AMP's Recommendations for Clinical Pharmacogenotyping Allele Selection

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AMP PGx Working Group

Recommendations for Clinical Pharmacogenetic Testing: Defining a Minimum Set of Variants that should be included in Genotyping assays

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- **Karen E. Weck** (Co-Chair), University of North Carolina
- **Larisa H. Cavallari,** University of Florida
- **Makenzie L. Fulmer,** ARUP Laboratories and University of Utah School of Medicine
- **Andrea Gaedigk,** Children's Mercy Kansas City, PharmVar representative
- **Houda Hachad**, AccessDx Laboratory
- **Yuan Ji**, ARUP Laboratories and University of Utah School of Medicine
- **Lisa V. Kalman**, Division of Laboratory Systems, Centers for Disease Control and Prevention
- **Reynold C. Ly,** Indiana University
- **Ann M. Moyer,** Mayo Clinic, CAP representative
- **Stuart A. Scott**, Stanford University Medical Center
- **Ron van Schaik,** Erasmus MC University Medical Center, ESPT and DPWG representative
- **Michelle Whirl-Carrillo,** Stanford University, CPIC and PharmGKB representative

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Goals

- To develop recommendations defining a minimum set of variants (a “Must-Test” list) that should be included in clinical genotyping assays
- Used by the clinical PGx testing community as a reference for test development

- **Members**
  - Subject matter expert representatives from the clinical PGx testing community (US and Europe), including organizational representation from CAP, CPIC, DPWG, ESPT, PharmGKB, and PharmVar
- **Projects – Allele Selection for Clinical Genotyping**
  - *CYP2C19* - **Pratt VM, et al. *JMD*, 2018;20:269-276**
  - *CYP2C9* - **Pratt VM, et al. *JMD*, 2019;21:746-755**
  - Warfarin-Related Genes - **Pratt VM, et al. *JMD*, 2020;22:847-859**
  - *CYP2D6* – **Pratt VM, et al. *JMD*, 2021;23:1047-1064**
  - *TPMT/NUDT15* - **Pratt VM, et al. *JMD*, 2022;24:1079-1088**
  - *CYP3A4/CYP3A5* **– Pratt VM, et al. *JMD*, 2023;25:619-629**
  - ***Recommendations for additional genes will be forthcoming***

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Considerations in PGx Test Interpretation

- Many genes are reported using a “star-allele” system to represent haplotypes rather than reporting each individual variant
  - Variants are typically named in the order of discovery
  - *1 indicates that no variants were identified (normal allele)
- For most genes, an individual will have 2 alleles, each with 1 copy of the gene
  - Reported as a diplotype (e.g., *1/*2)
  - Some individuals may have a gene deletion or duplication/multiplication involving one allele
- Diplotype is used to predict the phenotype (metabolizer status)
  - Ultrarapid, rapid, normal, intermediate, poor metabolizer
- **If no variants are detected – either because they are not present or they are not included in the assay design – the diplotype is often reported as *1/*1 and the phenotype as normal metabolizer**

PGx Genotyping Recommendations are Needed

GOAL: To promote standardization of PGx allele testing across clinical laboratories

- Inconsistent interpretation can lead to discordant therapeutic recommendations
- Publications show lack of consistency in alleles included in commercial platforms and clinical tests
- **Genomic Medicine X: Research Directions in Pharmacogenomics Implementation**
  - **NHGRI Meeting in 2017 - Call for assay standardization**

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PGx Genotyping Recommendations are Needed

- **How do we reduce the variability?**
  - Option 1: Test all known alleles
    - Not practical – for *CYP2D6*, there are currently >170 alleles (not counting sub-alleles)!!!
  - Option 2: Sequence instead of targeted genotyping
    - Likely coming in the future
    - Current state: Pharmacogenes are technically challenging by short-read NGS chemistry
      - Pseudogenes, homologous gene families, duplications, deletions, deeply intronic variants
      - Challenges in reporting genotypes
        - » What do you put on the clinical report when you otherwise have a *CYP2D6*2/*4,* but also see a rare variant that does not have a corresponding star allele?
      - Challenges in interpreting rare variants/alleles
  - **Option 3: Why not use a similar to ACMG recommendations for *CFTR* testing?**
    - Define a minimum set of variants based on multiethnic allele frequency in order to optimize diagnostic test rate

AMP PGx Working Group:
Expert Consensus Recommendation/Opinion Development

- **Tier 1 -** Minimum "must-test" alleles
  - Well-characterized effect on the function of the protein and/or gene expression
  - Appreciable minor allele frequency in a patient population
  - Available reference materials
  - Technical feasibility to detect variant in a clinical laboratory (NEW requirement for *CYP2D6*)
- **Tier 2 -** Extended panel
  - Meet at least one but not all the criteria for inclusion in Tier 1
- **Other**
  - Variants with unknown or uncertain function are not recommended for inclusion in clinical test panels

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AMP PGx Working Group - *CYP2C19*

- First deliverable: consensus expert opinion recommendations for clinical *CYP2C19* testing

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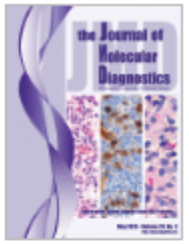
The Journal of Molecular Diagnostics

Volume 20, Issue 3, May 2018, Pages 269-276



Special article

Recommendations for Clinical *CYP2C19* Genotyping Allele Selection: A Report of the Association for Molecular Pathology

Victoria M. Pratt * †, Andria L. Del Tredici * ‡, Houda Hachad * §, Yuan Ji * ¶, Lisa V. Kalman * ‖, Stuart A. Scott * ** ††, Karen E. Weck * ‡‡ §§

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AMP PGx Working Group - *CYP2C9*

- Second deliverable: consensus expert opinion recommendations for clinical *CYP2C9* testing

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The Journal of Molecular Diagnostics

Volume 21, Issue 5, September 2019, Pages 746-755

Special article

Recommendations for Clinical *CYP2C9* Genotyping Allele Selection: A Joint Recommendation of the Association for Molecular Pathology and College of American Pathologists

Victoria M. Pratt * †, Larisa H. Cavallari * ‡, Andria L. Del Tredici * §, Houda Hachad * ¶, Yuan Ji * ‖, Ann M. Moyer * **, Stuart A. Scott * †† ‡‡, Michelle Whirl-Carrillo * §§, Karen E. Weck * ¶¶

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AMP PGx Working Group - Warfarin

- Third deliverable: consensus expert opinion recommendations for clinical warfarin testing

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The Journal of Molecular Diagnostics

Volume 22, Issue 7, July 2020, Pages 847-859

Special article

Recommendations for Clinical Warfarin Genotyping Allele Selection: A Report of the Association for Molecular Pathology and the College of American Pathologists

Victoria M. Pratt * † , Larisa H. Cavallari * ‡, Andria L. Del Tredici * §, Houda Hachad * ¶, Yuan Ji * ‖, Lisa V. Kalman **, Reynold C. Ly * ††, Ann M. Moyer * ‡‡, Stuart A. Scott * §§ ¶¶, Michelle Whirl-Carrillo * ‖‖, Karen E. Weck * ***

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AMP PGx Working Group - *CYP2D6*

- Fourth deliverable: consensus expert opinion recommendations for clinical *CYP2D6* testing

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The Journal of Molecular Diagnostics
Volume 23, Issue 9, September 2021, Pages 1047-1064

Special article

Recommendations for Clinical *CYP2D6* Genotyping Allele Selection: A Joint Consensus Recommendation of the Association for Molecular Pathology, College of American Pathologists, Dutch Pharmacogenetics Working Group of the Royal Dutch Pharmacists Association, and the European Society for Pharmacogenomics and Personalized Therapy

Victoria M. Pratt [*] [†], Larisa H. Cavallari [*] [‡], Andria L. Del Tredici [*] [§], Andrea Gaedigk [*] [¶], Houda Hachad [*] [‖], Yuan Ji [*] [**], Lisa V. Kalman [*] [††], Reynold C. Ly [*] [‡‡], Ann M. Moyer [*] [§§], Stuart A. Scott [*] [¶¶] [‖‖], R.H.N. van Schaik [*] [***] [†††], Michelle Whirl-Carrillo [*] [‡‡‡], Karen E. Weck [*] [§§§]

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AMP PGx Working Group - *TPMT/NUDT15*

- Fifth deliverable: consensus expert opinion recommendations for clinical *TPMT/NUDT15* testing

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The Journal of Molecular Diagnostics
Volume 24, Issue 10, October 2022, Pages 1051-1063

Special article

TPMT and *NUDT15* Genotyping Recommendations: A Joint Consensus Recommendation of the Association for Molecular Pathology, Clinical Pharmacogenetics Implementation Consortium, College of American Pathologists, Dutch Pharmacogenetics Working Group of the Royal Dutch Pharmacists Association, European Society for Pharmacogenomics and Personalized Therapy, and Pharmacogenomics Knowledgebase

Victoria M. Pratt [*] [†], Larisa H. Cavallari [*] [‡], Makenzie L. Fulmer [*] [§], Andrea Gaedigk [*] [¶], Houda Hachad [*] [‖], Yuan Ji [*] [§], Lisa V. Kalman [*] [**], Reynold C. Ly [*] [†], Ann M. Moyer [*] [††], Stuart A. Scott [*] [‡‡] [§§], R.H.N. van Schaik [*] [¶¶] [‖‖] [***], Michelle Whirl-Carrillo [*] [†††], Karen E. Weck [*] [‡‡‡] [§§§]

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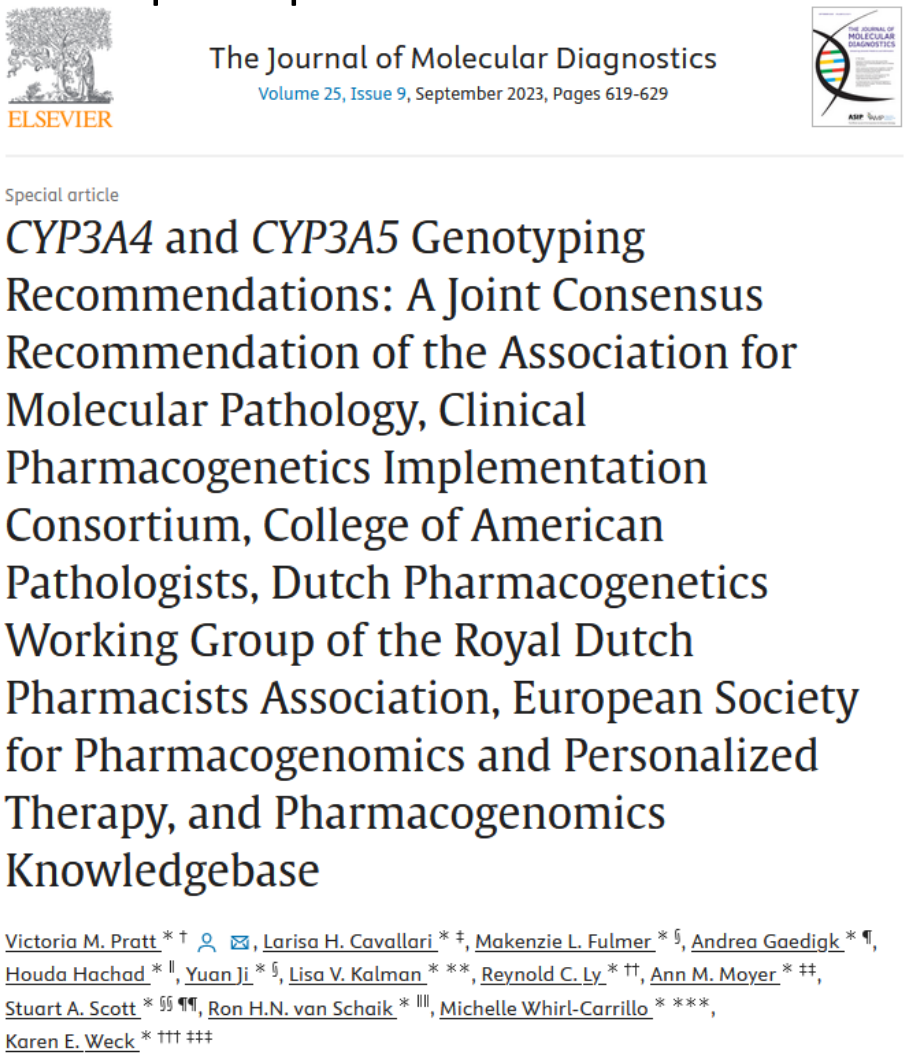
AMP PGx Working Group - *CYP3A4/CYP3A5*

- Sixth deliverable: consensus expert opinion recommendations for clinical *CYP3A4/CYP3A5* testing



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The Journal of Molecular Diagnostics
Volume 25, Issue 9, September 2023, Pages 619-629

Special article

CYP3A4 and *CYP3A5* Genotyping Recommendations: A Joint Consensus Recommendation of the Association for Molecular Pathology, Clinical Pharmacogenetics Implementation Consortium, College of American Pathologists, Dutch Pharmacogenetics Working Group of the Royal Dutch Pharmacists Association, European Society for Pharmacogenomics and Personalized Therapy, and Pharmacogenomics Knowledgebase

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AMP PGx Working Group Genotyping Recommendations

These recommendations are intended to:

- Promote standardization of PGx testing across different laboratories
- Inform clinical laboratory professionals when designing and validating clinical PGx assays
- Complement other clinical guidelines, such as those issued by CPIC and DPWG, *which primarily focus on the interpretation of genotyping results and therapeutic recommendations for specific drugs*

Future work:

- Other PGx genes with clinical relevance planned / in progress

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Please email Dr. Victoria (Vicky) Pratt (vicky.pratt@agenabio.com), Co-Chair of the AMP PGx Working Group, for feedback and suggestions!

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