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Clinical Oncology Update

Etoposide: Four Decades of Development of a Topoisomerase II Inhibitor

K.R. Hande

Vanderbilt University School of Medicine, Department of Medical Oncology, Nashville VA Medical Center, U.S.A.

Podophyllin-containing materials have been used as folk medicines for centuries. In the 1950s, scientists began a search to identify a more effective podophyllotoxin derivative. These efforts eventually resulted in the development of a new class of antineoplastic agents which target the DNA unwinding enzyme, topoisomerase II. The history of the development of one of the first identified topoisomerase II inhibitors, etoposide, is reviewed in this paper. Critical developments in etoposide's mechanism of action, pharmacology and administration schedule are summarised. The clinical benefits of the recently marketed etoposide prodrug, etoposide phosphate (Etopophos®) are also detailed. The current status of other clinically approved anticancer agents which target topoisomerase II is briefly reviewed. © **1998 Published by Elsevier Science Ltd. All rights reserved.**

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INTRODUCTION

DNA TOPOISOMERASES are nuclear enzymes which make transient DNA strand breaks allowing the cell to manipulate the topology of its DNA [1, 2]. DNA topoisomerases are essential for DNA replication, transcription, chromosomal segregation and DNA recombination. Two major topoisomerase forms are present in all cells: the type I enzyme which makes single-strand cuts in DNA and type II enzymes which cut and pass double-stranded DNA. DNA topoisomerase I was first described in 1971 [3] and DNA topoisomerase II in 1976 [4]. Several commercially available antineoplastic drugs are now known to be inhibitors of topoisomerase I (irinotecan, topotecan) or topoisomerase II (etoposide, teniposide, doxorubicin, daunorubicin, idarubicin, mitoxantrone). This review will concentrate primarily on the development of etoposide as an antineoplastic agent. Etoposide was the first agent recognised as a topoisomerase II inhibiting anticancer drug. Research on etoposide has helped the understanding of mechanisms by which drugs poison topoisomerase II. Recognition of the relationship of topoisomerase II inhibition and a resulting antineoplastic effect has stimulated development of other agents. Currently available topoisomerase II inhibitors are briefly described.

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ETOPOSIDE

Historical development

Podophyllotoxins have been used as medications by various cultures for over 1,000 years (Table 1) [5]. In the 19th century, podophyllin was found to be topically effective for skin cancers. In 1946, the antimitotic properties of podophyllin were established [6]. Clinical evaluation of podophyllotoxin and selected derivatives demonstrated modest antineoplastic activity. However, toxicity of podophyllin was prohibitive [7, 8]. In the 1950s, investigators at Sandoz Pharmaceuticals began synthesising a series of podophyllotoxin derivatives in the hope of identifying agents which retained antineoplastic activity, but had less toxicity. A series of aldehyde condensation products of the non-purified root of the Indian *Podophyllum* plant were found to have antitumour activity against L1210 leukaemia. After extensive isolation procedures, the most effective 'antileukaemic factor' was found to be 4′-demethylepipodophyllin benzylidene glucoside (DEPBG). Whilst previous antineoplastic *Podophyllum* compounds were spindle poisons which produced an increase in mitosis in fibroblast cultures, in tissue cultures treated with DEPBG mitotic figures were practically absent. Two analogues of DEPBG, with increased antineoplastic activity were subsequently synthesised, etoposide (VP-16) in 1966 and teniposide (VM-26) in 1967.

Remarkably, clinical testing of teniposide began within 2 years of drug synthesis (4 years for etoposide). Clinical trials

Table 1. *History of podophyllotoxins in medicine*

Drug	Year	Therapy for
Wild Chervil	900	Cancer
Podophyllum peltatum	1800	Emetic, cathartic
Podophyllin resin	1820	Scrofula, syphilis, cough
Podophyllin	1861	Skin cancer
Mandrake Root Issyk-Kul	1940s	Testicular cancer
Carter's Little Liver Pills	1945	Everything
Podophyllin/Analogues	1950	Lymphoma

Table 2. *Antineoplastic activity of etoposide as a single agent*

Tumour	Response rate (%)	Reference
Testicular	33	[10]
Small cell lung		[11]
Untreated	44	
Previously treated	13	
Non-Hodgkin's lymphoma	29	[12]
Hodgkin's Disease	14	[12]
Non-lymphocytic leukaemia	24	[13]
Ovarian	21	[14]
Gastric	21	[15]
Breast		[16]
Untreated	15	
Previously treated	6	
Non-small cell lung cancer	10	[17]
Kaposi's sarcoma	34	[17]

begun in 1971 and first reported in 1973–1974, demonstrated antineoplastic activity for etoposide and teniposide in AML (acute myeloid leukaemia), Hodgkin's disease, non-Hodgkin's lymphoma, lung cancer (both small cell and non-small cell), gastric cancer, breast cancer and ovarian cancer (Table 2). In 1978, Sandoz licensed development of etoposide and teniposide to Bristol-Myers Squibb. With the demonstrated antineoplastic activity of the drug, FDA approval was granted for etoposide (VePesid) in 1983. It is interesting that at the time of approval, the mechanism of action of this drug and its pharmacology were only beginning to be defined.

Mechanism of action

Studies in the late 1970s clearly demonstrated widespread antitumour activity for etoposide and led to FDA approval for clinical use. However, the mechanism by which etoposide exerted its antineoplastic effects remained unclear. Although etoposide, like podophyllin, can alter microtubule assembly, it does so only at concentrations several fold greater than those achieved *in vivo*. At concentrations achieved *in vivo*, etoposide caused dose-dependent single-strand and double-strand DNA breaks when incubated with cells [18]. When etoposide was removed, DNA breakage was quickly repaired. Incubation of etoposide with purified DNA did not produce DNA strand breaks. However, when etoposide and isolated nuclei were incubated, DNA strand breaks were seen [19]. Thus, something in the nuclei, in addition to DNA, was required to obtain DNA strand breakage.

The relationship between topoisomerase II inhibition and etoposide's antitumour activity was, in part, delayed as information about this important enzyme was being elucidated. It was not until 1979 that the name 'DNA topoisomerases' was introduced [1]. Extensive biochemical analysis of this group of enzymes was being undertaken at the same time etoposide was being brought to the clinic. In 1984, several laboratories demonstrated that mammalian topoisomerase II was the target for etoposide action. Topoisomerase II enzymes are multisubunit proteins, require ATP for overall catalytic activity and modulate DNA topology by passing an intact helix through a transient double-stranded break created in the DNA backbone [1, 2]. As a result of their double-stranded DNA passage reaction, type II topoisomerases are able to regulate over- and under-winding of the double helix and resolve nucleic acid knots and tangles. Etoposide and other topo II inhibitors do not kill cells by blocking topoisomerase catalytic function. Rather they poison these enzymes by increasing the steady-state concentration of their covalent DNA cleavage complexes. This action converts topoisomerases into physiological toxins that introduce high levels of transient protein-associated breaks in the genome of treated cells.

The potential lethality of these drug-induced cleavage complexes rises dramatically when replication machinery or helicases attempt to traverse the covalently bound topoisomerase roadblock in the DNA. This disrupts the cleavage complex and converts transient single- or double-strand breaks into permanent double-stranded fractures which are no longer held together by proteinaceous bridges. These breaks become targets for recombination, sister chromatid exchange, the generation of large insertions and deletions and the production of chromosomal aberrations and translocation. When these permanent DNA breaks are present at sufficient concentration, they trigger a series of events that ultimately culminates in cell death by apoptosis.

Etoposide pharmacology

The molecular weight of etoposide is 588. It is poorly soluble in water. To increase solubility, etoposide is formulated in vials containing 100 mg drug, 400 mg polysorbate 80, 3.25 mg polyethylene glycol 300, 10 mg citric acid and absolute alcohol to 5 ml. Unopened vials are stable for 24 months at room temperature. Etoposide is most stable at pH 5. In basic solutions, it epimerizes to the cis-lactone. Because of its limited water solubility, etoposide has a tendency to precipitate when diluted for intravenous (i.v.) administration. Visual inspection of precipitate in solution is a sensitive measure of stability. When mixed in either 5% dextrose or 0.9% sodium chloride, the duration of etoposide stability is dependent on concentration. (Table 3). Undiluted etoposide has been reported to crack plastic infusion devices, probably due to the polyethylene glycol contained in the solution [23].

Initial pharmacokinetic studies by P. Creaven and L. Allen [24] involved administration of tritiated etoposide intravenously

Table 3. *Etoposide stability in solution* (*from refs* [5])

Concentration (μg/ml)	Duration of stability (h)
0.2	96
0.4	72
0.5	24
0.6	18
0.75	8
1.0	5

to patients. Subsequent thin-layer chromatography of blood and urine samples was performed to distinguish unchanged drug from metabolites. In these important studies, approximately 50% of the total dose of the radiolabel was recovered in the urine, with two-thirds of urinary radioactivity present as unchanged drug. These studies documented several key points about etoposide pharmacology which have subsequently been confirmed in other studies [5, 17, 25]. First, approximately one-third of administered etoposide is excreted in the urine. Etoposide clearance, in part, related to creatinine clearance [26]. Second, little of the drug is excreted into the bile. Third, hepatic metabolism (to glucuronide and demethylmetabolites) accounts for one-third of drug clearance. Creaven and Allen performed similar studies with teniposide and demonstrated a longer drug half-life with this agent and more extensive biliary clearance when compared with etoposide [27].

Pharmacokinetic parameters determined from studies at Vanderbilt University are summarised in Table 4. Studies by other investigators have yielded similar results. The area under the concentration versus time curve (AUC) and peak plasma concentrations achieved following i.v. etoposide administration are linearly related to dose [28]. This linear dose-clearance relationship and the fact that myelosuppression is the primary dose-limiting etoposide toxicity, has made etoposide a popular component of high-dose, marrow-ablative chemotherapy regimens. Etoposide's steady-state volume of distribution ranges from 5 to 17 l/m^2. Etoposide is highly bound to plasma proteins with an average free plasma fraction of 6%. Total etoposide clearance is modestly decreased in patients with renal failure, but not in patients with hepatic obstruction [29]. The etoposide plasma binding ratio (the amount of bound drug/the amount of free drug) is directly related to the serum albumin concentration. Cancer patients, in particular those with hepatic involvement, often have reduced serum albumin concentrations. Since free drug is biologically active, conditions which decrease protein binding increase the pharmacological effect of a given dose. Patients with low serum albumin concentration have greater toxicity from a given drug dose than patients with normal serum albumin concentrations. Several studies [30, 31] have correlated plasma etoposide concentrations (or AUC) with toxicity. Measurement of free versus total drug provides better correlations. Correlations of drug concentration and tumour response have been suggested in some, but not all studies [32–34].

Schedule dependence of etoposide and oral etoposide

Several preclinical findings suggested that the duration of exposure of neoplastic cells to etoposide is important in producing maximal antitumour activity [35, 36]. Etoposide's target, topoisomerase II, is significantly expressed only in dividing cells during selected mitotic phases of the cell cycle [37]. Chronic scheduling may, therefore, be advantageous because it maximises the likelihood of exposing malignant cells to etoposide during sensitive periods of the cell cycle. Cytotoxicity of topoisomerase-II-targeting drugs relates not only to the magnitude of formation of drug-induced, enzyme-mediated DNA strand breaks, but also to the intracellular half-life of these lesions [38]. Therefore, antineoplastic agents or protracted scheduling schemes that prolong the presence of DNA strand breaks in the cell would be expected to result in superior efficacy.

Table 4. *Etoposide pharmacokinetic parameters (Vanderbilt data)*

Volume of distribution	13.8 ± 7.4 l/m^2
Clearance (plasma)	26.5 ± 9.6 ml/min/m^2
Renal clearance	9.3 ± 3.9 ml/min/m^2
Half-life	6.4 ± 2.4 h

Although the importance of drug administration scheduling on etoposide's antineoplastic activity was suggested in early preclinical and clinical trials, the most informative study on the importance of schedule dependence included patients with small cell lung cancer (SCLC) who received 500 mg/m^2 etoposide either as a 24-h i.v. infusion or as a daily 2-h infusion for 5 days [39]. Though both patient groups received the same total drug dose, differences in response rates were dramatic. In the 1 day treatment arm, 10% of patients responded to therapy, compared with an 89% response rate in the 5 day treatment arm. Pharmacokinetic data from this trial revealed no significant difference in total AUC measurements between these two treatment arms. However, prolonged maintenance of low serum etoposide concentrations (≥ 1 μg/ml) was associated with superior efficacy in the 5 day treatment arm. In a subsequent study in SCLC [40], five and eight day schedules were found to have equivalent antineoplastic activity. Haematological toxicity was greater in the 5 day arm. These two studies suggested that prolonged exposure to low concentrations of etoposide may improve etoposide's therapeutic ratio.

In 1987, the US Food and Drug Administration approved a soft gelatin capsule formulation of etoposide for clinical use. This allowed long-term drug administration. Etoposide capsules contain 50 mg etoposide in a solution of purified water, citric acid, glycerin and polyethylene glycol 400. The use of oral etoposide provides a convenient, tolerable treatment regimen which avoids the need for hospitalisation. There are, however, drawbacks to the use of oral etoposide. Once absorbed, there is no pharmacological difference between oral and i.v. etoposide with respect to mechanism of action drug, half-life, mode of drug elimination or type of toxicity. Bioavailability of oral etoposide ranges from 40 to 75% and varies with drug dose. Oral absorption is linear to doses up to 250 mg but decreases with doses greater than 300 mg [41]. Table 5 summarises a series of studies from Vanderbilt and compares the variability in apparent drug clearance as a function of the route of drug administration. Between-patient variability is significantly greater than within-patient variability. However, administration of drug by the oral route increases variability both within and between patients as compared to intravenous administration. Joel and colleagues [42] have tried to improve the bioavailability and variability of etoposide by concomitant administration of ethanol, bile salts, cimetidine, metaclopromide and propantheline without

Table 5. *Variation in etoposide clearance with different routes of administration*

Group	Administration	Coefficient of variation % (*n*)
Within same patient	i.v.	11% (12)
Within same patient	p.o.	16% (11)
Between patients	i.v.	29% (48)
Between patients	p.o.	38% (16)

i.v., intravenous; p.o., oral.

success. Preclinical studies [43] suggest that inhibition of P-glycoprotein may improve etoposide bioavailability.

To determine whether administration schedules longer than a standard 3–5 day treatment might further improve the therapeutic index of etoposide, prolonged oral dose-regimens have been developed [44]. Several phase II studies using either 50 mg or 50 mg/m^2 etoposide once or twice daily for 14–21 days have been undertaken involving patients with both previously treated and untreated lung cancer, lymphoma, previously treated germ cell tumours, soft tissue sarcomas, ovarian cancer, breast cancer, melanoma and renal cell carcinoma [45]. Responses have been seen in all tumour types, although uncommon in melanoma, sarcoma and renal cell carcinomas. Response rates are equal to or greater than those expected from historical data from similar patient populations. Several patients with SCLC, lymphomas and germ cell tumours who have responded to the chronic schedule were previously clinically resistant to standard doses and schedules.

The activity of oral etoposide in these phase II trials suggested that prolonged oral etoposide administration might be a more effective method of drug administration. Unfortunately, two randomised clinical trials have now been conducted which have failed to demonstrate any benefit to prolonged oral etoposide therapy, at least in the initial treatment of SCLC. Both the Cancer and Leukemia Group B (CALGB) [46] and British Medical Research Council [47] trials, which compared long-duration oral etoposide with more standard chemotherapy, failed to show any improvement in response rates or median survival with oral etoposide administration when compared with more standard treatment regimens.

Etoposide phosphate

As previously mentioned, etoposide is poorly soluble in water. Large doses of etoposide may, therefore, occasionally require administration of significant fluid volumes. This fluid load can cause heart failure in some patients and presents an obstacle to rapid administration of etoposide and to long-term home infusional administration regimens. Hypersensitivity reactions and hypotension may also occur with rapid administration of etoposide, perhaps due to the vehicles needed as solubilisers. To overcome solubility problems, attempts have been made to modify the molecule. Etoposide phosphate (Etopophos®, Bristol-Myers Squibb Co., Princeton, New Jersey, U.S.A.) (Figure 1) is an etoposide prodrug approved for intravenous use by the US Food and Drug Administration in 1996. It is soluble in water at concentrations up to 20 mg/ml. Several studies [48–51] have shown that etoposide phosphate:

(1) is rapidly (within 15 min) and completely converted to etoposide by the action of alkaline phosphatases in blood;
(2) can be safely administered over short (5–30 min) time periods;
(3) is pharmacokinetically equivalent to etoposide; and
(4) has toxicities identical to those seen with etoposide.

Conversion to etoposide is rapid and not saturated even at high drug doses (1.6 g/m^2) used in marrow transplantation regimens [52]. Etoposide phosphate appears to have equivalent antineoplastic activity to etoposide. Similar response rates have been seen in SCLC patients given etoposide/cisplatin versus etoposide phosphate/cisplatin [53].

The pharmacological properties of etoposide phosphate clearly make it more convenient than etoposide to administer. It can be given more rapidly and may, although not proven, reduce hypersensitivity reactions. However, since it is an etoposide prodrug, it should not be any more effective than etoposide. Recommendations for use of etoposide phosphate over etoposide depend, to a large extent, on cost savings. Since etoposide is now off patent, its costs have decreased. Table 6 lists the average wholesale price of etoposide and etoposide phosphate as well as the actual pharmacy costs at Vanderbilt University. At Vanderbilt, etoposide phosphate costs five times as much as etoposide. However, the convenience of drug administration can result in reduced administration time and decreased need for i.v. fluid. Depending on costs of nurses' time and overhead costs, etoposide phosphate may or may not be a more cost-effective treatment. Doyle and colleagues [54] have estimated the total

Table 6. Etoposide and etoposide phosphate pharmacy cost at Vanderbilt University

	Average wholesale price	Vanderbilt cost	Vanderbilt cost per 100 mg
VePesid® (1 gm)	$1245.00	$181.00	$18.10
VePesid® (500 mg)	$639.00	$90.50	$18.10
Etopophos® (100 mg)	$120.00	$97.16	$97.16

(£1 = $1.62.)

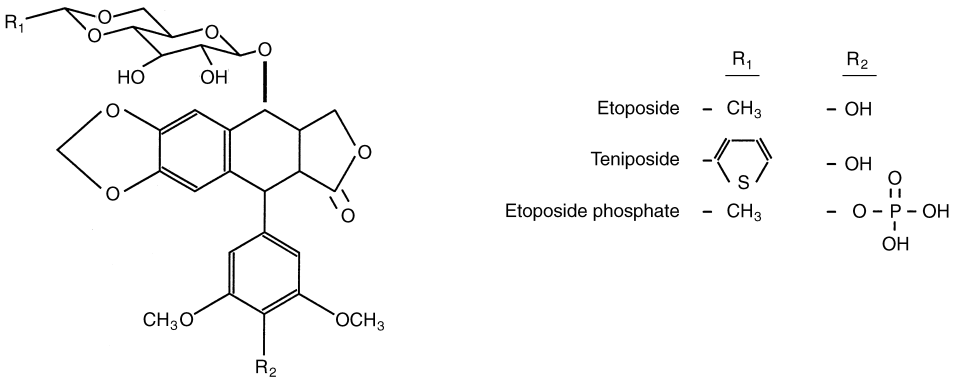

Figure 1. Chemical structure of etoposide, teniposide and etoposide phosphate.

costs of treating patients with SCLC with etoposide phosphate to be roughly equivalent to treatment with etoposide.

A potential advantage of etoposide phosphate may be a more predictable oral bioavailability. Although an oral formulation of etoposide phosphate is not yet commercially available, oral etoposide phosphate has been evaluated in phase I trials in Europe [55, 56]. Etoposide phosphate is rapidly absorbed and converted to etoposide. Bioavailability averages 68% at doses up to 150 mg/m^2/day. Interestingly, the variability in oral etoposide phosphate absorption (26% coefficient of variation) appears to be somewhat less than the variability in oral etoposide bioavailability when the data of Chabot and colleagues [56] are compared with data from Vanderbilt (Table 7). If these results are confirmed, the pharmacokinetic properties of oral etoposide phosphate may give it a significant advantage over etoposide.

OTHER TOPOISOMERASE II TARGETING ANTINEOPLASTIC AGENTS

In addition to etoposide, five other topoisomerase II targeting antineoplastic agents (teniposide, doxorubicin, daunorubicin, idarubicin and mitoxantrone) have been approved for clinical use in the US by the Food and Drug Administration (Table 8). Other topoisomerase II inhibitors (Table 9) which have demonstrated antitumour activity in animal or clinical studies have not been approved for general clinical use in the US.

Table 7. *Bioavailability of oral etoposide and etoposide phosphate*

	Bioavailability (%)	Coefficient of variation (%)	Reference
i.v. Etoposide	100	28	[45]
100 mg oral etoposide	72	30	[41]
400 mg oral etoposide	48	37	[41]
125 mg/m^2 oral etoposide phosphate	68	26	[56]

Table 8. *Clinical use of topoisomerase III inhibiting anticancer agents*

Drug	Trade name	Major indications
Doxorubicin	Adriamycin®	Lymphomas*
	Rubex®	Breast cancer*
	Doxil®	Sarcomas*
		Kaposi's sarcoma
		Leukaemias
Daunorubicin	Cerubidine®	Acute myelogenous leukaemia*
		Acute lymphocytic leukaemia
Idarubicin	Idamycin®	Acute myelogenous leukaemia
Mitoxantrone	Novantrone®	Acute leukaemia
		Breast cancer
		Lymphomas
Etoposide	VePesid®	Testicular cancer*
	Etopophos®	Small cell lung cancer*
		Lymphomas
		Ewing's sarcoma
		Kaposi's sarcoma
		Ovarian cancer
Teniposide	Vumon®	Poor prognosis acute lymphoblastic leukaemia*

*Drug of first choice for treatment of these diseases.

Teniposide

Teniposide is an analogue of etoposide. It was initially isolated and evaluated in patients before etoposide. However, early concerns about hypersensitivity reactions with teniposide administration and subsequent use of teniposide at inappropriately low doses led to a slower development of this drug when compared with etoposide. Teniposide was approved for clinical use in the US in 1993, some 30 years after its initial synthesis and 10 years after etoposide approval. It is used primarily in the treatment of leukaemias and lymphomas (primarily childhood). However, in the face of similar preclinical findings and dose-limiting toxicities (DLTs) when compared to etoposide, teniposide may be equivalent to etoposide. Few studies have been completed comparing the activity of these two agents.

In vitro, teniposide is 10-fold more active than etoposide in killing cancer cells. Since both teniposide and etoposide have relatively similar abilities to inhibit topisomerase II, the greater *in vitro* cytotoxic potency is believed to be related to better cellular uptake of teniposide [9]. Equitoxic teniposide doses are roughly one-third less than those of etoposide in patients. Teniposide toxicities are identical to those of etoposide: myelosuppression, hair loss, nausea and vomiting (mild) and mucositis. Teniposide is even less water soluble than etoposide and allergic reactions are seen more frequently with teniposide than etoposide [57]. Development of acute non-lymphocytic leukaemia with 11q23 chromosomal abnormalities, similar to cases seen with etoposide, have developed following teniposide use [58].

Anthracyclines

Anthracycline antibiotics, originally isolated from fermentation products of *Streptomyces peucetus*, were found to have antineoplastic activity over 4 decades ago, long before the identification of topoisomerase enzymes. It was not until 1984 that the anthracyclines were recognised as inhibitors of topoisomerase II [59]. Their ability to inhibit topoisomerase II appears to be the primary mechanism for tumour cytotoxicity. However, the anthracyclines produce a wide-range of biological reactions in addition to topoisomerase inhibition. First, the planar aglycone moiety of the anthracycline molecule can insert between base pairs of DNA (intercalation). This intercalation was originally felt to be the mechanism for anthracycline cytotoxicity. Secondly, the anthracyclines modify the ability of nuclear helicases to dissociate duplex DNA into single DNA strands thus hindering the process of strand separation [60]. Thirdly, all clinically active anthracyclines are anthraquinones. All quinones can undergo one

Table 9. *Investigational agents which inhibit topoisomerase II*

Amonifide	Genistein
Amsacrine	Intoplicine
Anthrapyrazoles	Makaluvamines
Aza IQD	Merbarone
Azatoxin	Menogaril
Bis (2,6-dioxopiperazine)	Naphthoquinones
Bulgarein	Nitroimidazole
Distamycin	Saintopine
Ditercalinium	Streptonigrin
Elliptinium	Suramin
Epirubicin	Whitangulatin
Ethidium bromide	

and two electron reductions producing reactive compounds which damage macromolecules and lipid membranes [61]. It is now known that reduced metals (such as iron) are critical components in the formation of these reactive intermediates [62]. Free-radical formation induced by the anthracyclines appears to be the primary mechanism by which this class of drugs causes damage to heart tissues.

Doxorubicin (adriamycin®) is the most commonly used anthracycline. It has antineoplastic activity when used as therapy for a wide variety of cancers. It is the drug primarily used in the treatment of patients with lymphomas, breast cancer and sarcomas. Doxorubicin has recently been formulated in liposomes with surface-bound segments of polyethylene glycol (pegylated liposomes). This slow-release form of doxorubicin can deliver high concentrations of doxorubicin to Kaposi's sarcoma lesions [63]. Daunomycin is another anthracycline, used less commonly than doxorubicin. It is the drug of choice for initial therapy of acute myelogenous leukaemia. Idarubicin is a daunorubicin analogue used only in the treatment of acute myelogenous leukaemia. Several studies have been performed to determine whether idarubicin is a better drug than daunorubicin for acute myelogenous leukaemia. Whilst there is some suggestion that idarubicin may be slightly better than daunorubicin [64, 65], these differences are small and probably not clinically significant. The cheaper drug should probably be used. There is no significant difference in toxicity. Idarubicin is better absorbed than doxorubicin and daunorubicin and this fact may allow for development of an orally administered anthracycline [66]. However, there is significant patient-to-patient variability in oral idarubicin absorption which may limit the clinical use of this method of drug administration. The oral form of idarubicin remains investigational in the US.

Cardiac damage is the toxicity unique to doxorubicin and other anthracyclines. It appears to result from formation of free-radical intermediates which kill myocardial cells lacking free-radical scavenging defence mechanisms. Cardiac toxicity is unrelated to topoisomerase II inhibition. The cardiac damage caused by anthracyclines is cumulative and not usually noted until several doses of drug have been given. At total administered doses of doxorubicin less than 500 mg/m^2 (several months of therapy), heart failure is seen in fewer than 7% of cases. Heart failure may develop many months after discontinuation of doxorubicin [68]. The risk of toxicity is related to the peak drug concentration achieved in blood following administration. Administration schedules giving less drug at any one time, such as long-term infusions, reduce the risk of heart failure. Dexrazoxane (Zinecard®) is an iron chelating agent which can protect the heart from anthracycline toxicity, by decreasing the formation of iron-catalysed free-radical formation [69].

Mitoxantrone

In searching for anthracycline analogues with lower toxicity than doxorubicin, a number of multiringed planar substances were synthesised and tested for antineoplastic activity. Of a series of anthracenediones, mitoxantrone was the most active and is the only compound of this class approved for clinical use [70]. It also inhibits topoisomerase II. Mitoxantrone has a significantly reduced potential to form free-radical intermediates when compared with anthracyclines and appears to have less cardiotoxicity. It is used for treatment of breast and prostate cancer, leukaemia (myelocytic) and lymphoma. Because of decreased toxicity compared with doxorubicin, mitoxantrone has been incorporated into selected chemotherapy regimens in place of doxorubicin for treatment of breast cancer and lymphoma, particularly in patients with a poor performance status who are believed to be at significant risk for doxorubicin toxicity [71]. In selected head-to-head comparison studies, response rates and survival rates appear to be similar when mitoxantrone is substituted for doxorubicin [72]. However, in other series, mitoxantrone has not been found to be as effective as doxorubicin [73]. Overall, it appears that mitoxantrone has less toxicity, but less antitumour activity when compared with doxorubicin.

The primary toxicities of mitoxantrone are similar to those seen with the anthracyclines: myelosuppression, nausea, vomiting and cardiac toxicity. At doses which produce equivalent drops in the white blood cell and platelet counts (75 mg/m^2 doxorubicin versus 15 mg/m^2 mitoxantrone), nausea and vomiting and hair loss are less with mitoxantrone when compared with doxorubicin [74]. With commonly used dosages, approximately twice as much mitoxantrone can be given before heart failure develops when compared to doxorubicin [71].

CONCLUSION

Four decades of research have led to the recognition that certain antineoplastic drugs poison topoisomerase II, thereby killing cancer cells. These studies have provided a new target site for drug development. Etoposide, the lead drug in this class, continues to be a widely used agent with activity against a wide range of cancers. It is a standard component of therapy for small cell lung cancer, testicular cancers and lymphomas. Optimal and convenient schedules have been devised over the past two decades. The pharmacology of oral and i.v. etoposide preparations has been determined. Etoposide phosphate may avoid some of the toxicities of etoposide and, although still under investigation, may reduce variability in oral drug absorption when compared with etoposide. Other topoisomerase inhibitors also play a prominent role in cancer chemotherapy.

Whilst currently approved topoisomerase II-active drug (anthracyclines, podophyllotoxins and mitoxantrone) constitute an important class of clinically active drugs, problems, other than toxicity, exist with their use [75, 76]. First, slow growing tumours with reduced topoisomerase II concentrations are more resistant to this class of drugs. Second, most topoisomerase II inhibiting agents are substrates for PgP (the transporter associated with the multidrug resistant phenotype) or are substrates for multidrug resistance associate protein. Finally, mutations or alterations in drug binding sites on topoisomerase II have been associated with development of resistant tumour cells. Based on current knowledge of the biochemistry, regulation and function of topoisomerase II, development of new inhibitors or mechanisms for preventing drug resistance may be forthcoming.

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