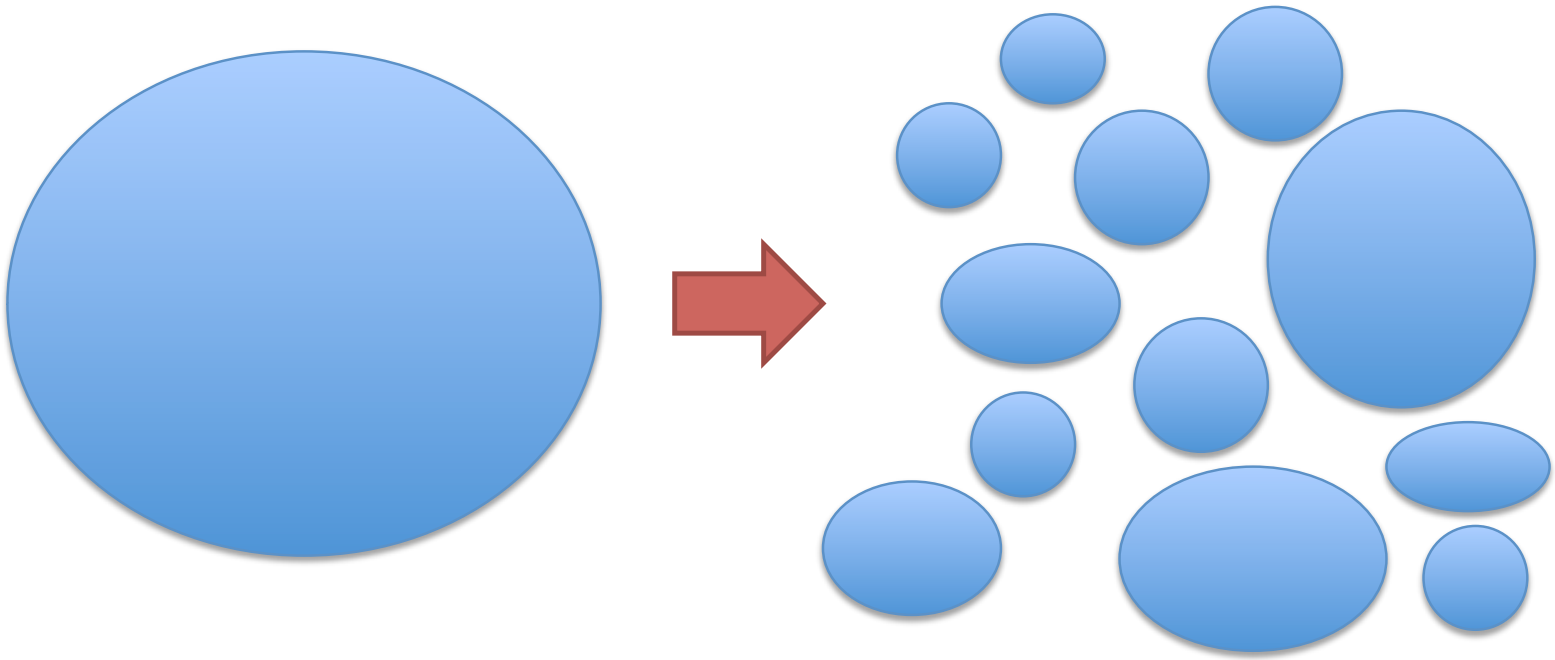
A suffix-array based algorithm for expression clustering

Scott Hazelhurst University of the Witwatersrand, Johannesburg (South Africa)

Zsuzsanna Lipták University of Salerno/University of Verona (Italy)

WAABD, Pisa, 21 Oct. 2011

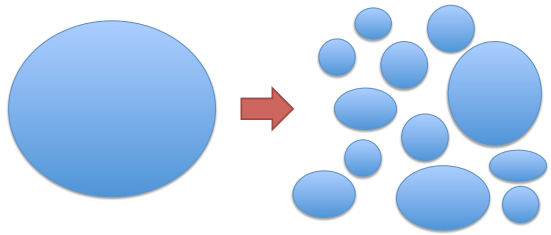
What is the problem?



Given:
Set of n sequences (10^5 or more)
of length m

Wanted:
clusters s.t. each cluster
corresponds to a gene

Using pairwise comparison



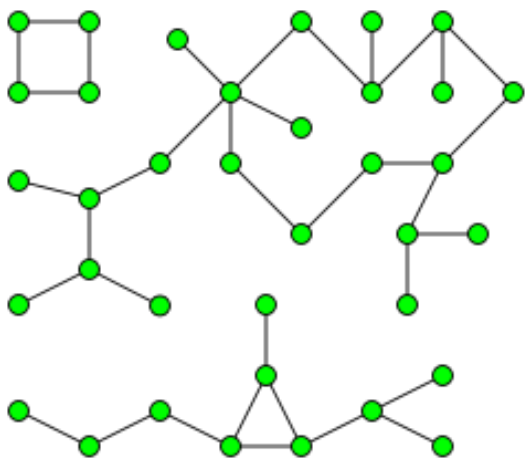
n = # seq's, m = ave. length

Single linkage

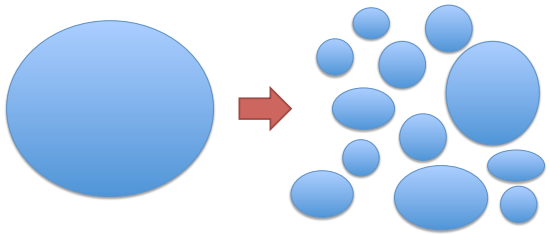
s, t similar $\Rightarrow$ s, t in same cluster

As a graph problem

Find connected components.
Remember: Edges have to be computed!



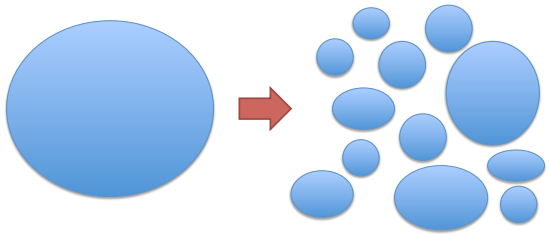
Using pairwise comparison



$n = \#$ seq's, $m =$ ave. length

- One comparison (e.g. edit dist.) costs m^2 $\rightsquigarrow$ $O(n^2 m^2)$ time

Using pairwise comparison



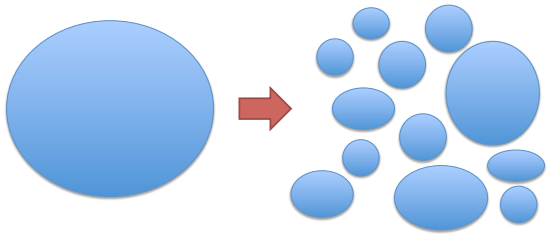
$n = \#$ seq's, $m =$ ave. length

- One comparison (e.g. edit dist.) costs m^2 $\rightsquigarrow$ $O(n^2 m^2)$ time

  Most pairs are not similar: use filters e.g. $O(m)$ time

  if x, y pass filter then compute $d(x, y)$

Using pairwise comparison



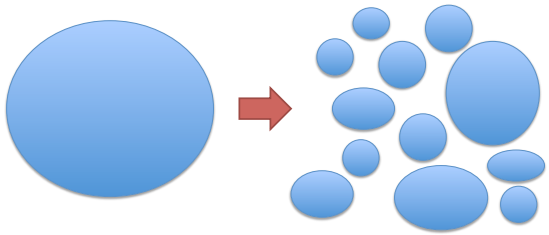
$n = \#$ seq's, $m =$ ave. length

- One comparison (e.g. edit dist.) costs m^2 $\rightsquigarrow$ $O(n^2 m^2)$ time

  Most pairs are not similar: use filters e.g. $O(m)$ time

  if x, y pass filter then compute $d(x, y)$
- If one filter test costs m $\rightsquigarrow$ $O(n^2 m)$ time + sg.

Using pairwise comparison



$n = \#$ seq's, $m =$ ave. length

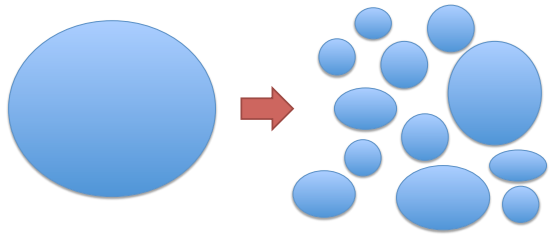
- One comparison (e.g. edit dist.) costs m^2 $\rightsquigarrow$ $O(n^2 m^2)$ time

  Most pairs are not similar: use filters e.g. $O(m)$ time

  if x, y pass filter then compute $d(x, y)$
- If one filter test costs m $\rightsquigarrow$ $O(n^2 m)$ time + sg.
- In general:

  If one filter test costs $f(m)$ $\rightsquigarrow$ $O(n^2 f(m))$ time + sg.

Using pairwise comparison



$n = \#$ seq's, $m =$ ave. length

- One comparison (e.g. edit dist.) costs m^2 $\rightsquigarrow$ $O(n^2 m^2)$ time

  Most pairs are not similar: use filters e.g. $O(m)$ time

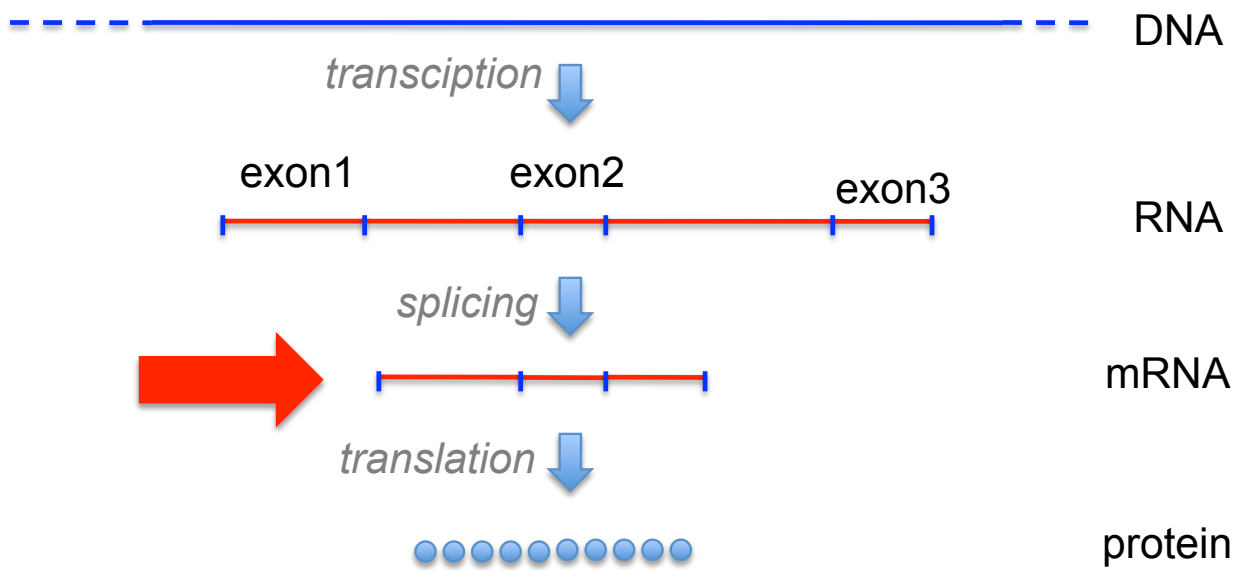
  if x, y pass filter then compute $d(x, y)$
- If one filter test costs m $\rightsquigarrow$ $O(n^2 m)$ time + sg.
- In general:

  If one filter test costs $f(m)$ $\rightsquigarrow$ $O(n^2 f(m))$ time + sg.

Here: We get rid of the n^2 factor!

Clustering expression data

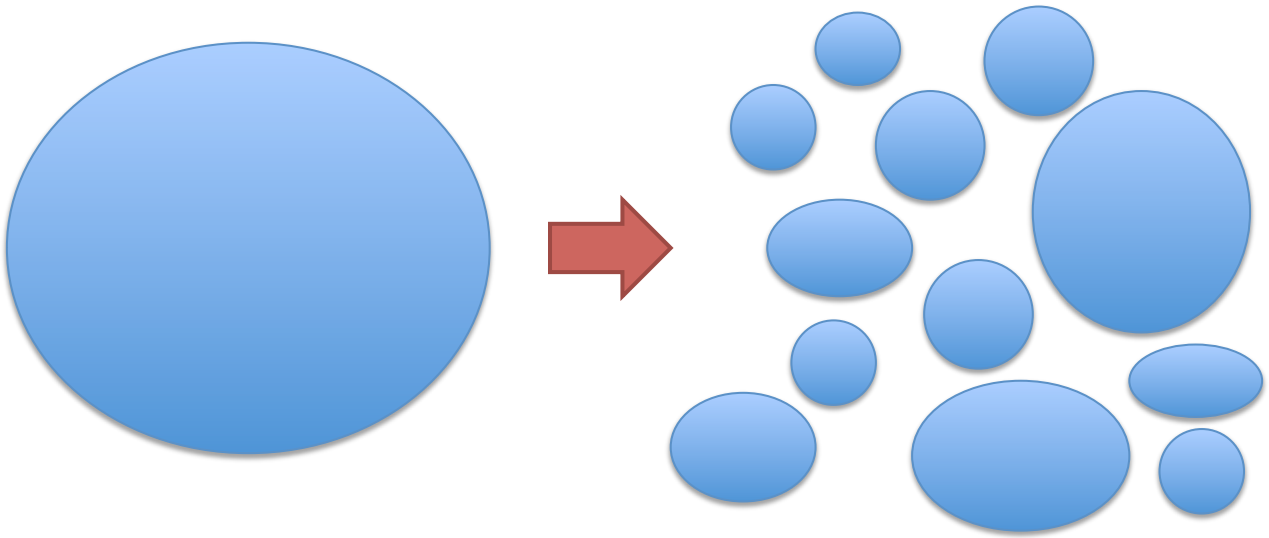
Expression data (formerly ESTs)



Transcriptome/Expression data

(ESTs = expressed sequence tags)
Partial cDNA copies of mRNAs (lab process)
both Sanger-style (ESTs) and NGS

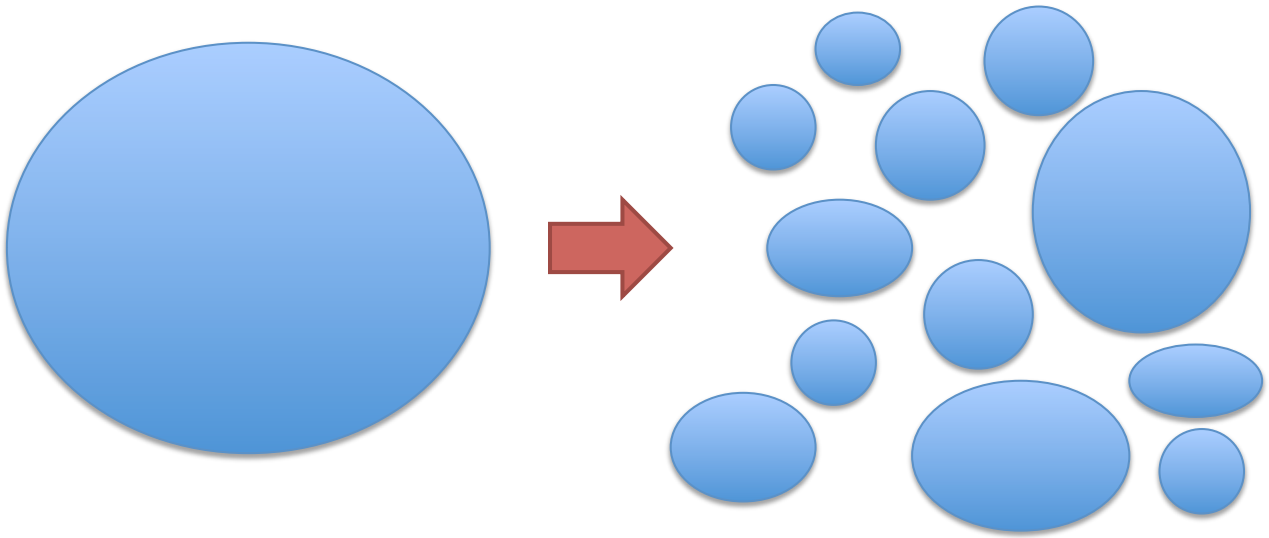
Problem statement



Desideratum

Given a set of sequences S, find a partition of S s.t. if s, t are products of the same gene, then s, t in same cluster.

Problem statement



Desideratum

Given a set of sequences S, find a partition of S s.t. if s, t are products of the same gene, then s, t in same cluster.

Formal Problem

Given a set of sequences S, find a partition of S s.t. if s, t are similar, then s, t in same cluster.

Expression clustering

- expression studies (e.g. diseased vs. healthy),
- gene discovery,
- SNP detection,
- discovery of products of alternative splicing,
- estimating no. of genes

We are not doing:

- consensus building
- alignment to genome

String similarity

Example

s = CAAGACAA, t = CAGAGCAC

- alignment / edit distance (N/W 1970, S/W 1981)
- q-gram-distance: $\sum_{|w|=q} |\text{freq(w,s) - freq(w,t)}|$ (Ukkonen 1992)
- d^2: $\sum_{|w|=q} (\text{freq(w,s) - freq(w,t)})^2$ (Torney et al. 1990)
- others (fingerprints, information theory based, . . .)

String similarity

Example

s = CAAGACAA, t = CAGAGCAC

- alignment / edit distance = 3 (N/W 1970, S/W 1981)
- q-gram-distance: $\sum_{|w|=q}$ |freq(w,s) - freq(w,t)| (Ukkonen 1992)
- d^2: $\sum_{|w|=q}$(freq(w,s) - freq(w,t))^2 (Torney et al. 1990)
- others (fingerprints, information theory based, . . .)

CAAGA–CAA
CA–GAGCAC

String similarity

Example

s = CAAGACAA, t = CAGAGCAC

- alignment / edit distance = 3 (N/W 1970, S/W 1981)
- q-gram-distance: $\sum_{|w|=q} |\text{freq(w,s) - freq(w,t)}|$ (Ukkonen 1992)
- d^2: $\sum_{|w|=q} (\text{freq(w,s) - freq(w,t)})^2$ (Torney et al. 1990)
- others (fingerprints, information theory based, . . .)

CAAGA–CAA
CA–GAGCAC

AA	2 0
AC	1 1
AG	1 2
CA	2 2
GA	1 1
GC	0 1

String similarity

Example

s = CAAGACAA, t = CAGAGCAC

- alignment / edit distance = 3 (N/W 1970, S/W 1981)
- q-gram-distance: $\sum_{|w|=q} |\text{freq(w,s) - freq(w,t)}| = 4$ (Ukkonen 1992)
- d^2: $\sum_{|w|=q} (\text{freq(w,s) - freq(w,t)})^2$ (Torney et al. 1990)
- others (fingerprints, information theory based, . . .)

CAAGA–CAA
CA–GAGCAC

AA	2	0	2
AC	1	1	0
AG	1	2	1
CA	2	2	0
GA	1	1	0
GC	0	1	1
			4

String similarity

Example

s = CAAGACAA, t = CAGAGCAC

- alignment / edit distance = 3 (N/W 1970, S/W 1981)
- q-gram-distance: $\sum_{|w|=q} |\text{freq(w,s) - freq(w,t)}| = 4$ (Ukkonen 1992)
- d^2: $\sum_{|w|=q} (\text{freq(w,s) - freq(w,t)})^2 = 6$ (Torney et al. 1990)
- others (fingerprints, information theory based, . . .)

CAAGA–CAA
CA–GAGCAC

AA	2	0	2	4
AC	1	1	0	0
AG	1	2	1	1
CA	2	2	0	0
GA	1	1	0	0
GC	0	1	1	1
			4	6

Similar windows

```
>1
tttattattatgggattaggcttctctctcgcgattctcttacgtt
ggttatatctttatattacttttttcttattgtataattagcttccg
tcctctaggtcccttctcta
>2
cccctctctgttggttatatctttatattacttttttcttattgtataa
cttcttattattctccgctctctctcttatcttccggaattctcgagg
ggatatatattatgcggcgccctcttatatctctatattctctc
a
```

Similar windows

```
>1
tttattattatgggattaggcttctctctcgcgattctcttacGTT
CGTTATATCTTTATATTACTTTTTCTTATTGTATAAttagcttccg
tcctctaggtcccttctcta
>2
cccctctctGTTGGTTATATCTTTATATTACTTCTTCTTATTGTATAA
cttcttattattctccgctctctctcttatcttccggaattctcgagg
ggatatatattatgcggcgccctcttatatctctatattctctc
a
```

Our string similarity measure

We use d^2, minimized over all pairs of windows of given size.

$$d(s,t) = \min_{s' \sqsubseteq s, t' \sqsubseteq t, |s|=|t|=\mu} \sum_{|w|=q} (\text{freq}(w,s') - \text{freq}(w,t'))^2$$

Parameters:

wordsize q $(= 6, 8)$, windowsize, threshold (s, t similar iff $d(s,t) < \theta$)

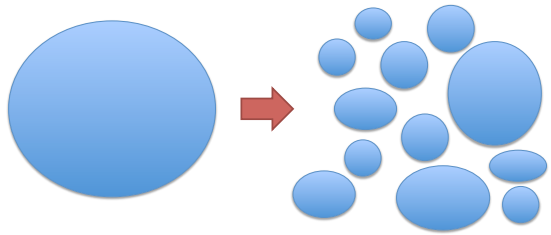
Computation time: $O(m^2)$

Speedup of previous tool

wcd + KABOOM! = wcd-express

The KABOOM-filter

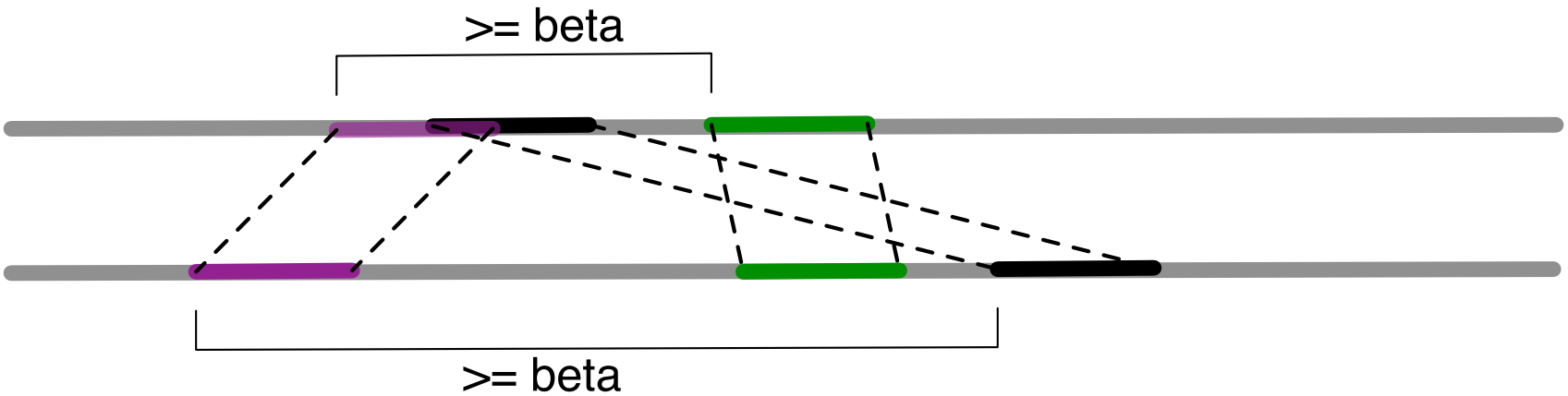
Filters for d^2



n = # seq's, m = ave. length

- Recall: we want to avoid $O(n^2m^2)$ runtime
- Filters with guarantee that need pairwise comparison only reduce m^2 factor
- One long exact match can be implemented with suffix array (linear in nm) – but too fragile!
- KABOOM-filter combines good running time with good filtering qualities

The KABOOM-filter

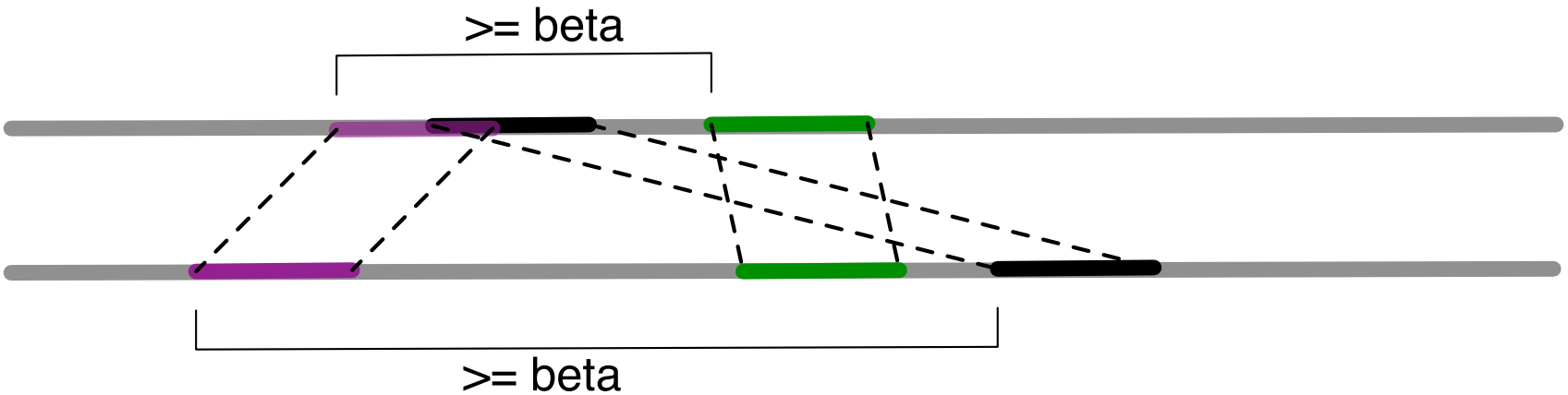


Definition

s, t are (k, α, β)-similar if they have at least α many k-words in common, at least β apart.

- k-words are counted with multiplicities (a.k.a. D^2)
- distance β between leftmost and rightmost occurrence, in both strings
- order of matches not important

The KABOOM-filter



Definition

s, t are (k, α, β)-similar if they have at least α many k-words in common, at least β apart.

- heuristic filter for d^2
- very good in practice: produces practically no FNs and reduces no. of d^2 comparisons dramatically
- $k > q$ (typically k between 12 and 20)

Implementation of the KABOOM-filter

for every sequence s_i
  for every k-word w in s_i
    find all s_j containing w
    $M = M \cup s_j$ (M: candidate sequences for s_i)
    update variables for s_j
  for every s_j in M
    check whether s_i, s_j fulfill (k, α, β)-criterion
    if yes, compute $d(s_i, s_j)$

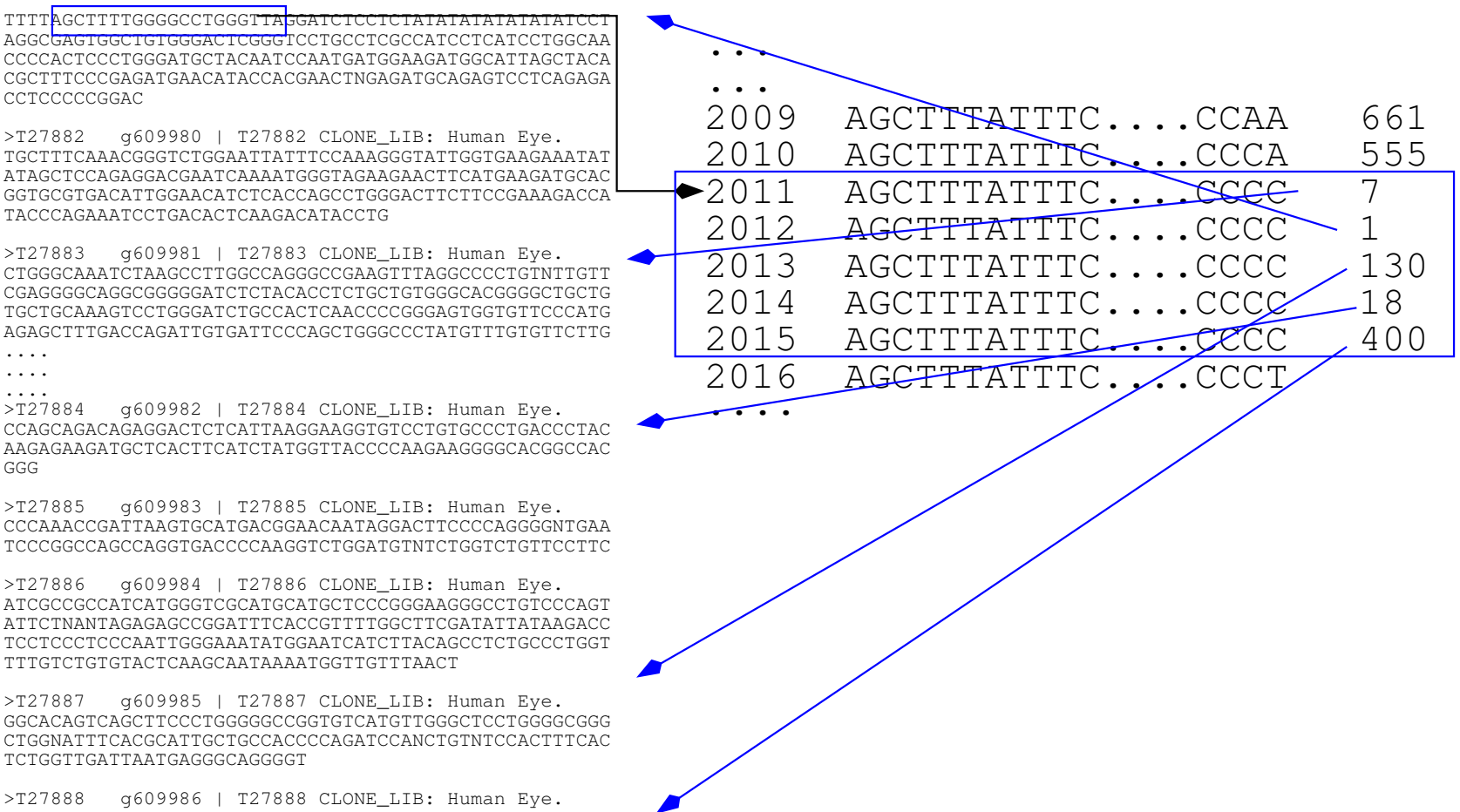
Implementation of the KABOOM-filter

for every sequence s_i
  for every k-word w in s_i
    find all s_j containing w
    $M = M \cup s_j$ (M: candidate sequences for s_i)
    update variables for s_j
  for every s_j in M
    check whether s_i, s_j fulfill (k, α, β)-criterion
    if yes, compute $d(s_i, s_j)$

We need

- suffix array of concatenated string
- inverse of suffix array

Implementation of the KABOOM-filter



Modified suffix array sa_k

aaa@aacggt@gttaaagt@tcggt@gttat@cgg@acggt@

i	sa	Text from sa[i]	sa_k
15	16	agt@tcggt@gttat@cgg@acggt@	16
16	29	at@cgg@acggt@	29
17	32	cgg@acggt@	6
18	37	cggt@	21
19	6	cggt@gttaaagt@t...gt@	32
20	21	cggt@gttat@cgg@acggt@	37
21	34	g@acggt@	34
22	33	gg@acggt@	33
23	38	ggt@	7
24	7	ggt@gttaaagt@t...t@	22
25	22	ggt@gttat@cgg@acggt@	38
26	39	gt@	8

Modified suffix array sa_k

aaa@aacggt@gttaaagt@tcggt@gttat@cgg@acggt@

i	sa	Text from sa[i]	sa_k
15	16	agt@tcggt@gttat@cgg@acggt@	16
16	29	at@cgg@acggt@	29
17	32	cgg@acggt@	6
18	37	cggt@	21
19	6	cggt@gttaaagt@t...gt@	32
20	21	cggt@gttat@cgg@acggt@	37
21	34	g@acggt@	34
22	33	gg@acggt@	33
23	38	ggt@	7
24	7	ggt@gttaaagt@t...t@	22
25	22	ggt@gttat@cgg@acggt@	38
26	39	gt@	8

Analysis

Time

- $\sum_{|w|=k} \text{freq}(w)^2$, which is between nm and n^2m^2
- in practice: very fast
- WE work efficiency: 1-(TP + FP)/(all pairs) above 99.5%

Space

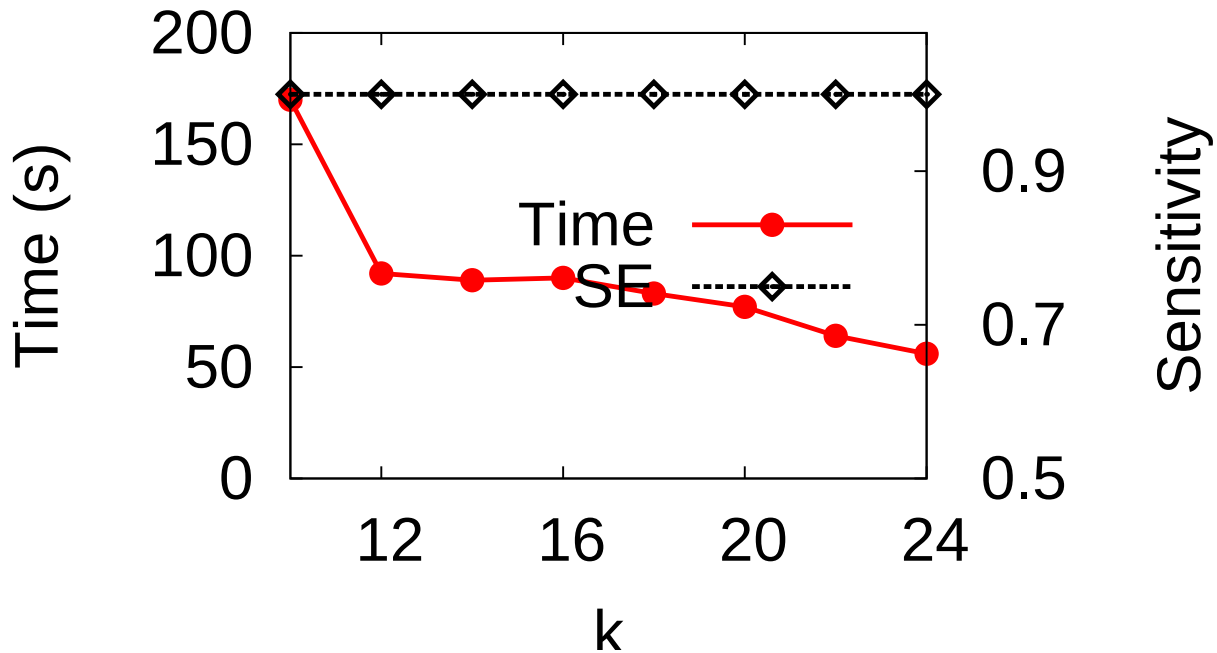
- text nm (nt)
- k-depth suffix array: $nm \log n$ (x2 for RC)
- inverse: $nm \log n$

Experimental results

Speedup w.r.t. wcd

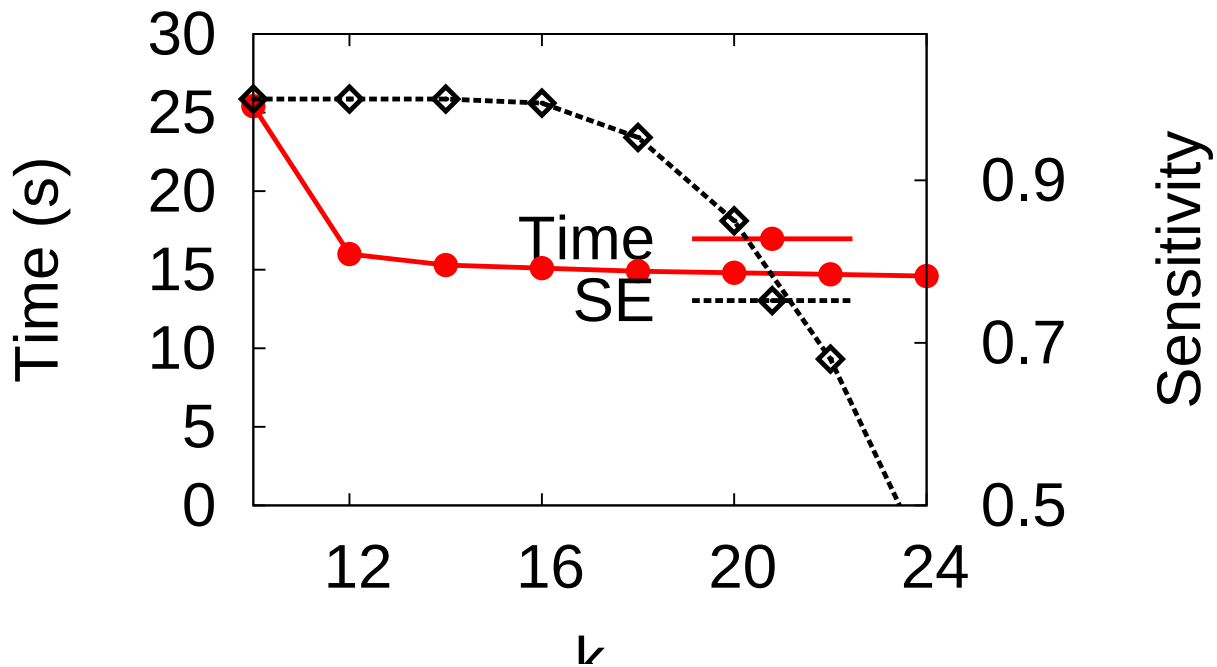
Data set	# seqs (K)	size Mb	γE γE	**wcd-exp (s)**	**wcd (s)**	Speed-up factor
A076941	77	32	17	**100**	**578**	5.7
A208	484	208	102	**1240**	**23983**	19.3
C10	126	56	355	**511**	**4512**	8.8
chlamy	190	100	139	**1000**	**5989**	6.0
Drosophila	25	86	68	**184**	**1542**	8.4
ecHuman	17	11	171	**135**	**496**	3.7
pubcot	30	17	34	**65**	**222**	3.4
ricinus	58	40	162	**840**	**1518**	1.8
xen	233	137	63	**855**	**9298**	10.8

Sensitivity to choice of k



Filtered cotton set, $\alpha = 3, \beta = 32, H = 48$.

Sensitivity to choice of k



Synthetic 454 data set, $\alpha = 3, \beta = 16, H = 32$.

Comparison with other tools

Issues

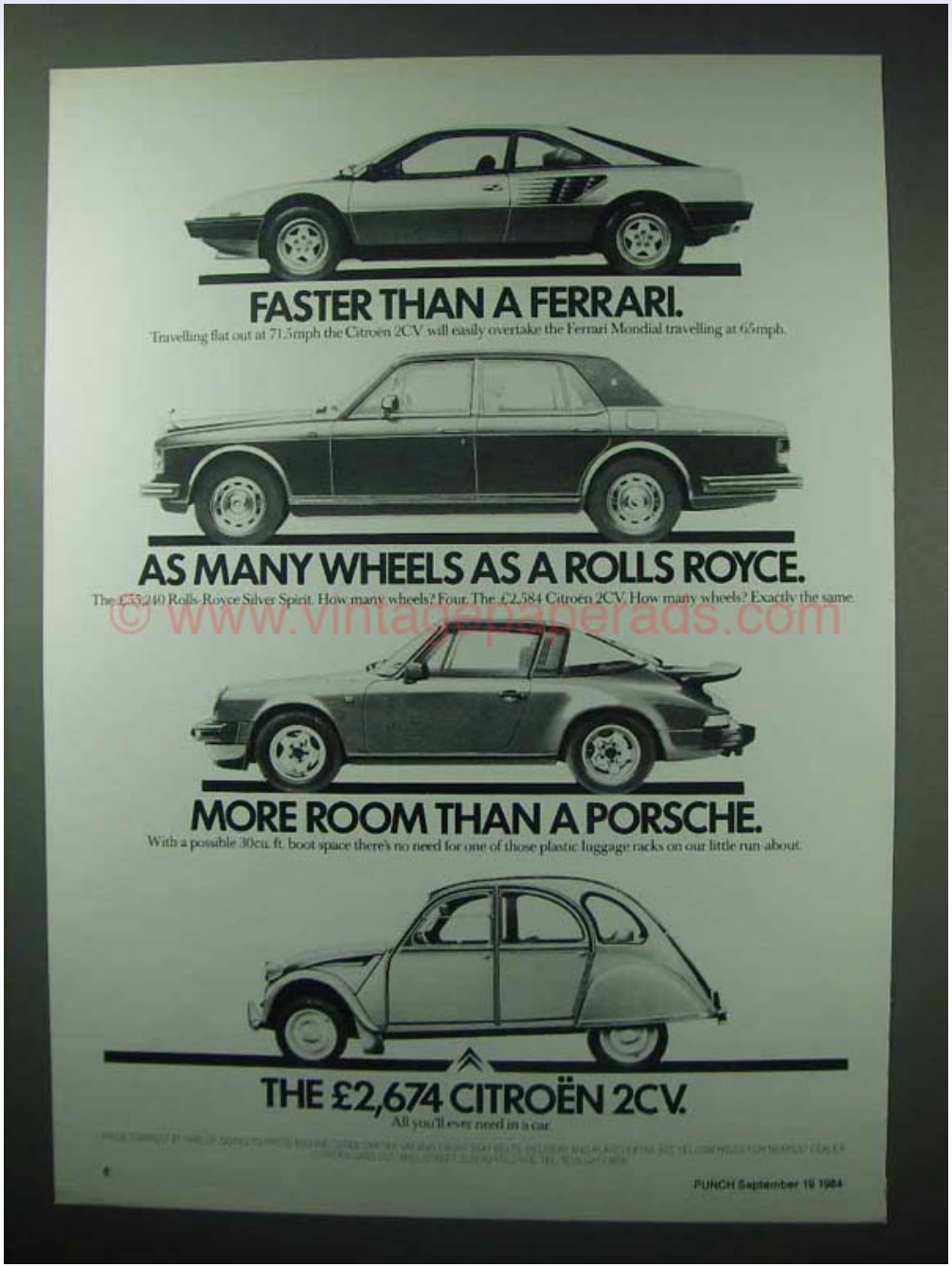
- choice of parameters
- pre-/postprocessing (here: filtering)
- quality: what's the truth?
- synthetic vs. real data

Comparison with other tools

Issues

- choice of parameters
- pre-/postprocessing (here: filtering)
- quality: what's the truth?
- synthetic vs. real data

Citroën/Ferrari comparison problem


FASTER THAN A FERRARI.
Travelling flat out at 71.5mph the Citroën 2CV will easily overtake the Ferrari Mondial travelling at 65mph.
AS MANY WHEELS AS A ROLLS ROYCE.
The £53,240 Rolls-Royce Silver Spirit. How many wheels? Four. The £2,584 Citroën 2CV. How many wheels? Exactly the same.
© www.vintagepaperads.com
MORE ROOM THAN A PORSCHE.
With a possible 30cu. ft. boot space there's no need for one of those plastic luggage racks on our little run-about.
THE £2,674 CITROËN 2CV.
All you'll ever need in a car.
PUNCH September 19 1984

Quality and runtime comparison

	Sensitivity		Jaccard Index		Time (s)	
	wcdx	PEACE	wcdx	PEACE	wcdx	PEACE
A076941	0.932	0.930	0.473	0.477	100	951
chlamy	0.949	0.949	0.513	0.513	907	8823
ecHuman	0.996	0.998	0.707	0.630	50	147
metasim454	0.793	0.714	0.765	0.689	16	66
metasimIllum	0.444	0.368	0.398	0.364	19	1975

SE = TP/(TP+FN), JI =TP/(TP+FP+FN), PPV = TP/(TP+FP)

PEACE: *NAR*, 2010

Quality of clustering using KABOOM-heuristic

"We can drive our car at least as fast as others have driven their car."

- ESTate
- PaCE
- PEACE
- TIGR

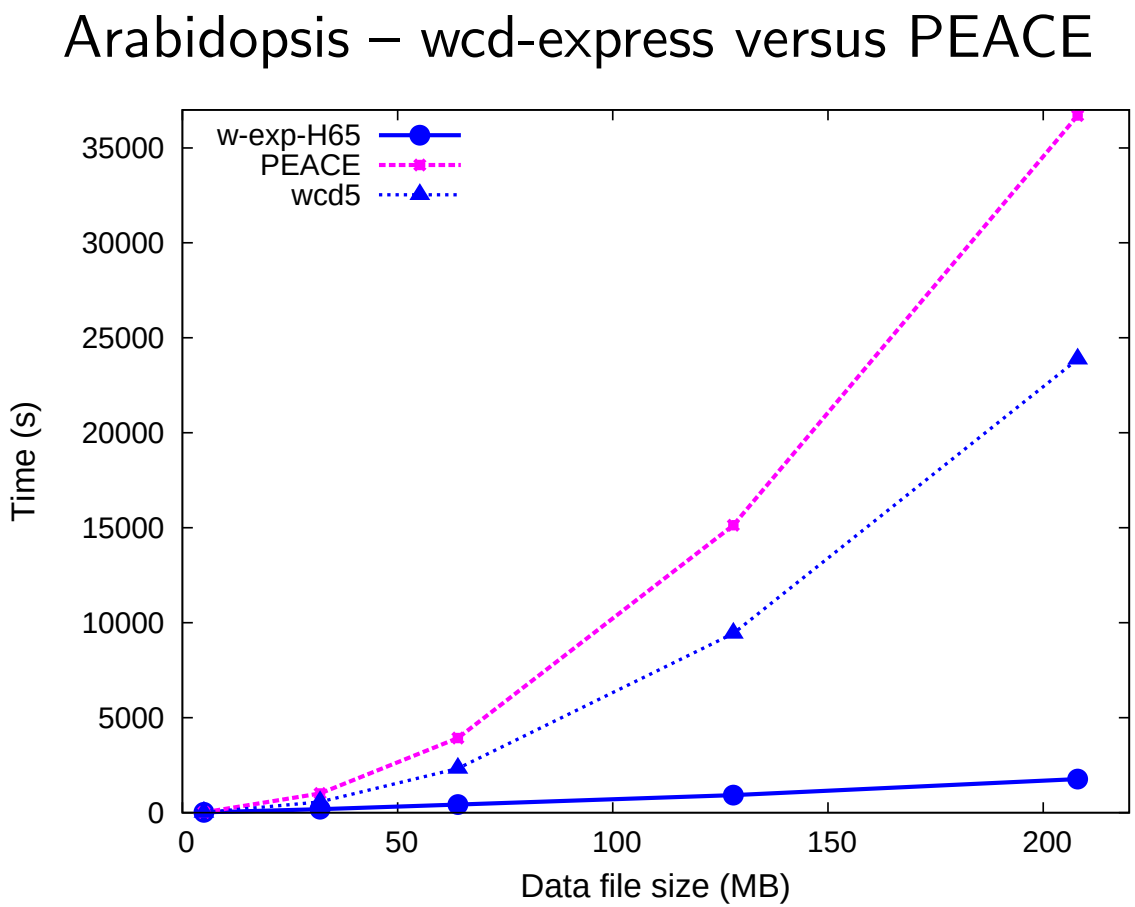
Quality of clustering using KABOOM-heuristic

"We can drive our car at least as fast as others have driven their car."

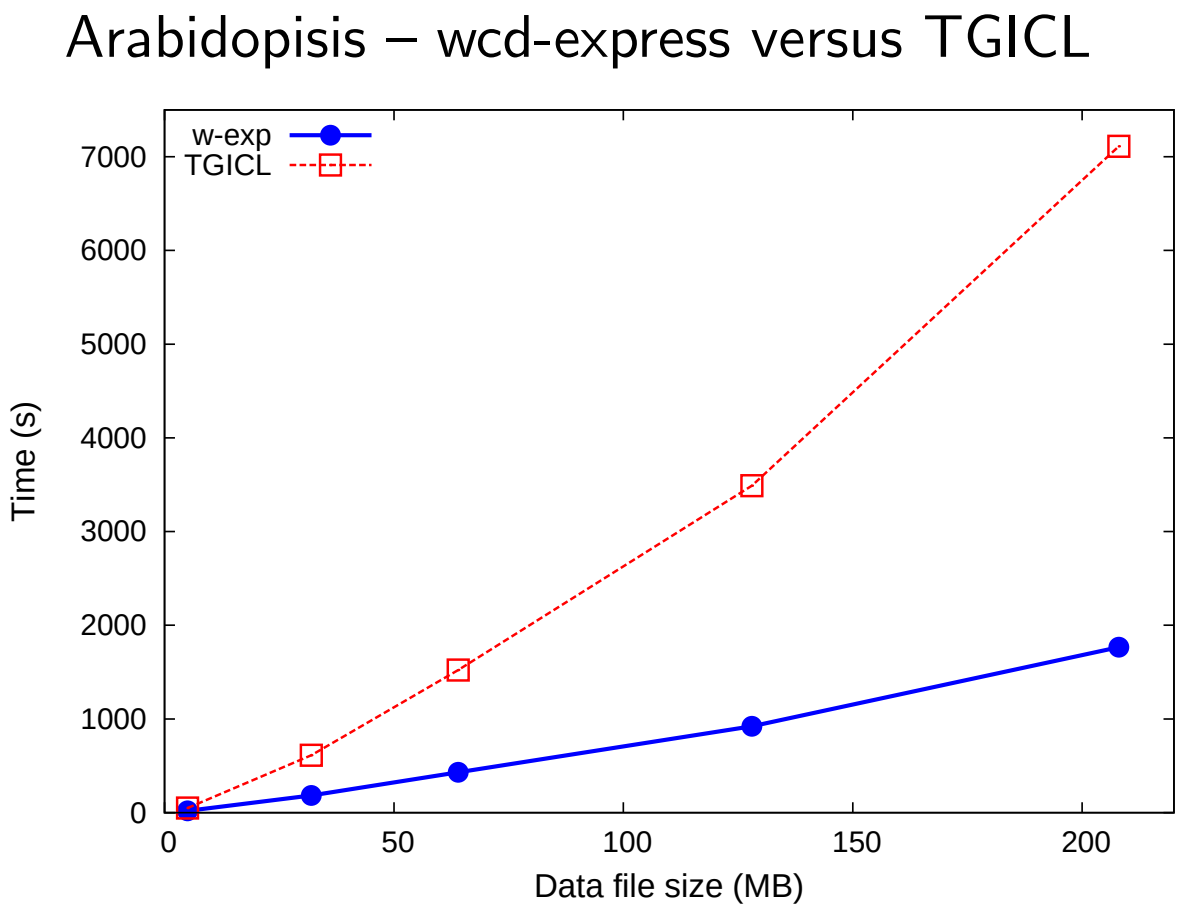
- ESTate
- PaCE
- PEACE
- TIGR

Performance matters.

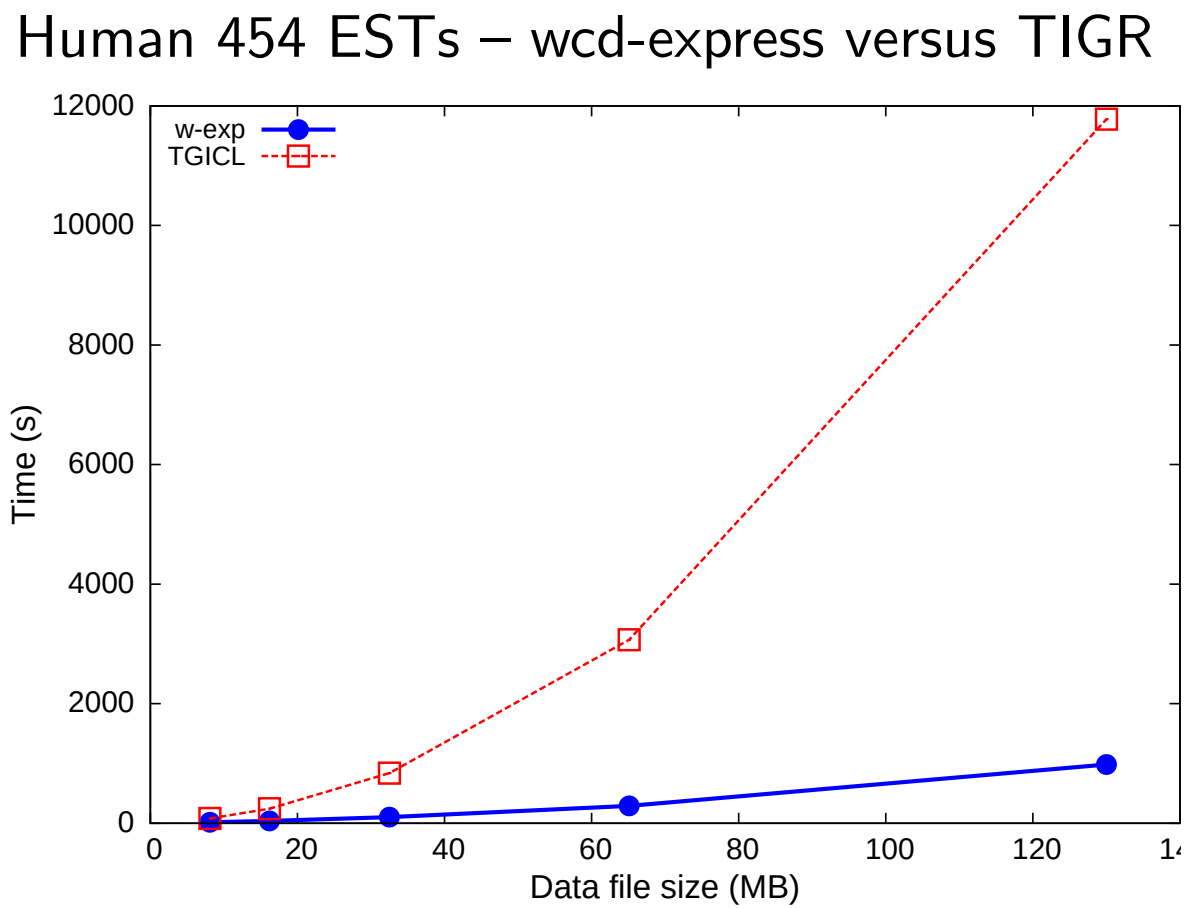
Runtime comparison



Runtime comparison



Runtime comparison



Conclusion

- wcd + KABOOM! = wcd-express
- Much faster and at least as good as other tools.
- Memory: very heavy memory usage (suffix array + inverse)

Conclusion

- wcd + KABOOM! = wcd-express
- Much faster and at least as good as other tools.
- Memory: very heavy memory usage (suffix array + inverse)

Ongoing/future work

- Memory needs to be reduced!
- Integrate wcd-express into workflow (filtering/masking, assembly)
- GUI?

Paper to appear in Bioinformatics.


KaBOOM!
®
it starts with a
playground.

Thank you.

zsuzsa@cebitec.uni-bielefeld.de

Scott.Hazelhurst@wits.ac.za