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Review

Localized paclitaxel delivery☆

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Abstract

While the search for new antineoplastic agents is in progress, optimization of delivery for existing drugs will remarkably improve the current scenario in the management of cancer. Paclitaxel, a new antineoplastic agent, is one such drug deserving attention in the field of regional drug delivery, offering immense pharmacokinetic as well as therapeutic advantage via localized delivery. The antiangiogenic activity of paclitaxel has been demonstrated using the chick chorioallantoic membrane model (CAM). This review focuses on the antiangiogenic activity of paclitaxel supported by the evidence that angiogenesis inhibitors display potential synergism with cytotoxic agents in the treatment of primary and metastatic cancers. Preclinical trials have confirmed that the biological and cytotoxic effects of paclitaxel on several tumor cell lines are enhanced by the increase in both the drug concentration and the duration of exposure. Sufficient experimental evidence has accumulated to state that localized delivery will exploit the multiple pharmacological effects of paclitaxel in the treatment of refractory and metastatic cancerous diseases. The drug delivery systems, namely, microspheres, surgical pastes and implants, fabricated for localized paclitaxel delivery are reviewed explaining the concept of increased tumor burden alleviating body burden as a consequence of such delivery systems. Some of the preclinical trials are very encouraging and speculate a promising future for these devices in the battle against solid tumors. Finally, the review briefs on the possibilities for better paclitaxel delivery and the future drug delivery systems for localized cancer chemotherapy.

Abbreviations: BBB, blood brain barrier; CAM, chick chorioallantoic membrane model; DSC, differential scanning colorimetry; EVA, ethylene vinyl acetate; GA, glycolic acid; GPC, gel permeation chromatography; IPM, isopropyl myristate; LA, lactic acid; MePEG, methoxy polyethylene glycol; MW, molecular weight; PCL, poly caprolactone; P(CPP-SA), poly[bis (*p*-carboxy phenoxy)propane-sebacic acid]; PDLLA, poly (D,L-lactic acid); PEG, poly ethylene glycol; P(FAD-SA), poly(fatty acid dimer-sebacic acid); PLA, poly lactic acid; PLGA, poly(L-glycolic acid); SEM, scanning electron microscopy.

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1. Introduction

Cancer may be defined as a disease characterized by uncontrolled multiplication and spread within the body of abnormal forms of body's own cells. There are three main approaches to deal with established cancer: surgical excision, irradiation and chemotherapy. Most anticancer drugs that are in clinical use are only antiproliferative but have no specific effect on invasiveness or the tendency to metastasize. Furthermore, they affect all rapidly dividing cells including normal tissues and show dose-limiting toxic effects. Hence, an alternative approach would be to use pharmacokinetic principles to optimize drug administration, such as targeted drug delivery.

Drug targeting can occur at different levels: first order targeting is increased delivery of drug to the body compartment harboring tumor; second order targeting is increased delivery of drug to tumor cells; and third order targeting implies selective intra-cellular delivery. Regional chemotherapy via localized drug delivery is a method of first order targeting by enhancing drug delivery to the organ or tissue harboring tumor. Localized drug delivery may be defined as a method of delivery of drug (for its local action) from a dosage form to a particular site in the biological system where its entire pharmacological effect is desired.

Usually in cancer therapy, surgical treatment is carried out for patients with resectable carcinoma. However, treatment failure due to local recurrence of primary tumors or metastatic spread often occurs during management. Hence, an integrated therapeutic approach is desired. In accordance with this concept, surgical adjuvant chemotherapy employing an anticancer agent during or immediately after surgery would be beneficial. A delivery system loaded with paclitaxel at tumor resection site will provide a high local concentration of the drug detrimental to malignant cells which may have survived surgery, thus preventing regrowth and metastasis of tumor.

The recent applications of molecular techniques to tumor biology has lead to an increasing understanding of some of the mechanisms underlying the crucial aspects of malignant phenotype, namely tissue invasion, angiogenesis and metastasis. An ideal molecule would be the one which can block all the underlying mechanisms involved in tumor pathogenesis (which is far from possible!!). However, the much promising paclitaxel aided by localized delivery seems to fit the situation better than the other currently available drugs. An ideal delivery system is one which not only improves patient compliance but also capitalizes on, and magnifies the nature-gifted versatility of paclitaxel eventually resulting in the termination of cancer.

2. Pathophysiology of cancer and paclitaxel drug delivery system

While drug delivery is by no means the only factor affecting the success or failure of therapy, attaining adequate drug levels at the tumor cell is of primary importance because inadequate tumor cell drug-burden will lead to low cell kill and to the potential for the early development of resistance to the drug.

In the eradication of cancer, one of the most difficult tasks of chemotherapy is extending the cytotoxicity of agents to tumor cells in G_0 resting phase. The unique mechanism of action of paclitaxel, i.e. stabilization of already existing microtubules and promotion of microtubule assembly (Schiff et al., 1979; Kuhn, 1994) makes the tumor cell in the resting phase relatively more susceptible to its cytotoxic effect, though to a lesser extent than those cells in G_2/M phases (Donaldson et al., 1994). Since tumor growth, progression and metastasis are angiogenesis dependent, the antiangiogenic property of paclitaxel must also be simultaneously exploited when cells are in dormant state, so that by the time they enter G_1, G_2 and M phases of cell cycle, vasculature is sufficiently

inhibited, making the environment unfavorable for cell growth and proliferation.

2.1. Ternary diagram

The multiple pharmacological effects of paclitaxel, namely, antiproliferative, antiangiogenic (Klauber et al., 1997), antimetastatic (Stearns and Wang, 1992) and apoptotic (Yen et al., 1996) properties, can be better explored and exploited via localized delivery as compared to systemic delivery. Targeting vasculature would be beneficial clinically due to lack of development of resistance by endothelial cells to antiangiogenic therapy (Augustin, 1998) and is complementary to existing therapies. Cytotoxic agents which act on dividing tumor cells at the growing edge of tumor mass are ineffective against tumor cells in the center of solid tumors, and antiangiogenics destroy tumors from the inside-out. It is imperative that all the attributes of the new drug molecule, like paclitaxel, are transferred to the drug delivery system for successful cancer chemotherapy. This phenomenon is depicted in the ternary diagram (Fig. 1), where each apex of the triangle represents one attribute of paclitaxel molecule. The area occupied by paclitaxel molecule in the diagram shows a perfect balance in the simultaneous display of its three attributes via localized delivery as against systemic delivery where only the antiproliferative activity of paclitaxel is transferred in appreciable magnitude to the drug delivery system. The concept is further explained by considering each of pharmacokinetics, antiangiogenic and antimetastatic effects of paclitaxel.

3. Pharmacokinetic considerations of localized drug delivery

Regional chemotherapy through localized drug delivery is based on the premise that anticancer agents display a steep dose response for both therapeutic effect and toxicity. Paclitaxel exhibits dose-dependent cytotoxicity, antimetastatic and antiangiogenic activities and the advantages of localized paclitaxel delivery are summarized in Table 1.

The regional drug exposure advantage (R_d) for agents with linear pharmacokinetics is related to the rate of drug elimination in the rest of the body and the blood perfusion rate into the target region (tumor) by the formula (Eckman et al., 1974; Dedrick, 1988):

Table 1
Localized paclitaxel delivery

Advantages	Comments
(a) Bone marrow suppression and cardiac rhythm disturbances are alleviated (Boye et al., 1995; Perez, 1998)	Greater tumor cell burden and reduced body burden of drug
(b) Less dose of drug is required to fill up the volume of distribution	Paclitaxel is extremely protein bound (95–98%) (Sonnichsen and Relling, 1994)
(c) First pass through the tumor compartment	Paclitaxel is extremely lipophilic and lipophilic drugs exhibit high extraction ratios (Ohkouchi et al., 1990)
(d) Antiangiogenic and antimetastatic properties of paclitaxel are dose dependent (Markman et al., 1992; Burt et al., 1995)	Angiogenesis preceeds metastasis
(e) Paclitaxel has dose and route-dependent pharmacokinetics (Gianni et al., 1995)	Prediction of plasma and tumor drug concentration profiles is difficult
(f) Induction of apoptosis by paclitaxel is concentration dependent (Yen et al., 1996)	Apoptotic fraction increases linearly with the logarithm of paclitaxel concentration
(g) Paclitaxel affect cells in G_0 phase as a function of exposure time (Donaldson et al., 1994)	It promotes polymerization of tubulin dimers into non-functional microtubules in absence of GTP and MAP
(h) Partially reduces or even obviates the need for premedication	Improved patient compliance and reduced cost of therapy

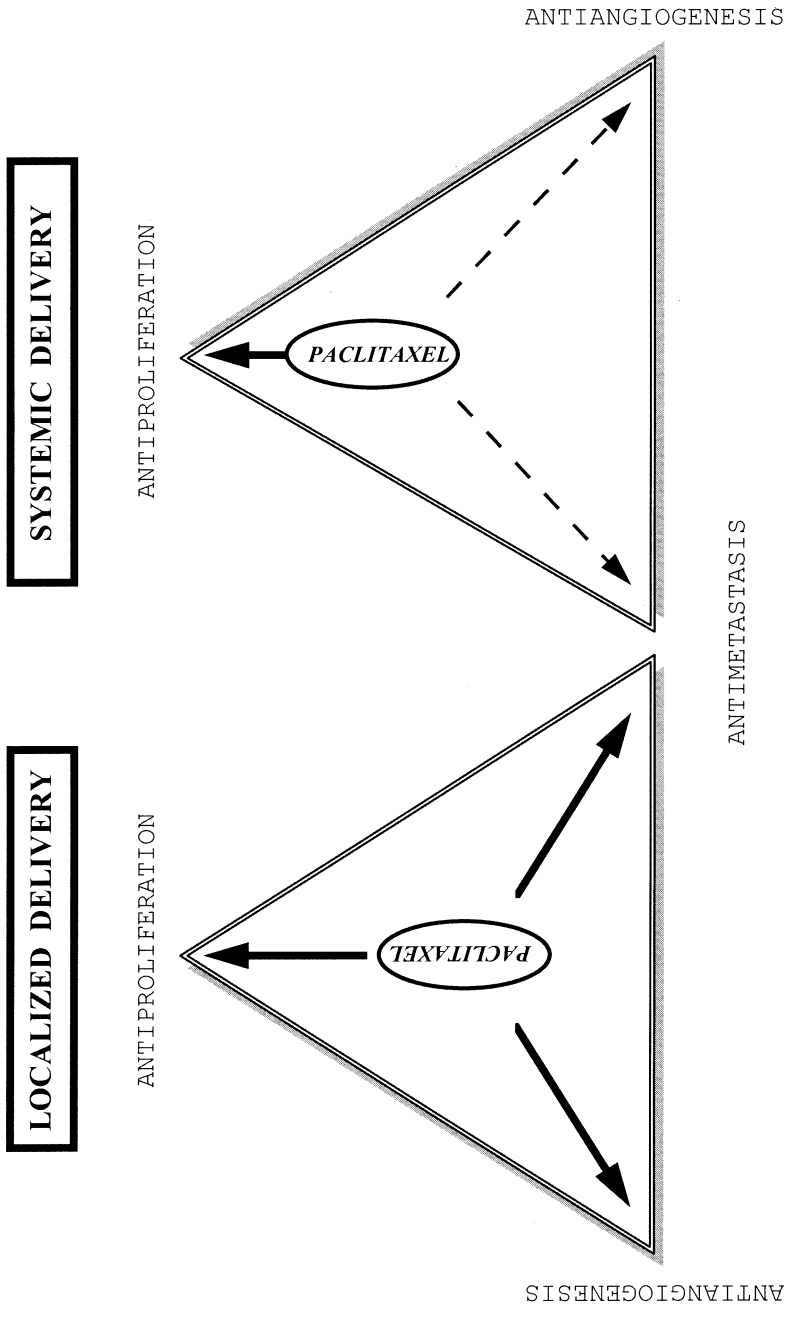


Fig. 1. Ternary diagram for paclitaxel delivery system.

regional advantage, $R_d = 1 + \frac{CL_T}{Q_R(1 - E_R)}$

where, CL_T is the total body drug clearance, Q_R is blood perfusion rate of the tumor and E_R is extraction ratio of drug by tumor.

The implications of the equation are that: (i) an ideal candidate for localized delivery would be a drug having high clearance rate and tumor extraction ratio; and (ii) an ideal drug delivery system would be the one which not only releases drug at the required rate but also practically holds all released doses at tumor the site by decreasing the blood perfusion of the tumor. The disposition of paclitaxel in vivo is characterized by a biexponential model. Alpha and beta half-lifes are rather short ranging from 0.27 to 0.32 h (mean $= 0.29$) and 1.3–8.6 h (mean $= 5.0$), respectively (Rowinsky et al., 1990) and the molecule is extremely lipophilic ($\text{Log } P_{O:W} = 3.5$, (Heimans et al., 1994)). The above values makes R_d of localized paclitaxel delivery apparent. The pharmacokinetic advantage of localized paclitaxel delivery is related to pharmacokinetic analysis in Fig. 2. The drug is extracted by the tumor during its first pass through the tumor compartment resulting in increased tumor drug-burden. Hence, the amount of drug available into systemic circulation and to peripheral compartment for toxic effect is considerably reduced alleviating the body burden due to drug.

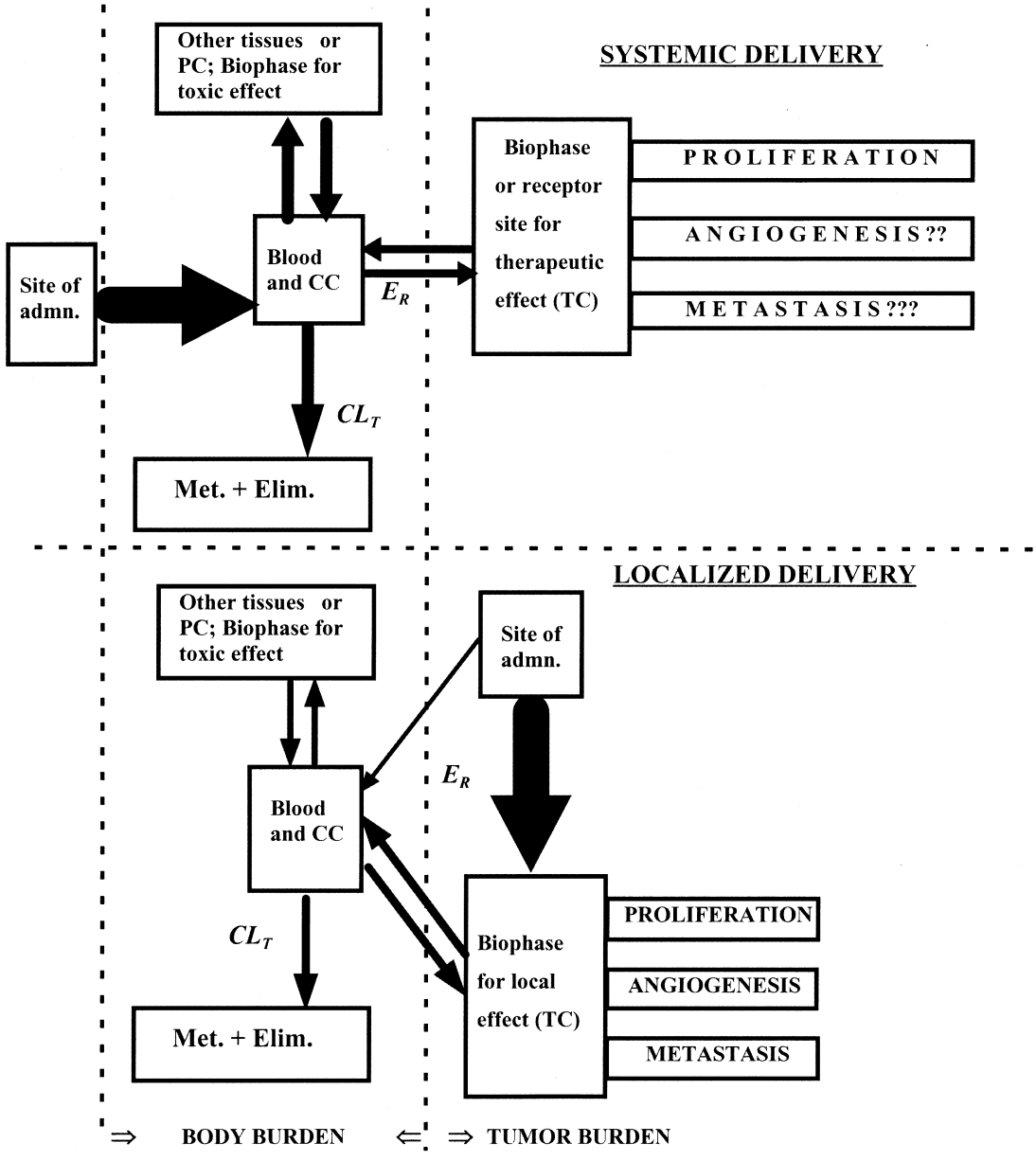
4. Angiogenesis and cancer

Angiogenesis may be defined as development of new blood vessels and it is a well established fact that tumors are angiogenesis dependent (Table 2). Just as normal organ development and growth is dependent on blood supply, so is the tumor for which reason it neovascularizes. Endothelial cell, a primary cell type in tumor micro-vasculature, is therefore, a new potential target to fight cancer. From a therapy point of view, it is advantageous to view the tumor in terms of two distinct cell populations, namely, tumor cell population and an endothelial cell population, wherein the multiplication of each cell type must be prevented. Endothelial cells of the parent vessel are stimulated to degrade the endothelial basement membrane (Ausprunk and Folkman, 1977), migrate into the perivascular stroma where they initiate a capillary sprout which subsequently expands into a tubular structure (Kalebic et al., 1983). A number of such tubular structures are organized into a functioning circulatory network. Migrating endothelial cells produce type IV collagenase and other proteinases for the degradation of extracellular matrix. Hence, specific inhibitors of these enzymes block endothelial cell invasion and angiogenesis (Mignatti et al., 1989).

4.1. Antiangiogenic response of paclitaxel

Paclitaxel has potent dose-dependent antiangiogenic activity as demonstrated by experiments using chick chorioallantoic memebrane (Burt et al., 1995; Dordunoo et al., 1995; Winternitz et al., 1996; Dordunoo et al., 1997), though the actual mechanism has not been clearly elucidated. Studies with transfilter coculture system that imitate normal vessel wall were very promising with paclitaxel; brief single dose exposure of paclitaxel to human endothelial cells and human smooth muscle cells (0.1–1 μmol; 20 min) exhibited a long-lasting antiproliferative effect (Axel et al., 1997), an important step in angiogenesis inhibition.

Combination therapy involving antiangiogenic agent with antiproliferative agents has shown enormous synergistic effect (Teicher et al., 1992; Kamei et al., 1993). Remodelling and growth of vasculature is a normal physiological process, as in wound healing and ischaemia (Tai-Ping et al., 1995) compelling us to look at tumor specific antiangiogenic agents. Moreover, a given dose of an antiangiogenic agent when given by systemic administration showed little antiangiogenic effect at tumor site, whereas the same dose when given by local delivery was very effective (Yanai et al., 1995). Since paclitaxel has both antiproliferative and antiangiogenic properties, localized paclitaxel delivery should display the dual advantage synergism.



CC=CENTRAL COMPARTMENT; PC=PERIPHERAL COMPARTMENT; TC=TUMOR COMPARTMENT

Fig. 2. Schematic diagram illustrating the pharmacokinetic advantage and reduced body burden of drug for localized paclitaxel delivery relative to systemic delivery. The thickness of arrows indicate the relative magnitude of burden and the length of the bar is inversely proportional to the inhibitory effect of drug on each of the events.

5. Metastasis and cancer

Experimental evidence suggests that metastasis is angiogenesis dependent (Lien and Ackerman, 1970). For a tumor cell to successfully metastasize, it must breach several barriers and be able to respond to specific growth factors. Briefly, tumor cells after gaining access to the vasculature in the

Table 2
Summary of the evidence that tumor growth, progression and metastasis is angiogenesis dependent[a]

Indirect evidence	
Angiogenesis is one of the many factors necessary for tumor to metastasize	Liotta et al., 1974
The diameter of tumors implanted on chick chorioallantoic membrane increased several-fold after the transition from avascular phase to vascular phase	Knighton et al., 1977
In transgenic mice, the progression from normal cells to hyperplasia to neoplasia was delayed until the onset of angiogenesis	Folkman et al. (1989)
The probability of appearance of distant metastasis of breast carcinoma increased with the vascularization of tumor	Weidner et al. (1991)
Angiogenesis is necessary both at the beginning and at the end of the cascade of events that promote metastasis	Fidler and Ellis (1994)
Tumors in isolated perfused organs showed limited growth due to the lack of proliferation of blood vessels. After transplantation into mice tumors vascularize and expand rapidly	Folkman et al. (1996)
Growth and metastasis of Bomirski Ab melanoma is suppressed by angiogenesis inhibitor TNP-470 in hamsters	Mysliwski et al. (1998)
Direct evidence	
A specific angiogenesis inhibitor, AGM-1470 inhibits tumor growth in-vivo but not in-vitro	Ingber et al. (1990)
Monoclonal antibody directed against bFGF causes remission of tumor size in mice	Hori et al. (1991)
Interferon-alfa induces remission of hemangioma in infants due to decreased FGF production	Ezekowitz et al. (1992)
Monoclonal antibody therapy against VEGF causes remission of tumor transplanted into mice	Kim et al. (1993)
A monoclonal antibody neutralizing the $\alpha_v\beta_3$ integrin blocks angiogenesis and tumor growth	Brooks et al. (1994)
Down-regulation of VEGF expression by adenovirus-mediated gene transfer inhibits angiogenesis in human colon cancer	Bouvet et al. (1998)

[a] Modified from Gasparini and Harris (1995).

primary tumor and surviving the circulation, arrest in the microvasculature of the target organ to make an entry. This is followed by induction of angiogenesis to establish itself in the target organ; angiogenesis is necessary at the beginning as well as at the end of the cascade of events. The metastatic process begins with the interaction of the differentiated tumor cell with basement membrane. After attachment, at the point of contact of the tumor cell, a lytic zone in the basement membrane is formed due to the secretion of enzymes which degrade the matrix (Brown et al., 1990; Gottesman, 1990). This is followed by invasion brought about by the motility of the cell across the basement membrane and stroma through the zone of broken matrix. Cell migration during invasion requires attachment and detachment of the cell and the lysed matrix around tumor paves path for invasion. Thus, tumor cells couple proteolytic activity with motility to achieve invasion. The degradative enzymes fall into three different classes—heparinases, serine-, thiol- and metal-dependent enzymes (Liotta et al., 1980; Wang and Stearns, 1988; Nakajima et al., 1989)—and a cascade of events including all the enzymes is probably involved in tumor invasion. Within the metalloproteinase family falls the type IV collagenase and agents which inhibit this enzyme prevent tumor cell invasion (Reich et al., 1988; Mignatti et al., 1989).

5.1. Evidence for antimetastatic potential of paclitaxel

The inhibitory effect of paclitaxel on the metastatic activity of tumor was studied using Matrigel (Stearns and Wang, 1992). Type IV collagenase secretion was inhibited by paclitaxel in a dose dependent manner coincidental with the microtubule bundling which might have interfered with their ability to mediate protease vesicle transport and secretion of the enzyme necessary for basement membrane disruption. The inhibitory effect of paclitaxel on penetration and invasion of tumor cells through Matrigel was proportionate to the dose and exposure time, and was irreversible as the exposure time increased. Micromo-

lar concentrations of paclitaxel completely inhibited attachment of cells to Matrigel. The multiple inhibitory effect of paclitaxel, i.e. interference with cell attachment/detachment and motility and protease secretion might have been responsible for the antimetastatic effect. The metastatic efficiency of paclitaxel treated cells was reduced in SCID mice after i.v. injection when compared to untreated cells and metastasis was completely blocked when exposure time and dose were increased; in general, paclitaxel exhibited long-term inhibitory effects.

6. Drug administration and problems

Paclitaxel is a low therapeutic index drug and therapy is always associated with toxic side effects (Nightingale, 1992). However, the potential benefits of paclitaxel therapy, in general, outweigh the possible risks. An excellent review describing different clinical aspects of paclitaxel was published (Rowinsky and Donehower, 1995). Paclitaxel is practically insoluble in water. Hence, the commercially available injection is a sterile solution of the drug in Cremophor® EL (polyethoxylated castor oil) and dehydrated alcohol (Mead Johnson Oncology Products, 1997). Five to six doses of paclitaxel are generally given at a dose of 135 or 175 mg/m^2 as a 3 or 24 h infusion, every 3 weeks (Kramer and Heuser, 1995).

Paclitaxel concentrate is a clear, colorless to slightly yellow viscous liquid. Paclitaxel is administered by i.v. infusion at a concentration of 0.3–1.2 mg/ml, after diluting the paclitaxel concentrate for injection with 0.9% sodium chloride injection or 5% dextrose injection or 5% dextrose and 0.9% sodium chloride injection, or 5% dextrose in Ringer's injection (Kramer and Heuser, 1995; Mead Johnson Oncology Products, 1997). After dilution in an infusion solution, the drug may exhibit haziness due to the additives of the formulation vehicle rather than precipitation of paclitaxel. Paclitaxel in aqueous solutions is chemically stable for 1–2 days. Inclusion of a hydrophilic, microporous inline filter of a pore size not more than 0.22 μm is necessary during paclitaxel infusion. Contact of undiluted paclitaxel concentrate for injection with plasticized polyvinyl chloride (PVC) equipment or devices used to prepare solutions for infusion is not recommended because Cremophor® EL causes leaching of diethylhexylphthalate (DEHP) from PVC containers. This leaching of DEHP is substantial and occurs in concentration-dependent manner and is also dependent on the type of administration set used (Allwood and Martin, 1996). In addition to plastic surfaces, rapid and nonspecific adsorption of paclitaxel also occurs with glass surfaces (Song et al., 1996). This problem can be overcome, to some extent, by increasing the organic component of the solvent system or using organic solvents. In order to minimize the exposure of patient to leached DEHP, diluted paclitaxel solutions preferably should be stored in glass or polypropylene bottles or in plastic (polypropylene or polyolefin) bags and administered through polyethylene lined administration sets (Allwood and Martin, 1996).

Paclitaxel therapy is associated with hypersensitivity reactions, therefore, premedication is mandatory before paclitaxel administration (Dabur, 1994; Mead Johnson Oncology Products, 1997). These hypersensitivity reactions may be due to Cremophor® EL, rather than the drug itself (Gregory and DeLisa, 1993). The premedication schedule includes; corticosteroids (e.g. dexamethasone), diphenhydramine or chlorphenaremine, H_2-receptor antagonists (e.g. ranitidine) and anti-emetics. Paclitaxel administration must be discontinued immediately in case of severe hypersensitivity reactions (Kohler and Goldspiel, 1994) and the patient must be treated with epinephrine and i.v. fluids. There is no known antidote for paclitaxel overdose (Panchagnula, 1998).

7. Drug delivery systems

Delivery of drugs via polymeric systems is a common technique, where drug release is regulated either by erosion of or diffusion through polymeric matrix. The biocompatible polymers used for parenteral therapy are either biodegradable or non-biodegradable. Biodegradable poly-

Table 3
Delivery systems developed for localized paclitaxel delivery

Polymeric device	Advantages	Disadvantages
Microspheres	Chemoembolization, no dose dumping	Usually first order release
Surgical pastes	Direct injection	Less consistent performance
Biodegradable implants	Large functional life-time	Possible risk of rejection and dose dumping

mers where degradation is hydrolytically (non-enzymatically) rather than enzymatically controlled are preferable in that there will be less subject-to-subject variation. The gamma irradiation of these polymeric devices to achieve terminal sterilization may result in dose-dependent chain scission and concomitant MW loss or cross-linking which may adversely affect the release kinetics and hence the performance. The release rates must be controlled such that drug levels in the tumor compartment are neither below the therapeutic window nor on the asymptotic portion of dose response curve where a large increase in the drug levels results in a very small increase in the rate of response and the excess drug is washed into the systemic circulation for toxic effects. Drug delivery systems explored so far for localized paclitaxel delivery are microspheres, surgical pastes and implants (Table 3). In order to develop a useful polymeric delivery system for paclitaxel, one needs to be able to control and manipulate the properties of the system both in terms of its physical and release characteristics. Hence, in the following discussion physical characterization of delivery system is detailed to correlate with, and comprehend the observed phenomenon.

7.1. Microspheres

Microspheres are monolithic systems containing a polymeric matrix in which drug substance is either dispersed and/or dissolved depending on its solubility. Transcatheter arterial chemoembolization achieved strong antitumor efficacy in the treatment of unresectable primary and metastatic tumors in liver (Gyves et al., 1983; Fujimoto et al., 1985; Ichihara et al., 1989) with fewer side-effects due to regional elevation of drug concentration (Kato et al., 1980).

A total of 1, 2 and 5% paclitaxel loaded microspheres were prepared by solvent evaporation method and evaluated (Dordunoo et al., 1995) with loading efficiency greater than 95%. Though scanning electron microscopy (SEM) micrographs of the microspheres showed no evidence of drug on the surface, in vitro release rate studies indicated a burst effect, which might have been due to the rough or pitted surface morphology apparently increasing the surface area for diffusion-mediated release of the drug. Higher drug loadings resulted in increased release rates due to the lower crystalline nature of the poly caprolactone (PCL) matrix and/or greater porosity created by the drug itself as it dissolved and diffused out of the matrix. Angiogenesis inhibition on the chick chorioallantoic membrane model (CAM) model was found to be dose-dependent as only those microspheres at 5% loading were effective.

Microspheres (0.6% paclitaxel) composed of a blend of biodegradable poly lactic acid (PLA) and a non-degradable ethylene vinyl acetate (EVA) copolymer were prepared by the solvent evaporation method (Burt et al., 1995). A blend of 1:1 EVA:PLA was selected as a composite material based on the observation that the microspheres could be easily prepared due to decreased problems of tackiness and coalescence and exhibited sustained drug release with increased functional life-time. Though the in vitro studies indicated an initial burst effect, the overall release rates were very slow. Therefore, characterization of the crystal form of the drug in the polymer might disclose a plausible reason for the slow release.

Further studies with isopropyl myristate (IPM) revealed that IPM significantly increased the release rate of paclitaxel in vitro (Wang et al., 1996) from Paclitaxel-IPM-poly(L-glycolic acid) (PLGA) microspheres and it was found that the release is governed by diffusion in the matrix. One more study was conducted (Wang et al., 1997) to examine the significance of molecular weight (MW) and copolymer ratio of PLGA on drug release rate and polymer degradation profiles. The

release rates increased with MW and lactic acid (LA) content of the PLGA matrix due to the greater number of micropores on the surface as evident from the SEM scans. LA being more hydrophobic than glycolic acid (GA) must have facilitated drug release. In the presence of IPM, polymer with both higher MW of PLGA and LA content precipitated faster during the preparation by the solvent evaporation method, accounting for the larger number of micropores on the surface. However, PLGA microspheres (LA:GA, 75:25, MW = 10 000) gave zero-order release for 3 weeks probably because of perfect balance between IPM mediated diffusion and matrix erosion.

7.2. Surgical pastes

Surgical pastes are monolithic drug containing devices prepared by thermal processing of polymeric materials having suitable glass transition temperatures or by in-situ solvent incompatibility, for direct injection at a desired site. Semisolid polyorthoester pastes have been used for bone induction in rats (Pinholt et al., 1991) and glaucoma filtration surgery (Merkli et al., 1995).

Polycaprolactone pastes of paclitaxel (1–30% w/w) were prepared and characterized with varying concentration of methoxy poly ethylene glycol (MePEG, 0–20%) (Winternitz et al., 1996). The preformulation studies conducted revealed that MePEG (MW = 350) blended more homogeneously with PCL matrix (MW = 20 000–27 000) reducing its melting point sufficiently for injection in the presence of drug as compared to other formulations containing other MW fractions of poly ethylene glycol (PEG), a blend of PCL:MePEG (4:1) melted at 50.4°C. Differential scanning colorimetry (DSC) studies revealed that both MePEG and paclitaxel decreased the melting point of PCL matrix but increased its crystallinity, thus increasing its functional life-time. Though tensile strength of the pastes was decreased by MePEG in a concentration dependent manner, they did not disintegrate, possibly due to the increased crystallinity. Microscopy showed the monolithic solution nature of surgical paste at loadings upto 10%, and at loadings greater than 5% they were monolithic dispersions, suggesting that the release mechanism of the former is different from that of the latter and is loading dependent. In vitro release studies showed that the incorporation of MePEG did not result in increased release rates. Hence, the increased crystallinity as confirmed from DSC studies retarded release due to decreased molecular diffusion coefficients. Gamma irradiation sterilization (30 k Gy) had no effect on the release, since it neither altered the MW nor the crystallinity of the polymer matrix as confirmed from GPC and DSC studies, respectively. Angiogenesis inhibition was found to be dose-dependent in the CAM model.

Biodegradable surgical pastes of poly (D,L-lactic acid) P(DLLA)–PEG–P(DLLA) copolymer and PDLLA + PCL blends of paclitaxel at 20% loading were fabricated (Zhang et al., 1996). As described previously, an increase in the PEG content resulted in the crystallinity of the block copolymer, but in vitro release studies indicated that release rates increased with PEG content in contrast to the earlier group. Pastes with 10% PEG had high melting point (> 60°C) unsuitable for injection, whereas those with greater than 40% disintegrated due to swelling of the incorporated PEG. This was attributed to the reduced polymer MW due to polymer breakdown and release of PEG as supported by the data from gel permeation chromatography (GPC). Moreover, NMR spectra also revealed a decrease in the PEG peak area indicating the dissolution of PEG after its dissociation from PDLLA. In vitro release studies from PDLLA + PCL blends showed an increase in the release rate with PDLLA content and pastes with PDLLA content greater than 80% disintegrated because PDLLA (MW = 800) degraded rapidly while PCL served as holding material. The formulations PDLLA:PCL (90:10) and PDLLA–PEG–PDLLA (30%PEG) caused tumor weight regression by 54 and 40%, respectively, in subcutaneously established MDAY-D2 tumors in mice after local delivery via injection at tumor site.

The problem of increased crystallinity could be overcome by replacing MePEG with coprecipitated microparticles of paclitaxel with various water soluble polymers into the PCL matrix by melt

technique at 65°C corresponding to a 20% loading of the drug (Dordunoo et al., 1997). In vitro release rate profiles followed the square root of time relationship indicating a diffusion related mechanism. Paclitaxel release from the PCL matrix increased in the presence of additives in the same order as the rate of swelling and with the proportion of additive and the size of drug-additive microparticles, but this also resulted in the disintegration of the matrix and therefore clinically useless formulations. The additives with a higher swelling nature exerted turmor pressure rupturing the polymer barrier between adjacent particles creating micro-channels and thus facilitating the escape of drug molecules from the matrix. The CAM model for the antiangiogenic study showed that the incorporation of additives markedly increased the diameter of the zone of avascularity compared to the pastes devoid of additives. A similar observation was made from in vivo studies after peri-tumoral injection of the molten paste with established palpable tumors. Paclitaxel–gelatin–PCL pastes (20:20:60) produced a reduction of $63 \pm 27\%$ in tumor mass.

7.3. Biodegradable implants

Drugs that readily cross the blood brain barrier (BBB) are more effective against CNS tumors than drugs that do not cross the BBB because of their size and lipophilicity. As a generalization, drugs that cross the BBB to produce appreciable brain drug levels are either hydrophilic with $MW \leq 160$ or hydrophobic $MW \leq 400$ (Levin, 1980). The much hydrophobic paclitaxel molecule ($MW = 853.9$; $\text{Log } P_{O:W} = 3.5$, (Heimans et al., 1994)) has poor BBB penetration capacity and hence, the use of implants for intracerebral delivery is justified.

Intracerebral chemotherapy is emerging as an important therapeutic approach in the treatment of recurrent malignant gliomas because of the pharmacokinetic advantage offered bypassing the BBB and also due to the fact that malignant gliomas recur within a few centimeters away from the tumor excision site. Drug loaded polymer discs (carmustine, 4-hydroxycyclophosphamide or paclitaxel) of poly[bis (*p*-carboxy phenoxy)propane-sebacic acid] (P(CPP-SA, 20:80) at 20% loading were implanted intracranially in cyanomolgus monkeys (Fung et al., 1998). The disks were made by compression molding (10 mm diameter; 1 mm thickness) and were gamma-irradiation sterilized. The rate of release of paclitaxel remained constant (~ 3 μg/day) over a 30 day period of in vitro study with 7% release in 100 days. The blood and CSF levels of paclitaxel were low in comparison to carmustine and 4-hydroxycyclophosphomide due its highly hydrophobic nature and low local concentrations attained at the polymer disk-tissue interface. Pharmacokinetic analysis of drug distribution data confirmed that interstitial fluid convection in addition to concentration gradients contributed to drug transport and distribution in the brain. Similar slow release of paclitaxel, 15% in 100 days, from P(CPP-SA) polyanhydrides (Jampel et al., 1991), and 17% in 100 days from poly(fatty acid dimer-sebacic acid) (P(FAD-SA), 1:1) (Park et al., 1998) were observed.

However, faster release rate (45–65% in 30 days) of paclitaxel was observed (Walter et al., 1994) from relatively more hydrophilic P(CPP-SA, 20–40% loading) and the implants were found to be promising in an experimental glioma model. The implants doubled to tripled the median survival of rats bearing tumor. Paclitaxel concentrations remain elevated for at least 1 month after implantation in brain and offered a good pharmacokinetic advantage due to prolonged exposure to therapeutic concentrations bypassing the BBB. Since paclitaxel is extremely lipophilic, the more the hydrophilicity of the polymeric device faster is the release rate, and the desired release rate can be achieved by judicious selection of the polymeric material.

The following inferences can be drawn from the above studies:

1. Erosion is found to be the main mechanism by which paclitaxel is released.
2. The performance of delivery system is mainly influenced by the molecular weight and relative hydrophilicity of the polymers.
3. The presence of additives with appropriate HLB values in the delivery system aid in achieving the desired paclitaxel release rate.

4. The antiangiogenic effect and tumor regression of paclitaxel are a dose-dependent phenomenon.
5. The pharmacokinetic advantage offered is demonstrated from the sustained therapeutic levels of paclitaxel produced in intracranial and subcutaneous tumor animal models.

8. Possibilities for better therapy

Therapy can be made much more convenient and better by the use of implantable pumps (Kwan, 1991). Alzet miniosmotic pumps were successfully used to deliver intratumorally antisense oligonucleotides in rats (Walker et al., 1998) and antiangiogenic agents in mice (Teicher et al., 1992). Infusaid pump is a mechanical pump powered by means of an expanding chamber containing fluorocarbon whose vapor pressure at ambient conditions is approximately 300 mmHg greater than atmospheric pressure. The pump can be refilled by percutaneous injection simultaneously recharging the power source. Meldtronic programmable pump is a battery powered pump and can be programmed for the desired flow rate. These pumps discharge contents via an outlet catheter at the tumor. In the near future, it is expected that the technological advancements in design and performance of implantable infusion and osmotic pumps will not only improve patient compliance, but also help in combating tumor drug resistance development preserving the clinical efficacy of unique lead molecules, like paclitaxel.

9. Conclusion and future directions

The limitations of traditional modes of therapy for anticancer drugs, namely non-specific distribution and systemic toxicity and rapid development of resistance are forcing one to look at new modalities in cancer therapy and identify molecular targets which makes tumor cell highly susceptible to such therapies. The highly lipophilic character, long-lasting antiproliferative activity attributed to the unique mechanism of action and, dose- and exposure time-dependent antiangiogenic, antimetastatic activities make paclitaxel a good candidate for local drug therapy of cancer. The potential of paclitaxel in the treatment of bladder cancer was evaluated after local administration to dogs. Paclitaxel was accumulated in urothelium in high concentrations and was long retained with insignificant systemic exposure. The preclinical trial suggested that paclitaxel is a good candidate for the treatment of intravesical bladder cancer via localized delivery (Song et al., 1997). Clinical trials have confirmed a major pharmacokinetic advantage (>1000-fold) for peritoneal cavity exposure following intraperitoneal delivery of paclitaxel compared to systemic administration (Markman et al., 1992, 1995).

Resistance to paclitaxel in vitro could be overcome through prolonged drug exposure (Lopes et al., 1993). Though, microspheres, surgical pastes and implants are promising formulations, localized paclitaxel delivery will be immensely fostered by the technological advancements in the design and performance of implantable infusion pumps, osmotic pumps and triggered drug delivery systems. The concept of localized paclitaxel drug delivery is still in its infancy and under investigational stages, yet all preliminary studies indicate that these approaches hold the potential of being explored and developed as full blown technologies for this purpose. Clinical trials suggested that paclitaxel is synergistic with cisplatin, etoposide, etc., in combination therapy (Spencer and Faulds, 1994). Combination of systemic with local mode of therapy with paclitaxel would be the ultimate solution to successful cancer therapy where systemic administration of other synergistic antiangiogenic/cytotoxic agents at relatively low doses would check metastases while localized delivery alleviates body drug-burden while keeping tumor drug-burden at its maximum.

10. Future drug delivery systems for localized delivery

Cancer is a disease characterized by amplification and hyper-expression of oncogenes and the encoded proteins of these cancer genes play an

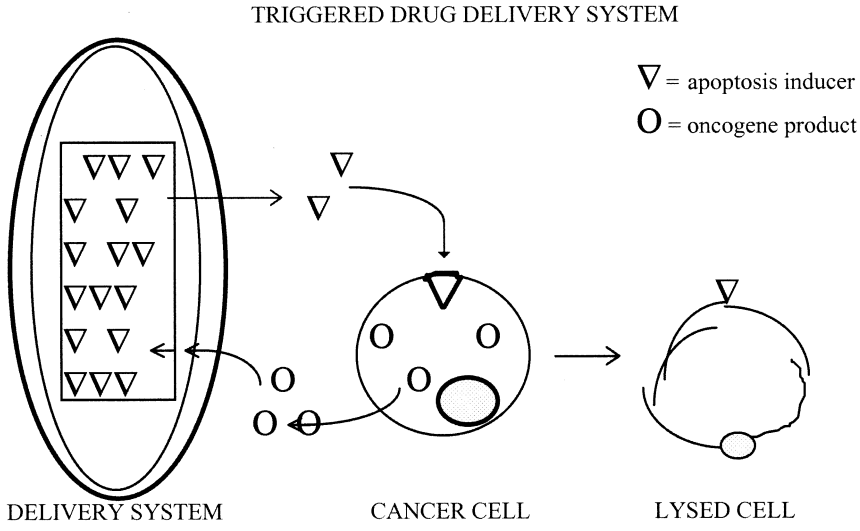


Fig. 3. Scheme of apoptotsis induction by an oncogene product triggered drug delivery system.

important role in signal transduction pathways involved in cell proliferation. Oncogene products which are expressed at high levels in cancer can be classified into four groups: growth factors, membrane receptors, intracellular transducers and nuclear transcription factors. These genes and their proteins can be looked upon as ideal targets for a rational drug hunting program, such as antisense oligonucleotide cancer therapy. In this context, the use of triggered drug delivery systems would add a new dimension to cancer therapy. A self regulated drug delivery system is defined as one capable of receiving feedback information and adjust drug output in response to that information, where the feedback signal either modulates the rate of drug release or triggers drug release from an otherwise passive device. The triggering can be accomplished, through a selective sensing mechanism, by the appearance of a specific substrate in the tissues surrounding the device. When one of the oncogene products can serve the specific substrate purpose and trigger an appropriately fabricated device, it releases drug and/or agents which are cytotoxic and/or induce apoptosis of the surrounding tumor cells either directly or through activation of cytotoxic T lymphocytes and natural killer cells (Fig. 3).

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