Title: Atypical risk variants in oncology and rare disease: Findings from a review of laboratory case studies and over 6000 ClinVar submissions

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INTRODUCTION: Interpretation of atypical risk variants (ARVs) such as pathogenic low penetrance (PLP) and hypomorphic variants is challenging for diagnostic laboratories,

clinicians, genetic counselors, and patients. Our field lacks guidance on how to identify, classify and describe them. These terms can influence weight applied to a particular line of evidence, or whether variants are reported at all. These variants also contribute to conflicting classifications in ClinVar which may impact medical actionability and patient care.

PURPOSE: This study reviewed laboratory case studies from oncology and rare disease, along with ClinVar submissions, to identify hallmarks of ARVs and call for improved guidance on terminology, variant classification and reporting.

METHODS: We reviewed select candidate ARVs identified in our diagnostic laboratory including variants in cancer-associated genes, such as BRCA2, MEN1, MLH1, and TSC2, and rare disease genes including MYBPC3, SHQ1, PIGG, KIAA0586 among others. We compared evidence prompting consideration for atypical risk and evaluated evidence applied in the final classification of each variant under the current 2015 ACMG guidelines.

We also analyzed search results for "hypomorphic" and "low penetrance" in ClinVar to identify commonalities and discrepancies terminology, interpretation, and reporting of ARVs.

RESULTS: We identified common features, including presence in population databases, clinical evidence of reduced penetrance, and supporting experimental data from protein functional and splicing assays. Most variants reached a Pathogenic classification, but reporting language was modified to convey atypical risk, penetrance, and/or function.

Our ClinVar search found 2,073 variants in 314 genes that mentioned hypomorphs. Of the 6,811 variants in ClinVar mentioning low penetrance, most are classified as VUS (30%) or conflicting (34%). We observed that even when published evidence for ARVs existed, it was inconsistently described, classified and/or cited.

CONCLUSION: Through case studies and exploratory analyses, we find that further guidance on ARVs is needed as multiple terms are often conflated and used interchangeably despite having different meanings. There is no discrete field for labeling this variant type in public databases like ClinVar. While Pathogenic (Low Penetrance) is an option, Pathogenic classifications are still used instead, due to uncertainty around risk being 'low' for their gene-disease relationship. As such, there is no reliable estimate of how often ARVs are encountered by diagnostic labs in practice, and the variants mentioned here are likely underestimates. Whether or not a variant is characterized as an ARV in ClinVar relies on laboratories including a descriptor, usually in the free text comment section of their submissions. There is limited guidance around best practices for

identifying, interpreting, terming and reporting these variant types. We propose "reduced risk Pathogenic" as a preferred term. Such guidance is critical for consistent variant classification across laboratories and utility of searching databases for clinicians.

How these variants are identified, assessed and reported has implications for clinical validity and utility. Consistent guidance would likely reduce the number of conflicting interpretations in ClinVar. Whether or not these variants are likely to result in disease may be highly context-dependent, which has a wide range of implications from follow-up care to carrier screening.