Alternative drug formulations of docetaxel: a review

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The anticancer drug docetaxel (Taxotere) is formulated in the nonionic surfactant polysorbate 80 (Tween 80). Early in the clinical development of docetaxel, it became clear that docetaxel administration is associated with the occurrence of unpredictable (acute) hypersensitivity reactions and cumulative fluid retention. These side-effects have been attributed, in part, to the presence of polysorbate 80 and have consequently initiated research focused on the development of a less-toxic, better-tolerated polysorbate 80-free formulation of docetaxel. More recently, there is an increasing interest in developing a (polysorbate 80-free) docetaxel formulation that selectively targets malignant tissue, thereby increasing efficacy while decreasing the occurrence of side-effects related to wide and nonspecific body distribution. This review aims to discuss the preclinical and clinical results of pharmaceutical strategies [PEGylated (immuno)liposomal docetaxel, docetaxel–fibrinogen-coated olive oil droplets, docetaxel encapsulated nanoparticle–aptamer bioconjugates, submicronic dispersion formulation] to develop an alternative, solvent-free, delivery form for docetaxel characterized by increased efficacy and decreased toxicity. *Anti-Cancer Drugs* 18:95–103 © 2007 Lippincott Williams & Wilkins.

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Introduction

The anticancer drug docetaxel (Taxotere; Sanofi-Aventis, Anthony Cedex, France) demonstrates significant antitumour activity against various human malignancies, and is approved for the treatment of patients with locally advanced or metastatic breast cancer, non-small-cell lung cancer, hormone refractory prostate cancer and advanced gastric cancer [1]. Docetaxel (Fig. 1) is a semisynthetic drug prepared by chemical modification of 10-deacetylbacattin III, an inactive precursor compound isolated from the needles of the European yew tree, *Taxus baccata*. Owing to its poor water solubility (3 μg/ml), docetaxel is dissolved in the nonionic surfactant polysorbate 80 (Tween 80), the major component of which is polyoxyethylene-20-sorbitan monooleate (Fig. 2), structurally similar to polyethylene glycols (PEGs). In early phase I clinical trials, docetaxel was supplied as a sterile solution containing 15 mg/ml docetaxel in 50% polysorbate 80 and 50% ethanol [2–6]. To decrease the amount of polysorbate 80 and ethanol administered to patients, this formulation was optimized and a new formulation was used in the phase II and III clinical trials, and subsequently marketed. The currently approved formulation contains 40 mg/ml docetaxel and 1040 mg/ml polysorbate 80 (i.e. 26 mg polysorbate 80/mg docetaxel) and requires further dilution with 13% ethanol before addition to the intravenous infusion solution.

Early in the clinical development of docetaxel, it became clear that docetaxel administration is associated with the occurrence of unpredictable (acute) hypersensitivity reactions, widely ranging in incidence (range 5–60%) and severity (from mild pruritus to systemic anaphylaxis). In addition, fluid retention resulting in weight gain, peripheral oedema and occasionally pleural or pericardial effusions was reported with an incidence increasing above 50% at cumulative docetaxel doses of 400 mg/m^2 or above [7–9]. The occurrence of hypersensitivity reactions has, in part, been attributed to intrinsic toxic effects of polysorbate 80, more specifically to oxidation products and oleic acid present in polysorbate 80, which are known to cause histamine release [10,11]. The role of histamine in the aetiology of polysorbate 80-induced hypersensitivity reactions is further supported by the fact that these side-effects are effectively ameliorated by premedication with corticosteroids and antihistamines [12,13]. Recently, however, the allergenic origin of these hypersensitivity reactions has been opposed and a pathogenetic mechanism, involving the release of vasoactive substances, has been suggested [14]. The occurrence of cumulative fluid retention may, in part, be explained by the fact that the formulation vehicle has been shown to increase membrane permeability [15]. This finding is in line with reports of increased filtration of fluid to the interstitial space following a decrease in plasma colloid osmotic pressure, which occurs upon multiple docetaxel courses [16,17]. Moreover, the fact that corticosteroids significantly reduce and delay the onset of fluid retention, allowing for a higher cumulative dose to be administered [13], may also indicate an effect of vessel

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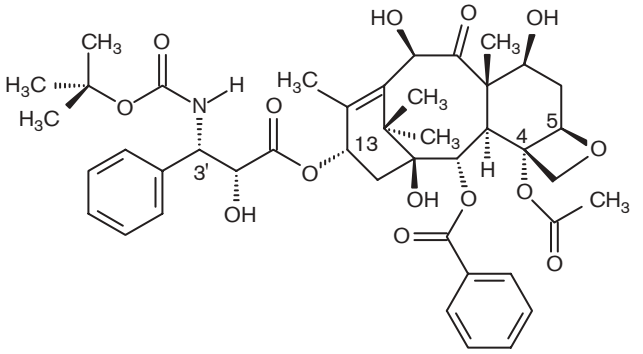
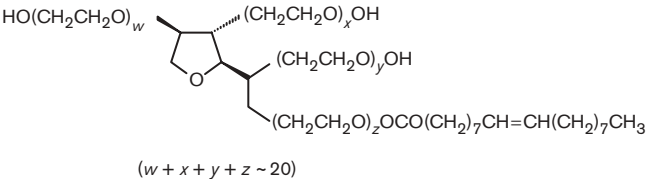
Fig. 1



Chemical structure of docetaxel.

Fig. 2



Chemical structure of the primary constituent of polysorbate 80: polyoxyethylene-20 sorbitan-monooleate.

wall permeability. Although of minor importance, it should be noted that the beneficial effects of prophylactic corticosteroid administration may come at the cost of treatment-related morbidity (e.g. hyperglycaemia, peptic/duodenal ulcers). Finally, polysorbate 80 has been shown to increase plasma viscosity and produce changes in erythrocyte morphology, effects which have been suggested to contribute to mechanisms related to docetaxel-mediated cardiovascular side effects [18].

The drawbacks associated with the presence of polysorbate 80 in the formulation of docetaxel initially initiated research focused on developing a formulation that enhances the solubilization of docetaxel while avoiding the use of polysorbate 80, thus resulting in a less toxic, better-tolerated polysorbate 80-free formulation. More recently, there is increasing interest in developing more advanced (solvent-free) formulations, which provide selective tumour delivery. Examples include targeted (immuno)liposomes and nanoparticles, which profoundly modify both the pharmacokinetics (PK) and pharmacodynamics (PD) of docetaxel, thereby increasing efficacy while decreasing the occurrence of side-effects related to wide and nonspecific body distribution (e.g. neurotoxicity, musculo-skeletal toxicity, neutropenia).

This review aims to discuss the preclinical and clinical results of pharmaceutical strategies to develop an alternative, solvent-free delivery form for docetaxel. Related strategies, including the development of novel, structurally related docetaxel analogues, although also aimed at optimizing the risk–benefit ratio (i.e. balance between treatment-related toxicity and efficacy) for docetaxel treatment, have been discussed previously elsewhere [19].

Prerequisites for a polysorbate 80-free formulation

A polysorbate 80-free docetaxel formulation is only desirable if polysorbate 80 does not substantially contribute to docetaxel's antitumour efficacy. Several reports have suggested that polysorbate 80 possesses intrinsic tumour activity *in vitro* and in animals models [20,21], possibly attributable to oleic acid, rapidly cleaved from polysorbate 80 upon serum carboxylesterase-mediated hydrolysis [22] and known to interfere with malignant cell proliferation [23]. The exact contribution of polysorbate 80 and/or oleic acid to clinically observed antitumour efficacy, however, has not (yet) been clarified. Furthermore, due to its extremely low volume of distribution [24,25], delivery of polysorbate 80 outside the central compartment to the tumour is believed to be insignificant, consequently excluding any substantial contribution to cytotoxicity. In addition, docetaxel is a well known substrate for the transmembrane transporter ABCB1 [P-glycoprotein; multidrug resistance (MDR-1)] [26]. ABCB1, which is expressed in tumours and normal tissues, acts as a drug/xenobiotic efflux pump [27] and its overexpression is implicated in the occurrence of MDR. Polysorbate 80-mediated inhibition of ABCB1 has been observed *in vitro* [28,29], although not confirmed *in vivo* [30]. Moreover, to date, clinical investigations evaluating docetaxel administration in combination with ABCB1 inhibitors (e.g. laniquidar, tariquidar, zosuquidar, elacridar) [19], in the hope of restoring or enhancing chemosensitivity, have been disappointing and have not (yet) yielded an adequate MDR-reversal agent. Furthermore, although ABCB1 plays a (major) role in the faecal disposition of docetaxel [31], its influence on docetaxel plasma PK is minimal to absent [32]. Overall, it is unlikely that a polysorbate 80-free docetaxel formulation will significantly affect docetaxel's cytotoxic properties.

Recent in-vitro and in-vivo data have demonstrated that low doses of docetaxel [33,34] and clinically achievable concentrations of polysorbate 80 [35] both exhibit antiangiogenic properties. Clinically relevant concentrations of polysorbate 80 (i.e. concentrations achieved at the end of docetaxel infusion) nullify docetaxel-mediated inhibition of angiogenesis [35], however, suggesting that a polysorbate 80-free docetaxel formulation may

potentially have a positive effect on the antiangiogenic capacity of docetaxel.

In-vitro studies have demonstrated that docetaxel is extensively bound to albumin and α-1 acid glycoprotein, and that the latter is the main determinant of variability in docetaxel serum binding [36]. Lately, the influence of polysorbate 80 on the PK of total and unbound docetaxel (the latter being the pharmacologically active fraction) has been extensively investigated [37]. At clinically relevant concentrations, polysorbate 80 significantly increased the fraction of unbound docetaxel *in vitro*, a finding that was confirmed *in vivo*. In addition, Baker *et al.* [25] demonstrated that there is a significant relationship between systemic exposure to polysorbate 80 and the clearance rate of unbound docetaxel; higher polysorbate 80 exposure is associated with reduced clearance of unbound docetaxel, consequently resulting in increased unbound drug exposure. Moreover, exposure to unbound docetaxel was more closely related to drug-induced severe haematological toxicity than total docetaxel exposure. The exact mechanistic basis for the decreased protein binding of docetaxel in the presence of polysorbate 80 is unknown, yet is presumably the result of micellar complexes formed by polysorbate 80 with serum proteins (albumin and α-1 acid glycoprotein) [38], thus leading to saturable protein binding of docetaxel [36] and/or the result of displacement of protein-bound docetaxel by rapidly generated polysorbate 80 degradation products (e.g. oleic acid) [39]. Overall, it is clear that the degree of docetaxel plasma binding and consequently the fraction of, and exposure to, unbound drug is, in part, influenced by the formulation vehicle. A polysorbate 80-free formulation, may thus, through a decrease in the fraction of unbound drug, reduce the incidence and severity of unpredictable neutropenia, thereby improving the risk–benefit ratio for docetaxel treatment. One could argue that such a formulation might compromise drug efficacy owing to decreased concentration of the pharmacologically active drug fraction in plasma and, more importantly, at the tumour site. To date, population PK/PD studies have identified systemic (total) docetaxel exposure as a significant predictor of (haematological) toxicity, yet the correlation between any measure of docetaxel exposure (i.e. total clearance, total systemic exposure, peak plasma level, duration of plasma levels greater than a certain value) and antitumour response is, however, much less clear [9]. Furthermore, data on intratumoural docetaxel PK are lacking and an improvement in the risk–benefit ratio, through a decrease in toxicity, may outweigh a decrease, to a yet unknown degree, in antitumour efficacy.

In conclusion, on the basis of the biological and pharmacological properties of polysorbate 80, it seems unlikely that a polysorbate 80-free docetaxel formulation will compromise docetaxel antitumour efficacy.

Alternative formulations – preclinical data

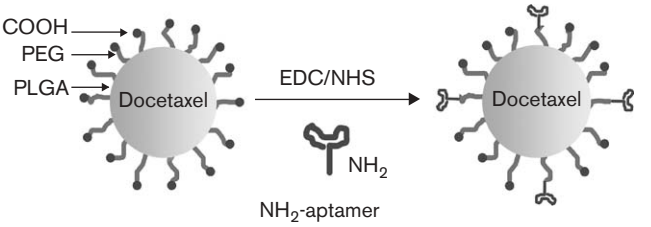
Avoiding the use of polysorbate 80 and at the same time developing a drug formulation that targets malignant tissue has received substantial interest recently, and has led to several alternative, solvent-free docetaxel formulations with varying potential to selectively deliver docetaxel to the tumour, thereby potentially enhancing efficacy and decreasing the occurrence of undesirable side-effects. One approach to avoid polysorbate 80 administration and selectively target the tumour is the use of fibrinogen microspheres as delivery vehicle, as previously investigated for other anticancer drugs [40,41]. Local fibrin(ogen) deposition occurs within the stroma of the majority of solid tumours, and is associated with tumour angiogenesis, growth and metastatic potential [42]. In addition, thrombin-mediated accumulation and retention of intravenously administered fibrinogen-coated olive oil droplets, at fibrin(ogen)-rich sites, has been demonstrated [43]. These features initiated the preparation of murine fibrinogen-coated micronized olive oil droplets loaded with docetaxel [44] and subsequently, evaluation of this formulations's antitumour activity upon intraperitoneal administration to mice bearing a fibrin(ogen)-rich ascites tumour [45]. Upon intraperitoneal treatment with the docetaxel–fibrinogen-coated olive oil droplet formulation (docetaxel dose around 20 mg/kg; mean olive oil droplet size around 12 μm), median survival increased approximately 2-fold compared with treatment with docetaxel solubilized in polysorbate 80. A preliminary toxicity assessment on the basis of the change in weight of healthy, tumour-free mice 15 days following intraperitoneal injection of either isotonic saline, docetaxel solubilized in polysorbate 80 or docetaxel-loaded fibrinogen-coated olive oil droplets demonstrated no significant differences. The association of docetaxel tumour cells was monitored by administering tumour-bearing mice with either docetaxel solubilized in polysorbate 80 or docetaxel-loaded fibrinogen-coated olive oil droplets, both spiked with [^{3}H]docetaxel. Docetaxel association with tumour cells, measured by liquid scintillation counting 48 h after treatment, was at least 10-fold increased upon intraperitoneal administration of docetaxel-loaded olive oil droplets compared with docetaxel solubilized in polysorbate 80. These findings suggest potential to improve the therapeutic efficacy of docetaxel treatment. Several issues, however, require to be further addressed, including the feasibility of intravenous administration which requires smaller droplet size, the influence of anticoagulants or fibrinolytic agents, which may potentially reduce the therapeutic efficacy of the fibrinogen-coated olive oil formulation, and toxicity aspects related to the observed significant antibody response (i.e. droplet-induced production of antifibrinogen antibodies), of which the long-term effects on effectiveness are yet unclear.

Recently, research has increasingly focused on nanotechnological devices for the development of (biomarker)-targeted delivery systems for multiple therapeutic agents [46]. Nanotechnology is a multidisciplinary field, which covers a diverse array of devices derived from engineering, biology, physics and chemistry. These nanotechnology devices (nanotherapeutics) include nanovectors aimed at improving the tumour-targeting efficacy of anticancer drugs [47]. An injectable drug-delivery nanovector is defined as a hollow or solid structure with a diameter in the 1–1000 nm range. It can be filled with anticancer drugs and targeting moieties can be attached to its surface, resulting in specific and differential uptake by the targeted cells, to deliver a constant dose of chemotherapy over an extended period of time. Probably the most well-known, simplest and earliest examples of nanovectors applied in cancer treatment are liposomes, which are a hollow type of nanovector, whereas nanoparticles are considered solid nanovectors. Liposomes are spherical particles (vesicles) consisting of one or more lipid bilayer membranes, which encapsulate an internal space in which notably hydrophilic agents can be entrapped; the lipid bilayer membrane of the liposome may serve as a reservoir for hydrophobic drugs. PEGylated liposomes [STEALTH (sterically-stabilized) liposomes] differ from conventional liposomes by a polymer (PEG) surface coating. These modified liposomes are characterized by reduced uptake by the reticulo-endothelial system, favourable PK (long circulating time, slow clearance rate, small volume of distribution), reduced accumulation in healthy tissues and, most importantly, by increased, preferential tumour uptake owing to their ability to extravasate through the hyperpermeable tumour vasculature, a tumour-targeting mechanism known as enhanced permeation and retention [48,49]. These distinct features make PEGylated liposomes an attractive drug carrier. Indeed, for anticancer drugs, the advantages of PEGylated liposomes are best illustrated by PEGylated liposomal doxorubicin (Caelyx, Doxil, Myocet). The wish to circumvent the use of polysorbate 80 and to improve the therapeutic index for docetaxel-based therapy through specific tumour targeting has led to the successful preparation of PEGylated liposomal docetaxel [50] without compromising cytotoxicity. Indeed, *in-vitro* cytotoxic activity of the PEGylated docetaxel formulation was almost equipotent to the nonliposomal docetaxel formulation. PK profiles for docetaxel solubilized in polysorbate 80 and docetaxel encapsulated in the PEGylated liposomes, assessed after a single intravenous bolus dose to mice, were both best described by a two-compartment model. The PK parameters, however, differed significantly; docetaxel terminal half-life was increased nearly 13-fold upon liposomal encapsulation, and clearance and volume of distribution were decreased more than 100- and 6-fold, respectively, compared with docetaxel solubilized in polysorbate 80. Further increase of the docetaxel concentration inside the PEGylated liposomes (currently 0.7 ± 0.2 mg/ml) is required before clinical trials can be initiated to determine if the improved PK features result in selective and efficient tumour uptake and reduced toxicity. Interestingly, in rats and mice [51,52], the PK of a second dose of PEGylated liposomes (devoid of encapsulated drug) was dramatically altered compared with the first dose in a time-interval-dependent manner. The most prominent difference was a major increase in clearance, hence the observation is referred to as the 'accelerated blood clearance' (ABC) phenomenon. Initially, the ABC effect was suggested to be caused by a considerable increase in hepatic accumulation, possibly involving certain serum factor(s) secreted into the blood after the first dose of PEGylated liposomes. Most recently evaluations have demonstrated that IgM is the major serum protein, which selectively binds to PEGylated liposomes upon repeated injection, and that these IgM-bound PEGylated liposomes can then activate the complement system [53], thus leading to accelerated clearance and enhanced hepatic uptake. Theoretically, the ABC phenomenon can potentially compromise therapeutic efficacy and the strongly increased drug uptake in the liver may cause severe undesirable liver toxicity. Moreover, repeated administration of PEGylated liposomes may lead to the occurrence of unexpected immune reactions. In clinical practice, however, the occurrence of immune reactions after repeated doses of PEGylated liposomal doxorubicin is rare (1–5%), suggesting that the observed ABC phenomenon for PEGylated docetaxel may only have a minor impact. Nevertheless, future research in the design and clinical use of PEGylated liposomal docetaxel, should determine the implications of these findings.

Covalent attachment of targeting ligands, such as monoclonal antibodies specific for antigens expressed on the surface of cancer cells, is another modification of the conventional liposome with the aim to improve selective tumour delivery. Docetaxel has been shown to enhance tumour response upon irradiation [54]; however, clinical application of this radiosensitizing potential is limited owing to side-effects associated with the drug's poor tumour selectivity. To increase tumour delivery and to evaluate the radiosensitizing properties of docetaxel, human colon adenocarcinoma cell lines expressing carcinoembryonic antigen were treated with irradiation and PEGylated docetaxel 'immunoliposomes', i.e. immunoliposomes prepared by coupling monoclonal antibodies against carcinoembryonic antigen to the PEG coating of the lipid membrane [55]. Specifically, cells were incubated (2 h, 37°C) with different concentrations of immunoliposomal docetaxel or liposomal docetaxel (range, 1–1000 nmol/l docetaxel) after which the cells were washed and further incubated (24–48 h, 37°C). Nonincubated cells received a series of test radiation doses ranging from 0 to 8 Gy to determine the degree of radiotoxicity; radiotoxicity was most pronounced at a dose

of 2 Gy. Consequently, this radiation dose was used to irradiate the cells incubated with immunoliposomal and liposomal docetaxel. Cytotoxicity, assessed using the colorimetric 3-(4,5-dimethylthiazol-2yl)-2,5-diphenyl tetrazolium bromide assay, was induced by immunoliposomal docetaxel in a dose- and time-dependent manner. Similar evaluation of the cytotoxic efficacy of the multimodality treatment demonstrated that the effects of immunoliposomal docetaxel were potentiated upon radiation compared with liposomal docetaxel with irradiation or only irradiation ($P < 0.05$). Furthermore, flow cytometric analysis demonstrated that upon treatment with immunoliposomal docetaxel combined with irradiation, apoptosis was significantly increased compared with the multimodality treatment for liposomal docetaxel. Further research should determine if this specific immunoliposomal docetaxel formulation offers potential to improve local radiotherapy in the treatment of colon cancer.

Fig. 3



Schematic representation of the coupling of amine-functionalized A10 PSMA aptamers to a docetaxel-encapsulated, pegylated nanoparticle by carbodiimide chemistry thus resulting in a docetaxel-encapsulated PEGylated-poly(D,L-lactic-*co*-glycolic acid) nanoparticle–aptamer bioconjugate. PEG, polyethylene glycol; COOH, carboxylic acid; PLGA, poly(D,L-lactic-*co*-glycolic acid); NH_2-aptamer, amine-functionalized A10 prostate-specific membrane antigen aptamer; EDC, 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide; NHS, *N*-hydroxysuccinimide.

As mentioned, an expanding number of nanovectors are currently under development for novel, optimized drug-delivery modalities [46,47,56]. Approaches include molecular targeting of nanovectors through conjugation of active recognition moieties to the surface of the nanovector (an approach characterized by potential advantages above conventional antibody-targeted therapy), intracellular targeting of nanoparticles by folate, dendritic polymers as multifunctional nanodevices, silicon and silica materials as materials for injectable nanovectors, metal-(e.g. gold) based nanovectors and polymer-based nanovectors, of which the latter seem to be the most promising for clinical translation. Recently, docetaxel-encapsulated nanoparticles formulated with biocompatible and biodegradable poly(D,L-lactic-*co*-glycolic acid)-block-PEG-copolymer and surface functionalized with A10 2′-fluoropyrimidine aptamers (i.e. RNA oligonucleotides; nucleic acid ligands) [57] that bind to the extracellular domain of the transmembrane prostate-specific membrane antigen, a well-characterized antigen expressed with high specificity on the surface of prostate cancer cells, have been successfully developed *in vitro* (Fig. 3) and their cytotoxicity evaluated using a xenograft nude mouse prostate cancer model [58]. Owing to the surface functionalization with the specific prostate-specific membrane antigen (PSMA) aptamers, these docetaxel-encapsulated nanoparticle–aptamer bioconjugates (Doc-Np-Apt) exert significantly enhanced cellular cytotoxicity *in vitro* resulting from targeted delivery and enhanced cell-specific uptake compared with nontargeted docetaxel-encapsulated nanoparticles (lacking the PSMA aptamer). A single intratumoural injection of Doc-Np-Apt (40 mg/kg) *in vivo* was significantly more efficacious regarding tumour size reduction and survival time compared with an equivalent dose of nontargeted docetaxel-encapsulated nanoparticles. The enhanced efficacy was attributed to delayed clearance from the target site owing to preferential binding to the PSMA proteins, leading to internalization and subsequent intracellular drug release. Mean body weight loss at nadir was significantly decreased (2-fold) for Doc-Np-Apt compared with nontargeted docetaxel-encapsulated nanoparticles, suggesting reduced treatment toxicity. Furthermore, there was no evidence of persistent haematological toxicity. Several aspects of this approach have the potential to facilitate translation into clinical practice, including the fact the poly(D,L-lactic-*co*-glycolic acid) is a component the Food and Drug Administration (FDA) has approved for clinical use, and the fact that the targeting molecules (aptamers) are small, relatively stable, nonimmunogenic and easy to produce on a large scale. However, before clinical application is possible several aspects, including potential sensitization reactions, biological/biophysical barriers impeding targeted delivery and the tailoring of dosing and administration schedules remain to be examined.

Alternative formulations – clinical data

Clinical studies evaluating the efficacy and safety profile of a polysorbate 80-free docetaxel formulation are limited to two phase I dose-finding studies [59,60]. Both trials evaluated a polysorbate 80-free submicronic dispersion formulation (Sanofi-Aventis) administered once every 3 weeks, as a 1-h intravenous infusion without corticosteroid or antihistamine premedication. In patients with advanced solid tumours ($N = 41$), the maximum tolerated dose was 145 mg/m^2 (without granulocyte colony-stimulating factor support), setting the recommended dose level at 130 mg/m^2, which is significantly higher than 100 mg/m^2, currently the highest recommended single-agent dose (http://www.taxotere.com). Despite the lack of premedication, no hypersensitivity reactions were observed and fluid retention occurred in 49% of the patients at a relatively high median cumulative dose (575 mg/m^2), yet was severe in less than 5% of the cases.

Dose-limiting toxicities (DLTs) did not differ from those seen with the current formulation of docetaxel (severe neutropenia and febrile neutropenia) nor did the spectrum of haematological and nonhaematological toxicities. Interestingly, a retrospective analysis concluded that at equimolar doses the solvent-free formulation appeared to result in a lower incidence and severity of infections, mucositis, diarrhoea and neuropathy, the latter finding supporting the notion that polysorbate 80, capable of producing vesicular degeneration [61], contributes in part, to the predominantly mild sensory neuropathy observed in a high proportion of patients treated with docetaxel. The overall response rate among 31 breast cancer patients who had received a dose of 100 mg/m^2 or above was 48.4% [95% confidence interval (CI) 30.2–66.9%)], which is comparable to the results of the pivotal trial leading to the approval of docetaxel for locally advanced or metatstatic breast cancer (overall response rate 47.8%; 95% CI 40.1–55.5%) [62]. A PK evaluation revealed that clearance was independent of dose and similar to previously observed values [9]. The polysorbate 80-free formulation was also administered in combination with doxorubicin as first-line chemotherapy to patients with metastatic breast cancer ($N = 38$). In this study, antitumour activity was observed at all dose levels, and the maximum tolerated dose was reached at 100 mg/m^2 polysorbate 80-free docetaxel and 60 mg/m^2 doxorubicin. Febrile neutropenia was again the DLT. The recommended dose for phase II evaluation was set at 85 mg/m^2 polysorbate 80-free docetaxel and 60 mg/m^2 doxorubicin, which is higher than the doses applied in a randomized phase III study evaluating docetaxel (75 mg/m^2) and doxorubicin (50 mg/m^2) treatment for the same indication [63]. In order to continue the development of this well-tolerated and active formulation, the manufacturing process was modified to accomodate large-scale (i.e. industrial) production. This upscaling process, however, led to some slight modifications of the formulation's physico-chemical properties that may have been the cause of toxicities (oedema, and hand and foot skin reactions) observed at lower doses and in earlier cycles compared with the initial solvent-free formulation. Although these toxicities were not severe enough to be classified as DLTs, they did often lead to treatment withdrawal and were the reason to discontinue further clinical development of this polysorbate 80-free formulation (personal communication, Sanofi-Aventis).

Discussion

Pharmaceutical excipients have an essential role in drug formulations. In contrast to earlier views, however, these excipients, including the nonionic surfactant polysorbate 80 used to solubilize the hydrophobic anticancer drug docetaxel, are not inert vehicles, but are able to affect drug disposition and toxicity patterns [61]. Consequently, acknowledging that a less-toxic (i.e. polysorbate 80-free) formulation of docetaxel is desirable, recent research has focused on developing alternative (solvent-free) drug delivery forms while continuing to maximize the drug's antitumour efficacy through preferential uptake of the drug at the site of action, i.e. the tumour. This pharmaceutical strategy has lead to several alternative drug formulations including fibrinogen-coated olive oil droplets loaded with docetaxel, PEGylated docetaxel liposomes, docetaxel immunoliposomes and docetaxel-encapsulated nanoparticle–aptamer bioconjugates, all currently in different preclinical stages of development and with different advantages and disadvantages (Table 1). Whether the observed advantages (improved PK, selective tumour uptake, increased survival) over the current docetaxel formulation translate to clinical benefits remains to be seen. Potential drawbacks that require to be overcome include the ABC effect observed with PEGylated liposomes, the occurrence of (unexpected) immune reactions, also seen with docetaxel–fibrinogen-coated olive oil droplets and, most importantly, overcoming the pharmaceutical challenge of achieving encapsulation of a therapeutically meaningful amount of drug in the liposomes [64]. It would seem that the use of monoclonal antibodies or the more recently introduced aptamers (DNA or RNA oligonucleotides) as targeting ligand has much potential given the increasing number of well defined biomarkers (e.g. antigens) expressed on the cancer cell surface. Which of these two targeting approaches proves to be more suitable remains to be examined. Initial problems related to specificity, purity, immunogenicity, relatively long development times and batch-to-batch variability upon large-scale biological production of (monoclonal) antibodies, although largely overcome, could still complicate their general application. On the other hand, aptamer synthesis does not rely on biological systems and is an entirely chemical process, which can easily be scaled up. Indeed, aptamer drug-targeting could potentially provide an adequate alternative to antibody-based drug-targeting techniques. Moreover, although the aptamer-based targeting approach is considered highly promising, it is expected that the greatest gain in optimizing therapeutic selectivity will be achieved by synergistic combinations of different nanotechnological targeting strategies.

Currently, only one polysorbate 80-free docetaxel formulation has been evaluated clinically in two phase I trials. Although this formulation demonstrated uncompromised cytotoxic activity, the occurrence of toxicities leading to treatment withdrawal resulted in the discontinuation of further clinical development of this solvent-free docetaxel formulation.

To date, attempts to improve taxane-based treatment have largely focused on, and have been most successful for, paclitaxel [65,66], the first taxane to be introduced (FDA approval 1992). Indeed, most recently paclitaxel

Table 1 A summary of the advantages and disadvantages of the alternative formulations of docetaxel currently in development compared to the current formulation of docetaxel

Formulation	Advantages	Disadvantages
Taxotere (Sanofi-Aventis)	FDA approved (since 1996) Substantial clinical experience Well-established PK/PD relationships	Docetaxel solubilized in polysorbate 80 Polysorbate 80 associated with: hypersensitivity reactions cumulative fluid retention
PEGylated liposomal docetaxel	Potential for selective tumour delivery Tumor targeting via EPR-phenomenon Equipotent *in-vitro* cytotoxicity[a] Improved PK features[a]: ↑ $t_{1/2}$, ↓ CL, ↓ V_d	Preclinical development stage ↑ docetaxel concentration in liposomes required Observed ABC phenomenon may: compromise efficacy lead to adverse (immune) reactions
PEGylated docetaxel immunoliposomes	Potential for selective tumour delivery Tumour targeting via monoclonal antibodies Increased radiosensitizing potential[a] Potential to improve local radiotherapy	*In-vitro* development stage Problems related to use and production of Mabs
Docetaxel–fibrinogen-coated olive oil droplets	Potential for selective tumour delivery Tumour targeting via fibrinogen microspheres ↑ median survival[a] (i.p.) ↑ docetaxel tumour concentration[a] (i.p)	Preclinical development stage i.v. dosing not yet evaluated Antifibrinogen antibodies observed Influence of anticoagulants to be determined
Docetaxel-encapsulated nanoparticle–aptamers	Potential for selective tumour delivery Tumour targeting via aptamers Aptamers: small, nonimmunogenic, stable, large-scale production possible Enhanced cytotoxicity *in vitro*[b], enhanced efficacy *in vivo*[b], reduced toxicity *in vivo*[b]	Preclinical development stage Problems related to use of PEG-copolymer

FDA, US Food and Drug Administration; PK/PD, pharmacokinetic/pharmacodynamic; EPR, enhanced permeation and retention; $t_{1/2}$, terminal half-life; CL, clearance; V_d, volume of distribution; ABC, accelerated blood clearance; i.p., intraperitoneal administration; i.v., intravenous administration; Mabs, monoclonal antibodies; PEG, polyethyleneglycol.
[a]Compared with docetaxel solubilized in polysorbate 80.
[b]Compared with nontargeted docetaxel-encapsulated nanoparticles.

nanoparticles conjugated to albumin molecules (Abraxane, Abraxis Oncology; FDA approval 2005), the latter enhancing the transport of the nanoparticles across the vascular endothelium, demonstrated an improved therapeutic index compared with paclitaxel (Taxol) in the treatment of patients with metastatic breast cancer [67]. Several reasons may explain why the majority of research has focused on improvement of the paclitaxel drug formulation, and include the earlier introduction and thus larger body of clinical experience, and the fact that a greater improvement may be made for current paclitaxel-based treatment with less effort, as the formulation vehicle cremophor EL used to solubilize paclitaxel presents more PK and PD drawbacks [61] than polysorbate 80. Docetaxel, however, is also a highly suitable candidate to concentrate on, given its superiority above paclitaxel in overall survival in patients with metastatic breast cancer [68], linear PK [69], single enzyme-mediated metabolism [70,71] and the existing extensive knowledge on PK/PD relationships [9]. The success of Abraxane suggests that developing merely a solvent-free formulation is not enough therapeutic improvement to warrant extensive clinical evaluation. The combination of a solvent-free docetaxel formulation with tumour-targeting characteristics ultimately shows the most promise of future therapeutic gain for this highly active drug.

In conclusion, preclinical research aimed at optimizing the risk–benefit ratio for docetaxel-based therapy through development of a less toxic, polysorbate 80-free formulation with tumour-targeting properties is ongoing and encouraging; however, it is unlikely that an alternative formulation will be available for clinical use in the near future. Although this may sound disappointing, it is clear that one is convinced of the need to optimize the risk–benefit ratio for docetaxel treatment and any advances that can be made in this area, however modest they may be, are worth further research.

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