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Perspective: Multidrug Resistance

Additional Transporter Characterization May Lead to New Pharmaceutical Opportunities

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Cancer cell resistance to chemotherapy is considered to be one of the major reasons for failure of therapy for a majority of cancer patients (1). Some tumors are intrinsically resistant to treatment whereas others acquire resistance with exposure to a variety of drugs that appear to be structurally unrelated. This phenomenon is called multidrug resistance (MDR). Studies have shown that these cancer cells overexpress membrane bound proteins that efflux drugs out of the cells. Two proteins in particular, P-glycoprotein (P-gp) and multidrug resistance-associated protein (MRP2), have been linked to MDR associated with a variety of cancers. Recently it was also discovered that these transporters are not only expressed in tumor cells but also in many normal tissue such as the kidney, lung, liver, intestine, blood brain barrier and other tissues (1,2). Furthermore, it is evident from the literature that compounds that interact with efflux pumps, like P-gp, represent a wide spectrum of chemical structures as well as different classes of drugs. Those classes include not only anticancer drugs, but also therapeutic agents such as other cytotoxic agents, HIV-protease inhibitors, detergents, antibiotics, immunosuppressives, antihypertensives and many more (1,2). More recently, it was discovered that these transporters play an important role in the pharmacokinetics and tissue distribution of many therapeutic drugs (1,2).

In the last 6–8 months several papers (3–5) have been published in *Pharmaceutical Research* that focus on the potential of P-gp modulators to enhance bioavailability as well as their possibility to modulate drug-drug interactions. In one of the papers Sandt *et al.* (3) discussed the effect of P-gp modulators on the integrity of the blood-brain barrier (BBB) *in vitro.* The paper also discussed the potential usefulness of the *in vitro* BBB coculture model to evaluate P-gp modulators *in vitro* as well as the models use to elucidate the potential of P-gp modulators to cause drug-drug interactions (3). The first of two papers written by Wang *et al.* (4) discussed the effect of grapefruit juice on the possible modulation of not only CYP3A4 but also P-gp. Furthermore, it speculates about the use of this modulator to enhance bioavailability without the need to increase the dose of therapeutics. The second paper by Wang *et al.* (5) investigated the previously observed clinical interaction of statins with other drugs, using an *in vitro* whole cell assay. It is suggested that the interactions of statins may, in part or all, be due to inhibition of P-gp.

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Additional studies will be of importance to improve and extend our understanding of MDR modulators. It is now well established that co-administration of drugs that are P-gp substrates with P-gp modulators can lead to a significant increase in systemic absorption following oral administration or in intracellular accumulation of the drug. Co-administration of a modulator can also lead to increases in unwanted side-effects from the drug itself due to changes in the drugs' pharmacokinetics and tissue distribution. Such changes could be especially important for narrow therapeutic index drugs. Drug-modulator co-administrations can potentially also lead to drug-drug interactions with concomitant medication. The use of MDR modulators may, thus, lead to more liabilities of the drug and should be pursued with caution, as a last resource for drugs with unmeet medical needs. Our ability to control such interaction in the future will depend on the development of more specific MDR modulators and our understanding of the large class of ABC transporters, many of which have not been identified and/or characterized. With increased knowledge of the specificity of the individual transporters we may be able to more effectively design combinations that increase selectivity of action at the desired tissue sites without the unintended, increased drug levels and site effect liability.

Another area of interest is the development of *in vitro* and *in vivo* screening assays for the recognition of P-gp substrates. Two papers have recently been published in *Pharmaceutical Research* discussing this issue (6,7). The first paper by Gao *et al.* (6) discusses the apparent binding of P-gp modulators to the Taxol binding site using caco-2 cells. Recent studies indicate a minimum of two binding sites within P-gp (8,9) and more recently there is even evidence for the possibility of a third binding site (10). This is a rapidly evolving area of research and soon we will hopefully have different assays for the selective binding sites. These type of assays will further help us in determining the potential of possible drug-drug interactions involving the specific binding sites as well as help us in developing more specific modulators. Furthermore, as more data are generated for the different binding sites, we may be able to build specific computer models that can predict the likelihood of a compound to be a substrate for each of the binding sites. The paper by Cisternino *et al.* (7) discusses the use of an *in situ* brain perfusion model in the P-gp deficient (mdr1a[–/–]) and wild-type mouse to determine if compounds are P-gp substrates (7). The results of these studies demonstrate the value of knockout models for transporter studies *in vivo*. The results demonstrate that chemical inhibition of P-gp is not as effective as the knockout mouse to determine the influence of this efflux pathway. Such knock-

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out models, however, need to be well characterized. The only difference between the knockout and the wild type should be the gene of interest and no concurrent up-regulation of other transporter systems should occur. It is unclear at present if this is the status of this knockout model.

Recent studies have indicated that transporters in the MRP family of transporters are also important in modulating bioavailability and disposition of drugs in the body. A recent publication by Han *et al.* (11) looked at important physicochemical properties for recognition of methotrexate analogs by the MRP2/cMOAT transporter. The results indicate that recognition by the transporter is dependent upon a balance between polar and non-polar surface properties. Future research in this area will no doubt involve looking at more diverse sets of substrates to see if this structure-activity relationship is the same.

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