Tutorial 1

June 5, 2017

Knots and links in DNA

- are formed during replication, packing, and reconnection events.
- can be problematic for the cell.
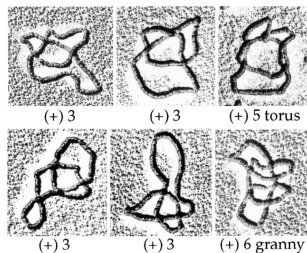- are resolved by enzymes (e.g. topoisomerases and recombinases)

Figure: Knots observed with electron microscopy [KRC+99].

Topoisomerases

Topoisomerases act by strand passage. This process is modeled by a *crossing change*.
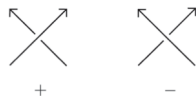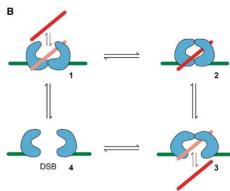

Figure: Two gate model of strand passage mechanism by type II topoisomerase [BBM11].

Figure: Crossing changes.

Site-specific Recombinases

Site-specific recombinases are enzymes that act by local reconnection at two specific sites on a DNA substrate.

- Examples: *Xer, Cre*, λ, *FLP; Gin, Hin*
- Recombination causes integration, excision or inversion of genetic material.
- Site-specific recombinases target short sites (e.g. Xer targets *psi* sites) on DNA specific to each enzyme.
- May be sensitive to the direction of the nucleotide sequence (*directly repeated* or *inversely repeated*).
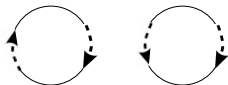

Figure: A direct repeat and an inverse repeat. The arrow indicates the recombination sites.

Recombinases

Most recombinases are classified by the action at the biochemical level

- **Serine recombinases:** Recombination is via a double-stranded break and rotation mechanism.
- **Tyrosine recombinases:** Recombination is via Holliday junction.
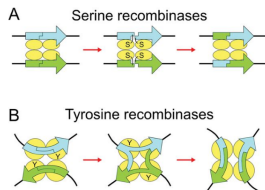

Figure: Figure from [Col13]

Either way: mathematically, site-specific recombination is a local reconnection event and is modeled via *band surgery*.

Band surgery models recombination by recombinases

A band b is an embedding $b(I \times I) \to S^3$. Band surgery produces new link M from L:

$$M = L - b(I \times \partial I) \cup b(\partial I \times I)$$

L $L \cup B = M \cup B$ M

- Recombination at directly repeated sites: coherent (oriented) band surgery.
- Recombination at inversely repeated sites: non-coherent (unoriented) band surgery.

# Band surgery models recombination by recombinases

Recombination at directly repeated sites is modeled by coherent band surgery.

- Orientation on product is well-defined
- If product is single component then substrate is two-component link.

Recombination at inversely repeated sites is modeled by non-coherent band surgery.

- Orientation on product not well-defined
- Substrate and product both single component knots
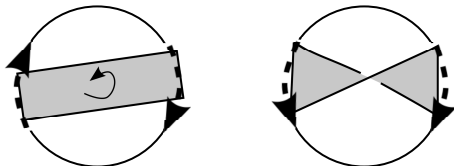
Band surgery realized as a skein move

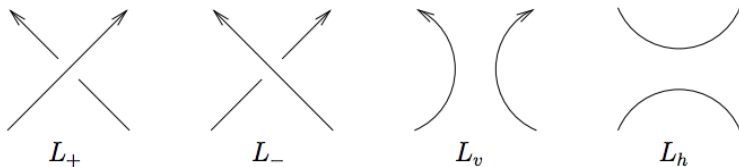L_+ L_- L_v L_h

Figure: Terms from oriented and unoriented skein triples
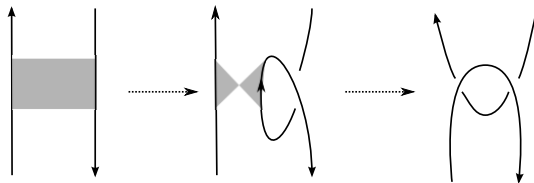
Figure: Coherent band surgery realized as an oriented skein resolution

Band surgery exercises
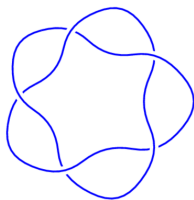
Figure: Find a coherent band surgery from 5_1 to $3_1\#$ Hopf.

Figure: Find non-coherent band surgeries from 8_{17} to 6_3 and 4_1.

Band surgery exercises

Figure: Coherent band surgery: $5_1 \mapsto 3_1 \# 2_1^2$ [AK14]

Figure: Non-coherent band surgeries: $8_{17} \mapsto 6_3, 4_1$ [Kan16]

Observe: coherent band surgery on a knot is oriented, product is link with two components. Non-coherent band surgery on knots is unoriented, product is knot.

Different notions of "Gordian" distance

Assume L, M unoriented links and band-surgery is an unoriented operation.
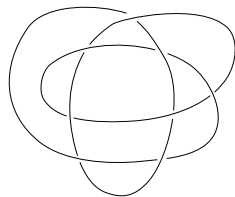
- Gordian distance $d(J, K)$ is the minimal number of crossing changes to deform knot J into knot K.
- Band Gordian distance $d_b(L, M)$ is the minimal number of band surgeries (either type) to deform link L into link M.
- Non-coherent band Gordian distance $d_2(J, K)$, is the minimal number of non-coherent band surgeries to deform knot J into knot K.
- Unknotting numbers: $u(K) = d(K, U)$, $u_b(K) = d_b(K, U)$ and $u_2(K) = d_2(K, U)$ where U is the unknot 0_1.

Note: $d_b(J, K) \leq d_2(J, K)$.

Band-Gordian distance examples

- $d(3_1, U) = u(3_1) = 1$
- $d(4_1, U) = u(4_1) = 1$.
- $d_2(3_1, U) = u_2(3_1) = 1$.
- $d_2(4_1, 3_1) = 1$ and $d_2(4_1, U) > 1$ (Lickorish, linking form), therefore $u_2(4_1, U) = 2$.
- $d_b(\text{Hopf link}) = 1$
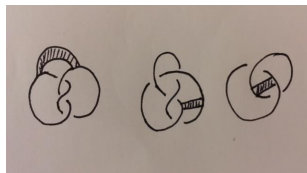
Nomenclature of knots

Warning: there are many different naming conventions for knots and links. Not all types of notation take into account mirrors or orientations.

- in the Rolfsen link table as $L4a1$
- as the torus link $T(p, q) = T(2, 4)$
- as the two-bridge link $b(\alpha, \beta) = b(4, 3)$
- in 4-plat notation for two-bridge knots as $C(1, 3)$
- Also referred to as 4_1^2, the 4-catenane or the '4-cat'

Other types of notation include braid representatives, Morse link presentations, and encodings of diagrams such as Gauss codes or Planar diagram codes.

Selected result 1: Tangle analysis and Xer recombination [VCS05]

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Tangle Analysis of Xer Recombination Reveals only Three Solutions, all Consistent with a Single Three-dimensional Topological Pathway

Mariel Vazquez[1]*, Sean D. Colloms[2] and De Witt Sumners[3]

[1]*Department of Mathematics University of California at Berkeley, Berkeley CA 94720-3840, USA*

[2]*Division of Molecular Genetics Institute of Biomedical and Life Sciences, University of Glasgow Glasgow G11 6NU, Scotland UK*

[3]*Department of Mathematics Florida State University Tallahassee FL 32306, USA*

The product of Xer recombination at directly repeated *psi* sites on a circular unknotted DNA molecule is a right-hand four-noded catenane. Here, we use tangle equations to analyze the topological changes associated with Xer recombination at *psi*. This mathematical method allows computation of all possible topological pathways consistent with the experimental data. We give a rigorous mathematical proof that, under reasonable biological assumptions, there are only three solutions to the tangle equations. One of the solutions corresponds to a synaptic complex with antiparallel alignment of recombination core sites, the other two correspond to parallel alignment of cores. We show that all three solutions can be unified into a single three-dimensional model for Xer recombination. Thus the three distinct mathematical solutions do not necessarily represent distinct three-dimensional pathways, and in this case the three distinct tangle solutions are different planar projections of the same three-dimensional configuration.

*Corresponding author

Selected result 2: XerCD-dif recombination unlinks stepwise [SIG+13]

FtsK-dependent XerCD-dif recombination unlinks replication catenanes in a stepwise manner.

Shimokawa K[1], Ishihara K, Grainge I, Sherratt DJ, Vazquez M.

⊕ **Author information**

Abstract

In Escherichia coli, complete unlinking of newly replicated sister chromosomes is required to ensure their proper segregation at cell division. Whereas replication links are removed primarily by topoisomerase IV, XerC/XerD-dif site-specific recombination can mediate sister chromosome unlinking in Topoisomerase IV-deficient cells. This reaction is activated at the division septum by the DNA translocase FtsK, which coordinates the last stages of chromosome segregation with cell division. It has been proposed that, after being activated by FtsK, XerC/XerD-dif recombination removes DNA links in a stepwise manner. Here, we provide a mathematically rigorous characterization of this topological mechanism of DNA unlinking. We show that stepwise unlinking is the only possible pathway that strictly reduces the complexity of the substrates at each step. Finally, we propose a topological mechanism for this unlinking reaction.

KEYWORDS: DNA topology; Xer recombination; band surgery; tangle method; topology simplification

Comment in
Mathematical validation of a biological model for unlinking replication catenanes by recombination. [Proc Natl Acad Sci U S A. 2013]

Project 1

Consider the system

$$
\begin{aligned}
N(O + P) &= 0_1 \\
N(O + R) &= L
\end{aligned}
$$

where L is a two-component link with unknotted components. For which links can we prove that O is rational?

Consider the system

$$
\begin{aligned}
N(O + P) &= 2_1^2 \text{ Hopf link} \\
N(O + R) &= T_n
\end{aligned}
$$

where T_n is a twist knot. For which knots can we prove that O is rational?

Does the proof technique above work? What possible Dehn surgery obstructions would we need?

Project 2

Consider coherent band surgeries taking single component knots to either the two-component unlink (0_1^2) or the Hopf link (2_1^2).

How do the neighborhoods of the Hopf link and two-component unlink differ in the coherent band Gordian graphs?

Can you characterize the neighborhoods of these links in the coherent band-Gordian graph (restricted to one and two-component links)?

Note: numerical work indicates the following transitions:

$$
\begin{aligned}
0_1^2 &\leftarrow 0_1, 6_1, 6_1^*, 8_{20}, 8_{20}^* \\
2_1^2 &\leftarrow 0_1, 3_1, 4_1, 5_1, 6_1^*, 6_2, 6_3, 7_2, 7_6, 7_7^*, 8_6, \\
&\quad\, 8_7, 8_{10}, 8_{11}, 8_{13}^*, 8_{14}, 8_{20}, 8_{21}, 9_{22}^*, 9_{44}, 9_{45}
\end{aligned}
$$

Project 3

Consider non-coherent band surgery from the unknot to any $T(2, 2n + 1)$ torus knot.

- What is different in the non-coherent band surgery case?
- Is it possible to prove in the tangle model that the tangle O is rational?

Consider all possible non-coherent band surgery pathways from $T(2, 2n + 1)$ to the unknot. Find the pathways which exist and obstruct the ones that don't.

Note: partial data from numerical experiments with non-coherent band surgery may be available.

Project 4

Computational project: conduct network analysis on the coherent
and non-coherent band Gordian graphs (adjacency matrix data to
be made available).

Can you make any meaningful observations?

Project 5

Determine all non-coherent bandings from $T(2, 5)$ to 6-crossing knots.

Determine all non-coherent bandings from $T(2, 7)$ to 8-crossing knots.

Selected result 1: Tangle analysis and Xer recombination

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Gel electrophoresis experiments carried out in [CBS97] show that Xer recombination at directly repeated psi sites on a circular unknotted DNA molecule yield a right-handed four-noded catenane.
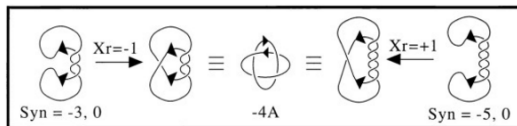

Figure: Model of recombination hypothesized in [CBS97].

Translation: the Xer enzyme appears to tie an unknot into a (2,4)-torus link, and based on the biology, it is expected to happen like in Figure 12.

Goal: Characterize topological pathways consistent with experimental date.

Tangle method

Recombination events are modeled by system of tangle equations:

$$N(O + P) = K_1$$

$$N(O + R) = K_2$$

Here:

- O, P and R are tangles (unknown)
- P contains the part changed during recombination
- R is the new part
- K_1 and K_2 are substrate and product of recombination (known)

When O, P, and R are assumed to be (sums of) rational tangles, system can be solved.

In the context of Xer acting on unknotted DNA:
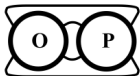
The set-up:

$$N(O + P) = \text{unknot} = b(1, 1)$$
$$N(O + R) = T(2, 4) = b(4, 3)$$

Theorem ([VCS05])

In the system

$$N(O + P) = unknot = b(1, 1)$$
$$N(O + R) = T(2, 4) = b(4, 3)$$

P and R are either prime or rational, and O is rational.

Theorem ([VCS05])

In the system

$$N(O + P) = unknot = b(1, 1)$$
$$N(O + R) = T(2, 4) = b(4, 3)$$

P and R are either prime or rational, and O is rational.

Claim

O, P and R are locally unknotted.

If O or P locally knotted, $N(O + P)$ not an unknot.
If R locally knotted, $N(O + R)$ not a link of unknots. $\square$

Claim: O is rational.

For this we need two facts and two lemmas.

Fact

1. A tangle is rational if and only if its branched double cover is a solid torus in S^3.

2. The branched double cover of the two-bridge knot $b(\alpha, \beta)$ is the lens space $L(\alpha, \beta)$.

Claim: O is rational.

Lemma (Lickorish)

If A and B are locally unknotted tangles and $N(A + B)$ is a 4-plat, at least one of A or B is rational.

Lemma (Hirasawa-Shimokawa)

No Dehn surgery on a non-trivial strongly invertible knot can produce a lens space $L(2k, 1)$.

A *strongly invertible* knot is one which is preserved set-wise by an involution on S^3. Here, the involution is the covering map.

Proof sketch

Suppose O is prime. By the first lemma, P is rational. Thus $\tilde{P}$ is a solid torus and $\tilde{O} = S^3 - \tilde{P}$ is the complement of some knot $K \subset S^3$.

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Now we have that $L(4, 3)$ is obtained by Dehn filling (i.e. gluing in a solid torus) along K, which is strongly invertible with respect to the covering transformation on S^3. By the second lemma K must be trivial (note that $L(4, 3) = L(4, 1)$). Thus $\tilde{O}$ is a solid torus, which implies O is rational. □

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Now tangle calculus (to be explained later this week) can be used to determine solutions to the system

$$N(O + P) = b(1, 1), N(O + R) = b(4, 3)$$

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