Breast Cancer Resistance Protein Substrate and Inhibition Evaluation: Why, When, and How?

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Dear Editor

We wish to thank Poirier et al. (2014) for their recently published manuscript, “The need for human breast cancer resistance protein substrate and inhibition evaluation in drug discovery and development: why, when, and how?" which attempted to contemporize many of the key concepts of breast cancer resistance protein (BCRP; ABCG2)-mediated drug disposition and drug interactions. Indeed, further contextualization of the relevance of BCRP is needed to improve the clinical translation and prediction of both perpetrator and victim drug-drug interactions related to this transporter. However, we noticed that the authors have missed some critically important concepts when referring to drugs that have complex permeability/efflux and atypical metabolism, whose disposition has been mechanistically uncovered by reverse translation (working from bedside back to the bench). These omissions resulted in the description of sulfasalazine as a poor BCRP substrate in humans, even though it is currently one of the two best available clinical BCRP probes along with rosuvastatin (Urquhart et al., 2008; Yamasaki et al., 2008; Kusuhara et al., 2012).

The authors selectively used one specific clinical study as "proof" that "sulfasalazine PK was not affected in individuals with impaired BCRP function, nor when coadministered with a BCRP inhibitor" (Poirier et al., 2014). The study in question by Adkison et al. (2010) used oral sulfasalazine enteric-coated tablets and, in fact, surprisingly showed no significant pharmacokinetic changes with oral coadministration of the BCRP inhibitor, pantoprazole, or a significant gene-dose effect of the BCRP 421 C>A functional polymorphism. These results stand in stark contrast to three independent clinical studies using oral immediate-release sulfasalazine tablets and/or suspension formulation, where significant 2- to 4-fold increases in sulfasalazine exposure were observed in carriers of the BCRP 421 C>A polymorphism and during coadministration of an oral BCRP inhibitor (Urquhart et al., 2008; Yamasaki et al., 2008; Kusuhara et al., 2012), as well as three independent preclinical studies in Bcrp-knockout mice and rats (Zaher et al., 2006; Shukla et al., 2009; Zamek-Gliszczynski et al., 2012). As discussed by Adkison et al. (2010), an accidental flaw in the execution of their study demonstrated the importance of proper formulation selection (suspension or immediate release, but not extended release), rather than disproving the utility of sulfasalazine as a marker of BCRP activity in humans.

Please allow us to elaborate on some subtle and important, although perhaps not apparently obvious, study caveats from the Adkison et al. (2010) study that are used by Poirier et al. (2014) as "proof" that sulfasalazine is not sensitive to BCRP efflux in humans. The Adkison et al. (2010) study was confounded because the clinical site used an enteric-coated delayed-release formulation, which allowed sulfasalazine to bypass the majority of small-intestinal BCRP (ABCG2). Mechanistically, if a delayed-release tablet is administered, then sulfasalazine will not be available until the distal small intestine (ileum)/proximal large intestine (cecum), where BCRP expression is lower on a relative expression basis (Englund et al., 2006), but more importantly, is far less concentrated per unit of surface area (Pang, 2003), such that the absorption process will be altered from that observed following administration of an immediate-release formulation or suspension. As such, using a delayed-release formulation would be altered from that observed following administration of an immediate-release formulation or suspension. As such, using a delayed-release formulation would also impair the ability to detect a clinical drug-drug interaction focusing on sulfasalazine BCRP-limited absorption as an endpoint.

Furthermore, we find the following interpretation of the curcumin-sulfasalazine interaction by Poirier et al. (2014) to not represent the totality of available data: “…the mechanism of the sulfasalazine/curcumin interaction is not only an inhibition of BCRP but also an inhibition of the bacterial degradation of sulfasalazine in the intestine. This would also explain the lack of interaction seen between pantoprazole and sulfasalazine mentioned earlier.” Curcumin effects on sulfasalazine pharmacokinetics can only be attributed to BCRP, because curcumin increased sulfasalazine oral exposure 5- to 13-fold in wild-type mice, but had absolutely no effect on sulfasalazine pharmacokinetics in Bcrp-knockout mice (Shukla et al., 2009). As discussed earlier, the reason Adkison et al. (2010) did not observe an effect of the BCRP inhibitor, pantoprazole, on sulfasalazine pharmacokinetics is likely because the enteric-coated formulation was used, which bypassed BCRP in the small intestine.

We would also like to comment on the authors’ conclusion that sulfasalazine is not as sensitive to BCRP modulation in humans as knockout rodents based on the apparent discordance between the magnitude of oral sulfasalazine exposure increase in Bcrp-knockout rodent studies [23- to 111-fold (Zaher et al., 2006; Shukla et al., 2009; Zamek-Gliszczynski et al., 2012)] and that in human clinical studies [2- to 4-fold (Urquhart et al., 2008; Yamasaki et al., 2008; Kusuhara et al., 2012)]. This difference in Bcrp function between complete genetic ablation in Abcg2−/− animals and partial Bcrp inhibition by gefitinib was also acknowledged in the original publication by Zaher.

This Letter to the Editor is in response to ‘‘The Need for Human Breast Cancer Resistance Protein Substrate and Inhibition Evaluation in Drug Discovery and Development: Why, When, and How?’’ by Poirier et al., found in Drug Metab Dispos 2014, 42:1466–1477. dx.doi.org/10.1124/dmd.114.060970.

ABBREVIATIONS: BCRP (ABCG2), breast cancer resistance protein.

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et al. (2006) and later confirmed by Shukla et al. (2009). Indeed, this difference would be expected based on pharmacokinetic principles given that ABCG2 421 C>A polymorphism or coadministration of oral BCRP inhibitors represents partial inhibition of efflux function, and not complete ablation as in the gene-knockout models (Zamek-Gliszczynski et al., 2013). Furthermore, the contribution of intestinal versus systemic BCRP must be considered when comparing genetic-knockout animals to oral inhibitors in the clinic because, as pointed out by the authors, intestinal BCRP is more prone to inhibition (i.e., exposed to far greater inhibitor concentrations) than systemic BCRP in humans (Kalvass et al., 2013; Zamek-Gliszczynski et al., 2013). In this regard, it merits pointing out that in Bcrp-knockout mice, sulfasalazine exposure was increased an order of magnitude following intravenous administration (systemic BCRP contribution) and up to two orders of magnitude following oral administration (intestinal + systemic BCRP contribution) (Zaher et al., 2006). These data indicate that in rodents, there is maximally a one order of magnitude window for sulfasalazine exposure increase that can be elicited by intestinal BCRP inhibition. Differentiation between systemic and intestinal BCRP contribution, along with the pharmacokinetic effect of partial inhibition versus complete genetic ablation of BCRP, help reconcile the apparent disconnect between knockout rodent and clinical drug-drug interaction studies using sulfasalazine as a BCRP victim probe substrate. As such, we urge the authors to reconsider the conclusion that “A significant amount of discordance is apparent when comparing the observations in Bcrp knockout mice with that of clinical findings, the most significant of which is for sulfasalazine” (Poirier et al., 2014). This claimed “discordance” is an artifact of directly comparing partial intestinal BCRP inhibition to complete genetic ablation of both intestinal and systemic BCRP, a subject recently addressed by the International Transporter Consortium (Kalvass et al., 2013; Zamek-Gliszczynski et al., 2013).

If the goal is to clarify when, why, and how BCRP is important in drug discovery and development, we recommend that an integrated approach to transporter biology remain a priority, where the totality of data (in vitro, preclinical, and clinical experiences) is used to predict and understand BCRP-mediated drug disposition and drug-drug interactions.

Authorship Contributions
Wrote or contributed to the writing of the manuscript: Ware, Urquhart, Sugiyama, Zamek-Gliszczynski.

References

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