HAL
open science

OpenCYP: An open source database exploring human variability in activities and frequencies of polymophisms for major cytochrome P-450 isoforms across world populations

Susanna Vichi, Franca Maria Buratti, Emma Di Consiglio, Laura Turco, Leonie S. Lautz, Keyvin Darney, Jean-Lou Christian Michel Dorne, Emanuela Testai

To cite this version:

Susanna Vichi, Franca Maria Buratti, Emma Di Consiglio, Laura Turco, Leonie S. Lautz, et al.. OpenCYP: An open source database exploring human variability in activities and frequencies of polymophisms for major cytochrome P-450 isoforms across world populations. Toxicology Letters, 2021, 10.1016/j.toxlet.2021.07.019 . anses-03319826

HAL Id: anses-03319826

https://anses.hal.science/anses-03319826v1

Submitted on 26 Nov 2021

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Toxicology Letters 350 (2021) 267–282

Contents lists available at ScienceDirect

Toxicology Letters

journal homepage: www.elsevier.com/locate/toxlet

OpenCYP: An open source database exploring human variability in activities and frequencies of polymophisms for major cytochrome P-450 isoforms across world populations

Susanna Vichi[a,*], Franca Maria Buratti[a], Emma Di Consiglio[a], Laura Turco[a], Leonie S. Lautz[b], Keyvin Darney[b], Jean-Lou Christian Michel Dorne[c], Emanuela Testai[a]

[a] Istituto Superiore di Sanità, Environment & Health Department, Viale Regina Elena 299, Roma, Italy
[b] Risk Assessment Department, French Agency for Food, Environmental and Occupational Health & Safety (ANSES), 14 rue Pierre et Marie Curie, Maisons-Alfort, F-94701, France
[c] European Food Safety Authority, Via Carlo Magno 1A, 43126, Parma, Italy

HIGHLIGHTS

- Extensive literature research on CYP1A2, CYP2A6, CYP2D6, CYP3A4/3A5, CYP3A7 activity.
- Open source OpenCYP database provides data for healthy adult variability in CYP activities.
- Quantitative comparisons are hampered by different experimental conditions used.
- Data gaps identified on specific age groups (e.g. elderly, children and neonates).
- OpenCYP database as a promising tool for PBk modelks and chemical risk assessment.

ARTICLE INFO

Article history:
Received 31 March 2021
Received in revised form 22 July 2021
Accepted 27 July 2021
Editor: Dr. Angela Mally
Available online 2 August 2021

Keywords:
Cytochrome P450
Human variability
Enzymatic activity
Genetic polymorphisms
PBK models
Human risk assessment

ABSTRACT

The open source database "OpenCYP database" has been developed based on the results of extensive literature searches from the peer-reviewed literature. OpenCYP provides data on human variability on baseline of activities and polymophism frequencies for selected cytochrome P-450 isoforms (CYP1A2, CYP2A6, CYP2D6, CYP3A4/3A5 and CYP3A7) in healthy adult populations from world populations. CYP enzymatic activities were generally expressed as the metabolic ratio (MR) between an unchanged probe drug and its metabolite(s) in urine or plasma measured in healthy adults. Data on other age groups were very limited and fragmented, constituting an important data gap. Quantitative comparisons were often hampered by the different experimental conditions used. However, variability was quite limited for CYP1A2, using caffeine as a probe substrate, with a symmetrical distribution of metabolic activity values. For CYP3A4, human variability was dependent on the probe substrate itself with low variability when data considering the dextromethorphan/demethilathed metabolite MR were used and large variability when the urinary 6β-hydroxycortisol/cortisol ratio was used. The largest variability in CYP activity was shown for CYP2D6 activity, after oral dosing of dextromethorphan, for which genetic polymorphisms are well characterised and constitute a significant source of variability.

It is foreseen that the OpenCYP database can contribute to promising tools to support the further development of QIVIVE and PBK models for human risk assessment of chemicals particularly when combined with information on isoform-specific content in cells using proteomic approaches.
© 2021 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

1. Introduction

In vitro methods and physiologically-based modelling have been identified as key research priorities for the international research community involved in toxicological research and chemical risk assessment at the regulatory level. Such methods

* Corresponding author.
E-mail address: susanna.vichi@iss.it (S. Vichi).

http://dx.doi.org/10.1016/j.toxlet.2021.07.019
0378-4274/© 2021 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Table 1
List of queries used for the ELS (formatted for Scopus) for CYP1A2, CYP2A6, CYP2D6, CYP3A4, CYP3A5, CYP3A7.

STUDY QUESTION	Objective: Data collection of human of: CYP1A2, CYP2A6, CYP2D6, CYP3A4, CYP3A5, CYP3A7.
Search string	TITLE-ABS-KEY (("population distribution" OR "tissue distribution " OR "tissue localization" OR intestine OR liver OR kidney OR lung OR "expression level*" OR "gene expression" OR "genetic polymorphism*" OR "individual susceptibility" OR "gene environment" OR "ethnic variability" OR caucasian OR asian OR "Afro American" OR hispanic OR "race difference*" OR "age difference*" OR "gender difference*" OR "sex difference*" OR ontogenesis OR "foetal stage" OR neonate* OR children OR "elderly people" OR adult* OR genotype OR induction OR inhibition)) AND TITLE-ABS-KEY ((human* W/50 ("CYP3A4" OR "CYP3A5" OR "CYP3A7" OR "CYP1A2" OR "CYP2A6" OR "CYP2D6"))) AND NOT ("cell line*" OR "cell culture*" OR HEPArg OR HEPG2 OR recombinant OR supersome*)

and models provide opportunities to a more mechanistic understanding of toxicokinetic (TK) and toxicodynamic (TD) processes allowing quantitative *in vitro in vivo* extrapolations (QIVIVE) and ultimately, with a growing commitment from research and scientific advisory bodies, reducing animal testing. In the food and feed safety area, the European Food Safety Authority (EFSA) published in 2014 a scientific report addressing "modern methodologies and tools for human hazard assessment of chemicals" including *in vitro* and *in silico* methods, integrated test strategies, OMICs and physiologically-based kinetic and dynamic modelling (i.e. PBK and PBD models). The report identified, through consultations of national and international public health agencies, key priorities to implement such new approach methodologies (NAMs) in the risk assessment process (EFSA, 2014). From these key priorities, EFSA launched a number of collaborative research projects including the creation of open source toxicological databases such as EFSA OpenFoodTox and related *in silico* models (Dorne et al., 2017, 2021; Benfenati et al., 2020) as well as generic open source PBK models for human health, animal health and ecological risk assessment (Grech et al.;, 2019; Baas et al., 2018; Lautz et al., 2020a,b,c).

A key strategy to implement these tools in chemical risk assessment is to move from external exposure metrics to internal dose metrics using TK parameters. Human variability in phase I and phase II metabolic pathways as well as transporters and renal excretion, from which the internal dose is strictly dependent, is critical to obtain reliable model estimates (Bessems et al., 2014). Characterisation of human variability in Absorption, Distribution, Metabolism and Excretion (ADME) processes requires the mapping of human isoforms involved in xenobiotic metabolism, frequencies of known polymorphisms across populations of different geographical origin, and the derivation of variability distributions for internal exposure dose metrics using peak concentrations (Cmax), area-under the plasma-concentration curve (AUC) or clearance, which, depending on the chemical and its toxicological profile, can represent markers of acute or chronic exposure. This ultimately allows the integration of TK processes with TD data, reflecting dose-response relationships for markers of adverse effects in relation to the Modes of action (MoA) and Adverse Outcome Pathway (AOP) of chemicals as a quantitative continuum.

The present work is part of an EFSA funded project "Modeling human variability in toxicokinetic and toxicodynamic processes using Bayesian meta-analysis, physiologically-based modeling and in vitro systems". The project aims to develop predictive tools for human health risk assessment of single and multiple chemicals compounds by integrating human variability in TK and TD processes and physiologically-based kinetic models to increase robustness, transparency and openness of EFSA scientific assessments (Hardy et al., 2015). In the frame of different EFSA funded projects, analysis of human variability has been recently published for a range of phase I isoforms (CYP3A4, CYP2C19, CYP2C19, CYP2D6, paraoxonase-1, carboxylesterases), Phase II enzymes (UDP-glucuronyltransferases, glutathione-S-transferases) and transporters (Quignot et al., 2021; Darney et al., 2019, 2020a, b,2021 submitted; Kasteel et al., 2020a,b; Buratti et al., 2021; Di Consiglio et al., 2021 submitted).

Amongst phase I xenobiotic biotransformation enzymes, Cytochrome P450s (CYPs) have a predominant role. Human variability in CYP activity is strongly affected by genetic polymorphisms, although such metabolic activities can be also modulated through differences in enzyme espression influenced by a number of variables including age, health status, life-style, and the geographical origin of the population (Ingelman-Sundberg, 2004; Dorne et al., 2003; Darney et al., 2019, 2021; Quignot et al., 2018, 2021). Overall, any shift in the metabolic capacity of CYP isoforms may lead to a change in the internal dose of toxicologically relevant parent compounds or metabolite(s) and hence to a decreased or increased response to therapeutic drugs as reported and retrievable in available central repositories (e.g PharmGKB and PharmVar Consortium) or sensitivity to adverse effects resulting from exposure to xenobiotics. In order to address the variability in the metabolic activity of CYP isoforms, this manuscript describes the results of extensive literature searches and critical data collection to investigate human variability in metabolic activities and relevant genetic polymorphisms in world populations of CYP isoforms in healthy adults, namely CYP1A2, CYP2A6, CYP2D6, CYP3A4/3A5 and CYP3A7. Results are presented in the open-source database OpenCYP. Future perspectives conclude particularly to explore the use of such database to develop QIVIVE extrapolations models fro chemical risk assessment.

2. Materials and methods

Extensive Literature Searches (ELS) were performed using the methodology described in the EFSA Systematic Review Guidance (EFSA Guidance, 2010). The ELS was performed (January 1989 –October 2019) from the primary peer-reviewed scientific literature on CYP isoform activities looking into databases and multidisciplinary platforms of international coverage, namely Scopus, Web of Science, PubMed, Food Sience Source and Agricola. The ELS were carried out through the formulation of appropriate queries, exploiting the use of specific key terms for each topic and Boolean operators (eg. AND, OR, NOT), using different specific syntax for each database, designed to include as many studies as possible relevant to the scope and to minimise the bias (Table 1).

2.1. Primary screening

The primary screening was carried out using titles and abstracts, after removal of duplicates. Scientific studies reporting *in vivo or ex-vivo* activity of the selected human CYP isoforms (i.e. CYP1A2, CYP2A6, CYP2D6, CYP3A4/3A5 and CYP3A7) in the English language were included (inclusion criteria).

Papers belonging to one of the following categories were excluded (exclusion criteria):

- Studies on species other than human
- *in vitro* studies

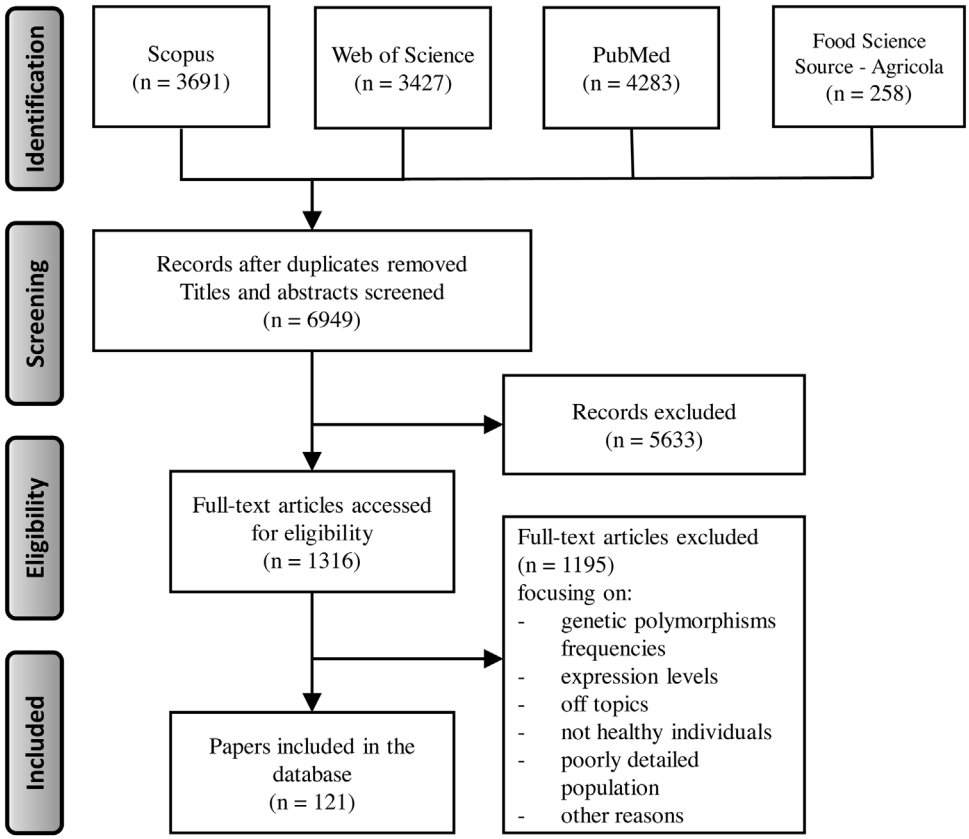

Fig. 1. Prisma flow diagram illustrating the results from the extensive literature searches on activities of major CYP isoforms (CYP1A2, CYP2A6, CYP2D6, CYP3A4, CYP3A5, CYP3A7) in humans.

- Studies devoted to set up an analytical method
- Modelling studies only
- TD data only or drug development
- Data from studies on unhealthy individuals (patients)
- Data from studies on individuals using medication or on occupationally exposed individuals.

Only entries meeting the inclusion/exclusion criteria were imported into EndNote™ reference software for further evaluation.

2.2. Secondary screening

The secondary screening involved an in-depth analysis of the content and quality of each individual peer reviewed paper, taking into account study design, methods, results, statistical analysis and reporting. Factors which may affect the quality of results, such as insufficient description of methods, results reported without adequate descriptive statistics, poor description of the human population were excluded; proceedings, letters to editors, conference papers, short communications, for which an adequate quality check could not be carried out, were also excluded. The reliability of the data was evaluated by applying the Klimisch scoring system (Klimisch et al., 1997), when applicable, so that studies classified in category 4 were excluded from further analysis due to poor quality. For population studies for which application of the Klimish score was not appropriate, the scoring system reported in Darney et al. (2019) was applied to exclude papers reporting a) no or insufficient information on the enrolled population (number, age, health status, geographical origin of the population), b) insufficient description of the methodology used and c) pharma/toxicokinetic data without descriptive statistics.

2.3. Data extraction

Data from eligible studies after the secondary screening were extracted and captured in a data extraction form, organised in a structured Excel database, adapted from OECD harmonised templates (OHTs) from an EFSA's data model allowing consistency with EFSA's chemical hazards database (OpenFood-Tox) (Dorne et al., 2017). Reviews, book chapters and other sources not reporting primary datasets for CYP activity were not considered for data extraction to avoid multiple inclusion of the same dataset from different references.

The database was structured as a single excel sheet showing in sequential order the data extracted from the selected papers: each study was identified with an ID number and information extracted included references, CYP isoform activity, details of the study design and method used (e.g. probe substrate(s), methods for quantification of the parent/metabolite(s), matrix in which the metabolites were measured, time of treatment), number of individuals, summary statistics of *in vivo* parameters (e.g. sample size, arithmetic or geometric mean, median, standard deviation, confidence intervals, etc.) and factors impacting on variability (e.g. world populations, presence of polymorphic enzymes) (Supplementary Material A).

Whenever papers compared data between i) healthy adults (e.g. control group) *vs* individuals affected by specific diseases, or ii) healthy adults (e.g. control group) *vs* individuals taking specific medication or occupationally exposed, data from the control group were used, while data referring to the patients or 'exposed' people were flagged into the database by using a separate note column (pathologies or exposure type, respectively) as additional information.

The final database contains the metabolic activities for the selected CYP isoforms: CYP1A2, CYP2A6, CYP2D6, CYP3A4, CYP3A5, CYP3A7, across healthy adults and world populations.

2.4. Meta-analyses of TK data

For the in vivo probe substrates identified from the ELS, data from human PK studies following oral (single) or intravenous (bolus) exposure to derive kinetic parameters such as Cmax, area under the curve-AUC and clearance were collected for CYP2A6 and 1A2. The search was carried out with the above mentioned criteria, to evaluate the possibility of performing a meta-analysis, aimed at estimating the distributions of variability for these two CYP activities in non-phenotyped subjects for each probe substrate and deriving the related variability distributions and uncertainty factors, following the method described in Darney et al. (2019).

3. Results and discussion

3.1. Overview of the OpenCYP database

The whole data collection strategy and information flow of information for the six selected CYP isoforms namely CYP1A2, CYP2A6, CYP2D6, CYP3A4, CYP3A5, CYP3A7 is summarised in Fig. 1. As expected, the largest datasets were available for the major CYP3A subfamily and CYP3A4 isoform, CYP1A2 and CYP2D6, while datasets for CYP2A6 and CYP 3A5/3A7 isoforms were much more limited (Fig. 2).

Overall, a total number of 121 papers providing quantitative data on isoform-specific CYPs activities and polymorphism frequencies across world populations were extracted and computed in the OpenCYP excel database (Supplementary material A and DOI for EFSA knowledge junction: 10.5281/zenodo.5031737). The complete list of relevant articles available in the database is provided in Supplementary material B.

Baseline enzymatic activities for healthy adults, measured using isoform-specific probe substrates for each CYP, were preferentially collected using stable and reproducible parameters such as the metabolic ratio (MR), defined as the ratio(s) between the amount of an unchanged probe substrate and its metabolite(s) measured in urine, saliva, serum or plasma (Pelkonen et al., 1998, 2008). This approach is widely used to investigate inter-individual differences in CYP phenotypes, and implies that probes substances comply with key requirements to be applied in human *in vivo* studies, such as being non-toxic, easily available and measurable in biological fluids with analytical methods that are sufficiently specific and sensitive. MRs were stratified on the basis of CYP genetic polymorphisms for a large number of studies to take into account the impact of the genotype

3.2. Isoform-specific CYP enzymatic activities and consideration of geographical ancestry in human populations

Characterisation of the geographical ancestry across human world populations were often poorly described and constitute a potential bias to quantify isoform-specific CYP enzymatic activities from the literature, even though the country in which the subjects were recruited for each study was systematically spelt out. Such a bias is particularly relevant for societies of cosmopolitan nature, where mixed populations from a range of geographical ancestry exist. Therefore, when geographical ancestry of the individuals was not specified, or a heterogeneous group was considered, a comparable picture of the isoform-specific CYP activities could not be reached for different world populations. Overall, when the geographical ancestry was indicated, the majority of isoform-specific CYP activities were available for Caucasian and Asiatic populations (Table 2).

In addition, information on the age of the recruited subjects almost exclusively referred to healthy adult populations, reported the data were heterogeneously reported either as ranges or average value (with or without the standard deviation), or even without summary statistics i.e 'young adults'. Finally, in some instances, the "adult population" spanned over a very wide age group (e.g. 36–85 years) which also included the elderly). For such peer reviewed articles, this inconsistency limited options to compare healthy adults and the elderly subgroup. Overall, data for other age groups including the elderly, children and newborns are very limited.

In addition to the MR, taken as the preferable metrics, many other parameters were reported for the quantification of the isoform-specific CYP activities as follows (Table 3):

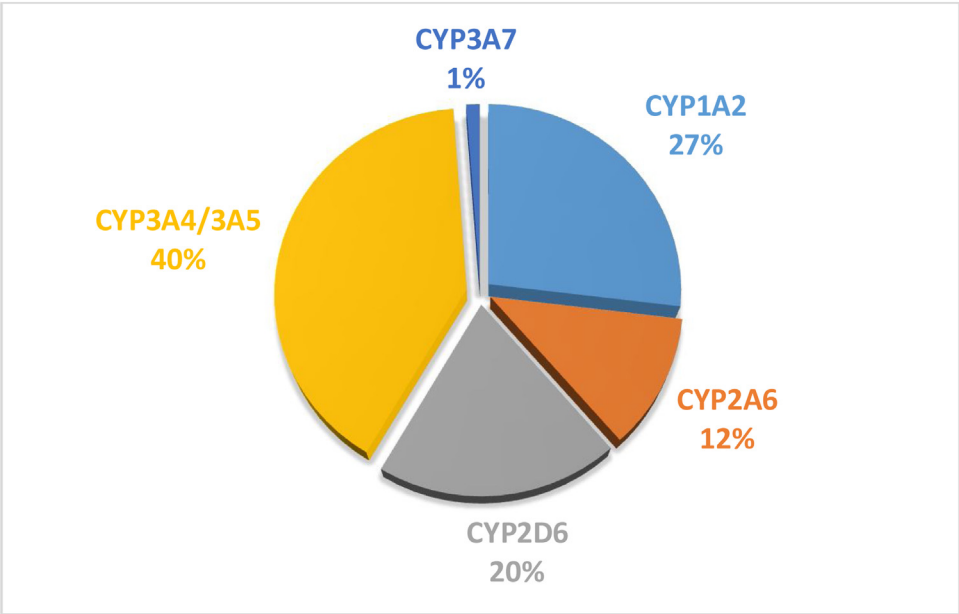

Fig. 2. Relative contribution of CYP isoform activities data to the OpenCYP database.

Table 2
Geographical ancestry of human populations for isoform-CYP activities included in the database.

	Geographical origin	n. of papers	% values
CYP1A2			
	No infornation available	21	46
	Mixed geographical ancestry	5	11
	Caucasian	10	22
	Asiatic	10	22
	Other	0	0
	Tot	46	100
CYP2A6			
	Asian	6	30
	Afro-American	3	15
	no info	5	25
	white/Caucasian	3	15
	Other	3	15
	Tot	20	100
CYP2D6			
	No info	17	50
	Caucasian	10	29
	Chinese/Japanese/Asian	4	12
	Mixed	3	9
	Tot	34	100
CYP3A			
	no info	37	52
	Chinese/Japanese/Asian	14	20
	Caucasian	12	17
	mixed	8	11
	Tot	71	100

- **CYP1A2** activity was quantified using kinetic parameters for probe substrates (e.g. caffeine and theophylline clearance), caffeine breath test and methoxy-resorufin-O-dethylase (MROD) assays.
- **CYP2A6** activity was measured through coumarin hydroxylation rate.
- **CYP2D6** activity was quantified using debrisoquine recovery ratio, dextromethorphan (DEX) O-demethylation or DEX measurements, desipramine or metoprolol clearance.
- **CYP3A** activity was measured with 6-β-hydroxycortisol/cortisol ratio and the DEX/MM ratio as well as with other selected probe substances (e.g. midazolam, alprazolam or testosterone) and as the percentage of erythromycin metabolised per hour (references in Supplementary material A).

Overall, isoform-specific activity and MRs evidenced high variability which essentially reflects differences in the experimental conditions tested, according to probe concentrations, times of measurement, biological matrix (e.g. plasma or urine), exposure routes and, when available, characteristics of the population enrolled in the study (e.g. gender, age, geographical origin).

3.2.1. CYP1A2 activity

For **CYP1A2,** the most widely used probe substrate was caffeine (94 % of the extracted papers) (Table 4). It is involved in the main route of caffeine metabolism in humans (70–80 %) that is the N-3-demethylation to paraxanthine (Thorn et al., 2012), even though other enzymes are known to be involved in further reactions to a much lesser extent (xanthinoxidase, N-acetyl transferase and with lesser extent also CYP2E1, CYP2A6, CYP3A4, CYP3A5) (Dorne et al., 2001). However, caffeine kinetics is very well characterised in humans (i.e. complete absorption, high clearance and short half-life). Median value of caffeine tested doses was shown to be 100 mg (min = 45 mg; max = 300 mg) with detection in biological matrices, primarily urine and plasma (see references in Supplementary material A). Each peer reviewed paper reported the mean or the median of MR value after oral caffeine administration (100−300 mg): they were spread over a wide range (0.1–27.6) with the minimum measured in urine after oral dosing of 100 mg caffeine in a female Chinese population

Table 3
Validated Human assays for the measurement of CYP3A, CYP2D6, CYP1A2 and CYP2A6 isoform-specific activities.

Different parameters used to describe the enzymatic activity	n. of papers	% values
CYP1A2		
Caffeine Metabolic Ratio	38	79
PK parameters (e.g. clearance)	6	13
Caffeine Breath test	3	6
MROD assay	1	2
Tot	48	100
CYP2A6		
nicotine metabolic ratio	7	41
caffeine metabolic ratio	6	35
coumarin hydroxylation rate	4	24
Tot	17	100
CYP2D6		
DEX/DOR ratio	20	54
debrisoquine recovery ratio	5	14
metoprolol/hydroxymetoprolol ratio	4	11
sparteine metabolic ratio	2	5
other (DEX clearance, encainide/(ODE + MODE) ratio, DEX O-demethylation, (+)-tramadol/(+)(R,R-O-demethyltramadol) ratio, desipramine clearance, metoprolol clearance)	6	16
Tot	37	100
CYP3A		
pharmacokinetics variables of midazolam	23	28
6-β-hydroxycortisol/cortisol ratio	15	19
DEX/MM ratio	10	12
% of erytromycin metabolized per hour	9	11
pharmacokinetics variables of alprazolam	5	6
pharmacokinetics variables of testosterone	3	4
Other * (n < 3)	16	20
Tot	81	100

Table 4
List of suitable probes for CYP1A2, CYP2A6, CYP2D6 and CYP3A metabolic activity assessment.

Probes	n. of papers	% values
CYP1A2		
Caffeine	43	94
methoxyresorufin	1	2
tizanidine	1	2
theophylline	1	2
Tot	46	100
CYP2A6		
nicotine	8	44.4
caffeine	6	33.3
coumarin	4	22.2
Tot	18	100
CYP2D6		
DEX/DOR	22	61
debrisoquine	5	14
metoprolol/hydroxymetoprolol	4	11
sparteine	2	6
other (encainide/(ODE + MODE), (+)- tramadol/(+)(R,R-O-demethyltramadol), desipramine)	3	8
Tot	36	100
CYP3A		
midazolam	23	27
6-β-hydroxycortisol	16	19
DEX/MM	11	13
14C N-methyl erytromycin	9	11
alprazolam	5	6
testosterone	3	4
Other *	18	21
Tot	85	100

* Other: Alfentanil, Budesonide, Buspirone, Caffeine, Cyclosporine, Dapsone, Erythromicin, Omeprazole, Prednisolone, Quinidine, Quinine, Simvastatin, Tacrolimus, Verapamil).

(0–12 h collection) (Chen et al., 2011) and the maximum in a Jordanian population of both sexes (24 h collection) (Hakooz and Hamdan, 2007), respectively. When data from different papers were grouped according to the matrix and time of collection, MR values were:

1) Mean Urine MR 4–6 hours after dosing: 6.05 ± 2.26 (SD) and a median value = 6.11 (n = 466)
2) Mean Urine MR 12–24 hours after dosing: 17.48 ± 6.01 (SD) and a median value = 18.3 (n = 90)
3) Mean Plasma MR 4–6 hours after dosing: 0.51 ± 0.27 (SD) and a median value = 0.48 (n = 950)

Overall, variability was relatively limited, despite heterogenous data from different human populations (gender, different age, diet and geographical origin, and likely genetic make up) with similar means and median values indicating a symmetric distribution of values (Gaussian). However, it can be considered that data variability was averaged out by the mean (or median) value reported in each paper. In contrast, a paper on a Tunisian group of individuals genotyped for 6 different CYP1A2 variants reported a MR in plasma higher than the mean value cited above (range 1.97–2.55) even though variants did not impair caffeine metabolism (Imène et al., 2015).

The Bayesian meta-analysis methodology applied in Darney et al. (2019) for CYP3A4, for the derivation of isoform-specific human variability and related uncertainty factors, has not been applied to CYP1A2. Indeed, looking at the retrieved literature, no new significantly relevant literature on CYP1A2 probe substrates was available since Dorne et al. (2001) and such an analysis was considered unnecessary without prior information (non-informative distribution).

3.2.2. CYP2A6 activity

Caffeine and nicotine (the latter tested at the median dose of 2 mg in plasma and urine) were also the most common probes used to address **CYP2A6** activity (respectively in 33 % and 44 % of the extracted papers). Caffeine was selected for the high specifity of the conversion of the caffeine metabolite paraxantine (1,7-dimethylxanthine, 17X) to 1,7-dimethyl uric acid (17U) exclusively catalyzed by CYP2A6, which activity can be measured through the comparison of the urinary molar ratios of these metabolites (Nowell et al., 2002). Nicotine is instead frequently used for a routine CYP2A6 phenotyping, since the essay employs a non-invasive method (references in Supplementary material A). For nicotine, the minimum value of MR was 0.08, measured in plasma in a population of African-Canadians after oral dosing of 4 mg (4,5 h collection) (Al Koudsi et al., 2009), while the maximum value was 11.5 measured in plasma after oral dosing of 2 mg (2 h collection) and reported for an Afro-American population (Fukami et al., 2004). For 17X/17U, the minimum MR = 0.06 was measured in urine after oral dosing of 100 mg (24 h collection) (Hakooz and Hamdan, 2007), while the maximum value was 0.48 after oral dosing of 106–118 mg (6 h collection) (Begas et al., 2017).

As for CYP1A2, the Bayesian meta-analysis methodology applied in Darney et al. (2019) to derive isoform specific uncertainty factors was not applied to CYP2A6, since looking at the retrieved literature no significant new data were available since Dorne et al. (2004) and the variability derived in that paper is still considered accurate (21–34 % in healthy adults).

3.2.3. CYP2D6 activity

Dextromethorphan (DEX) has been used extensively over the years as a probe substrate to assess the *in vivo* metabolic activities of **CYP2D6** and also **CYP3A**, based on its mediated metabolism to dextrorphan (DOR), or 3-methoxymorphinan (3-MM). Here, the ELS showed that DEX is a CYP2D6 probe substrate and CYP3A4 probe substrate in 61 % and 13 % of the extracted papers respectively and this is particularly valid for historical papers from the 1990s. More recently, DEX has been dismissed as a CYP2D6 probe substrate, after its recognition as a psychotropic substance carrying a potential for abuse and dependence (Martinak et al., 2017). Median values of the DEX tested oral doses in humans were shown to be around 30 mg for both CYP2D6 (range 25–80 mg) and CYP3A (range 30–80 mg) (see references in supplementary material).

For CYP2D6, MRs for DEX/DOR were very widely distributed ranging from very low values with a minimum of $\simeq$ 0.002 indicating efficient biotransformation typical of extensive metabolisers (EMs) to a maximum of 0.59 associated with poor metabolisers (PMs) (Funck-Brentano et al., 1992; Wenk et al., 2004; McCune et al., 2001; Markowitz et al., 2003a,b,c; o'Connell et al., 2006; Tushar et al., 2007; Al-Jenoobi et al., 2014a, b, 2015; Saruwatari et al., 2014). Historically, a urinary MR (DEX/DOR) $\geq$0.3 has been generally applied to define PM phenotypes. From this database, the Funck-Brentano et al. (1992) enrolled 110 individuals of which 52 individuals were also genotyped: results indicated that both CYP2D6 EM and PM phenotypes were identified from the DEX/DOR MR calculated in urine. On the other hand, other studies showed that dextromethorphan metabolic ratio did not reliably discriminate heterozygous and homozygous EMs (Evans and Relling, 1991).

Grouping of datasets using the average DEX oral dose (30 mg), the mean urinary MRs for DEX (collected after 8 h) were 0.14 $\pm$ 0.50 with a median value of 0.01. Human subpopulations included both male and female genders, different age groups, individuals with a range of diets, geographical origin and likely a range of CYP2D6 genotypes and phenotypes. Overall, human variability in CYP2D6 was substantial, as also indicated by the SD reported in each study, generally higher than the mean value itself. Therefore, although data variability could be expected to be averaged out by the mean (or median) value, reported in each paper, this is not the case for CYP2D6. The fact that mean and median value differ by more than one order of magnitude indicates that the distribution of values is asymmetrical, with a distribution shifting to lower values. Data on human variability in kinetics for CYP2D6 including inter-phenotypic differences have been described elsewhere (Darney et al., 2021), using the Darney et al. (2019) Bayesian meta-analysis method.

3.2.4. CYP3A4 activity

Since **CYP3A4** is the predominant enzyme of CYP3A subfamily and plays a major role in drug metabolism, a significant number of probe substrates are available (Table 4). From the retrieved literature, the most frequently probe susbtrates were identified as:

1) midazolam (27 % of publications) through its 1-hydroxylation which is mainly (but not exclusively) responsible for clearance in humans. Midazolam *in vivo* kinetic parameters related to 1-hydroxymidazolam formation (i.e. AUC, Cmax, tmax, $t_{1/2}$, Vmax/Km) have been previously applied for meta-analysis of human CYP3A4 variability (Darney et al., 2019);

2) 6β-hydroxycortisol which has been historically often selected as a relatively non-invasive endogenous marker for CYP3A4 *in vivo* induction and inhibition, even if its sensitivity and selectivity is still controversial.

With regards to the variability of the baseline level of CYP3A4 activity, it was not possible to draw robust conclusions based on the midazolam/hydroxymidazolam MR, due to the limited number of individuals enrolled and the diferent experimental conditions used for its measurement (see Supplementary material A). Using urinary MR 6β-hydroxycortisol/cortisol as index of CYP3A4/A5 activity, interindividual differences, including differences across world populations, were shown with lowest MR value reported in 13 Asian females (2.1) whereas MR in Caucasians individuals from the same study were nearly 3-fold higher (5.6). Overall, highest MR values were reported for 36 egyptian males (23.3), while for females (n = 29) MR values were nearly 4-fold lower (mean = 6.5). However, no bimodality in either gender was observed and none of the studies carried population genotyping (Lin et al., 1999; El Desoky et al., 2005). Grouping of the datasets while considering urinary 6β-hydroxycortisol/cortisol MR (collected in the morning the day after dosing), the MR distribution was 11.24 $\pm$ 17.25 (median value = 6.5). Conversely, when selecting CYP3A4/A5 activity data from urinary DEX/3-MM ratio (25−30 mg DEX collected between 0−8 hours after oral dosing), variability in MR was lower with mean/SD values of 3.27 $\pm$ 2.86 and a median value of 2.95.

Some authors suggested that there are no 'ideal' markers to predict CYP3A4 enzymatic activity with high accuracy, since none

Table 5
Frequencies of most common genetic polymorphisms in human world populations for CYP1A2.

Gene	Change	Polymorphic allel	SNP rs	MAF[a] Asian	MAF[a] - Caucasian	MAF[a] - African/ African American	MAF[a] – other	additional notes	References
CYP1A2	−163C > A	*1 F (or *1 F, *1 J and *1 K)	rs762551	0.30−0.61	0.27−0.34	0.40−0.68		gMAF, global MAF 0.35	Preissner et al., 2013; Pilgrim et al., 2012; Zanger and Schwaby, 2013; Xue et al., 2014; Zhou et al., 2009.
	−2464T→delT	*1D	rs35694136	0.42−0.707	0.048−0.24 in Caucasian; 0.92 in Turkish	0.075−0.40			Preissner et al., 2013; Zhou et al., 2009.
	−3860G > A	*1C	rs2069514	0.063−0.27	0.008−0.08	0.071−0.40	0.20−0,30 Hispanic	gMAF, global MAF 0.19	Preissner et al., 2013; Zanger and Schwaby, 2013; Ren et al., 2016; Xue et al., 2014; Zhou et al., 2009.
	−739T > G	*1E, *1 G, *1 J, *1K		0.027−0.10	0.004−0.09	0.03−0.13			Zhou et al., 2009
	−729T > C	*1K	rs12720461	0−0.04	0.03−0.05	0.030			Zhou et al., 2009
	5347 T > C	*1B, *1H, *1 G, *3, *8, *15, *16		0.19−0.2	0.392				Zhou et al., 2009

[a] = minor allele frequency.

of them are uniquely metabolised by the isoform (Benet, 2005) and the possibility that multiple substrates are simultaneously bound to the isoform cannot be excluded (Ekroos and Sjogren, 2006). As expected, a data gap emerged regarding the in vivo activity data for CYP3A7; only few in vitro were data collected from the screened literature, that were exceptionally entered into the inventory file.

3.3. Genetic polymorphisms of CYP isoforms

Genetic polymorphisms of CYP isoforms strongly contribute to human variability in CYP activities including differences across populations of different geographical ancestry and also constitute a major source of inter-individual differences in xenobiotic kinetics and dynamics. Such human variability has been extensively investigated in the area of pharmaceuticals to pave the way for personalised medicine (Sim and Ingelman-Sundberg, 2011; Lauschke and Ingelman-Sundberg, 2016). Moreover, CYP isoforms are also involved in the bioactivation of pro-carcinogens and cellular signaling molecules and such extensively studied polymorphisms are well characterised as potential susceptibility factors in the aetiology and onset of a range of cancers and other diseases. While considering the impact of these isoform-specific genetic polymorphisms on CYP1A2, 2A6, 2D6 and 3A4/3A5/3A7 activities, the available peer-reviewed literature was large (n = 418)

Table 6
Frequencies of most common genetic polymorphisms in human world populations for CYP2A6.

Gene	Polymorphic allele	Change	SNP rs	MAF[a] - Asian	MAF[a] - Caucasian	MAF[a] - African/ African American	MAF[a] - other	additional notes	References
CYP2A6	*1B	gene conversion with CYP2A7		0.267–0.542	0.276–0.335	0.112–0.198			Preissner et al., 2013; López-Flores et al., 2017
	*1D	−1013A > G		0.10–0.20	0.267–0.40	>0.5			López-Flores et al., 2017
	*1F	5717C > T		0	0.018	0			López-Flores et al., 2017
	*1G	5717C > T, 5825A > G		0	0.012	0.123–0.133			López-Flores et al., 2017
	*1H	−745A > G		0.016–0.045	<0.11	0.098			López-Flores et al., 2017
	*2	1799T > A	rs1801272	0–0.011	0.011–0.053	0–0.01		0.013 gMAF	Zanger and Schwaby, 2013; Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017
	*4	crossover with CYP2A7		0.049–0.256	0.003–0.04	0.015–0.020			Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017
	*5	6582G > T	rs5031017	0–0.146	0-<0.001	0-<0.001			López-Flores et al., 2017; Zhou et al., 2017
	*7	6558T > C	rs5031016	0.02–0.13	<0.001	0-<0.001		0.04 gMAF	Zanger and Schwaby, 2013; Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017
	*8	6600G > T	rs28399468	0.003–0.05	0–0.003	0–0.003			López-Flores et al., 2017; Zhou et al., 2017
	*9	−48T > G	rs28399433	0.144–0.27	0.040–011	0.040–0.12		0.13 gMAF	Preissner et al., 2013; Zanger and Schwaby, 2013; Naidoo et al., 2014; Murphy, 2017; Zhou et al., 2017
	*10	two point mutations		0–0.043	0.-<0.001	0.-<0.001			López-Flores et al., 2017; Zhou et al., 2017
	*12	several SNPs		0–0.013	0.029	0–0.004	0.035–0.047 Hispanics	0.008–0.035 gMAF	Preissner et al., 2013; Murphy, 2017; López-Flores et al., 2017
	*13	−48T > G, 13 G > A		0–0.015	0	0			López-Flores et al., 2017
	*14	86G > A		0–0.035	0.014–0.052	0.008–0.014			López-Flores et al., 2017; Zhou et al., 2017
	*15	−48T > G, 2134A > G		0–0.022	0	0			López-Flores et al., 2017
	*16	2161C > A		0-<0.001	0–0.036	<0.001–0.017			López-Flores et al., 2017; Zhou et al., 2017
	*17	several point mutations		0	0	0.073–0.112			Naidoo et al., 2014; Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017
	*18	three subvariants		0–0.014	0.003–0.022	0–0.006			López-Flores et al., 2017; Zhou et al., 2017
	*19	multiple changes		0.005–0.014	0.-<0.001	0-<0.001			López-Flores et al., 2017; Zhou et al., 2017
	*20	multiple changes		0.000	0.000	0.01–0.017			López-Flores et al., 2017
	*21	51G > A, 6573A > G		0–0.019	0.005–0.028	0.002–0.007			López-Flores et al., 2017; Zhou et al., 2017
	*23	2161C > T		0–0.001	0	0.01–0.02			López-Flores et al., 2017; Zhou et al., 2017
	*24	two subvariants		0-<0.001	0	0.007–0.013			López-Flores et al., 2017; Zhou et al., 2017
	*35	two subvariants		0.005–0.128	0–0.149	0.025–0.059			López-Flores et al., 2017

[a] = minor allele frequency.

Table 7
Frequencies of the most common genetic polymorphisms in human world populations for CYP2D6.

Gene	Polymorphic allele	Change	SNP rs	MAF[a] - Asian	MAF[a] - Caucasian	MAF[a] - African/ African American	MAF[a] - other*	additional notes	References
CYP2D6	*2	2850C > T, 4180 G > C and other variants	rs16947, rs1135840	0.131–0.362	0.041–0.343	0.141–0.267	0.215		Gaedigk et al., 2017; Zhou et al., 2017
	*3	2549delA	rs35742686	0.–0.001	0.013–0.041	0–0.003	0.007	gMAF 0.009	Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017
	*4	1846 G > A	rs3892097	0.004–0.116	0.150–0.250	0.034–0.119	0.105	gMAF 0.106	Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017
	*4D	100C > T; 1039C > T; 1661 G > C; 1846 G > A; 4180 G > C			0.034				Preissner et al., 2013
	*4 L	100C > T; 997C > G; 1661 G > C; 1846 G > A; 4180 G > C			0.045				Preissner et al., 2013
	*5	Recombination		0.02–0.065	0.029–0.041	0.04–0.062	0.024		Gaedigk et al., 2017; Preissner et al., 2013; Zhou et al., 2017
	*6	1707delT	rs5030655	0–0.001	0.009–0.022	0–0.003	0.004	gMAF 0.01	Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017
	*7	2935A > C	rs5030867	0–0.008	0–0.01	<0.001	0.000		Gaedigk et al., 2017; Preissner et al., 2013; Zhou et al., 2017
	*8	1758G > T	rs5030865	0-<0.001	0	<0.001	0.001		Gaedigk et al., 2017; Zhou et al., 2017
	*9	2615–2617delAAG	rs5030656	0–0.009	0.016–0.021	0.001–0.005	0.012		Gaedigk et al., 2017; Preissner et al., 2013; Zhou et al., 2017
	*10	100C > T	rs1065852	0.065–0.70	0.024–0.080	0.032–0.120		gMAF 0.260	Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017
	*12	124G > A	rs5030862	0	0	<0.001	0.042		Gaedigk et al., 2017; Zhou et al., 2017
	*13 revised	hybrid allele		0	0.002	0.005	0		Gaedigk et al., 2017
	*14	1758G > A	rs5030865	0–0.016	0	0–0.002	0.003		Gaedigk et al., 2017; Zhou et al., 2017
	*15	137insT; 137_138insT			0		0.006		Gaedigk et al., 2017
	*16	See CYP2D6*13			0.002				Gaedigk et al., 2017
	*17	1023C > T; 2850C > T	rs28371706; rs16947	0–0.001	<0.001–0.004	0.14–0.24	0.026	gMAF 0.049 for 1023C > T	Gaedigk et al., 2017; Zanger and Schwaby, 2013; Zhou et al., 2017
	*18	4133dupGTGCCCACT		<0.001					Gaedigk et al., 2017
	*19	2539_2542delAACT		0			0.003		Gaedigk et al., 2017
	*21	2573insC		0.005					Gaedigk et al., 2017
	*27	3853G > A		0.001					Gaedigk et al., 2017
	*28	19G > A; 1661 G > C; 1704C > G; 2850C > T; 4180 G > C		<0.001					Gaedigk et al., 2017
	*29	1659G > A; 1661 G > C; 2850C > T; 3183 G > A; 4180 G > C		<0.001	0–0.001	0.065–0.092	0.019		Gaedigk et al., 2017; Zhou et al., 2017
	*31	multiple changes		0	0.001		0.005		Gaedigk et al., 2017
	*34	2850C > T		0.002	0.030		0.001		Gaedigk et al., 2017
	*35	multiple changes		0.001	0.064		0.018		Gaedigk et al., 2017
	*36	multiple changes		0.015	0		0.002		Gaedigk et al., 2017
	*39	1661 G > C, 4180 G > C	rs1135840	0.002	0.069	0.004	0.004		Gaedigk et al., 2017
	*40	1863_1864 in.(TTT CGC CCC)2		0	0	0.006–0.017	0.001		Gaedigk et al., 2017
	*41	2988G > A	rs28371725	0.022–0.135	0.03–0.090	0.01–0.097	0.036	gMAF 0.055	Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017
	*42	3259insGT; 3259_3260insGT		0	0	0–0.004	0.004		Gaedigk et al., 2017; Zhou et al., 2017
	*43	77G > A		<0.001–0.008	<0.001	0.007–0.02			Gaedigk et al., 2017; Zhou et al., 2017
	*44	2950G > C		0.001					Gaedigk et al., 2017
	*45	multiple changes		0		0.052–0.068	0		Gaedigk et al., 2017
	*46	multiple changes		0		0–0.011	0		Gaedigk et al., 2017
	*45 or *46	multiple changes		0	0	0.029–0.040	0.011		Gaedigk et al., 2017
	*47	multiple changes		0.001					Gaedigk et al., 2017
	*48	972C > T		0.001					Gaedigk et al., 2017
	*49	multiple changes		0.007		0			Gaedigk et al., 2017
	*50	1720A > C		0.001		0			Gaedigk et al., 2017
	*51	multiple changes		0.001					Gaedigk et al., 2017
	*52	multiple changes		0.003					Gaedigk et al., 2017
	*54			0.001					Gaedigk et al., 2017

Table 7 (*Continued*)

Gene	Polymorphic allele	Change	SNP rs	MAF[a] - Asian	MAF[a] - Caucasian	MAF[a] - African/ African American	MAF[a] - other*	additional notes	References
		100C > T; 1039C > T; 1661 G > C; 2556C > T; 4180 G > C							
	*56A/B	multiple changes		0	0.007	0.0021–0.0025	0		Gaedigk et al., 2017
	*58	multiple changes		0		0–0.002			Gaedigk et al., 2017
	*59	1661 G > C; 2291 G > A; 2850C > T; 2939 G > A; 4180 G > C		0	0.007				Gaedigk et al., 2017
	*60	1887insTA; 2303C > T		<0.001					Gaedigk et al., 2017
	*63	multiple changes		<0.001					Gaedigk et al., 2017
	*64	multiple changes		0		0.003			Gaedigk et al., 2017
	*65	multiple changes		0.011		0.002			Gaedigk et al., 2017
	*66	See CYP2D6*13			0.001	0–0.00255			Gaedigk et al., 2017
	*69	multiple changes		0.006					Gaedigk et al., 2017
	*71	−1584C > G; 125 G > A; 1494 T > C		0.005					Gaedigk et al., 2017
	*73	multiple changes		0		0.002			Gaedigk et al., 2017
	*74	974C > A, 3609 G > T		0		0.002			Gaedigk et al., 2017
	*82	multiple changes		0			0.023		Gaedigk et al., 2017
	*100	multiple changes					0.002		Gaedigk et al., 2017
	*101	multiple changes					0.002		Gaedigk et al., 2017

[a] = minor allele frequency.

and included a range of populations of different geographical ancestry. Data collection was performed using reviews and meta-analyses. Relevant and informative reviews include Gaedigk et al. (2017) and Zhou et al. (2017) to whom the reader is referred for full details. The latter reported a global distribution map of CYP alleles with clinical relevance and integrated whole-genome and exome sequencing data from 56,945 unrelated individuals for five major human world populations to potentially develop population-adjusted pharmacological dosing (Zhou et al., 2017).

Tables 5–8 illustrates the frequencies of polymorphisms for *CYP 1A2, 2A6, 2D6* and *3A4/3A5/3A7* in healthy adults from different geographical ancestry.

3.3.1. Allelic frequency of CYP1A2 polymorphisms

The ELS search yielded articles on 13 different *CYP1A2* alleles with a variable occurrence depending on geographical origin and is summarised in Table 5. The most frequent alleles described were: *CYP1A2*1F* (-163 C > A), which is often reported as rs762551, but is also part of several other haplotypes (e.g. *1 F, *1 J and *1 K), with a frequency range of 0.27–0.34 in Caucasians, 0.30–0.61 in Asians and 0.40–0.68 in Africans/African Americans (Preissner et al., 2013; Pilgrim et al., 2012; Zanger and Schwaby, 2013; Xue et al., 2014; Zhou et al., 2009). The range values were narrow for Caucasians and wider for Asians and Africans and up to 2-fold higher compared to that in Caucasians. *CYP1A2* (5347 T > C), associated to the *1B, *1H, *1 G, *3, *8, *15, *16 haplotype, was absent in Africans/African Americans with a frequency of 0.19–0.20 in Asians, and showed highest value (0.392) in Caucasians (Zhou et al., 2009). *CYP1A2*1D* (-2464T→delT) had the lowest prevalence in Africans/African Americans, ranging from 0.075 to 0.40, in a similar fashion to that in Caucasians (0.048 to 0.24) and in contrast, frequencies were up to 10-fold higher (0.42–0.707) in Asians, with very high frequencies in Turkish populations for which around 92 % of the population is characterised by this CYP1A2 polymorphism (Preissner et al., 2013; Zhou et al., 2009). *CYP1A2*1C* (-3860G > A) has the lowest allele frequency in Caucasians (0.008–0.08), with the exception of Hispanics (0.20–0.30), the latter being higher than that reported in Asians (0.063–0.27), and in Africans/African Americans 0.071–0.40 (Preissner et al., 2013; Zanger and Schwaby, 2013; Ren et al., 2016; Xue et al., 2014; Zhou et al., 2009).

To date, a number of studies have investigated the association between genetic polymorphisms of CYP1A2 and its enzymatic activities. However, the functional consequences of such polymorphisms on the metabolism of CYP1A2 substrates is still controversial. In a recent review assessing the effects of genetic polymorphisms on CYP1A2 activity by caffeine metabolism (3570 subjects), the functional importance of −163C > A polymorphism (rs762551) was demonstrated. In this study, inter-individual differences were more pronounced amongst smokers, showing a higher enzymatic activity in homozygous or heterozygous individuals compared that in individuals expressing the CYP1A2 wild-type (Koonrungsesomboon et al., 2018). Conversely, no altered caffeine metabolic ratios were clearly demonsterd for other CYP1A2 polymorphisms.

3.3.2. Allelic frequency of CYP2A6 polymorphisms

To date, more than 40 alleles and some uncharacterised haplotypes have been identified at the CYP2A6 locus, rising from single nucleotide polymorphism (SNP), gene conversion, gene deletion and/or gene duplication (López-Flores et al., 2017). The distribution of CYP2A6 allelic variants has a typical pattern in populations of different geographical ancestry, as described for the other CYPs. Results from the retrieved review articles and meta-analyses showed a high prevalence in world populations of the *CYP2A6*1B* allele, generated by a gene conversion with CYP2A7 locus, ranging from 0.267 to 0.542 in Asians, from 0.276 to 0.335 in Caucasians and from 11.2 to 19.8 in Africans/African Americans (Preissner et al., 2013; López-Flores et al., 2017). The *CYP2A6*1D* (-1013A > G) has a high frequency especially in Africans/African Americans (>0.5), but it is common also in Caucasians, ranging from 0.267 to 0.40, and lower in Asian populations with a frequency of 0.10–0.20 (López-Flores et al., 2017). *CYP2A6*4* is one of the most widely studied CYP2A6 allele in human populations and rises from an homologous unequal crossover with CYP2A7, leading to the whole gene deletion that accounts for the majority of PMs. It is prevalent in Asians (in particular Japanese) with an allele frequency of 0.049–0.256, while it shows a frequency ≤ 0.04 in European descents and even a lower one in Africans/African Americans (Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017). *CYP2A6*5* and *7 variants are also common in Asians (frequency up to 0.146 and 0.130, respectively), but they are rare in

Table 8
Frequencies of most common genetic polymorphisms in human world populations for CYP 3A4, 3A5, 3A7.

Gene	Polymorphic allele	Change	SNP rs	MAF[a] - Asian	MAF[a] - Caucasian	MAF[a] - African/African American	MAF[a] - other	additional notes	References
CYP3A4	*2	644 T > C	rs55785340	0	0.011–0.027	0			Werk and Cascorbi, 2014; Zhou et al., 2017; Preissner et al., 2013
	*3	1334 T > C	rs4986910	0	0–0.04	0–0.033			Werk and Cascorbi, 2014; Zhou et al., 2017
	*4	352A > G	rs55951658	<0.001–0.006	0	0			Werk and Cascorbi, 2014; Zhou et al., 2017
	*5	653C > G	rs55901263	<0.001–0.006	0	0			Werk and Cascorbi, 2014; Zhou et al., 2017
	*6	830_831insA	rs4646438	<0.001–0.005	0	0			Werk and Cascorbi, 2014; Zhou et al., 2017
	*7	167G > A	rs56324128	0	0.001–0.014	0			Werk and Cascorbi, 2014; Zhou et al., 2017
	*8	389G > A	rs72552799	<0.001–0	0.003–0.014	0–0.04			Werk and Cascorbi, 2014; Jarrar et al., 2016; Zhou et al., 2017
	*9	508G > A	rs72552798	0	0–0.002	0			Werk and Cascorbi, 2014; Zhou et al., 2017
	*10	520G > C or A	rs4986908	<0.001–0.001; 0.012 for A	<0.001; 0.003 for C	0.002			Werk and Cascorbi, 2014; Zhou et al., 2017
	*11	1088C > T	rs67784355	<0.001–0.002	0–0.003	<0.001		gMAF 0.001	Werk and Cascorbi, 2014; Zhou et al., 2017
	*12	1117C > T	rs12721629	0-<0.001	0–0.004	0.003–0.007		gMAF 0.001	Werk and Cascorbi, 2014; Zhou et al., 2017
	*13	1247C > T	rs4986909	0–0.012	0–0.004	0–0.021			Werk and Cascorbi, 2014; Zhou et al., 2017
	*14	44T > C	rs12721634					gMAF 0–0.003	Werk and Cascorbi, 2014
	*15	485 G > A	rs4986907	0-<0.001	0	0–0.042		gMAF 0.014	Werk and Cascorbi, 2014; Zhou et al., 2017
	*16	554C > G	rs12721627	0–0.014	0	0		gMAF 0.005	Werk and Cascorbi, 2014; Zhou et al., 2017
	*17	566T > C	rs4987161	0	0–0.020	0			Werk and Cascorbi, 2014; Preissner et al., 2013
	*18	878 T > C	rs28371759	0–0.028	0	0.002		gMAF 0.01	Werk and Cascorbi, 2014; Zhou et al., 2017
	*19	1399C > T	rs4986913	0–0.012	0–0.022	0			Werk and Cascorbi, 2014; Zhou et al., 2017
	*20	1461_1462insA	rs67666821	0	0-<0.006	<0.001			Werk and Cascorbi, 2014; Zhou et al., 2017
	*21	956A > G	rs201821708	0.005					Werk and Cascorbi, 2014
	*22	15389C > T	rs35599367	0–0.043	0.025–0.083	<0.001–0.043		gMAF 0.021	Werk and Cascorbi, 2014; Zanger and Schwaby, 2013; Zhou et al., 2017
	*23	484C > T	rs57409622					gMAF 1	Werk and Cascorbi, 2014
	*24	600A > T	rs113667357					gMAF 1	Werk and Cascorbi, 2014
	*1B	UTR −392A > G	rs2740574	0	0.030–0.050	0.50–0.82	Hispanics 0.064–0.146	gMAF 0.20	Keshava et al., 2004; Werk and Cascorbi, 2014; Zanger and Schwaby, 2013
CYP3A5	*2	1289C > A	rs28365083	0	0.001–0.10	<0.001			Jarrar et al., 2016; Zhou et al., 2017
	*3	6986A > G	rs776746	0.66–0.78	0.77–0.96	0.12–0.50		gMAF 0.312	Jarrar et al., 2016; Zanger and Schwaby, 2013; Preissner et al., 2013; Naidoo et al., 2014; Zhou et al., 2017
	*4	14665A > G	rs56411402	0-<0.01	0	0			Park et al., 2014; Zhou et al., 2017
	*5	12952T > C		<0.001–0.007	0	0			Park et al., 2014; Zhou et al., 2017
	*6	14,690 G > A	rs10264272	0	0–0.3	0.12–0.25		gMAF 0.045	Park et al., 2014; Zanger and Schwaby, 2013; Naidoo et al., 2014; Zhou et al., 2017
	*7	27131_27132insT	rs41303343	0-<0.001	0	0.1			Park et al., 2014; Naidoo et al., 2014; Zhou et al., 2017
	*8	3699C > T	rs55817950	0-<0.001	0	0–0.04			

Table 8 (*Continued*)

Gene	Polymorphic allele	Change	SNP rs	MAF[a] - Asian	MAF[a] - Caucasian	MAF[a] - African/African American	MAF[a] - other	additional notes	References
									Park et al., 2014; Zhou et al., 2017
	*9	19,386 G > A	rs28383479	0.02					Park et al., 2014
	*10	6986A > G; 29,753 T > C; 31611C > T			0.02				Park et al., 2014; Preissner et al., 2013
CYP3A7	*2	26041C > G		0.28	0.08	0.62	0.17 Saudi Arabian		Preissner et al., 2013
	*3						0.003 in koreans		Lee et al., 2010
	*1C		at least 5 SNPs		0.03	0.06			Dapia et al., 2017

[a] = minor allele frequency.

individuals of European or African origin (Zanger and Schwaby, 2013; Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017). *CYP2A6*9* is a SNP at the promoter region (−48T > G) that leads to a decreased enzyme expression, reported in 0.144−0.27 of Asians while its frequency varies from 0.04 to 0.12 in other ethnic groups (Preissner et al., 2013; Zanger and Schwaby, 2013; Naidoo et al., 2014; Murphy, 2017; Zhou et al., 2017). *CYP2A6*17* was reported in Africans/African Americans at a frequency up to ∼ 0.11, whereas it is absent in Caucasians and Asians (Naidoo et al., 2014; Murphy, 2017; López-Flores et al., 2017; Zhou et al., 2017). Distributions of CYP2A6 alleles across world populations are listed in Table 6.

3.3.3. Allelic frequency of CYP2D6 polymorphisms

The ELS search highlighted the widely polymorphic nature of CYP2D6, owing to the existence of more than 300 different allelic variants and a series of subvariants, giving rise to decreased, normal, or increased enzyme activities amongst individuals and world populations. While certain polymorphisms show similar frequencies across world populations, others have been detected sporadically in specific populations or their prevalences widely differ amongst world populations. Some polymorphisms (including presence of multiple copies of the wild type gene) are fully functional, some exhibit reduced functionality and some are null and therefore do not affect CYP2D6 activity. This range of polymorphisms and functional changes can explain the variability of enzymatic activities i.e from no/low activity in PMs, to intermediate, extensive and ultra-rapid metabolisers. Table 7 displays CYP2D6 genetic polymorphisms within at least one of the major world population. In this subset, the *CYP2D6*4* (1846 G > A) is the most frequent CYP2D6 allele in Caucasians, where it is present at a frequency of 0.15−0.25, and leads to the absence of the protein in the liver, while it has a minor frequency in Asians (0.004−0.116) and in Africans/African Americans (0.034−0.119) (Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017). Another commonly detected allelic variant with a normal function is the *CYP2D6*2* (2850C > T, 4180 G > C and others) reported in 0.131−0.362 of Asians, in 0.141−0.267 of Africans/African Americans and in 0.041−0.343 of Caucasians (Gaedigk et al., 2017; Zhou et al., 2017). In contrast, the *CYP2D6*5* gene deletion allele is present at a frequency of 0.020 to 0.062 in all the major world population groups (Gaedigk et al., 2017; Preissner et al., 2013; Zhou et al., 2017) whereas about 0.065 to 0.70 of Asians are carriers of the *CYP2D6*10* (100C > T) decreased-function allele, while it has a lower prevalence in Caucasians (0.024−0.080) and in Africans/African Americans (0.032−0.120) (Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017). The frequency of the partially defective *CYP2D6*17* variant (1023C > T; 2850C > T) occurs in up to 24 % of Africans/African Americans, whereas it is rare in the other world populations (Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017). CYP2D6 duplications due to unequal crossing have been also identified in combination with the *29, *41, *45 alleles and the overall frequency of the gene duplications in all world populations is variable but generally ≤ 0.10 (Gaedigk et al., 2017; Preissner et al., 2013; Zanger and Schwaby, 2013; Zhou et al., 2017). Frequencies at the CYP2D6 locus in human world popualtions are illustrated in Table 7.

3.3.4. Allelic frequency of CYP3A4, 3A5, 3A7 polymorphisms

Genes coding for the CYP3A subfamily, namely *CYP3A4*, *CYP3A5* and *CYP3A7* are clustered on chromosome 7. At least 40 allelic variants have been described in the CYP3A4 gene which is, at the protein level, by far the major CYP isoform expressed in human hepatocytes (Jarrar et al., 2016). *CYP3A4*1B* is considered the most common SNP in CYP3A4 and the most extensively studied. However, its significance is still not yet clear due to contrasting results on its impact on enzymatic activity. In Africans/African American, frequency of *CYP3A4*1B* is in the range of 0.50−0.82, whereas it is absent in Japanese and Chinese populations and has a low frequency (0.030−0.050) in Caucasians (Keshava et al., 2004; Werk and Cascorbi, 2014; Zanger and Schwaby, 2013). However, amongst all the other CYP3A4 variants currently identified, almost all fall into the category of rare polymorphisms, showing a frequency between 0.01 and 0.03, or mutations, having a frequency < 0.01. Of these variants, however numerous, the *CYP3A4*2* (644 T > C) shows a frequency between 0.011 and 0.027 in Caucasians and is absent in other world populations (Werk and Cascorbi, 2014; Preissner et al., 2013; Zhou et al., 2017). *CYP3A4* 3* (1334 T > C) has been found only in Caucasians (0−0,04) and in Africans/African Americans (0−0,03) (Werk and Cascorbi, 2014; Zhou et al., 2017) and *CYP3A4*15* (485 G > A) has been only described in Africans/African Americans with a maximum frequency of 0.042 (Werk and Cascorbi, 2014; Zhou et al., 2017). *CYP3A4*18* (878 T > C) was described in 0.028 of Asians and in 0.002 of Africans/African Americans (Werk and Cascorbi, 2014; Zhou et al., 2017).

Finally, the intronic variant *CYP3A4*22* (15389C > T) has been reported with a frequency of 0.025−0.083 in Caucasians and up to 0.043 both in Asians and in Africans/African Americans (Werk and Cascorbi, 2014; Zanger and Schwaby, 2013; Zhou et al., 2017). The relevance of interindividual differences for the *CYP3A4*22* variant has been demonstrated in relation to the pharmacokinetics of a number of drugs used in cancer therapies. In addition, it has been associated with reduced clearances of tacrolimus and cyclosporin A in renal transplant patients. On the other hand, differences in kinetics may also result in more efficient therapeutic responses as indicated by treatment with fluticasone propionate or lipid-

lowering agents, expecially when administered to patients which do not express CYP3A5 (Werk and Cascorbi, 2014).

CYP3A5 is expressed in extrahepatic tissues with more than 25 allelic variants (Jarrar et al., 2016). The *CYP3A5*3* genetic polymorphism (intronic transition 6986A > G) is the most common SNP, leading to the loss of CYP3A5 activity due to the disruption of the correct splicing of CYP3A5 transcripts. It has been reported in 0.77–0.96 of Caucasians, in 0.66–0.78 of Asians and in 0.12–0.50 of Africans/African Americans (Jarrar et al., 2016; Zanger and Schwaby, 2013; Preissner et al., 2013; Naidoo et al., 2014; Zhou et al., 2017). Variability in the frequency of *CYP3A5*3* across world populations is at the basis of the marked differences in the metabolism of drugs that are CYP3A5 substrates (Lamba et al., 2002). *CYP3A5*2* (1289C > A) is a coding SNP reported in 0.001–0.10 of Caucasians and in <0.001 of Africans/African Americans (Jarrar et al., 2016; Zhou et al., 2017). *CYP3A5*6* (14,690 G > A), *7 (27131_27132insT) and *8 (3699C > T) have been mostly found in Africans/African Americans, respectively at the frequencies of 0.12–0.25, 0.1, and 0–0.04 (Park et al., 2014; Zanger and Schwaby, 2013; Naidoo et al., 2014; Zhou et al., 2017). Apart from CYP3A5*3, the only genetic polymorphism reported in Asians with an incidence slightly >1% is the *CYP3A5* 9* (19,386 G > A) (Park et al., 2014).

CYP3A7 is the major hepatic CYP enzyme expressed in the foetus. Generally, its expression decreases and shifts to CYP3A4 after birth; however, it continues to be expressed at significant levels in some adult individuals carrying the *CYP3A7*1C* allele, influencing levels of circulating endogenous sex hormones and clinical outcome from various malignancies as lymphocytic leukemia, breast and lung cancer (Johnson et al., 2016). *CYP3A7*1C* allele has been reported to be defined by at least 5 SNPs, at a frequency of 0.03 in Caucasian populations and 0.06 in Africans/African Americans (Dapia et al., 2017). The most common genetic polymorphism was shown to be the *CYP3A7*2* (26041C > G), found in 0.62 of Africans/African Americans, in 0.28 of Asians and in 0.08 of Caucasians (Preissner et al., 2013), while the *CYP3A7*3* allele has been reported only in Koreans at the frequency of 0.003 (Dapia et al., 2017; Lee et al., 2010). Distributions of CYP3A4, 3A5 and 3A7 alleles across world populations are summarised in Table 8.

4. Conclusions and future perspectives

The manuscript presents the results of large extensive literature searches from the peer-reviewed scientific literature and the creation of the OpenCYP database which will be public (DOI: 10.5281/zenodo.5031737) (Supplementary material A and B): an open source database providing data for human variability in the baseline activities and frequencies of polymophisms for CYP1A2, CYP2A6, CYP2D6, CYP3A4/3A5 and CYP3A7 across world populations. This database presents a collection of quantitative data on CYP enzymatic activities extracted from in vivo studies conducted on ethnically heterogeneous populations. This peculiarity represents its added value with respect to what is currently avaialble, given that the majority of search engines that are freely accessible generally provide only qualitative information on enzymatic activities for specific substrates or, in most cases, indicate only the coherent reference literature.

As relevant examples, reliable and high quality databases addressing data collection of human genotypes and phenotypes are already available, as the central repositories PharmGKB (http://www.pharmgkb.org) and PharmVar Consortium (https://www.pharmvar.org/). They both represent an important open-access source of information, especially directed to clinical applications, considering different responses to drugs based on genetic variability.

The former, structured around four specific entities: variants, genes, drugs and diseases, provides clinical information, including dosing guidelines and drug labels, potentially clinically relevant gene-drug association, and also information on how genetic variants can be involved in the PK or PD pathways of a specific therapeutic drug. Analogously the Pharmacogene Variation (PharmVar) Consortium reports pharmacogene (PGx) variation with a focus on haplotype structure and allelic variation, supporting the interpretation of pharmacogenetic test results to guide precision medicine.

Besides clinical applications, having available data on human variability on the activity of phase I, phase II enzymes and transporters involved in ADME processes, represents critical data needs to move towards the development of PBK and *in silico* tools that provide reliable estimates when used in the risk assessment process. Having information on the isoform-specific biotransformation of a chemical obtained *in vitro*, the possibility to integrate data on human variability of the active enzyme (as the ones included in the OpenCYP database) could allow for improvement and testing the prediction power of PBK generic models.

The OpenCYP database, as an open source database, aims to provide CYP-specific enzymatic activity for healthy adults and their genetic polymorphism frequencies across world populations. In addition, additional efforts have supported the generation of human variability distributions for a range of Phase I, Phase II and transporters using human *in vivo* kinetic parameters and bayesian meta-analyses (e.g. CYP3A4 and paraoxonoase-1, carboxylesterases, CYP2D6) (Darney et al., 2019, 2020a,b, 2021-submitted). For these enzymes, pathway-related uncertainty factors were derived for populations of different geographical ancestry (CYP3A4, CYP2D6, paraoxonoase-1 and carboxyl esterases) and inter-phenotypic differences (paraoxonoase-1) associated with genetic polymorphisms (Darney et al., 2019, 2020a,b, 2021; Di Consiglio et al., 2021).

While compiling the OpenCYP EFSA database, we conclude that, despite the high number of collected papers, discrepancies in the reporting of the age and demographic characteristics of enrolled individuals were noted across the datasets and constituted a challenge to perform a full statistical analysis of CYP-specific activities and variabilities in relation to geographical origin. In addition, when the same probe substrate was used, the lack of standardisation in the experimental conditions (dosing, time of sampling, matrix collected) hampered the possibility to perform a full statistical analysis. Data investigating age groups other than adults such as the elderly, children and neonates were very limited and fragmented constituting an additional important data gap. Moreover, few papers reported both phenotyping and genotyping results. These considerations highlight the need to use standardised methodologies in the experimental design and reporting, whenever data on enzymatic activity in humans are investigated within a specific population.

The use of MR measured in urine or plasma was considered an adequate metrics for possible comparison and allowed to derive variability of CYP enzymatic activities within healthy individuals.

When data were sub-divided according to similar testing conditions, our analysis indicates that variability was quite limited for CYP1A2 (assessed with caffeine as a probe substrate) and a symmetrical distribution of metabolic activities values was observed. CYP3A4 activity showed also a limited variability, when assessed by using the urinary DEX/3-MM ratio. Conversely, when the urinary 6β-hydroxycortisol to cortisol ratio was used as index of CYP3A4 activity, large interindividual/interethnical differences were shown. This could be the result of the overall variability due to both CYP3A4 and to the genetic polymorphism of transporters responsible for the renal excretion (clearance) of cortisol. Genetic variants in kidney membrane carriers can cause considerable interindividual variation in physiological processes as well as in pharmacotherapy (Tracy et al., 2016; Yin et al., 2004), and

specifically 6β-hydroxycortisol/cortisol ratio may be affected when the renal organic anion transporter 3 (OAT3) function is altered, since 6β-hydroxycortisol is a substrate of OAT3As (Imamura et al., 2014). As a consequence, the use of the 6β-hydroxycortisol/cortisol ratio does not reflect only the CYP3A4 variability. Another possible additional explanation could be limited comparability of the populations included into the collected studies. The highest variability was observed for CYP2D6, after oral dosing of DEX, for which genetic polymorphisms are presumed to act as a significant additional source of variability.

In conclusion, OpenCYP constitutes a useful collection of baseline isoform-specific activities in adults and frequencies of polymophisms for selected P-450 isoforms (CYP1A2, CYP2A6, CYP2D6, CYP3A4/3A5 and CYP3A7) across world populations. It is proposed to extend the database for other CYP isoforms and phase I enzymes (CYP2C9, CYP2C19, ADH, paraoxonases, esterases) as well as phase II enzymes (UGTs, GSTs, sulphotransferases, methyltransferases, glycine conjugation etc) and transporters (P-glycoprotein, OATPs, OATs etc), while including absolute expression levels of the isoforms using proteomic analyses. It is foreseen that the integration of databases such as OpenCYP with the available large databases on human *in vivo* variability in markers of acute and chronic exposure, as well as absolute expression levels of the isoforms from recent proteomic experiments, would support the further development of QIVIVE and PBK models to be applied in chemical risk assessment (Darney et al., 2019, 2020a,b, 2021; Kasteel et al., 2020a, b; Buratti et al., 2021; Di Consiglio et al., 2021). Future work also includes testing these models with case studies for food and feed relevant compounds integrating *in vitro* kinetic data, variability distributions and frequencies of polymorphisms in virtual populations for model calibration and validation and ultimately implement their routine use in food and feed safety.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This project was supported by the European Food Safety Authority (EFSA) under the grant agreement no. GA/EFSA/SCER/2015/01. Views expressed in this paper are the authors only and do not represent the views of the European Food Safety Authority.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.toxlet.2021.07.019.

References

Al Koudsi, N., Ahluwalia, J.S., Lin, S.K., Sellers, E.M., Tyndale, R.F., 2009. A novel CYP2A6 allele (CYP2A6*35) resulting in an amino-acid substitution (Asn438Tyr) is associated with lower CYP2A6 activity in vivo'. Pharm. J. 9 (4), 274–282. doi: http://dx.doi.org/10.1038/tpj.2009.11.

Al-Jenoobi, F.I., Al-Thukair, A.A., Alam, M.A., Abbas, F.A., Al-Mohizea, A.M., Alkharfy, K. M., Al-Suwayeh, S.A., 2014a. Effect of garden cress seeds powder and its alcoholic extract on the metabolic activity of CYP2D6 and CYP3A4. Evid. Complement. Alternat. Med. 2014, 634592 doi:http://dx.doi.org/10.1155/2014/634592.

Al-Jenoobi, F.I., Al-Thukair, A.A., Alam, M.A., Abbas, F.A., Al-Mohizea, A.M., Alkharfy, K.M., Al-Suwayeh, S.A., 2014b. Modulation of CYP2D6 and CYP3A4 metabolic activities by Ferula asafetida resin. Saudi Pharm. J. 22 (6), 564–569. doi:http://dx.doi.org/10.1016/j.jsps.2014.03.004.

Al-Jenoobi, F.I., Al-Thukair, A.A., Alam, M.A., Abbas, F.A., Al-Mohizea, A.M., Alkharfy, K.M., Al-Suwayeh, S.A., 2015. Effect of curcuma longa on CYP2D6- and CYP3A4-mediated metabolism of dextromethorphan in human liver microsomes and healthy human subjects. Eur. J. Drug Metab. Pharmacokinet. 40 (1), 61–66. doi: http://dx.doi.org/10.1007/s13318-014-0180-2.

Baas, J., Augustine, S., Marques, G.M., Dorne, J.L., 2018. Dynamic energy budget models in ecological risk assessment: from principles to applications. Sci. Total Environ. 628-629, 249–260. doi:http://dx.doi.org/10.1016/j.scitotenv.2018.02.058.

Begas, E., Tsioutsiouliti, A., Kouvaras, E., Haroutounian, S.A., Kasiotis, K.M., Kouretas, D., Asprodini, E., 2017. 'Effects of peppermint tea consumption on the activities of CYP1A2, CYP2A6, xanthine oxidase, N-acetyltranferase-2 and UDP-glucuronosyltransferases-1A1/1A6 in healthy volunteers', 2017. Food Chem. Toxicol. 100, 80–89. doi:http://dx.doi.org/10.1016/j.fct.2016.12.021.

Benet, L.Z., 2005. There are no useful CYP3A probes that quantitatively predict the in vivo kinetics of other CYP3A substrates and no expectation that one will be found. Mol. Interv. 5 (2), 79–83. doi:http://dx.doi.org/10.1124/mi.5.2.5.

Benfenati, E., Carnesecchi, E., Roncaglioni, A., Baldin, R., Ceriani, L., Ciacci, A., Kovarich, S., Sartori, L., Mostrag, A., Magdziarz, T., Yang, C., 2020. Maintenance, update and futher development of EFSA's chemical hazards: OpenFoodTox 2.0. EFSA Supporting Pub. 2020, 36. doi:http://dx.doi.org/10.2903/sp.efsa.2020.EN-1822 EN-1822.

Bessems, J.G., Loizou, G., Krishnan, K., Clewell 3rd, H.J., Bernasconi, C., Bois, F., Coecke, S., Collnot, E.M., Diembeck, W., Farcal, L.R., Geraets, L., Gundert-Remy, U., Kramer, N., Küsters, G., Leite, S.B., Pelkonen, O.R., Schröder, K., Testai, E., Wilk-Zasadna, I., Zaldívar-Comenges, J.M., 2014. PBTK modelling platforms and parameter estimation tools to enable animal-free risk assessment: recommendations from a joint EPAA–EURL ECVAM ADME workshop. Regul. Toxicol Pharmacol. RTP 68 (1), 119–139. doi:http://dx.doi.org/10.1016/j.yrtph.2013.11.008.

Buratti, F.M., Darney, K., Vichi, S., Turco, L., Di Consiglio, E., Lautz, L.S., Béchaux, C., Dorne, J.C.M., Testai, E., 2021. Human variability in glutathione-S-transferase activities, tissue distribution and major polymorphic variants: meta-analysis and implication for chemical risk assessment. Toxicol. Lett. 337, 78–90. doi: http://dx.doi.org/10.1016/j.toxlet.2020.11.007.

Chen, Y., Xiao, C.Q., He, Y.J., Chen, B.L., Wang, G., Zhou, G., Zhang, W., Tan, Z.R., Cao, S., Wang, L.P., Zhou, H.H., 2011. Genistein alters caffeine exposure in healthy female volunteers. Eur. J. Clin. Pharmacol. 67 (4), 347–353. doi:http://dx.doi.org/10.1007/s00228-010-0964-5.

Dapia, I., Tabakov, A., Espinosa Román, L., Díaz, L., Arias Lajara, P., Melgosa, M., Melgar, A., Fernández, C., Herranz Estellés, A., Lapunzina, P., Borobia, A.M., Carcas, A.J., 2017. Effect of the CYP3A5, CYP3A4, CYP3A7, ABCB1, POR and NR1I2 Genes in the Pharmacokinetics of Tacrolimus in a Pediatric Cohort with Stable Serum Concentrations after Renal Transplantation: Study Protocol. doi:http://dx.doi.org/10.24217/2530-4984.17v1.00005.

Darney, K., Testai, E., Buratti, F.M., Di Consiglio, E., Kasteel, E.E.J., Kramer, N., Turco, L., Vichi, S., Roudot, A.C., Dorne, J.L.C.M., Béchaux, C., 2019. Inter-ethnic differences in CYP3A4 metabolism: a Bayesian meta-analysis for the refinement of uncertainty factors in chemical risk assessment. Comput. Toxicol. 12 (2019) 100092 doi:http://dx.doi.org/10.1016/j.comtox.2019.100092.

Darney, K., Kasteel, E.E.J., Buratti, F.M., Turco, L., Vichi, S., Bechaux, C., Roudot, A.C., Kramer, N., Testai, E., Dorne, J.L.C.M., Di Consiglio, E., Lautz, L.S., 2020a. Bayesian meta-analysis of interphenotypic differences in human serum paraoxonase-1 activity for chemical risk assessment. Environ. Int. 138, 105609 doi:http://dx.doi.org/10.1016/j.envint.2020.105609.

Darney, K., Turco, L., Buratti, F.M., Di Consiglio, E., Vichi, S., Roudot, A.C., Béchaux, C., Testai, E., Dorne, J., Lautz, L.S., 2020b. Human variability in influx and efflux transporters in relation to uncertainty factors for chemical risk assessment. Food Chem. Toxicol. 140, 111305 doi:http://dx.doi.org/10.1016/j.fct.2020.111305.

Darney, K., Lautz, L.S., Béchaux, C., Wiecek, W., Testai, E., Amzal, B., Dorne, J.L.C.M., 2021. Human variability in polymorphic CYP2D6 metabolism: implications for the risk assessment of chemicals in food and emerging designer drugs. Environ. Int. 156, 106760 doi:http://dx.doi.org/10.1016/j.envint.2021.106760.

Di Consiglio, E., Darney, K., Buratti, F.M., Turco, L., Vichi, S., Testai, E., Lautz, L.S., Dorne, J. L.C.M., 2021. Human variability in carboxylesterases and carboxylesterase-related uncertainty factors for chemical risk assessment. Toxicol. Lett. 2021 doi:http://dx.doi.org/10.1016/j.toxlet.2021.07.005 Published on line July.

Dorne, J.L.C.M., Walton, K., Renwick, A.G., 2001. Uncertainty factors for chemical risk assessment: human variability in the pharmacokinetics of CYP1A2 probe substrates. Food Chem. Toxicol. 39 (7), 681–696. doi:http://dx.doi.org/10.1016/s0278-6915(01)00005-9.

Dorne, J.L.C.M., Walton, K., Renwick, A.G., 2003. Human variability in CYP3A4 metabolism and CYP3A4-related uncertainty factors for risk assessment. Food Chem. Toxicol. 41 (2), 201–224. doi:http://dx.doi.org/10.1016/S0278-6915(02)00209-0.

Dorne, J.L.C.M., Walton, K., Renwick, A.G., 2004. Human variability for metabolic pathways with limited data (CYP2A6, CYP2C9, CYP2E1, ADH, esterases, glycine and sulphate conjugation). Food Chem. Toxicol. 42 (3), 397–421. doi:http://dx.doi.org/10.1016/j.fct.2003.10.003.

Dorne, J.L.C.M., Richardson, J., Kass, Georges, Georgiadis, G., Monguidi, M., Pasinato, L., Cappe, S., Verhagen, H., Robinson, T., 2017. Editorial: openfoodtox: EFSA's open source toxicological database on chemical hazards in food and feed. EFSA J. 15 (1), 3. doi:http://dx.doi.org/10.2903/j.efsa.2017.e15011 2017 e15011.

Dorne, J.L.C.M., Richardson, J., Livaniou, A., Carnesecchi, E., Ceriani, L., Baldin, R., Kovarich, S., Pavan, M., Saouter, E., Biganzoli, F., Pasinato, L., Zare Jeddi, M., Robinson, T.P., Kass, G.E.N., Liem, A.K.D., Toropov, A.A., Toropova, A.P., Yang, C., Tarkhov, A., Georgiadis, N., Di Nicola, M.R., Mostrag, A., Verhagen, H., Roncaglioni, A., Benfenati, E., Bassan, A., 2021. EFSA's OpenFoodTox: an open source toxicological database on chemicals in food and feed and its future developments. Environ. Int. 146, 106293 doi:http://dx.doi.org/10.1016/j.envint.2020.106293.

EFSA Guidance, 2010. Application of systematic review methodology to food and feed safety assessments to support decision making. EFSA J. 8 (6), 1637. doi:http://dx.doi.org/10.2903/j.efsa.2010.1637 2010.

EFSA Scientific Report, 2014. Modern methodologies and tools for human hazard assessment of chemicals. EFSA J. 12 (4), 3638. doi:http://dx.doi.org/10.2903/j.efsa.2014.3638 2014.

Ekroos, M., Sjogren, T., 2006. Structural basis for ligand promiscuity in cytochrome P450 3A4. Proc. Natl. Acad. Sci. U. S. A. 103 (37), 13682–13687. doi:http://dx.doi.org/10.1073/pnas.0603236103.

El Desoky, E.S., Mohamed, H.O., Farghaly, W.M.A., Hamed, S.A., Hedaya, M.A., Siest, J. P., 2005. Study of urinary 6β-hydroxycortisol/cortisol ratio in spot urine sample as a biomarker of 3A4 enzyme activity in healthy and epileptic subjects of Egyptian population. Pharmacol. Res. 51 (6), 575–580. doi:http://dx.doi.org/10.1016/j.phrs.2005.02.014.

Evans, W.E., Relling, M.V., 1991. Concordance of P450 2D6 (debrisoquine hydroxylase) phenotype and genotype: inability of dextromethorphan metabolic ratio to discriminate reliably heterozygous and homozygous extensive metabolizers. Pharmacogenetics 1 (3), 143–148.

Fukami, T., Nakajima, M., Yoshida, R., Tsuchiya, Y., Fujiki, Y., Katoh, M., McLeod, H.L., Yokoi, T., 2004. A novel polymorphism of human CYP2A6 gene CTP2A6*17 has an amino acid substitution (V365M) that decreases enzymatic activity in vitro and in vivo. Clin. Pharmacol. Ther. 76 (6), 519–527. doi:http://dx.doi.org/10.1016/j.clpt.2004.08.014.

Funck-Brentano, C., Thomas, G., Jacqz-Aigrain, E., Poirier, J.M., Simon, T., Béréziat, G., Jaillon, P., 1992. Polymorphism of dextromethorphan metabolism: relationships between phenotype, genotype and response to the administration of encainide in humans. J. Pharmacol. Exp. Ther. 263 (2), 780–786.

Gaedigk, A., Sangkuhl, K., Whirl-Carrillo, M., Klein, T., Leeder, J.S., 2017. Prediction of CYP2D6 phenotype from genotype across world populations. Genet. Med. 19 (1), 69–76. doi:http://dx.doi.org/10.1038/gim.2016.80.

Grech, A., Tebby, C., Brochot, C., Bois, F.Y., Bado-Nilles, A., Dorne, J.L.C.M., Quignot, N., Beaudouin, R., 2019. Generic physiologically-based toxicokinetic modelling for fish: integration of environmental factors and species variability. Sci. Total Environ. 651 (1) doi:http://dx.doi.org/10.1016/j.scitotenv.2018.09.163.

Hakooz, N., Hamdan, I., 2007. 'Effects of dietary broccoli on human in vivo caffeine metabolism: a pilot study on a group of Jordanian volunteers. Curr. Drug Metab. 8 (1), 9–15. doi:http://dx.doi.org/10.2174/138920007779315080.

Hardy, A., Dorne, J.L.C.M., Aiassa, E., Alexander, J., Bottex, B., Chaudhry, Q., Germini, A., Nørrung, B., Schlatter, J., Verloo, D., Robinson, T., 2015. Editorial: increasing robustness, transparency and openness of scientific assessments. EFSA J. 13 (3), 3. doi:http://dx.doi.org/10.2903/j.efsa.2015.e13031 2015 e13031.

Imamura, Y., Tsuruya, Y., Damme, K., Heer, D., Kumagai, Y., Maeda, K., Murayama, N., Okudaira, N., Kurihara, A., Izumi, T., Sugiyama, Y., Kusuhara, H., 2014. 6β-Hydroxycortisol is an endogenous probe for evaluation of drug-drug interactions involving a multispecific renal organic anion transporter, OAT3/SLC22A8, in healthy subjects. Drug Metab. Dispos. 42, 685–694.

Imène, A., Maurice, A.J., Arij, M., Sofia, P., Saad, S., 2015. Breast cancer association with CYP1A2 activity and gene polymorphisms - A preliminary case-control study in Tunisia. Asian Pac. J. Cancer Prev. 16 (8), 3559–3563. doi:http://dx.doi.org/10.7314/apjcp.2015.16.8.3559.

Ingelman-Sundberg, M., 2004. Human drug metabolising cytochrome P450 enzymes: properties and polymorphisms. Naunyn Schmiedebergs Arch. Pharmacol. 369 (1), 89–104. doi:http://dx.doi.org/10.1007/s00210-003-0819-z.

Jarrar, M., Behl, S., Manyam, G., Ganah, H., Nazir, M., Nasab, R., Moustafa, K., 2016. Cytochrome allelic variants and clopidogrel metabolism in cardiovascular diseases therapy. Mol. Biol. Rep. 43 (6), 473–484. doi:http://dx.doi.org/10.1007/s11033-016-3983-1.

Johnson, N., De Ieso, P., Migliorini, G., Orr, N., Broderick, P., Catovsky, D., Matakidou, A., Eisen, T., Goldsmith, C., Dudbridge, F., Peto, J., Dos-Santos-Silva, I., Ashworth, A., Ross, G., Houlston, R.S., Fletcher, O., 2016. Cytochrome P450 allele CYP3A7*1C associates with adverse outcomes in chronic lymphocytic leukemia, breast, and lung cancer. Cancer Res. 76 (6), 1485–1493. doi:http://dx.doi.org/10.1158/0008-5472.CAN-15-1410.

Kasteel, E.E.J., Darney, K., Kramer, N.I., Dorne, J.L.C.M., Lautz, L.S., 2020a. Human variability in isoform-specific UDP-glucuronosyltransferases metabolism: from markers of acute and chronic exposure to polymorphisms and uncertainty factors. Arch. Toxicol. 94 (8), 2637–2661. doi:http://dx.doi.org/10.1007/s00204-020-02765-8 2020.

Kasteel, E.E.J., Nijmeijer, S.M., Darney, K., Lautz, L.S., Dorne, J.L.C.M., Kramer, N.I., Westerink, R.H.S., 2020b. Acetylcholinesterase inhibition in electric eel and human donor blood: an in vitro approach to investigate interspecies differences and human variability in toxicodynamics. Arch. Toxicol. 94 (12), 4055–4065. doi:http://dx.doi.org/10.1007/s00204-020-02927-8.

Keshava, C., McCanlies, E.C., Weston, A., 2004. CYP3A4 polymorphisms–potential risk factors for breast and prostate cancer: a HuGE review. Am. J. Epidemiol. 160 (9), 825–841. doi:http://dx.doi.org/10.1093/aje/kwh294.

Klimisch, H.J., Andreae, M., Tillmann, U.A., 1997. A systematic approach for evaluating the quality of experimental toxicological and ecotoxicological data. Regul. Toxicol. Pharmacol. 25 (1), 1–5. doi:http://dx.doi.org/10.1006/rtph.1996.1076.

Koonrungsesomboon, N., Khatsri, R., Wongchompoo, P., Teekachunhatean, S., 2018. The impact of genetic polymorphisms on CYP1A2 activity in humans: a systematic review and meta-analysis. Pharmacogenomics J. 18 (6), 760–768. doi:http://dx.doi.org/10.1038/s41397-017-0011-3.

Lamba, J.K., Lin, Y.S., Schuetz, E.G., Thummel, K.E., 2002. Genetic contribution to variable human CYP3A-mediated metabolism. Adv. Drug Deliv. Rev. 54 (10), 1271–1294. doi:http://dx.doi.org/10.1016/s0169-409x(02)00066-2.

Lauschke, Volker M, Ingelman-Sundberg, Magnus, 2016. The Importance of Patient-Specific Factors for Hepatic Drug Response and Toxicity. Int J Mol Sci 17 (10), 1714.

Lautz, L.S., Dorne, J.L.C.M., Oldenkamp, R., Hendriks, A.J., Ragas, A.M.J., 2020a. Generic physiologically based kinetic modelling for farm animals: part I. Data collection of physiological parameters in swine, cattle and sheep. Toxicol. Lett. 319, 95–101. doi:http://dx.doi.org/10.1016/j.toxlet.2019.10.021.

Lautz, L.S., Hoeks, S., Oldenkamp, R., Hendriks, A.J., Dorne, J.L.C.M., Ragas, A.M.J., 2020b. Generic physiologically based kinetic modelling for farm animals: part II. Predicting tissue concentrations of chemicals in swine, cattle, and sheep. Toxicol. Lett. 318, 50–56. doi:http://dx.doi.org/10.1016/j.toxlet.2019.10.008.

Lautz, L.S., Nebbia, C., Hoeks, S., Oldenkamp, R., Hendriks, A.J., Ragas, A.M.J., Dorne, J. L.C.M., 2020c. An open source physiologically based kinetic model for the chicken (Gallus gallus domesticus): calibration and validation for the prediction residues in tissues and eggs. Environ. Int. 136, 105488 doi:http://dx.doi.org/10.1016/j.envint.2020.105488.

Lee, S.S., Jung, H.J., Park, J.S., Cha, I.J., Cho, D.Y., Shin, J.G., 2010. Identification of a null allele of cytochrome P450 3A7: CYP3A7 polymorphism in a Korean population. Mol. Biol. Rep. 37 (1), 213–217. doi:http://dx.doi.org/10.1007/s11033-009-9608-1.

Lin, Y., Anderson, G.D., Kantor, E., Ojemann, L.M., Wilensky, A.J., 1999. Differences in the urinary excretion of 6-β-hydroxycortisol/cortisol between Asian and Caucasian women. J. Clin. Pharmacol. 39 (6), 578–582. doi:http://dx.doi.org/10.1177/00912709922008182.

López-Flores, L.A., Pérez-Rubio, G., Falfán-Valencia, R., 2017. Distribution of polymorphic variants of CYP2A6 and their involvement in nicotine addiction. EXCLI J. 16, 174–196. doi:http://dx.doi.org/10.17179/excli2016-847.

Markowitz, J.S., DeVane, C.L., Chavin, K.D., Taylor, R.M., Ruan, Y., Donovan, J.L., 2003a. Effects of garlic (Allium sativum L.) supplementation on cytochrome P450 2D6 and 3A4 activity in healthy volunteers. Clin. Pharmacol. Ther. 74 (2), 170–177. doi:http://dx.doi.org/10.1016/S0009-9236(03)00148-6.

Markowitz, J.S., Donovan, J.L., DeVane, C.L., Sipkes, L., Chavin, K.D., 2003b. Multiple-Dose Administration of Ginkgo biloba Did Not Affect Cytochrome P-450 2D6 or 3A4 Activity in Normal Volunteers. J. Clin. Psychopharmacol. 23 (6), 576–581. doi:http://dx.doi.org/10.1097/01.jcp.0000095340.32154.c6.

Markowitz, J.S., Donovan, J.L., DeVane, C.L., Taylor, R.M., Ruan, Y., Wang, J.S., Chavin, K.D., 2003c. Multiple doses of saw palmetto (*Serenoa repens*) did not alter cytochrome P450 2D6 and 3A4 activity in normal volunteers. Clin. Pharmacol. Ther. 74 (6), 536–542. doi:http://dx.doi.org/10.1016/j.clpt.2003.08.010.

Martinak, B., Bolis, R.A., Black, J.R., Fargason, R.E., Birur, B., 2017. Dextromethorphan in cough syrup: the poor man's psychosis. Psychopharmacol. Bull 47 (4), 59–63 15.

McCune, J.S., Lindley, C., Decker, J.L., Williamson, K.M., Meadowcroft, A.M., Graff, D., Sawyer, W.T., Blough, D.K., Pieper, J.A., 2001. Lack of gender differences and large intrasubject variability in cytochrome P450 activity measured by phenotyping with dextromethorphan. J. Clin. Pharmacol. 41 (7), 723–731. doi:http://dx.doi.org/10.1177/00912700122010627.

Murphy, S.E., 2017. Nicotine metabolism and smoking: ethnic differences in the role of P450 2A6. Chem. Res. Toxicol. 30 (1), 410–419. doi:http://dx.doi.org/10.1021/acs.chemrestox.6b00387.

Naidoo, P., Chetty, V.V., Chetty, M., 2014. Impact of CYP polymorphisms, geographical origin and sex differences in metabolism on dosing strategies: the case of efavirenz. Eur. J. Clin. Pharmacol. 70 (4), 379–389. doi:http://dx.doi.org/10.1007/s00228-013-1634-1.

Nowell, S., Sweeney, C., Hammons, G., Kadlubar, F.F., Lang, N.P., 2002. CYP2A6 activity determined by caffeine phenotyping: association with colorectal cancer risk. Cancer Epidemiol. Biomarkers Prev. 11 (4), 377–383.

O'Connell, M.B., Frye, R.F., Matzke, G.R., St. Peter, J.V., Willhite, L.A., Welch, M.R., Kowal, P., LaValleur, J., 2006. Effect of conjugated equine estrogens on oxidative metabolism in middle-aged and elderly postmenopausal women. J. Clin. Pharmacol. 46 (11), 1299–1307. doi:http://dx.doi.org/10.1177/0091270006292249.

Park, J.Y., Cha, Y.J., Kim, K.A., 2014. CYP3A5*3 polymorphism and its clinical implications and pharmacokinetic role. Transl Clin Pharmacol. 22 (1), 3–7. doi:http://dx.doi.org/10.12793/tcp.2014.22.1.3 2014.

Pelkonen, O., Maenpaa, J., Taavitsainen, P., Rautio, A., Raunio, H., 1998. Inhibition and induction of human cytochrome P450 (CYP) enzymes. Xenobiotica 28 (12), 1203–1253. doi:http://dx.doi.org/10.1080/004982598238886.

Pelkonen, O., Turpeinen, M., Hakkola, J., Honkakoski, P., Hukkanen, J., Raunio, H., 2008. Inhibition and induction of human cytochrome P450 enzymes: current status. Arch. Toxicol. 82 (10), 667–715. doi:http://dx.doi.org/10.1007/s00204-008-0332-8.

Pilgrim, J.L., Ruiz, Y., Gesteira, A., Cruz, R., Gerostamoulos, D., Carracedo, A., Drummer, O.H., 2012. Characterization of single nucleotide polymorphisms of cytochrome p450 in an Australian deceased sample. Curr. Drug Metab. 13 (5), 679–692. doi:http://dx.doi.org/10.2174/1389200211209050679.

Preissner, S.C., Hoffmann, M.F., Preissner, R., Dunkel, M., Gewiess, A., Preissner, S., 2013. Polymorphic cytochrome P450 enzymes (CYPs) and their role in personalized therapy. PLoS One 8 (12) e82562 doi:http://dx.doi.org/10.1371/journal.pone.0082562.

Quignot, N., Wiecek, W., Amzal, B., Dorne, J.L., 2018. The Yin-Yang of CYP3A4: a Bayesian meta-analysis to quantify inhibition and induction of CYP3A4 metabolism in humans and refine uncertainty factors for mixture risk assessment. Arch. Toxicol. 93 (1), 107–119. doi:http://dx.doi.org/10.1007/s00204-018-2325-6.

Quignot, N., Więcek, W., Lautz, L., Dorne, J.L., Amzal, B., 2021. Inter-phenotypic differences in CYP2C9 and CYP2C19 metabolism: bayesian meta-regression of

human population variability in kinetics and application in chemical risk assessment. Toxicol. Lett. 337, 111–120. doi:http://dx.doi.org/10.1016/j.toxlet.2020.11.016.

Ren, J., He, B.Z., Zhang, T.S., Lu, S.P., Yan, T., 2016. Meta-analysis of correlation between the CYP1A2-3860 G & A polymorphism and lung cancer risk. Genet. Mol. Res. 15 (2) doi:http://dx.doi.org/10.4238/gmr.15028353.

Saruwatari, J., Takashima, A., Yoshida, K., Soraoka, H., Ding, T.B., Uchiyashiki, Y., Tsuda, Y., Imamura, M., Oniki, K., Miyata, K., Nakagawa, K., 2014. Effects of seijo-bofu-to, a traditional Japanese herbal medicine containing furanocoumarin derivatives, on the drug-metabolizing enzyme activities in healthy male volunteers. Basic Clin. Pharmacol. Toxicol. 115 (4), 360–365. doi:http://dx.doi.org/10.1111/bcpt.12224.

Sim, S.C., Ingelman-Sundberg, M., 2011. Pharmacogenomic biomarkers: new tools in current and future drug therapy. Trends Pharmacol. Sci. 32 (2), 72–81. doi:http://dx.doi.org/10.1016/j.tips.2010.11.008.

Thorn, C.F., Aklillu, E., McDonagh, E.M., Klein, T.E., Altman, R.B., 2012. PharmGKB summary: caffeine pathway. Pharmacogenet. Genomics 22 (5), 389–395.

Tracy, T.S., Chaudhry, A.S., Prasad, B., Thummel, K.E., Schuetz, E.G., Zhong, X.B., Tien, Y.C., Jeong, H., Pan, X., Shireman, L.M., Tay-Sontheimer, J., Lin, Y.S., 2016. Interindividual variability in cytochrome P450-Mediated drug metabolism. Drug Metab. Dispos. 44 (3), 343–351. doi:http://dx.doi.org/10.1124/dmd.115.067900.

Tushar, T., Vinod, T., Rajan, S., Shashindran, C., Adithan, C., 2007. Effect of honey on CYP3A4, CYP2D6 and CYP2C19 enzyme activity in healthy human volunteers. Basic Clin. Pharmacol. Toxicol. 100 (4), 269–272. doi:http://dx.doi.org/10.1111/j.1742-7843.2006.00039.x.

Wenk, M., Todesco, L., Krähenbühl, S., 2004. Effect of St John's Wort on the activities of CYP1A2, CYP3A4, CYP2D6, N-acetyltransferase 2, and xanthine oxidase in healthy males and females. Br. J. Clin. Pharmacol. 57 (4), 495–499. doi:http://dx.doi.org/10.1111/j.1365-2125.2003.02049.x.

Werk, A.N., Cascorbi, I., 2014. Functional gene variants of CYP3A4. Clin. Pharmacol. Ther. 96 (3), 340–348. doi:http://dx.doi.org/10.1038/clpt.2014.129.

Xue, H., Lu, Y., Xue, Z., Lin, B., Chen, J., Tang, F., Huang, G., 2014. The effect of CYP1A1 and CYP1A2 polymorphisms on gastric cancer risk among different ethnicities: a systematic review and meta-analysis. Tumour Biol. 35 (5), 4741–4756. doi:http://dx.doi.org/10.1007/s13277-014-1620-y.

Yin, O.Q., Shi, X., Tomlinson, B., Chow, M.S., 2004. Interindividual and intraindividual variability of the urinary 6beta-Hydroxycortisol/Cortisol ratio in Chinese subjects: implications of its use for evaluating CYP3A activity. J. Clin. Pharmacol. 44 (12), 1412–1417. doi:http://dx.doi.org/10.1177/0091270004269760.

Zanger, U.M., Schwaby, M., 2013. Cytochrome P450 enzymes in drug metabolism: regulation of gene expression, enzyme activities, and impact of genetic variation. Pharmacol. Ther. 138 (1), 103–141. doi:http://dx.doi.org/10.1016/j.pharmthera.2012.12.007.

Zhou, S.F., Yang, L.P., Zhou, Z.W., Liu, Y.H., Chan, E., 2009. Insights into the substrate specificity, inhibitors, regulation, and polymorphisms and the clinical impact of human cytochrome P450 1A2. AAPS J. 11 (3), 481–494. doi:http://dx.doi.org/10.1208/s12248-009-9127-y.

Zhou, Y., Ingelman-Sundberg, M., Lauschke, V.M., 2017. Worldwide distribution of cytochrome P450 alleles: a meta-analysis of population-scale sequencing projects. Clin. Pharmacol. Ther. 102 (4), 688–700. doi:http://dx.doi.org/10.1002/cpt.690.