Poster eP368

A Streamlined Process for Assessing the Strength of a Relationship Between a Gene and Specific Disease

C Lisa Kurtz[1] and Michelle Paczosa[1]
Quest Diagnostics, Secaucus, NJ. [1] These authors contributed equally to this work.

ACMG Annual Clinical Genetics Meeting
Nashville, TN | March 23-25, 2022

Correspondence: C Lisa Kurtz
Catherine.L.Kurtz@QuestDiagnostics.com

Introduction

- Assessing the strength of the association between a gene and a phenotype is essential for variant classification.
- This categorization determines whether certain types of evidence can be applied when scoring variants, which can help reduce the frequency of "variant of uncertain significance" classifications.
- Existing methods for assessing gene-disease relationships are often laborious and time-consuming. Being able to return results faster, while retaining standardization, would improve patient care and accelerate research.
- Here, we developed a streamlined, quantitative process to categorize the strength of a gene-disease relationship and compared it to existing methods.

Process

- First, the gene, phenotype, and mode of inheritance are specified.
- Next, if ≥72 total variants (or ≥18 loss-of-function [LOF] variants, in genes where LOF is the mechanism of disease) are associated with the specified phenotype in HGMD, the relationship is considered **Automatic Strong**.
- Otherwise, the number of probands and additional evidence in the primary literature are quantified (**Table 1**), and the relationship is categorized according to the number of total points awarded (**Figure 1**).

Process (continued)

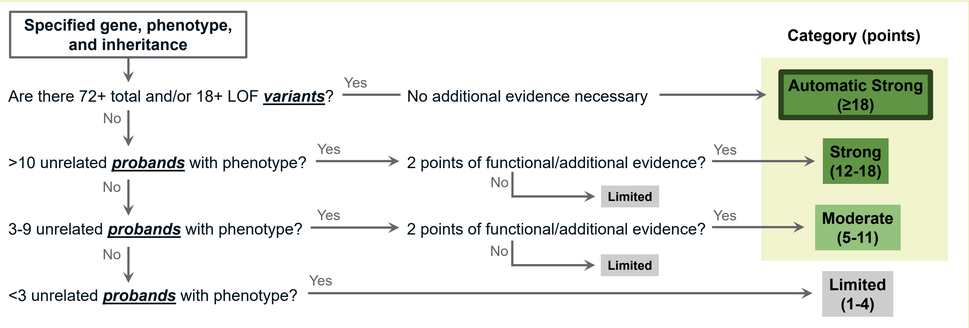

Figure 1. Streamlined process for categorizing gene-disease relationship strength.

- **Strong:** >10 probands +2 points of additional evidence
- **Moderate**: 3-9 probands +2 points of additional evidence
- **Limited:** <3 probands
- Relationships with <2 points of additional evidence are also categorized as Limited, regardless of the number of probands.
- Gene-disease relationships of Moderate strength or stronger are considered well-established, and supporting evidence can be applied toward variant pathogenicity.

Table 1. Scoring of Evidence for Gene-Disease Relationship Assessment[1]

Evidence type	Conditions	Points
Proband	Each unrelated individual with the phenotype of interest	+1
Evidence categories below are capped at +2 points total		
Functional study[a]	Alteration of normal function consistent with disease mechanism	+2
	Alteration of normal function consistent with disease but experimental design not optimal or results only consistent with a subset of phenotypic features	+1
Segregation[b]	A variant segregates with disease with a LOD score > 2 in multiple families	+2
	A variant segregates with disease with a LOD score between 1.0 (0.9 in a dominant gene) and 2.0 or >2 in a single family	+1
Association[b]	Association is statistically significant in multiple families with the same variant, $P<0.05$	+2
	Association is statistically significant in only a single family, $P<0.1$ or $P<0.05$	+1
De novo	Each confirmed *de novo* occurrence	+1
	Each unconfirmed *de novo* occurrence	+0.5
Co-occurrence	(Same) variant in AR disorder co-occurs with P/LP variant in same gene, ≥ 6 unrelated patients	+2
	(Same) variant in AR disorder co-occurs with P/LP variant in same gene, ≥ 3 unrelated patients	+1

AR, autosomal recessive; LOD, logarithm of the odds; P/LP, pathogenic/likely pathogenic.
[a] Functional studies are recommended as additional evidence, but family and/or co-occurrence data can be used if functional studies are not available.
[b] Scoring and assigned point values complement the Quest Variant Scoring and Classification matrix[1] when possible.

Methods

- Two scientists independently assessed 203 gene-disease relationships with a broad range of phenotypes, modes of inheritance, and relationship strengths (96 previously assessed by ClinGen,[2] 107 not assessed by ClinGen).
- The scientists' categorizations were compared (1) to each other to assess standardization and (2) to ClinGen to assess concordance with established methods.[3]
- Categorizations that differed when the same evidence was used were considered discordant; those that differed because of new data were considered concordant.
- Internal Automatic Strong/Strong and ClinGen Definitive/Strong categorizations were all considered equivalent when assessing concordance between our process and ClinGen.

Results

- Using the streamlined process, categorizations of gene-disease relationship strength were 100% concordant (203/203) between the 2 scientists.
- These internal categorizations were 93% concordant (89/96) with corresponding categorizations in ClinGen (**Figure 2**).
- When the 7 discordant categorizations were examined in-depth (**Table 2**), 2 primary explanations were found:
  1. Use of professional judgement
     - In 2 cases, although the collected points placed the gene-disease relationships in the Strong category, the ClinGen Expert Panels used professional judgement to downgrade the associations to Moderate.
  2. Differences in data use
     - **a. Number of probands:**
       - In 2 cases (*DOCK7*, *ORAI1*), having <10 probands made the internal categorization more conservative compared to ClinGen.
     - **b. Definition of additional evidence:**
       - In 2 cases (*SIK1*, *FGFR1*), we included de novo occurrences as additional evidence, while ClinGen separates genetic and functional evidence.
- In 1 case (*TWIST1*), both factors contributed. The ClinGen Expert panel gave a more conservative categorization because only 3 cases of Sweeney-Cox syndrome and minimal functional evidence had been published.
- In 5 of 7 discordant cases, the final categorization did not change the types of evidence that could be used to score variants identified in the given gene.

Table 2. Discordant Gene-Disease Categorizations Between the Internal Streamlined Process and ClinGen

Gene	Disease	MOI	Internal categorization	ClinGen categorization (approval date)	Reason for discordance
POLR2A	Neurodevelopmental syndrome	AD	Strong	Moderate (modified) (2020)	Professional judgement
ZNF711	Intellectual disability	XL	Strong	Moderate (modified) (2020)	Professional judgement
DOCK7	Epileptic encephalopathy	AR	Moderate	Definitive (2021)	Data used differently (P)
ORAI1	Tubular aggregate myopathy	AD	Moderate	Definitive (2020)	Data used differently (P)
FGFR1	Hartsfield-Bixler-Demyer syndrome	AD	Strong	Moderate (2021)	Data used differently (AE)
SIK1	Developmental and epileptic encephalopathy	AD	Moderate	Limited (2018)	Data used differently (AE)
TWIST1	Sweeney-Cox syndrome	AD	Moderate	Limited (2021)	Data used differently (P, AE)

AE, additional evidence; AD, autosomal dominant; AR, autosomal recessive; MOI, mode of inheritance; P, proband count; XL, X-linked.

Results (continued)

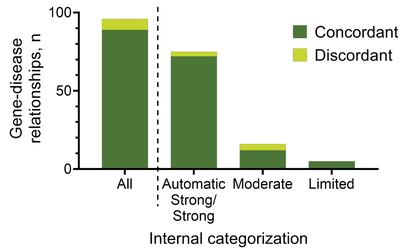

Figure 2. Concordance of internal categorizations with ClinGen categorizations.

Conclusions

- We developed an accurate and well-defined process for streamlined analysis and categorization of gene-disease relationships, which may help increase standardization and reduce workload compared to existing methods.
- Overall, categorizations from our streamlined process and those from ClinGen Expert panels were highly concordant.
- Challenges included phenotype specificity and comprehensive literature collection, but these are not unique to the streamlined process.
- Gene-disease relationships used in clinical settings should be reassessed periodically—particularly those in the "Limited" category. In several instances, recently published data caused the strength of a gene-disease association to increase between assessments.

References

1. Karbassi I, Maston GA, Love A, et al. A standardized DNA variant scoring system for pathogenicity assessments in Mendelian disorders. *Hum Mutat*. 2016;37(1):127-34. doi:10.1002/humu.22918
2. ClinGen – Clinical Genome Resource. Accessed January, 2020. https://www.clinicalgenome.org/
3. Strande NT, Riggs ER, Buchanan AH, et al. Evaluating the clinical validity of gene-disease associations: an evidence-based framework developed by the Clinical Genome Resource. *Am J Hum Genet*. 2017;100(6):895-906. doi:10.1016/j.ajhg.2017.04.015