

DNA **methyl transferases** Enzyme Proteins **CG** specific **Cytosine C5** methylation :

A. **practical**: ChemScape MDLi  RasMol  MAGE  FireFox applications.

B. Lunch htdocsLocal <http://aris.gusc.lv/ChemFiles/hhaiDNAmethylCtransferKeny/methmast.htm>

RSU Aris Kaksis 2023 experimental research **solutions** :

DNA Methyltransferase DNMT3, DNMT1, HhaI: DNMT3 adds methyl groups to **Cytosine** bases in DNA during development of an organism in different types of cells. DNMT1 adds methyl groups to **Cytosine** bases in newly replicated DNA. HhaI adds methyl groups to DNA in bacterial cells, as part of the restriction/modification system that protects the cell from viruses.

3. **N**-terminus amino acids is Met..... and **C**-terminus amino acid is Tyr.....

What is total number of amino acids on **6MHT** polypeptide chain ...?

5. What four bases are a legitimate **DNA** bases: **G**-.....
C-.....**A**-.....**T**-.....

6. What four bases are a legitimate **RNA** bases: **G**-.....
C-.....**A**-.....**U**-.....

7. What double stranded DNA sequence recognizes DNMT1 DNA m5c-methyltransferase shown as **3PT6**? 5'-.....-3'

8. What double stranded DNA sequence recognizes HhaI DNA m5c-methyltransferase shown as **1MHT**? ? 5'-.....-3'

11. **N**-terminus amino acid is Met..... and **C**-terminus amino acid is Ser.....

What is total number of amino acids on **3PT6** polypeptide chain $1600-651+1=...$?

13. What mechanism methylation endogenous or flip out DNA strand **C cytosine**?

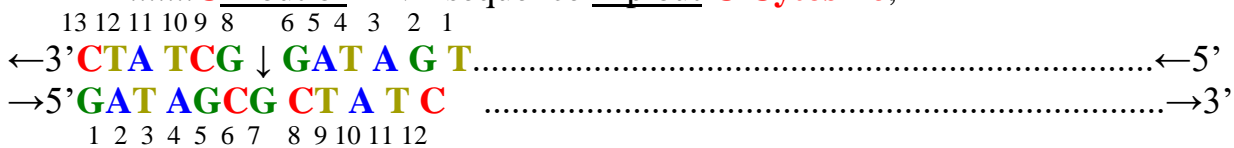
DNA strand

14. What DNA fragment length in **1MHT.pdb** in angstroms? Å Å

14a. What base pairs numbers constitute the DNA fragment **1MHT.pdb**? base pairs +1

14b. Methylation cytosine base nummber**C** of DNA fragment **1MHT.pdb** on sequence :

.....**C** out of DNA sequence flip out **C Cytosine**;



15. What amino acid in before reaction (Equation) bind **cytosine** ⁴²⁷**C** with hydrogen bonds?

Glu..... hydrogen bonds start interaction with target 427

16. What amino acid with a nucleophilic attack on the sixth cytosine carbon C6 start reaction?

Cys..... on the cytosine carbon C.....

2 Nucl. Acids Res. (1997) 25 (14):2773-2783. © 1997 Oxford University Press **6MHT HhaI**
>**6MHT ; 1MHT | P05102** · **MTH1_HAEPH** Haemophilus parahaemolyticus **327 AA**

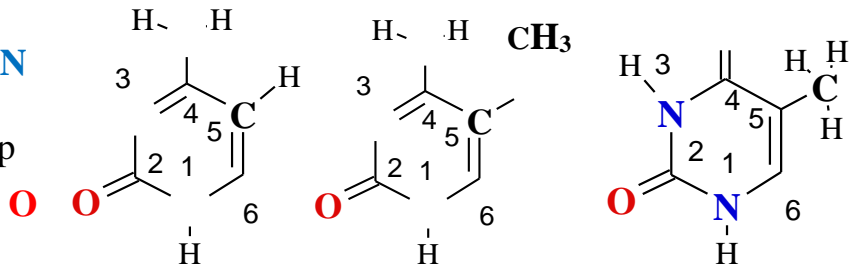
1 60
MIEIKDKQLTGLRFIDLFAGLGGFRLALESCGAECVYSNEWDKYAQEVYEMNFGEKPEGD
61 120
ITQVNEKTIPDHDILCAGFPCQAFSISGKQKGFEDSRGTLFFDIARIVREKKPKVVFEMEN
121 180 DNS
VKNFASHDNGNTLEVVKNTMNELDYSFHAKVLNALDYGIPQKRERIYMICFRNDLNIQNF
181 240
QFPKPFELNTFVKDLLLPDSEVEHLVIDRKDLVMTNQEIEQTTPKTVRLGIVGKGGQGER
241 300
IYSTRGIAITLSAYGGGIFAKTGGYLVNGKTRKLRHPRECARVMGYPDSYKVHPSTSQAAYK
301 327 360
QFGNSVVINVLQYIAYNIGSSLNFKPY

2 Science 25 February 2011: Vol. 331 no. 6020 pp. 1036-1040 **human DNA+3PTA human; DMT1**
P26358 DNMT1_HUMAN; 3PTA **646-1600** , 3PT6 **1600-651**

1 60 70 80
MPARTAPARVPTLAVPAISLPDDVRRRLKDLERDSLTEKECVKEKLNLLHEFLQTEIKNQLCDLETCLRKEELSEEGYLA
81 60 70 160
KVKSLLNKDLSLENGAHAYNREVNGLRENGNQARSEARRVGMADANSPPKPLSKPRTPRRSKSDGEAKPEPSPSPRITRK
161 60 70 240
STRQTTITSHFAKGPAPKRKPQEESEAKSDESIKEEDKDQDEKRRRVTSRERVARPLPAEPPERAKSGTRTEKEEERDEK
241 60 70 320
EEKRLRSQTKEPTPKQKLKEEPPDREARAGVQADEDEDGDEKDEKHKRSQPKDLAAKRRPEEKEPEKVNQIISDEKDEDEK
321 60 70 400
EEKRRKTTPKEPTEKKMARAKTMNSKTHPPKCIQCGQYLDDPDLKYQHPPDAVDEPQMLTNEKLSIFDANESGFESYE
401 60 70 480
ALPQHKLTCFSVYCKHGHLCPIDTGLIEKNIELFFSGSAKPIYDDDPGLEGGVNGKNLGPINEWWITGFDGGEKALIGFS
481 60 70 560
TSFAEYIILMDPSPEYAPIFGLMQEKIYISKIVVEFLQNSDSTYEDLINKIETTVPSPGLNLRFTEDSLLRHAQFVVEQ
561 60 70 640
VESYDEAGDSDEQPIFLTPCMRDLIKLAGVTLGQRRQAARRQTIHSTREKDRGPTKATTTKLVYQIFDTFFAEQIEKDD
641 **646** **651** 60 70 720
REDKEN**NAFKRR**RCGVCEVCQQPECGKCKACKDMVKFGGSGRSKQACQERRCPNMAMKEADDDEEVDDNIPEMPSPKKMHQ
721 60 70 800
GKKKKQKNRISWVGEAVKTDGKKSYYKVCIDAETLEVGDVSVIPDDSSKPLYLARVTALWEDSSNGQMFHAHWFCAG
801 60 70 880
TDTVLGATSDPLELFLVDECEDMQLSYIHSKVKVIYKAPSENWAMEGGMDPESLLEGDDGKTYFYQLWYDQDYARFESPP
881 60 70 960
KTQPTEDNKFKFCVSCARLAEMRQKEIPRVLEQLEDLSRVLYYSATKNGILYRVGDGVYLPPEAFTFNKLSPPVKRPR
961 60 70 1040
KEPVDEDLYPEHYRKYSDYIKGSNLDAPEPYRIGRIKEIFCPKKSNGRPNETDIKIRVNFYRPENTHKSTPASYHADIN
1041 60 70 1120
LLYWSDEEAVVDFKAVQGRCTVEYGEDLPECVQVYSMGGPNRFYFLEAYNAKSKSFEDPPNHARSPGNKGGKGGKGGKGP
1121 60 70 1200
KSQACEPSEPEIEIKLPLKRLTLDVFGCGGLSEGFHQAGISDTLWAIEMWDPAAQAFRLNPGSTVFTEDCNILLKLVMA
1201 60 70 1360
GETTNSRGQRLPQKGDVEMLCGGPPCQGFSGMNRFNRSRTYSKFNLSLVSFLSYCDYRPRFFLENVRNFVSKRSMVL
1361 60 70 1440
KLTLRCLVRMGYQCTFGVLQAGQYGAQTRRRRAIILAAAPGEKLPFPEPLHVFAPRACQLSVVDDKFKVSNITRLSSG
1441 60 70 1520
PFRTITVRDMSDLPEVRNGASALEISYNGEPQSWFQRQLRGAQYQPILRDHICKDMSALVAARMRHIPLAPGSDWRDLF
1521 60 70 1600
NIEVRLSDGTMARKLRYTHHDRKNGRSSSGALRGVCSCEVAGKACDPAARQFNTLIPWCLPHTGNRHNHWAGLYGRLEWD
1601 60 70 1680
GFFSTTVTNPEPMGKQGRVLHPEQHRVSVRECARSQGFPTYRLEFGNILDKHRQVGNVPPPLAKAIGLEIKLCMLAKA
1681 1691 1697
RESASAKIKEEEAAKD
HETNAM SAH **S-ADENOSYL-L-HOMOCYSTEINE** ZN ZINC ION
FORMUL 4 SAH C14 H20 N6 O5 S
FORMUL 5 ZN 4(ZN 2+)

17. Put in **C**, **m5C** **C** and **T**

N N N N N N
 heterocyclic structures nitrogen
 atoms **1N**, **3N**, and side amino group
H₂N-4 at carbon **4C**, oxygen =**O**.
 Put in Methyl group **-CH₃** mc5
 cytosine to carbon **5C**!



(Equation)

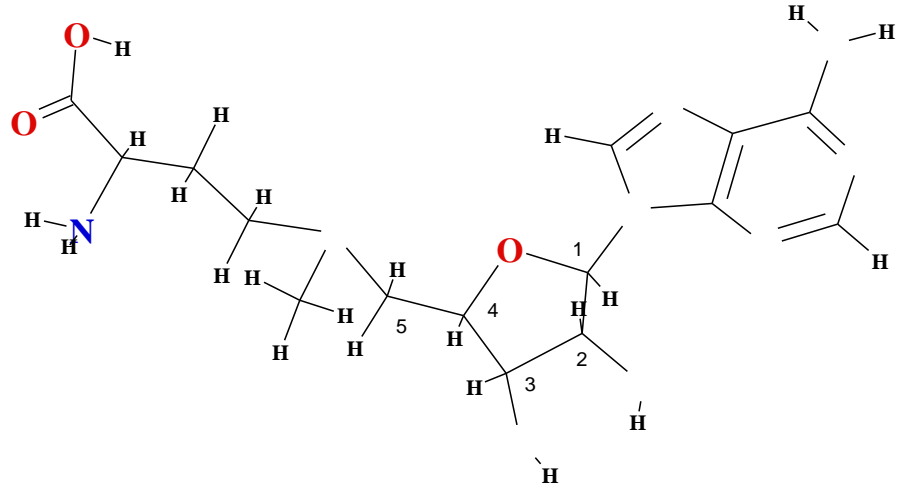
C

m5CC

T

18. Put in **S⁺**-adenosyl-L-
 methionine with short cut
 name AdoMet five **N** atoms
 and methyl sulfur **S⁺**-called
 AdoMet! Put in two ribose
 oxygen atoms at C2 and C3
 of molecule!

O O
N N N N N S⁺

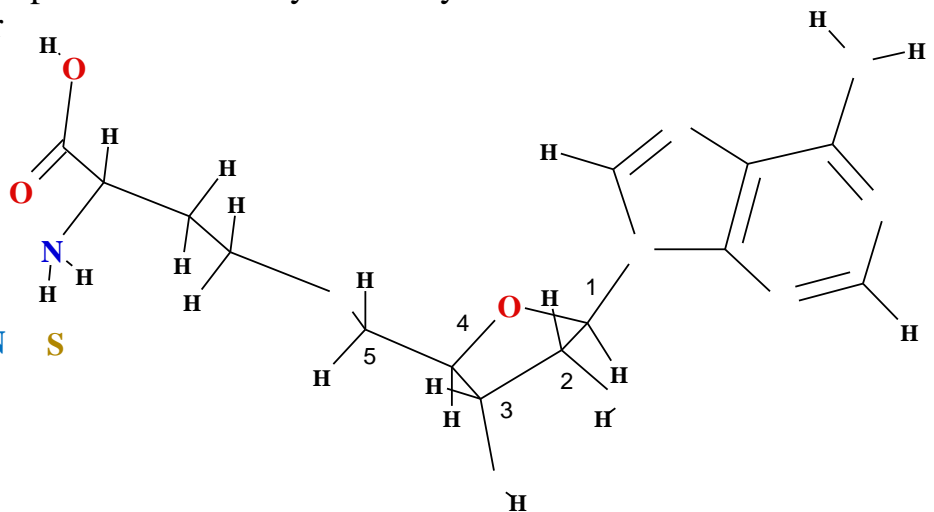


19. Put in **C** and **A** methylation product **S**-adenosyl homo cysteine with short cut name AdoHcy!
 Put in five **N** atoms and sulfur
S-adenosyl homo cysteine
 connected to ribose carbon
 atom C5!

Put in two ribose oxygen
 atoms at C2 and C3 of
 molecule!

N N N N N S

O O



20. Choose on left frame button **Protein** and MDL (select **Mouse click Action**) **distance** to
 measure 3D size of **1MHT.pdb** molecule in angstroms? Å.....ÅÅ

21. What three domains connected by **1MHT-6MHT.pdb** methyl transferase?

connected by.....**domain** and..... **domain**, with.....**region**

22. Between what is located pronounced cleft in **hinge region**?

between.....**domain** and..... **domain**

23. What four **DNA** base pairs are capable bound the cleft in **hinge region**?

cleft is capable binding double stranded **B-DNA** at 5'-.....,,,-3'

3'-.....,,,-5'

24. What type secondary structures dose contains the **large domain** of methyl transferase? ...

..... structures and..... **helices**

25. Choosing by right button “select’ and “Highlight Selection” what **beta sheets** and how many **beta strands** constitute methyl transferases and what position of **hairpin-loop**?
beta sheet with..... **parallel** and **strand with hairpin-loop**.
26. What **alpha helices** constitute methyl transferase molecule? sandwich between.....
alpha helices plus**beta strand as butter** between **second****alpha helices**
and..... **alpha helix** thatin front of the **beta sheet core**
27. What type secondary 2° structures dose contains the **small domain** of methyl transferase?
..... as **sheet**.
28. What **beta sheets** and how many **beta strands** constitute methyl transferase molecule?
.....**beta strands** are configured in an anti parallel formation.
and are arranged in a circular formation that resembles a (spirālveida ritenis)
29. What secondary structures dose contains the **hinge region** of methyl transferase?
hinge region is composed of an structure
that connectsand..... **domains**
30. What conserved motifs comprise **hinge region** of methyltransferase?
hinge region is comprised withand..... conserved motifs
the first half motif IX**domain** second half X locates in**domain**
31. What conserved motifs labeled **variable region** spans the entire length of the methyl
transferase?..... **region** betweenandmotifs
32. What two units within **variable region** between VIII and IX motifs of methyl transferase
responsible for sequence 5'-**GCGC**-3’ specific recognition and?
target base selection recognitionto**sub region**
33. What twenty six amino acids bind AdoMet in site a **hydrophobic pocket**?.....
5AA: Phe.....,Ala.....,Gly.....,Leu.....,Gly....., 11: Gly.....,Phe.....,Ala.....,Trp.....,
Pro.....,Gly.....,Ile.....,Ile.....,Cys.....,Ala.....,Gly....., 10: Phe.....,Pro.....,Gly.....,
Leu.....,Phe.....,Phe.....,Val.....,Val.....,Val.....,Val.....

38. What type methyltransferase adds **methyl** groups to **Citozins** bases in DNA in bacterial cells, as part of the restriction/modification system that protects the cell from viruses?

.....-1MHT.pd,6MHT.pdb

39. What type methyltransferase adds **methyl** groups to **Cytosine** bases in DNA in newly replicated DNA. These **methyl** groups are important for replicating controlling genes in different types of newly reproducing cells copies?

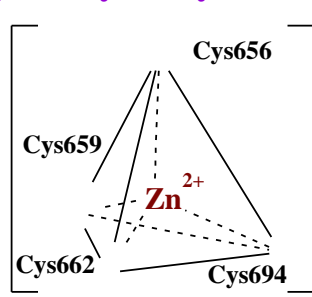
.....-3PT6.pdb.
40. What type methyltransferase adds **methyl** groups to **Cytosine** bases in DNA during development of an organism cell. These **methyl** groups are important for **epigenetic** controlling genes in differentiation development of cell types?

Fresh replicate DNA methylase DNMT1 PDB 3PT6

41. What four amino acids in **zinc(II)** ion finger coordination sphere **Zn2** and **Zn4** 3PT6.pdb?

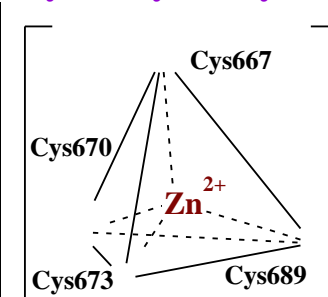
Cys.....Cys.....Cys.....Cys.....; Zn4 Cys.....Cys.....Cys.....Cys.....

42. Put in **Zn²⁺**
coordinative Geometry
Tetragon Nr2 with 4 sulfur
S⁻ ions and complex charge!
3PT6Zn2



S⁻ S⁻ S⁻ S⁻ 2-

43. Put in **Zn²⁺**
coordinative
Geometry Tetragon
Nr4 with 4 sulfur **S⁻**
ions and complex
charge! **3PT6Zn4**

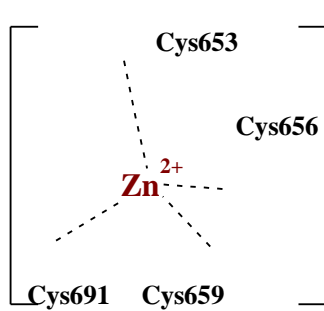


S⁻ S⁻ S⁻ S⁻ 2-

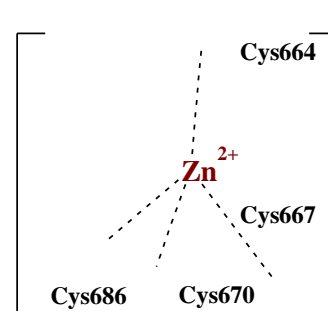
44. What four amino acids in **zinc(II)** ion finger coordination sphere **Zn2** and **Zn5** 3PTA.pdb?

Cys.....Cys.....Cys.....Cys.....; Zn5 Cys.....Cys.....Cys.....Cys.....

45. Put in **Zn²⁺**
coordinative Geometry
Tetragon Nr2 with
4 sulfur **S⁻** ions and
complex charge!
3PTAZn2



46. Put in **Zn²⁺**
coordinative Geometry
Tetragon Nr5 with
4 sulfur **S⁻** ions and
complex charge!
3PTAZn5

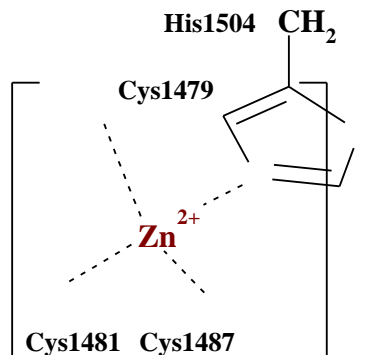


47. What four amino acids in **zinc(II)** ion finger coordination sphere **Zn1702-1703** 4DA4.pdb?

Zn1702 Cys.....Cys.....Cys.....His.....;

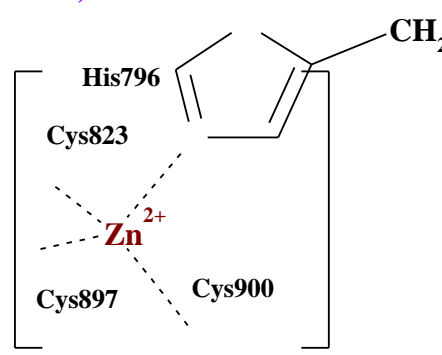
; Zn²⁺ 1703 HIS.....Cys.....Cys.....Cys.....

48. Put in **Zn²⁺**
coordinative
Geometry
Tetragon Nr2 with
3 sulfur **S⁻** ions,
His1504 two **N** atoms
and complex charge!
4DA4Zn1702



S⁻ S⁻ S⁻ N N 1-

49 Put in **Zn²⁺**
coordinative
Geometry Tetragon
Nr2 with
3 sulfur **S⁻** ions,
His796 two **N** atoms
and complex
charge!**4DA4Zn1703**



S⁻ S⁻ S⁻ N N 1-