ALFA: allele frequency data for variant analysis and interpretation

- Closed captioning: www.captionedtext.com & enter 4376574
- The **recording of this webinar** will be on our YouTube channel in the Webinars playlist in a few days: <youtube>/user/NCBINLM/playlists
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NCBI's ALFA: allele frequency data for variant analysis and interpretation

Lon Phan – NCBI dbSNP resource lead

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Outline

- ALFA Introduction
- Data Release Summary
- Data Access Tutorials
  - Web
  - API
  - FTP
- Conclusion and Q&A

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Allele Frequency Aggregator (ALFA)

https://www.ncbi.nlm.nih.gov/snp/docs/gsr/alfa/

- The goal is to provide allele frequency from more than 1 million subjects dbGaP subjects
- Aggregating heterogenous data from dbGaP studies include chip array, exome, and genomic sequencing
- Variant, population, and frequency data normalized and provided in standard exchange formats.
- The data will be integrated with dbSNP regular build release with assigned RS accessions for variants and available for access by web, FTP, and API.

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ALFA Process Overview

Preprocessing → **Computation** → **Product Release**

Preprocessing
- 'Approved' Study selection
- Study QA/QC
- GRAF-Pop ancestry calculation
- Data conversion to VCF

Computation
- Normalization of variants
  - SPDI[1] conversion
  - Remapping
  - VOCA[1]
  - RefSNP assignment
- Aggregation
  - Individuals and genotype
  - Populations
- Compute frequency

Product Release
- RefSNP Web page
- API
- FTP

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[1]Holmes et al. https://pubmed.ncbi.nlm.nih.gov/31738401/

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ALFA Release Summary (March 10,2020)

Inputs	
Studies	42
Subjects	98,494
Genotypes	551,345,630,054

Outputs	
Unique Variants	446,909,594
• Exist in dbSNP	442,539,572
• Novel	4,370,022

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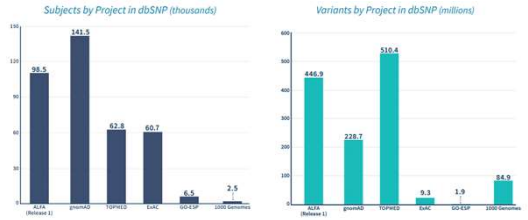
P[4 Improved precision and accurarcy of RS assignment; reducing duplicates.

Phan, Lon (NIH/NLM/NCBI) [E], 4/21/2020

dbSNP Allele Frequency



ALFA (release 1) has ~10% of dbGaP 1M potential input subjects with genotype analyzed by Graf-Pop

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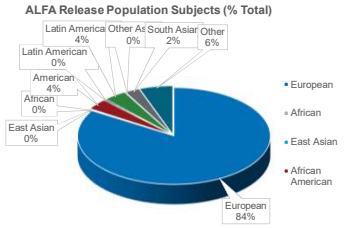
Population Summary



Population	Subjects
European	82,475
African[1]	114
East Asian	153
African American	3,555
Latin American 1	354
Latin American 2	3,801
Other Asian	84
South Asian	2,459
Other	5,499
ALFA Total	**98,494**
dbGaP Total	**2,491,605**
dbGaP Total (Unique)	**1,397,069**

[1] Included both African and African-Americans

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https://www.ncbi.nlm.nih.gov/snp/docs/gsr/data_inclusion/#population

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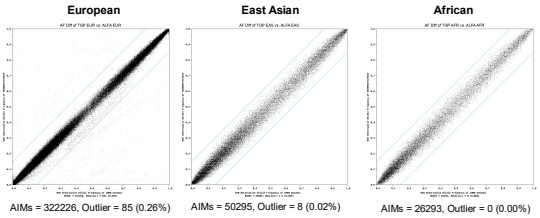
Concordance between AFLA and 1000 Genomes Populations



AIMs = 322226, Outlier = 85 (0.26%)

AIMs = 50295, Outlier = 8 (0.02%)

AIMs = 26293, Outlier = 0 (0.00%)

- > 95% concordance with GnomAD and TopMed

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ALFA Variants of Interest

Variant Source	ALFA RS Count	Percent(%) of Total	Total RS
ClinVar	229963	59	387427
GTR	366	77	474
GWAS Catalog (p-value < 10^-5)	21494	98	21972
Pubmed Cited	188809	82	231502

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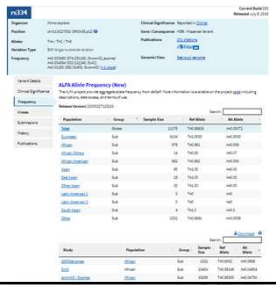
ALFA frequency reported on RefSNP Page



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Search dbSNP Web

https://www.ncbi.nlm.nih.gov/snp/docs/entrez_help/

- RS number
- Position (GRCh37 and GRCh38)
- Gene
  - Symbol
  - ID
- And other attributes
  - Frequency
  - ClinVar clinical significance
  - Many others...

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P[5 Variants of high interest

Phan, Lon (NIH/NLM/NCBI) [E], 4/21/2020

Sample Variant Search

https://www.ncbi.nlm.nih.gov/snp/docs/entrez_help/

- ClinVar Variant
- Conflicting interpretations of pathogenicity
- Cited in PubMed
- Missense variant
- Cluster Validation (observed by multiple submitters)
- NO frequency from other projects (1000G, GnomAD, etc.)

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Generate Query Term

(((conflicting interpretations of pathogenicity[Clinical_Significance] AND snp_pubmed_cited[Filter] AND missense variant[Function_Class] AND by cluster[Validation]))) NOT "by frequency"[Validation Status]

- Using Filters
- Using Advance Search

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Search dbSNP using eUtils

https://www.ncbi.nlm.nih.gov/snp/docs/entrez_help/

Use same web query syntax

```
eShandle = Entrez.esearch(db="snp",  # search dbSNP
                          #complex query for missense and pathogenic variants in LPL gene with global MAF betweeen 0 and 0.01.
                          term='(((conflicting interpretations of pathogenicity[Clinical_Significance] AND snp_pubmed_cited[Filt
er] AND missense variant[Function_Class] AND by cluster[Validation]))) NOT "by frequency"[Validation Status]',
                          usehistory="y", #cache result on server for download in batches
                          retmax=20 # return 20 RSID max
                         )
```

https://github.com/ncbi/dbsnp/blob/master/tutorials/eUtils.ipynb

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NCBI Variation Service API

www.ncbi.nlm.nih.gov/variation/v0/

GET	/metadata/frequency	Retrieve frequency metadata
GET	/interval/{seq_id}:{position}:{length}/overlapping_frequency_records	Retrieve frequencies for variants overlapping a position range
GET	/refsnp/{rsid}/frequency	Lookup frequency data for specified RSID

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NCBI Variation Service API Tutorials

https://github.com/ncbi/dbsnp/tree/master/tutorials/Variation%20Services/Jupyter_Notebook

- Launch `by_gene.ipynb` notebook on Binder: launch binder

Retrieve frequency data by gene using eUtils and Variation Service

- Launch `by_rsid.ipynb` notebook on Binder: launch binder

Retrieve frequency data by rsid in JSON format

- Launch `frequencies_for_vcf.ipynb` notebook on Binder: launch binder

Compare frequencies for two populations for variations from a VCF file

- Launch `metadata_as_hash.ipynb` notebook on Binder: launch binder

Retrieve and transform ALFA project and population meta data

- Launch `plot.ipynb` notebook on Binder: launch binder

Plot population minor allele frequencies (MAFs) for variants in a gene location

- Launch `querying_subsets_ftp.ipynb` notebook on Google Colab

Retrieving subsets of VCF data from remote NCBI FTP site

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Conclusions

- New source of frequency data from dbGaP growing catalog of studies
- Quarterly updates with new dbSNP and dbGaP data
  - New unrestricted studies
  - Novel variants
  - Updated population frequency
- Normalized data with improved quality and precision reported in standard formats
- A beacon to data of interest in dbGaP

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- URLs & Places to Learn More -

- NCBI Insights Blog: ncbiinsights.ncbi.nlm.nih.gov
- Learn: www.ncbi.nlm.nih.gov/home/learn.shtml
- Factsheets: ftp.ncbi.nih.gov/pub/factsheets/
- NCBI YouTube Channel: www.youtube.com/ncbinlm
- NCBI Helpdesk: info@ncbi.nlm.nih.gov

Questions?

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