Complex-Network Modelling and Inference

Lecture 6: Application: Genome Reconstruction

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Section 1

Genome Reconstruction

Genomes

Definition

A *genome* is the genetic material in an organism.

- Consists of DNA for us (RNA for viruses)
- Made up of sequences of nucleotides
  - Humans have about 3 billion nucleotides in our genome
- (roughly) DNA has made up of sequences of 4 molecules (the "bases")
  - **A**denine
  - **G**uanine
  - **C**ytosine
  - **T**hymine

  which appear in a double-helix, but in fixed pairs, so we can write a DNA sequence as a sequence of these letters, *e.g.*,

$$AAGCTTAAGTC$$

Genome sequencing

- We can't just read the sequence
- There are various approaches to reading, but generally
  - they can only see a small part at a time: a "read"
  - we don't know where in the sequence a read comes from
  - if we get lots, then they will have lots of overlaps
- Practicalities
  - real reads contain errors
  - we can't guarantee that reads cover all the genome
  - you don't know which side of the DNA a read comes from

But we will ignore these problems for the moment

Terminolgy

- A sequence of k symbols is called a *k-mer*. Also called an n-gram
  - *e.g.,* in English, 3-mers are sequences like "abc", "rtb", ...
  - *e.g.,* in binary, 3-mers are sequences like "010", "111", ...
  - *e.g.,* in DNA, 3-mers are sequences like "ATG", "TTT", ...

  Usually we are interested in sub-sequences from a longer sequence.
- A prefix is the start of a sequence, and a suffix the end
  - when we are dealing with k-mers, we will mean a $k-1$-prefix or suffix
- Given n symbols, how many possible k-mers are there?

Example 1

Assume our reads provide all 3-mers from a DNA sequence

ATG

AAT

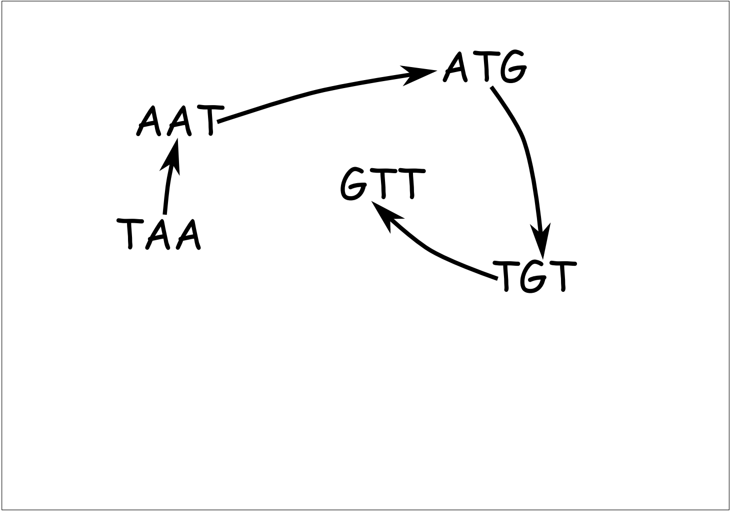
GTT

TAA

TGT

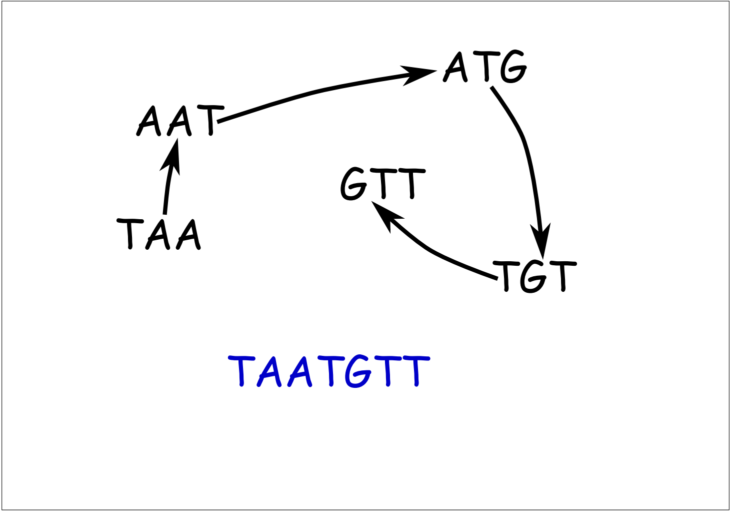
Example 1

Create a graph by linking suffix $\rightarrow$ prefix



Example 1

The sequence is just the path through this graph



$$\begin{tikzcd}
TAA \arrow[r] & AAT \arrow[r] & ATG \arrow[r] & TGT \arrow[r] & GTT
\end{tikzcd}$$

Are we done?

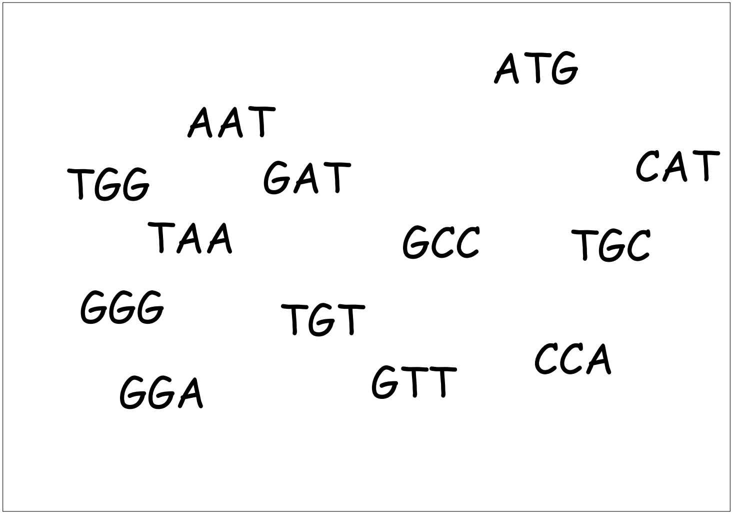
No!

- *k*-mers are repeated in a long sequence
  - the above assumed that each appeared exactly once in the sequence
  - there can be more than one prefix/suffix match
- Above also assumes
  - we have all *k*-mers
  - no errors

  but for the moment, lets ignore these issues, as the first one is big enough

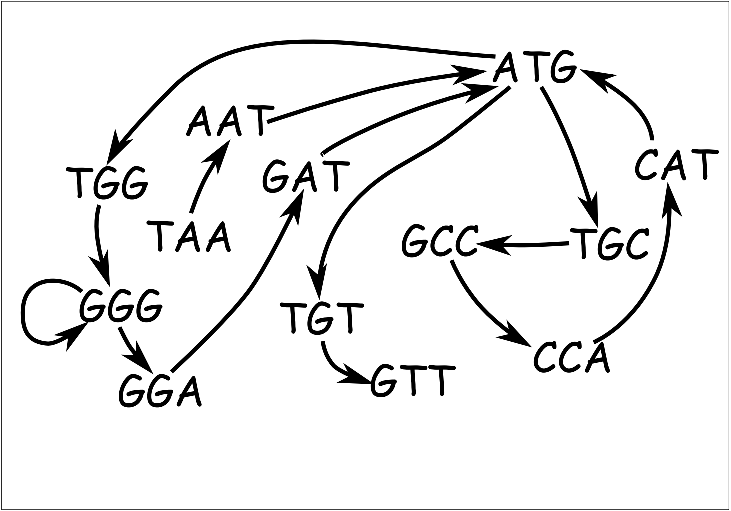
Example 2

A (slightly) bigger example



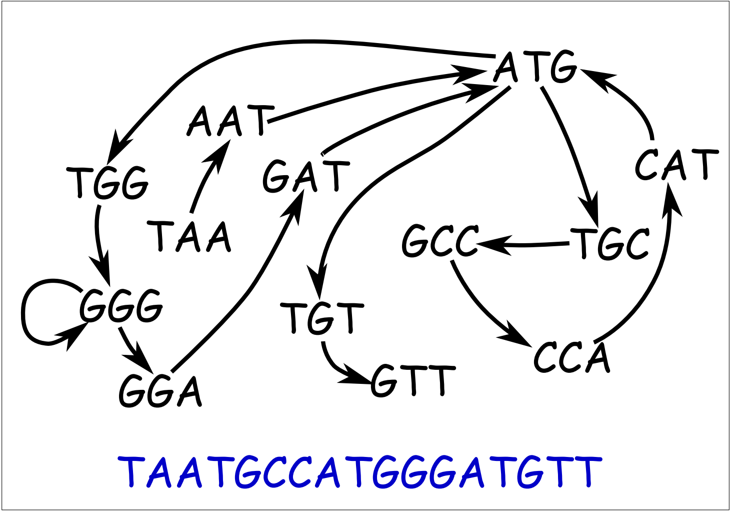
Example 2

Harder to spot the "walk" (it isn't cycle-free, so not a "path")



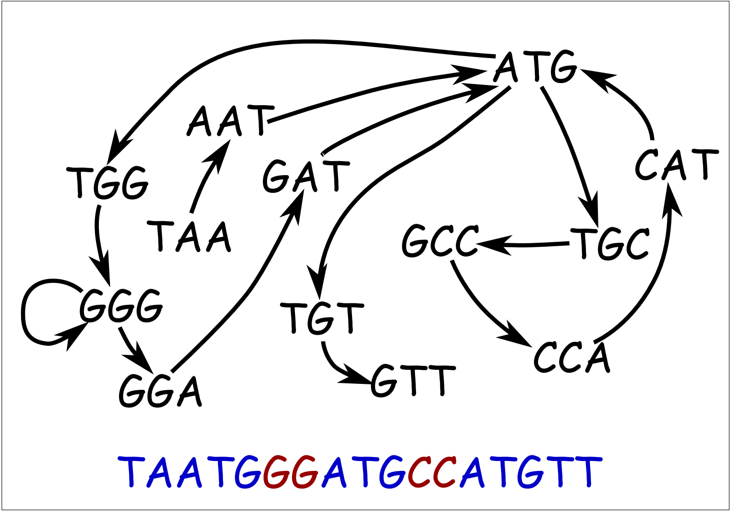
Example 2

Here is one possibility



Example 2

But there is ambiguity



A how to

- If we knew multiplicity of ATG we would create 3 nodes.
  - this simplifies some problems
    - *e.g.*, we know we don't loop through GGG
  - it makes the graph quite a bit more complicated
    - 2 extra nodes
    - lots of extra links
  - it doesn't resolve all ambiguities
    - only way to avoid is to have longer sequences
- Given this new graph the problem becomes one of finding a *Hamiltonian path*

Hamiltonian paths and cycles

Definition

A *Hamiltonian path* is a path that visits each node exactly once. A *Hamiltonian cycle* visits each node exactly once, and then returns to the start.

The problem of finding a Hamiltonian path is *NP-complete*

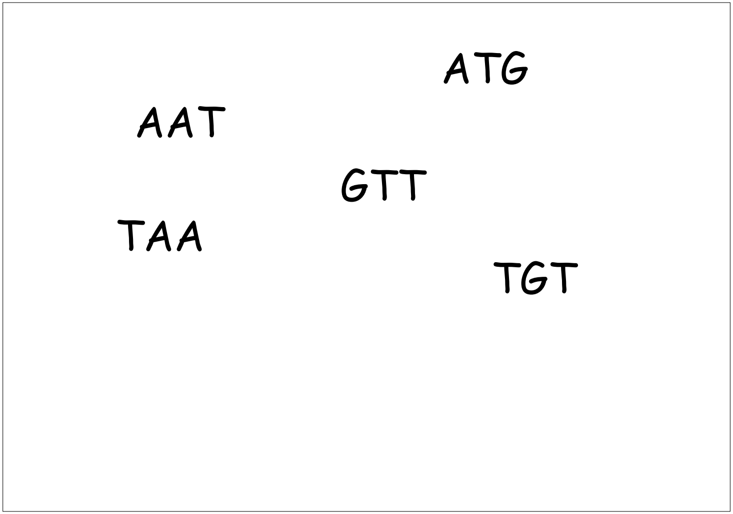
- this is a hard class of problems (computationally)
- there are no known polynomial-time algorithms
  - but it is easy to check a given cycle is correct
- this is the problem early genome sequencers attempted, but you can't (practically) solve big NP-complete problems
- so they came up with an alternative graph: the de Brujin graph

de Brujin graph

- Often one set of data can be represented by multiple different graphs
- This is a classic example
  - the obvious graph is not the right one to work with
- *de Bruijin graph*
  - the nodes are prefixes and suffixes
  - the edges are the k-mers
    - ★ they link their prefix to their suffix

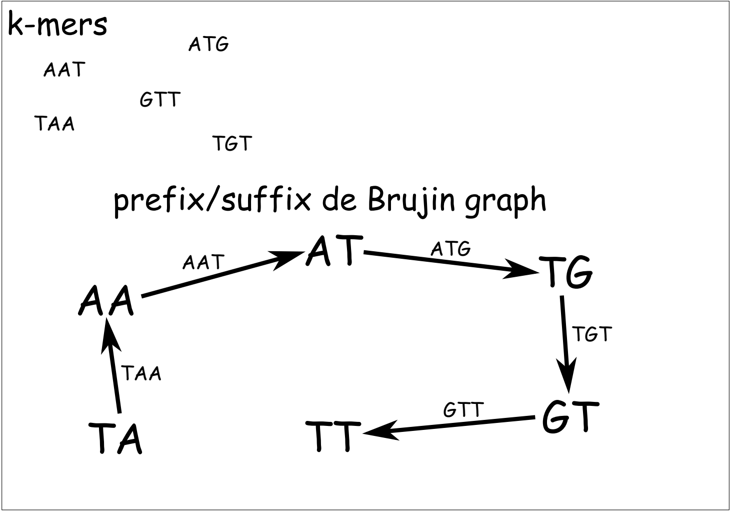
Example 1

Construct a de Brujin graph



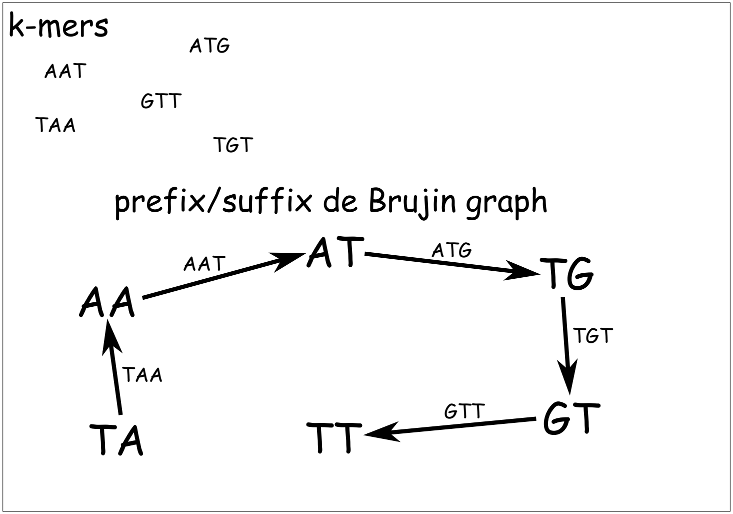
Example 1

Construct a de Brujin graph



Example 1

Now we just look for an Eulerian path in this graph



$$\begin{tikzcd}
 & AT \arrow[r, "ATG"] & TG \arrow[d, "TGT"] \\
AA \arrow[ur, "AAT"] & & GT \arrow[dl, "GTT"] \\
TA \arrow[u, "TAA"] & TT &
\end{tikzcd}$$

Example 2

de Brujin graph of second example

$$\begin{tikzcd}
 & & & CC \arrow[dl, "CCA"'] & & & \\
 & & CA \arrow[d, "CAT"] & & GC \arrow[ul, "GCC"'] & & \\
TA \arrow[r, "TAA"] & AA \arrow[r, "AAT"'] & AT \arrow[rr, bend left, "ATG"] \arrow[rr, "ATG"] \arrow[rr, bend right, "ATG"] & & TG \arrow[u, "TGC"'] \arrow[r, "TGT"'] \arrow[d, "TGG"] & GT \arrow[r, "GTT"'] & TT \\
 & & GA \arrow[u, "GAT"] & & GG \arrow[ll, "GGA"'] \arrow[loop right, "GGG"] & &
\end{tikzcd}$$

Eulerian paths

- The sequence on the de Brujin graph can be found by taking finding an Eulerian path
  - a path that goes along each edge exactly once
  - need to include multiplicity of edges in construction
- Eulerian paths are easy to construct
- We need a minor adaptation here for directed graphs

Definition

A digraph is *balanced* if the in-degree of each node is the same as its out degree.

Theorem

An Eulerian cycle exists on a digraph if and only if it is balanced and strongly connected.

I leave the proof as an exercise, as well as the extension to an Eulerian path.

Further reading I

Phillip Compeau and Pavel Pevzner, *Bioinformatics algorithms: An active learning approach*, Active Learning Publishers, 2014.