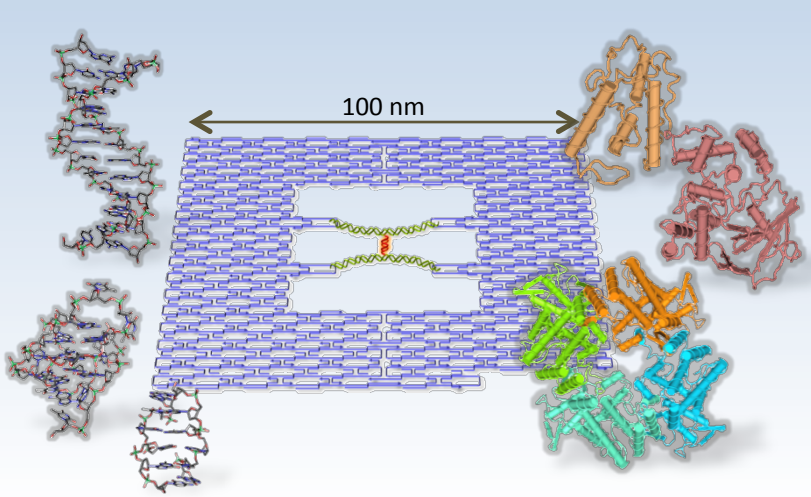
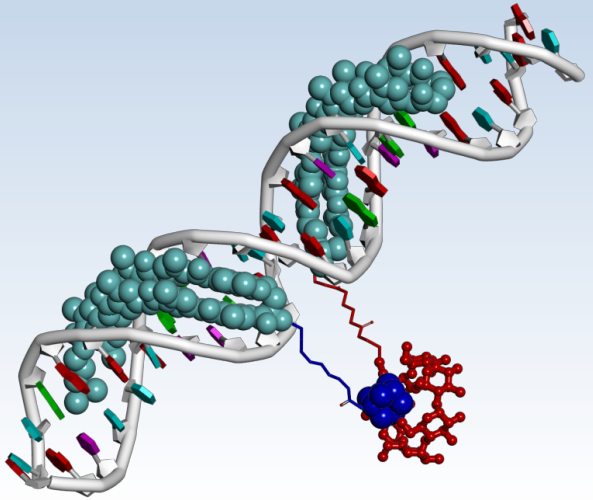
生体分子機能論 2018-9
エピジェネティックス1

Advanced Course in Molecular Biology and Biochemistry





Hiroshi Sugiyama
Department of Chemistry, Graduate School of Science
Institute for Integrated Cell-Material Sciences (iCeMS)
Kyoto University

Basics

1 Basic elements of nucleic acids and their synthesis
2 Sequencing of DNA
3 3D structure of DNA 1
4 3D structure of DNA 2

Chemistry

5) DNA alkylation
6) Hydrogen abstraction 1
7) Hydrogen abstraction 2
8) Charge transfer

Biology

9) Epigenetics 1
10) Epigenetics 2
11) ATRX

遺伝子発現の階層的な制御

１）ジェネティクス制御（塩基配列特異的結合）

２）エピジェネティックス制御

エピジェネティックス：

DNAの塩基配列に依存しない遺伝子発現制御

Gene Regulation in Prokaryotes.

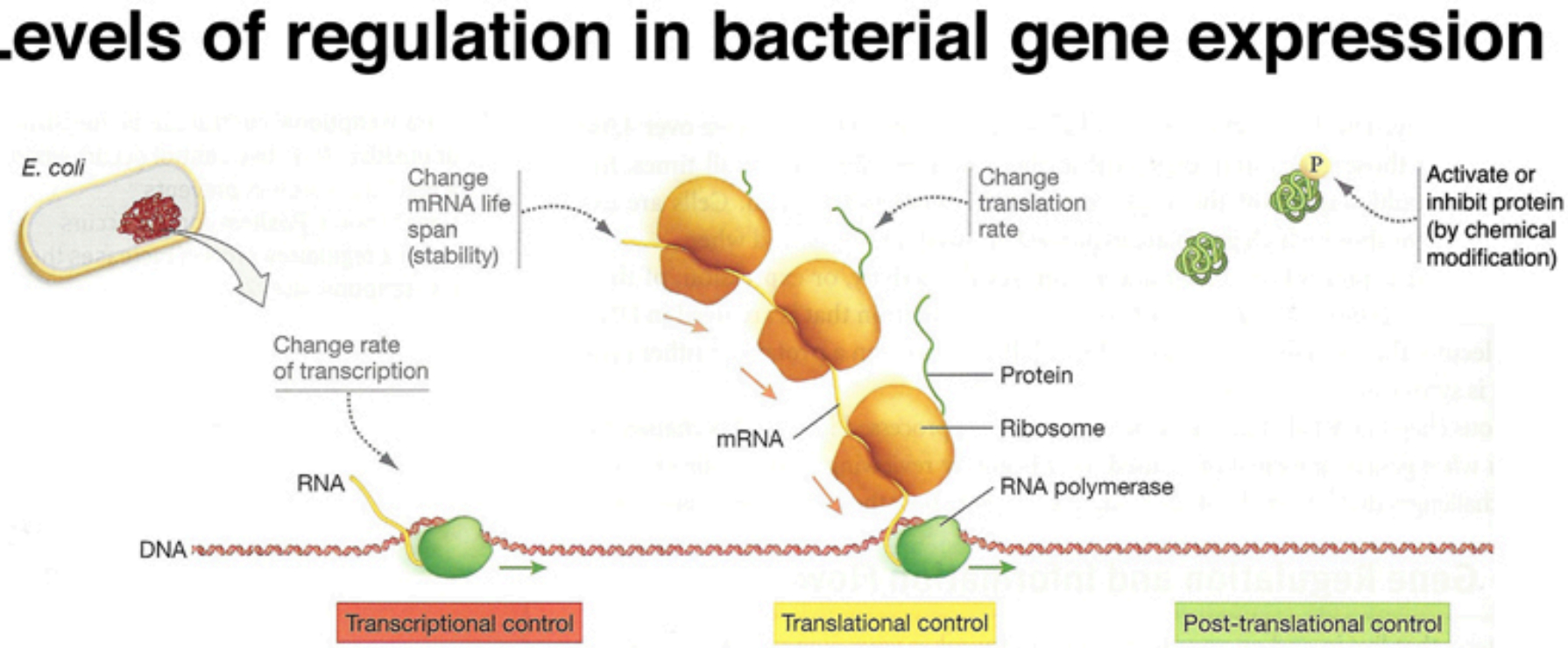
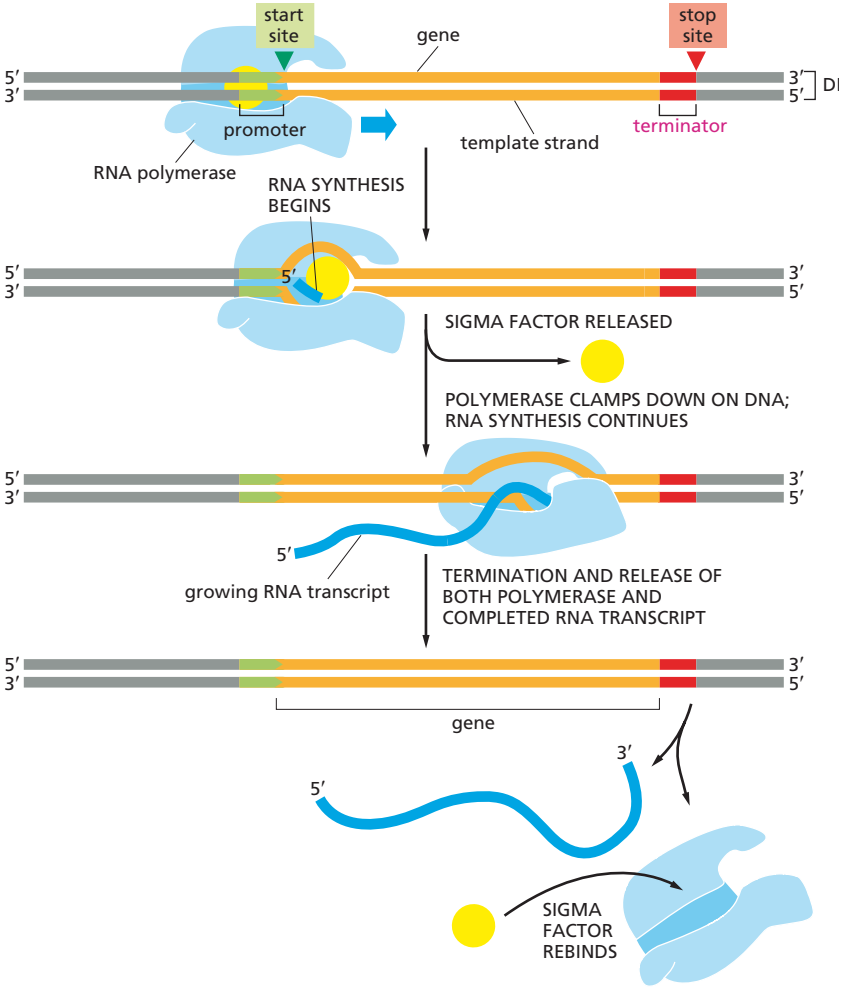


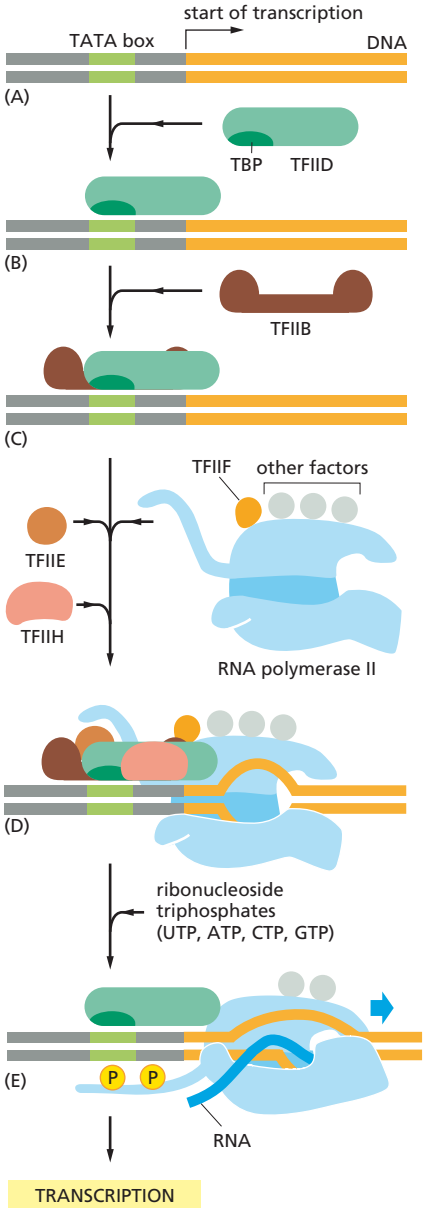
FIGURE 17.1 Gene Expression in Bacteria Can Be Regulated at Three Levels.

http://www.discoveryandinnovation.com/BIOL202/notes/lecture16.html

Transcription



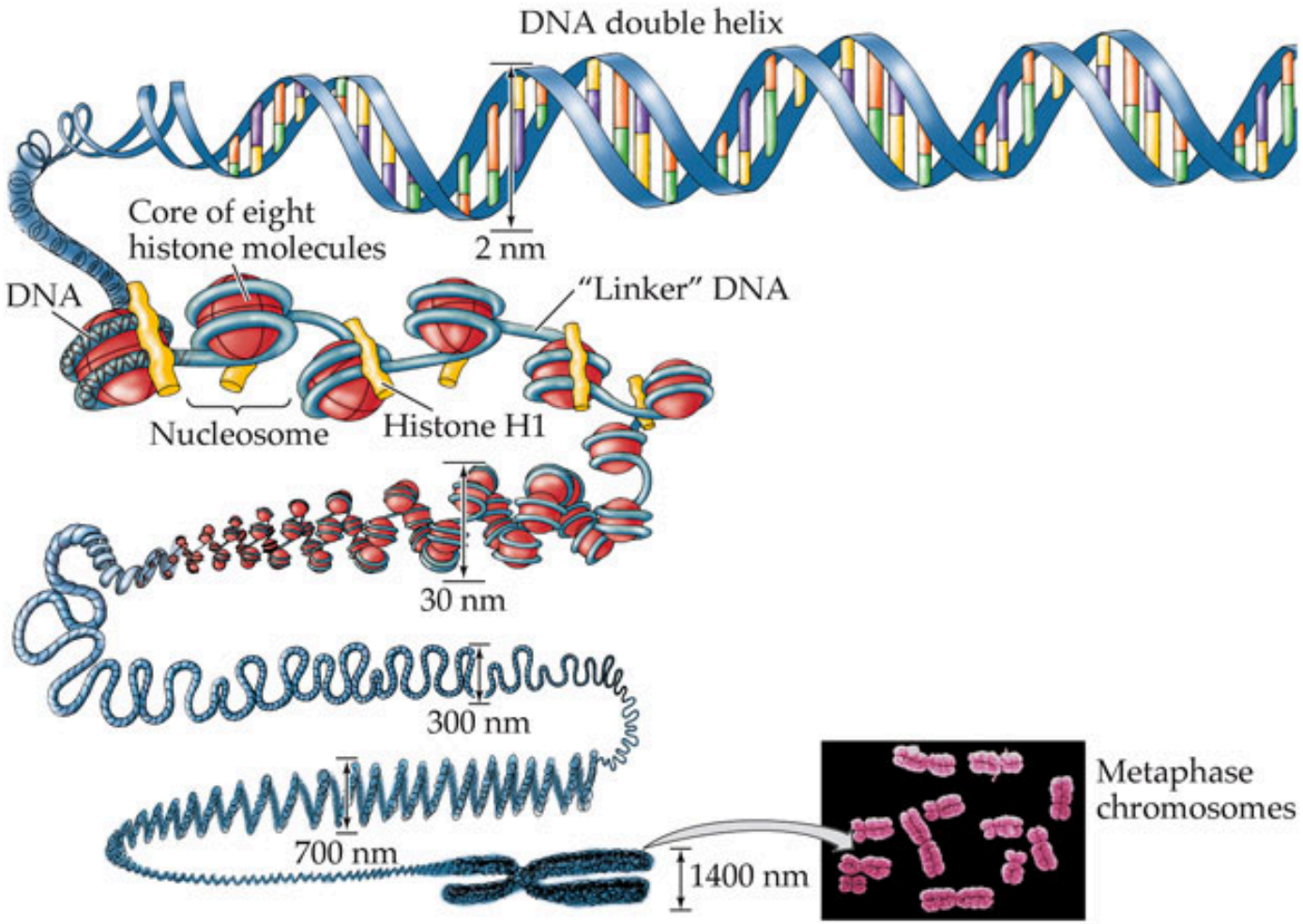
Bacteria



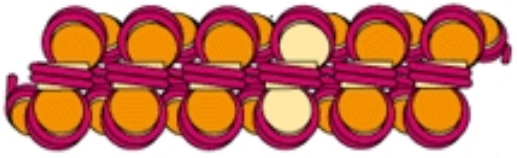
Eukaryote

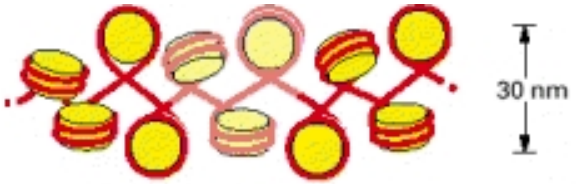
Gene Regulation in Eukaryotes

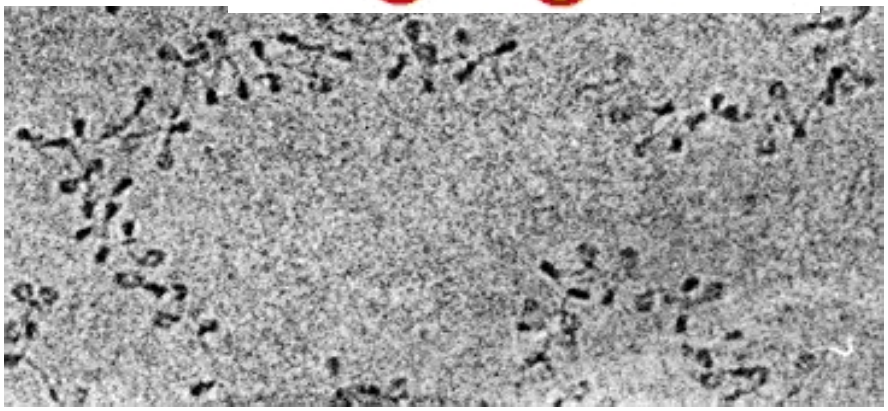
Eukaryotic gene expression is so complicated process and it is tightly regulated by complex epigenetics, and also at the transcriptional, post-transcriptional, translational, and post-translational levels.



Chromatin Structures

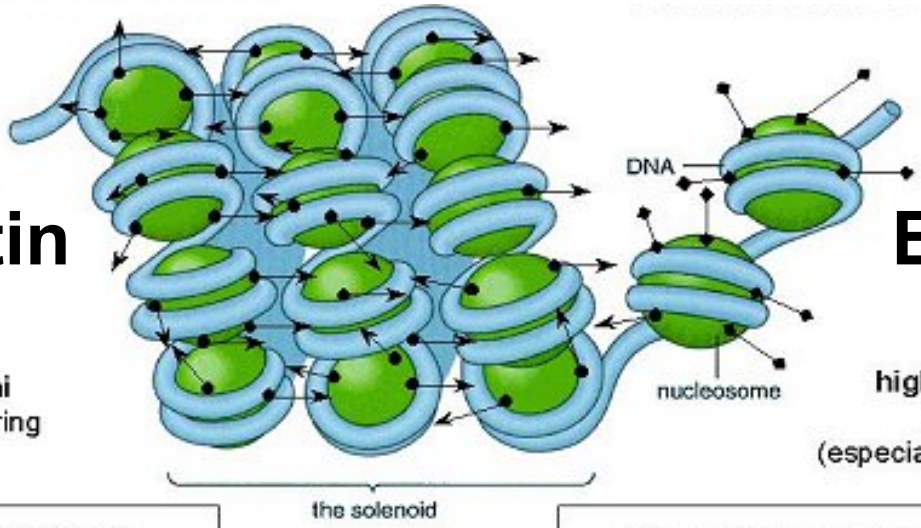






Closed Heterochromatin

Open Euchromatin



(+) charged N termini
(bind DNA on neigboring nucleosomes)

highly acetylated core histones
(especially H3 and H4)

- HIGH level of histone H1
- NO gene transcription

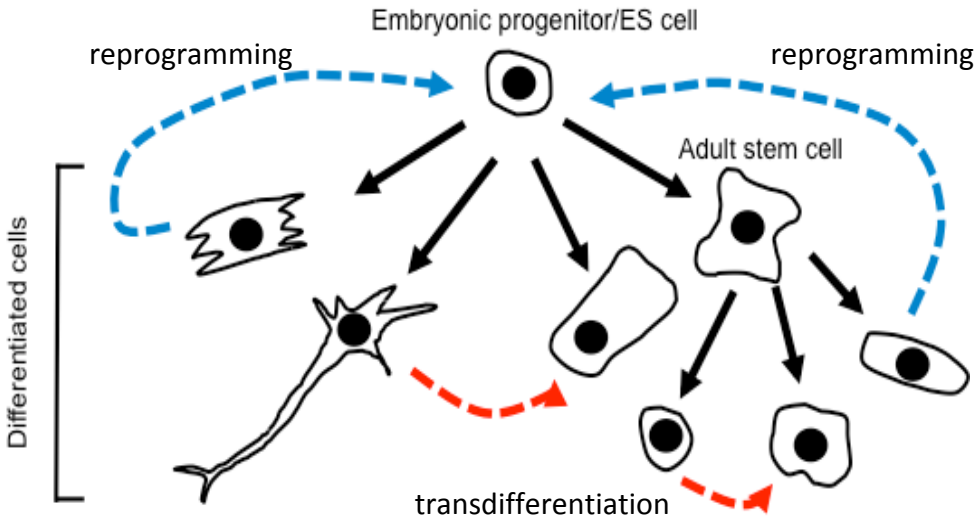
- Reduced level of histone H1
- Gene transcription possible

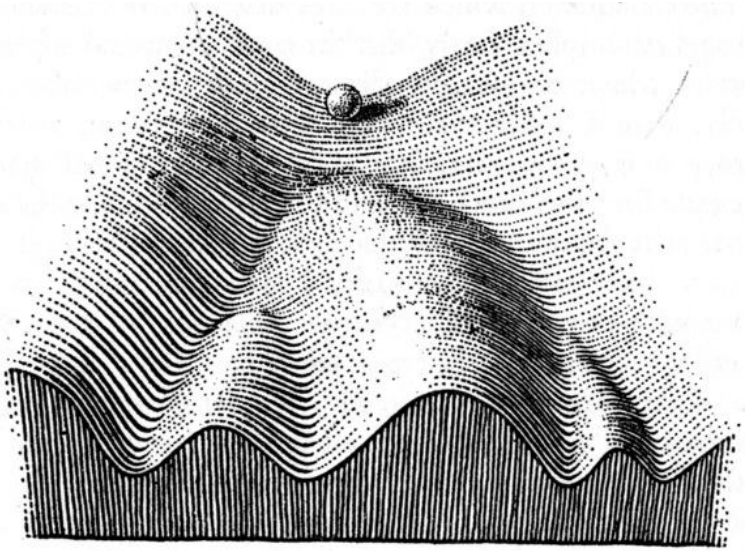
http://sgi.bls.umkc.edu/waterborg/chromat/chroma09.html

Epigenetics

Prof. C. H. Waddington
1905-1975

- Epi- (greek: “over, above”) genetic: heritable changes in gene expression via mechanisms without a change in the DNA sequence and regulated by environmental signals.
- C.H. Waddington coined the term epigenetics to mean above or in addition to genetics to explain differentiation.
- The Epigenetic Code regulates Chromatin Structure and Gene Transcription.
- Unique epigenetic signatures characterize specific cell lineage.





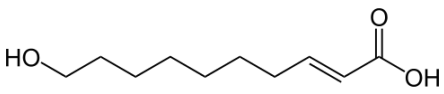
Epigenetic landscape

Epigenetics

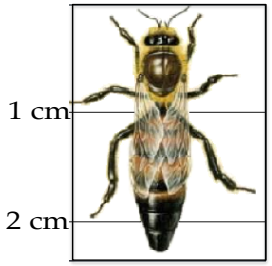
Tortoiseshell cats

Tortoiseshell is a cat coat coloring named for its similarity to tortoiseshell material. Tortoiseshell cats are almost exclusively female.

The cells of female cats, which like other mammalian females have two X chromosomes (XX), undergo the phenomenon of X-inactivation, in which one or the other of the X-chromosomes is turned off at random in each cell in very early development.



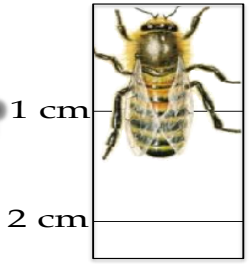
Queen bee acid
Royal jelly acid



Queen

Royal Jelly

Worker Jelly



Worker

478 | NATURE | VOL 473 | 26 MAY 2011

Royalactin induces queen differentiation in honeybees

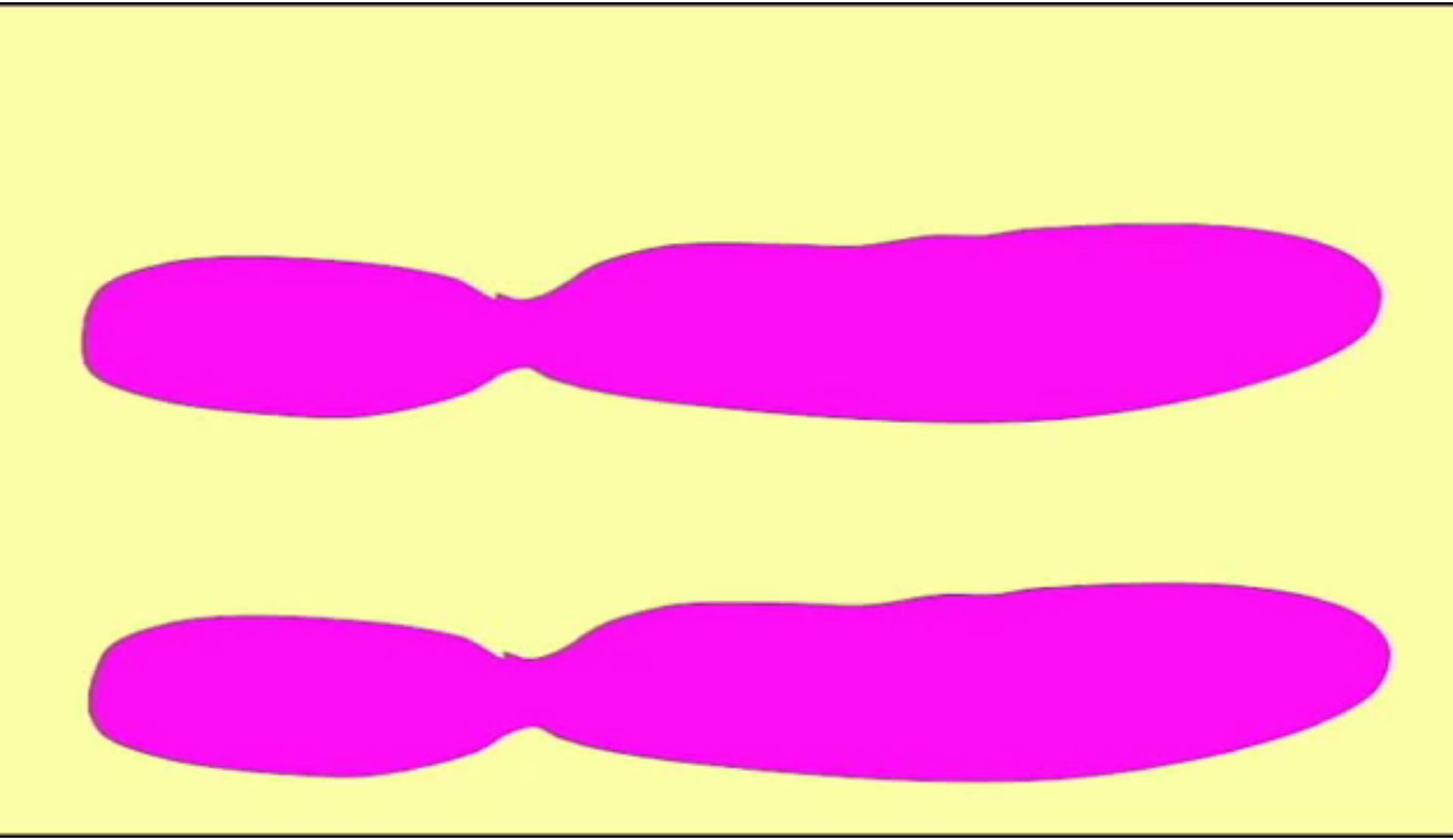
Masaki Kamakura[1]

22 SEPTEMBER 2016 | VOL 537 | NATURE | E10

Royalactin is not a royal making of a queen

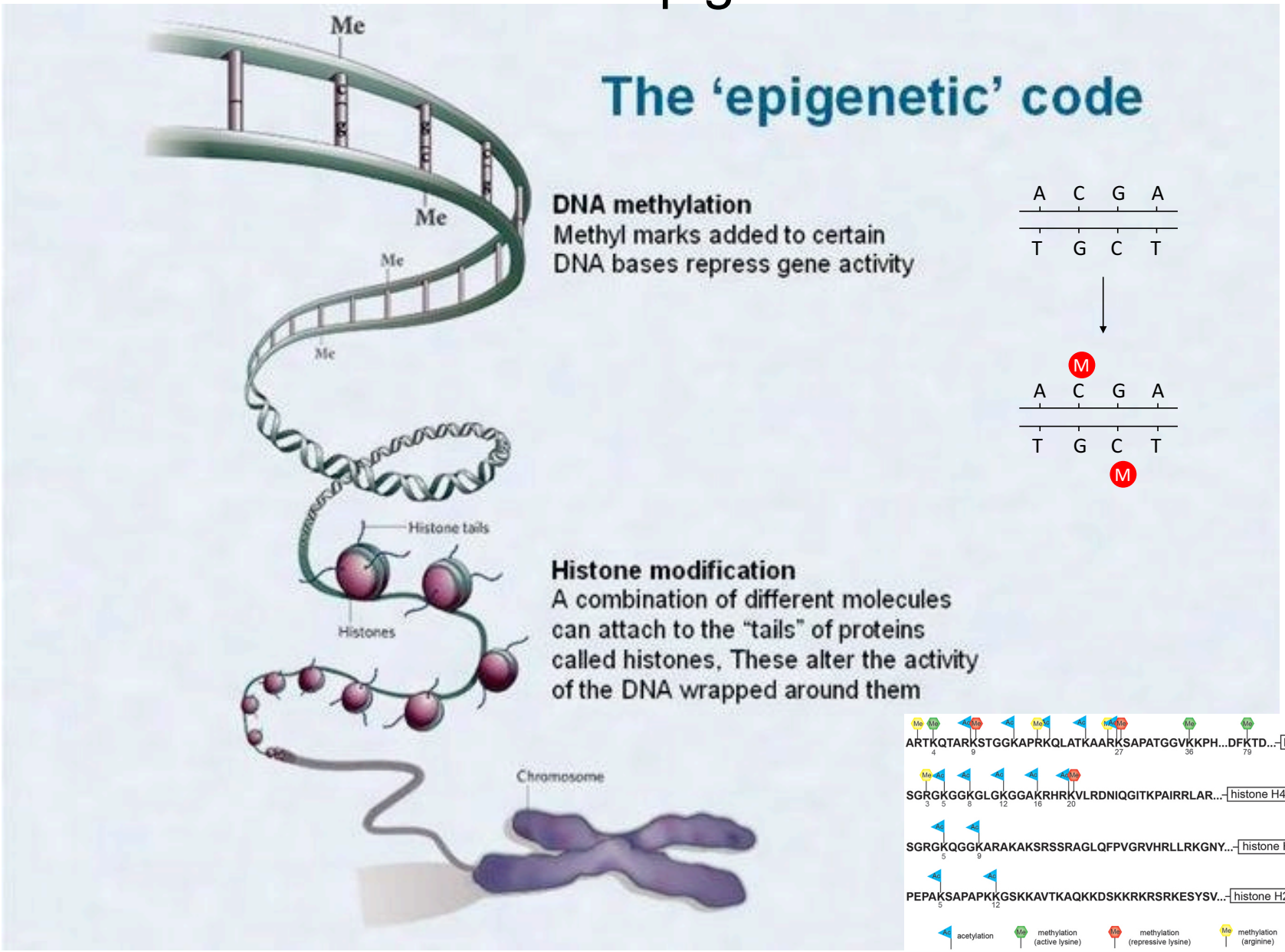
ARISING FROM M. Kamakura *Nature* **473,** http://dx.doi.org/10.1038/nature10093 (2011)

X Chromosome Inactivation (XCI)



https://www.youtube.com/watch?v=Vfk45_VH-HY

DNA Methylation & Histone Modifications Form the Epigenetic Code



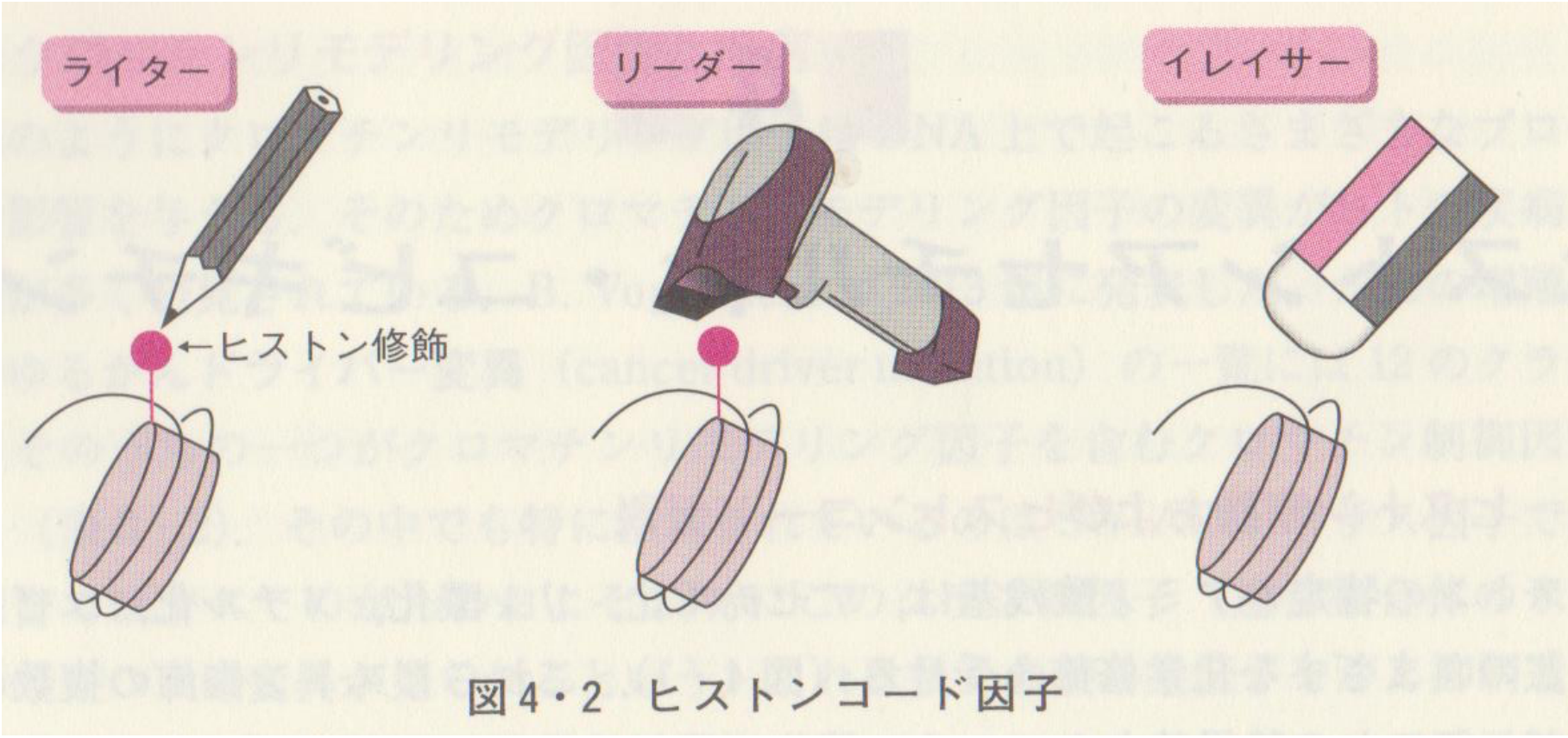


図4・2　ヒストンコード因子

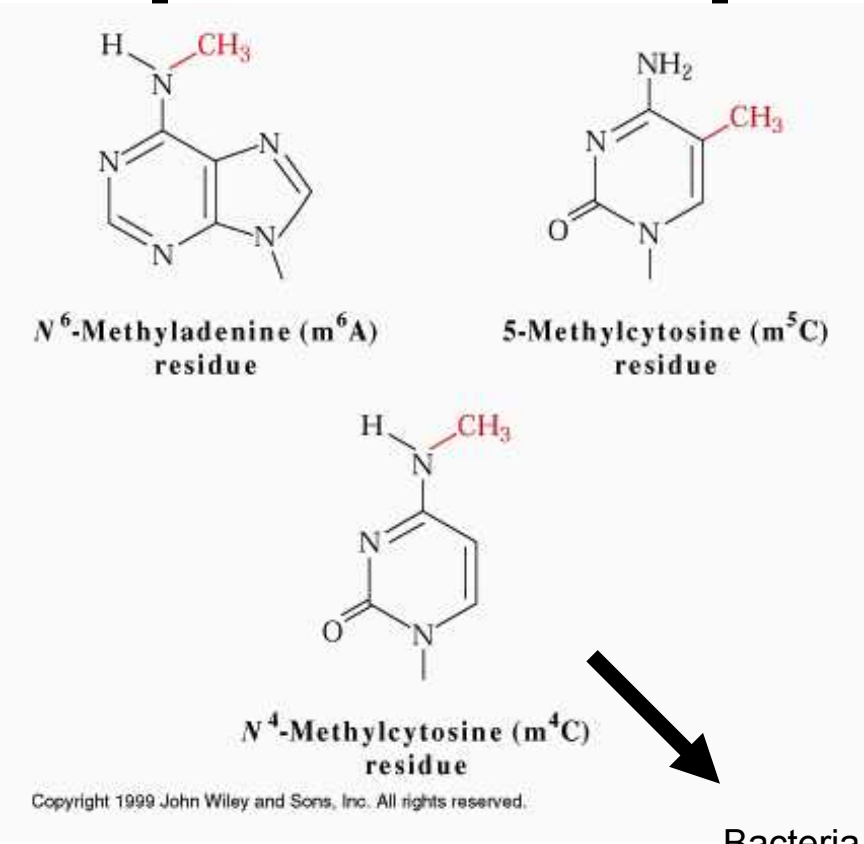
DNA Methylation

Bacteria - wide range of functions

Protista, Plantae, (Mammalia?)

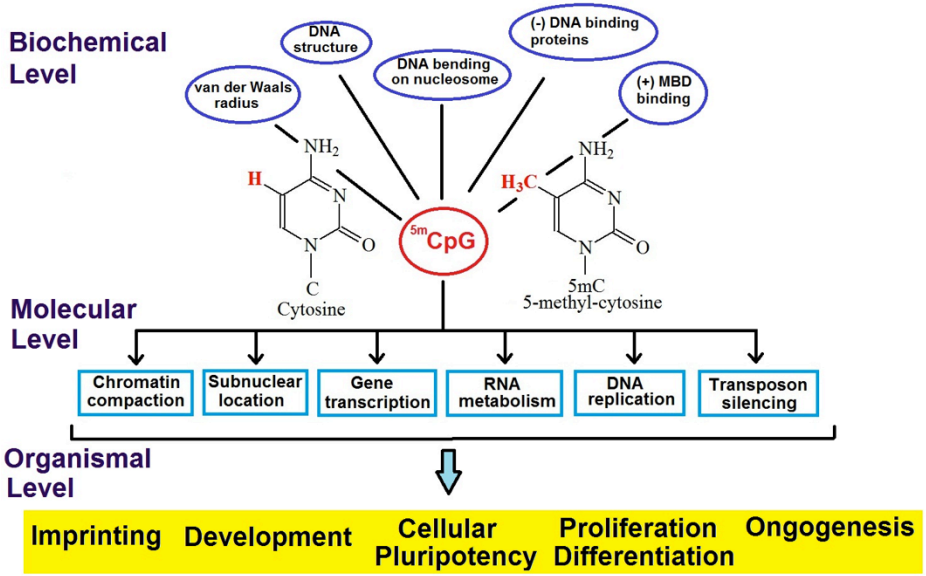
Eukaryota - numerous effects

Bacteria - protection against RE

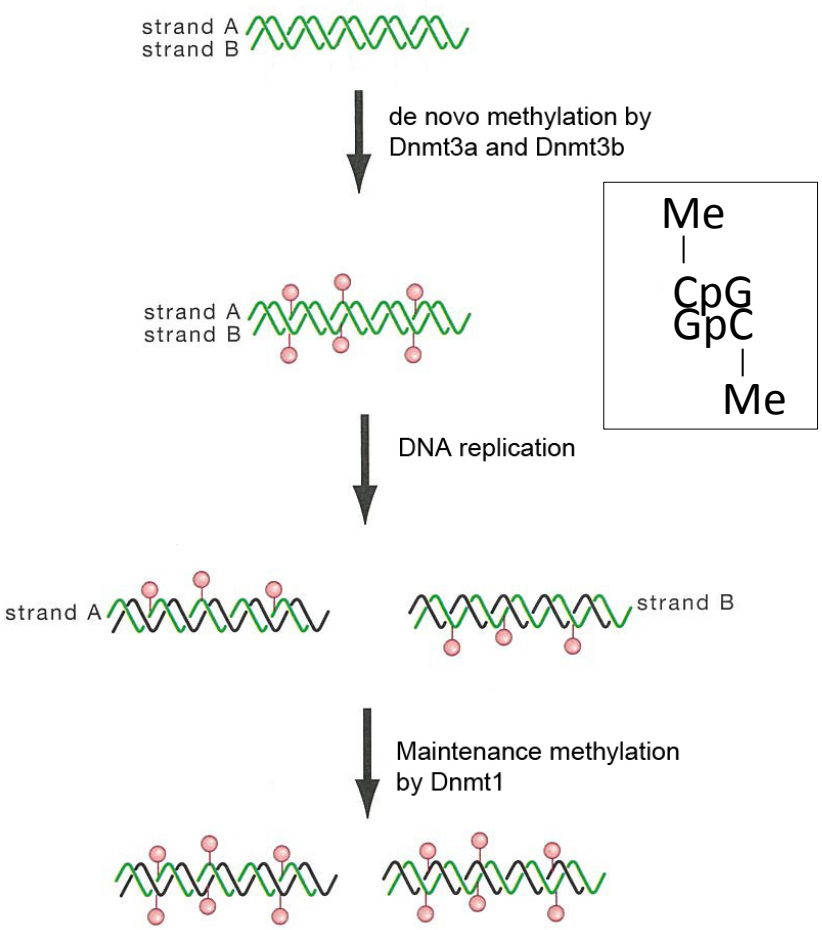


Bacteria - protection against RE

Heritable gene silencing by CpG DNA methylation



Franchini et al. 2012



- Methylation patterns are established by Dnmt3a/b in early development.

- Faithfully maintained through DNA replication (Dnmt1).

Brockdorff Talks

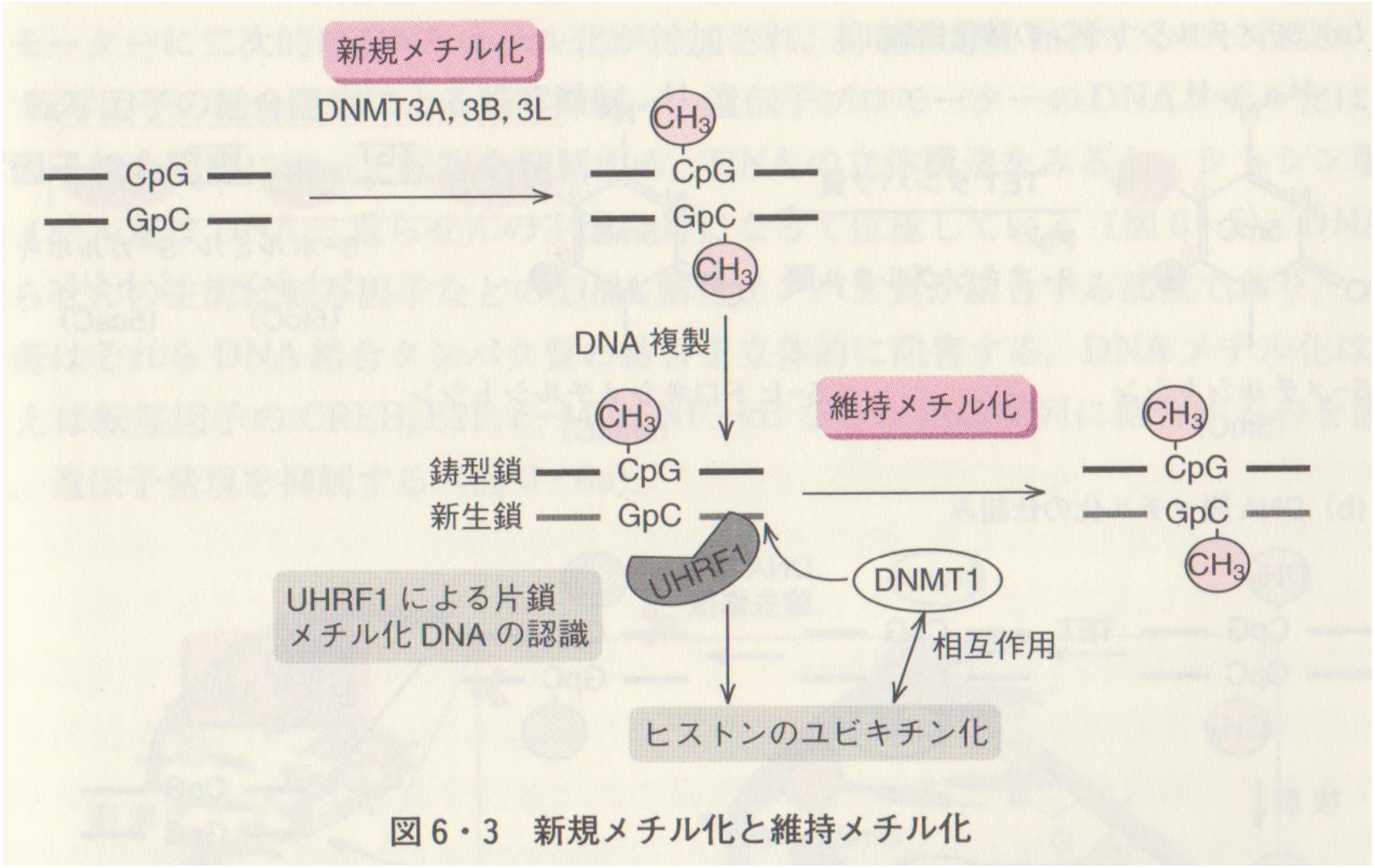
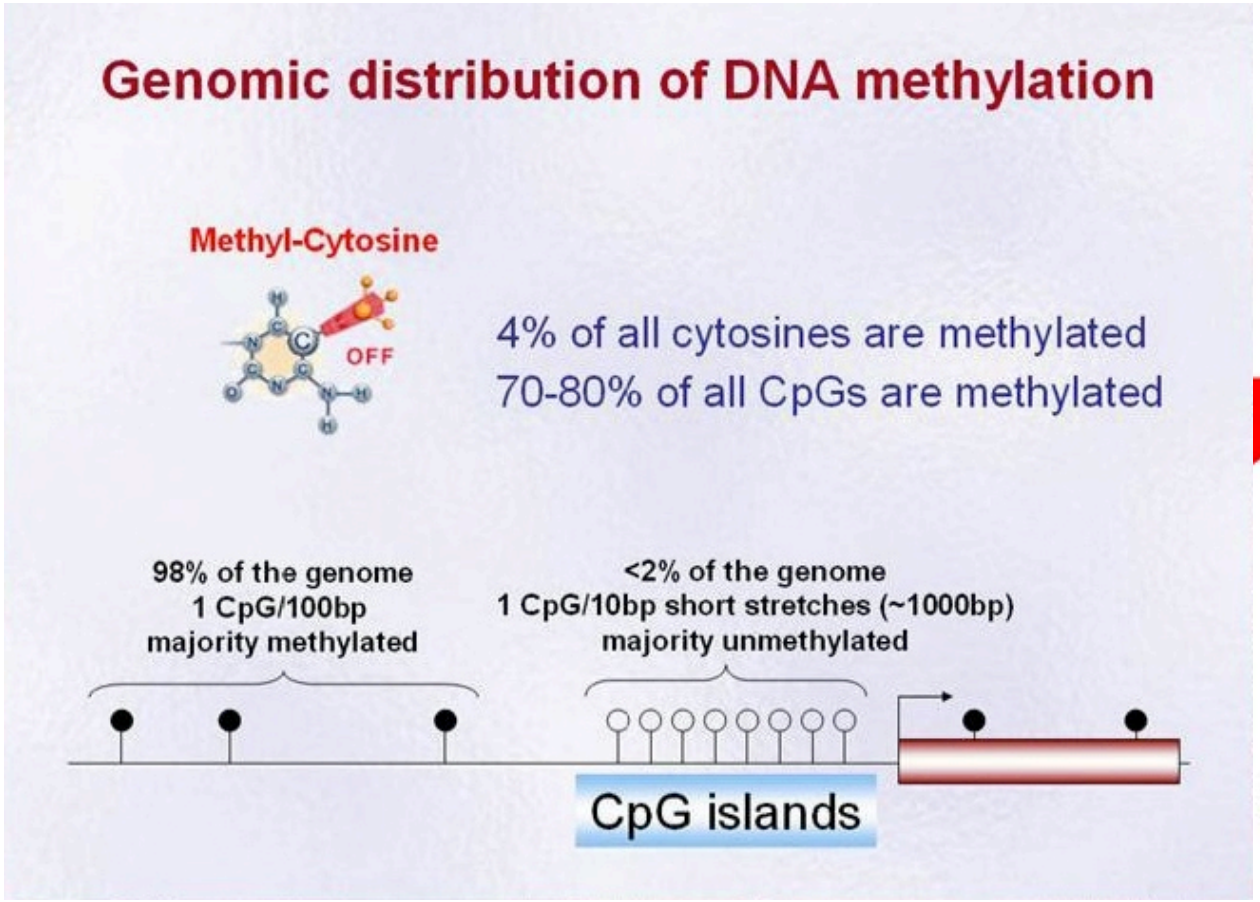
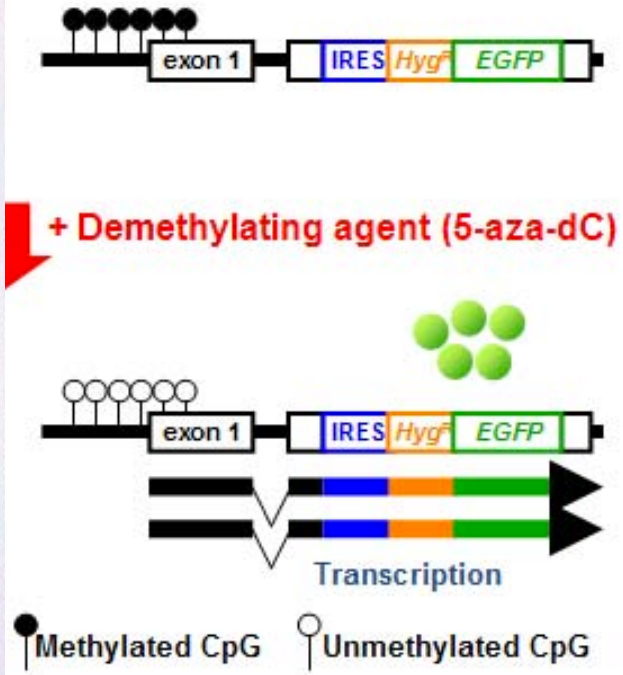


図 6・3　新規メチル化と維持メチル化

Critical CpG Sequences in CpG Islands Near Promoters





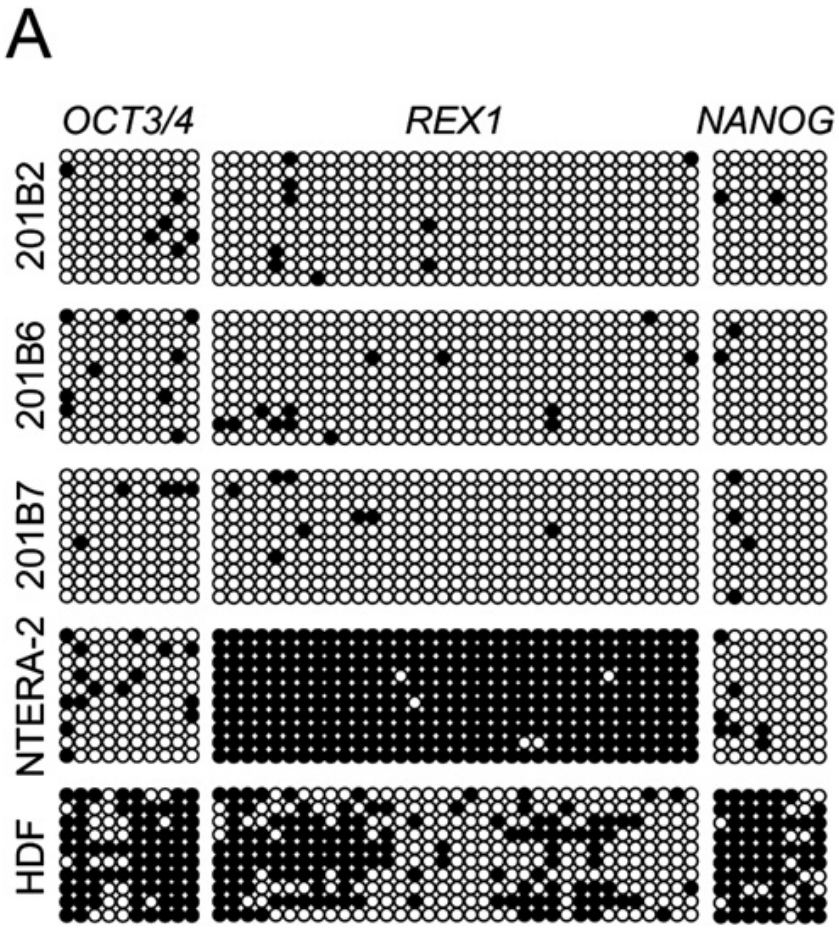
Cell *131*, 861–872, November 30, 2007

Induction of Pluripotent Stem Cells from Adult Human Fibroblasts by Defined Factors

Kazutoshi Takahashi,[1] Koji Tanabe,[1] Mari Ohnuki,[1] Megumi Narita,[1,2] Tomoko Ichisaka,[1,2] Kiichiro Tomoda,[3] and Shinya Yamanaka[1,2,3,4,*]

SUMMARY

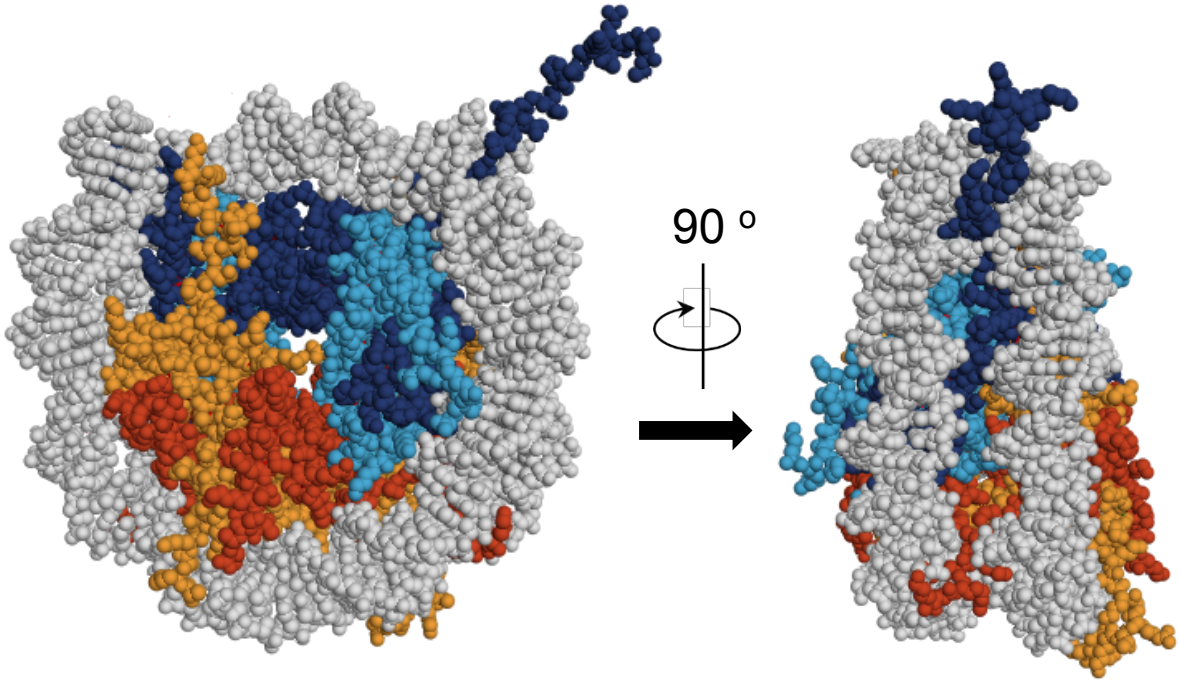
Successful reprogramming of differentiated human somatic cells into a pluripotent state would allow creation of patient- and disease-specific stem cells. We previously reported generation of induced pluripotent stem (iPS) cells, capable of germline transmission, from mouse somatic cells by transduction of four defined transcription factors. Here, we demonstrate the generation of iPS cells from adult human dermal fibroblasts with the same four factors: Oct3/4, Sox2, Klf4, and c-Myc. Human iPS cells were similar to human embryonic stem (ES) cells in morphology, proliferation, surface antigens, gene expression, epigenetic status of pluripotent cell-specific genes, and telomerase activity. Furthermore, these cells could differentiate into cell types of the three germ layers in vitro and in teratomas. These findings demonstrate that iPS cells can be generated from adult human fibroblasts.



(A) Bisulfite genomic sequencing of the promoter regions of *OCT3/4*, *REX1*, and *NANOG*.

Nucleosome

- Histone octamer is composed of pairs of H2A, H2B, H3, and H4 proteins
- About 147 bp of DNA is wrapped around histone octamer to form nucleosome

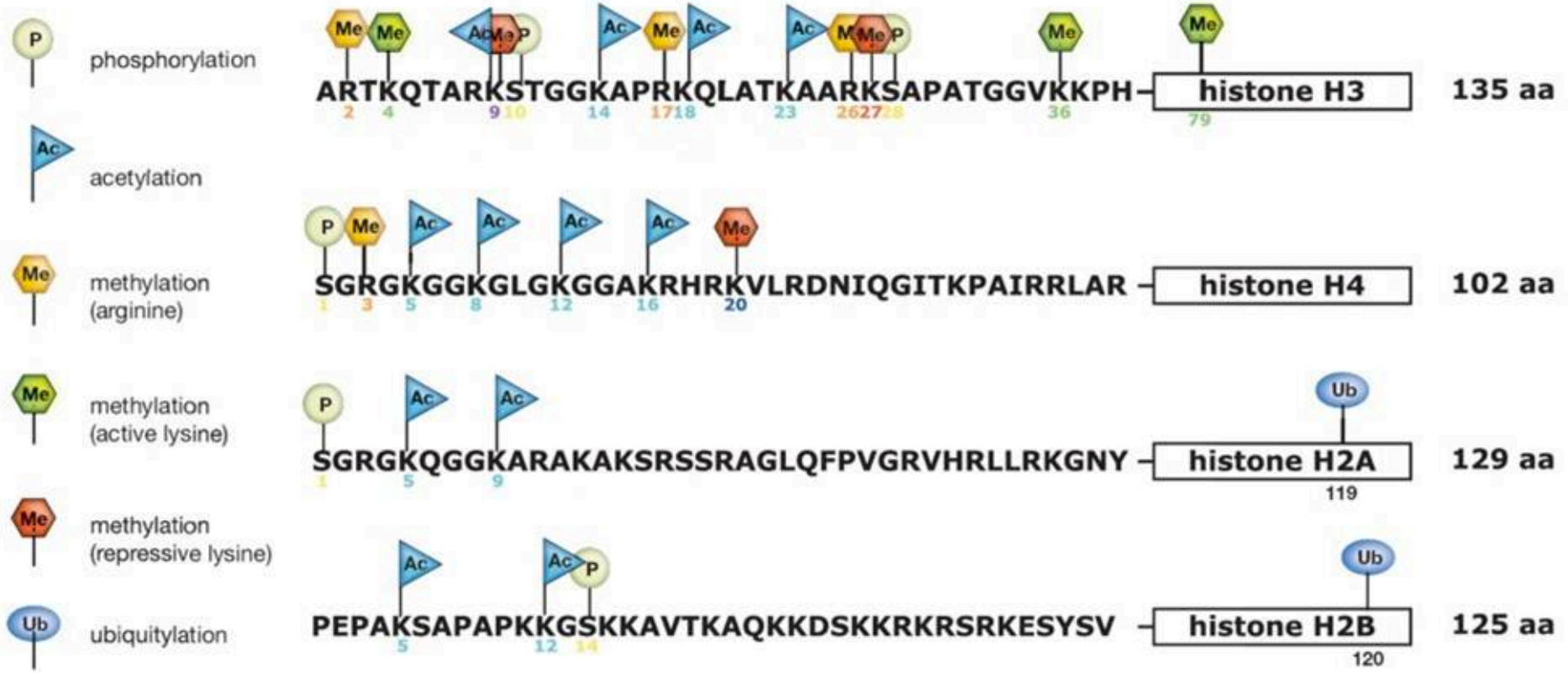


Lugar, K. *et al. Nature* **1997**, *389*, 251-60.

H2A H2B H3 H4

The Histone Code

Post-translational modifications of histones, along with deposition of histone variants, form a “histone code” of activation or repression of transcription in euchromatin or heterochromatin, respectively.

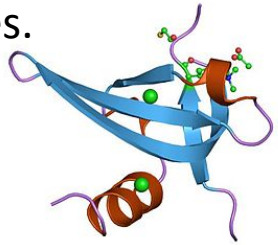


http://www.shechterlab.org/2009/science/chromatin-and-the-histone-code/

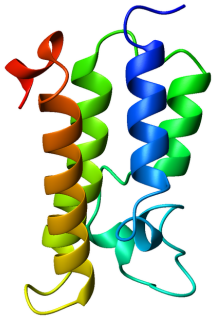
The Histone Code

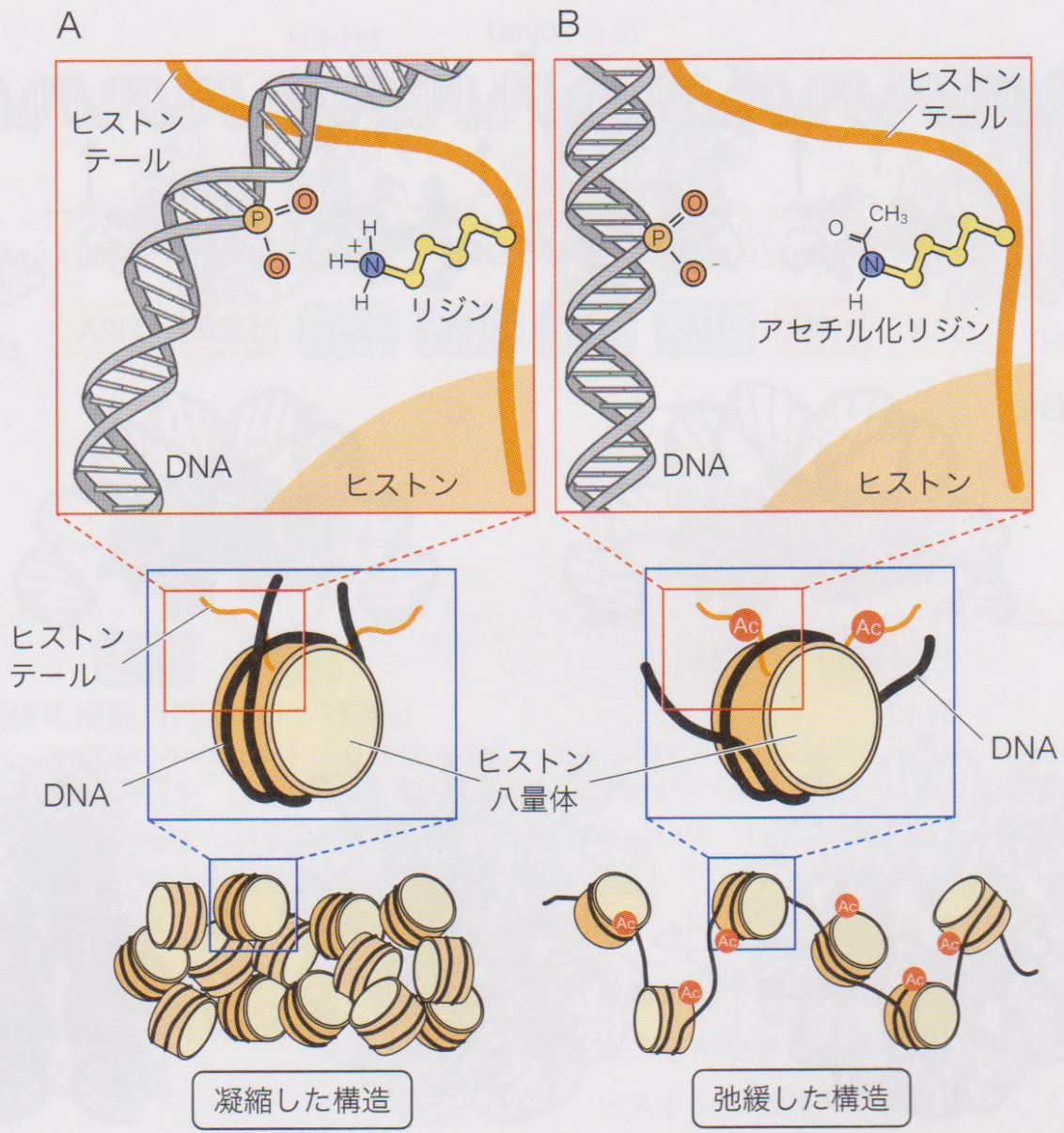
		Reader	Writer
Tri methylation			
H3 K4	Activation	Chromodomain	MLL, Set
H3 K9	Repression	Chromodomain	Suv39h G9a
H3 K27	Repression	Ezh2, G9a	Ezh2
H3 K36	Activation		
Mono methylation			
H3 K4	Enhancer mark		
Acethylation			
H3 K9	Activation	Bromodomain	Gcn5 PCAF
H3 K14	Activation	Bromodomain	p300 CBP
H3 K27	Activation		

A chromodomain (chromatin organization modifier) is a protein structural domain of about 40-50 amino acid residues.



A bromodomain is an approximately 110 amino acid protein domain that recognizes acetylated lysine residues.





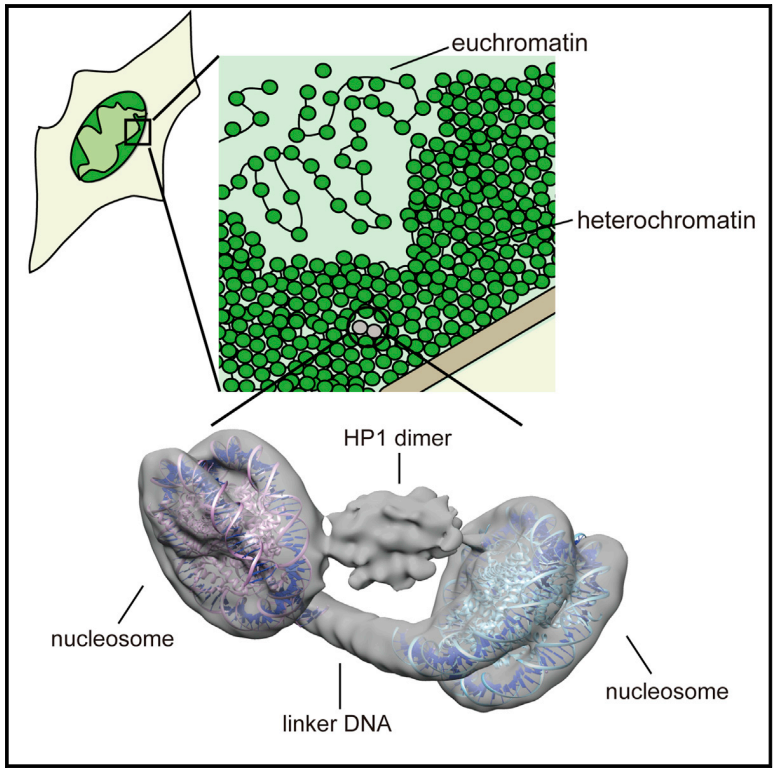
ヒストンのアセチル化修飾がクロマチン構造に与える影響

A）未修飾のリジンは正電荷をもつため，DNAのリン酸骨格の負電荷と静電相互作用により結合して，凝縮したクロマチン構造をとりやすい．B）リジンがアセチル化修飾を受けると，リジンの正電荷が失われてDNAとの静電相互作用が弱くなるため，クロマチンは緩んだ構造をとりやすくなる．

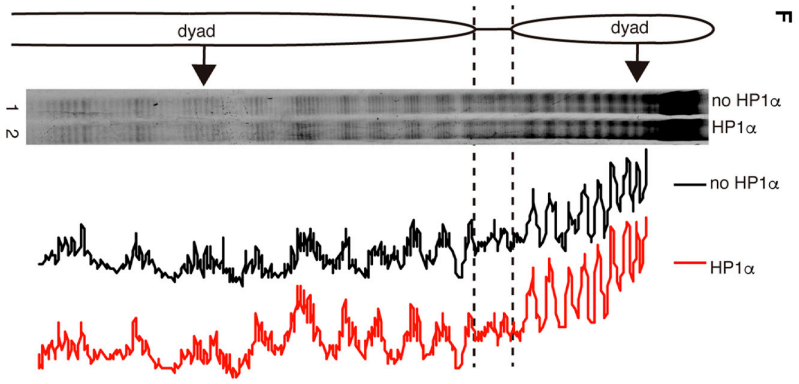
Molecular Cell *69*, 385–397, February 1, 2018

Structural Basis of Heterochromatin Formation by Human HP1

Shinichi Machida,[1,5] Yoshimasa Takizawa,[2,5] Masakazu Ishimaru,[1] Yukihiko Sugita,[2] Satoshi Sekine,[1] Jun-ichi Nakayama,[3] Matthias Wolf,[2,*] and Hitoshi Kurumizaka[1,4,6,*]



- The HP1-H3K9me3 dinucleosome complex structure determined by the cryo-EM method
- HP1 forms a symmetric dimer and bridges two H3K9me3 nucleosomes in the complex
- The HP1 chromoshadow domain dimer exists in an accessible location in the complex
- The linker DNA between nucleosomes does not directly interact with HP1



HP1 isoforms: HP1α, HP1β, HP1γ

H3.2 K9 trimethylated K9C/C110A mutant

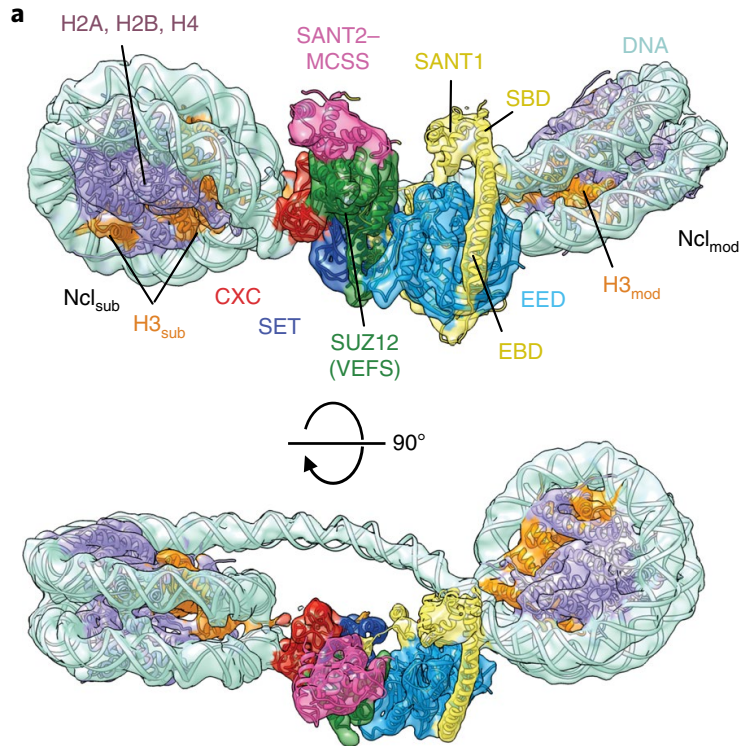
(2-bromoethyl)trimethylammonium bromide

15 bp linker DNA

NATURE STRUCTURAL & MOLECULAR BIOLOGY | VOL 25 | FEBRUARY 2018 | 154–162

Cryo-EM structures of PRC2 simultaneously engaged with two functionally distinct nucleosomes

Simon Poepsel[1,2], Vignesh Kasinath[1,2] and Eva Nogales[1,2,3,4*]

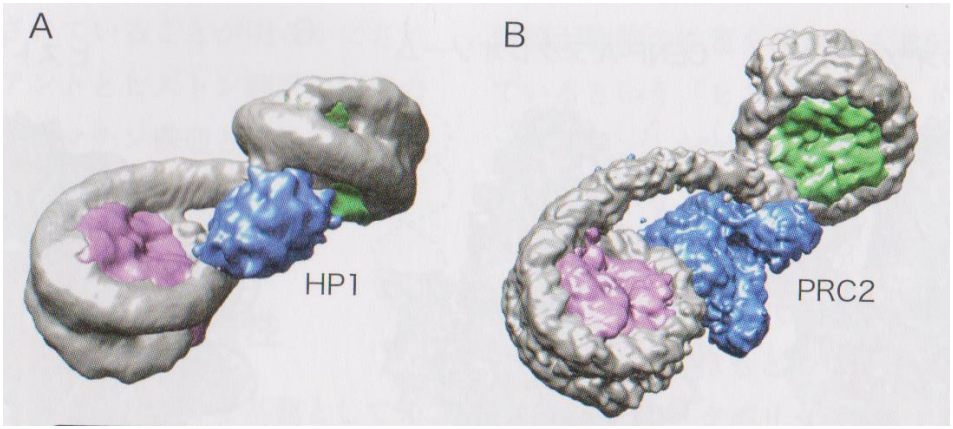


H3K27me3 K27C/C110A mutant

Polycomb repressive complex 2 (PRC2): EZH2, EED, SUZ12, RBAP48, and AEBP2

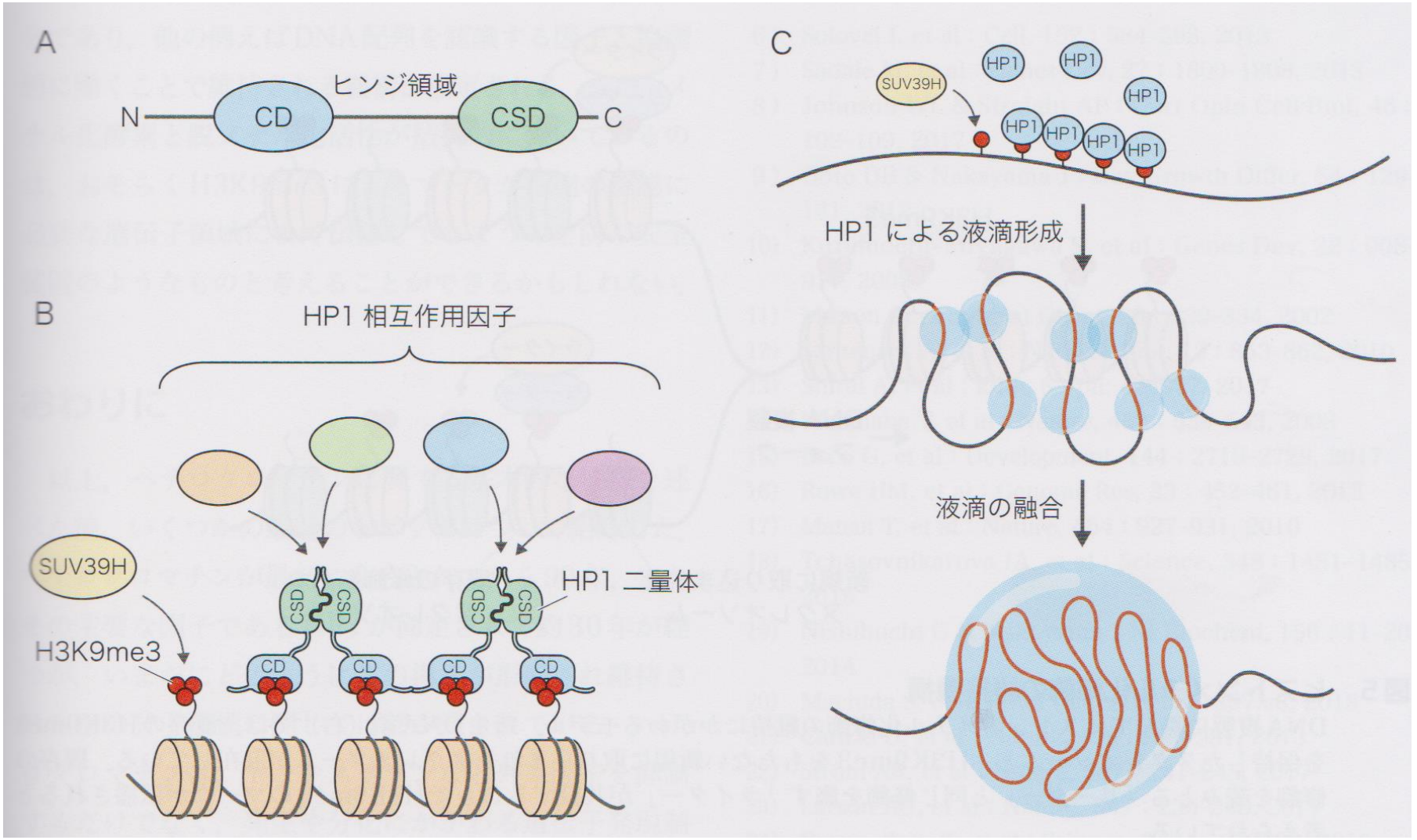
Polycomb repressive complex 2 (PRC2) binds to H3K27me3

35 bp linker DNA



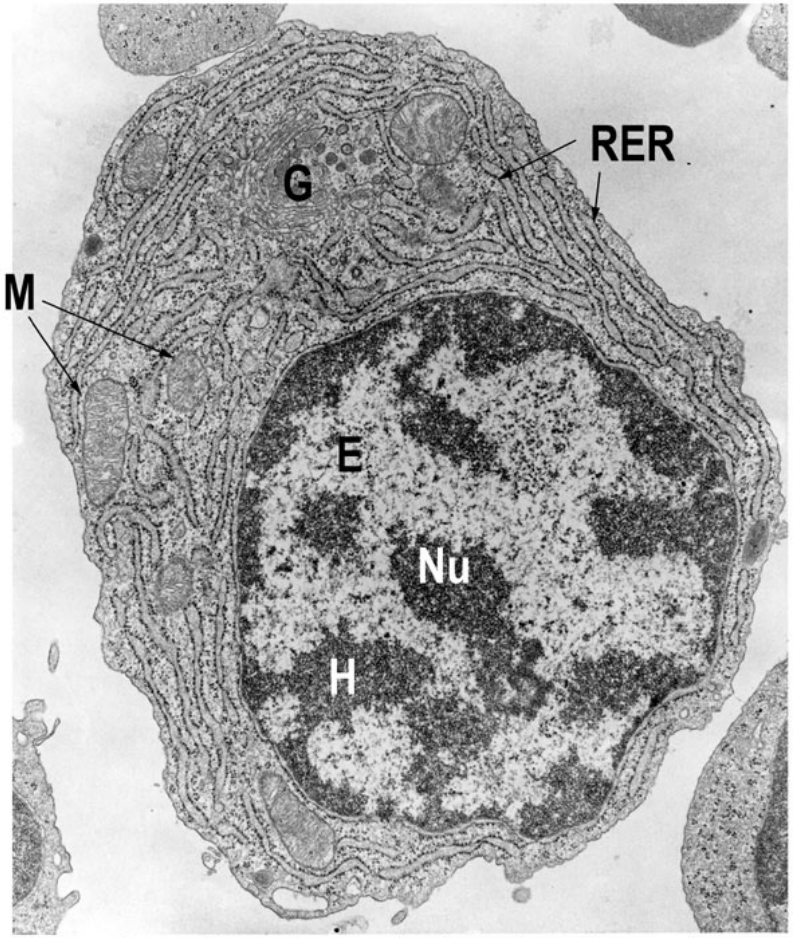
Constitutive heterochromatin

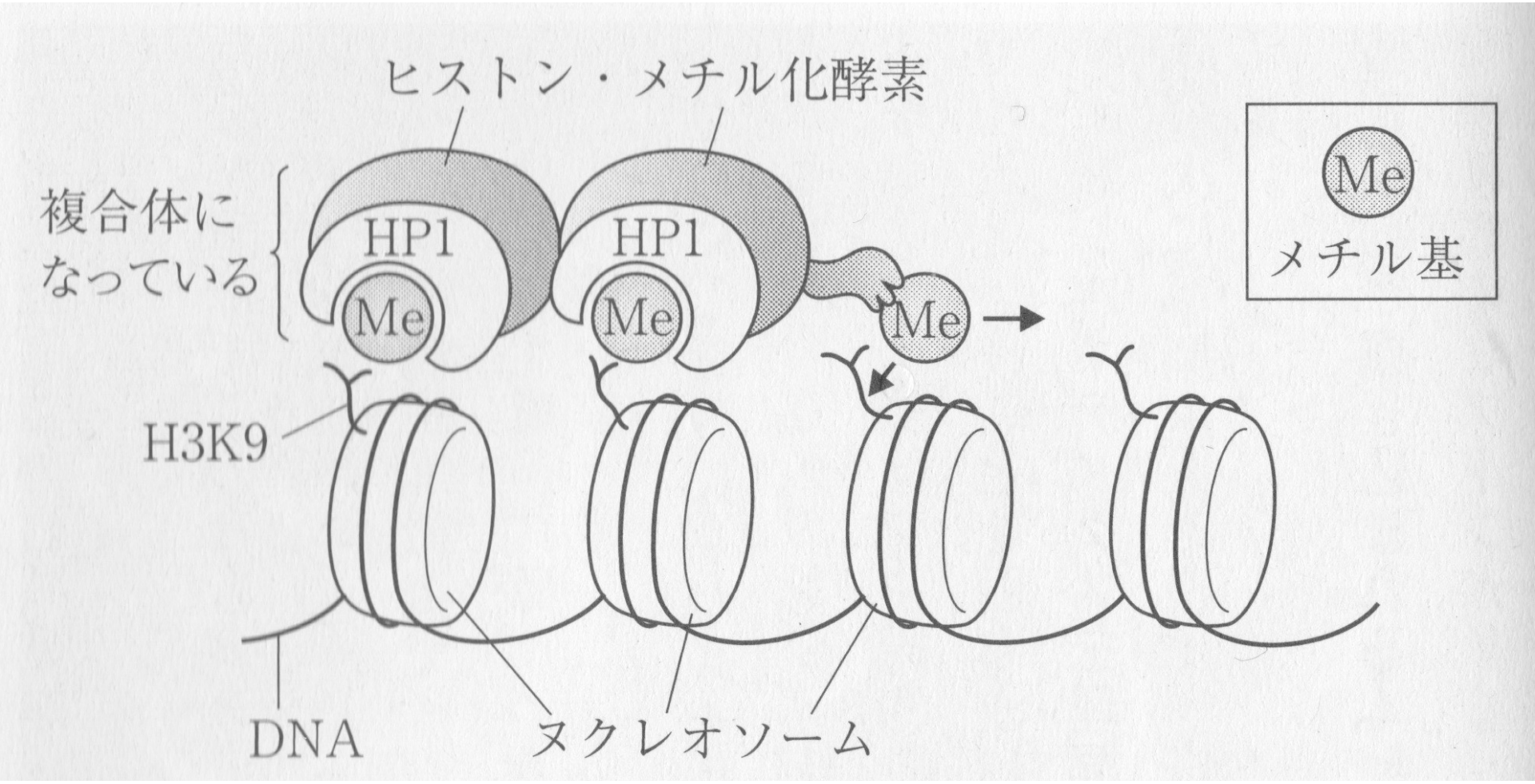
Facultative heterochromatin


A
N
CD
ヒンジ領域
CSD
C
B
HP1 相互作用因子
SUV39H
H3K9me3
CSD
CD
HP1 二量体
C
SUV39H
HP1
HP1 による液滴形成
液滴の融合

Euchromatin and Heterochromatin

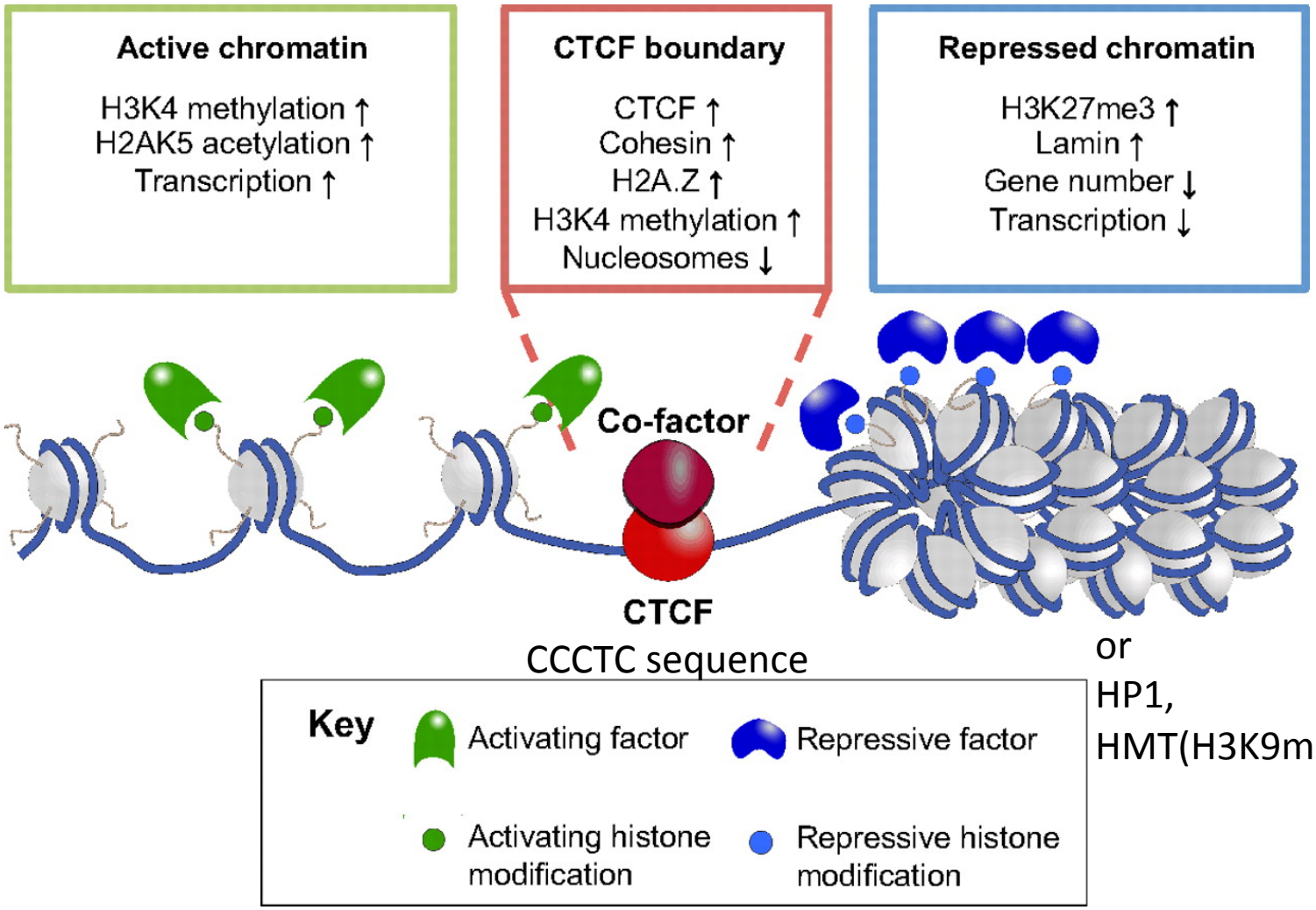
- In electro microscopic image of cell section, **euchromatin** region (E) and **heterochromatin** region (H) can be observed




ヒストン・メチル化酵素
複合体に
なっている
HP1
HP1
Me
Me
Me
Me
メチル基
H3K9
DNA
ヌクレオソーム

CTCF boundary elements are associated with specific chromatin features.

Centromere, X chromosome



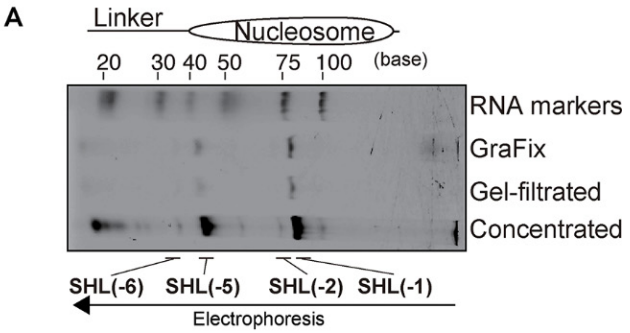
or
HP1,
HMT(H3K9me3)

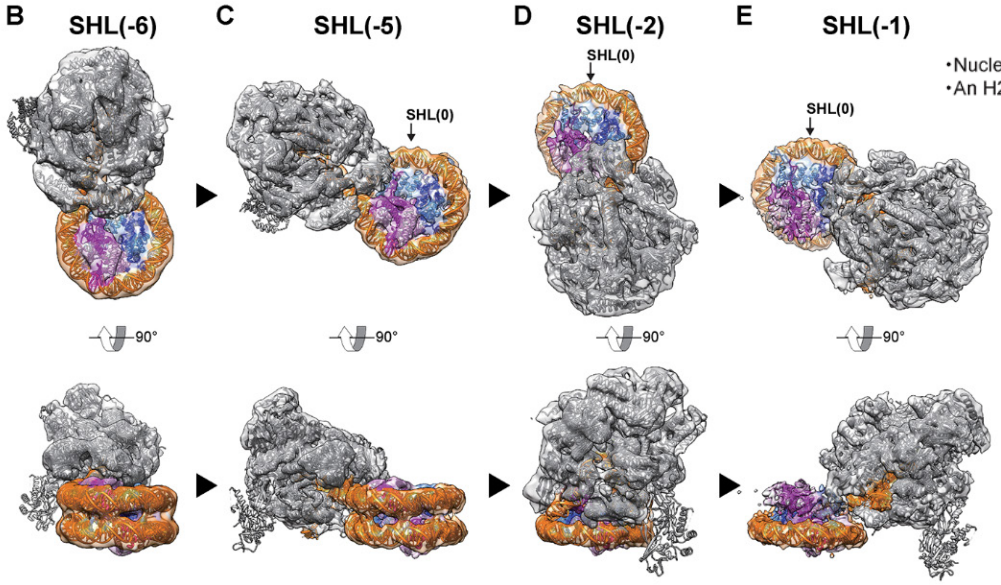
Martin Herold et al. Development 2012;139:1045-1057

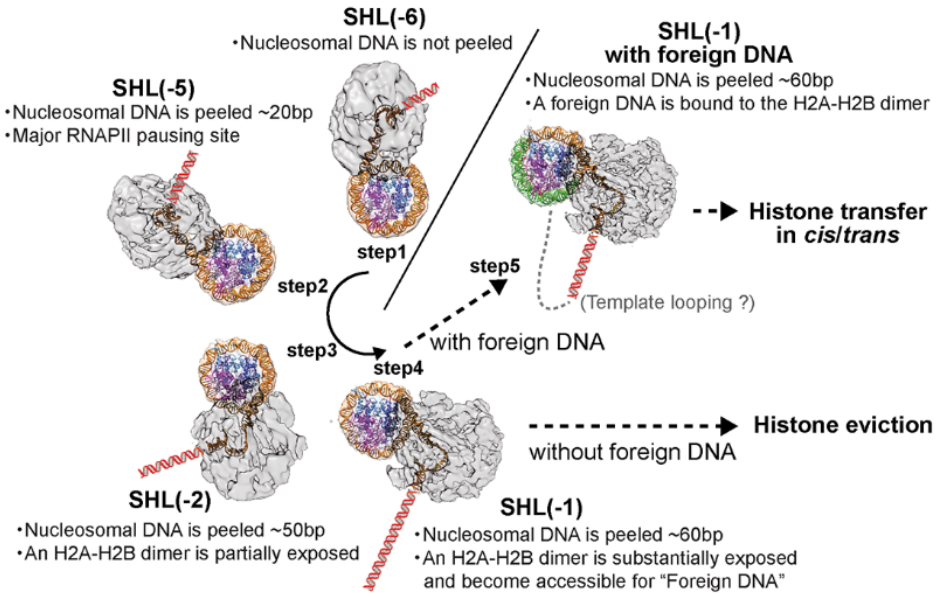
Structural basis of the nucleosome transition during RNA polymerase II passage

Cite as: T. Kujirai *et al.*, *Science* 10.1126/science.aau9904 (2018).

Tomoya Kujirai[1,2]*, Haruhiko Ehara[2]*, Yuka Fujino[1,3], Mikako Shirouzu[2], Shun-ichi Sekine[2]†, Hitoshi Kurumizaka[1,2,3]†

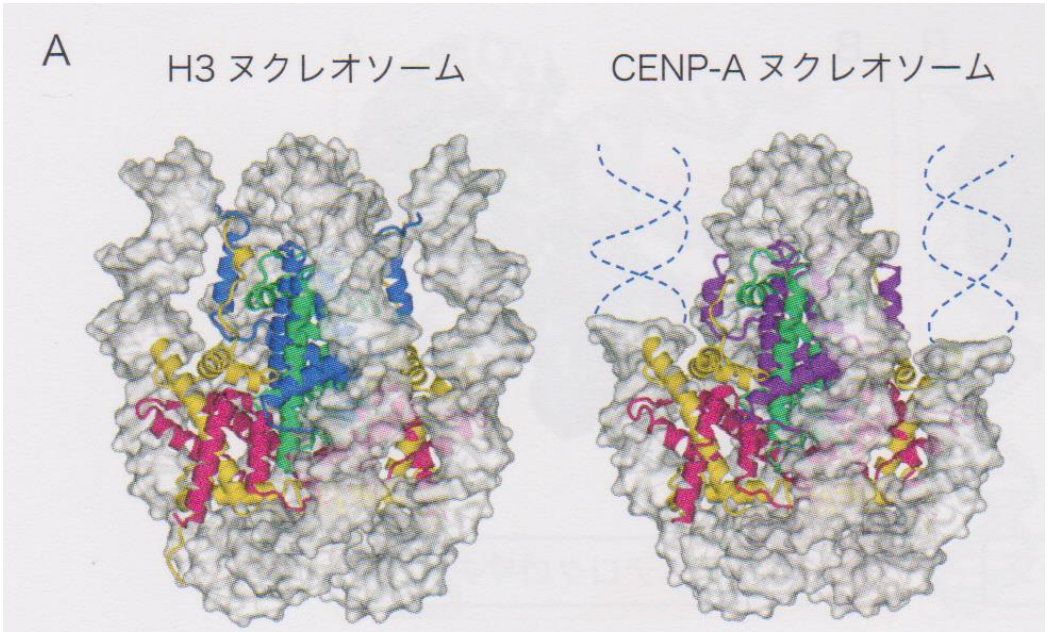






RNA polyamerase II pauses at superhelical locations, SHL(-6), SHL(-5), SHL(-2), and SHL(-1) of the nucleosome.

Understanding chromatin transcription and epigenetic regulation.



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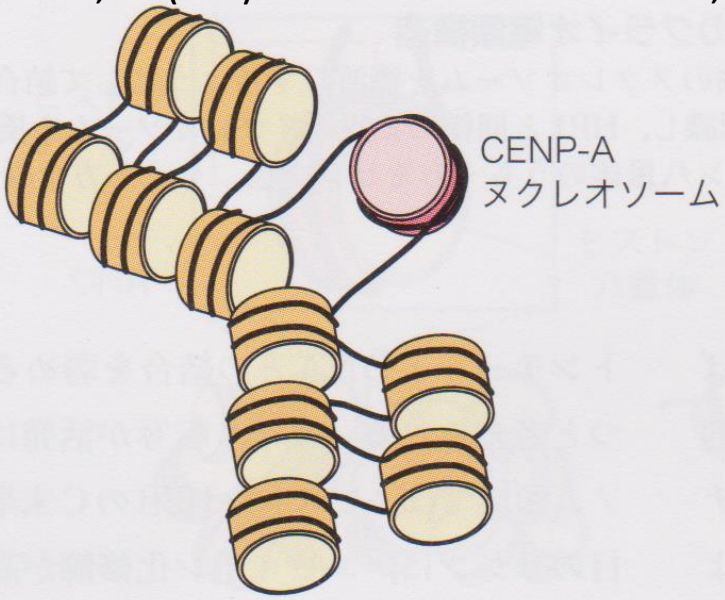

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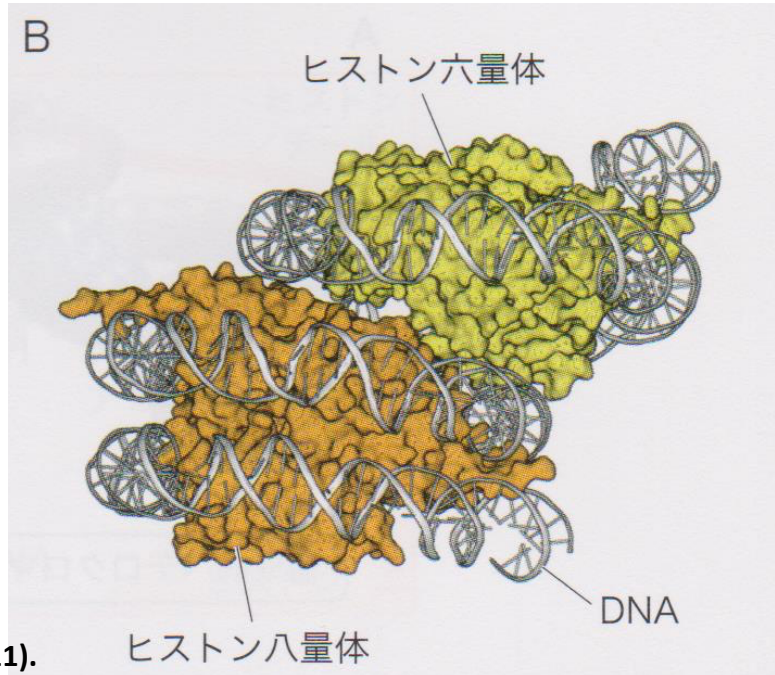

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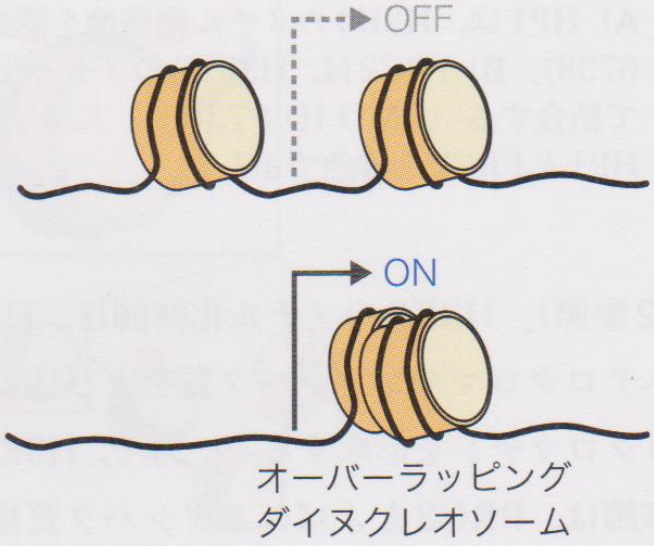

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