Chapter 11
The Role of Transporters in Drug Development: Regulatory Science Perspectives from the FDA

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Abstract Pharmacokinetic drug interactions can lead to altered systemic exposure and varied drug response. Evaluation of a new molecular entity's (NME's) drug–drug interaction (DDI) potential is therefore an integral part of drug development and regulatory review prior to its market approval. Transporters are expressed in varying abundance in all tissues in the body where they govern the access of molecules to cells or their exit from cells, thereby controlling the overall distribution of drugs to their intracellular site of action. Clinically relevant interactions mediated by transporters are of increasing interest in drug development. Research in this emerging area has revealed that drug transporters, acting alone or in concert with drug metabolizing enzymes, can play an important role in modulating drug absorption, distribution, metabolism, and excretion, thus affecting the pharmacokinetics and/or pharmacodynamics of a drug. The newly published draft drug interaction guidance by the Food and Drug Administration (FDA) in 2012 includes updated recommendations in addressing transporter-mediated drug interactions with various decision trees to help guide drug development and regulatory review. This chapter discusses, from a scientific perspective, role of transporters in drug development with a focus on transporter-mediated DDIs. First, transporter-related recommendations in the recent FDA's draft drug interaction guidance are discussed. Second, additional transporters that are emerging to be important in drug disposition are discussed. Third, recent review examples and transporter-related labelings are presented. Finally, future directions are discussed.

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Abbreviations

ABC	ATP-binding cassette
ADME	Absorption, distribution, metabolism, and excretion
AUC	Area under the plasma concentration-time curve
BCRP	Breast cancer resistance protein
BSEP	Bile salt export pump
CL_r	Renal clearance
CL_{total}	Total clearance
C_{max}	Maximal plasma concentration
cMOAT	Canalicular multispecific organic anion transporter (also named MRP2)
DDI	Drug–drug interaction
F_aF_g	Fraction of dose of inhibitor which is absorbed
FDA	Food and Drug Administration
f_u	Fraction unbound
GFR	Glomerular filtration rate
$[I]_1$	Mean steady state total (free and bound) C_{max} following the highest proposed clinical dose
$[I]_2$	Dose of inhibitor (in mol)/250 mL
$I_{in,max}$	Estimated maximum inhibitor concentration at the inlet to the liver
ITC	International transporter consortium
K_a	Absorption rate constant
LST	Liver-specific transporter
MATE	Multidrug and toxic compound extrusion transporter
MRP	Multidrug resistance-associated protein
NDA	New drug application
NME	New molecular entity
OAT	Organic anion transporter
OATP	Organic anion transporting polypeptide
OCT	Organic cation transporter
PD	Pharmacodynamics
PFiC2	Progressive familial intrahepatic cholestasis type 2
P-gp	P-glycoprotein
PK	Pharmacokinetics
PMC	Postmarketing commitment
PMR	Postmarketing requirement
Q_h	Estimated hepatic blood flow
SGLT2	Sodium-glucose cotransporter 2
SLC	Solute carrier
TEA	Tetraethyl ammonium

11.1   Introduction

Transporters are membrane-bound proteins that control the access of endogenous substances and xenobiotics (drugs) to various sites of the human body. In contrast to metabolizing enzymes, which are largely concentrated in the liver and intestine, transporters are present in all tissues in the body and play important roles in drug absorption, drug distribution, tissue-specific drug targeting, and elimination, thus influencing drug pharmacokinetics (PK) and pharmacodynamics (PD) (both benefit and risk). Transporters can also work in concert with metabolizing enzymes in affecting a drug's PK and PD. As discussed in previous chapters, similar to metabolizing enzymes, transporters have binding sites that are saturable and can be inhibited or induced.

Transporters are expressed in varying abundance in all tissues in the body where they govern the access of molecules to cells or their exit from cells, thereby controlling the overall distribution of drugs to their intracellular site of action. Changes in transporter expression or activity via either genetic factors or drug interactions can contribute to variability in drug exposure and response. Many drug–drug interactions (DDIs) that cannot be explained by interactions at the metabolizing enzyme level may be mediated by transporters (Giacomini et al. 2010; Zhang et al. 2010; Zhang et al. 2011). One example is the interaction between cyclosporine and rosuvastatin. Cyclosporine increased rosuvastatin exposure by sevenfold in heart transplantation patients as compared to healthy subjects (Simonson et al. 2004). This interaction was "unexpected" because rosuvastatin is not extensively metabolized by Cytochrome P450 enzymes and it is also not transported by P-glycoprotein (P-gp), both of which are inhibited by cyclosporine. Recent studies suggested that OATP1B1 (organic anion transporting polypeptide 1B1), OATP1B3, and BCRP (breast cancer resistance protein) are involved in rosuvastatin disposition and may have contributed to this interaction (Ho et al. 2006; Niemi 2010; Xia et al. 2007).

From the human genome project, more than 400 transporters are identified. These transporters belong to one of two superfamilies: ATP-binding cassette (ABC) and solute carrier (SLC). The advancement of molecular cloning of various transporters and in vitro cell overexpression system to study interaction of drugs at the transporter level has enabled researchers to examine the underlying mechanism of DDIs. This improved understanding has provided the foundation to potentially predict in vivo drug interaction based on in vitro assays. For example, it was found that many statin drugs are OATP1B1 substrates; their interactions with cyclosporine (an OATP1B1 inhibitor) are therefore "anticipated." In addition, recent findings that many HIV protease inhibitors are OATP1B1/OATP1B3 inhibitors (Annaert et al. 2010) are critical in the design of needed clinical drug interaction studies in order to manage myriad of potential drug interactions between HIV protease inhibitors and other concomitantly administered drugs, including statins (http://www.fda.gov/Drugs/DrugSafety/ucm293877.htm).

Transporters have been drug targets for novel treatments. For example, drugs have been developed to be selective inhibitors of sodium-glucose cotransporter 2 (SGLT2) as potential therapeutics for treatment of type 2 diabetes (Pfister et al. 2011). Drugs that are substrates for a highly concentrative, broad-specific amino acid transporter (SLC6A14) may be developed for cancer treatment, e.g., estrogen-receptor positive breast cancer (Karunakaran et al. 2011).

This chapter discusses, from a regulatory science perspective, role of transporters in drug development with a focus on transporter-mediated DDIs. Transporter-related recommendations in the recent Food and Drug Administration's (FDA's) revised draft drug interaction guidance are discussed. Additional transporters that are emerging to be important in drug development are also discussed. In addition, recent review examples and transporter-related labeling are presented. Finally, future directions are discussed.

11.2 Regulatory Perspectives with Regard to Studying of Transporters During Drug Development

11.2.1 FDA Guidance Development Regarding Evaluation of Transporters During Drug Development

As part of the drug development strategy, transporters have been studied and evaluated during drug development (Chap. 14). This evaluation has become an important part of the new drug submission and approval process because transporters can play a key role in safety by mediating DDIs. The importance of transporter-mediated drug interactions has been discussed at FDA advisory committee meetings in recent years (November 3–4, 2004, http://www.fda.gov/ohrms/dockets/ac/cder04.html#PharmScience and October 18–19, 2006, http://www.fda.gov/ohrms/dockets/ac/cder06.html#PharmScienc). Based on these committee discussions, in the 2006 FDA draft drug interaction guidance, the FDA recommended that sponsors study certain types of transporter-mediated drug interactions (http://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/DrugInteractionsLabeling/ucm093606.htm). Particular attention was paid to P-gp, which was thought to be a key transporter affecting the pharmacokinetics of digoxin, a narrow therapeutic index drug. Studying a new molecular entity's (NME's) inhibition or induction potential with P-gp is clinically relevant to appropriate dosing with digoxin and other molecules with similar characteristics.

Within more than 400 transporters in human genome, only about 30 transporters are found to date to be involved in absorption, distribution, metabolism, and excretion (ADME) processes and can be subjected to drug interactions. Besides P-gp, a reasonable question is what other transporters may be important and should be studied during the course of drug development? In 2007, an International Transporter

Consortium (ITC), which includes members from academia, industry, and the FDA, was formed with the goal of determining transporters that are of emerging importance in clinical drug interactions, establishing standards for in vitro evaluation of transporter-based interactions that may reduce the need for in vivo studies, and achieving, where possible, a consensus on the current knowledge of transporters in drug development (Huang and Woodcock 2010; Huang et al. 2010). The ITC organized an FDA critical path initiative-funded transporter workshop in October 2008 and authored a transporter whitepaper that was published in Nature Reviews Drug Discovery in March 2010 (Giacomini et al. 2010). The publication shared experiences, stimulated further discussions, and provided strategic directions in the following scientific areas: key transporters with clinical implications, in vitro methodologies, and decision trees on key transporters as to when to conduct in vitro and in vivo drug interaction evaluations (Giacomini et al. 2010).

The 2010 ITC paper discussed the following transporters as established and may be appropriate to evaluate during drug development: P-gp, BCRP, OATP1B1, OATP1B3, Organic Cation Transporter 2 (OCT2), and organic anion transporters (OAT1 and OAT3) (Giacomini et al. 2010). This was based on compelling clinical evidence that these transporters are involved in drug absorption, disposition, and/or drug interactions. Recent research indicates the important role of various transporters in the absorption (e.g., intestinal P-gp and BCRP), distribution (e.g., P-gp at the blood–brain barrier, OATP1B1, and OATP1B3 in the liver), and excretion (e.g., organic anion and cation transporters, OAT1, OAT3, and OCT2 in the kidney) of drugs. Several drugs (e.g., quinidine, verapamil, itraconazole) can increase plasma levels of digoxin by inhibiting the efflux transporter, P-gp, at the intestinal level. Plasma levels of many statins, including simvastatin, atorvastatin, pravastatin, and rosuvastatin, can be increased by inhibitors of hepatic uptake transporters (OATP1B1), such as cyclosporine, lopinavir/ritonavir, or eltrombopag. In addition, probenecid, a general inhibitor of OAT1/3, has been shown to increase the plasma levels of furosemide, tenofovir, and ciprofloxacin, possibly by the inhibition of the active renal tubular secretion of these substrates. Table 11.1 lists selected clinically relevant transporter-based DDIs. Many of these interactions resulted in two- to threefold increases in systemic exposure of the substrates; some were more than ninefold (e.g., when comparing pravastatin levels in the presence of cyclosporine in transplant patients with those without cyclosporine in healthy subjects). Other interactions resulted in decreases in plasma levels (e.g., tipranavir/ritonavir and loperamide). It is therefore important to consider, during drug development, which transporters can affect the ADME of an investigational drug and how the investigational drug can affect other drug's ADME due to its effect on transporters.

Following the publication of the ITC whitepaper, FDA discussed transporter-mediated drug interactions at an advisory committee meeting in March 2010 (http://www.fda.gov/AdvisoryCommittees/Calendar/ucm201691.htm) to seek committee members' advice on whether the current science, technology, and clinical importance related to transporters would support the recommendation to evaluate the above-mentioned major transporters routinely during drug development. The seven transporters listed in the whitepaper were considered by the FDA

Table 11.1 Selected transporter-mediated clinically significant drug–drug interactions (modified from tables in Giacomini et al. 2010 and various literature sources)

Gene	Aliases[a]	Tissue	Function	Interacting drug	Substrate (affected drug)	Changes in substrate plasma AUC (AUC ratios)	References
ABC transporters of clinical importance in the absorption, disposition, and excretion of drugs							
ABCB1	P-gp, MDR1	Intestinal enterocyte, kidney proximal tubule, hepatocyte (canalicular), brain endothelia	Efflux	Dronedarone	Digoxin	2.6-fold	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm Rameis (1985), Hager et al. (1979)
				Quinidine	Digoxin	1.7-fold	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm, Jerling (2006)
				Ranolazine	Digoxin	1.6-fold	
				Tipranavir/ritonavir	Loperamide	0.5-fold	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm, Mukwaya et al. (2005)
				Tipranavir/ritonavir	Saquinavir/ritonavir	0.2-fold	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm
ABCG2	BCRP	Intestinal enterocyte, hepatocyte (canalicular), kidney proximal tubule, brain endothelia, placenta, stem cells, mammary gland (lactating)	Efflux	GF120918	Topotecan	2.4-fold	Kruijtzer et al. (2002)
				Curcumin	Sulfasalazine	3-fold	Kusuhara et al. (2012)
SLC transporters of clinical importance in the disposition and excretion of drugs							
SLCO1B1	OATP1B1	Hepatocyte (sinusoidal)	Uptake	Lopinavir/ritonavir	Bosentan	5–48-fold[b]	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm
	OATP-C			Cyclosporine	Pravastatin	9.9-fold	Neuvonen et al. (2006), Hedman et al. (2004)
	OATP2 LST-1			Rifampin (single dose)	Glyburide	2.3-fold	Zheng et al. (2009)

Gene	Transporter	Direction	Inhibitor	Substrate	Effect	Reference
SLCO1B3	OATP1B3, OATP-8 (Hepatocyte (sinusoidal))	Uptake	Cyclosporine	Rosuvastatin	7.1-fold[c,d,e]	Simonson et al. (2004)
			Cyclosporine	Pitavastatin	4.6-fold[e]	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm
			Lopinavir/ritonavir	Rosuvastatin	2.1-fold[e]	http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm, Kiser et al. (2008)
SLC22A2	OCT2 (Kidney proximal tubule)	Uptake	Cimetidine	Dofetilide	1.5-fold	Abel et al. (2000)
			Cimetidine	Pindolol	1.5-fold	Somogyi et al. (1992)
			Cimetidine	Metformin	1.4-fold[f]	Somogyi et al. (1987), Somogyi and Muirhead (1987)
SLC22A6	OAT1 (Kidney proximal tubule, placenta)	Uptake	Probenecid	Cephradine	3.6-fold	Li et al. (2006), Mischler et al. (1974)
			Probenecid	Cidofovir	1.5-fold	Li et al. (2006), Cundy (1999)
			Probenecid	Acyclovir	1.4-fold	Li et al. (2006), Laskin et al. (1982)
SLC22A8	OAT3 (Kidney proximal tubule, choroid plexus, brain endothelia)	Uptake	Probenecid	Furosemide	2.9-fold[g]	Li et al. (2006)

BCRP breast cancer resistance protein, *P-gp* P-glycoprotein, *MDR* multidrug resistance, *LST* liver-specific transporters, *OATP* organic anion transporting polypeptide, *OCT* organic cation transporter, *OAT* organic anion transporter

[a] Implicated transporter refers to the likely transporter; however, because the studies are in vivo, it is not possible to assign definitively specific transporters to these interactions

[b] Minimum predose plasma level (Ctrough) data from day 4 (48-fold), day 10 (5-fold) after coadministration

[c] Interaction could be partly mediated by BCRP

[d] Based on cross-study comparisons

[e] Interaction could be partly mediated by OATP1B1

[f] Interaction could be partly mediated by MATE-1/MATE-2K

[g] Interaction could be partly mediated by OAT1

Advisory Committee as being well demonstrated to play a role in DDIs in humans and should be considered for routine evaluation during drug development. The FDA's 2012 draft drug interaction guidance (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf) included recommendations on when to evaluate transporter-based drug interactions.

11.2.2 Highlights of FDA's 2012 Draft DDI Guidance on Transporter Evaluation During Drug Development

The purpose for transporter-mediated DDI studies is to ensure patient safety for marketed drugs and NMEs during clinical trials. Two important questions should be addressed before determining whether a transporter study is to be conducted:

1. Will the new drug (NME) become unsafe if a marketed drug inhibits a transporter that affects NME's exposure levels, i.e., does the NME's level depend on a given transporter?
2. Will the NME make other marketed drugs unsafe by inhibiting a transporter that a marketed drug is a substrate for?

The first question can be addressed by assessing whether the NME is a substrate of major transporters and the second by studying whether the NME is an inhibitor of major transporters.

To study an NME as a substrate of transporters, the ADME properties (major route of elimination) of the NME and the location of major transporters in human organs are key points to consider (see Fig. 11.1). For example, if the NME is highly metabolized or mainly eliminated by biliary secretion, liver transporters (OATP1B1, OATP1B3, P-gp, and BCRP) may be important for its disposition. Conversely, if a drug is mainly eliminated by renal elimination, then renal transporters such as OAT and OCT should be considered. In addition to OAT and OCT, multidrug and toxin extrusion transporters (MATEs) in the kidney may be important as well (see more discussion below in Sect. 11.2.3). The NME's physicochemical properties and structure can also be important determinants for its being a substrate for certain transporters. For example, OCTs and OATs mainly recognize low molecular weight cationic drugs and anionic drugs, respectively. However, there are exceptions; for example, cimetidine is recognized as a substrate for both OCT2 and OAT3.

Decision models to determine if an NME is a substrate for various transporters are included in the 2012 FDA draft drug interaction guidance (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf). In the FDA recommendations, an in vitro overexpression cell system that is transfected with a specific transporter tested with appropriate positive and negative control drugs can be one of systems to evaluate the NME as a substrate for that transporter. In vivo contribution of the transporter to the NME's disposition can be evaluated with either a specific in vivo inhibitor or in subjects with different
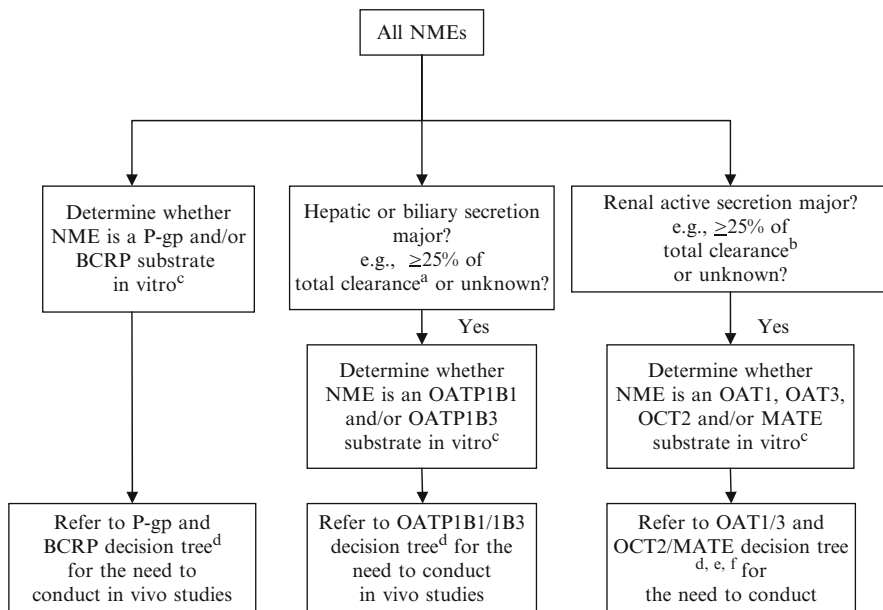
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                          ┌─────────────┐
                          │  All NMEs   │
                          └─────────────┘
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| Determine whether NME is a P-gp and/or BCRP substrate in vitro[c] | Hepatic or biliary secretion major? e.g., ≥25% of total clearance[a] or unknown? | Renal active secretion major? e.g., ≥25% of total clearance[b] or unknown? |

Yes / Yes

| | Determine whether NME is an OATP1B1 and/or OATP1B3 substrate in vitro[c] | Determine whether NME is an OAT1, OAT3, OCT2 and/or MATE substrate in vitro[c] |

| Refer to P-gp and BCRP decision tree[d] for the need to conduct in vivo studies | Refer to OATP1B1/1B3 decision tree[d] for the need to conduct in vivo studies | Refer to OAT1/3 and OCT2/MATE decision tree[d, e, f] for the need to conduct |

Fig. 11.1 Evaluation of new molecular entities (NMEs) as substrates for P-gp, BCRP, OATP1B1, OATP1B3, OAT1, OAT3, and OCT2/MATE transporters (modified from Giacomini et al. 2010; http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf)

[a]Biliary secretion can be estimated from preclinical data, in vitro hepatocyte uptake data or radio-labeled ADME data, and non-renal clearance data

[b]% Active renal secretion was estimated from $(CL_r - fu*GFR)/CL_{Total}$

[c]The sponsor has the option to use in vitro tools first for the evaluation

[d]http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf

[e]http://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/DrugInteractionsLabeling/ucm080499.htm

[f]Zamek-Gliszczynski, MJ et al. 2012

genotypes if the transporter has known polymorphism. A Biopharmaceutics Classification System (BCS) Class 1 drug that is highly soluble and highly permeable is not likely to be limited by an efflux transporter for its absorption even if it is recognized as a substrate in vitro. Whether this may apply for other BCS class drugs (i.e., Class 2 drugs) needs to be determined further.

To determine whether an NME is an inhibitor of major transporters, the 2012 FDA draft drug interaction guidance has also recommended decision trees to help determine when an in vivo clinical study is needed based on in vitro data (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf). The trees that were constructed included relatively conservative criteria for each decision point so as to avoid false negatives, i.e., a conclusion of no drug interaction when there may be drug interactions. The FDA's recommendations in the 2012 draft guidance differ from those from the ITC whitepaper in that the total maximum inhibitor concentration instead of unbound concentration
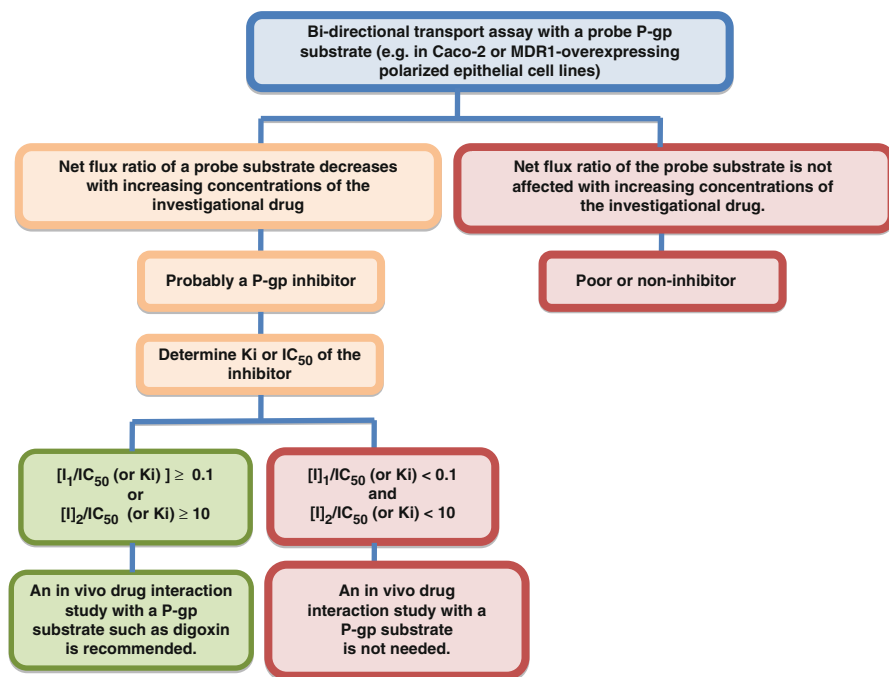
Fig. 11.2 P-gp/BCRP inhibition tree. Decision tree to determine whether an investigational drug is an inhibitor of P-gp and when an in vivo clinical study is needed. A similar model can be applied to a BCRP inhibitor (modified from figure in reference (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf)). $[I]_1$ represents the mean steady-state total (free and bound) C_{max} following administration of the highest proposed clinical dose. $[I]_2$= Dose of inhibitor (in mol)/250 mL (if IC_{50} is in a molar unit). For IC_{50} determination, a unidirectional assay (e.g., B to A) based on the probe substrate can also be considered

was used in the P-gp/BCRP and OATP decision trees (Figs. 11.2 and 11.3). In addition, in the OATP inhibition tree, a cut-off value of 1.25 for "*R*" instead of 2 was recommended. These modifications were based on limited data available to date for drugs that have both in vitro and in vivo interaction data (Agarwal et al. 2012; Vaidyanathan et al. 2012). Compared to the criteria proposed in the ITC whitepaper, the new criteria proposed in the FDA draft drug interaction guidance showed a lower number of false negative cases (Agarwal et al. 2012; Vaidyanathan et al. 2012). For example, in Table 11.2 we compared the criteria used in the ITC whitepaper and the FDA guidance for the OATP1B1 inhibition decision tree. Based on a dataset of 28 inhibitor–substrate pairs that have both in vitro OATP1B1 inhibition and in vivo interaction data, the ITC criteria (two steps) showed an overall 7 false negative cases and 2 false positive cases and the FDA criteria (two steps) showed an overall 4 false negative cases and 2 false positive cases. Interestingly, using the total C_{max}/IC_{50} in the first step as proposed in the FDA decision tree, there was 0 false negative and 2 false positives. By using an *R* of 1.25 in the second step in the two-step
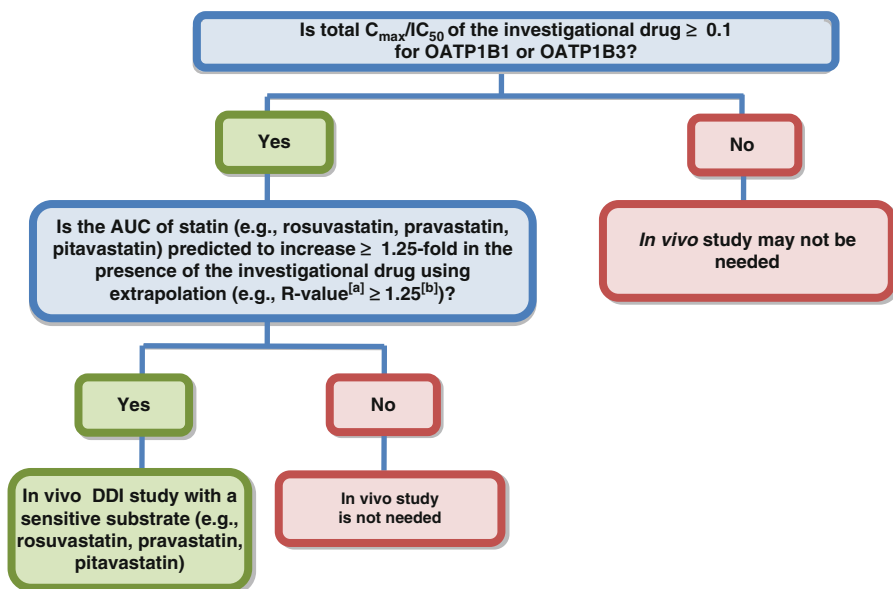
Fig. 11.3 OATP inhibition tree. Decision tree to determine whether an investigational drug is an inhibitor of OATP1B1 or OATP1B3 and when an in vivo clinical study is needed (modified from figures in reference (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf))

[a]R-value = 1+ (fu × $I_{in,max}$/IC_{50}), where, $I_{in,max}$ is the estimated maximum inhibitor concentration at the inlet to the liver and is equal to: C_{max} + (k_a × Dose × F_a F_g/Qh). C_{max} is the maximum systemic plasma concentration of inhibitor; Dose is the inhibitor dose; F_aF_g is the fraction of the dose of inhibitor which is absorbed; k_a is the absorption rate constant of the inhibitor and Qh is the estimated hepatic blood flow (e.g., 1500 mL/min). If Fa Fg values and ka values are unknown, use 1 and 0.1 min^{-1} (Ito et al. Pharmacol Rev. 50:387–412, 1998) for FaFg and ka, respectively because the use of theoretically maximum value can avoid false-negative prediction. For drugs whose fu values are less than 0.01 or fu cannot be accurately determined due to high protein-binding, then assume fu = 0.01, to err on the conservative side to avoid false negative predictions

[b]These are the suggested values according to the upper limit of equivalence range. We are open to discussion based on sponsors' interpretation

process, 2 false positives remained and there were 4 false negative cases. The available, limited data may suggest that one step instead of two steps may be sufficient as an initial screening to avoid false negative cases. However, the false negative cases based on two-step approaches may be attributed to mechanisms other than OATP1B1 inhibition for the interaction observed or to the variability in IC_{50} values determined in various laboratories as well as potential substrate-dependent IC_{50} values. For example, the cyclosporine–pitavastatin interaction represented one of the false negative cases based on *R* calculations. Using the FDA criteria, cyclosporine is classified as an inhibitor of OATP1B1 in vitro using total C_{max}/IC_{50} in the first step; however, it was not classified as an inhibitor of OATP1B1 in vitro based on *R* in the second step (i.e., *R* < 1.25). In vivo interaction data showed a significant increase in

Table 11.2 OATP1B1 inhibition potential prediction: comparison of total C_{max} vs. unbound C_{max} in the calculation of step 1 and R of step 2 vs. observed in vivo interactions based on ITC and FDA-proposed criteria

Inhibitor	Affected drug	In vivo AUC fold change[a]	IC_{50} in μM	In vitro substrate used for IC_{50} determination	ITC step 1: unbound $C_{max}/IC_{50} \geq 0.1$? (value)	FDA step 1: total $C_{max}/IC_{50} \geq 0.1$? (value)	Step 2: predicted R value[a] ≥ 2 (ITC)? or ≥ 1.25 (FDA)?	Category based on ITC two steps	Category based on FDA two steps	Category based on FDA first step only
Atazanavir	Pitavastatin	1.31	1.7	Cholyl-glycylamido-fluorescein (CGamF)[b]	Yes (0.6)	Yes (4.5)	2.1	TP	TP	TP
Atorvastatin	Repaglinide	1.14	0.7	Estradiol-17-β-glucuronide[b]	No (0.0006)	No (0.03)	1.0	TN	TN	TN
Cyclosporin	Atorvastatin	7.44–15.30	0.021	Atorvastatin	Yes (3.7)	Yes (53)	8.7	TP	TP	TP
Cyclosporin	Bosentan	1.97	0.3	Bosentan	Yes (0.22)	Yes (3.2)	1.5	FN	TP	TP
Cyclosporin	Cerivastatin[c]	3.80–4.75	0.238	Cerivastatin	Yes (0.26–0.46)	Yes (3.6–6.5)	1.5–1.7	FN	TP	TP
Cyclosporin	Fluvastatin	1.89–3.55	0.021	Atorvastatin[b]	Yes (1.5)	Yes (21)	4.5	TP	TP	TP
Cyclosporin	Pitavastatin	4.51	0.7	Pitavastatin	No (0.045)	Yes (0.64)	1.09	FN	FN	TP
Cyclosporin	Pravastatin	9.92–22.83	0.021	Atorvastatin[b]	Yes (1.5)	Yes (21)	4.5	TP	TP	TP
Cyclosporin	Rosuvastatin	7.08	0.31	Rosuvastatin	Yes (0.14)	Yes (2.1)	1.28	FN	TP	TP
Cyclosporin	Repaglinide	2.43	0.021	Atorvastatin[b]	Yes (1.8)	Yes (26)	4.0	TP	TP	TP
Cyclosporin	Simvastatin[c]	2.55–7.97	0.021	Atorvastatin[b]	Yes (2.7)	Yes (38)	6.6	TP	TP	TP
Darunavir/ritonavir	Pitavastatin	0.74	3.1	Cholyl-glycylamido-fluorescein (CGamF)[b]	Yes (0.14)[e]	Yes (2.9)[e]	2.4[e]	FP	FP	FP

Erythromycin	Bromocriptine	3.7[d]	Estradiol-17-β-glucuronide[b]	9.2	No (0.08)	Yes (0.52)	1.13	FN	FN	TP
Gemfibrozil	Atorvastatin	1.34	Atorvastatin	32	Yes (0.1)	Yes (3)	1.25	FN	TP	TP
Gemfibrozil	Pitavastatin	1.45	Pitavastatin	38	No (0.08)	Yes (2.7)	1.21	FN	FN	TP
Gemfibrozil	Repaglinide	7.42	Rosuvastatin[b]	4	Yes (0.77)	Yes (25)	2.96	TP	TP	TP
Gemfibrozil	Rosuvastatin	1.88	Rosuvastatin	4	Yes (0.77)	Yes (25)	2.96	TP	TP	TP
Gemfibrozil	Bosentan	5.22	CGamF[b]	0.5	Yes (0.7)[f]	Yes (47)[f]	2.97[f]	TP	TP	TP
Lopinavir/ritonavir	Pitavastatin	0.8	CGamF[b]	0.5	Yes (0.7)[f]	Yes (47)[f] (false positive)	2.97[f]	FP	FP	FP
Lopinavir/ritonavir	Rosuvastatin	1.6–2.07	CGamF[b]	0.5	Yes (0.7)[f]	Yes (47)[f]	2.97[f]	TP	TP	TP
Rifampin	Atorvastatin	4.30–9.33	Atorvastatin	3.25	Yes (0.96)	Yes (3.8)	2.32	TP	TP	TP
Rifampin	Bosentan	6.00	Bosenten	3.2	Yes (0.96)	Yes (3.9)	2.34	TP	TP	TP
Rifampin	Caspofungin	1.61	Estradiol-17-β-glucuronide[b]	0.6	Yes (5.2)	Yes (20.9)	8.14	TP	TP	TP
Rifampin	Pravastatin[c]	2.27	Estradiol-17-β-glucuronide[b]	0.6	Yes (5.2)	Yes (20.9)	8.14	TP	TP	TP
Sildenafil	Bosentan	1.33	Bosentan	1.5	No (0.02)	Yes (0.6)	1.05	FN	FN	TP
Simvastatin	Talinolol	0.99	Estradiol-17-β-glucuronide[b]	5	No (0.00002)	No (0.0004)	1.00	TN	TN	TN
Tipranavir/ritonavir	Atorvastatin	9.36	Estradiol-17-β-glucuronide[b]	1.1	Yes (1.72)[g]	Yes (86)[g]	3.73[g]	TP	TP	TP
Tipranavir/ritonavir	Rosuvastatin	1.37	Estradiol-17-β-glucuronide[b]	1.1	Yes (1.72)[g]	Yes (86)[g]	3.73[g]	TP	TP	TP

(continued)

Table 11.2 (continued)

Summary

N=28	ITC 2-steps	FDA 2-steps	FDA 1 step (i.e., C_{max}/IC_{50})
FN TP	8 16	2 20	0 24
TN FP	2 2	2 4	2 2
Accuracy=(TN+TP)/N*100 %	64 %	79 %	93 %

TP true positive, *TN* true negative, *FP* false positive, *FN* false negative

[a] AUCR ≥ 1.25 is considered positive interaction

[b] IC_{50} values determined using a different substrate as the in vivo-affected drug

[c] Two different assays were used to determine cyclosporin concentrations (C_{max} following single dose and multiple dose were used)

[d] Interaction likely to be mediated by CYP3A

[e] Data shown are based on darunavir. R value from ritonavir was lower, 1.2. $F_a = 1$ and $K_a = 0.1$ min^{-1} were used in the calculation. If darunavir and ritonavir are combined, $R_{total} = 1 + (1.4 + 0.2) = 2.6$

[f] Data shown are based on lopinavir. R value from ritonavir was lower, 1.13. $F_a = 1$ and $K_a = 0.1$ min^{-1} were used in the calculation. If lopinavir and ritonavir are combined, $R_{total} = 1 + (1.97 + 0.13) = 3.1$

[g] Data shown are based on tipranavir. R value from ritonavir was lower, 1.27. $F_a = 1$ and $K_a = 0.1$ min^{-1} were used in the calculation. If tipranavir and ritonavir are combined, $R_{total} = 1 + (2.73 + 0.27) = 4$

pitavastatin plasma concentration when it was co-administered with cyclosporine that was thought to be mediated by OATP1B1 (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm) (see Table 11.2). One possible reason is that IC_{50} determined in vitro may be higher than in vivo potency. Alternatively, the interaction between these two drugs may be mainly due to BCRP inhibition in vivo rather than OATP1B1 inhibition. The BCRP decision tree, in this case, may be better in projecting the interaction potential between cyclosporine and pitavastatin. The I_1/IC_{50} and I_2/IC_{50} of cyclosporine for BCRP were estimated to be >0.1 and 10, respectively (Xia et al. 2007), indicating a positive interaction using the BCRP decision tree (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatory Information/Guidances/UCM292362.pdf). Therefore, multiple transporter decision trees may need to be considered for drugs that are substrates or inhibitors for multiple transporters to avoid false negatives. Mechanistic understanding of drug interaction is key when applying decision trees to predict drug interactions. In vitro assays will help provide information on a drug's potential as a substrate or inhibitor for certain transporters.

Concerns were expressed that an abundance of false positives will have a detrimental effect on the development of new drugs because of conducting studies that do not need to be conducted (i.e., these drugs have no drug interaction risk). Thus, a data-driven balance between false positive and false negative in deriving the cut-off criteria in the decision trees is clearly needed. The evolution and appropriate application of these decision trees will require constant monitoring and they should be revised as the knowledge base increases over time (Giacomini et al. 2010; Agarwal et al. 2012; Tweedie et al. 2013; http://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/DrugInteractionsLabeling/ucm080499.htm).

Drug interactions can occur by various mechanisms. Understanding the roles of metabolizing enzymes as well as transporters in a drug's ADME will provide a starting point to evaluate drug interactions during drug development. In vitro models to predict drug interaction potential are well established for CYP enzyme-based mechanisms, and are evolving for transporter-based mechanisms. The decision trees as described (in Figs. 11.1, 11.2, and 11.3 and in FDA draft guidance (http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM292362.pdf)) provide directions to link the in vitro data (as a substrate or an inhibitor) or other drug characteristics (e.g., physicochemical properties, in vivo ADME data) with their interaction potential. Various technologies and standards developed for the P450 enzymes have enabled us to understand possible P450-mediated drug interaction during drug development to support strategies intended to manage drug interactions to optimize the benefit/risk ratio in patient populations. Similarly, with the advancement of our understanding of transporters and development of various tools and standards to study transporters, we will be able to better predict transporter-mediated interactions.

Finally, the clinical significance of transporter-mediated DDIs needs to be interpreted in the framework of exposure–response relationship. Understanding transporters and their interactions will provide a mechanistic approach to explain

variability in a drug's pharmacokinetics, pharmacodynamics, efficacy and safety in human subjects, and to identify subgroups of patients at risk of developing adverse events or being undertreated.

11.2.3 *Emerging Transporters*

In March 2012, the ITC organized a second transporter workshop that included discussions of emerging transporters such as MATEs, multidrug resistance-associated proteins (MRPs), and bile salt export pumps (BSEPs) (Zamek-Gliszczynski et al. 2012; Hillgren et al. 2013).

11.2.3.1 MATEs

The multidrug and toxin extrusion transporter MATE1 (*SLC47A1*) is expressed in both kidney and liver cells at the apical side of the cell membrane, whereas MATE2-K (*SLC47A2*) is mainly expressed in the kidney (Giacomini et al. 2010; Damme et al. 2011; Masuda et al. 2006; Otsuka et al. 2005). Two isoforms of MATE2 have been identified, one of which, MATE2-K, has been characterized as a membrane transporter in the kidney (Masuda et al. 2006). Various drugs, including metformin, as well as endogenous substances, such as guanidine, have been shown to be substrates of MATE1 (Otsuka et al. 2005). MATE2-K, like MATE1, appears to transport an array of structurally diverse compounds, including many cationic drugs and endogenous compounds (Tanihara et al. 2007). Komatsu et al. (2011) characterized isoform 1 of MATE2 and showed that both human MATE2 (isoform 1) and MATE2-K (isoform 2) (1) operate in the kidney as electroneutral H⁺/organic cation exchangers; (2) express and localize in the kidney, with MATE2-K being slightly more abundant than MATE2; (3) transport tetraethyl ammonium (TEA); and (4) have similar inhibitor specificities. Since some substrates (e.g., metformin) or inhibitors (e.g., cimetidine) recognized by OCT2 are also recognized by MATEs (Tanihara et al. 2007), MATEs may act in concert with OCT2 to mediate the excretion of some drugs (Choi et al. 2011; Kusuhara et al. 2011). More evidence has shown that MATEs may play a role in the elimination of organic cationic drugs or in DDIs that was thought to be mediated by OCT2 in the kidney (Zamek-Gliszczynski et al. 2012; Masuda et al. 2006). Therefore, when evaluating drugs for their interaction potential with OCT2, their potential as a MATE substrate or inhibitor needs to be considered. MATEs (*SLC47A*) may be considered for prospective investigation along with OCT2 in drug development (Zamek-Gliszczynski et al. 2012; Hillgren et al. 2013).

11.2.3.2 MRPs

The MRP (*ABCC*) family of transporters is closely related and structurally similar to the MDR family. MRP transporters constitute nine members of the

ATP-binding cassette C subfamily (*ABCC1–6*, *10–12*). Other transporters in the ABCC subfamily are the cystic fibrosis transmembrane conductance regulator (*ABCC7*) and two sulfonylurea receptor isoforms (*ABCC8 and -9*) (Keppler 2011; Klaassen and Aleksunes 2010). Cloning, functional characterization, and cellular localization of most MRP subfamily members have identified them as ATP-dependent efflux pumps that transport a broad spectrum of endogenous and xenobiotic anionic substances across cellular plasma membranes (Keppler 2011; Klaassen and Aleksunes 2010).

MRP1 (*ABCC1*), MRP2 (*ABCC2*), and MRP4 (*ABCC4*) have been the most widely studied members of the MRP family in the context of PK and drug response. MRP1 was initially identified in lung cells which were known to not express P-gp and pumps anionic compounds (Cole et al. 1992). Substrates for MRP1 include anionic endogenous products; glutathione, glucuronosyl, and sulfate conjugates; and, in some cases, neutral molecules coupled to glutathione transport without conjugation. MRP2 (*ABCC2*) is similar to MRP1 except in its tissue distribution and localization. It is expressed on the canalicular membrane of hepatocytes and was formerly known as the canalicular multispecific organic anion transporter (cMOAT). The hepatobiliary and renal elimination of many drugs and their metabolites is mediated by MRP2 in the hepatocyte canalicular membrane and by MRP4 as well as MRP2 in the luminal membrane of proximal renal tubules. Therefore, inhibition of these efflux pumps affects PK unless compensation is provided by other ATP-dependent efflux pumps with overlapping substrate specificities. Genetic mutations in MRP2 cause Dubin–Johnson syndrome, a disease characterized by hyperbilirubinemia resulting from reduced transport of conjugated bilirubin into bile (Paulusma et al. 1997). MRP3 has been recently shown to transport phenolic glucuronide conjugates of acetaminophen, etoposide, methotrexate, and morphine from the basolaterol surface of hepatocytes into blood (Zelcer et al. 2005). MRP4 (*ABCC4*) has been shown to transport a number of endogenous substrates, such as eicosanoids, urate, conjugated steroids, folate, bile acids, and glutathione, as well as many drug substrates including cephalosporines, methotrexate, and nucleotide analog reverse transcriptase inhibitors (Schuetz et al. 1999; Russel et al. 2008). Based on preclinical or clinical data, a drug or its conjugates as MRP2 substrates may need to be considered along with other efflux transporters in the liver. The determination for studying a drug's inhibition on MRP2 may be based on preclinical and clinical observations of liver toxicity (Zamek-Gliszczynski et al. 2012; Hillgren et al. 2013).

11.2.3.3 BSEP

Enterohepatic circulation of bile acids is mediated by specific transporters in the hepatocytes and enterocytes (Klaassen and Aleksunes 2010). BSEP (*ABCB11*) is a transporter that is expressed exclusively on the canalicular side of hepatocytes and is involved in the biliary efflux of monovalent bile acids whereas MRP2 exports divalent and sulfated and/or glucuronidated bile acids and other conjugated anions including Phase II drug metabolites. Although BSEP primarily transports bile acids,

it can also transport drugs such as pravastatin (Hirano et al. 2005). A number of BSEP inhibitors have been identified (e.g., cyclosporine A, rifampicin, gliben-clamide) (Byrne et al. 2002).

Altered expression or function of bile acid transporters can be either a cause or a consequence of cholestasis. Progressive familial intrahepatic cholestasis type 2 (PFIC2) is caused by mutations in the *ABCB11* gene, which encodes BSEP (Jansen et al. 1999; Strautnieks et al. 1998). Mutations in the ABCB11 gene can lead to a rapid progressive hepatic dysfunction in early infancy. In such patients, the biliary bile salt levels can be reduced to less than 1 % of that in normal subjects. These defects or inhibition of BSEP may contribute to certain types of drug-induced cho-lestasis or other liver injury (Noe et al. 2005; Ogimura et al. 2011). Further research is needed to determine how drugs can be studied early in their development to assess their BSEP-related safety liabilities (Morgan et al. 2010). The determination for studying a drug's inhibition on BSEP may be based on preclinical and clinical observations of liver toxicity such as cholestasis (Zamek-Gliszczynski et al. 2012; Hillgren et al. 2013).

11.3 Inclusion of Transporter Information in Labeling of Recently Approved NMEs by the FDA

Labeling is an important communication tool for health care practitioners to understand risk–benefit profile of a drug. Managing DDI is an important compo-nent for minimizing adverse events related to polypharmacy. Agarwal et al. recently conducted a survey on the inclusion of transporter-related information in the package inserts (also referred to as PIs or labeling) of 183 NMEs (excluding biologics) approved between 2003 and 2011 (Agarwal et al. 2011, 2013). These analyses indicate that with recent advancement in the transporter research area, the FDA's 2006 draft drug interaction guidance may have encouraged drug com-panies to evaluate the role of transporters in a drug's ADME and incorporate transporter-related information in new drug applications (NDAs) as suggested by the increased percentage of NMEs with transporter information in the PIs approved in 2007–2011 (56 %) as compared to those of 2003–2006 (23 %) (Table 11.3 and Fig. 11.4). In vivo drug interaction studies with digoxin (without prior in vitro assessments) were conducted less frequently in the 2007–2011 period (3 %, 3/95), as compared to those in 2003–2006 period (15 %, 13/88), as indicated in the labelings. This finding may indicate that the FDA-proposed in vitro P-gp decision tree in the 2006 draft drug interaction guidance may have influenced the decision-making and negative in vitro inhibition data have reduced the need for in vivo digoxin drug interaction studies during drug development (Agarwal et al. 2012).

Table 11.3 Summary of transporter information in the NME labeling (2003–2011)

	2003–2006	2007–2011	Total
Total number of all approved NMEs	88	95	183
Number (%) of NME labelings that have information with specific transporter name(s)	21 (24 %, 21/88)	53 (56 %, 53/95)	74 (40 %, 74/183)
(a) P-gp	15 (75 %, 15/20)	49 (92 %, 49/53)	64 (88 %, 64/73)
(b) Other transporters	7 (33 %, 7/20)	20 (38 %, 20/53)	27 (36 %, 27/73)
Number (%) of NME labelings with in vivo digoxin DDI study data (without mentioning the involvement of specific transporters)	14 (16 %, 14/88)	3 (3 %, 3/95)	17 (9 %, 17/183)
Number (%) of NME labelings that include transporter-related information in the "Highlights" section of PI	6 (7 %, 6/88)	14 (15 %, 14/95)	20 (11 %, 20/183)

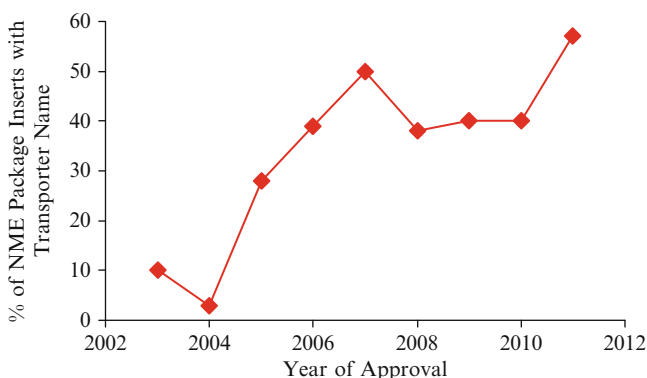Modified from Supplemental Table 1 in Agarwal et al. (2013)

Fig. 11.4 Percentage of NME labelings with transporter information (2003–2011)

11.4 Transporter-Related Postmarketing Requirement/ Postmarketing Commitment (PMR/PMC)

In the past, FDA has used the word *postmarketing commitment* to cover both required and not required studies and clinical trials that sponsor conducted post drug approval. In 2007, a new section 505(o) of the Federal Food, Drug, and Cosmetic Act (the Act) (21 U.S.C. 355(o)) was added to section 901 of the Food and Drug Administration Amendments Act of 2007 (FDAAA) in 2007. Section 505(o)

(3) authorizes FDA to require certain postmarketing studies and clinical trials for prescription drugs and biological products. Under FDAAA, PMR studies are required by the FDA "if FDA becomes aware of new safety information," and PMC is agreed upon studies between FDA and the applicant but it is not required (Guidance for Industry 2011). The results from postmarketing studies or trials can help further refine the safety, efficacy, or optimal use of a product.

As the field of transporter research is evolving rapidly, transporter-related studies may not have been conducted during drug development or considered in the premarketing approval decision, especially for drugs in therapeutic areas for which there are major unmet medical needs, such as oncology. Therefore, the FDA has recently asked for postmarketing studies of potential transporter-mediated DDIs when appropriate. A review of recent PMC and PMR studies included in the NME approval letters (2007–2011) indicated that there were more than 20 PMC and PMR studies that were related to evaluation of transporter-based DDIs either in vitro or in vivo (Fan et al. 2012). Results from these studies, when completed, will provide helpful information in the label. Transporters could be an important determinant for safe and effective use of a drug. Because there is a lag time between drug approval and PMR/PMC fulfillment, the sponsors are encouraged to consider and collect relevant information related to transporters, when appropriate, during drug development and make it part of their drug development plan.

11.5   Recent Review Examples

We present some recent examples related to P-gp in NDA reviews in this section to illustrate how in vitro data may have helped to determine the need for in vivo DDI studies and be included in the labeling for guiding the drug usage.

Table 11.4 lists four NME examples (boceprevir, rilpivirine, ezogabine, and ticagrelor that were approved in 2011). All four NMEs contain in vitro inhibition data on P-gp (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm). Based on the P-gp decision tree (Fig. 11.2), i.e., using the ratio of the in vivo exposure (C_{max} or dose) and their corresponding in vitro parameters (IC_{50} values) as an indicator for their potential to inhibit P-gp in vivo, all four of them suggested positive inhibition. Among these four drugs, only ticagrelor had a follow-up in vivo study with digoxin (a P-gp substrate) during drug development and the information (positive in vivo data) was included in the "Highlights" of the labeling (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm). The other three drugs had a PMR study with digoxin in their approval letter. However, the in vitro P-gp inhibition data were presented variably in the labeling. Ezogabine's information is in the "Highlights" section on the metabolite inhibition on digoxin based on in vitro data, while boceprevir's in vitro P-gp inhibition information is in the "Drug Interactions" section and rilprivirine's labeling does not mention P-gp or digoxin. Considerations of each drug's therapeutic areas, other clinical pharmacology information (e.g., other significant drug interactions), and related clinical practice may have affected

Table 11.4 Selected examples of NME approved in 2011 with labeling information related to P-gp and/or with postmarketing requirement (PMR) studies

Drug name (brand name)	I_1/IC_{50} (>0.1?)	I_2/IC_{50} (>10?)	In vivo study with digoxin conducted at the time of approval?	Labeling (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm)	Regulatory outcome (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm)
Boceprevir (VICTRELIS)	0.13 (Yes)	246 (Yes)	No	Section 7.2: P-gp inhibitor based on in vitro "Boceprevir is a potential inhibitor of P-glycoprotein (P-gp) based on in vitro studies. The potential for a drug interaction with sensitive substrates of P-glycoprotein (e.g., digoxin) has not been evaluated in a clinical trial"	A PMR study with digoxin is recommended: "Conduct an in vivo drug-drug interaction trial between boceprevir and a sensitive substrate of P-glycoprotein (e.g. digoxin)"
Rilpivirine (EDURANT)	0.05 (No)	27 (Yes)	No	No mentioning of P-gp or digoxin	A PMR study with digoxin is recommended: "Conduct a clinical trial in healthy subjects to evaluate the effect of rilpivirine at steady state on the single dose pharmacokinetics of digoxin. The pharmacokinetics of digoxin when coadministered with rilpivirine (test arm) will be compared to the pharmacokinetics of digoxin by itself (reference arm). The primary digoxin pharmacokinetic parameters that will be evaluated are $AUC_{(0-\infty)}$, $AUC_{(0-t)}$, and C_{max}"

Table 11.4 (continued)

Drug name (brand name)	I_1/IC_{50} (>0.1?)	I_2/IC_{50} (>10?)	In vivo study with digoxin conducted at the time of approval?	Labeling (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm)	Regulatory outcome (http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm)
Ezogabine (POTIGA)	<0.06 (IC_{50} >100 μM) for the parent drug (no) ~0.6 for metabolite NAMR[a] (Yes)	<53 (IC_{50} >100 μM) for the parent drug (indeterminant) Not Applicable for metabolite.	No	In Highlights: "*N*-acetyl metabolite of ezogabine may inhibit renal clearance of digoxin, a P-glycoprotein substrate. Monitor digoxin levels. (7.2)" Also in Sections 7.2 (Drug Interactions/Digoxin) and 12.3 (Clinical Pharmacology/ Drug Interactions)	A PMR study with digoxin is recommended: "A clinical trial to evaluate the acetyl metabolite of ezogabine (NAMR) as an inhibitor of P-glycoprotein using digoxin as a probe substrate. Refer to the Agency's Guidance (http://www.fda.gov/downloads/Drugs/ GuidanceComplianceRegulatoryInformation/ Guidances/UCM072101.pdf) for more detailed recommendations regarding transporter-based drug-drug interactions"
Ticagrelor (BRILINTA)	0.17 (Yes)	88 (Yes)	Yes Positive interaction observed (AUC ↑ 28 %, C_{max} ↑75 %)	In Highlights: "Monitor digoxin levels with initiation of or any change in BRILINTA. (7.4)" Also in Section 7.5 (Drug Interactions/Digoxin): "Digoxin: Because of inhibition of the P-glycoprotein transporter, monitor digoxin levels with initiation of or any change in BRILINTA therapy [*see Clinical Pharmacology (12.3)*]." And in Clinical Pharmacology Section of the labeling	N/A

PMR postmarketing requirement, *NAMR N*-acetyl metabolite of ezogabine

[a]The major metabolite (NAMR) was found to be an in vitro inhibitor of P-gp

these labelings. The publication of the new draft drug interaction guidance and the clinical pharmacology labeling guidance may help to have consistent labeling.

11.6   Conclusions and Future Directions

Currently, transporters are being studied to varying degrees during drug development across the industry. More research is needed to develop and optimize various technologies (e.g., in vitro, in silico, imaging) to better study transporters and drug interactions. Specific guidelines and decision trees have been provided in a white paper and FDA's draft DDI guidance for assessing transporter-mediated drug interactions for transporters with demonstrated clinical relevance: P-gp, BCRP, OCT2, OAT1, OAT3, OATP1B1, and OATP1B3 (Giacomini et al. 2010; http://www.fda. gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/ UCM292362.pdf). With the advancement of molecular biology and availability of various cell lines expressing transporters, the in vitro systems have been increasingly used and will have a greater utility. However, many challenges remain despite recent progress. For example, in vitro assays have their physiological limitations in mimicking the in vivo situation, in particular, the interplay between enzymes and transporters and the possible compensatory increase in activities of one or more other transporters when the activity of a transporter is suppressed. Development of best practices of in vitro assays will facilitate greater utility of in vitro studies, minimize inter-laboratory variability, and increase data quality and data interpretation (Brouwer et al. 2013).

For in vivo studies, monitoring plasma concentrations in drug interaction studies may not reveal the interaction effect at the tissue level as interactions with transporters can affect a drug's tissue uptake and local concentrations leading to undesirable effects with no or little change in systemic exposure (Watanabe et al. 2010). Physiologically based pharmacokinetic (PBPK) modeling has been increasingly used during drug development and regulatory review in addressing DDIs (Zhao et al. 2011; Huang et al. 2013). PBPK models incorporating multiple processes involving both transporters and enzymes and other intrinsic and extrinsic patient factors may be developed to help address these complex questions (Huang and Rowland 2012). Furthermore, clear labeling and education efforts are needed in order to provide useful information to health care providers and patients about their individual risk of drug interactions related to transporters, and how to identify patients at risk.

Continual collaboration among academia, industry, and government agencies including regulatory agencies, such as the model set by ITC, is key to move the science forward in the spirit of the critical path initiative of FDA (Huang and Woodcock 2010).

Disclaimer The views presents in this chapter are those of authors and do not necessarily reflect the official view of the FDA.

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