

Genome Editing and Engineering

Course No: BT-637



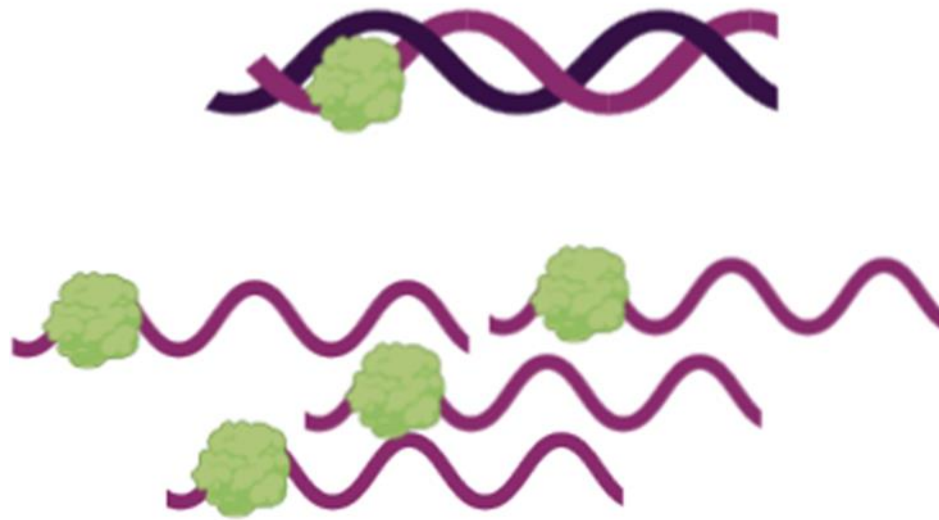
LECTURE-10

Dr. Kusum K. Singh

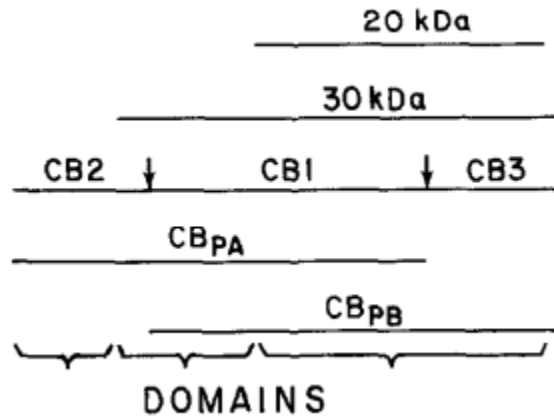
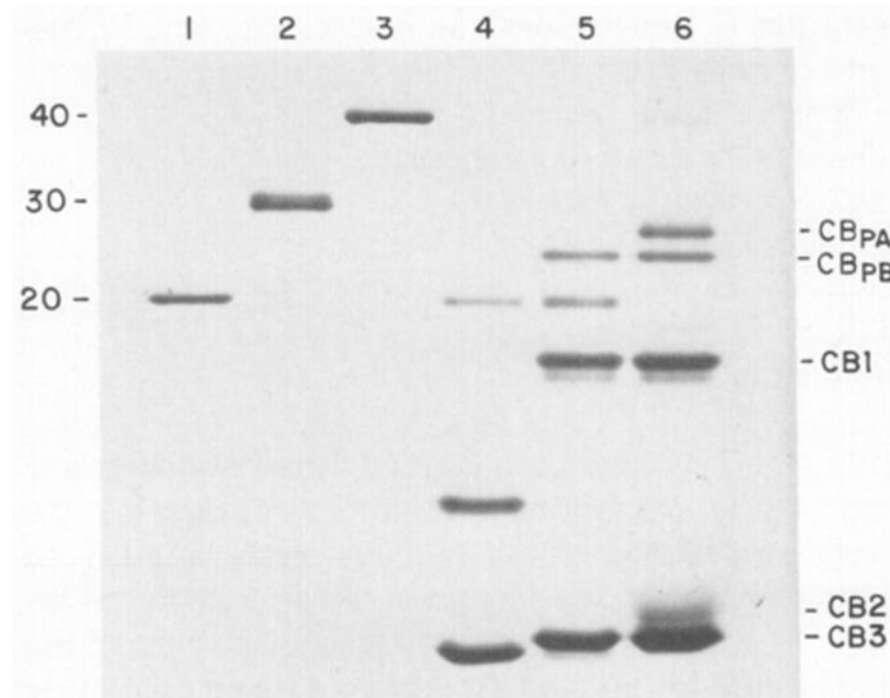
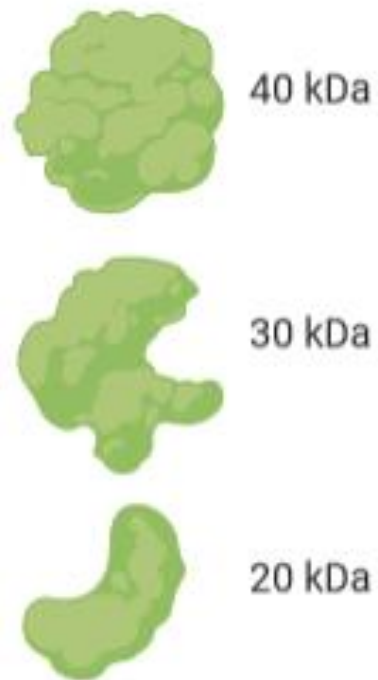
Department of Biosciences and Bioengineering

Indian Institute of Technology Guwahati

Introduction



Introduction

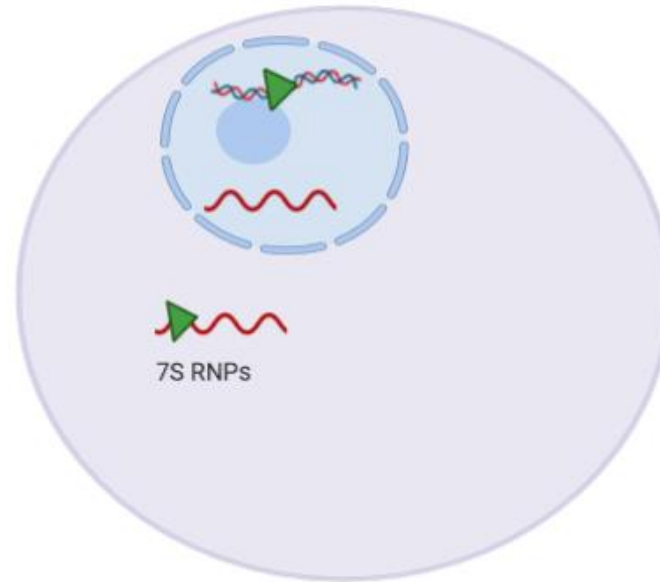


Repetitive zinc-binding domains in the protein transcription factor IIIA from *Xenopus* oocytes

J. Miller, A.D. McLachlan and A. Klug

The EMBO Journal

MRC Laboratory of Molecular Biology, Hills Road, Cambridge CB2 2QH,
UK

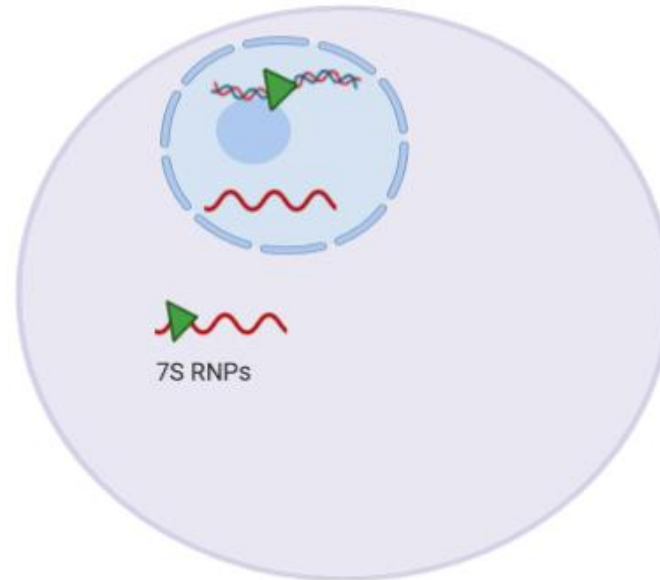


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0.1 mM DTT

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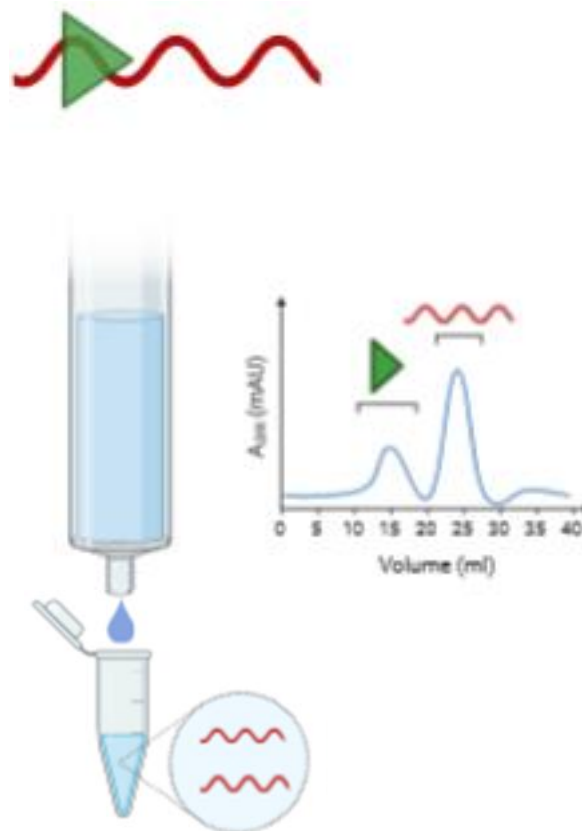
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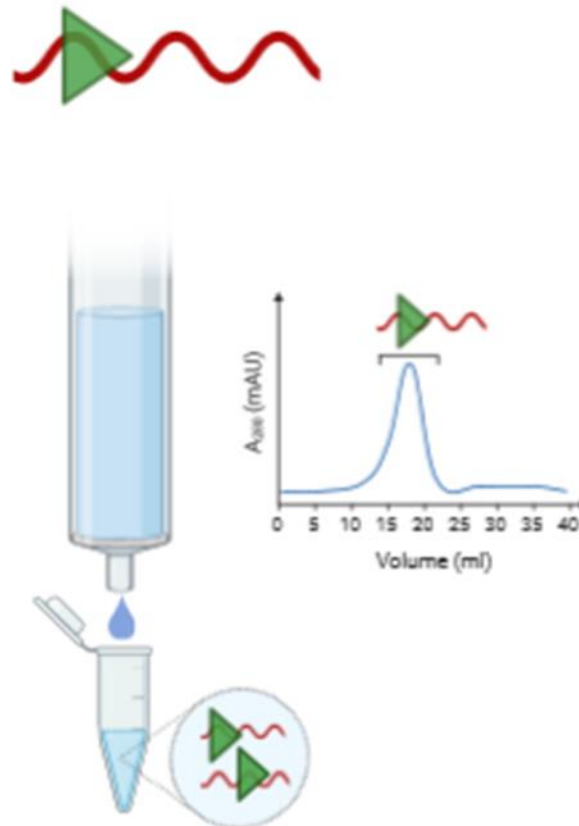


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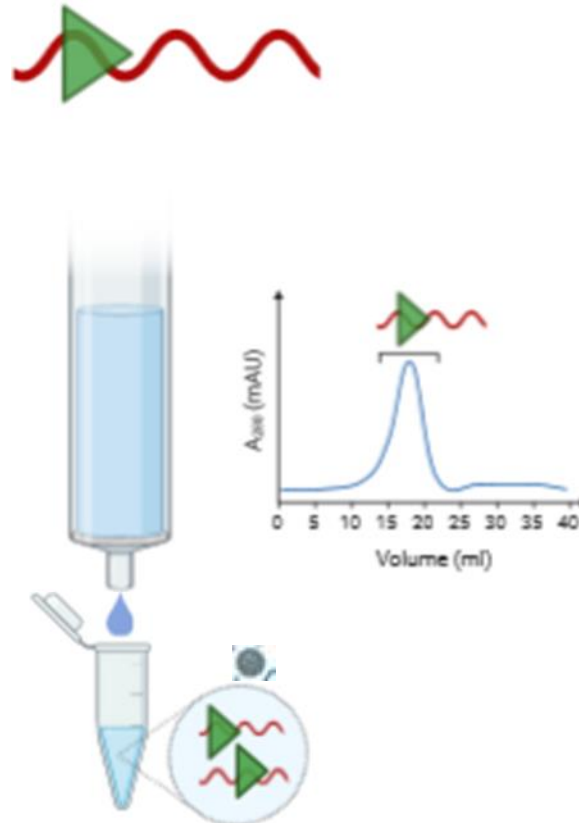


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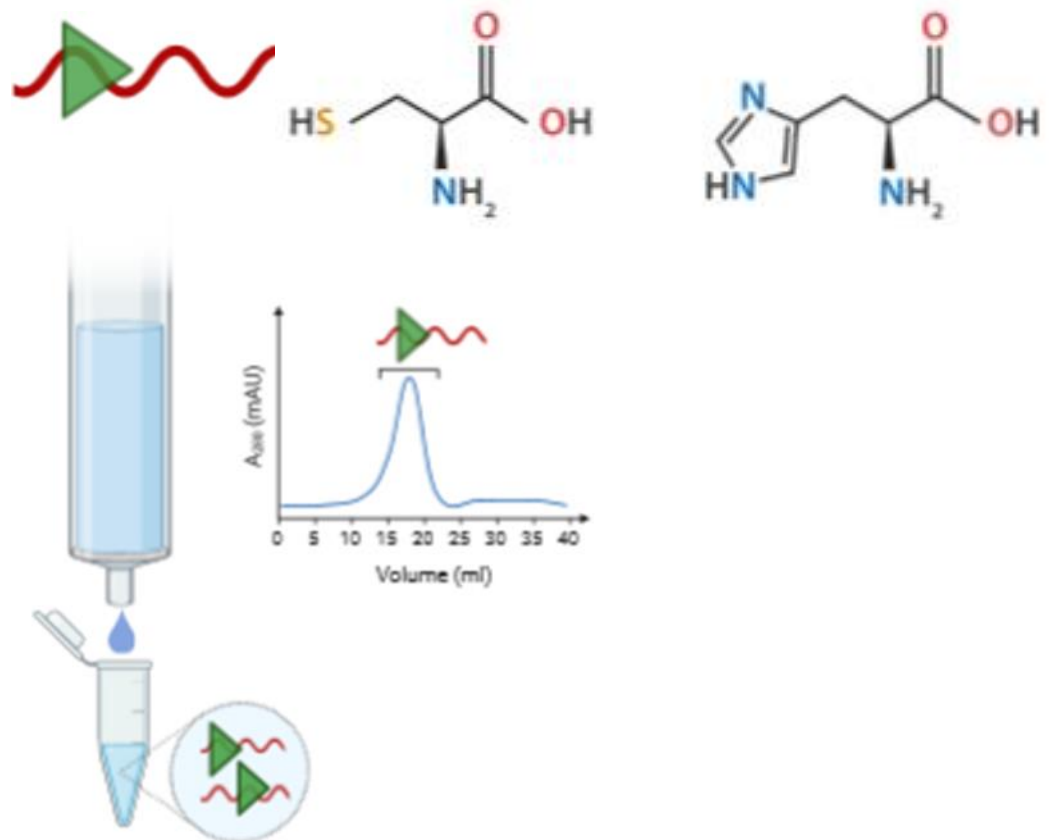
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- DTT

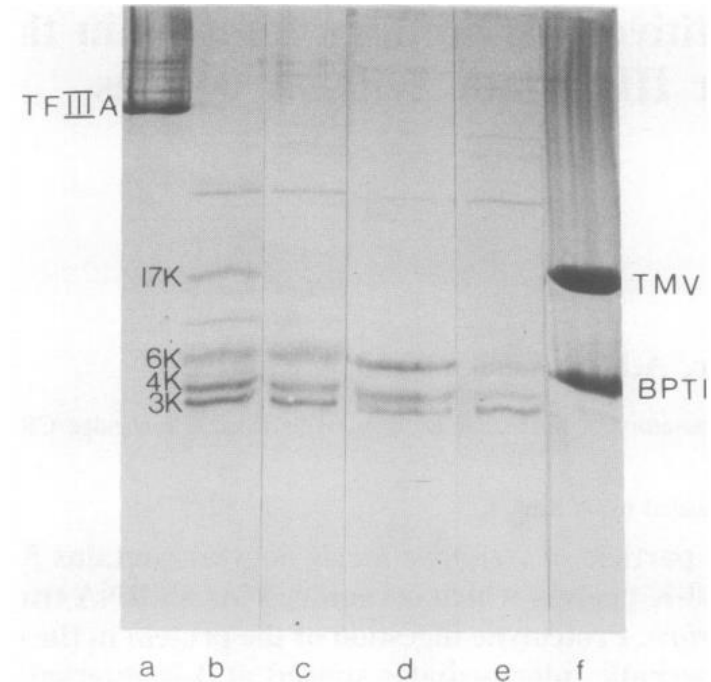
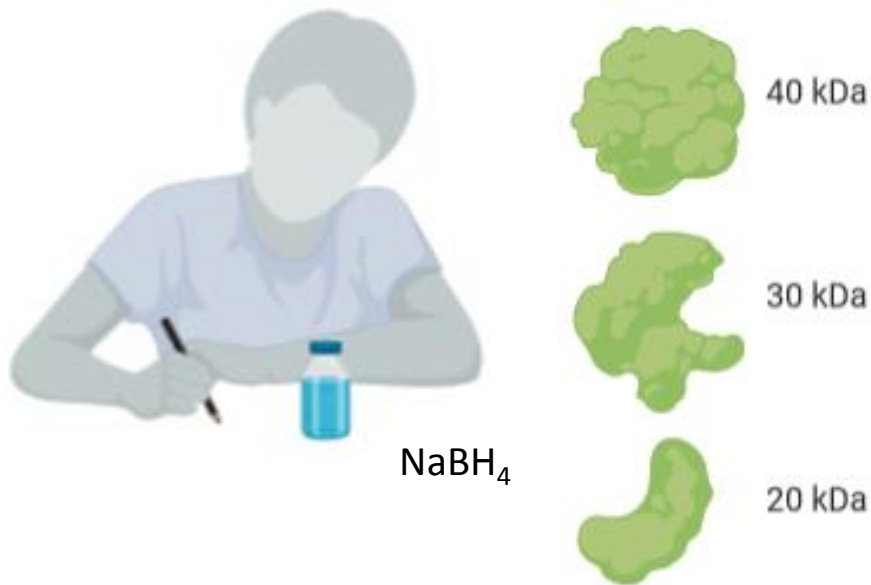


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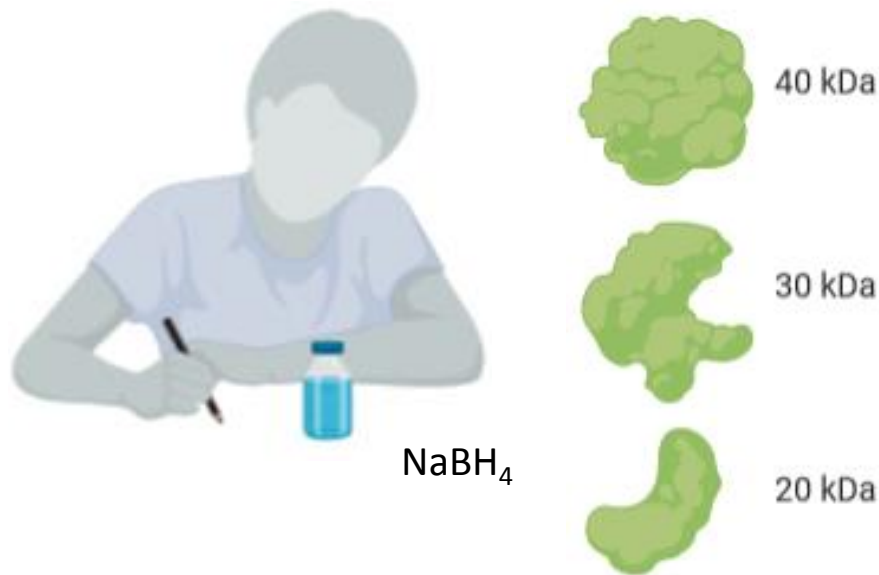


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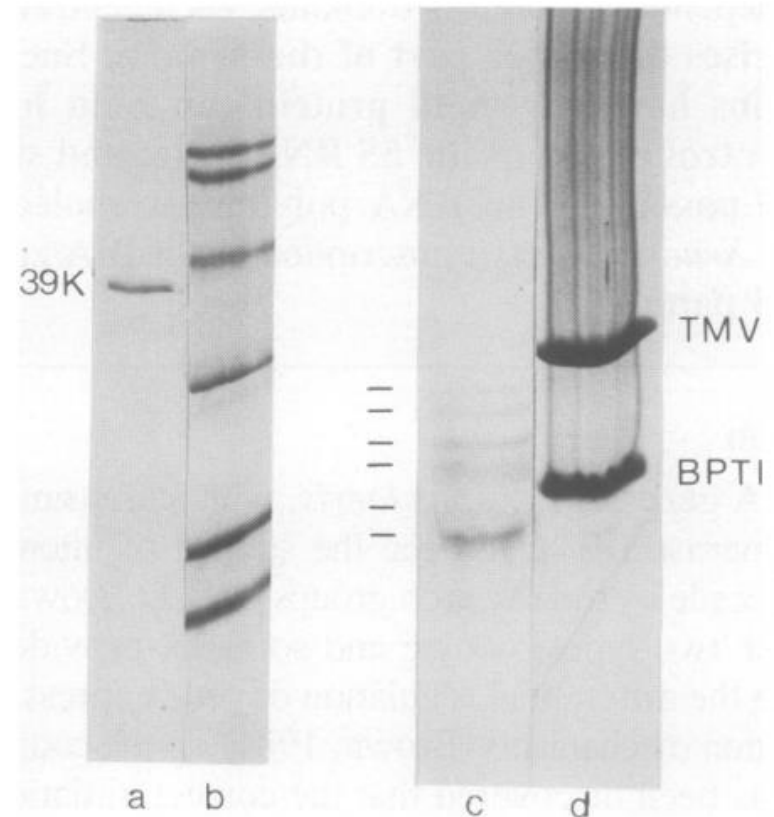
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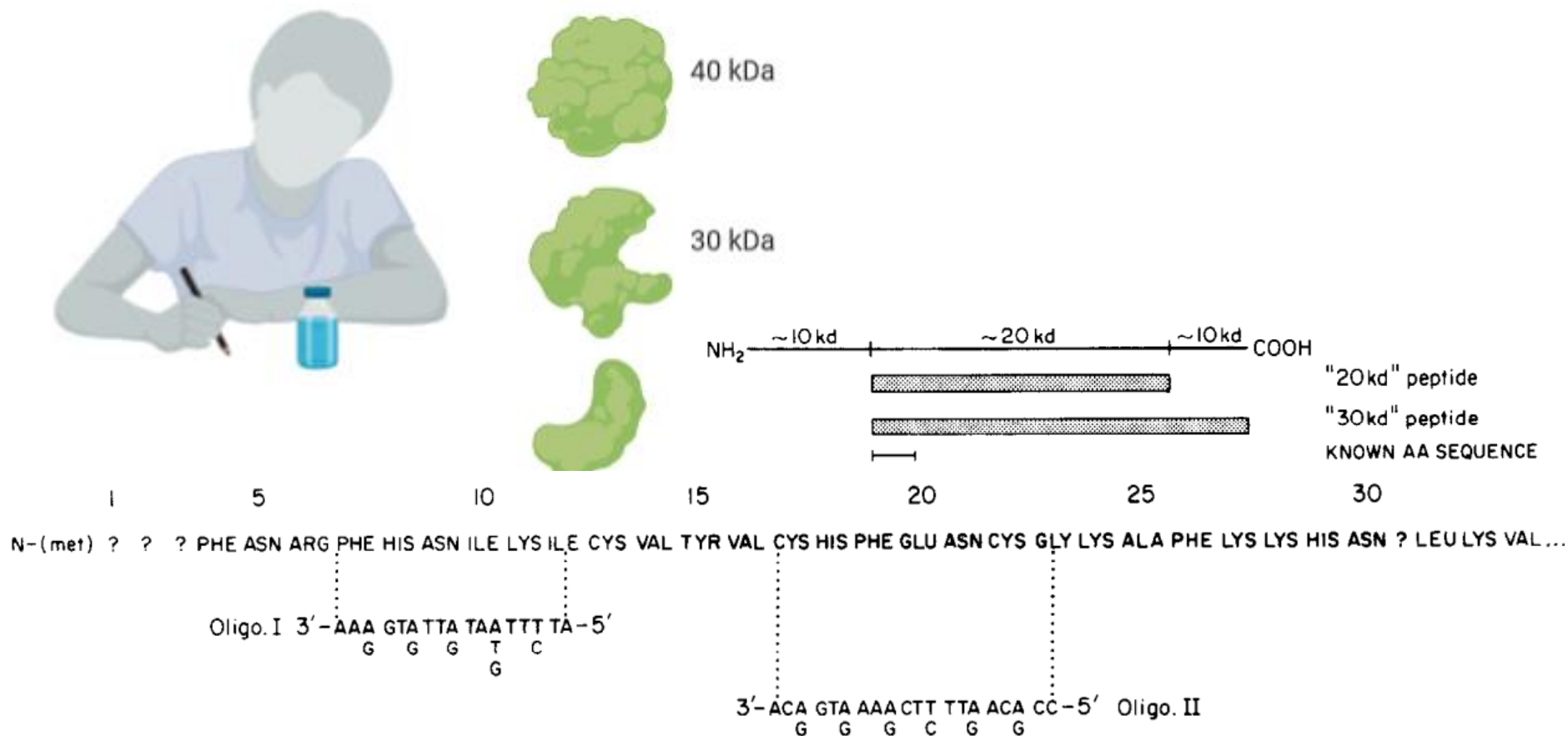
- Periodic intermediates &
- persistence of small fragments



Xenopus 5S Gene Transcription Factor, TFIIIA: Characterization of a cDNA Clone and Measurement of RNA Levels throughout Development

Ann M. Ginsberg, Balas O. King, and
Robert G. Roeder

Cell, Vol. 39, 479–489, December 1984



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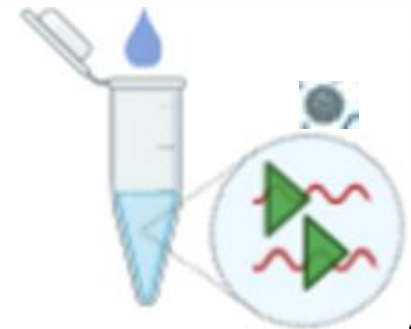
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1 8 13 17 23

=====

| T G E K * P (Y) V (C) . . D G (C) D K R (F) T K K . . (L) K

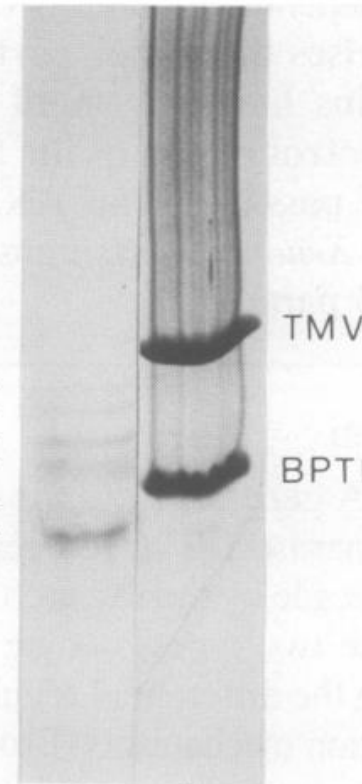
=====



1 (M G E K A L P V V Y K R)

.2

1	(Y) I (C) S F A D (C) G
2	T G E K * P (F) P (C) K E E G (C) E
3	T G E K * N (F) T (C) D S D G (C) D
4	N I K I C V (Y) V (C) H F E N (C) G
5	T Q Q L * P (Y) E (C) P H E G (C) D
6	A G - - * - (Y) P (C) K K D D S (C) S
7	Q D - - * L A V (C) - - D V (C) N
8	E K E R T V (Y) L (C) P R D G (C) D
9	E E Q R * P (F) V (C) E H A G (C) G



* A (H) L C * K (H)	37
* R (H) S L * T (H)	67
* K (H) F N R F (H)	98
* V (H) Q F * S (H)	129
* R (H) E K * V (H)	159
L K (H) V A E C (H)	188
* D (H) Q K * T (H)	214
* S (H) I Q S F (H)	246
* R (H) S V * V (H)	276

277

(D P E K R K L * K E K C P R P K R S I

S V S G T E K T D S L V K N K P S G T E I (N) G S L V L D K L T I Q)

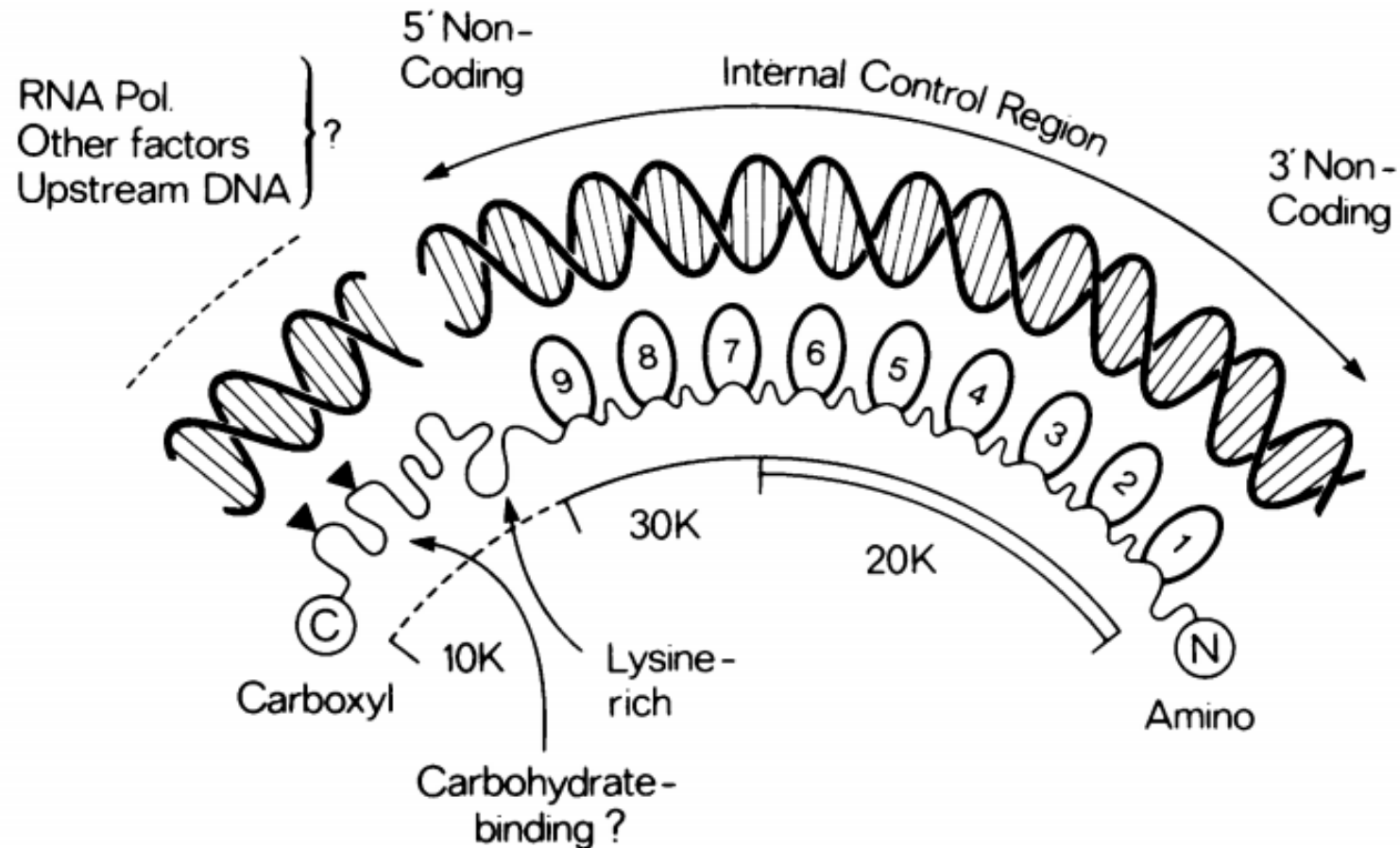
P K S K E K (N) A 311

344

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1 8 13 17 23 26 30

=====

T G E K * P (Y) V (C) . . D G (C) D K R (F) T K K . . (L) K * R (H) . . * . (H)

=====

1 (M G E K A L P V V Y K R)

12

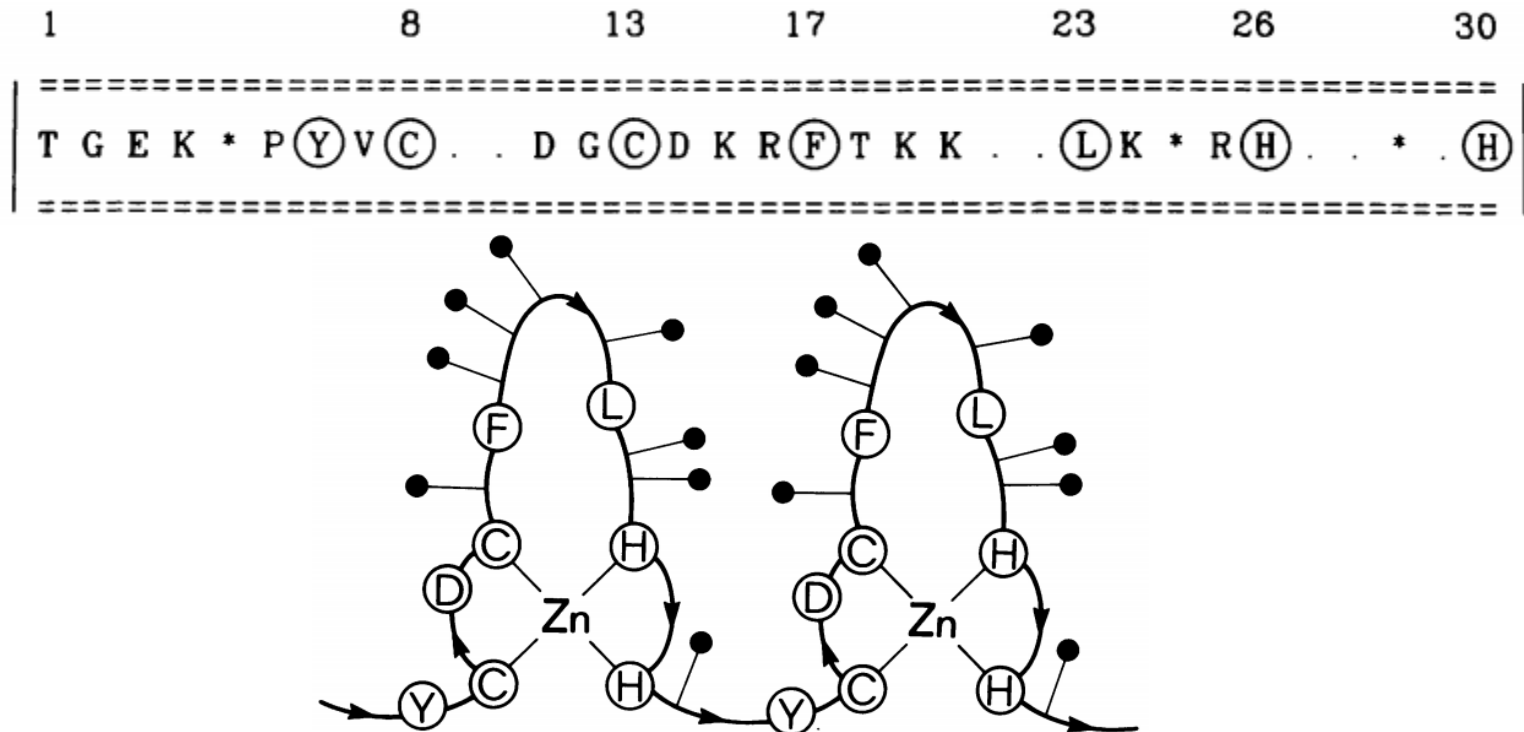
1	(Y) I (C) S F A D (C) G A A (Y) N K N W K (L) Q * A (H) L C * K (H)	37
2	T G E K * P (F) P (C) K E E G (C) E K G (F) T S L H H (L) T * R (H) S L * T (H)	67
3	T G E K * N (F) T (C) D S D G (C) D L R (F) T T K A N M K * K (H) F N R F (H)	98
4	N I K I C V (Y) V (C) H F E N (C) G K A (F) K K H N G (L) K * V (H) Q F * S (H)	129
5	T Q Q L * P (Y) E (C) P H E G (C) D K R (F) S L P S R (L) K * R (H) E K * V (H)	159
6	A G - - * - (Y) P (C) K K D D S (C) S (F) V G K T W T (L) Y L K (H) V A E C (H)	188
7	Q D - - * L A V (C) - - D V (C) N R K (F) R H K D Y (L) R * D (H) Q K * T (H)	214
8	E K E R T V (Y) L (C) P R D G (C) D R S (Y) T T A F N (L) R * S (H) I Q S F (H)	246
9	E E Q R * P (F) V (C) E H A G (C) G K C (F) A M K K S (L) E * R (H) S V * V (H)	276

277

(D P E K R K L * K E K C P R P K R S L A S R L T G Y I P P K S K E K (N) A 311

S V S G T E K T D S L V K N K P S G T E T (N) G S L V L D K L T I Q) 344

Repetitive zinc-binding domains in the protein transcription factor IIIA from *Xenopus* oocytes



- Key points = a new protein fold for nucleic acid binding
- A novel principle of DNA recognition
- As design of specific DNA recognition was distinct from helix turn helix motifs

Conclusions of Lecture-10

- TF IIIA binds 5S RNA (7S) and 5S DNA.
- TF contain three structural domains: further digestion 3K
- TFIIA contains repetitive Zinc binding domains.
- Amino acid sequence revealed 9 similar units.
- Each unit is of approximately 30 residues.
- These 30 residues contain two invariant pairs of Cys and His.
- Each unit fold independently and is centered on Zn ion.

Questions??