

Title: Performance of SDHB-specific variant classification rules: An interim pilot analysis

Authors: Lauren Desrosiers-Battu (Baylor College of Medicine, Texas Children's Hospital), Roderick Clifton-Bligh (The University of Sydney), Jean-Pierre Bayley (Leiden University Medical Center), Terri McVeigh (Royal Marsden NHS Foundation Trust), Paul Newey (University of Dundee), Aparna Prasad (Invitae/LabCorp), Susan Richter (University of Auckland), Jamie Weyandt (Ambry Genetics), Tobias Else (University of Michigan), Endocrine Tumor Predisposition Variant Curation Expert Panel (Clinical Genome Resource)

Abstract

Background: The 2015 American College of Medical Genetics and Genomics (ACMG) and Association for Molecular Oncologists sequence variant classification (SVC v3) provides general guidance, however, gene-disease expertise is required to optimize classification. Many labs that offer genomic sequencing report on ACMG secondary findings genes, including *SDHB*, however may not have deep expertise in endocrine tumor predisposition (Endo-TPS).

Methods: The ClinGen Endo-TPS Variant Curation Expert Panel (VCEP) evaluated performance and recommended modifications to SVC v3 for *SDHB*-related TPS. Specific codes, e.g. PP3, were calibrated with odds of pathogenicity (OddsPath) calculations using known pathogenic/likely pathogenic (P/LP) (n=14) and benign/likely benign (B/LB) (n=7) variants. *SDHB* specifications for SVC v3 were piloted for P/LP, B/LB, and uncertain or conflicting variants (VUS).

Results: Eighteen variants were evaluated via VCEP rules, with 6 classified as P/LP, 7 as VUS and 5 as B/LB. All P/LP VCEP classifications (n=6) were consistent with the ClinVar classification summary, i.e. aggregate classification. One variant with conflicting classifications in ClinVar was downgraded to LB. All 3 variants with conflicts introduced were B/LB in ClinVar and VUS by VCEP rules. PS4, PP3 and BP4 were most frequently applied. PS4 was applied at various strengths dependent on available data with 1 variant very strong (VS), 3 strong (S) and 2 moderate (M). Of 11 missense variants evaluated based on *SDHB* PP3/BS4 calibration data, PP3_M was applied for 5 variants with REVEL scores >0.857 and BP4 was applied as supporting evidence for 3 variants with scores <0.627.

Discussion: Performance of VCEP SVC v3 specifications for *SDHB*-related TPS in a pilot of 18 variants was comparable to aggregate classification in ClinVar. Of those that differed, none generated new clinically-significant conflicts in ClinVar and 1 provided more certainty (VUS to LB). Application of PP3 and BP4 differed from ClinGen genome-wide calibration of REVEL scores. None of the 3 variants that met the VCEP BP4 rule would have had BP4 applied based on the ClinGen REVEL calibration as *SDHB* variants, regardless of

classification, tend to have high REVEL scores. Additionally, hypomorphic *SDHB* variants are associated with recessive mitochondrial disease, which challenges variant classification. Functional data, particularly for B/LB variants, will be critical for classification of these variants.