influenced by the amount of anionic phospholipids in the membrane, since binding of doxorubicin decreases the concentration of the free, transportable form of the drug.[2] We found that since the addition of verapamil specifically decreased the binding of doxorubicin the rate of passive diffusion across model membranes containing anionic phospholipids was increased. It can be concluded that besides the reported decrease of P-gp mediated efflux at least two other effects can account for a rise of the internal doxorubicin concentration in the presence of verapamil, namely a decrease of binding to anionic phospholipids in plasma- and intracellular membranes and an increase of the rate of import across the plasma membrane. Furthermore, it is tempting to speculate that the specific decrease of the doxorubicin concentration at the headgroup region of the inner leaflet of the plasma membrane of a cancer cell strongly affects the availability of the doxorubiin for the P-gp.

1. De Wolf FA, Staffhorst RWHM, Smits H-P, Onwezen MF and De Kruijff B. *Biochemistry* 1993; 32: 6688–95.
2. Speelmans G, De Wolf FA, Staffhorst RWHM and De Kruijff B, submitted.

49
The addition of cyclosporin A (CsA) results in markedly elevated tissue levels of doxorubicin (DOX) in the SCID mouse

WT Bellamy, YM Peng, A Odeleye, E Huizenga, MJ Xu, TM Grogan and RS Weinstein

Department of Pathology and Arizona Cancer Center, University of Arizona, Tucson, AZ 85724, USA.

CsA has been demonstrated to reverse P-glycoprotein (P-gp)-mediated multidrug resistance *in vitro* and clinical trials are underway to study its efficacy in reversing P-gp-mediated drug resistance. An acute toxicity in the SCID mouse model was previously reported,[1] which was associated with the combination of CsA and DOX and not observed with either drug alone, nor with cremaphor EL (crem), the vehicle for CsA. This acute toxicity was characterized by death of the animals between days 3 and 5 of a planned q 4d ×3 dosing regimen and was associated only with the combination of CsA and DOX and was not observed with either drug alone, nor with combinations of verapamil and DOX or cremaphor EL, the vehicle for cyclosporin, and DOX. The current study was undertaken to determine if this acute toxicity was due to increased tissue levels of doxorubicin in target organs. It has been reported that CsA alters the pharmacokinetics of VP-16[2] as well as DOX.[3] In the current study, SCID mice were dosed with DOX (1.5 mg/kg, i.p.) and CsA (50 mg/kg, i.m.), crem (50 mg/kg, i.m.) or verapamil (40 mg/kg, i.p.). Animals were sacrificed at either 24 or 48 h following drug dosing and tissues harvested for analysis. Tissue levels of DOX were assessed by HPLC analysis. Results revealed a statistically significant elevation of tissue DOX levels when combined with CsA. At 24 h following dosing with DOX and CsA, DOX levels in the liver, kidney and heart were elevated 2- to 3-fold compared to the same organs from animals receiving DOX and saline. Tissue levels were increased with verapamil but not to the same magnitude as with CsA. Both verapamil and cyclosporin resulted in increased levels of DOX in the brain. Tissue levels of DOX in those animals receiving crem and DOX were elevated only slightly. It was concluded that the elevations in tissue DOX levels observed with the drug combination of DOX and CsA may explain the acute toxicities observed in the *in vivo* SCID mouse model.

Supported in part by the Phoenix Friends of the Arizona Cancer Center.

1. Bellamy WT *et al. Proc Am Ass Cancer Res* 1993; 34: 319.
2. Bellamy WT *et al. J Clin Oncol* 1992; 10: 1635.
3. Bellamy WT *et al. Cancer Res* 1993; 53: 4837.

50
Effects of the cyclosporine SDZ-PSC 833 (PSC 833) on the pharmacokinetics and toxicity of doxorubicin in mice

O Gonzalez, T Colombo, L Imperatori, M Zucchetti, M de Fusco and M d'Incalci

Istituto 'Mario Negri', Via Eritrea, 62, 20157 Milan, Italy.

PSC 833 is a cyclosporin A analog which is under clinical investigation in combination with doxorubicin (DOX) or other anti-cancer agents as a MDR1 revertant agent. The present study is focussed on the effects of PSC 833 on the distribution and toxicity of DOX in non-tumor bearing BDF male mice. Mice were given PSC 833 i.p. 30 min before i.v. DOX treatment. DOX was determined by a HPLC assay at different times during the 72 h following DOX treatment in serum, heart, intestine, liver, kidney, adrenals and brain. In all tissues except brain DOX AUC values were much greater in mice receiving 10 mg/kg DOX in combination with 12.5 or 25 mg/kg PSC 833 than in mice receiving DOX alone. The highest increase in DOX concentrations was found in intestine, liver, kidney and adrenals. Low, but still significant differences were found in the heart. PSC-833 did not appear to influence either urinary and fecal DOX elimination or DOX metabolism. Doses of PSC 833 devoid of any toxicity potentiated the acute and delayed toxicity of DOX dramatically. The mechanism for the enhanced toxicity has not been elucidated yet, but it is likely related to an increased tissue retention of DOX due to inhibition of the P-170 pump by PSC 833, as recently proposed for cyclosporin A by this laboratory.[1] Studies are in progress to establish whether in spite of the high toxicity observed, the combination of DOX and PSC 833 may still have advantageous results for the therapy of resistant tumors overexpressing P-170.

1. *J Pharm Exp Pharmacol* 1994; in press.