Issues regarding data reanalysis for actionable variants

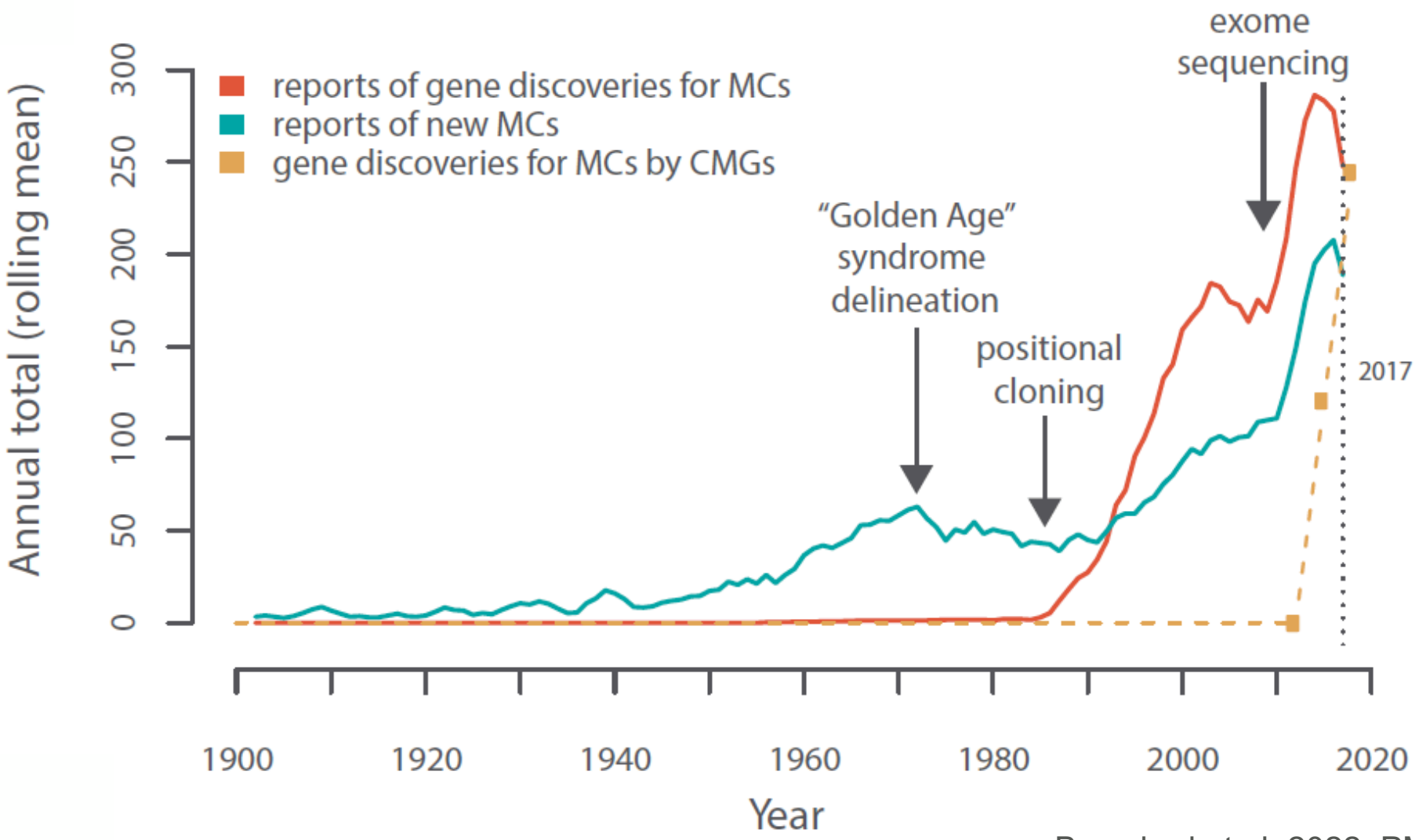
Natasha Strande, PhD, DABMGG, FACMG
Assistant Professor
Dept. Genomic Health and Autism & Developmental Medicine Institute
Clinical Laboratory Director, MyCode

Geisinger

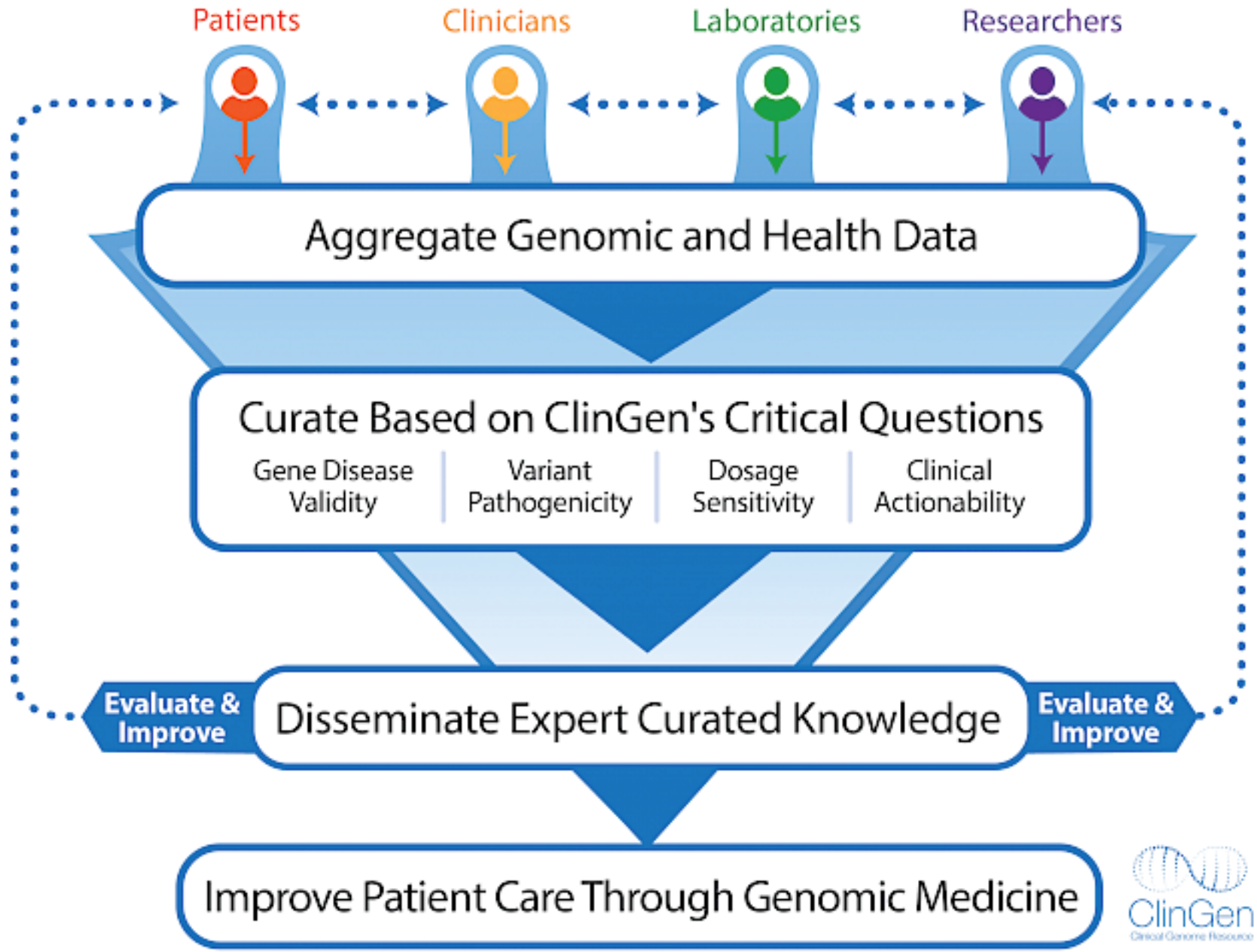
Outline

- Review variant and gene classification schemes and evolution
- Rate of variant re-classification over time
- MyCode reanalysis experience with updating gene lists
- Policies regarding reanalysis

Continued Gene Discovery



Bamshad et al, 2022; PMID: 31491408


Patients
Clinicians
Laboratories
Researchers
Aggregate Genomic and Health Data
Curate Based on ClinGen's Critical Questions
Gene Disease Validity
Variant Pathogenicity
Dosage Sensitivity
Clinical Actionability
Evaluate & Improve
Disseminate Expert Curated Knowledge
Evaluate & Improve
Improve Patient Care Through Genomic Medicine
ClinGen

Gene Disease Validity

GENE/DISEASE PAIR:				
Assertion criteria	**Genetic Evidence (0-12 points)**	**Experimental Evidence (0-6 points)**	**Total Points (0-18)**	**Replication Over Time (Y/N)**
Description	Case-level, family segregation, or case-control data that support the gene-disease association	Gene-level experimental evidence that support the gene-disease association	Sum of Genetic & Experimental Evidence	> 2 pubs w/ convincing evidence over time (>3 yrs.)
Assigned Points	A	B	C	D
CALCULATED CLASSIFICATION		**LIMITED**	**0.1-6**	
		MODERATE	**7-11**	
		STRONG	**12-18**	
		DEFINITIVE	**12-18 & Replicated Over Time**	
Valid contradictory evidence (Y/N)*	**List PMIDs and describe evidence:** E			
CURATOR CLASSIFICATION	F			
FINAL CLASSIFICATION	G			

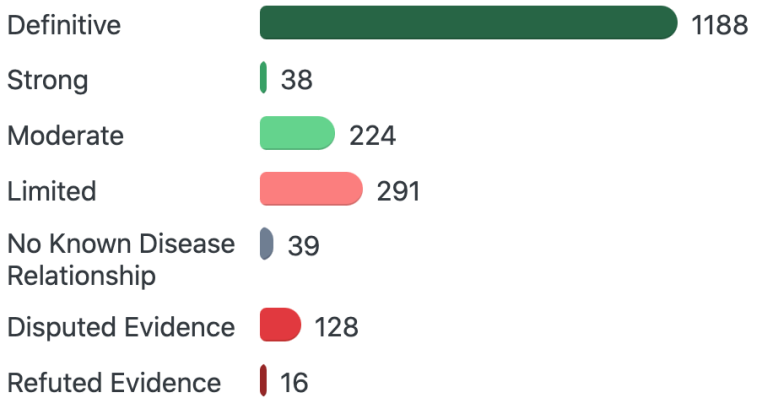
Strande & Riggs et al., 2017; PMID: 28552198

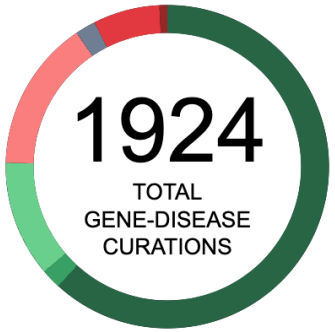
Gene-Disease Clinical Validity Statistics

The ClinGen Gene-Disease Clinical Validity curation process involves evaluating the strength of evidence supporting or refuting a claim that variation in a particular gene causes a particular disease.

Classification Statistics

Gene-Disease Clinical Validity has **1924 curations** encompassing **1586 genes**.

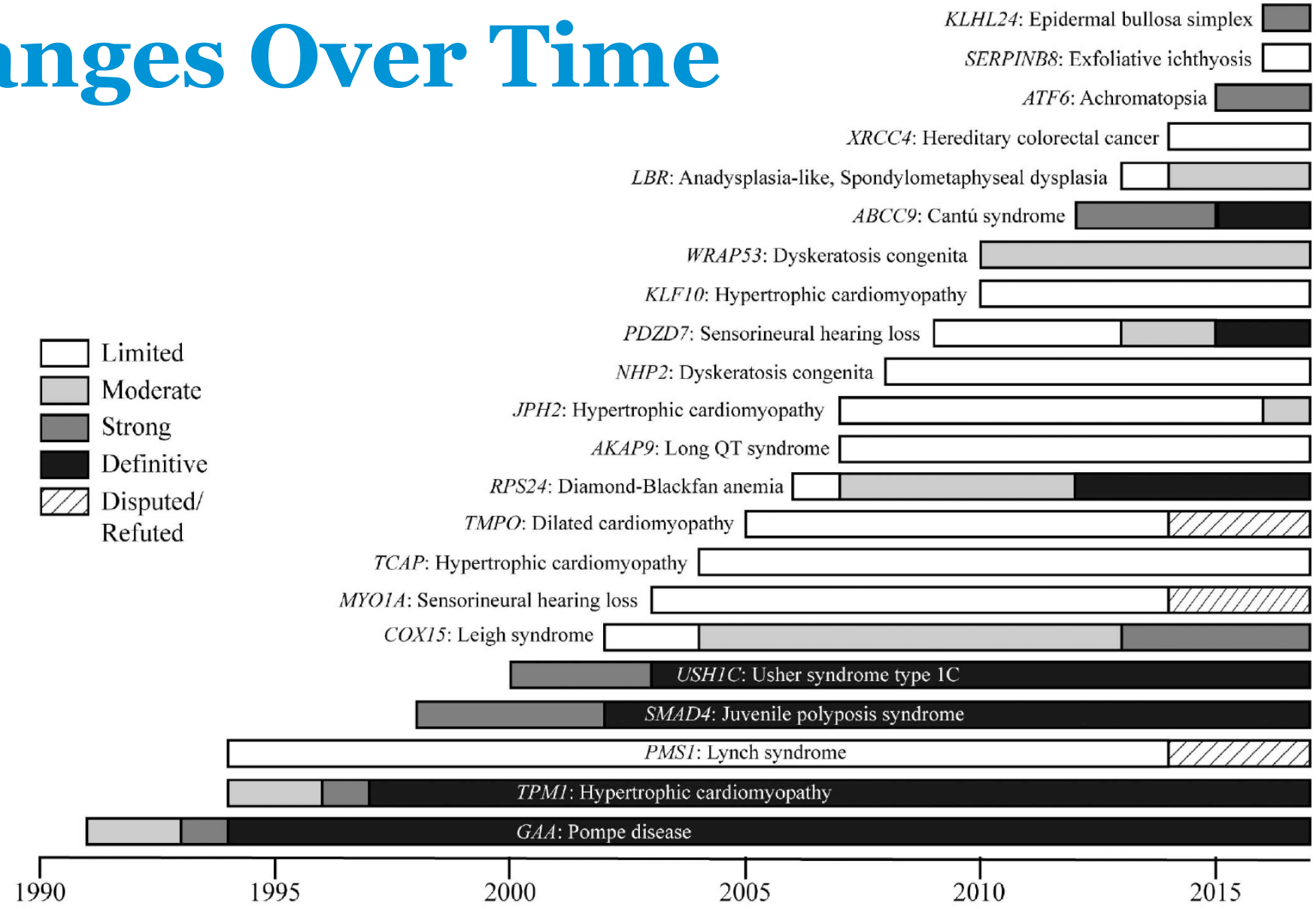




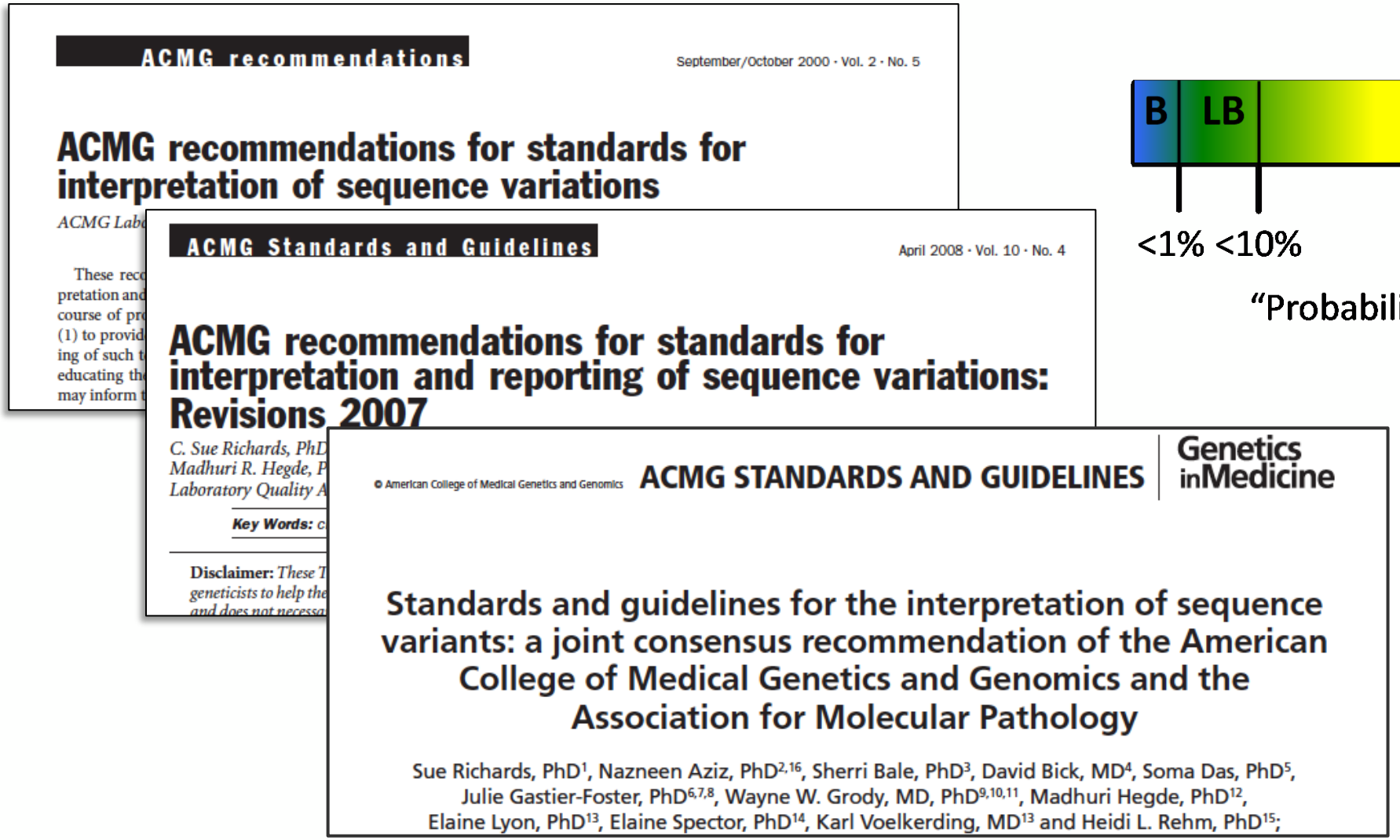
ClinGen
Clinical Genome Resource

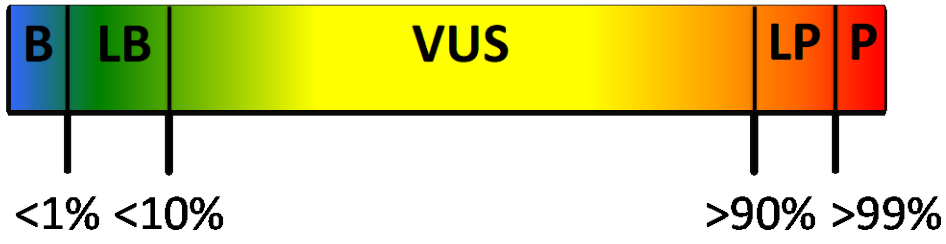
McGlaughon et al., 2018; PMID: 30311372

Changes Over Time



Variant Classification Guidelines





"Probability of pathogenicity"

Classification Criteria

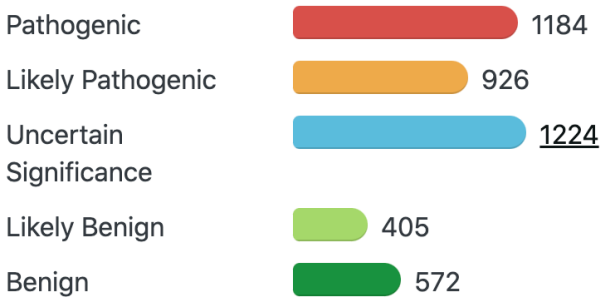
		BENIGN CRITERIA		PATHOGENIC CRITERIA			
Strength of evidence		Strong	Supporting	Supporting	Moderate	Strong	Very Strong
Odds of Pathogenicity*		–18.7	–2.08	2.08	4.33	18.7	350.0
Evidence Category and Corresponding ACMG/AMP Codes	Population Data	*BA1*+ BS1 BS2			PM2	PS4	
	Allelic Evidence & Cosegregation Data	BS4	BP2 BP5	PP1 →	PM3 PM6	PS2	
	Computation & Predictive Data		BP1 BP3 BP4 BP7	PP2 PP3	PM1 PM4 PM5	PS1	PVS1
	Functional Data	BS3				PS3	
	Other		BP6	PP4 PP5			

Strande, et al, 2018; PMID: 29988079

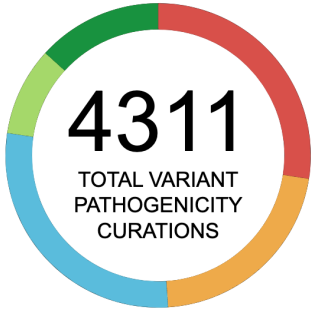
Evolving Classification Guidelines

Classification Statistics

Variant Pathogenicity has **4311 curations**.



21 Approved ClinGen Variant Curation Expert Panels



ClinGen
Clinical Genome Resource

Expert Panel Name	Type	CDWG	Status
ACADVL Variant Curation Expert Panel	VCEP	Inborn Errors of Metabolism CDWG	
Brain Malformations Variant Curation Expert Panel	VCEP	Neurodevelopmental Disorders CDWG	
Cardiomyopathy Variant Curation Expert Panel	VCEP	Cardiovascular CDWG	
CDH1 Variant Curation Expert Panel	VCEP	Hereditary Cancer CDWG	
Cerebral Creatine Deficiency Syndromes Variant Curation Expert Panel	VCEP	Inborn Errors of Metabolism CDWG	
DICER1 and miRNA-Processing Gene Variant Curation Expert Panel	VCEP	Hereditary Cancer CDWG	
Familial Hypercholesterolemia Variant Curation Expert Panel	VCEP	Cardiovascular CDWG	
FBN1 Variant Curation Expert Panel	VCEP	Cardiovascular CDWG	
Glaucoma Variant Curation Expert Panel	VCEP	Ocular CDWG	
Hearing Loss Variant Curation Expert Panel	VCEP	Hearing Loss CDWG	
Hereditary Breast, Ovarian and Pancreatic Cancer Variant Curation Expert Panel	VCEP	Hereditary Cancer CDWG	
Lysosomal Storage Disorders Variant Curation Expert Panel	VCEP	Inborn Errors of Metabolism CDWG	
Malignant Hyperthermia Susceptibility Variant Curation Expert Panel	VCEP	Other	
Mitochondrial Disease Nuclear and Mitochondrial Variant Curation Expert Panel	VCEP	Inborn Errors of Metabolism CDWG	
Monogenic Diabetes Variant Curation Expert Panel	VCEP	Inborn Errors of Metabolism CDWG	
Myeloid Malignancy Variant Curation Expert Panel	VCEP	Hereditary Cancer CDWG	
Phenylketonuria Variant Curation Expert Panel	VCEP	Inborn Errors of Metabolism CDWG	
Platelet Disorders Variant Curation Expert Panel	VCEP	Hemostasis/Thrombosis CDWG	
PTEN Variant Curation Expert Panel	VCEP	Hereditary Cancer CDWG	
RASopathy Variant Curation Expert Panel	VCEP	RASopathy CDWG	
Rett and Angelman-like Disorders Variant Curation Expert Panel	VCEP	Neurodevelopmental Disorders CDWG	
TP53 Variant Curation Expert Panel	VCEP	Hereditary Cancer CDWG	

Rates of Variant Reclassification

Variable Rates

Volume 21 | Number 10 | October 2019 | GENETICS in MEDICINE

Variant classification changes over time in *BRCA1* and *BRCA2*

Chloe Mighton, BSc[1,2], George S. Charames, PhD FACMG[3,4,5], Marina Wang, MD[4], Kathleen-Rose Zakoor, MBinf[4,5], Andrew Wong, MSc[4], Salma Shickh, MS CGC[1,2], Nicholas Watkins, MSc CGC/CCGC[4], Matthew S. Lebo, PhD FACMG[6,7], Yvonne Bombard, PhD[1,2] and Jordan Lerner-Ellis, PhD FACMG[3,4,5]

12.4% variants

Analyzing and Reanalyzing the Genome: Findings from the MedSeq Project

22% participants

Kalotina Machini,[1,2,3] Ozge Ceyhan-Birsoy,[1,7] Danielle R. Azzariti,[1,4] Himanshu Sharma,[1] Peter Rossetti,[1] Lisa Mahanta,[1] Laura Hutchinson,[1] Heather McLaughlin,[1,8] The MedSeq Project, Robert C. Green,[3,4,5] Matthew Lebo,[1,2,3,4,9] and Heidi L. Rehm[1,2,3,4,6,9,*]

The American Journal of Human Genetics *105*, 177–188, July 3, 2019 **177**

Highly dependent on the type and date of initial classification type

JAMA | Original Investigation

6.4% variants

Prevalence of Variant Reclassification Following Hereditary Cancer Genetic Testing

Jacqueline Mersch, MS, CGC; Nichole Brown, MS, CGC; Sara Pirzadeh-Miller, MS, CGC; Erin Mundt, MS, CGC; Hannah C. Cox, PhD; Krystal Brown, PhD; Melissa Aston, BS; Lisa Esterling, PhD; Susan Manley, MS, CGC, MBA; Theodora Ross, MD, PhD

Analysis of hereditary cancer gene variant classifications from ClinVar indicates a need for regular reassessment of clinical assertions

0.6 – 6.4% variants

Aimee L. Davidson[1,2] | Olga Kondrashova[1] | Conrad Leonard[1] | Scott Wood[1] | Emma Tudini[1,3] | Georgina E. Hollway[1] | John V. Pearson[1] | Felicity Newell[1] | Amanda B. Spurdle[1] | Nicola Waddell[1,2]

Directionality of Changes

Table 1 Summary of classification and reclassification from ClinVar (Jan 2016–July 2019)

Starting classification (n)	Percentage reclassified (n)	Reclassification type (n)	Percentage of initial classification group	Percentage of all reclassifications
Pathogenic (63,658)	0.17% (110)	P → LP (64)	58.2%	1.4%
		P → VUS (41)	37.3%	0.91%
		P → LB (1)	0.91%	0.02%
		P → B (4)	3.6%	0.09%
Likely pathogenic (36,808)	2.16% (796)	LP → P (625)	78.5%	13.9%
		LP → VUS (165)	20.7%	3.7%
		LP → LB (4)	0.50%	0.09%
		LP → B (2)	0.25%	0.04%
Uncertain significance (272,581)	0.95% (2584)	VUS → P (171)	6.6%	3.8%
		VUS → LP (486)	18.8%	10.8%
		VUS → LB (1586)	61.4%	35.2%
		VUS → B (341)	13.2%	7.6%
Likely benign (140,779)	0.71% (996)	LB → P (2)	0.20%	0.04%
		LB → LP (2)	0.20%	0.04%
		LB → VUS (66)	6.6%	1.5%
		LB → B (926)	93.0%	20.6%
Benign (58,024)	0.03% (15)	B → P (1)	6.7%	0.02%
		B → LP (3)	20.0%	0.07%
		B → VUS (1)	6.7%	0.02%
		B → LB (10)	66.7%	0.22%

Abbreviations: *B* Benign, *LB* Likely benign, *LP* Likely pathogenic, *P* Pathogenic, *VUS* Variant of uncertain significance

Harrison and Rehm, 2019; PMID: 31752965

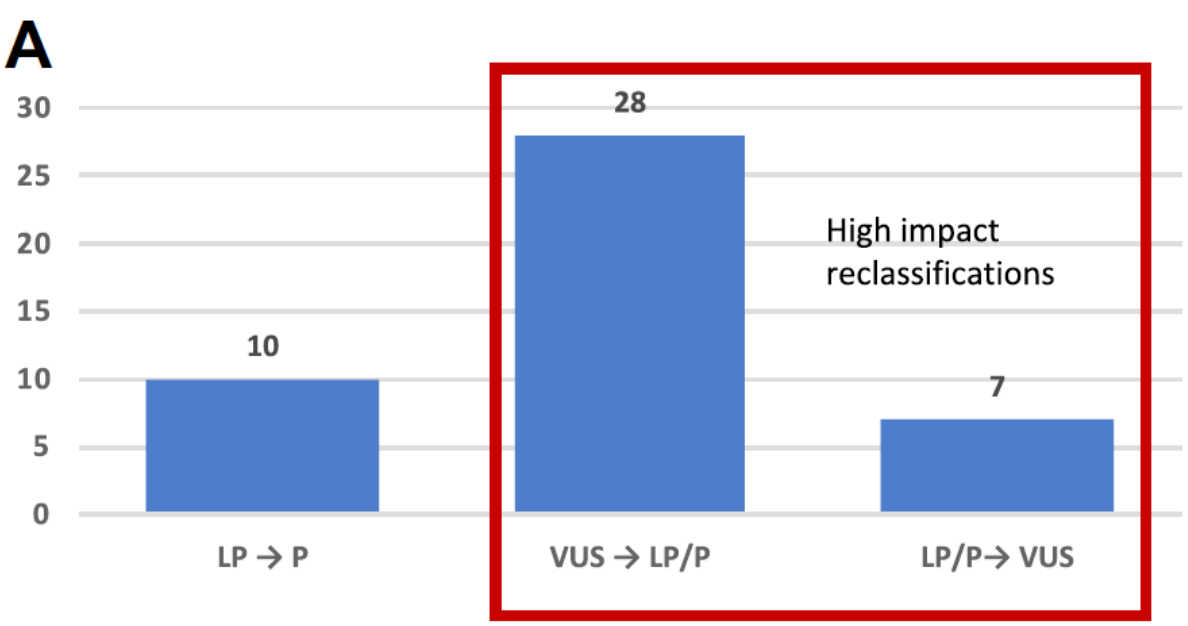
Table 1 Number of *BRCA1/2* variants that AMDL had submitted to ClinVar for which there were discordant submissions

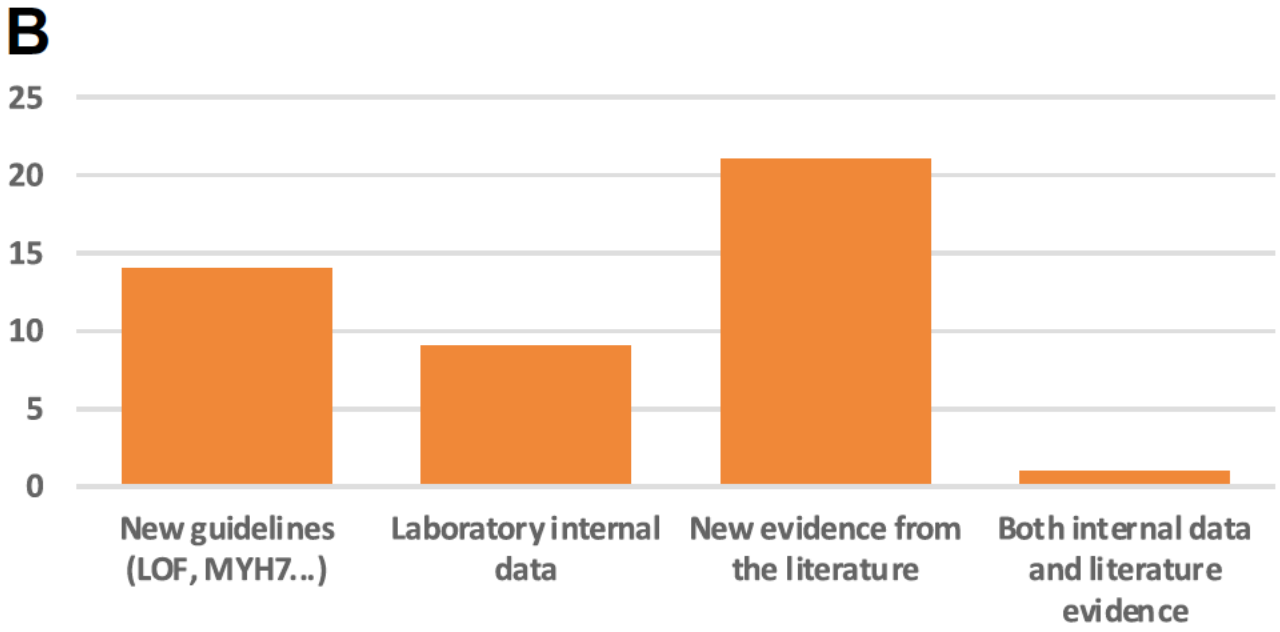
	Number of variants with discordant ClinVar submissions (total $n = 488$)
Discrepancy across two ACMG/AMP levels	
Likely Benign/Benign	14.8% (72/488)
Likely Pathogenic/ Pathogenic	9.2% (45/488)
Discrepancy across three ACMG/AMP levels	
Benign/Likely Benign/ VUS	68.6%% (335/488)
Pathogenic/Likely Pathogenic/VUS	6.1%% (30/488)
Discrepancy across five ACMG/AMP levels	
Pathogenic/VUS/Likely Benign/Benign	0.6% (3/488)
Different classification system	
Pathogenic/Risk Factor	0.6% (3/488)

ACMG/AMP American College of Medical Genetics and Genomics/Association for Molecular Pathology, *ADML* Advanced Molecular Diagnostics Laboratory, *VUS* variant of uncertain significance.

Mighton et al, 2019; PMID: 31043710

Reanalysis of eMERGE data





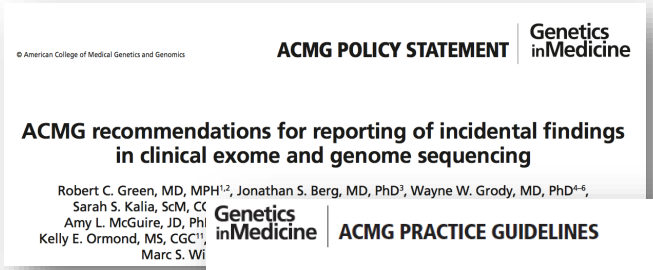
Zouk et al, 2022; PMID: 34906510

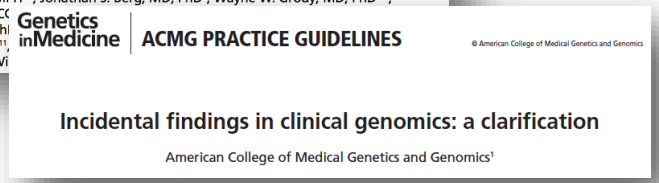
Evolving ACMG SF Recommendations

Presidential Commission *for the* Study of Bioethical Issues

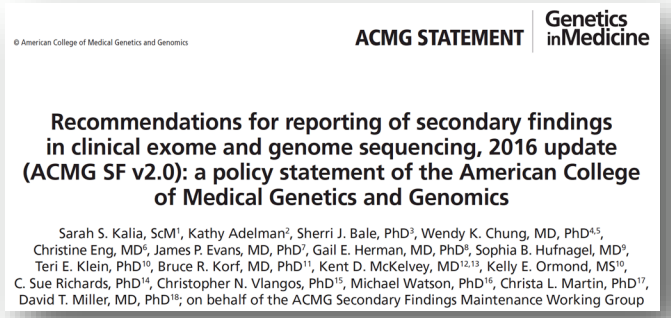
Dec. 2013

July — 56 genes



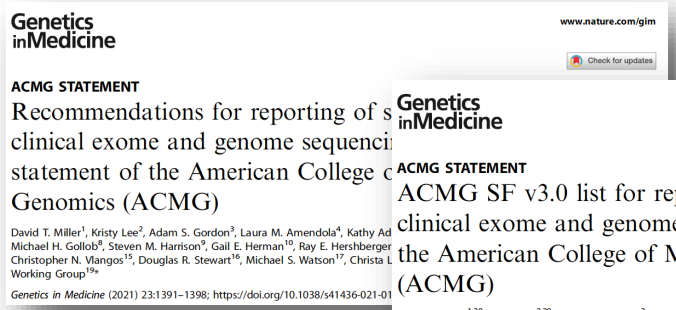


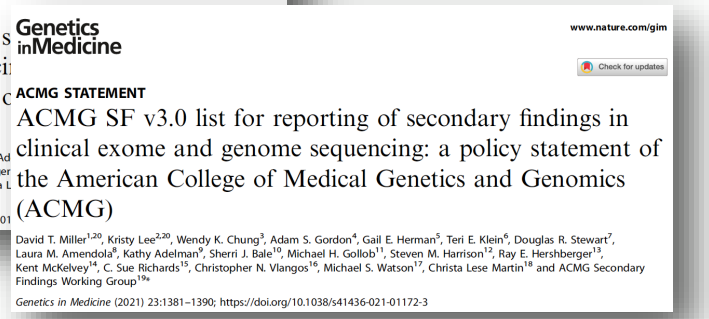
Feb. 2017

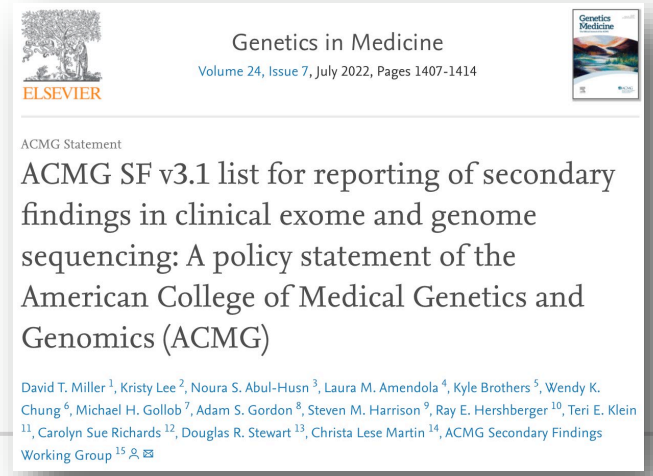


2021

March



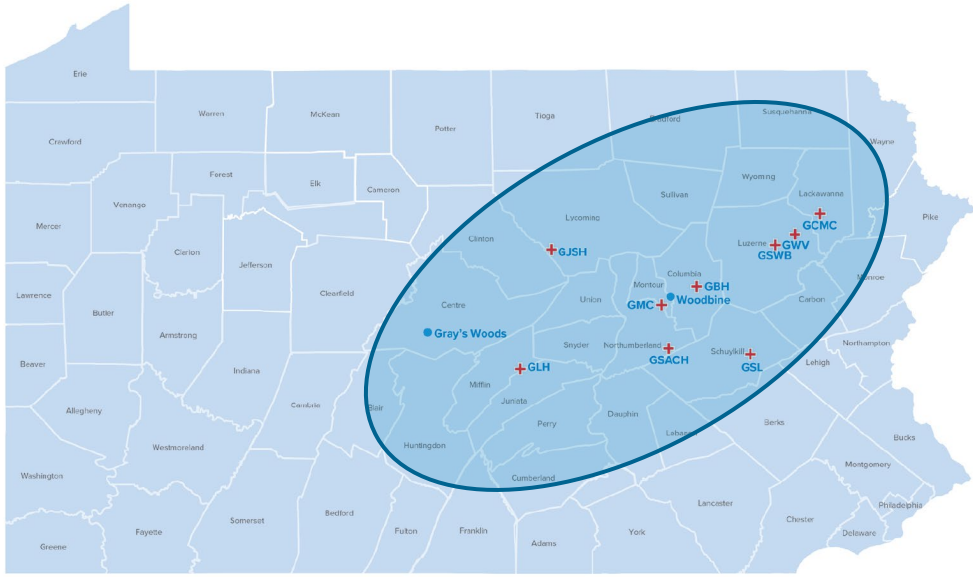




Experiences from Geisinger's MyCode Biobank

Geisinger

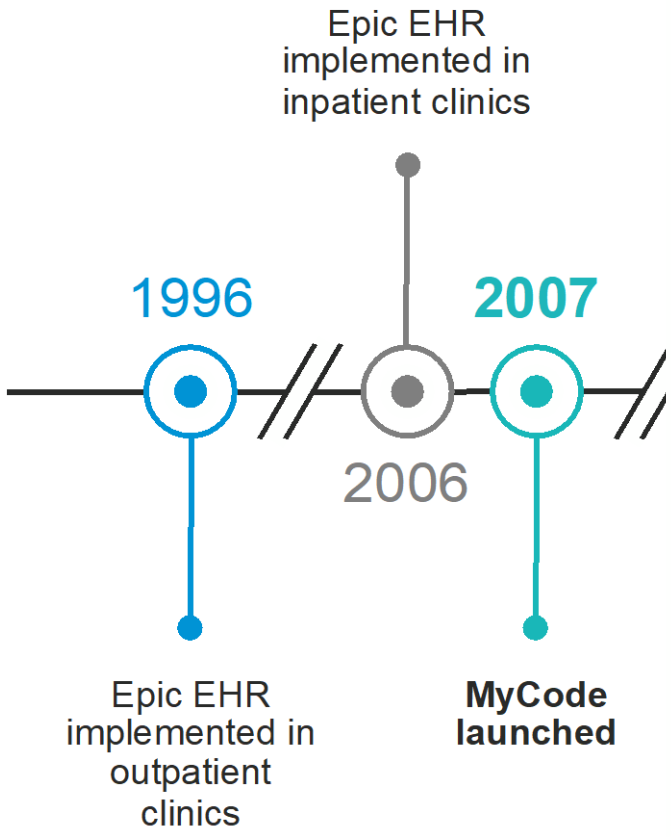
- Integrated healthcare system in Central and Northeast Pennsylvania
- Large, stable population of >3M patients, including many multi-generation families
- Longstanding EHR with comprehensive clinical data
- Strong, trusting relationship between patients and Geisinger



PENNSYLVANIA

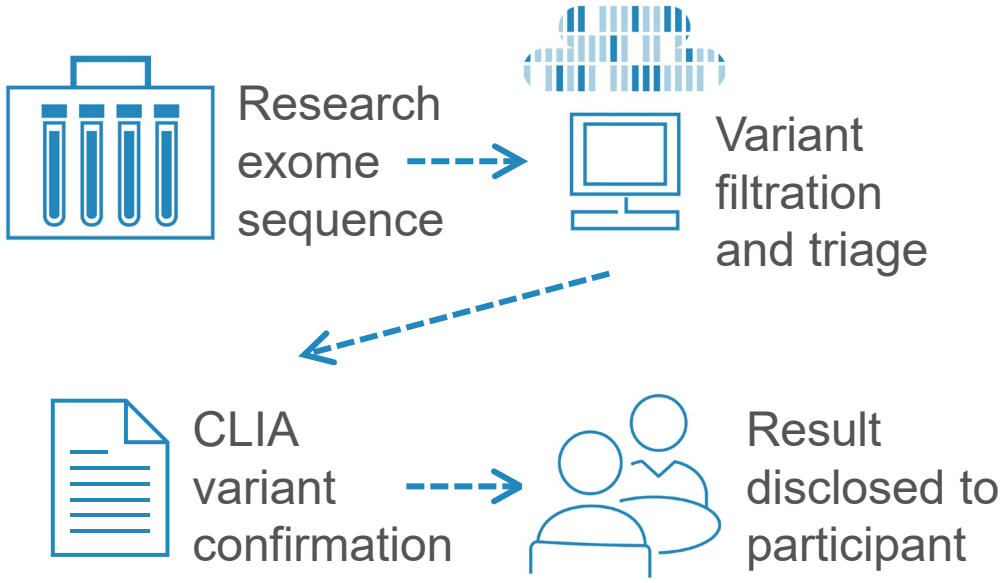
Adapted from Kelly et al. 2021 *Am J Med Genet C* PMID:33576083

MyCode Timeline



1. MyCode Community Health Initiative is a **precision medicine research project** at Geisinger
2. Includes a system-wide **biobank** designed to store blood and other samples for research use by Geisinger and Geisinger collaborators

Genomic Screening via MyCode



Opportunistic Screening

Final Gene List

Gene Validity
Is gene associated with disease?

Access to Care
Is care available locally?

Clinical Utility
Is this information actionable?

Clinical Expertise
Are there local clinical experts?

Secondary Findings
Are there SF recommendations?

Newly Added SF v3.0 Genes

Cancer

Hereditary breast and ovarian cancer: *BRCA1/2*, ***PALB2***

Lynch syndrome:

MLH1, *MSH2*, *MSH6*, *PMS2*

Familial Adenomatous Polyposis: *APC*

Endocrine tumor syndromes:

6 genes + ***MAX*** & ***TMEM127*** for pheochromocytoma & paragangliomas

+ 10 other cancer conditions

Cardiovascular

Vasculopathies: 7 genes

Cardiomyopathies

(HCM, DCM, ARVC):

16 genes + ***FLNC, TTN*** for DCM

Inherited arrhythmias:

4 genes + ***CASQ2*** & ***TRDN*** (AR) for CPVT

Familial hypercholesterolemia: *APOB*, *LDLR*, & *PCSK9*

Miscellaneous

Malignant hyperthermia: *RYR1* & *CACNA1S*

Wilson disease (AR): *ATP7B*

Hemochromatosis: *HFE* C282Y homozygotes

Hereditary Hemorrhagic telangiectasia: *SMAD4*, ***ACVRL1*** & ***ENG***

MODY: ***HNF1A***

Retinopathy (AR): ***RPE65***

Metabolic

Ornithine transcarbamylase deficiency: *OTC*

Fabry Disease (XL): *GLA*

Biotinidase deficiency (AR): ***BTD***

Pompe Disease (AR): ***GAA***

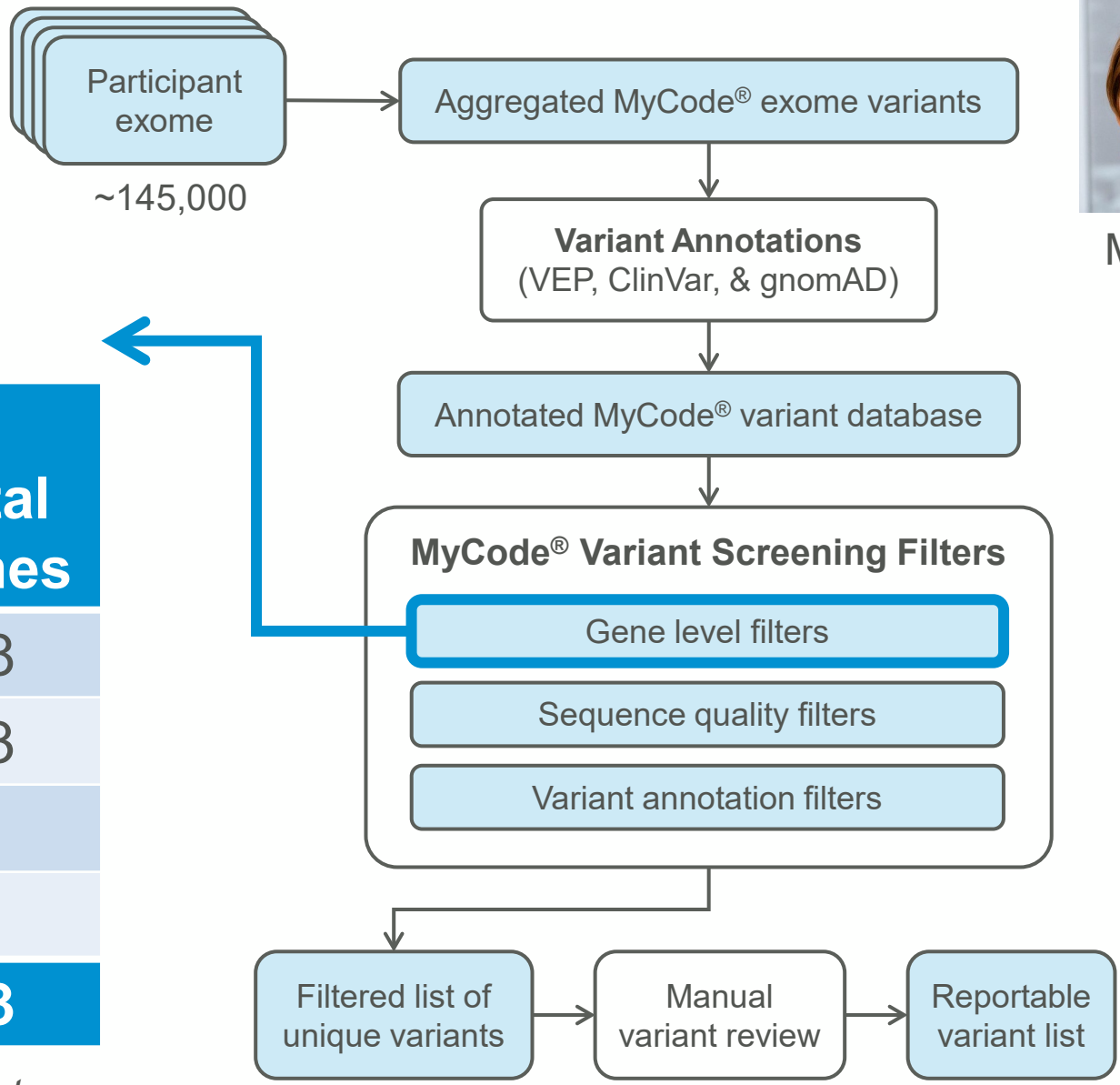
Screening Approach

Melissa Kelly

Genes: **ACMG SF Lists**

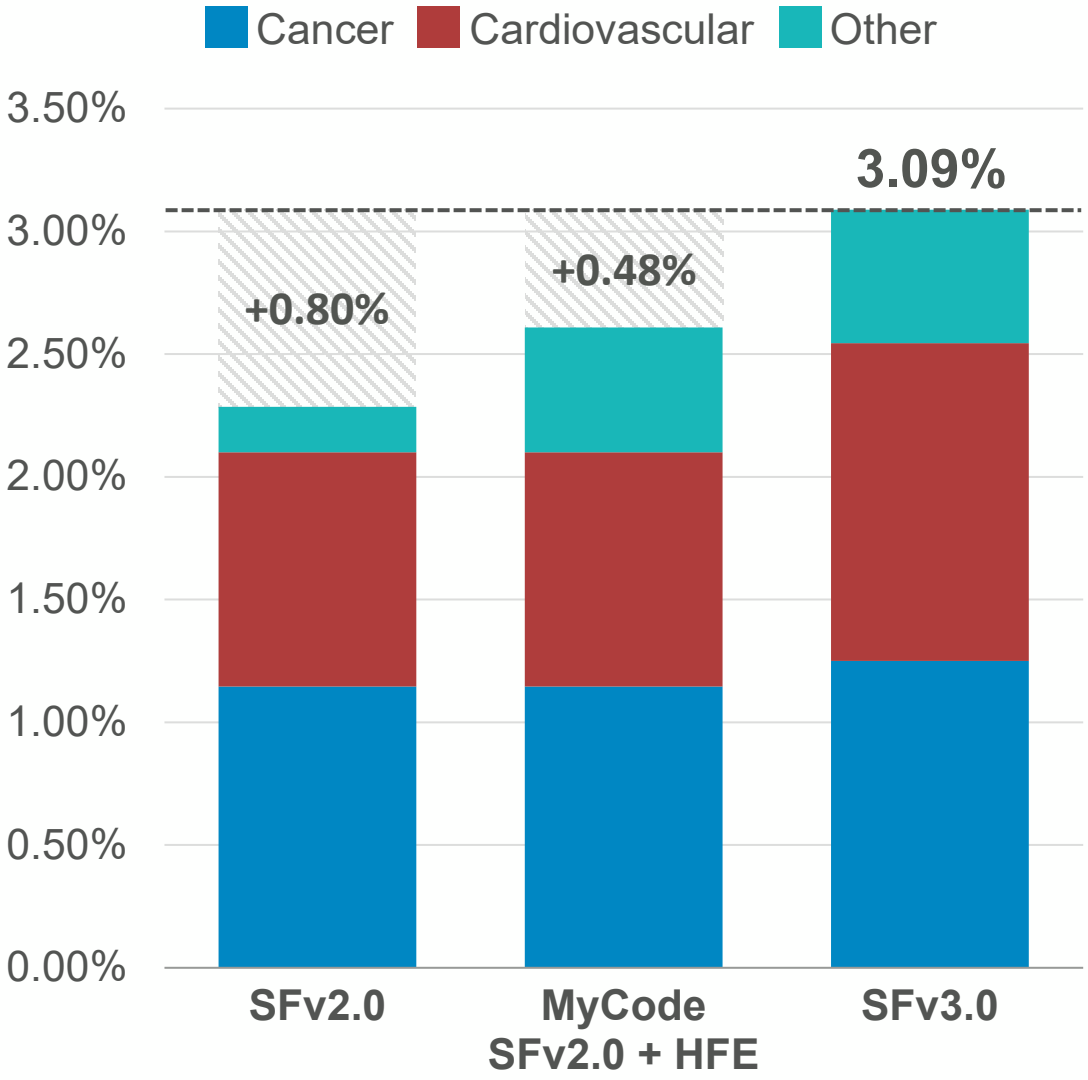
Clinical Domain	v2.0 Genes	v3.0 New Genes	Total Genes
Cancer:	25	+3	28
Cardio:	29	+4	33
Metabolic:	2	+2	4
Misc:	3	+5	8
Total:	**59**	**+14**	**73**

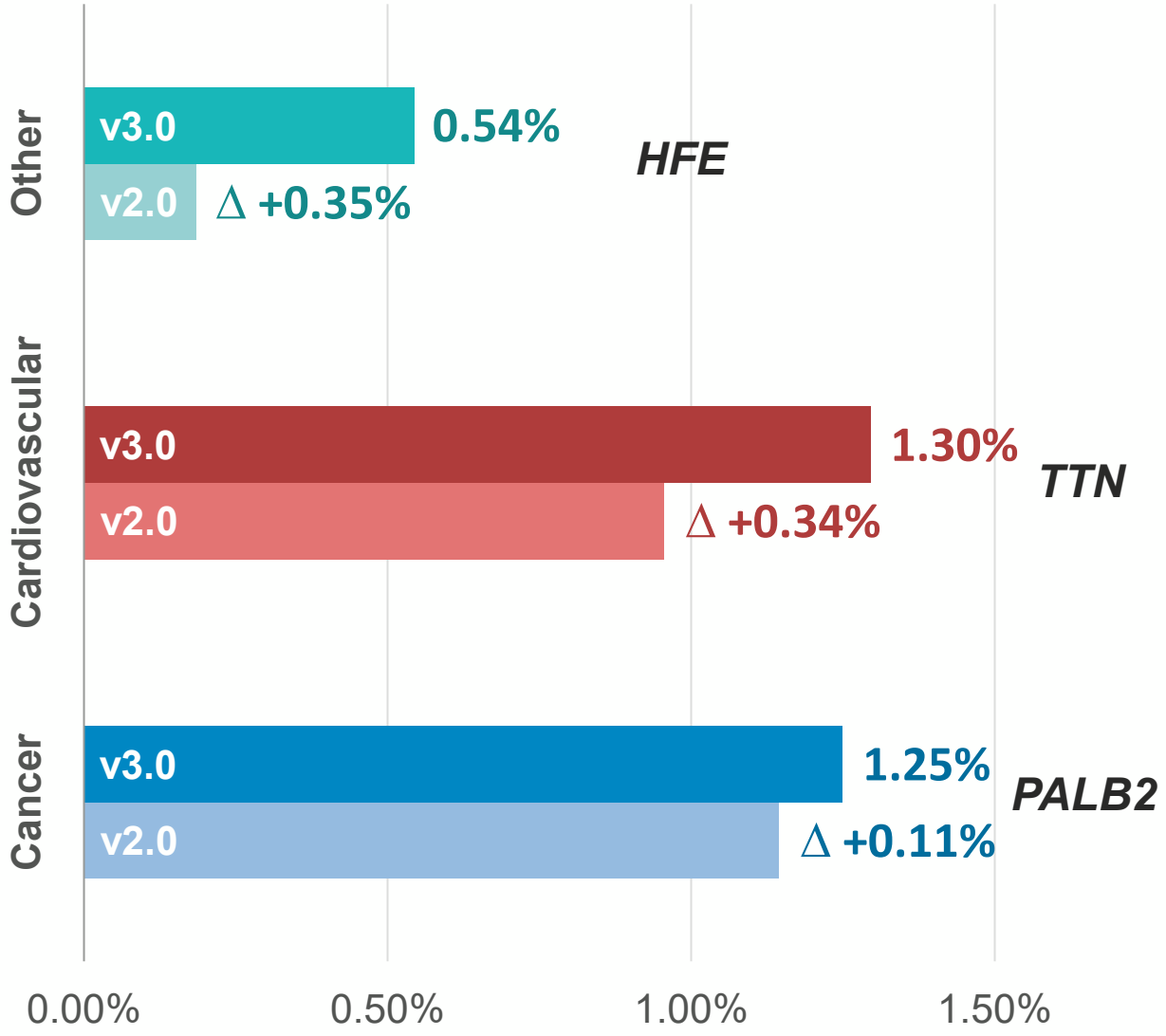
**** Disclaimer: ACMG SF recommendations were not intended for population screening ****



Adapted from Kelly et al. 2021 *Am J Med Genet C.* PMID:33576083

Increase in detection rate with v3.0





ACMG SF v3.1 Genes

Cancer

Hereditary breast and ovarian cancer: *BRCA1/2*, ***PALB2***

Lynch syndrome:

MLH1, *MSH2*, *MSH6*, *PMS2*

Familial Adenomatous Polyposis: *APC*

Endocrine tumor syndromes:

6 genes + ***MAX*** & ***TMEM127*** for pheochromocytoma & paragangliomas

+ 10 other cancer conditions

Cardiovascular

Vasculopathies: 7 genes

Cardiomyopathies

(HCM, DCM, ARVC):

16 genes + ***FLNC, TTN, BAG3, DES, RBM20,*** & ***TNNC1*** for DCM

Inherited arrhythmias:

4 genes + ***CASQ2*** & ***TRDN*** (AR) for CPVT

Familial hypercholesterolemia: *APOB*, *LDLR*, & *PCSK9*

Miscellaneous

Malignant hyperthermia: *RYR1* & *CACNA1S*

Wilson disease (AR): *ATP7B*

Hemochromatosis: *HFE* C282Y homozygotes

Hereditary Hemorrhagic telangiectasia: *SMAD4*, ***ACVRL1*** & ***ENG***

MODY: ***HNF1A***

Retinopathy (AR): ***RPE65***

Hereditary transthyretin amyloidosis: ***TTR***

Metabolic

Ornithine transcarbamylase deficiency: *OTC*

Fabry Disease (XL): *GLA*

Biotinidase deficiency (AR): ***BTD***

Pompe Disease (AR): ***GAA***

Policies Regarding Reanalysis

© American College of Medical Genetics and Genomics

ACMG STATEMENT | Genetics inMedicine

Points to consider in the reevaluation and reanalysis of genomic test results: a statement of the American College of Medical Genetics and Genomics (ACMG)

Joshua L. Deignan, PhD[1], Wendy K. Chung, MD, PhD[2], Hutton M. Kearney, PhD[3], Kristin G. Monaghan, PhD[4], Catherine W. Rehder, PhD[5] and Elizabeth C. Chao, MD[6]; on behalf of the ACMG Laboratory Quality Assurance Committee

Molecular Diagnosis & Therapy (2021) 25:529–536
https://doi.org/10.1007/s40291-021-00541-7

CURRENT OPINION

Clinical Exome Reanalysis: Current Practice and Beyond

Jianling Ji[1,2] · Marco L. Leung[3,4] · Samuel Baker[5] · Joshua L. Deignan[6] · Avni Santani[7,8]

Reclassification of clinically-detected sequence variants: Framework for genetic clinicians and clinical scientists by CanVIG-UK (Cancer Variant Interpretation Group UK)

ACMG Points to Consider

- Policies needed to address how reanalysis will be handled
  - *Variant-level reevaluation* -interrogation and potential reclassification of previously reported variants.
  - *Case-level reanalysis* - involves the review of all variants in an exome or genome, both reported and unreported.
- Respond to external requests for reanalysis
- Reports should clearly state the possibility of variant classification changes over time

Summary

- Evolving list of actionable genes/conditions (v2 to v3 = 35% increase)
- Frequency of changes in variant classifications varies (6.4% – 15%)
- Multiple reasons for reclassifications