Commentary

Liposome-based drug delivery in breast cancer treatment

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Abstract

Drug delivery systems can in principle provide enhanced efficacy and/or reduced toxicity for anticancer agents. Long circulating macromolecular carriers such as liposomes can exploit the 'enhanced permeability and retention' effect for preferential extravasation from tumor vessels. Liposomal anthracyclines have achieved highly efficient drug encapsulation, resulting in significant anticancer activity with reduced cardiotoxicity, and include versions with greatly prolonged circulation such as liposomal daunorubicin and pegylated liposomal doxorubicin. Pegylated liposomal doxorubucin has shown substantial efficacy in breast cancer treatment both as monotherapy and in combination with other chemotherapeutics. Additional liposome constructs are being developed for the delivery of other drugs. The next generation of delivery systems will include true molecular targeting; immunoliposomes and other ligand-directed constructs represent an integration of biological components capable of tumor recognition with delivery technologies.

Keywords: drug delivery, immunoliposomes, liposomes, monoclonal antibody, polymers

Introduction

Drug delivery systems offer the potential to enhance the therapeutic index of anticancer agents, either by increasing the drug concentration in tumor cells and/or by decreasing the exposure in normal host tissues. This commentary will discuss some of the recent trends in liposome-based drug delivery systems for breast cancer therapy.

Solid tumors such as breast cancer have historically provided many challenges to systemic therapy. Theoretical barriers to drug penetration in solid tumors include heterogeneous vascular supply and high interstitial pressures within tumor tissue, particularly in necrotic zones. Delivery systems can even exacerbate these problems due to the slow diffusion of macromolecular agents through tumor tissue.

However, delivery systems have been developed to exploit a feature of tumor microphysiology often referred to as the 'enhanced permeability and retention' effect [1]. This effect is a consequence of the dysregulated nature of tumor angiogenesis, which characteristically involves structural and physiologic defects leading to hyperpermeability. Macromolecular agents with highly restricted volumes of distribution and the capacity for greatly prolonged circulation will preferentially extravasate from these abnormal vessels and accumulate in tumor tissue. The leading examples of such 'passively targeted' agents include long circulating liposomal drugs.

Delivery of anthracyclines

Anthracyclines exemplify the case of potent anticancer activity that is constrained by highly problematic systemic toxicities. For this reason, the most studied drug delivery applications in oncology have involved anthracyclines to preserve or to enhance efficacy against tumor cells while limiting exposure to critical target sites such as myocardium and bone marrow.

mAb = monoclonal antibody.

Anthracyclines have been encapsulated in a number of liposomal constructs. Current versions utilize ion trapping methods to achieve extremely efficient loading of doxorubicin or daunorubicin within the aqueous interior of unilamellar (single bilayer) liposomes, reaching 10^4 drug molecules per liposome particle. Liposome-encapsulated doxorubicin (TLC D-99, Myocet™; Elan Pharmaceuticals, Inc., Cedar Knolls, NJ, USA) was developed as a multiple vial kit, with individual vials containing moderately sized (~190 nm) liposomes, lyophilized doxorubicin, and citric acid buffer. The components are mixed at point of care, resulting in highly efficient (>99%) loading of doxorubicin into the liposomes. Myocet™ provides a limited degree of prolonged circulation as compared with free drug. However, the liposome encapsulation significantly alters the biodistribution of doxorubicin, resulting in some reduction in toxicities.

A randomized trial of Myocet™ versus doxorubicin, with both in combination with cyclophosphamide for first-line treatment of metastatic breast cancer, showed comparable response rates for the two regimens. However, Myocet™ was associated with a decreased frequency of cardiotoxicity and neutropenia [2]. While the decrease in toxicities was clearly shown, there remains controversy as to whether Myocet™ has demonstrated equivalent efficacy to doxorubicin [3].

Liposomes capable of markedly prolonged circulation and enhanced tumor accumulation have been developed, using a series of modifications to liposome structure that act to retard uptake by mononuclear phagocytes. Liposomal daunorubicin (DaunoXome®; Gilead Sciences, Inc., Forest City, CA, USA) provides extended circulation with its very small size (~45 nm) and its rigid bilayer, and it is clearly active against Kaposi's sarcoma and other tumors [4].

Pegylated liposomal doxorubicin (Doxil®, Caelyx®; Alza Pharmaceuticals, San Bruno, CA, USA) has achieved the most prolonged circulation to date, with a terminal half-life of 55 hours in humans. These are small (~100 nm) and rigid liposomes, onto which a polymeric coat of polyethylene glycol has been grafted. These pegylated (also referred to as sterically stabilized or 'Stealth') liposomes display inhibited interaction with plasma proteins and mononuclear phagocytes, and they consequently display greatly prolonged circulation time.

These pharmacokinetics in turn facilitate accumulation in tumor tissue, which has been demonstrated in assays of tumor biopsies in Kaposi's sarcoma [5] and skeletal metastases of breast cancer [6]. Furthermore, the altered biodistribution and pharmacokinetics substantially mitigate many of the typical toxicities of bolus doxorubicin administration, such as nausea and alopecia, while inducing alternate toxicities. The most notable of these is palmar–plantar erythrodysesthesia ('hand–foot syndrome'), which has generally been associated with continuous infusion schedules of doxorubicin rather than bolus administration. Palmar–plantar erythrodysesthesia is the usual dose-limiting toxicity of Doxil®, and it renders problematic any substantial dose escalation that liposome delivery might otherwise provide.

Doxil® has shown efficacy against a number of solid tumor types, including Kaposi's sarcoma and refractory ovarian cancer. In metastatic breast cancer, phase II trials of Doxil® monotherapy as first-line or second-line treatment have observed response rates of 31% [7]. Ongoing phase III studies include a direct comparison of Doxil® versus doxorubicin in metastatic breast cancer. Polychemotherapy studies involving Doxil® have included Doxil® plus cyclophosphamide as first-line treatment of metastatic breast cancer, which was associated with a response rate of 65% [8]. Doxil® has also been tested in combination with gemcitabine [9] or docetaxel [10], with these regimens showing good tolerability and activity.

The biodistribution of liposomes can reduce the cardiotoxic effects of anthracyclines. As already mentioned, Myocet™ has demonstrated reduced cardiotoxicity in combination with cyclophosphamide. A study of Doxil® in which cardiac biopsies were performed in 10 treated patients observed significantly less histopathologic evidence of myocardial injury than in matched patients receiving somewhat higher doses of free doxorubicin [11]. Anthracycline cardiotoxicity has attracted renewed interest with the advent of trastuzumab, a recombinant humanized mAb against the product of the HER2 (erbB2, neu) proto-oncogene [12]. Trastuzumab in combination with doxorubicin or epirubicin is highly efficacious but is associated with an extremely high frequency of cardiotoxicity [12]. For this reason, clinical trials are evaluating the combination of trastuzumab with liposomal anthracyclines, including two separate trials with Myocet™ and Doxil®.

Polymer-based drug delivery represents another strategy to favorably alter the pharmacokinetics and biodistribution of derivatized drugs. One example is PK-1, a conjugate of the water-soluble, acrylamide-based, copolymer *N*-(2-hydroxypropyl)methacrylamide and doxorubicin [13]. Polymer-based anthracyclines have not achieved the same degree of prolonged circulation as sterically stabilized liposomal doxorubicin. However, their circulation does significantly surpass that of free doxorubicin, resulting in tumor accumulation via the 'enhanced permeability and retention' effect.

The optimal role for liposomal anthracyclines in breast cancer treatment remains to be determined. The most prominent feature of these agents has been a decrease in many of the toxicities of anthracycline therapy rather than an increase in potency. Hence, liposomal anthracyclines

(particularly Doxil®, based on its pharmacology and existing clinical data) could be considered for inclusion in first-line or second-line regimens in place of standard anthracyclines. Greater utility may be achieved via molecular targeting for increased potency (discussed later).

Liposomal delivery of other drugs

The most active drugs against breast cancer are currently the anthracyclines and taxanes (paclitaxel and docetaxel). Strategies for the delivery of taxanes are under active investigation to increase tumor exposure and/or to reduce adverse effects such as neurotoxicity, edema, asthenia, and alopecia. In addition, special issues with the taxanes provide further rationale for application of delivery systems. Both paclitaxel and docetaxel are poorly soluble in aqueous solutions, and have consequently been formulated with vehicles Cremaphor EL and polysorbate 80 (TWEEN), respectively. These formulations are highly allergenic, require extensive premedication, and are responsible for most of the acute toxicities observed with taxane therapy, rather than the taxanes themselves. Delivery strategies in clinical trials include liposome-encapsulated paclitaxel [14] and poly(L-glutamic acid)–paclitaxel, a polymer conjugate [15].

Other applications for delivery systems in breast cancer include approved chemotherapy drugs such as vinca alkaloids, platinums, and camptothecins. In each case, it is possible that delivery systems such as liposomes or polymers could improve pharmacokinetics, could increase tumor accumulation, and/or could reduce limiting toxicities.

While delivery systems for standard anticancer compounds may increase their clinical utility, there is also intense interest in developing delivery strategies for novel anticancer agents that cannot be used by themselves as drugs. Delivery systems can potentially overcome many common pharmacologic problems such as those involving solubility, *in vivo* stability, pharmacokinetics, tumor uptake, and toxicity. The increasing repertoire of sophisticated delivery systems may thus allow new classes of potent anticancer agents to reach clinical application. This includes not only drug delivery, but also liposome-derived systems for nucleic acid-based agents, such as antisense oligonucleotides and gene therapy constructs.

Towards molecularly targeted drug delivery

As discussed, currently approved liposomal drug delivery systems provide stable formulation, provide improved pharmacokinetics, and provide a degree of 'passive' or 'physiological' targeting to tumor tissue. However, these carriers do not directly target tumor cells. The design modifications that protect liposomes from undesirable interactions with plasma proteins and cell membranes, and which contrast them with reactive carriers such as cationic liposomes, also prevent interactions with tumor cells. Instead, after extravasation into tumor tissue, liposomes remain within tumor stroma as a drug-loaded depot. Liposomes eventually become subject to enzymatic degradation and/or phagocytic attack, leading to release of drug for subsequent diffusion to tumor cells. The next generation of drug carriers under development features direct molecular targeting of cancer cells via antibody-mediated or other ligand-mediated interactions.

Immunoliposomes, in which mAb fragments are conjugated to liposomes, represent a strategy for molecularly targeted drug delivery (for a review, see [16]). Anti-HER2 immunoliposomes have been developed with either Fab′ or scFv fragments linked to long circulating liposomes [17]. In preclinical studies, anti-HER2 immunoliposomes bound efficiently to and internalized in HER2-overexpressing cells, resulting in efficient intracellular delivery of encapsulated agents [17–20]. Anti-HER2 immunoliposomes loaded with doxorubicin displayed potent and selective anticancer activity against HER2-overexpressing tumors, including significantly superior efficacy versus all other treatments tested (free doxorubicin, liposomal doxorubicin, free mAb [trastuzumab], and combinations of trastuzumab plus doxorubicin or liposomal doxorubicin) [21]. Anti-HER2 immunoliposomes are currently undergoing scale up for clinical studies.

The immunoliposome approach offers a number of theoretical advantages as compared with other antibody-based strategies. Anti-HER2 immunoliposome delivery of doxorubicin may circumvent the prohibitive cardiotoxicity associated with combined trastuzumab plus doxorubicin treatment. Anti-HER2 immunoliposomes can be constructed using scFv that, unlike trastuzumab, lack antiproliferative activity, are incapable of antibody-dependent cellular cytotoxicity, and require threshold levels of HER2 expression for delivery [20,21]. In contrast to drug immunoconjugates, which consist of a small number of drugs (typically <10 drugs per mAb) directly coupled via linkers to selected residues on the mAb, immunoliposomes exploit the exponentially greater capacity of drug-loaded liposomes (up to 10^4 drugs per liposome). Immunoliposomes also appear to be nonimmunogenic and capable of long circulation even with repeated administration.

Antibody-based targeting is also being developed in conjunction with polymer systems. Similarly, ligand-based targeting using growth factors, hormones, vitamins (e.g. folate), peptides or other specific ligands is being pursued in conjunction with both liposomes and polymers.

Breast cancer involving the chest wall

A special problem in the management of advanced breast cancer is that of chest wall recurrence/metastasis, which is typically highly morbid and difficult to treat. Photodynamic therapy, in which systemically administered

photosensitive compounds are activated by an external light source, can be considered a related strategy to drug delivery and is under investigation for the treatment of superficial breast tumors.

In a multimodality strategy, hyperthermia (which is currently used to enhance the efficacy of external beam radiation for chest wall metastasis) has been used to modulate delivery of liposomal drugs. In animal models, the application of hyperthermia to subcutaneous tumors concomitant with or prior to intravenous administration of long circulating liposomes results in a marked increase in liposome accumulation within tumor tissue [22,23]. The mechanism for this hyperthermia-enhanced liposome delivery appears to be via heat-induced changes in vascular permeability and microcirculatory dynamics, which further facilitate liposome extravasation from tumor vessels.

In a phase I/II clinical trial of metastatic breast cancer involving the chest wall, sequential hyperthermia followed by pegylated liposomal doxorubicin (Doxil®) was very well tolerated and produced objective responses in the majority of chest wall tumors [24]. It was notable that all patients had extensive prior treatment, and most had recurred or progressed after radiation and multiple chemotherapy regimens. Hyperthermia can also be used in conjunction with thermosensitive liposomes, which are being developed for heat-triggered release of encapsulated drug in tumor tissue [25].

Conclusions

Drug delivery systems are designed to stably and efficiently carry anticancer agents to tumor sites. This has been accomplished with some success by liposomal versions of anthracyclines. Current liposomal anthracyclines provide improved pharmacokinetics, provide reduced toxicities to a number of organ sites, and provide potentially increased tumor uptake. The next generation of delivery systems in development combine these features with tumor cell recognition, and include antibody-targeted and cell-internalizing systems. Such systems will enable drug delivery to move beyond pharmacokinetic-driven and biodistribution-driven mechanisms to true molecular targeting. This trend can also be viewed as an integration of biological therapeutics and drug delivery technologies. It is probable that such integrated approaches will include biologically targeted delivery systems for small molecule drugs as well as for biological agents with anticancer activity, and will be an increasingly important theme in the development of new treatments of breast cancer.

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