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Paclitaxel and its formulations

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Abstract

Paclitaxel (Taxol®) is a promising anti-tumor agent with poor water solubility. It is effective for various cancers especially ovarian and breast cancer. Intravenous administration of a current formulation in a non-aqueous vehicle containing Cremophor EL may cause allergic reactions and precipitation on aqueous dilution. Moreover, the extensive clinical use of this drug is somewhat delayed due to the lack of appropriate delivery vehicles. Due to this there is a need for the development of alternate formulation of paclitaxel having good aqueous solubility and at the same time free of any side effects. Various approaches employed so far include cosolvents, emulsions, micelles, liposomes, microspheres nanoparticles, cyclodextrins, pastes, and implants etc. which are discussed in this paper.

Keywords: Paclitaxel; Taxol; Formulation; Solubility

1. Introduction

Paclitaxel, the first of a new class of microtubule stabilizing agents, has been hailed by National Cancer Institute (NCI) as the most significant advance in chemotherapy of the past 15–20 years. Paclitaxel was not a chance discovery but was the outcome of the investigation of over 12 000 natural compounds for anticancer activity (Appendino, 1993)

Paclitaxel is a diterpenoid pseudoalkaloid (Fig. 1) having molecular formula $C_{47}H_{51}NO_{14}$, corresponding to molecular weight of 853 Da. For anti-tumor activity it is required that entire taxane molecule (Fig. 2) be present, since the ester and the tetraol formed by a low temperature cleavage of paclitaxel are found to be essentially inactive (Wall and Wani, 1996). Paclitaxel was isolated in early 1960s from the bark of Pacific Yew (*Taxus brevifolia*; family Taxaceae), one of the geographical varieties of yew (Wani et al., 1971). Paclitaxel was obtained in a pure form in 1969 and its structure was published in 1971, after many complexities due to its low concentration and structure complexities (Wani et al., 1971; Bingham, 1994).

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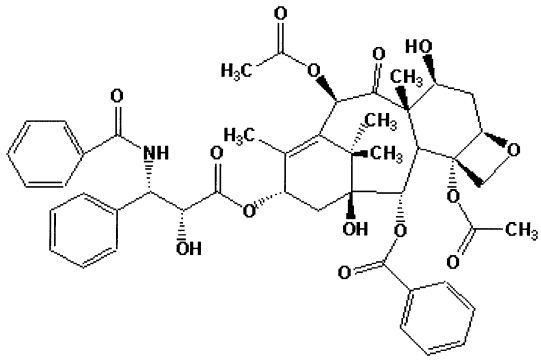

Fig. 1. Structure of paclitaxel (5β,20-Epoxy-1,2α,4,7β,13α-hexahydroxytax-11-en-9-one4,10-diacetate2-benzoate13-ester with (2*R*,3*S*)-*N*-benzoyl-3-phenyllisoserine).

The importance of paclitaxel was not recognized until the late 1970s, since, it is difficult to obtain and also due to its low solubility it has a formulation problem. It was in 1979, that Susan Horwitz discovered that paclitaxel has a unique mechanism of action and interest was further stimulated when impressive activity was demonstrated in NCI tumor screening.

Unlike other microtubule agents, such as *Vinca* alkaloids, which induce the disassembly of microtubules, paclitaxel promotes the polymerization of tubulin (Schiff et al., 1979; Hamel et al., 1981; Parness and Horwitz, 1981; Schiff and Horwitz, 1981; Rowinsky et al., 1990). The microtubule formed in presence of paclitaxel are extraordinarily stable and dysfunctional, thereby causing the death of the cell by disrupting the normal tubule dynamics required for cell division and vital interphase process. Paclitaxel has neoplastic activity particularly against primary epithelial ovarian carcinoma, breast cancer, colon, head, non-small cell lung cancer, and AIDS related Kaposi's sarcoma (Horwitz, 1992; Spencer and Faulds, 1994; Rowinsky and Donehover, 1995; Alshowaier and Nicholls, 1997). This has led to the approval of the drug in many countries for its use as second line treatment of ovarian and breast cancers. In phase II trials with patients treated previously with high dose chemotherapy, the response rate in advanced and refractory ovarian cancer was 30% (Mcguire et al., 1989). The overall response rate in phase I trials in previously treated patients with metastatic breast cancer was 56% (Holmes et al., 1991). In our present paper, we are going to elaborate on the various formulation aspects of paclitaxel, problems associated with the existing formulation and the promising formulation aspects of paclitaxel.

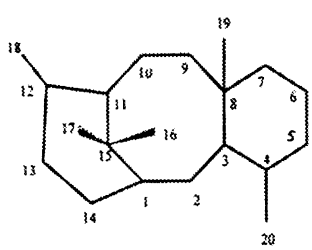

Fig. 2. Taxane nucleus.

2. Physical properties and pharmacokinetics

Paclitaxel is white to off-white crystalline powder. It is highly lipophilic, insoluble in water and melts at around 216–217 °C. Its disappearance from plasma is found to be biphasic (Wiernik et al., 1987b). The initial rapid decline represents distribution to the central compartment and elimination of the drug and the later phase is due in part, to the efflux of the drug from the peripheral compartment (Brown et al., 1991). The generally accepted dose is 200–250 mg m^{-2} and is given as 3 and 24 h infusion. Pharmacokinetics of paclitaxel shows wide variability. Terminal half-life was found to be in the range of 1.3–8.6 h (mean 5 h) (Rowinsky and Donehower, 1993) and the steady-state volume of distribution was found to be ~ 87.1 m^{-2}. The drug undergoes an extensive P-450 mediated hepatic metabolism and less than 10% drug in the unchanged form is excreted in the urine (Rizzo et al., 1990). Most of the drug is eliminated in feces. More than 90% of the drug binds rapidly and extensively to plasma proteins (Wiernik et al., 1987a; Rowinsky et al., 1990). The highest concentration of the paclitaxel following a 6-h infusion in rats was found to be in lung, liver, kidney and spleen and was essentially excluded from brain and testes (Rowinsky and Donehower, 1993).

3. Limitations

Clinical application of paclitaxel is accompanied by twofold problems.

3.1. Availability

Although the extraction of paclitaxel has increased yields to 0.04% w/w, four trees have to be sacrificed to produce 2 g of the drug for the chemotherapy of one patient. This is not affordable from the environmental point of view (Oliver, 1993; Si-Shen and Guofeng, 2001). The total synthesis of paclitaxel and its analogs represent formidable challenges due to its structural complexity. Though scientists world over are trying very hard and various approaches of its total synthesis has been followed and published, a practical method for its synthesis is yet to be developed (Nicolaou et al., 1994; Holton et al., 1994; Wander et al., 1995). An appropriate solution, thereby, seems to be a semi-synthesis, which involves extraction of paclitaxel from needles and twigs of more abundant English yew trees, or Chinese red bran yew tree (Witherup et al., 1990).

3.2. Solubility

Paclitaxel is poorly soluble in an aqueous medium, but can be dissolved in organic solvents. Its solutions can be prepared in a milimolar concentration in a variety of alcohols, such as methanol, ethanol, tertiary-butanol as well as in DMSO. Non-aqueous solubility is found to be ~ 46 mM in ethanol, ~ 20 mM in methylene chloride or acetonitrile, ~ 14 mM in isopropanol (Adams et al., 1993). Numbers of reports have been published on the solubility of paclitaxel and acceptable value of aqueous solubility is 0.6 mM (Tarr and Yalkowsky, 1987; Swindell and Krauss, 1991). Moreover, paclitaxel lacks functional groups that are ionisable in a pharmaceutically useful range and therefore manipulation in pH does not enhance its solubility. Furthermore, common approaches to improve solubility like addition of charged complexing agents or by producing alternate salts of the drug are not feasible in the case of paclitaxel (Straubinger, 1995).

Prodrug synthesis has also been extensively studied to increase the aqueous solubility of paclitaxel (Burt et al., 1995). The preferred position for the preparation of prodrug of paclitaxel is 2′ position since many 2′-acyl-paclitaxel derivatives hydrolyze fairly rapidly back to paclitaxel in blood compartments (Mellado et al., 1984). Since the configuration of C-7 hydroxyl group does not seem to be a factor in determining cytotoxicity, C-7 prodrug ester has also been synthesized (Deutsch et al., 1989). Nicholaou designed the paclitaxel ester with a strong electron withdrawing substituents, such as alkoxy in the α-position of the ester, in order to accelerate the hydrolytic cleavage (Nicolaou et al., 1993). In vitro, these prodrugs have been shown to possess cytotoxic activity against tumor cell lines comparable to those of paclitaxel. In addition, human plasma catalyzes the release of active paclitaxel. Polyethylene glycol (PEG) is an amphiphilic macromolecule that, in the molecular weight range of 2–12 kDa, it imparts greater aqueous solubility to conjugates of hydrophobic organic compounds or proteins, augmenting circulation half life and increasing immunogenicity (Monfardini and Veronese, 1998). A prodrug strategy employing PEG as a solubilising agent has been successfully demonstrated in case of paclitaxel (Greenwald et al., 1994, 1996).

4. Paclitaxel dosage form

Paclitaxel is currently formulated in a vehicle composed of 1:1 blend of Cremophor EL (polyethoxylated castor oil) and ethanol which is diluted with 5–20-fold in normal saline or dextrose solution (5%) for administration. This formulation is stable in unopened vials for 5 years at 4 °C. However, lots of problems employing this vehicle have been reported.

(1) One of the substantial problems associated with this formulation is that the ethanol: Cremophor vehicle required to solubilize it is toxic (Dorr, 1994). Although it has been used to administer other drugs such as cyclosporine (Howrie et

al., 1985) and teniposide (O'Dwyer et al., 1986), the amount of Cremophor necessary to deliver the required doses of paclitaxel is significantly higher than that of administered with any other marketed drug (Rowinsky et al., 1992). The side effects caused by Cremophor EL include hypersensitivity reactions, nephrotoxicity and neurotoxicity. Cremophor EL also has an influence on the function of endothelial and vesicular muscles and causes vasodilation, labored breathing, lethargy and hypotension (Friedland et al., 1993; Lilley and Scott, 1993). The vehicle has been shown to cause fatal hypersensitivity reactions at nearly every step in the development path, in both preclinical and clinical testing (Lorenz et al., 1977; Dye and Watkins, 1980; Weiss et al., 1990). One mediator of hypersensitivity reaction is histamine release and prophylaxis to counteract histaminergic mechanism reduces the incidence of hypersensitivity reaction. Hypersensitivity reactions are the most prevalent with bolus administration and shorter infusion schedules (Weiss et al., 1990; Rowinsky et al., 1993). Premedication with corticosteroids (dexamethasone) and antihistamine (both H_1 and H_2-receptor antagonist) is used with paclitaxel to increase safety and reduce intensity and the incidence of serious hypersensitivity reactions associated with paclitaxel administration in Cremophor (Weiss et al., 1990; Lam et al., 1997). Clinically pharmacological intervention may be less desirable than a safer and better-tolerated formulation. Several possible mechanisms exist by which the various drugs administered, either to support patients during paclitaxel administration, or to treat their cancer or the side effects of chemotherapy could contribute to interactions that may affect paclitaxel efficacy or toxicity. The doses of dexamethasone given are relatively large, and certain H_2-receptor antagonist such as cimetidine and ranitidine is well known inhibitors of the cytochrome P-450 metabolic pathway (Powell and Donn, 1983) and the later is thought to be involved in the metabolism of paclitaxel (Monsarrat et al., 1990; Jamis-Dow et al., 1993; Rowinsky and Donehower, 1993; Cresteil et al., 1994; Klecker et al., 1994), suggesting the potential for interaction.

(2) The recommended concentration of the drug in the properly diluted clinical formulation is 0.3–1.2 mg ml^{-1} (0.35–14 mM) and has only short-term physical stability as some particles slowly tend to precipitate out of the aqueous media. The stability of appropriately diluted paclitaxel was estimated at 12–24 h and its use was recommended within 12 h of dilution in the aqueous media. It was observed that over a period of 24 h, small number of particles may be observed in the diluted form and the number of particles appeared to be greater in the solution of more than 0.6 mg ml^{-1} (≥ 0.7 mM) paclitaxel (Adams et al., 1993). However, the results of the analysis indicated to have 98–100% of their original drug content in the solution obtained after removal of these particles by filtration (Waugh et al., 1991), which suggests that either the total amount of paclitaxel precipitated was small or that the precipitate was not of paclitaxel. Hence, an in-line filter is recommended for the intravenous line and it is suggested that drug is administered promptly after dilution.

(3) Paclitaxel/ethanol: Cremophor formulation also shows an incompatibility with the components of the infusion sets. It was reported (Venkataraman et al., 1986; Pfeifer and Hale, 1993; Allwood and Martin, 1996; Song et al., 1996; Xu et al., 1998) that both ethanol and Cremophor leach diethylhexylpthalate (DHEP) from the polyvinylchloride (PVC) infusion bags and administration sets. The manufacturers of paclitaxel thus recommended the use of glass, polypropylene or polyolefin containers for its storage. These recommendations, however, pose a number of practical problems since the availability of these types of containers is severely limited and as such the medical staff may be unfamiliar in its handling. The amount of DEHP extracted depends on the concentration of paclitaxel vehicle, the length of contact time between the injection vehicle and the container and the type of administration set used for its administration.

(4) Further issue of compatibility arises from the fact that patients undergoing paclitaxel therapy frequently receive other medications also and a potential exists for some to be administered concomitantly through the same intravenous

catheter. In such cases, it is required to verify physical and chemical stability in the presence of other drugs and their excipients. Interaction with nearly 60 common drugs or excipients (Trissel and Martinez, 1992, 1993) was investigated by observing turbidity after mixing with paclitaxel in dextrose/ethanol: Cremophor.

(5) The chemical stability of paclitaxel could be altered not only by the interaction with other drug molecules, but also by the condition of handling and administration. According to one study, the solutions of paclitaxel in the range of pH 4–8 were stable for ≥ 72 h (Waugh et al., 1991). On the other hand, a rapid degradation occurred at pH 11 and numerous degradation products were observed within 1.5 h, and its degradation was virtually complete in ≤ 72 h.

(6) In addition to the possibility of metabolic interactions, paclitaxel is highly bound in the blood (~ 90–95%) (Longnecker et al., 1987; Wiernik et al., 1987a; Kumar et al., 1993). Some of the agents used for prophylaxis likewise are also highly bound to plasma proteins and therefore there exists the potential for the displacement of bound paclitaxel or co-administered drug from blood components, which could alter the anti-tumor activity, or toxicity of paclitaxel or of co-administered drug.

(7) The use of Cremophor as a vehicle also appears to alter the biochemical properties of lipoproteins, such as high-density lipoproteins (Kongshang et al., 1991; Sykes et al., 1994). It has also been shown to partially mediate the cytotoxic activity of paclitaxel in primary cultures of tumor cells from patients and to reverse P-glycoprotein mediated multidrug resistance (Lebmann et al., 1994; Nygren et al., 1995).

Out of the above mentioned limitations, the most serious problem clinically is that of hypersensitivity reactions. These problems were observed during clinical trials and were found to be critical point in development of paclitaxel. Paclitaxel was approved for phase II trials in April 1985, only after including premedication with antihistamines in the regimen along with 24 h continuous infusion to reduce peak concentration of both Cremophor and taxol.

5. Alternative formulations

The primary goal of formulation development for paclitaxel is to eliminate the Cremophor vehicle by reformulation of the drug in a better-tolerated vehicle. Reformulation also provides the possibility of improving the efficacy of paclitaxel based anticancer therapy. A great deal of effort is being directed towards the development of aqueous based formulations for paclitaxel, including soluble semi-synthetic paclitaxel derivatives that do not require solubilisation by Cremophor and that decrease the systemic clearance of the drug. The approaches (Fig. 3) being used for a desired formulation of paclitaxel by various methods are, co-solvency, emulsification, micellisation, liposome formation, non-liposomal lipid carriers (microspheres, nanocapsules etc.), cyclodextrins (CDs), local drug delivery devices and miscellaneous.

5.1. Co-solvency

Weak electrolytes and non-polar drug molecules frequently have poor water solubility. Their solubility can be increased by the addition of water miscible solvent in which the drug has a good solubility. The solvents used in combination to increase the solubility of the solute are known as co-solvents (Boylan, 1987). Commonly, water-miscible co-solvents are used as the method of formulating intravenous non-water-soluble drugs. However, often the drug precipitation occurs (Flynn, 1984) upon the addition of the co-solvent mixture to intravenous fluids or blood and in such cases extremely slow infusion is required to prevent the precipitation. It was found that the vehicle consisting of ethanol and polysorbate-80, to be diluted in glucose solution before use, was suitable for the administration of paclitaxel. The same vehicle has been previously used for the administration of toxetere (Docetaxel) (Bissery et al., 1991).

Another co-solvent system consisting of ethanol, polysorbate (Tween) 80, and the surfactant Pluronic L64, in the ratios of 3:1:6 (v/v/v) for paclitaxel formulations was reported (Tarr and Yalkowsky, 1987). The drug was solubilized at a

concentration of 5 mg ml^{-1}. No change in chemical and physical stability was observed over 3 months but the solution on dilution with water to a concentration of 3.45 mM was found to be physically stable for only 3 days.

Paclitaxel, inherently possessing limited aqueous solubility can be rendered water soluble by using principle of co-solvency, commonly used solvents being polysorbate 80, ethanol etc. Though the precipitation of the drug on dilution with the aqueous media is an important factor to be considered.

5.2. Emulsification

The emulsion (O/W) type of delivery system has also attracted much attention lately. Tarr and Yalkowsky (1987) formulated paclitaxel in an oil-in-water emulsion using triacetin as the internal phase. They found that triacetin is an excellent solubilizing vehicle for this compound. The solubility of paclitaxel in triacetin is 75 mg ml^{-1}. In spite of this the drug slowly precipitates out of the solution when the emulsion is diluted approximately nine times with dextrose (5%) in an intravenous piggyback system. Wheelar et al. manufactured a blend of emulsion and liposome with corn oil, egg phosphotidylcholine (EPC), cholesterol and paclitaxel (Wheeler et al., 1994). Microscopic examination of taxol containing emulsion revealed no crystalline drug at concentration as high as 1–15 mg of oil and taxol concentration in excess of 5 mg ml^{-1} were easily obtained in the emulsion formulation. Moreover, the formulation was found to be stable after IV administration as judged by the plasma clearance behavior of individual lipid components. An emulsion of paclitaxel made with triolein, dipalmityl-phospatidylcholine and polysorbate-80 with PEG coated at emulsion surfaces was found to prolong or extend the circulation time (Lunberg, 1997). Another emulsion dosage form containing paclitaxel, oil blend, EPC, Tween 80 and glycerol solution was found to have a very good stability at 4 °C and the paclitaxel containment efficiency was maintained above 95% with a mean emulsion diameter around 150 nm for at least 3 months (Kan et al., 1999). It showed cytotoxicity against HeLa cells with IC50 at 30 nM. Further, another group of workers observed no degradation of the drug from its two parentral (IV) drug delivery systems in an emulsion form employing benzyl benzoate and tributyrin after storing for at least 3 months at 4 and 25 °C (Simamora et al., 1998). Moreover, these products were devoid of any irritation effect and phlebitis in rabbits com-

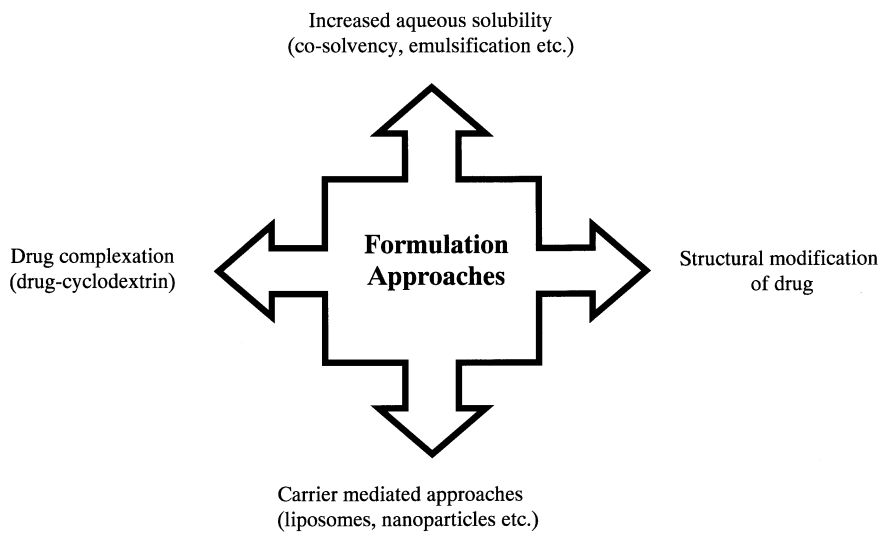

Fig. 3. Alternative approaches for paclitaxel formulation.

monly encountered with Cremophor used as vehicle. It was also found that these emulsions were compatible with several common additives and showed minimum side effects. Kaufman et al. got the emulsion formulation of paclitaxel (O/W type emulsion) comprising of paclitaxel in safflower oil and water patented. Its solution in oil obtained after dissolving the drug in oil with isopropanol as a cosolvent and then separating the later, was then dispersed in water using lecithin as a surfactant, to form a stable O/W emulsion. Glycerin was added to adjust the osmolarity. They also reported its compatibility with several additives and also demonstrated that the product possessed minimum side effects (Kaufman et al., 1997). Thus emulsification of paclitaxel into O/W system can be an important approach towards the aqueous formulation system.

5.3. Micellisation

Micellisation is an important approach capable of solubilizing a hydrophobic drug in a hydrophilic environment comprising of biodegradable drug carrier micelle and a hydrophobic drug wherein the drug is physically entrapped and not covalently bonded to the polymeric drug carrier micelle.

Polymer micelles are convenient passive targeting carrier systems of anticancer drugs since they are structurally strong and unlike liposomes are not captured by the reticuloendothelial cell system (RES) because of their particle size (20–100 nm) (Yokoyama et al., 1990; Kataoka et al., 1993). Polymers used should be non-toxic, biodegradable and should metabolize in the body. Miwa et al. (1998) prepared micelles of paclitaxel using *N*-lauryl-carboxymethyl-chitosan (LCC) having the size less than 100 nm. This particle size was considered effective for passive targeting for tumors. The concentration of paclitaxel in micellar solution was very high, with a maximum of 2.37 mg ml^{-1}. This concentration was 1000 times above that in saturated solution of paclitaxel at pH 7.4. LCC-entrapped paclitaxel was more effective in cytostatic activity than a free paclitaxel in low concentrations. Polymeric micellar paclitaxel formulation using amphiphilic diblock copolymers have also been utilized (Ramaswamy et al., 1997).

Onyuksel et al. solubilized paclitaxel in aqueous medium with the use of bile salt/phospholipid mixed micelles. The solubilisation potential of the mixed micelles increased as the total lipid concentration and the molar ratio of bile salt/phospholipid increased. Precipitation of the drug was avoided with the spontaneous formation of the drug-loaded liposomes from the mixed micelles. The formulation can be stored in a freeze-dried form as mixed micelles to achieve optimum stability and the liposomes can be prepared by simple dilution just prior to administration. Using panel of cell cultured lines; it was found that the cytotoxic activity of paclitaxel was retained when formulated as mixed-micellar solution. Further, for the same solubilisation potential, the mixed-micellar vehicle appeared to be less toxic than the standard non-aqueous vehicle of paclitaxel containing Cremophor EL (Onyuksel et al., 1994). Micelle encapsulated paclitaxel is thereby water soluble and in addition devoid of common side effects associated with cremophor vehicle.

5.4. Liposome formation

Among the drug carrier systems, liposomes represent a mature versatile technology with a considerable potential for encapsulation of both the lipophilic and the hydrophilic drugs and are in clinical trials for a number of neoplastic and infectious diseases. Encapsulation in liposomes often results in distinct changes in the pharmacokinetic and the pharmacodynamic properties of the drugs, in some cases causing a marked decrease in toxicity or increase in potency (Sharma et al., 1993). Liposomes consist of one or more aqueous compartments contained within lipid membrane bilayer. Because the liposomes contain a hydrophilic domain, a hydrophobic domain and an interfacial region, they may accommodate therapeutic agents having diverse physical characteristics. Sampedro et al. prepared multilamellar vesicles employing different combinations of phospholipids like L-dimyristoyl phosphatidylcholine (DMPC) and L-Dimyristoylphosphatidyl glycerol (DMPG) and cholesterol by the standard evaporation/hydration methods. In general, mixtures of DMPC:DMPG, at a molar ratio 7:3 and

9:1, and the addition of 5% cholesterol (w/w) gave the optimum results. In vitro cytotoxicity of liposomal drugs against L1210 cells was found to be more than that of a free drug in the case of paclitaxel (Sampedro et al., 1993). Later, the liposomal drug delivery systems containing paclitaxel and phospholipid in molar ratio of 1:33 from phosphatidylglycerol and phosphatidylcholine (1:9 molar ratio) was developed (Sharma and Straubinger, 1994) and was found to be stable for more than 2 months at 4 °C, and for 1 month at 20 °C. These formulations retained growth inhibitory activity of the free drug in-vitro against a variety of tumor cell lines. In mice, these were found to be well tolerated when administered in bolus doses by both intravenous and intraperitoneal (IP) routes. The maximum tolerated dose (MTD) was >200 mg kg^{-1}; which exceeded for that of free paclitaxel having MTD of 30 mg kg^{-1} by IV or 50 mg kg^{-1} by IP route. Recently, Ceruti et al. compared conventional paclitaxel loaded liposomes with the stealth long circulating PEGylated liposomes containing paclitaxel and determined the encapsulation efficiency, physical stability and the drug leakage profile in human plasma. They found that PEGylated liposomes were less stable during storage than the conventional liposomes and had a lower drug release in plasma at 37 °C. After 2 and 48 h, the conventional liposomes had the same cytotoxicity as the free drug, while PEGylated liposomes were found to be as active as free drug only after 48 h. Drug encapsulation in the conventional liposomes produced marked differences in pharmacokinetic compared with that of free drug. The PEGylated liposomes were long circulating, with an elimination half-life of 48.6 h, compared with 9.27 h for conventional liposomes. Biodistribution studies also showed a considerable decrease in the drug uptake in the mononuclear phagocytic system (MPS) such as liver and spleen at 0.5 and 3 h after injection of PEGylated liposomes when compared with conventional liposomes. Further, increased concentrations of paclitaxel were entrapped in liposomes by incorporating water-soluble prodrugs such as 2′-succinyl-2′-methyl pyridinium acetate and 2′-mPEG ester paclitaxel derivative in lipid vesicle. Liposomes containing 2′-mPEG (5000)-paclitaxel showed the best performance in terms of stability, entrapment efficiency and drug concentration (6.5 mg ml^{-1}) (Ceruti et al., 2000). The in vitro cytotoxic activity of this liposomal prodrug was similar to that of the parent drug. The most important change in the pharmacokinetics of the prodrug versus free drug liposomal formulations was plasma lifetime, which was longer in liposomes containing 2′-mPEG (MW 5000)-paclitaxel (Crosasso et al., 2000). As a result of advances both in pharmaceutical technology and in therapeutic rationale, liposomes have advanced to human testing in growing number of clinical trials. But the use of liposomes in drug targeting is found to be limited mainly due to problem of low entrapment efficiency, drug instability, rapid drug leakage and poor storage stability. Though the problems like stability has been overcome by the development of new vesicular drug delivery systems like non-ionic surfactant vesicles (niosomes), but still further research work is required in the area of liposomal delivery to make it an ideal approach for drug delivery.

5.5. Non-liposomal drug carrier systems

Use of biodegradable polymeric micro/nanocapsules for controlled delivery of anticancer agent has an advantage in enhancing the therapeutic efficacy and reducing the systemic side effects. It has been reported that nanospheres show a significant advantage over microspheres. Nanospheres enable intravenous as well as intramuscular injection and subcutaneous administration by minimizing possible irritant reactions. Wang et al. studied the anti-tumor activity of paclitaxel encapsulated in liposomes or in nanocapsules and observed that nanocapsules showed comparable in vitro activity but proved to be toxic apparently due to their composition. They prepared paclitaxel loaded microspheres using a lactic-co-glycolic acid copolymer with a lower molecular weight from which faster release of paclitaxel is expected. Moreover, an additive, isopropyl myristate was incorporated into the microspheres to further enhance the release rate of paclitaxel. The mechanism of release of the drug from these microspheres was dominated by the drug diffusion in matrix (Wang et al., 1996).

Recently, Si-Shen Feng and Guofeng Huang developed the paclitaxel-loaded nanospheres of biodegradable polymers with freeze dry solvent extraction/evaporation technique. Among the emulsifiers employed, dipalmitoyl-phosphatidylcholine (DPPC) was found to provide more complete coating on the surface of the products, which thus results in higher emulsifying efficiency compared with that of polyvinyl alcohol (PVA). The data also indicated that phospholipids with short and saturated chains have excellent emulsifying effects (Si-Shen and Guofeng, 2001). Substantially water insoluble pharmacologically active agent can also be delivered in the form of microparticles that are suitable for parentral administration in aqueous suspensions. Microparticle suspension dosage form allows some degree of targeting to organs like liver, spleen, lungs, lymphatic circulation and the like, through the use of particles of varying size, and through administration via various routes (Soon-Shiong et al., 1996). This method of delivery further allows the administration of substantially water insoluble pharmacologically active agents by using a much smaller volume of liquid and requiring greatly reduced administration time relative to administration volumes and the time required by prior art delivery systems (e.g. intravenous infusion of $\sim$1–2 l of fluid over a 24 h period are required to deliver a typical human dose of 200–400 mg of paclitaxel).

Polymeric nanoparticles possess the advantages gained by the use of liposomal delivery, additionally overcoming the limitations faced by them, including low entrapment efficacy, drug instability and leakage. Moreover, nanoparticles due to their extremely small size can extravasate at the pathological site such as solid tumors through passive targeting mechanism. A very recent U.S. patent reinstates this fact where the inventors have prepared the nanoparticles of copolymeric micelles which are nanometer size particles of micellar aggregation of amphiphillic polymers. The polymers used were vinyl pyrrolidone and *N*-isopropyl acrylamide to achieve nanoparticles in the size range of 30–60 nm in diameter (Maitra et al., 2001)

5.6. Cyclodextrins

CDs are cyclic oligosaccharides, which have been used extensively to increase the solubility, dissolution rate and the bioavailability of poorly soluble drugs (Uekama and Otagiri, 1987; Szejtli, 1991a,b). They have also been used to increase the stability of labile drugs (Uekama et al., 1983) and improve the performance of intravenous formulations (Etes et al., 1991). An enhancement in solubility of paclitaxel from an inclusion complex with hydroxy propyl-β-CD (HPβCD) was demonstrated by Cserhati et al. They stated that the complex formed was more hydrophilic than the uncomplexed drug indicating that the solubility of the drug can be enhanced by HPβCD. They also prepared complexes of paclitaxel with alpha-CDs (Cserhati et al., 1995), but taking into account the structure of paclitaxel, with a large fused, taxane ring system and bulky phenyl group in C13 esterified side chain, CDs with large cavities such as β- and γ-CD, were thought to be more suitable. It was found that β-CDs increased paclitaxel solubility by 950-fold or more, and clinically useful concentration (1–4 mM) of paclitaxel can be easily achieved (Sharma et al., 1995). Later it was also reported that paclitaxel was found to be more stable in CD solution than in buffer solution of comparable pH. Maximum stability of paclitaxel occurred at pH 3-5 range. Moreover, it was also found that paclitaxel formed predominantly second order complexes with the CDs and the complexes with HPβCD were found to be more stable than those of HPγCD or γCD. Further increase in solubility of paclitaxel was observed with addition of ethanol (Dordunoo and Burt, 1996). CDs though widely in use for new drug delivery systems possess serious limitations in case of paclitaxel. The drug tends to precipitate out of the cyclodexrin-drug complex on dilution with the aqueous media. To overcome this, increased concentration of CD will have to be used; but at this concentration, CDs may cause haemolysis. Thereby, this approach requires a lot of further research and consideration.

5.7. Local drug delivery

One of the therapeutic approaches to solid tumor is the surgical removal followed by irradiation and/or systemic chemotherapy to kill malignant cells which may have survived the surgery, and prevent metastasis and re-growth of tumor. Implanting a biodegradable device loaded with anti-neoplastic agent in the cavity created by the tumor provides high local concentration of the drug killing the malignant cells which survived the surgery and also prevent the systemic side-effects of the chemotherapy normally associated with the intravenous administration. Paclitaxel, which exhibits a significant systemic toxicity if given in form of prolonged local drug delivery following the surgical removal of the tumor will not only improve the efficacy but also reduce the systemic toxicity associated with it. Park et al. (1998) developed the disk shaped implants of polyanhydride P (FAD–SA, 50:50 w/w) loaded with 10% w/w of paclitaxel. It was found that paclitaxel was released very slowly and only 15% of the drug was released in 77 days and based on first order kinetics, it can be predicted that 44 months will be required to release 100% of the drug from the device, which is too prolonged for any clinical condition. However, the faster release of paclitaxel (45–65% in 30 days) was observed from a relatively more hydrophilic P (CPP–SA, 20:80 w/w) and the implants were found to be promising in an experimental malignant glioma model. Drug loaded polymeric disks using a biodegradable polyanhydride have been formulated for implantation into the cavity of resected brain tumors (Wu et al., 1994). Local interstitial delivery of paclitaxel in a rat model of malignant glioma resulted in high paclitaxel concentrations in the brain and increased median survival time (Walter et al., 1994). In addition, patients with malignant gliomas were treated with drug-polymer implants inserted into the tumor using a computerized tomography (CT) guided stereotactic method and showed a marked reduction in the tumor mass (Kubo et al., 1994). Recently (Dordunoo et al., 1997), a surgical paste based on poly (ξ-caprolactone) (PCL), a low melting (about 44–56 °C) biodegradable, and biocompatible polymer was developed. This paste is applied from a syringe in the molten state following gentle heating. Since, paclitaxel release is very slow (Winternitz et al., 1996) from PCL as it has a long degradation half-life and so they studied the effect of incorporation of various water-soluble additives into PCL matrix on release of paclitaxel. On in vivo studies they found that formulation consisting of paclitaxel/gelatin/PCL paste significantly reduced the tumor mass in mice. Factors such as the type of water-soluble agent, the microparticle size and the proportion of the additives were shown to influence the release characteristics of the drug.

5.8. Miscellaneous

Other formulation approaches include the conjugation of paclitaxel to the stable macromolecular drug carrier. Paclitaxel-albumin conjugate has been prepared in which the drug is covalently linked to human serum albumin through succinyl spacer (Dosio et al., 1997). Physico-chemical properties and anti-tumor activity of these macromolecular conjugates were evaluated in vitro and their pharmacokinetic properties and toxicities were studied in vivo in mice. The conjugates were stable in physiological solution and in serum, whereas the presence of proteases or liver extract caused the drug to be released in a linear fashion. When tested with three different cell lines, it was found that conjugates maintained high cytotoxicity with efficient cell binding and internalization followed by release of the drug inside the cell. The conjugate continuously released the drug to the plasma over the prolonged periods of time, thus providing a depot effect. Acute toxicity observed with the standard formulation of paclitaxel was greatly reduced in the albumin-conjugated preparation in mice.

6. Conclusion

Paclitaxel (Taxol®), the first taxane in clinical trials, is active against a broad range of cancers that are generally considered to be refractory to conventional therapy. This has lead to the regulatory approval of paclitaxel in the U.S. and many

other countries for its use in the palliative therapy of patients with ovarian and breast cancer resistant to chemotherapy. The challenge now is to develop strategies using paclitaxel in the initial therapy of cancer in which cure/improved survival may be an achievable goal. Considering all the above mentioned approaches; 'carrier delivery systems', including liposomes, micelles and particulate drug delivery systems, seem to be the best possible approach towards the ideal dosage form which would bypass all the present limitations and provide a desirable means and cure.

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