

Is Likely Pathogenic Really 90% Likely?

A Look at the Data

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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

	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 PMS Protein length changing variant PMA	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	Nonegregation with disease BS4		Covegregation 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			

Table 5 Rules for combining criteria to classify sequence variants

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

Pathogenic
Likely pathogenic
Uncertain significance
Likely benign
Benign

ACMG/AMP guidelines suggests

Likely Pathogenic = 90% certainty of being Pathogenic

Although there is no quantitative definition of the term “likely,” guidance has been proposed in certain variant classification settings. A survey of the community during an ACMG open forum, however, suggested a much wider range of uses of the term “likely.” Recognizing this, we propose that the terms “likely pathogenic” and “likely benign” be used to mean greater than 90% certainty of a variant either being disease-causing or benign to provide laboratories with a common, albeit arbitrary, definition.

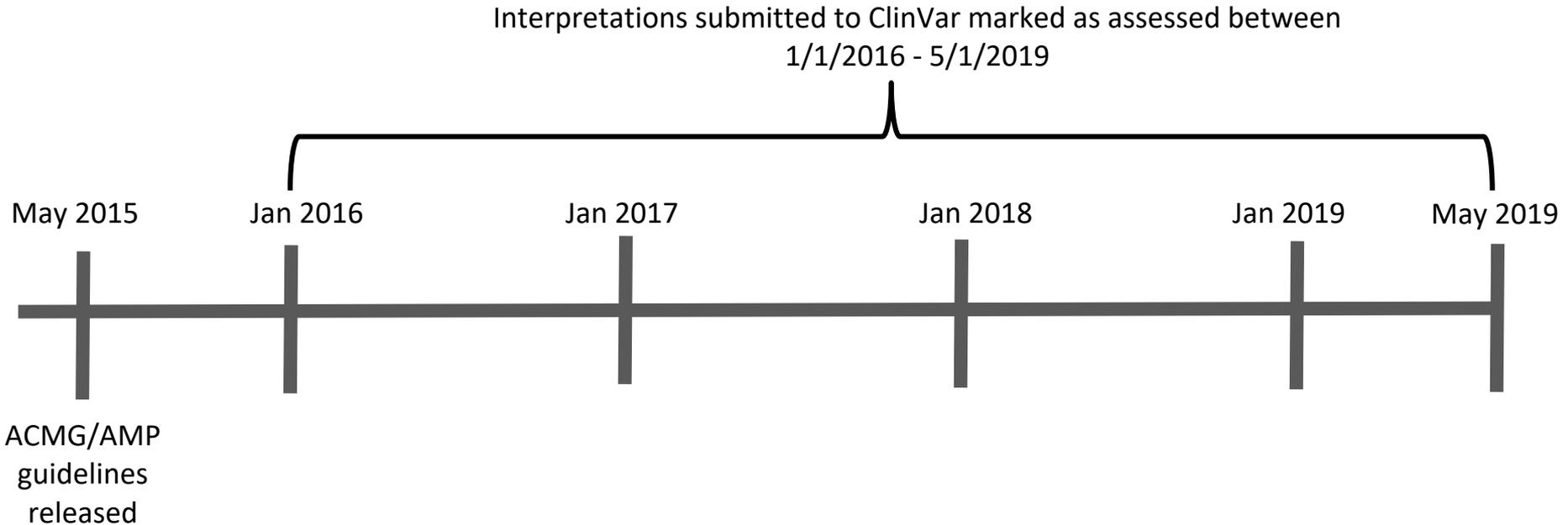
Rationale for 90%

The ACMG/AMP committee felt that 90% represented a confidence that was high enough to warrant physicians taking action as well as high enough that downgrade reclassifications (to VUS, LB, Benign) would not be frequent.

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- Understanding LP classification confidence is necessary as many clinicians treat LP and P classifications equally, meaning LP reclassifications to VUS/LB/Benign are likely unanticipated and thus clinicians and patients are less likely to check in with the lab for an update.

We sought to understand current practice by calculating the reclassification rates of variants submitted to ClinVar, choosing variants assessed after Jan 2016 (and reassessed by May 2019) in hopes of restricting to variants classified with the 2015 ACMG/AMP guideline.

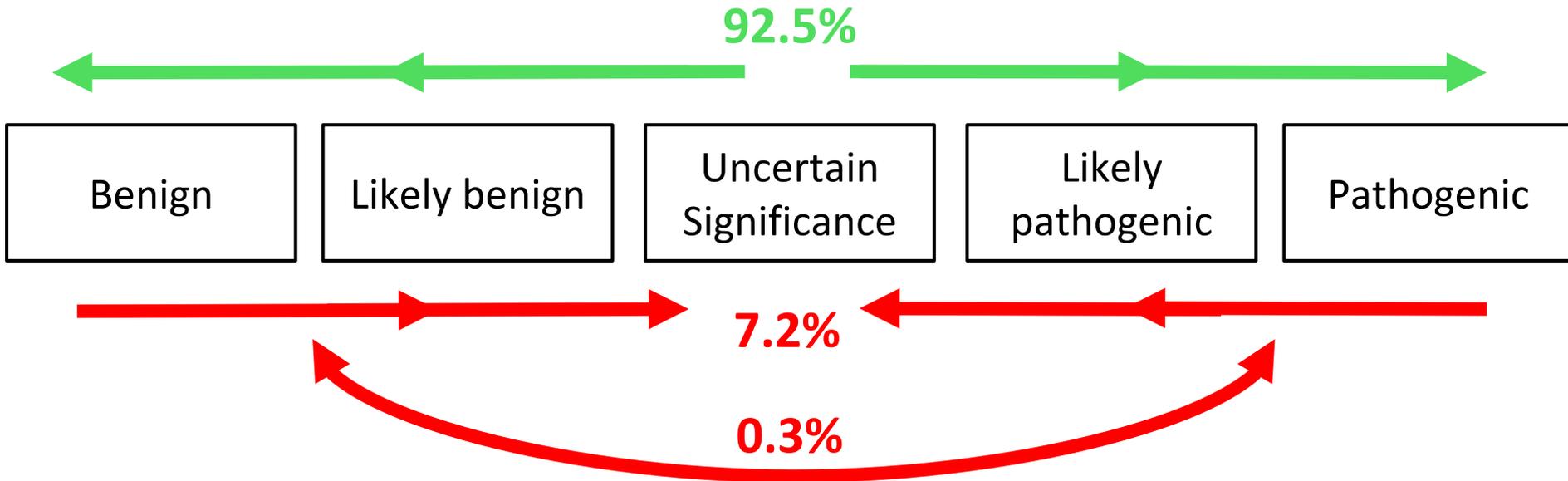


Identified all classifications in ClinVar marked as “last evaluated” after 1/1/2016

(goal to have variants classified with current guidelines)

Interpretation (Last evaluated)	Review status (Assertion criteria)	Condition (Inheritance)	Submitter	Supporting information (See all)
 Pathogenic (Aug 20, 2015)	criteria provided, single submitter (Variant Classification) Method: clinical testing	Left ventricular noncompaction cardiomyopathy	Blueprint Genetics , Accession: SCV000264100.2 Submitted: (Jan 15, 2016)	Evidence details Publications PubMed (2)
 Likely pathogenic (Feb 01, 2017)	criteria provided, single submitter (GeneDx Variant Classification (06012015)) Method: clinical testing	Not Provided	GeneDx Accession: SCV000208635.10 Submitted: (Nov 28, 2017)	Evidence details Comment: The E1801K likely pathogenic variant in the MYH7 gene has been reported in a Moldavian family with early onset distal myopathy and later onset, severe ... (more)
 Pathogenic (Nov 25, 2014)	criteria provided, single submitter (LMM Criteria) Method: clinical testing	Cardiomyopathy (Autosomal dominant inheritance)	Laboratory for Molecular Medicine, Partners HealthCare Personalized Medicine Accession: SCV000059616.5 Submitted: (Feb 23, 2018)	Evidence details Publications PubMed (4) Comment: The p.Glu1801Lys variant in MYH7 has been reported in 1 individual with early onset distal myopathy and late onset DCM (Udd 2009) as well as ... (more)

- Between **Jan 2016 - May 2019**, there were **552,134 classifications** submitted to ClinVar assessed on or after 1/1/2016
 - **4,445 (0.8%)** reclassified and updated in ClinVar by **5/1/2019**
 - **92.5%** moved to a classification category of more certainty
 - **7.5%** moved to a less certain (7.2%) or opposing (0.3%) category.



Reclassification Type	#	% of all reclass
Uncertain significance to Likely benign	1586	35.68%
Likely benign to Benign	959	21.57%
Likely pathogenic to Pathogenic	589	13.25%
Uncertain significance to Likely pathogenic	470	10.57%
Uncertain significance to Benign	341	7.67%
Uncertain significance to Pathogenic	165	3.71%
Likely pathogenic to Uncertain significance	145	3.26%
Likely benign to Uncertain significance	65	1.46%
Pathogenic to Likely pathogenic	63	1.42%
Pathogenic to Uncertain significance	36	0.81%
Benign to Likely benign	11	0.25%
Pathogenic to Benign	4	0.09%
Likely pathogenic to Likely benign	4	0.09%
Benign to Likely pathogenic	3	0.07%
Likely pathogenic to Benign	1	0.02%
Likely benign to Likely pathogenic	1	0.02%
Benign to Pathogenic	1	0.02%
Benign to Uncertain significance	1	0.02%

Change to more certainty

Change to less certainty or opposite category

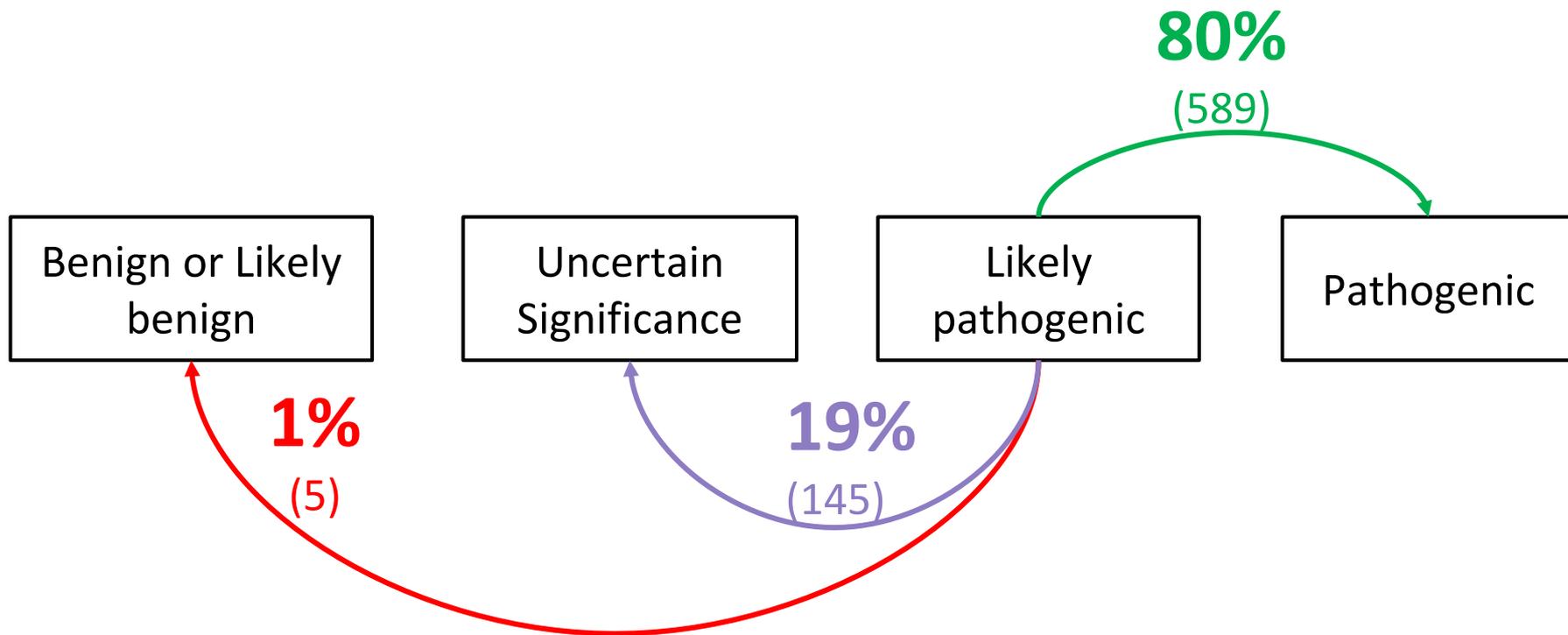
Likely pathogenic classifications have highest reclassification rate

	# Assertions submitted to ClinVar	% Not Reclassified	% Reclassified
Pathogenic	61,338	99.83% (61,235)	0.17% (103)
Likely pathogenic	35,731	97.93% (34,992)	2.07% (739)
Uncertain significance	262,611	99.02% (260,045)	0.98% (2,566)
Likely benign	136,164	99.25% (135,137)	0.75% (1,027)
Benign	56,290	99.97% (56,273)	0.03% (17)

...however vast majority of classifications did not change between 1/1/2016 - 5/1/2019

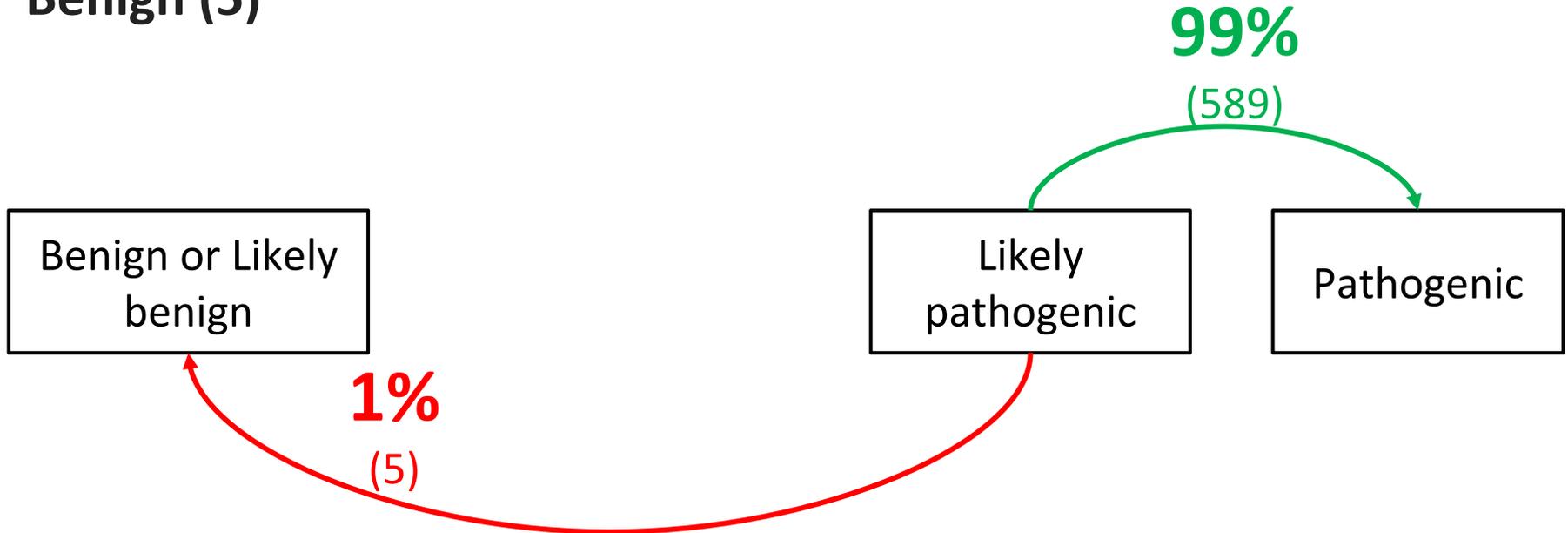
Likely Pathogenic Reclassifications

739 Likely pathogenic classifications completed after 1/1/2016 had been reclassified and updated in ClinVar by 5/1/2019



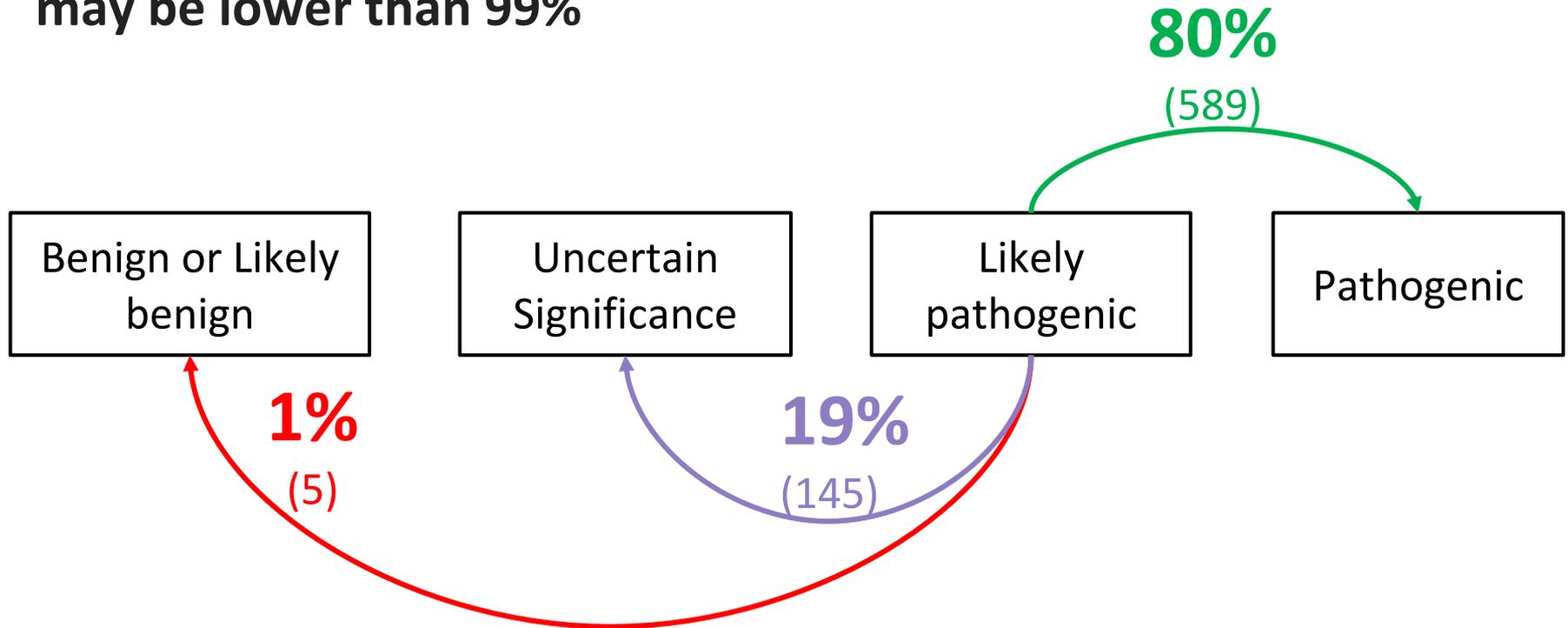
Likely Pathogenic Reclassifications

If only including LP reclassifications to a more definitive category (LP to P or LP to LB/B), **Likely pathogenic reclassification rates suggests a 99% (589/594) certainty of being Pathogenic (589) compared to Benign (5)**



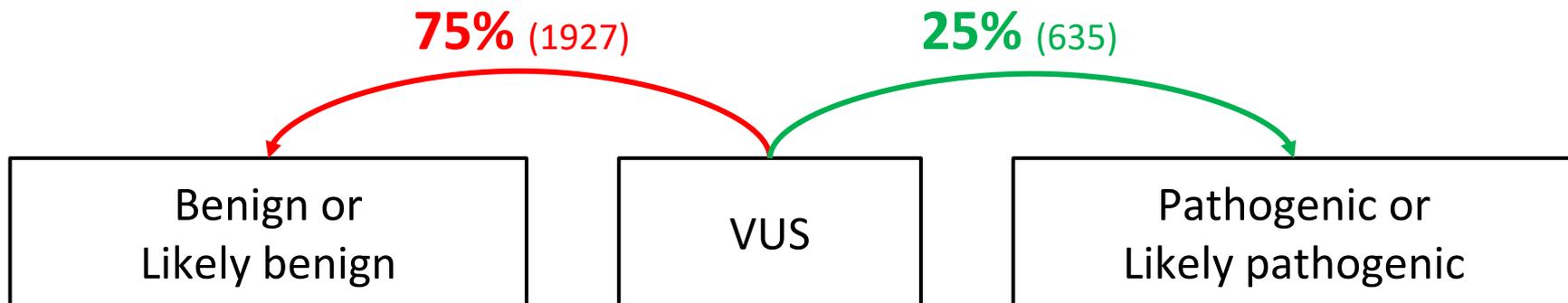
Likely Pathogenic Reclassifications

However, **19% (145/739)** of LPs dropped to VUS suggesting that some of these may eventually move to LB/B and the rate of LP to P may be lower than **99%**



Uncertain Significance Reclassifications

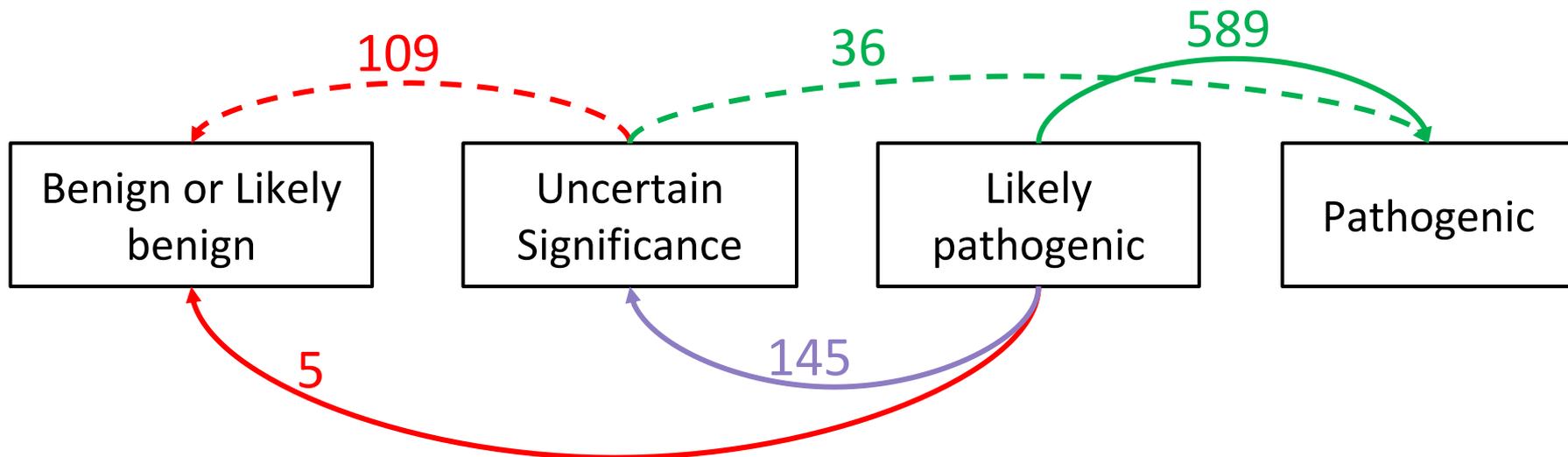
2,562 Uncertain significance classifications completed after 1/1/2016 had been reclassified and updated in ClinVar by 5/1/2019



Can extrapolate reclassification of “LP to VUS” variants from VUS reclassification trends...

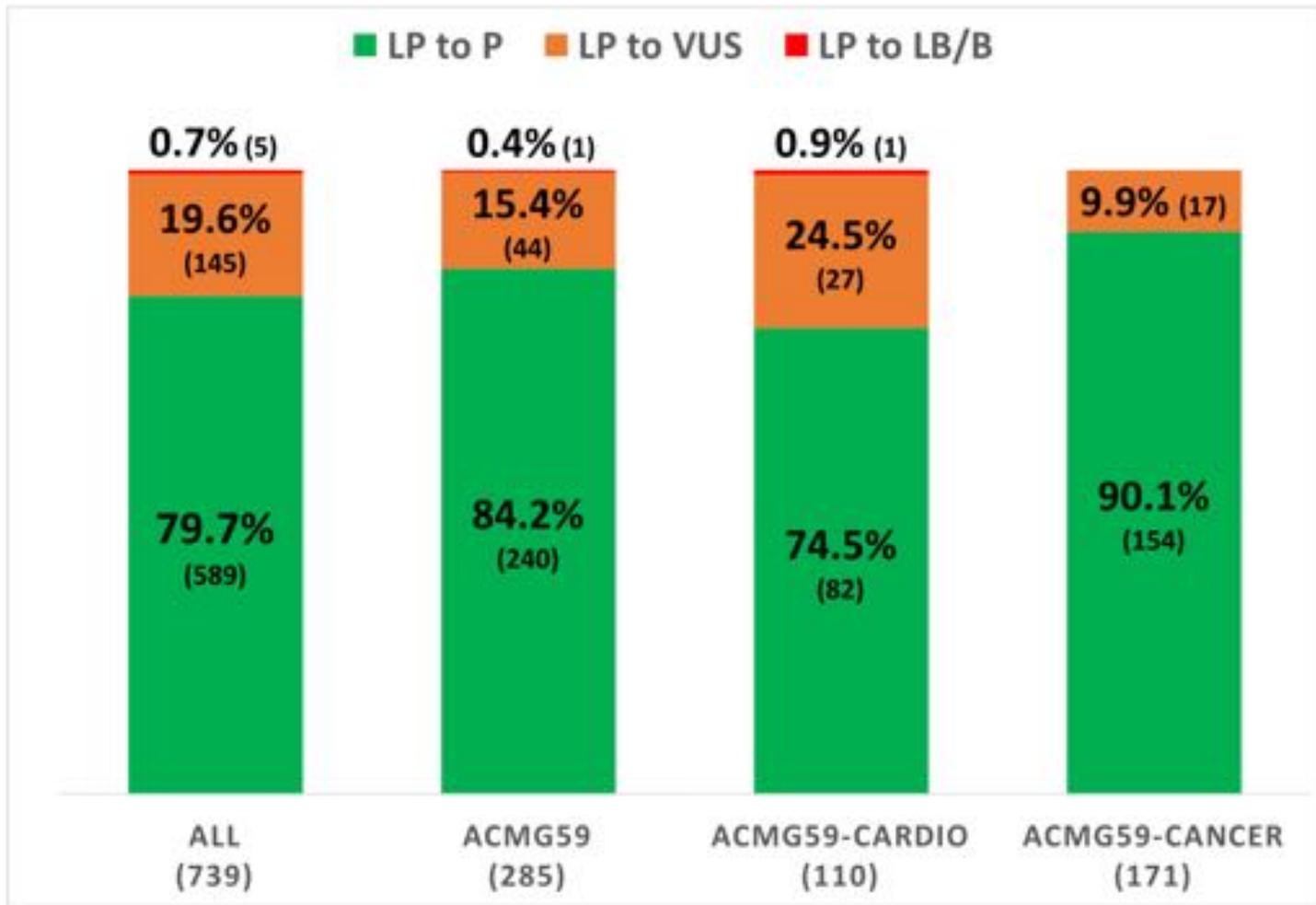
Likely Pathogenic Reclassifications (*Worst Case*)

Applying VUS reclassification rate to “LP to VUS” variants shows overall extrapolated **Likely pathogenic rate of 85% certainty of being Pathogenic (589+36) compared to Benign (5+109)**

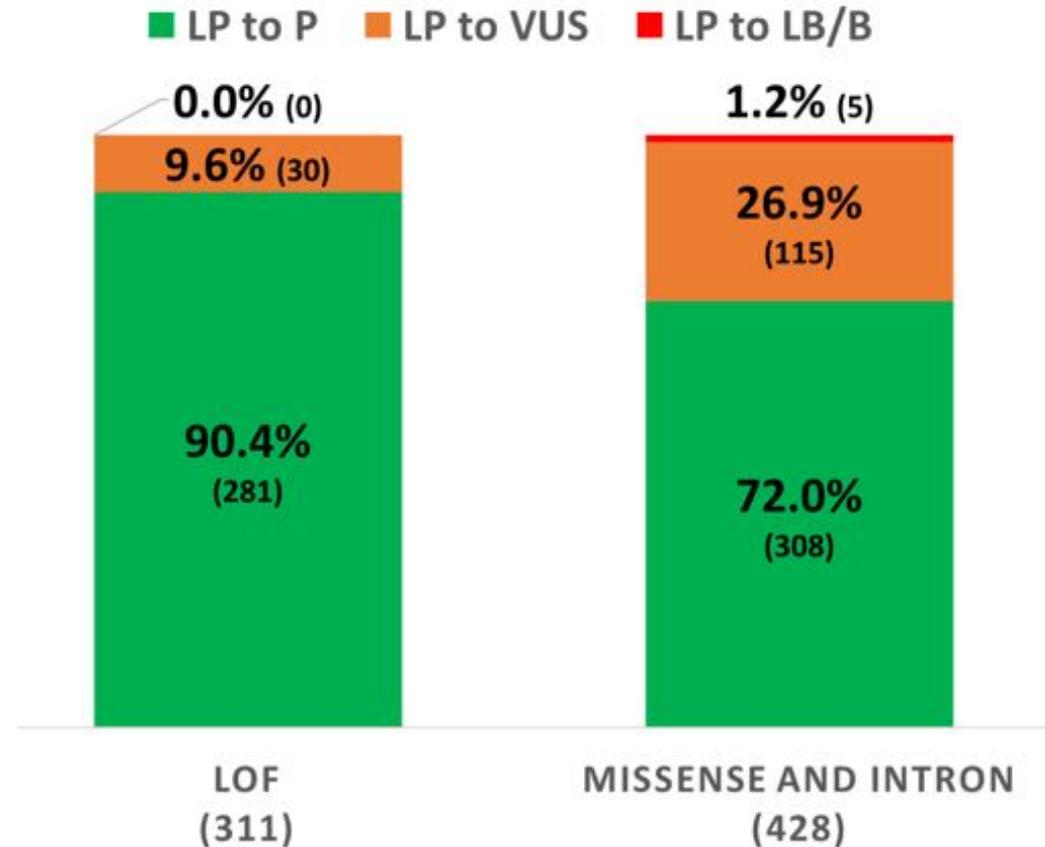


However, the rate of LP to VUS to LB/B is probably smaller than the rate of VUS to LB/B

Likely Pathogenic reclassification rates by disease area



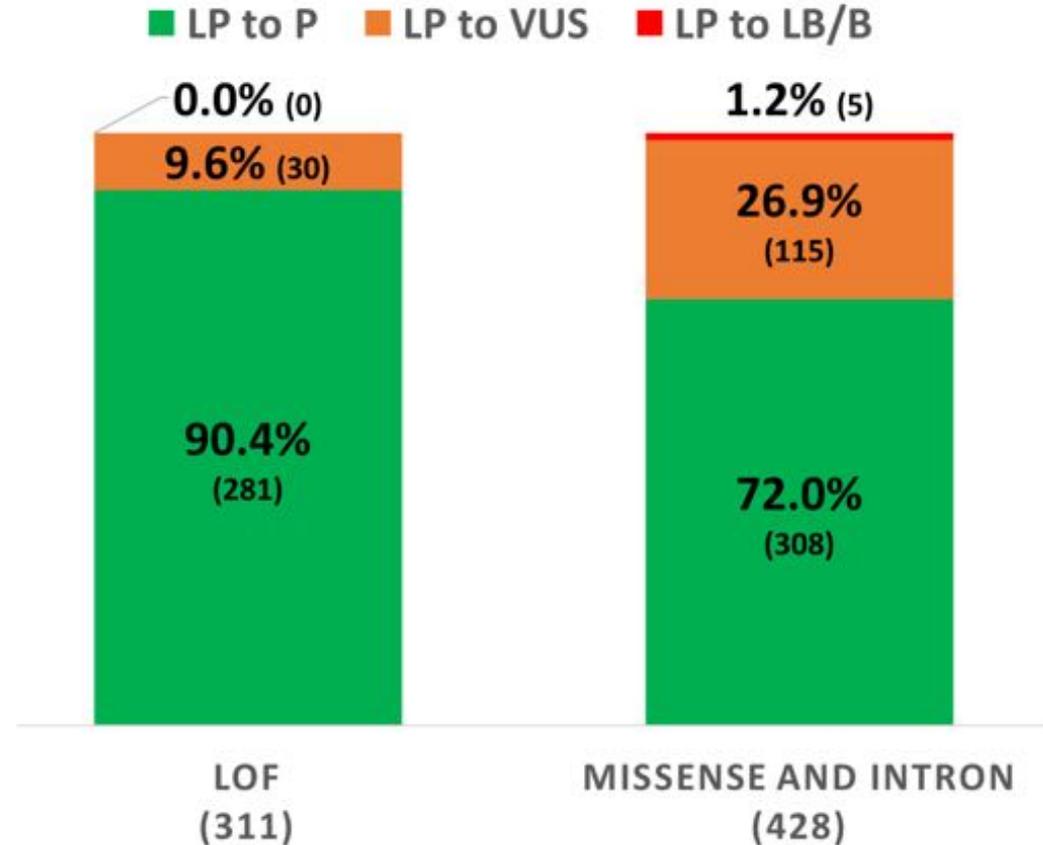
Likely Pathogenic reclassification rates by variant type



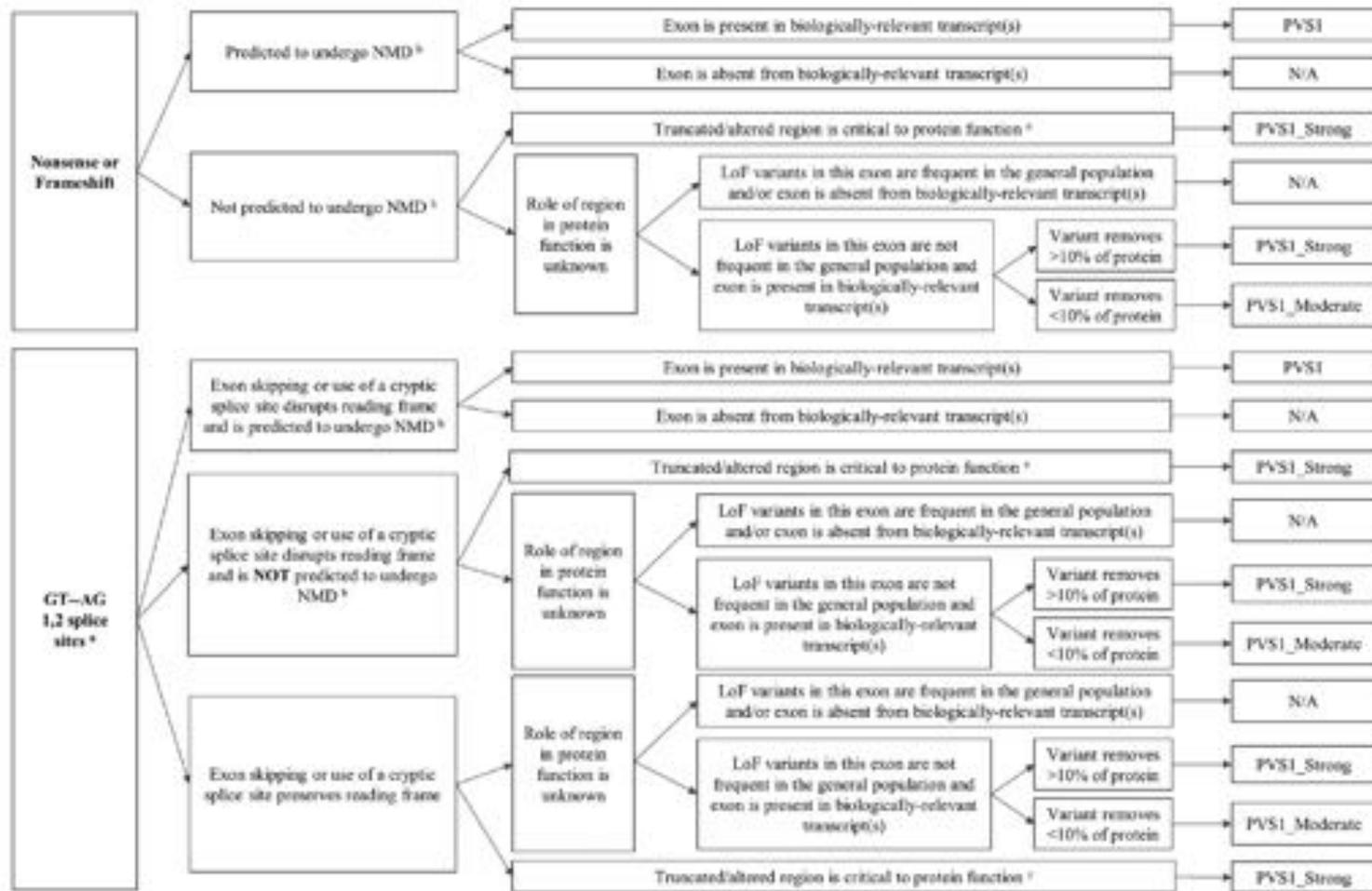
Likely Pathogenic reclassification rates by variant type

Of 30 LoF variants reclassified from LP to VUS, a review of the submitted evidence summaries suggests:

- **30%** (9) downgraded due to uncertainty in gene/disease evidence/mutation mechanism
- **57%** (17) downgraded due to uncertainty in variant impact
 - **20%** (6) rescue by alt splice site
 - **20%** (6) occur in last exon
 - **13%** (4) gene uses alt start site



PVS1 Decision Tree (Abou Tayoun 2018; PMID:30192042)



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- Reclassification data from ClinVar shows that **99% of LP classifications move to P compared to LB/B**, suggesting that application of the term is consistent with the intended confidence level.
- Vast majority of LPs still remain as LP within a three year window and a small subset (0.4%) dropped to VUS suggesting that more data and a longer time of analysis will be needed to more robustly evaluate the rate of LP reclassification

Acknowledgements

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ClinVar Submitters

