BLENDED GENOME EXOME

Capturing genomic diversity with a novel whole-exome plus low-pass whole genome product

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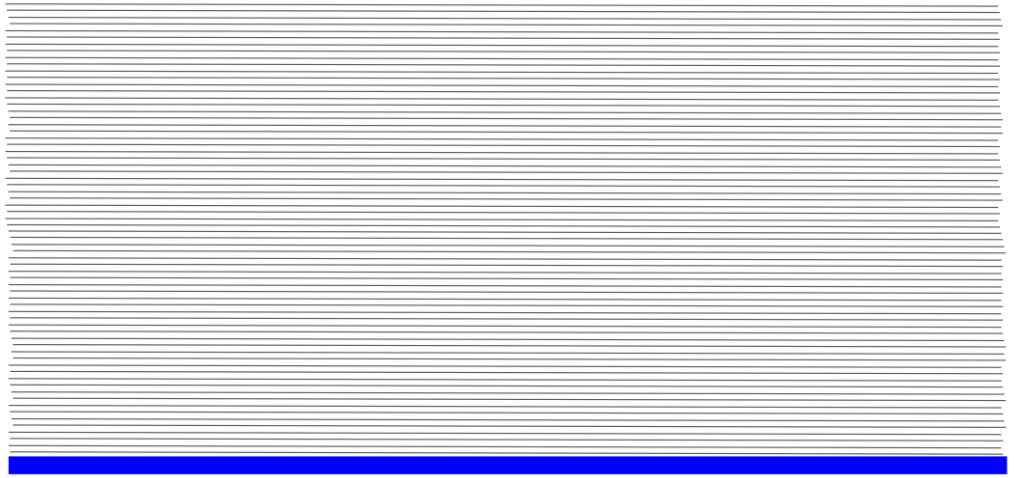
Disclosures

- none

How do we effectively capture the diversity of the genome at scale?

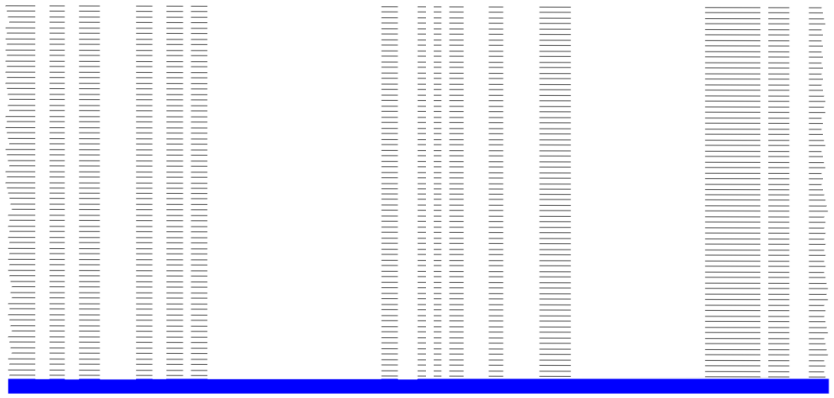
- **Option 1**: Deep whole genomes



- **Problem:** not yet cost-effective to sequence high-coverage whole genomes for large cohorts

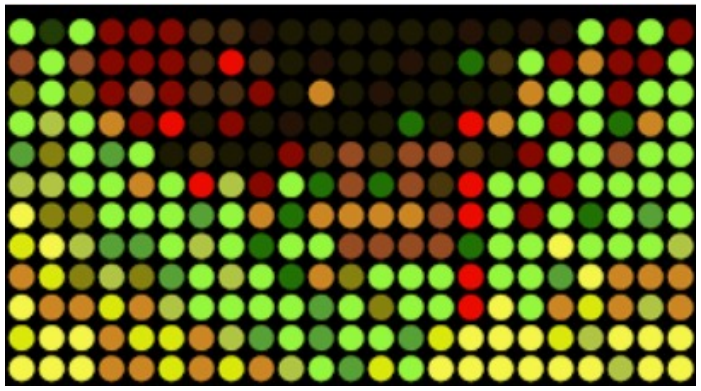
How do we effectively capture the diversity of the genome at scale?

- **Option 2**: Deep exome + GWAS array imputation



Rare coding variants

+



Common variant backbone for imputation

- **Problems**
  - All but the most expensive GWAS arrays biased towards SNPs discovered/common in European ancestries
  - Logistical challenges in harmonizing analyses from separate technologies

Blended Genome Exome (BGE) technology offers a new solution

Solutions:

- Unbiased common variant capture
- Exome and genome in the same sequence run
- Single CRAM/gVCF
- Cost effective alternative to deep WGS or exome + array

$150 per sample

*discounts at bulk sample size

The name? BGE won over:

- *GenEx Hybrid*
- *BEST capture*
- *Blendome*
- *Genxome*
- *Genome McExome Face*

The high-throughput technology behind BGE

Can run over 60 samples through a single lane of sequencing!

Gory details:

- Enzymatic fragmentation (NEBNext Ultra II FS kit)
  - NEB – New England Biosciences
- Quarter reaction volumes
- 384 sample batches (have 192 indexed adapters now)
- 384 well SPRI cleanups
  - SPRI – Solid Phase Reversible Immobilization
- Multiple additions of sample + bead to magnet
- Reduced cost exome capture
- Tempest for fast non-contact dispense destination normalization (384 in minutes!)

Lessons learned from Covid Dx and Covid Seq!

Low pass imputation using GLIMPSE software

GLIMPSE Home Overview Installation Documentation Benchmark GitHub

glimpse

Genotype Likelihoods IMputation and PhaSing mEthod

CATCH A GLIMPSE OF YOUR LOW DEPTH SEQUENCING DATA

GLIMPSE is a phasing and imputation method for large-scale low-coverage sequencing studies.

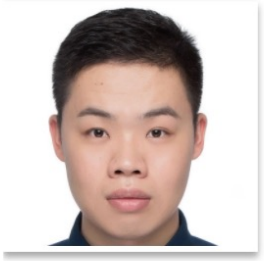
Main features of the method:

1. **Accurate imputed genotype calls.** Our method takes advantage of reference panels to produce high quality genotype calls.
2. **Accurate phasing.** GLIMPSE outputs accurate phased haplotypes for the low-coverage sequenced dataset.
3. **Low-coverage sequencing outperforms SNP arrays.** Imputation using low-coverage sequencing data is competitive to SNP array imputation. Results for European and African-American populations are interactively available on the website.
4. **A cost-effective paradigm.** GLIMPSE realises whole genome imputation from the HRC reference panel for less than 1$.

GLIMPSE tools is available under the MIT licence on the Github repository https://github.com/odelaneau/GLIMPSE.

HUGE thanks to Kai Yuan for being our GLIMPSE workflow expert

THE HUANG LAB



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Kai Yuan is a postdoctoral fellow in the Massachusetts General Hospital and the Broad Institute, advised by Dr. Hailiang Huang. He obtained his PhD in computational biology from the Partner Institute for Computational Biology, Chinese Academy of Sciences. During his PhD training, he worked on population genetics, especially for the admixed populations and developed several methods to infer population

Low pass imputation using GLIMPSE software

"Variable position" = SNP in reference panel, not in sequence data

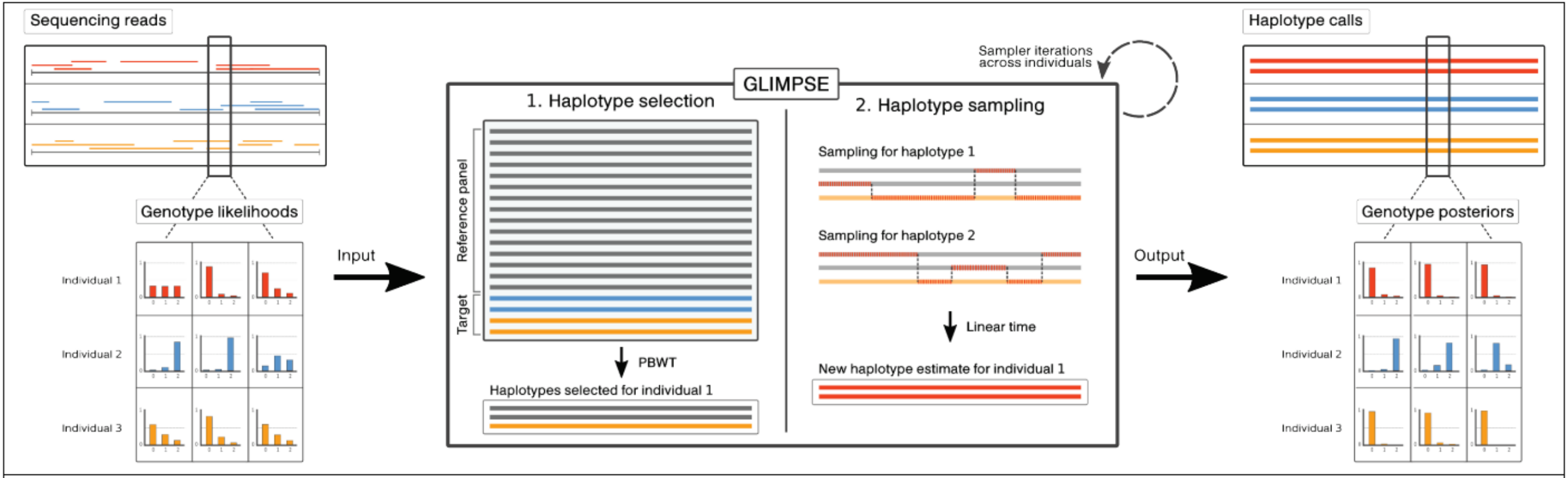
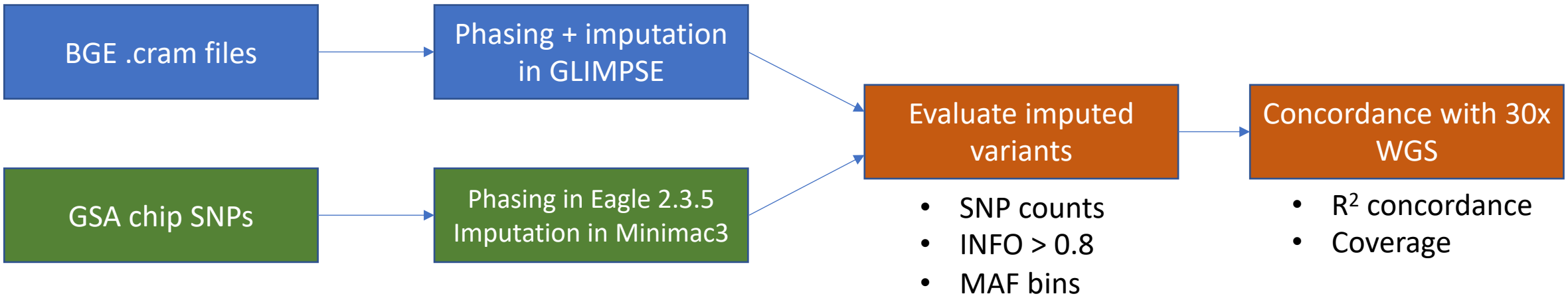


Figure1: GLIMPSE method overview. The input of the method is a matrix of genotype likelihoods defined at all variable positions obtained directly from the sequencing reads (left). GLIMPSE refines the genotype likelihoods using a Gibbs sampler scheme. At each iteration a new pair of haplotypes for each individual is estimated (middle). This involves two main steps: (1.) the haplotype selection using a reference panel and the current estimate of all other target haplotypes (middle, left) and (2.) a linear time sampling algorithm based on the Li and Stephens model (middle, right). As an output, GLIMPSE produces consensus-based haplotype calls and genotype posteriors at every variable position (right).

Evaluating BGE performance using deep whole genomes

- Using high quality calls from 30x genomes as our “truth” dataset
  - Compare low-pass GLIMPSE imputation against Global Screening Array (GSA) chip

- Pilot sample sets
  - Early rounds: 31 to 62 Hispanic samples
  - Later rounds: 23 African samples from PUMAS (Ethiopia and South Africa)



Iterating towards the BGE product

- 31-62 Hispanic samples
- Blood derived DNA
- HRC imputation
- GRCh37/hg19

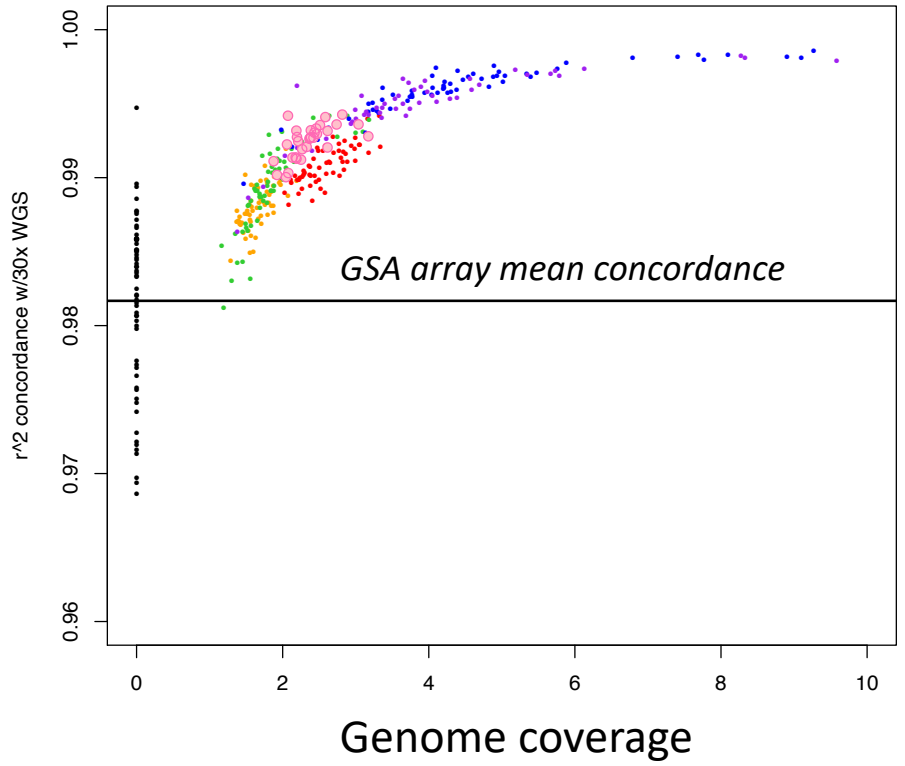
- Started as additional ~200k baits to the exome content (exome + array baits) at different mixtures
- 1x WGS + exome was **not** the focus

Condition	High quality imputed SNPs	Fully concordant SNP count
50-50 mixture	4.8 million	3.3 million
66-34 mixture	4.5 million	2.9 million
80-20 mixture	3.3 million	1.6 million
1x WGS + GLIMPSE	**11.3 million**	**7.6 million**
GSA chip	7.8 million	5.8 million

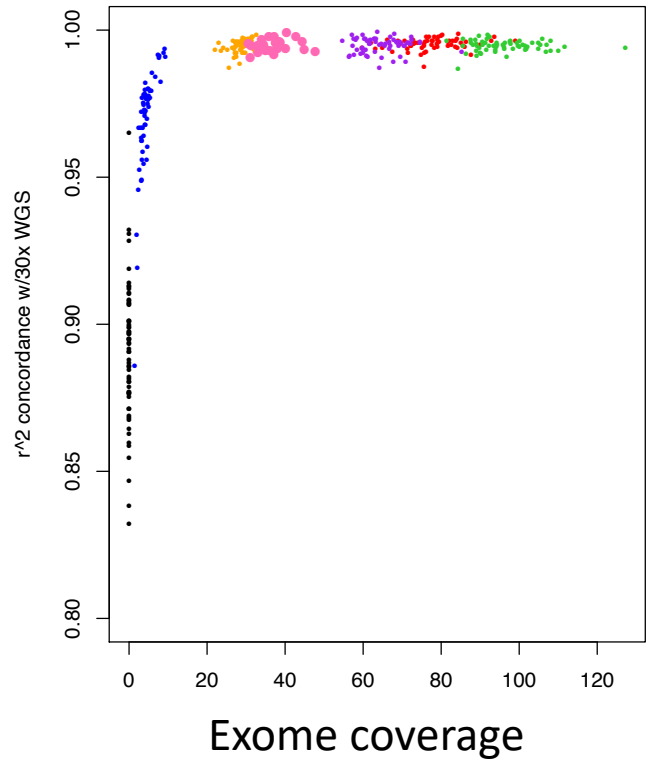
Finding the optimal genome/exome coverage

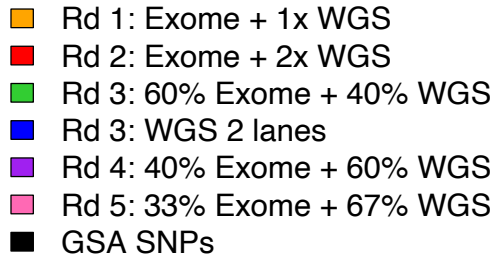
- 31-62 Hispanic samples
- Blood derived DNA
- HRC imputation
- GRCh37/hg19

Genome concordance



Exome concordance

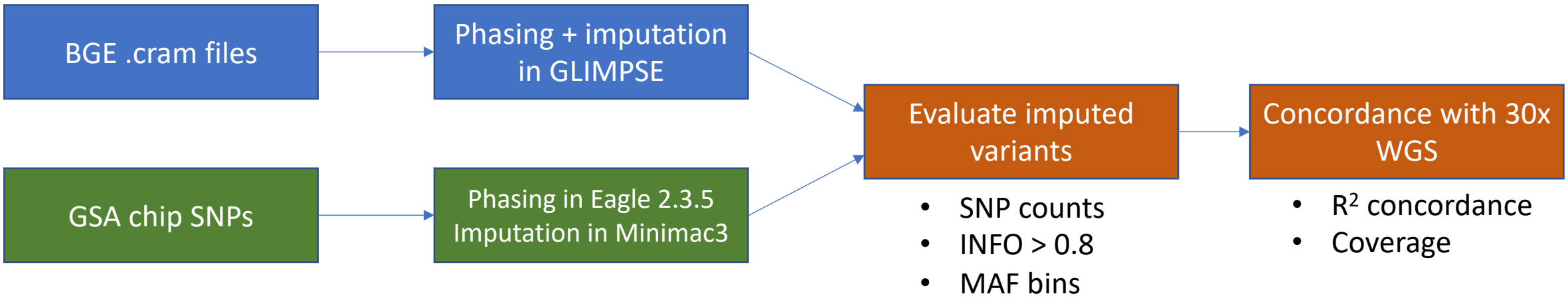




Results from the BGE pilot dataset

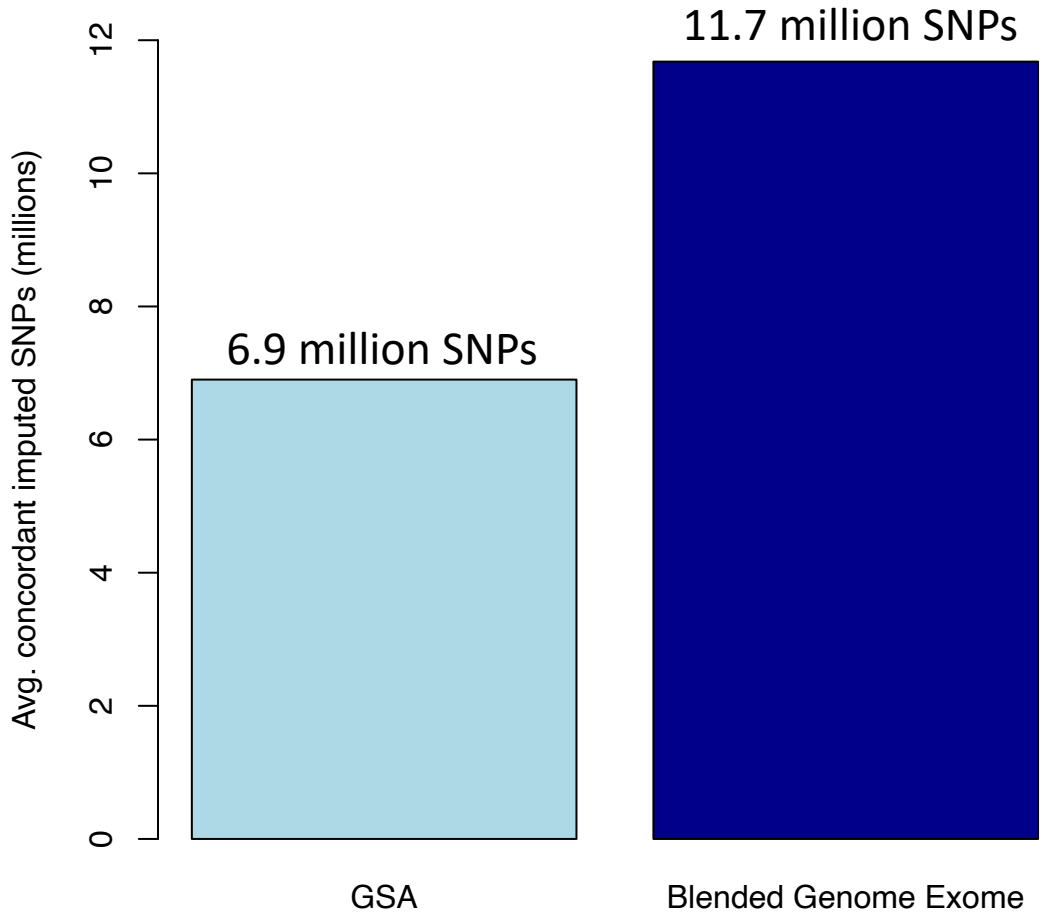
- 764 participants
  - 64 NeuroGAP South Africa (UCT)
  - 317 NeuroGAP Ethiopian (AAU)
  - 381 China (BioX)

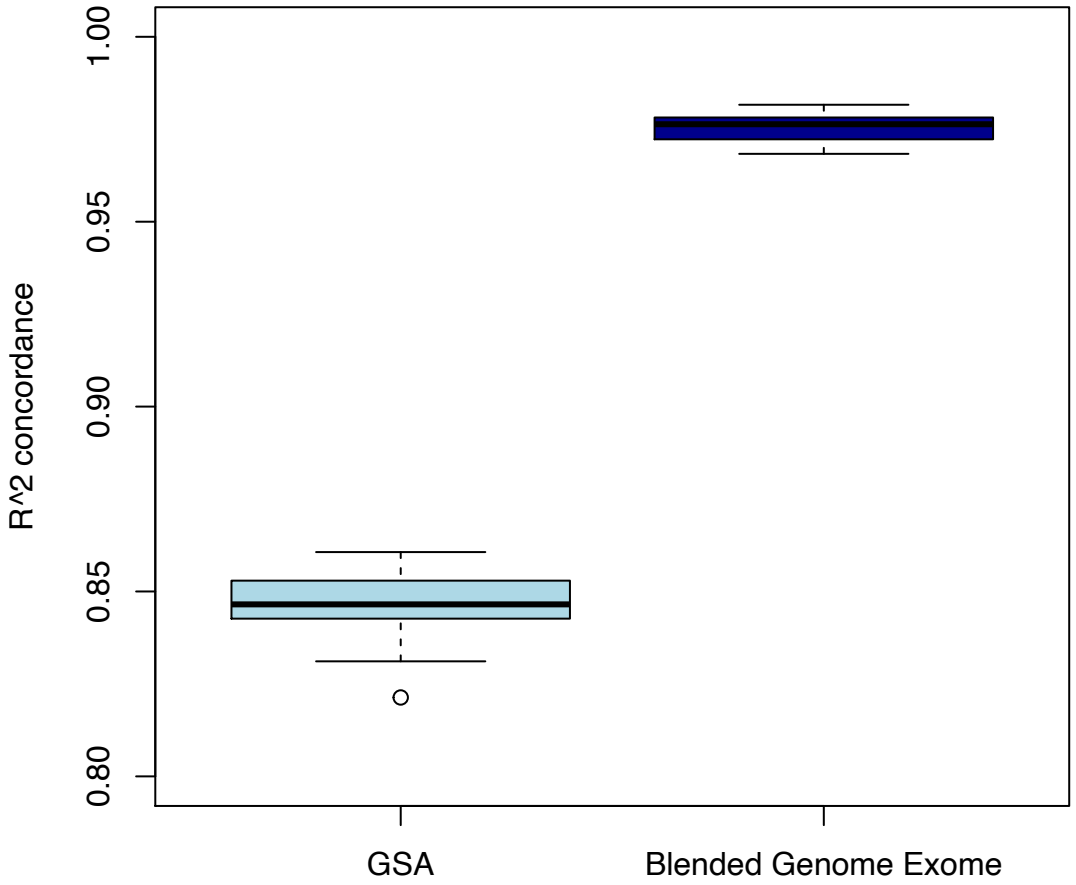
- 23 participants also have deep WGS (30x coverage) for concordance comparison
  - 13 NeuroGAP South Africa (UCT)
  - 10 NeuroGAP Ethiopian (AAU)



BGE has more SNPs and higher concordance than current GSA platform

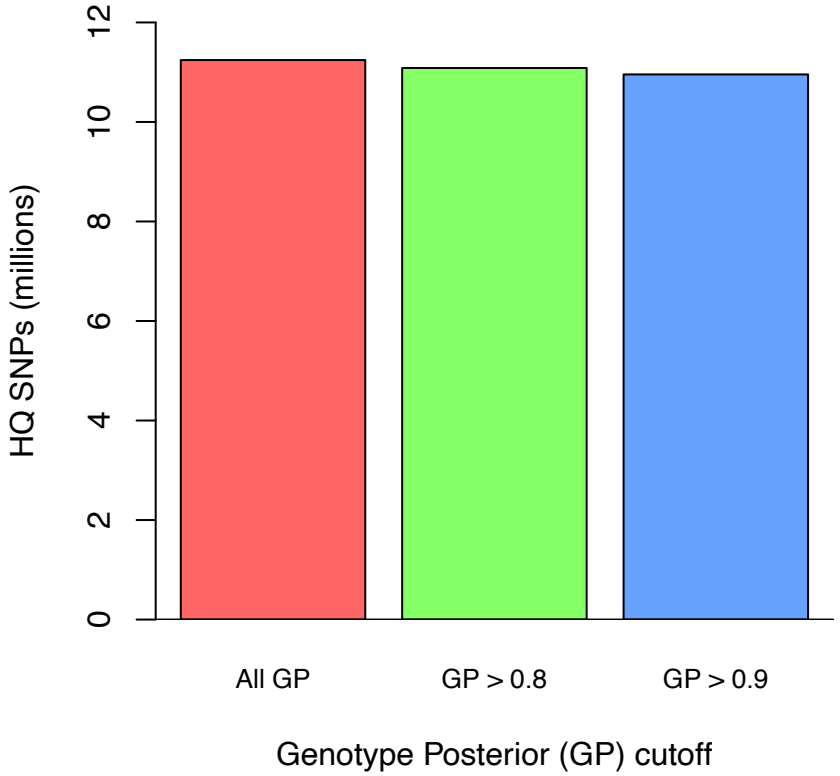
- 23 African samples
- Saliva derived DNA
- HRC imputation
- GRCh38/hg38

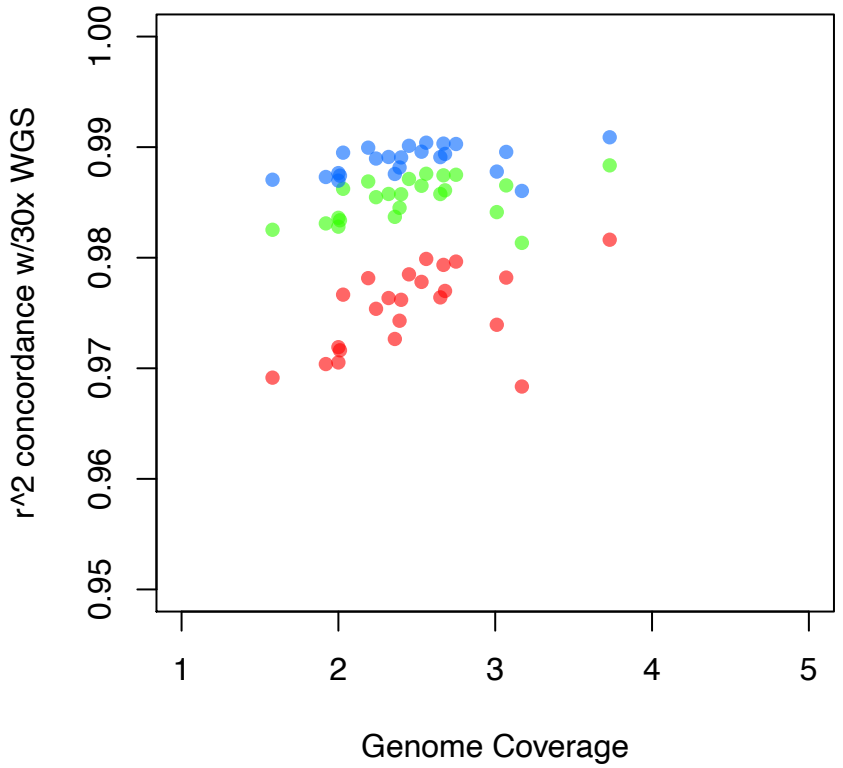


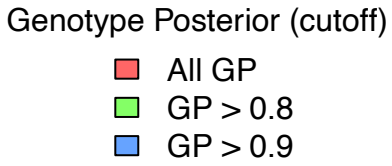


Restricting to higher genotype posterior cutoffs improves concordance with 30x genomes

- 23 African samples
- Saliva derived DNA
- HRC imputation
- GRCh38/hg38



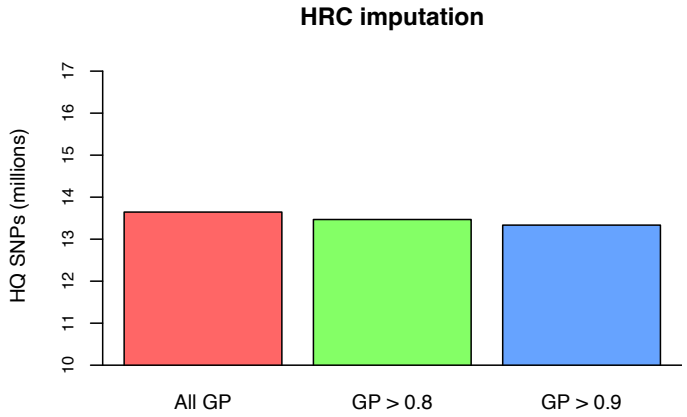


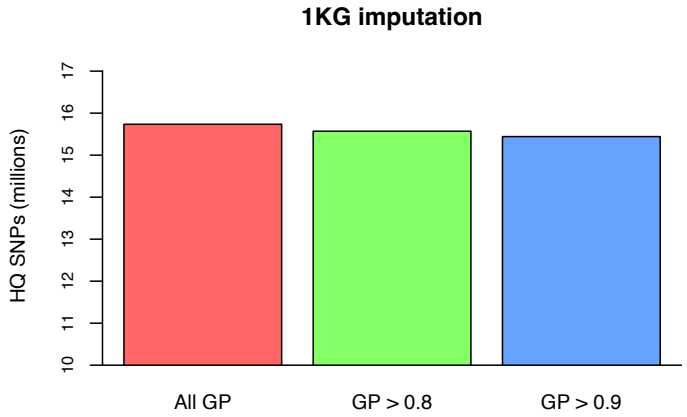


Choice of reference panel matters
1000 genomes (1KG) gives us 3 million more SNPs than HRC

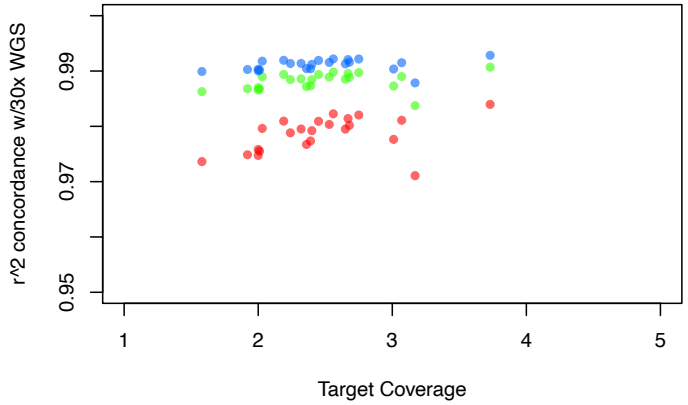
- 23 African samples
- Saliva derived DNA
- HRC/1KG imputation
- GRCh38/hg38

NOTE: Moving to calls from full 764 sample pilot cohort

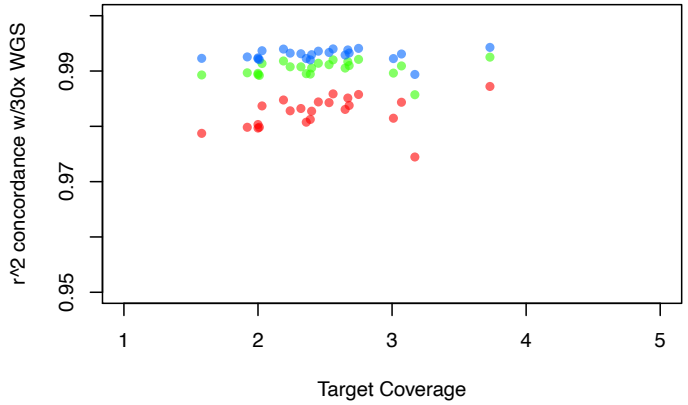


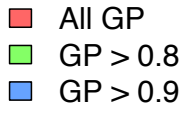


HRC ref panel
39 million SNPs



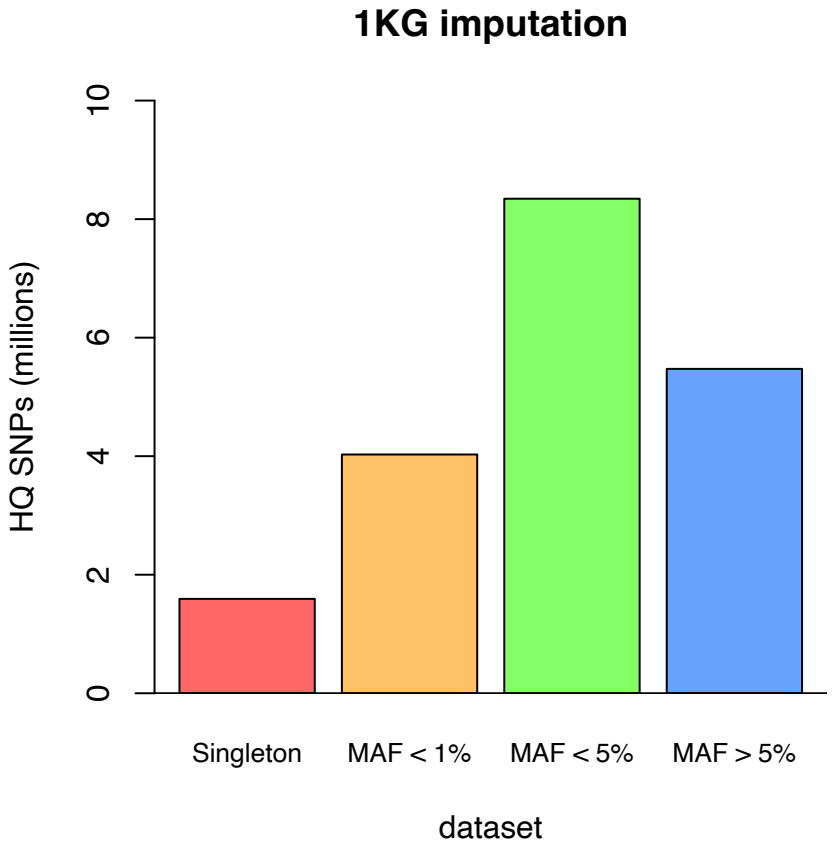
1KG ref panel
62 million SNPs

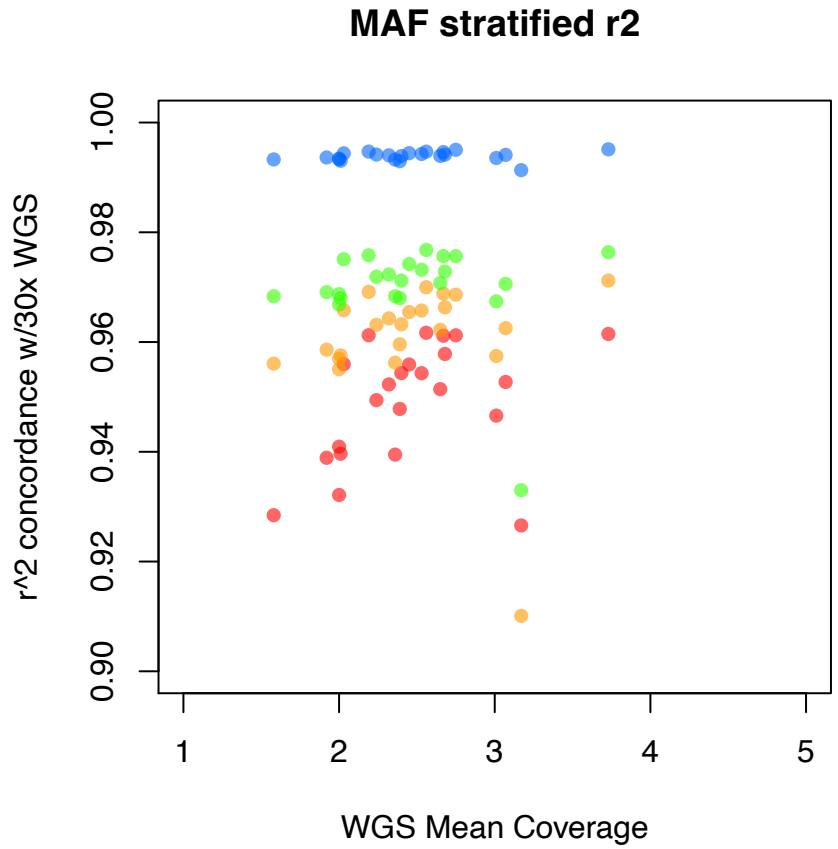




Are we capturing lower frequency variants well?

- 23 African samples
- Saliva derived DNA
- 1KG imputation
- GP > 0.9 calls



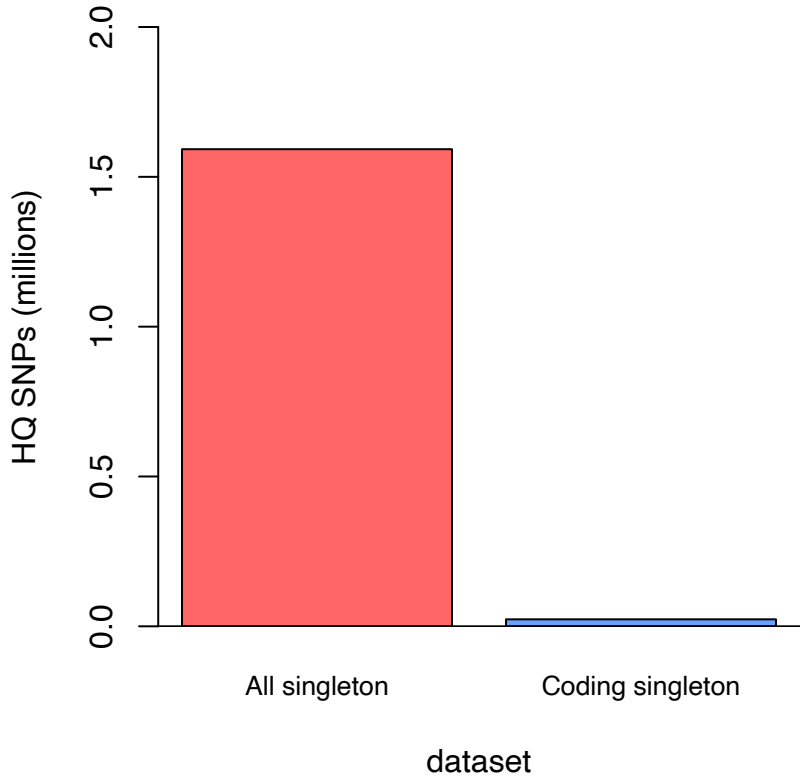


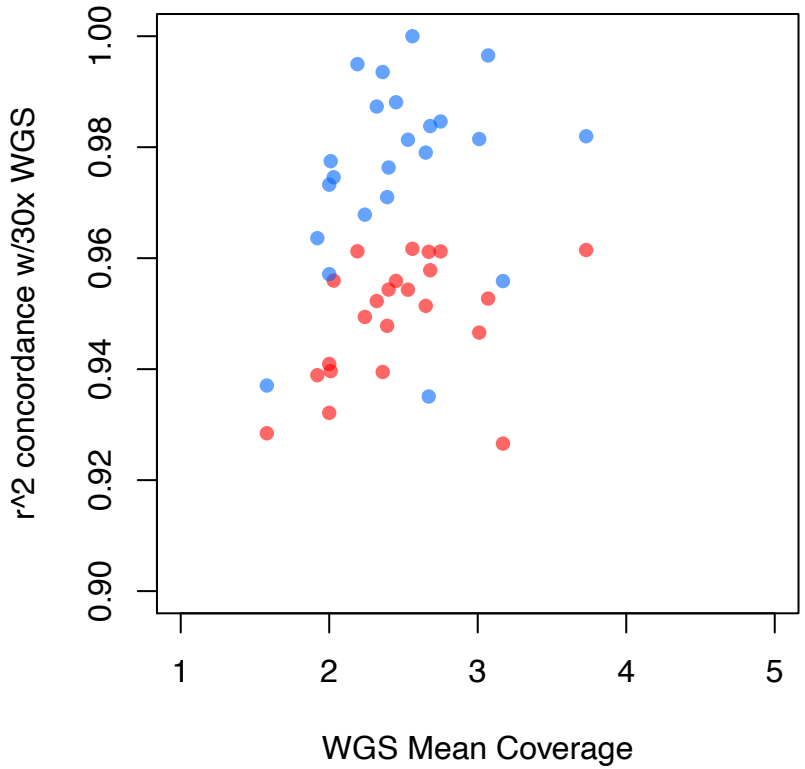
Singleton
MAF < 1%
MAF < 5%
MAF > 5%

MAF estimates from 371 NeuroGAP African samples

Are we imputing rare coding variants well?

- 23 African samples
- Saliva derived DNA
- 1KG imputation
- GP > 0.9 calls



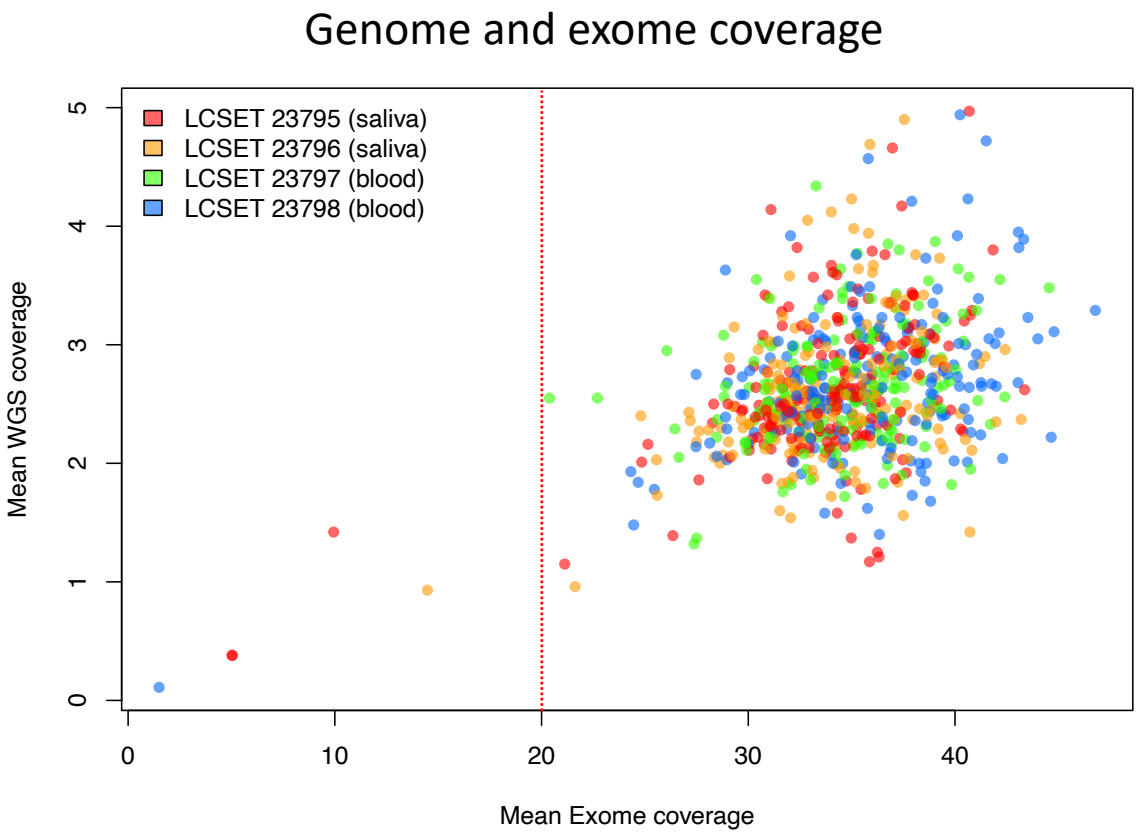


All singleton
Coding singleton

61% of coding singletons captured in 1KG ref panel

GLIMPSE imputation across ancestry subsets

- 764 pilot samples
- 1KG imputation
- All calls



SNPs (in millions)

Location	Sample size	INFO > 0.6	INFO > 0.8	INFO > 0.9
South Africa	61	17.9	16.5	14.5
Ethiopia	317	22.8	20	17.2
East Asia	381	19.3	16.6	13.9
Combined	**764**	**25.1**	**21**	**17.3**

GLIMPSE runtime is robust to sample size

- 764 pilot samples
- 1KG imputation
- All calls

Full Sample run time - each job capped at 8GB max RAM

GLIMPSE step	Jobs submitted	Run Time (CPU hrs)
cram2GL	16808	1370.36
VcfCombine	22	184.75
GetSiteInfo	22	3.20
GenomeChunked	22	0.06
ChunkImpute	952	8732.44
ChunkLigate	22	7.40
Phase	22	2.01

Cohort-specific run time
Jobs capped at 4GB max RAM

Location	Sample size	Run Time per sample
South Africa	61	**12.5**
Ethiopia	317	**12.1**
East Asia	381	**11.3**

BLENDED GENOME EXOME

- Unbiased common variant capture
- Deep whole exome
- More SNPs and better accuracy than standard array
- Cost effective

BGE core dev team

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BGE core analysis/feedback

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- Laura Gauthier
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- Karesten Koenen
- Rocky Stroud
- Anne Stevenson
- NeuroGAP participants
- BioX participants

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