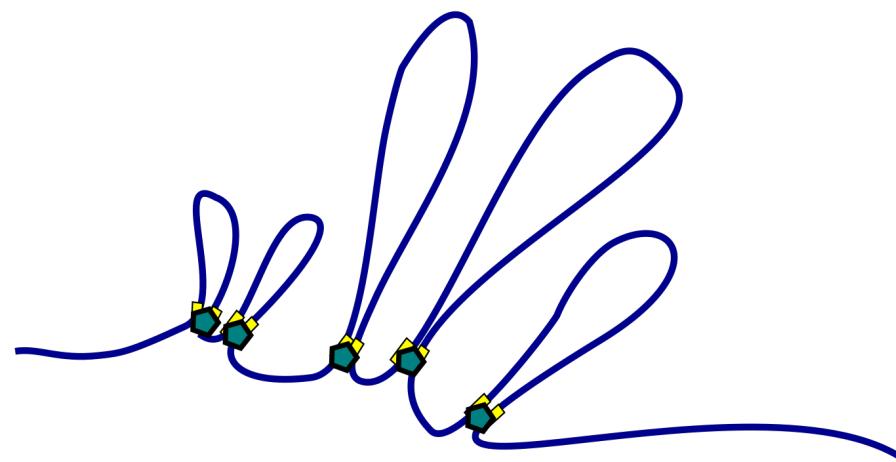


Hackathon ENGiE - Pasteur

Où sont les boucles chromosomiques?

*Unité Régulation Spatiale des Génomes,
Institut Pasteur*



4, 5 Octobre 2018, COMET, Paris

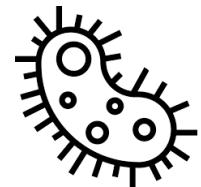
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- ▶ Présentation de notre équipe et de notre activité de recherche
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- ▶ Les boucles chromosomiques et le défi lancé!
- ▶ Session interactive: prise en main des données et visualisation

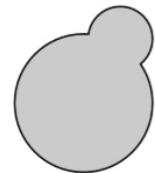
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L'équipe Régulation Spatiale des génomes



Bactérie



Levure

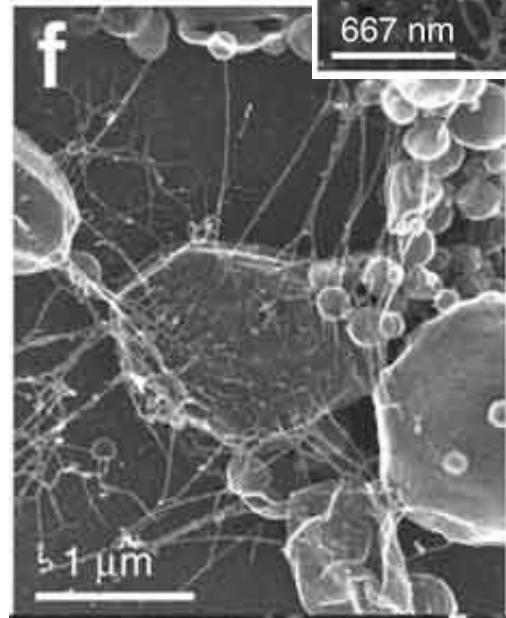
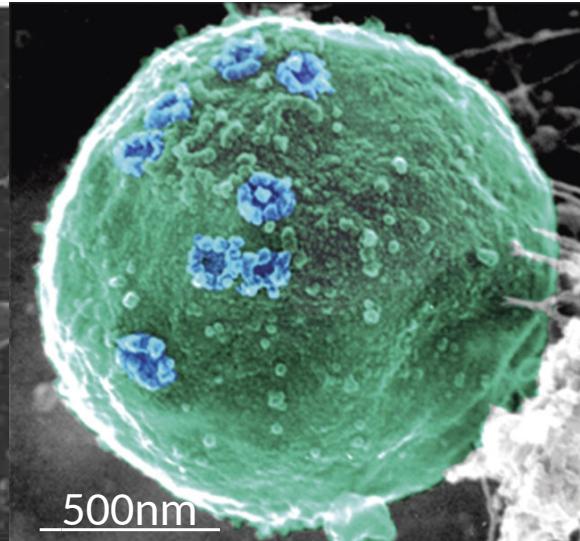
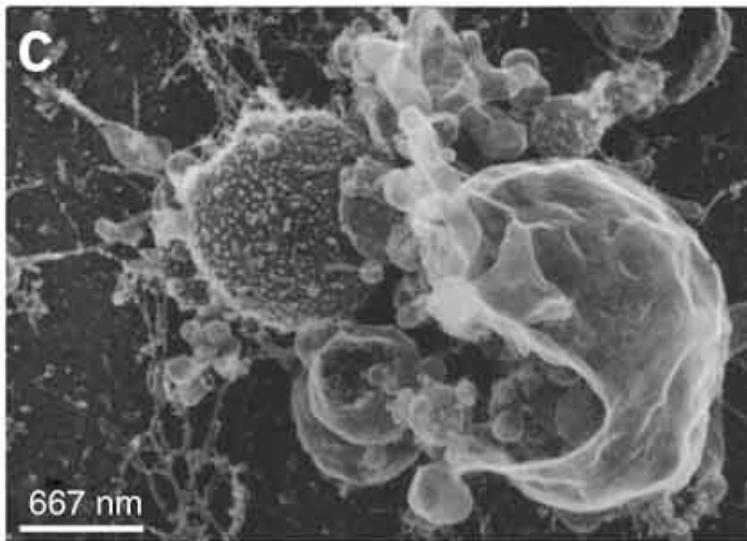
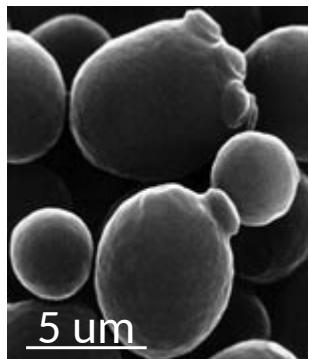


Humain

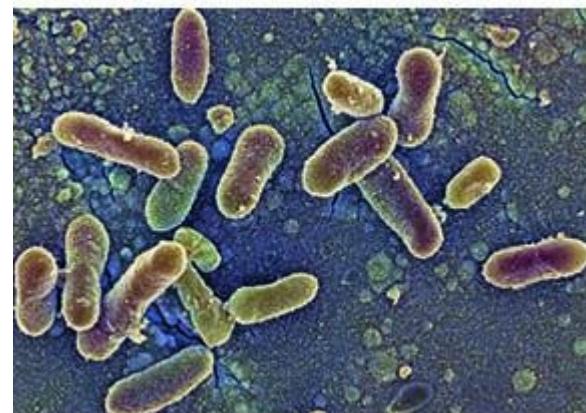


Institut Pasteur

Exemples de photos de levures



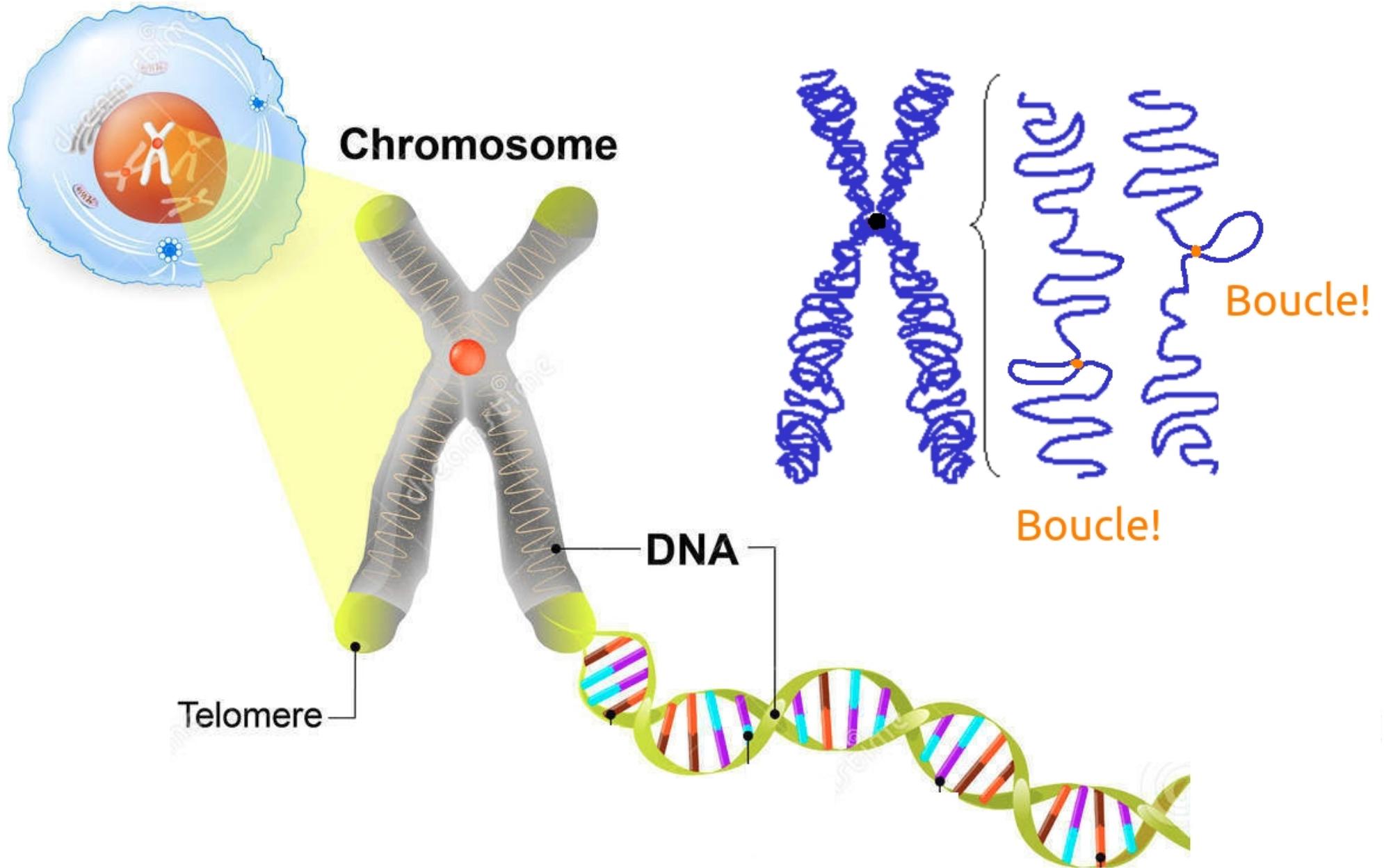
Kiseleva et al., 2007
Scanning electron microscopy



Exemple de photos de bactérie

Votre cellule

Rappels



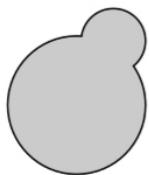
Les chromosomes sont les molécules (ADN enroulés avec d'autres protéines) porteuses de l'information génétiques.

Quelques chiffres



Bactérie

1 chromosome, 5 millions de bases, 4,000 gènes



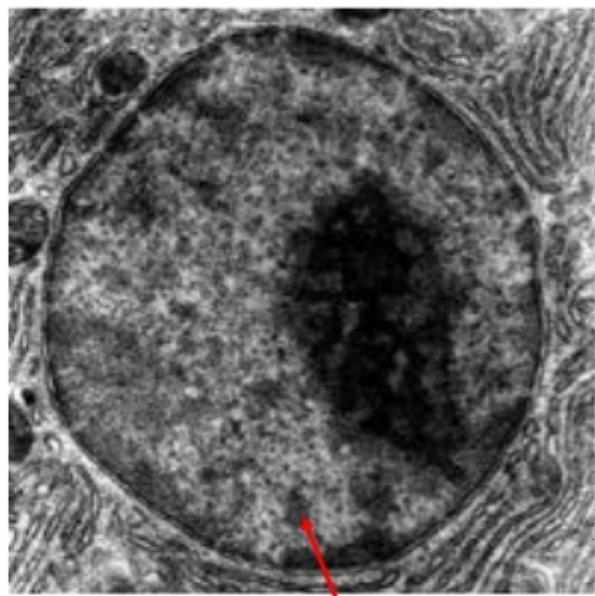
Levure

16 chromosomes, 12 millions de bases, 6,000 gènes



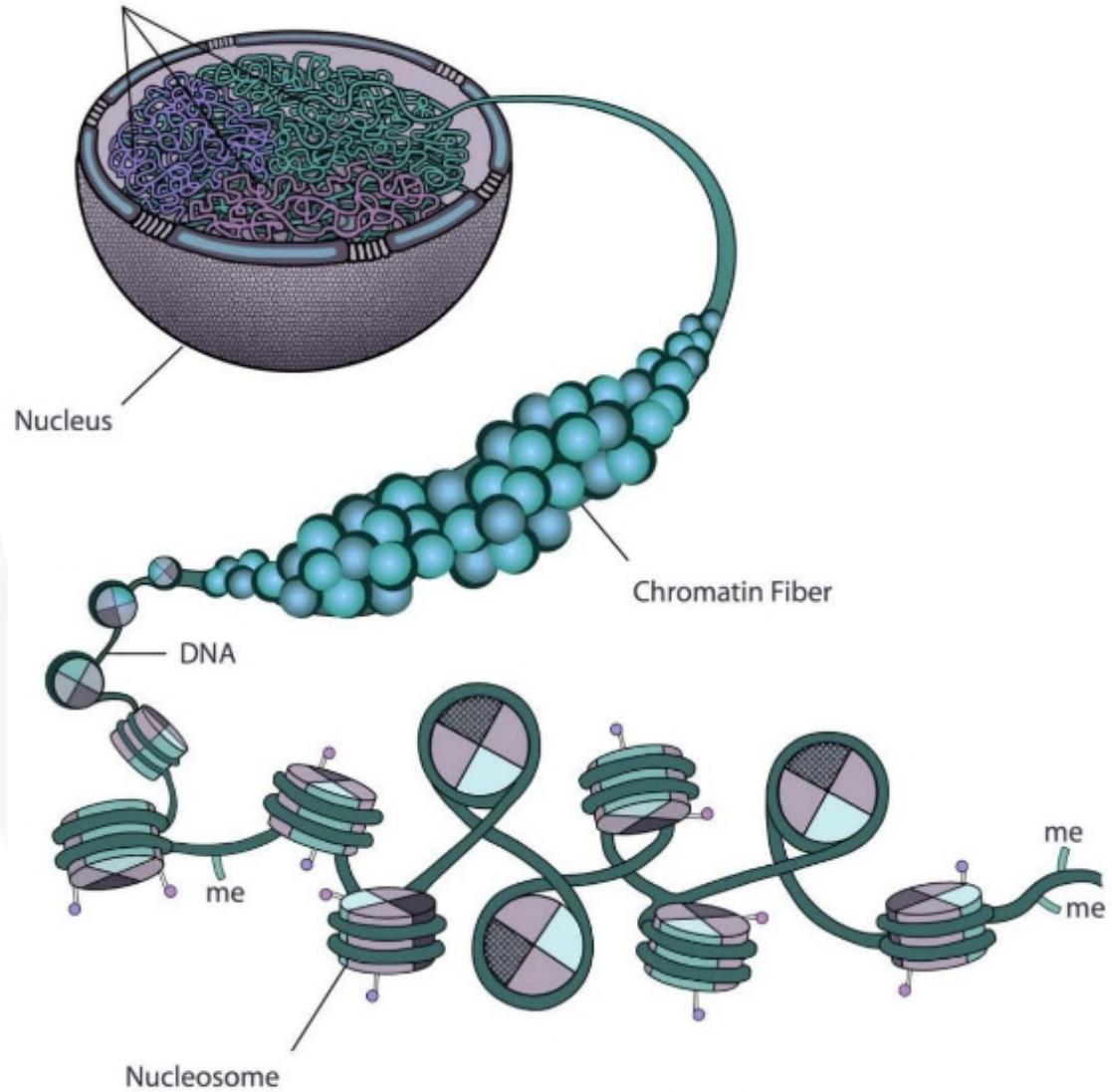
Humain

24 paires de chromosomes, 3,5 milliards de bases, 25,000 gènes



~2 meters
of DNA

~ 10 μm



Rosa & Shaw, 2013

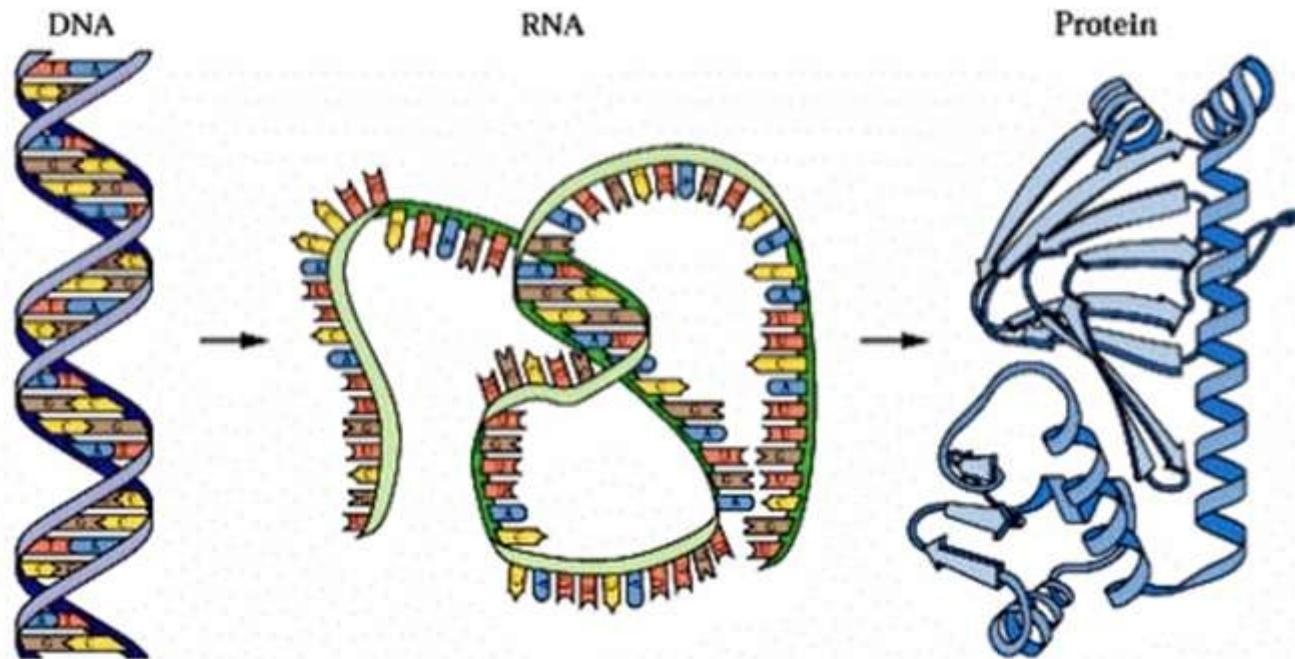
→ Nécessité de compacter la longue molécule d'ADN!

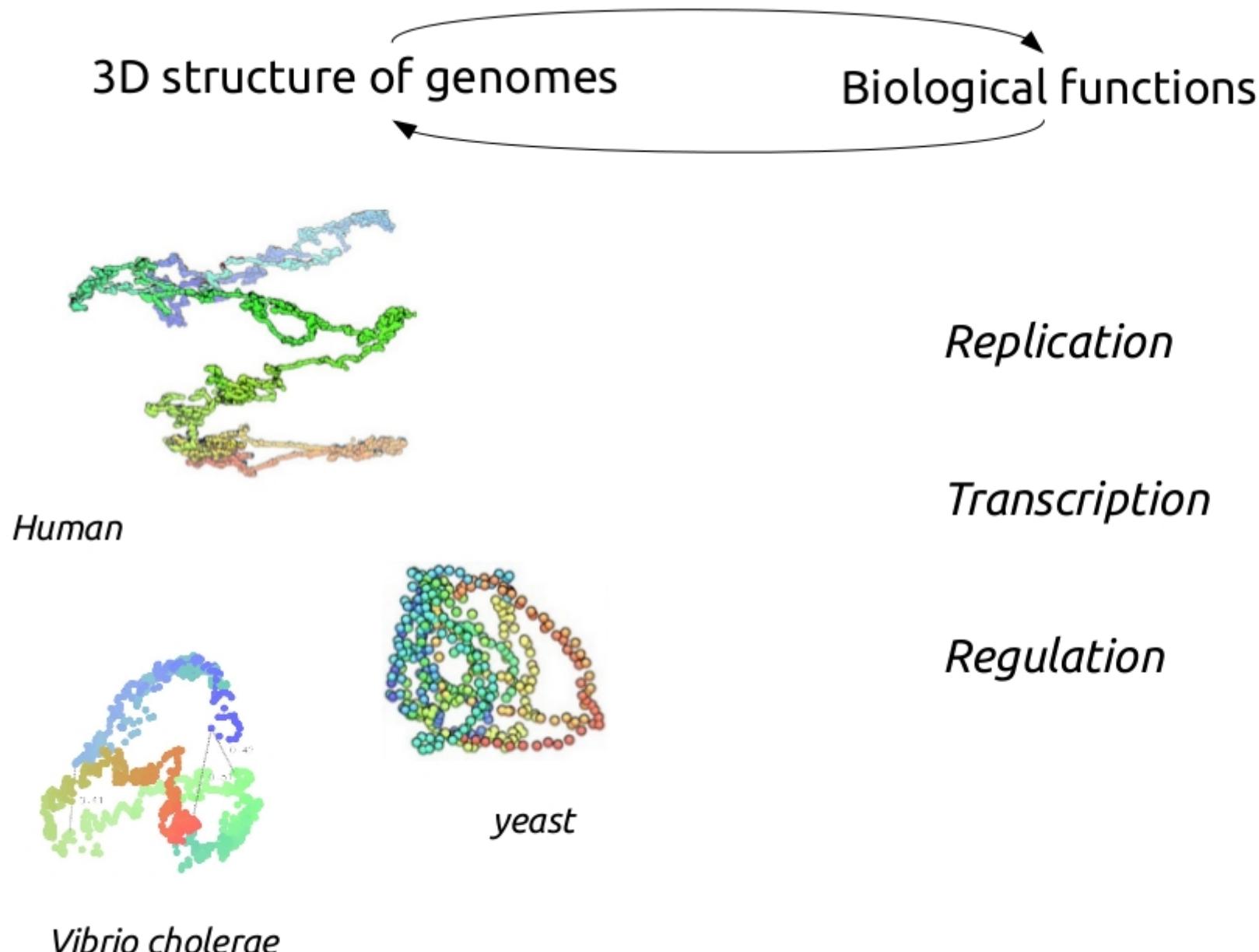
Rappels: dogme central de la biologie moléculaire

ADN → ARN → Protéine

Transcription

Traduction

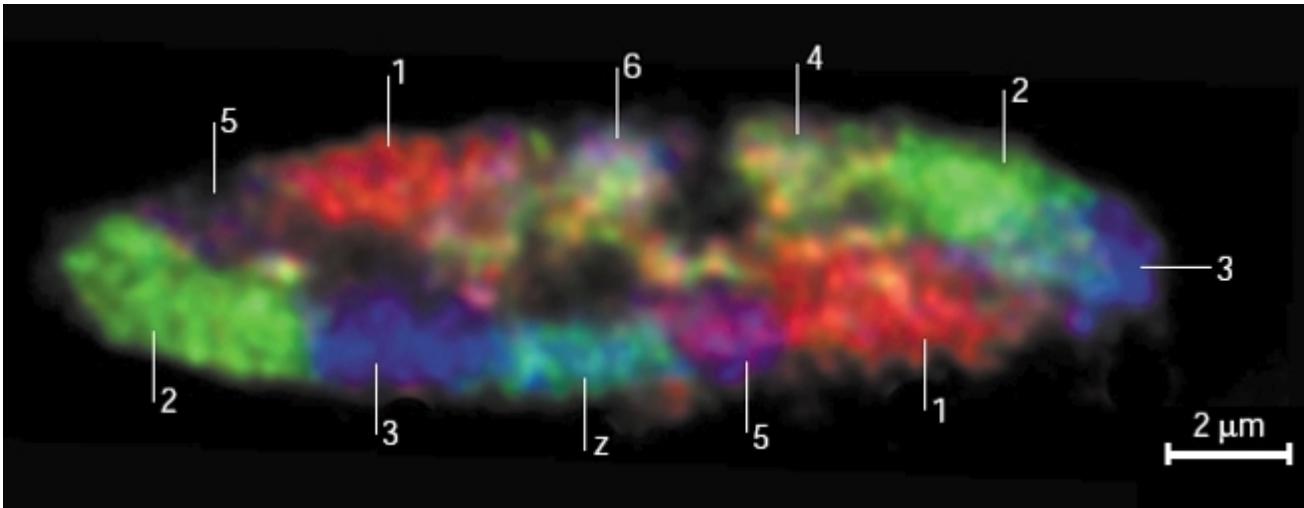




Notre équipe s'intéresse aux liens entre la structure 3D des chromosomes et certaines fonctions biologiques.

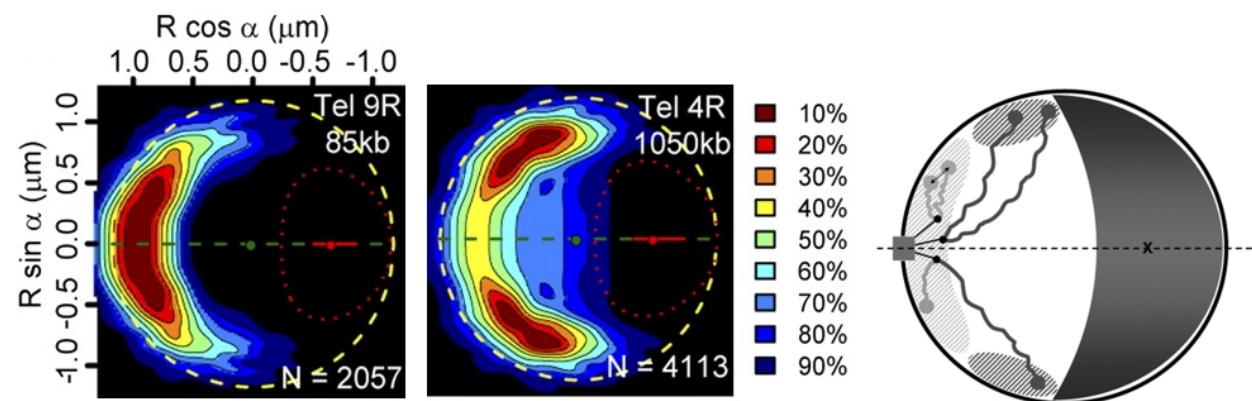
Comment visualiser la structure des chromosomes ?

Utilisation de la fluorescence et microscopie



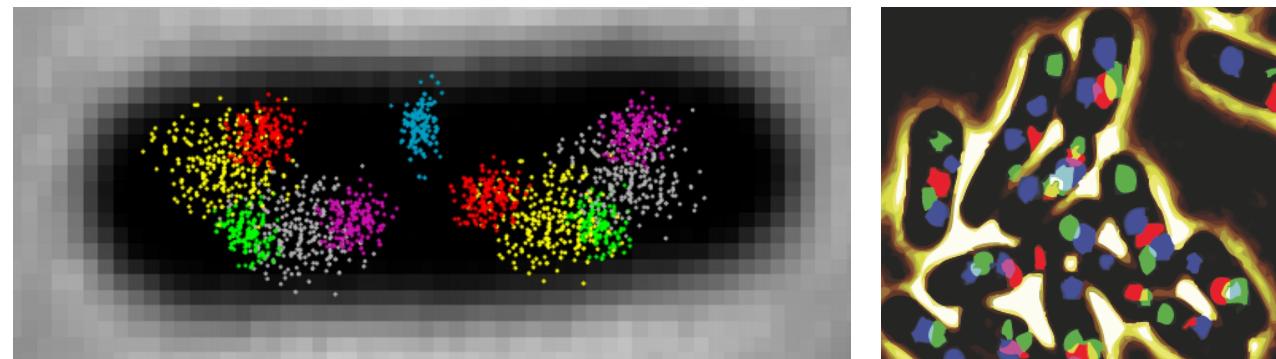
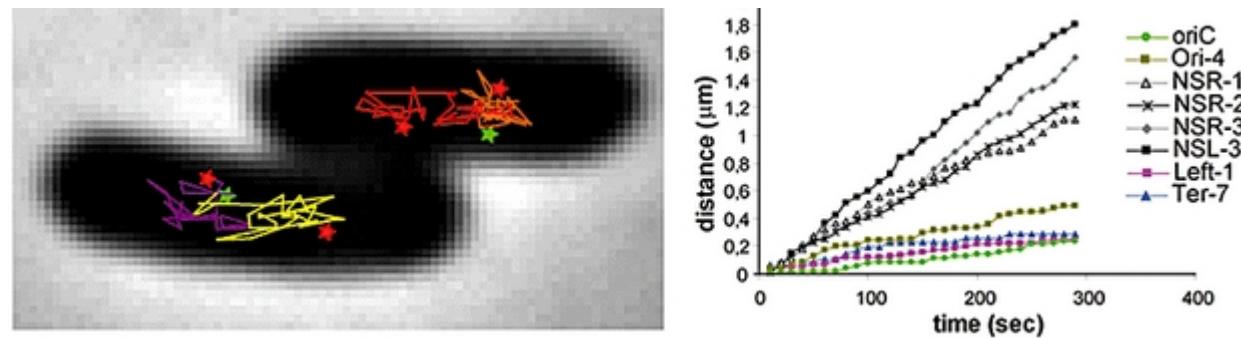
Chromosome territories in the chicken studied by FISH.

T. Cremer & C. Cremer Nature Reviews Genetics 2001



Effect of Chromosome arm length in yeast.
P. Thérizol et al. PNAS 2009

Des expériences pour observer la mobilité de gènes chez la bactérie E.Coli



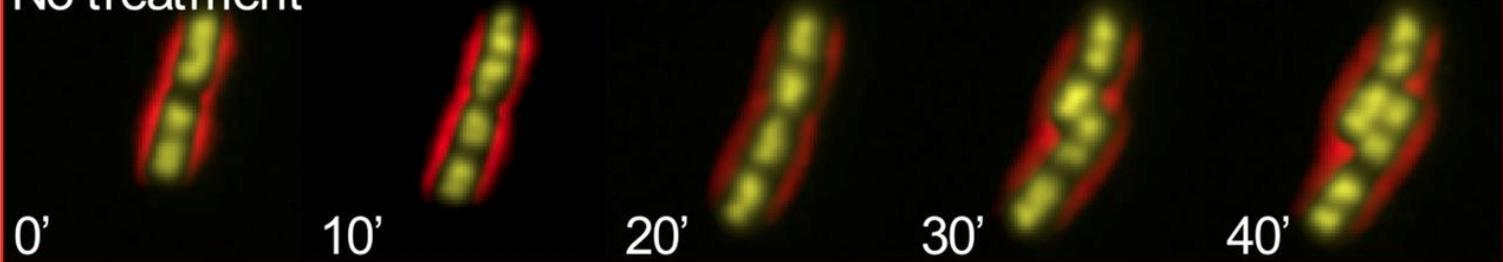
O. Espeli et al., Mol. Mic. 2008

Certains gènes bougent plus que d'autres!

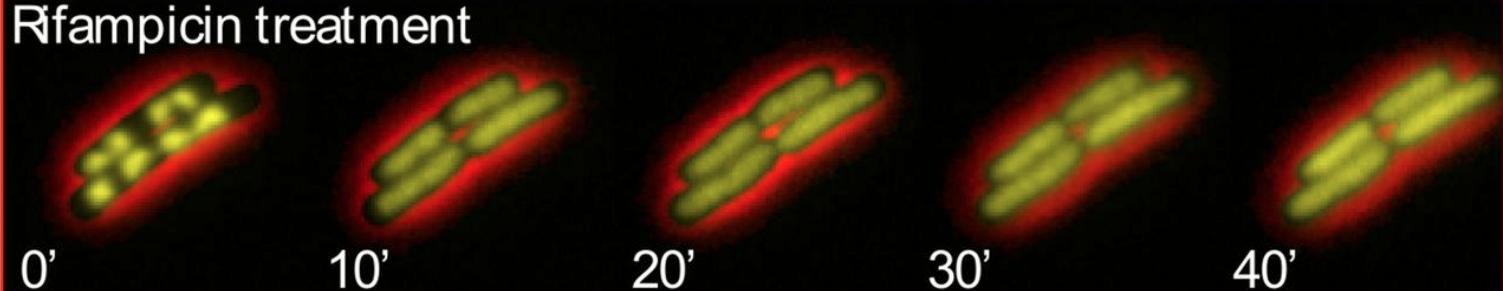
Effet de la transcription ou de la traduction sur le chromosome d'E.Coli

Timelapse of HU-mCherry strains growing on LB pads at 30°C.

No treatment

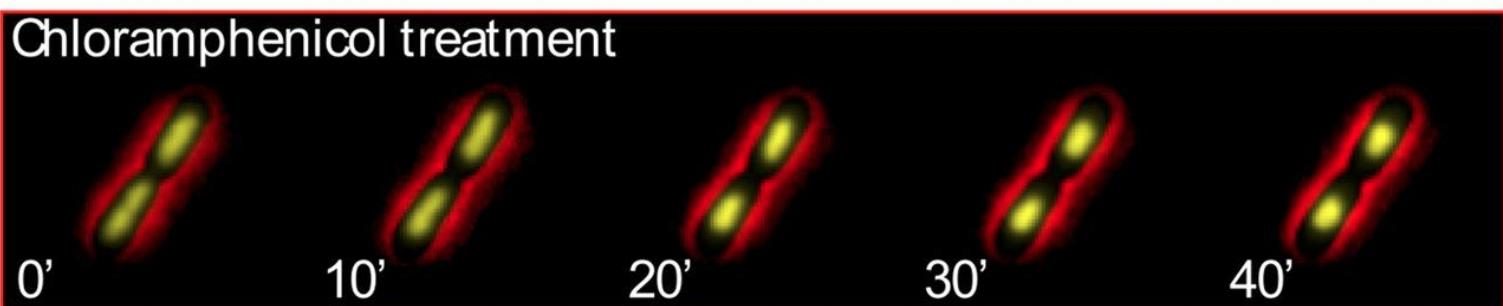


Rifampicin treatment



No transcription

Chloramphenicol treatment

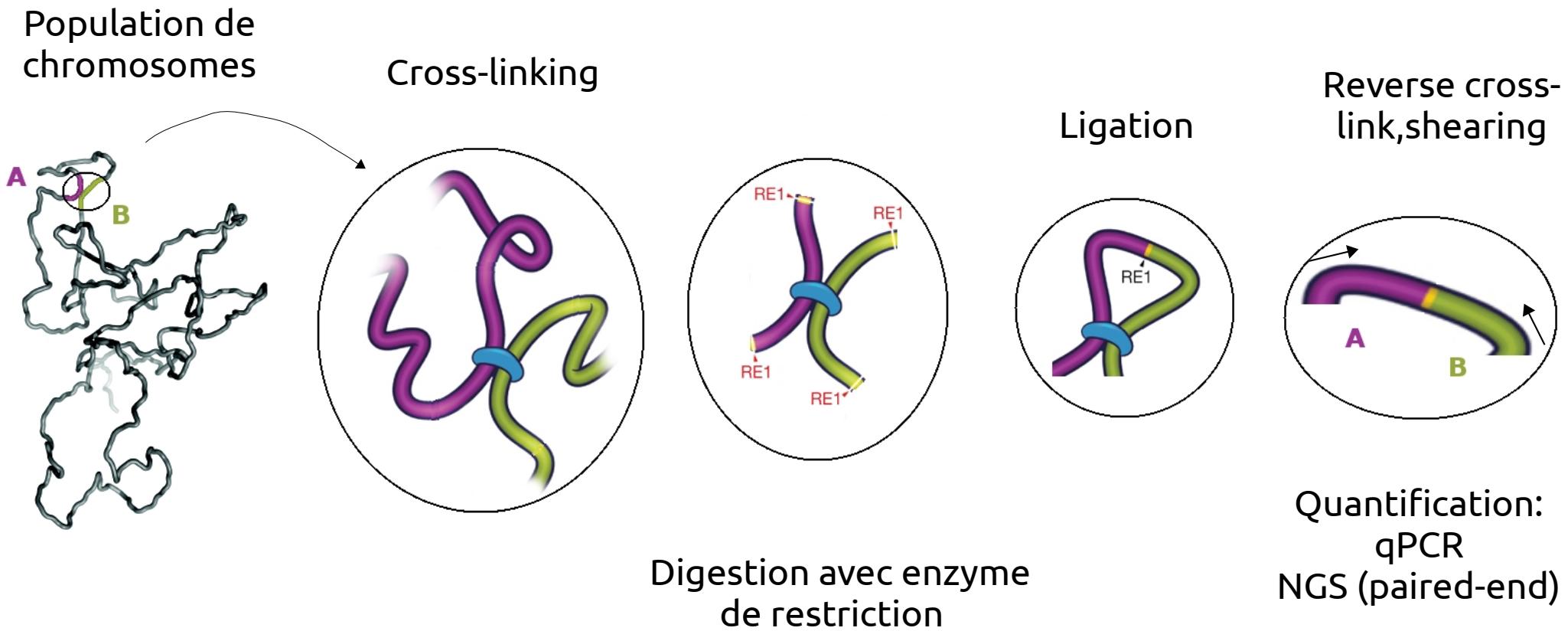


No traduction

Présentation introductory

- ▶ Présentation de notre équipe et de notre activité de recherche
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Hi-C, 3C (Conformation Chromosome Capture) technologies



La technique du 3C piège les fragments d'ADN qui sont physiquement proches à 3D.

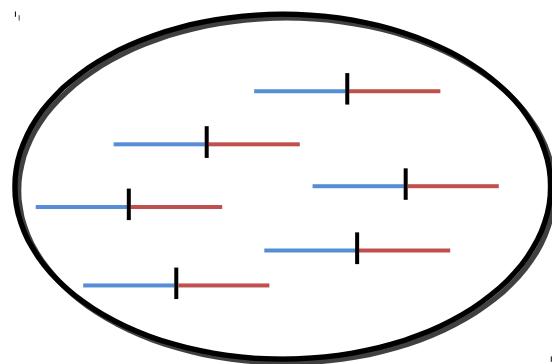
Dekker et al. Science 2002,
Lieberman-Aiden, van Berkum et al., 2009

A la pailasse

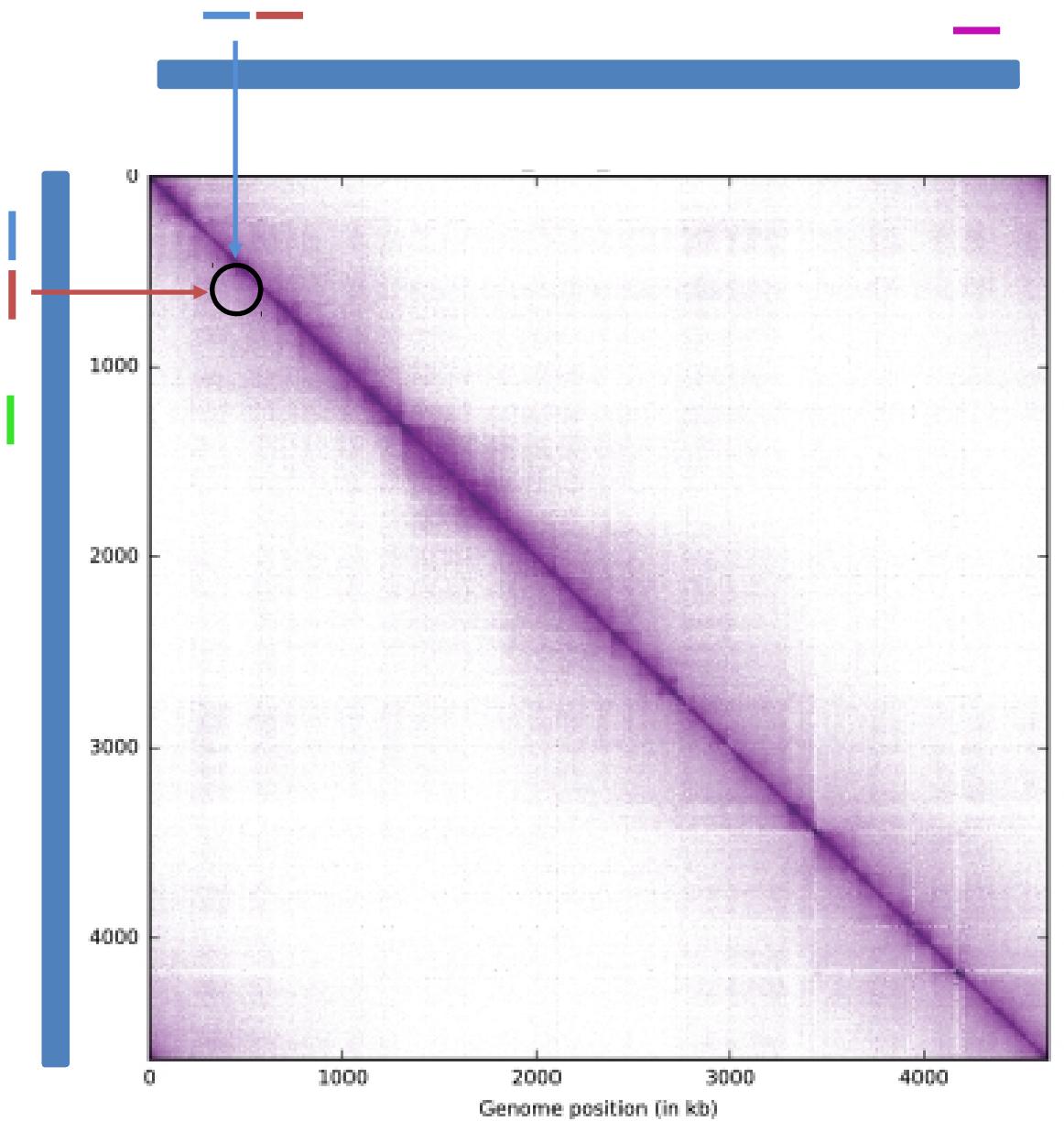
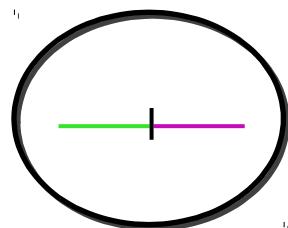


Example de Contact Map: *Escherichia coli* chromosome

Close proximity → Frequent contacts



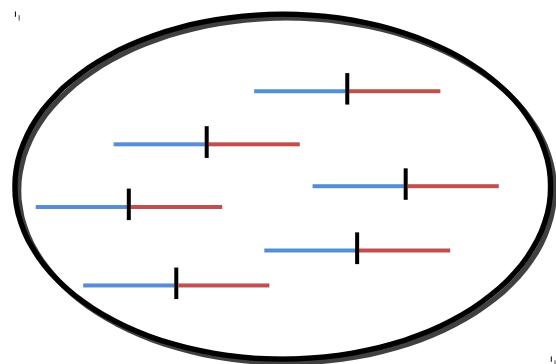
Far away → Rare contacts



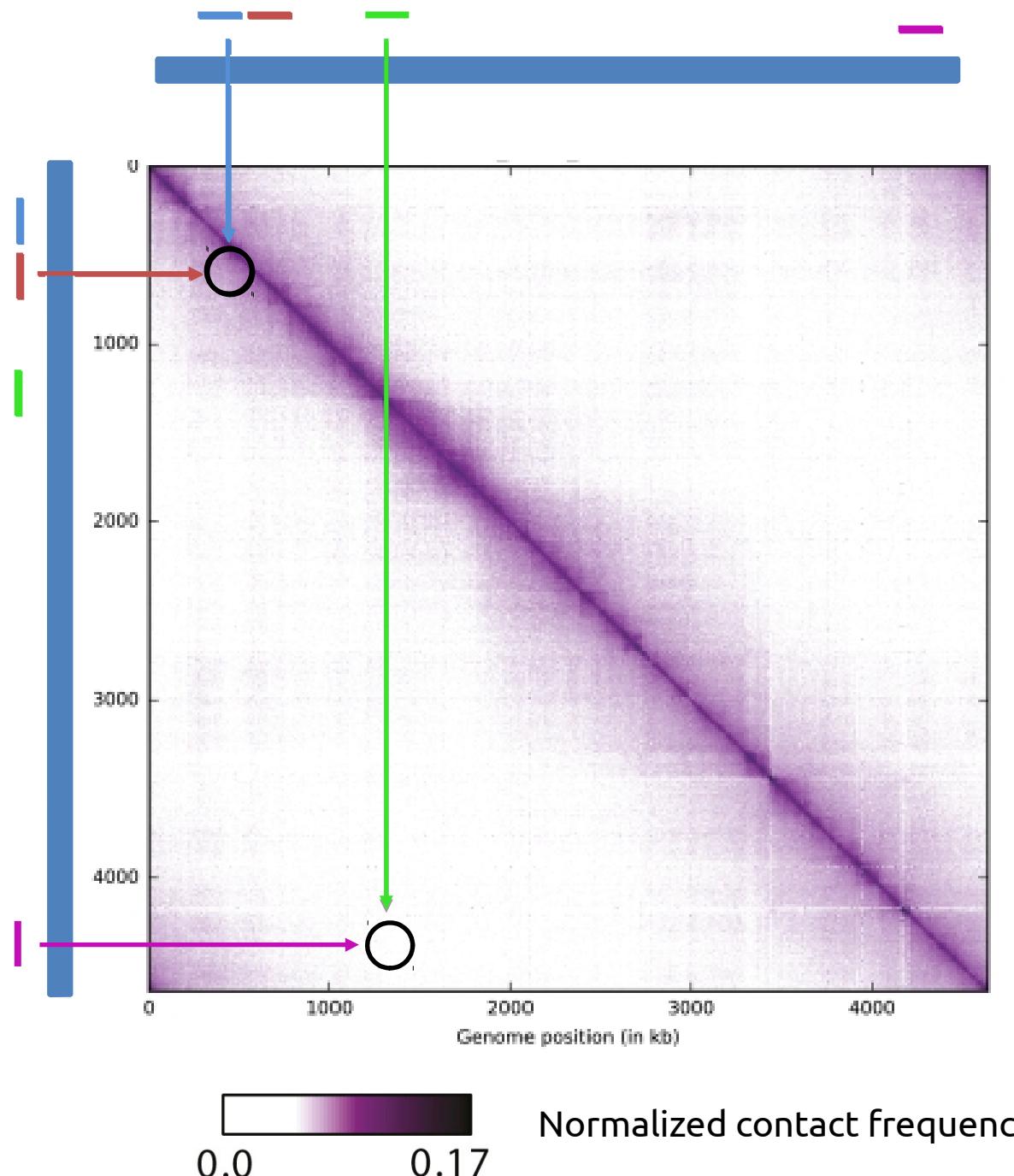
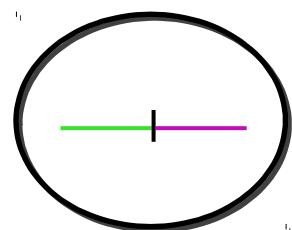
Normalized contact frequency

Example of Contact Map: *Escherichia coli* chromosome

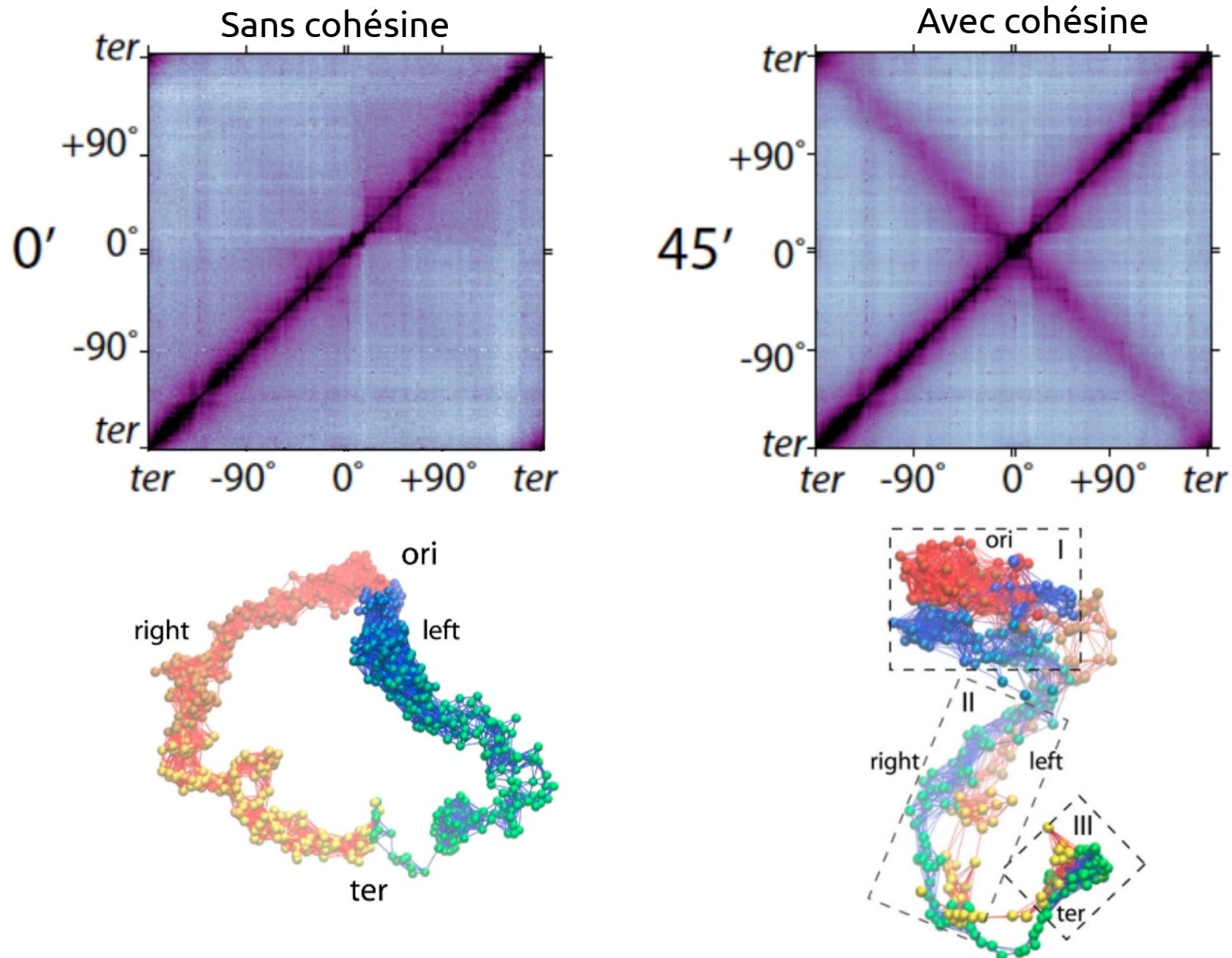
Close proximity → Frequent contacts



Far away → Rare contacts

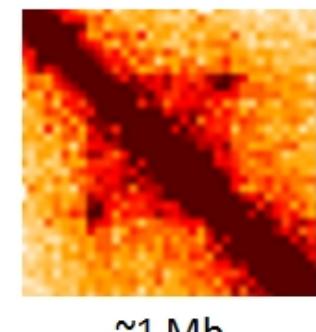
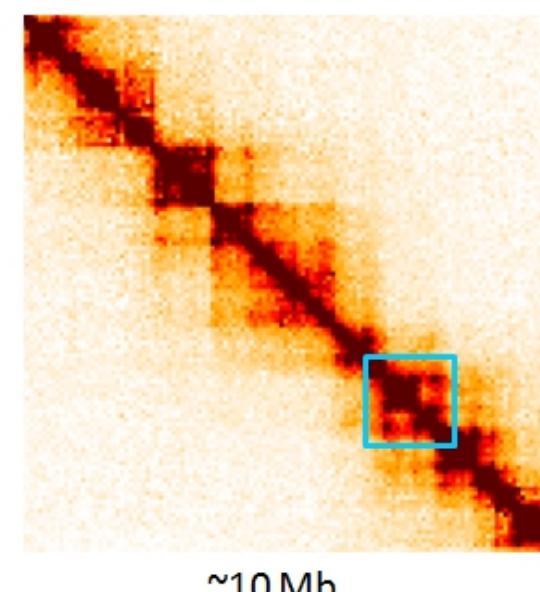
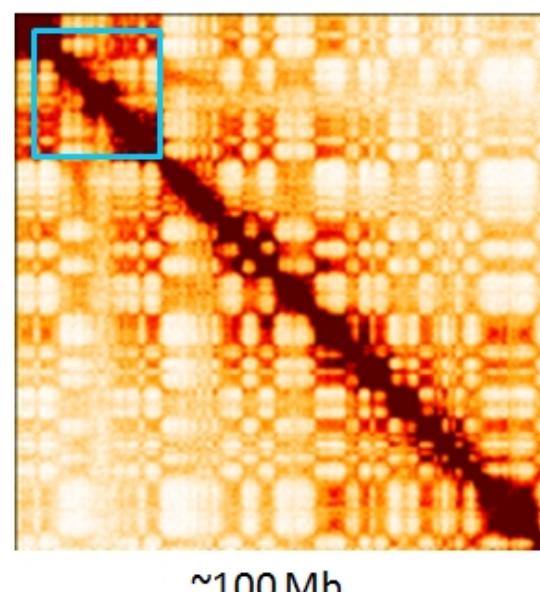
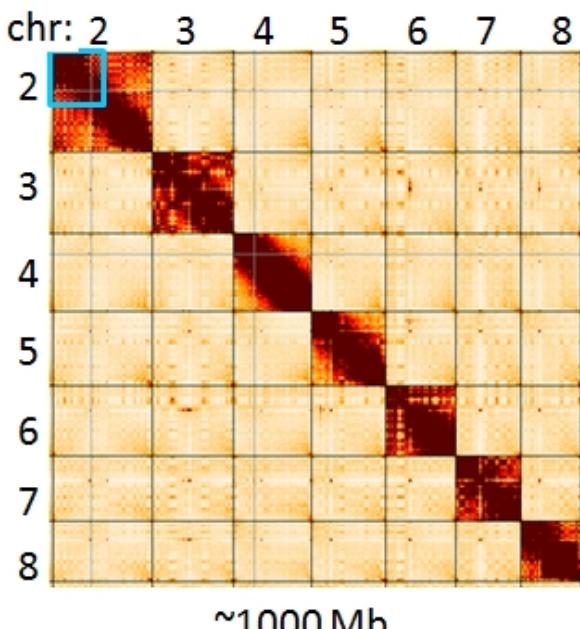


Exemple de découverte faite grâce à la technologie 3C

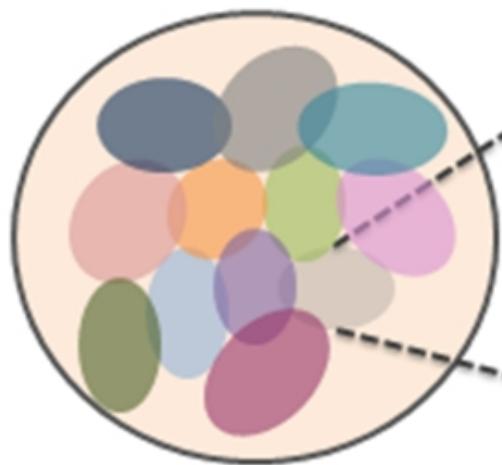


TB Le et al. Science 2013, X Wang et al. Science 2017
M Marbouty, A Le Gall et al. Mol Cell, 2015

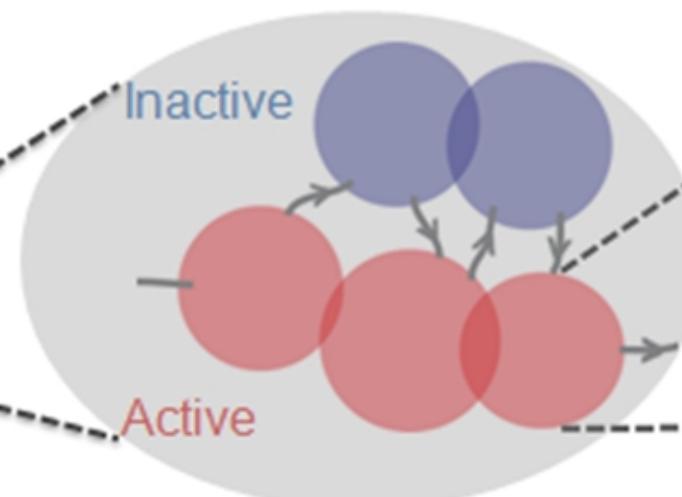
Exemples de découvertes faites grâce à la technologie 3C



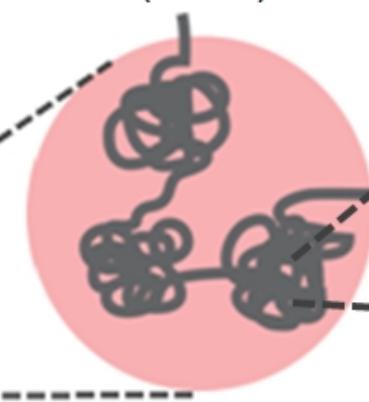
Chromosome Territories



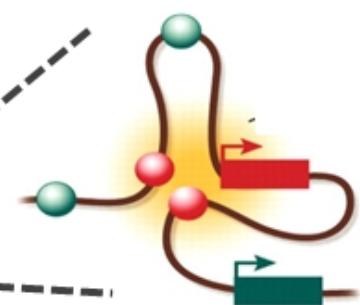
Compartments



Topological Domains (TADs)



Enhancer-Promoter Loops



La technologie 3C a révélé une organisation hiéarchique des chromosomes.

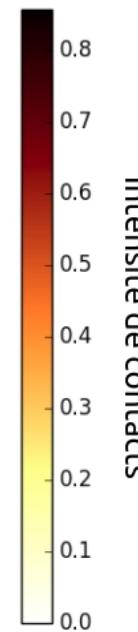
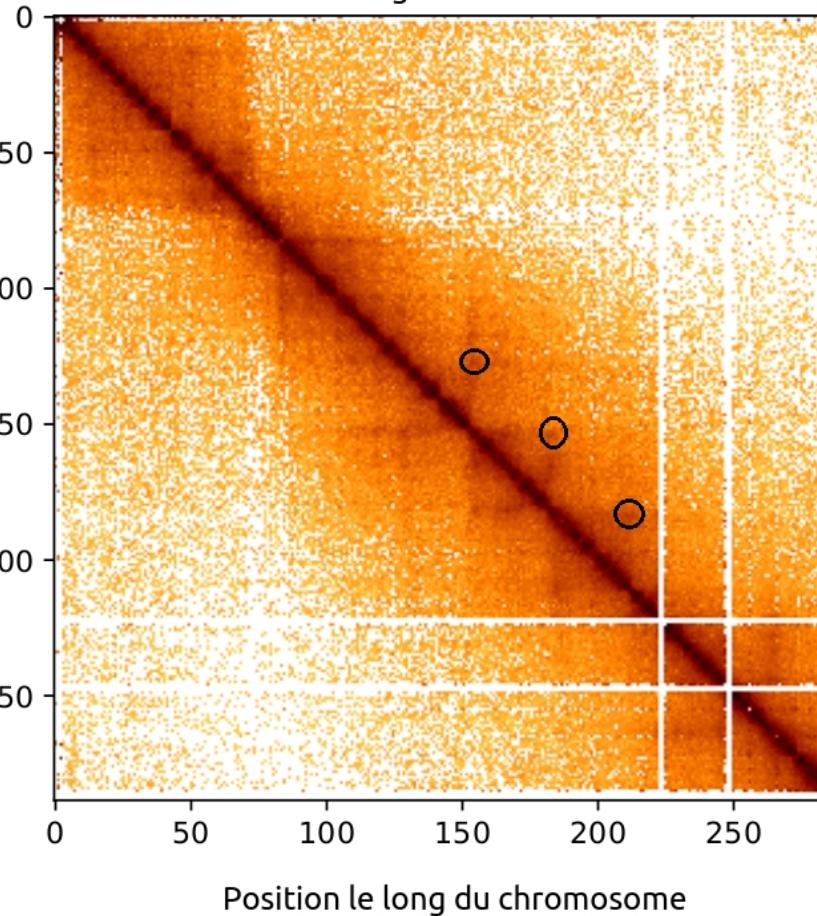
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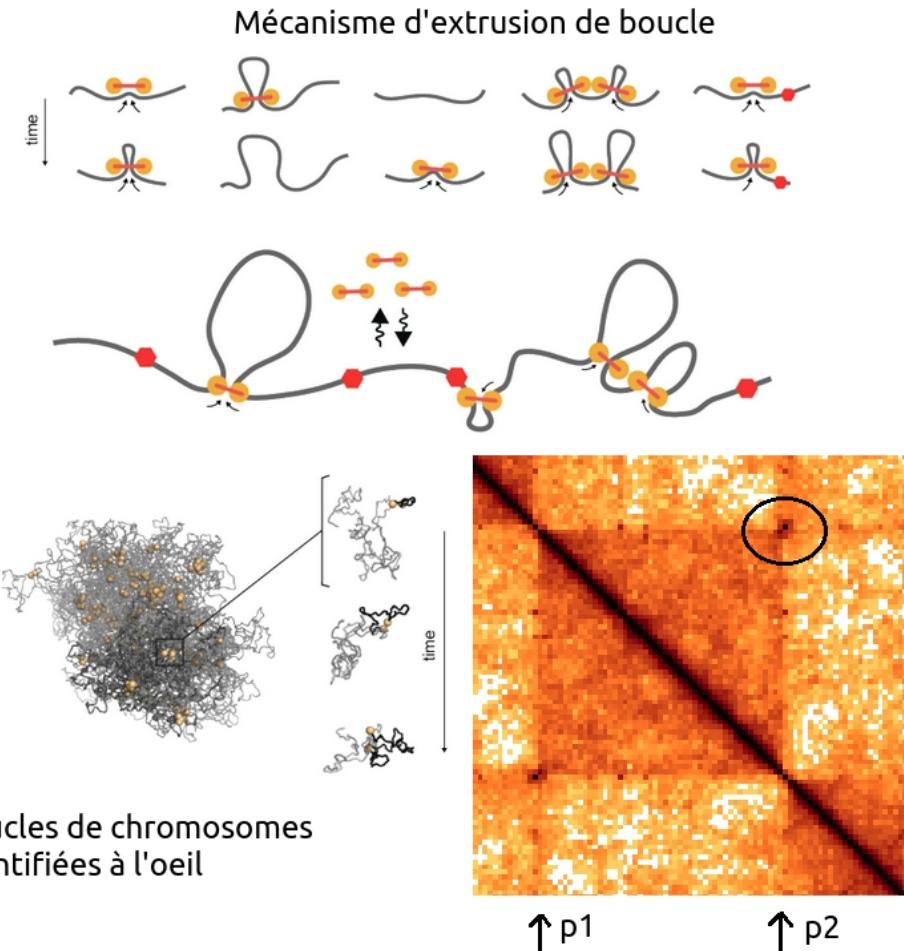
Les boucles chromosomiques

Carte de contacts expérimentale du chromosome 5
de la levure du boulanger

Position le long du chromosome



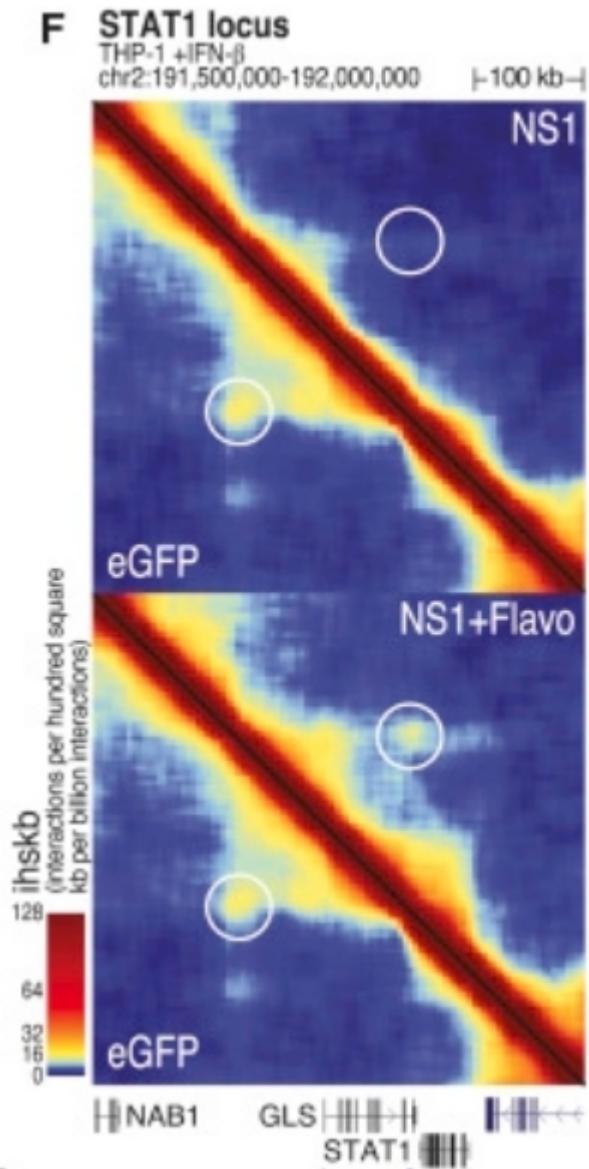
○ boucles de chromosomes
identifiées à l'oeil



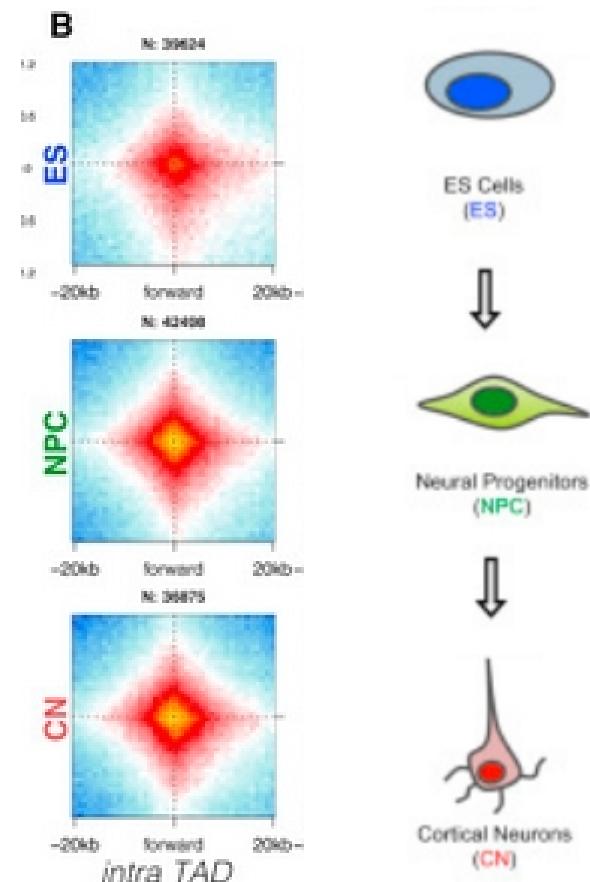
Carte de contact issue de
simulations avec une boucle
liant les positions p1 et p2

boucle chromosomique = contact stable entre deux régions éloignées sur le chromosome

L'importance des boucles chromosomiques dans plusieurs champs de la biologie contemporaine

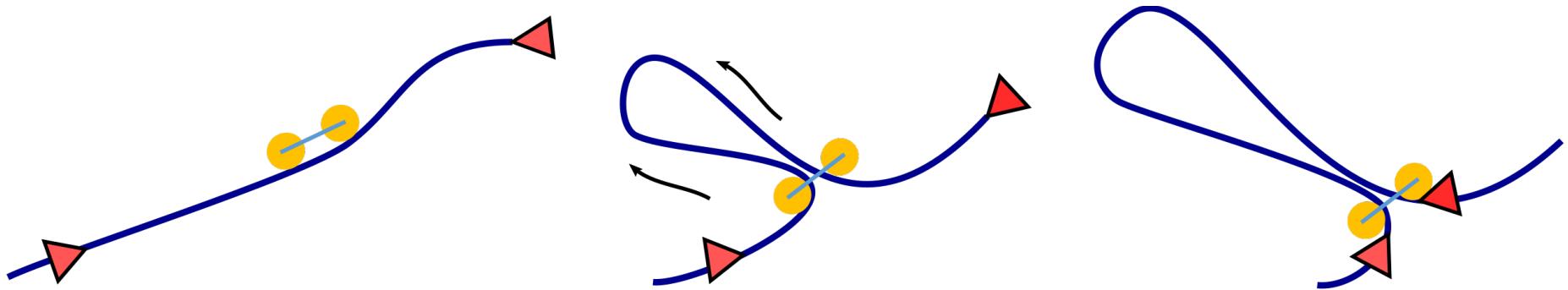


Certaines boucles disparaissent
lors d'une infection virale.
Heinz et al., Cell 2018

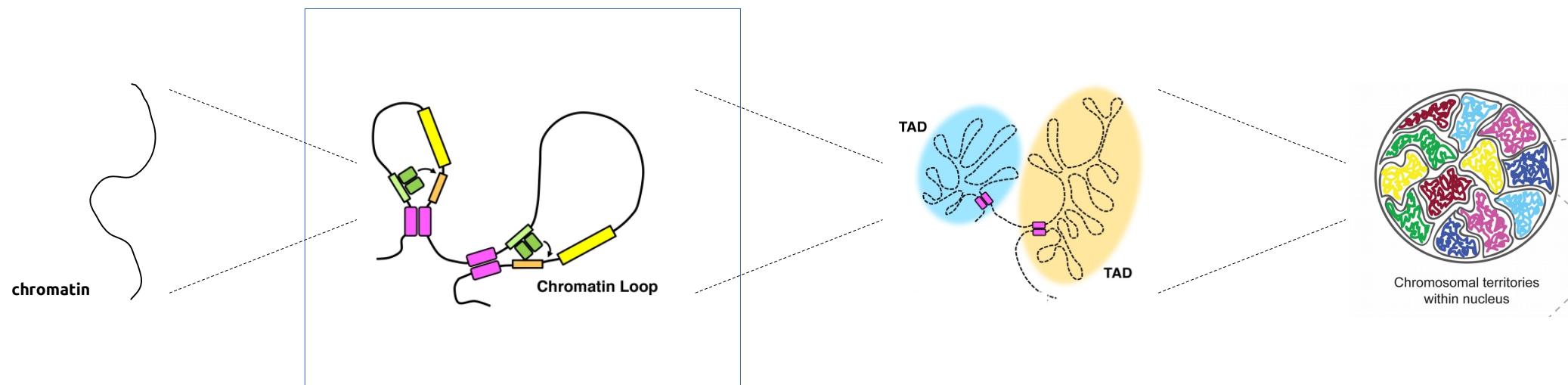


Certaines boucles deviennent plus
fortes durant la différenciation
cellulaire.
Bonev et al. Cell 2017

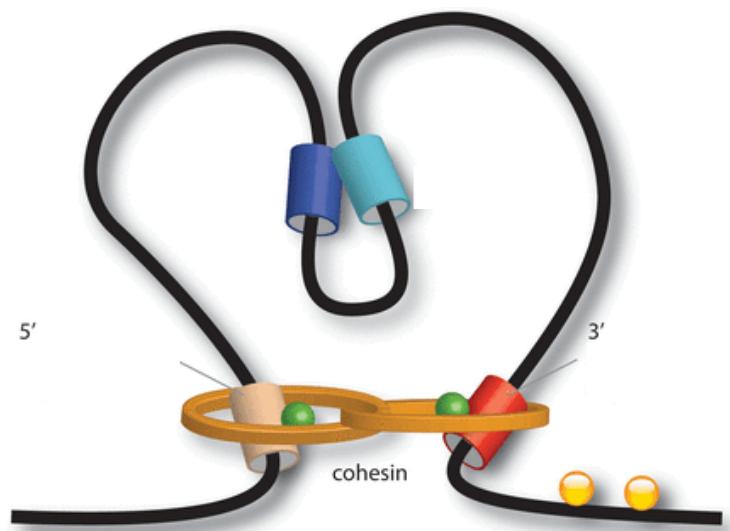
Un des méchanismes pour faire des boucles chromosomiques: “Loop extrusion”



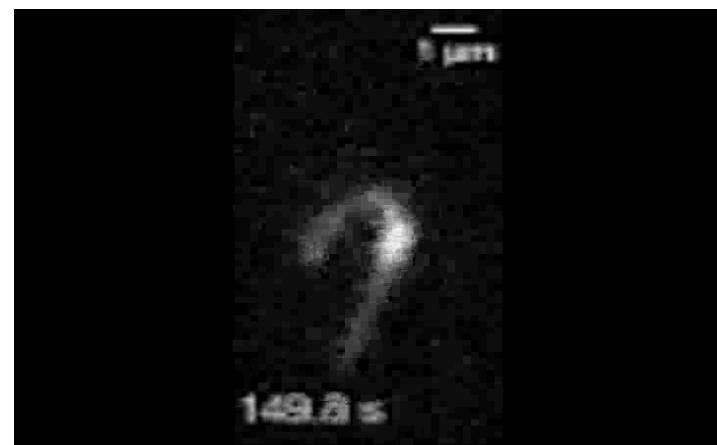
Les protéines appelées **Cohésines** sont de véritables moteurs moléculaires capables de générer des boucles



From <https://www.endotext.org/> and
Matharu and Ahituv, Plos Genet 2015

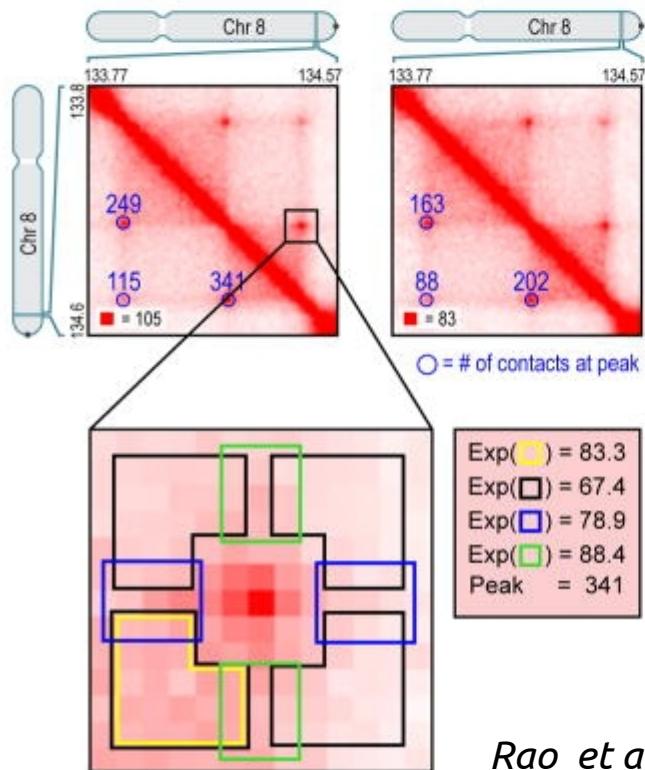


Feeney et al, Biochem J 2010

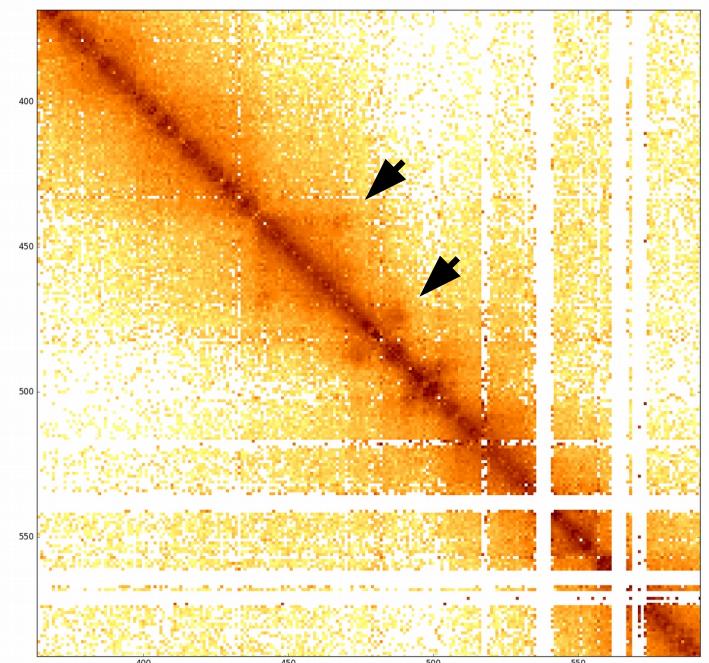


Ganji et al,
Science 2018

Cas des mammifères



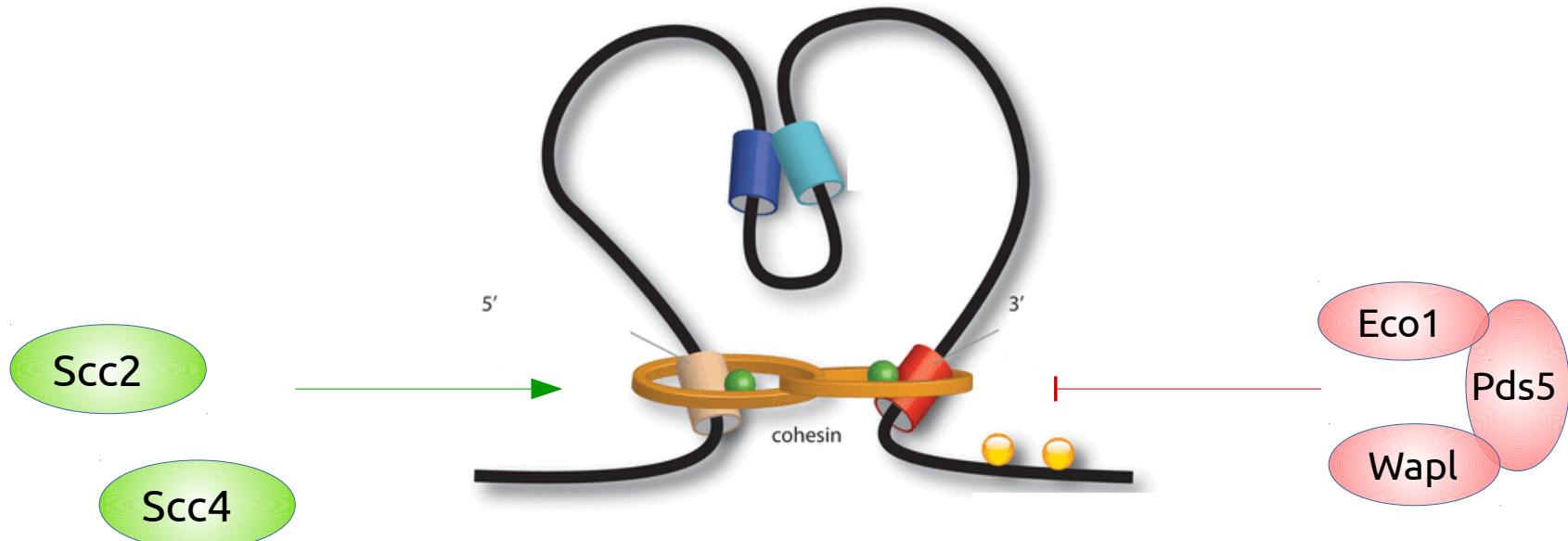
Problème plus complexe: Levure



- Outil: hiccup (software: Juicer)
- pixels enrichis / 4 voisins
+ FDR correction (+ clustering)
- Les outils classiques ne fonctionnent pas
- Solution(s) ?

Étude de la régulation des boucles chromosomiques chez la levure

Un de nos projets actuels dans l'équipe



- Certaines protéines vont agir sur la présence, la taille des boucles chromosomiques.

Le **défi** pour le hackathon ENGiE - Pasteur

→ Concevoir **et développer** un programme pour identifier les positions de boucles chromosomiques à partir d'une carte de contacts.

Le défi pour le hackathon ENGiE - Pasteur

- ▶ 1 Training set: 2000 cartes, ~10 boucles / carte

25102 boucles de chromosomes ainsi que 47235 positions de frontières de domaines.

→ Pour tester les algorithmes développés ou pour une approche de Machine Learning.

- ▶ 1 Mysterious set: 2000 cartes, ~10 boucles / carte

25152 boucles de chromosomes ainsi que 47276 positions de frontières de domaines.

→ Jeu de positions à identifier pour le challenge!

- ▶ 1 set de données expérimentales récemment générées au labo.

Pour aller plus loin et tester sur de vraies données!

Le lot pour l'équipe vainqueure



Carton de six bouteilles de
Brouilly AOC « Le Jardin des R »
Médaille d'Argent Paris 2012

Une parcelle des vignes appartient à
l'Institut Pasteur grâce au leg de
Madame Rampon.

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Session interactive : charger et visualiser les données

<https://github.com/axelcournac/Hackathon-ENGiE-PASTEUR>

[Code](#)[Issues 0](#)[Pull requests 0](#)[Projects 0](#)[Wiki](#)[Insights](#)[Settings](#)

This repository contains data, exemples of codes, references for the Hackathon Engie-Pasteur.

[Edit](#)[Manage topics](#)[33 commits](#)[1 branch](#)[0 releases](#)[2 contributors](#)Branch: [master](#)[New pull request](#)[Create new file](#)[Upload files](#)[Find file](#)[Clone or download](#)

 axelcournac	Add files via upload	Latest commit ee71c88 5 hours ago
 Training_Set	Update readme	7 hours ago
 articles	Add files via upload	7 days ago
 data	Add files via upload	5 hours ago
 exemples_codes	Rename vizmap.py to exemples_codes/vizmap.py	6 days ago
 pictures	Add files via upload	6 days ago
 README.md	Update README.md	6 hours ago
 hackathon_challenge.pdf	Add files via upload	13 days ago

[README.md](#)

Hackathon ENGiE-Pasteur

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Communiquer

Réfléchir

Echanger

Créer

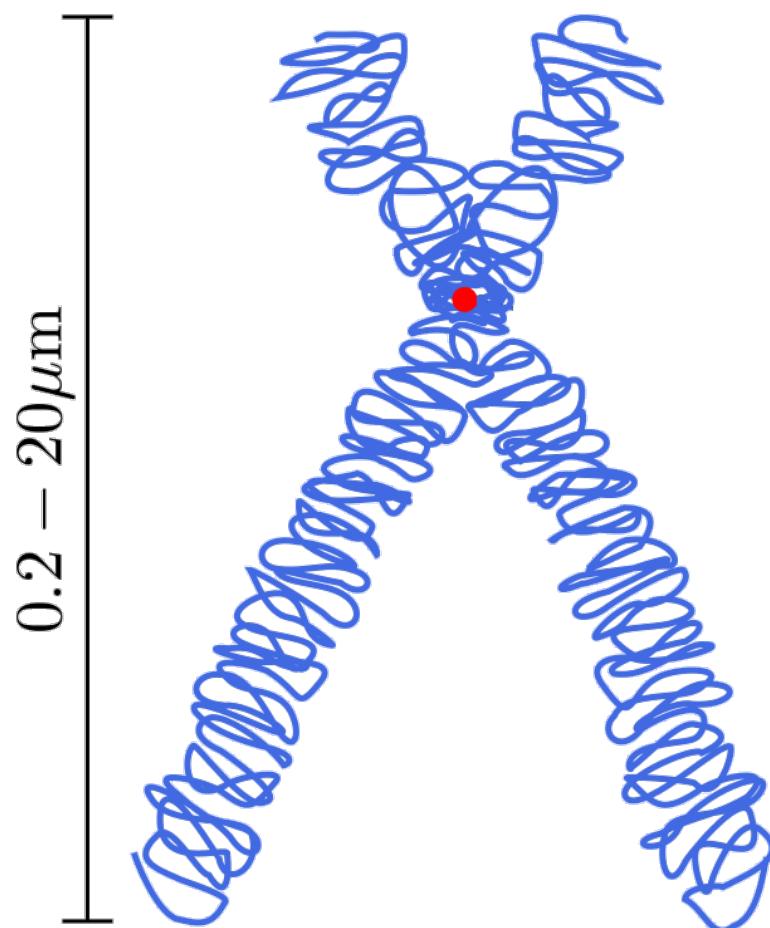
Innover!

Have fun

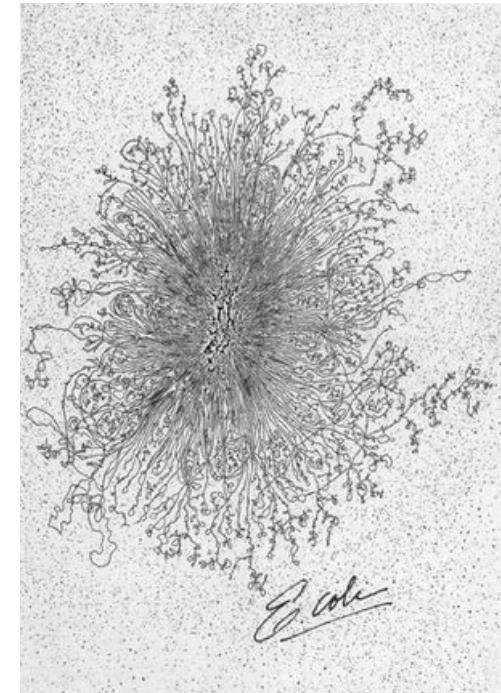
Bonne chance à tous !

Potential questions

3D structure of chromosomes



From *Molecular Biology* textbooks



From old microscopy pictures



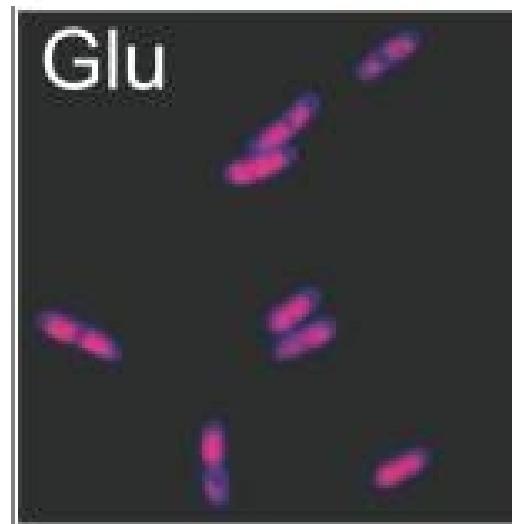
From Italian physicist talk

MukBEF (SMC family) as a mayor role in condensing the E.Coli chromosome

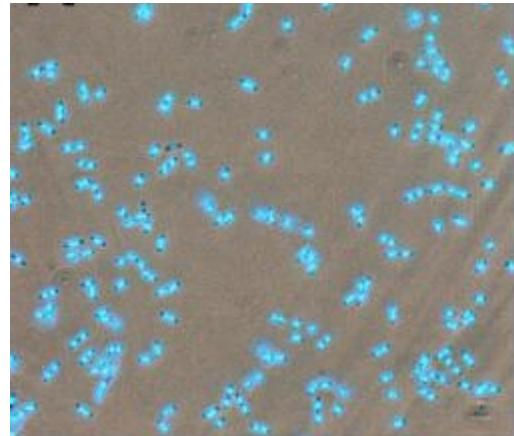


S. Cuylen et al.,
Trends Cell Biol., 2011

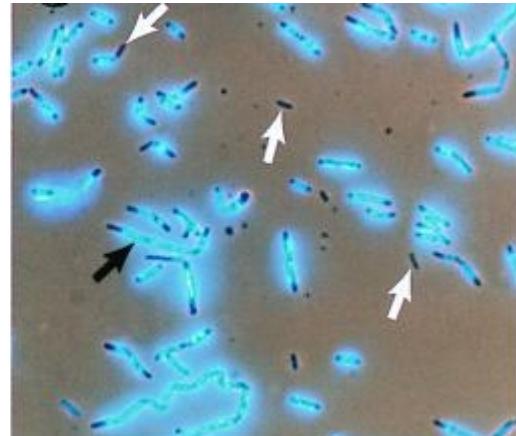
MukBEF overproduced



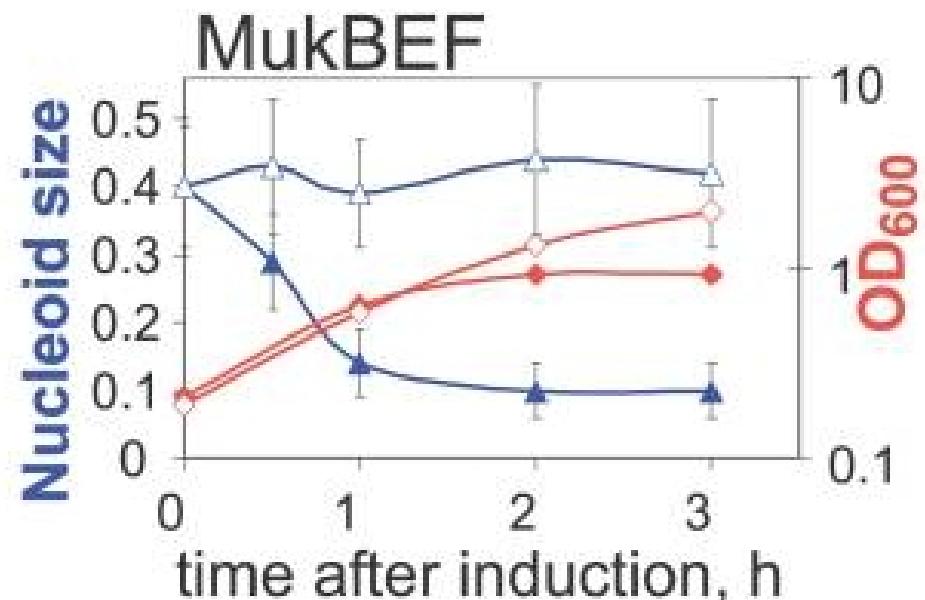
WT

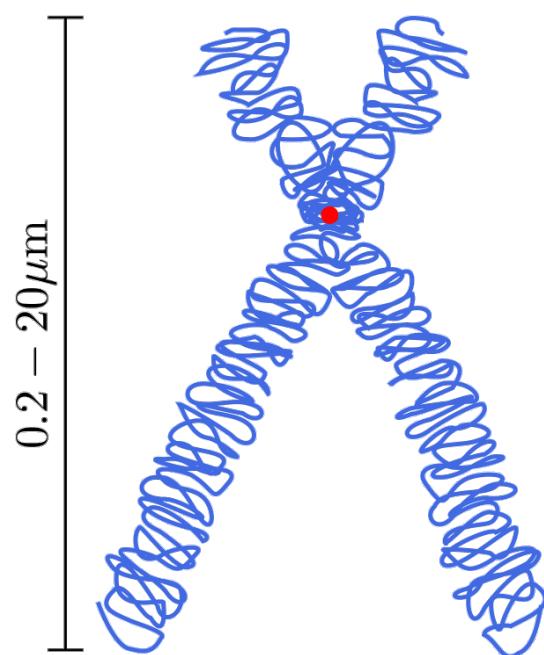


Delta MukBEF

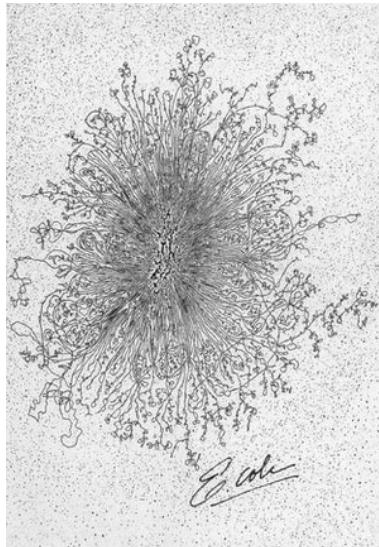


Anucleate cells





From *Molecular Biology*
textbooks



From old electronic
microscopy
pictures



From Italian physicist talk

Our current vision of chromosome



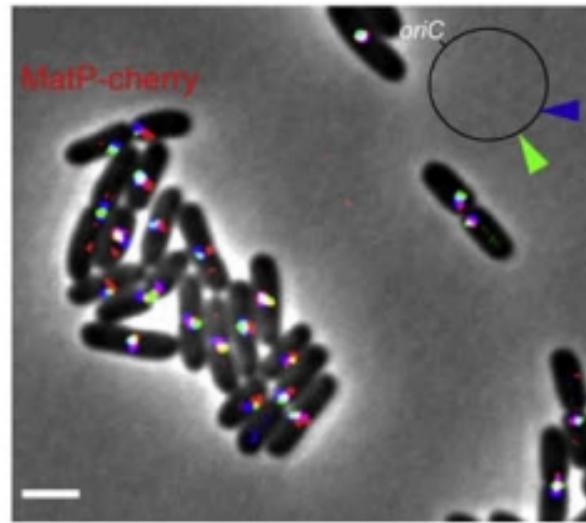
Intersection of highways.



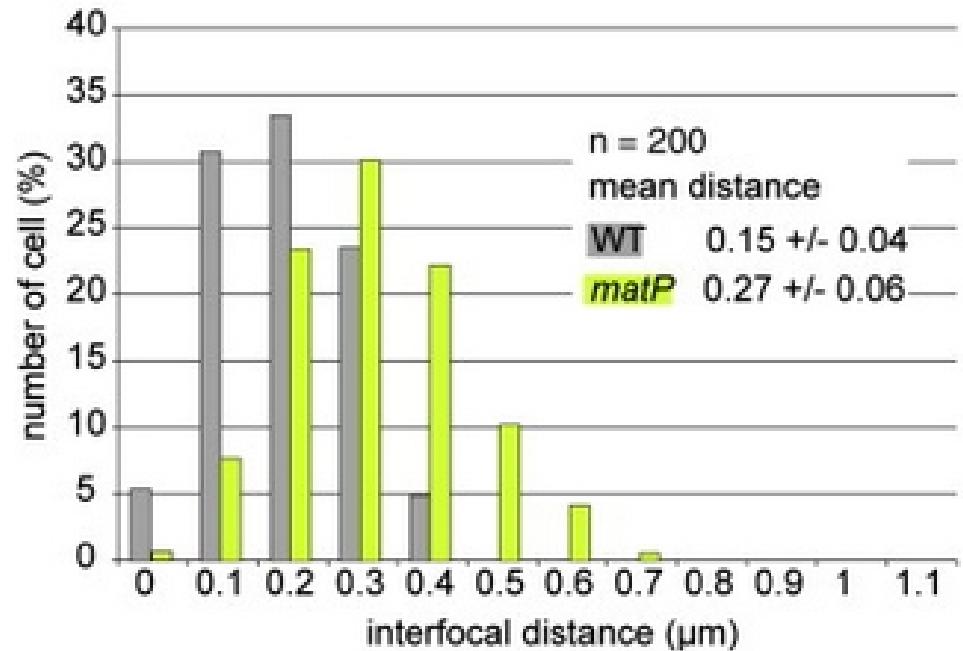
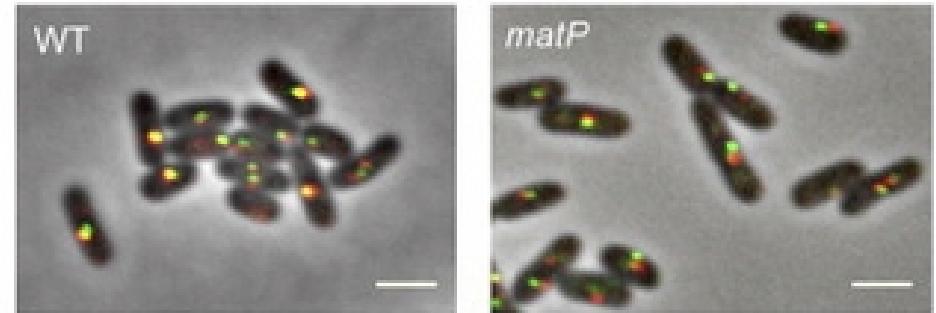
Snake nest.

A chromosome appears to be a very living molecule constantly condensed and decondensed by the action of molecular motors.

The protein MatP has a major role in the organization of the Ter macrodomain



MatP colocalise with TER macrodomain



Deletion of MatP increases
interfocales distances in TER

The use of **Molecular Dynamics** simulations to decipher the mechanisms behind contact maps

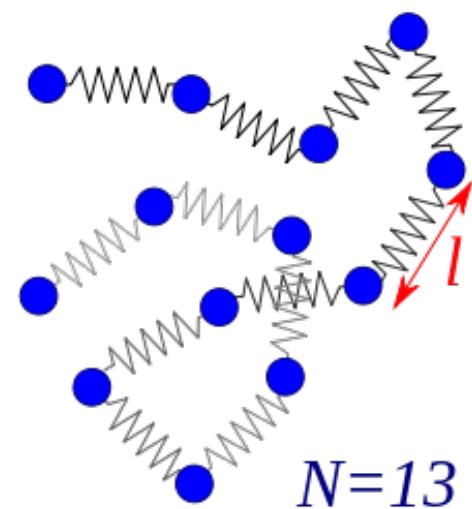
Langevin dynamics

The method : simulate the trajectory of each particle

(eg : monomer of a polymer = 500 bp)

$$M\ddot{X} = -\nabla U(X) - \gamma\dot{X} + \sqrt{2\gamma k_B T} R(t)$$

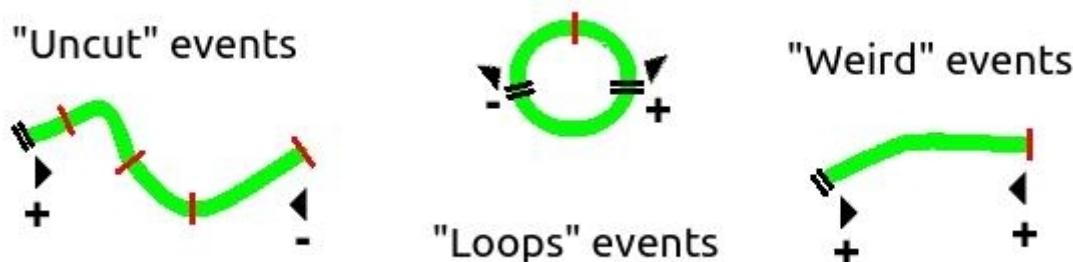
Viscous Force
Several potentials Stochastic Force



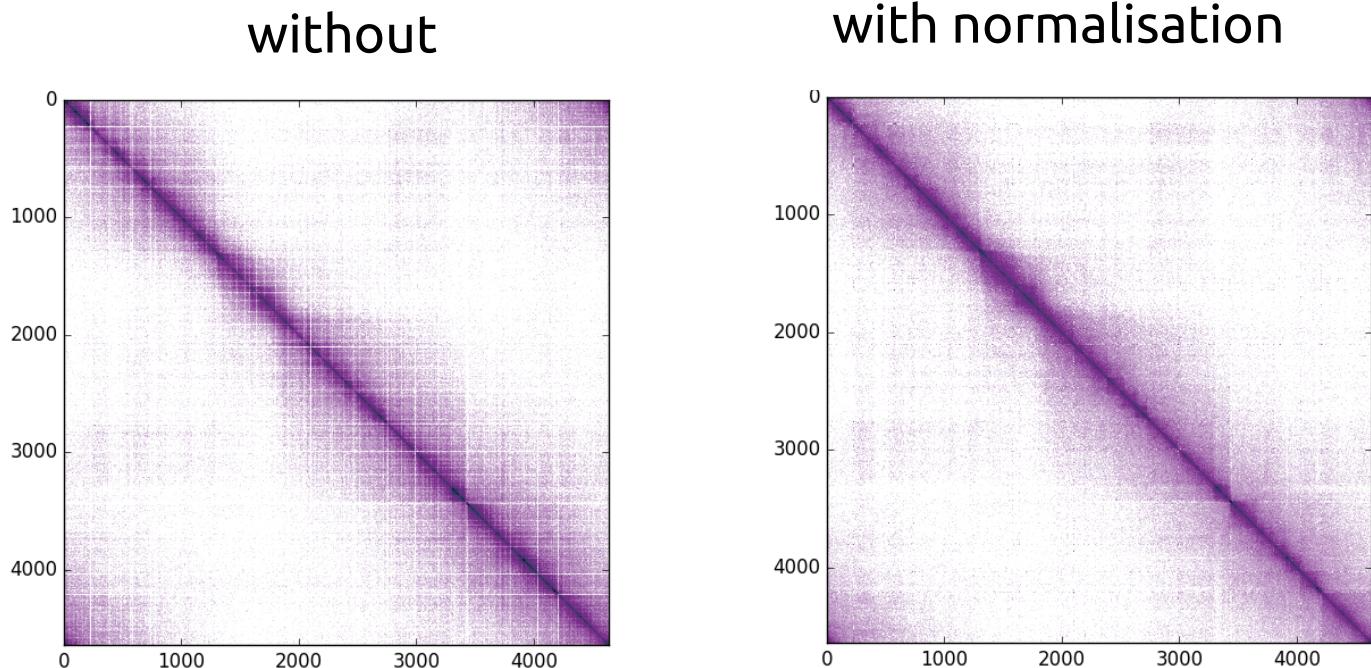
2 examples of Computational Methods to extract biological meaning from contacts data

Method 1: Sequential Component Normalisation (SCN)

① Careful classification of sequenced events



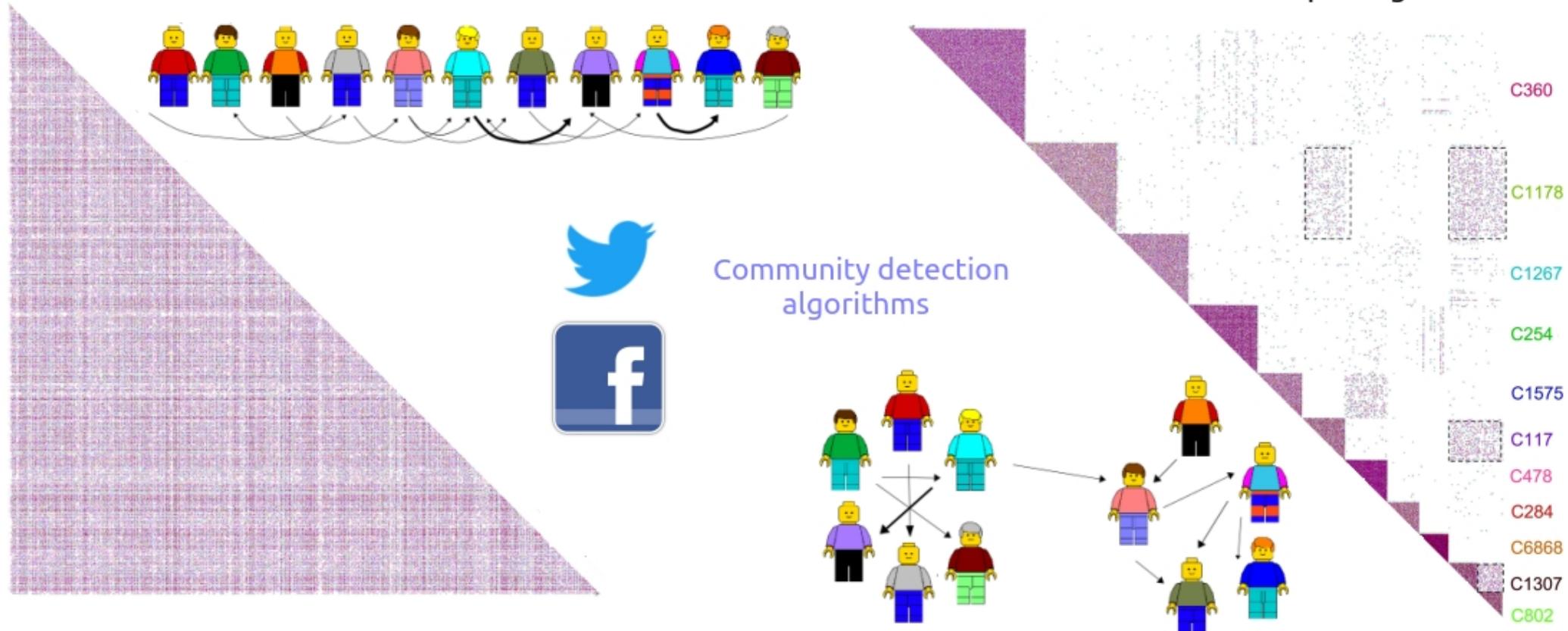
② Iterative averaging procedure



A. Cournac et al, BMC Genomics 2012

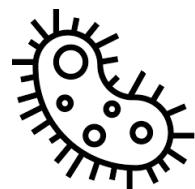
Codes and tutorial available: https://github.com/axelcournac/3C_tutorial

Method 2: Network based approach for *meta3C*

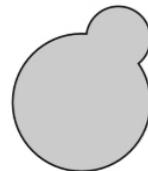


Personal scientific questions

- What is the impact of biological functions like transcription or replication on chromosome structure?
- Which proteins dictate the precise folding of chromosomes and at which scales?
- Do the repeated or similar sequences could also have a particular role in structuring chromosomes?



Bacteria

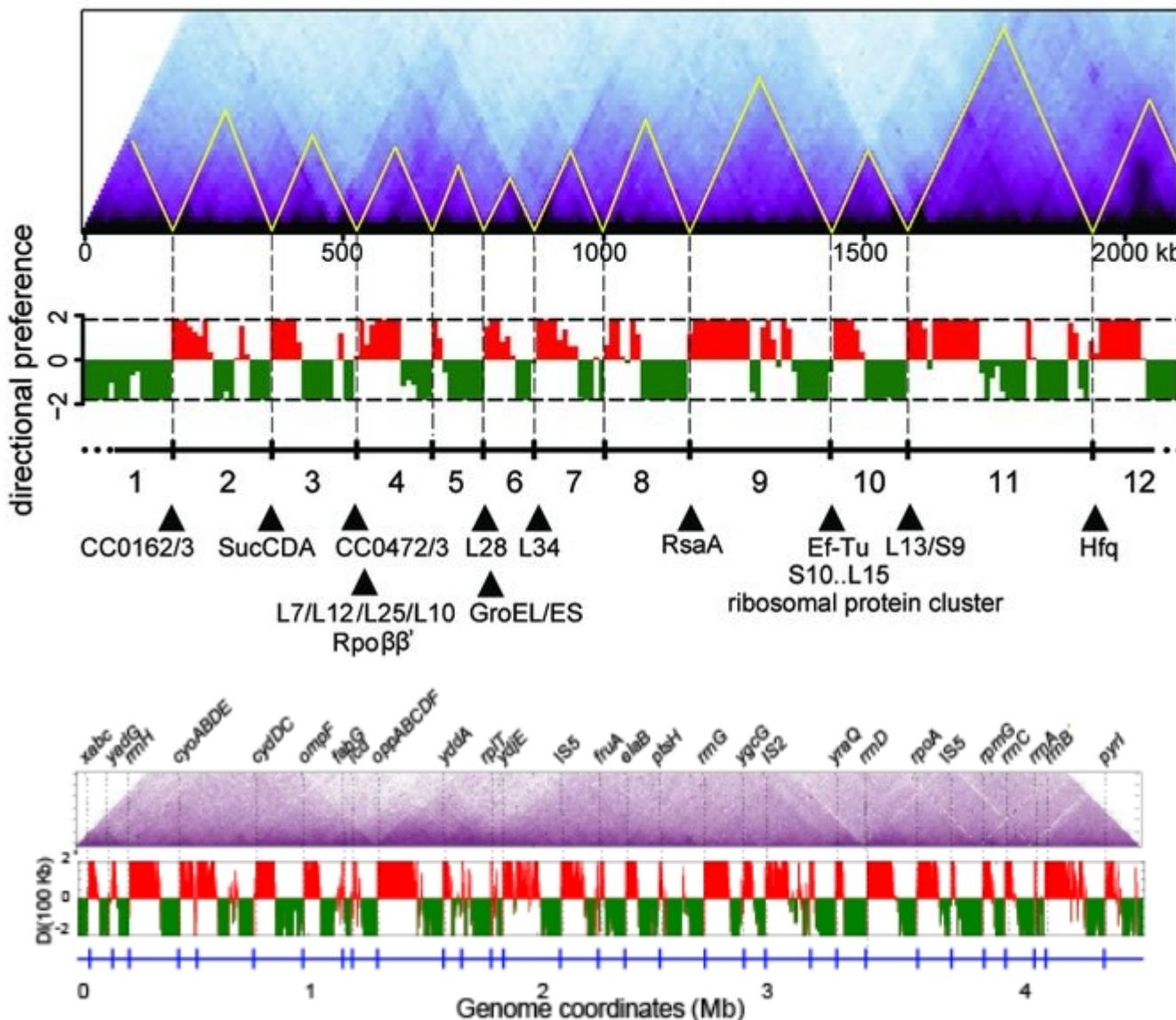


Yeast



Human

Transcription level has a role in the segmentation into Chromosomal Interaction Domains (CIDs), size ~ 100 kb



TB Le et al. Science 2013

We detected 31 chromosomal domains (size: from 40 kb to ~300 kb)
 Highly transcribed genes are enriched in borders regions.

The use of Molecular Dynamics simulations to decipher the mechanisms behind contact maps

Langevin dynamics

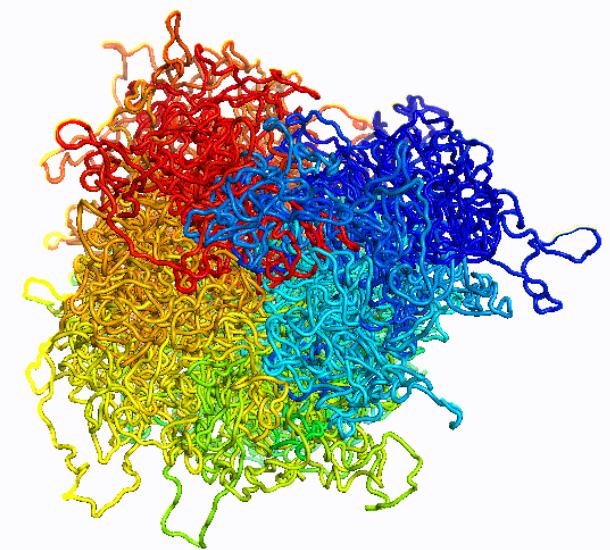
The method : simulate the trajectory of each particle

(eg : monomer of a polymer = 500 bp)

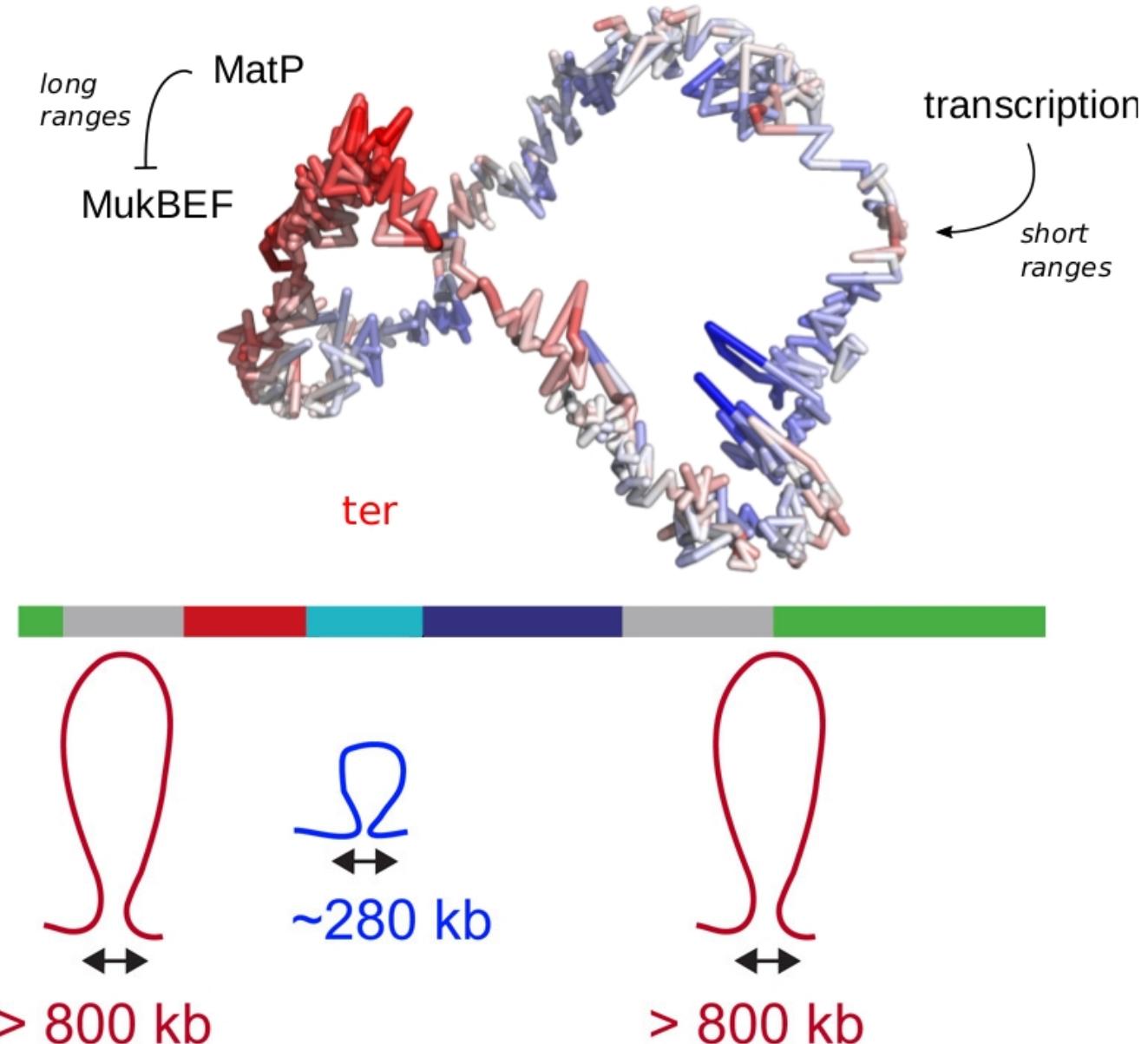
$$M\ddot{X} = -\nabla U(X) - \gamma\dot{X} + \sqrt{2\gamma k_B T} R(t)$$

Several potentials

Stochastic Force



Main message



MatP can restrict the range of action of the condensin MukBEF in the TER macro-domain.

V Lioy, A Cournac et al., *Cell* 2018