# Inhibition of P-glycoprotein transport function by modulating its conformational and topological states

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## Summary

1. We have tested several Pgp transported drugs by the *a*ntibody *c*ompetition *t*est (ACT). The tested compounds could be clearly classified into the ACT-positive or ACT-negativ groups, therefore we can conclude that this kind of dichotomy can be a general feature of many Pgp substrates/modulators.
2. The partial inhibition of the pump by the UIC2 mAb could be increased by ACT-positive agents to near-complete inhibition that prevailed after the removal of the drugs. The inhibitory binding of UIC2 could be induced by low, in itself ineffective concentrations of the modulators.
3. The coadministration of UIC2 mAb and these modulators could more effectively reduce the resistance of MDR cells than either the mAb or the modulators alone, also reflected in the *in vitro* cytotoxicity test.
4. We have investigated the possible causes of increased binding of UIC2 by the transported drugs (UIC2-shift). We have found that besides a small change in the dissociation constant the elevated UIC2 binding is brought about by the increase in the number of available binding sites.
5. The UIC2 mAb reached and was bound to the cell surface Pgps of solid tumors *in vivo*. The amount of bound UIC2 could be increased by the coadministration of low-dose, ACT-positive cyclosporin A. The mAb binding elicited increased daunorubicin accumulation in tumor cells *in vivo* and this was accompanied by an elevated daunorubicin uptake.
6. The raft association of Pgps could be decreased by membrane cholesterol depletion or increased by cholesterol saturation in MDR cells. Cholesterol depletion caused Pgp inhibition in living cells, while cholesterol saturation induced an increased rate of Pgp internalization.
7. Although cholesterol modulation was accompanied by decreased cell viability, we didn't observe a lowered ATP content in the dye-excluding, apparently viable cells that might have been the result of nonspecific membrane permeabilization. Thus, we could confirm that Pgp inhibition was effected by the altered lipid environment that perturbed drug transport efficiency and not to the toxicity of the cell treatment.

**Keywords**

Multidrug resistance, P-glycoprotein, UIC2 monoclonal antibody, xenotransplantation, cholesterol, ATP, cytotoxicity