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Pharmacogenomic Implementation Fails When Molecular Certainty Exceeds Clinical Certainty

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ABSTRACT

Pharmacogenomics is often implemented as though certainty accumulates monotonically from variant detection to prescribing action. In practice, these levels can separate. A genotype may be analytically secure while its functional consequence remains substrate-dependent, altered by copy-number structure or comedication, or incompletely transferable across populations. A gene–drug association may be pharmacologically compelling while the magnitude of clinical benefit remains dependent on comparator therapy, endpoint, treatment phase, and the availability of alternatives. Guideline organizations may additionally translate the same underlying evidence into different actions. This Current Opinion argues that pharmacogenomic implementation should therefore not be triggered by molecular certainty alone. Instead, implementation decisions should distinguish analytical certainty, functional certainty, gene–drug association certainty, clinical effect-size uncertainty, and therapeutic context. High-certainty examples such as selected DPYD–fluoropyrimidine and TPMT/NUDT15–thiopurine relationships demonstrate that clinically actionable pharmacogenomics is achievable, but they should not be treated as templates for every candidate gene–drug pair. The article develops a proposed molecular-to-clinical certainty architecture in which downstream uncertainty remains visible rather than being hidden behind a binary “actionable” label. This approach reframes implementation as a decision problem requiring convergence between reliable molecular information and a sufficiently clear clinical consequence.

Keywords: Pharmacogenomics, Genotype-to-phenotype translation, Phenoconversion, Clinical actionability, Gene–drug interactions, Precision pharmacotherapy

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INTRODUCTION

Pharmacogenomic implementation is frequently discussed as a problem of evidence accumulation: identify a reproducible variant, establish a biological effect, associate that effect with drug exposure or response, and translate the finding into prescribing guidance. That sequence is necessary but incomplete. Decisions about whom to test, when to test, and whether a result should alter treatment also depend on the severity and preventability of the outcome, the probability that the result will become relevant, the availability of therapeutic alternatives, turnaround time, and the consequences of acting on an uncertain recommendation [1]. Contemporary implementation barriers likewise extend beyond molecular validity to reimbursement, electronic clinical decision support, laboratory quality, equity, regulation, clinician education, and evidence interpretation [2]. The practical

problem is therefore not simply whether pharmacogenomic information is true, but whether the level of certainty achieved at one inferential layer is sufficient for the clinical decision at hand.

Population-level exposure data make this distinction important. Real-world analyses suggest that substantial numbers of patients encounter drugs linked to actionable pharmacogenomic recommendations, creating a large theoretical opportunity for pre-emptive or targeted testing [3]. Yet exposure to an actionable drug and carriage of an actionable genotype do not establish that testing will change management or improve an outcome for a particular patient. Conversely, the PREPARE implementation trial showed that a structured multigene strategy can reduce clinically relevant adverse drug reactions when pharmacogenomic information is integrated into prescribing across defined gene–drug pairs [4]. Pharmacogenomic implementation therefore cannot be dismissed as clinically unproven in general, but neither can successful panel-level evidence be transferred automatically to every gene, drug, population, or treatment setting.

The distinction becomes sharper when genotype is compared with direct or context-sensitive measures of drug metabolism. Pharmacogenomics, therapeutic drug monitoring, and emerging endogenous or circulating phenotype measures capture different determinants of pharmacokinetic variability [5]. Genotype is stable, whereas realized enzyme activity may not be. The core argument developed here is consequently that molecular certainty and clinical certainty are related but non-equivalent quantities. Implementation fails when a highly confident molecular result is allowed to suppress uncertainty that reappears at the functional, gene–drug, clinical-effect, therapeutic-context, or guideline-translation level.

Analytical certainty

Analytical certainty concerns what has actually been measured and how completely the assay represents the pharmacogene. It is tempting to treat a reported diplotype as an unambiguous molecular fact, but the reliability of that statement depends on locus complexity and assay architecture. Long-read pharmacogenomic sequencing can resolve structural variants, complex haplotypes, phasing relationships, and previously unresolved star-allele configurations that may be ambiguous or incompletely captured by conventional short-read approaches [6]. This improves molecular description, particularly at difficult loci, but it does not by itself establish that the additional analytical detail changes therapy. Analytical resolution is therefore an upstream condition for reliable interpretation rather than a surrogate for clinical utility.

CYP2D6 provides a particularly important example because copy-number variation and hybrid or complex allele structures can change the phenotype assigned to a patient. In a large psychiatric pharmacogenomic cohort, allele-specific copy-number information and medication-informed phenoconversion altered the interpretation of CYP2D6 activity for clinically relevant subsets of patients [7]. A report that omits relevant copy-number structure can consequently appear precise while representing an incomplete molecular state. The appropriate distinction is not “genotyped” versus “not genotyped,” but whether the assay has sufficient resolution for the variant architecture capable of changing the downstream clinical interpretation.

Mature pharmacogenomic tests can retain additional boundaries. Implementation of in-house DPYD genotyping before fluoropyrimidine therapy demonstrated that a clinically established assay can alter starting-dose decisions and can be integrated into routine oncology workflows [8]. Nevertheless, commonly used DPYD variant panels have historically emphasized variants characterized in populations of European ancestry. A systematic review of severe fluoropyrimidine toxicity in non-European populations identified additional variants and emphasized that the transferability of a fixed panel depends on population-specific allele representation and supporting functional evidence [9]. Analytical certainty can therefore be population-dependent even when the biological target—dihydropyrimidine dehydrogenase deficiency—is well established.

Standardization can reduce avoidable interpretation noise. CPIC and the Dutch Pharmacogenetics Working Group developed consensus recommendations for CYP2D6 genotype-to-phenotype translation to harmonize activity-score interpretation [10]. Such consensus is valuable because laboratories should not produce different phenotype labels merely because they apply incompatible translation rules. But harmonization solves a reproducibility problem, not every biological problem. A standardized genetically predicted phenotype remains a prediction of functional capacity under defined assumptions; it is not proof that the same functional state exists for every substrate, medication combination, or moment in treatment.

Functional certainty

Functional certainty asks whether the molecular state predicts the biological activity that matters for the drug being considered. For CYP2D6, this distinction is increasingly important because variant function is not always

well represented by a single substrate-independent activity value. Translational work examining variant haplotypes has shown that catalytic consequences can differ between substrates and that explicitly accounting for substrate-specific function can improve prediction of CYP2D6-mediated metabolism [11]. A phenotype category may therefore be useful while still concealing drug-specific variation. Functional interpretation should remain tied to the actual substrate whenever evidence indicates that variant effects are not interchangeable across drugs.

The functional state can also change without any change in genotype. In multimedicated geriatric patients, endogenous CYP2D6 phenotyping demonstrated genotype–phenotype mismatch consistent with phenoconversion, particularly in the presence of interacting medications and high medication burden [12]. This is clinically consequential because an individual genetically classified as having substantial CYP2D6 activity can behave functionally more like a lower-activity phenotype when an inhibitor is present. Comedication should therefore not be treated as an annotation appended after genotype interpretation; when strong enough to change enzyme activity, it becomes part of the phenotype itself.

The gap between biological plausibility and clinical actionability is also visible in antidepressant pharmacogenomics. CPIC provides prescribing recommendations for selected CYP2D6, CYP2C19, and CYP2B6 relationships but concluded that available evidence for SLC6A4 and HTR2A does not support using those genotypes to guide antidepressant prescribing [13]. This distinction is critical. A gene can participate in a biologically plausible pathway, be associated with a pharmacodynamic phenotype, and still fail to meet the evidentiary standard required for a treatment recommendation. Functional certainty must therefore refer to a drug-relevant, reproducible functional consequence rather than to pathway membership alone.

The Dutch Pharmacogenetics Working Group similarly demonstrates why function must remain drug-specific. Its recommendations for non-SSRI and non-tricyclic antidepressants vary according to the particular drug, CYP2D6 or CYP2C19 phenotype, and the expected consequences for exposure or treatment [14]. A broad label such as “CYP2D6 poor metabolizer” does not identify a universal clinical action. The same functional phenotype can justify dose adjustment, selection of an alternative, monitoring, or no change depending on the drug and the therapeutic circumstances.

Gene–drug association certainty

Some pharmacogenomic relationships are sufficiently mature that uncertainty at the association level is substantially reduced. Thiopurine treatment provides one such case. The updated CPIC guidance for TPMT and NUDT15 translates decreased or absent enzymatic function into specific starting-dose modifications because impaired thiopurine metabolism is strongly linked to severe toxicity risk [15]. This is a high-certainty pharmacogenomic relationship, but it still does not mean genotype completely predicts efficacy or toxicity. It means that one component of dose selection is sufficiently established to warrant action.

The contrast within beta-blocker pharmacogenomics is instructive. CPIC concluded that available evidence supported CYP2D6-guided recommendations for metoprolol but was insufficient to support prescribing actions for several evaluated pharmacodynamic candidate genes, including ADRB1, ADRB2, ADRA2C, GRK4, and GRK5 [16]. The important lesson is not that pharmacodynamic pharmacogenomics is intrinsically weak. It is that mechanistic plausibility and repeated association do not automatically cross the threshold required for prescribing. Evidence certainty can differ sharply within the same therapeutic class.

Drug specificity can persist even when the pharmacogene is the same. Updated CPIC recommendations for CYP2D6 and 5-HT₃ receptor antagonists preserve differences between individual agents rather than treating the antiemetic class as pharmacogenetically uniform [17]. Similarly, CYP2C19 loss-of-function variation has sufficient mechanistic and clinical support to guide clopidogrel therapy in defined cardiovascular settings [18]. The relevant inference is therefore gene–drug–indication specific rather than simply gene specific.

Other guidelines show the same pattern. Pharmacogenomic recommendations for statin-associated musculoskeletal symptoms depend on the particular statin and on genes influencing transport or metabolism, while acknowledging that musculoskeletal symptoms remain multifactorial [19]. For opioid therapy, CPIC supports CYP2D6-guided decisions for selected drugs such as codeine and tramadol but finds substantially weaker evidence for using OPRM1 or COMT variation as prescribing determinants [20]. DPYD-guided fluoropyrimidine dosing represents another comparatively mature relationship in which reduced predicted enzyme activity warrants a lower starting dose or avoidance in specified circumstances [21]. These cases support a hierarchy of gene–drug certainty, but they do not justify a universal threshold based solely on statistical association or mechanistic plausibility.

Clinical effect-size uncertainty

Even when the gene–drug relationship is credible, the size and durability of clinical benefit can remain uncertain. A recent randomized trial in depressive disorders reported higher response and remission with pharmacogenomic-guided treatment than with clinician-directed treatment without pharmacogenomic guidance [22]. Such findings establish that pharmacogenomic guidance can improve psychiatric outcomes under some study conditions. They do not establish a stable class-wide effect because commercially or institutionally implemented tools differ in genes, algorithms, medication coverage, and how clinicians use recommendations.

A 2024 systematic review and meta-analysis of randomized antidepressant pharmacogenomic trials illustrates this problem. The pooled evidence suggested modest improvements in efficacy, but the apparent benefits weakened when analyses were restricted to trials with lower risk of bias, and a clear tolerability advantage was not demonstrated [23]. Thus, molecularly plausible drug–gene relationships can coexist with a clinical-effect estimate that is small, heterogeneous, or method-sensitive. The clinically relevant question is not simply whether an effect exists, but how large, reproducible, and decision-changing that effect is in the intended treatment environment.

The PRIME Care trial provides an especially useful separation between process and outcome. Making commercial pharmacogenomic results available to clinicians substantially shifted prescribing away from medications with predicted drug–gene interactions, yet the difference in remission was small and was not sustained as a significant difference at week 24 [24]. Pharmacogenomic information therefore influenced clinician behavior more clearly than it changed the longer-term patient outcome. A reduction in predicted drug–gene conflicts should not be treated as interchangeable with evidence of durable clinical benefit.

Comparable uncertainty appears outside psychiatry. In the GIFT trial, genotype-guided warfarin initiation reduced a composite of clinically important outcomes in patients beginning therapy during hip or knee arthroplasty care [25]. By contrast, TAILOR-PCI showed a directionally favorable ischemic result for genotype-guided P2Y12 selection among CYP2C19 loss-of-function carriers but did not meet statistical significance for its prespecified primary comparison [26]. POPular Genetics demonstrated that genotype-guided de-escalation after primary percutaneous coronary intervention could preserve thrombotic safety while reducing bleeding compared with universal use of more potent P2Y12 inhibition [27]. These trials are not contradictory. They show that pharmacogenomic effect size is partly a property of the comparator strategy, endpoint, treatment phase, and clinical setting.

Treatment alternatives and therapeutic context

Clinical actionability emerges only after asking what will be done differently because of the result. The same genotype can have high value when it redirects treatment away from a high-risk exposure, modest value when multiple similarly effective options exist, and little immediate value when the relevant drug is unlikely to be used. The pharmacogenomic result therefore cannot be evaluated independently of therapeutic alternatives.

This dependence is visible in oncology implementation. A pragmatic panel-based pharmacogenomic study in supportive cancer care demonstrated that testing and clinical decision support were feasible, but relatively few medication changes were attributable to the pharmacogenomic result, and the study was not positioned to establish a clear quality-of-life advantage [28]. A technically actionable result is therefore distinct from an action-triggering clinical encounter. If the patient never receives the relevant drug, if the treatment course is short, or if an alternative is already preferred for non-genetic reasons, molecular information may remain clinically dormant.

DPYD illustrates the opposite configuration. Fluoropyrimidines can cause severe toxicity in patients with substantially reduced dihydropyrimidine dehydrogenase activity, clinically recognized genotype-based dosing guidance exists, and treatment can be modified before exposure. Yet even here implementation requires laboratory availability, turnaround time, clinical workflows, decision support, clinician and patient communication, and jurisdiction-specific handling of recommendations [29]. The strength of the molecular relationship does not make these system variables disappear.

A similar contextual principle applies to phenoconversion. A CYP2D6 genotype established years earlier may remain analytically correct while becoming an incomplete guide to current metabolism after a strong inhibitor is introduced. In such situations, treatment context does not merely “modify” the recommendation; it can change the effective phenotype relevant to the prescribing decision. Therapeutic context must therefore be represented as an active part of pharmacogenomic interpretation rather than as information consulted only after a genotype-derived recommendation has been produced.

The interaction among molecular evidence, functional state, therapeutic alternatives, and the immediate clinical decision is easier to recover visually than from sequential prose. **Figure 1** therefore reconciles these evidence

streams around the boundary at which a pharmacogenomic result either becomes a treatment-changing action or remains clinically indeterminate.

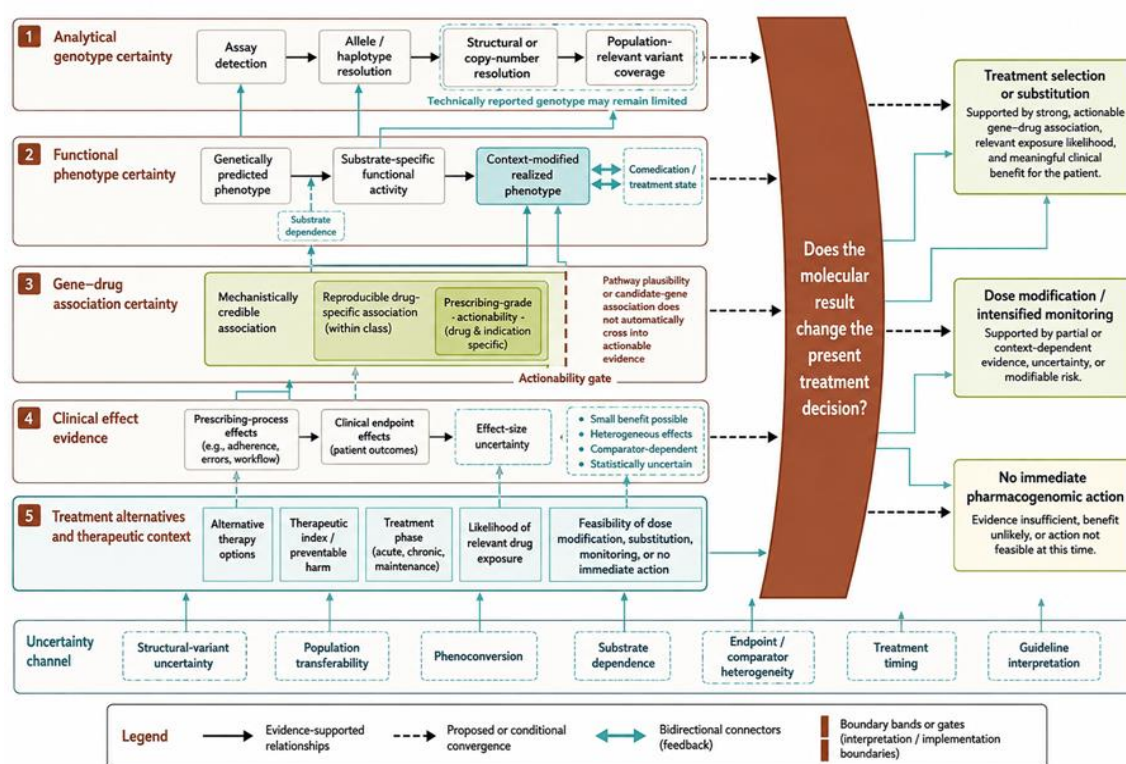


Figure 1. Treatment context determines whether molecular pharmacogenomic evidence becomes a clinical action

Guideline discordance

Guideline discordance represents another point at which molecular certainty can exceed clinical certainty. Pharmacogenomic consortia frequently evaluate overlapping evidence but differ in remit, evidence grading, phenotype translation, therapeutic alternatives, or the threshold at which a recommendation becomes actionable. A cross-sectional comparison of CPIC and DPWG guidance in Irish primary care found substantial agreement overall but identified 22 drug–gene interactions with recommendation differences and a smaller subset in which the recommended clinical action itself differed [30]. Discordance should therefore not be exaggerated as wholesale disagreement; its importance lies in the cases where two credible frameworks can translate the same broad evidence base into materially different prescribing decisions.

Differences can also arise between professional guidelines and regulatory labeling. The ClinGen Pharmacogenomics Working Group has highlighted the need to distinguish clinical validity from clinical actionability and has documented examples in which CPIC, DPWG, PharmGKB-associated resources, and regulatory labeling do not occupy the same position on whether or how a drug–gene association should change treatment [31]. Such disagreement does not necessarily indicate that one organization has interpreted the science incorrectly. Regulatory labels, prescribing guidelines, evidence-curation frameworks, and implementation resources perform different functions and may require different evidentiary thresholds.

A further interpretive layer is added by commercial reporting. Comparison of commercial antidepressant pharmacogenetic reports with CPIC guidance identified discrepancies in genotype-to-phenotype translation and in the recommendations attached to some reported results [32]. Molecular accuracy at the laboratory level therefore does not guarantee consistency at the phenotype or prescribing-recommendation level. The complete clinical object received by a prescriber is not merely a genotype; it is a genotype filtered through allele calling, phenotype translation, evidence classification, medication annotation, and a reporting algorithm.

Standardization remains essential. CPIC has expanded internationally and provides a widely used infrastructure for converting existing pharmacogenomic results into prescribing guidance [33]. Yet guideline adoption does not prove that every covered recommendation produces equivalent patient benefit, nor does it answer who should undergo testing before treatment. The clinically relevant task is to identify where discordance affects the decision

and why. **Table 1** therefore separates evidence certainty from the particular source of discordance and from the clinical decision that remains unresolved.

Table 1. Decision-changing forms of pharmacogenomic guideline discordance and the clinical uncertainty they leave behind

Observed configuration		Mechanism and competing diagnosis	Corrective discrimination
Comparison anchor / Evidence certainty	source of discordance / Unresolved implementation question	interpretation state	Clinical decision point / Monitoring or reassessment requirement
Comparison anchor: Established pharmacokinetic gene–drug relationship with convergent professional guidance Evidence certainty: Comparatively high for the defined gene–drug state	Principal source of discordance: Usually residual differences in wording, phenotype boundaries, or local workflow rather than the core action Unresolved implementation question: Does testing occur early enough for the agreed recommendation to change treatment?	Harmonized high-actionability state	Clinical decision point: Modify starting dose, select an alternative, or avoid a clearly high-risk exposure Monitoring or reassessment requirement: Confirm current drug, phenotype assignment, interacting medications, and clinical suitability of the recommended action
Comparison anchor: Two guidance systems recommend broadly similar management but use different evidence framing or phenotype terminology Evidence certainty: Moderate to high for action; variable for mechanism or effect magnitude	Principal source of discordance: Evidence grading, terminology, activity-score translation, or emphasis on pharmacokinetic versus clinical evidence Unresolved implementation question: Is apparent agreement masking different assumptions that matter in a boundary case?	Action-concordant but rationale-discordant state	Clinical decision point: Same broad prescribing direction despite different justification Monitoring or reassessment requirement: Document which framework generated the recommendation and reassess if phenotype or drug context changes
Comparison anchor: Credible guidance systems evaluate the same drug–gene relationship but recommend different clinical actions Evidence certainty: Intermediate or context-dependent	Principal source of discordance: Different actionability thresholds, comparator assumptions, endpoint weighting, or interpretation of evidence sufficiency Unresolved implementation question: Which recommendation best matches the present clinical decision rather than merely the strongest molecular association?	Action-level discordance state	Clinical decision point: Continue standard treatment versus adjust dose, substitute therapy, or increase monitoring Monitoring or reassessment requirement: Explicitly reconcile alternative guidance with therapeutic index, treatment phase, available alternatives, and patient-specific risk
Comparison anchor: Professional pharmacogenomic recommendation compared with product labeling or regulatory communication Evidence certainty: Molecular association may be well supported while formal actionability differs	Principal source of discordance: Different institutional remit, regulatory evidentiary threshold, labeling history, or update cycle Unresolved implementation question: How should clinicians act when professional guidance is more directive than regulatory labeling?	Guideline–regulatory mismatch state	Clinical decision point: Whether genomic information should alter prescribing beyond label-directed practice Monitoring or reassessment requirement: Preserve the source and status of each recommendation; do not collapse them into a single “approved” category
Comparison anchor: Commercial laboratory phenotype or medication recommendation compared with consensus guideline translation Evidence certainty: Underlying genotype may be certain while phenotype	Principal source of discordance: Proprietary translation rules, allele-content differences, report version, medication algorithm, or non-guideline genes Unresolved implementation question: Is disagreement caused by the molecular result, the	Report-layer discordance state	Clinical decision point: Whether the report should be accepted directly or reinterpreted Monitoring or reassessment requirement: Verify diplotype/phenotype translation and identify whether the

or recommendation is less certain	phenotype translation, or the treatment algorithm?		recommendation is guideline-based or proprietary
Comparison anchor: Static genotype-derived recommendation evaluated against current comedication or changing physiology Evidence certainty: High molecular certainty but reduced functional certainty	Principal source of discordance: Phenoconversion, inhibitor exposure, polypharmacy, changing organ function, or treatment phase Unresolved implementation question: Has the patient's functional state moved far enough from the genetically predicted phenotype to alter the decision?	Functionally unstable state	Clinical decision point: Whether a previously valid recommendation remains applicable at the current encounter Monitoring or reassessment requirement: Reassess interacting drugs and other determinants of realized phenotype before acting
Comparison anchor: Candidate gene or biologically plausible pathway lacks prescribing-grade support Evidence certainty: Low or indeterminate for treatment action	Principal source of discordance: Association evidence does not establish reproducible drug-specific clinical consequence Unresolved implementation question: What additional functional or clinical evidence is required before this association should influence treatment?	Insufficient-evidence state	Clinical decision point: Whether to act on an association despite absence of guideline-level evidence Monitoring or reassessment requirement: Avoid deterministic prescribing changes; monitor emerging evidence where clinically relevant

Note: The register is a proposed synthesis derived from the manuscript's evidence distinctions. It does not rank guideline organizations and does not assign numerical certainty scores. "Evidence certainty" refers to the level relevant to the stated clinical decision, not to analytical confidence in the genotype alone.

The molecular-to-clinical certainty gap

The central implementation problem emerges when certainty is examined across rather than within pharmacogenomic layers. Variant detection, genotype-to-phenotype translation, functional interpretation, gene–drug association, clinical effect, and prescribing utility answer different questions. Confidence at an upstream level does not necessarily propagate intact to the next. A correctly detected allele can have uncertain functional consequences for a particular substrate; an established functional effect can produce heterogeneous clinical outcomes; and an outcome association can become less useful when a safe, inexpensive, or already preferred treatment alternative changes the decision.

Timing introduces another separation. Pre-emptive pharmacogenomic testing can make results available before an actionable medication is prescribed. In pediatric oncology, early screening demonstrated that clinically relevant pharmacogenomic information can be generated in advance of later drug exposure, increasing the opportunity for results to be present when treatment decisions occur [34]. Information availability, however, is not identical to decision validity. A pre-emptively determined genotype may still require reinterpretation when the eventual drug, dose, comedication profile, organ function, or treatment objective becomes known. Pre-emption can solve the logistical problem of result availability without solving every downstream inferential problem.

This Current Opinion therefore proposes a molecular-to-clinical certainty gap rather than a single continuum of pharmacogenomic evidence. The gap is not a claim that certainty necessarily declines at every step. Selected gene–drug pairs maintain high confidence across several layers. Instead, it identifies points at which evidence must be re-evaluated before certainty is transferred downstream. Analytical certainty asks whether the relevant molecular state was measured correctly. Functional certainty asks what that state means for the drug and current biological context. Gene–drug association certainty asks whether the functional difference consistently alters pharmacokinetics, pharmacodynamics, toxicity, or efficacy. Clinical effect-size certainty asks how large and reproducible the treatment consequence is. Decision certainty finally asks whether acting on that information is preferable to the available alternatives.

These layers can become uncoupled. CYP2D6 is analytically sophisticated but functionally dynamic because substrate dependence, copy-number architecture, and phenoconversion complicate a static phenotype assignment. By contrast, DPYD and fluoropyrimidines illustrate a setting in which molecular, functional, toxicity, and prescribing evidence converge more strongly. Antidepressant pharmacogenomics occupies an intermediate position: selected metabolic gene–drug relationships are sufficiently established for prescribing recommendations, while randomized evidence for broad multigene testing shows smaller and more method-sensitive clinical effects. The implication is not that one field is "certain" and another "uncertain." Certainty must be attached to the specific inference being made.

A second implication concerns accumulation of contextual variables. Population-specific variant representation can constrain analytical transferability; comedications can alter functional phenotype; the index drug determines whether a variant matters; the clinical comparator influences observed treatment benefit; and the health system determines whether the result can be delivered in time to affect care. Increasing molecular resolution addresses only some of these uncertainties. Once an upstream limitation is resolved, new downstream questions become visible rather than disappearing.

A threshold framework for implementation

A clinically useful implementation threshold should therefore require convergence rather than molecular confidence alone. This article proposes that implementation be considered across four decision-critical domains: analytical reliability, functional interpretability, gene–drug actionability, and clinical decision consequence. Existing evidence demonstrates that these domains can diverge: formal guidelines may support some metabolic gene–drug relationships while withholding recommendations for biologically plausible candidate genes, and clinical trials can show substantially weaker patient-level effects than the molecular rationale might imply [13, 16, 23]. The proposed framework is conceptual and intentionally nonquantitative. It does not assign scores, certainty grades, or numerical cutoffs that have not been prospectively validated.

The first threshold concerns analytical sufficiency. The assay must reliably detect the molecular states capable of changing interpretation in the intended population. This requirement is stricter than technical validity in an abstract laboratory sense. If clinically relevant structural variation, copy-number states, rare functional alleles, or population-specific variants are systematically missed, a highly reproducible result can still be incomplete. Analytical sufficiency is therefore gene-, platform-, and population-dependent.

The second threshold concerns functional interpretability. A genotype-derived phenotype should be sufficiently stable for the drug and clinical state being considered. Where substrate-specific activity, strong inhibition, induction, organ dysfunction, or other sources of phenoconversion materially alter function, the implementation pathway should acknowledge that instability rather than present the genetically predicted phenotype as immutable. Functional uncertainty does not necessarily prevent action; it determines whether genotype alone is enough.

The third threshold concerns gene–drug actionability. Evidence must connect the relevant molecular or functional state to a drug-specific consequence that can plausibly alter treatment. This requirement prevents pathway plausibility or candidate-gene association from being mistaken for clinical readiness. It also allows highly developed relationships to move forward without requiring identical evidentiary histories across all pharmacogenes.

The fourth threshold concerns the clinical consequence of acting. The magnitude of expected benefit, severity of preventable harm, feasibility of an alternative, therapeutic index, treatment duration, likelihood of drug exposure, and cost of a wrong decision all modify implementation value. In this sense, the threshold for implementing a test to avoid severe, preventable toxicity may reasonably differ from the threshold for using a test intended to produce a modest average improvement in symptom control. DPYD demonstrates how clinical toxicity evidence, prescribing recommendations, and implementation requirements can converge around a high-stakes decision [8, 21, 29]. Even in such a setting, however, genotype-guided starting-dose recommendations remain one component of treatment rather than a substitute for clinical monitoring.

The framework also proposes a weakest decision-critical layer rule. When one indispensable layer is substantially uncertain, confidence at other layers should not simply compensate for it. Strong molecular evidence cannot repair absence of a drug-specific functional relationship. A compelling pharmacokinetic effect cannot establish a patient-important benefit when clinical-effect evidence is insufficient. Conversely, modest uncertainty in a noncritical dimension need not block implementation when the relevant treatment decision is strongly supported and the consequence of inaction is serious. This rule is deliberately qualitative because the importance of each layer changes with the gene–drug pair and decision.

Pre-emptive testing adds a timing threshold. Testing early is most defensible when actionable exposure is sufficiently likely, the result remains interpretable over time, and possessing the information before prescribing offers an advantage over reactive or pretreatment testing. High population exposure to actionable medications can support pre-emptive strategies, but exposure prevalence alone should not be treated as proof that every tested individual will benefit. The decision is a balance between informational readiness and the probability that the information will become clinically relevant.

Communication of residual uncertainty

Implementation also depends on how uncertainty is communicated. A report that compresses a complex evidence chain into a categorical label such as “use with caution” or “increased risk” can make molecular certainty appear broader than it is. The report should distinguish, where clinically important, confidence in the genotype, confidence in the inferred phenotype, confidence in the gene–drug association, and confidence that a particular treatment change improves the outcome relevant to the patient.

This separation is particularly important for phenoconversion. A genotype may remain correct for life while its functional interpretation changes with medication exposure. Reports and clinical decision-support systems should therefore avoid implying that every phenotype label is permanently equivalent to realized enzyme activity. When strong inhibitors or other major contextual modifiers are present, a concise statement that the genetically predicted phenotype may not represent the current functional phenotype can be more informative than adding further molecular detail.

Guideline disagreement should likewise remain visible when it changes treatment. CPIC, DPWG, regulatory resources, and commercial interpretation systems may differ because they use different evidence thresholds, remits, translation rules, or therapeutic assumptions. Guideline, regulatory, and commercial-report discordance should therefore be identified as distinct sources of uncertainty rather than collapsed into a single recommendation [30-32]. The aim is not to transfer the burden of resolving every disagreement to the prescriber. It is to make clear whether disagreement concerns the genotype, phenotype assignment, evidence grading, or action recommended. Clinical-effect uncertainty also deserves explicit communication. A strong gene–drug association should not be presented as though it predicts the magnitude of benefit for an individual patient. PRIME Care illustrates why this distinction matters: pharmacogenomic information can substantially alter medication selection while producing a smaller and less persistent difference in the downstream clinical endpoint. Conversely, the absence of a large average benefit in one trial does not invalidate a well-supported gene–drug relationship used to avoid a specific high-risk exposure. Reports should state the inference they support rather than borrowing certainty from another level.

Communication should finally be decision-centered. The clinically useful question is rarely “Is this pharmacogenomic result valid?” in isolation. It is “How certain are we that this result should change this treatment for this patient now?” That formulation forces residual uncertainty into the same frame as alternatives, treatment phase, comedication, expected benefit, preventable harm, and reversibility of the decision. Such transparency may make pharmacogenomic recommendations appear less absolute, but it makes their clinical meaning more defensible.

CONCLUSION

Pharmacogenomic implementation becomes unreliable when confidence in a molecular result is allowed to stand in for confidence in a clinical decision. Analytical validity, functional interpretation, gene–drug actionability, clinical effect size, therapeutic alternatives, timing, and guideline translation are related but separable components of implementation certainty.

The practical consequence is not to demand perfect certainty before using pharmacogenomics. Some gene–drug relationships already support consequential treatment decisions despite residual uncertainty, while others remain unsuitable for routine action even when their molecular biology is persuasive. The relevant threshold depends on what the test can measure, how reliably genotype predicts the current functional state, how strong and drug-specific the association is, what benefit or harm is expected from acting, and what alternatives are available.

The proposed molecular-to-clinical certainty architecture therefore treats implementation as convergence across decision-critical layers rather than as the automatic downstream consequence of molecular precision. Its purpose is not to create another numerical evidence score, but to expose where certainty is being transferred without sufficient justification. Pharmacogenomics is most clinically credible when residual uncertainty is preserved, contextual modifiers remain visible, and the recommendation states precisely which inference the evidence supports. Molecular certainty is valuable; implementation becomes defensible only when clinical certainty is sufficient for the decision it is being asked to change.

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