Open and closed pangenomes with *k*-mer counting

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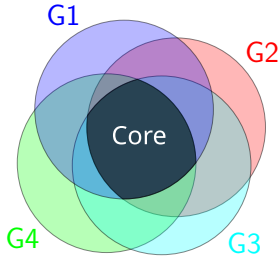
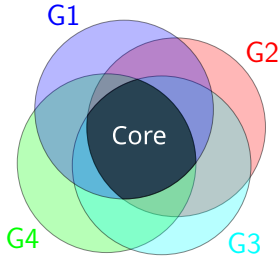
Pangenome (Tettelin et al., 2005)

- **Pangenome:** the set of all distinct genes present in a species
  - **Core genes**: present in all genomes
  - **Dispensable genes**: present in some genomes

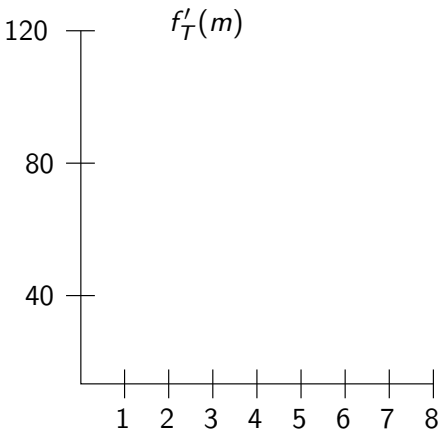


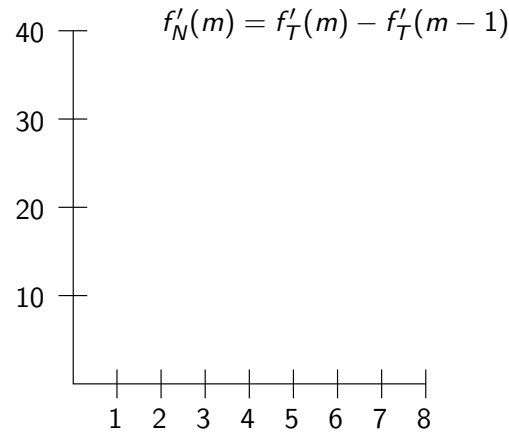
Pangenome (Tettelin et al., 2005)

- **Pangenome:** the set of all distinct genes present in a species
  - **Closed pangenome**
    The number of distinct genes is asymptotic
  - **Open pangenome**
    The number of distinct genes keeps increasing



G_1 G_2 G_3 G_4 G_5 G_6 G_7 G_8





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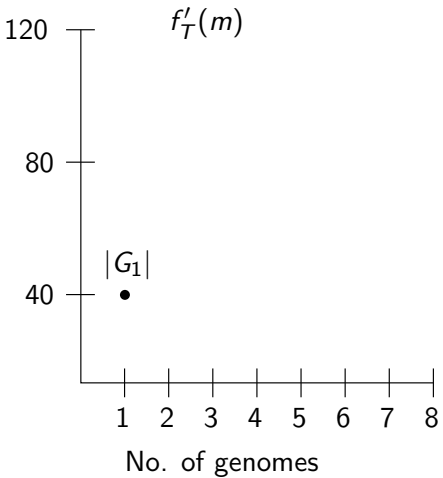

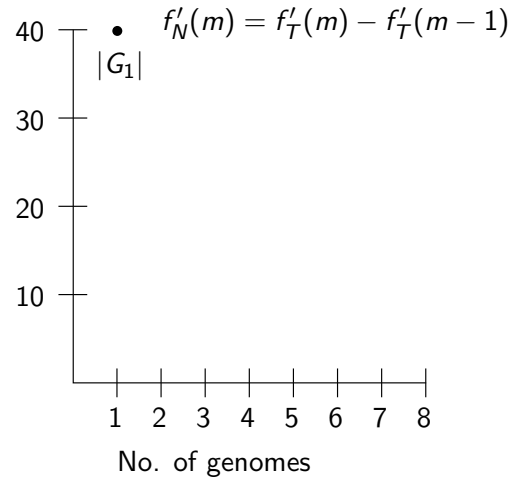

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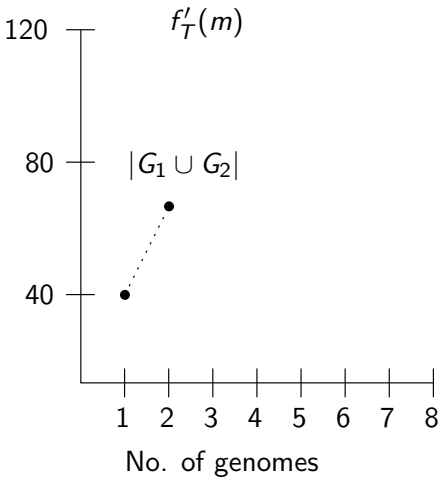

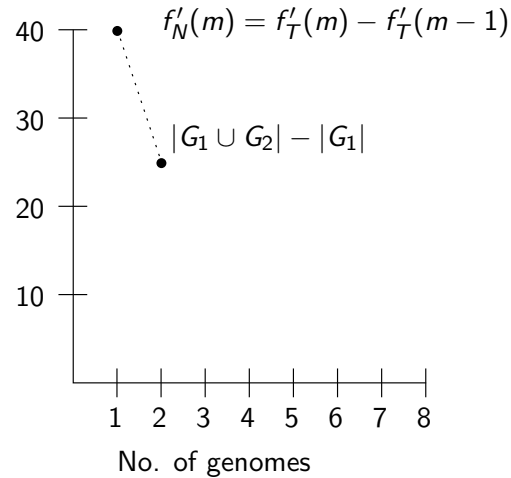

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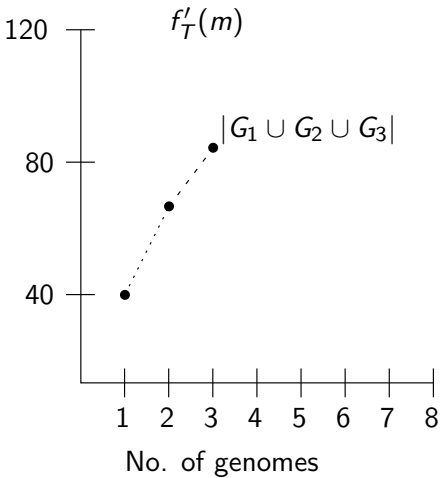

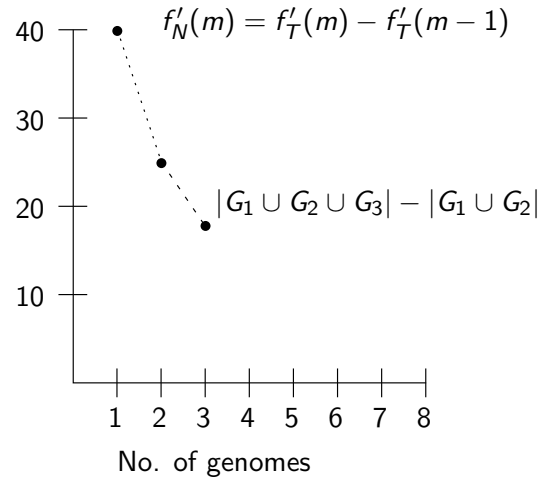

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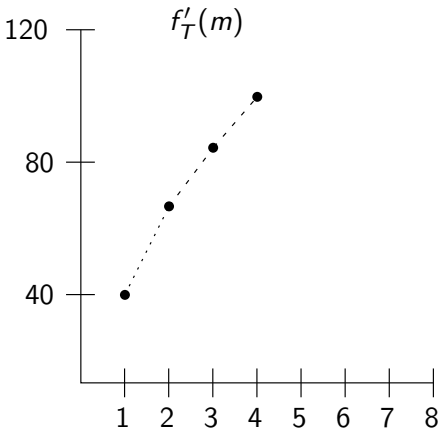

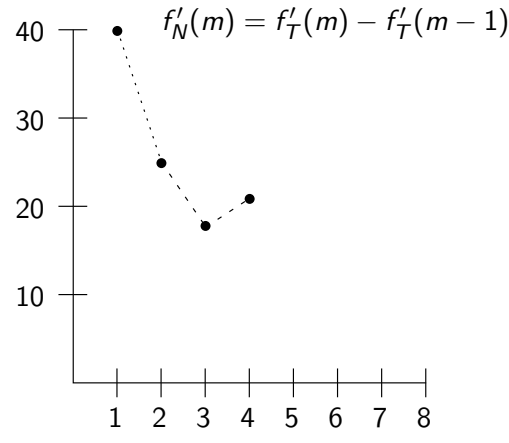

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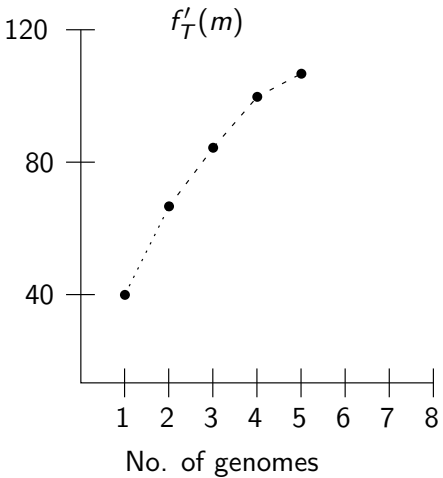

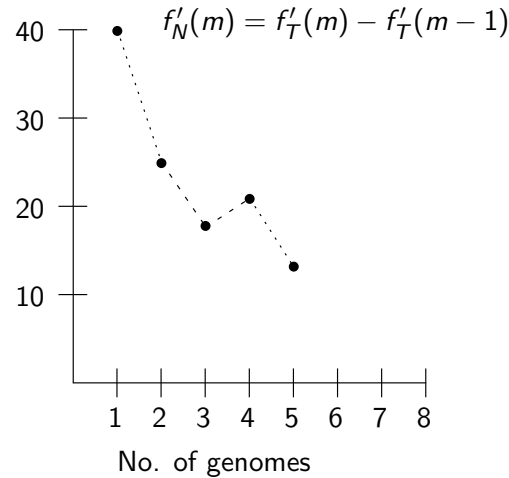

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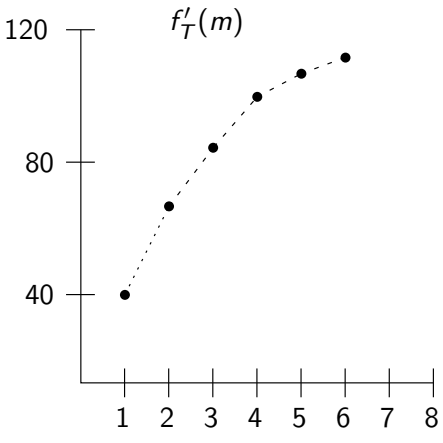

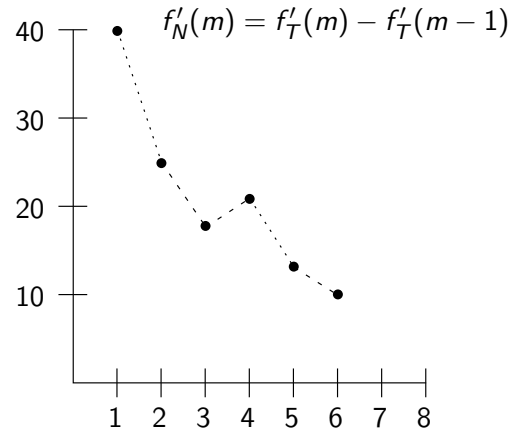

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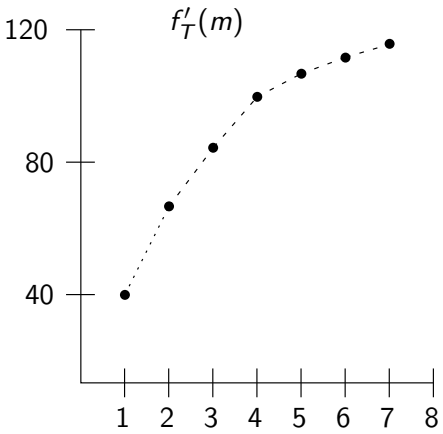

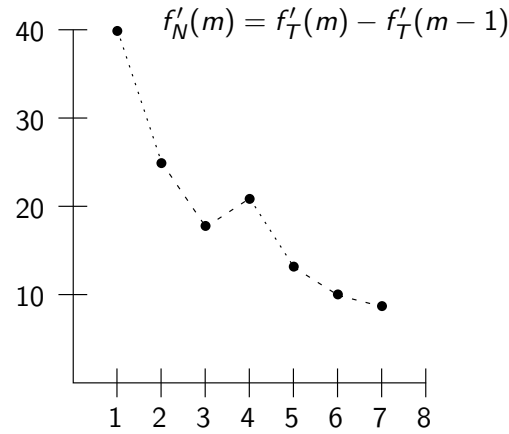

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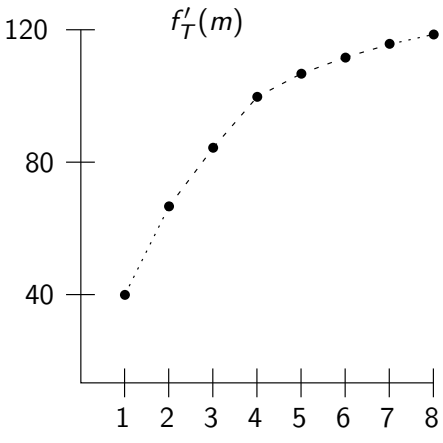

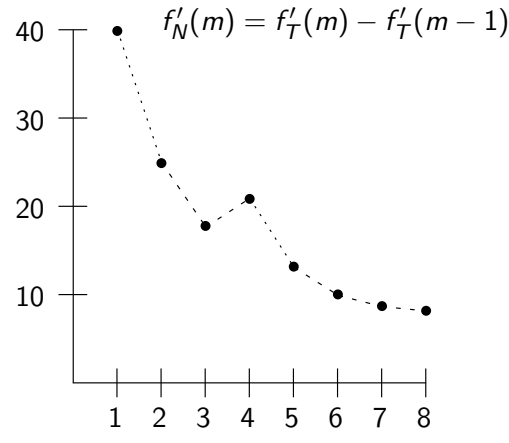

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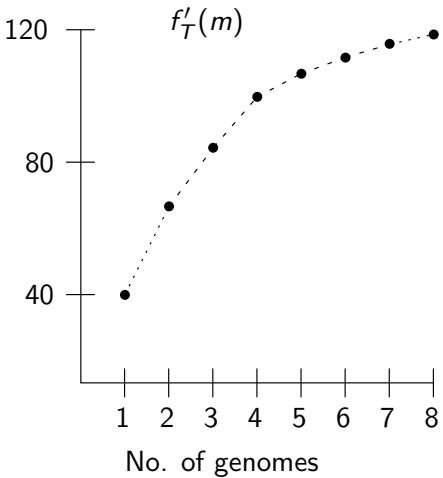

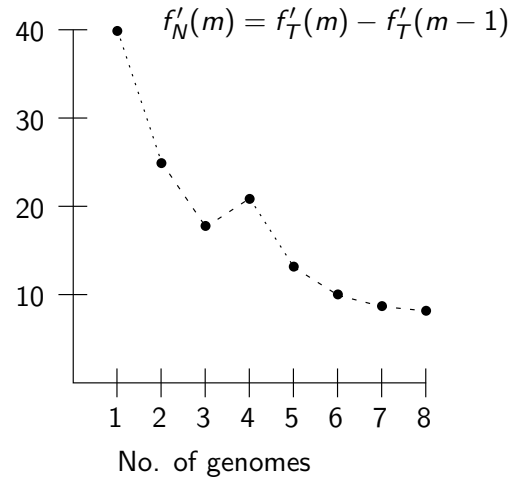

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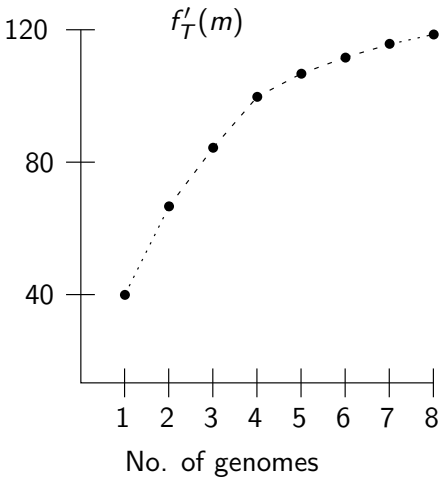

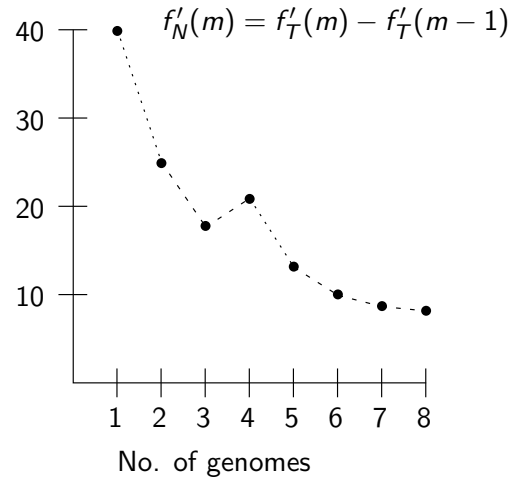

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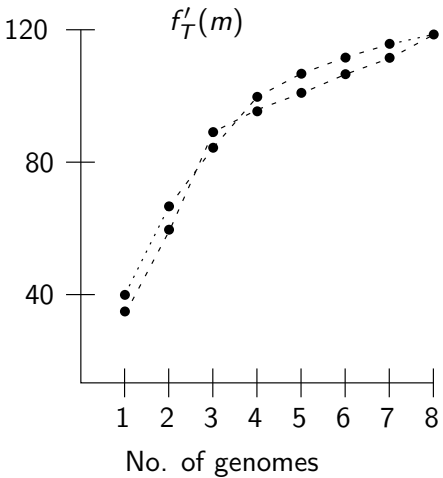

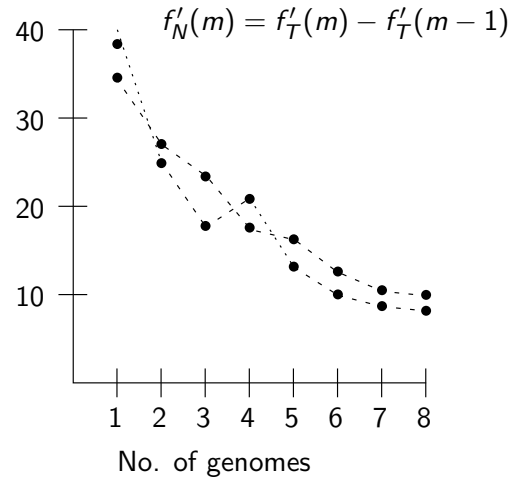

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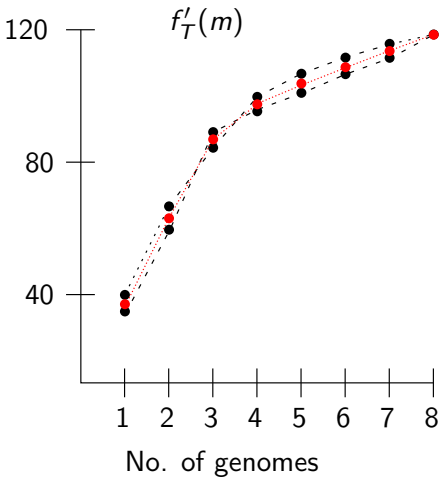

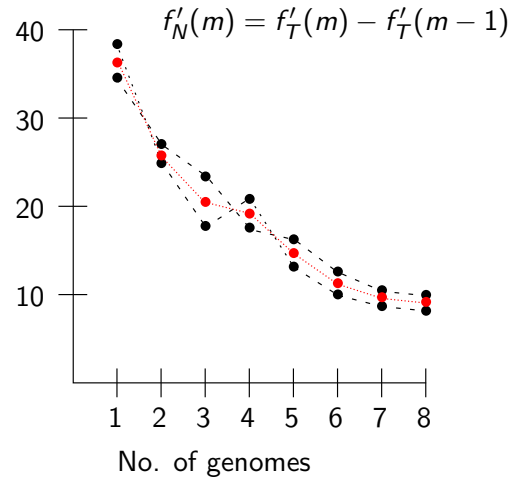

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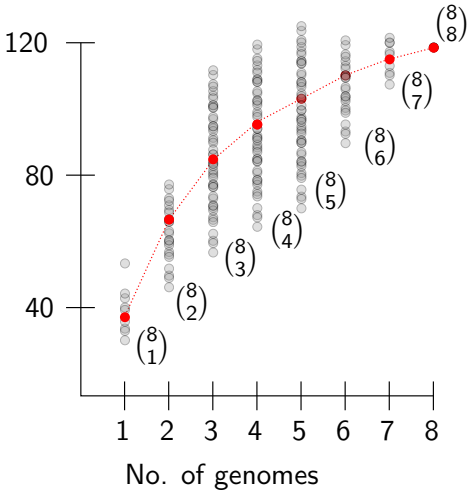
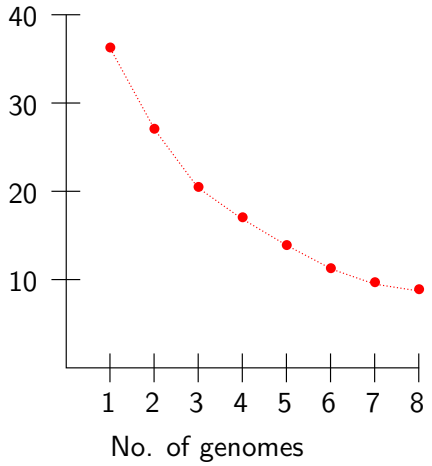

120
80
40
1
2
3
4
5
6
7
8
No. of genomes


40
30
20
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2
3
4
5
6
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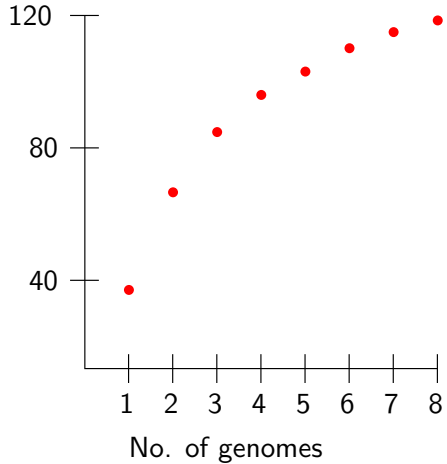
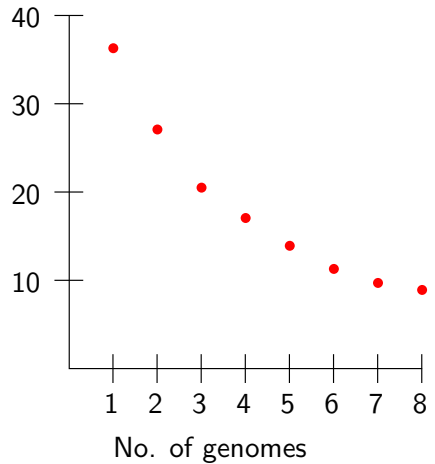
Tettelin et al., 2008 - Open closed Pangenome

Heap's law





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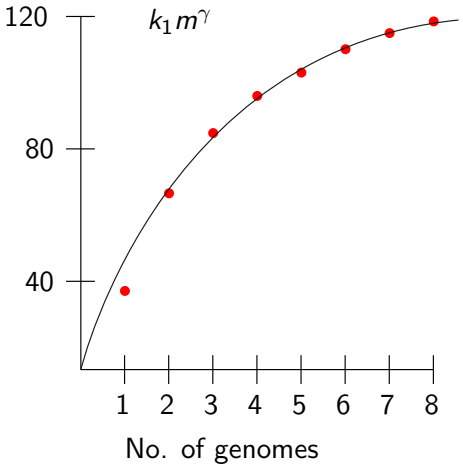

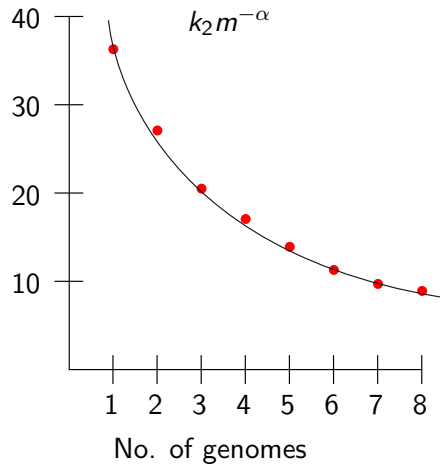

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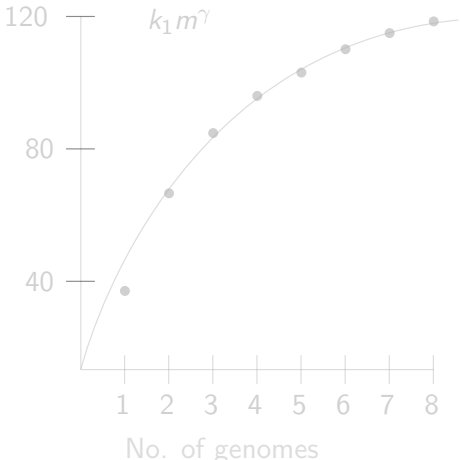

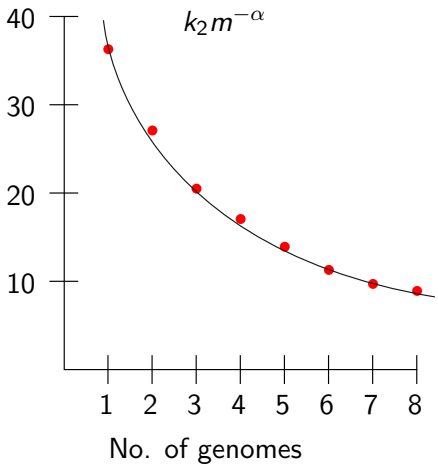

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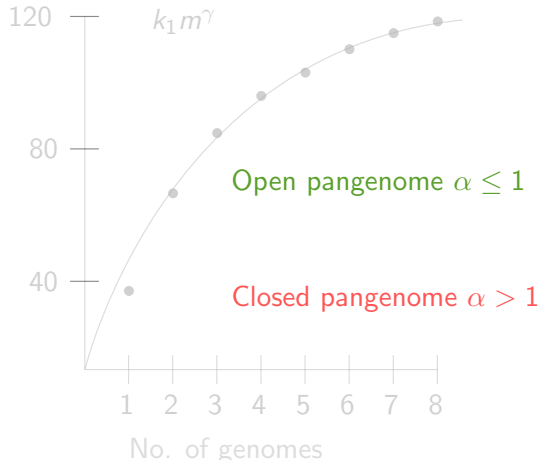

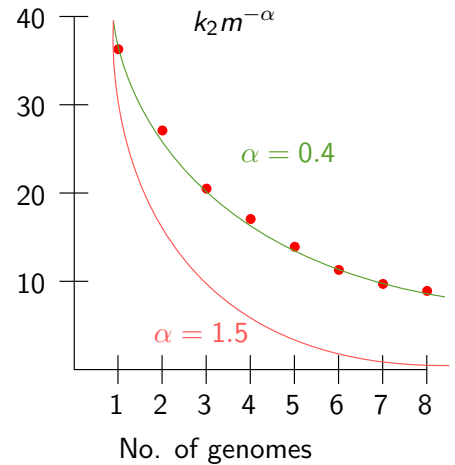

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$$\mathcal{P}(\{G_1, G_2, G_3\}) = \{\emptyset, \{G_1\}, \{G_2\}, \{G_3\}, \{G_1, G_2\}, \{G_1, G_3\}, \{G_2, G_3\}, \{G_1, G_2, G_3\}\}$$

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- $\mathcal{G}_m = \{S \in \mathcal{P}(\mathcal{G}) \mid |S| = m\}$ the set of subsets of $\mathcal{G}$ of cardinality m

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- The computation of $f_T(m)$ requires taking the average of $\binom{n}{m}$ values,

$$f_T(m) = \frac{1}{\binom{n}{m}} \sum_{S \in \mathcal{G}_m} \left| \bigcup_{G \in S} G \right|$$

$$\text{e.g., } f_T(2) = \frac{|G_1 \cup G_2| + |G_1 \cup G_3| + |G_2 \cup G_3|}{\binom{3}{2}}$$

- Pangenome growth:

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- Average number of new genes that are added when adding the m-th genome:

$$f_N(m) = \begin{cases} 0 & \text{if } m = 0 \\ f_T(m) - f_T(m-1) & \text{otherwise} \end{cases}$$

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- Choice of *k*

Obtaining f_T

Genes

- Pan-matrix

	G_1	G_2		...			G_n
gene_1	0	0	1	1	0	1	1	1
gene_2	1	1	0	1	1	0	0	0
...				...				
gene_l	0	1	0	0	1	0	0	0

- Sampling with different orderings of $\mathcal{G}$

Obtaining f_T **efficiently**

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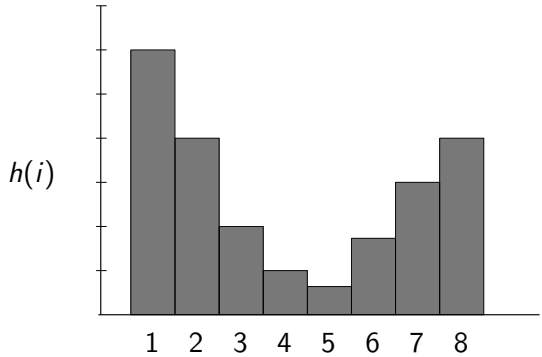
- How:

$$h(i) = \text{number of items occuring in } \textbf{exactly } i \text{ genomes}$$



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$$f_T(3) = (|G_1 \cup G_2 \cup G_3| + |G_1 \cup G_2 \cup G_4| + |G_1 \cup G_2 \cup G_5| + |G_1 \cup G_3 \cup G_4| + |G_1 \cup G_3 \cup G_5| + |G_1 \cup G_4 \cup G_5| + |G_2 \cup G_3 \cup G_4| + |G_2 \cup G_3 \cup G_5| + |G_2 \cup G_4 \cup G_5| + |G_3 \cup G_4 \cup G_5|) \frac{1}{\binom{5}{3}}$$

Obtaining f_T efficiently

- $\mathcal{G} = \{G_1, G_2, G_3, G_4, G_5\}$
- The item x is present in G_1, G_2

$$f_T(3) = (|G_1 \cup G_2 \cup G_3| + |G_1 \cup G_2 \cup G_4| + |G_1 \cup G_2 \cup G_5| + |G_1 \cup G_3 \cup G_4| + |G_1 \cup G_3 \cup G_5| + |G_1 \cup G_4 \cup G_5| + |G_2 \cup G_3 \cup G_4| + |G_2 \cup G_3 \cup G_5| + |G_2 \cup G_4 \cup G_5| + |G_3 \cup G_4 \cup G_5|) \frac{1}{\binom{5}{3}}$$

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- Percent of $S \in \mathcal{G}_m$ that **do not have item** x

$$\binom{n-i}{m} \Big/ \binom{n}{m}$$

Obtaining f_T efficiently

- $\mathcal{G} = \{G_1, G_2, G_3, G_4, G_5\}$
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- Percent of $S \in \mathcal{G}_m$ that **do not have item** x

$$\binom{n-i}{m} \Big/ \binom{n}{m} = \frac{(n-i)^{\underline{m}}}{n^{\underline{m}}}$$

$$n^{\underline{m}} = \overbrace{n(n-1)\dots(n-m+1)}^{m \text{ factors}}$$

Obtaining f_T efficiently

$$f_T(m) = \sum_{i=1}^{n} h(i)\left(1 - \frac{(n-i)^{\underline{m}}}{n^{\underline{m}}}\right)$$

Obtaining f_T efficiently

$$
\begin{aligned}
f_T(m) &= \sum_{i=1}^{n} h(i)\left(1 - \frac{(n-i)^{\underline{m}}}{n^{\underline{m}}}\right) \\
&= \sum_{i=1}^{n} h(i) - \frac{1}{n^{\underline{m}}} \sum_{i=1}^{m} (n-i)^{\underline{m}}
\end{aligned}
$$

Obtaining f_T efficiently

$$
\begin{aligned}
f_T(m) &= \sum_{i=1}^{n} h(i)\left(1 - \frac{(n-i)^{\underline{m}}}{n^{\underline{m}}}\right) \\
&= \sum_{i=1}^{n} h(i) - \frac{1}{n^{\underline{m}}} \sum_{i=1}^{m} (n-i)^{\underline{m}}
\end{aligned}
$$

- $(n-i)^{\underline{j+1}} = (n-i-j+1)(n-i)^{\underline{j}}$

Obtaining f_T efficiently

$$
\begin{aligned}
f_T(m) &= \sum_{i=1}^{n} h(i)\left(1 - \frac{(n-i)^{\underline{m}}}{n^{\underline{m}}}\right) \\
&= \sum_{i=1}^{n} h(i) - \frac{1}{n^{\underline{m}}} \sum_{i=1}^{m} (n-i)^{\underline{m}}
\end{aligned}
$$

- $(n-i)^{\underline{j+1}} = (n-i-j+1)(n-i)^{\underline{j}}$
- Time complexity: $O(n^2)$

How to obtain $h(i)$

Genes

Pan-matrix

	G_1	G_2			…			G_n
gene_1	0	0	1	1	0	1	1	1
gene_2	1	1	0	1	1	0	0	0
…					…			
gene_l	0	1	0	0	1	0	0	0

How to obtain $h(i)$

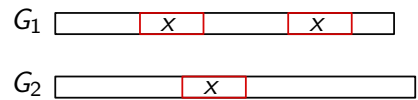
Genes

Pan-matrix

	G_1	G_2		...		G_n
gene_1	0	0	1	1	0	1	1	1
gene_2	1	1	0	1	1	0	0	0
...				...				
gene_l	0	1	0	0	1	0	0	0

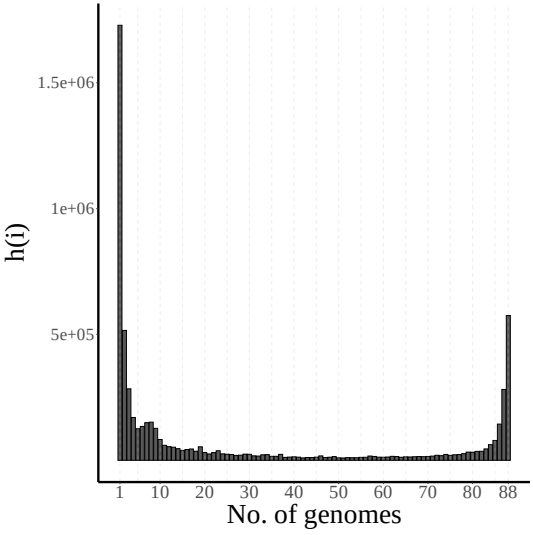
k-mers

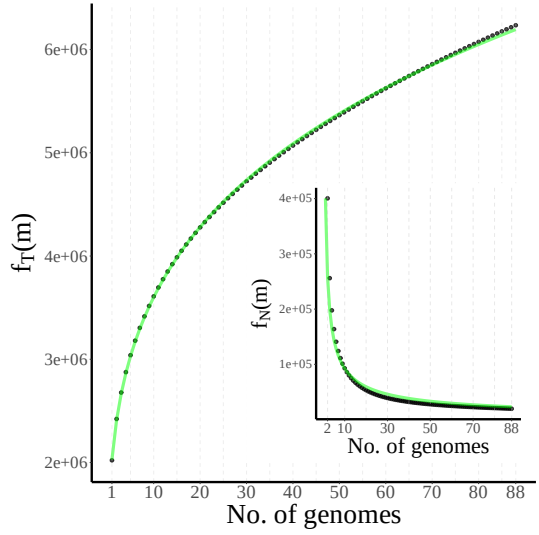
Modified version of YAK[1](yak-hist)



- k-mer x has multiplicity 2

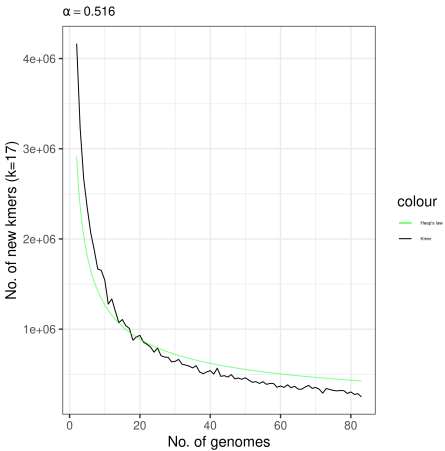
[1]https://github.com/lh3/yak


h(i)
1.5e+06
1e+06
5e+05
1
10
20
30
40
50
60
70
80
88
No. of genomes

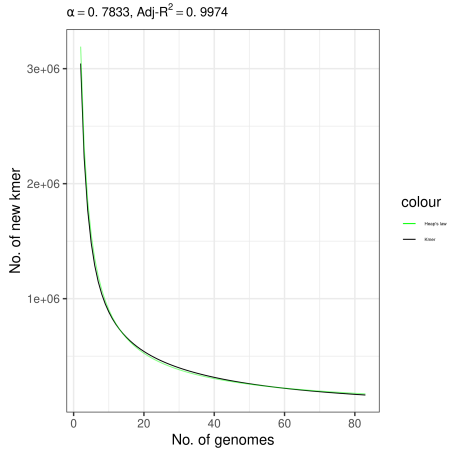

$f_T(m)$
6e+06
5e+06
4e+06
3e+06
2e+06
1
10
20
30
40
50
60
70
80
88
No. of genomes
$f_N(m)$
4e+05
3e+05
2e+05
1e+05
2
10
30
50
70
88
No. of genomes

Results

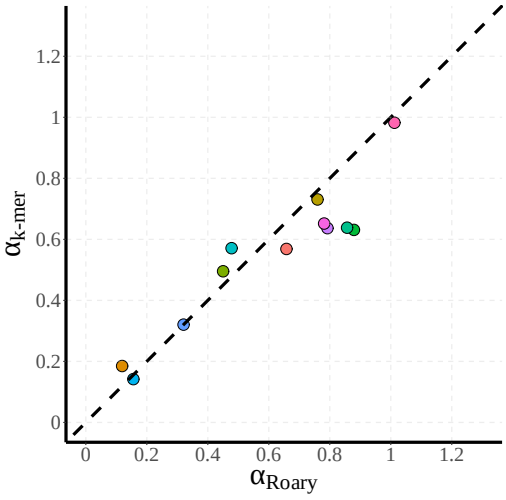
Permutation



Without permutation



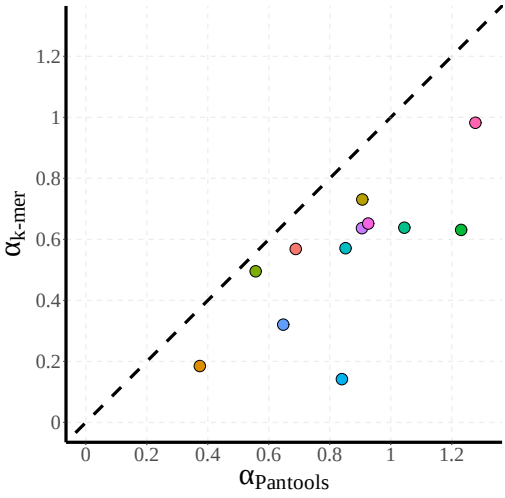
Results

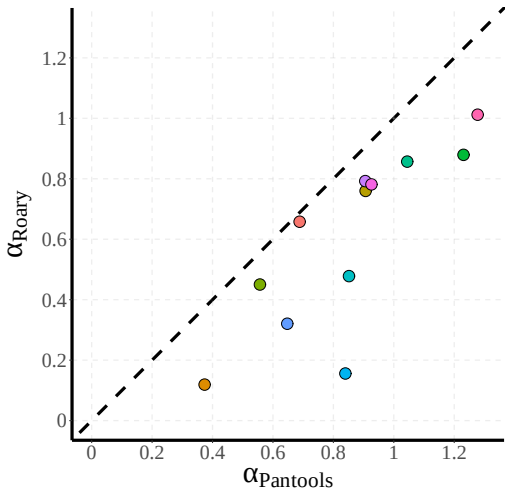


species

- Bacillus_cereus
- Buchnera_aphidicola
- Campylobacter_jejuni
- Clostridium_botulinum
- Coxiella_burnetii
- Francisella_tularensis
- Helicobacter_pylori
- Prochlorococcus_marinus
- Rhodopseudomonas_palustris
- Streptococcus_pneumoniae
- Streptococcus_pyogenes
- Yersinia_pestis

Results


1.2
1
0.8
0.6
0.4
0.2
0
0
0.2
0.4
0.6
0.8
1
1.2
$\alpha_{\text{k-mer}}$
α_{Pantools}

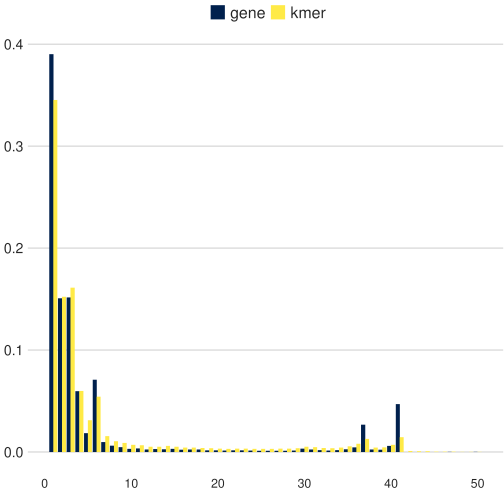

1.2
1
0.8
0.6
0.4
0.2
0
0
0.2
0.4
0.6
0.8
1
1.2
α_{Roary}
α_{Pantools}

Normalized Kendal Tau distance

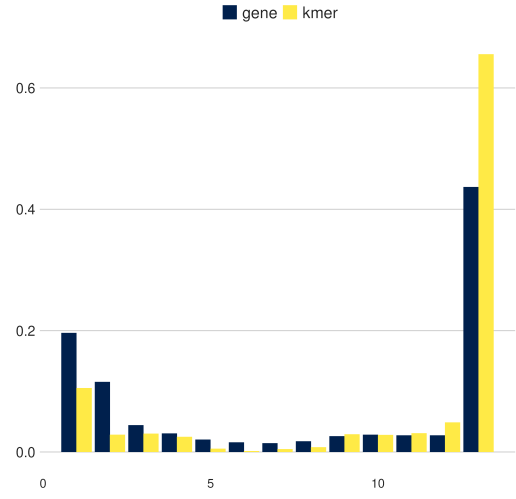
Counts the number of **pairwise disagreements** between two ranking lists divided by the total number of pairwise comparison, $n(n-1)/2$.

- Roary vs. k-mer: 0.167
- Roary vs. Pantools: 0.106
- k-mer vs. Pantools: 0.182

Histrogram, $h(i)$ of genes (Roary) and k-mers



Clostridium botulinum



Coxiella burnetii

Jensen-Shannon divergence

Species	**JSD**
Francisella tularensis	0.065
Coxiella burnetii	0.061
Yersinia pestis	0.11
Streptococcus pneumoniae	0.019
Helicobacter pylori	0.048
Clostridium botulinum	0.023
Streptococcus pyogenes	0.042
Prochlorococcus marinus	0.0048
Campylobacter jejuni	0.054
Buchnera aphidicola	0.023
Bacillus cereus	0.026
Escherichia coli	0.014

Core

$$f_C(m) = \frac{1}{\binom{n}{m}} \sum_{S \in \mathcal{G}_m} \left| \bigcap_{G \in S} G \right|$$

Compute f_C efficiently

$$f_C(m) = \frac{1}{n^{\underline{m}}} \sum_{i=m}^{n} h(i) i^{\underline{m}}$$

Quorum, q

$$f_C(m,q) = \frac{1}{\binom{n}{m}} \sum_{\lceil q*m \rceil}^{n} h(i) \sum_{j=\lceil q*m \rceil}^{i} \frac{\binom{i}{j}}{\binom{n-i}{m-j}}$$

Thank you for your attention