Restoration of Taxol Sensitivity of Multidrug-Resistant Cells by the Cyclosporine SDZ PSC 833 and the Cyclopeptolide SDZ 280-446

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Background: Taxol, a promising agent for the treatment of cancer, has entered phase II clinical trials. Nevertheless, it belongs to the class of compounds that show impaired retention in multidrug-resistant cells expressing P-glycoprotein (Pgp), a drug efflux pump. Chemosensitizers like verapamil modulate multidrug resistance by interfering with the efflux action of Pgp and thus can decrease drug resistance or can restore drug sensitivity by restoring normal drug accumulation and distribution within the multidrug-resistant tumor cell. The two strongest, nearly equipotent chemosensitizers identified to date are the cyclosporine derivative SDZ PSC 833 and the semisynthetic cyclopeptolide SDZ 280-446. *Purpose:* This study was designed to investigate the capacities of verapamil, SDZ PSC 833, and SDZ 280-446 to decrease resistance of two multidrug-resistant cell lines to taxol. *Methods:* We studied in vitro the growth of two multidrug-resistant tumor cell lines displaying high resistance to taxol: multidrug-resistant Chinese hamster ovary cells and murine monocytic leukemia P388 cells. We determined the taxol concentration that produced 50% inhibition of cell growth (IC_{50}) in the two multidrug-resistant cell lines and in the parent cell lines, in the presence of a range of chemosensitizer concentrations (0-30 μM). IC_{50} values were determined in the presence and in the absence of verapamil, SDZ PSC 833, or SDZ 280-446. *Results:* At nontoxic concentrations (0.3-1 μM), SDZ PSC 833 and SDZ 280-446 produced an almost complete reversal of the high taxol resistance of the multidrug-resistant tumor cells, whereas only partial restoration of sensitivity to taxol was achieved with verapamil. *Conclusion:* SDZ PSC 833 and SDZ 280-446 can restore the normal taxol sensitivity of highly resistant multidrug-resistant tumor cells. *Implications:* The combination of taxol with SDZ PSC 833 or SDZ 280-446 may be recommended for treatment of multidrug-resistant cancers. [J Natl Cancer Inst 85:478–483, 1993]

Taxol (NSC-125973) is a new anti-cancer agent (*1,2*) presently undergoing phase I and II clinical trials (*3,4*). This drug is isolated from trees of the genus *Taxus*, and its ability to arrest cell proliferation in the G_2 and M phases of the cell cycle is well documented (*5,6*). This capacity correlates with its unusual properties of interaction with tubulin in vitro (hyperstabilization of microtubules) (*1*).

Unfortunately, a major mechanism of cellular resistance to taxol is the increased drug efflux out of cells that display the "classic" multidrug-resistant phenotype. An increased capacity to pump out a variety of drugs with unrelated structures and intracellular targets (e.g., vinca alkaloids, doxorubicin, etoposide, dactinomycin, and colchicine) correlates with increased levels of mdr gene-encoded membrane glycoproteins (P-glycoprotein) (*7,8*).

A variety of agents can decrease in vitro the drug resistance of multidrug-resistant tumor cells and sometimes completely restore their sensitivity to chemotherapeutic agents. Such chemo-sensitizers belong to various structural classes. By interfering with the efflux function of P-glycoprotein (Pgp), they can decrease drug resistance or can restore drug sensitivity by restoring the normal drug accumulation and distribution within the multidrug-resistant tumor cell (*7,8*). Among these chemosensitizers, the calcium channel blocker verapamil (*7*) has been the most commonly studied. Nevertheless, very potent chemosensitizers have been most commonly identified among cyclosporines (*9,10*) and cyclopeptolides (*11*) derived from a Fungi imperfecti (*Septoria* sp.). Extensive studies of chemically modified derivatives of cyclosporines and cyclopeptolides led to the selection of the two strongest, nearly equipotent chemosensitizers identified to date: the cyclosporine derivative SDZ PSC 833 and the semisynthetic cyclopeptolide SDZ 280-446 (*11,12*).

How taxol is processed by Pgp is unknown, and whether SDZ PSC 833 and SDZ 280-446 could restore a normal sensitivity to taxol was not predictable. The present study was, thus, aimed at (*a*) measuring the level of taxol resistance displayed by two multidrug-resistant tumor cell lines extensively studied in our laboratory and (*b*) testing whether taxol resistance could be modulated by verapamil, SDZ PSC 833, and SDZ 280-446.

Materials and Methods

Cell Lines

Parental and multidrug-resistant Chinese hamster ovary (CHO) cells (*13*) were obtained from Dr. V. Ling (Ontario Cancer Institute, Toronto, Canada). Parental and multidrug-resistant murine monocytic leukemia P388 cells (*14*) were obtained from Dr. M. Grandi (Farmitalia Carlo Erba Research Center, Milan, Italy). The multidrug-resistant P388 cell line has been selected by growth in the presence of doxorubicin for the multidrug-resistant phenotype (*14*). The multidrug-resistant CHO cell line has been selected by growth in the presence of colchicine for the multidrug-resistant phenotype (*13*). Detailed information on the properties of these cell lines (i.e., sensitivity to various drugs and effects of various chemosensitizers) has been published by our laboratory (*9-12*).

The parental CHO cell line (AuxB$_1$ subclone ABISII) and the multidrug-resistant CHO line (CH^{R}C5 subclone C5S3.2) (*13*) have been extensively studied with regard to the capacity of various cyclosporines (*9*) and other chemosensitizers (*10*) to reverse the multidrug-resistant phenotype in vitro. These studies led to the development of the novel cyclosporine derivative SDZ PSC 833 and the novel semisynthetic cyclopeptolide SDZ 280-446 (*11,12*).

The parental P388 line (P388N) and the multidrug-resistant P388 line (P388-Dox^R) (*14*) have been used to compare in vitro the resistance-modulating activity of FK-506 and cyclosporin A (*15*), to evaluate the restoration of daunomycin retention in vitro by a variety of chemosensitizers (*16*), and to select the SDZ PSC 833 (*12,17*) and SDZ 280-446 (*11*) development compounds.

Anticancer Drugs and Chemosensitizers

For preliminary experiments aimed at testing the taxol resistance of our cell lines, we obtained taxol (from *Taxus baccata*) from the National Cancer Institute, National Institutes of Health,

Bethesda, Md. For subsequent experiments aimed at reversing the taxol resistance of our multidrug-resistant cell lines, we purchased taxol (from *Taxus brevifolia*) from Calbiochem AG, Buchs, Switzerland. The taxol was prepared as a 10-mg/mL stock solution in pure ethanol. Verapamil was purchased from Sigma Chemical Co. (St. Louis, Mo.) and prepared as a 100-mM stock solution in pure ethanol. SDZ PSC 833 and SDZ 280-446 were synthesized by Sandoz Pharma Ltd. (Basel, Switzerland) and were prepared as 10-mM stock solutions in ethanol.

Chemosensitizer Capacity Assay

Cell cultures were prepared as described previously (*9*). Briefly, the cultures were made to a final volume of 0.2 mL in 96-well microplates at 37 °C in a humidified atmosphere of 7% CO_2 and 93% air. Three-day cultures of P388 cells were initiated with 2000 parental P388 cells or 5000 multidrug-resistant P388 cells per well. Five-day cultures of CHO cells were initiated with 400 parental CHO cells or 800 multidrug-resistant CHO cells per well. Growth was measured by a colorimetric assay, as described previously (*9*).

The cell growth inhibitory effect of the chemosensitizer alone, in the presence of the taxol solvent (ethanol), was expressed as percent of the growth level obtained in the absence of both chemosensitizer and taxol, but in the presence of their solvent (ethanol) (*10,12*).

The taxol IC$_{50}$ (the concentration producing 50% inhibition of cell growth) was calculated from the dose–response curves obtained by plotting the measured growth versus the taxol concentration, as shown in Fig. 1. A whole range of IC$_{50}$ values were obtained in the presence of the chemosensitizer (verapamil, SDZ PSC 833, or SDZ 280-446A) (i.e., IC$_{50}$$^+$); values were also obtained in the absence of chemosensitizer, but in the presence of its solvent (*10,12*) (i.e., IC$_{50}$$^-$). The increases of taxol sensitivity, or "gains" in sensitivity, were given by the ratio IC$_{50}$$^-$/IC$_{50}$$^+$, and a gain was calculated for each chemosensitizer concentration (*11*).

Results

Taxol Resistance

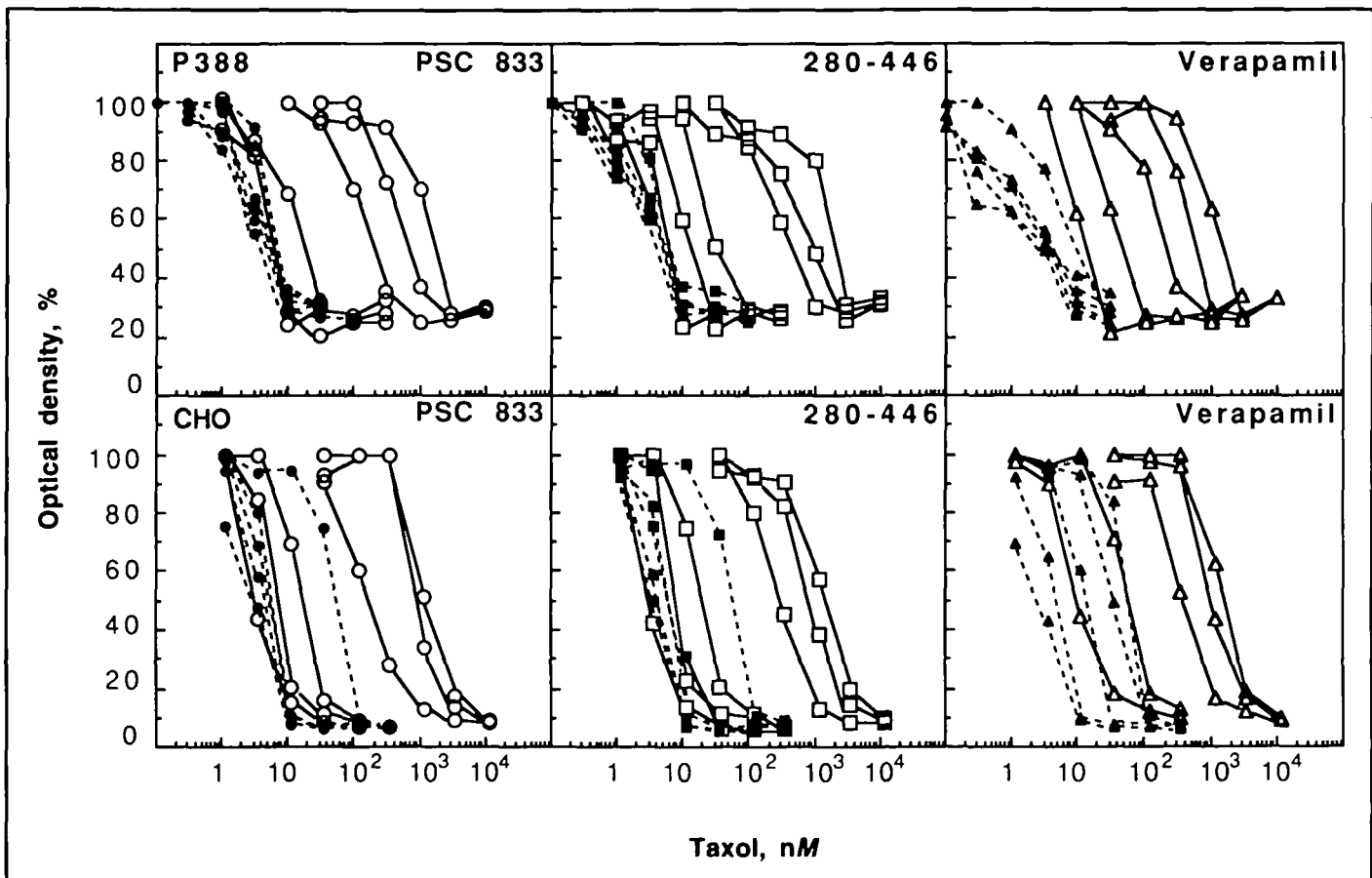The multidrug-resistant P388 cells were approximately 283-fold more re-

Fig. 1. Taxol-mediated cell growth inhibition. Results from a typical experiment showing cell growth (as percent of control growth; Y-axes) as a function of the taxol concentration (as n*M*; X-axes). Cell growth was measured by the optical density and taken as 100% in control culture wells (cells exposed to solvent only, run on each microculture plate). Dose–response curves obtained with P388 and CHO cell lines are shown, respectively, at the top and bottom of the figure (open symbols and solid lines for multidrug-resistant cells, and closed symbols and broken lines for parental cells). On each graph, curves from right to left were obtained in the presence of increasing concentrations of SDZ PSC 833 or SDZ 280-446 (0, 0.01, 0.03, 0.1, 0.3, and 1 μ*M*) or of verapamil (0, 1, 3, 10, and 30 μ*M*).

sistant to taxol than the parental P388 cells (Table 1). This increase compares with mean values of 130-fold for colchicine, 158-fold for daunomycin, 101-fold for doxorubicin, 140-fold for etoposide, and 158-fold for vincristine (12).

The multidrug-resistant CHO cells were approximately 36-fold more resistant to taxol than the parental CHO cells (Table 1). This increase compares with mean values of 54-fold for colchicine, 93-fold for daunomycin, 47-fold for etoposide, and 28-fold for vincristine (12). Higher relative resistance indices (Table 1) were observed for the P388 cells than for the CHO cells. This result may be partly due to the fact that the parental P388 cells were not sensitizable by Pgp-directed chemosensitizers (15,16) and did not express immunodetectable levels of Pgp (16), whereas the parental CHO cells themselves displayed a low level of innate resistance (9,10); this low-level resistance is presumably associated with their expression of low levels of Pgp (18,19).

Chemosensitization of P388 Cells

No sensitization of the parental P388 cells could be obtained by the chemosensitizers tested (verapamil, SDZ PSC 833, and SDZ 280-446). The taxol IC_{50}

Table 1. Anticancer drug resistance of P388 and CHO cell lines

Anticancer drug	Cell line*	
	P388	CHO
Colchicine†	130	54
Daunomycin†	158	93
Doxorubicin†	101	ND
Etoposide†	140	47
Vincristine†	158	28
Taxol	283	36
Parent cell IC_{50}‡	6.1 ± 0.7	50.8 ± 13.6
Multidrug-resistant cell IC_{50}‡	1729 ± 272	1828 ± 26

* Unless otherwise specified, numbers = relative resistance index (multidrug-resistant cell IC_{50}/parent cell IC_{50}). ND = not determined.

† Indices calculated using IC_{50} values previously obtained and published by our group (12).

‡ Mean values ± SD of 20 determinations within four individual experiments.

remained in the 4-6 nM range for concentrations of SDZ PSC 833 and SDZ 280-446 of 0.01 to 1.0 μM. There was also no decrease in the taxol IC_{50} of the parental P388 cells for verapamil concentrations up to 30 μM. Similarly, no major growth inhibition of the parental P388 cells was caused by the chemosensitizers alone at concentrations below 1 μM (Fig. 2). Thus, our strongest chemosensitizers (SDZ PSC 833 and SDZ 280-446), did not significantly sensitize (less than twofold increase) the parental P388 cells to taxol, as has been found with other drugs (colchicine, vincristine, daunomycin, doxorubicin, and etoposide) and with Pgp probes (rhodamine 123 and others) (11,12,16).

In the case of the multidrug-resistant P388 cells, a virtually complete reversal of taxol resistance could be achieved with SDZ PSC 833 and SDZ 280-446 but not verapamil. Indeed, a taxol IC_{50} of about 1800 nM, as obtained in the absence of any chemosensitizer, could be reduced to about 6 nM in the presence of 1 μM of SDZ

PSC 833 or SDZ 280-446 (Table 2). These chemosensitizer concentrations were not inhibitory (<20% inhibition) by themselves for multidrug-resistant P388 cell growth (Fig. 2). At a concentration at which it was not substantially inhibitory by itself (1 μM), verapamil could sensitize the cells only weakly (taxol IC_{50} of about 700 nM); at a concentration (30 μM) at which it slightly inhibited multidrug-resistant P388 cell growth by itself (Fig. 2), verapamil still could not bring the taxol IC_{50} to its expected baseline of 4-6 nM (Table 2). (The expected baseline was the range of IC_{50} values obtained with this type of assay with a panel of sensitive cell lines.)

Chemosensitization of CHO Cells

The taxol IC_{50} of about 50 nM shown by the parental CHO cells in the absence of chemosensitizer could be reduced to the apparent baseline of 2-4 nM by the three chemosensitizers (Table 2). SDZ PSC 833 and SDZ 280-446 were stronger chemosensitizers
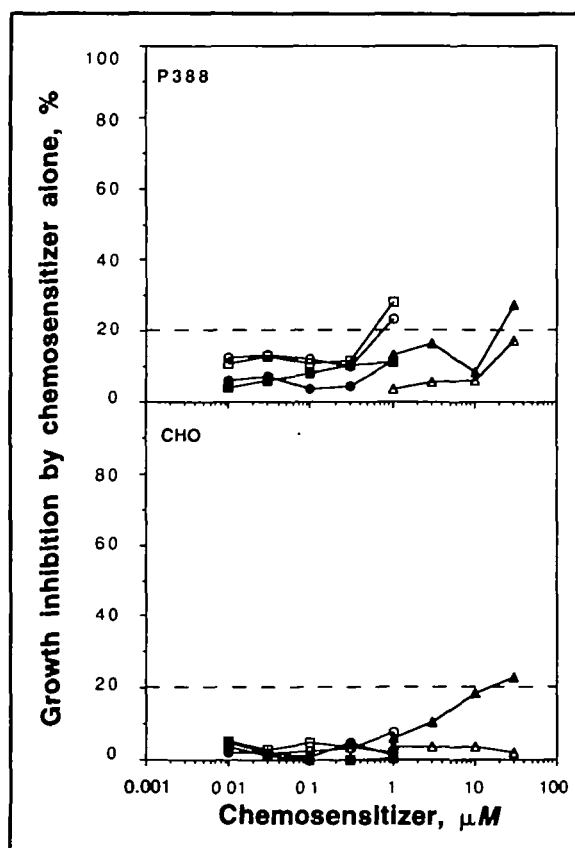

Fig. 2. Chemosensitizer-induced cell growth inhibition. Cell growth inhibition (as percent of control growth; Y-axes) is shown as a function of chemosensitizer concentration (as μM; X-axes). Growth inhibition was calculated as the difference between growth in the absence and presence of chemosensitizer. Open symbols represent parental cells, and closed symbols represent multidrug-resistant cells (circles for SDZ PSC 833, squares for SDZ 280-446, and triangles for verapamil). Values are means of two to four experiments.

Table 2. Taxol resistance-modulating activity of SDZ PSC 833, SDZ 280-446, and verapamil for P388 and CHO cell lines

Chemosensitizer	Concentration,* µM	Taxol IC$_{50}$, nM			
		P388 cells		CHO cells	
		Parent	Multidrug-resistant	Parent	Multidrug-resistant
SDZ PSC 833	0	6.3 ± 0.9	1812 ± 151	50.2 ± 13.0	1662 ± 344
	0.01	5.5 ± 0.2	665 ± 218	5.5 ± 0.4	994 ± 170
	0.03	4.9 ± 0.2	241 ± 179	3.9 ± 1.7	195 ± 52
	0.1	5.2 ± 1.1	38.5 ± 24	4.0 ± 1.5	18.3 ± 2.7
	0.3	4.5 ± 0.9	10.9 ± 7.1	3.7 ± 1.3	7.3 ± 0.9
	1	4.5 ± 1.8	6.3 ± 1.6	3.4 ± 1.3	3.0 ± 0.5
SDZ 280-446	0	6.0 ± 0.8	1760 ± 195	49.7 ± 14.6	1886 ± 223
	0.01	5.1 ± 0.2	975 ± 141	8.3 ± 0.5	835 ± 48
	0.03	4.7 ± 0.6	518 ± 116	5.7 ± 0.9	221 ± 115
	0.1	4.5 ± 0.4	69 ± 33	4.4 ± 1.8	25.3 ± 5.2
	0.3	4.7 ± 1.1	22.3 ± 8.9	3.9 ± 1.6	10.8 ± 3.8
	1	5.2 ± 1.2	7.2 ± 1.0	3.5 ± 1.3	3.8 ± 0.9
Verapamil	0	5.9 ± 0.7	1613 ± 428	52.4 ± 17	1950 ± 87
	1	4.7 ± 1.2	742 ± 311	31.7 ± 20	1226 ± 325
	3	3.9 ± 1.5	246 ± 91	22.3 ± 25	352 ± 60
	10	4.2 ± 1.3	60.7 ± 16	4.2 ± 1.6	66 ± 7.6
	30	3.7 ± 1.6	18.5 ± 3.3	2.7 ± 0.8	15 ± 5.6

*The same solvent concentration was present in all samples. Mean value of two to four experiments ± SD.

than verapamil, inasmuch as this baseline was approached by 0.03 µM SDZ PSC 833 and 0.1 µM SDZ 280-446 but required 10 µM verapamil. The parental CHO cells were slightly resistant to the multidrug resistance-modifying agents, with higher drug IC$_{50}$s than for parental tumor cell lines of other origins (e.g., L1210 and KB-3.1), as observed earlier with several other drugs (e.g., colchicine and etoposide) and with these and other chemosensitizers (e.g., amiodarone and quinidine) (9-12). The resistance of the parental CHO cells can be abrogated by very low concentrations of strong chemosensitizers or by higher doses of weak chemosensitizers (unpublished data from the study of nearly 1000 compounds). This chemosensitizing activity can probably be explained by the inhibition of the small amounts of Pgp expressed by the parental CHO cells (18,19). With little Pgp function to neutralize, the maximum gains in sensitivity were, of course, correspondingly low (<20-fold) (9-12).

The multidrug-resistant CHO cells were highly resistant to taxol, with an IC$_{50}$ of about 1800 nM, but were completely sensitizable by 1 µM SDZ PSC 833 or SDZ 280-446 (Table 2). Those chemosensitizer concentrations gave no detectable inhibition of multidrug-resistant CHO cell growth (Fig. 2). Moreover, even 30 µM

verapamil could not bring the taxol IC$_{50}$ to the apparent 2-4 nM baseline, although it produced a taxol IC$_{50}$ of about 2.7 nM in untreated parental CHO cells.

Discussion

Taxol is a promising anticancer drug (20,21), as shown by recent clinical studies (2-4), but it belongs to the class of compounds that can be effluxed by multidrug-resistant cells expressing Pgp (1). Fortunately, as shown in Table 2 and Fig. 2, taxol resistance can be virtually neutralized by nontoxic concentrations (0.3-1 µM) of SDZ PSC 833 or SDZ 280-446.

In the present study, gains in taxol sensitivity of 250- to 300-fold could be reached with the multidrug-resistant P388 cells. (For calculation of gains, see the "Materials and Methods" section.) In equimolar comparisons, SDZ PSC 833 seemed to be slightly more active than SDZ 280-446 for the reversal of taxol resistance, but the difference was small (about twofold). Much higher concentrations of verapamil were required to obtain comparable gains.

With the multidrug-resistant CHO cells, gains of about ninefold and about 550-fold could be obtained by 0.03 µM and 1 µM, respectively, of either SDZ PSC 833 or SDZ 280-446. These

chemosensitizers appeared to be equipotent for chemosensitizing this cell line and much stronger than verapamil: A maximum gain of sensitization of 130-fold required 30 µM verapamil.

The two multidrug-resistant tumor cell lines in the present study displayed a level of taxol resistance similar to their levels of resistance shown for several other drugs of unrelated structure and mode of action (Table 1). In contrast, a taxol-resistant J774.2 cell subline, obtained by growing the cells with taxol (22), displayed a much higher resistance to taxol (833-fold), and showed clearly weaker cross-resistance to colchicine (58-fold), vinblastine (43-fold), puromycin (97-fold), doxorubicin (128-fold), and dactinomycin (88-fold). These cells exhibited a decreased accumulation of taxol in comparison with sensitive cells, which could be correlated with the presence of a 135-kd phosphoprotein (22).

The multidrug-resistant phenotype of tumor cells, even when mediated only by Pgp overexpression (i.e., with no other known resistance mechanism), does not display constant features: Each Pgp-expressing multidrug-resistant cell line can display its own pattern of cross-resistance (7,8). Therefore, it could be expected that a given multidrug-resistant cell line may also show different degrees of chemosensi-

tization by different chemosensitizers for different drugs.

Distinct drug binding–transport sites on the Pgp might indeed exist for different drugs or groups of drugs. Colchicine does not inhibit the in vitro photolabeling of Pgp by a vinblastine analog, whereas vinca alkaloids, verapamil, and diltiazem were able to do so (23,24). Both calcium channel blockers, verapamil and diltiazem, were also able to inhibit Pgp photolabeling by a cyclosporine analog, whereas colchicine in similar concentrations had no effect (25). Thus, the colchicine binding–transport site on Pgp might be distinct from the vinca alkaloid site.

In a recent study, the two most commonly used chemosensitizers, verapamil and quinidine, could not restore the retention of rhodamine 123 in the same multidrug-resistant P388 cell line as that used in the present study (26), although they could restore daunomycin retention (16); in contrast, amiodarone, cyclosporin A, SDZ 280-446, and SDZ PSC 833 could restore the retention of both Pgp-function probes (16,26). These results show that different chemosensitizers may differently affect drug transport by Pgp molecules.

Besides this variable efficacy of chemosensitizers mediated by the wild-type Pgp of multidrug-resistant cells, an additional variability in the gains of sensitivity may depend on Pgp polymorphism resulting from somatic mutations in response to selective pressure. Typically, a series of multidrug-resistant KB epidermoid carcinoma (ATCC CCL17) cell lines, which had been obtained by independent selection with colchicine, vinblastine, or doxorubicin, displayed a definitely preferential relative resistance to the drug used for selection, although they displayed a broad cross-resistance to the other drugs (8). Single point mutations sufficient to dramatically affect the profile of resistance conferred by the wild-type Pgp, such as a marked alteration of the relative resistance to colchicine, vinblastine, and doxorubicin, were caused by a single glycine–valine interchange (27). Similarly, mutations in the hamster pgp1 gene conferred a distinctive multidrug-resistant profile to dactinomycin-selected sublines of multidrug-resistant DC/3F cells: Highest resistance was now for dactinomycin in-

stead of colchicine as in the parental line (28). Thus, while one mutation increased colchicine resistance in one instance (27), a different mutation decreased it in the other (28). Finally, multidrug-resistant J774.2 sublines selected by growth in the presence of colchicine, vinblastine, or taxol developed heterogeneous forms of multidrug resistance-associated glycoproteins (29-31). Such glycoproteins of the taxol-resistant sublines were found to be similar but not identical to the classic Pgp expressed in vinblastine- and colchicine-resistant sublines derived from the same parent (29,30).

Eventually, the taxol-specific molecular variants (24,25) of the multidrug resistance-associated glycoproteins might display altered chemosensitization profiles in comparison with the bona fide mdr1-encoded Pgp, although the taxol resistance of resistant CHO cells could be reversed by verapamil (32). In this respect, it would thus be interesting to study how active SDZ PSC 833 and SDZ 280-446 are with such taxol-induced variants of multidrug resistance-associated glycoproteins.

In conclusion, precisely how taxol is processed by Pgp molecules remains unknown. Nonetheless, tumor cell resistance to taxol can be abrogated by either the cyclosporine SDZ PSC 833 or the cyclopeptolide SDZ 280-446. The combination of taxol with either SDZ PSC 833 or SDZ 280-446 may be recommended for the treatment of multidrug-resistant cancers.

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Notes

Growth Inhibition of Human Lymphoma Cell Lines by the Marine Products, Dolastatins 10 and 15

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Background: Dolastatins 10 and 15
are small peptides isolated from the
marine sea hare *Dolabella auricu-
laria*. In vitro studies of these pep-
tides have demonstrated antimitotic
and antiproliferative activity and
growth inhibition in hematopoietic
progenitor cells. *Purpose:* The pur-
pose of our in vitro study was to
determine the biological effects of
these marine peptides on growth of
human lymphoma cell lines and to
investigate mechanisms by which the
dolastatins may act. *Methods:* Cell
lines DB, HT, RL, and SR were
grown from the ascites or pleural
effusion of four patients with lym-
phoma. The DB, HT, and RL cell
lines are of B-cell origin, and the SR
cell line appears to be a less differen-
tiated lymphoid cell type. Cells from
these lines were cultured in the
presence of vincristine or dolastatin
10 or 15. [^{3}H]Thymidine-uptake as-
says were used to measure effects on
DNA synthesis. Cell cycle analysis
using propidium iodide was per-
formed to measure drug-induced cell-
cycle arrest. DNA fragmentation was
used as an assay for drug-induced
apoptosis and was measured by
agarose gel electrophoresis. *Results:*
In the three B cell lines, dolastatin 10
was more effective than dolastatin
15. Values for concentrations re-
quired for inhibition of proliferation
by 50% (IC_{50}) were .00013-.0013 nM
for dolastatin 10 in each cell line;
values for dolastatin 15 were approx-
imately .13 nM in DB and HT cells
and .0013-.013 nM in RL cells. SR
cells were more sensitive to dola-
statin 15 than to dolastatin 10 (IC_{50}
= .00013-.0013 nM versus .0013-.013
nM). Both dolastatins arrested more
than 70% of cells in mitosis in all cell
lines. This effect was reversed if the
drug was removed by 4 hours, but by
8 hours of exposure, reversal was not
possible. Both dolastatins 10 and 15
produced apoptosis in DB and HT
cells but not in the other two cell
lines. *Conclusions:* We have demon-
strated that dolastatins 10 and 15
have a profound antiproliferative
effect on four different human
lymphoma cell lines and that the
dolastatins are approximately 3-4
logarithms more effective as anti-
proliferative compounds, on a molar
basis, than vincristine—a clinically
useful, antiproliferative agent. These
data support the hypothesis that
apoptosis, as measured by DNA
fragmentation, appears to be a cell-
specific response and may not be
directly related to the antimitotic
effect of the dolostatins. *Implications:*
Our results suggest that these com-
pounds may be good candidates for
development as antineoplastic agents.
[J Natl Cancer Inst 85:483–488,
1993]

In the past few years, there has been
an intensive search for new anti-
neoplastic agents derived from natural
plant or animal sources. These efforts
have revealed many potentially useful
new compounds isolated from marine
animals, including the bryostatins (*1-3*),
halichondrin B and homohalichondrin
B (*4*), and the dolastatins (*5-8*). Among
these natural compounds, the dola-
statins appear to have the greatest
therapeutic potential. The dolastatins
are small peptides or depsipeptides
isolated from the East Indian Ocean sea
hare, *Dolabella auricularia*; at least 15
unique molecules have been identified
(*5-8*).

In this report, we have analyzed the
biological effect of two of these
unusual peptides, dolastatins 10 and 15
(*5,6*), on a panel of human lymphoma
cell lines. Dolastatin 10 consists of four
amino acid residues (dolavaline, valine,
dolaisoleuine, and dolaproine) and has