SVJedi-graph: using a variation graph to improve structural variant genotyping with long reads

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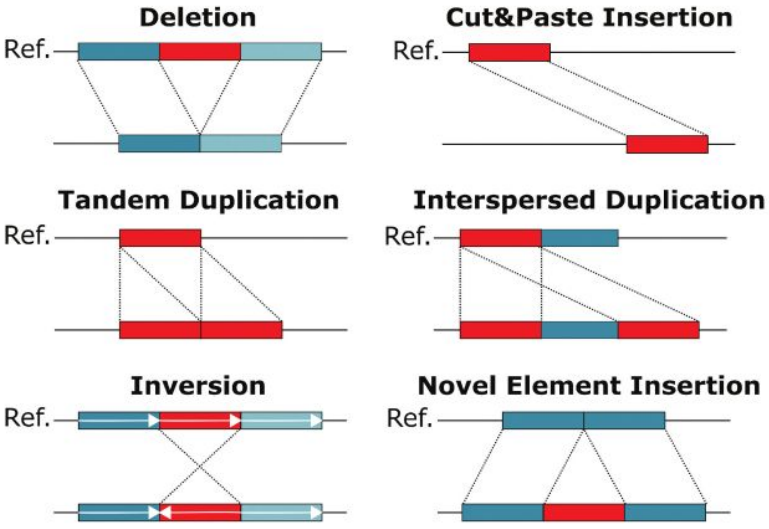
Structural variants



Heller and Vingron, 2019

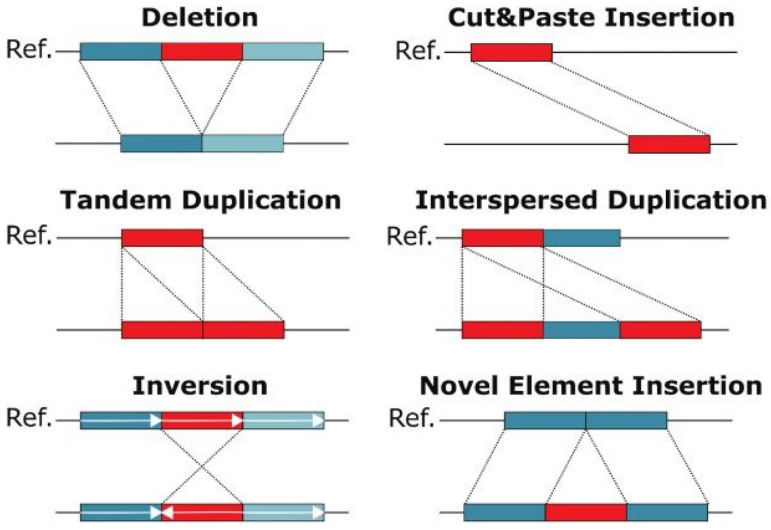
Defined:

as rearrangements ≥ 50 bp

relatively to a reference genome

by
- breakpoints
- sequence

Structural variants



Heller and Vingron, 2019

Defined:

as rearrangements ≥ 50 bp

relatively to a reference genome

by
- breakpoints
- sequence

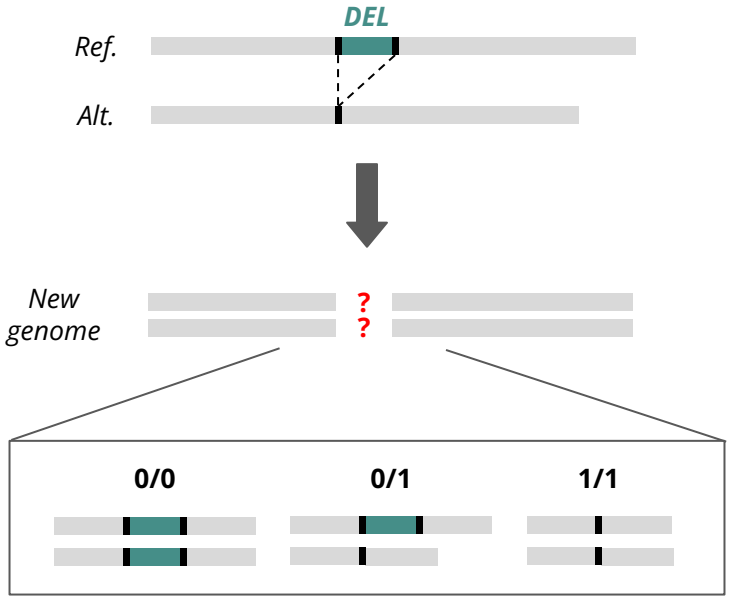
Impact:

depends on genomic context

can lead to
- diseases
- polymorphism in agronomic key traits

Genotyping structural variants

- After SV identification
  - type
  - position
  - sequence for INS
- Presence of the SVs on the haplotypes ?



Approaches:

- Mapping *vs*. “mapping-free”
- Short reads *vs*. long reads

State of the art

Mapping-based genotypers:

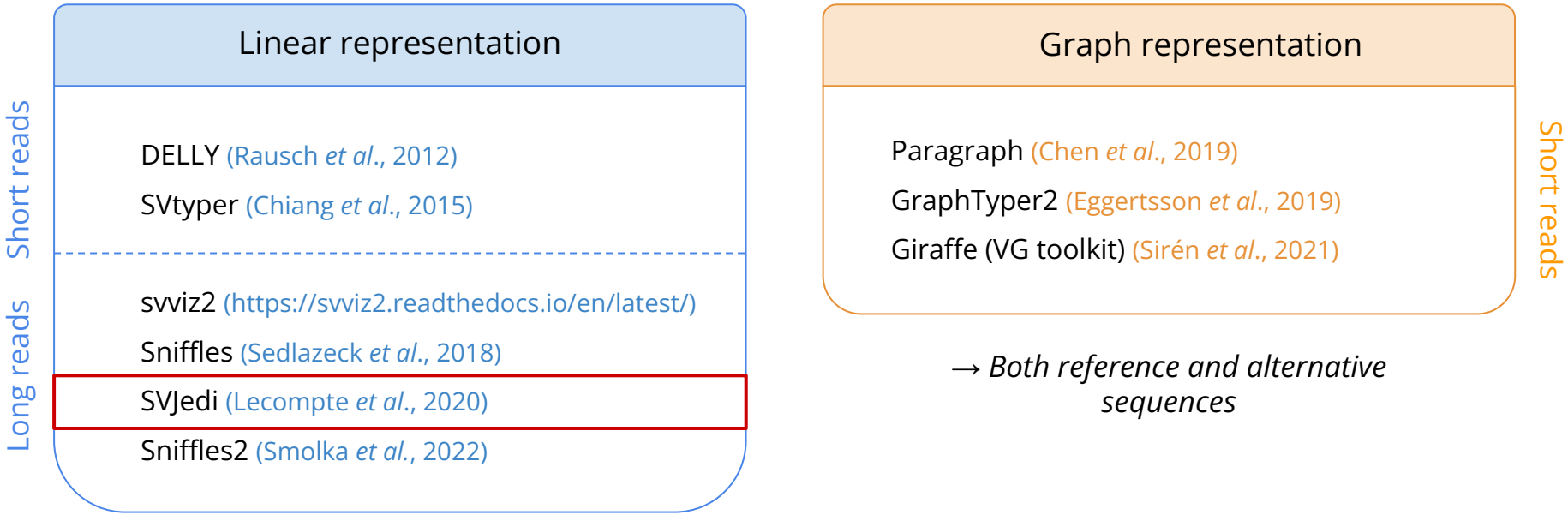
	Linear representation	Graph representation
Short reads	DELLY (Rausch *et al.*, 2012) SVtyper (Chiang *et al.*, 2015)	Paragraph (Chen *et al.*, 2019) GraphTyper2 (Eggertsson *et al.*, 2019) Giraffe (VG toolkit) (Sirén *et al.*, 2021)
Long reads	svviz2 (https://svviz2.readthedocs.io/en/latest/) Sniffles (Sedlazeck *et al.*, 2018) SVJedi (Lecompte *et al.*, 2020) Sniffles2 (Smolka *et al.*, 2022)	

→ *Reference bias*

→ *Both reference and alternative sequences*

State of the art

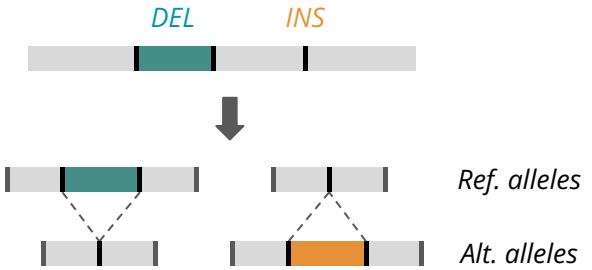
Mapping-based genotypers:



→ *Reference bias*

→ *Both reference and alternative sequences*

SVJedi (Lecompte *et al.*, 2020)



Principle: Representing both alleles for each SV in linear reference

→ *Reduce reference bias*

Tool	Genotyping accuracy	Genotyping rate	Time
SVJedi	**92.2**	90.3	**2h25m**
Sniffles -Ivcf	82.0	**99.8**	17h16m
svviz2	65.9	**100**	5days
Sniffles (discovery mode)	43.6	48.1	18h04m
pbsv	77.9	65.3	5h29m

from Lecompte et al., 2020

Limitation: Drop of genotyping rate with close/overlapping SVs

→ ⚠ *Sequence redundancy*

https://github.com/llecompte/SVJedi

Our contribution: SVJedi-graph

Long read SV genotyper using a variation graph representation

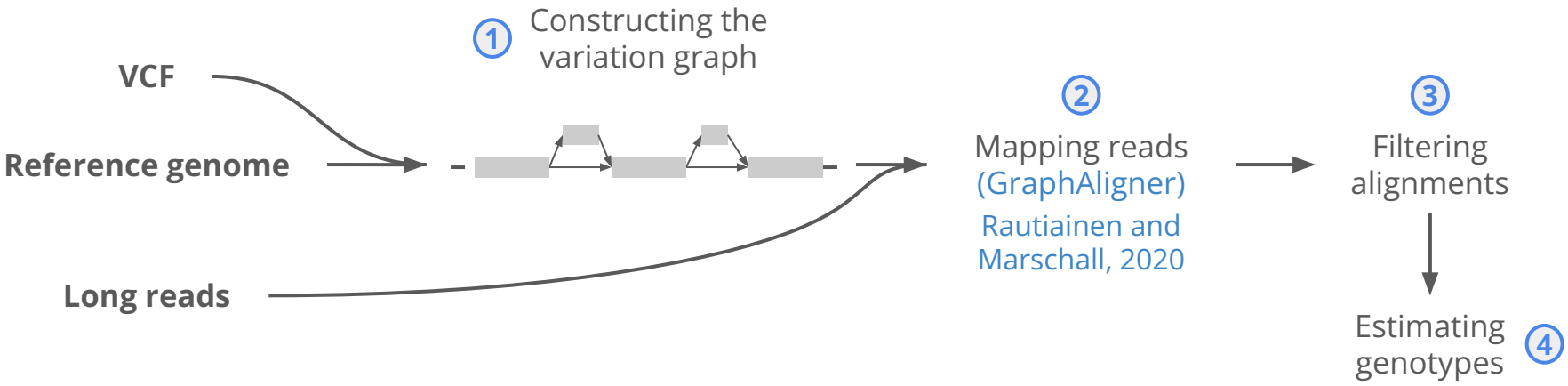
- Improve close SV genotyping by using a variation graph

- Represent the whole genome sequence

Method

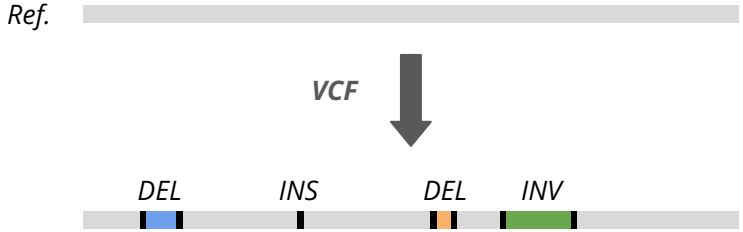
Overview of SVJedi-graph

Input: reference genome, SV set, long reads



Output: genotyped SV set

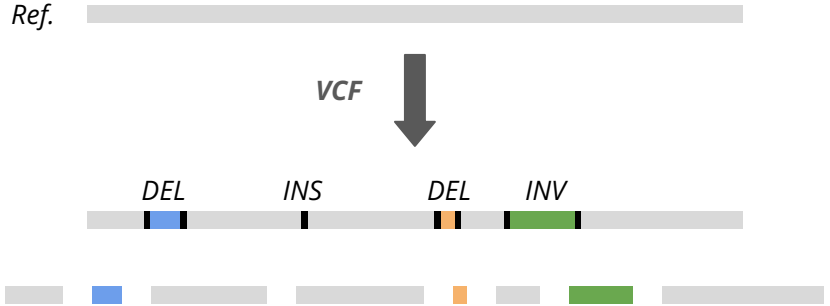
(1) Constructing the variation graph



For each chromosome:

1. List & sort breakpoint positions

(1) Constructing the variation graph

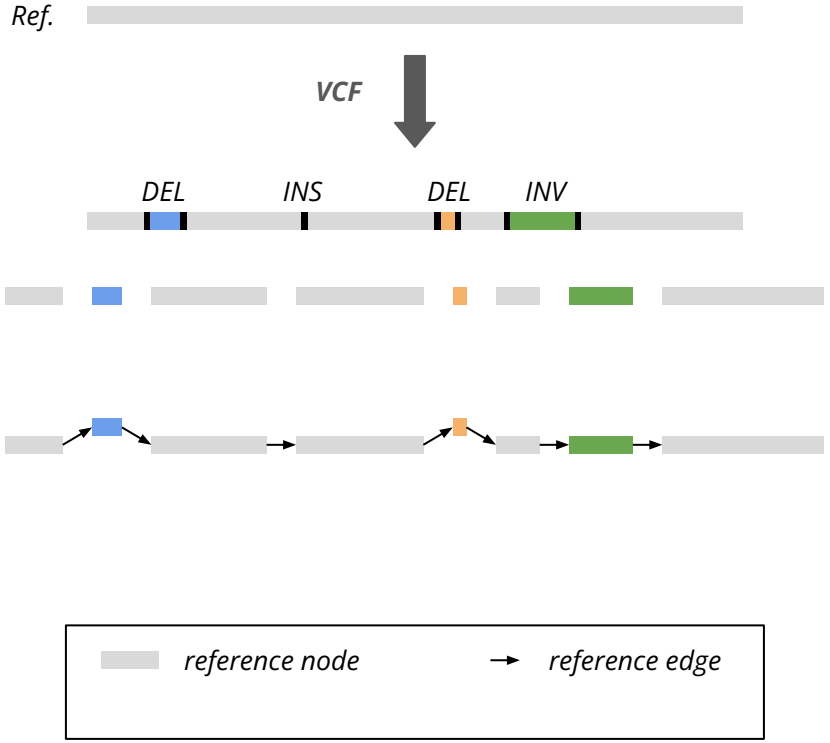


For each chromosome:

1. List & sort breakpoint positions
2. Split sequence at each breakpoint

1 fragment = 1 node

(1) Constructing the variation graph

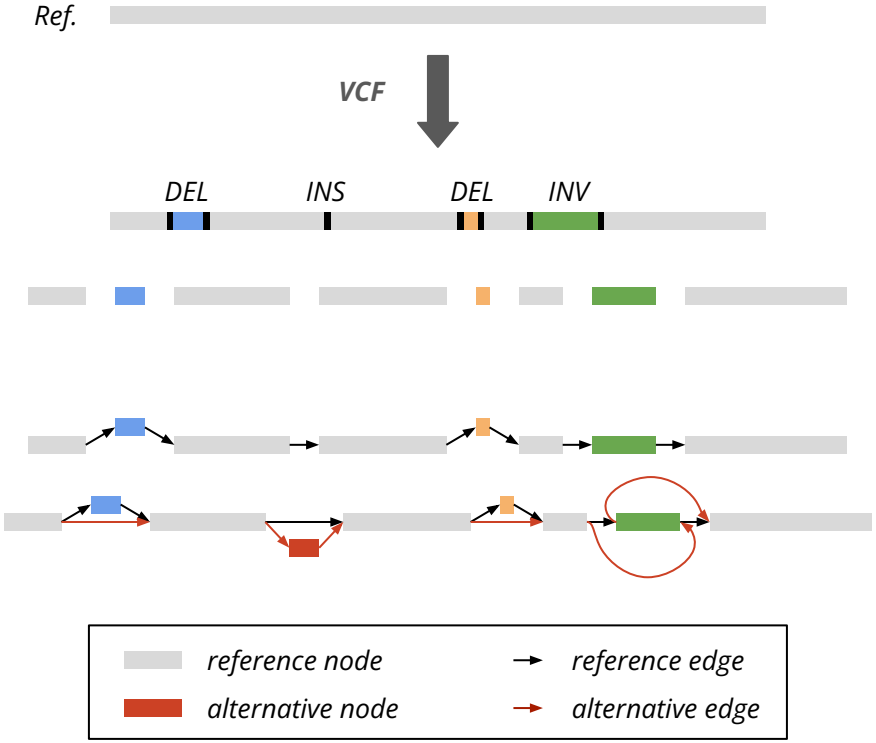


For each chromosome:

1. List & sort breakpoint positions
2. Split sequence at each breakpoint

   1 fragment = 1 node
3. Add reference edges

(1) Constructing the variation graph



For each chromosome:

1. List & sort breakpoint positions
2. Split sequence at each breakpoint

   1 fragment = 1 node
3. Add reference edges
4. Add alternative edges

   + alternative nodes for insertions

(2)(3) Mapping the reads and filtering the alignments

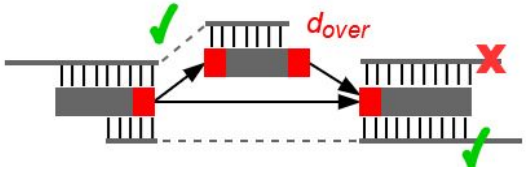
Mapping: GraphAligner (Rautiainen and Marschall, 2020)

Alignments filters:

➢ Number of nodes in the alignment path >= 2 → *Filtering alignments to analyse*

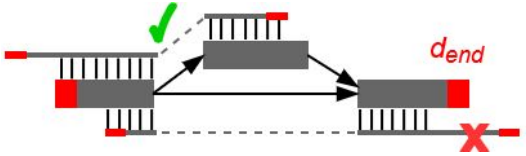
➢ Breakpoints overlap

→ *Confidence in supported allele*



➢ Alignment semi-globality

→ *Accuracy of mapping location*



(4) Predicting the genotype

- Count supporting reads for each allele
- Normalize by allele length ratio
- Compute likelihood for each genotype

$$\ell(0/0) = (1 - err)^{c_0^*} \times err^{c_1} \times C_{c_0^*+c_1}^{c_0^*}$$

$$\ell(1/1) = err^{c_0^*} \times (1 - err)^{c_1} \times C_{c_0^*+c_1}^{c_0^*}$$

$$\ell(0/1) = \left(\tfrac{1}{2}\right)^{c_0^*+c_1} \times C_{c_0^*+c_1}^{c_0^*}$$

Reused from SVJedi (Lecompte et al., 2020)

Evaluation on simulated datasets

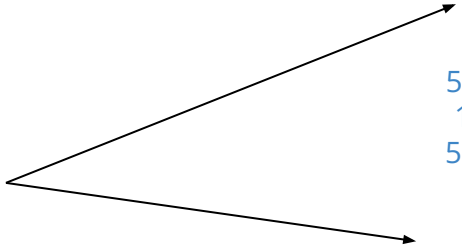
The simulated datasets

Reference: human chromosome 1 (GRCh37.p13)

SV sets generation:

- ➢ 1,000 deletions from dbVar
- ➢ + close/overlapping deletions

  (⅓ 0/0 - ⅓ 0/1 - ⅓ 1/1)



6 sets w/ close deletions

5k → 10k	100 → 500
1k → 5k	50 → 100
500 → 1k	0 → 50

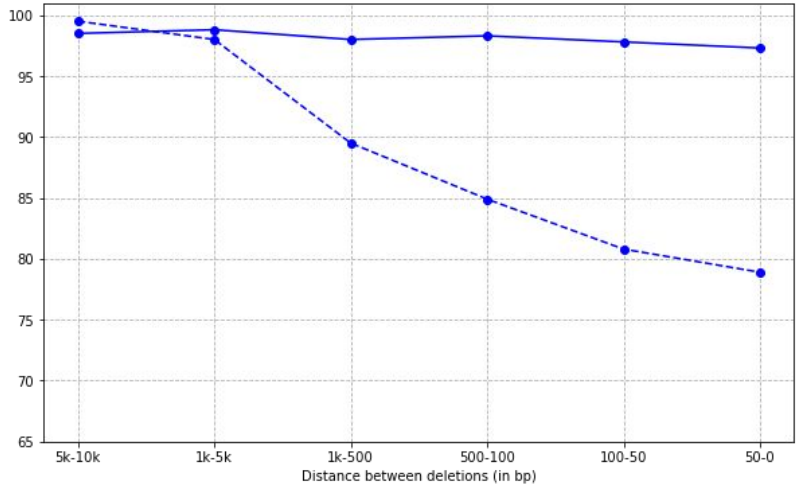
6 sets w/ overlapping deletions

0 → 50	200 → 300
50 → 100	300 → 400
100 → 200	400 → 500

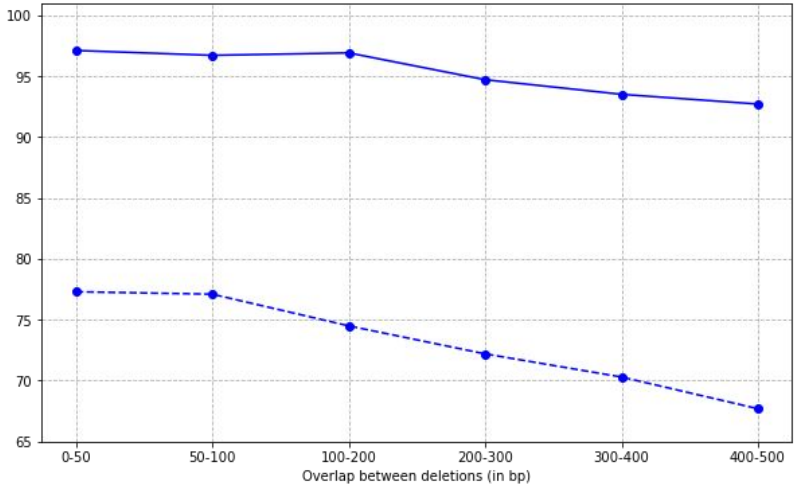
Reads simulation: PacBio, 16 % error rate (SimLoRD)

The simulated datasets - Results (SVJedi)

Close deletions



Overlapping deletions



genotyping accuracy

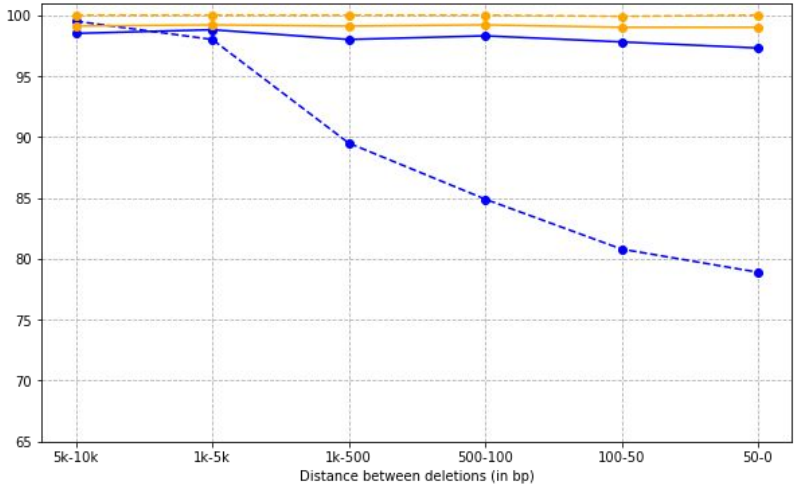
genotyping rate

Rate: % of SVs genotyped / all SVs

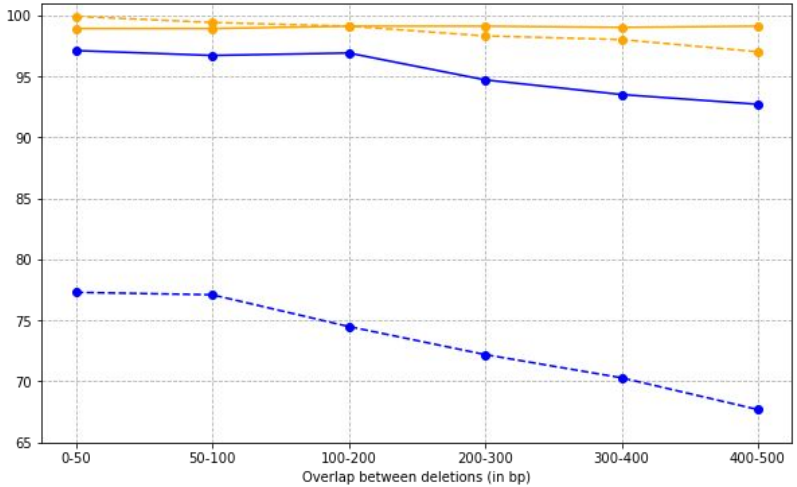
Accuracy: % of SVs accurately genotyped / genotyped SVs

The simulated datasets - Results (SVJedi-graph)

Close deletions



Overlapping deletions



genotyping accuracy

genotyping rate

SVJedi

SVJedi-graph

➢ **Recovery of genotyping rate**

Evaluation on real dataset

The GIAB dataset

Reference: human reference genome (GRCh37.p13)

Reads: PacBio from HG002 (GIAB dataset)

SV set: HG002 Tier 1 (Zook *et al.*, 2019)

- ➢ 5,464 deletions
- ➢ 7,281 insertions

} *with ground truth genotypes*

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pbsv	77.9	65.3	5h29m
SVJedi-graph	**92.9**	97.4	15h28m

→ *Time cost of mapping on graph*

New SV calling dataset from GIAB data

Reference: human reference genome (GRCh37.p13)

Reads: PacBio from HG002 (GIAB dataset)

SV calling: NGMlr + Sniffles (Sedlazeck *et al*., 2018)

- ➢ 7,922 deletions
- ➢ 9,529 insertions
- ➢ 202 inversions

Dataset	all SVs	“close” SVs
GIAB “gold standard”	12,721	581 (4.6%)
New SV calling set	**17,624**	**2,205 (12.5%)**

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	Genotyping rate
SVJedi	51 %
SVJedi-graph	**98 %**

Concluding remarks

Implemented in python

Availability:

 https://github.com/SandraLouise/SVJedi-graph

 (soon)

Work in progress:

Evaluating genotyping accuracy on the GIAB dataset

Improve read mapping time

Genotyping translocations

Acknowledgements

Mobility grant

Access to computing cluster

And my PhD supervisors: Claire Lemaitre and Fabrice Legeai

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