Variant Classification: ACMG recommendations

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Overview

- Introduction
- ACMG-AMP Classification System
- Evaluation of inter-laboratory concordance in variant classification

Link for download the ACMG Standards and Guidelines

https://www.acmg.net/docs/Standards_Guidelines_for_the_Interpretation_of_Sequence_Variants.pdf

search: “acmg standards and guidelines”

Why do we need classification systems

Published in final edited form as:
Hum Mutat. 2008 November ; 29(11): 1282–1291. doi:10.1002/humu.20880.

Sequence variant classification and reporting: recommendations for improving the interpretation of cancer susceptibility genetic test results

Sharon E. Plon[1,*,#], Diana M. Eccles[2,*], Douglas Easton[3], William D. Foulkes[4], Maurizio Genuardi[5], Marc S. Greenblatt[6], Frans B.L. Hogervorst[7], Nicoline Hoogerbrugge[8], Amanda B. Spurdle[9], and Sean Tavtigian[10] for the IARC Unclassified Genetic Variants Working Group[†]

Proposed Classification System for Sequence Variants Identified by Genetic Testing

Class	Description	Probability of being Pathogenic
5	Definitely Pathogenic	>0.99
4	Likely Pathogenic	0.95–0.99
3	Uncertain	0.05–0.949
2	Likely Not Pathogenic or of Little Clinical Significance	0.001–0.049
1	Not Pathogenic or of No Clinical Significance	<0.001

5 classes linked to validated quantitative measures of causality/ pathogenicity

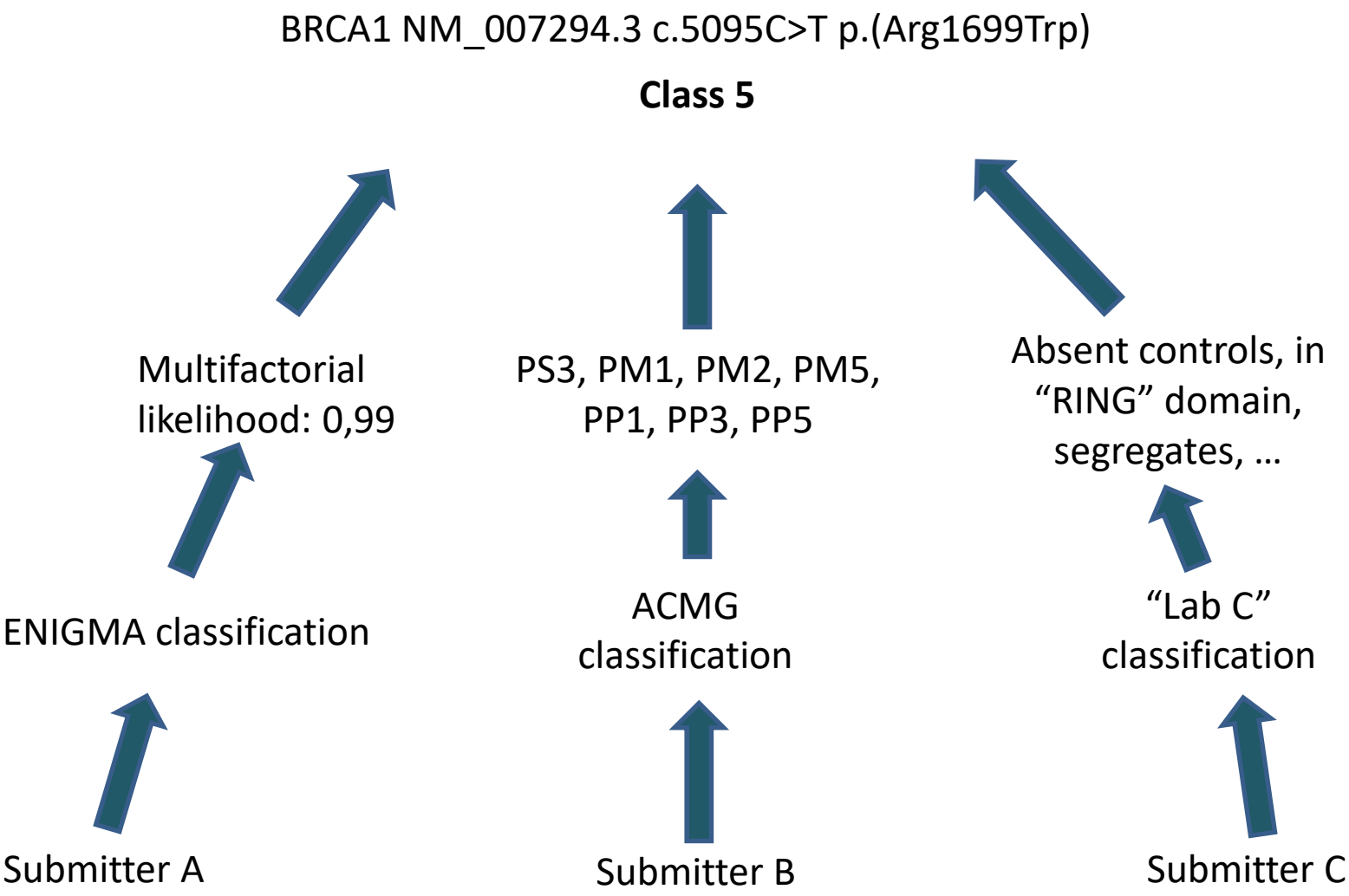
Class	Clinical Testing	Surveillance Recommendations if At-Risk Relative is Positive	Research Testing of Family Members
5	Test at-risk relatives for variant	Full high-risk surveillance guidelines	Not indicated
4	Test at-risk relatives for variant*	Full high-risk surveillance guidelines	May be helpful to further classify variant
3	Do not use for predictive testing in at-risk relatives*	Based on family history (and other risk factors)	May be helpful to further classify variant
2	Do not use for predictive testing in at-risk relatives*	Treat as "no mutation detected" for this disorder	May be helpful to further classify variant
1	Do not use for predictive testing in at-risk relatives*	Treat as "no mutation detected" for this disorder	Not indicated

All 5 classes are linked to clinical recommendations

Goal of IARC: To give actionable clinical recommendations to genetic data

Accurate and consistent variant classification is prerequisite for Dx & Precision Medicine

Why do we need classification systems



Example of pathogenic variant BRCA1 p.Cys61Gly (ClinVar)

Clinical significance (Last evaluated)	Review status (Assertion method)	Collection method	Condition(s) (Mode of inheritance)	Origin	Citations	Submitter - Study name
Pathogenic (Aug 10, 2015)	reviewed by expert panel • ENIGMA BRCA1/2 Classification Criteria (2015)	curation	Breast-ovarian cancer, familial 1 [MedGen | OMIM]	germline	• PubMed (1) [See all records that cite this PMID] • Other citation	Evidence-based Network for the Interpretation of Germline Mutant Alleles (ENIGMA) Study description
Pathogenic (Nov 3, 2014)	criteria provided, single submitter • ACMG Guidelines, 2015 • ACMG Guidelines, 2015	clinical testing	Breast-ovarian cancer, familial 1 [MedGen | OMIM]	germline		Michigan Medical Genetics Laboratories,University of Michigan
Pathogenic (Feb 11, 2016)	criteria provided, single submitter • LMM Criteria	clinical testing	Hereditary breast and ovarian cancer syndrome (Autosomal dominant inheritance) [MedGen | Orphanet]	germline	• PubMed (8) [See all records that cite these PMIDs]	Laboratory for Molecular Medicine,Partners HealthCare Personalized Medicine
Pathogenic (Dec 30, 2014)	criteria provided, single submitter • ACMG Guidelines, 2015 • ACMG Guidelines, 2015	clinical testing	Hereditary cancer-predisposing syndrome [MedGen]	germline		Color Genomics, Inc.,
Pathogenic (Feb 18, 2015)	criteria provided, single submitter • Quest pathogenicity assessment criteria	clinical testing	Breast-ovarian cancer, familial 1 (Autosomal dominant inheritance) [MedGen | OMIM]	germline	• PubMed (11) [See all records that cite these PMIDs]	Quest Diagnostics Nichols Institute San Juan Capistra
Likely pathogenic (Jul 1, 2016)	criteria provided, single submitter • ACMG Guidelines, 2015 • ACMG Guidelines, 2015	clinical testing	Familial cancer of breast [MedGen | Orphanet | OMIM]	germline		GeneKor MSA
Pathogenic (Oct 2, 2015)	criteria provided, single submitter • CIMBA Mutation Classification guidelines May 2016	clinical testing	Breast-ovarian cancer, familial 1 [MedGen | OMIM]	germline		Consortium of Investigators Modifiers of BRCA1/2 (CIM c/o University of Cambridg
Pathogenic	criteria provided, single submitter • Carraro et al. (PLoS One. 2013)	research	Breast cancer [MedGen]	germline	• PubMed (2) [See all records that cite these PMIDs]	Laboratory of Genomics and Molecular Biology,A. C. Camargo Cancer Center Study description
Pathogenic (Feb 22, 2016)	criteria provided, single submitter • Ambry Autosomal Dominant and X-Linked criteria (10/2015)	clinical testing	Hereditary cancer-predisposing syndrome [MedGen]	germline		Ambry Genetics
Pathogenic (Jan 20, 2017)	criteria provided, single submitter • GeneDx Variant Classification (06012015)	clinical testing	not provided [MedGen]	germline		GeneDx
Pathogenic (Apr 4, 2013)	criteria provided, single submitter • ACMG guidelines, 2007	clinical testing	Hereditary breast and ovarian cancer syndrome [MedGen | Orphanet]	germline		Genetics Diagnostic Laboratory,Children's Hospital of Eastern Ontario Study description
Pathogenic (May 13, 2015)	criteria provided, single submitter • EGL Classification Definitions	clinical testing	Breast-ovarian cancer, familial 1 [MedGen | OMIM]	germline	• Other citation	Emory Genetics Laboratory,Emory University
Pathogenic (Jan 18, 2017)	criteria provided, single submitter • Invitae Variant Classification Sherloc (09022015)	clinical testing	Hereditary breast and ovarian cancer syndrome [MedGen | Orphanet]	germline	• PubMed (2) [See all records that cite these PMIDs]	Invitae
Pathogenic (Feb 23, 2017)	criteria provided, single submitter • ACMG Guidelines, 2015 • ACMG Guidelines, 2015	clinical testing	Familial cancer of breast (Autosomal dominant inheritance) [MedGen | Orphanet | OMIM]	germline		Baylor Miraca Genetics Laboratories Study description

ACMG-AMP Classification System

Different Classification Systems

14 submissions / 10 different classification systems

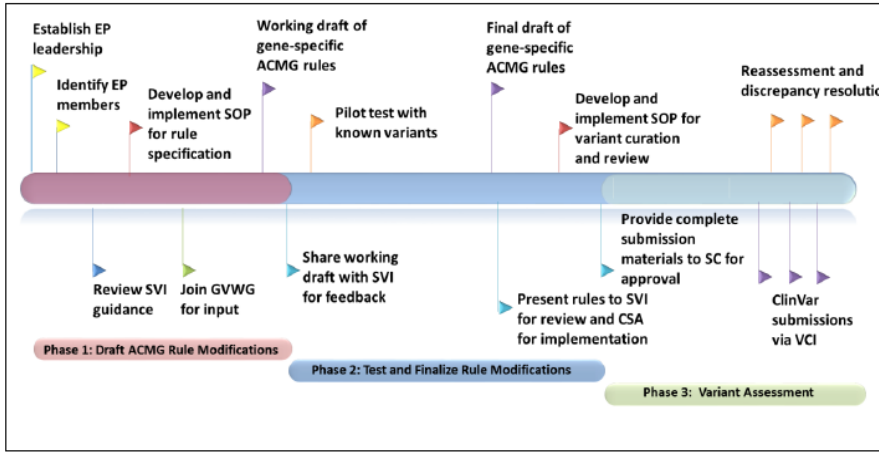
Expert Panel Classification

https://www.clinicalgenome.org/

ClinGen promotes formation of gene/ disease specific Expert Panels (EP)

ClinGen
Clinical Genome Resource

Figure 2: Expert Panel milestones



3.1 Overarching Goals

i. Define the set of conditions and associated genes that fall within the Clinical Domain WG.
   a. Evaluate the clinical validity (strength of evidence) of gene-disease associations for condition(s) within the working group domain (see 3.2).
   b. Prioritize genes and conditions for attention by the WG, considering those that have not been sufficiently evaluated and annotated for clinical use as potential priorities.
   c. Identify other groups with overlapping interests in gene-disease associations relevant to the conditions that are the responsibility of the WG in order to coordinate efforts.

ii. Facilitate deposition of variants from clinically relevant genes into ClinVar (see 3.3).
   a. Identify existing professional guidelines and community-organized efforts that are curating variants in genes related to the specific disease domain.
   b. Serve as a liaison to locus specific databases (LSDBs) and similar research efforts in order to facilitate reciprocal exchange of data between LSDBs and ClinVar.
   c. Identify clinical laboratories that perform testing in the clinical domain and facilitate interactions with ClinGen staff for data submission to ClinVar.

iii. Encourage development of Expert Panels to evaluate the clinical significance of genetic variants for submission to ClinVar.
   a. Identify and encourage external groups that are already involved in curating genetic variants within the domain, and coordinate with them to avoid duplicating effort.
   b. Review and evaluate the information provided from external curation groups for 3-star "Expert Panel" status in ClinVar based on the review process developed by the ClinGen Steering Committee.
   c. Establish ClinGen Expert Panels to focus on conditions deemed important by the CDWG but not currently served by an existing community effort (see 3.4).
   d. Perform specification of the ACMG/AMP sequence variant interpretation guidelines framework for variant classification to the respective diseases/genes.

Expert Panel Classification

https://www.clinicalgenome.org/

Clinical Domain WG	Expert Panel	Status
Cardiovascular CDWG	Brugada Syndrome Gene Curation Expert Panel	
	Cardiovascular Dilated Cardiomyopathy Gene Curation Expert Panel *(In progress)*	
	Cardiovascular Familial Hypercholesterolemia Variant Curation Expert Panel *(In progress)*	
	Cardiovascular KCNQ1 Variant Curation Expert Panel *(In progress)*	
	Cardiovascular LQTS Gene Curation Expert Panel *(In progress)*	
	Familial Thoracic Aortic Aneurysm and Dissection Gene Curation Expert Panel	
	Hypertrophic Cardiomyopathy Gene Curation Expert Panel	
	Inherited Cardiomyopathy Variant Curation Expert Panel	
Hearing Loss CDWG	Hearing Loss Gene Curation Expert Panel	
	Hearing Loss Variant Curation Expert Panel *(In progress)*	
Hemostasis/Thrombosis CDWG	Platelet Disorders Expert Panel	N/A
Hereditary Cancer CDWG	Breast and Ovarian Cancer Gene Curation Expert Panel	
	CDH1 Variant Curation Expert Panel *(In progress)*	
	Colon Cancer and Polyposis Gene Curation Expert Panel	
	Hereditary Cancer Gene Curation Expert Panel *(In progress)*	
	Myeloid Malignancy Variant Curation Expert Panel *(In progress)*	
	PTEN Variant Curation Expert Panel *(In progress)*	
	Somatic/Germline Variant Curation Group *(In progress)*	
	TP53 Variant Curation Expert Panel *(In progress)*	
Inborn Errors of Metabolism CDWG	Aminoacidopathy Gene Curation Expert Panel *(In progress)*	
	Fatty Acid Oxidation Gene Curation Expert Panel *(In progress)*	
	Mitochondrial Disease Gene Curation Expert Panel *(In progress)*	
	Mitochondrial Disease Variant Curation Expert Panel *(In progress)*	
	PAH Variant Curation Expert Panel	
	Storage Diseases Variant Curation Expert Panel *(In progress)*	
Monogenic Diabetes CDWG	Monogenic Diabetes Variant Curation Expert Panel *(In progress)*	
Neurodevelopmental Disorders CDWG	Autism and Intellectual Disability Gene Curation Expert Panel	
	Epilepsy Gene Curation Expert Panel	
	Rett Angelman Variant Curation Expert Panel *(In progress)*	
RASopathy CDWG	RASopathy Gene Expert Panel	

MOAC

Mother Of All Classification Systems

ACMG recommendations

September/October 2000 · Vol. 2 · No. 5

ACMG recommendations for standards for interpretation of sequence variations

ACMG L

ACMG Standards and

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I. Interpre

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variations

1. **Sequence variation is previously reported and is a re ognized cause of the disorder.** Review of the literatur central mutation databases, e.g., Human Gene Mutatio Database (HGMD), or the locus-specific database, to a sess the current degree of certainty that the sequence variation causative of the disorder should be undertaken prior to r porting. Concordance studies between phenotype and g notype within a family may provide acceptable criteria i the absence of more definitive functional studies.
2. **Sequence variation is previously unreported and is the type which is expected to cause the disorder.** Exam ples include variation that is predicted to shift the mRN reading frame; result in the introduction of a stop codo..

ACMG recommen interpretation an Revisions 2007

C. Sue Richards, PhD[1], Sherri Bale, Ph
Madhuri R. Hegde, PhD[6], Elaine Lyon
Laboratory Quality Assurance Commi

Key Words: *clinical genetic testi*

ACMG STANDARDS AND GUIDELINES | Genetics inMedicine

Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology

Sue Richards, PhD[1], Nazneen Aziz, PhD[2,16], Sherri Bale, PhD[3], David Bick, MD[4], Soma Das, PhD[5], Julie Gastier-Foster, PhD[6,7,8], Wayne W. Grody, MD, PhD[9,10,11], Madhuri Hegde, PhD[12], Elaine Lyon, PhD[13], Elaine Spector, PhD[14], Karl Voelkerding, MD[13] and Heidi L. Rehm, PhD[15]; on behalf of the ACMG Laboratory Quality Assurance Committee

Disclaimer: These ACMG Standards and Guidelines were developed primarily as an educational resource for clinical laboratory geneticists to help them provide quality clinical laboratory services. Adherence to these standards and guidelines is voluntary and does not necessarily assure a successful medical outcome. These Standards and Guidelines should not be considered inclusive of all proper procedures and tests or exclusive of other procedures and tests that are reasonably directed to obtaining the same results. In determining the propriety of any specific procedure or test, the clinical laboratory geneticist should apply his or her own professional judgment to the specific circumstances presented by the individual patient or specimen. Clinical laboratory geneticists are encouraged to document in the patient's record the rationale for the use of a particular procedure or test, whether or not it is in conformance with these Standards and Guidelines. They also are advised to take notice of the date any particular guideline was adopted and to consider other relevant medical and scientific information that becomes available after that date. It also would be prudent to consider whether intellectual property interests may restrict the performance of certain tests and other procedures.

ACMG-AMP Guidelines

ACMG STANDARDS AND GUIDELINES | Genetics inMedicine

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- Qualitative evaluation of different data types (28 defined criteria with assigned code)
- Each code is assigned a weight (stand-alone, very strong, strong, moderate, or supporting) and direction (benign or pathogenic)
- Variants then can be assigned in one of 5 classes (IARC 5-tier system)
- If not enough lines of evidence are invoked to classify a variant as P, LP, LB, or B, or there are valid but contradictory lines of evidence, a variant is interpreted as a VUS

ACMG-AMP Guidelines

	Benign		Pathogenic			
	Strong	**Supporting**	**Supporting**	**Moderate**	**Strong**	**Very strong**
Population data	MAF is too high for disorder BA1/BS1 **OR** observation in controls inconsistent with disease penetrance BS2			Absent in population databases PM2	Prevalence in affecteds statistically increased over controls PS4	
Computational and predictive data		Multiple lines of computational evidence suggest no impact on gene /gene product BP4 Missense in gene where only truncating cause disease BP1 Silent variant with non predicted splice impact BP7 In-frame indels in repeat w/out known function BP3	Multiple lines of computational evidence support a deleterious effect on the gene /gene product PP3	Novel missense change at an amino acid residue where a different pathogenic missense change has been seen before PM5 Protein length changing variant PM4	Same amino acid change as an established pathogenic variant PS1	Predicted null variant in a gene where LOF is a known mechanism of disease PVS1
Functional data	Well-established functional studies show no deleterious effect BS3		Missense in gene with low rate of benign missense variants and path. missenses common PP2	Mutational hot spot or well-studied functional domain without benign variation PM1	Well-established functional studies show a deleterious effect PS3	
Segregation data	Nonsegregation with disease BS4		Cosegregation with disease in multiple affected family members PP1	Increased segregation data →		
De novo data				De novo (without paternity & maternity confirmed) PM6	De novo (paternity and maternity confirmed) PS2	
Allelic data		Observed in *trans* with a dominant variant BP2 Observed in *cis* with a pathogenic variant BP2		For recessive disorders, detected in trans with a pathogenic variant PM3		
Other database		Reputable source w/out shared data = benign BP6	Reputable source = pathogenic PP5			
Other data		Found in case with an alternate cause BP5	Patient's phenotype or FH highly specific for gene PP4			

ACMG-AMP Guidelines

Pathogenic	(i) 1 Very strong (PVS1) AND a) *≥1 Strong (PS1–PS4) OR* b) *≥2 Moderate (PM1–PM6) OR* c) *1 Moderate (PM1–PM6) and 1 supporting (PP1–PP5) OR* d) *≥2 Supporting (PP1–PP5)*
	(ii) ≥2 Strong (PS1–PS4) OR
	(iii) 1 Strong (PS1–PS4) AND a) *≥3 Moderate (PM1–PM6) OR* b) *2 Moderate (PM1–PM6) AND ≥2 Supporting (PP1–PP5) OR* c) *1 Moderate (PM1–PM6) AND ≥4 supporting (PP1–PP5)*
Likely pathogenic	(i) 1 Very strong (PVS1) AND 1 moderate (PM1–PM6) OR
	(ii) 1 Strong (PS1–PS4) AND 1–2 moderate (PM1–PM6) OR
	(iii) 1 Strong (PS1–PS4) AND ≥2 supporting (PP1–PP5) OR
	(iv) ≥3 Moderate (PM1–PM6) OR
	(v) 2 Moderate (PM1–PM6) AND ≥2 supporting (PP1–PP5) OR
	(vi) 1 Moderate (PM1–PM6) AND ≥4 supporting (PP1–PP5)
Benign	(i) 1 Stand-alone (BA1) OR
	(ii) ≥2 Strong (BS1–BS4)
Likely benign	(i) 1 Strong (BS1–BS4) and 1 supporting (BP1–BP7) OR
	(ii) ≥2 Supporting (BP1–BP7)
Uncertain significance	(i) Other criteria shown above are not met OR
	(ii) the criteria for benign and pathogenic are contradictory

ACMG-AMP Guidelines

ACMG STANDARDS AND GUIDELINES | Genetics inMedicine

General Considerations:

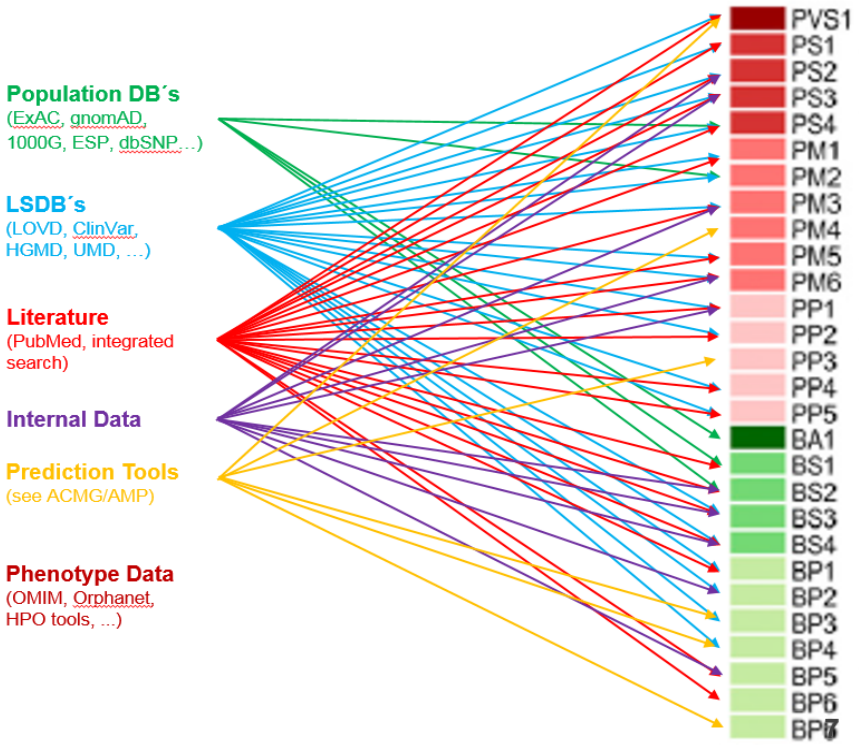
- for variants in all Mendelian genes (single gene, gene panel, exome, genome or transcriptome)
- not for somatic, Px, multigenic/complex disorders and not for low/ moderate penetrance variants
- be carefull with candiate genes („genes of uncertain significance“; „GUS“; Aug 2018: **OMIM 3.948 genes**)
- the terms „mutation“ and „polymorphism“ should not be used
- instead use „variant“ (pathogenic (5); likely pathogenic (4); uncertain significance (3); likely benign (2); benign (1))
- variants should be reported using the HGVS nomenclature (http://www.hgvs.org/mutnomen)
- to provide flexibility: some criteria listed as one weight can be moved to another weight using professional judgment, depending on the evidence collected (multiple observations of a variant in trans with path. variant – PM3 to PS)

„Pathogencity should be determined by the entire body of evidence in aggregate, including all cases studied, arriving at a single conclusion“

ACMG-AMP Guidelines

What are the requirements?

- Detailed population frequency data (ExAC, 1000G, now gnomAD)
- Clinical databases / LSDB´s
- Thorough literature search (find AND correctly interprete the literature)
- Access to your internal DB (hopefully these data are soon published!)
- Bioinformatic prediction integrated (protein, splice sites)



27 ACMG/AMP criteria from strong pathogenic to supporting benign
(Richards et al.; Genet Med. 2015)

Information

Selected SNP

SNP (TP53:NM_000546:c.704A>G:p.Asn235Ser ; het; AD) Benign (Iana, 2016-06-27 17:53:33)

Google | OMIM | Gene Reviews | LOVD

PatientID	Coverage	Quality	Subpanel
103274	423	222.0	Cancer_MammaErweitert

Associated Disease	Source
lung cancer alveolar cell carcinoma, included;;adenocarcinoma of lung, inclu...	OMIMGENE
li-fraumeni syndrome 1; lfs1;;sarcoma family syndrome of li and fraumeni;;sb...	OMIMGENE
esophageal cancer esophageal squamous cell carcinoma, susceptibility to, i...	OMIMGENE
squamous cell carcinoma, head and neck; hnscc	OMIMGENE
adrenocortical carcinoma, hereditary; adccadrenocortical carcinoma, pediatri...	OMIMGENE
papilloma of choroid plexus; cpp;;choroid plexus papillomachoroid plexus ca...	OMIMGENE
basal cell carcinoma, susceptibility to, 7; bcc7	OMIMGENE
b-cell chronic lymphocytic leukemia	Orphanet
small cell lung cancer	Orphanet
precursor b-cell acute lymphoblastic leukemia	Orphanet
gliosarcoma	Orphanet
giant cell glioblastoma	Orphanet

Search Associated Diseases

PP5??

Allele Frequency is greater than expected for disorder

TP53 p.(Asn235Ser)

- ExAC / ESP MAF = 0,0002 (= **0,02%**)
- Prevalenz Li-Fraumeni: 1:20.000 (= 0,5:10.000) or 0,00005 (= **0,005%**)
- highly penetrant and early onset

BP4

J Clin Invest. 1995 Apr;95(4):1606-11.

Germline p53 mutations are frequently detected in young children with rhabdomyosarcoma.

Diller L[1], Sexsmith E, Gottlieb A, Li FP, Malkin D.

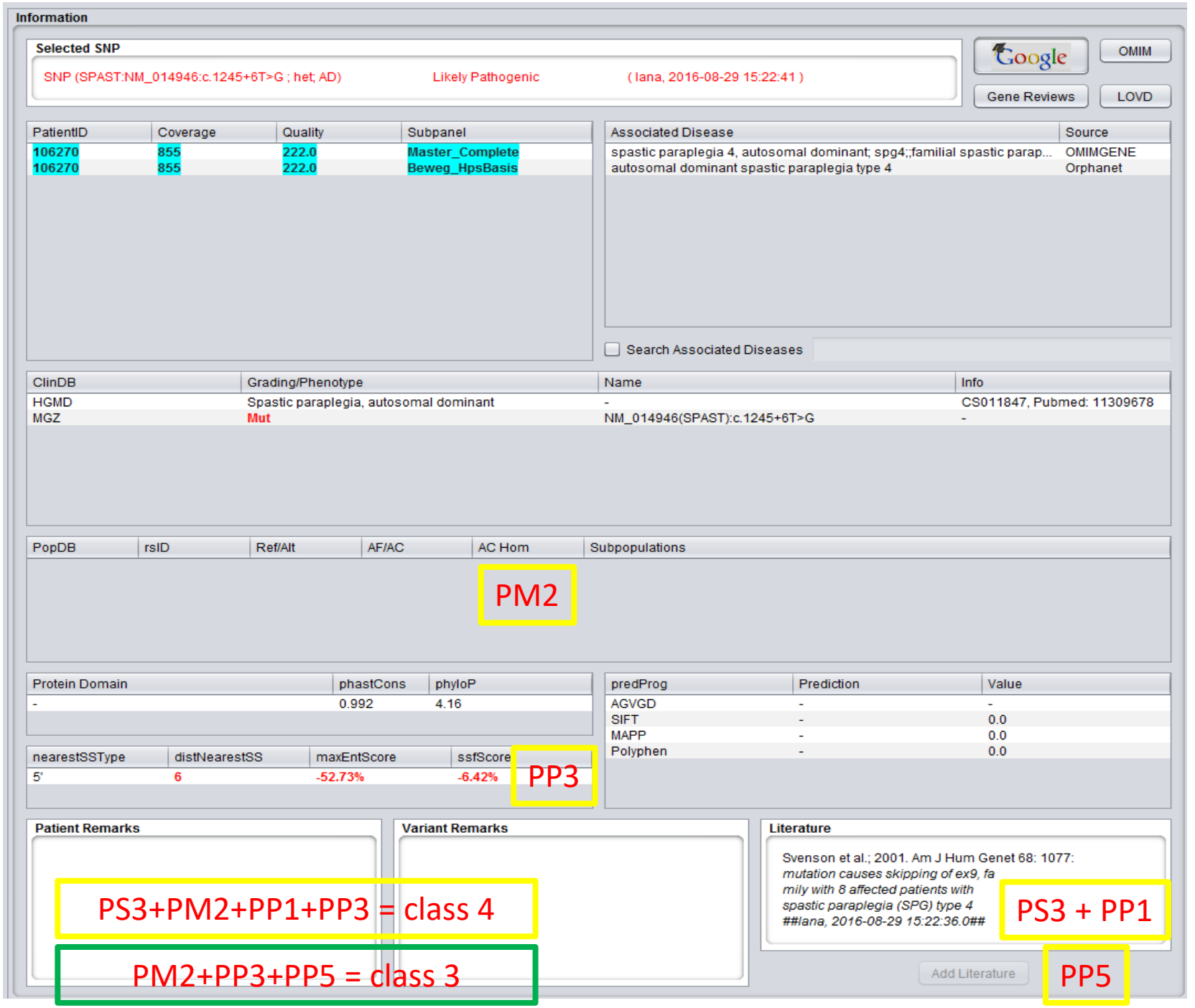
Author information

Abstract

We investigated the possibility that a proportion of children with sporadic rhabdomyosarcoma (RMS) carry constitutional mutations of the p53 tumor suppressor gene. 33 patients with sporadic RMS at two large outpatient pediatric oncology clinics submitted blood samples. Genomic DNA was extracted from peripheral blood leukocytes and PCR was used to amplify exons 2-11 of the p53 gene. Amplified genomic DNA was screened for the presence of germline p53 mutations using single-strand conformation polymorphism (SSCP) analysis. The DNA sequence of those samples that showed aberrant migration of bands on SSCP analysis was determined to identify the precise nature of the gene mutations. Patient records were reviewed to assess clinical correlates of the mutant p53 carrier state. Heterozygous constitutional mutations were detected in 3/33 patient samples screened. Two of these missense mutations are located in exon 7 and one in exon 8 of the p53 gene.

BS1+BS3+BS4+BP2+BP4 = class 1

BP2+BS4+BS3


Information
Selected SNP
SNP (SPAST:NM_014946:c.1245+6T>G ; het; AD)
Likely Pathogenic
(lana, 2016-08-29 15:22:41)
Google
OMIM
Gene Reviews
LOVD
PatientID
Coverage
Quality
Subpanel
106270
855
222.0
Master_Complete
106270
855
222.0
Beweg_HpsBasis
Associated Disease
Source
spastic paraplegia 4, autosomal dominant; spg4;;familial spastic parap...
OMIMGENE
autosomal dominant spastic paraplegia type 4
Orphanet
Search Associated Diseases
ClinDB
Grading/Phenotype
Name
Info
HGMD
Spastic paraplegia, autosomal dominant
-
CS011847, Pubmed: 11309678
MGZ
Mut
NM_014946(SPAST):c.1245+6T>G
-
PopDB
rsID
Ref/Alt
AF/AC
AC Hom
Subpopulations
PM2
Protein Domain
phastCons
phyloP
-
0.992
4.16
predProg
Prediction
Value
AGVGD
-
-
SIFT
-
0.0
MAPP
-
0.0
Polyphen
-
0.0
nearestSSType
distNearestSS
maxEntScore
ssfScore
5'
6
-52.73%
-6.42%
PP3
Patient Remarks
Variant Remarks
Literature
Svenson et al.; 2001. Am J Hum Genet 68: 1077:
mutation causes skipping of ex9, fa
mily with 8 affected patients with
spastic paraplegia (SPG) type 4
##lana, 2016-08-29 15:22:36.0##
PS3 + PP1
PS3+PM2+PP1+PP3 = class 4
PM2+PP3+PP5 = class 3
Add Literature
PP5

ACMG-AMP Guidelines

Criteria for classifying pathogenic variants (Tabelle 1)

Evidence of pathogenicity		Category
Supporting	PP1	Co segregation with disease in multiple affected family members in a gene definitively known to cause the disease *Note: May be used as stronger evidence with increasing segregation data*
	PP2	Missense variant in a gene that has a low rate of benign missense variation and in which missense variants are a common mechanism of disease
	PP3	Multiple lines of computational evidence support a deleterious effect on the gene or gene product (conservation, evolutionary, splicing impact, etc.) • *Caveat: Because many in-silico algorithms use the same or very similar input for their predictions, each algorithm should not be counted as an independent criterion. PP3 can be used only once in any evaluation of a variant.*
	PP4	Patient's phenotype or family history is highly specific for a disease with a single genetic etiology
	PP5	Reputable source recently reports variant as pathogenic, but the evidence is not available to the laboratory to perform an independent evaluation

- Choose more than one which are based on different algorithms
- Create rule how to use/interpret the results of the predictors (e.g. 3 from 5 must agree; one outlier is accepted, etc.)
- Difficult to get consensus between labs

ACMG-AMP Guidelines

Cold Spring Harbor Laboratory

bioRxiv beta

THE PREPRINT SERVER FOR BIOLOGY

New Results

Evaluation of *in silico* algorithms for use with ACMG/AMP clinical variant interpretation guidelines

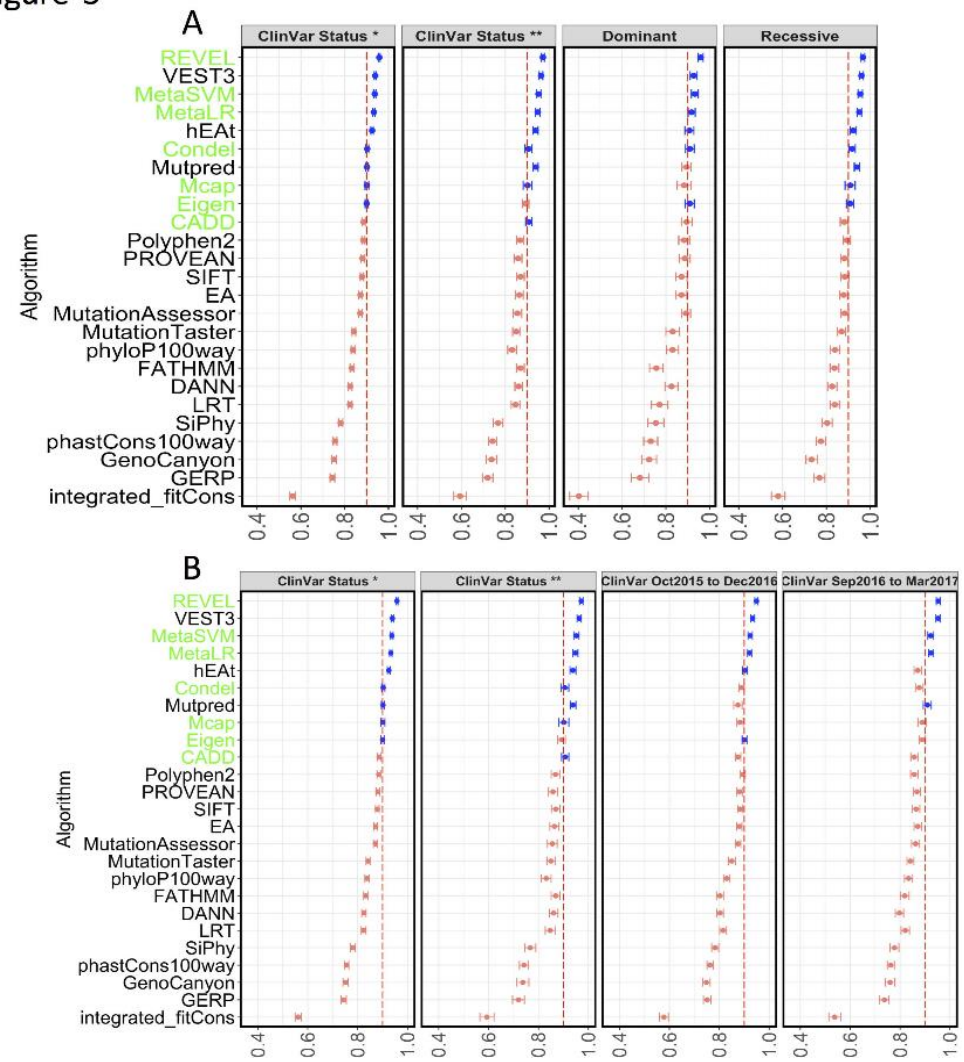
Rajarshi Ghosh, Ninad Oak, Sharon E. Plon

doi: https://doi.org/10.1101/146100

Abstract

The ACMG/AMP variant classification guidelines for clinical reporting recommend complete concordance of predictions among all in silico algorithms used without specifying the number or types of algorithms. The subjective nature of this recommendation contributes to discordance of variant classification among clinical laboratories. Using 14,819 benign or pathogenic missense variants from the ClinVar database, we compared performance of 25 algorithms across datasets differing in distinct biological and technical variables. There was wide variability in concordance among different combinations of algorithms with particularly low concordance for benign variants. We identified recently developed algorithms with high predictive power and robust to variables like disease mechanism, gene constraint and mode of inheritance, although poorer performing algorithms are more frequently used based on review of the clinical genetics literature (2011-2017). We describe high performing algorithm combinations with increased concordance in variant assertion, which should lead to more informed *in silico* algorithm usage by diagnostic laboratories.

Figure 3



ACMG-AMP Guidelines

Criteria for classifying benign variants (Tabelle 2)

Evidence of benign impact		Category
Stand alone	BA1	Allele frequency is >5% in Exome Sequencing Project, 1000 Genomes Project, or Exome Aggregation Consortium
Strong	BS1	Allele frequency is greater than expected for disorder (see Table 6)
	BS2	Observed in a healthy adult individual for a recessive (homozygous), dominant (heterozygous), or X-linked (hemizygous) disorder, with full penetrance expected at an early age
	BS3	Well-established in vitro or in vivo functional studies show no damaging effect on protein function or splicing
	BS4	Lack of segregation in affected members of a family • Caveat: The presence of phenocopies for common phenotypes (i.e., cancer, epilepsy) can mimic lack of segregation among affected individuals. Also, families may have more than one pathogenic variant contributing to an autosomal dominant disorder, further confounding an apparent lack of segregation.
Supporting	BP1	Missense variant in a gene for which primarily truncating variants are known to cause disease
	BP2	Observed in trans with a pathogenic variant for a fully penetrant dominant gene/disorder or observed in cis with a pathogenic variant in any inheritance pattern
	BP3	In-frame deletions/insertions in a repetitive region without a known function
	BP4	Multiple lines of computational evidence suggest no impact on gene or gene product (conservation, evolutionary, splicing impact, etc.) • Caveat: Because many in silico algorithms use the same or very similar input for their predictions, each algorithm cannot be counted as an independent criterion. BP4 can be used only once in any evaluation of a variant.
	BP5	Variant found in a case with an alternate molecular basis for disease
	BP6	Reputable source recently reports variant as benign, but the evidence is not available to the laboratory to perform an independent evaluation
	BP7	A synonymous (silent) variant for which splicing prediction algorithms predict no impact to the splice consensus sequence nor the creation of a new splice site AND the nucleotide is not highly conserved

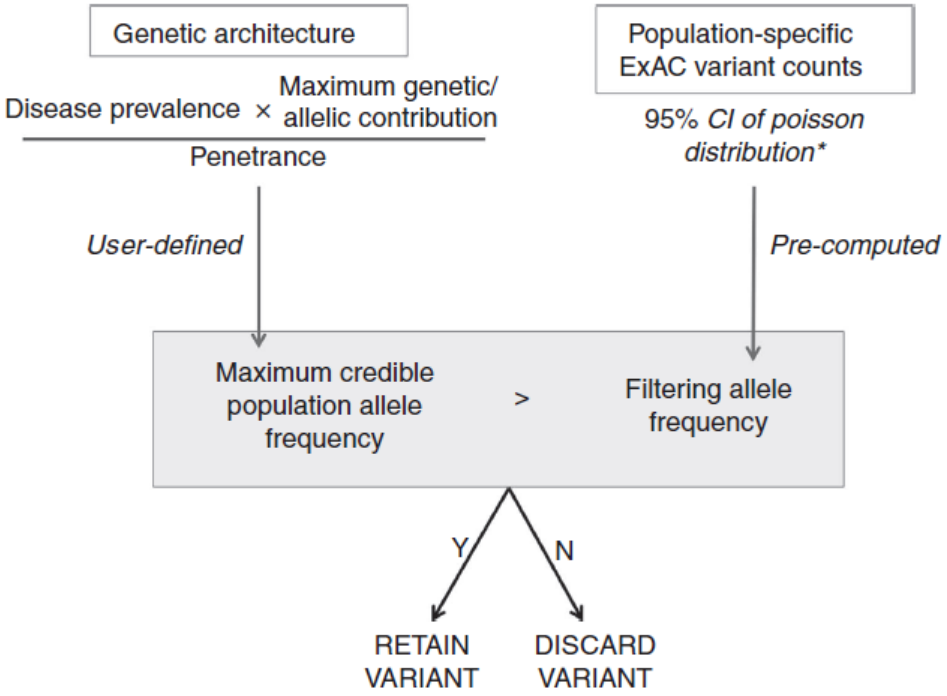
ACMG-AMP Guidelines

Official journal of the American College of Medical Genetics and Genomics **ORIGINAL RESEARCH ARTICLE** Genetics inMedicine

Open

Using high-resolution variant frequencies to empower clinical genome interpretation

Nicola Whiffin, PhD[1,2], Eric Minikel, MS[3,4], Roddy Walsh, MSc[1,2], Anne H. O'Donnell-Luria, MD, PhD[3,4], Konrad Karczewski, PhD[3,4], Alexander Y. Ing, MS, CGC[5,6], Paul J.R. Barton, PhD[1,2], Birgit Funke, PhD, FACMG[5,6], Stuart A. Cook, PhD, MRCP[1,2,7,8], Daniel MacArthur, PhD[3,4,9] and James S. Ware, PhD, MRCP[1,2,4,10]



ACMG-AMP Guidelines

Variant: 15:48725102 C / T

Filter Status	PASS
dbSNP	rs112084407
Allele Frequency	0.0007913
Filtering AF	0.001042 (European (Non-Finnish))
Allele Count	96 / 121322
UCSC	15-48725102-C-T
ClinVar	Click to search for variant in Clinvar

Filtering allele frequency (AF): a threshold for filtering variants that are too common to plausibly cause disease. If the variant filtering AF is greater than the maximum credible population AF for the disease of interest, the variant is too common to be causative and may be filtered. Click here to see the filtering AF calculator app and citation.

Site Quality Metrics

Annotations

This variant falls on 5 transcripts in 1 genes:

missense

- FBN1 Transcripts

3' UTR

- FBN1 - ENST00000537463

Note: This list may not include additional transcripts in the same gene that the variant does not overlap.

Population Frequencies

Population	Allele Count	Allele Number	Number of Homozygotes	Allele Frequency
European (Non-Finnish)	84	66710	0	0.001259
Latino	10	11534	0	0.000867
South Asian	2	16512	0	0.0001211
African	0	10406	0	0
East Asian	0	8638	0	0
European (Finnish)	0	6614	0	0
Other	0	908	0	0
Total	**96**	**121322**	**0**	**0.0007913**

Table 2 Maximum credible population frequencies and maximum tolerated ExAC allele counts for variants causative of exemplar inherited cardiac conditions, assuming a penetrance of 0.5 throughout

Disease	Maximum allelic contribution	Prevalence	Penetrance	Maximum population frequency	Maximum tolerated ExAC allele count
Marfan	0.015	1/3,000	0.5	5.0×10^{-6}	2
Noonan	0.10	1/1,000	0.5	1.0×10^{-4}	18
CPVT	0.10	1/10,000	0.5	1.0×10^{-5}	3
Classic Ehlers-Danlos	0.40	1/20,000	0.5	2.0×10^{-5}	5

CPVT,catecholaminergic polymorphic ventricular tachycardia; ExAC, Exome Aggregation Consortium database.
Prevalence estimates (taken as the highest value reported) were obtained from Marfan,[40] Noonan,[18] CPVT,[19] and classical Ehlers-Danlos.[20]

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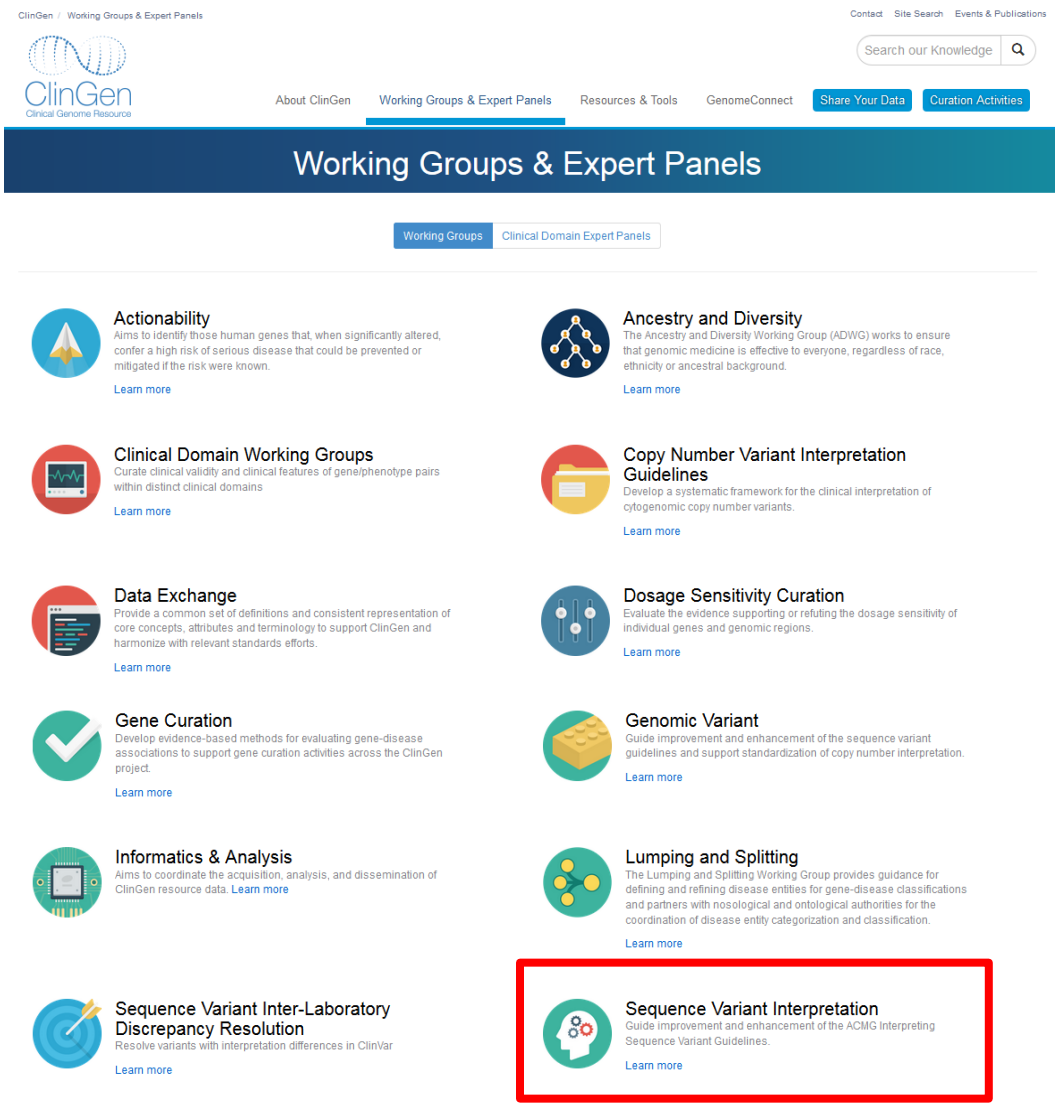

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ClinGen
Clinical Genome Resource

About ClinGen | Working Groups & Expert Panels | Resources & Tools | GenomeConnect | Share Your Data | Curation Activities

Sequence Variant Interpretation

The goal of the Sequence Variant Interpretation (SVI) Working Group is to support the refinement and evolution of the ACMG/AMP Interpreting Sequence Variant Guidelines to develop quantitative approaches to variant interpretation.

The Sequence Variant Interpretation Task Team also consults with and supports Expert Panel groups to develop gene- and disease-specific refinements of the ACMG/AMP Interpreting Sequence Variant Guidelines to increase the uniformity and consistency of the Expert Panel recommendations.The SVI Working Group has representation from the following ClinGen curation groups: Brain Malformations Expert Panel, Cardiovascular CDWG, Hereditary Cancer CDWG, Hereditary Hearing Loss CDWG, Inborn Errors of Metabolism CDWG, Mitochondrial Expert Panel, Monogenic Diabetes Expert Panel, Neurodevelopmental CDWG, RASopathy Expert Panel, Biocurators WG, CNV Interpretation WG and the Variant Curation Interface development team.

MEMBERS

SVI Publications

- Modeling the ACMG/AMP variant classification gudielines as a Bayesian classification framework

SVI Approved Expert Panel Specified ACMG/AMP Criteria

SVI approved ACMG/AMP guidelines specified by ClinGen Expert Panels.

- MYH7 - Inherited Cardiomyopathy
- RASopathy

SVI General Recommendations for Using ACMG/AMP Criteria

SVI provides general recommendations for using the ACMG/AMP criteria to improve consistency in usage and transparency in classification rationale.

- Guidance on how to rename criteria codes when strength of evidence is modified
- BA1: Updated Recommendation for the ACMG/AMP Stand Alone Pathogenicity Criterion for Variant Classification
  - BA1 Exception List (July 2018)
  - BA1 Exception List Nomination Form
- PVS1: Recommendation for interpreting the loss of function PVS1 ACMG/AMP critiera (preprint)
- PS2/PM6: Recommendation for de novo PS2 and PM6 ACMG/AMP critiera (Version 1.0)
- PP5/BP6: Recommendation for reputable source PP5 and BP6 ACMG/AMP criteria

Leadership

Leslie G. Biesecker, MD

Steven Harrison, PhD

Coordinators

Please contact a coordinator if you have questions.

Danielle Azzariti, MS, CGC
dazzarit@broadinstitute.org

Membership

Membership in this committee spans many fields, including genetics, medical, academia, and industry. [View Members]
For more information, please contact:
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dazzarit@broadinstitute.org

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New Results

Recommendations for Interpreting the Loss of Function PVS1 ACMG/AMP Variant Criteria

Ahmad Abou Tayoun, Tina Pesaran, Marina DiStefano, Andrea Oza, Heidi Rehm, Leslie Biesecker, Steven Harrison, ClinGen Sequence Variant Interpretation Working Group

doi: https://doi.org/10.1101/313718

This article is a preprint and has not been peer-reviewed [what does this mean?].

Abstract

The 2015 ACMG/AMP sequence variant interpretation guideline provided a framework for classifying variants based on several benign and pathogenic evidence criteria. This guideline includes a pathogenic criterion (PVS1) for predicted loss of function variants. However, the guideline did not elaborate on the specific considerations for the different types of loss of function variants, nor did it provide decision-making pathways assimilating information about the variant type, its location within the gene, or any additional evidence for the likelihood of a true null effect. Furthermore, the ACMG/AMP guideline did not take into account the relative strengths for each evidence type and the final outcome of their combinations with respect to PVS1 strength. Finally, criteria specifying the genes for which PVS1 can be used are still missing. Here, as part of the Clinical Genomic Resource (ClinGen) Sequence Variant Interpretation (SVI) Working Groups goal of refining ACMG/AMP criteria, we provide recommendations for applying the PVS1 rule using detailed guidance addressing all the above-mentioned gaps. We evaluate the performance of the refined rule using heterogeneous types of loss of function variants (n= 56) curated by seven disease-specific groups across ten genes. Our recommendations will facilitate consistent and accurate interpretation of predicted loss of function variants.

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Criteria for classifying pathogenic variants (Tabelle 1)

Evidence of pathogenicity		Category
Very strong	PVS1	Null variant (nonsense, frameshift, canonical ±1 or 2 splice sites, initiation codon, single or multiexon deletion) in a gene where LOF is a known mechanism of disease. *Caveats:* • *Beware of genes where LOF is not a known disease mechanism (e.g., GFAP, MYH7)* • *Use caution interpreting LOF variants at the extreme 3' end of a gene* • *Use caution with splice variants that are predicted to lead to exon skipping but leave the remainder of the protein intact* • *Use caution in the presence of multiple transcripts*

- PVS1 is a very powerful criteria
- PVS1 + PM2 (absent from controls) = class 4 (likely pathogenic)
- If criteria is not met (e.g. 3´end of gene, *in frame* exon skipping) PVS1 could not be provoked at all, these variants fall into class 3

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Determination of disease mechanism is critical

TABLE 1. Criteria for LoF disease mechanism.

- **Follow PVS1 Flowchart if:**
  - Clinical validity classification of gene is STRONG or DEFINITIVE

    AND
  - 3 or more LOF variants are Pathogenic without PVS1 AND >10% of variants associated with the phenotype are LOF (must be across more than 1 exon)

- **Decrease final strength by *one* level (i.e. VeryStrong to Strong) if:**
  - Clinical validity classification of gene is at least MODERATE

    AND
  - 2 or more LOF variants have been previously associated with the phenotype (must be across more than 1 exon)

    AND
  - Null mouse model recapitulates disease phenotype

- **Decrease final strength by *two* levels (i.e. VeryStrong to Moderate) if:**
  - Clinical validity classification is at least MODERATE

    AND EITHER
  - 2 or more LOF variants have been previously associated with the phenotype (must be across more than 1 exon)

    OR
  - Null mouse model recapitulates disease phenotype

- **If there is no evidence that LOF variants cause disease, PVS1 should not be applied at any strength level.**

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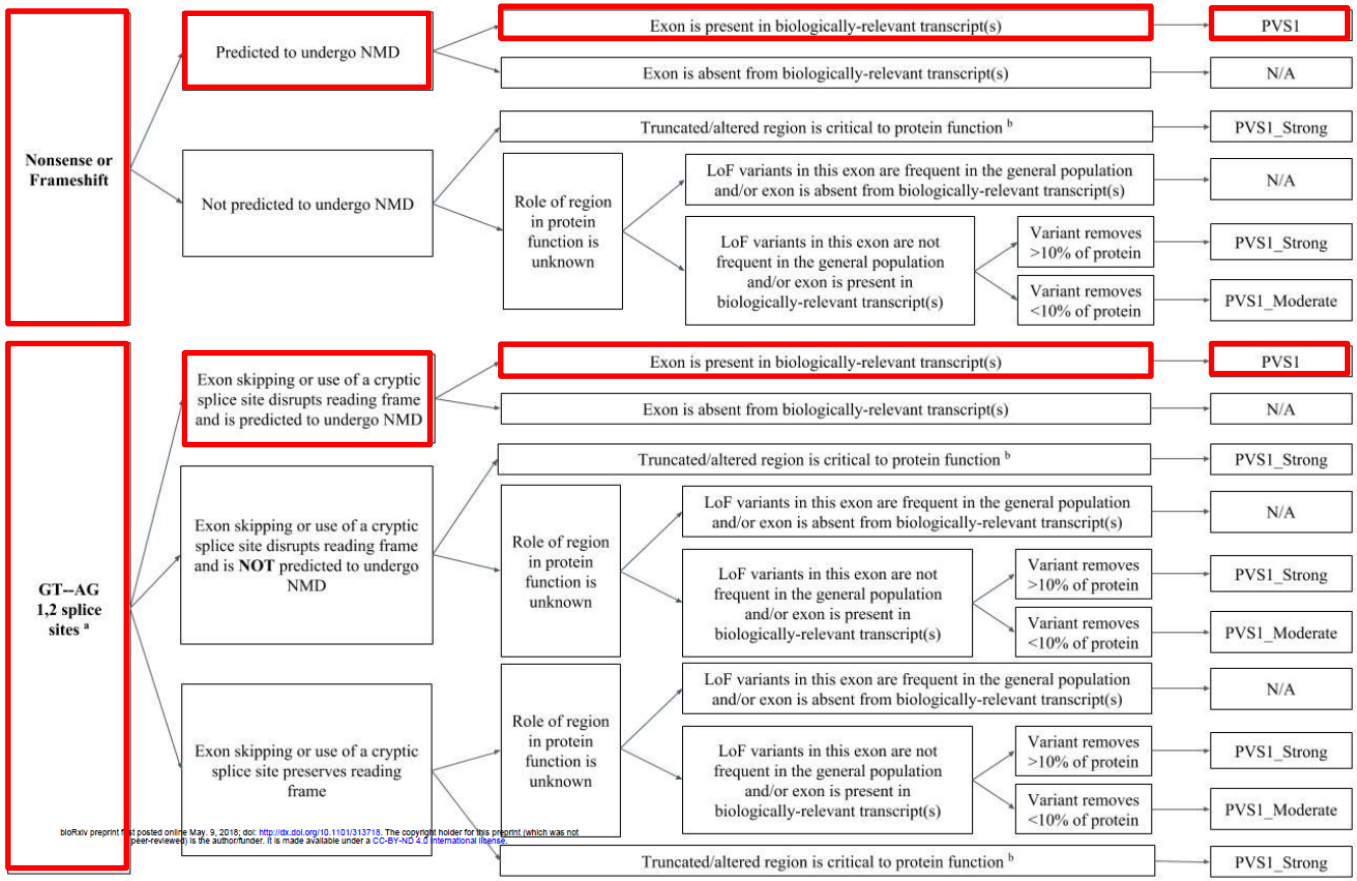

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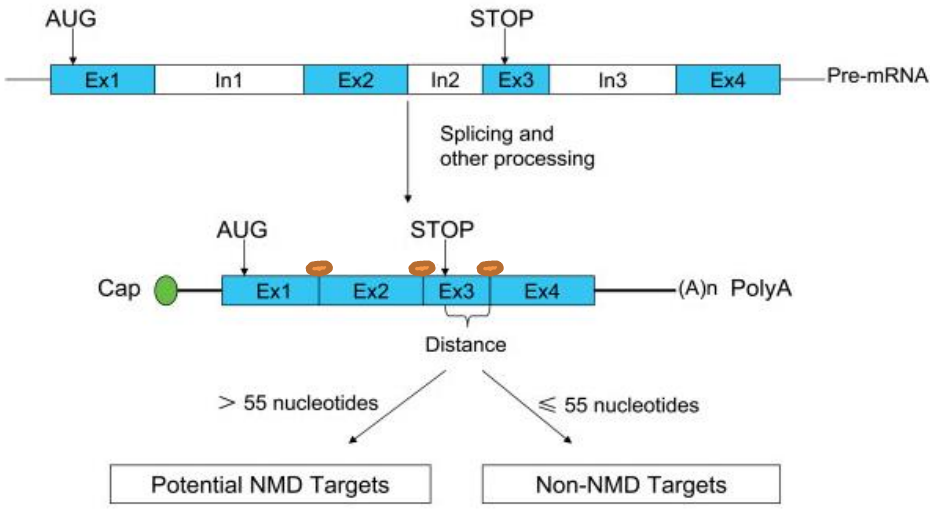
NMD or not NMD, that is the question ….

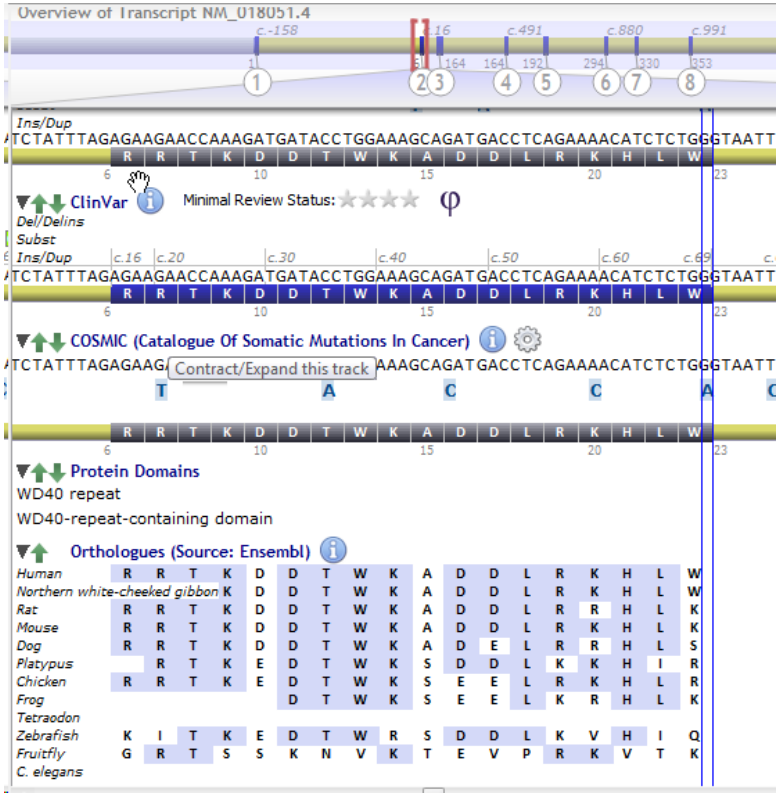
PTC in last exon or 55 nucl. of second last exon



Ex*: Exons **In*:** Introns **AUG**: start codon **STOP**: termination codon

EJC (Exon Junction Complex)

WDR60 NM_018051:c.69G>A p.(Trp23*)



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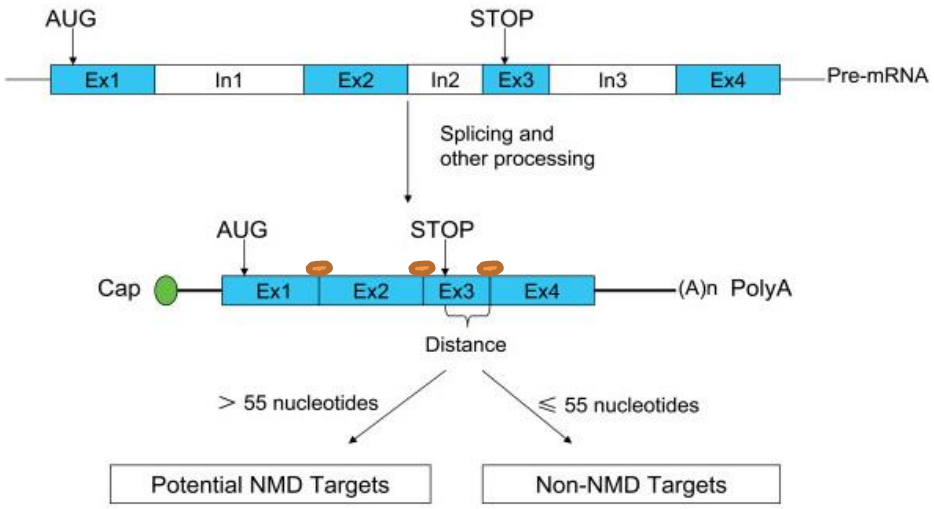

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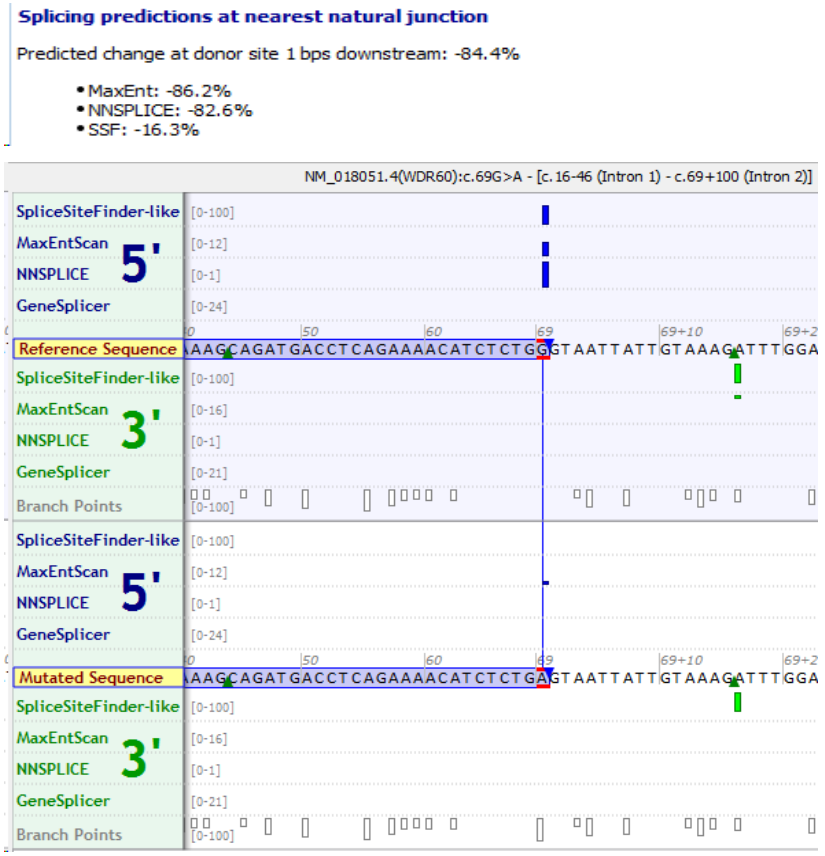

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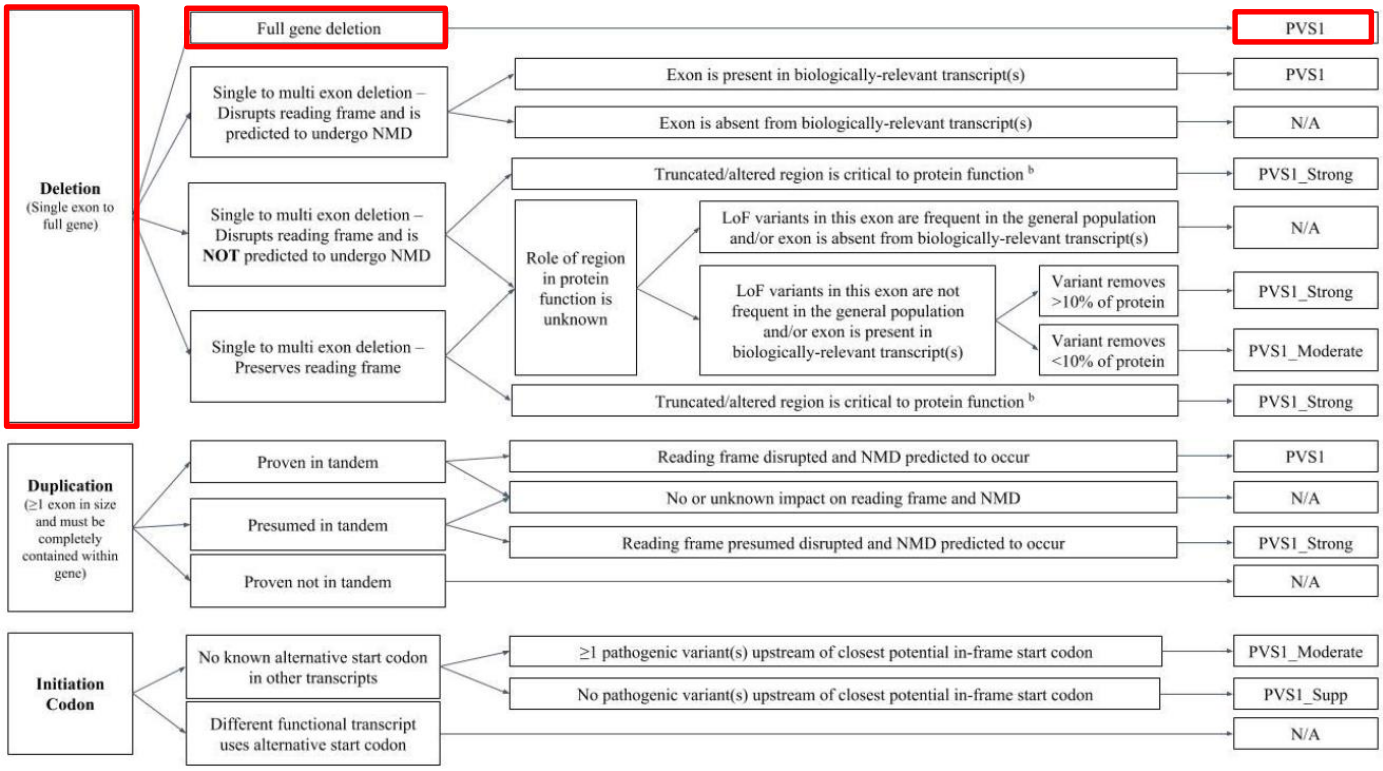

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Supporting	PP1	Co segregation with disease in multiple affected family members in a gene definitively known to cause the disease *Note: May be used as stronger evidence with increasing segregation data*
	PP2	Missense variant in a gene that has a low rate of benign missense variation and in which missense variants are a common mechanism of disease
	PP3	Multiple lines of computational evidence support a deleterious effect on the gene or gene product (conservation, evolutionary, splicing impact, etc.) • *Caveat: Because many in-silico algorithms use the same or very similar input for their predictions, each algorithm should not be counted as an independent criterion. PP3 can be used only once in any evaluation of a variant.*
	PP4	Patient's phenotype or family history is highly specific for a disease with a single genetic etiology
	PP5	Reputable source recently reports variant as pathogenic, but the evidence is not available to the laboratory to perform an independent evaluation

SVI

The ACMG/AMP reputable source criteria for the interpretation of sequence variants

To the Editor: In 2015, the American College of Medical Genetics and Genomics and the Association for Molecular Pathology (ACMG/AMP) promulgated recommendations for the interpretation of sequence variants.[1] These recommendations have been widely implemented and shown to be useful for improving variant classification consistency.[2–4] From the beginning, they were recognized to be a starting point for further future refinements and extensions. The Clinical Genome (ClinGen) Resource is focused on curating the genome for use in molecular diagnosis.[5] One such effort is the Sequence Variant Interpretation Working Group, which has taken on the task of refining and evolving the current ACMG/AMP recommendations. This working group meets regularly and also, as individuals, interacts widely with the clinical testing community. Through these interactions, the working group has received input from multiple sources that two related criteria in the original recommendations should be considered for removal from the ACMG/AMP framework:

- PP5 "Reputable source recently reports variant as pathogenic, but the evidence is not available to the laboratory to perform an independent evaluation."
- BP6 "Reputable source recently reports variant as benign, but the evidence is not available to the laboratory to perform an independent evaluation."

It is our strongly held view that primary data are far preferable to expert opinion without access to primary data. The PP5 and BP6 criteria rely on assertions that are not directly linked to the evidence on which they were based. These criteria might have been appropriate in 2015 as they were originally intended as a bridge to allow the community to benefit from clinical laboratory expertise and locus-specific research databases, prior to the wider use of resources such as ClinVar that provide mechanisms for laboratories and researchers to share underlying primary data. Indeed, ClinVar has been even more successful than hoped and now includes 580,831 assertions for over 375,106 variants (ClinVar website accessed 8 January 2018). Submissions with "assertion criteria provided" review status, designating that the submitter either provided their evidence to ClinVar or indicated a willingness to provide evidence upon inquiry, account for 81% (470,245-/580,831) of all submissions to ClinVar (ClinVar website accessed 8 January 2018). Therefore, there is less need to rely on assertions from reputable sources in the absence of primary evidence.

A second rationale for these two criteria was to support the efforts of the Sharing Clinical Reports Project (https://www.clinicalgenome.org/data-sharing/sharing-clinical-reports-project-scrp/), in which clinicians collected the test reports (including variant interpretation) produced by a large commercial laboratory that for the past decade has consistently declined to share underlying data or to submit assertions to ClinVar. As data for hereditary breast and ovarian cancer susceptibility alleles have increasingly been forthcoming from other laboratories, the necessity of this secondary information has declined and the currency of these data has receded.

Finally, we are concerned that these two criteria may be commonly misused by laboratories that incorporate primary data into variant assessment (e.g., functional data, criteria PS3 and BS3) and at the same time invoke criteria PP5 and BP6 for existing classifications that are based on the same set of data, which may lead to double counting, and potentially lead to errors in classification.

Based on these considerations, we propose that laboratories discontinue the use of criteria PP5 and BP6 as soon as that is practically achievable. We have removed these criteria from the ClinGen Variant Curation Interface. However, as with all types of evidence, interpretation of variants is the responsibility of the clinical testing laboratory director, who should account for the entirety of evidence and the sources of the data, and these recommendations should not be interpreted otherwise.

DISCLOSURE

L.G.B. is an uncompensated adviser for Illumina. S.M.H. declares no conflict of interest.

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REFERENCES

1. Richards S, Aziz N, Bale S, et al. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet Med* 2015;17:405–424.

Supporting	PP1
	PP2
	PP3
	PP4
	PP5

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evaluation

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Criteria for classifying benign variants (Tabelle 2)

Evidence of benign impact		Category
Stand alone	BA1	Allele frequency is >5% in Exome Sequencing Project, 1000 Genomes Project, or Exome Aggregation Consortium

- BA1 (stand alone) means … class 1
- Cumulative evidence suggests that this does not apply to all subpopulations
- Founder alleles in subpopulations could have higher frequencies

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Gene	Variant	Classification	ACMG/AMP Criteria applied (not including BA1 or BS1)	ClinVar ID	ClinGen Allele Registry ID	Chr	Position	Ref	Alt	ExAC Source Pop	ExAC Source Pop MAF	ClinVar disease entry
ACAD9	NM_014049.4: c.-44_-41dupTAAG	VUS	PS3_Supporting; BS2	1018	CA114709	3	128,598,490	C	CTAAG	AFR	0.1261	Deficiency of Acyl-CoA dehydrogenase family, member 9
GJB2	NM_004004.5: c.109G>A (p.Val37Ile)	Pathogenic	PS4; PP1_Strong; PM3_VeryStrong; PS3_Moderate	17023	CA172210	13	20,763,612	C	T	EAS	0.07242	Deafness, autosomal recessive
HFE	NM_000410.3: c.187C>G (p.His63Asp)	Pathogenic*	PS4	10	CA113797	6	26,091,179	C	G	NFE	0.1368	Hereditary hemochromatosis
HFE	NM_000410.3: c.845G>A (p.Cys282Tyr)	Pathogenic*	PS4; PP3	9	CA113795	6	26,093,141	G	A	NFE	0.05135	Hereditary hemochromatosis
MEFV	NM_000243.2: c.1105C>T (p.Pro369Ser)	VUS	PM3; PM5	2551	CA280114	16	3,299,586	G	A	EAS	0.07156	Familial Mediterranean fever
MEFV	NM_000243.2: c.1223G>A (p.Arg408Gln)	VUS	PM3; PM5	2552	CA280116	16	3,299,468	C	T	EAS	0.05407	Familial Mediterranean fever
PIBF1	NM_006346.2: c.1214G>A (p.Arg405Gln)	VUS	PM3; BS2	217689	CA210261	13	73,409,497	G	A	AMR	0.09858	Joubert syndrome
ACADS	NM_000017.3: c.511C>T (p.Arg171Trp)	VUS	PS3_Moderate; PM3; PP3	3830	CA312214	12	121,175,678	C	T	FIN #	0.06589	Deficiency of butyryl-CoA dehydrogenase
BTD	NM_000060.4: c.1330G>C (p.Asp444His)	Pathogenic	PS3; PM3_Strong; PP3; PP4	1900	CA090886	3	15,686,693	G	C	FIN #	0.05398	Biotidinase deficiency

*ACMG/AMP criteria selected does not match the classification as these variants are common low-penetrant variants and the ACMG/AMP guidelines are not designed for this variant type

Detected at >5% MAF only in Finnish population (see text).

Genomic coordinates on build GRCh37

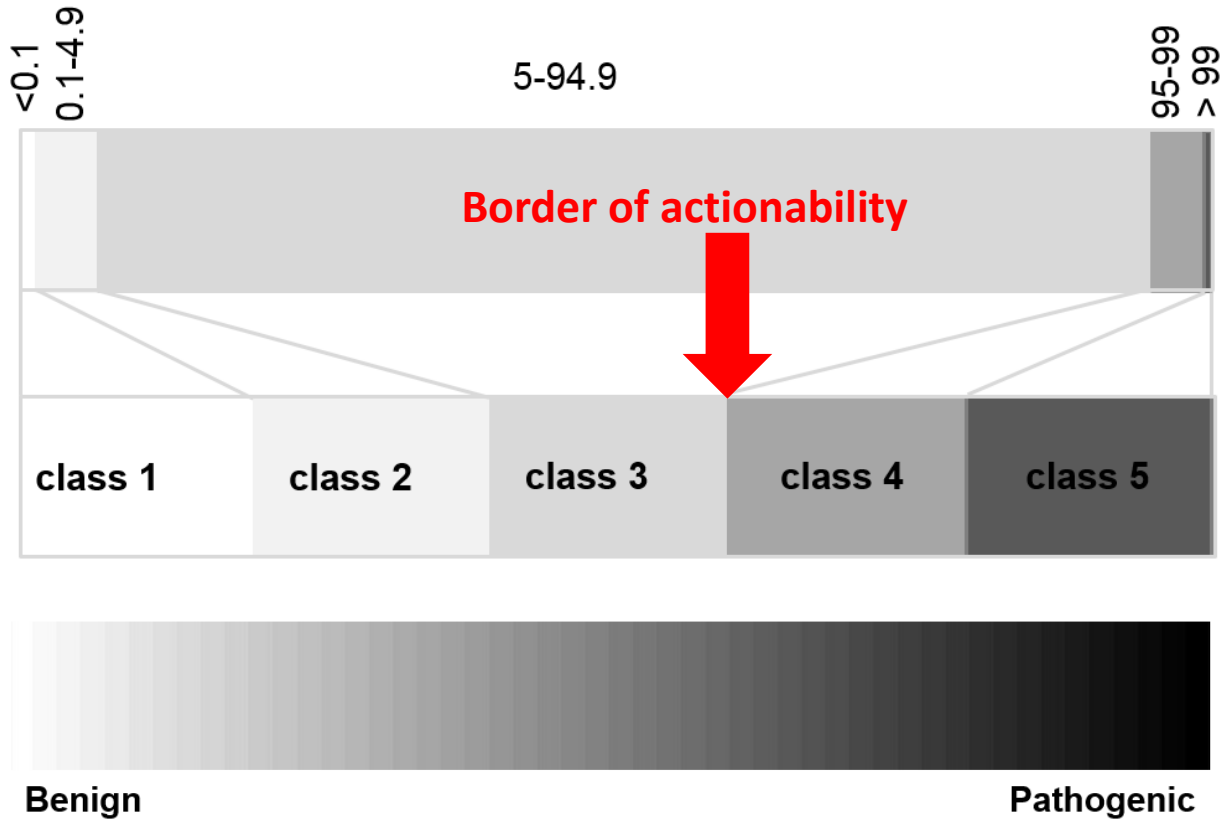
AFR: African/African American, EAS: East Asian, NFE: Non-Finnish European, AMR: Latino, FIN=Finnish

Evaluation of inter-laboratory concordance

Study	Description	Observed Concordance	Reasons for Inconsistency	Remarks
Amendola et al. Am J Hum Genet 2016	Comparison of concordance of 9 CSER-labs classifying 99 variants	**34% before** and **71% after** consensus discussion / only 5% of differences are clinically relevant	Correct use of several ACMG rules was not clear / challenging variants	training is necessary for consistent classification / underscores importance of not only having a standardized approach to variant assessment but also sharing variant interpretations for identifying and resolving discordance
Harrison et al. Genet Med 2017	ClinVar Laboratory comparison and consistency assessment	**83% initially concordant** **87% of discordant** variants **could be resolved**	ACMG rules not applied to ClinVar variants (53%) Internal data not published (33%) Differences in use/ weighting of data (14%)	Participating laboratories increased their overall concordance from 88.3 to 91.7%, sharing variant interpretations in ClinVar is critical to moving toward more consistent variant interpretations
Pepin et al. Genet Med 2016	Comparison and evaluation of consistent variant classifications (outside labs vs in house) in a distinct disease field (COLx)	**29% complete**, **29% „moderate“** **58% not actionable**	Lack of reference of the biology (48%) Lack of access to unpublished data (33%)	In diseases with a „special biology“ expert knowledge is important for accurate classification / unpublished data are a major source of inconsistent classification
Balmana et al. J Clin Oncol 2016	ClinVar study comparing variant classifications of 603 variants in non-BRCA cancer genes	**74% concordance** **11% clinically relevant**	many observed differences were because of variants in low-penetrance genes (RR<2)	Conflicting interpretation of genetic findings is frequent and may have implications for medical management decision
Yang et al. Genet Med 2017	ClinVar search of discordant actionable classifications, evaluation of reasons for inconsistencies	**96% major consensus** **94% complete consensus**	Non-clinical lab subm. Clinical areas differ Old data points Literature citations	Recent variant classifications from clinical testing laboratories have high overall concordance.

Are there really large inconsistencies in ClinVar ?

This depends on what you compare …



InSiGHT: posterior probability of pathogenicity derived by multifactorial likelihood analysis https://www.insight-group.org/

Are there really large inconsistencies in ClinVar ?

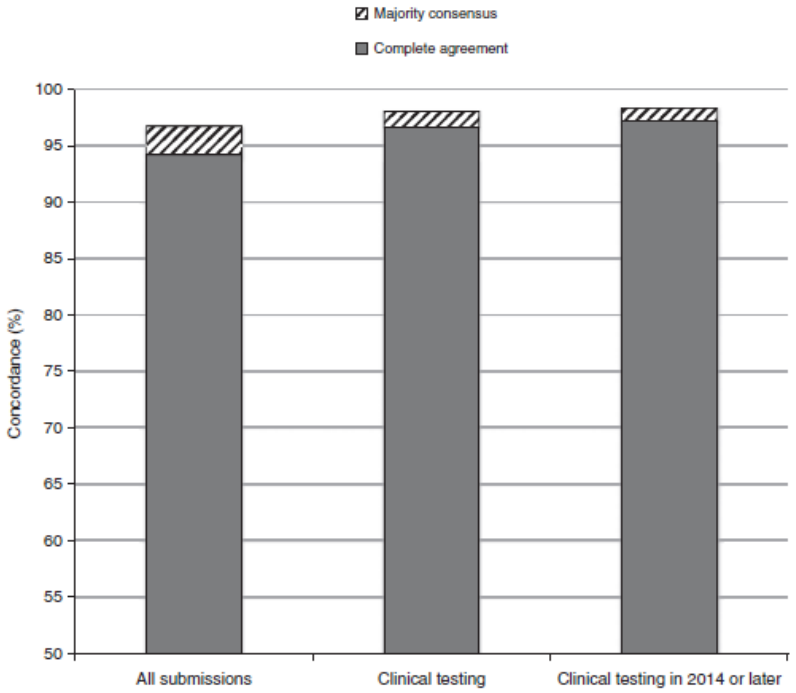
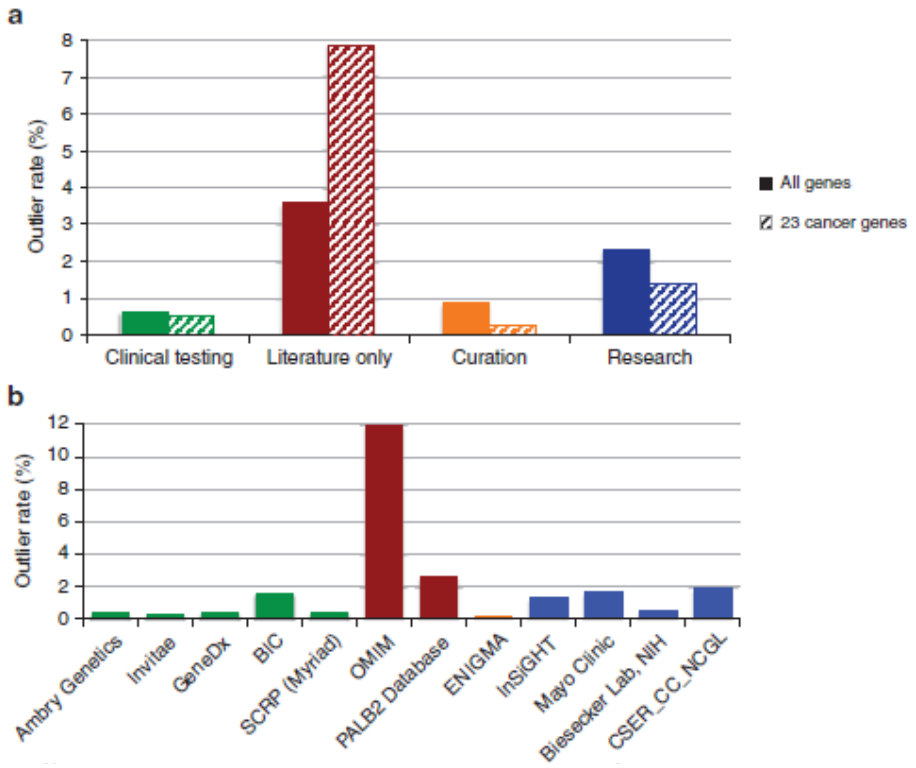


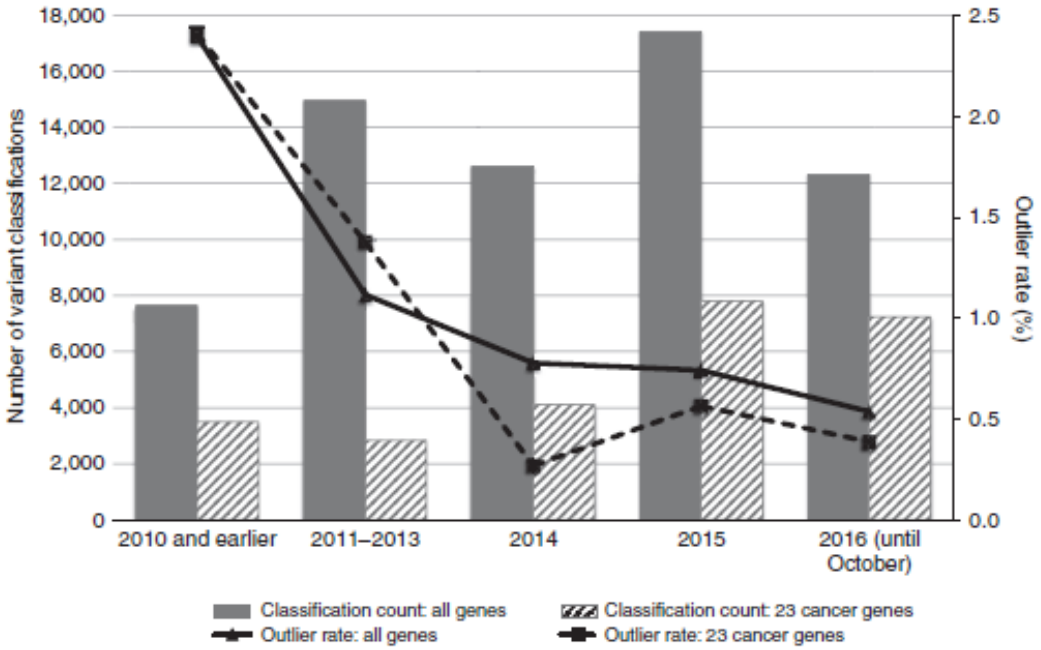
Figure 5 Concordance for ClinVar and subsets. Variant classification concordance measured as a fraction of variants for all of ClinVar and for subsets of ClinVar filtered by submission type and classification date. Concordance is calculated on an actionability basis (see text).

Are there really large inconsistencies in ClinVar ?



Are there really large inconsistencies in ClinVar ?

ORIGINAL RESEARCH ARTICLE



Summary …

Like every new method/ technology the ACMG-AMP classification guidelines need training and time

Eventually we will get used to it …..