

Fully Dynamic de Bruijn Graphs

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April 10, 2017

- 1 Introduction
- 2 Data Structure
- 3 Implementation

Genome assembly: De-Bruijn graph

De-Bruijn graph: Reconstructing a string from a set of its k -mers

- ① Data structure method on genome assembly.
- ② Consist of multiple K -mers which generated by genome sequence.
- ③ Same vertices, $K-1$ mers are glue together in the final step.

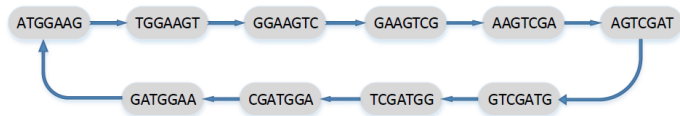


Fig. 1. The de Bruijn graph constructed from string ATGGAAGTCGATGGAAG, with $k = 7$.

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- Compared to 'overlap-consensus-layout' method, De Bruijn graph-based assembly approach handles the assembly of repetitive regions better

Application problem

- Using de Bruijn graph in practice is the high memory occupation for certain organisms
- Human genome encoded in a de Bruijn graph with a k-mer size of 27 requires 15GB to store the node sequences
- Bulges and whirls occur because of sequencing errors or repeats in the genome, so would like to be able to efficiently add and remove edges from graph

How to efficiently update graph while maintaining memory space efficiency?

Fully Dynamic de Bruijn Graphs(Belazzougui et al. (2016))

Introduces compact, dynamic representation of De Bruijn graph

- Nodes and edges can be inserted and deleted efficiently
- k-mers are represented by integers using a combination of Karp-Rabin hashing and minimal perfect hashing.
- A partition of the graph into a forest allows efficient membership queries with no error.

Project

- 1 Implement the data structure from the paper.
- 2 Evaluate our data structure on graphs built from real sequencing data.
- 3 Compare our data structure with alternative approaches for De Bruijn graphs

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- Static hash function

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- De Bruijn graph and forest

Hash function f

- 1 The hashing function we use is the combination of **Karp-Rabin** and **minimal perfect hashing**.

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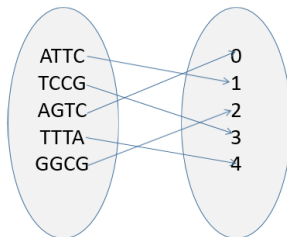
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- 2 **Karp-Rabin**: Given a prime P and base r , a Rabin-Karp hash function f is a function defined over the space of all strings of length k such that $f(x_1 \dots x_k) = (\sum_{i=1}^n x_i r^i) \bmod P$.

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Hash function properties

Lemma

Given a static set N of n k -tuples over an alphabet Σ of size σ , with high probability in $O(kn)$ expected time we can build a function $f: \Sigma^k \rightarrow \{0, \dots, n-1\}$ with the following properties:

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- ③ given a k -tuple v , we can compute $f(v)$ in $O(k)$ time.
- ④ given u and v , such that suffix of u of length $k-1$ is the prefix of v of length, or vice versa, we can compute $f(v)$ in $O(1)$ time if we already computed $f(u)$.

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- Suppose we only have two k-mers, one qualified minimal perfect hashing function f needs to ensure that $f(\text{"ATTC"}) = 0$ and $f(\text{"TTCG"}) = 1$
or
 $f(\text{"ATTC"}) = 1$ and $f(\text{"TTCG"}) = 0$.

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- 4 insertions and deletions take $O(k)$ amortized expected time.
- 5 the data structure may work incorrectly with very low probability (inverse polynomial in n).

Representation of edges in de Bruijn graph

- 1 The edges (E) of G are stored in two binary matrices, IN and OUT , each of size $n \times |\Sigma|$.
- 2 These two matrices are used to maintain the IN and OUT edge of each vertex. We can move each vertex forward and backward using this information.

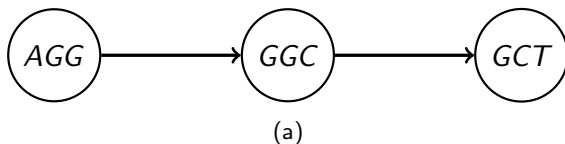
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- 3 The IN and OUT matrices can be constructed as:

$$(u = ba_1 \dots a_{k-1}, v = a_1 a_2 \dots a_{k-1} c) \in E \\ \iff OUT(f(u), c) = 1, IN(f(v), b) = 1.$$

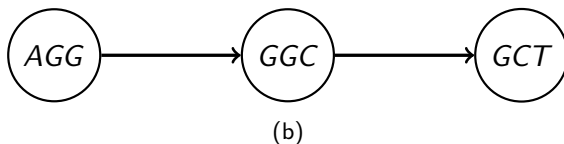
Example of *IN* and *OUT* matrices

Here is a simple de bruijn graph:



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Suppose $f(AGG) = 0, f(GGC) = 1, f(GCT) = 2$, the *IN* and *OUT* matrices can be initialized as:

Example of *IN* and *OUT* matrices

IN	A	G	C	T
0(AGG)	0	0	0	0
1(GGC)	1	0	0	0
2(GCT)	0	1	0	0

OUT	A	G	C	T
0(AGG)	0	0	1	0
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(a)

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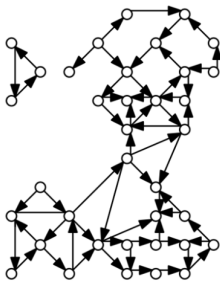
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- 3 Our strategy is to sample a subset of nodes for which we store the plain-text k-tuple and connect all the un-sampled nodes to the sampled ones.
- 4 Given a start point, we can move forward and backward using *IN* and *OUT* matrices. Once we reached the root, we can check if the resulting k-mer matches with root k-mer.

De Bruijn graph and Forest

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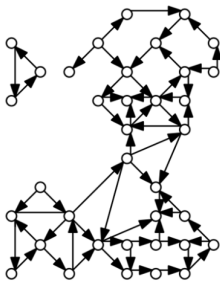
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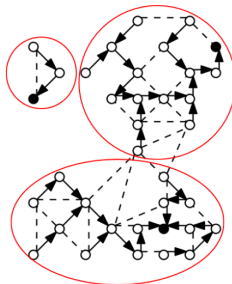
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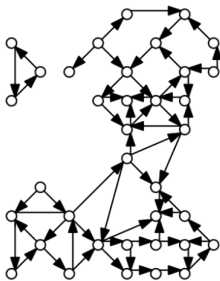
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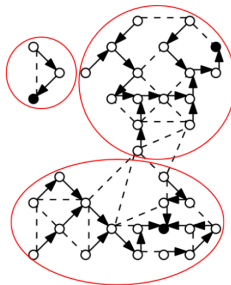
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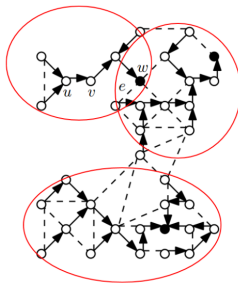
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Adding an edge

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- 3 listing the edges incident to a node we are visiting takes $O(\sigma)$ time, and crossing an edge takes $O(1)$ time.

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- K -mer representation
- Hash function implementation
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- “Semi-dynamic De Bruijn Graph”

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- Example:

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- How do we construct the Karp-Rabin hash function?

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- For $K = 27$ on *E. coli*, lower bound for prime is

16584693176107222092.

Max value in unsigned 64-bit integer:

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- Note that the hash function can hash any kmer, but is bijective when restricted to kmers that actually exist in our De Bruijn graph.
- That completes the generation of the hash function.

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Construction of Hash function

```
1: procedure GENERATEHASH
   Input  $S$ , a set of  $n$   $k$ -tuples over an alphabet  $\Sigma$  of size  $\sigma$ 
2:    $R = \max(\sigma, kn^2)$ 
3:    $P = \text{getPrime}(R)$ 
4:    $r = \text{randomNumber}(0, P - 1)$ 
5:    $f = \text{rabinHash}(r, P)$ 
6:   while  $\text{isInjective}(f, S)$  is FALSE do
7:      $r = \text{randomNumber}(0, P - 1)$ 
8:      $f = \text{rabinHash}(r, P)$ 
9:   end while
10:   $g = \text{minimalPerfectHash}(f(S))$  ;
11:  return  $g \circ f$  ;
12: end procedure
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$$f(n) = \frac{f(m) - first \cdot r}{r} + last \cdot r^K$$

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- What is the problem with naively implementing this update?

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- Solution to second problem: precompute $r^{-1} \bmod P$ and store it. (Requires generalized Euclidean algorithm)
- Solution to first problem? Hint: What is $P \bmod P$?

$$f(n) = \frac{(f(m) - \text{first} \cdot r)}{r} + \text{last} \cdot r^K$$

- First problem is that since $f(m)$ was computed mod P , first term might be negative.
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- Add $(4P - \text{first} \cdot r)$ to $f(m)$, as this will always be nonnegative.

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- This will slow data access down since multiple operations are required for each access, but much more memory efficient.

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- Example with two ints (each containing 32 bits).

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- With this class, only waste at most 31 bits in memory per instance of the class.

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- So we guarantee to use only $n\sigma + 31$ bits of memory for each of IN,OUT.
- To construct, simply read through edge $k + 1$ -mers and set the correct index of each of IN, OUT.

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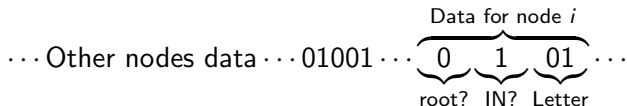
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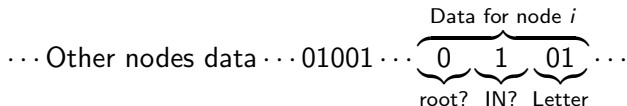
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- This is how we will do membership queries (explained more later).

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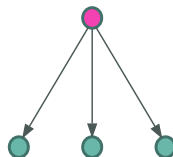
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- Add its hash and kmer to the map.



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De Bruijn breadth first search

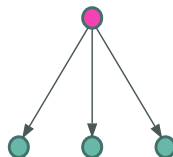
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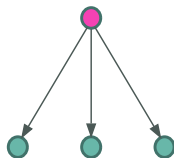
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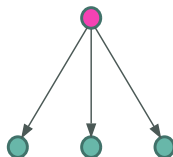
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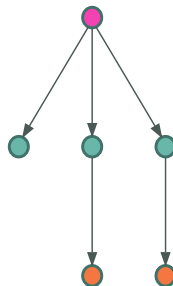
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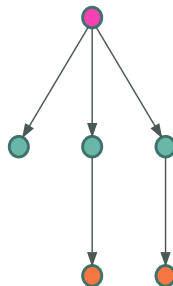
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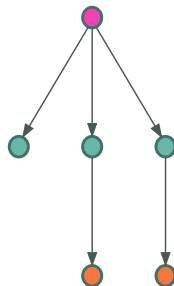
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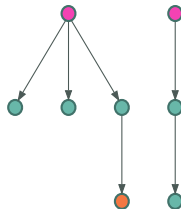
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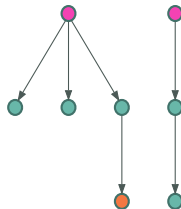
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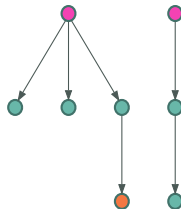
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- Continue on until the entire De Bruijn graph has been visited.



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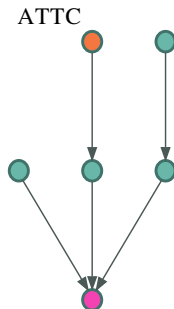
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- We travel up the tree to the root ...

Membership Query

Membership of "ATTC"

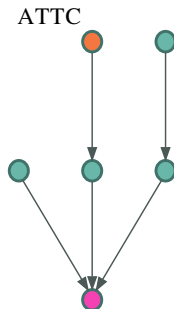
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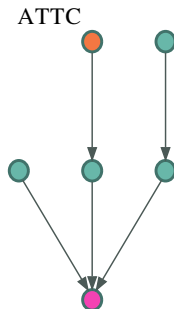
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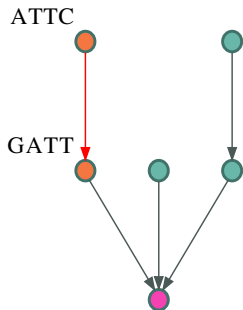
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- Note:
Even if the kmer is not in our De Bruijn graph, we can still hash it and get a place in the forest.



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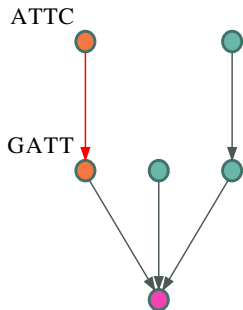
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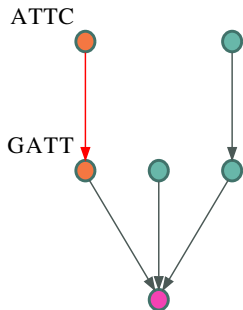
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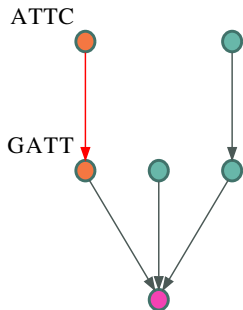
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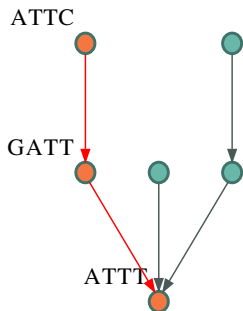
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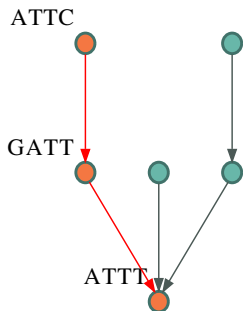
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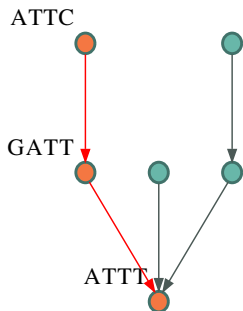
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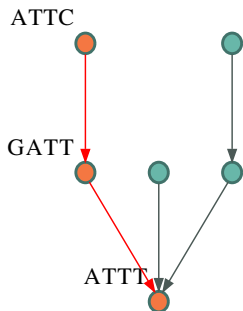
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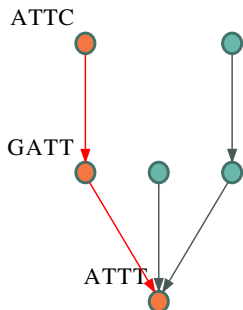
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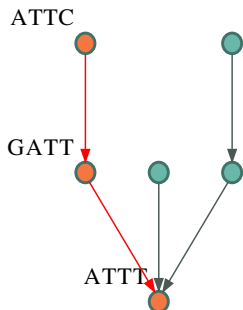
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- We rely on the library BBHash for our minimal perfect hash function, but we need a dynamic perfect hash function.

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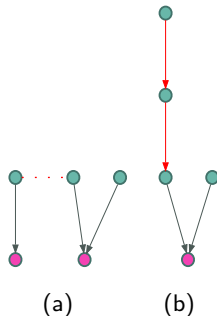
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- We may be able to combine the trees so that we have trees with heights in the desired range.

Dynamic Edges

Adding an Edge

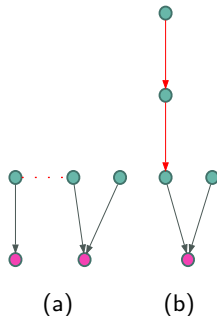
- Suppose that the edge (dotted) is between two trees in the forest below the minimum height, shown in (a).



Dynamic Edges

Adding an Edge

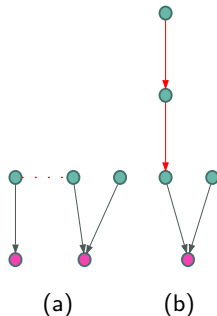
- Suppose that the edge (dotted) is between two trees in the forest below the minimum height, shown in (a).
- We can combine the trees into one tree of height less than the maximum.



Dynamic Edges

Adding an Edge

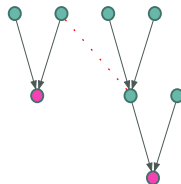
- Suppose that the edge (dotted) is between two trees in the forest below the minimum height, shown in (a).
- We can combine the trees into one tree of height less than the maximum.
- You have to add another edge to the forest, reverse the direction of some forest edges, then get rid of one of the tree's roots.



Dynamic Edges

Adding an Edge

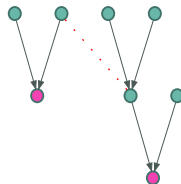
- Now suppose that only one of the trees are below the desired minimum height and we add an edge (dotted).



Dynamic Edges

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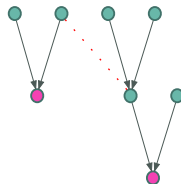
- Now suppose that only one of the trees are below the desired minimum height and we add an edge (dotted).
- What we do depends on the height of the node in the edge from the taller tree.



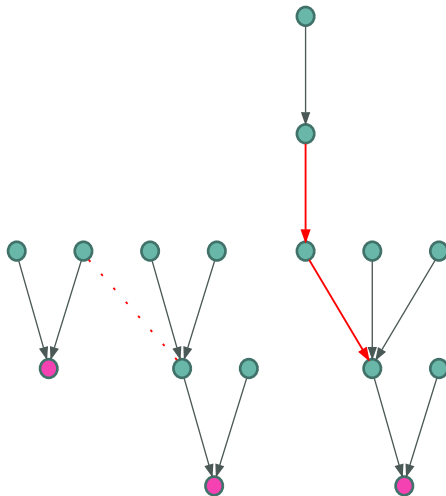
Dynamic Edges

Adding an Edge

- Now suppose that only one of the trees are below the desired minimum height and we add an edge (dotted).
- What we do depends on the height of the node in the edge from the taller tree.
- If the height is less than α , we can change the trees similar to before.



Adding Edges



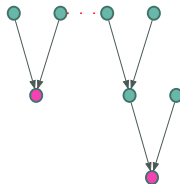
(a) $\text{Height} < \alpha$

(b)

Dynamic Edges

Adding an Edge

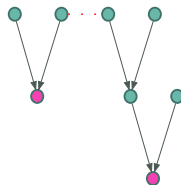
- Now suppose that the height is greater than or equal to α



Dynamic Edges

Adding an Edge

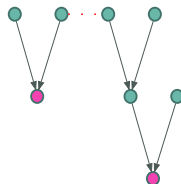
- Now suppose that the height is greater than or equal to α
- We can't necessarily just combine the trees like before, we could end up with a tree greater than the maximum height.



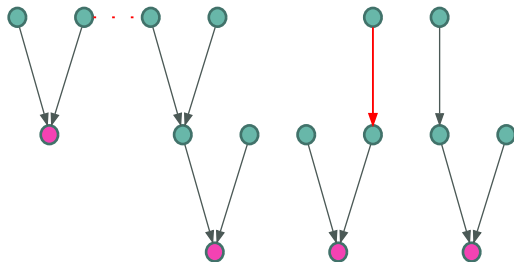
Dynamic Edges

Adding an Edge

- Now suppose that the height is greater than or equal to α
- We can't necessarily just combine the trees like before, we could end up with a tree greater than the maximum height.
- We break off some part of the bigger tree into the smaller.



Adding Edges



(a) Height $\geq \alpha$

(b)

Removing Edges

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- If the edge is in our tree, then removing it breaks up a tree.
- We need to find a new root for one of the trees
- If a tree is too short, use similar techniques to above to produce a taller tree.

Adding and Removing Nodes

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Adding and Removing Nodes

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- The primary reason is that the minimal perfect hash function library we used, BBHash, isn't dynamic.
- We would need to implement our own dynamic perfect hash function.

Experiments

Datasets:

Dataset	Chromosomes	Read count	Read length	Size
S288C ⁴	17	-	-	12M
<i>E. coli</i>	1	$27 \cdot 10^6$	101	3.3G

Platform:

- Intel(R) Xeon(R) CPU E5-2697 v4 @ 2.30GHz (18 cores) with 396 GB RAM

The results are tested by one run, which could be affected by computational resource conflicts.

⁴<http://www.yeastgenome.org/strain/S288C/overview>

Live Demo

- 1 We have developed a web-based demo to allow users do multiple types of comparisons, which makes the evaluation easier.
- 2 The link is here: <http://128.227.162.189:9999/>. Visit and play with it!

Fully Dynamic de Bruijn Graph Running Demo

This demo runs some examples and compare results.

K value:

of queries:
Dataset:

Go »View »

Comparative Algorithms

- ❶ **Bloom Filter:** Standard Bloom filter
- ❷ **KBF1:** One-sided Bloom filter improves false positive rate three fold without using any additional storage.
- ❸ **KBF2:** Two-sided Bloom filter improves FPR by an order of magnitude while using very little additional memory⁵.

⁵<https://github.com/Kingsford-Group/kbf>

Query k -mer Generation

- 1 We generate query k -mers with three different number of queries:

-	1	2	3
number of queries (million)	0.5	1	10

- 2 We use two random ways to generate query k -mers
 - Muting one base of randomly extracted from the input k -mers
 - ~~Purely random k -mer generation.~~ However, it does not result in obvious difference compared with the first one.

FDBG Data Structure Information

Table: The FDBG data structure information on E coli.

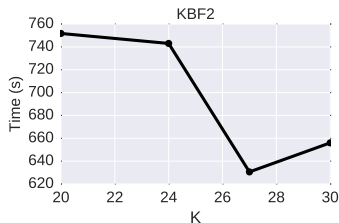
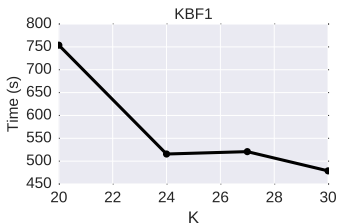
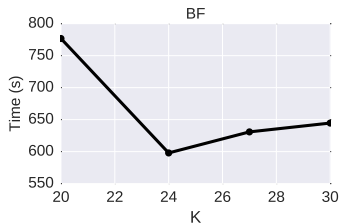
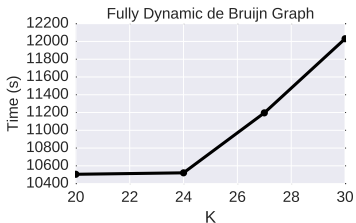
k	k -mers	RAM (MB)	Trees	Avg. height
20	770,956,037	1,485	5,492,320	48.14
24	784,990,222	1,519	6,412,386	50.45
27	783,739,686	1,517	6,532,142	47.28
30	776,321,600	1,505	6,752,622	48.46

- RAM = IN and OUT matrices + forest + minimal perfect hashing
- An example when $k = 20$, $N = 770956037$, $\sigma = 4$:

$$\begin{aligned} & N \times \sigma \times 2 + N \times 4 \times 2 + \text{mph} \\ &= (735 + 735 + 15) \text{MB} \times (1024 \times 1024 \times 8) \end{aligned}$$

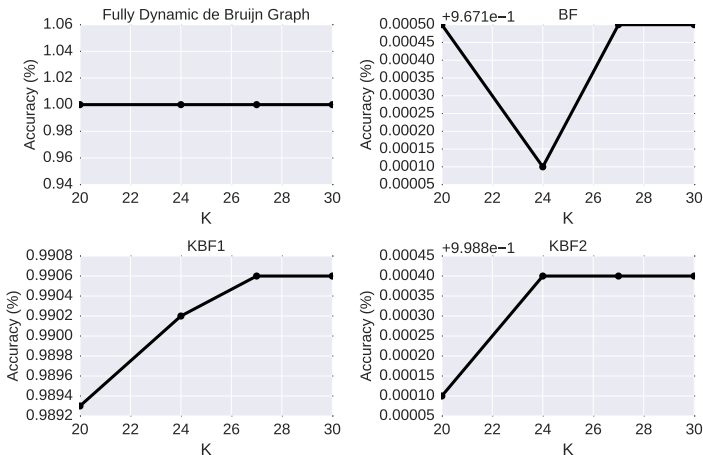
Populate Time w.r.t k

Populate time



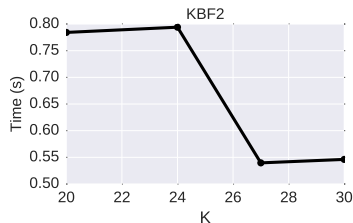
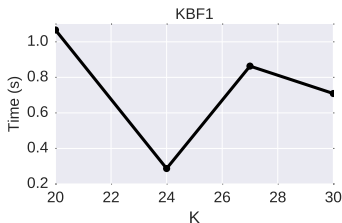
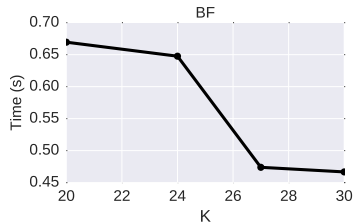
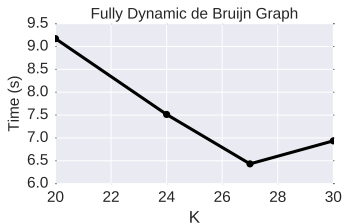
Query Accuracy w.r.t k

Query Accuracy



Query Time w.r.t k

Query time



Query Time w.r.t Tree Height

Table: Query time w.r.t the tree height.

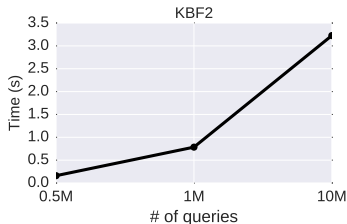
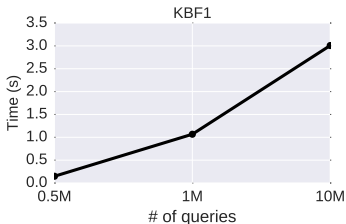
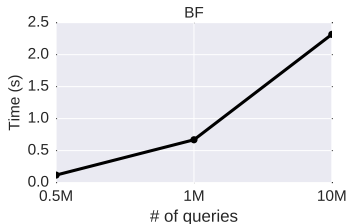
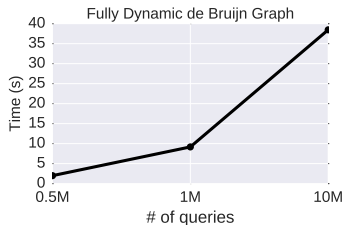
k	Avg. height	Query time (s)
20	48.14	6.69 ⁶
24	50.45	7.51
27	47.28	6.43
30	48.46	6.93

Lower tree height gives rise to lower query time, because it needs fewer steps to trace the tree.

⁶Averaged by two runs.

Query Time w.r.t Number of Queries

Query time



Conclusion

- 1 Have implemented static De Bruijn graph data structure and will complete edge insertion and deletion.
- 2 We have tested the algorithms on two gene datasets. Compared with all Bloom filter based methods, ours has exactly 100% accuracy.
- 3 We have compared with three different methods.
- 4 We have made a website and a live demo.

End

Thanks and Questions?