Advances of Paclitaxel Formulations Based on Nanosystem Delivery Technology

C. Luo, Y. Wang, Q. Chen, X. Han, X. Liu, J. Sun* and Z. He*

School of Pharmacy, Shenyang Pharmaceutical University, 103 Wenhua Road, Shenyang 110016, China

Abstract: In this paper, an overview of recent advances of paclitaxel formulations based on nanosystems is provided. Paclitaxel is very effective in the treatment of various cancers especially ovarian and breast cancer, but it demonstrates poor aqueous solubility, which results in the difficulty challenging the development of paclitaxel parenteral formulations, so its clinical application is greatly restricted. The conventional paclitaxel formulation uses Cremophor EL and ethanol to solubilize paclitaxel, which could cause severe side effects. Nanotechnology has been widely exploited in the field of antitumor research, and paclitaxel is no exception. In recent decades, a series of novel formulations of paclitaxel based on nanotechnology have been developed, including albumin-bound paclitaxel, polymeric micelle-formulated paclitaxel, polymer-paclitaxel conjugates, liposome encapsulated paclitaxel *etc*. The common advantage shared with these novel injectable formulations is that they are developed based on nanotechnology and Cremophor EL-free. In addition, these nanoformulations can significantly reduce toxicities of paclitaxel and greatly promote its antitumor efficiency.

Keywords: Paclitaxel, poor water solubility, solvent related side effects, paclitaxel formulations, cremophor EL-free, nanosystems, nanotechnology, antitumor efficiency.

INTRODUCTION

Paclitaxel has a special chemical structure (Fig. **1**), a complex diterpene having a taxane ring with a four-membered oxetane ring and an ester side chain at position C-13. Paclitaxel has a unique mechanism of action, enhances the polymerization of tubulin to stabilize microtubules and interacts with microtubules to prevent depolymerization. It is inimitable among all chemotherapeutic agents, contributing to its specific binding site on microtubule, no need guanosine triphosphate and microtubule-associated proteins to protein to polymerize tubulin [1].

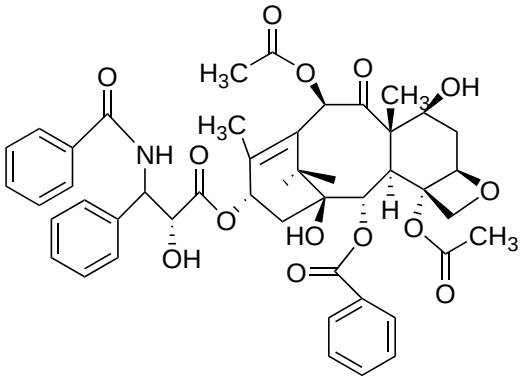


Fig. (1). Chemical structure of paclitaxel.

*Address correspondence to this author at the Department of Biopharmaceutics, School of Pharmacy, Shenyang Pharmaceutical University, 103 Wenhua Road, Shenyang 110016, China;
Tel: +86-24-23986321; Fax: +86-24-23986321;
E-mail: hezhonggui@gmail.com, sunjin66@21cn.com

The administration of many anticancer drugs is limited by their unsatisfactory properties, such as poor solubility, narrow therapeutic window, drug toxicity, side effect and resistance. In a little over 15 years, paclitaxel has become among the most clinically useful chemotherapy agents [2]. Paclitaxel has shown encouraging antitumor activity and plays a major role in cancer chemotherapy. Unfortunately, it shows poor aqueous solubility and multi-drug resistance. Previous studies [3-6] showed that paclitaxel resistance has been linked to: (a) alterations in tubulin; (b) expression of the P-gp 170 drug-efflux pump; (c) high Raf-1 kinase activity; and (d) over expression of HER-2/neu. Due to its poor aqueous solubility, the conventional paclitaxel formulation was firstly prepared using Cremophor EL and ethanol to solubilize paclitaxel. However, Cremophor EL related toxicity is serious in clinic, including histamine release, severe anaphylaxis, hyperlipidemia, abnormal lipoprotein patterns, aggregation of erythrocytes, and prolonged, sometimes irreversible sensory neuropathy [11].

Given all the disadvantages of the paclitaxel and its conventional formulation, there is significant interest to develop novel Cremopher EL free formulations that can selectively enhance their antitumor activity, reduce toxicity profile, target tumor tissues and perform pharmacological properties. Many efforts have been made to improve its solubility and antitumor activity, and this paper summarized several high-quality and high-impact formulations (Fig. **2**). Among these, some formulations have successfully put in the market, including Abraxane (paclitaxel albumin-stabilized nanoparticle), Genexol-PM (polymeric micelle-formulated paclitaxel), Xyotax™ (poly(L-glutamic acid)-paclitaxel), Lipusu (liposome encapsulated paclitaxel) and Nanoxel (nanomicelles). Some other formulations entered into the stage of clinical study. All these novel formulations

1875-5607/12 $58.00+.00

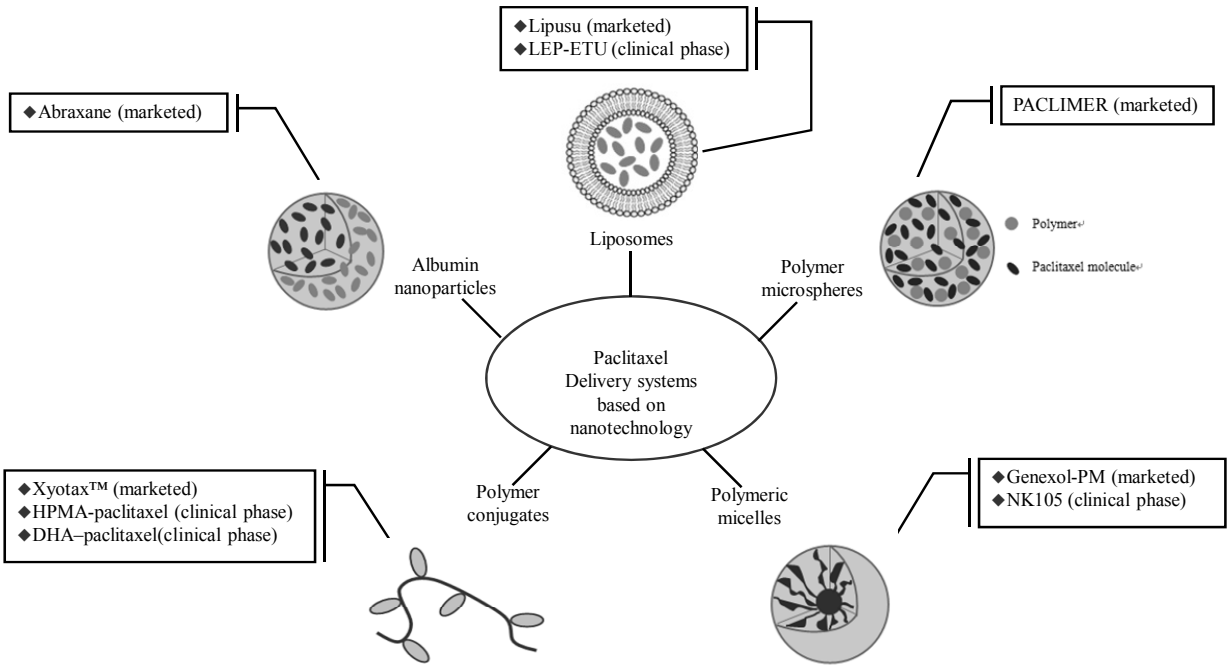


Fig. (2). Schematic illustration of various paclitaxel delivery systems based on nanotechnology and some formulations.

demonstrated greater antitumor activity, overcame poor solubility of paclitaxel, and reduced side effect. What is more, these formulations revealed more satisfactory pharmacokinetic profiles and performed greater therapeutic activity than the conventional paclitaxel formulations.

Colloidal drug delivery systems have been widely developed for their use in cancer therapy. The superiority of colloidal disperse systems for drug delivery can be attributed to their small size, enhanced solubility , reduced toxicity, controlled release of the drug and improvement of drug pharmacokinetics and biological distribution. With the development of biodegradable materials and nanothechnology, Nanosystems have become promising drug delivery systems for the delivery of drug and gene. Used as drug delivery, nanocarriers are generally less than 1000 nm in at least one dimension, nano-vehicles can overcome many problems in drug delivery, which can been taken up by cells more efficiently and can control the fate of a drug entering the biological environment [7]. Nanocarriers are well-suited vehicles for targeted tumor delivery. The ability of nanoparticles to gather at a target site is dependent on nano vehicles, properties of target sites and the interactions between them. The physicochemical properties of the nano-vehicles itself and the selection of targeting ligands to the surface of nano-carriers still are key design parameters to improve therapeutic efficacy [8]. In most pharmaceutical applications, nanosystems are applied as passive drug vehicles for systemic delivery, limited penetration across the vascular endothelium and uptake by the reticuloendothelial system (RES) are two major challenges to nanosystems delivery into tissues. To achieve long circulation and penetration more drugs into target tissues, Surface Modification of Nanoparticles such as grafting hydrophilic polymers onto the nanoparticle and aiming at specific receptors or antigens expressed on the surface of tumor tissue and cells shows great superiority [9].

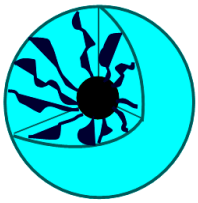
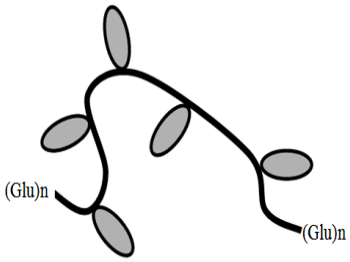
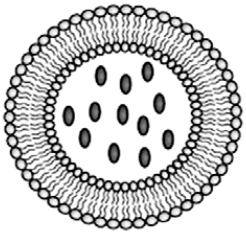
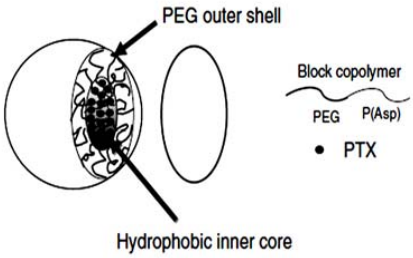
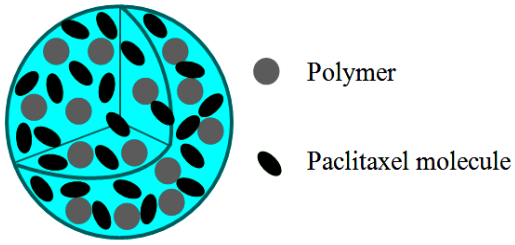
Nanotechnology-based drug delivery systems have played a key role in these novel and Cremophor EL-free paclitaxel formulations. In recent decades, many drug delivery nanosystems have been developed, including nanosuspension, nanoemulsion, polymeric micelles, polymeric conjugates, liposomes, nanocrystalline *etc*. The nanosystems are appropriate for encapsulating insoluble and soluble drugs, and even for macromolecules, proteins and genes. In summary, nanosystems are capable of enhancing solubility and dispersion, improving oral absorption and bioavailability, reducing toxicity and solvent related side effects, targeting selectively the tumor area, controlling drug release and even gene engineering.

Nanotechnology then provides new drug delivery platform for tumor treatment, and has been becoming the hottest research region of drug delivery system. paclitaxel formulations based on nanotechnology have achieved much success. In this paper, we review the development of paclitaxel and its formulations on the market, the physicochemical properties (Table **1**), antitumor activity and pharmacokinetic profiles of paclitaxel formulations. The unique features and promising advantages are highlighted, and we have analyzed the safety and effectiveness of these novel formulations by comparing their antitumor ability, related toxicity and pharmacokinetics profiles with the conventional paclitaxel formulation.

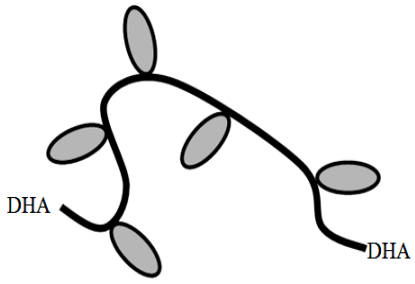
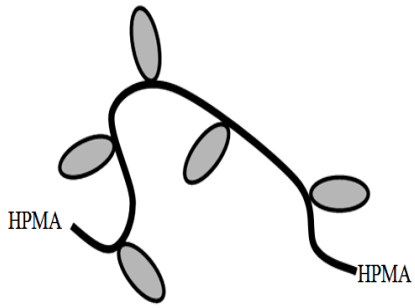
THE DEVELOPMENT OF PACLITAXEL AND ITS FORMULATIONS ON THE MARKET

As clearly shown in Fig. (**3**), development of paclitaxel began in 1960s, when America chemists M.C.Wani and

Table 1. Structures and Main Physicochemical Properties of Some Paclitaxel Formulations

Structures	Physicochemical properties	Reference
 Genexol-PM	The polymeric micelles are composed of hundreds of amphiphilic diblock copolymers and have a diameter of 20–50 nm.	[17]
 Xyotax™	Conjugated paclitaxel represent~37%, by weight, of the conjugate, equivalent to about one paclitaxel ester linkage per11glutamic acid residues of the polymer.	[22]
 Lipusu	Lipusu is prepared in form of liposome.	[24]
 NK105	NK105 contains 23% (w/w) of paclitaxel. The weight-average diameter of the nanoparticles was approximately 85 nm ranging from 20 to 430 nm.	[31]
 PACLIMER	PACLIMER, paclitaxel (10%) is encapsulated in a proprietary polymer in the form of microspheres.	[32]

(Table 1). Contd…..

Structures	Physicochemical properties	Reference
 DHA-paclitaxel	DHA-paclitaxel, a novel conjugate formed by linking the natural fatty acid DHA to paclitaxel	[37]
 HPMA-paclitaxel	the polymer: paclitaxel ratio was approximately 19:1 w/w. which is very water soluble (>2 compared to >0.0001 mg/ml for paclitaxel)	[40]

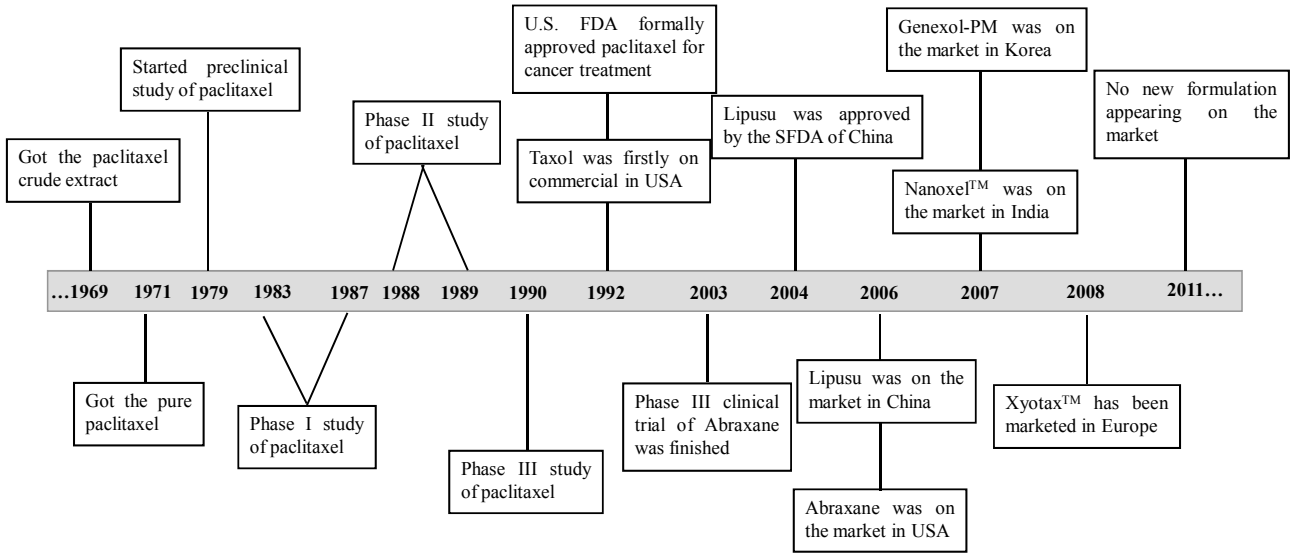


Fig. (3). Timeline of paclitaxel and its formulation commercially, general events are presented.

Monre C.Wall got the paclitaxel crude extract from pacific yew. And in 1969, researchers found that paclitaxel is the active agent of the crude extract. In 1971, Wani and Wall got the pure paclitaxel from pacific yew bark. However, the importance of paclitaxel was not recognized until the late 1970s, because it is difficult to obtain and very poor soluble. In 1979, paclitaxel was selected as a candidate to start preclinical study, and paclitaxel was observed to be effective for breast cancer, colon cancer, bronchial cancer, and endometrial cancer. In the same year, Susan Horwitz discovered that paclitaxel has a unique mechanism of action on the tumor. The most impressive anti-tumor activity was demonstrated in NCI tumor screening [2]. Phase I study of paclitaxel began in 1983, and determined the maximum human tolerated dose and dose limiting toxicity between 1983 and 1987. Phase II study was carried out between 1988 and 1989, and phase II clinical trial results showed that paclitaxel have a significant effect for advanced ovarian

cancer and metastatic breast cancer. Phase III clinical trial started in 1990, mainly determined the dosage, time and effectiveness of paclitaxel.

Due to its poor solubility, the early available commercial formulation Taxol contains Cremophor EL (polyethoxylated castor oil) and ethanol as excipients to solubilize paclitaxel. Food and Drug Administration (FDA) of American formally approved Taxol for the treatment of advanced ovarian cancer, lung cancer, uterine cancer in 1992. However, Cremophor EL can cause severe side effects, for example, hypersensitivity reactions, nephrotoxicity and neurotoxicity [9]. In addition, paclitaxel is a substrate for the P-gp transporter which prevents it effectively gaining access to tumor tissue [10-11]. Therefore, to develop novel formulations for paclitaxel delivery is of a great essence, and some formulations have successfully reached the market over the last decades.

Taxol was the first formulation of paclitaxel on the market, in which Cremophor EL was used to deliver paclitaxel. Taxol was firstly approved in America in 1992. Although some Cremophor EL related side effects were observed in clinical study, Taxol can allow paclitaxel prepared into the clinically used dosage form. Abraxane, a representative of nanotechnology-based formulations on the market, is paclitaxel albumin-bound nanosuspension. Its phase III clinical trial was finished in 2003, and FDA had approved it mainly for treatment for breast cancers in 2006. Abraxane is really a breakthrough in the development of paclitaxel formulations, which owes the first formulation using nanotechnology to construct formulation on the market. Lipusu is a well characterized novel lyophilized liposome-based paclitaxel formulation, which has been approved by the State Food and Drug Administration of China in 2004. It is the first paclitaxel liposome injection which had been on the market in China in 2006. Samyang Corporation (Seoul, Korea) has developed a novel paclitaxel formulation-Genexol-PM, which is a sterile lyophilized polymeric micelle composed of methoxy poly(ethylene glycol)-L-poly(lactide) [mPEG-PLA] loaded paclitaxel without Cremophor EL, and Genexol-PM had been on the market in Korea in 2007. Xyotax™-Poly (L-glutamic acid)-paclitaxel (PGA-TXL) is a water-soluble paclitaxel conjugate. Xyotax™ has been marketed in Europe in 2008, and now in the clinical study in American. Nanoxel^{TM} is also a Cremophor EL-free soluble formulation, which had got an approval from the Drug Controller General of India (DCGI) for its anti-cancer drug delivery formulation, and had been approved early by the DCGI in 2006 for the treatment of breast cancer.

FORMULATIONS ON THE MARKET

Paclitaxel has become the most commonly used chemotherapeutic agent in the world, after the identification of it as the active constituent extract of the Pacific yew Taxus brevifolia [12], and the characterization of its novel microtubule activity [13]**.** Novel formulations of paclitaxel have been on the market including Abraxane, Genexol-PM, Xyotax™, Nanoxel and Lipusu. All these formulations possess the common superiorities as follows: (a) free of Cremophor EL and avoiding the related side effects; (b) high loading dose of paclitaxel without additional toxicity; (c) tumor targeting ability of nanosystems and fewer side effects. Due to the above improvements, these formulations have been used as first-line chemotherapy agents. In clinical and pharmacokinetic studies, these novel formulations permit the administration of higher paclitaxel doses than conventional paclitaxel, receiving greater antitumor efficiency but fewer side effects. Antitumor activity and pharmacokinetics profiles of some formulations on the market are shown in Table **2**. The achievement of a higher dose in the case of Cremophor EL -free paclitaxel formulations may be explained by the absence of the Cremophor EL -associated influence of the pharmacokinetics of paclitaxel, and the properties of nanosystems put great influence on the pharmacokinetics of paclitaxel. Paclitaxel has been prepared with a variety of formulations to improve its solubility and pharmacokinetics. Clinical and pharmacokinetics study fully demonstrated the superiorities of these formulations.

Abraxane

Among these formulations on the market recent years, Abraxane developed by American BioScience, Inc., Santa Monica, California, is a Cremophor EL-free, 130-nm particle form of albumin nanosuspension loading paclitaxel. The use of albumin as a vehicle avoids the solvent-related toxicities. Furthermore, Abraxane has the potential to increase drug delivery to tumors by initiating albumin receptor (gp60)-mediated transcytosis across endothelial cells and accumulating the drug in tumors due to binding of albumin to secreted protein, acidic and rich in cysteine [14]. Another research has shown that albumin-bound paclitaxel greatly improved the efficacy of radiotherapy for two murine cancer (OCa-I and MCa-4) when the combined use of albumin-bound paclitaxel and radiation [15]. Abraxane employed as paclitaxel albumin-bound nanosuspension, had been approved for metastatic breast cancer by the Food and Drug Administration in the USA. A phase I study had been done, the novel formulation was safely administered as a short i.v. infusion without steroid or antihistamine premedication [16]. The Maximum tolerated dose (MTD) was 300 mg/m^2 given over 30 min once every 3 weeks. Alopecia occurred in all patients, neurotoxicity and mucositis were dose limiting. Grade IV granulocytopenia and one episode of superficial keratopathy were seen at 375 mg/m^2. In a phase III clinical research, 3-week cycles of Abraxane (260 mg/m^2 IV over 30 min without premedication) was compared with Taxol (175 mg/m^2 IV over 3 h with premedication) in 454 patients with metastatic breast cancer [11]**.** Response rates and time to tumor progression (TTP) were significantly higher with Abraxane than with Taxol. The incidence of grade IV neutropenia in the phase III study was significantly lower with Abraxane, despite a higher paclitaxel dose. Abraxane pharmacokinetic parameters were linear over the clinically relevant dose range. The AUC for Abraxane was lower than that of the same dose of conventional paclitaxel formulation, which potentially predicts improved tissue distribution of albumin-based paclitaxel. The clinical researches and pharmacokinetics profiles indicated the improvement in the field of safety and therapeutic efficiency.

Genexol-PM

Genexol-PM, another novel paclitaxel nanotechnology-based formulation, demonstrated significant anti-tumor activity in patients with nonsmall-cell-lung cancer (NSCLC). Genexol-PM is one kind of polymeric micelles, which is composed of methoxy poly(ethylene glycol)-L-poly(lactide) [mPEG-PLA] loaded paclitaxel without Cremophor EL. Genexol-PM has been shown to be effective in targeting deliver the chemotherapeutic agent to tumor tissue. Genexol-PM is believed to be superior to the conventional paclitaxel formulation in terms of the absence of premedication and the delivery of higher paclitaxel doses without additional toxicity [17]. Targeting effect permits a higher administrated dose with less toxicity. Due to membrane transport *via* endocytosis, Genexol-PM polymeric micelles would accumulate in lysosomes around nuclear, thereby chemotherapeutic agents released from the polymeric micelles closer to nuclear targets, which may improve cellular toxicity. Genexol-PM allowed administration of higher doses of paclitaxel than Taxol. The biodistribution of Genexol-PM showed 2–3-folds higher levels in various tissues, including liver, spleen, kidney, and lung, and more importantly in tumors. The MTD of Genexol-PM was 390mg/m^2, and which of Abraxane was 300mg/m^2 [16]. The Dose limiting toxicities (DLTs) were myalgia, sensory and motor neuropathy, and neutropenia. What is more, Genexol-PM plus cisplatin combination chemotherapy showed significant antitumor activity with relatively low incidence of toxicity in patients with advanced NSCLC [18]. Pharmacokinetic behaviors of Genexol- PM displayed a tendency to be linear, except at a dose of 230 mg/m^2. As compared with conventional paclitaxel formulation, Genexol-PM showed lower AUC and a shorter plasma $t_{1/2}$, accounting for the increased collection of Genexol-PM into the tumor tissues. This enhancement is certified by the discovering that the higher paclitaxel concentration was found in the tumor in a preclinical study of Genexol-PM [19].

Xyotax™

Polymer-drug conjugate has been developed as one approach to overcome multi-drug resistance and to improve the therapeutic efficacy. Xyotax™-Poly (L-glutamic acid)-paclitaxel (PGA-TXL) is a water-soluble paclitaxel conjugate by linking paclitaxel to poly (l-glutamic acid) *via* ester bonds. PGA-TXL was prepared by carbodiimide-mediated, ester coupling of hydroxyl groups of paclitaxel and carboxyl groups of glutamic acid (as PGA) [20]**.** L-glutamic acid was chosen as a potential carrier unit for paclitaxel based on the following rationale [21]: (a) PGA contains a large number of side chain carboxyl functional groups for drug attachment: (b) PGA can be readily degraded by lysosomal enzymes due to its nontoxic basic component, L-glutamic acid; and (c) sodium glutamate has been reported to prevent manifestations of neuropathy induced by paclitaxel, thus enabling higher doses of paclitaxel to be tolerated. In a phase I and pharmacokinetic study, 19 patients were investigated on the 3-weekly phase Ia study and 11 patients on the 2-weekly phase Ib study. In the phase Ia study, the maximum tolerated dose was 233 mg/m^2, two partial responses were observed, and DLTs were neutropenia and neuropathy. Pharmacokinetic study indicated a prolonged half-life of >100 hours and limited volume of distribution for conjugated paclitaxel. The plasma concentrations of conjugated paclitaxel were 10-to100-fold higher than those of unconjugated paclitaxel [22]. The pharmacokinetic data of conjugated paclitaxel indicates that the distribution of conjugated paclitaxel is mostly collected to plasma.

Nanoxel™

Based on principles of nanotechnology, Nanoxel™ is a Cremophor EL-free soluble formulation, which is proved as an effective and safe chemotherapeutic agent for advanced breast, non-small-cell lung and ovarian carcinomas. Smooth spherical particles of 80 -100 nm were observed by scanning electron microscopy. Paclitaxel was found to retain the *in vitro* cytotoxicity in nanomicellar particles, and Nanoxel™ showed up to three-fold higher uptake of paclitaxel in target cancer cells as compared to the conventional formulation, which resulted in an increase in tubulin stabilization and induction of apoptosis. Transmission electron microscopy demonstrated that the nanomicelles were endocytosized and entrapped in the acidic spherical compartment. Just as Genexol-PM, Nanoxel™ permitted a higher administrated dose without additional toxicity. Hundreds of patients have benefited from this novel formulation. The phase I data showed that Nanoxel™ had linear pharmacokinetics and can be administered without premedication. The phase II data in metastatic breast cancers showed comparable efficacy and better safety profile than the traditional paclitaxel formulation [23].

Lipusu

Among the novel drug delivery systems, liposomes represent a mature, versatile technology and possess considerable potential for entrapment of both lipophilic and hydrophilic drugs. Lipusu was a well characterized novel lyophilized liposome-based paclitaxel formulation. The mean particle size was about 150 nm before and after lyophilization, and the drug entrapment efficiency was more than 90%, and Lipusu has a stable size distribution and high encapsulation efficiency. Stability data indicated that the lyophilized Lipusu was physically and chemically stable for at least 12 months at 25℃ [24]. Lipusu was found to have the similar anti-tumor activities *in vitro* and *in vivo*, but its toxicity is lower than that of paclitaxel injection under the same dosage [25]. Kong *et al.* showed that the response rate (RR) is 39.1% in the treatment of NSCLC patients with Lipusu and cisplatin [26]. Chen *et al.* compared Lipusu with conventional paclitaxel on treatments of breast cancer and NSCLC and demonstrated that both of them have similar efficacy but the former reduces the incidence of serious hypersensitive reactions significantly more than the latter [27]. Lipusu has demonstrated the ability to modulate multi-drug resistance in human ovarian cancer cell lines and antitumor activity in mice models [28]. Straubinger *et al.* had shown that the MTD of paclitaxel liposome is 200 mg/kg, while the conventional paclitaxel dosage is 30 mg/kg [29]. In a clinical study on the premedication of paclitaxel liposome, 53 patients were involved in the research, 27 patients experienced grade I-II, and 2 grade III-IV anemia. Grade III

Table 2. Antitumor Activities and Pharmacokinetics Profiles of Formulations on the Market

Trade name	MTD	DLTs	Antitumor activity	PK	Number of patient	reference
Abraxane	300 mg/m^2	Neurotoxicity and mucositis	The ORR for Abraxane was 16.3%, 7 confirmed partial responses and the disease control rate was 48.8%	linear pharmacokinetics over the dose range of 135–300 mg/m^2	46	[16]
Genexol-PM	390 mg/m^2	neuropathy, myalgia, and neutropenia	3 partial responses (14%), 6 patients (28%)remained stable	excluding 230 mg/m^2, the pharmacokinetics was linear, The AUC and C_{max} of Genexol-PM revealed lower values than equivalent doses of Taxol	21	[17]
Xyotax™	233 mg/m^2	neutropenia	2 partial responses	Showed longer halftime, and high paclitaxel peak concentrations (C_{max}) are avoided	19	[22]
Lipusu	200 mg/kg	Neutropenia, nausea, vomiting	The median number of Lipusu based cycles was 3, toxicities were mild.	$t_{1/2}$was approximately 10-fold greater, V_t was more than 2-fold greater	53	[25]

and grade IV leucopenia occurred in 9.4% (5/53 patients). The major nonhematologic toxicity was the evaluation of ALT, which was observed in 30.2% (16/53) of the patients. Grade I and II nausea and vomiting occurred in 16.9% (9/53) and diarrhea occurred in 9.4% (5/53) of the patients, it demonstrated higher efficacy but less toxicities [29].

All above the formulations entered in market all have their own advantages, which make them special and superior in cancer treatment. Abraxane is prepared in the form of nanosuspension, with paclitaxel in forms of nanocrystalline. Genexol-PM and Nanoxel^{TM} are prepared as nanomicelles, small size of particles also allows greater water solubility, higher doses and targeting result in excellent antitumor activity. Xyotax™ is prepared by directly conjugating paclitaxel with poly (l-glutamic acid) to form polymeric conjugates. Lipusu-delivery paclitaxel is based on a liposome-based paclitaxel formulation, with stable size distribution, high encapsulation efficiency and better stability. The dose for therapy of Toxal is175mg/m^2, but these novel formulations all permit higher dose without serious toxicity for cancer treatment. What is more, these formulations make paclitaxel gather in tumor tissues, resulting in greater antitumor efficiency and smaller toxicities toward healthy tissues. Prolonging half-life produces longer effect of treatment, as the half-life of Xyotax™ was prolonged more than 100 hours. Pharmacokinetics profiles of these formulations tend to be liner, which is not the case for free paclitaxel.

FORMULATIONS STAYING IN CLINICAL STUDY

Striking progresses have been achieved in the paclitaxel delivery systems for cancer treatment, and lots of works have been staying in clinical studies. They illustrated more excellent properties and receive more satisfied efficiency than formulations before. These formulations are constructed in various novel nanosystems, which further improve the safety, efficiency and targeting. Here the constructions, characteristics, antitumor efficacy and pharmacokinetics profiles (Table **3**) of some new formulations which stay in clinical studies are reviewed.

NK105

Recently, paclitaxel was incorporated into the polymeric micelles from the PEG-b-P (Asp) modified with 4-phenyl-1-butanolate, and this formulation was termed as NK105. NK105 was obtained as a freeze-dried formulation and contained 23% (w/w) of paclitaxel. The weight-average diameter of the nanoparticles was approximately 85 nm ranging from 20 to 430 nm [9]. Due to the coating of PEG in the outer shell of the micelles, NK105 is soluble in water. In the phase I study, NK105 was administered as a 1-h intravenous infusion every 3 weeks, without antiallergic premedication, and NK105 was well tolerated with 180 mg/m^2 as the MTD, and the DLT was neutropenia [30]. Pharmacokinetic profiles had shown that the plasma AUC of NK105 at 150 mg/m^2 was approximately 15-fold higher than what of the conventional paclitaxel formulation. NK105 exhibited slower clearance from the plasma than free paclitaxel. Furthermore, the half-life at the terminal phase ($t_{1/2}$) was 4–6 times longer for NK105 than for the conventional paclitaxel formulation. The maximum plasma concentration (C_{max}) and AUC of NK105 were approximately 3 and 15 times higher than for the conventional paclitaxel formulation, respectively [30]. In addition, besides its enhancement of antitumor activity, NK105 also can enhance radiotherapy activity [31]. Combined NK105 treatment with radiation showed significantly superior antitumor activity as compared to combined free paclitaxel treatment with radiation. This effect has been attributed to its effect of preventing microtubules depolymerization and inducing cell cycle block at the G2/M phase, the most radiosensitive phase of the cell cycle.

Paclical®

A disadvantage of Taxol is that the side effects of Cremophor EL limit the administration dose. An excipient which does not cause side effects on its own would make it possible to administer higher doses of paclitaxel, hereof Oasmia Pharmaceutical AB Uppsala, Sweden has developed a novel excipient that resembles Vitamin A (XR-17). With the utilization of XR-17, Oasmia has managed to produce a water soluble formulation of Paclitaxel (Paclical®), which does not require premedication. Cremophor EL can cause related side effects, but XR-17 reveals its advantage that it does not cause side effects on its own. The main indication is ovarian cancer. Other planned indications are lung cancer (NSLC) and malignant melanoma [32]. An initial study with Paclical® in patients was carried out by Oasmia Pharmaceutical AB, Uppsala, Sweden. Totally 34 patients with advanced cancers were treated, the maximum tolerable dose of the candidate was set to 250 mg/m^2, no unexpected side-effects or hypersensitivity reactions were observed, and all dosages of Paclical® were well tolerated by most patients. The reported adverse events were expected relating with the paclitaxel, nothing to do with the XR-17. The stabilisation of the cancer diseases was seen in about half of the patients treated with 3 cycles [32].

PACLIMER

PACLIMER, containing 10% (w/w) paclitaxel encapsulated in polymer in the form of microspheres, is a controlled release formulation of paclitaxel. In this form, paclitaxel was incorporated into the biodegradable polymer poly (D, L-lactide co-ethyl phosphate) as the form of microspheres. The size of the microspheres ranges from 20 to 200 um in diameter, with a median diameter of 53 um [33]. In a phase I and pharmacokinetic study, 13 patients were treated. PACLIMER was designed to provide a sustained-release form of paclitaxel after intraperitoneal (IP) administration [34]. They found that IP administration of paclitaxel microspheres was well tolerated up to 1200 mg/m^2 without defining MTD. DLTs consisted of abdominal pain, ileus and bowel obstruction. Patients received up to 1200 mg/m^2 without further evidence of DLT. Pharmacokinetic analysis showed a trend toward a dose-dependent effect of IP paclitaxel microspheres. Sustained paclitaxel levels were maintained throughout all 8 weeks of therapy, however, paclitaxel concentrations were below the plasma concentrations associated with toxicity. In human lung cancer xenografts, intratumoral injection of paclitaxel microspheres demonstrated that paclitaxel was released slowly from the microspheres with<80% released after 90 days [33]. The efficacy of systemic chemotherapy for non-small cell lung cancer (NSCLC) has been improved with newer systems and agents.

Taxoprexin

Docosahexaenoic acid (DHA)-paclitaxel, a novel conjugate formed by linking the natural fatty acid DHA to paclitaxel, was designed as a prodrug targeting tumor tissues. DHA-paclitaxel is a 2-O-acyl conjugate of the natural fatty acid Docosahexaenoic Acid linked to paclitaxel *via* an ester linkage. Preclinical study suggests that there is an increased fatty acid uptake in tumors, presumably for use as biochemical precursors and energy sources [35]. Taxoprexin significantly enhanced tumor distribution and antitumor activity in various tumor models as compared with paclitaxel solution [36]. DHA-paclitaxel is a 4-fold weaker substrate for P-glycoprotein than paclitaxel and may be active in some drug-resistant tumors that over express P-glycoprotein [36]. The relative conversion of DHA-paclitaxel to paclitaxel was 21-fold higher in tumor than in plasma [37]. In a phase I and pharmacokinetic study, 24 patients received 78 cycles of DHA-paclitaxel over five dose levels (200–1100 mg/m^2), and neutropenia was the most frequent DLT after administration of DHA-paclitaxel as a 2-h infusion repeated every 21 days. DHA-paclitaxel exhibited a long terminal half-life (112 h). DHA-paclitaxel administration resulted in a dramatically altered paclitaxel plasma pharmacokinetic profile. Although paclitaxel AUC values were similar, DHA-paclitaxel administration produced about 10-fold lower paclitaxel C_{max} values and 5-fold longer apparent half-life for paclitaxel. Prolonged exposure to low paclitaxel concentrations may produce greater antitumor activity [38].

PNU166945

PNU166945 is a novel polymer-conjugated of paclitaxel. The agent contains a HPMA polymer, which is linked through an amino acid chain at the 2' position of paclitaxel. The polymer-drug is water-soluble, it can be dissolved and administered without Cremophor EL. No signs of hypersensitivity, neurotoxicity or cardiovascular toxicity were observed in the acute and subchronic toxicity studies with PNU166945 in rodents or dogs [39]. Preclinical results from mice studies have been reported for several HPMA bound drugs, but the results of clinical studies with HPMA copolymers are limited. Toxicity studies with PNU166945 in mice, rats and dogs indicated that the DLT was myelotoxicity, with no signs of hypersensitivity, neurotoxicity or cardiovascular toxicity [39]. In a phase I and pharmacokinetics study, 12 patients were accrued for the study. PNU166945 administered as a 1-h i.v. infusion was well tolerated and the highest investigated dose of PNU166945 was 196 mg/m^2, no DLTs were observed at this dose level. Pharmacokinetic study showed a linear increase was observed in the AUC. What is more, the pharmacokinetics results suggest an improved pharmacokinetic behavior with potential controlled release of paclitaxel [40].

NanoTax

Precipitation with compressed antisolvents (PCA) has been widely used as a technique to produce nanoparticles. Paclitaxel nanoparticles, termed as Nanotax®, ware successfully gained from acetone solution by using compressed CO_2 as an antisolvent for the drug based on PCA [41, 42]. Nanoparticles of paclitaxel, rang in size from 600-800 nm. Intravenous delivery of NanoTax suspension formed in physiological saline exhibited the same therapeutic effect in the treatment of mice bearing ovarian cancer as Taxol®, but without the adverse effects of Cremophor®. However, cancer-bearing mice treated intraperitoneally with NanoTax suspension performed much better effect as compared to similar treatments with either

Table 3. Antitumor Activities and Pharmacokinetics Profiles of Some Formulations Staying in Clinical Study

Name	MTD	DLT	Antitumor activity	PK	Number of patient	Reference
NK105	180 mg/m^2	neutropenia	1 partial response, and the size of the liver metastasis decreased by more than 90%	The C_{max} and AUC increased as the doses were escalated from 10 to180 mg/m^2	19	[30]
HPMA-paclitaxel conjugates	196 mg/m^2	No DLTs were observed	1 partial response, 2 patients at a dose of 140 mg/m^2 had stable disease	Showed dose proportionality, a linear increase was observed in the AUC	12	[40]
Paclical	250 mg/m^2	No unexpected side-effects were observed	about half of the patients shown a stabilisation of the cancer diseases	Shown linear pharmacokinetics	34	[32]
PACLIMER	1200 mg/m^2 without defining MTD	Abdominal pain, ileus and bowel obstruction	3 patients were progression-free for at least 6 months after completing therapy	Showed dose-dependent effect of IP administration, sustained paclitaxel levels were maintained 8 weeks with low toxicity	13	[34]
DHA–paclitaxel conjugate	1100 mg/m^2	Neutropenia	3 for adverse events, antitumor activity was confirmed in 1 patient at 1100 mg/m^2	C_{max} and AUC(24 h) increased in near proportion with dose level, and exhibited a small Vss, a long terminal half-life	24	[38]

macroparticulate paclitaxel or Taxol® [43]. The results of preclinical studies were very encouraging, and Nanotax is currently enrolling patients in a phase I clinical trial for peritoneal cancers to evaluate the safety, pharmacokinetics and preliminary efficacy of an intraperitoneally administered suspension [44].

In general, among these paclitaxel formulations staying in clinical study, NK105 is prepared as polymeric micelles, due to the PEG outside, it has greater water solubility, PEG also confers a stealth property to the formulation, which allows the micellar drug preparation to be less taken up by the reticuloendothelial system (RES) and to be retained in the circulation for a longer time. And the enhanced permeability and retention (EPR) effect causes accumulation of paclitaxel in tumor tissues. PACLIMER is prepared in forms of microspheres, paclitaxel was released from the PACLIMER delivery system primarily by degradation of the polymer. DHA-paclitaxel and HPMA-paclitaxel both are conjugates with paclitaxel, the advantage of polymeric drug delivery systems is that they enable tumor targeting, and accumulation may occur at the tumor tissues due to the increased permeability of tumors [45]. Enhanced local target can be achieved, and the drug is able to release itself from the carrier at the tumor tissues. Chemotherapy drugs conjugated to fatty acids could enhance tumor targeting, this concept led to the synthesis of the new chemical entity DHA-Paclitaxel. And these copolymer carriers usually resulted in long-circulating conjugates delivery systems, which may result in sustained release or controlled release. Depending on novel nanosystems, higher dose without greater toxicity for cancer therapy has become a trend for cancer treatment, just as PACLIMER was well tolerated up to 1200 mg/m^2 without defining MTD and the dose of DHA-paclitaxel for treatment also reached to 1100 mg/m^2, however, paclitaxel concentrations were well below the plasma concentrations associated with toxicity. They had longer $T_{1/2}$ than conventional formulation even than those ones on commercial. Among these, PACLIMER released slowly and performed as a sustained-release formulation.

PACLITAXEL FORMULATIONS BEFORE CLINICAL STUDY

Researches on nanosystems to deliver paclitaxel are still continued, researchers are mainly focus on its safety and efficiency. Extended release and targeting towards tumor tissues can successfully improve this situation. Moreover, active targeting therapy is significantly superior to passive targeting therapy based on the EPR (enhanced permeability and retention) effects. A lot of works focus on constructing nanosystems and enhancing targeting have been done in recent years. Novel extended release delivery system include chitosan-polymethacrylic acid copolymers [46], paclitaxel-loaded c(RGDyK)-Poly(ethylene glycol)-blockpoly(lactic acid) micelle (c(RGDyK)-PEG-PLA-PTX) [47], oral microemulsions of paclitaxel [48], and poly(ethylene oxide)-block-poly(ε-caprolactone) (PEO-b-PCL) copolymers bearing paclitaxel *etc*. Active targeting related researches including Anti-HIF-1α antibody-conjugated pluronic triblock copolymers encapsulated Paclitaxel [49], peptide-conjugated biodegradable nanoparticles [50], *etc*. Which have the following advantages: (a) to deliver anti-cancer drugs to objective tumor tissues; (b) to decrease therapeutic dose; (c) to reduce toxicity of normal cells and to reduce the side effects. Therefore, developments of active targeted and long effecting formulations have a profound significance in clinical tumor therapy.

CONCLUSIONS AND FUTURE DIRECTIONS

Antitumor drug would produce damage to normal tissues as it was administrated to kill tumor. The threat of severe side effects caused by blind distribution of the drugs has meant that maximum dosages are needed to be restricted. However, using nano-technology to design novel drug delivery systems is promising to solve this problem. Among these reviewed formulations in this paper mostly are nanosystems, and they mainly deliver active agents by using drug carriers, the property of which significantly affect the *in vivo* behavior of drug. The advantages of nanosystems are apparent: increasing the solubility and stability of drug, enhancing the administration safety and improving pharmacokinetic behavior.

Although nanosystems have achieved much success to deliver drug, there still are many shortcomings, and there will be a lot of work need to be done in the future. Some aspects of nanotechnology are not yet mature. For example, most nanosystems exist some problems such as low drug loaded. Moreover, studies on *in vivo* destiny of nano-formulations should be continued. Therefore, efforts to enhance drug loadings and design long effecting and active targeting drug carrier will be very meaningful. The receptor and ligand, antibody and antigen, and characteristics of tumor tissues can be used to achieve the purpose of targeting. Using antibody and antigen is restricted to tissues expressing high levels of related antigens. However, As more new-type polymers being synthetized and novel preparations and technologies being developed, designing active target drug delivery system by modifying nanocarriers based on the characteristics of tumor tissues will greatly improve the efficiecy of cancer treatment.

CONFLICT OF INTEREST

None declared.

ACKNOWLEDGEMENTS

We would like to acknowledge the support of this work by Key Project for Drug Innovation (2009ZX09301-012 and 2010ZX09401-304) and the National Basic Research Program of China (973 program, 2009CB930300).

REFERENCES

[1] Horwitz, S.B. Taxol (paclitaxel): mechanisms of action. *Ann. Oncol*., **1994**, *6*, 3-6.

[2] Anil, K. S.; Alka, G.; Deeppika, A. Paclitaxel and its formulations. *Int. J. Pharm*., **2002**, *235*, 179-192.

[3] Malathi, H.; Frank, L.; Tami, A.; Tan, X.Z.; Sylvia, M.; Daniel, B.M.; James, H.N.; James, P.S.; Lee, M.G. Paclitaxel-resistant cells have a mutation in the paclitaxel-binding region of β-tubulin (Asp26Glu) and less stable microtubules. *Mol. Cancer Ther*., **2006**, *5*, 270-278.

[4] Charles, D.; George E.; Duran, K. A. S. Resistance mechanisms in human sarcoma mutants derived by single-step exposure to paclitaxel (Taxol). *Cancer Res*., **1996**, *56*, 1091-1097.

[5] Eugene, M.; Ainura, K.; Svetlana, Z.; Hum, K. Levels of multidrug resistance (MDR1) P-glycoprotein expression by human breast cancer correlate with *in vitro* resistance to Taxol and doxorubicin. *Clin. Cancer Res*., **1998**, *4*, 389-398.

[6] Aghdass, R.-N.; Dan, L.; Sherry, P.; Richard, A. B. High Raf-1 kinase activity protects human tumor cells against paclitaxel-induced cytotoxicity. *Clin. Cancer Res*., **1998**, *4*, 1111-1116.

[7] Sarabjeet, S.S.; Hicham, F.; Baljit, S. Nanotechnology-based drug delivery systems. *J. Occup. Med. Toxicol*. **2007**, 2:16.

[8] Margaret, A.P.; Martin, L.G.; Nicholas, A.P. Targeted nanodelivery of drugs and diagnostics. *Nano. Today*, **2010**, *5*, 143–159.

[9] Adrian, C.; Kerri, A.M.; Jan, E.S. Overcoming *in vivo* barriers to targeted nanodelivery. *WIREs. Nanomed. Nanobi*. **2011**, *3*, 421–437.

[10] Eric, K. Rowinsky, P. J.; Barke, J. E. K.; Robert, W. T.; David, S. E.; Ross, C. D. Phase I and pharmacodynamic study of Taxol in refractory acute leukemias. *Cancer. Res*., **1989**, *49*, 4640-4647.

[11] Alex, S.; Judith V. A.; Ulrich, M.; Alfred, H. S.; Johan, W. S.; Dirk, K. F. M.; Piet, B.; Willem, J. N.; Jos, H. B.; Olaf, V. T. Limited oral bioavailability and active epithelial excretion of paclitaxel (Taxol) caused by P-glycoprotein in the intestine. *Proc. Natl. Acad. Sci. USA*., **1997**, *94*, 2031-2035.

[12] Mansukhlal, C. W.; Harold, L. T.; Monroe, E. W.; Philip, C.; Andrew, T. M. Plant antitumor agents. VI. Isolation and structure of Taxol, a novel antileukemic and antitumor agent from Taxus brevifolia. *J. Am. Chem. Soc*., **1971**, *93*, 2325-2327.

[13] Peter, B. S.; Jane, F.; Susan, B. H. Promotion of microtubule assembly *in vitro* by Taxol. *Nat*., **1979**, *277*, 665-667.

[14] Green, M.R.; Manikhas, G.M.; Orlov, S.; Afanasyev, B.; Makhson, A.M.; Bhar, P.; Hawkins, M.J. Abraxane, a novel Cremophor-free, albumin-bound particle form of paclitaxel for the treatment of advanced non-small-cell lung cancer. *Ann. Oncol*., 2006, *17*, 1263-1268.

[15] Nicole, W.; David, V.; Nancy, H.; Stephen, H.; Thomas, A. B.; Luka, M.; Kathryn, A. M. 130-nm Albumin-Bound Paclitaxel Enhances Tumor Radiocurability and Therapeutic Gain. *Clin. Cancer Res*., **2007**, *13*, 6.

[16] Nuhad, K. I.; Neil, D.; Sewa, L.; Patrick, S.-S.; Richard, L. T.; Edgardo, R.; Bita, E.; Sigrid, E. R.; Agop, B.; Gabriel, N. H.; Julie, A. E. Phase I and pharmacokinetic study of ABI-007, a Cremophor-free, protein-stabilized, nanoparticle formulation of paclitaxel. *Clin. Cancer Res*., **2002**, *8*, 1038-1044.

[17] Kim, T.-Y.; Kim, D.-W.; Chung, J.-Y.; Sang, G. S.; Kim, S.-C.; Dae, S. H.; Noe, K. K.; Bang, Y.-J. Phase I and Pharmacokinetic Study of Genexol-PM, a Cremophor-Free, Polymeric Micelle-Formulated Paclitaxel, in Patients with Advanced Malignancies. *Clin. Cancer Res*., **2004**, *10*, 3708-3716.

[18] Kim, D.-W.; Kim, S.-Y.; Kim, H.-K.; Kim, S.-W.; Shin, S.W.; Kim, J.S.; Park, K.; Lee, M.Y.; Heo, D.S. Multicenter phase II trial of Genexol-PM, a novel Cremophor-free, polymeric micelle formulation of paclitaxel, with cisplatin in patients with advanced non-small-cell lung cancer. *Ann. Oncol*., **2007**, *18*, 2009-2014.

[19] Sung, C. K.; Dong, W. K.; Yong, H. S.; Joon, S. B.; Hun, S. O.; Sung, W. K.; Min, H. S. *In vivo* evaluation of polymeric micellar paclitaxel formulation: toxicity and efficacy. *J. Control Release*, **2001**, *72*,191-202.

[20] Li, C.; Yu, D.-F.; Robert, A. N.; Fernando, C.; Stephens, L.C.; Nancy, H.; Luka, M.; Sidney, W. Complete regression of well-established tumors using a novel water-soluble poly (L-glutamic acid)-paclitaxel conjugate. *Cancer Res*., **1998**, *58*, 2404-2409.

[21] Boyle, F.; Monk, R.; Davey, R.; Wheeler, H.; Shenlield, G. Prevention of experimental paclitaxel neuropathy with glutamate. *Proc. Am. Assoc. Cancer Res*., **1996**, *37*, 290-297.

[22] Alan, V. B.; E. R. Plummer.; Radha, T.; Julieann, S.; Melanie, G.; Lesley, R.; James, C.; Donald, B.; Alberto, B.; Mark, W. V.; A. H. Calvert. A Phase I and Pharmacokinetic Study of Paclitaxel Poliglumex (XYOTAX), Investigating Both 3-Weekly and 2-Weekly Schedules. *Clin. Cancer Res*., **2005**, *11*, 7834-7840.

[23] Devi, T. S. R.; Gayathri, S. Estimation of Paclitaxel drugs by HPLC method. *Der. Pharm. Chem*., **2010**, *2*, 109-115.

[24] Zhang, J. A.; Gopal, A.; Ma, L.; Sydney, U.; Xuan, T.; Tommaso, S.; Imran, A. Development and characterization of a novel Cremophorw EL free liposome-based paclitaxel (LEP-ETU) formulation. *Eur. J. Pharm. Biopharm.*, **2005**, *59*, 177-187.

[25] Yang, A.; Li, J.; Xu, H.; Chen, H. A study on antitumor effect of liposome encapsulated paclitaxel *in vivo* and *in vitro*. *Bull. Chin. Cancer*, **2006**, *15*, 862-864. (in Chinese)

[26] Caeo, S.-K.; Walter, B.; Otilia, D.; Jan, F.; Jaap, H.; Paul, H.; Anne, K.; Mia, K.; Ben, M.; Arie, N.; Alain, R.; Patrick, R.; Lon, S.-U.; Sculier, J.-P.; Nico, I.; Harry, B. Effect of paclitaxel combined with cisplatin in treatment of non small cell lung cancer. *N. Engl. J. Med*., **1992**, *326*, 524-530.

[27] Chen, Q.; Zhang, Q.Z.; Liu, J.; Li, L.Q.; Zhao, W.H.; Wang, Y.J.; Zhao, Q.H.; Li, L. Multi-center prospective randomized trial on

paclitaxel liposome and traditional taxol in the treatment of breast cancer and nonsmall-cell lung cancer. *Chinese J. Oncol*., **2003**, *25*, 190-192.

[28] Cabanes, A.; Briggs, K.; Gokhale, P.; Treat, J.; Rahman, A. Comparative *in vivo* studies with paclitaxel and liposome encapsulated paclitaxel. *Int. J. Oncol*., **1998**, *12*, 1035-1040.

[29] Straubinger, R.; Sharma, A.; Murray, M.; Mayhew, E. Novel taxol formulations: taxol-containing liposomes. *J. Nati. Cancer Inst. Monogr.,* **1993**, *15*, 69-78.

[30] T. Hamaguchi.; K. Kato.; H. Yasui.; C. Morizane.; M. Ikeda.; H. Ueno.; K. Muro.; Y. Yamada.; T. Okusaka.; K. Shirao.; Y. Shimada.; H. Nakahama.; Y. Matsumura. A phase I and pharmacokinetic study of NK105, a paclitaxel-incorporating micellar nanoparticle formulation. *Brit. J. Cancer*, **2007**, *97*, 170-176.

[31] T. Negishi.; F. Koizumi.; H. Uchino.; J. Kuroda.; T. Kawaguchi.; S. Naito.; Y. Matsumura. NK105, a paclitaxel-incorporating micellar nanoparticle, is a more potent radiosensitising agent compared to free paclitaxel. *Brit. J. Cancer*, **2006**, *95*, 601-606.

[32] Oasmia. Available at: http://www.oasmia.com (Accessed March 27, **2011**).

[33] Elizabeth, H.; Wenbin, D.; Rena, G. L.; Robert, I. G.; Enhanced Efficacy of a Novel Controlled Release Paclitaxel Formulation (PACLIMER Delivery System) for Local-Regional Therapy of Lung Cancer Tumor Nodules in Mice. *Clin. Cancer Res*., **1999**, *5*, 4242-4248.

[34] Deborah, K.; Armstrong.; Gini, F.F.; Maurie, M.; Howard, H.B. A phase I trial of intraperitoneal sustained-release paclitaxel microspheres (Paclimer®) in recurrent ovarian cancer: A Gynecologic Oncology Group study. *Gynecol. Oncol*., **2006**, *103*, 391-396.

[35] Leonard, A. S.; J. Webster Stayman III.; Robert, T. D. Amino acid, glucose, and lactic acid utilization *in vivo* by rat tumors. *Cancer Res*., **1982**, *42*, 4090-4097.

[36] Matthews, O.B.; Nigel, L.W.; Forrest, H.A.; Prabu, D.; Philip, A.W.; S. Hemamalini.; Madhavi, C.C.; Sharyn, D.B.; He, L.F.; Susan, B.H.; Charles, S.S. Tumor targeting by covalent conjugation of a natural fatty acid to paclitaxel. *Clin. Cancer Res*., **2001**, *7*, 3229-3238.

[37] Alex, S.; Antonio, C.W.; Jaap, V.; Yelena, Z.; Desiree, M.V.Z.; Gregory, L.M.; Charles, S.S.; Ross, C.D.; Sharyn, D.B. Disposition of DHA-paclitaxel, a novel taxane, in blood: *in vitro* and clinical pharmacokinetic studies. *Clin. Cancer Res*., **2003**, *9*, 151-159.

[38] Antonio, C.W.; Ross, C.D.; M.K. Carducci.; Michael, A.C.; Julie, R.B.; Yelena, Z.; Matthews, O.B.; Forrest, H.A.; Charles, S.S.; Philip, A.W.; Nigel, L.W.; Sharyn, D.B. Phase I Study of Docosahexaenoic Acid-Paclitaxel: a Taxane-Fatty Acid Conjugate with a Unique Pharmacology and Toxicity Profile. *Clin. Cancer Res*., **2003**, *9*, 3589-3597.

[39] *Pharmacia & Upjohn*. Investigator's brochure FCE 28161, copy no. 001, June **1996**.

[40] Jetske, M.M.T.; Wim, W.; Jan, H.M.S.; Margaret, S.; Ingrid, A.M.M.; Maria, G. Z.; Marurizio, R.; Hilde, R.; Franciska, J. K.; Jos, H.B. Phase I clinical and pharmacokinetic study of PNU166945, a novel water-soluble polymer-conjugated prodrug of paclitaxel. *Anticancer Drugs*, **2001**, *12*, 315-323.

[41] Subramaniam, B.; Saim, S.; Rajewski, R.; Stella, V. J. Methods and apparatus for particle precipitation and coating using near-critical and supercritical antisolvents. U.S. Patent 5,833,891, November 10, 1998.

[42] Subramaniam, B.; Bochniak, D. J.; Rajewski, R. Methods for continuous particle precipitation and harvesting. U.S. Patent 6,113,795, September 5, 2000.

[43] Niu, F.H.; Katherine, F. R.; Roger, A. R.; Charles, D.; Bala, S. *Paclitaxel Nanoparticles: Production Using Compressed CO_2 as Antisolvent: Characterization and Animal Model Studies*.; Polymeric Drug Delivery II.; ACS Symposium Series, **2006**; Vol. *924*, pp. 262-277.

[44] National Institutes of health. National Cancer Institute: clinical trails (PDQ®). Available at: http://www.cancer.gov/clinicaltrials (Accessed on November 10, **2011**).

[45] Duncan, R. Drug-polymer conjugates: potential for improved chemotherapy. *Anticancer Drugs*, **1992**, *3*, 175-210.

[46] Mohammad, R.S.; Roya, M.T.; Abel, M.; Mohammad, A.R. Design and characterization of chitosan nanoparticles as delivery systems for paclitaxel. *Carbohyd. Polym*., **2010**, *82*, 466-471.

[47] Zhan, C.Y.; Gu, B.; Xie, C.; Li, J.; Liu, Y.; Lu, W.Y. Cyclic RGD conjugated poly(ethylene glycol)-co-poly(lactic acid) micelle enhances paclitaxel anti-glioblastoma effect. *J. Control Release*, **2010**, *143*, 136-142.

[48] Adwoa, O.N.; HaiAn, Z.; Luciana, B.L.; Boris, J.-R.; Kurunthachalam, K.; Rachel, R. Oral microemulsions of paclitaxel: In situ and pharmacokinetic studies. *Eur. J. Pharm. Biopharm*., **2009**, *71*, 310–317.

[49] Song, H.; He, R.; Wang, K.; Ruan, J.; Bao, C.C.; Li, N.; Ji, J.J.; Cui, D.X. Anti- HIF-1α antibody-conjugated pluronic triblock copolymers encapsulated with Paclitaxel for tumor targeting therapy. *Biomaterials*, **2010**, *31*, 2302-2312.

[50] Yu, D.-H.; Lu, Q.; Xie, J.; Fang, C.; Chen, H.-Z. Peptide-conjugated biodegradable nanoparticles as a carrier to target paclitaxel to tumor neovasculature. *Biomaterials*, **2010**, *31*, 2278-2292.

Received: April 12, 2011 Revised: November 09, 2011 Accepted: November 15, 2011