Genotyping structural variants in pangenome graphs using the vg toolkit

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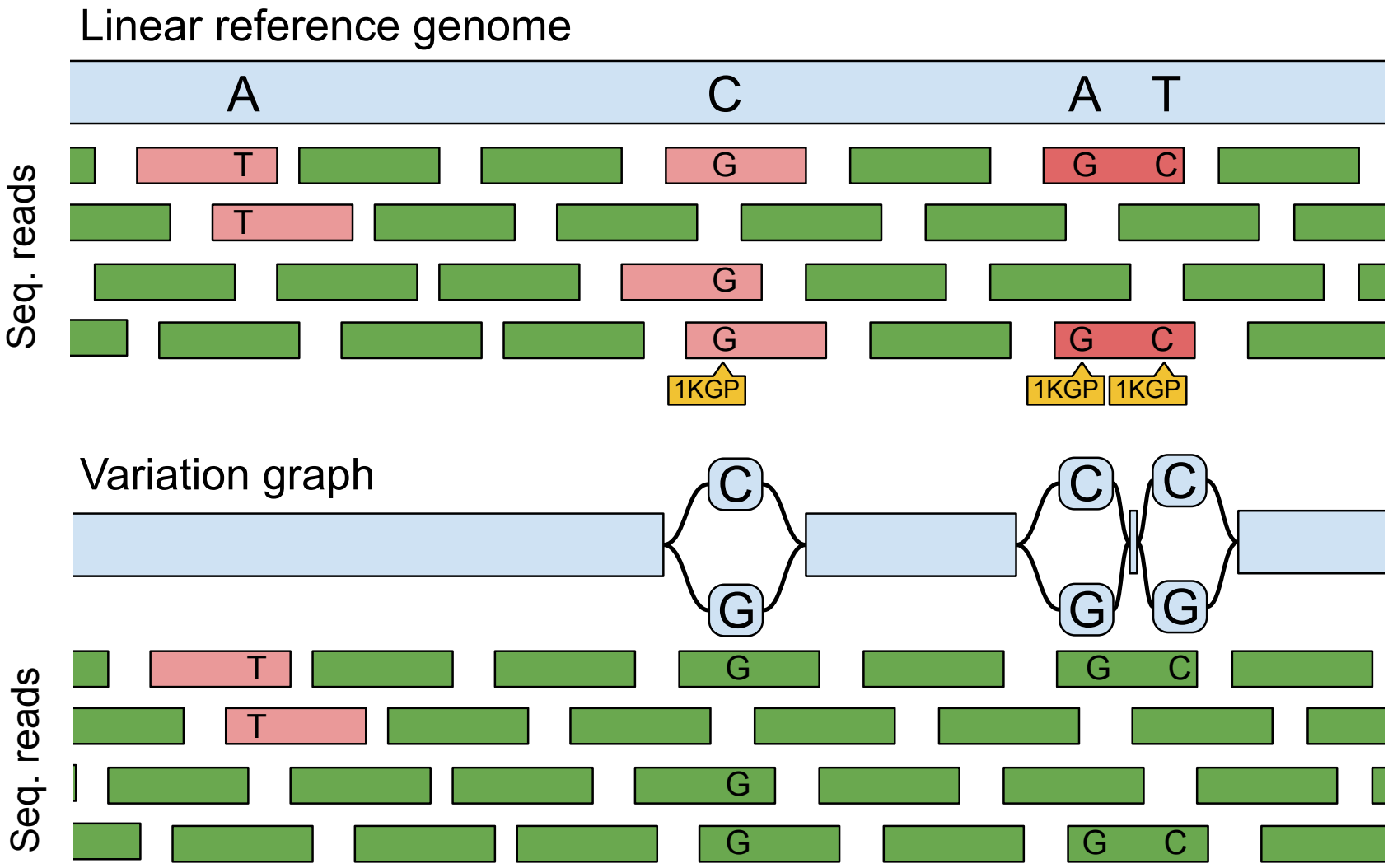
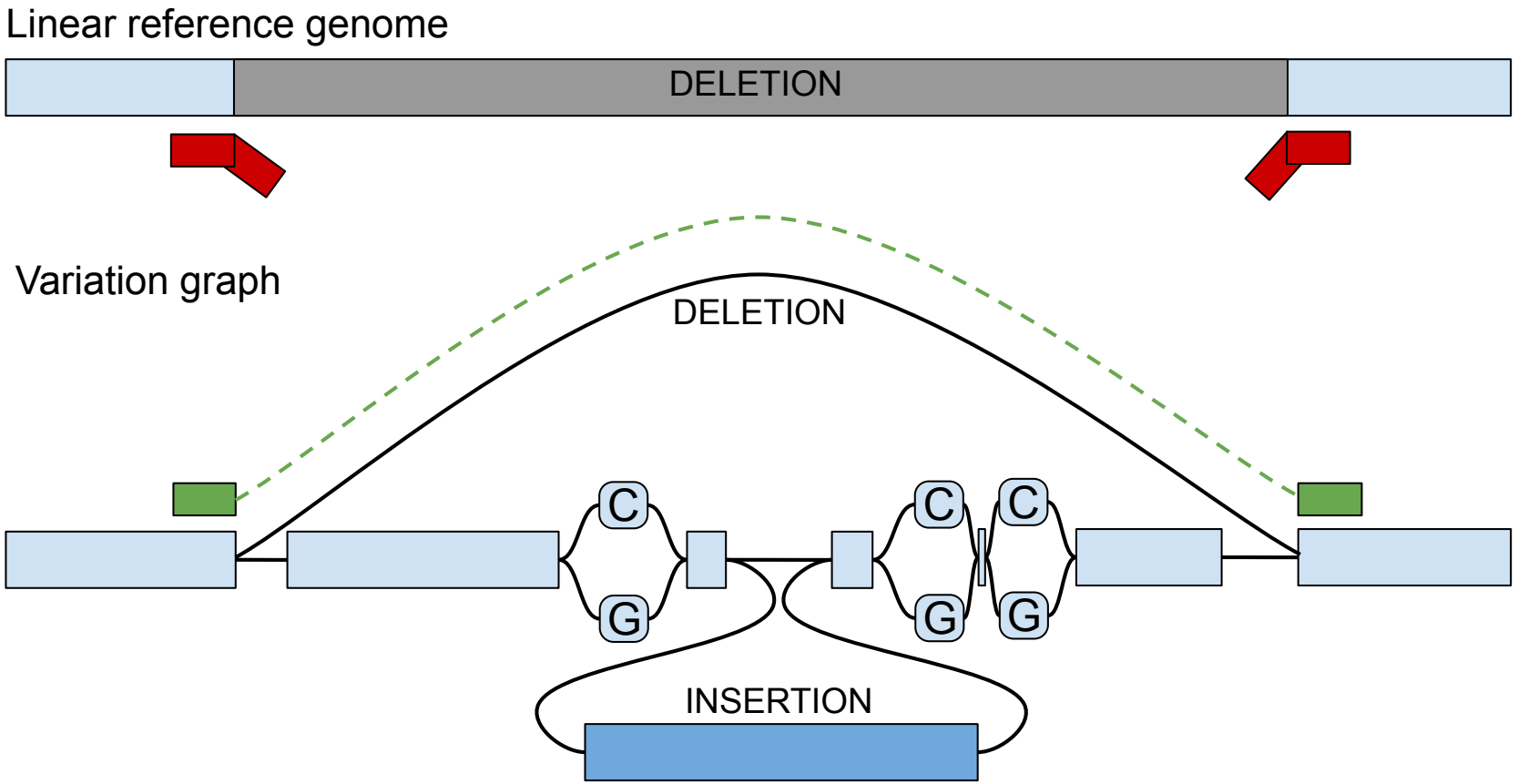
Genome Informatics

Pangenome graphs and variant-aware read mapping



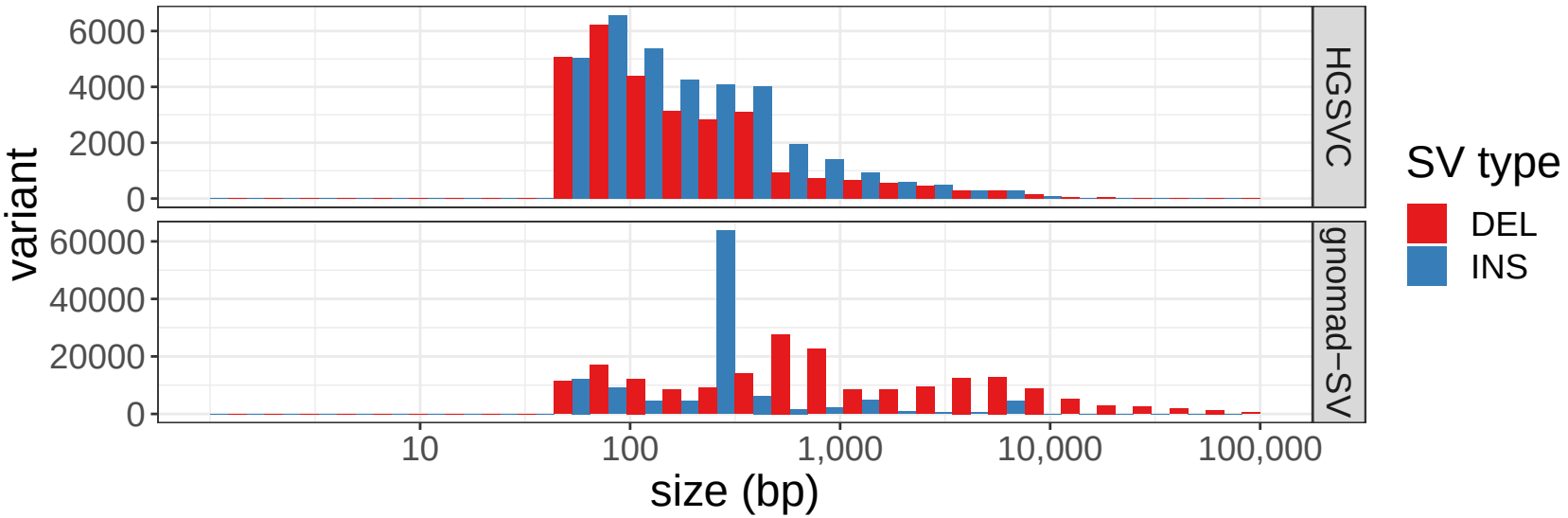
Mapping reads across structural variants

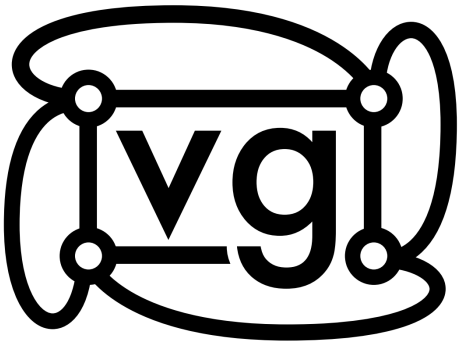
Structural variants are genomic variants larger than 50 bp, e.g. insertions, deletions, inversions translocations.



SV catalogs from long-read sequencing studies

Ref.	Project	Samples
Chaisson et al. 2019	Human Genome Structural Variation Consortium (**HGSVC**)	3
Audano et al. 2019	**SVPOP**	15
Zook et al. 2019	Genome in a Bottle (**GIAB**)	1





The **vg toolkit** is a complete, open source solution for **graph construction**, **read mapping**, and **variant calling**.

`https://github.com/vgteam/vg`

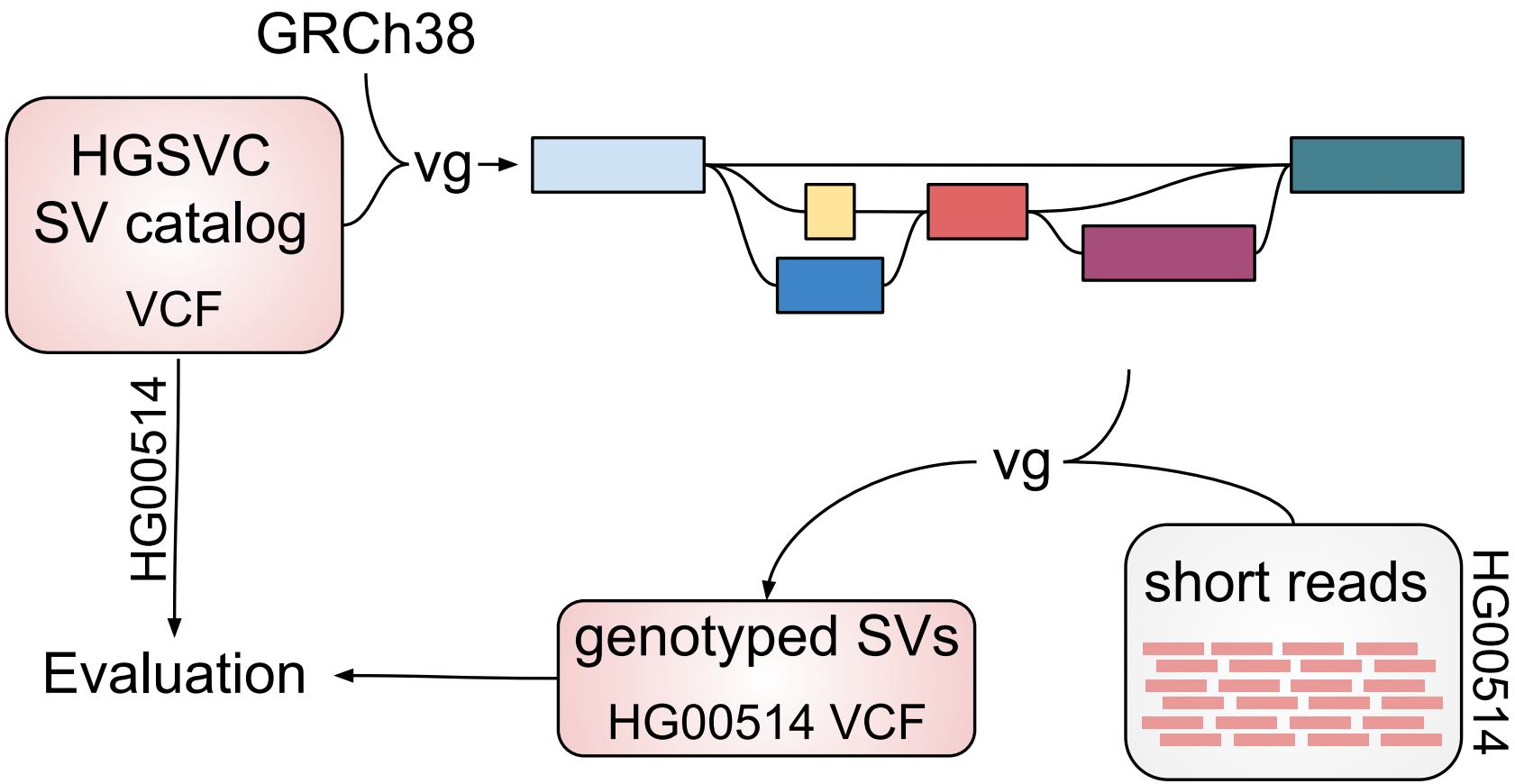
Garrison et al. Nature Biotech 2018

Can we genotype SVs from short-read sequencing datasets with the vg toolkit?

Starting from public SV catalogs or *de novo* assemblies.

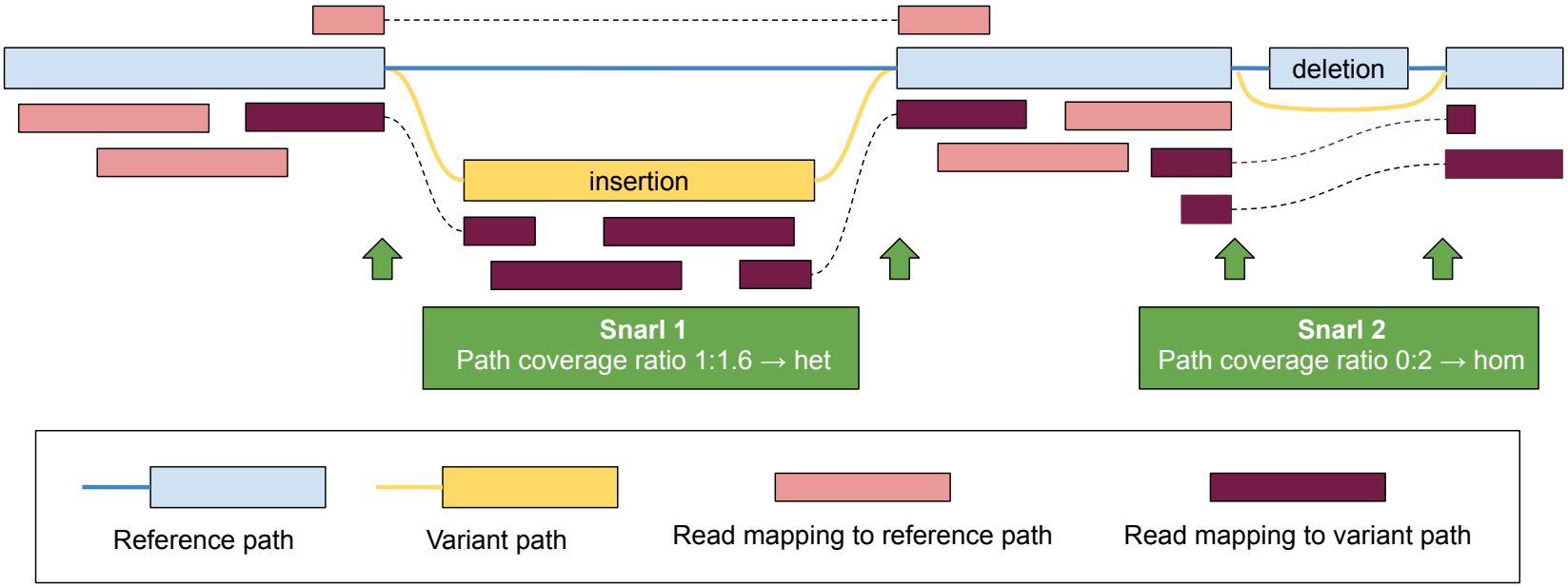
Hickey et al. bioRxiv 2019

Genotyping public SV catalogs in human



Evaluate genotype predictions for a sample from the truth set (e.g. HG00514).

Genotyping variants in vg

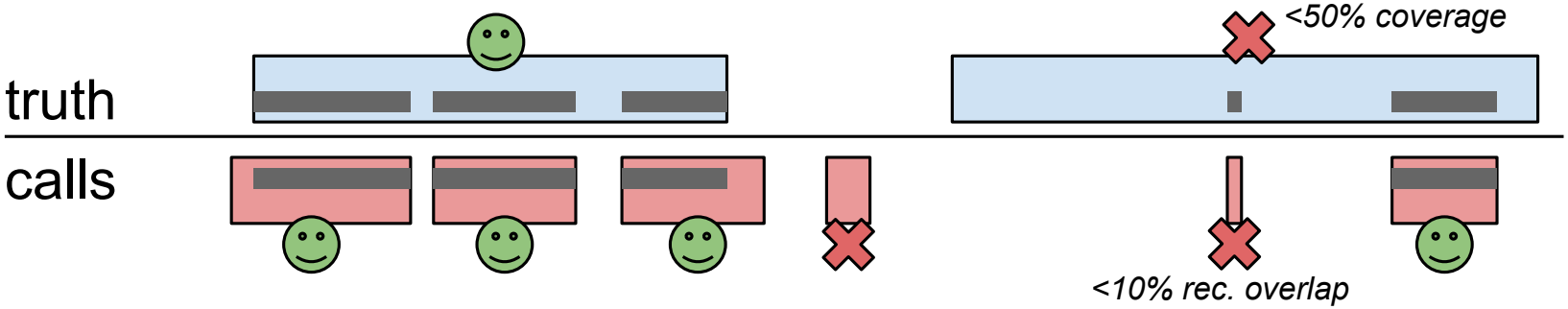


- Genotyping is based on the path coverage.
- A snarl is a variant site in the graph, a “bubble”.

Evaluating SV genotypes with a truth set

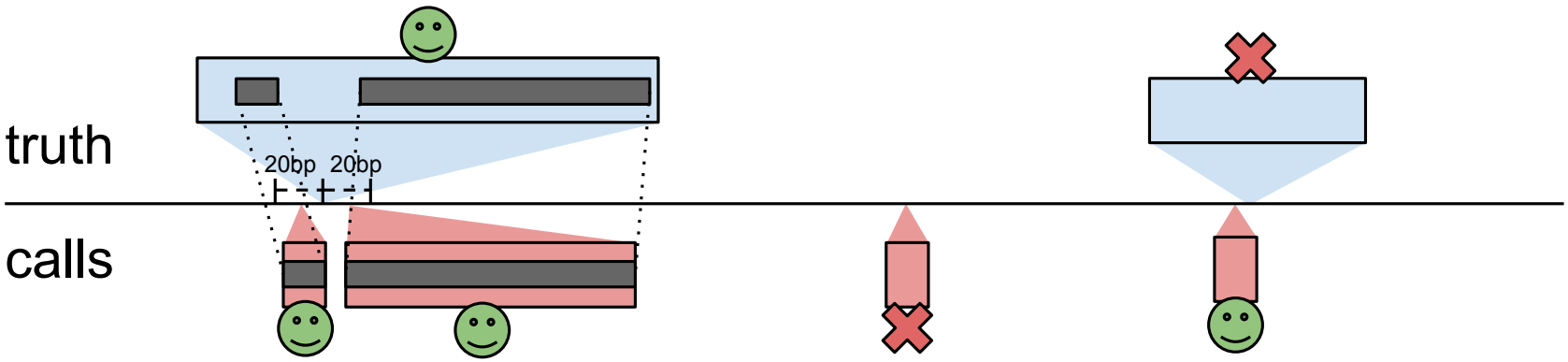
Deletions/Inversions

At least 50% coverage and 10% reciprocal overlap



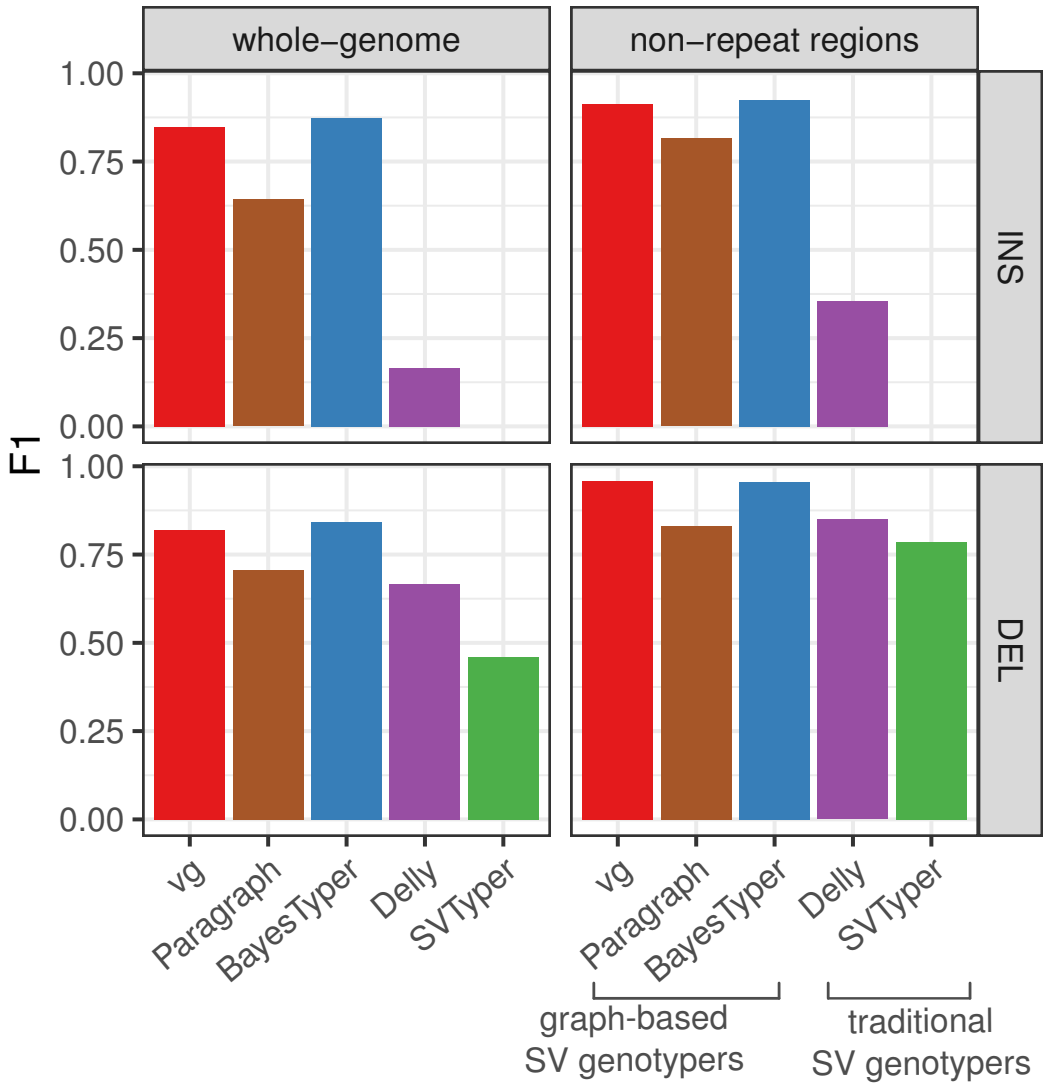
Insertions

At least 50% of inserted sequence matching nearby insertions



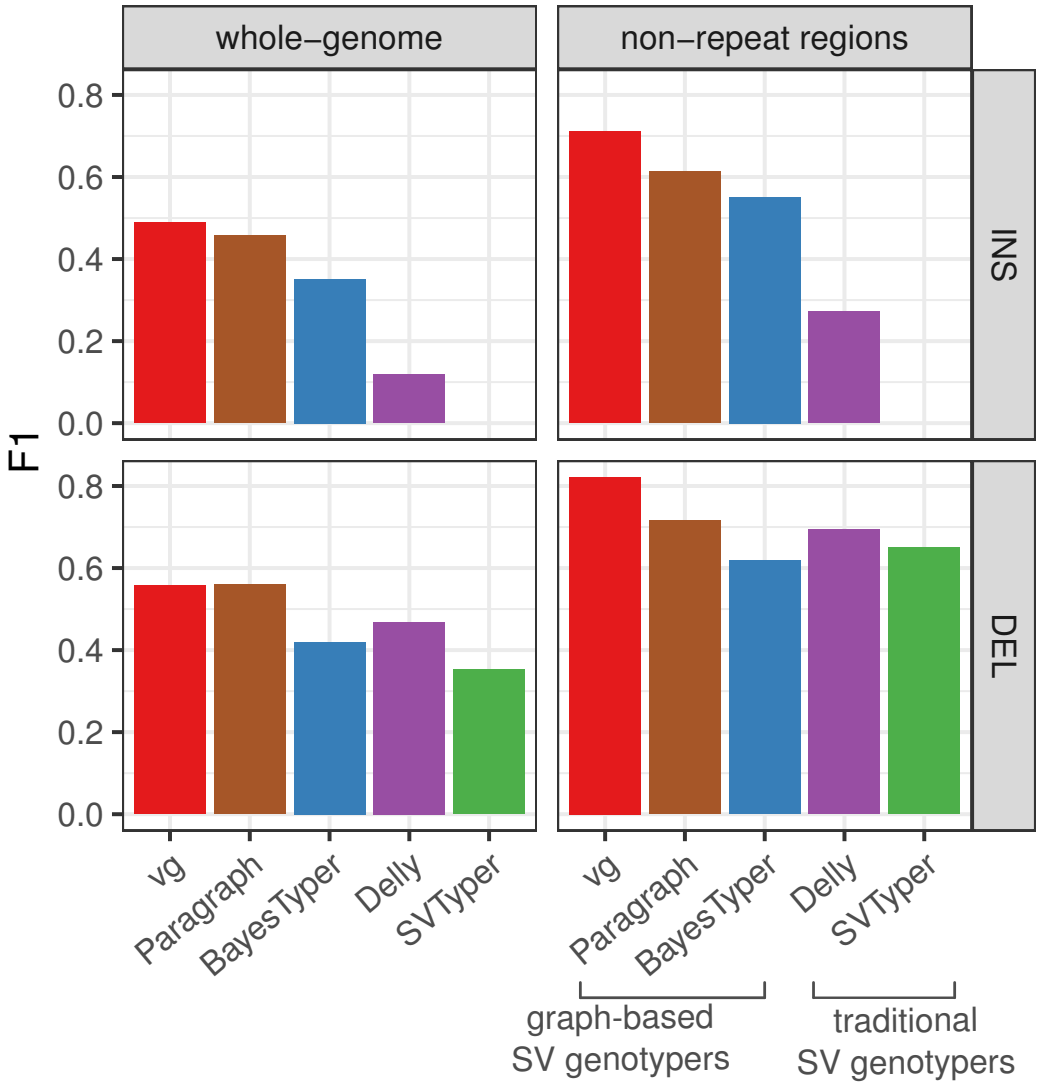
R package: `https://github.com/jmonlong/sveval`

Results on HGSVC - Simulated reads



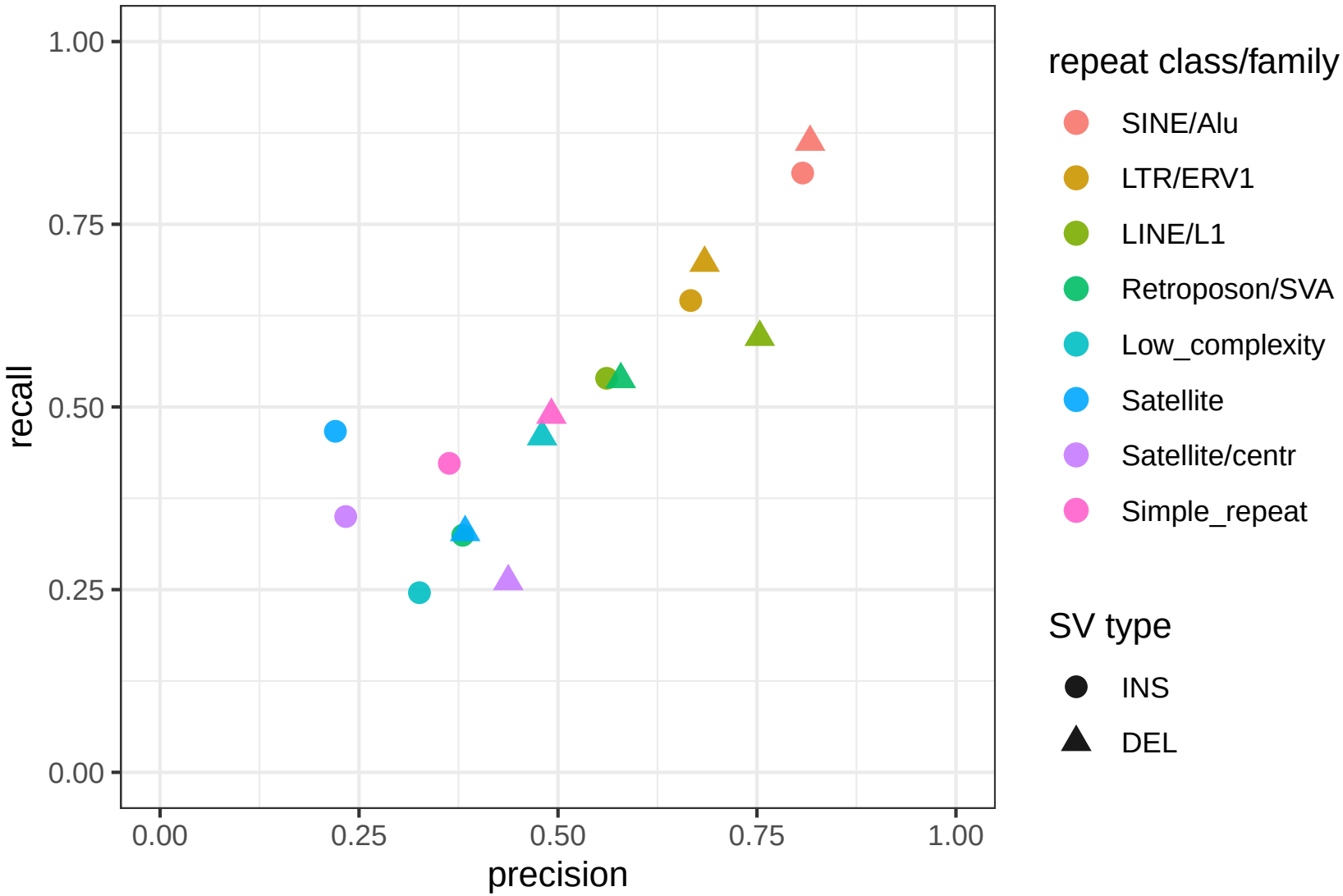
Non-repeat regions: regions not overlapping segmental duplications or simple repeats

Results on HGSVC - Real reads



Non-repeat regions: regions not overlapping segmental duplications or simple repeats

Simple repeat/low complexity regions are challenging



SV sequence annotated with RepeatMasker. Class assigned if covered ≥80% by a repeat element.

Challenges with the VCF format

Multiple equivalent representations, over-simplification, impractical.

Figure 8: Inversion

can be described equivalently in two ways. Either one uses the short hand notation described previously (recommended for simple cases):

#CHROM	POS	ID	REF	ALT	QUAL	FILTER	INFO
2	321682	INV0	T	<INV>	6	PASS	SVTYPE=INV;END=421681

or one describes the breakends:

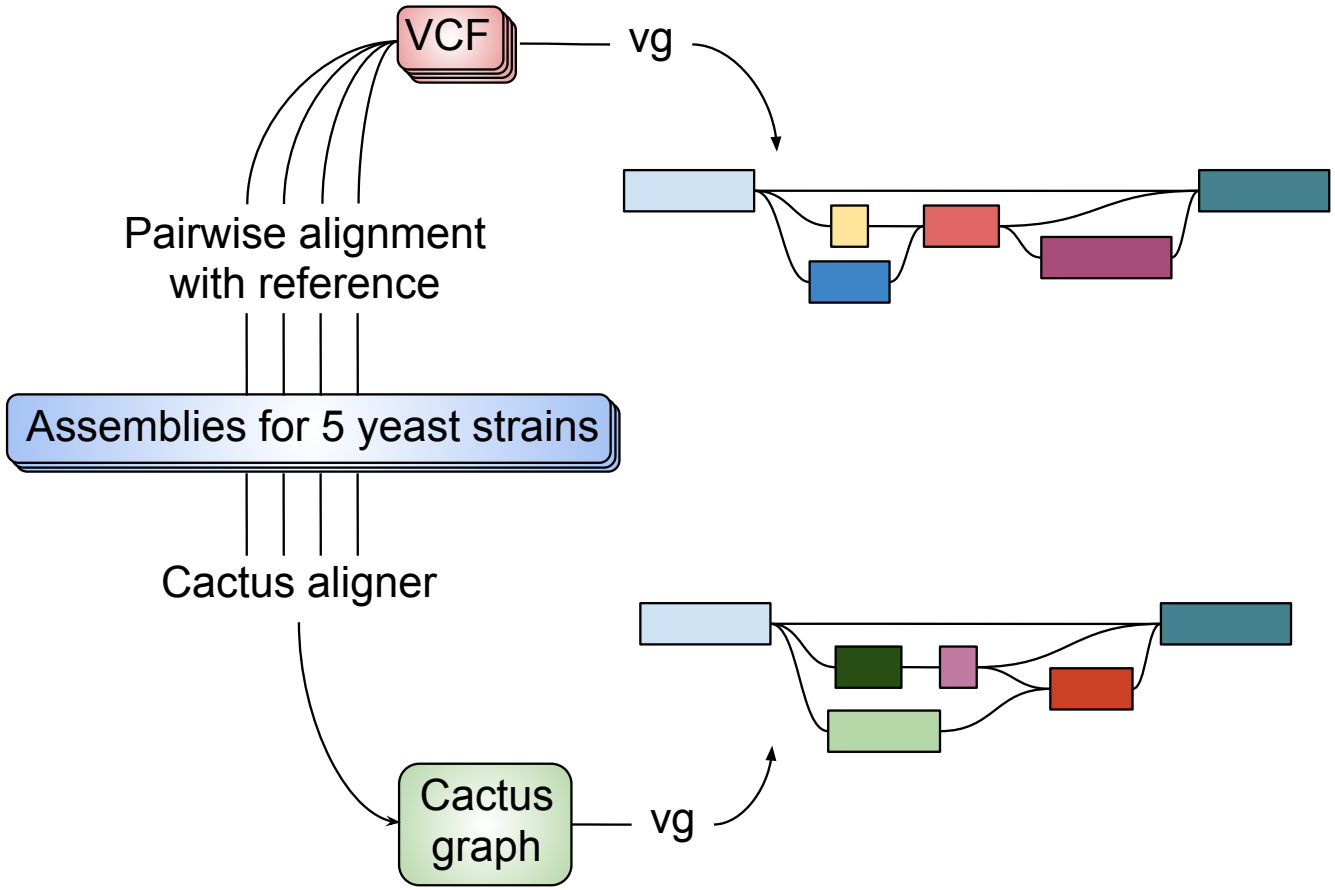
#CHROM	POS	ID	REF	ALT	QUAL	FILTER	INFO
2	321681	bnd_W	G	G]2 : 421681]	6	PASS	SVTYPE=BND;MATEID=bnd_U;EVENT=INV0
2	321682	bnd_V	T	[2 : 421682[T	6	PASS	SVTYPE=BND;MATEID=bnd_X;EVENT=INV0
2	421681	bnd_U	A	A]2 : 321681]	6	PASS	SVTYPE=BND;MATEID=bnd_W;EVENT=INV0
2	421682	bnd_X	C	[2 : 321682[C	6	PASS	SVTYPE=BND;MATEID=bnd_V;EVENT=INV0

VCF v4.2 specs

Why not start directly from de novo assemblies?

Analysis of 12 yeast strains from 2 clades

Selected 5 strains to build graph: one reference + 2 per clade.



Evaluating SV genotyping using mapping statistics

- No gold-standard to compare with.

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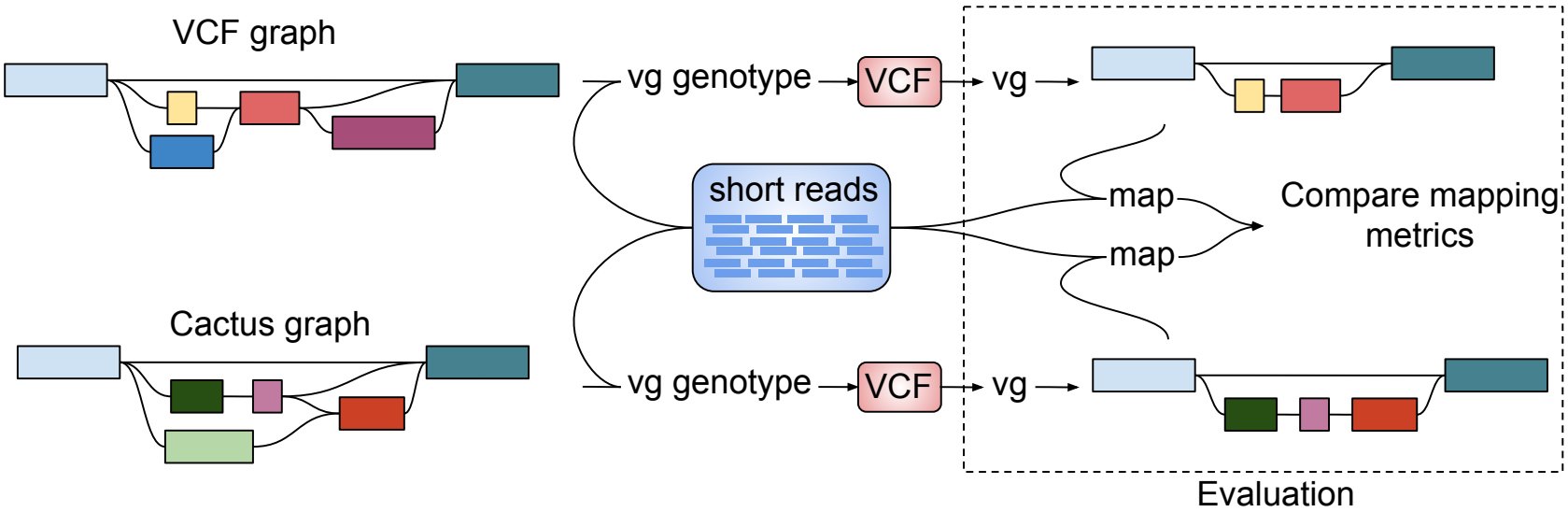
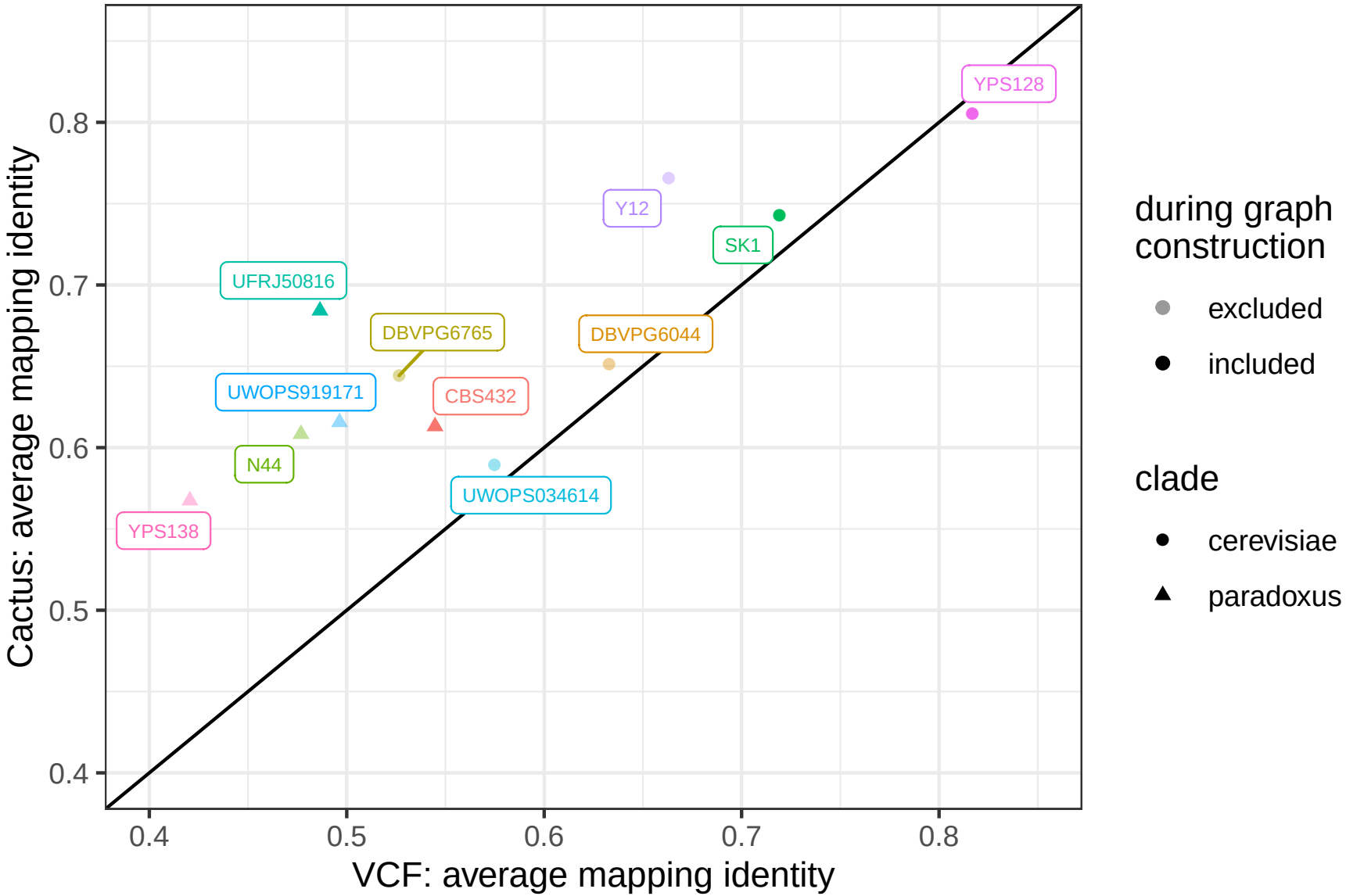
- No gold-standard to compare with.
- Map reads to a sample graph built from the SV calls:



- Mapping quality $\sim$ Sample graph quality $\sim$ SV calls quality.

Better mapping for SVs called in the cactus graph

Analysis restricted to reads at variation sites.



Conclusions

- The vg toolkit can integrate and genotype SVs.
- Graphs from *de novo* assemblies alignment performs better.

Hickey et al. bioRxiv 2019 `https://jmonlong.github.io/manu-vgsv/`

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Future directions

- Experiment with **high-quality human de novo assemblies** (e.g. the Human PanGenome Project).
- **Combine** public SV catalogs and **genotype** SVs in a large and diverse cohort.

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W.M. KECK FOUNDATION

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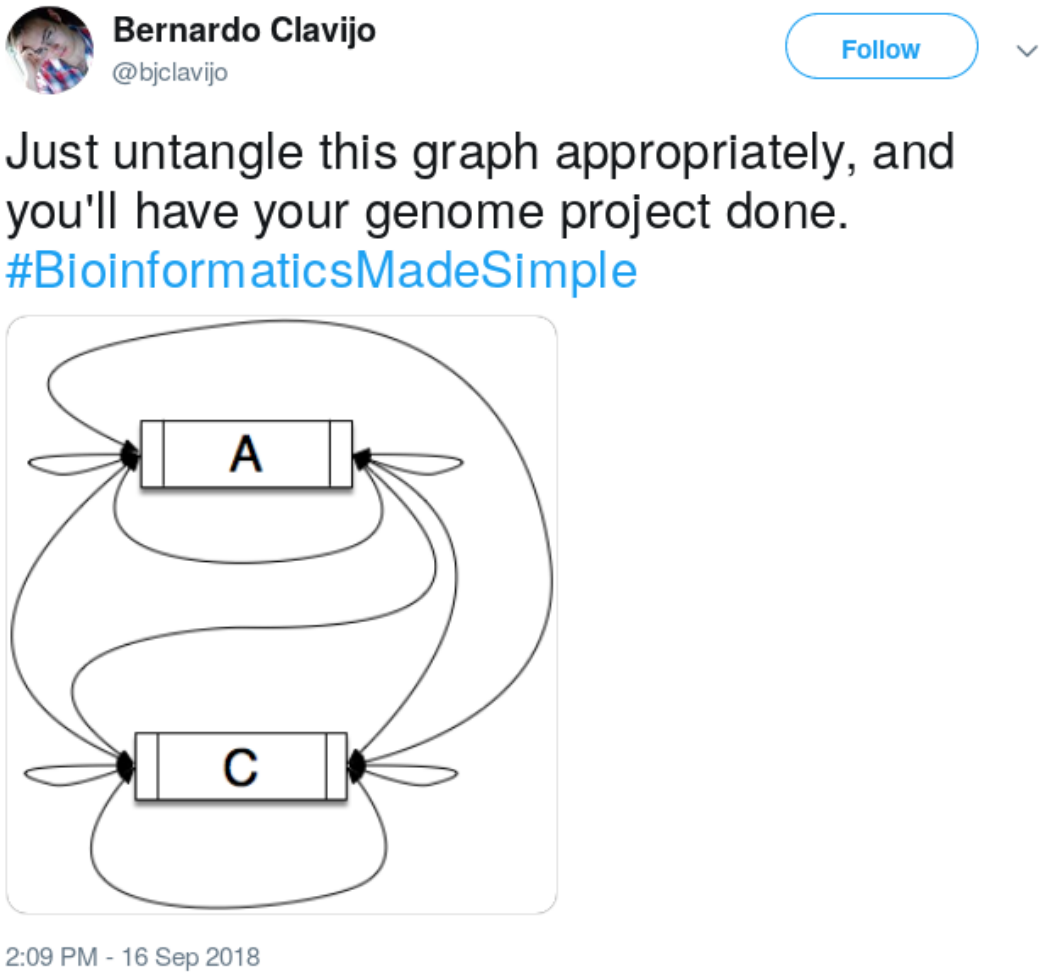
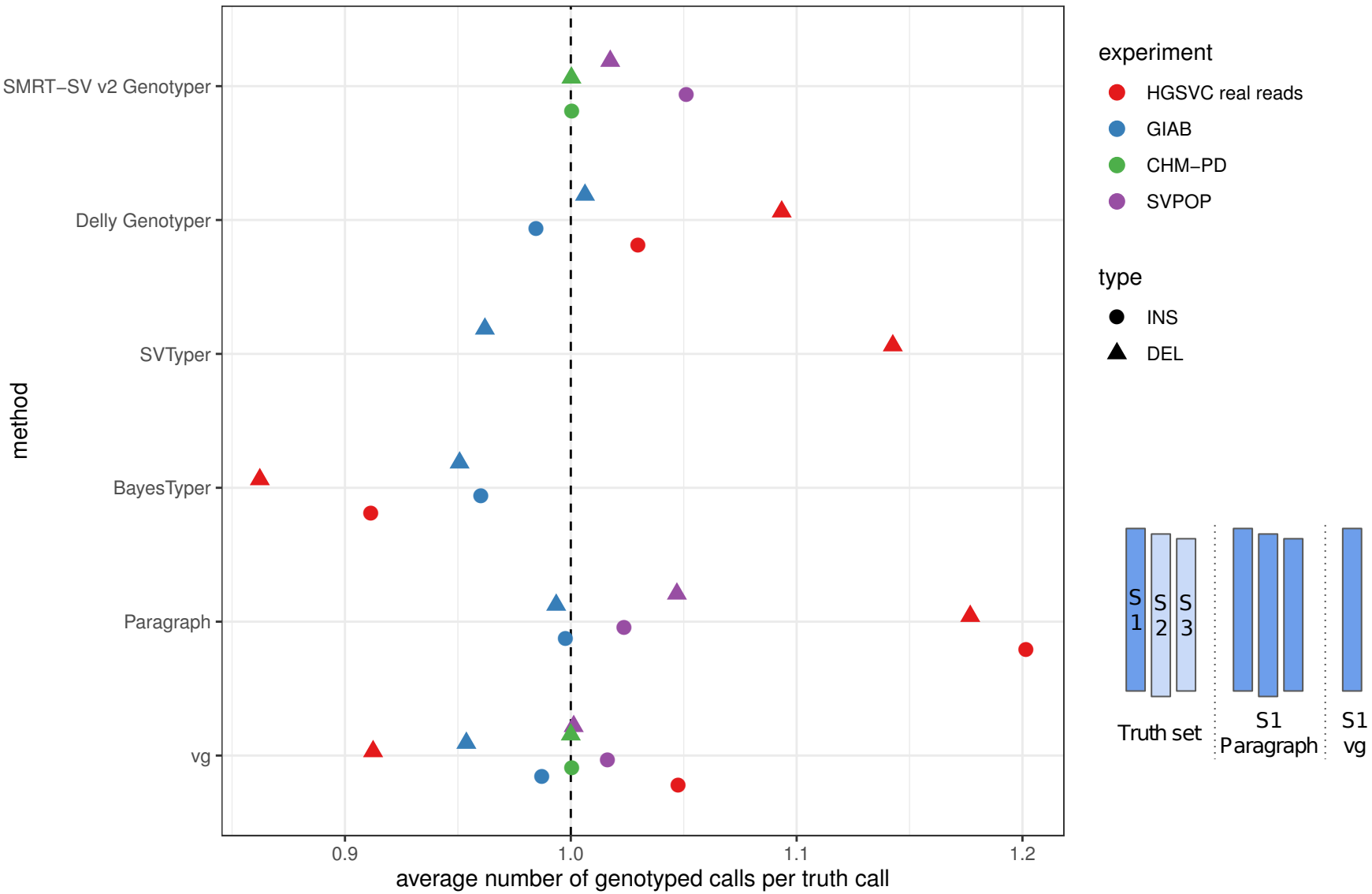
NHGRI

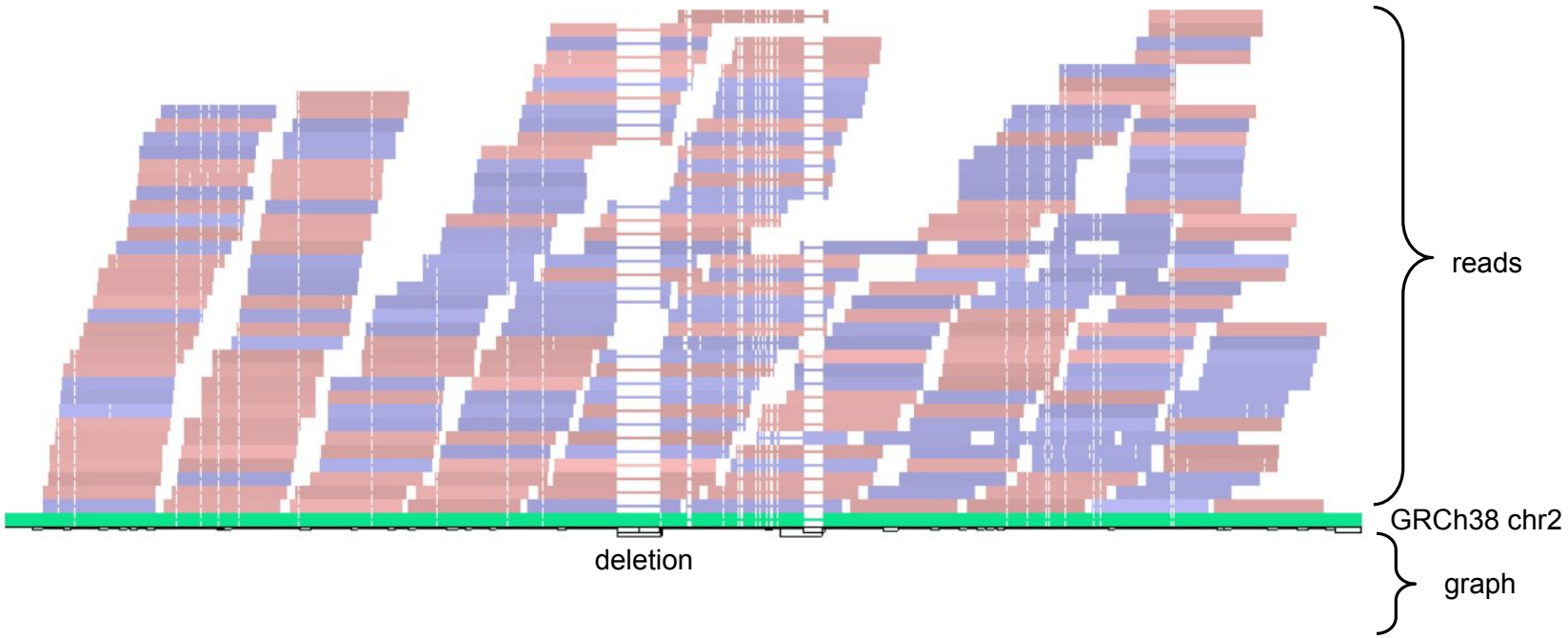
Universal genome graph



Some methods "over-genotype" similar variants

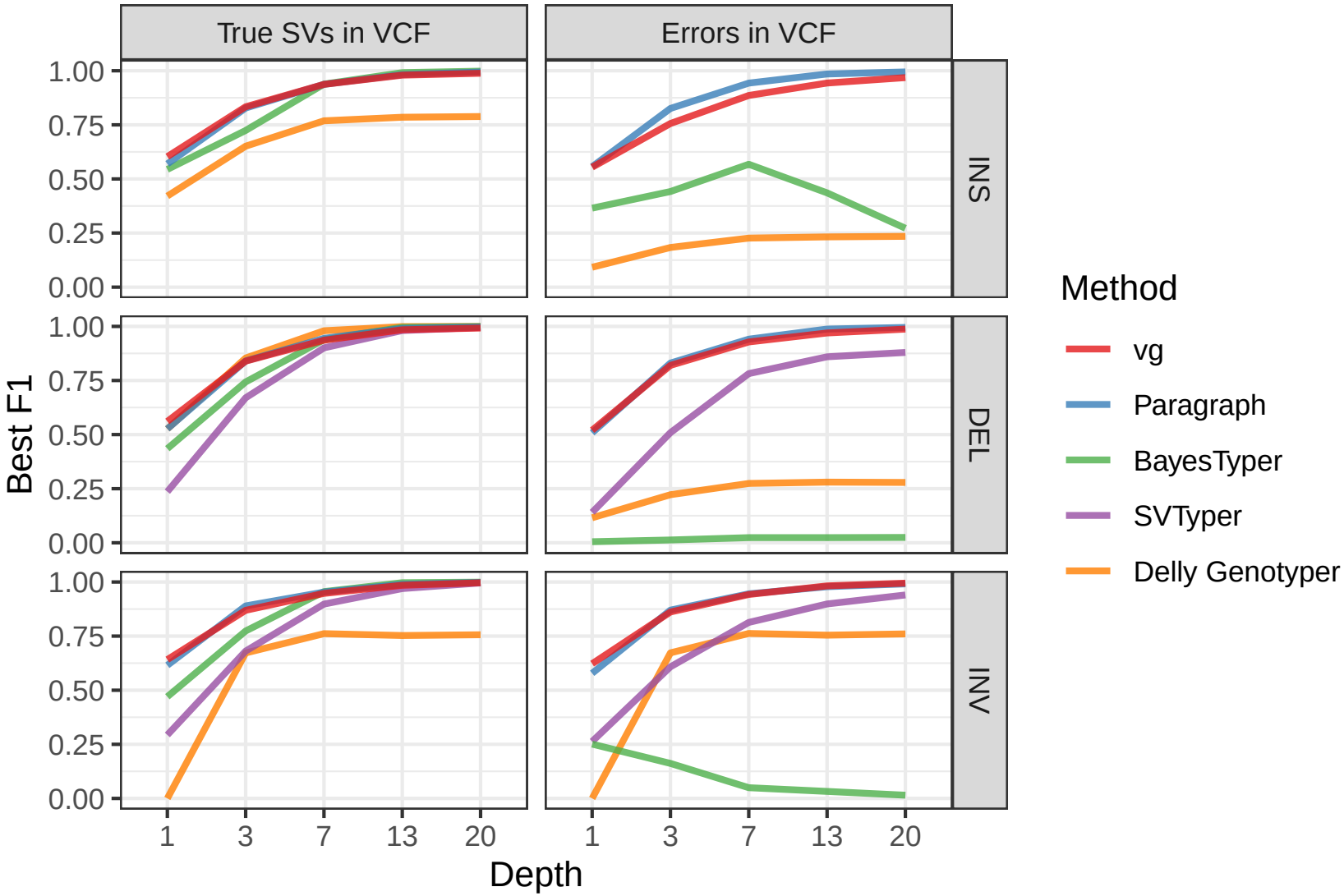


Deletion correctly genotyped by vg

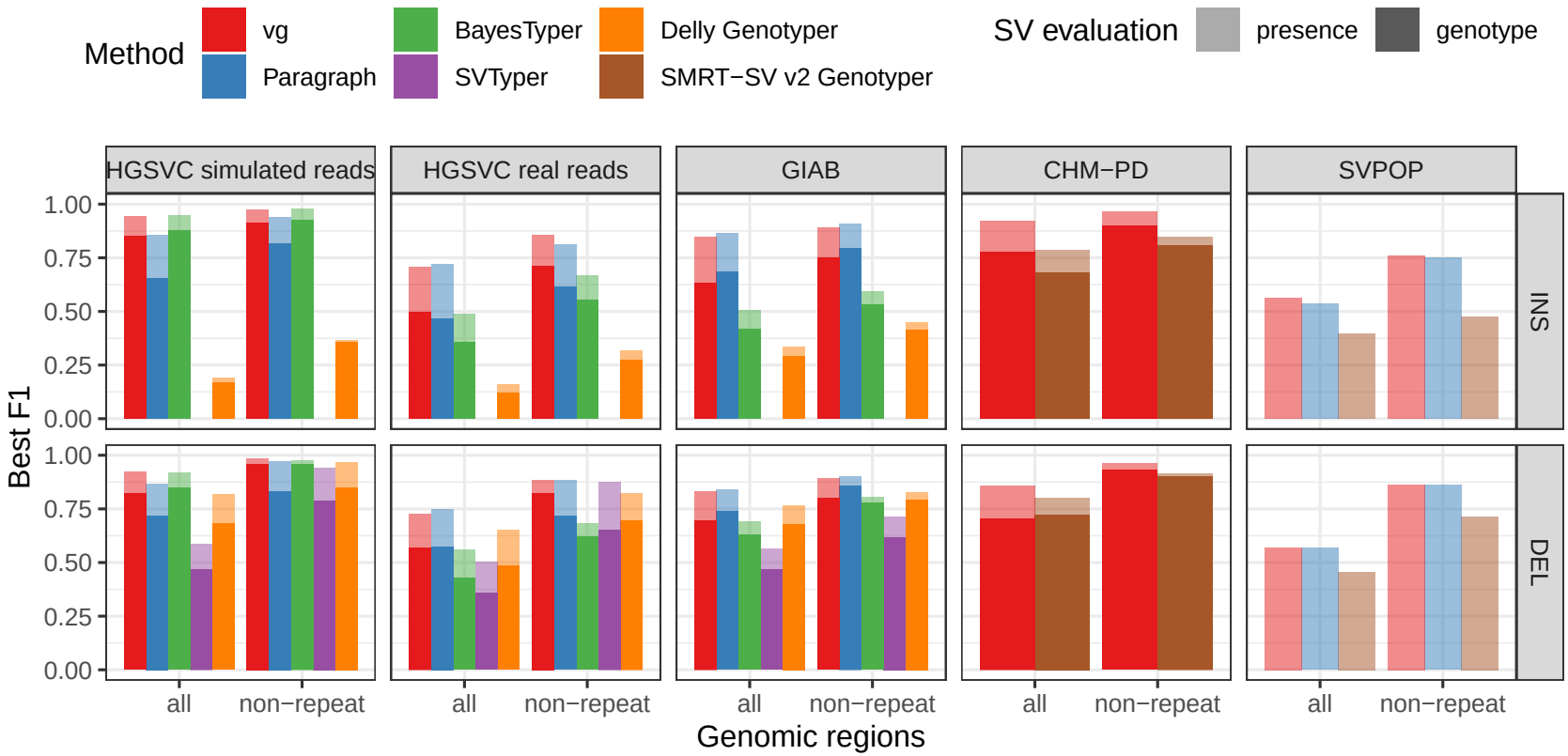


51 bp homozygous deletion in the 3' UTR of the LONRF2 gene.

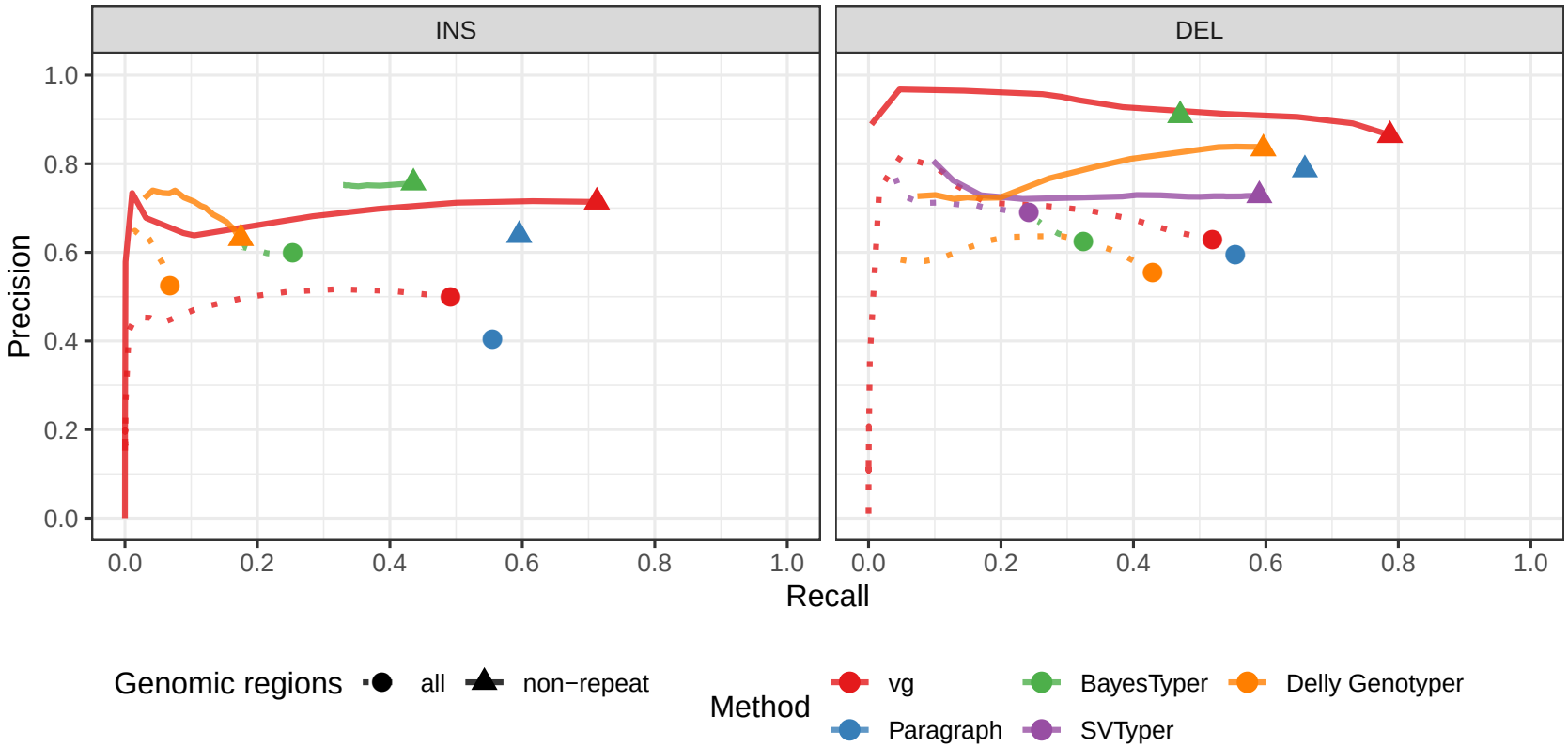
Simulation experiment



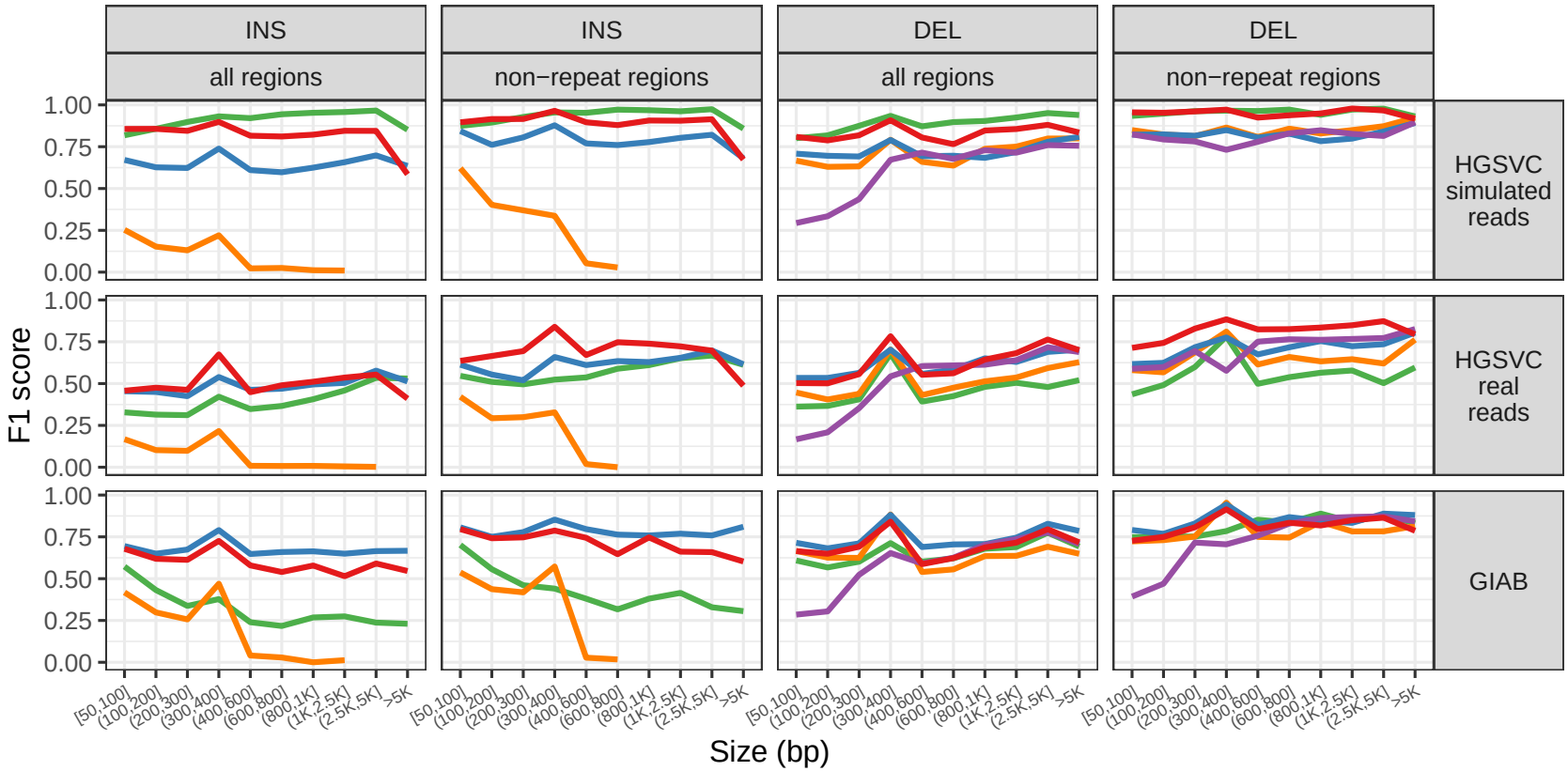
SV catalog summary results



Precision-recall curve



Evaluation per SV size



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