therapeutics directed against topoisomerases have a high initial efficiency in the treatment of cancer. However, an important problem is the development of resistance towards the employed compounds. The phenomenon is often associated with resistance towards a number of compounds, giving the cells a multidrug resistant (MDR) phenotype. One form of multidrug resistance is called altered topoisomerase MDR (at MDR), and is either caused by a reduced level of topoisomerase in the cells or by functional alterations in the enzymatic properties of the topoisomerase, rendering it drug resistant. Using topoisomerase from both resistant and sensitive cell lines, the alterations in enzymatic properties can be investigated. For these studies a new technique has been developed which allows investigation of the effect of topoisomerase inhibitors on the individual steps comprising the catalytic cycle of the enzymes. Profound differences have been found in the mode of action of different topoisomerase targeting agents.[1–4]

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Isolation of non-P-glycoprotein mediated multidrug resistant cells from human KB cells and reversal of the resistance

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Human KB cell lines resistant to high levels of adriamycin were isolated in a selection medium containing increasing concentrations of adriamycin, 1 μg/ml cepharanthine and 100 nM mezerein. The adriamycin-resistant KB cell line, C-A500, was cross-resistant to the standard drugs in the classical multidrug resistance phenotype, such as vincristine, actinomycin D, VP-16 and colchicine. The accumulation of adriamycin and vincristine was reduced in C-A500 cells but not in its revertant, C-AR, compared with that in parental KB cells. Adriamycin accumulation in C-A500 cells was decreased. The efflux of adriamycin from C-A500, but not from the revertant cells, was enhanced compared with that from KB-3-1 cells. These adriamycin resistant KB cells did not contain detectable levels of P-gp and did not overexpress MDR1. Multidrug resistance-associated protein (MRP) and MRP mRNA was expressed in the adriamycin-resistant KB cells, C-A12O, C-A500 and CA-1, but not in parental KB-3-1 and revertant C-AR cells. DNA topoisomerase II levels were considerably decreased in C-A500 and CA-1 and slightly decreased in C-A120. Cepharanthine and verapamil that reverse multidrug resistance in classical MDR cells could not reverse the drug resistance in C-A500 cells. However, a pyridine analog, PAK-104P, considerably increased the sensitivity of C-A500 cells to adriamycin. These results indicate that MRP overexpressed in the resistant cells may actively efflux adriamycin and vincristine from the cells and that both the increased expression of MRP and decreased level of topoisomerase II are responsible for the drug resistance in C-A120, C-A500 and CA-1 cells.

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The role of intercellular interactions in the development of tumor multidrug resistance

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The role of cellular interaction in the sensitivity of cultured cell populations to colchicine (CH) and in efficiency of P-gp function was studied. Mixtures of CH-resistant and CH-sensitive Djungarian hamster tumor cells were treated with CH and sensitivity to it was measured by colony forming assay. Comparisons were made with unmixed populations. Identification of individual subpopulations was possible via a genetic marker, cell resistance to 6-thioguanine (6-TG). CH-sensitive, 6-TG-sensitive cells had an HGPRT-positive phenotype detected by the ability of the cells to grow in HAT medium. CH-resistant with P-gp-mediated MDR, 6-TG-resistant (HGPRT-negative) variants could grow in 6-TG supplemented medium, but not in HAT. The survival of CH-sensitive cells in the CH supplemented medium was significantly increased after cocultivation with resistant counterparts for 3 days. To measure P-gp function the fluorescent dye rhodamin-123 (RH123) and FACScan analysis was used. P-gp-mediated RH123 efflux increased after 3 days' co-cultivation of sensitive cells with drug resistant variants. These data suggest that P-gp expressing cells in mixed populations influence both cell sensitivity to cytostatic cells and the function of P-gp in sensitive cells. The information may be useful for understanding of the development of multidrug resistance in tumors and new approaches to chemotherapy of human cancer.

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Role of MDR1 gene and P-glycoprotein expression in chemoresistance of human melanoma cell lines

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Metastatic malignant melanoma is considered a chemotherapy-refractory malignancy. A few previous studies have