**Box S1.** Details of the author’s 20-year journey on CYP2D6 genotyping in psychiatry

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| **1990s (“The Fear Stage”)**  Pharmaceutical companies were not interested in pharmacogenetics and new genetic technologies. Risperidone’s marketer described risperidone as metabolized by CYP2D6. Using a single-dose study in 12 healthy males, of which 2 were CYP2D6 PMs [S1], they proposed that CYP2D6 PM status was irrelevant for risperidone treatment [S2].  Meanwhile, due to the development of new technologies, the genetic revolution had begun [S3]. Affymetrix [S4] had developed the GeneChip by introducing oligonucleotide sequences in a microarray; the use of a laser fluorescence scanning system allowed for testing thousands of gene variants at the same time.  Pharmacologists were still developing CYP2D6 science [S5] and most psychiatrists had not paid any attention to the CYP2D6 gene polymorphic variations that led to two major phenotypes: PMs with no CYP2D6 and UMs with too much CYP2D6. In 1996, the author, an “unusual” psychiatrist, decided to move to Kentucky to work at a state psychiatric hospital in collaboration with a pharmacologist interested in using CYP2D6 to personalize psychiatric prescriptions [1]. A pilot study [2], including 100 psychiatric patients who were genotyped using the rudimentary genetic technology of the time, required each sample to be tested 8 times, one per each CYP2D6 allele studied. With the help of Affymetrix, the 100 samples [3] were retested using their GeneChip for CYP2D6 testing; it identified 12 PMs who did not have any active CYP2D6 in their bodies and 3 UMs who had at least 3 active copies of the CYP2D6 gene. At that time many of the psychiatric patients were taking first-generation psychiatric drugs including the TCAs and first-generation antipsychotics that are heavily dependent on CYP2D6 for their metabolism and are narrow therapeutic drugs with propensity to cause toxicity. Thus, this pilot cost-effectiveness study indicated that CYP2D6 PMs and UMs tend to cost more due to greater duration of admission [3]. The author was unable to get funding for extending the pilot study to a large sample in Kentucky psychiatric state hospitals.  In a case report he described how carbamazepine, a potent CYP3A4 inducer, influenced risperidone TDM by increasing its metabolism and proposed that risperidone was also metabolized by CYP3A4 [S6]. Later, other clinical DDI studies [S7, S8] and, more importantly, in vitro studies [S9, S10] verified that CYP3A4 was important in risperidone metabolism, although it has lower affinity than CYP2D6. In spite of the lack of interest in DDIs by risperidone’s marketer, a case-series study indicated that CYP2D6 PM status, taking CYP2D6 and/or CYP3A4 inhibitors, and taking CYP3A4 inducers were relevant for risperidone metabolism [S7]. |
| **2000s (“The Failure Stage”)**  The diagnostic section of Roche (Roche Molecular Diagnostics) decided to market the Affymterix GeneChip technology to test for CYP2D6 and CYP2C19. The product was marketed in early 2006 [4, 5, S11] and tested for 27 CYP2D6 alleles:  -12 alleles with no activity (\*3, \*4, \*5, \*6, \*7, \*8, \*11, \*15, \*19, \*20, \*40 and \*4xn)1  -2 with very low activity (\*10 and \*36),  -5 with low activity (\*9, \*17, \*29, \*41 and \*10xn),  -3 with activity lower than normal (\*29xn, \*41xn and \*17xn),  -2 with normal activity (\*2 and \*35), and  -3 with increased activity (\*1xn, \*2xn, and \*35xn).  Allele\*1 was considered the “default” allele; it is used when none of the tested alleles is identified.  The FDA had been pushing for pharmacogenetic testing [1] but had no authority to regulate pharmacogenetic testing and any laboratory demonstrating reasonable analytical validity could provide CYP2D6 genetic testing. Roche decided to market the AmpliChip CYP450 test with FDA approval to demonstrate that their test was superior to any other CYP2D6 genotyping test.  In 2006, first-generation psychiatric drugs were rarely used and second-generation drugs were metabolized by different CYPs and had wider therapeutic windows. Therefore, when Roche finally funded a large study, the study demonstrated that you can genotype >4,000 patients for CYP2D61 and CYP2C19 using the new technology but the testing was no longer clinically relevant for a great majority of the psychiatric patients [5]. The AmpliChip CYP450 test was relatively expensive and psychiatrists were uninterested and uneducated in CYP genotyping. The AmpliChip CYP450 test and other tests from the first wave of pharmacogenetic testing in psychiatry failed commercially since they were rarely used and then became unavailable [6, S12].  A case-control risperidone study indicated that after controlling for confounders, CYP2D6 PMs had more risk of risperidone ADRs and discontinuation due to ADRs [S13]. Risperidone TDM indicated that CYP2D6 PMs have compromised ability to eliminate risperidone [S14]. The lack of interest of risperidone’s marketer in DDIs led to a partial failure of the RCT that studied its use as adjunctive therapy in mania for patients taking carbamazepine [S15]. Once more, the author failed to obtain funding for a prospective RCT using CYP2D6 genotyping to dose risperidone.  Case reports indicated that CYP2D6 UMs may have ADRs when taking drugs activated by CYP2D6 such as codeine-like opioids [S16] or diphenhydramine [S17]. An initial attempt to publish guidelines for CYP genotyping was made. The guidelines were based on clinical experience and pharmacokinetic mechanisms [S18]. |
| **2010s (“The Hype Stage”)**  As someone who has pushed for pharmacogenetic testing in psychiatry for quite some time, the author has become appalled by the current status of the field, which has become dominated by marketing instead of scientific thinking [1]. Many psychiatrists became confused about the role of pharmacogenetic testing; they had been contacted by company representatives asking them to use pharmacogenetic testing for their patients, as this was the latest advance in psychiatric treatment. Psychiatrists doubted that this was a real advance but they did not have enough experience and knowledge to deny the utility of these tests. So they asked the author’s opinion about them or asked him to lecture on the clinical utility of pharmacogenetic testing in psychiatry. He developed a PowerPoint lecture to summarize the status of pharmacogenetic testing in psychiatry and lectured in several countries against using the non-validated pharmacogenetic tests [1].  The author has published reviews on the limitations of pharmacogenetic tests in general [1, S19] and the limited benefits of CYP2D6 genotyping [9] for atomoxetine [10, S20] and long-acting risperidone [S21]. Moreover, he made an attempt to incorporate CYP2D6 genotyping and knowledge of DDIs to guide risperidone dosing by using TDM [9]. |

ADR: adverse drug reaction; DDIs: drug-drug interactions; FDA: Food and Drug Administration; PM: poor metabolizer; RCT: randomized clinical trial; TCA: tricyclic antidepressant; TDM: therapeutic drug monitoring; UM: ultrarapid metabolizer.

1The version used in the study genotyping 4,562 patients also included the \*6xn allele [5].

**Box S2.** The author’s view of CYP2D6 pharmacology

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| **Influences on CYP2D6 activity** [S22]  An ideal gene for pharmacogenetics research since environmental influences are very limited:  1. CYP2D6 inhibitors can:  1.1. mimic CYP2D6 PMs (phenoconversion) if they completely inhibit CYP2D6 activity or  1.2. at least decrease the activity in the remaining patients.  Several psychiatric drugs are clinically relevant inhibitors:  -Potent: fluoxetine and paroxetine  -Moderate: bupropion, duloxetine and TCAs  -Mild: asenapine, fluvoxamine, and sertraline1  Any CYP2D6 substrate can behave as a competitive inhibitor, but this is rarely clinically  relevant. Competitive inhibition can be relevant in situations of polypharmacy and when  metabolism for several CYPs is compromised.  2. There are no CYP2D6 inducers. CYP2D6 activity appears ↑ in pregnancy (repressor is removed). |
| **CYP2D6 polymorphism** [12, s23]  1. PMs: have no CYP2D6 PM activity due to the presence of 2 non-functional alleles  2. IMs: have decreased activity due to different allele combinations  2.1. With very low activity, they are close to PMs and can easily become phenotypical PMs  through inhibition. They are particularly frequent in East Asians.  2.2. With activity close to normal, they are called normal slow (N slow) below.  3. NMs: have normal activity due to having at least 1 fully functional allele  4. UMs: have increased activity from at least 3 functional copies/alleles |
| **percentages of CYP2D6 phenotypes worldwide** [12]  African-Am Africans Americans East Asians Europeans Middle-East SC Jewish  or Oceanian Asians .  PMs 2 3 2 <1 5 1 1 6  IMs low 2 11 3 5 5 5 4 11  N slow 14 14 2 23 1 9 13 5  NMs 78 68 88 71 86 74 79 67  UMs 4 4 5 1 3 11 3 11 |
| **Psychiatric drugs dependent on CYP2D6 for their metabolism**  **Antidepressants**  -First-generation: TCAs: CPIC guideline [7]  (amitriptyline, imipramine, clomipramine; also CYP2C19)  -Second-generation: fluoxetine, fluvoxamine, paroxetine: CPIC guideline [8]  venlafaxine, vortioxetine [9]  **Antipsychotics**  -First-generation: haloperidol, zuclopenthixol (and possibly phenothiazines) [9]  -Second-generation: aripiprazole, brexpiprazole, iloperidone and risperidone (also CYP3A42) [S24]  **Other**  -Atomoxetine [10, S25] |
| **DRUGS ACTIVATED BY cyp2d6**  **codeine-like opioids**  -Codeine and tramadol: CPIC guideline [S26]  -Hydrocodone and oxycodone: more research is needed [S27]  **Other**  **-**Diphenhydramine: more research is needed, CYP2D6 UMs may show paradoxical excitation [S17] |

ADR: adverse drug reaction; CPIC: Clinical Pharmacogenetics Implementation Consortium; CYP: cytochrome P450; IM: intermediate metabolizer; NM: normal metabolizer; N: normal; PM: poor metabolizer; SC: south central; TCA: tricyclic antidepressant; TDM: therapeutic drug monitoring; UM: ultrarapid metabolizer.

1Sertaline may be a dose-related inhibitor; evidence of CYP2D6 inhibition may require high doses.

2CYP3A4 inducers (carbamazepine, phenobarbital and phenytoin) increase the metabolism of aripiprazole, brexpiprazole, iloperidone and risperidone by inducing CYP3A4 and increasing the percentage of these antipsychotics metabolized by CYP3A4.

**Box S3.** The author’s current view of pharmacogenetic test science and marketing

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| **SCIENCE FOR MARKETING GENETIC TESTS**  In 2006, a committee of US scientists proposed three aspects of genetic test evaluation [S12]:  -analytic validity: whether the test provides an accurate, reliable measurement of the genotype  -clinical validity: the ability of the test to detect or predict the associated disorder  -clinical utility: the risks and benefits of the test’s use in the clinical environment |
| **THE FDA’S FAILED ATTEMPTS TO REGULATE PHARMACOGENETIC TESTS**  -In the 1990s, the FDA started pushing for increased pharmacogenetic testing despite their role as the regulatory agency only for drug marketing, not for diagnostic test marketing [1]. In a “me, too” drug market, the reluctant pharmaceutical companies eliminated CYP2D6 drugs from their pipelines [1].  -In 2005, the FDA provided a voluntary guideline for pharmacogenetic testing for pharmaceutical companies in which CYP2D6 was described as a “valid biomarker” [1, S12].  -In 2008, the FDA issued a draft guideline for “In Vitro Diagnostic Multivariate Index Assays” indicating an intent to require these assays to meet premarket and postmarket device requirements under FDA regulations [S12]. This guidance was never implemented.  -In 2014, the FDA issued a draft trying to regulate diagnostic testing [S19]. This was received very  poorly by US laboratory experts [S28].  -In 2017, the FDA, in a discussion paper on “Laboratory Developed Tests”, acknowledged the public health need for greater oversight of these tests but stated the FDA will not try to regulate them [S29].  -In the uncertain political future that the US government and US Congress is facing in 2017, the author does not see how US regulation addressing the current legal vacuum which is allowing the marketing of non-validated pharmacogenetic tests in the US can be improved in the next few years. |
| **CURRENT US REGULATIONS FOR PHARMACOGENETIC TESTS** [S12, S19]:  CLIA regulations, which are administered by CMS, regulate quality standards for US clinical laboratories. Accreditation by CAP, JCAHO or state Health Departments assures full compliance with CLIA regulations and allows any laboratory to legally offer pharmacogenetic testing.  CLIA regulations focus on basic aspects of analytical validity at the laboratory level but were not developed for multivariate assays. They do not address clinical validity or clinical utility. |
| **FDA RECOMMENDATIONS FOR PHARMACOGENETIC TESTS** [15, S19]  The large prospective RCTs needed to establish the classic proof of concept and the cost-benefit ratio of pharmacogenomics in psychiatry will not occur due to very high costs and the lack of a funding mechanism. However, studies with comparisons to historical data have been used by the FDA to recommend pharmacogenomic tests in specific contexts [15]. |
| **OTHER COUNTRIES** [13, S30]  It is not clear how other countries approve the marketing of genetic testing. In a review [13] of 22 pharmacogenetic tests for CYP2D6 genotyping: 7 were not commercialized in US. |
| **TECHNOLOGICAL ADVANCES**  High-throughput sequencing technologies are beginning to be used in pharmacogenetics testing, in general, and in CYP2D6 genotyping, in particular. For CYP2D6 genotyping high-throughput sequencing technologies provide a promising time-efficient and cost-effective alternative to currently used genotyping techniques but they face several obstacles such as: 1) high sequence similarity and genetic recombinations between CYP2D6 and evolutionarily related pseudogenes CYP2D7 and CYP2D8, and 2) high copy number variation among individuals and 3) short read lengths generated by these technologies. Numanagić et al. proposed an algorithm to computationally infer CYP2D6 genotype from results obtained by high-throughput sequencing technologies [S31]. |
| **ADVANCES IN PHARMACOKINETIC KNOWLEDGE FOR FUTURE PSYCHIATRISTS**  The recent publication of the third edition of German guidelines for TDM in psychiatry [S32] has given the author hope that today's psychiatry residents may be more ready to use TDM to learn CYP science, to use CYP genotyping and become familiar with drug-drug interactions [S33]. |

CAP: College of American Pathologists; CLIA: Clinical Laboratory Improvement Amendments; CMS: Centers for Medicare & Medicaid; FDA: Food & Drug Administration; JCAHO: Joint Commission on Accreditation of Healthcare Organizations; RCT: randomized clinical trial; TDM: therapeutic drug monitoring; US: United States.

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