Interaktionen von RNAs und Proteinen

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DNA-DNA Interactions: Nuclear Organization

A. Interphase nucleus

- Chromosome territory (CT borders)
- Euchromatin
- Heterochromatin
- Nuclear pore
- Nucleoli

B. Prometaphase nucleus

- Chromosome
- Nuclear membrane
Nuclear Bodies Associated and Their Function

- Nucleolus: Associated with rDNA transcription and post-translational modifications.
- Nuclear Speckle: Involved in RNA splicing and stress response.
- Nuclear Stress Body: Related to stress, HS gene, and Pol II.
- Transcription Factory: Involved in transcription.
- Cajal Body: Associated with pre-snRNA and snRNP biogenesis.
- Gemini of Cajal Body: Unknown function.
- Histone Locus Body: Involved in histone pre-mRNA synthesis.
- Paraspeckle: Associated with A-to-I edited RNA and nuclear RNA retention.

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**Organization of Nuclear Bodies by Long Noncoding RNAs**

**arcRNAs**: architectural long noncoding RNAs

- nascent arcRNAs serve as scaffold for RNA binding proteins
- results in strong local enrichment of specific factors
Chromosome Territories

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Attachment of Chromatin to Nuclear Envelope

- S/MARs: Scaffold/Membrane attachment regions

Key:
- Green: Active gene
- Red: Repressed gene
- Blue: Enhancer
- Yellow: Promoter gene
- Gray: Lamin
A Special Territory: The Inactive X Chromosome

Dosage compensation in placental mammals

WHAT is happening?

- random X-inactivation
- female: $X, X \rightarrow X_a$ (active), $X_i$ (inactive)
- male: $X, Y \rightarrow X_a$
X Chromosome Inactivation (XCI)

HOW is it happening?

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XCI – Counting and Choosing

A (a) Autosomes work as a diploid pair to create a single blocking factor

X chromosomes bound by blocking factor remains active
Remaining X chromosome(s) inactivates

(b) Autosomes create a swarm of blocking factors

X chromosome with blocking factor remains active
X chromosomes bound by competence factor inactivate

B

X chromosomes create competence factors
Blocking factor neutralizes one of two competence factors

C

X chromosomes

serveral model try to explain Interaktionen von RNAs und Proteinen
XCI – Main Player(s) Xist (and Tsix)

- IncRNA **Xist** (X-inactive specific transcript) expressed from inactive X chromosome is responsible for silencing the X in *cis* → $X_i$ (inactive)
- IncRNA **Tsix** (antisense of Xist) expressed from active X chromosome is responsible for silencing Xist in *cis* → $X_a$ (active)
The X Inactivation Center (Xic)

- **Xist**: X-inactivation specific transcript
  - 17kb, spliced and polyadenylated transcript
- **Tsix**: antisense RNA of Xist
- genes (RNA or protein coding) promoting Xist expression
- genes (RNA or protein coding) suppressing Xist expression
Proposed mechanisms

- Xist has binding sites for proteins (similar to arcRNAs)
- SPEN binds “repeatA” on Xist
  interacts with SMRT-HDAC3 \( \rightarrow \) deacetylation
  may recruit PRC2 \( \rightarrow \) H3K27me3 (repressive mark)
  may recruit PRC1 \( \rightarrow \) H2AK119ub (repressive mark)
  excludes PolII from Xist-coated chromosome
- RBM15-WTAP interacts with Xist
  recruits m\(^6\)A machine that methylates Xist
  YTHDC1 reads m\(^6\)A enables transcriptional silencing
- **LBR** (lamina binding receptor) links Xist-coated chromatin to lamin in the lamina
Xist Spreading and Lamina Localization

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TADs at the Xic

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Measuring Chromatin Interactions

Chromosome Conformation Capture

- a method to measure chromatin-chromatin interactions

Hi-C Method
Topologically Associated Domains (TADs)

Structure of TADs

- Inactive TAD 1
- Active TAD
- Inactive TAD 2

Subdomains

Boundary Interactions
CTCF motifs
H3K36me3
H3K27me3

Interactions
Enhancers
Genes

Current Opinion in Genetics & Development
Topologically Associated Domains (TADs)

Functional/Regulatory Relevance
van Bemmel et al.  
*The bipartite TAD organization of the X-inactivation center ensures opposing developmental regulation of Tsix and Xist*  

Cerase et al.  
*Phase separation drives X-chromosome inactivation: a hypothesis*  
Nature Structural and Molecular Biology 26, 331–334 (2019)

Chen et al.  
*Xist recruits the X chromosome to the nuclear lamina to enable chromosome-wide silencing*  
Science 6311(354), 468–472 (2016)

Wang et al.  
*Comment on “Xist recruits the X chromosome to the nuclear lamina to enable chromosome-wide silencing”*  
Science 6343(356), eaal4976 (2017)

Pollex et al.  
*Nuclear positioning and pairing of X-chromosome inactivation centers are not primary determinants during initiation of random X-inactivation*  

da Rocha et al.  
*Novel players in X inactivation: insights into Xist-mediated gene silencing and chromosome conformation*  
Nature Structural and Molecular Biology volume 24, pages 197–204 (2017)