Development of population frequency thresholds specific for rare neurodevelopmental disorders

Dana Burow, Katie Yergert, Claire Watson, Melissa Samons, Matthew Johnson, Laura Hudish, **Audrey O'Neill**, Steven Harrison, Wendy Alcaraz

Ambry Genetics, Aliso Viejo, CA | aoneill@ambrygen.com

BACKGROUND

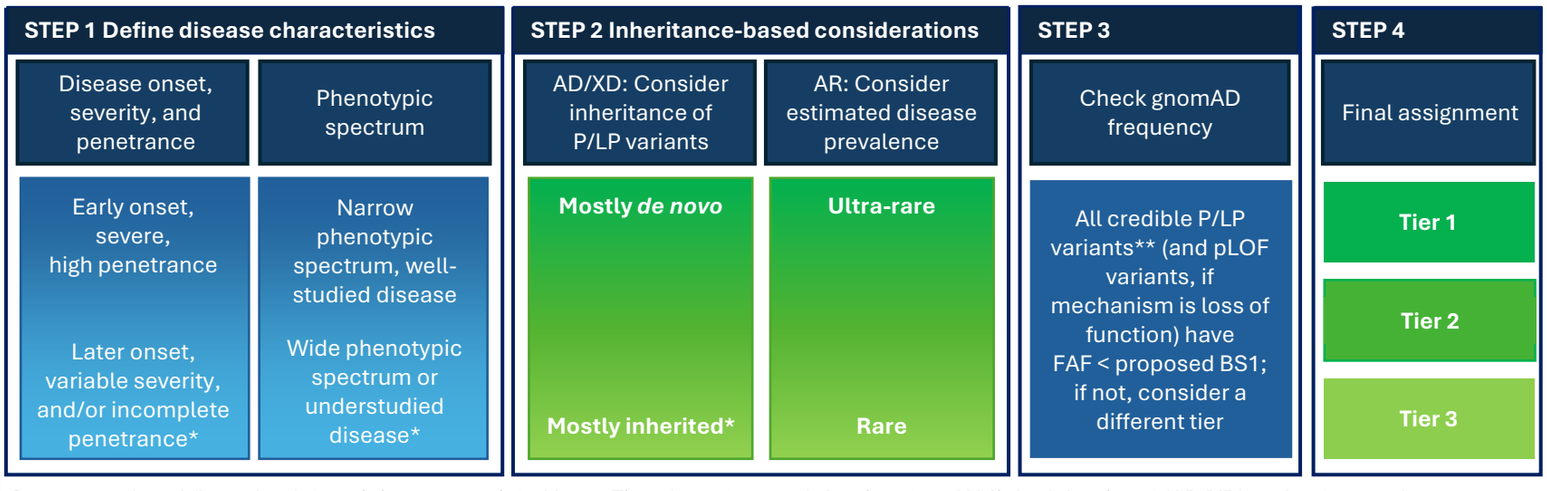
- Accurate population frequency thresholds are critical for application of benign evidence and determination of rarity
- Current approaches for setting specific population frequency thresholds rely on knowledge of gene-disease architecture (e.g., disease prevalence and penetrance) and/or characterized pathogenic variants[1,2]
- These approaches are not well suited to many neurodevelopmental disorders (NDD), which tend to be rare, understudied, and/or characterized by variable expressivity, high genetic heterogeneity, and relatively non-specific clinical features

TABLE 1: NEURO TIER THRESHOLDS

Tier	Rarity MAF or AC	BS1 FAF or AC	BA1 FAF or AC
AD/XD tier 1	0	2	5
AD/XD tier 2	1*	0.00001 or 5	10
AD/XD tier 3	0.000001	0.00001	0.0001
AR tier 1	0.000105	0.000105	0.00105
AR tier 2	0.00033	0.00033	0.0033
AR tier 3	0.00105	0.00105	0.0105

*if condition is XD, this allele should be heterozygous (not hemizygous)

FIGURE 1: WORKFLOWS FOR SELECTING THRESHOLDS



Summary of workflows for determining appropriate Neuro Tiers for autosomal dominant and X-linked dominant (AD/XD) and autosomal recessive (AR) gene-disease relationships

*if affected individuals are NOT expected to be excluded from gnomAD, Neuro Tiers are likely not appropriate

** known founder variants may be excluded

METHODS

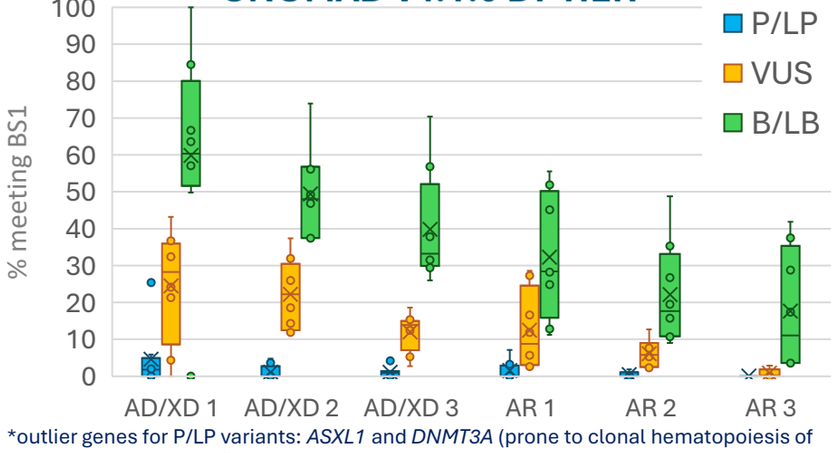
Development of Neuro Tiers:

- We developed a set of population frequency bins, termed "Neuro Tiers", to categorize gene-disease relationships based on inheritance pattern and phenotypic severity (see Figure 1).
- A panel of internal experts discussed and refined the criteria for each bin for BA1, BS1, and PM2 thresholds, based on GrpMax filtering allele frequency total (FAF) or minor allele frequency (MAF) and/or allele count (AC) (see Table 1). Tiers are assigned on a per-gene basis during variant assessment and/or gene curation (see Figure 1). Thresholds were tested across 80 genes using over 400 internally classified variants and gnomAD v4.1.0 data.

gnomAD/ClinVar pilot study:

- For each Neuro Tier, 8 genes were randomly selected.
- Variants classified as Pathogenic/Likely Pathogenic (P/LP), Uncertain Significance (VUS), and Benign/Likely Benign (B/LB) in ClinVar for the MANE transcript for each gene were quantified and compared to gnomAD v4.1.0 (filtered variants excluded).
- The AC and/or FAF for each P/LP, VUS, or B/LB variant was compared to the Neuro Tier threshold for BS1. Percents of variants in a gene meeting BS1 are shown in Figure 2.

FIGURE 2: VARIANTS MEETING BS1 IN GNOMAD v4.1.0 BY TIER



*outlier genes for P/LP variants: *ASXL1* and *DNMT3A* (prone to clonal hematopoiesis of indeterminate potential[3]); variants in these genes undergo extra gnomAD quality checks

TAKE HOME POINTS

- Tiered population frequency thresholds for NDD genes distinguish between pathogenic and benign variants (Fig 2)
  - On average, **1.5% of P/LP variants and 37% of B/LB** variants meet BS1
  - Note that most P/LP variants are absent in gnomAD
  - An average of **13% of VUS meet BS1** and can likely be downgraded
- **Use of gnomAD v4.1.0 data** can greatly expand the number of variants eligible for downgrade (Fig 2)
- **Caveat:** use of ClinVar and gnomAD data in Fig 2 is subject to various sources of error

REFERENCES

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