Variant Classification Discordance: A real-world experience of genetic test results in a community-based setting

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BACKGROUND

- Accurate interpretation of hereditary cancer germline genetic variants is critical to ensuring appropriate care.
- Myriad Genetics has developed tools that are instrumental in the accurate classification of variants, including a previously described history-weighting algorithm (Pheno), mutation co-occurrence statistical analysis (MCO), and in trans haplotype analysis. (See Table 1 for publications and abstracts on Myriad's tools.)
- Differences in classification are known to occur among commercial testing laboratories, however the rate at which a single provider may observe variants with a different classification has not been reported.
- Here, we compared genetic test results from multiple laboratories ordered by a single surgical community-based practice with the classifications from Myriad.

Table 1. Myriad publications and abstracts detailing laboratory validations.

Variants	Publications
BRCA1, BRCA2	Pruss D, et al. Development and validation of a new algorithm for the reclassification of genetic variants identified in the *BRCA1* and *BRCA2* genes. Breast Cancer Res Treat. 2014 147(1):119-132.
MLH1, MSH2, MSH6	Morris B, et al. Classification of genetics variants in genes associated with Lynch syndrome using a clinical history weighting algorithm. BMC Genet. 2016 17(1):99.
BRCA1, BRCA2	Bowles KR, et al. A clinical history weighting algorithm accurately classifies *BRCA1* and *BRCA2* variants. Presented ASHG, 2013.
MLH1, MSH2, MSH6	Bowles KR, et al. Development of a novel history weighting algorithm for the reclassification of genetic variants identified in genes associated with Lynch syndrome. Presented ACMG, 2015.
BRCA1, BRCA2, MLH1, MSH2, MSH6, ATM, CHEK2, PALB2	Bowles KR, et al. Reclassification of uncertain variants identified in high and moderate cancer risk genes using history weighting analysis. Presented ACMG, 2016.
BRCA1, BRCA2, ATM, CHEK2, PALB2	Bowles KR, et al. Enhancement of history weighting analysis to accurately classify variants in high and moderate risk cancer panel genes. Presented International Symposium on HBOC, 2016.
BARD1, RAD51C	Added summer 2017 – Data on file

METHODS

- Variants initially reported as a "variant of uncertain significance" (VUS) on hereditary cancer test results from multiple commercial laboratories ordered by a single surgeon at a community-based, comprehensive breast center from June 2013 to May 2021 were evaluated and compared to the classifications from Myriad.
- In total, 212 variants were submitted for comparison.
- After review, 42 variants were excluded because they had not been observed previously in Myriad-tested patients.
- Therefore, 170 variants were eligible for comparison.
- Variants were classified as pathogenic/likely pathogenic, VUS, and benign/likely benign for comparison.
- Descriptive statistics were used for analysis.

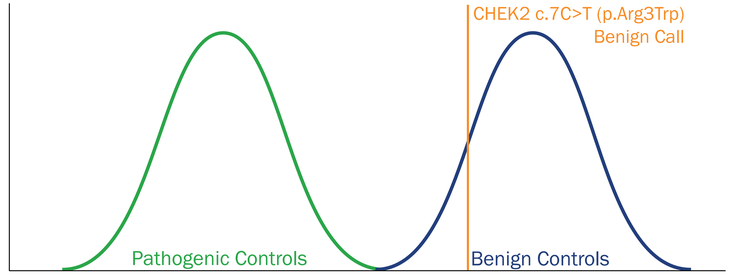
RESULTS

- Discordant classification was observed between Myriad and other testing laboratories for 28.2% (48/170) of the variants compared.
- Initially, all 170 variants were classified and reported by other testing laboratories as VUS, however, 15 variants (8.8%) were subsequently reclassified by other testing laboratories (N=13 were reclassified as benign/likely benign; N=2 were reclassified as pathogenic/likely pathogenic).
- Among all variants compared, 23.5% (40/170) were definitively classified by Myriad as benign/likely benign (Table 2).
- For 90.0% (36/40) of these variants, evidence driving the classification relied upon Pheno (32 variants) (Figure 1), MCO (4 variants), and in trans haplotype analysis (8 variants), with some variants having multiple lines of evidence.
- Other in-house specific rules were used for the remaining classifications (4/40; 10%).
- Of the 13 variants reclassified by other testing laboratories as benign/likely benign, 6 were classified as VUS by Myriad (Table 2).
- Notably, 2 variants classified by Myriad as VUS were reclassified by other laboratories as pathogenic/likely pathogenic (Table 2).
- 55% (22/40) of the discordant classifications were seen in high-risk genes including:

  APC, BRCA1/2, MLH1, MSH2, MSH6, PMS2, PALB2 and *STK11*
- 45% (18/40) were in moderate-risk genes including:

  ATM, BARD1, BRIP1, CHEK2, and *RAD51C*

Figure 1. Pheno combines the severities of all probands carrying the same variant into a pheno score.



Pathogenic Variant: Proband scores are mostly severe, Moderate scores are possible, benign scores are more rare

Benign Variant: Proband scores are a more even mixture of severe, moderate, and benign

Table 2. Variant classifications comparison between Myriad laboratory and other laboratories.

170 Variants	Myriad Laboratory	Other Laboratories
115	VUS	VUS
7	Benign/Likely Benign	Benign/Likely Benign
40	Benign/Likely Benign	VUS
6	VUS	Benign/Likely Benign
2	VUS	Pathogenic/Likely Pathogenic

CONCLUSIONS

- These data indicate that even in a single practice, significant discordance in variant classification exists based on the chosen laboratory.
- Myriad definitively classified nearly one-quarter of variants classified as VUS by other laboratories, likely due to the use of Myriad's laboratory-developed classification tools.
- The degree of discordance observed here reflects the need for continuous laboratory investment in variant classification tools and evaluation of genetic variants, enabling physicians and patients to receive accurate results to facilitate appropriate medical management decisions.

Contact scumming@myriad.com with questions.