Do Anti-P-Glycoprotein Antibodies Have a Future in the Circumvention of Multidrug Resistance?

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Discovering how to circumvent or overcome cancer drug resistance has become the Holy Grail of experimental chemotherapists and clinicians. It is clear that patients' tumors can become resistant to all the therapies they receive, and outwitting the tumor cells' resistance mechanisms has become an increasingly difficult challenge. In recent years, we have learned that the phenomenon of multidrug resistance, characterized by cross-resistance to many natural product anticancer drugs, decreases drug accumulation and retention and that P-glycoprotein (Pgp) expression (1) may be an important factor in some of the clinical failures of cancer chemotherapy. A number of tumors have been shown to express Pgp either de novo or after chemotherapy (2,3), and recent results suggest that Pgp expression may be an important indicator of therapeutic outcome (4). While non-Pgp forms of natural-product multidrug resistance have been described experimentally (5,6), they have not yet been shown in patients' tumors, principally because appropriate reagents that detect them are presently unavailable. Accordingly, efforts directed at circumventing multidrug resistance are focused on inhibition or modulation of Pgp function or expression and include screening and rational design of Pgp-inhibitory chemicals, as well as the use of modulator combinations, inhibitory antibodies, and antisense oligonucleotides (7,8).

Most experimental and clinical efforts to circumvent multidrug resistance have used drugs such as verapamil, quinidine, amiodarone, and cyclosporine that likely block the efflux function of Pgp (8). This approach has not yet proven satisfactory in the clinical setting in part because of the limitation of available modulators and because of their toxic effects (9,10). Planned or in progress are clinical trials of compounds (11,12) that appear to have a far more favorable therapeutic index than some of the "first generation" modulators (such as verapamil) and may be effective in maximizing the chemotherapy of drug resistant disease associated with the expression of Pgp.

Among the other possibilities to circumvent Pgp-associated multidrug resistance, several would seem to hold promise for the near future. Application of modulator combinations, which has provided some encouraging preliminary results in cell culture (13,14), has the advantage of maximum inhibition of Pgp without the dose-limiting toxic effects seen with single modulator treatment. This approach, however, may also suffer from lack of specificity and nonspecific toxicity. By contrast, the use of anti-Pgp antibodies to circumvent Pgp-multidrug resistance offers the prospect of specificity, since the antibodies should only target Pgp; toxicity would only be that associated with the administration of protein. It is obvious that only an antibody that recognizes surface determinants of Pgp will have a chance of modulating Pgp-multidrug resistance in vivo.

Anti-Pgp antibodies have been administered in vivo alone, in combination with anticancer drugs, and coupled to a toxin. Tsuruo et al. developed two antibodies that recognize surface-exposed epitopes of Pgp (15) and showed that one, MRK-16 monoclonal antibody, blocked the growth of multidrug resistant human tumor xenografts in athymic mice (16). In this issue of the Journal, Pearson et al. (17) have taken the experiment one step further: using a transplantable human colon carcinoma cell line infected with a retrovirus carrying a complementary DNA containing the MDR1 gene (also known as Pgy1), the authors also showed that MRK-16 monoclonal antibody was able to overcome Pgp-multidrug resistance in vitro and, more importantly, in xenografts in vivo when used in combination with vincristine. Of interest, neither the antibody alone nor vincristine alone were able to enhance the survival of mice bearing the Pgp-expressing xenografts. A preliminary account of similar findings was reported last year by Rittman-Grauer et al. (18) using the other antibody that recognizes surface determinants of Pgp, HYB-241. By contrast, Fitzgerald et al. (19) coupled MRK-16 monoclonal antibody to ricin and showed that this immunotoxin was effective in selectively killing tumor cells that expressed Pgp.

While these studies provide some encouraging preliminary results, they raise many questions about specificity, toxicity, mechanism of action, and feasibility. For example, is anti-Pgp antibody a more specific modulator of Pgp-multidrug resistance than drugs such as verapamil or cyclosporine? It is true that this effector has only one target—Pgp—and presumably one mechanism of action, compared with the modulator drugs. However, while all tumor cells that express Pgp are likely to be targets for enhanced drug accumulation by this antibody, so too are the normal cells of such secretory tissues of the kidneys, liver, intestine, and adrenal glands, as well as the capillary endothelial cells of the brain, as they all highly express Pgp (20). This potential binding of the antibody to normal tissue Pgp raises an important question of systemic toxicity. What will be the consequences to the patient of inhibition of normal tissue Pgp? Will inhibition of normal cell Pgp by antibodies be more of a concern than inhibition with Pgp-inhibitory drugs? Can administration of the antibody be modeled pharmacokinetically to produce preferential inhibition of tumor cell Pgp? One mitigating factor in favor of tumor cell selectivity of anti-Pgp antibodies may be related to the fact that Pgp appears to be distributed on the luminal surfaces of normal secretory tissues (kidneys, adrenal glands, small intestine, etc.) and may not be accessible to the antibody (21), whereas it would be accessible to modulator drugs such as verapamil. Since anti-Pgp antibodies may have different inhibitory effects on tumor versus normal tissue Pgp because of differences in Pgp density and subcellular localization, antibody...
distribution, blood supply, etc., it will be important to understand the pharmacologic disposition of these agents.

Use of murine monoclonal antibodies as therapeutic agents or adjuncts to chemotherapy is not without other intrinsic problems associated with antibody administration, including rapid elimination from the body, generation of immune responses, and toxic reactions (22). One way to circumvent some of these problems is to use F(ab')2 fragments, and another is to use human or mouse-human chimeric antibodies constructed by recombinant methods that join the antigen-recognizing variable regions of the mouse antibody to the constant regions of a human antibody. Compared with murine antibodies, such chimeras resulted in an increase in plasma half-life, decreased immunogenicity, and decreased toxicity (23). An anti-Pgp chimera was constructed using the variable region of MRK-16 monoclonal antibody and shown to be more effective than MRK-16 monoclonal antibody itself in killing multidrug resistant cells (24).

The mechanism of MRK-16 monoclonal antibody cytotoxicity was shown to be related to complement- and antibody-mediated lysis (16), and this was the case for the MRK-16 monoclonal antibody chimeras as well (24). The mechanism by which this antibody enhances the therapeutic efficacy of vincristine in the study by Pearson et al. (17) is somewhat less clear. In its initial description by Hamada and Tsuruo (15), MRK-16 monoclonal antibody enhanced the cytotoxicity of vincristine and dactinomycin but not doxorubicin in a human leukemic multidrug resistant cell line, and appeared to be related to the antibody's differential effects on drug accumulation. Similar results were reported by Broxterman et al. (25) using MRK-16 monoclonal antibody against a multidrug resistant human ovarian carcinoma cell line, but in this study, drug accumulation, including doxorubicin, was increased by exposure of the multidrug resistant cells to verapamil with MRK-16 monoclonal antibody. It is curious that in these studies MRK-16 monoclonal antibody alone did not enhance doxorubicin accumulation or cytotoxicity in vitro, whereas Pearson et al. (17) note (but show no data) that doxorubicin cytotoxicity was enhanced by the antibody in their system. The differences in the two results may be due in part to dissimilarities in amounts of Pgp expression and corresponding degrees of resistance between the cell types used in each study. These contrasting results raise the possibility that therapeutic efficacy of the antibody may depend strongly on the amount of Pgp expression and degree of resistance of the tumor cell. Pgp presentation by tumor cells relative to that in normal tissues may also be important.

Not clear in any of these studies is how the antibody, which binds to an external epitope of Pgp, is able to enhance drug accumulation; antibody effects on drug efflux have not been described. Current dogma is that drugs such as anthracyclines, vinca alkaloids, and epipodophyllotoxins enter cells by passive diffusion, bind to Pgp, and are then effluxed from the cells by this protein (26). Preliminary studies from several laboratories suggest that drug binding sites on Pgp are on an internal (cytoplasmic) site (27,28). If so, then one must postulate that the anti-Pgp antibody, which binds to the cell surface, may somehow block the "pore" or "channel" likely formed by Pgp, thereby blocking drug efflux. Another explanation is that the antibody enhances drug accumulation, rather than blocks drug efflux, possibly through antibody-dependent or complement-mediated cytolytic mechanisms. Detailed studies of the mechanism of enhanced drug accumulation by anti-Pgp antibodies are needed.

Finally, having raised questions about specificity, toxicity, and mechanism of action, the issue of efficacy of anti-Pgp antibodies needs to be addressed. The effects of MRK-16 monoclonal antibody and HYB-241 to enhance drug cytotoxicity and accumulation in vitro are not dramatic: Pearson et al. (17) showed only a twofold decrease in vincristine cytotoxicity in vitro. However, the increase in mean survival time by the antibody-vincristine combination was 160% of controls, suggesting that large decreases in IC50 may not be necessary to achieve an effect in vivo and that other mechanisms, such as complement- or antibody-mediated cytotoxicity played a role in in vivo cytotoxicity. Antibody efficacy might be enhanced in vivo by the combined use of other modulators like cyclosporine or verapamil, as was done in vitro by Broxterman et al. (25). Efficacy will also be influenced by antibody specificity for tumor cells and its potential pathological effects on normal tissue Pgp. There is also the concern that the effect of anti-Pgp antibodies may be restricted to only a few tumor types or xenografts and will not have general applicability, since an antibody directed toward the same antigen in different tumors may have different effects (29). Moreover, different xenografts may have distinct effects on host immune responses that could influence the effect of the anti-Pgp antibody. Studies with different xenograft models are clearly warranted. Lastly, it must be noted that administration of anti-Pgp antibodies will not be useful against those malignancies that express different (i.e., non-Pgp) forms of multidrug resistance or in which Pgp is not expressed on the surface.

So, is it feasible to use anti-Pgp antibodies to modulate clinical Pgp-multidrug resistance? Will this approach be effective? The preliminary results in the paper by Pearson et al. (17) in this issue of the Journal raise more questions than they answer, but therein lies their significance. Since the goal of studies focused on the circumvention of multidrug resistance is the improvement of the therapeutic index of the anticancer drugs, all current approaches are needed. The success of present and future clinical applications of Pgp modulators depends to a large extent on the form of multidrug resistance expressed by the tumor and the innovative use of all strategies to modulate it (7). We will follow the development of this aspect of multidrug resistance modulation with great interest.

References


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(7) BECK WT: Strategies to circumvent multidrug resistance due to P-glycoprotein or to altered DNA topoisomerase II. Bull Cancer 77:1131–1141, 1990
(11) Thimmaiah KN, Horton JK, Qian XD, ET AL: Structural determinants of phenoxazine type compounds required to modulate the accumulation of vinblastine and vincristine in multidrug-resistant cell lines. Cancer Commun 2:249–259, 1990
(20) CORDON-CARDO C, O'BRIEN JP, CASALS D, ET AL: Multidrug-resistance gene (P-glycoprotein) is expressed by endothelial cells at blood-brain barrier sites. Proc Natl Acad Sci USA 86:695–698, 1989

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