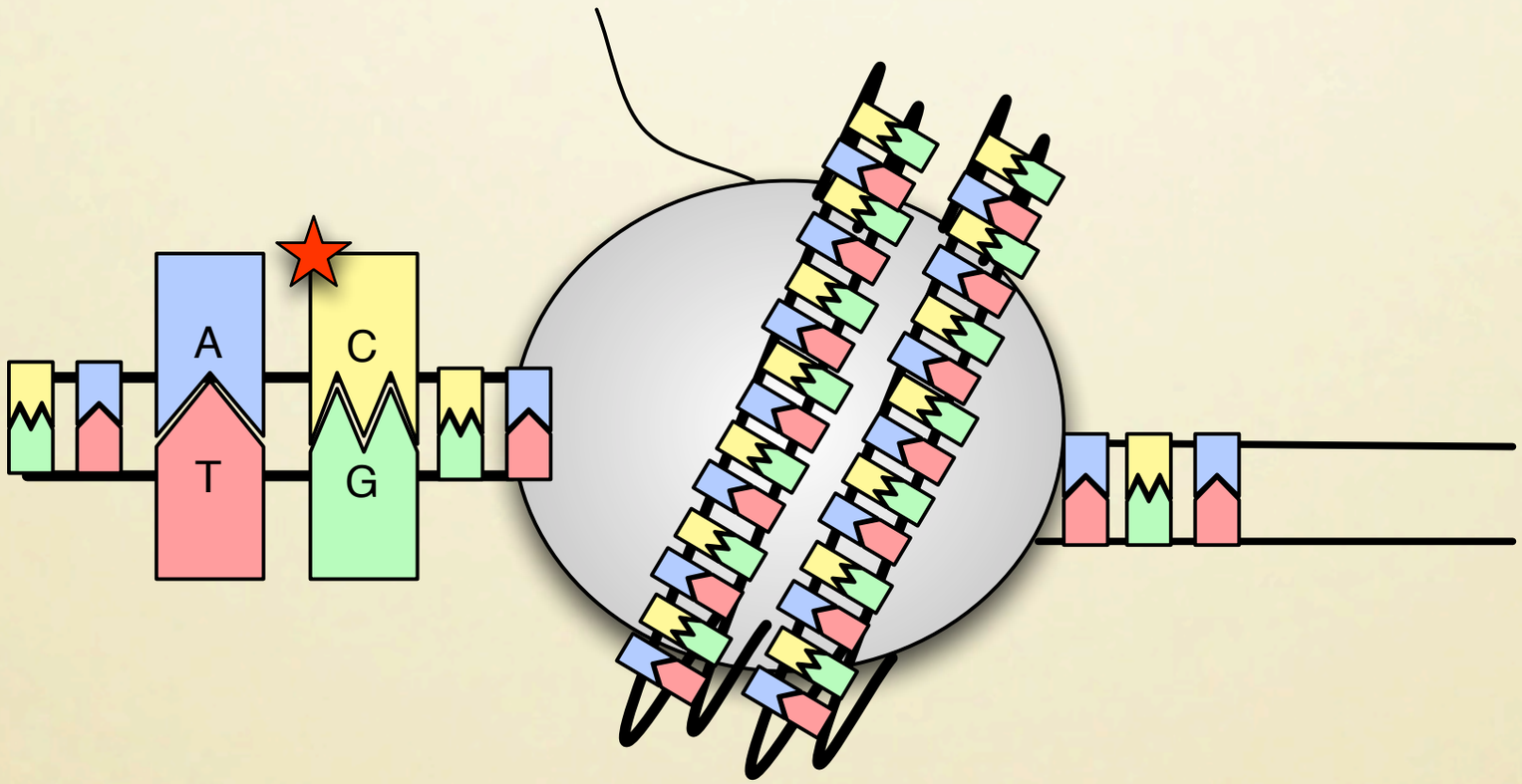

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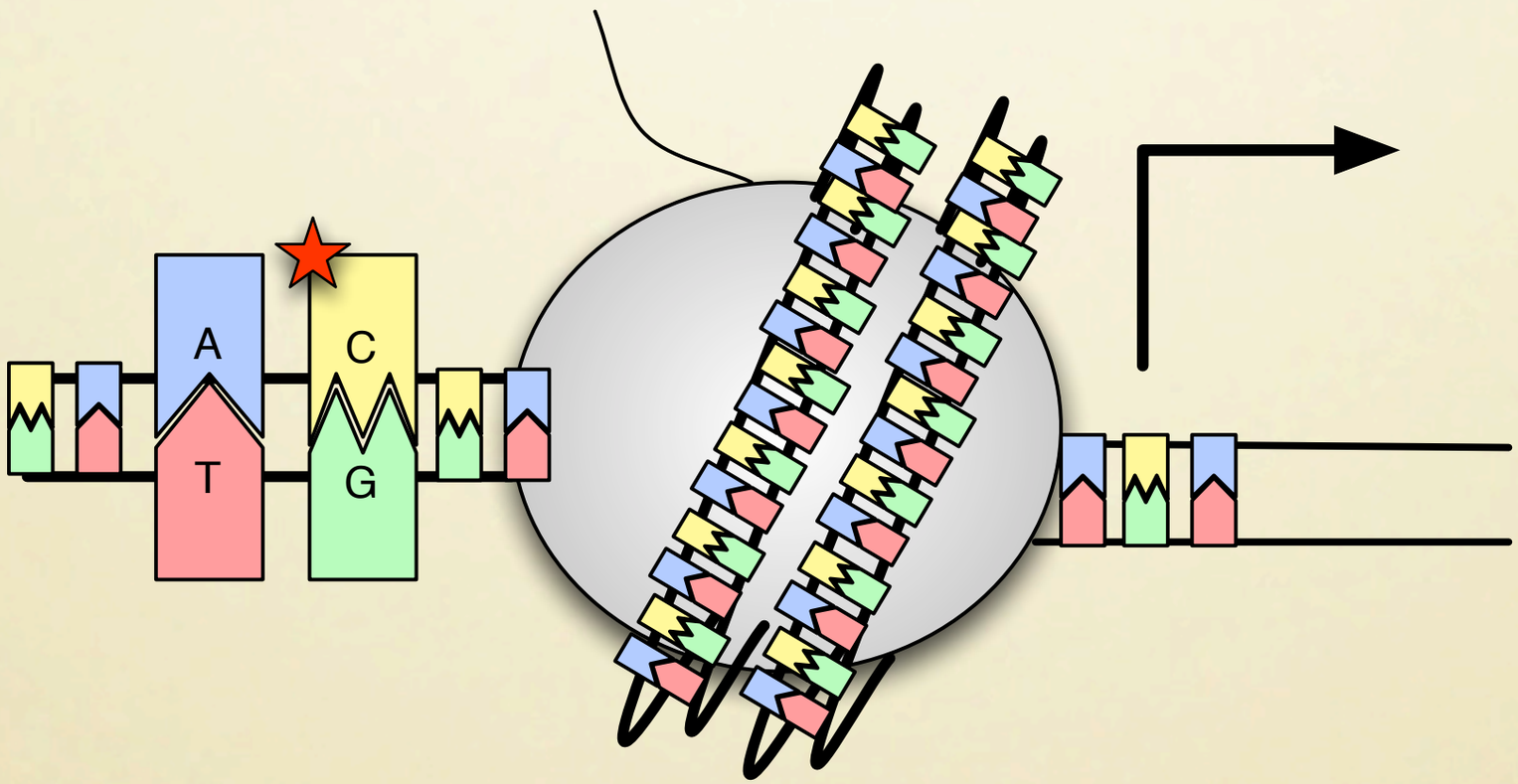
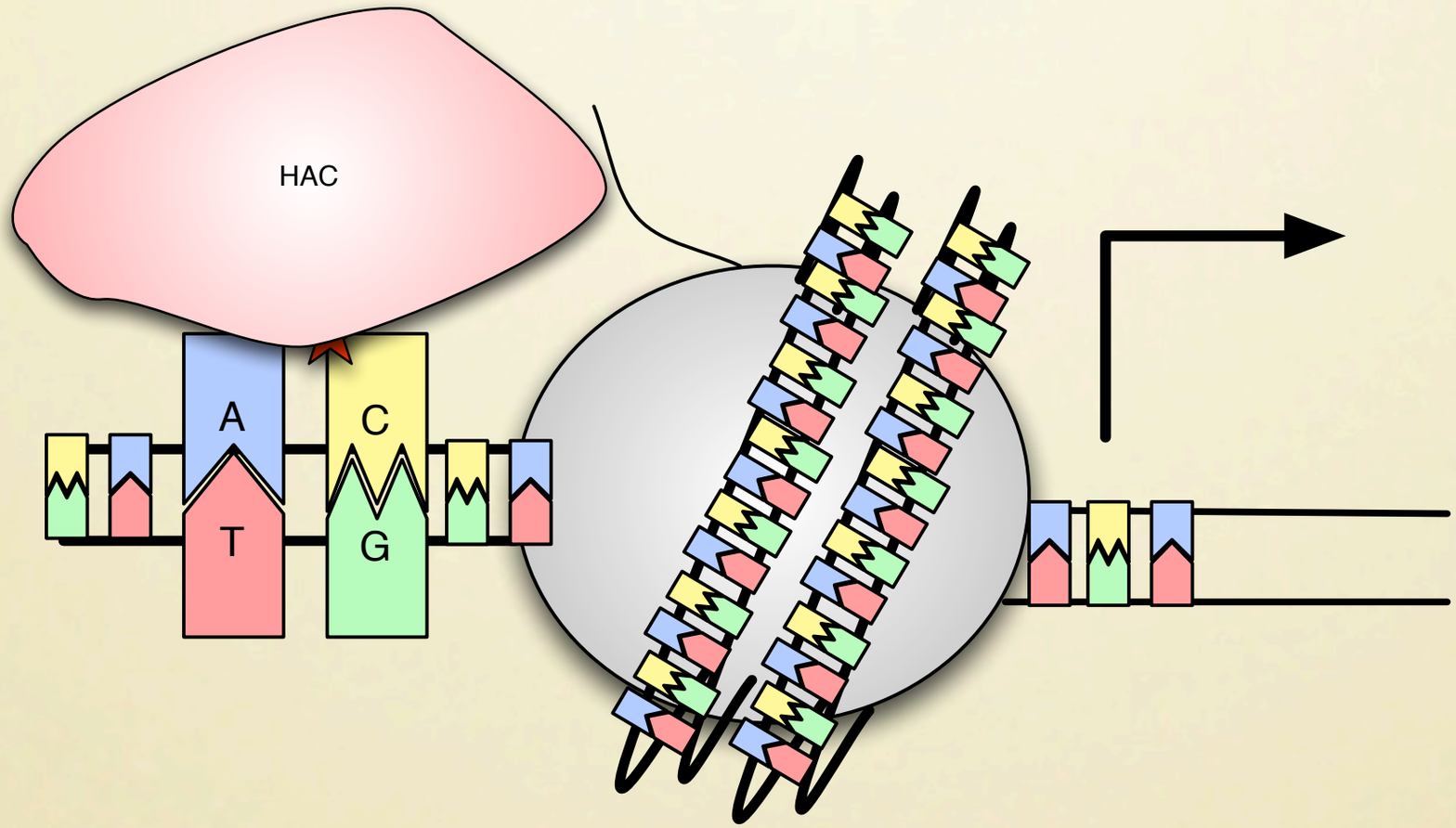
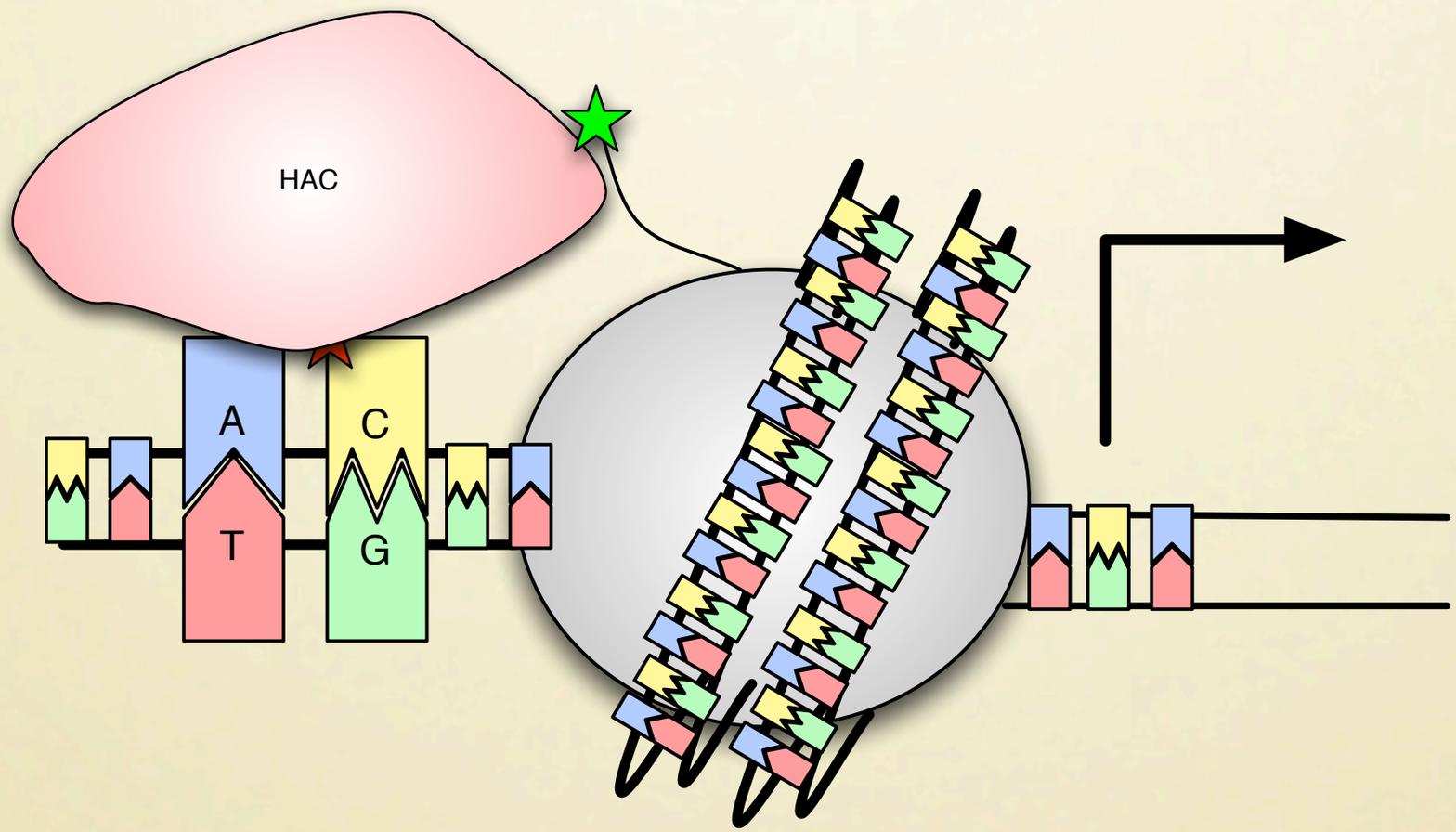


Tug o' war



Tug o' war



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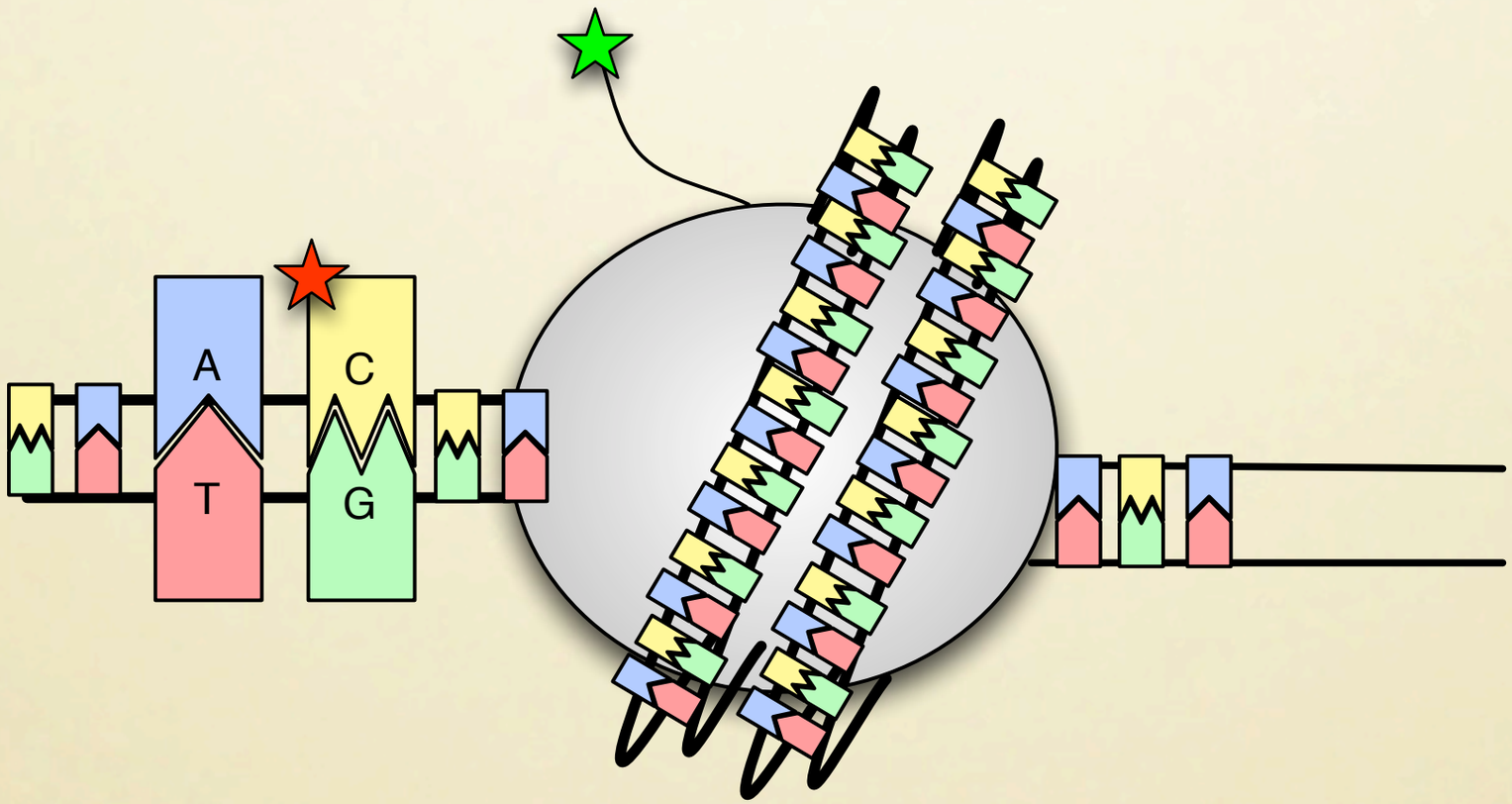


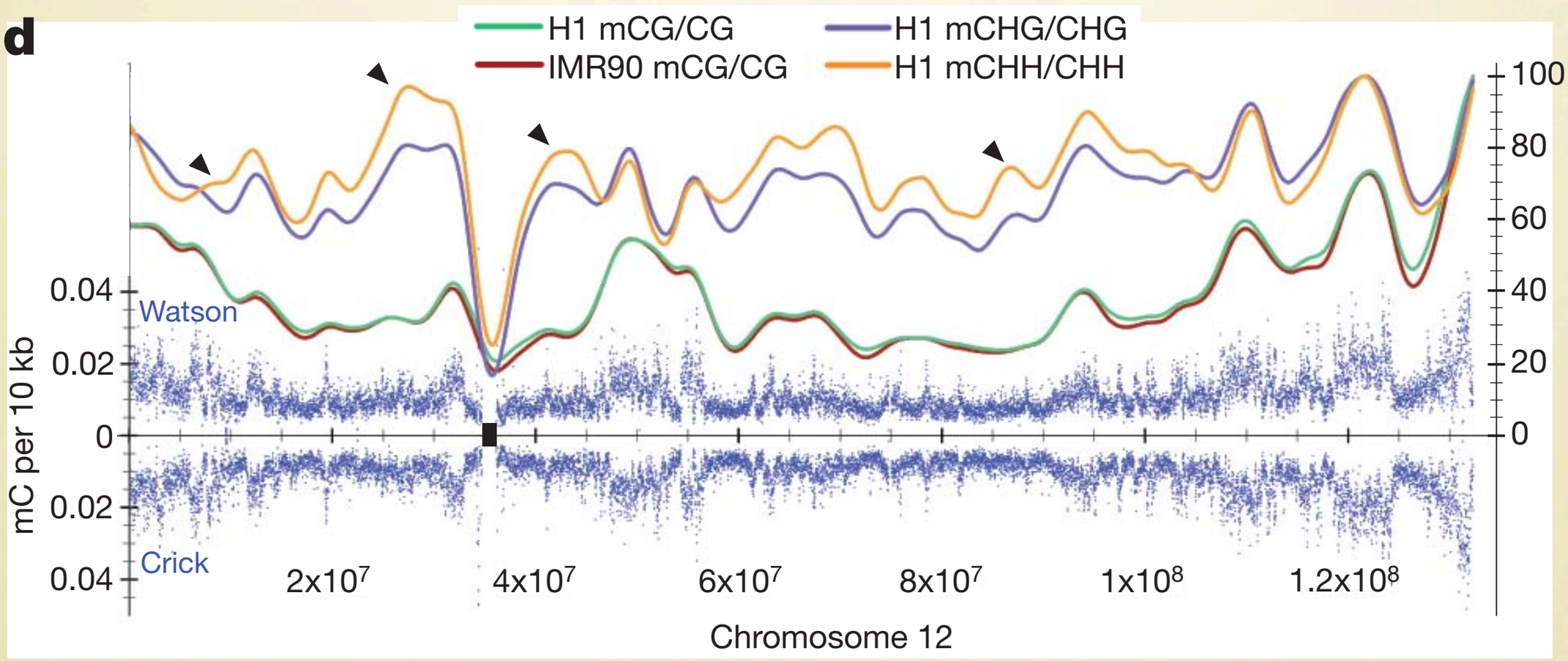
Methylation patterns

- Markers for gene silencing
- Epigenetics (not directly on the code)!
- Source of phenotypic heterogeneity

Arrives...

methyl C seq!

Methylome

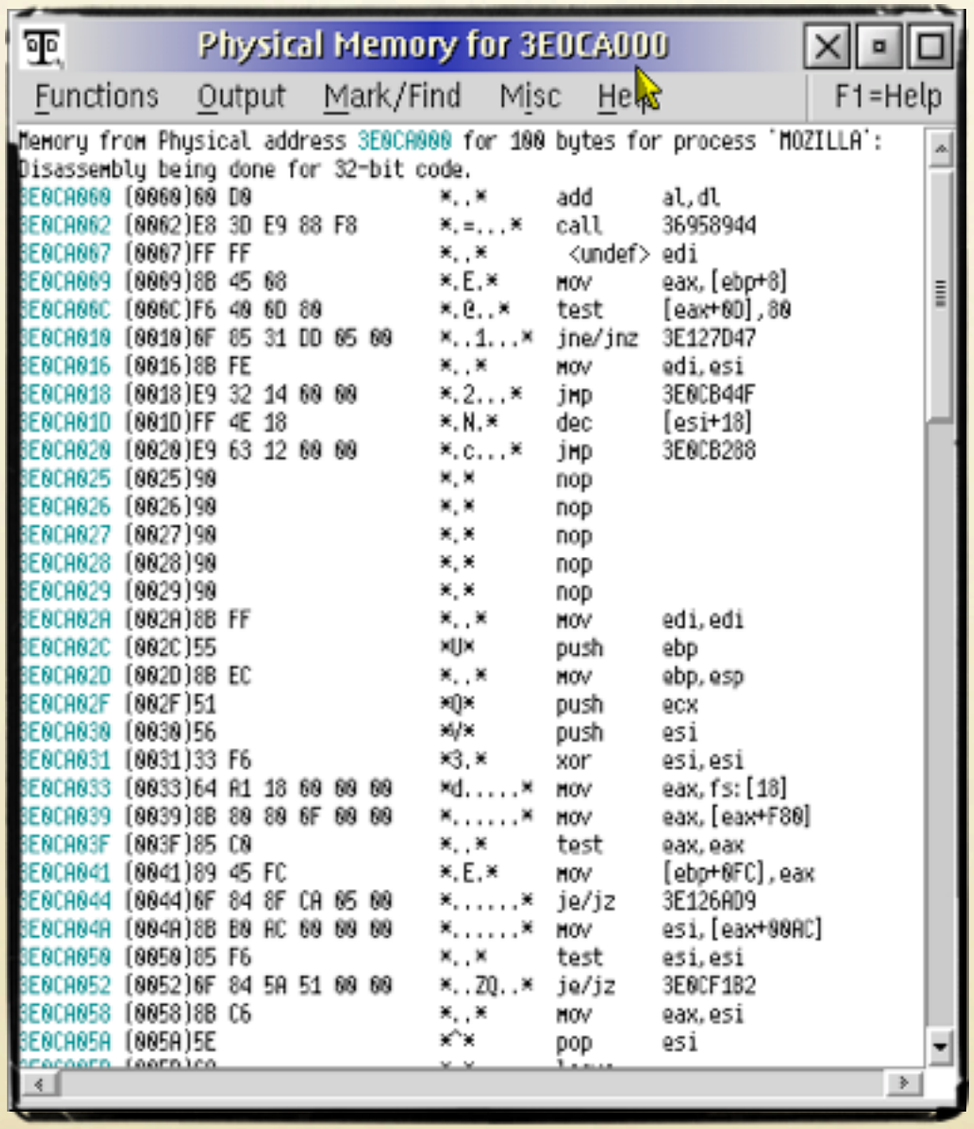


Human DNA methylomes at base resolution show widespread epigenomic differences

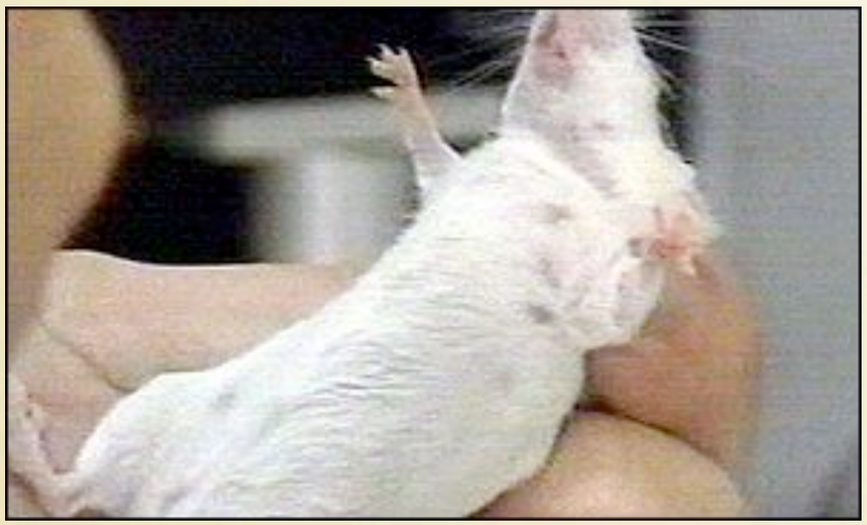
Ryan Lister[1]*, Mattia Pelizzola[1]*, Robert H. Dowen[1], R. David Hawkins[2], Gary Hon[2], Julian Tonti-Filippini[4], Joseph R. Nery[1], Leonard Lee[2], Zhen Ye[2], Que-Minh Ngo[2], Lee Edsall[2], Jessica Antosiewicz-Bourget[5,6], Ron Stewart[5,6], Victor Ruotti[5,6], A. Harvey Millar[4], James A. Thomson[5,6,7,8], Bing Ren[2,3] & Joseph R. Ecker[1]

For computing

Genes are primitive instructions not the program!



Genetic markers of acquired features!

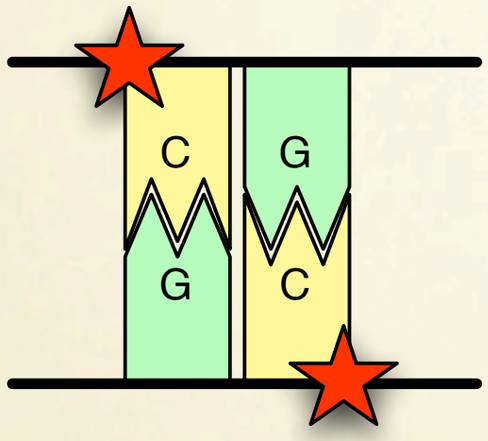


Cool mice

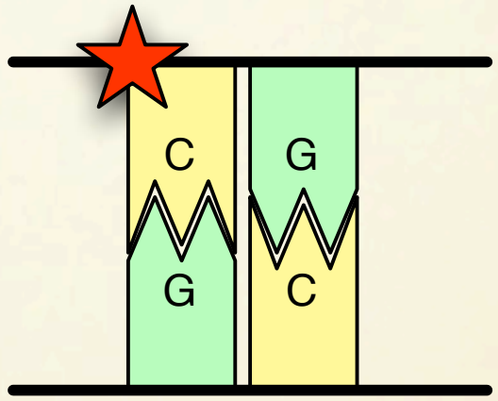
Stressed mice

The Maintenance Issue

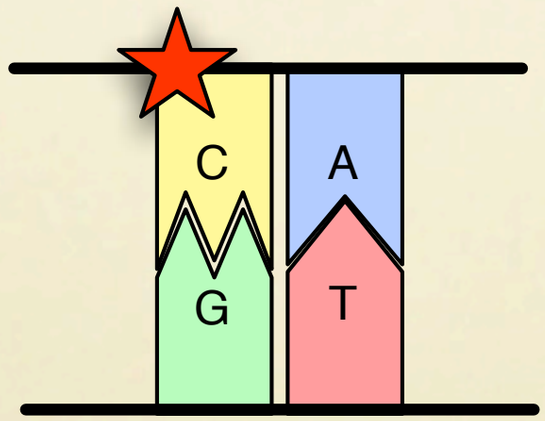
Methylation contexts



methylated CpG island

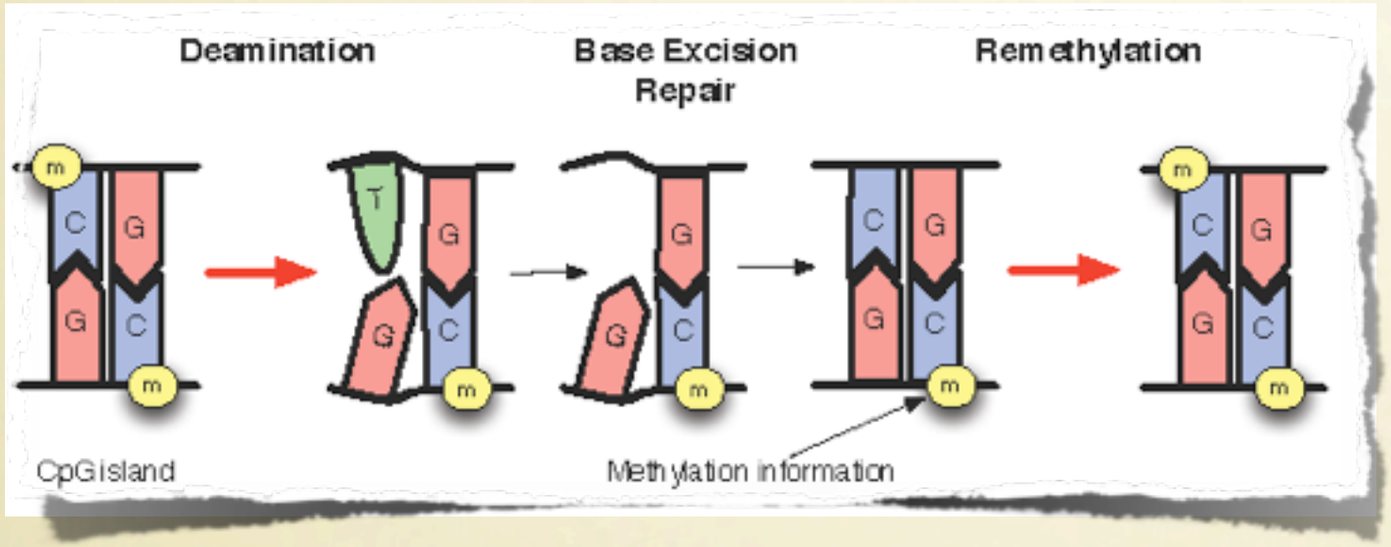


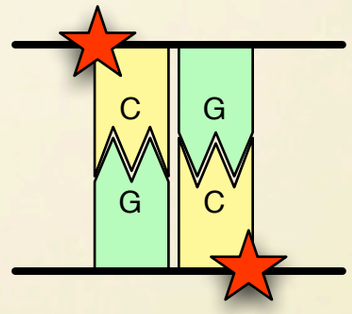
hemi-methylated CpG island

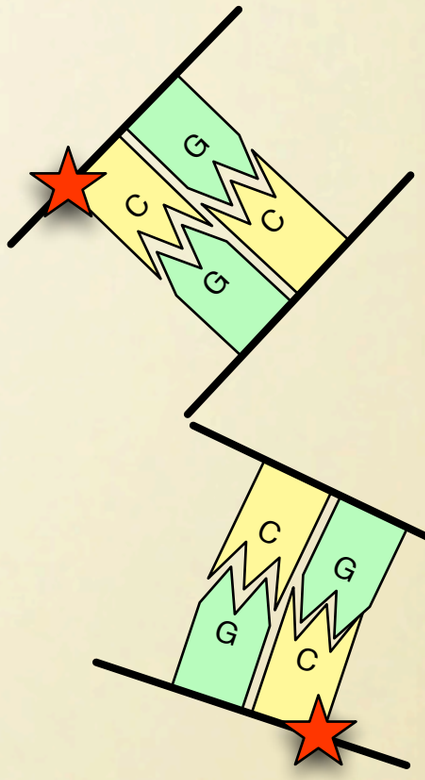


methylated CpH context

Symmetry is stability



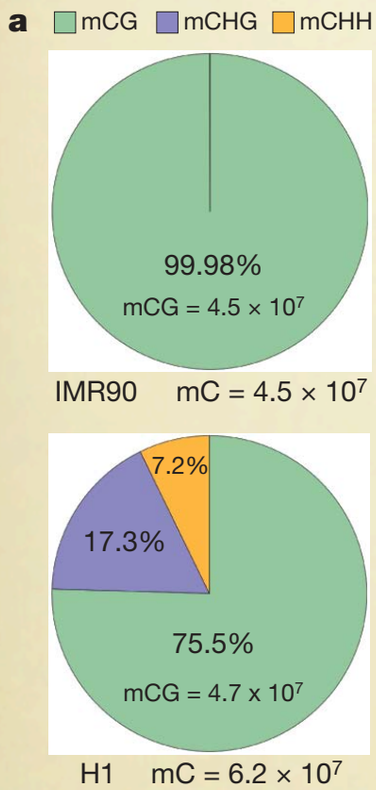


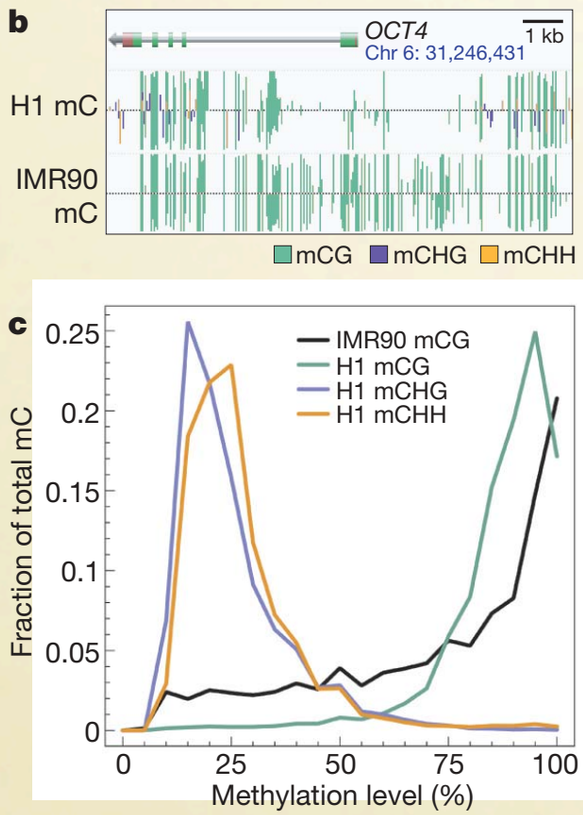


‘Maintenance’ DNA methyl transferase : DNMT1

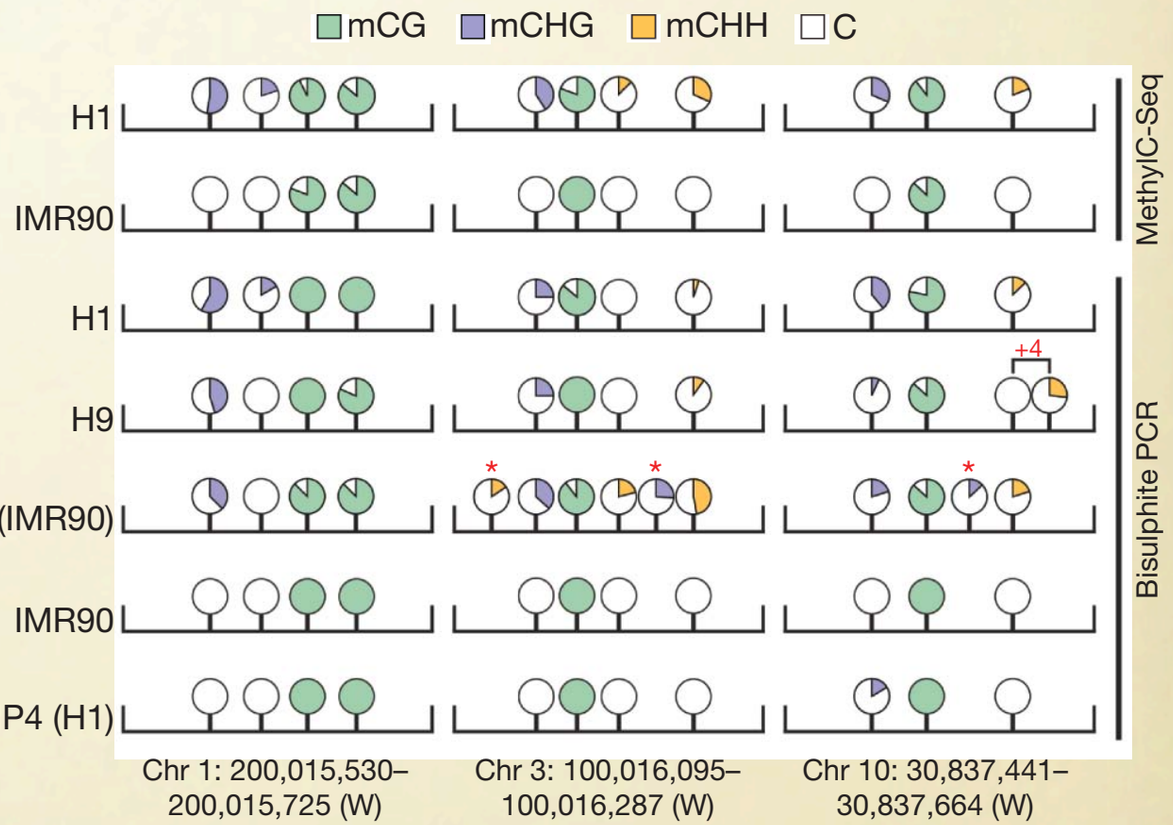
What about asymmetric contexts?

Stem cell methylome





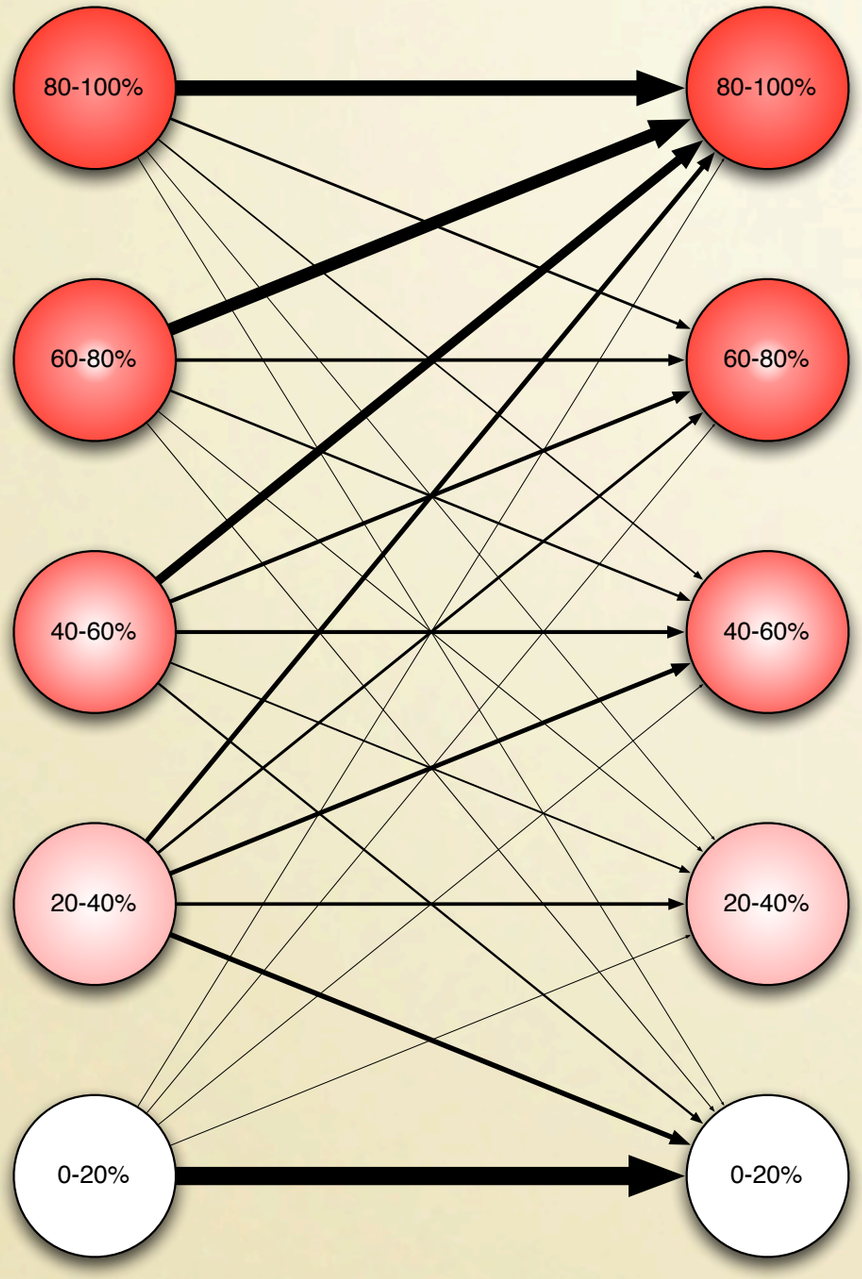
induced stem cell!



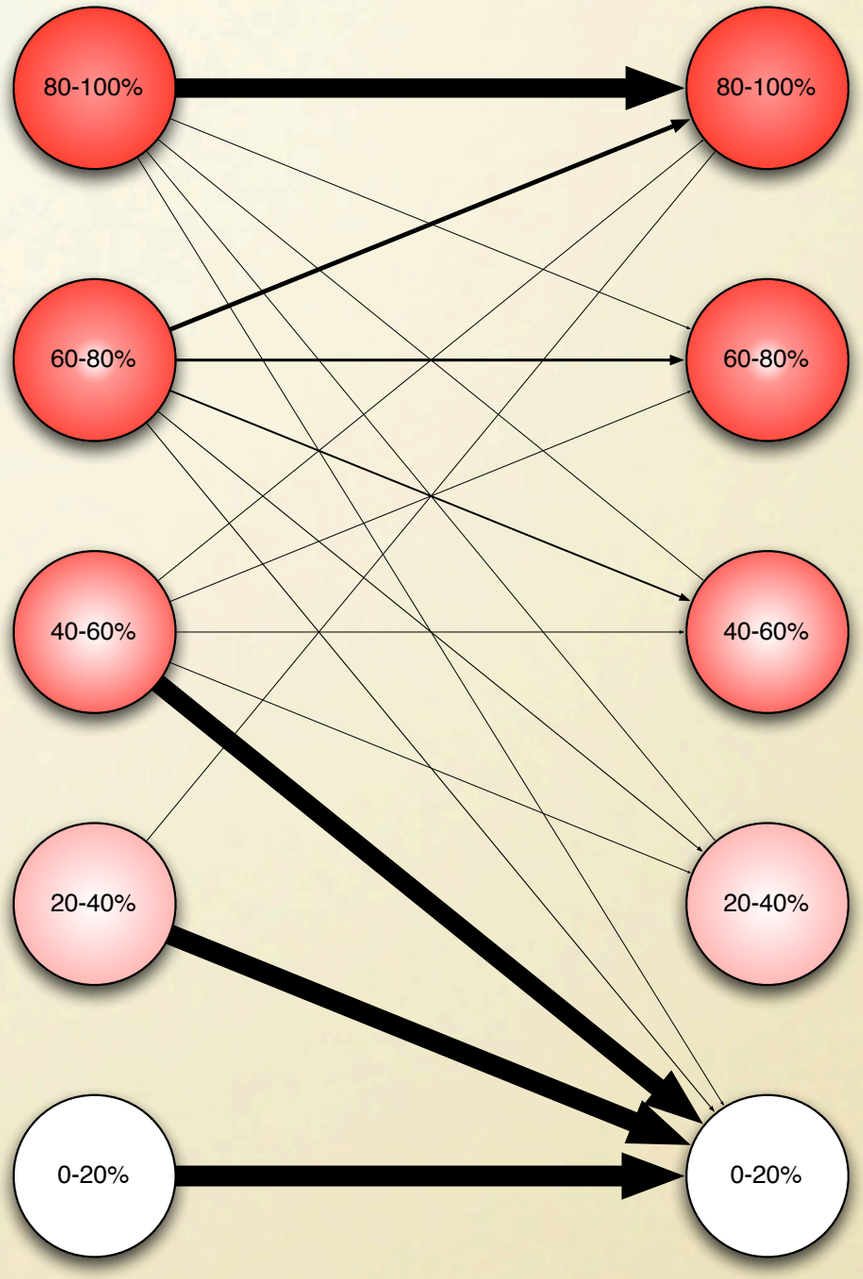
Human DNA methylomes at base resolution show widespread epigenomic differences

Ryan Lister[1]*, Mattia Pelizzola[1]*, Robert H. Dowen[1], R. David Hawkins[2], Gary Hon[2], Julian Tonti-Filippini[4], Joseph R. Nery[1], Leonard Lee[2], Zhen Ye[2], Que-Minh Ngo[2], Lee Edsall[2], Jessica Antosiewicz-Bourget[5,6], Ron Stewart[5,6], Victor Ruotti[5,6], A. Harvey Millar[4], James A. Thomson[5,6,7,8], Bing Ren[2,3] & Joseph R. Ecker[1]

Observation 1

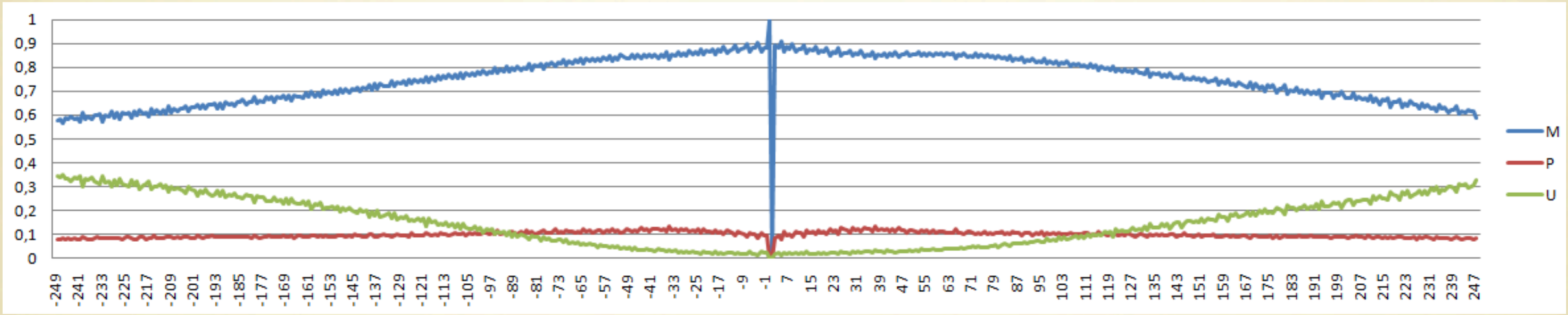


mCpG

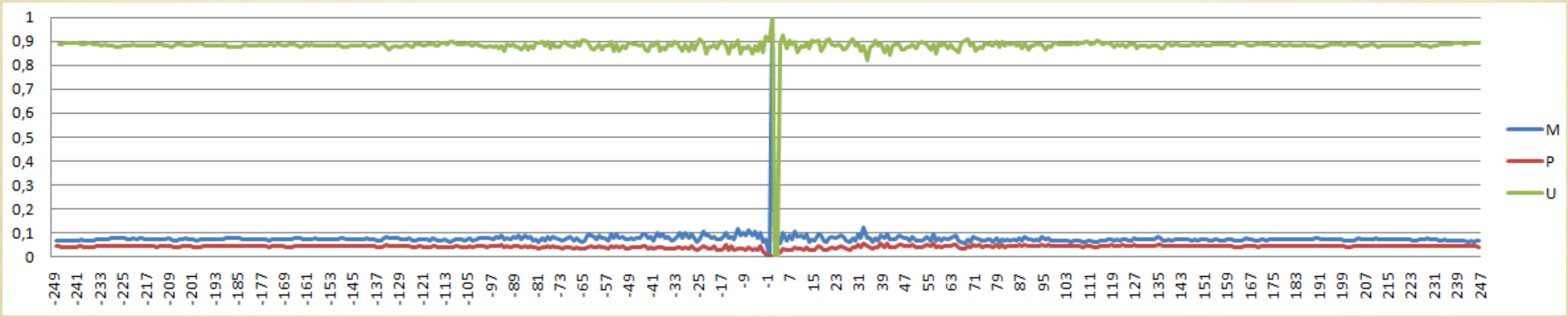


mCpH

Methylation profile 1

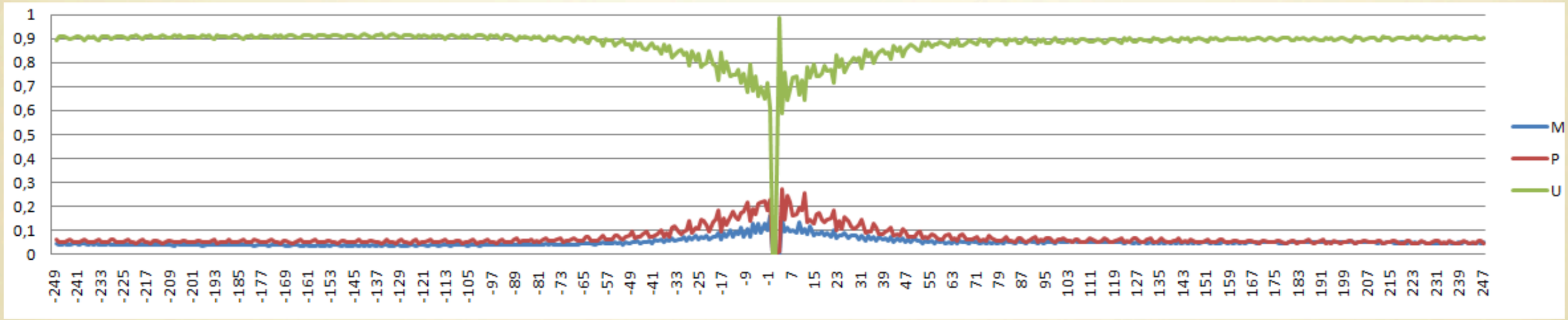


Methylation profile around a 80% methylated CpT (positive strand)

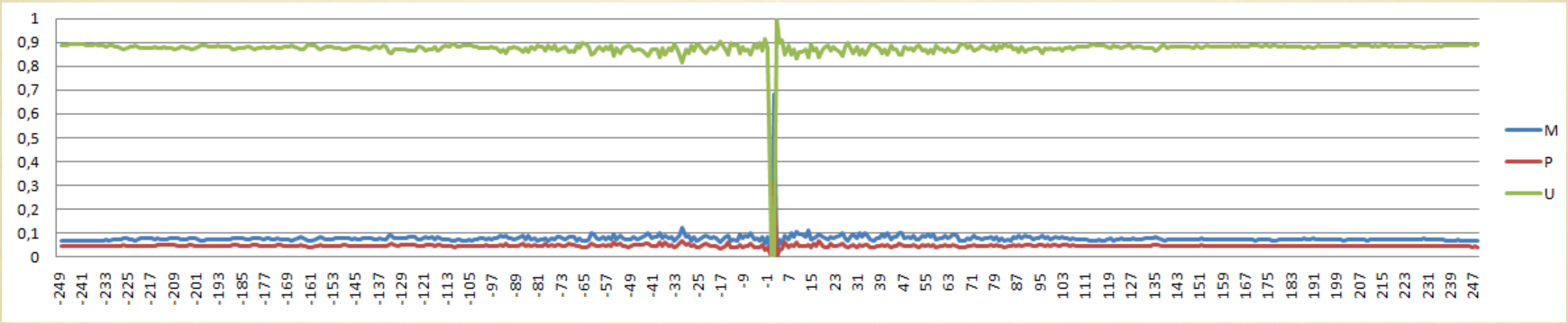


Methylation profile around a 80% methylated CpG (positive strand)

Methylation profile 2

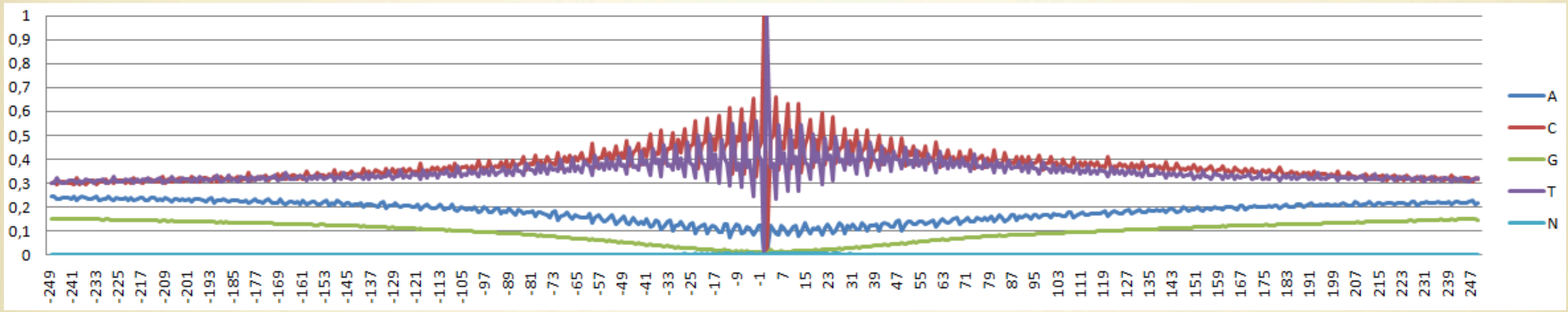


Methylation profile around a 80% methylated CpT (negative strand)

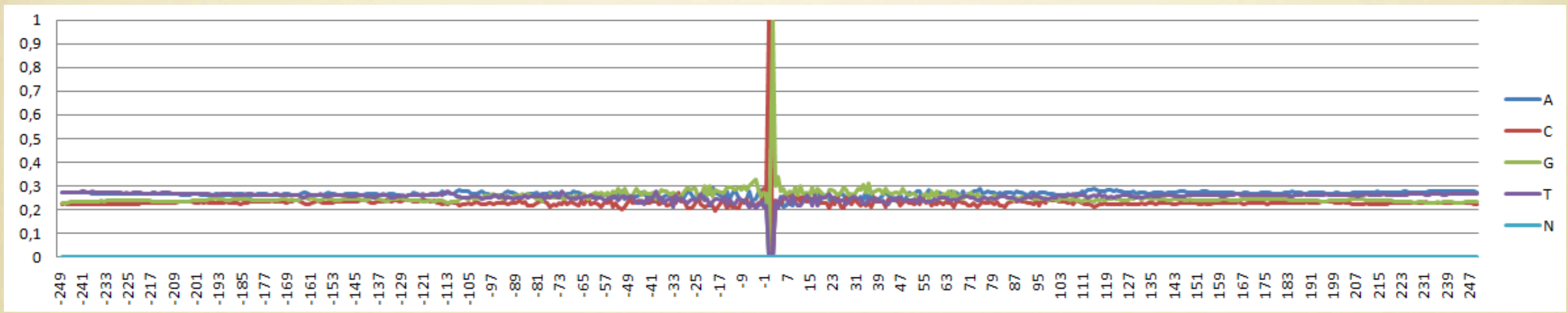


Methylation profile around a 80% methylated CpG (negative strand)

Nucleotide profile



Nucleotide probability profile given around a 80% methylated CpT



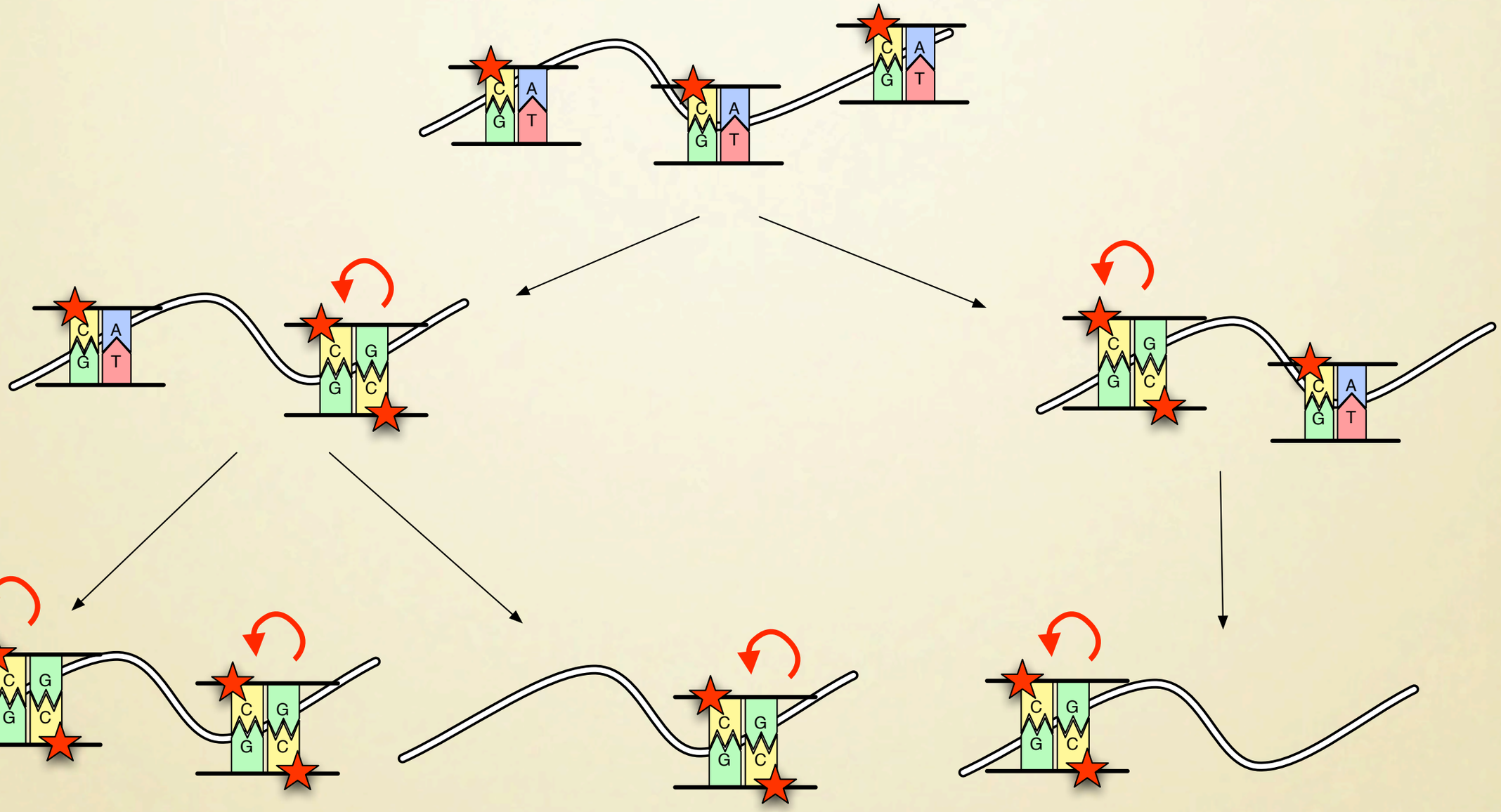
Nucleotide probability profile given around a 80% methylated CpG

Possible semantics...

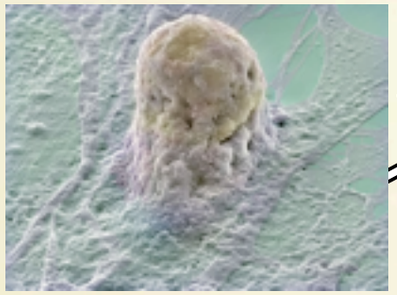
Specs

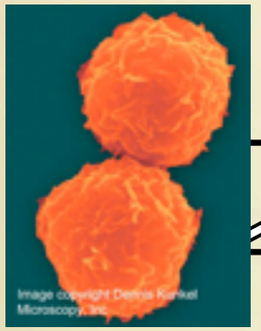
- mCpH contexts are absent from somatic cells and constitute about 25% of methylation contexts in stem cells
- Survive division
- Progressive loss?

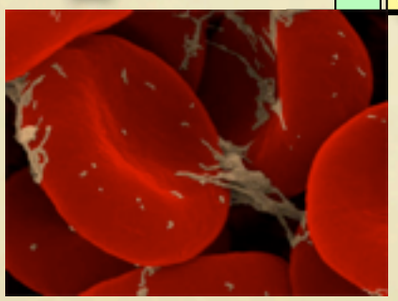
Plasticity vs stability

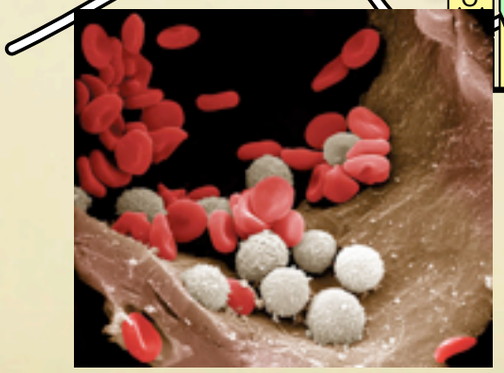


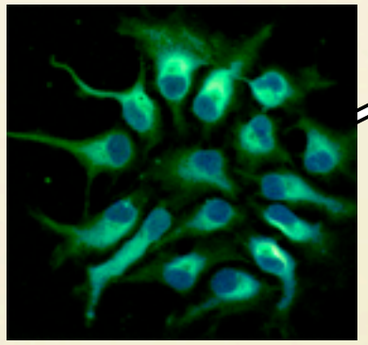
Plasticity vs stability

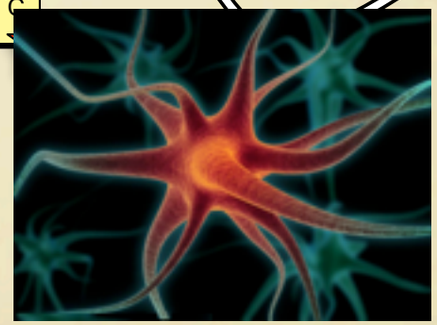










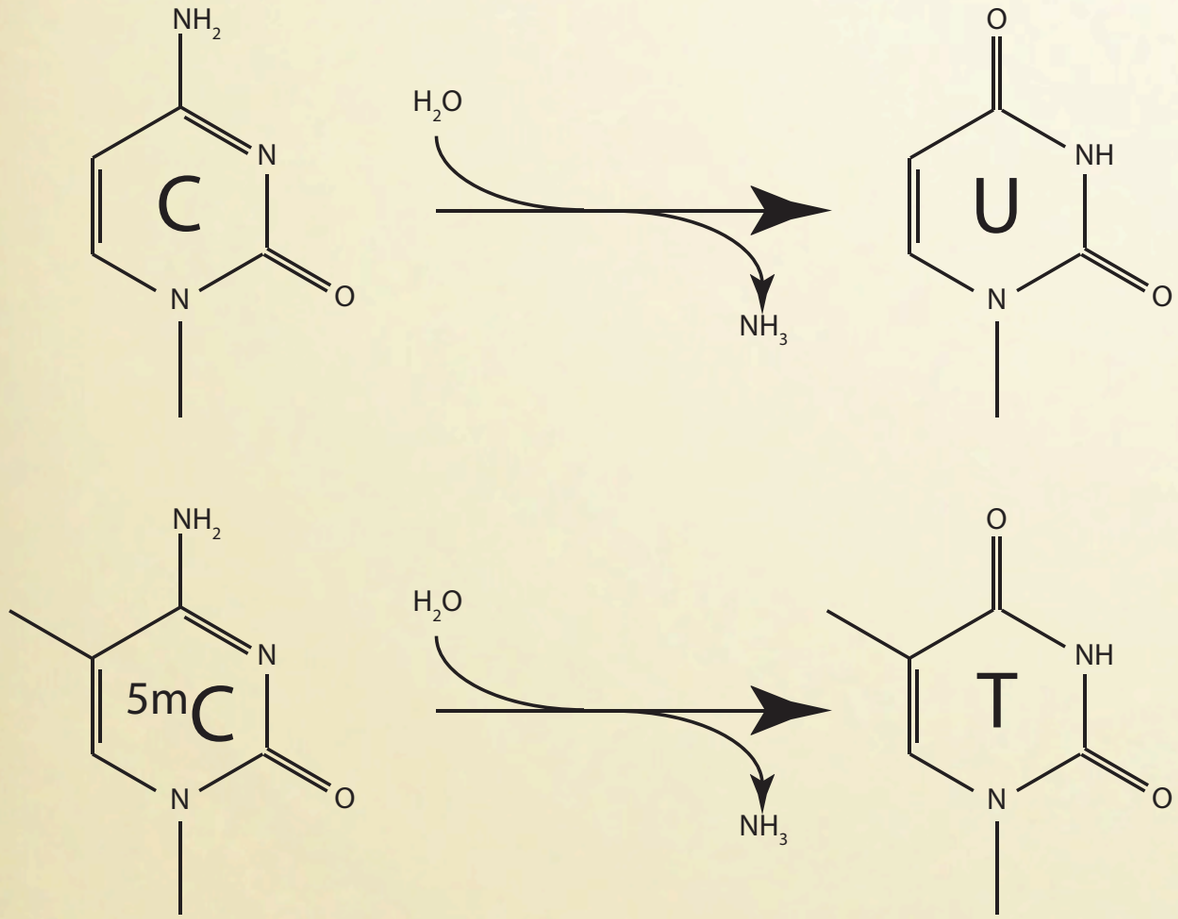


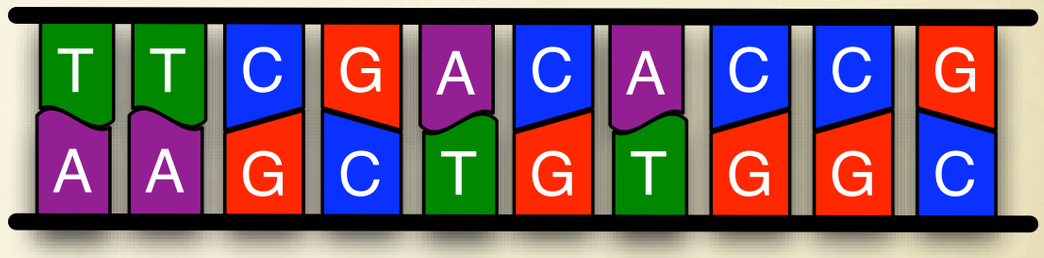
Model for Shifting Patterns

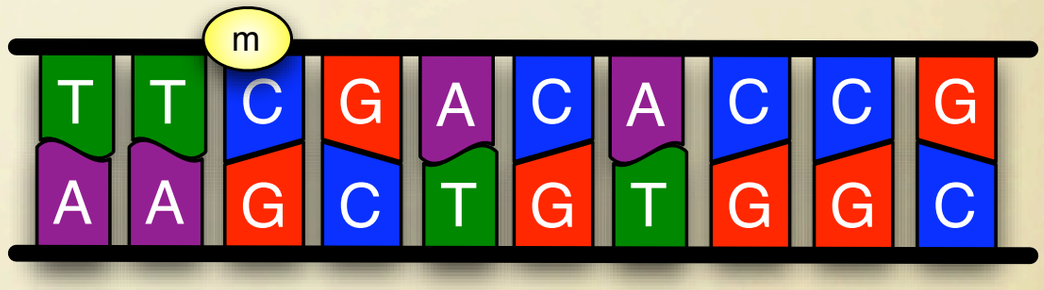
Bio facts

- No (known) enzyme can de-methylate DNA
- about 5% bases methylated in the genome (more in stem cells, less in somatic cells)
- X-chromosome is silenced by methylation
- deregulation of methylation is involved in most cancers (causes over/under gene expression)
- DNA methylation is specific to pluri-cellular organisms

Losing methylation pattern

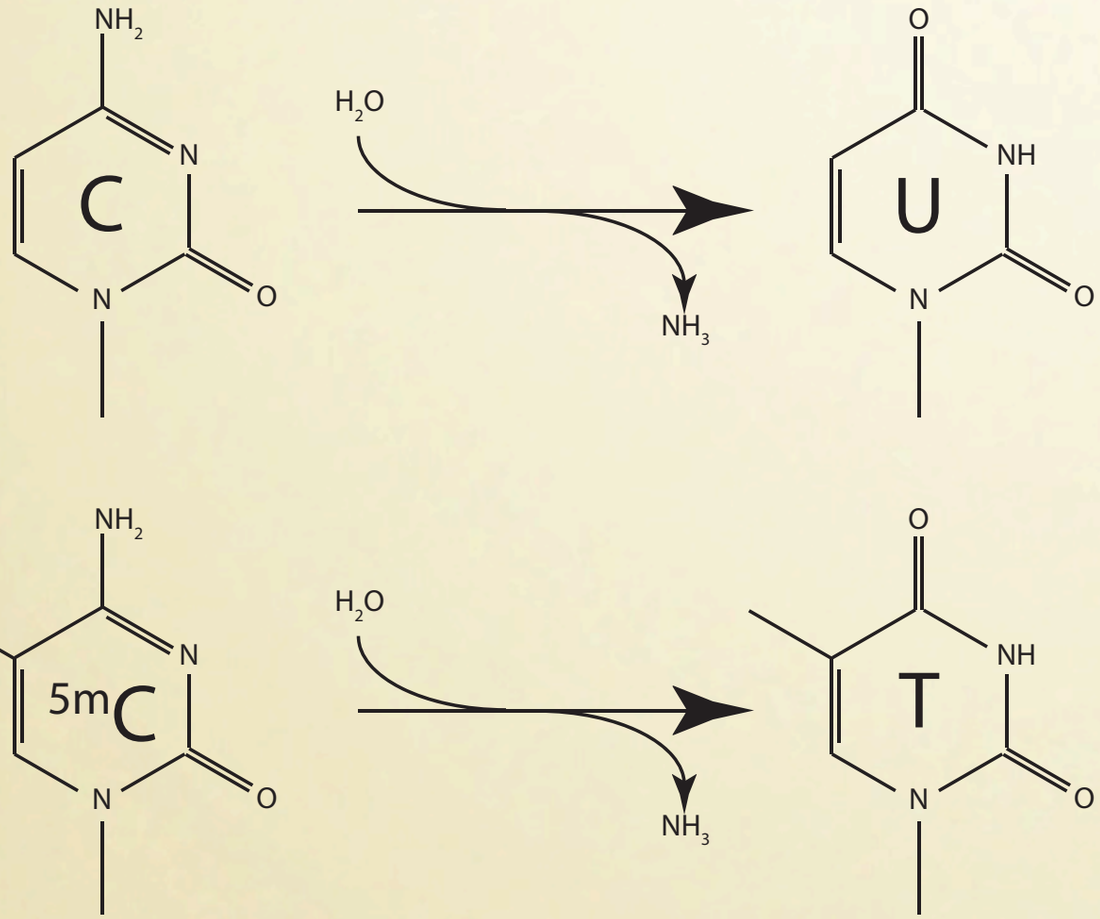


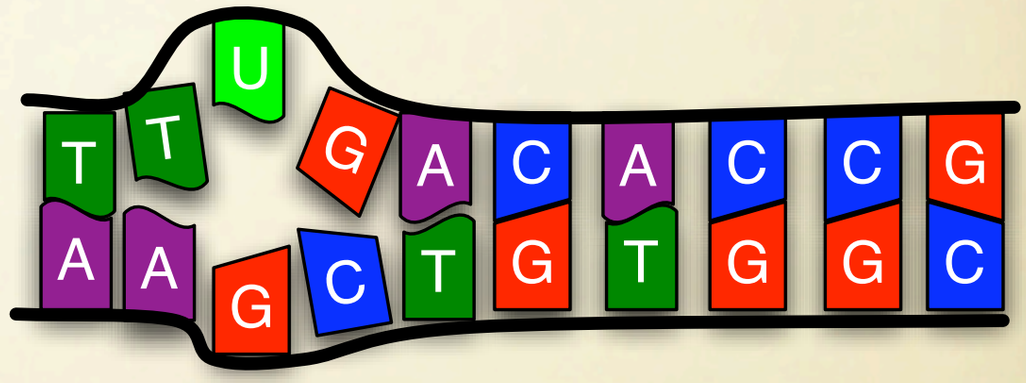




Repair must go back to initial state!

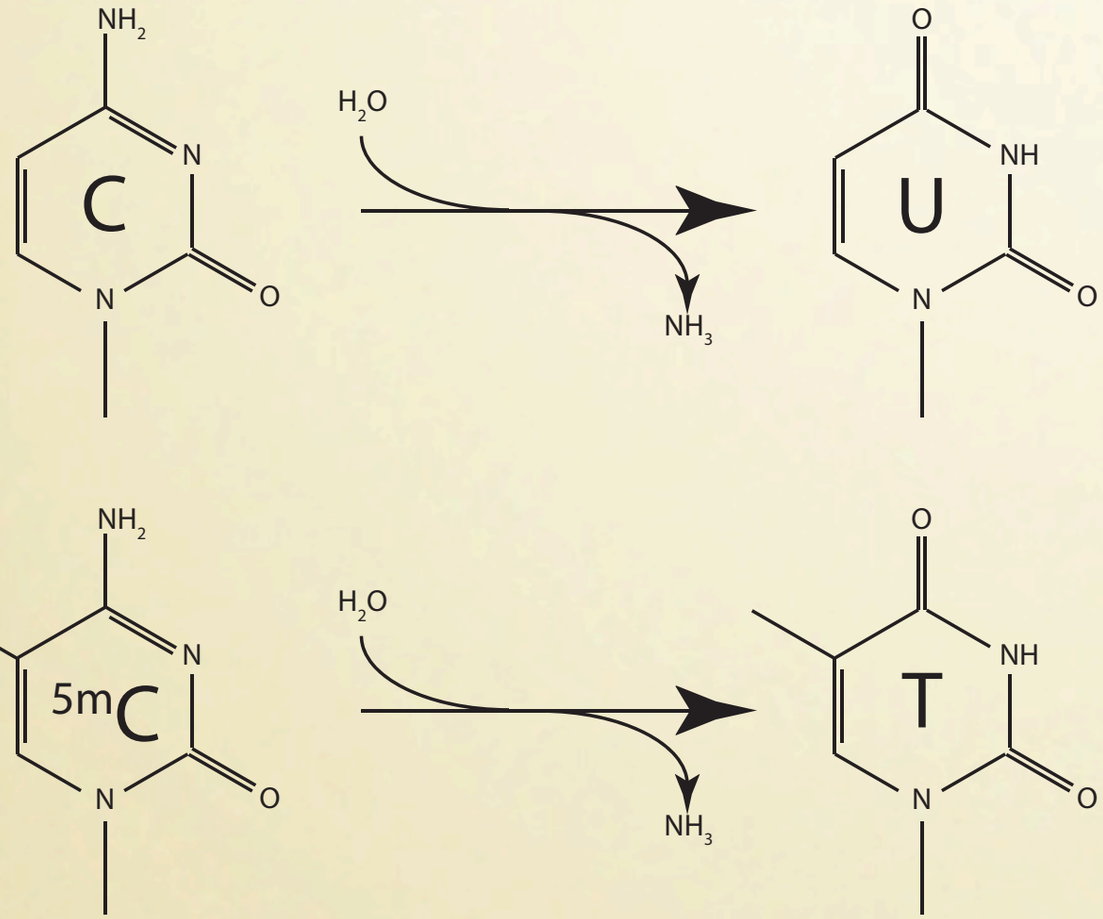
Losing methylation pattern

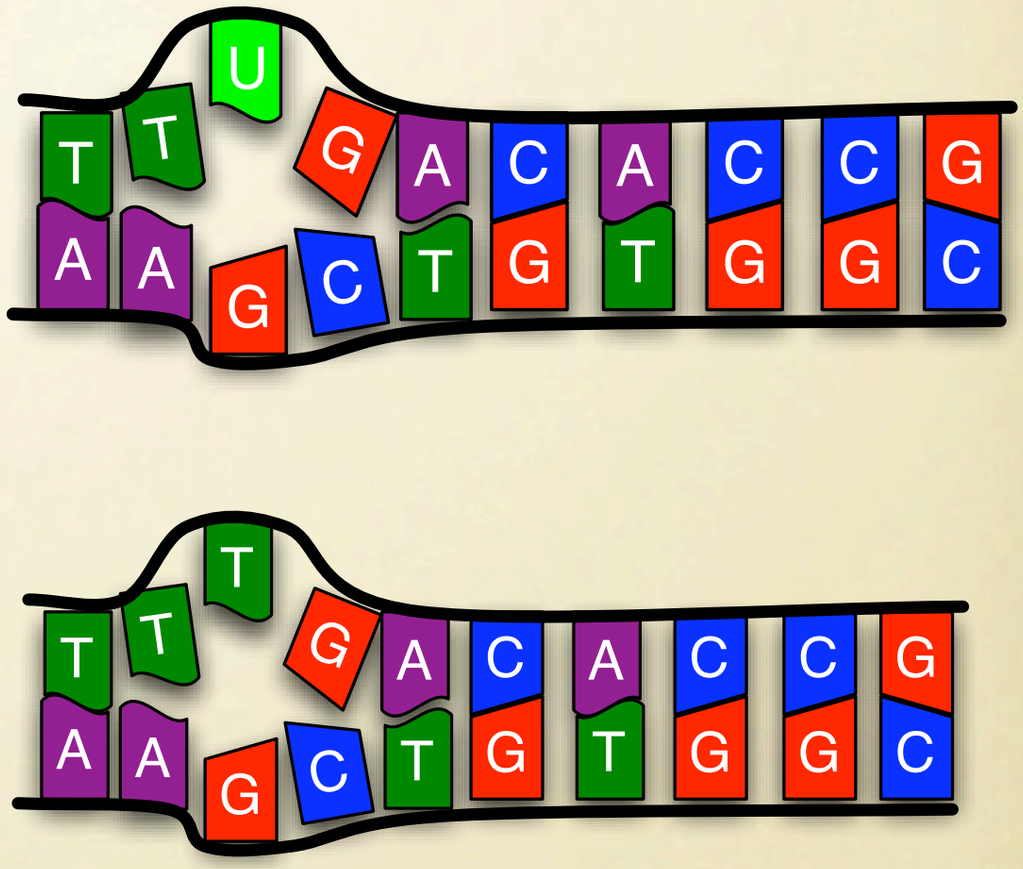




Repair must go back to initial state!

Losing methylation pattern





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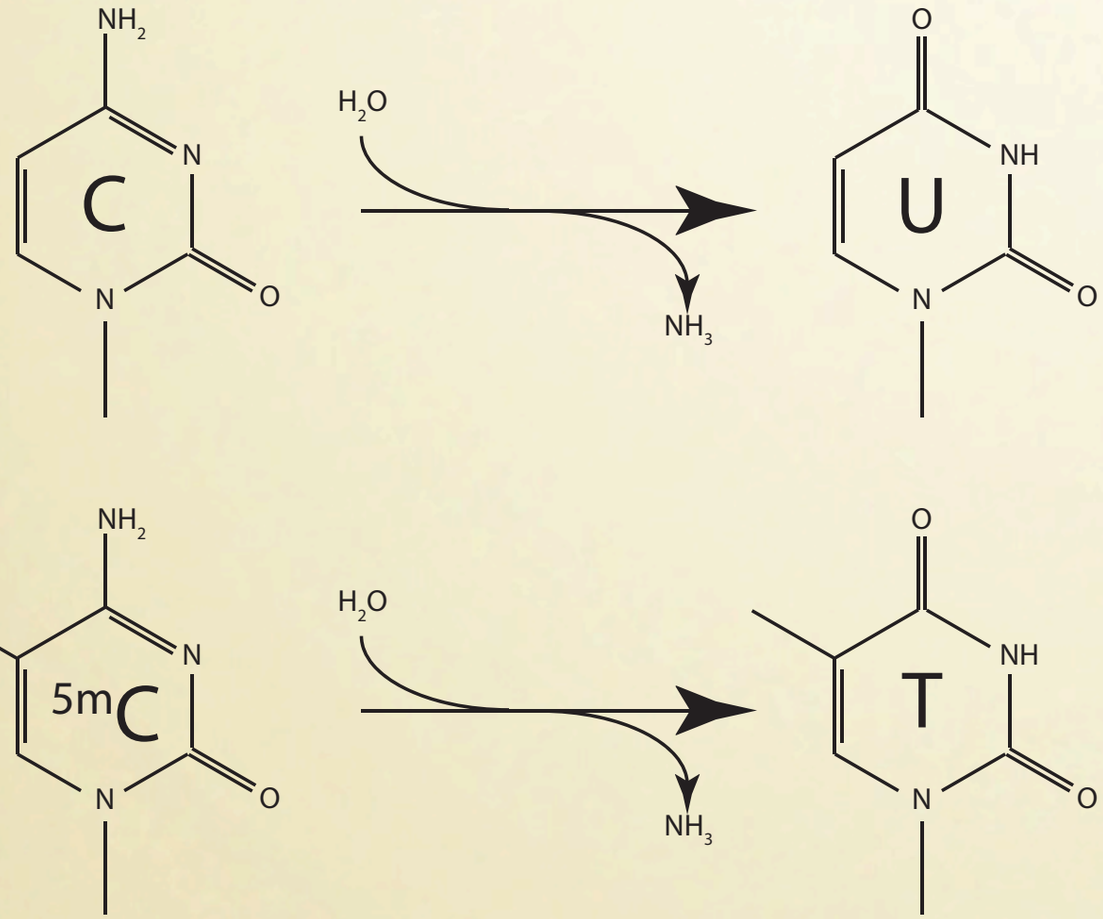

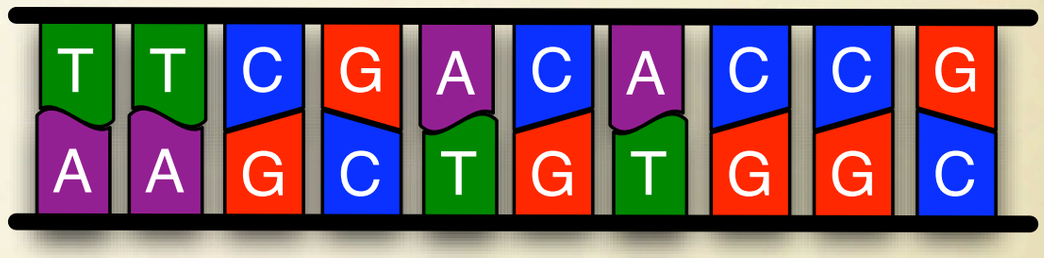

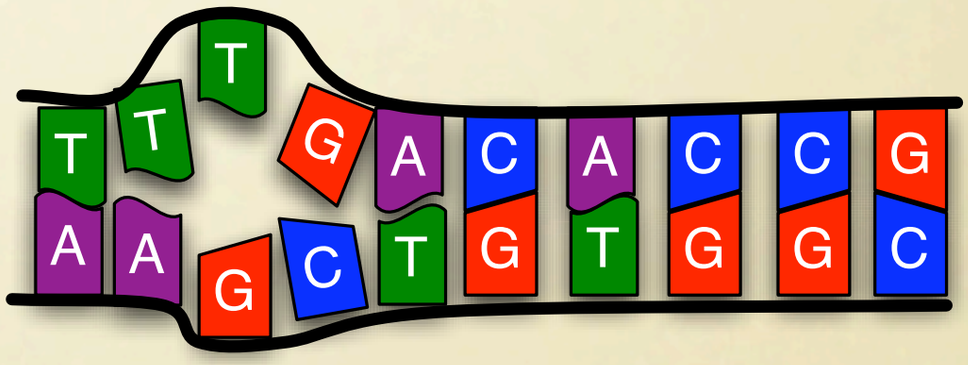

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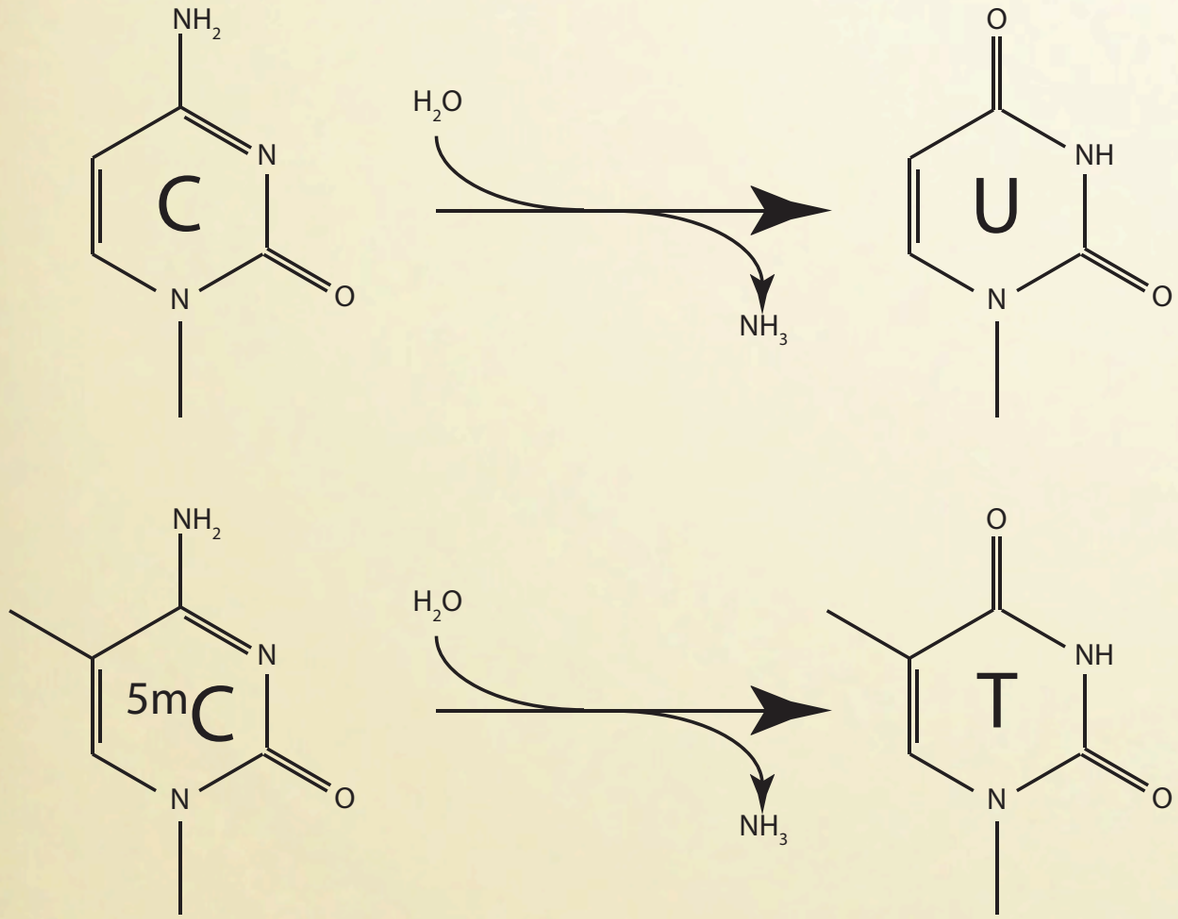
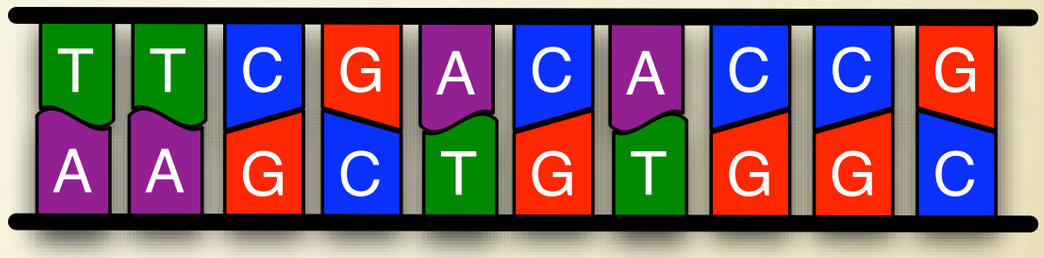
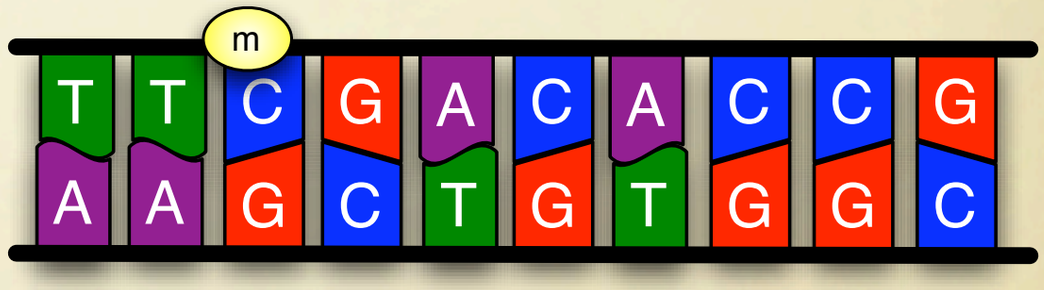
Losing methylation pattern

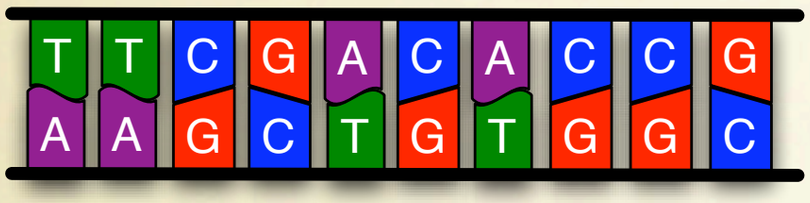


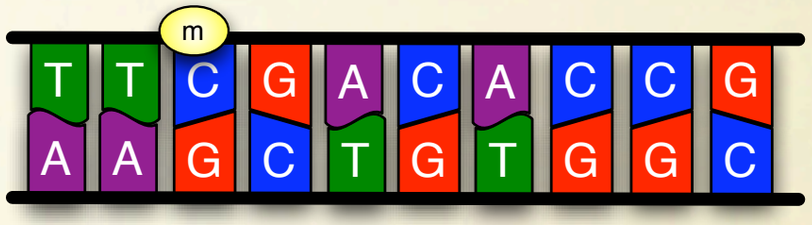




Repair must go back to initial state!

Maintenance





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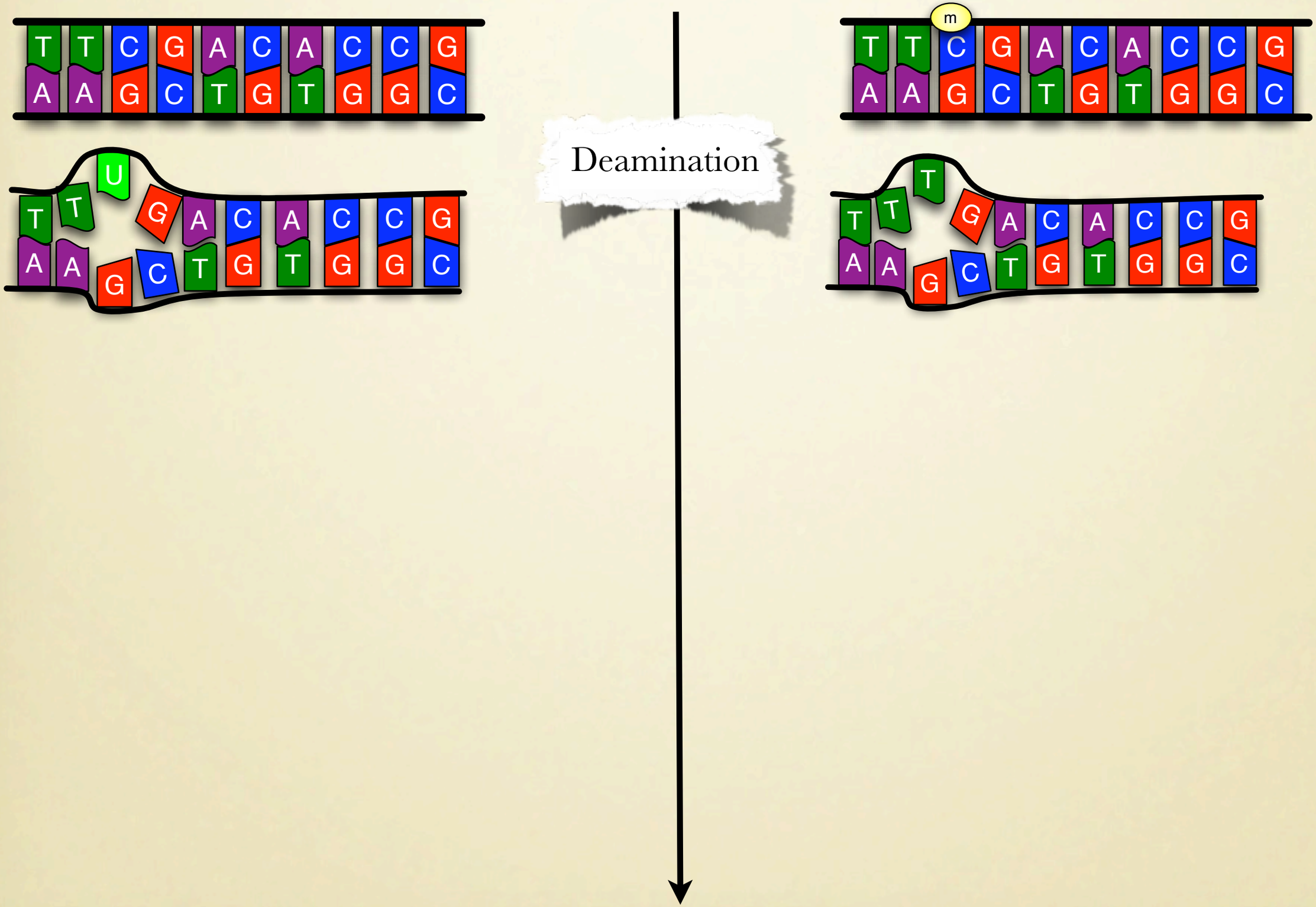

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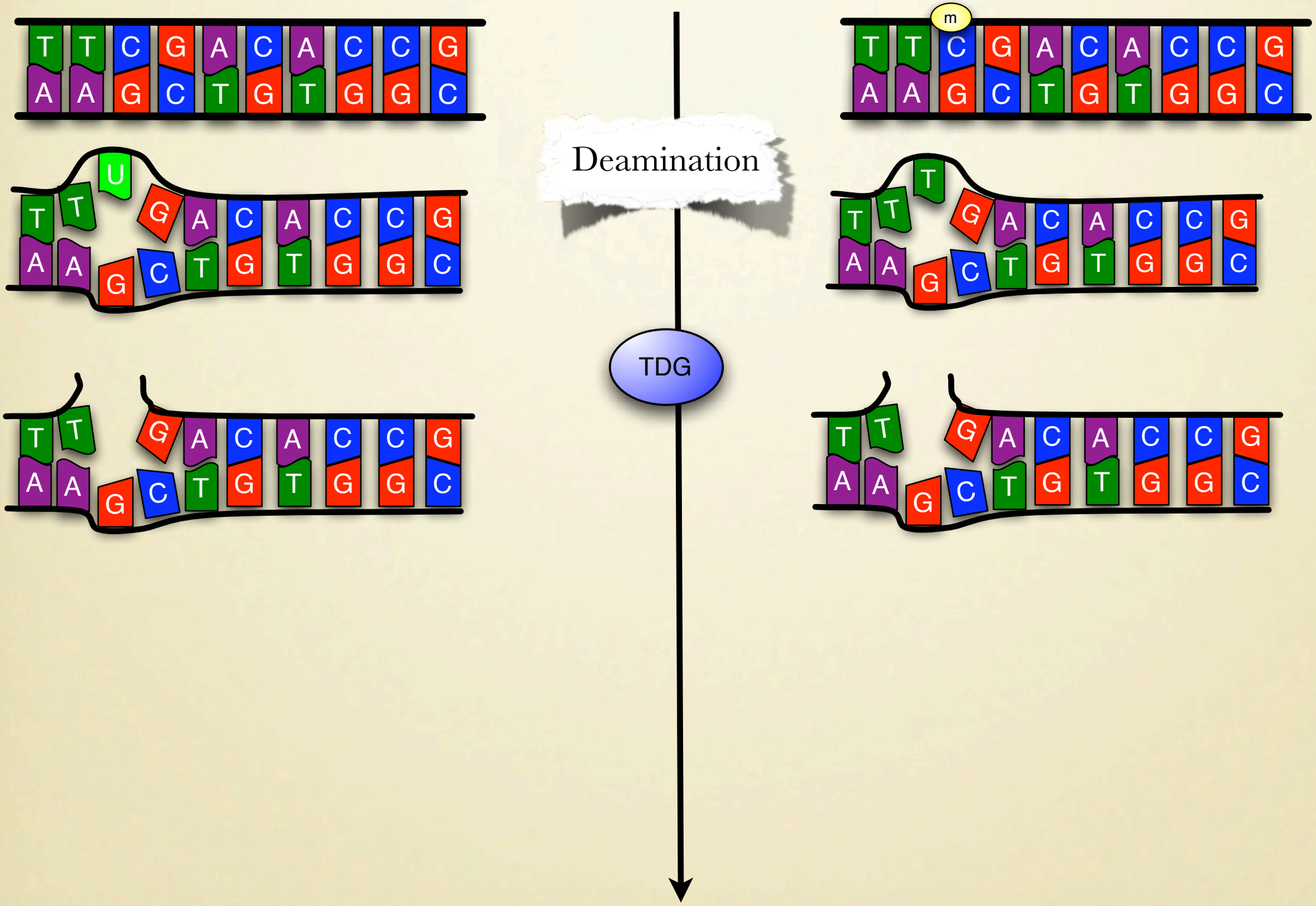

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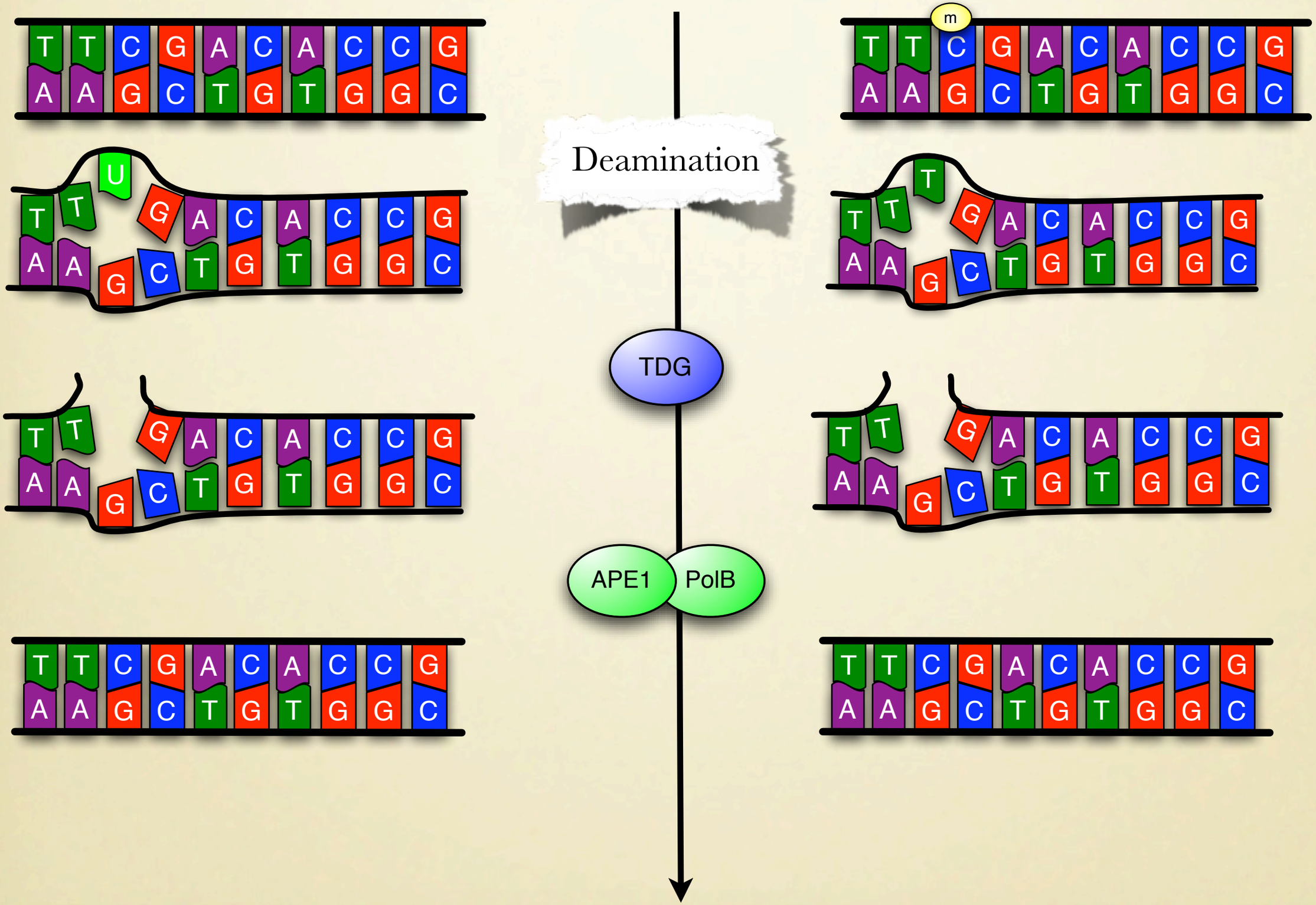

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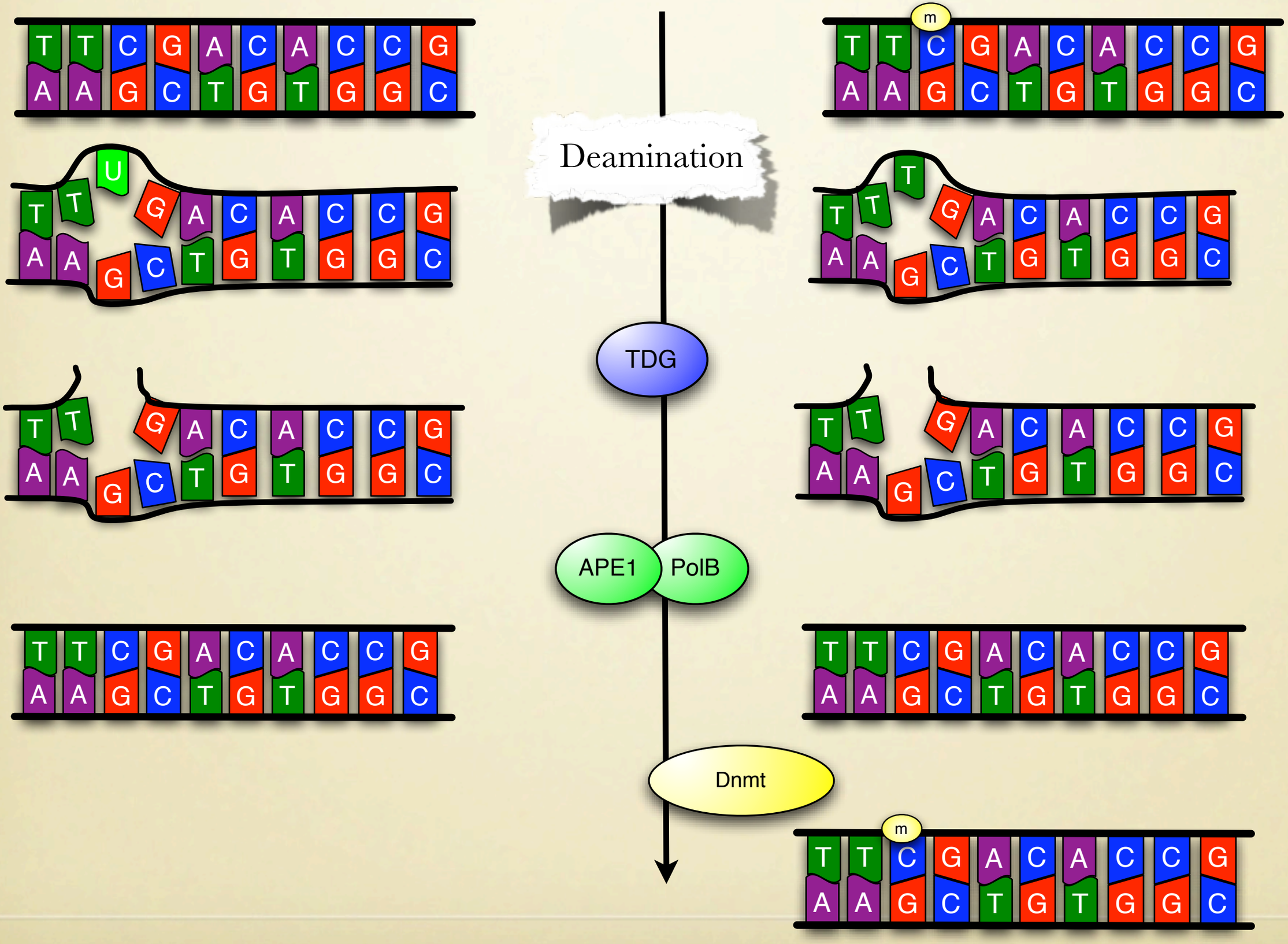

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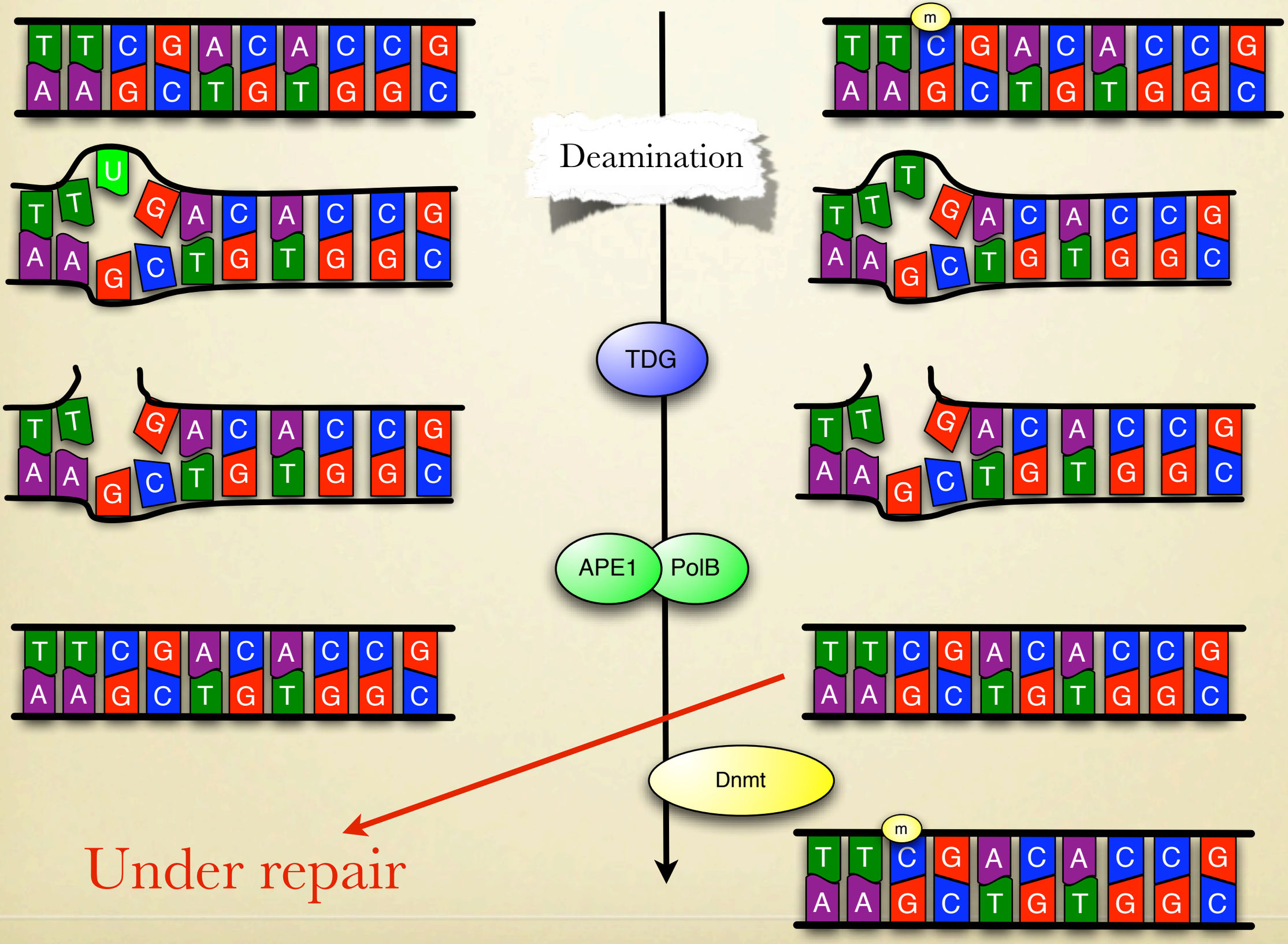

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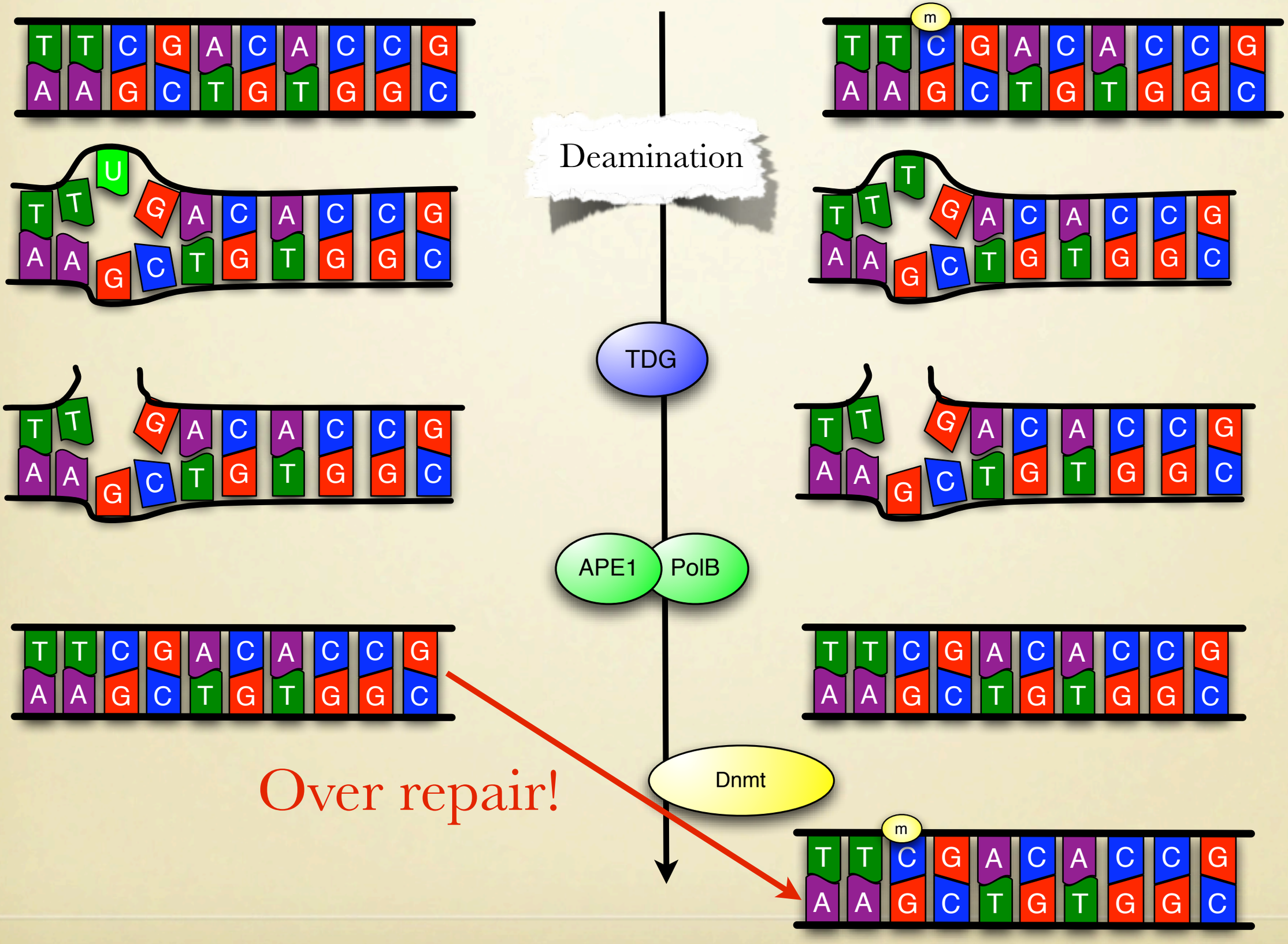

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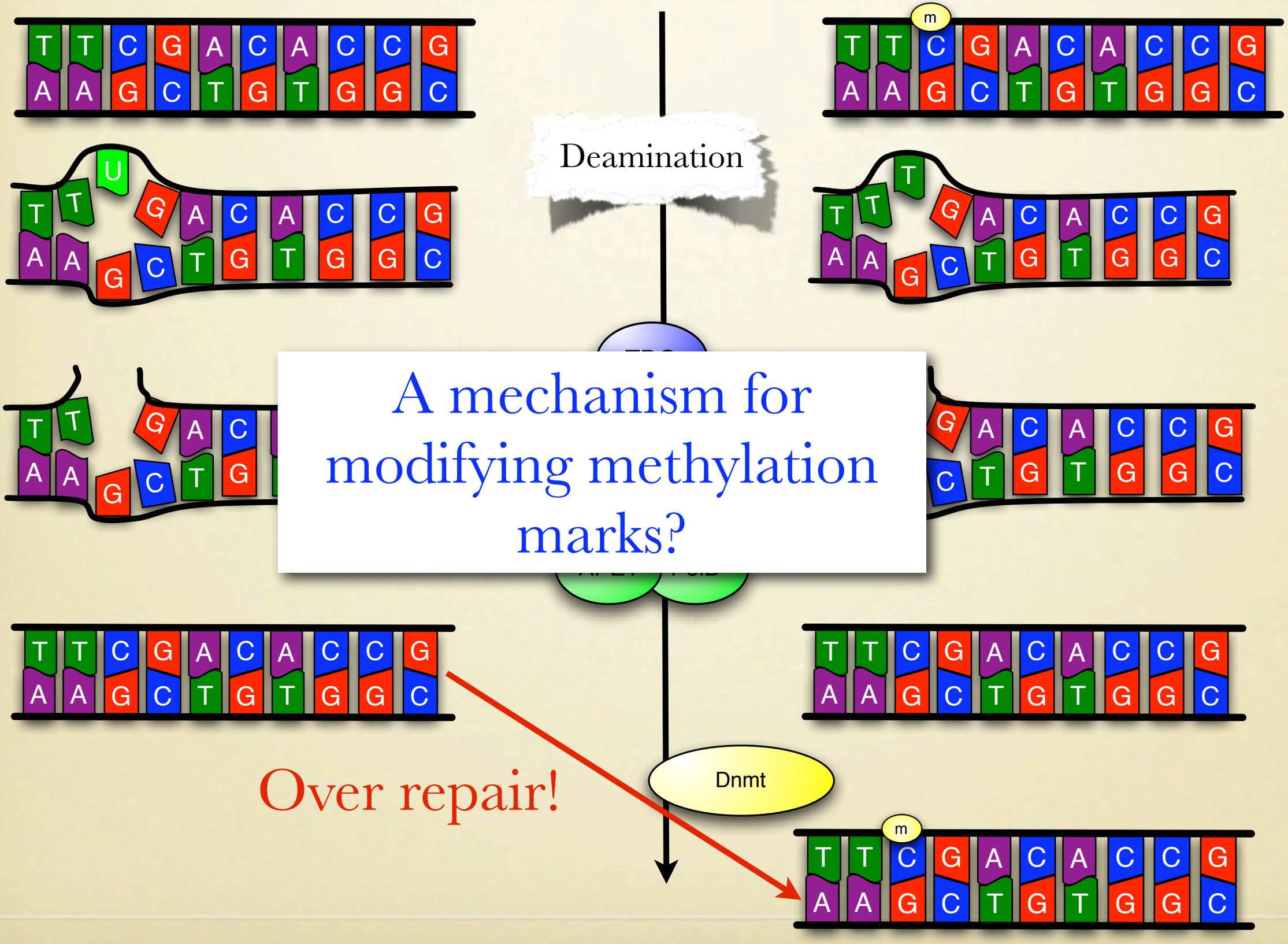

Investigating…

- What is known about these maintenance enzyme?
- How does epigenetic information changes with cell differentiation?

Witness 1

Table 7. **Sequence specificity of the DNA methylation activities of recombinant Dnmt1, Dnmt3a and Dnmt3b.**

	CpG	CpA	CpT	CpC
Dnmt1	272 (100)[1]	0 (0)	7.8 (2.9)	0 (0)
Dnmt3a	263 (100)	10.5 (4.0)	0 (0)	0 (0)
Dnmt3b	195 (100)	13.9 (7.1)	26.8 (13.7)	6.0 (3.1)
No enzyme	0	3.0	0	0

J. Biochem. **133**, 737–744 (2003)
DOI: 10.1093/jb/mvg095

Distinct Enzymatic Properties of Recombinant Mouse DNA Methyltransferases Dnmt3a and Dnmt3b

Isao Suetake, Junko Miyazaki, Chikako Murakami, Hideyuki Takeshima and Shoji Tajima*

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Witness 2

TABLE 1

Kinetic Parameters for hTDG

Substrate	k_{max} [a]	-Fold change relative to CpG·X	-Fold change relative to CpG·T
	Min^{-1}		
G·X			
CpG·T	0.22 ± 0.04	1	1
TpG·T	0.0060 ± 0.0001	0.027	0.027
GpG·T	0.0023 ± 0.0002	0.010	0.010
ApG·T	0.00038 ± 0.00005	0.0017	0.0017
CpG·U	2.6 ± 0.3	1	12
TpG·U	0.79 ± 0.04	0.303	3.6
GpG·U	0.88 ± 0.11	0.340	4.0
ApG·U	0.117 ± 0.003	0.045	0.5
CpG·FU	202 ± 16	1	918
TpG·FU	113 ± 1	0.558	513
GpG·FU	125 ± 11	0.618	568
ApG·FU	18 ± 1	0.089	75
CpG·ClU	120 ± 6	1	546
TpG·ClU	20.9 ± 0.5	0.174	95
GpG·ClU	11.1 ± 0.3	0.093	51
ApG·ClU	1.46 ± 0.15	0.012	6.7
CpG·BrU	11.6 ± 1.0	1	53
TpG·BrU	1.2 ± 0.1	0.106	5.6
GpG·BrU	0.44 ± 0.06	0.038	2.0
ApG·BrU	0.15 ± 0.02	0.013	0.7

Excision of 5-Halogenated Uracils by Human Thymine DNA Glycosylase

ROBUST ACTIVITY FOR DNA CONTEXTS OTHER THAN CpG*

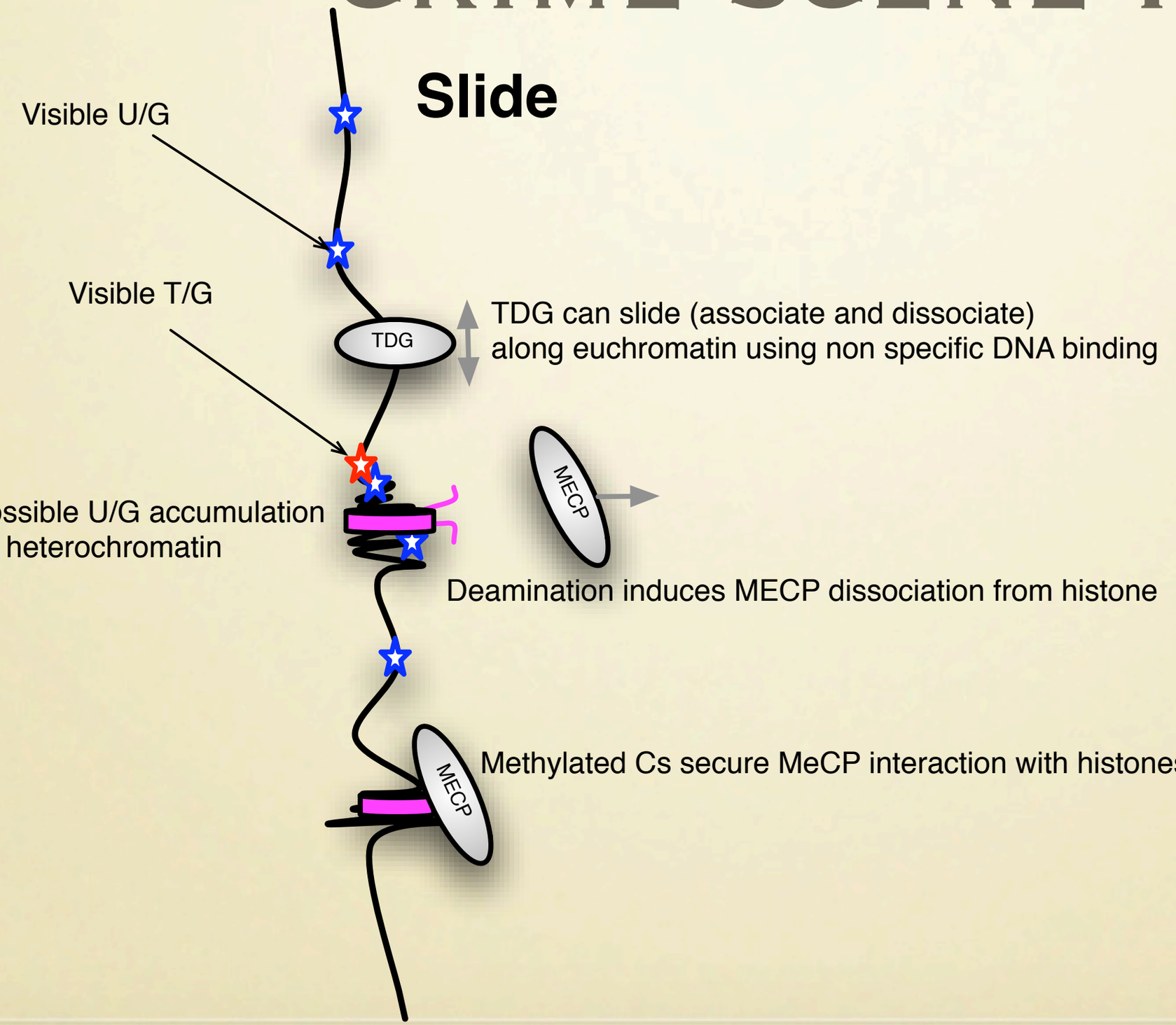
Received for publication, May 23, 2007, and in revised form, June 29, 2007 Published, JBC Papers in Press, June 29, 2007, DOI 10.1074/jbc.M704253200

Michael T. Morgan, Matthew T. Bennett, and Alexander C. Drohat[1]

THE JOURNAL OF BIOLOGICAL CHEMISTRY VOL. 282, NO. 38, pp. 27578–27586, September 21, 2007

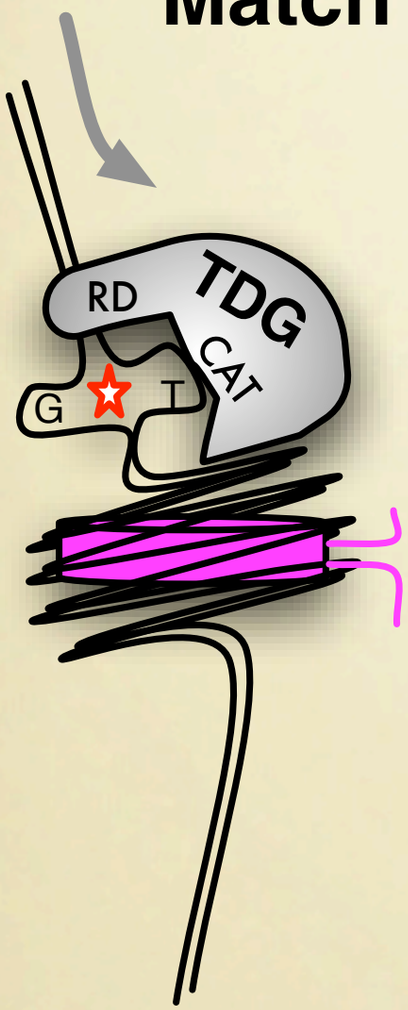
Future work...

Reconstructing a crime scene I



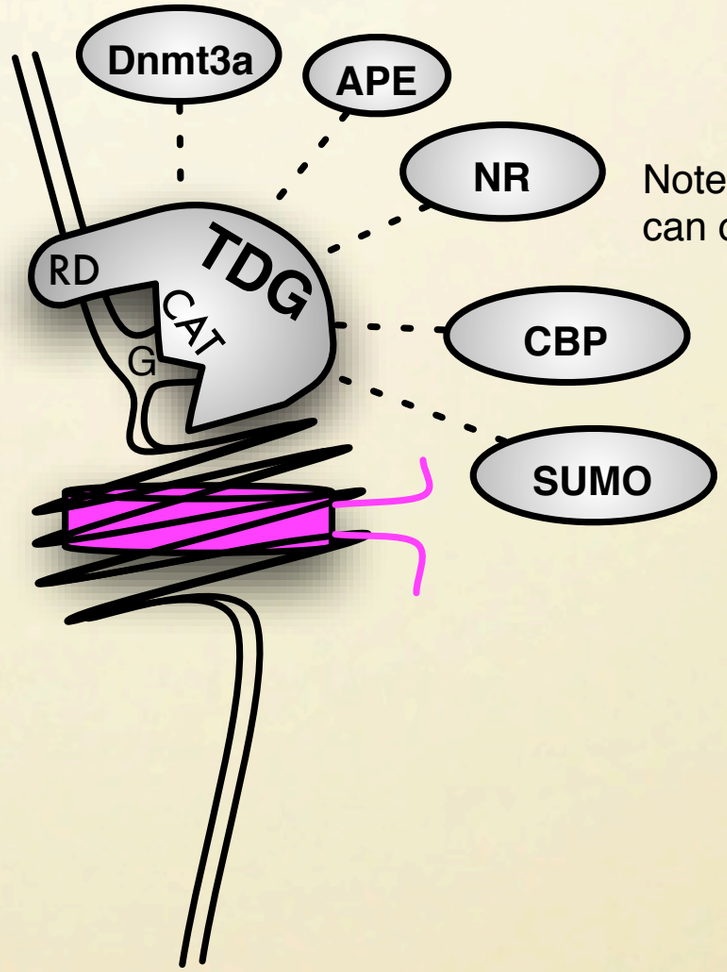
Reconstructing a crime scene II

Match



Upon encountering a G/T mismatch
TDG undergoes a conformational change
which opens its RD arm

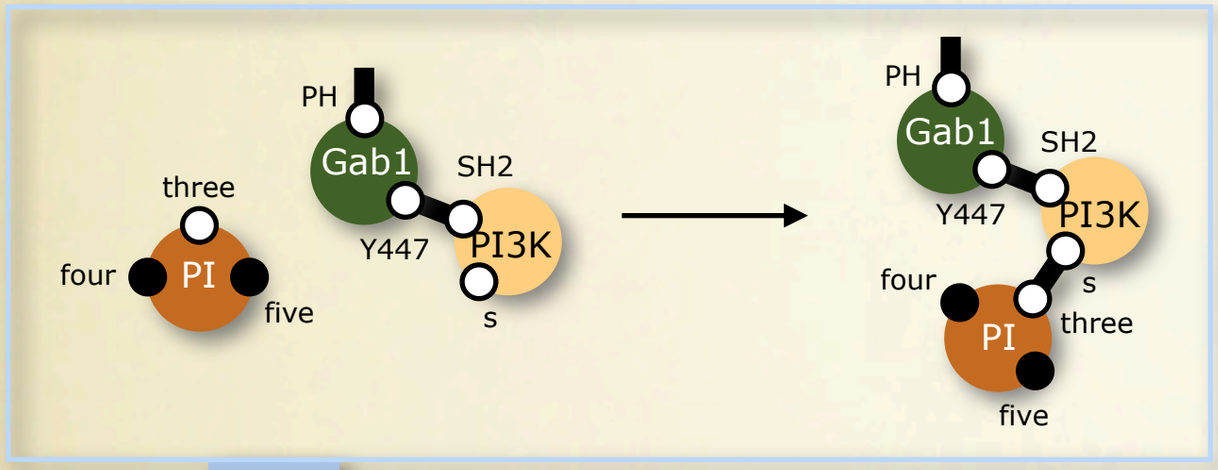
Dock



Note that no repair nor transcription can occur in that state

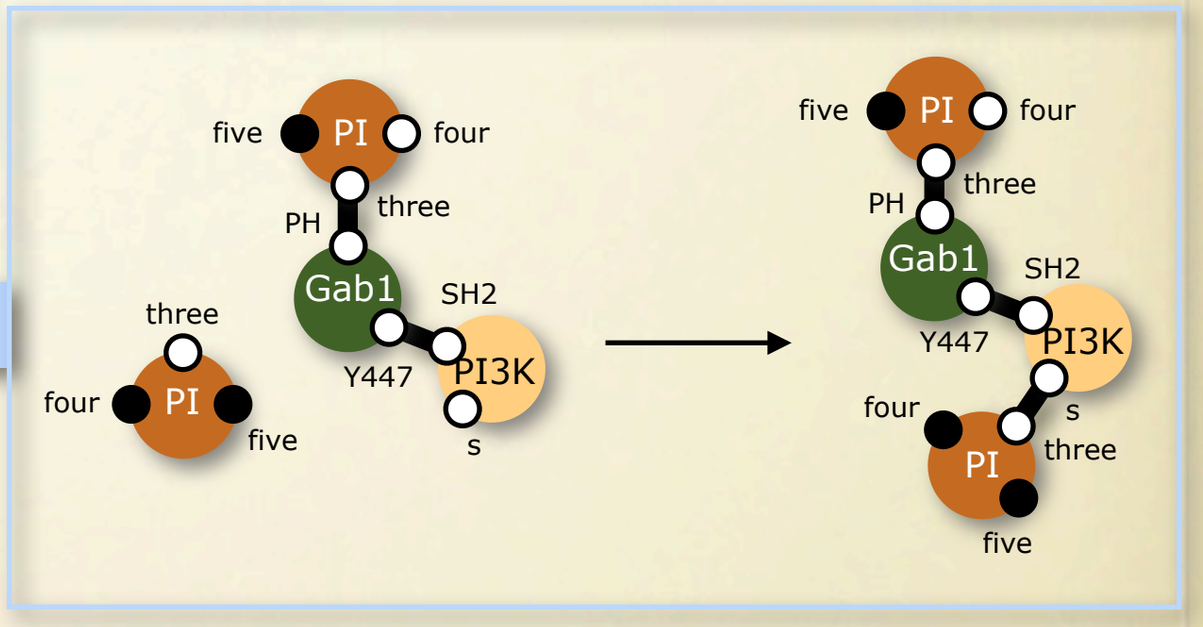
TDG excises the T base and enters a very stable configuration,
via the binding of the catalytic domain to the opposite G base,
where it starts recruiting possible partners for the next steps

Modeling...

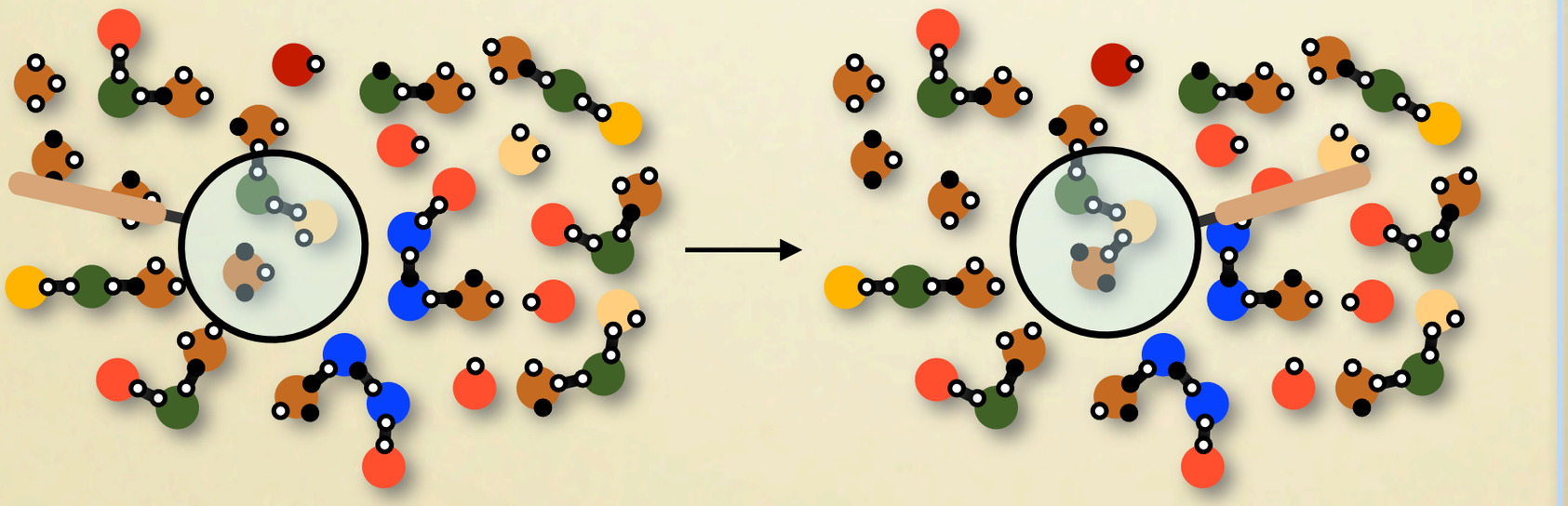


rule

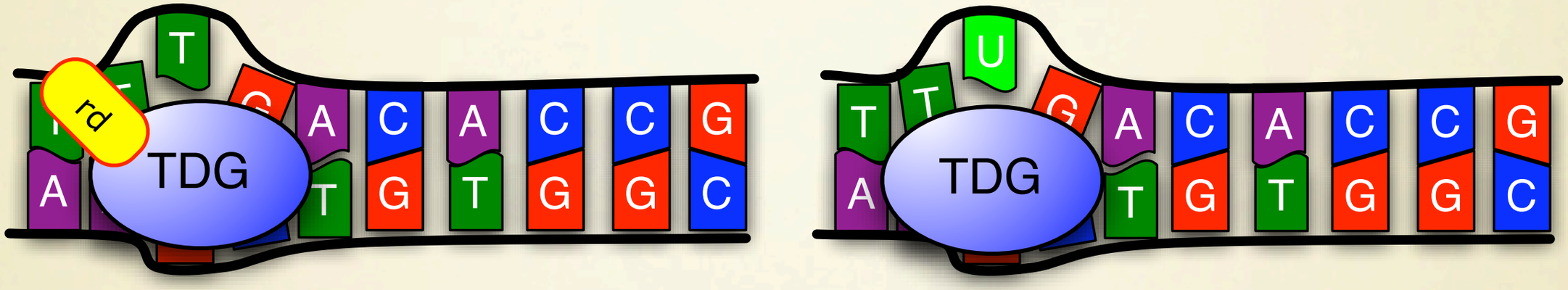
instance



event



Kappa model...



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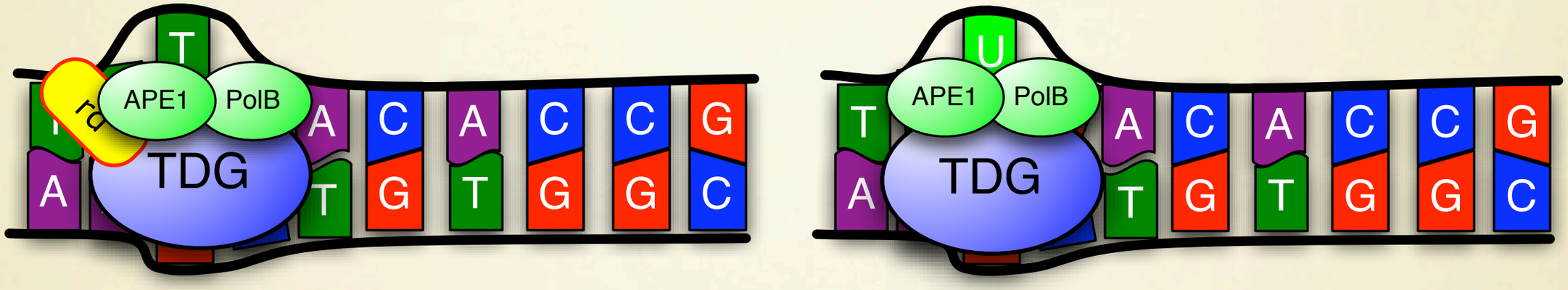
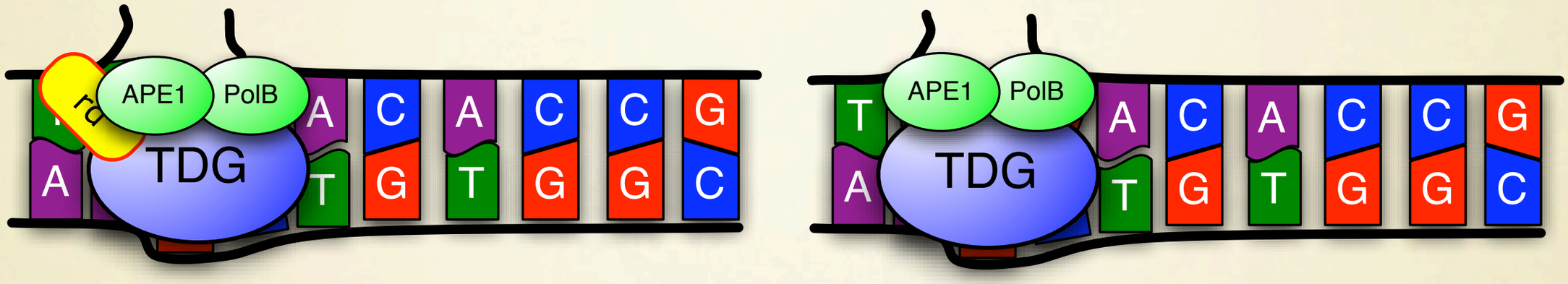
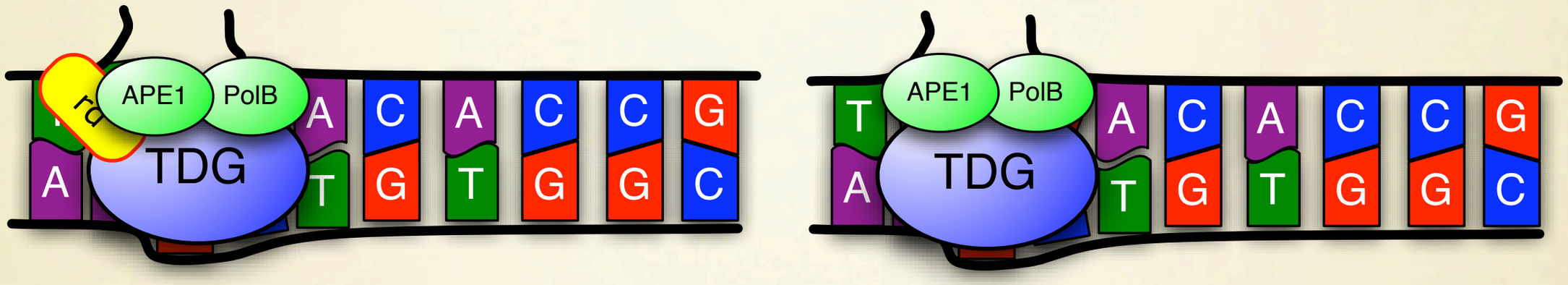

T
rd
APE1
PolB
TDG
A
A C A C C G
T G T G G C
U
APE1
PolB
TDG
T
A
A C A C C G
T G T G G C

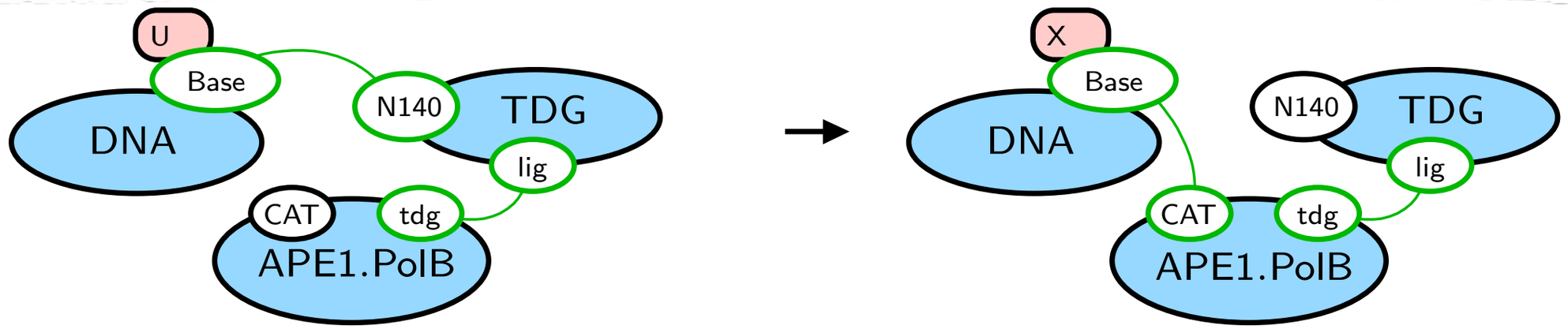
Kappa model...



Kappa model...



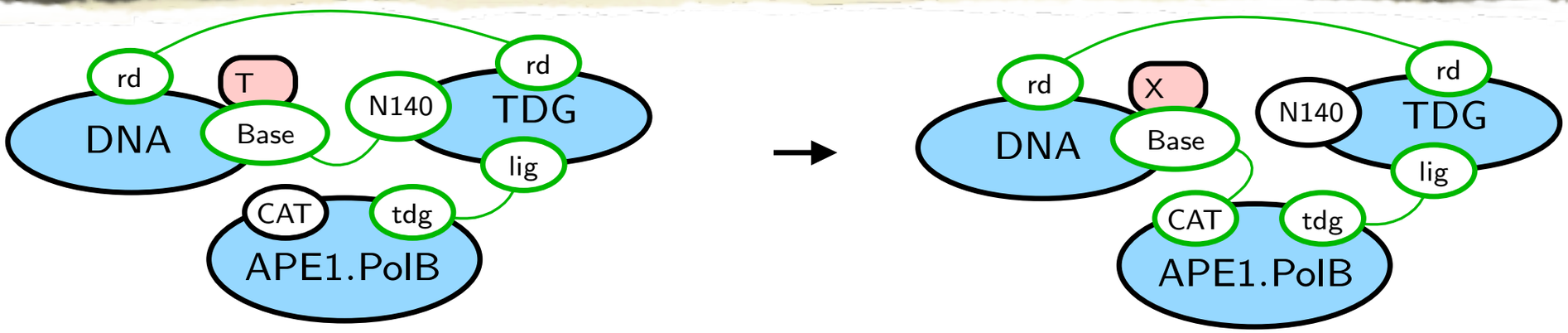
C



'Repair U with swap:'

$\mathrm{DNA}(\mathrm{base}^{0}{\sim}\mathrm{U}),\ \mathrm{TDG}(\mathrm{N140}^{0},\ \mathrm{lig}^{1}),\ \mathrm{APE1.PolB}(\mathrm{tdg}^{1},\ \mathrm{CAT}) \longrightarrow \mathrm{DNA}(\mathrm{base}^{0}{\sim}\mathrm{X}),\ \mathrm{TDG}(\mathrm{N140},\ \mathrm{lig}^{1}),\ \mathrm{APE1.PolB}(\mathrm{tdg}^{1},\ \mathrm{CAT}^{0})$

D



'Repair T with swap:'

$\mathrm{DNA}(\mathrm{base}^{0}{\sim}\mathrm{T},\ \mathrm{rd}^{2}),\ \mathrm{TDG}(\mathrm{N140}^{0},\ \mathrm{lig}^{1},\ \mathrm{rd}^{2}),\ \mathrm{APE1.PolB}(\mathrm{tdg}^{1},\ \mathrm{CAT}) \longrightarrow \mathrm{DNA}(\mathrm{base}^{0}{\sim}\mathrm{X},\ \mathrm{rd}^{2}),\ \mathrm{TDG}(\mathrm{N140},\ \mathrm{lig}^{1},\ \mathrm{rd}^{2}),\ \mathrm{APE1.PolB}(\mathrm{tdg}^{1},\ \mathrm{CAT}^{0})$