Universität Bielefeld

Computational Methods for the Analysis of the Diversity and Dynamics of Genomes

Efficient querying of colored de Bruijn graphs

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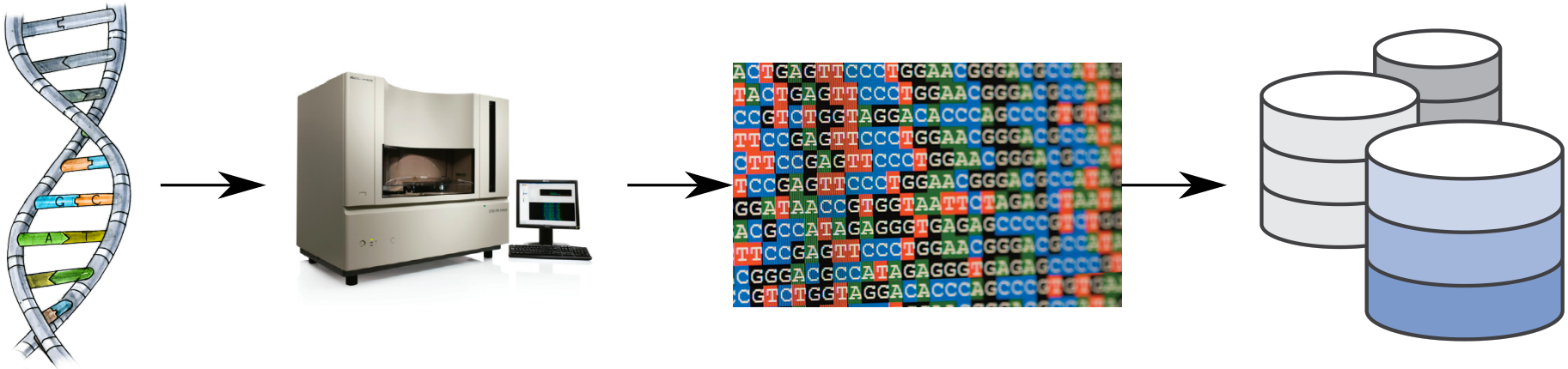
Contents

Motivation

DNA sequencing in former times:



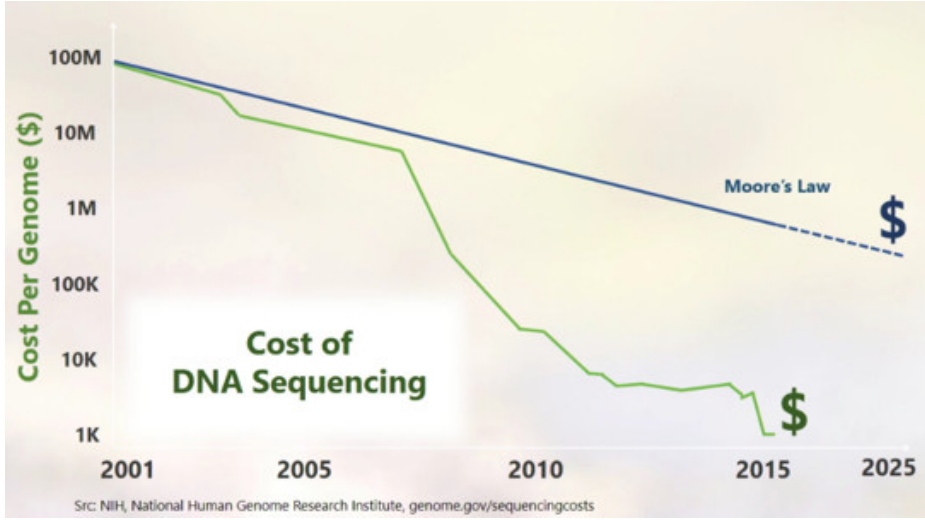
Especially, short DNA sequences were sequenced

- Single genes
- Viral genomes
- Small bacterial genomes

Motivation

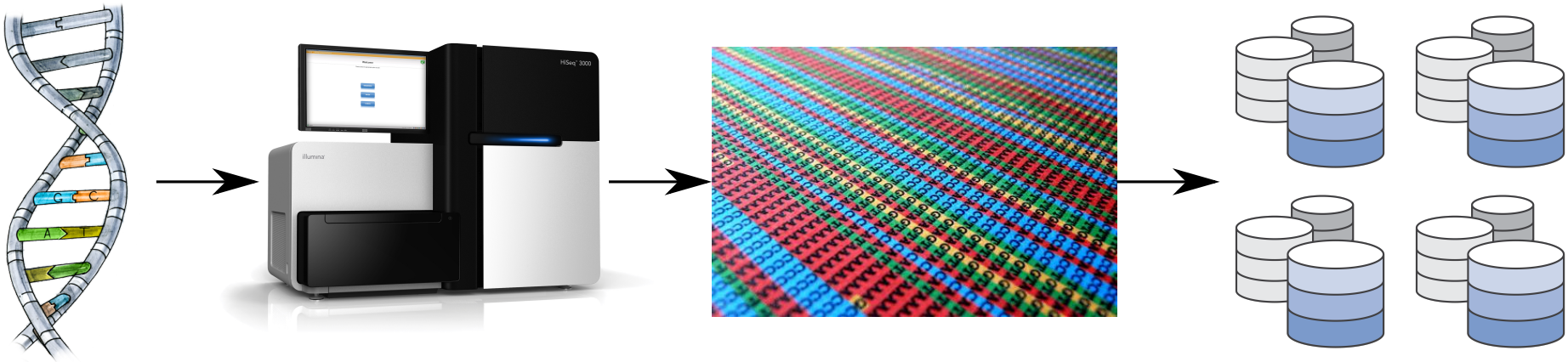
Current development:

- Sequencing technologies improve rapidly
- Sequencing becomes cheaper
- More and more sequencing projects are started



Motivation

DNA sequencing at the moment:

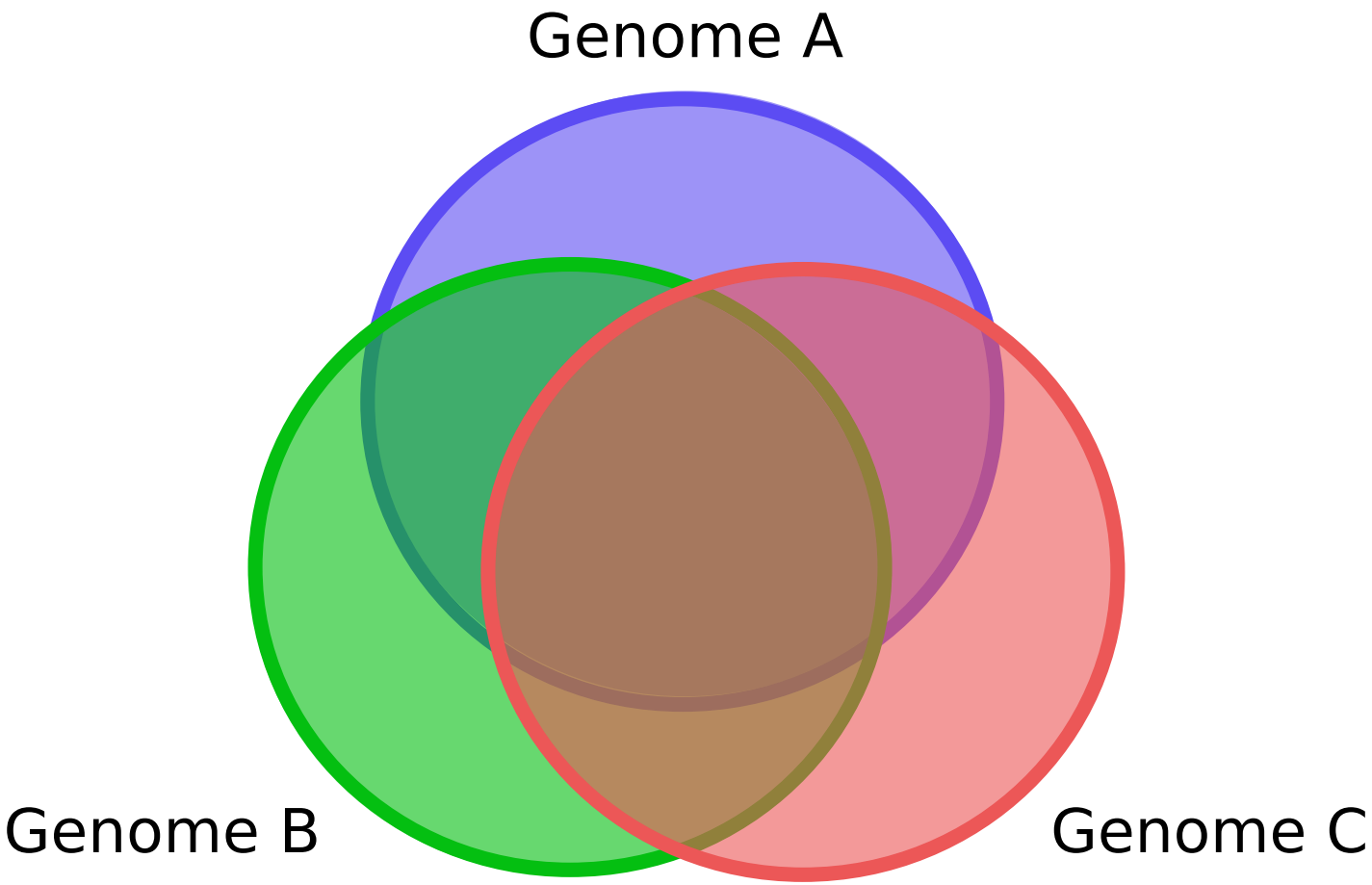


Long DNA sequences are sequenced:

- Whole genomes of complex plants and animals
- Many genomes per species

Motivation

How to store many highly similar DNA sequences, e. g. pan-genomes?



Colored de Bruijn graphs

Colored de Bruijn graphs

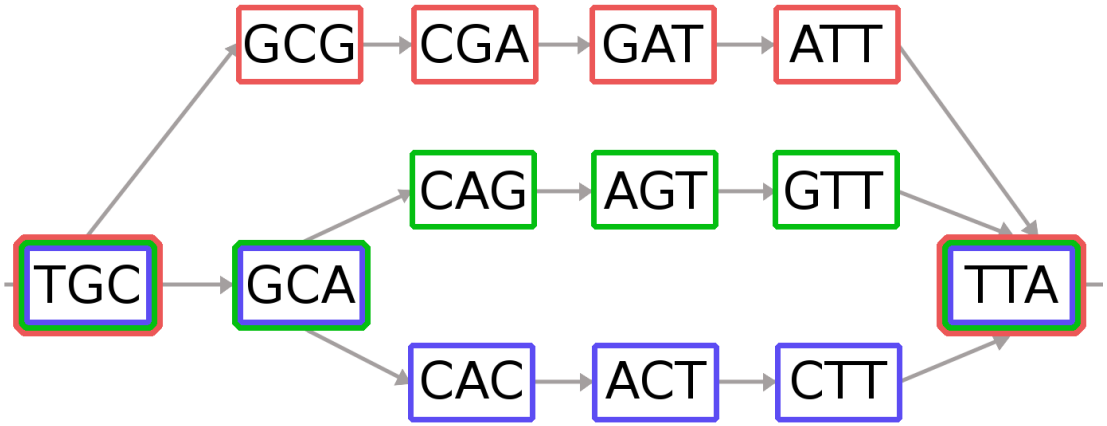
Colored de Bruijn graph (C-DBG)

$k = 3$

S_1 : TGCGATTA

S_2 : TGCAGTTA

S_3 : TGCACTTA



- Vertices represent substrings of length k (k-mers)
- Associated color represents sequence of origin
- Edges between vertices that share a $k-1$ overlap
- No explicit representation of edges necessary

Colored de Bruijn graphs

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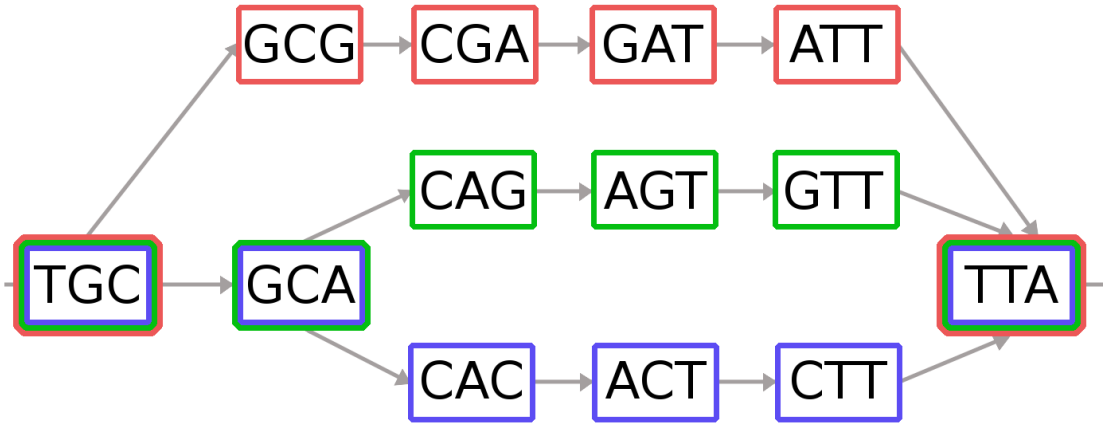

Graph allows:

1. Efficient storage of shared sequence parts
2. Reconstruction of sequences by graph traversal

Colored de Bruijn graphs

Compacted colored de Bruijn graphs

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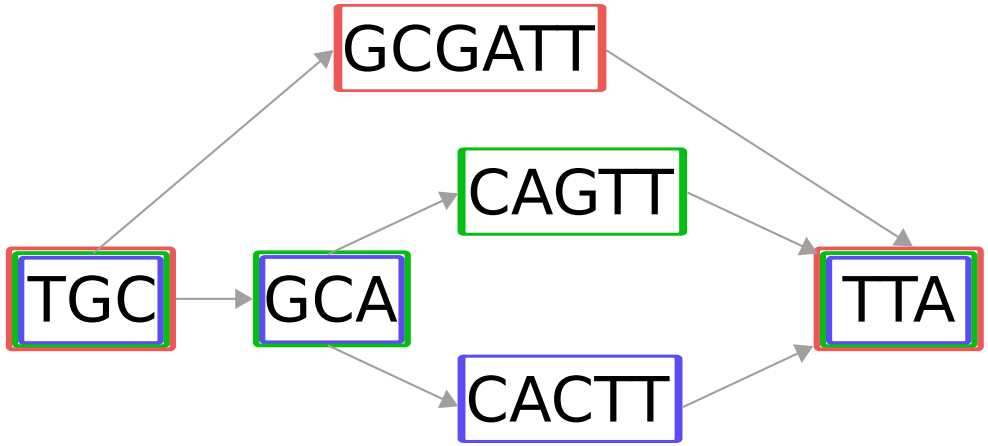

- Vertices of unique paths are replaced by single vertex (unitig)
- Label of unitig is sequence spelled by the path

Colored de Bruijn graphs

Compacted colored de Bruijn graphs

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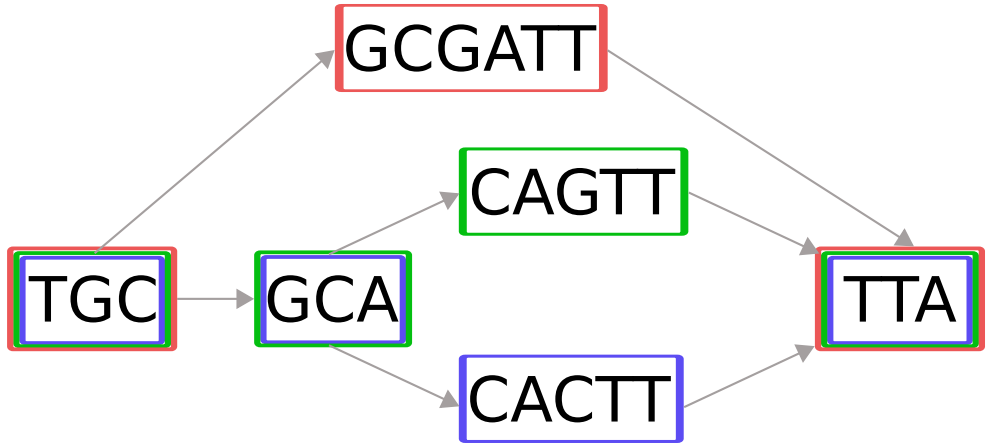


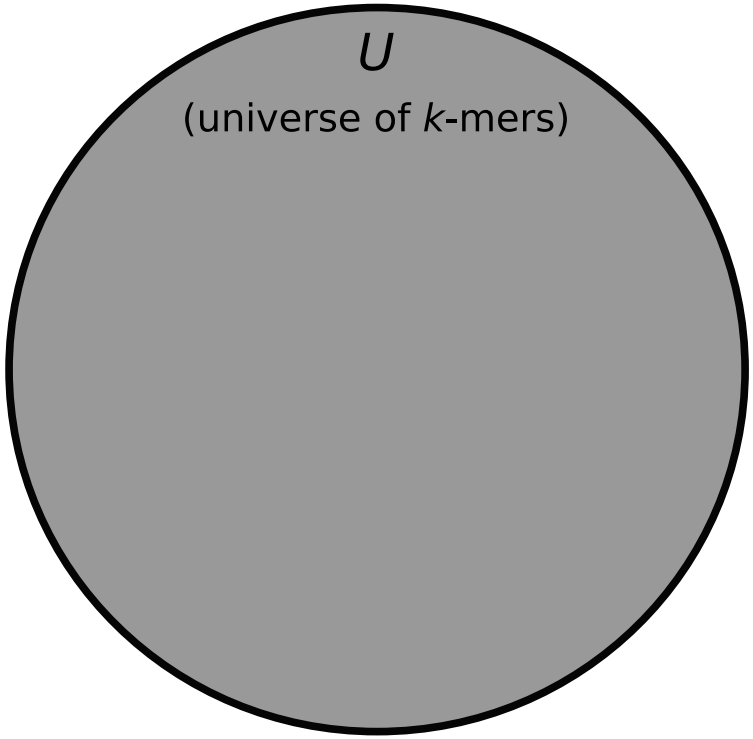
Extension allows:

1. Even more efficient storage
2. Faster sequence querying

Bloom Filter

Bloom Filter

Introduction Bloom filter

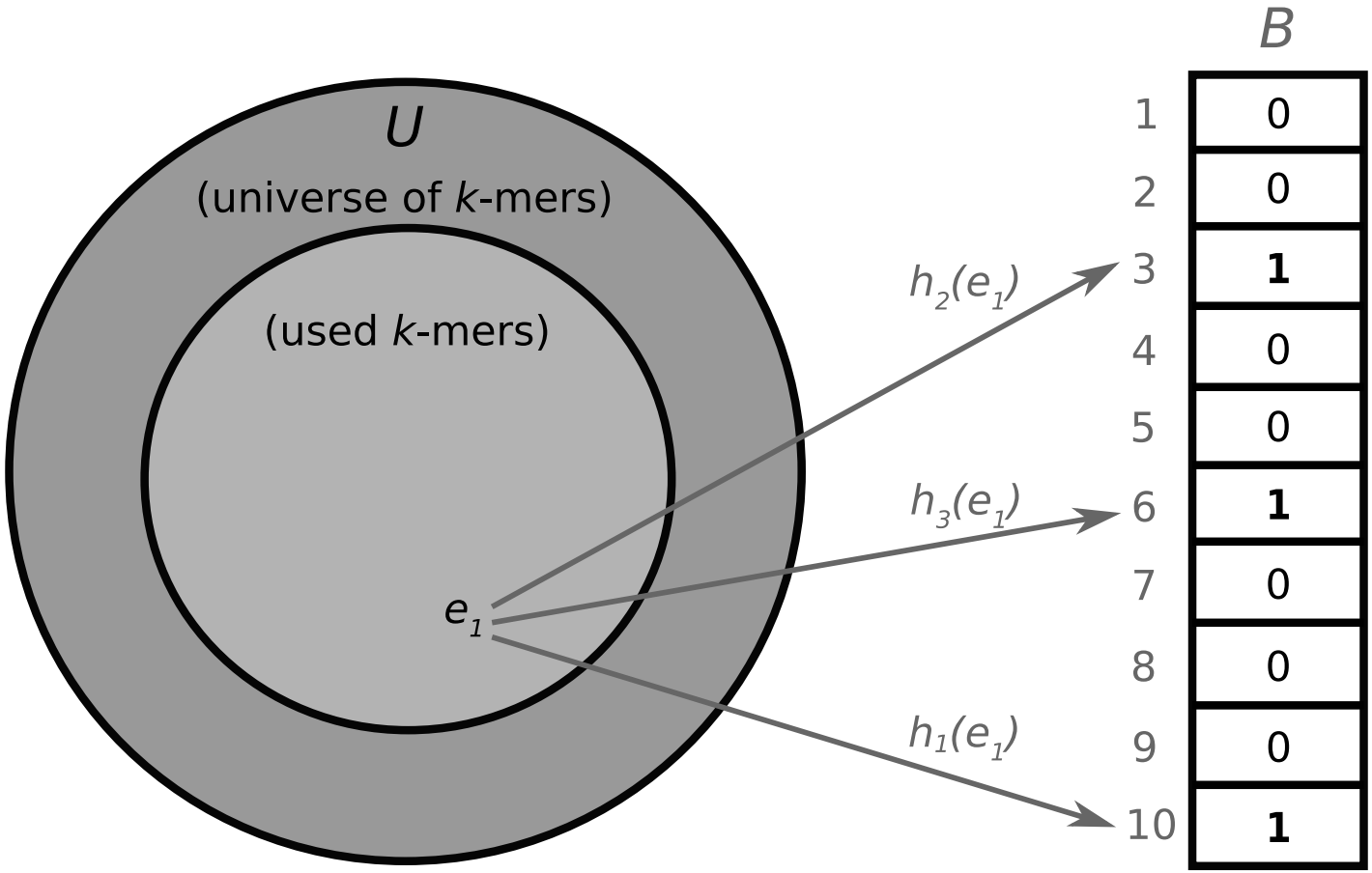


$h_1()$
$h_2()$
$h_3()$

	B
1	0
2	0
3	0
4	0
5	0
6	0
7	0
8	0
9	0
10	0

Bloom Filter

Introduction Bloom filter



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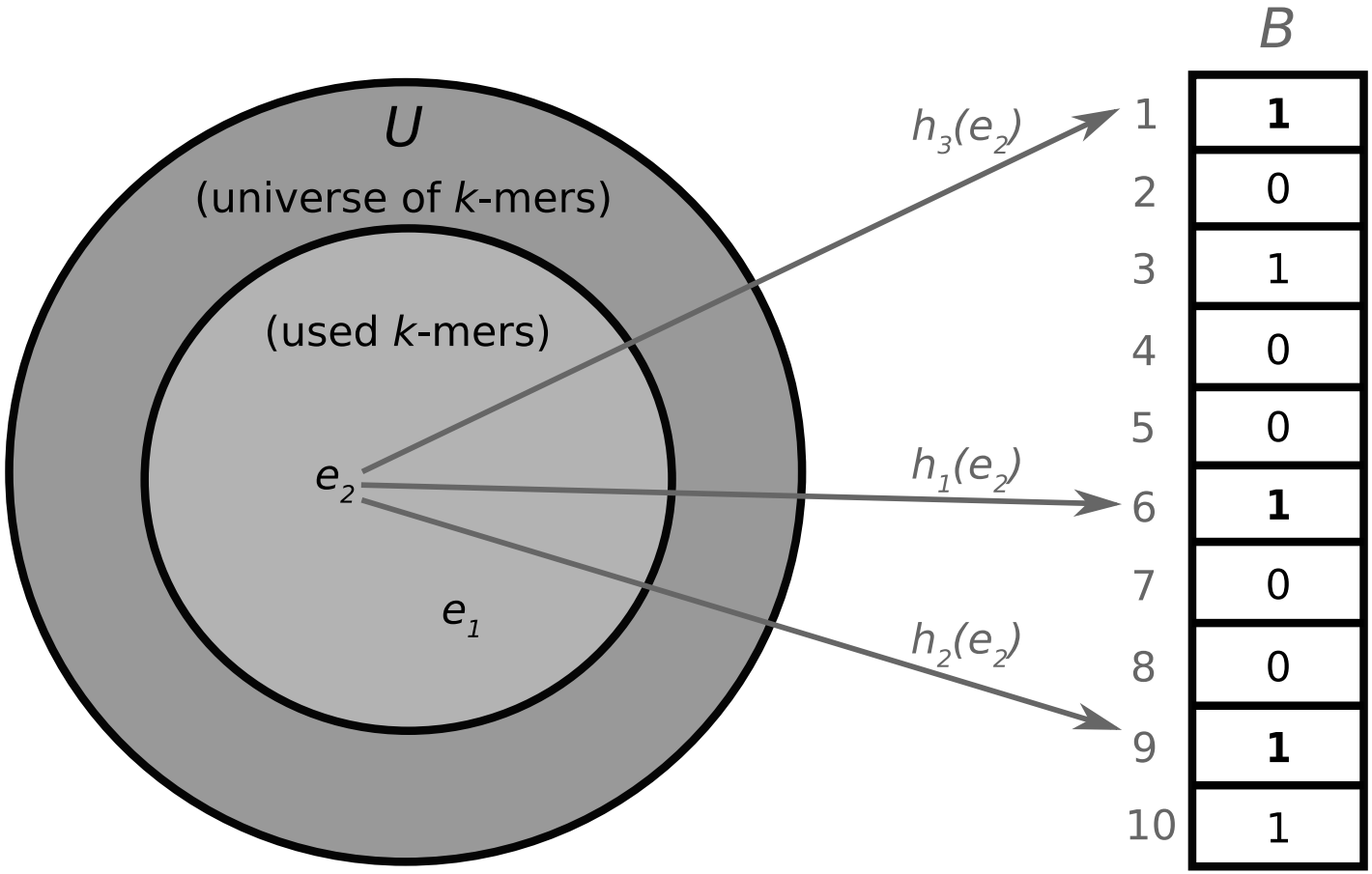

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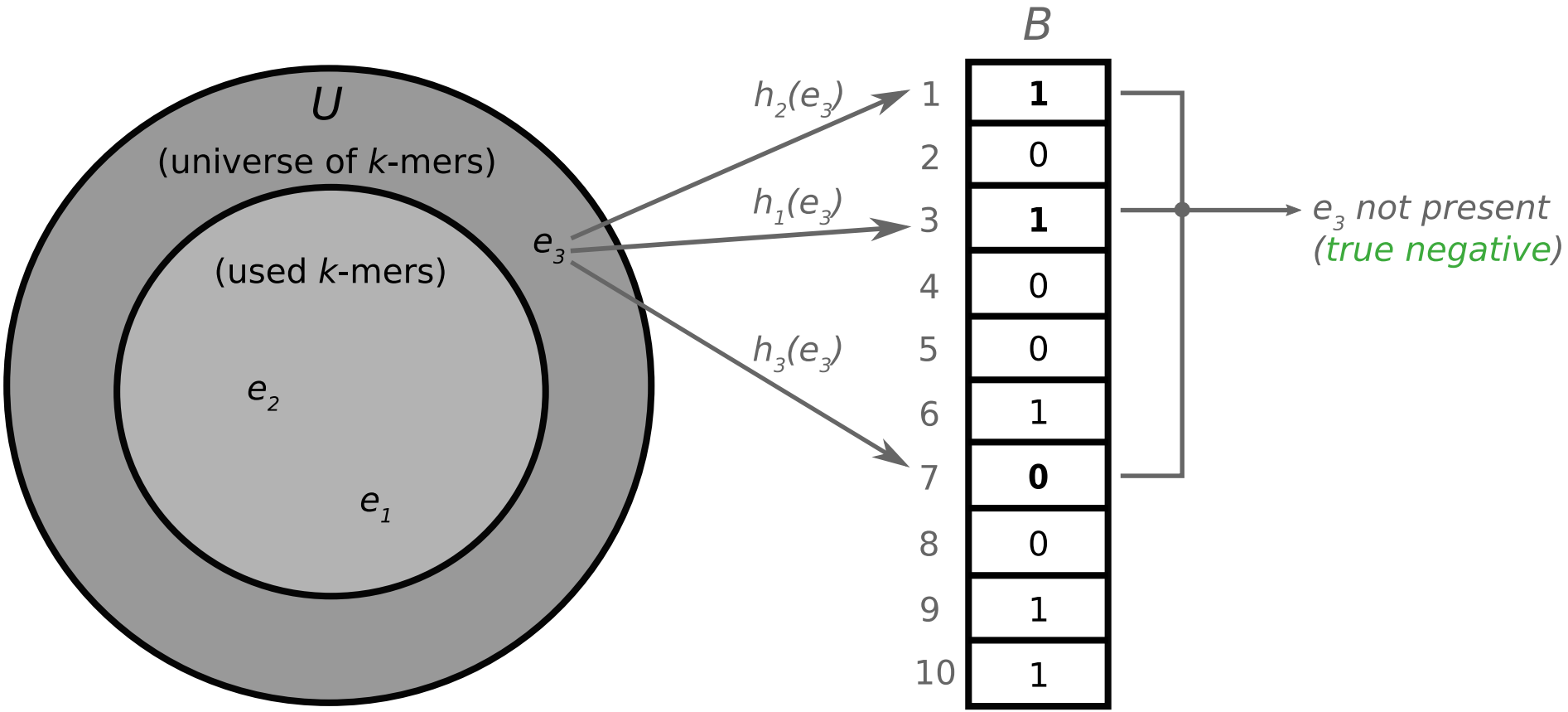

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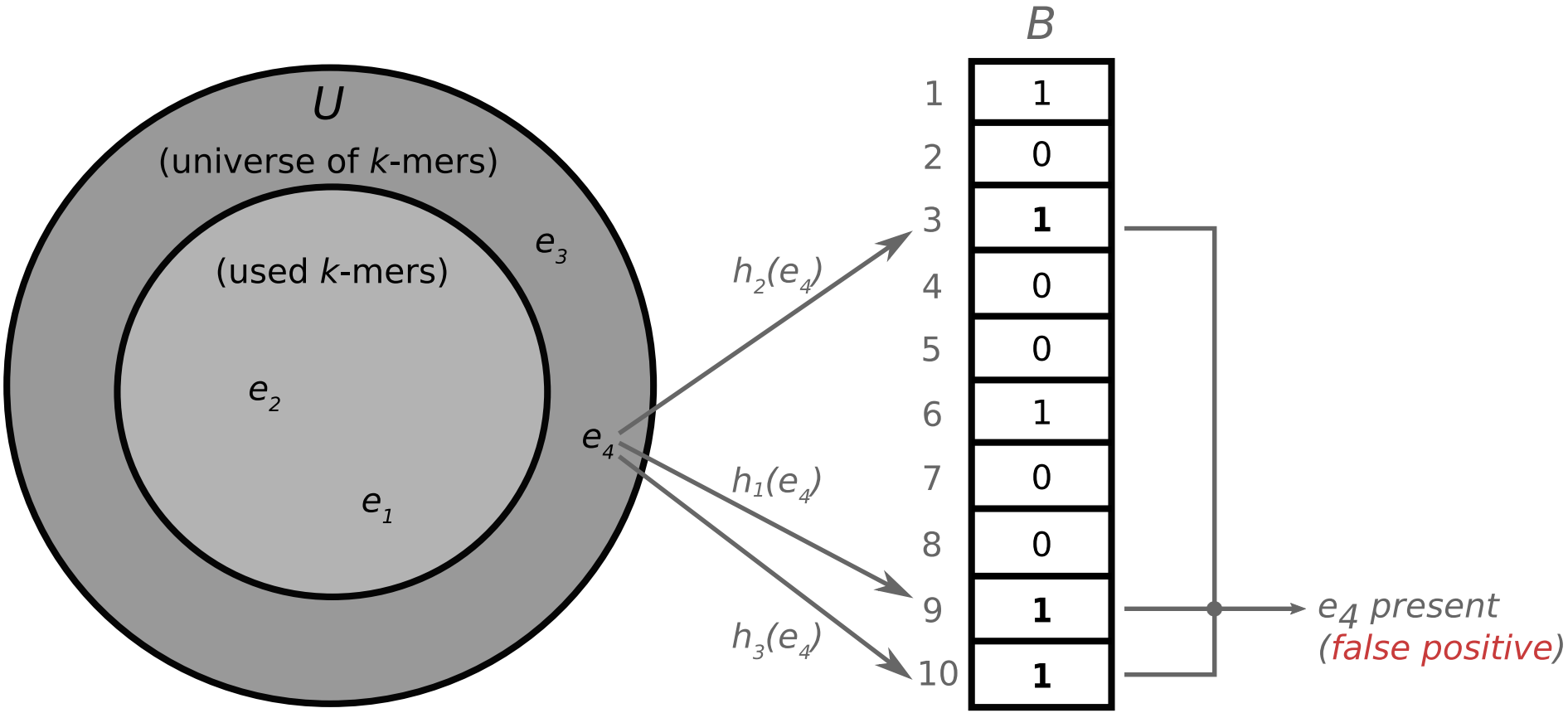


Note: Not possible to store color sets

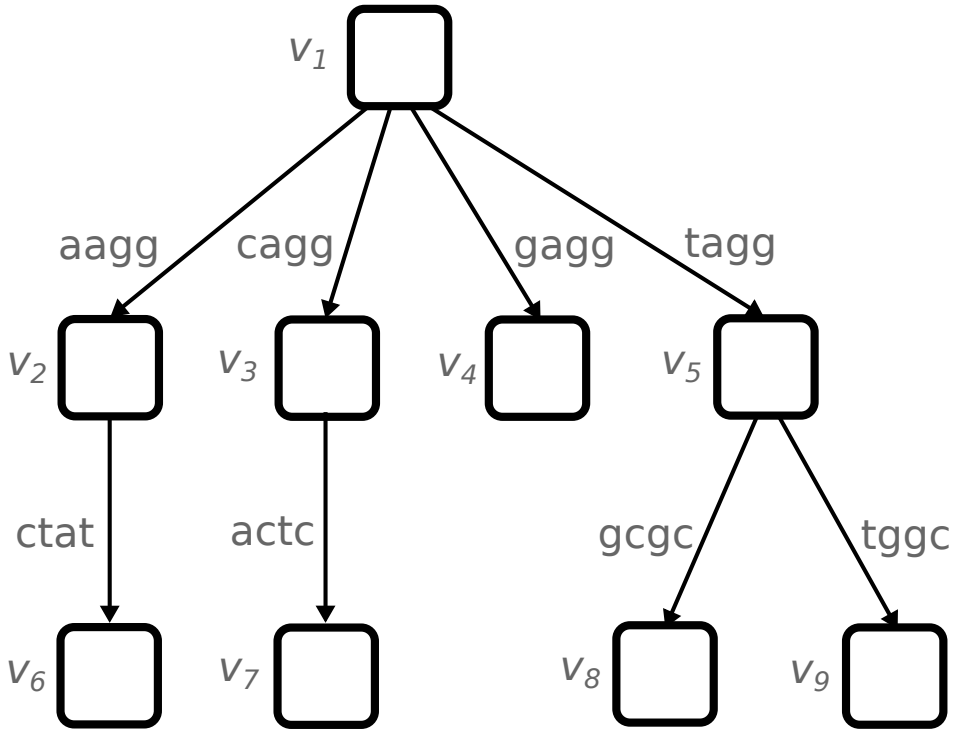
Bloom Filter Trie

Bloom Filter Trie

The Bloom Filter Trie – BFT (Holley *et al.*, 2016)

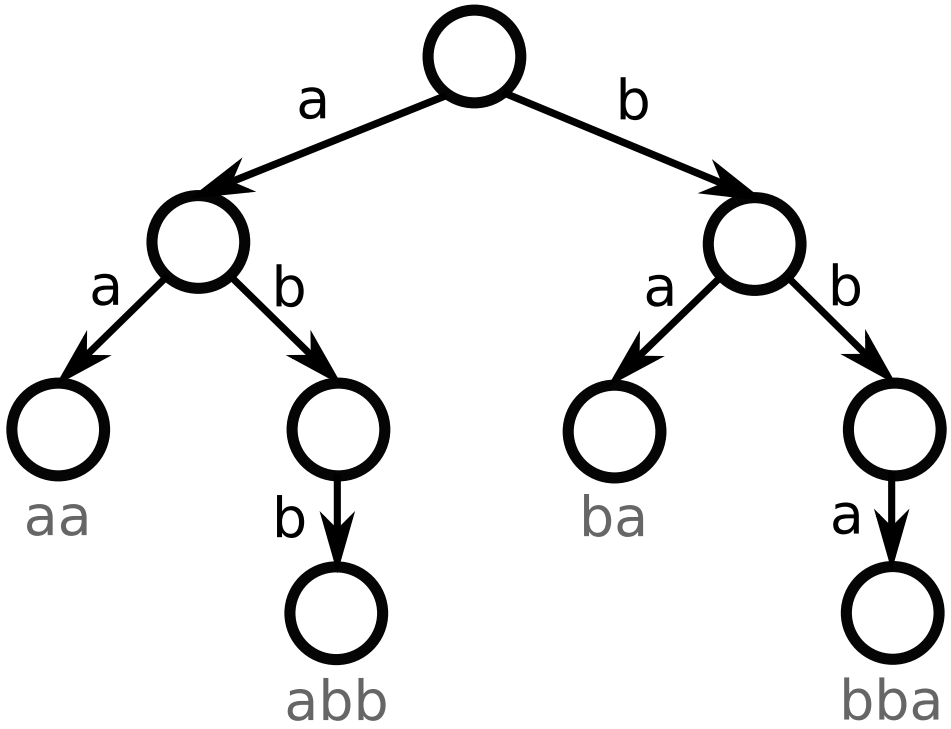
Implementation of a C-DBG

Only k-mers associated with colors are stored in a burst trie



Bloom Filter Trie

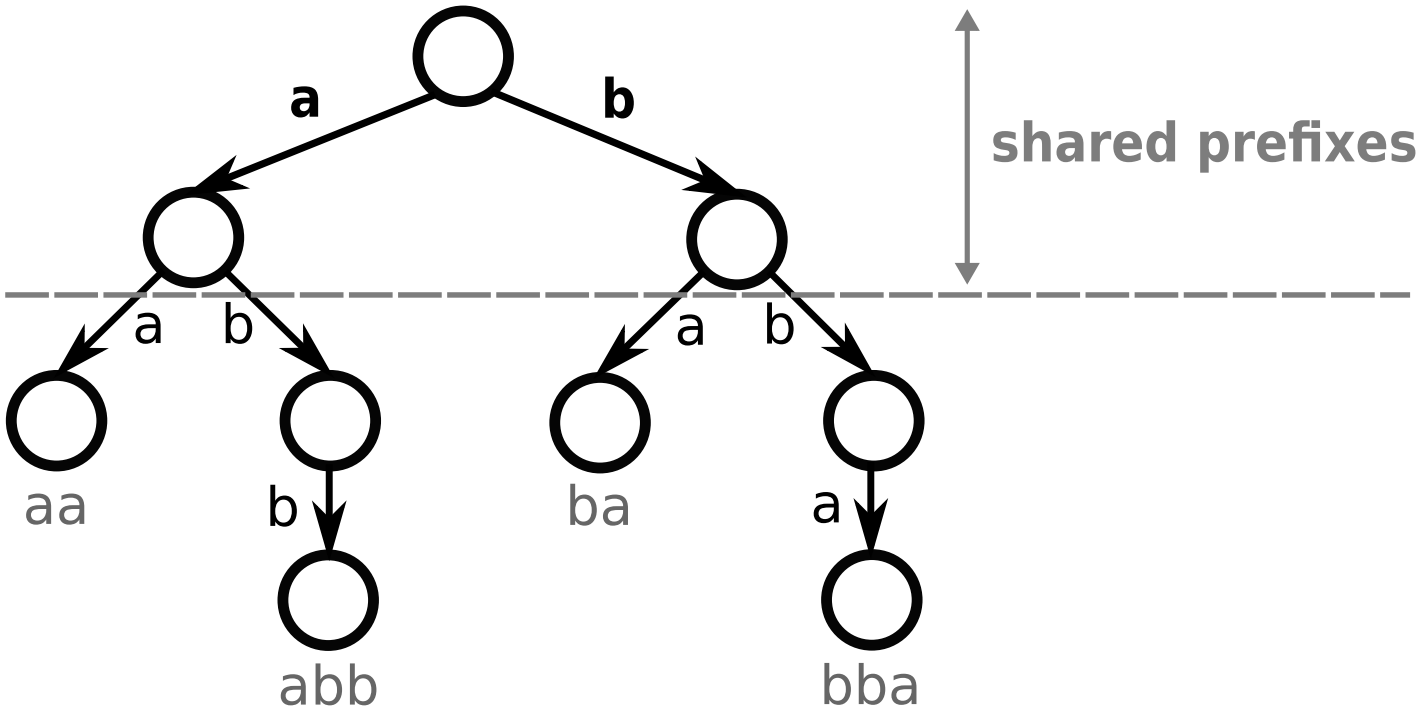
Introduction Trie



Trie of string set $\{aa, abb, ba, bba\}$

Bloom Filter Trie

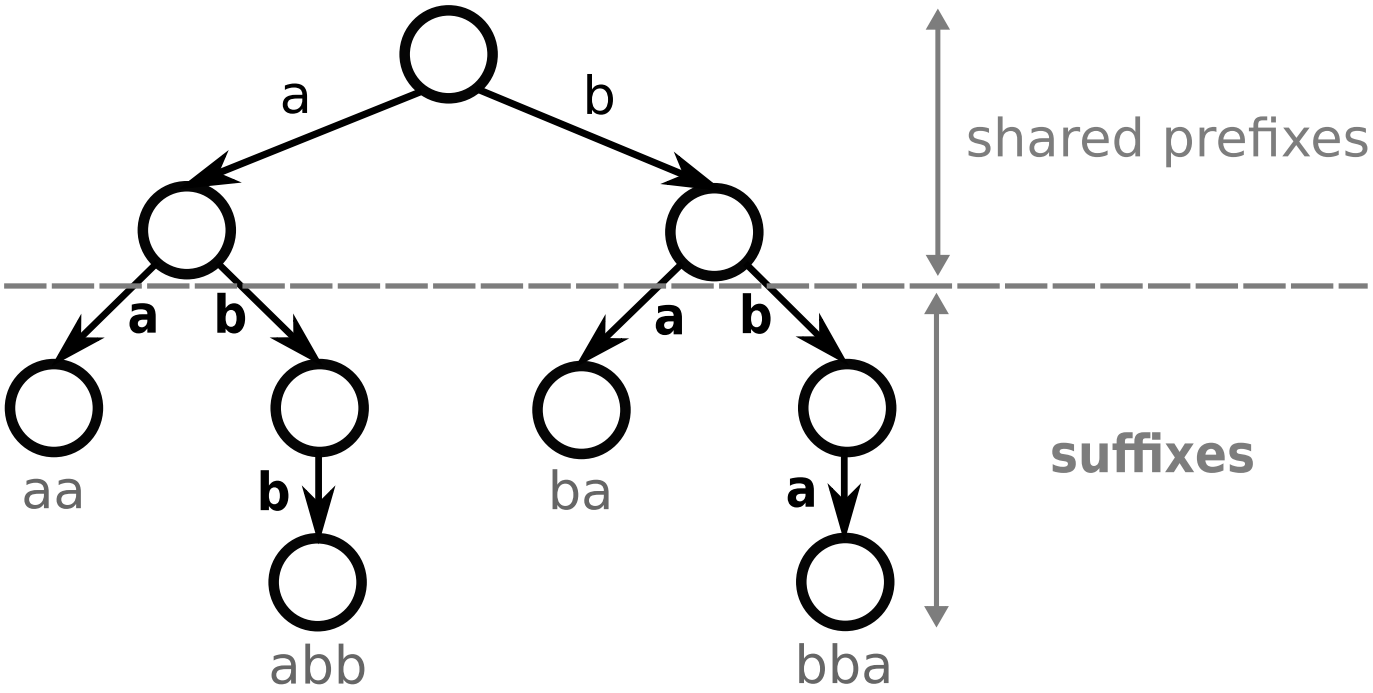
Introduction Trie



Trie of string set $\{\mathbf{a}\text{a}, \mathbf{a}\text{bb}, \mathbf{b}\text{a}, \mathbf{b}\text{ba}\}$

Bloom Filter Trie

Introduction Trie



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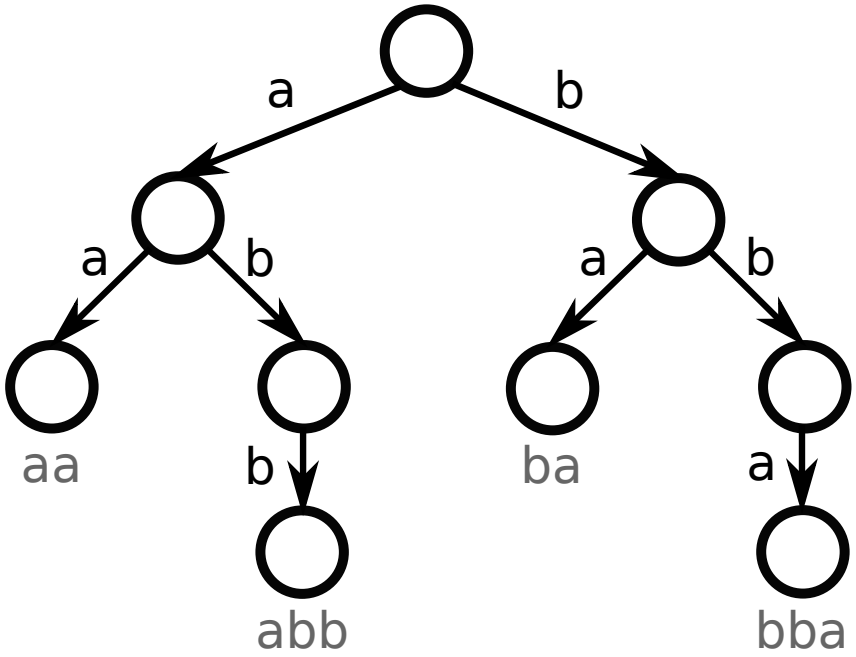
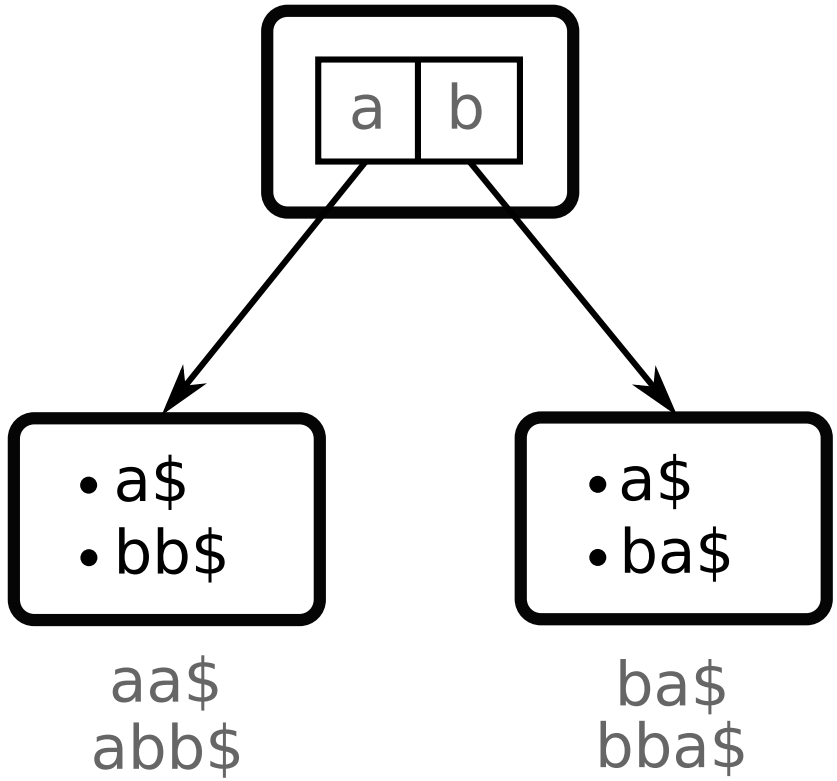
Bloom Filter Trie

Introduction Trie



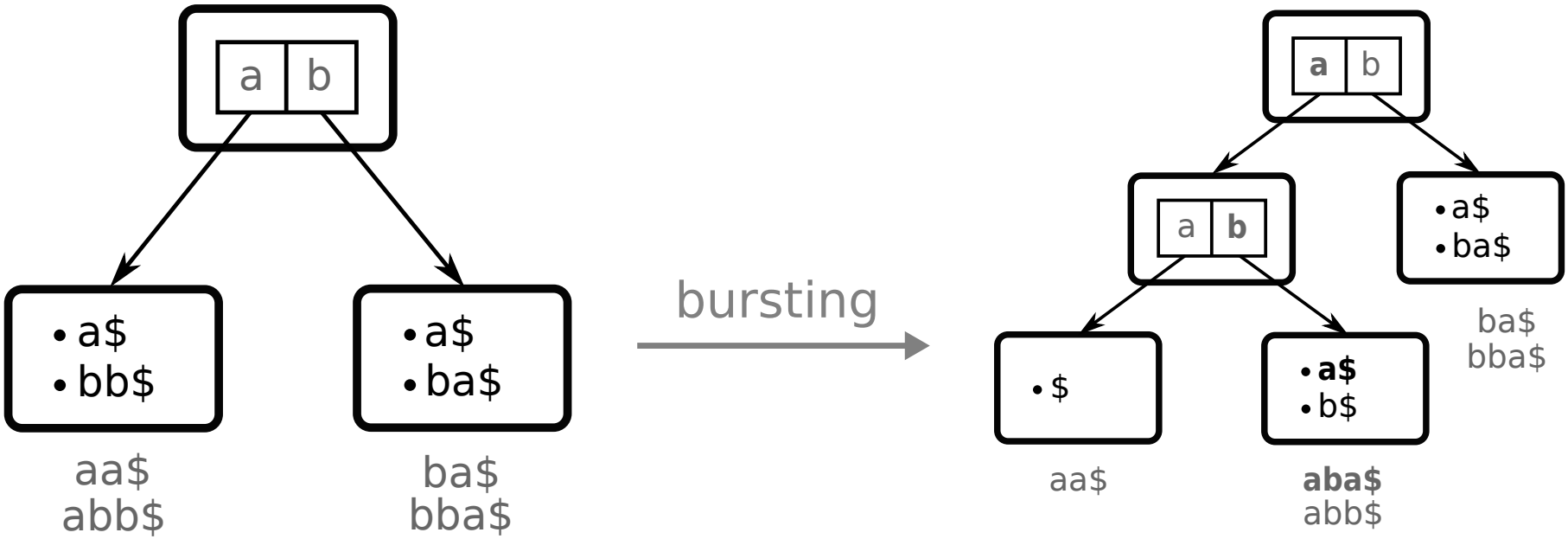
Trie



Burst-trie

Bloom Filter Trie

Introduction Trie

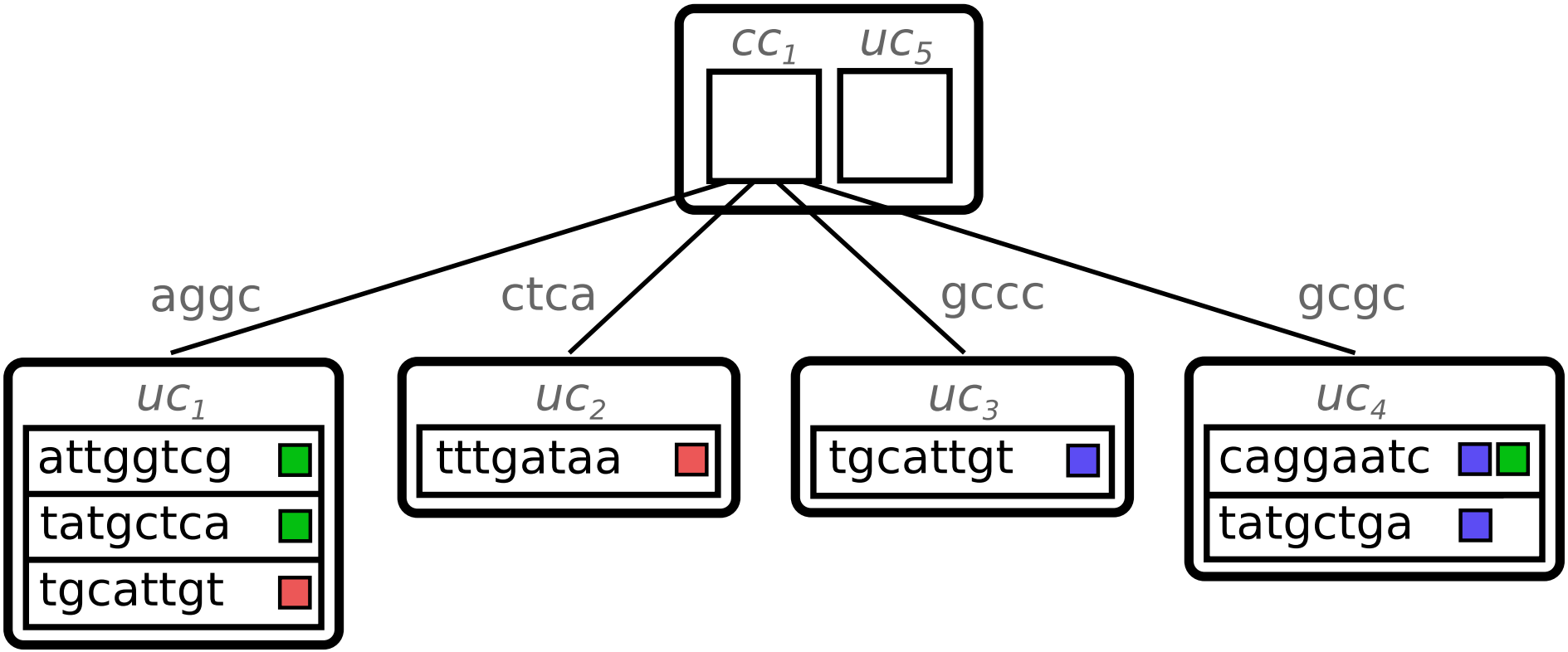


{aa, abb, ba, bba}

{aa, **aba**, abb, ba, bba}

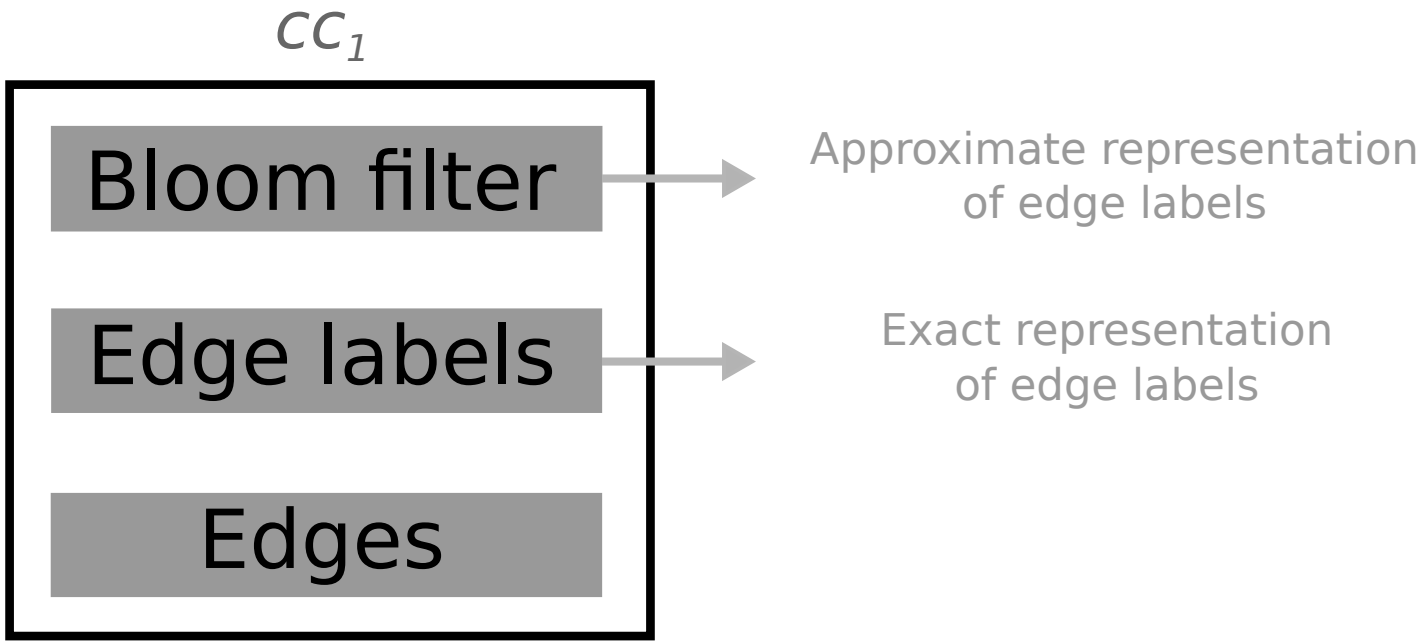
Bloom Filter Trie

Vertices of the Bloom Filter Trie



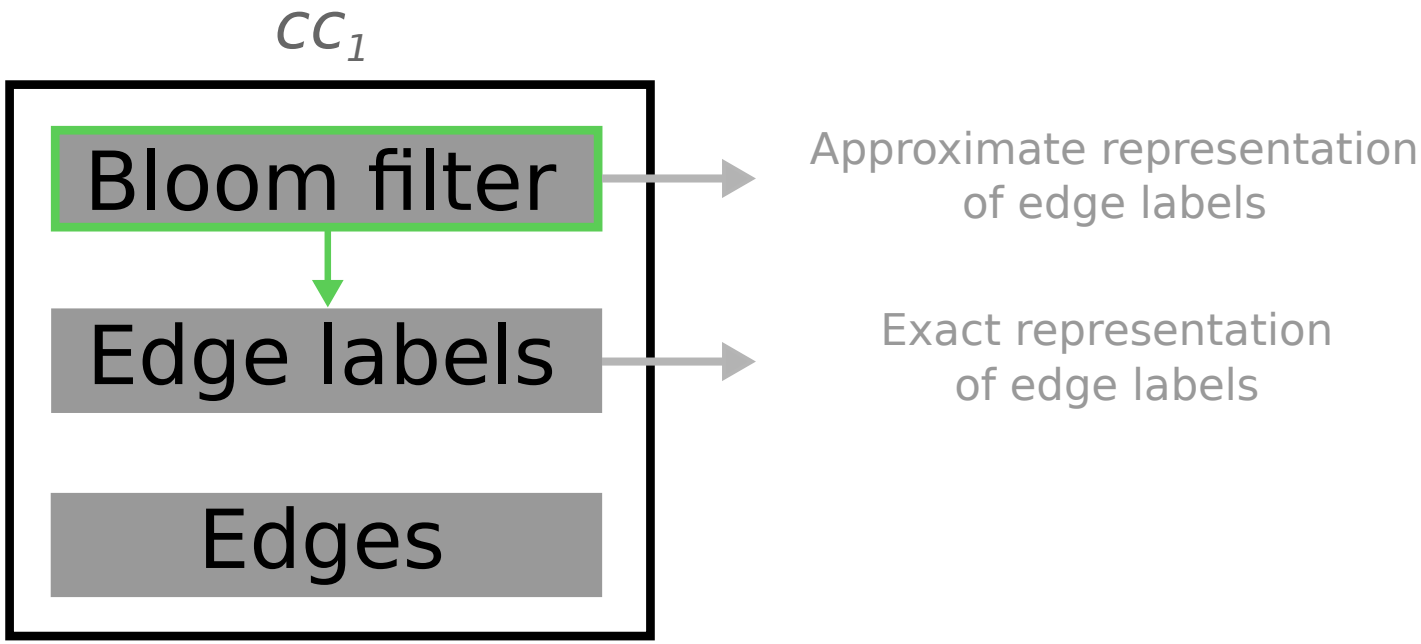
Bloom Filter Trie

Vertices of the Bloom Filter Trie



Bloom Filter Trie

Vertices of the Bloom Filter Trie



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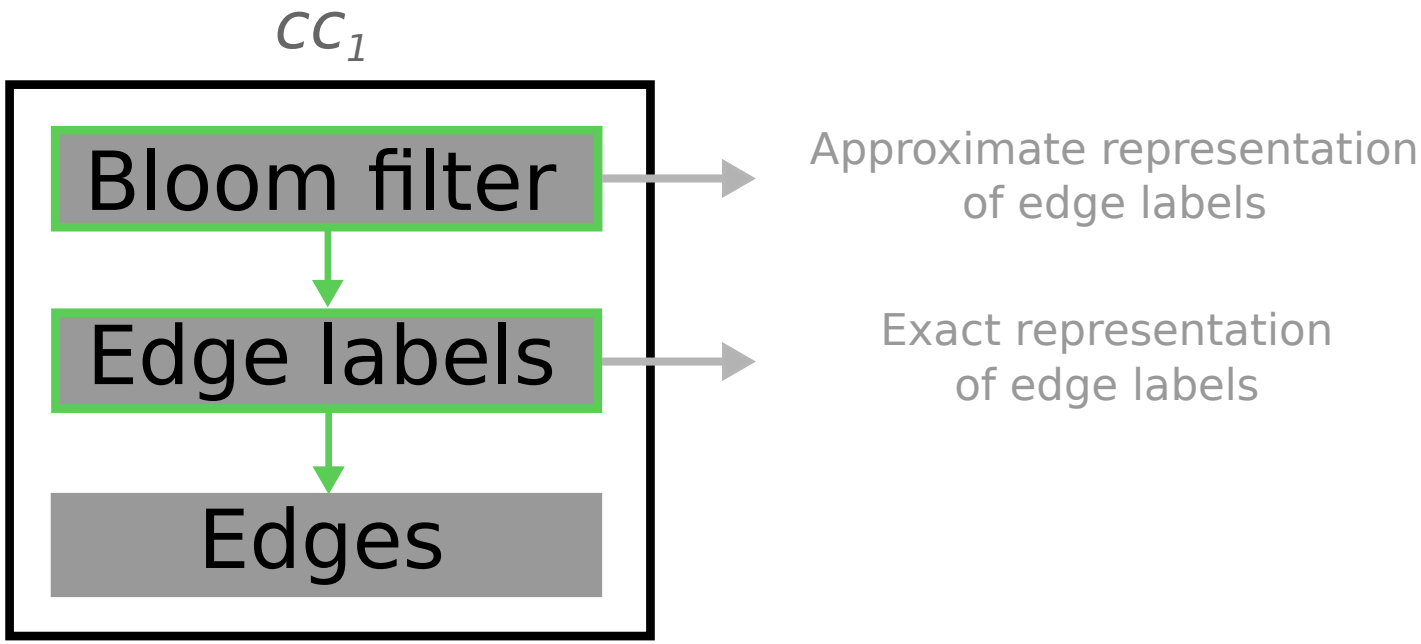

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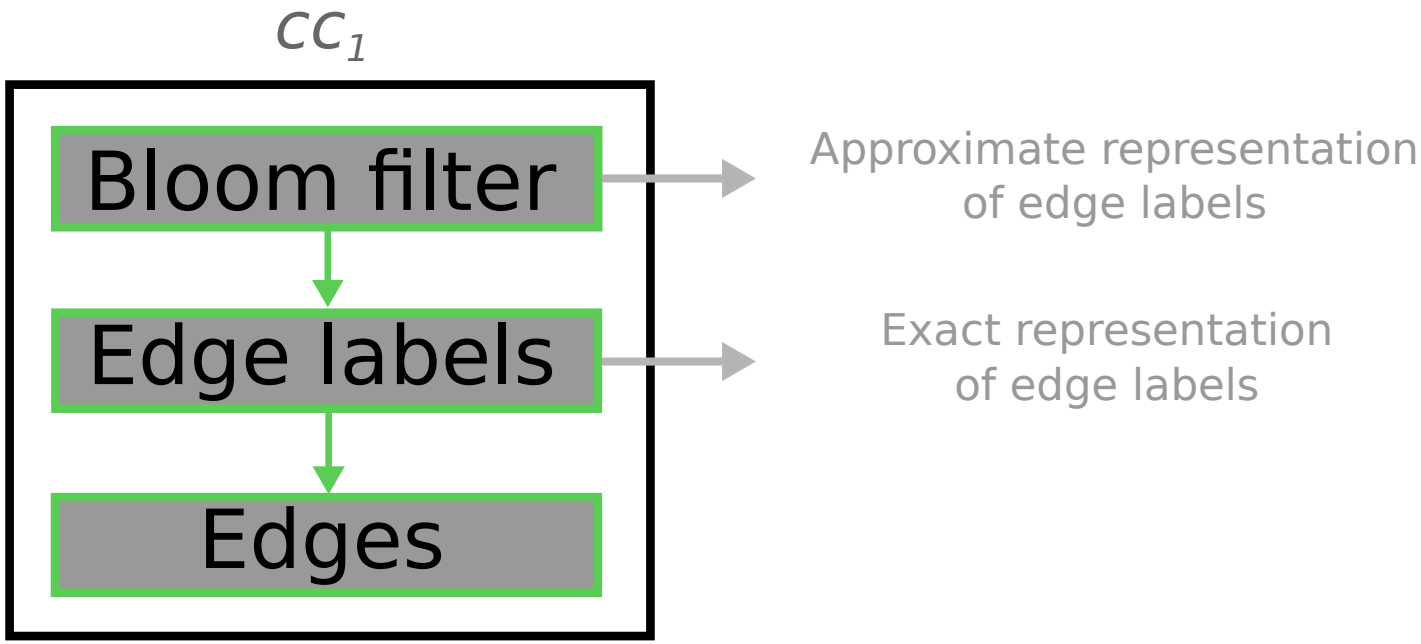

Bifrost

Bifrost

Bifrost (`https://github.com/pmelsted/bfgraph`)

Implementation of a compacted C-DBG

Currently under development

Unitigs can be accessed via minimizers they contain

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Unitigs can be accessed via minimizers they contain

Minimizer: lexicographically smallest ℓ-mer of a sequence and its reverse complement

$s = \text{TGACTTCCATGT}$
$\ell = 4$

ℓ-mer		rev. comp.	
TGAC	TCCA	GTCA	TGGA
GACT	CCAT	AGTC	ATGG
ACTT	CATG	AAGT	CATG
CTTC	ATGT	GAAG	ACAT
TTCC		GGAA	

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ℓ-mer		rev. comp.	
TGAC	TCCA	GTCA	TGGA
GACT	CCAT	AGTC	ATGG
ACTT	CATG	**AAGT**	CATG
CTTC	ATGT	GAAG	ACAT
TTCC		GGAA	

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Implementation of a compacted C-DBG

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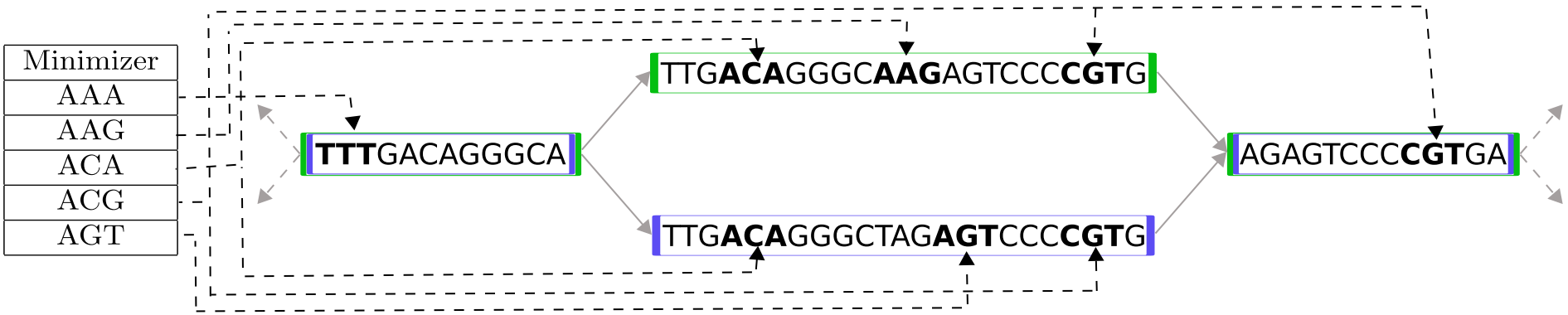
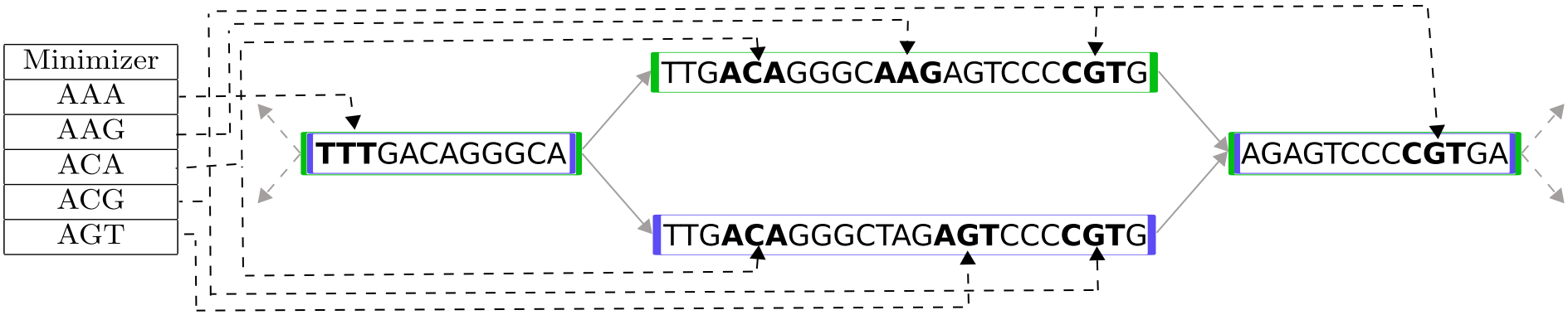


Bifrost

Why does it make sense to use minimizers?

- Many consecutive k-mers have the same minizer
- Minimizer index much smaller than k-mer index



Querying a sequence DB - BLAST

Querying a sequence DB - BLAST

BLAST – Basic Local Alignment Search Tool

Developed by Stephen Altschul *et al.* in 1990 (second version in 1997)
Is used to query databases of DNA and protein sequences

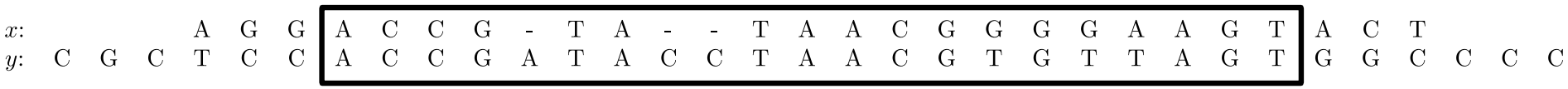
Different flavors are available:

- DNA→DNA (blastn)
- protein→protein (blastp)
- translated DNA→protein (blastx)
- protein→translated DNA (tblastn)
- ...

Querying a sequence DB - BLAST

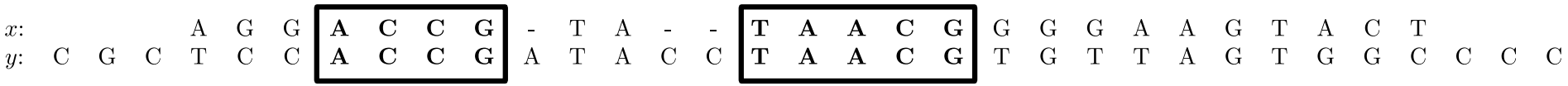
Problem statement:

Given a query sequence $x \in \Sigma^*$ and a sequence database Y, find highest scoring local alignments between x and $y \in Y$ above a certain significance score and return them along with score.



Querying a sequence DB - BLAST

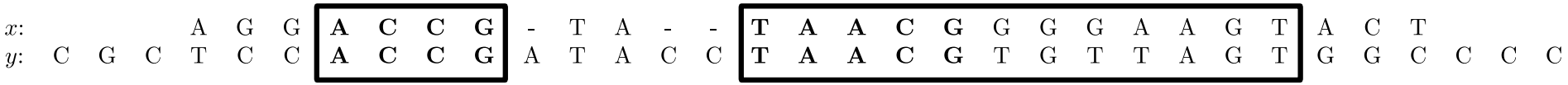
Algorithm procedure (blastn with default parameters):
1. Finding BLAST hits:
Find exact matches of minimal length w between x and all $y \in Y$.



$w = 4$

Querying a sequence DB - BLAST

2. Extension:
Extend hits to left and right using the *X-drop algorithm*.



Querying a sequence DB - BLAST

X-drop algorithm (extension to the right):
INPUT: query x, database sequence y, hit $h = (i, j)$, score s, drop-off parameter X
OUTPUT: extended hit h, increased score s

1: $s' \leftarrow 0$
2: $j' \leftarrow j + 1$
3: **while** end of x and y with reached **do**
4: **if** $s' += Score($next character in x and $y) > 0$ **then**
5: $s \leftarrow s'$
6: $j \leftarrow j'$
7: $s' \leftarrow 0$
8: **else if** $s' < X$ **then return** s and h
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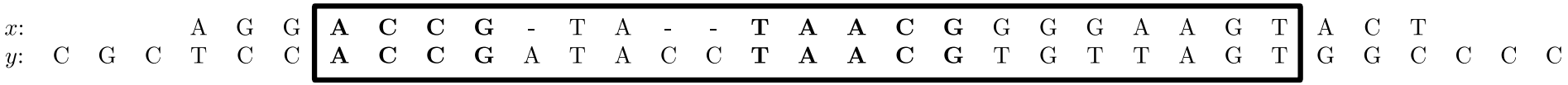
Querying a sequence DB - BLAST

3. Combination of results:
Closely located, extended hits within the same sequence are combined.



Querying a colored de Bruijn graph - PLAST

Querying a colored de Bruijn graph - PLAST

Efficient methods to query a C-DBG are yet unknown

Idea: Develop a method that queries a C-DBG in a BLAST-like manner to find local alignments between a query sequence and the sequences stored in the C-DBG.

$\rightarrow$ Pan-genome Local Alignment Search Tool – PLAST

Querying a colored de Bruijn graph - PLAST

Problem statement: Given a set of sequences Y represented as a C-DBG G and a query sequence x, find all $s \in S$ and their local alignments with x that are scored highest and above a certain threshold value.

Procedure of PLAST:

1. Seed detection
2. Seed extension
3. Combination of results

Querying a colored de Bruijn graph - PLAST

1. Seed detection:

x: AGG**ACCG**TA**TAACG**GGGAAGTACT

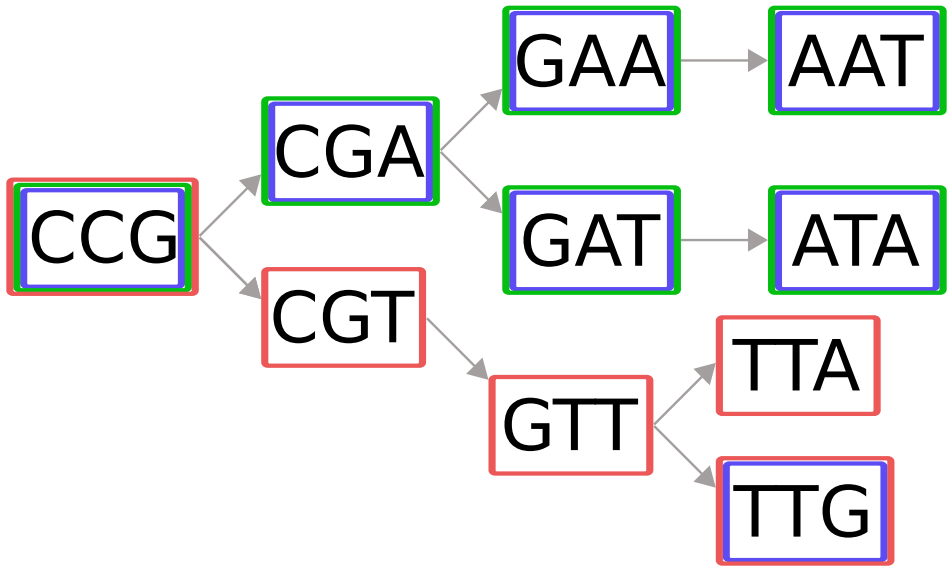
exact match length $w = 4$

Problems:

- Not efficiently possible with the BFT if $k > w$ (expected case)
- Minimizer size can be chosen arbitrarily with Bifrost, but is fixed after C-DBG creation and affects the computation time

Querying a colored de Bruijn graph - PLAST

2. Seed extension:

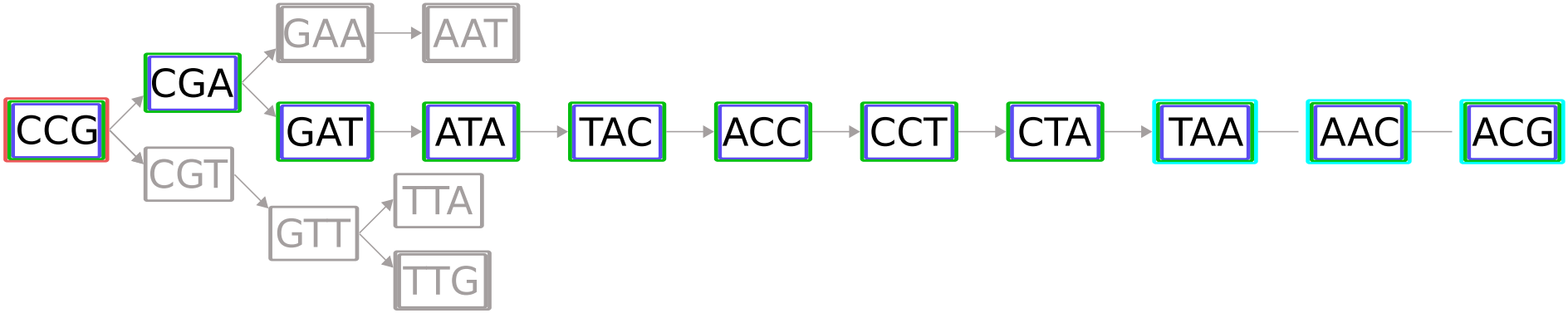


x: AGG**ACCG**TA**TAACG**GGGAAGTACT

Challenge: Smart graph traversal is crucial (especially if using the BFT)

Querying a colored de Bruijn graph - PLAST

3. Combination of results:



Same as extension step, but with gaps

Conclusion

- Current improvements in sequencing technologies necessitate the development of memory efficient ways to store sequencing data
- C-DBGs seem to be a suitable data structure to efficiently store highly similar sequences
- Fast and sensitive methods to query C-DBGs are required to allow their usage
- The development of PLAST is challenging but looks promising

End

Thank you for your attention!