MITOXANTRONE

1. Exposure Data

1.1 Chemical and physical data

1.1.1 *Nomenclature*

Mitoxantrone

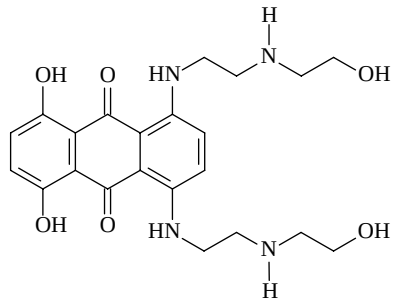
Chem. Abstr. Serv. Reg. No.: 65271-80-9
Chem. Abstr. Name: 1,4-Dihydroxy-5,8-bis[{2-[(2-hydroxyethyl)amino]ethyl}-amino]- 9,10-anthracenedione
IUPAC Systematic Name: 1,4-Dihydroxy-5,8-bis-[{2-[(2-hydroxyethyl)amino]-ethyl}-amino]anthraquinone
Synonyms: DHAQ; dihydroxyanthraquinone; mitoxanthrone; mitozantrone

Mitoxantrone dihydrochloride

Chem. Abstr. Serv. Reg. No.: 70476-82-3
Chem. Abstr. Name: 1,4-Dihydroxy-5,8-bis[{2-[(2-hydroxyethyl)amino]ethyl}-amino]- 9,10-anthracenedione, dihydrochloride
IUPAC Systematic Name: 1,4-Dihydroxy-5,8-bis-[{2-[(2-hydroxyethyl)amino]-ethyl}-amino]anthraquinone dihydrochloride
Synonyms: CL 232315; DHAD; mitoxanthrone dihydrochloride; mitoxantrone hydrochloride

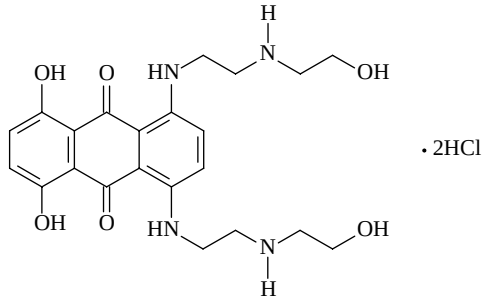
1.1.2 *Structural and molecular formulae and relative molecular mass*

Mitoxantrone



$C_{22}H_{28}N_4O_6$ Relative molecular mass: 444.49

Mitoxantrone dihydrochloride



$C_{22}H_{28}N_4O_6.2HCl$ Relative molecular mass: 517.41

1.1.3 *Chemical and physical properties of the pure substances*

Mitoxantrone

(*a*) *Description*: Crystalline solid (Budavari, 1996)
(*b*) *Melting-point*: 160–162 °C (Budavari, 1996)
(*c*) *Solubility*: Sparingly soluble in water; slightly soluble in methanol; practically insoluble in acetone, acetonitrile and chloroform (Budavari, 1996)

Mitoxantrone dihydrochloride

(*a*) *Description*: Hygroscopic blue–black solid (Budavari, 1996)
(*b*) *Melting-point*: 203–205 °C (Budavari, 1996)
(*c*) *Spectroscopy data*: Ultraviolet, infrared, nuclear magnetic resonance (proton and ^{13}C) and mass spectral data have been reported (Beijnen *et al*., 1988)
(*d*) *Solubility*: Sparingly soluble in water; slightly soluble in methanol; practically insoluble in acetone, acetonitrile and chloroform (Budavari, 1996)
(*e*) *Dissociation constants*: pK_a, 5.99, 8.13 (Gennaro, 1995)

1.1.4 *Technical products and impurities*

Mitoxantrone hydrochloride is the common name for the dihydrochloride salt. It is available mainly as 5-, 10-, 12.5- and 15-mL solutions for intravenous infusion containing 2.33 mg/mL mitoxantrone hydrochloride, equivalent to 2 mg/mL of mitoxantrone. The injection solution may also contain acetic acid, sodium acetate, sodium chloride, sodium metabisulfite and sodium sulfate as excipients (Gennaro, 1995; Canadian Pharmaceutical Association, 1997; British Medical Association/Royal Pharmaceutical Society of Great Britain, 1998; Editions du Vidal, 1998; LINFO Läkemedelsinformation AB, 1998; Rote Liste Sekretariat, 1998; Thomas, 1998; US Pharmacopeial Convention, 1998a).

The following impurities in mitoxantrone hydrochloride are limited by the requirements of *The European Pharmacopoeia*: 1-amino-5,8-dihydroxy-4-[{2-[(2-hydroxyethyl)amino]ethyl}amino]anthracene-9,10-dione; 5-hydroxy-1,4-bis[{2-[(2-hydroxy-

ethyl)amino]ethyl}amino]anthracene-9,10-dione; 2-chloro-1,4-dihydroxy-5,8-bis[{2-[(2-hydroxyethyl)amino]ethyl}amino]anthracene-9,10-dione; and 8,11-dihydroxy-4-(2-hydroxyethyl)-6-[{2-[(2-hydroxyethyl)amino]ethyl}amino]-1,2,3,4-tetrahydronaphtho-[2,3-*f*]quinoxaline-7,12-dione (Council of Europe, 1998).

Trade names for mitoxantrone hydrochloride include Mitoxantrona Filaxis, Mitoxantrona Raffo, Mitoxantron AWD, Mitoxantrone, Novanthrone, Novantron, Novantrone and Pralifan (Swiss Pharmaceutical Society, 1999).

1.1.5 *Analysis*

Several international pharmacopoeias specify infrared absorption spectrophotometry with comparison to standards as the method for identifying mitoxantrone hydrochloride; liquid chromatography is used to assay its purity. In pharmaceutical preparations, mitoxantrone hydrochloride is identified by ultraviolet absorption spectrophotometry, and liquid chromatography is used to assay for its content (US Pharmacopeial Convention, 1994; Council of Europe, 1998; US Pharmacopeial Convention, 1998b).

High-performance liquid chromatography is the most useful analytical tool for analysing mitoxantrone and its metabolites in biological matrices. Ion-pair chromatography and radioimmunoassay have also been used (Beijnen *et al*., 1988).

1.2 Production

The synthesis of mitoxantrone hydrochloride involves reacting leuco-1,4,5,8-tetrahydroxyanthraquinone with 2-[(2-aminoethyl)amino]ethanol to form 1,4-dihydroxy-6,7-dihydro-5,8-bis[{2-[(2-hydroxyethyl)amino]ethyl}amino]-9,10-anthracenedione. This product is aromatized with chloranil as the oxidant, and it is converted into mitoxantrone hydrochloride by treatment with hydrogen chloride in ethanol (Beijnen *et al*., 1988).

Information available in 1999 indicated that mitoxantrone and mitoxantrone hydrochloride were manufactured and/or formulated in seven and 31 countries, respectively (CIS Information Services, 1998; Royal Pharmaceutical Society of Great Britain; 1999; Swiss Pharmaceutical Society, 1999).

1.3 Use

Mitoxantrone is a cytotoxic drug used in the treatment of malignant disease in humans and animals. It is an inhibitor of DNA topoisomerase II. In the 1970s, analogues of the anthracenedione dyes (originally developed for use in the textile industry) were investigated as possible cytotoxic agents on the basis of the ability of the parent compounds to intercalate DNA. Mitoxantrone, a dihydroxyanthracenedione derivative, was the most active of a series of compounds synthesized (Zee-Cheng & Cheng, 1978; Dunn & Goa, 1996).

Mitoxantrone entered clinical trials in 1980 and has been used in cancer treatment since the mid-1980s. It was found to have anti-tumour activity in advanced breast cancer (often in patients in whom other treatments have failed), non-Hodgkin lymphoma and certain leukaemias. It is still most commonly used in these tumours, typically in combination with other cytotoxic drugs, and has also been used in the treatment of other cancers such as ovarian, prostate and lung cancer (Faulds *et al*., 1991). The typical dose is the equivalent of 12–14 mg/m^2 mitoxantrone once every three weeks in patients with lymphomas and tumours of solid tissues, and 12 mg/m^2 per day for five days in patients with leukaemia. When mitoxantrone is used in combination with other cytotoxic drugs, these doses are often lower (Dunn & Goa, 1996; Royal Pharmaceutical Society of Great Britain, 1999). In recent years, mitoxantrone has been used to a limited extent in the treatment of multiple sclerosis, typically at doses lower than those used in malignant disease and on a monthly schedule (Gonsettte, 1996; Millefiorini *et al*., 1997).

1.4 Occurrence

Mitoxantrone is not known to occur as a natural product. No data on occupational exposure were available to the Working Group.

1.5 Regulations and guidelines

Mitoxantrone hydrochloride is included in the European and US pharmacopoeias (Council of Europe, 1998; Swiss Pharmaceutical Society, 1999).

2. Studies of Cancer in Humans

The Working Group considered only studies in which mitoxantrone was given to patients who did not receive treatments with alkylating agents, with the exception of low doses of cyclophosphamide.

2.1 Case reports

Detourmignies *et al*. (1992) described two cases of acute promyelocytic leukaemia in France in patients who had previously received mitoxantrone. A woman, 51 years old, with a primary breast tumour had received a combination of mitoxantrone, vincristine, 5-fluorouracil, cyclophosphamide and radiotherapy (chest and axillary); she developed acute promyelocytic leukaemia nine months later. Another woman, 42 years old, with a primary breast tumour had received a combination of mitoxantrone, vincristine, 5-fluorouracil and radiotherapy (chest) and developed acute promyelocytic leukaemia after

17 months. [The Working Group noted that there was no information on dose, treatment schedule or duration of mitoxantrone treatment.]

Philpott *et al*. (1993) reported two cases of acute myeloid leukaemia following treatment of advanced breast cancer in the United Kingdom. The first case was that of a woman (aged 56 years) who received eight cycles of mitoxantrone (7 mg/m^2), methotrexate and mitomycin, local radiotherapy to the breast and axilla and tamoxifen. She was disease-free for 18 months but then developed acute myeloid leukaemia. The second patient (aged 39 years) was also treated with eight cycles of mitoxantrone (7 mg/m^2), methotrexate and mitomycin and in addition received radiotherapy to the breast. Thirteen months later she developed acute myeloid leukaemia.

Melillo *et al*. (1997) described three cases of acute myeloid leukaemia in 1996 in Italy in women who were treated with five to seven intravenous courses of methotrexate (30 mg/m^2 every three weeks), mitoxantrone (8 mg/m^2 every three weeks) and mitomycin (8 mg/m^2 every six weeks) for recurrent breast cancer at the age of 44, 52 and 60. They had previously received radical mastectomy and either cyclophosphamide, methotrexate and 5-fluorouracil or radiotherapy or both. Treatment with methotrexate, mitoxantrone and mitomycin was followed by tamoxifen, medroxyprogesterone acetate or medroxyprogesterone acetate and radiation therapy. Acute myeloid leukaemia (one case of acute monoblastic leukaemia, one of acute promyelocytic leukaemia and one of acute undifferentiated leukaemia) occurred 12–30 months after the start of treatment with the mitoxantrone-containing regimen. [The Working Group noted that the standard treatment for breast cancer with cyclophosphamide (< 20 g/m^2), methotrexate and 5-fluorouracil has not been associated with leukaemia (Curtis *et al.*, 1992) and that the bone marrow is unlikely to have been affected by radiotherapy of the breast area.]

Vicari *et al*. (1998) reported on a 36-year-old man in Italy with acute promyelocytic leukaemia after treatment with mitoxantrone for multiple sclerosis. The patient had been treated with high doses of corticosteroids during exacerbation of the multiple sclerosis. Five years before the diagnosis of acute promyelocytic leukaemia, the patient had received an intravenous dose of mitoxantrone (10 mg/m^2) once a month for five months (total dose, 87.5 mg). The patient was reported to have no history of exposure to known leukaemogenic risk factors or a personal or family history of malignancy.

Partridge and Lowdell (1999) reported the development of myelodysplastic syndrome in a 62-year-old woman treated for advanced breast cancer with five courses of mitoxantrone (7 mg/m^2), methotrexate and mitomycin. In addition, she had received radiotherapy to the breast and axilla and tamoxifen. After 22 months, myelodysplastic syndrome was diagnosed.

2.2 Cohort study

In a pilot study to determine the toxicity of adjuvant treatment for early-stage breast cancer in a single hospital in Ireland, Cremin *et al.* (1996) reported on cases of acute myeloid leukaemia in 59 premenopausal women (32–54 years of age at

diagnosis) with early-stage breast cancer who were treated between 1986 and 1992 with adjuvant regimens containing mitoxantrone and methothrexate with or without mitomycin. The planned doses for the intravenous regimen that included mitomycin (n = 30) were: mitoxantrone, 8 mg/m^2 every three weeks (total dose, 64 mg); mitomycin, 8 mg/m^2 every six weeks (total dose, 32 mg) and methothrexate, 30 mg/m^2 every three weeks (total dose, 240 mg). The planned doses for the intravenous regimen that did not include mitomycin (n = 29) were: mitoxantrone, 12 mg/m^2 every three weeks (total dose, 96 mg) and methothrexate, 35 mg/m^2 every three weeks (total dose, 280 mg). During follow-up for a median of 72 months, two cases of acute myeloid leukaemia (one of acute myelomonocytic leukaemia and one of acute myeloblastic leukaemia) and one case of myelodysplastic syndrome occurred. All three patients had received treatment without mitomycin in combination with tamoxifen (three cases), radiotherapy (one case) or other cytostatic drugs (one case). The interval between treatment and diagnosis was 17 and 18 months for the cases of acute myeloid leukaemia and 36 months for the case of myelodysplastic syndrome. The frequency of acute myeloid leukaemia and myelodysplastic syndrome was 3/59 (5%) in the two treatment groups combined and 3/29 in the group given treatment without mitomycin, who had received a higher dose of mitoxantrone and a slightly higher dose of methotrexate than the group treated with mitomycin. [The Working Group noted that the risk for leukaemia was not compared with the risk of the general population; however, it is clear that 2/59 is substantially more than the expected number. On the basis of an incidence of 3–4 per 100 000 persons per year (Parkin *et al.*, 1997), the Working Group calculated a relative risk of about 200. The cumulative risk for leukaemia at five years was not reported. The dose of mitoxantrone associated with leukaemia was higher than that usually given in the treatment of advanced breast cancer.]

[The Working Group was aware of a number of cohort studies (Powles *et al.*, 1991; Stein *et al.*, 1992; Smith & Powles, 1993; Gregory *et al.*, 1997) in which the combination of methotrexate, mitoxantrone and mitomycin was used in the second- or third-line treatment of advanced breast cancer. These were not considered further because the follow-up was rarely longer than one year and the patients would previously have been treated with leukaemogenic agents and/or radiation.]

3. Studies of Cancer in Experimental Animals

No data were available to the Working Group.

4. Other Data Relevant to and Evaluation of Carcinogenicity and its Mechanisms

4.1 Absorption, distribution, metabolism and excretion

4.1.1 *Humans*

The pharmacokinetics of mitoxantrone in humans has been reviewed (Batra *et al*., 1986; Ehninger *et al*., 1990; Dunn & Goa, 1996). There are no published data on the bioavailability of orally administered mitoxantrone in humans, but a number of studies have reported the pharmacokinetics of mitoxantrone given as an intravenous infusion over 3–60 min at doses of 1–80 mg/m^2. All showed an initial rapid phase representing distribution of the drug into blood cells, with a half-time of about 5 min (range, 2–16 min) and a long terminal half-time of about 30 h (range, 19–72 h) (Savaraj *et al*., 1982; Alberts *et al*., 1983; Smyth *et al*., 1986; Van Belle *et al*., 1986; Larson *et al*., 1987; Hu *et al*., 1992; Richard *et al*., 1992; Feldman *et al*., 1993). Many early studies reported much shorter terminal half-times, but suitably sensitive assays may not have been used or adequate numbers of late samples collected. Tri-exponential elimination has been reported, the second distribution phase having a half-time of about 1 h (Alberts *et al*., 1983; Smyth *et al*., 1986; Van Belle *et al*., 1986; Hu *et al*., 1992), representing re-distribution from blood cells into tissues. The extent of the distribution into blood cells is illustrated by the observation that at the end of a 1-h infusion, the concentrations of mitoxantrone in leukocytes were 10 times higher than those in plasma (Sundman-Engberg *et al*., 1993), while 2–5 h after the dose the leukocyte or leukaemic cell concentrations were 350 times higher than those in plasma. The typical peak plasma concentration after a 30–60-min infusion of 12 mg/m^2 was about 500 ng/mL (Smyth *et al*., 1986; Van Belle *et al*., 1986; Larson *et al*., 1987). The rapid disappearance from plasma results in a total plasma clearance rate of about 500 mL/min, while the large volume of distribution of 500–4000 L/m^2 indicates tissue sequestration of the drug (Savaraj *et al*., 1982; Alberts *et al*., 1983; Smyth *et al*., 1986; Hu *et al*., 1992; Richard *et al*., 1992; Feldman *et al*., 1993).

Studies of patients given mitoxantrone at doses up to 80 mg/m^2 (standard dose, 12 mg/m^2) suggest that the kinetics is linear up to this dose (Alberts *et al*., 1983; Feldman *et al*., 1993).

Studies of the urinary excretion of mitoxantrone concur that little of the administered dose is cleared renally. Urinary recovery has been reported variously as < 7% within 24 h (Savaraj *et al*., 1982; Alberts *et al*., 1983; Van Belle *et al*., 1986), 3.3% within 48 h and < 8% within 72 h (Alberts *et al*., 1983; Smyth *et al*., 1986). In one study, urinary recovery of radiolabel after intravenous administration of [^{14}C]mitoxantrone accounted for 6.7% of the dose after 24 h and 10% after 72 h, with 3.9% and 5.1% as unchanged drug, respectively (Savaraj *et al*., 1982). Urinary recovery of intra-

venously administered mitoxantrone over five days accounted for 6.5% of the dose, most of which (90%) was recovered during the first 24 h (Alberts *et al.*, 1983).

The elimination half-time of mitoxantrone in two patients with impaired liver function was 63 h, whereas that in patients with normal liver function was 23 h (Smyth *et al*., 1986); in five patients with hepatic dysfunction, ascites or oedema, the terminal half-time was 71 h, while that in control patients was 37 h; and in two patients with oedema, the integrated area under the curve (AUC) of plasma concentration–time was double that of control patients (Rentsch *et al*., 1998). Faecal recovery of radiolabel after a single 12 mg/m^2 dose was 18% (range, 14–25%) over five days (Alberts *et al*., 1983). These results suggest that the liver is important in the elimination of mitoxantrone and that patients with impaired liver function or an abnormal fluid compartment may be at increased risk for toxic effects.

With infusion of mitoxantrone at a dose of 1.2 mg/m^2 per day for 21 days instead of the standard regimen of 12 mg/m^2 every three weeks, although the plasma concentrations reached a plateau after 35 h, the concentrations of mitoxantrone in leukocytes continued to increase throughout the 21 days suggesting that prolonged infusion may increase intracellular exposure to the drug (Greidanus *et al*., 1989).

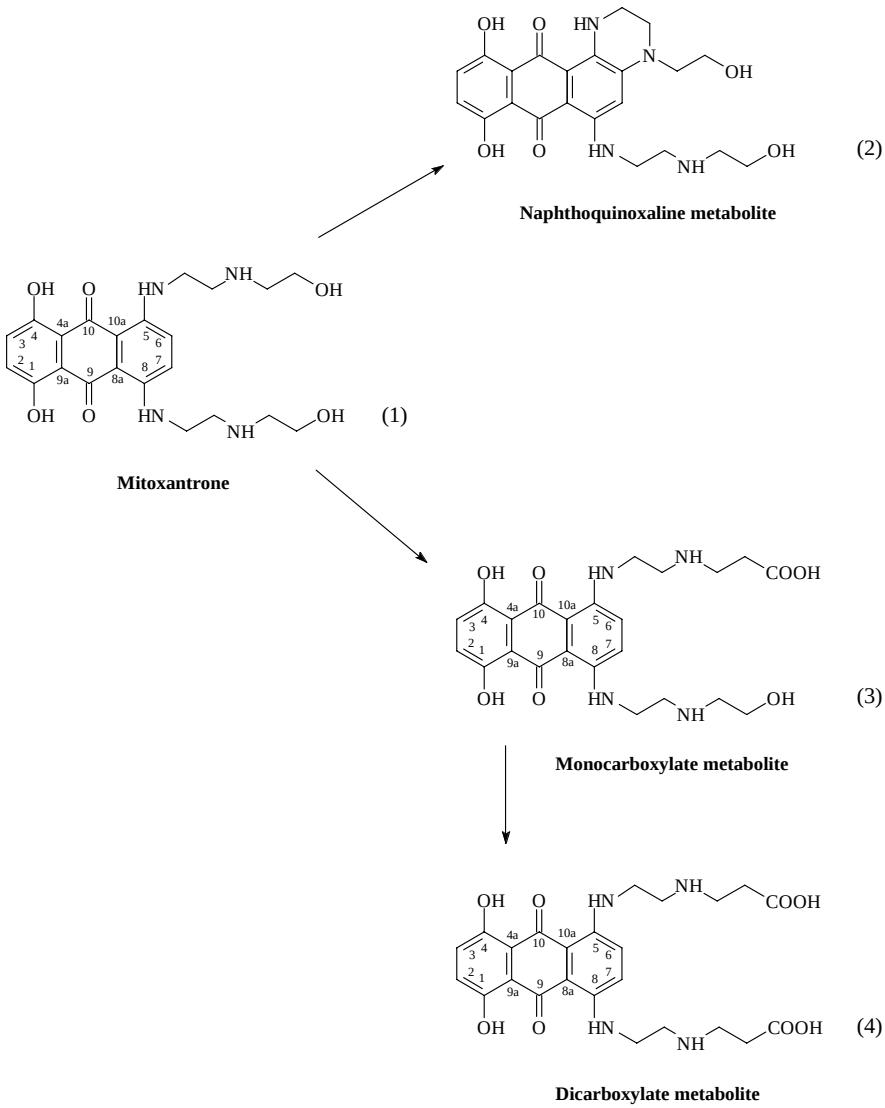
After intraperitoneal administration of mitoxantrone, high peritoneal concentrations but low systemic availability were reported, with a ratio of the AUC for peritoneal fluid to that for plasma of about 1000:1 (Alberts *et al*., 1988; Nagel *et al*., 1992).

The sequestration of mitoxantrone by body tissues results in retention of the drug for long periods. The characteristic blue–green colour of mitoxantrone has been observed on the surface of the peritoneum more than one month after intraperitoneal administration, and the concentrations in peritoneal tissue 6–22 weeks after intraperitoneal dosing ranged from < 0.1 to 14 μg/g tissue (Markman *et al*., 1993). Mitoxantrone was readily detectable in post-mortem tissue samples from all 11 patients who had received mitoxantrone intravenously between 10 and 272 days before death. The highest concentrations were found in the thyroid, liver and heart and the lowest in brain tissue (Stewart *et al*., 1986). In one patient given [^{14}C]mitoxantrone intravenously, who died 35 days after the dose, as much as 15% of the administered dose could be accounted for in the liver, bone marrow, lungs, spleen, kidney and thyroid glands (Alberts *et al*., 1983).

Limited data are available on the protein binding of mitoxantrone in humans. In one study, the fraction of unbound drug in plasma at the end of a 30-min infusion was only 3.0% (Hu *et al*., 1992).

Because of its limited urinary excretion, little information is available on the metabolism of mitoxantrone. Two inactive metabolites were identified in urine as the mono- and dicarboxylic acid derivatives resulting from oxidation of the terminal hydroxy groups of the side-chains (Figure 1) (Chiccarelli *et al*., 1986; Rentsch *et al*., 1998). The concentrations of mitoxantrone in urine were not altered by pre-incubation with a β-glucuronidase or sulfatase, suggesting that the drug is not excreted renally as either the glucuronide or sulfate conjugate (Smyth *et al*., 1986).

Figure 1. Metabolism of mitoxantrone



Adapted from Ehninger *et al.* (1986) and Mewes *et al.* (1993)

Studies with HepG2 hepatoma cells and rat hepatocytes suggest that mitoxantrone can be oxidized to an active naphthoquinoxaline metabolite which can bind covalently to RNA and DNA (Blanz *et al*., 1991; Mewes *et al*., 1993; Panousis *et al*., 1995). This metabolite has been identified in the urine of patients given mitoxantrone (Blanz *et al*., 1991), and two studies of cell systems suggest that it may contribute to the cytotoxic activity of mitoxantrone (Duthie & Grant, 1989; Mewes *et al*., 1993).

A 28-year-old woman in whom acute promyelocytic leukaemia was diagnosed during the 24th week of pregnancy was treated successfully with a variety of drugs and was given mitoxantrone prior to caesarean section at 34 weeks of gestation. After two further courses of 6 mg/m^2 mitoxantrone, her breast milk contained 120 ng/mL mitoxantrone 3–4 h after dosing and 18 ng/mL by five days, and the concentration remained at this level for 28 days. This finding indicates that the drug is slowly released from a deep tissue compartment (Azuno *et al*., 1995).

4.1.2 *Experimental systems*

There are no published studies of the pharmacokinetics of mitoxantrone given orally to laboratory animals. The drug was not developed for oral use, and in a review mitoxantrone was described as being poorly absorbed when administered orally [species not mentioned] (Batra *et al*., 1986).

In rats, dogs and monkeys, the disappearance of intravenously administered [^{14}C]-mitoxantrone from plasma was rapid, followed by a slow terminal elimination phase (James *et al*., 1983). In monkeys, the terminal half-time was eight days. Extensive tissue binding was indicated, with 50, 25 and 30% of the dose still retained 10 days after intravenous administration in rats, dogs and monkeys, respectively. In beagle dogs, triexponential elimination from plasma was reported, with a very rapid initial distribution phase with a half-time of 6.5 min, a longer distribution phase of 1.3 h and a slow terminal elimination of 28 h. Less than 4% of the dose was excreted in the urine during the first 48 h. Extensive tissue retention was again reported, the higher concentrations 24 h after dosing being found in the liver, kidney and spleen. Plasma protein binding was reported to be 70–80%. Two metabolites were detected, accounting for 30% of the radiolabel in plasma and 50% in urine, but were not identified (Lu *et al*., 1984).

A rapid distribution and a slow elimination phase were also observed in mice, with retention in body tissues, particularly liver and kidney (Rentsch *et al*., 1997), and in rabbits (Hulhoven *et al*., 1983).

A naphthoquinoxaline metabolite of mitoxantrone has been reported in rats and pigs, resulting from the oxidation of the phenylenediamine substructure (Blanz *et al.*, 1991; Figure 1). In general, mitoxantrone is believed to be active in mammalian cells *in vitro* in the absence of exogenous metabolic activation; however, inhibition of cytochrome P450 mixed-function oxidase by metyrapone in HepG2 hepatoxic cells and rat hepatocytes blocked the cytotoxic activity of mitoxantrone, suggesting that conversion to reactive species might be important (Duthie & Grant, 1989; Mewes *et al.*, 1993).

Panousis *et al*. (1995) reported that mitoxantrone is readily oxidized by myeloperoxidase or hydrogen peroxide and that the metabolites bind covalently to DNA. Various metabolic species and various DNA adducts have been chemically characterized, but their mutagenic properties have not been studied (Mewes *et al*., 1993; Dackiewicz *et al*., 1995).

4.2 Toxic effects

4.2.1 *Humans*

The acute toxicity of mitoxantrone has been reviewed (Smith, 1983; Faulds *et al*., 1991; Dunn & Goa, 1996). Leukopenia is the main dose-limiting effect, the lowest leukocyte counts typically being found 10–14 days after a single dose, with recovery by day 21. Thrombocytopenia occurs but is less common. Of 505 patients treated in phase II studies at the standard dose of 12–14 mg/m^2, 5% experienced very severe leukopenia (leukocyte count, $< 1 \times 10^9$/L), and 0.8% experienced very severe thrombocytopenia (platelet count, $< 25 \times 10^9$/L) (Smith, 1983). Patients with AIDS and Kaposi sarcoma treated at a dose of 12 mg/m^2 all experienced severe neutropenia, possibly related to impaired bone-marrow function (Kaplan & Volberding, 1985).

Like other anthracyclines, mitoxantrone is associated with cardiotoxicity. In large phase II studies, summarized by Smith (1983), 15 of 543 patients experienced cardiotoxicity (3%), reported as congestive cardiac failure, pulmonary oedema or unspecified. In a large European trial, seven of 264 patients experienced cardiac abnormalities (3%). Risk factors that may be predictive of the cardiotoxicity of this drug are previous anthracycline therapy, mediastinal radiotherapy and a history of cardiovascular disease (Crossley, 1983). The number of cardiotoxic events increases with cumulative doses of mitoxantrone > 120 mg/m^2 in patients who have previously been treated with anthracyclines, and > 160 mg/m^2 in patients who were not previously treated. Henderson *et al*. (1989) reported moderate to severe decreases in left-ventricular ejection fraction in eight of 132 patients treated with mitoxantrone, all of whom had received cumulative doses > 100 mg/m^2. The cumulative dose at which a patient has a 50% probability of having to discontinue treatment because of cardiotoxicity was estimated to be 182 mg/m^2, representing approximately 13 courses of treatment.

Other toxic effects seen with standard doses of mitoxantrone (12–14 mg/m^2) include nausea and vomiting (in approximately 50% of patients), diarrhoea (15%), stomatitis and mucositis (20%) and alopecia (50%), although these effects are usually mild and transient (Crossley, 1983). As the drug is an intense blue colour, discolouration of urine and skin is not uncommon. Cases of onycholysis have been reported (Creamer *et al*., 1995).

With higher doses (40–90 mg/m^2 or 12 mg/m^2 on days 1–3), the toxic effects are typically more severe, and hepatotoxicity has been reported (Feldman *et al*., 1993; Ballestrero *et al*., 1997; Feldman *et al*., 1997), manifest as transient increases in serum bilirubin concentration and in the activity of liver enzymes, and becoming more common with increasing dose (Feldman *et al*., 1993).

After intraperitoneal dosing, peritonitis is the dose-limiting toxic effect (Alberts *et al*., 1988).

4.2.2 *Experimental systems*

Much of the information on the toxicity of mitoxantrone in experimental animals has not been published in detail. The LD_{50} was reported to be about 5 mg/kg bw in rats and 10 mg/kg bw in mice, and the single minimum lethal dose in beagle dogs was 10 mg/m^2 (Henderson *et al*., 1982).

Many of the studies of the toxicity of mitoxantrone have focused on its cardiac effects, particularly in comparison with doxorubicin, another anthracycline known to be toxic to the heart. Beagle dogs given half the lethal dose (5.15 mg/m^2) had decreased leukocyte counts, haematocrit and haemoglobin concentration, diarrhoea, cutaneous sores and inactivity, one animal dying of pneumonia, but there was no evidence of cardiotoxicity at this dose (Henderson *et al*., 1982). At doses > 2 mg/kg bw, mitoxantrone induced cardiovascular and renal toxicity in rabbits (Hulhoven *et al*., 1983). In cats given mitoxantrone at doses of 2.5–6.5 mg/m^2 for the treatment of malignant tumours, the most common toxic effects were vomiting, anorexia, diarrhoea, lethargy, sepsis secondary to myelosuppression and seizures. Two cats died of complications that may have been attributable to mitoxantrone: one of cardiomyopathy and the other of pulmonary oedema (Ogilvie *et al*., 1993).

Hepatoxicity was observed in mice given 15 mg/kg bw (Llesuy & Arnaiz, 1990).

Mitoxantrone at clinically relevant concentrations was toxic to cultured heart cells from Sprague-Dawley rats, an effect that could be prevented by the chelating agent ICRF-187 (Shipp & Dorr, 1991; Shipp *et al*., 1993).

4.3 Reproductive and prenatal effects

4.3.1 *Humans*

A woman aged 39 who was treated for 14 weeks with mitoxantrone at 20 mg intravenously every three weeks for cystic adenoma of the adenoids became amenorrhoeic and had hot flushes after a cumulative dose of 100 mg. She had received no other treatment and had not taken hormones or oral contraceptives. Measurement of luteinizing hormone, follicle-stimulating hormone and oestradiol in her blood showed that their concentrations were in the menopausal range (Shenkenberg & Von Hoff, 1986). [The Working Group noted that ovarian biopsy was not performed and that the lack of ovarian function might have been related to the woman's age.]

A 26-year-old woman in the 20th week of pregnancy was treated with cytarabine and daunorubicin for acute myeloblastic leukaemia. Three weeks later, she received mitoxantrone at 12 mg/m^2 for three days in combination with cytarabine. The pregnancy continued, with normal fetal growth, for 60 days when she had complete remission. She was then given idarubicin and cytarabine for two days. Two days later, the fetus was found to be dead *in utero* (Reynoso & Huerta, 1994).

4.3.2 *Experimental systems*

In a brief review of the toxicology of mitoxantrone, an increased frequency of fetal resorptions and decreased fetal body weights were observed in pregnant rats dosed intravenously with 0.25 mg/kg bw, but no effects were observed in rabbits at intravenous doses up to 0.5 mg/kg bw [no details of treatment times or numbers of animals used were given] (James *et al.*, 1983).

4.4 Genetic and related effects

4.4.1 *Humans*

There are few studies of the genotoxicity of mitoxantrone in humans. Liang *et al.* (1993) evaluated the induction of chromosomal breaks and sister chromatid exchange in cultured lymphocytes from 42 patients with Hodgkin disease, collected before and during treatment with mitoxantrone in combination with vincristine, vinblastine and prednisone. The authors found no evidence of increased frequencies of chromosome or single-stranded DNA breaks or sister chromatid exchange.

Vicari *et al.* (1998) detected the PML-RAR-α fusion transcript, consistent with translocation t(15;17), in a case of acute promyelocytic leukemia that occurred five years after a cumulative dose of 50 mg/m^2 mitoxantrone administered as a single agent for the treatment of multiple sclerosis. It should be noted, however, that this translocation occurs in nearly all cases of de-novo acute promyelocytic leukaemia. In a review, Quesnel *et al.* (1993) mentioned a case of acute myeloid leukaemia with a complex karyotype including t(8;21) after treatment with mitoxantrone as a single agent for breast cancer.

Leblanc *et al.* (1994) identified a *MLL* gene rearrangement by Southern blot analysis in a case of acute monoblastic leukaemia with translocation t(1;11)(q13;q23) that followed chemotherapy with a mitoxantrone-containing protocol for primary acute myeloid leukaemia with inv(16). The therapy for the primary leukaemia included induction with cytarabine and mitoxantrone, two consolidations with cytarabine, daunorubicin and etoposide and then cytarabine and amsacrine followed by maintanence therapy with 6-mercaptopurine and cytarabine. The regimen thus contained three DNA topoisomerase II inhibitors. Which, if any, can be linked to the translocation t(1;11)(q13;q23) is uncertain.

Similarly, Bredeson *et al.* (1993) observed a case of acute monoblastic leukaemia with t(9;11)(p22;q23) 47 months after the initiation of treatment for primary non-Hodgkin lymphoma with a mitoxantrone-containing regimen that also included alkylating agents and other DNA topoisomerase II inhibitors.

Pedersen-Bjergaard and Philip (1991) reported a balanced translocation involving chromosome band 21q22 in a case of acute myeloid leukaemia that followed mitoxantrone-containing therapy.

Izumi *et al*. (1996) described a case of acute myeloblastic leukaemia with t(2;21)(q21;q22), t(8;21)(q22;q22) and add (13)(q34) after treatment for non-Hodgkin lymphoma with mitoxantrone, other DNA topoisomerase II inhibitors and alkylating agents.

Detourmignies *et al.* (1992) reported a case of acute promyelocytic leukaemia that occurred nine months after initiation of treatment of breast cancer with mitoxantrone, vincristine, cyclophosphamide and 5-fluorouracil, but karyotype analysis was not performed. Melillo *et al*. (1997) observed three cases of acute myeloid leukaemia after therapy with mitoxantrone, mitomycin and methotrexate for advanced breast cancer. In one case, the karyotype was normal. In the second case, which was an acute promyelocytic leukaemia, the karyotype revealed t(15;17)(q22;q12), typical of this leukaemia, and the PML-RAR-α fusion transcript was detected by reverse transcriptase polymerase chain reaction. The karyotype of the third case showed del(3)(q12;q25), –7, –19, +mar.

4.4.2 *Experimental systems*

General reviews on the mutagenicity of inhibitors of DNA topoisomerase II enzymes, including mitoxantrone, have been published (Anderson & Berger, 1994; Ferguson & Baguley, 1994, 1996; Baguley & Ferguson, 1998; Ferguson, 1998). Jackson *et al*. (1996) collated a genetic activity profile for this drug. The results are summarized in Table 1.

Limited data are available on the mutagenic effects of mitoxantrone in microbial assays, but, by analogy with other DNA topoisomerase II inhibitors, it probably gives weak or negative responses in assays in prokaryotes and lower eukaryotes. Mitoxantrone weakly induced reverse mutation in *Salmonella typhimurium* TA98 and TA1537, and these effects did not depend on metabolic activation. It did not induce reverse mutation in *S. typhimurium* TA1535 or TA100 in the presence or absence of metabolic activation. In another study, mitoxantrone induced reverse mutation in strains TA98, TA1538 and TA1978 in the absence of metabolic activation.

Mitoxantrone is a potent inducer of DNA breakage in mammalian cells *in vitro* and *in vivo*. It caused protein-masked DNA double-strand breaks and DNA–protein cross-links in various animal and human cell lines at a concentration of about 1 μmol/L. These strand breakage effects could be enhanced in T-47D human breast cancer cells by prior stimulation with oestrogen. In a single study, mitoxantrone did not induce DNA breakage in peripheral blood cells obtained from leukaemia patients treated with the drug.

Mitoxantrone was highly effective in causing chromosomal aberrations in cultured Chinese hamster cells and in human peripheral blood lymphocytes in tissue culture. These effects were reduced when an exogenous metabolic activation system was added to the cells. Mitoxantrone induced chromosomal aberrations in the bone marrow of rats.

Table 1. Genetic and related effects of mitoxantrone

Test system	Result[a]		Dose[b] (LED or HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Salmonella typhimurium TA1537, TA98, reverse mutation	+	+	32.5 mmol/L[c]	Au *et al*. (1981)
Salmonella typhimurium TA100, TA1535, reverse mutation	–	–	130 mmol/L[c]	Au *et al*. (1981)
Salmonella typhimurium TA1538, reverse mutation	+	NT	50 µg/plate	Matney *et al.* (1985)
Salmonella typhimurium TA98, reverse mutation	(+)	NT	200 µg/plate	Matney *et al.* (1985)
Salmonella typhimurium TA1978, reverse mutation	+	NT	20 µg/plate	Matney *et al.* (1985)
Drosophila melanogaster, somatic mutation and recombination	+		2000 in feed	Clements *et al*. (1990)
*Drosophila melanogaste*r, somatic mutation and recombination	+		445 in feed	Frei *et al*. (1992)
Drosophila melanogaster, sex-linked recessive lethal mutation	(+)		1335 in feed	Frei *et al*. (1992)
DNA single-strand breaks, Chinese hamster ovary cells *in vitro*	+	NT	0.05	Štetina & Veselá (1991)
DNA–protein cross-linkage, Chinese hamster ovary and UV-5 (excision repair-deficient) cells *in vitro*	+	NT	0.5	Štetina & Veselá (1991)
Unscheduled DNA synthesis, Sprague-Dawley rat primary hepatocytes *in vitro*	+	NT	0.1	Manandhar *et al*. (1986)
Gene mutation, L5178Y mouse lymphoma cells, *Tk* locus *in vitro*	+	+	0.0003	Manandhar *et al*. (1986)
Sister chromatid exchange, Chinese hamster ovary cells *in vitro*	+	+	0.022	Au *et al*. (1981)
Sister chromatid exchange, Chinese hamster ovary cells *in vitro*	+	NT	0.0004	Nishio *et al*. (1982)
Sister chromatid exchange, Chinese hamster ovary cells *in vitro*	+	+	0.0003	Manandhar *et al*. (1986)
Chromosomal aberrations, Chinese hamster ovary cells *in vitro*	+	(+)	0.004	Au *et al*. (1981)
Chromosomal aberrations, Chinese hamster ovary cells *in vitro*	+	NT	0.0004	Nishio *et al*. (1982)
Chromosomal aberrations, Chinese hamster ovary cells *in vitro*	+	NT	0.1	Rosenberg & Hittelman (1983)
Chromosomal aberrations, Chinese hamster lung cells *in vitro*	+	NT	0.01	Suzuki & Nakane (1994)
Polyploidy, Chinese hamster ovary cells *in vitro*	+	NT	9	Sumner (1995)
Cell transformation, C3H 10T1/2 mouse cells	–	NT	0.0005	Manandhar *et al*. (1986)

Table 1 (contd)

Test system	Result[a]		Dose[b] (LED or HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
DNA double-strand breaks, human leukaemia and lymphoma cell lines *in vitro*	+	NT	0.1	Ho *et al*. (1987)
DNA double-strand breaks, T-47D human breast cancer cells *in vitro*	+	NT	0.05	Epstein & Smith (1988)
DNA single and double-strand breaks, human LoVo cell line *in vitro*	+	NT	0.11	Capolongo *et al*. (1990)
DNA single-strand breaks, human lung carcinoma cells *in vitro*	+	NT	0.04	De Isabella *et al.* (1993)
DNA double-strand breaks, NCI-H69 human cells *in vitro*	+	NT	0.125	Smith *et al*. (1990)
DNA double-strand breaks, human astrocytoma and glioblastoma cell lines *in vitro*	+	NT	0.01	Senkal *et al*. (1997)
Chromosomal aberrations, human lymphocytes *in vitro*	+	NT	12×10^{-6}	Medeiros & Takahashi (1994)
Chromosomal aberrations, Sprague-Dawley rat bone-marrow cells *in vivo*	+		1 ip × 5	Manandhar *et al.* (1986)
Dominant lethal mutation, male and female Sprague-Dawley rats	–		1 ip × 5	Manandhar *et al.* (1986)
DNA single-strand breaks, human leukaemia cells *in vivo*	–		5 mg/m^2 iv × 3	Heinemann *et al.* (1988)

[a] +, positive; (+), weak positive; –, negative; NT, not tested

[b] LED, lowest effective dose; HID, highest ineffective dose; in-vitro tests, µg/mL; in-vivo tests, mg/kg bw per day; d, day

[c] Unclear what this concentration refers to

Mitoxantrone induced sister chromatid exchange in Chinese hamster cells. It also induced mutation and somatic recombination in the *Drosophila white–ivory* test for somatic mutation and in the wing spot test.

Mitoxantrone induced primarily small colony mutants at the *Tk* locus in mouse lymphoma L5178Y cells, in the presence or absence of exogenous metabolic activation. Small colony mutants in L5178Y cells are generally considered to be caused by chromosomal mutations (DeMarini *et al*., 1987).

Some discrepancies with regard to the activity of mitoxantrone have been found in various assays. Although apoptosis may eliminate cells that appear to be mutated (Ferguson & Baguley, 1994), short-term (2–6 h) exposure of the human myeloid leukaemia line HL-60 and of MOLT-4 cells to 0.02–0.4 μg/mL mitoxantrone induced cell cycle arrest rather than apoptosis in experiments reported by Del Bino and Darzynkiewicz (1991). In other studies, incubation of the human myeloid leukaemia lines HL-60 and KG-1 with mitoxantrone for only 1 h at concentrations between 0.1 and 10 μmol/L gave clear indications of apoptosis (Bhalla *et al*., 1993). The culture media and conditions differed between the two laboratories, and serum levels and types of media affect apoptosis (Ferguson & Baguley, 1996). Mitoxantrone-induced apoptosis was demonstrated in cultured B lymphocytes from a patient with chronic lymphoblastic leukaemia (Bellosillo *et al*., 1998).

Mitoxantrone-induced polyploidy was demonstrated by cytogenetic techniques in Chinese hamster ovary cells by Sumner (1995). Although not directly studied for induction of aneuploidy, mitoxantrone inhibited the polymerization of microtubule assembly (Ho *et al*., 1991) and almost certainly would act as an aneuploidogen. It did not induce transformation in C3H 10T1/2 cells (Manandhar *et al*., 1986).

Equivocal results were found in studies of germ cells in *Drosophila*. A clinical preparation of mitoxantrone weakly induced sex-linked recessive mutation, but the response failed to reach statistical significance (Frei *et al*., 1992). Similarly, assays for dominant lethal mutation in male and female Sprague-Dawley rats showed signs of reduced pregnancy rates but no clear statistical trend in dominant lethal events in either sex. [The Working Group noted that, given the unusual timing of the effects of etoposide on germ cells (Russell *et al*., 1998), these experiments would bear repetition with different schedules, as the available data must be considered equivocal.]

4.5 Mechanistic considerations

Mitoxantrone has three properties that are likely to induce mutation.

1. It inhibits DNA topoisomerase II enzymes: Mitoxantrone is a DNA topoisomerase II poison that has been shown to resemble etoposide and teniposide in promoting DNA cleavage, with a strong preference for C or T at position –1 (Capranico *et al.*, 1993; De Isabella *et al.*, 1993). Most of the mutational events reported in mammalian cells, including point mutations, chromosomal deletions and exchanges and aneuploidy, can be explained by this activity. Mitoxantrone does not inhibit bacterial topoisomerases

and may not mutate bacterial cells by the same mechanism as mammalian cells. Instead, it has two other activities that may be responsible for other types of mutation.

2. It possesses readily oxidizable functions: Oxidation of the substituted anthraquinone skeleton leads to biotransformation of mitoxantrone (Mewes *et al*., 1993). Panousis *et al*. (1995) reported that myeloperoxidase oxidizes mitoxantrone to metabolites that bind covalently to DNA. Dackiewicz *et al*. (1995) reported formation of several different DNA adducts when mitoxantrone was incubated with a peroxidase/hydrogen peroxide system. Nevertheless, none of the mutations seen with mitoxantrone is of the type usually associated with oxygen radicals.

3. It intercalates into, but does not covalently interact with, DNA: By analogy with other frameshift mutagens, mitoxantrone causes the frameshift mutagenicity seen in bacteria by DNA intercalation (Ferguson & Denny, 1990). A frameshift event was observed in both strains of *S. typhimurium* in which it caused reverse mutation (Au *et al.*, 1981).

Mitoxantrone-containing regimens are associated with chromosomal translocations in leukaemic cells similar to those observed with other DNA topoisomerase II inhibitors. The role of DNA topoisomerase II inhibitors in translocations associated with leukaemia is unknown. Two possibilities are plausible. The first is that mitoxantrone itself causes the translocations. It has been proposed that DNA cleavage induced directly by DNA topoisomerase II or by the drug-induced apoptotic cellular response is responsible for the nonrandom chromosomal translocations that lead to leukaemogenesis (Dassonneville & Bailly, 1998). The second possibility for the role of mitoxantrone in causing translocations is that it selects for cells that already have translocations. Indeed, *MLL* tandem duplications, a form of translocation, have been identified in peripheral blood and bone marrow of healthy adults (Schnittger *et al*., 1998). Chemotherapy has profound effects on the kinetics of the bone marrow; it causes cell death, forcing many bone-marrow stem cells to divide, which might select for the rare stem cells with a translocation (Knudson, 1992). In favour of the first possibility is the specificity of the association between DNA topoisomerase II inhibitors, but not other forms of chemotherapy that cause cell death in the bone marrow (such as alkylating agents), and leukaemias characterized by translocations.

5. Summary of Data Reported and Evaluation

5.1 Exposure data

Mitoxantrone is a synthetic DNA topoisomerase II inhibitor of the anthracenedione class that has been used in cancer treatment since the mid-1980s. It is used mainly in the treatment of advanced breast cancer, non-Hodgkin lymphoma and certain leukaemias. Recently, it has been used in the treatment of multiple sclerosis.

5.2 Human carcinogenicity data

In the one available, small cohort study of women with early-stage premenopausal breast cancer who had been treated with mitoxantrone in the absence of known or suspected leukaemogenic agents, a substantially increased risk for acute myeloid leukaemia was observed.

Case reports of acute myeloid leukaemia developing in patients treated with mitoxantrone are compatible with the association found in the cohort study.

5.3 Animal carcinogenicity data

No data were available to the Working Group.

5.4 Other relevant data

In humans, mitoxantrone is eliminated biphasically or triphasically, with a terminal half-time of 19–72 h. The drug is rapidly taken up by blood cells and is extensively distributed in body tissues. The pharmacokinetics of mitoxantrone is linear up to 80 mg/m^2 (standard dose, 12 mg/m^2). The elimination half-life was prolonged in patients with impaired hepatic function and in patients with ascites or oedema. Urinary recovery of mitoxantrone as the parent drug or radiolabel is low (< 10%), and significant amounts are still present in body tissues weeks or months after dosing. Few data are available on the metabolism of mitoxantrone in humans, but two inactive metabolites have been reported.

A long elimination phase and tissue retention are also seen in animal species. Active naphthoquinoxaline mitoxantrone metabolites have been reported in some experimental systems.

The main dose-limiting toxic effect of mitoxantrone is myelosuppression, manifest mostly as leukopenia. Other toxic effects include nausea and vomiting, diarrhoea, stomatitis, mucositis and alopecia. Cardiotoxicity is reported in about 3% of patients and is more common with cumulative doses of 160 mg/m^2 in previously untreated patients and 120 mg/m^2 in previously treated patients, particularly in those who have received anthracyclines.

Mitoxantrone can mutate cells through one of three mechanisms. It intercalates into DNA and causes frameshift mutations in bacteria through that mechanism. Although the drug *per se* does not interact covalently with the DNA, it is readily oxidized to a species which does form DNA adducts; however, there is currently little evidence that DNA adduct formation is critical for mutagenic events in mammalian cells. The drug is orders of magnitude more toxic in mammalian than in microbial cells. Most of the effects in mammals arise because mitoxantrone is an effective poison of DNA topoisomerase II enzymes. The predominant effects seen to date involve the deletion and/or interchange of large DNA segments. Additionally, mitoxantrone induces polyploidy.

Chromosomal translocations characteristic of those that occur after administration of DNA topoisomerase II inhibitors have been observed in leukaemic cells of patients treated with mitoxantrone-containing regimens. The mode of action of this compound is similar to that of others for which evidence of a leukaemogenic effect is more compelling.

5.5 Evaluation

There is *limited evidence* in humans for the carcinogenicity of mitoxantrone.

There is *inadequate evidence* in experimental animals for the carcinogenicity of mitoxantrone.

Overall evaluation

Mitoxantrone is *possibly carcinogenic to humans (Group 2B).*

6. References

Alberts, D.S., Peng, Y.-M., Leigh, S., Davis, T.P. & Woodward, D.L. (1983) Disposition of mitoxantrone in patients. *Cancer Treat. Rev*., **10** (Suppl. B), 23–27

Alberts, D.S., Surwit, E.A., Peng, Y.-M., McCloskey, T., Rivest, R., Graham, V., McDonald, L. & Roe, D. (1988) Phase I clinical and pharmacokinetic study of mitoxantrone given to patients by intraperitoneal administration. *Cancer Res*., **48**, 5874–5877

Anderson, R.D. & Berger, N.A. (1994) Mutagenicity and carcinogenicity of topoisomerase-interactive agents. *Mutat. Res.*, **309**, 109–142

Au, W.W., Butler, M.A., Matney, T.S. & Loo, T.L. (1981) Comparative structure–genotoxicity study of three aminoanthraquinone drugs and doxorubicin. *Cancer Res*., **41**, 376–379

Azuno, Y., Kaku, K., Fujita, N., Okubo, M., Kaneko, T. & Matsumono, N. (1995) Mitoxantrone and etoposide in breast milk (Letter to the Editor). *Am. J. Hematol*., **48**, 131–132

Baguley, B.C. & Ferguson, L.R. (1998) Mutagenic properties of topoisomerase-targeted drugs. *Biochim. biophys. Acta*, **1400**, 213–222

Ballestrero, A., Ferrando, F., Garuti, A., Basta, P., Gonella, R., Esposito, M., Vannozzi, M.O., Sorice, G., Friedman, D., Puglisi, M., Brema, F., Mela, G.S., Sessarego, M. & Patrone, F. (1997) High-dose mitoxantrone with peripheral blood progenitor cell rescue: Toxicity, pharmacokinetics and implications for dosage and schedule. *Br. J. Cancer*, **76**, 797–804

Batra, V.K., Morrison, J.A., Woodward, D.L., Siverd, N.S. & Yacobi, A. (1986) Pharmacokinetics of mitoxantrone in man and laboratory animals. *Drug Metab. Rev*., **17**, 311–329

Beijnen, J.H., Bult, A. & Underberg, W.J.M. (1988) Mitoxantrone hydrochloride. In: Florey, K., ed., *Analytical Profiles of Drug Substances*, New York, Academic Press, Vol. 17, pp. 221–258

Bellosillo, B., Colomer, D., Pons, G. & Gil, J. (1998) Mitoxantrone, a topoisomerase II inhibitor, induces apoptosis of B-chronic lymphocytic leukaemia cells. *Br. J. Haematol*., **100**, 142–146

Bhalla, K., Ibrado, A.M., Tourkina, E., Tang, C., Grant, S., Bullock, G., Huang, Y., Ponnathpur, V. & Mahoney, M.E. (1993) High-dose mitoxantrone induces programmed cell death or apoptosis in human myeloid leukemia cells. *Blood*, **82**, 3133–3140

Blanz, J., Mewes, K., Ehninger, G., Proksch, B., Waidelich, D., Greger, B. & Zeller, K.-P. (1991) Evidence for oxidative activation of mitoxantrone in human, pig, and rat. *Drug Metab. Dispos.*, **19**, 871–880

Bredeson, C.N., Barnett, M.J., Horsman, D.E., Dalal, B.I., Ragaz, J. & Phillips, G.L. (1993) Therapy-related acute myelogenous leukemia associated with 11q23 chromosomal abnormalities and topoisomerase II inhibitors: Report of four additional cases and brief commentary. *Leuk. Lymphoma*, **11**, 141–145

British Medical Association/Royal Pharmaceutical Society of Great Britain (1998) *British National Formulary*, No. 36, London, p. 373

Budavari, S., ed. (1996) *The Merck Index*, 12th Ed., Whitehouse Station, NJ, Merck & Co., p. 1064

Canadian Pharmaceutical Association (1997) *CPS Compendium of Pharmaceuticals and Specialties*, 32nd Ed., Ottawa, pp. 1071–1072

Capolongo, L., Belvedere, G. & D'Incalci, M. (1990) DNA damage and cytotoxicity of mitoxantrone and doxorubicin in doxorubicin-sensitive and -resistant human colon carcinoma cells. *Cancer Chemother. Pharmacol.*, **25**, 430–434

Capranico, G., De Isabella, P., Tinelli, S., Bigioni, M. & Zunino, F. (1993) Similar sequence specificity of mitoxantrone and VM-26 stimulation of in vitro DNA cleavage by mammalian DNA topoisomerase II. *Biochemistry*, **32**, 3038–3046

Chiccarelli, F.S., Morrison, J.A., Cosulich, D.B., Perkinson, N.A., Ridge, D.N., Sum, F.W., Murdock, K.C., Woodward, D.L. & Arnold, E.T. (1986) Identification of human urinary mitoxantrone metabolites. *Cancer Res*., **46**, 4858–4861

CIS Information Services (1998) *Worldwide Bulk Drug Users Directory 1997/98 Edition*, Dallas, TX [CD-ROM]

Clements, J., Howe, D., Lowry, A. & Phillips, M. (1990) The effects of a range of anti-cancer drugs in the *white–ivory* somatic mutation test in *Drosophila*. *Mutat. Res*., **228**, 171–176

Council of Europe (1998) *European Pharmacopoeia*, 3rd Ed., Supplement 1998, Strasbourg, pp. 387–388

Creamer, J.D., Mortimer, P.S. & Powles, T.J. (1995) Mitozantrone-induced onycholysis. A series of five cases. *Clin. exp. Dermatol*., **20**, 459–461

Cremin, P., Flattery, M., McCann, S.R. & Daly, P.A. (1996) Myelodysplasia and acute myeloid leukaemia following adjuvant chemotherapy for breast cancer using mitoxantrone and methothrexate with or without mitomycin (Short report). *Ann. Oncol*., **7**, 745–746

Crossley, R.J. (1983) Clinical safety and tolerance of mitoxantrone (Novantrone). *Cancer Treat. Rev*., **10**, 29–36

Curtis, R.E., Boice, J.D., Jr, Stovall, M., Bernstein, L., Greenberg, R.S., Flannery, J.T., Schwartz, A.G., Weyer, P., Moloney, W.C. & Hoover, R.N. (1992) Risk of leukemia after chemotherapy and radiation treatment for breast cancer. *New Engl. J. Med.*, **326**, 1745–1751

Dackiewicz, P., Skladanowski, A. & Konopa, J. (1995) ^{32}P-postlabelling analysis of adducts formed by mitoxantrone and ametantrone with DNA and homopolydeoxyribonucleotides after enzymatic activation. *Chem.-biol. Interactions*, **98**, 153–166

Dassonneville, L. & Bailly, C. (1998) [Chromosomal translocations and secondary leukaemias induced by topoisomerase II inhibitors.] *Bull. Cancer*, **85**, 254–261 (in French)

De Isabella, P., Capranico, G., Palumbo, M., Sissi, C., Krapcho, A.P. & Zunino, F. (1993) Sequence selectivity of topoisomerase II DNA cleavage stimulated by mitoxantrone derivatives: Relationships to drug DNA binding and cellular effects. *Mol. Pharmacol.*, **43**, 715–721

Del Bino, G. & Darzynkiewicz, Z. (1991) Camptothecin, teniposide, or 4′-(9-acridinylamino)-3-methanesulfon-*m*-anisidide, but not mitoxantrone or doxorubicin, induces degradation of nuclear DNA in the S phase of HL-60 cells. *Cancer Res.*, **51**, 1165–1169

DeMarini, D.M., Brock, K.H., Doerr, C.L. & Moore, M.M. (1987) Mutagenicity and clastogenicity of teniposide (VM-26) in L5178Y/TK$^{+/-}$-3.7.2C mouse lymphoma cells. *Mutat. Res.*, **187**, 141–149

Detourmignies, L., Castaigne, S., Stoppa, A.M., Harousseau, J.L., Sadoun, A., Janvier, M., Demory, J.L., Sanz, M., Berger, R., Bauters, F., Chomienne, C. & Fenaux, P. (1992) Therapy-related acute promyelocytic leukemia: A report on 16 cases. *J. clin Oncol.*, **10,** 1430–1435

Dunn, C.J. & Goa, K.L. (1996) Mitoxantrone. A review of its pharmacological properties and use in acute nonlymphoblastic leukaemia. *Drugs Aging*, **9**, 122–147

Duthie, S.J. & Grant, M.H. (1989) The role of reductive and oxidative metabolism in the toxicity of mitoxantrone, adriamycin and menadione in human liver derived Hep G2 hepatoma cells. *Br. J. Cancer*, **60**, 566–571

Editions du Vidal (1998) *Dictionnaire Vidal 1998*, 74th Ed., Paris, OVP, p. 1321

Ehninger, G., Proksch, B., Heinzel, G. & Woodward, D.L. (1986) Clinical pharmacology of Mitoxantrone. *Cancer Treat. Rep.*, **70**, 1373–1378

Ehninger, G., Schuler, U., Proksch, B., Zeller, K.-P. & Blanz, J. (1990) Pharmacokinetics and metabolism of mitoxantrone. A review. *Clin. Pharmacokinet.*, **18**, 365–380

Epstein, R.J. & Smith, P.J. (1988) Estrogen-induced potentiation of DNA damage and cytotoxicity in human breast cancer cells treated with topoisomerase II-interactive antitumor drugs. *Cancer Res.*, **48**, 297–303

Faulds, D., Balfour, J.A., Chrisp, P. & Langtry, H.D. (1991) Mitoxantrone. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in the chemotherapy of cancer. *Drugs*, **41**, 400–449

Feldman, E.J., Alberts, D.S., Arlin, Z., Ahmed, T., Mittelman, A., Baskind, P., Peng, Y.-M., Baier, M. & Plezia, P. (1993) Phase I clinical and pharmacokinetic evaluation of high-dose mitoxantrone in combination with cytarabine in patients with acute leukemia. *J. clin. Oncol.*, **11**, 2002–2009

Feldman, E.J., Seiter, K., Damon, L., Linker, C., Rugo, H., Ries, C., Case, D.C., Jr, Beer, M. & Ahmed, T. (1997) A randomized trial of high- vs standard-dose mitoxantrone with cytarabine in elderly patients with acute myeloid leukemia. *Leukemia*, **11**, 485–489

Ferguson, L.R. (1998) Inhibitors of topoisomerase II enzymes: A unique group of environmental mutagens and carcinogens. *Mutat. Res.*, **400**, 271–278

Ferguson, L.R. & Baguley, B.C. (1994) Topoisomerase II enzymes and mutagenicity. *Environ. mol. Mutag.*, **24**, 245–267

Ferguson, L.R. & Baguley, B.C. (1996) Mutagenicity of anticancer drugs that inhibit topoisomerase enzymes. *Mutat. Res.*, **355**, 91–101

Ferguson, L.R. & Denny, W.A. (1990) Frameshift mutagenesis by acridines and other reversibly-binding DNA ligands. *Mutagenesis*, **5**, 529–540

Frei, H., Clements, J., Howe, D. & Würgler, F.E. (1992) The genotoxicity of the anti-cancer drug mitoxantrone in somatic and germ cells of *Drosophila melanogaster*. *Mutat. Res.*, **279**, 21–33

Gennaro, A.R. (1995) *Remington: The Science and Practice of Pharmacy*, 19th Ed., Easton, PA, Mack Publishing, Vol. II, p. 1257

Gonsette, R.E. (1996) Mitoxantrone immunotherapy in multiple sclerosis. *Multiple Sclerosis*, **1**, 329–332

Gregory, R.K., Powles, T.J., Chang, J.C., & Ashley, S. (1997) A randomized trial of six versus twelve courses of chemotherapy in metastatic carcinoma of the breast. *Eur. J. Cancer*, **33**, 2194–2197

Greidanus, J., de Vries, E.G.E., Mulder, N.H., Sleijfer, D.T., Uges, D.R.A., Oosterhuis, B. & Willemse, P.H.B. (1989) A phase I pharmacokinetic study of 21-day continuous infusion mitoxantrone. *J. clin. Oncol.*, **7**, 790–797

Heinemann, V., Murray, D., Walters, R., Meyn, R.E. & Plunkett, W. (1988) Mitoxantrone-induced DNA damage in leukemia cells is enhanced by treatment with high-dose arabinosylcytosine. *Cancer Chemother. Pharmacol.*, **22**, 205–210

Henderson, B.M., Dougherty, W.J., James, V.C., Tilley, L.P. & Noble, J.F. (1982) Safety assessment of a new anticancer compound, mitoxantrone, in beagle dogs: Comparison with doxorubicin. I. Clinical observations. *Cancer Treat. Rep.*, **66**, 1139–1143

Henderson, I.C., Allegra, J.C., Woodcock, T., Wolff, S., Bryan, S., Cartwright, K., Dukart, G. & Henry, D. (1989) Randomized clinical trial comparing mitoxantrone with doxorubicin in previously treated patients with metastatic breast cancer. *J. clin. Oncol.*, **7**, 560–571

Ho, A.D., Seither, E., Ma, D.D.F. & Prentice, H.G. (1987) Mitozantrone-induced toxicity and DNA strand breaks in leukaemic cells. *Br. J. Haematol.*, **65**, 51–55

Ho, C.-K., Law, S.-L., Chiang, H., Hsu, M.-L., Wang, C.-C. & Wang, S.-Y. (1991) Inhibition of microtubule assembly is a possible mechanism of action of mitoxantrone. *Biochem. biophys. Res. Commun.*, **180**, 118–123

Hu, O.Y.-P., Chang, S.-P., Law, C.-K., Jian, J.-M. & Chen, K.-Y. (1992) Pharmacokinetic and pharmacodynamic studies with mitoxantrone in the treatment of patients with nasopharyngeal carcinoma. *Cancer*, **69**, 847–853

Hulhoven, R., Dumont, E. & Harvengt, C. (1983) Acute cardiovascular and renal changes induced by mitoxantrone in rabbits. A pharmacokinetic approach. *Toxicol. Lett.*, **18**, 19–26

Izumi, T., Ohtsuki, T., Ohya, K.-I., Ogawa, Y., Yoshida, M., Muroi, K., Imagawa, S., Hatake, K., Kuriki, K., Saito, K. & Miura, Y. (1996) Therapy-related leukemia with a novel 21q22 rearrangement. *Cancer Genet. Cytogenet.*, **90**, 45–48

Jackson, M.A., Stack, H.F. & Waters, M.D. (1996) Genetic activity profiles of anticancer drugs. *Mutat. Res.*, **355**, 171–208

James, V., Chiccarelli, F., Dougherty, W., Hall, C., Henderson, B., Iatropoulos, M., Morrison, J., Nicolau, G., Noble, J., Sparano, B., Gordon, G. & Wu, H. (1983) Preclinical toxicity studies on mitoxantrone and bisantrene. In*:* Rozencweig, M., Von Hoff, D.D. & Staquet, M.J., eds, *New Anticancer Drugs: Mitoxantrone and Bisantrene*, New York, Raven Press, pp. 47–69

Kaplan, L. & Volberding, P.A. (1985) Failure (and danger) of mitozantrone in AIDS-related Kaposi's sarcoma (Letter to the Editor). *Lancet*, **ii**, 396

Knudson, A.G. (1992) Stem cell regulation, tissue ontogeny, and oncogenic events. *Semin. Cancer Biol.*, **3**, 99–106

Larson, R.A., Daly, K.M., Choi, K.E., Han, D.S. & Sinkule, J.A. (1987) A clinical and pharmacokinetic study of mitoxantrone in acute nonlymphocytic leukemia. *J. clin. Oncol*., **5**, 391–397

Leblanc, T., Hillion, J., Derré, J., Le Coniat, M., Baruchel, A., Daniel, M.-T. & Berger, R. (1994) Translocation t(11;11)(q13;q23) and HRX gene rearrangement associated with therapy-related leukemia in a child previously treated with VP16. *Leukemia*, **8**, 1646–1648

Liang, J.C., Bailey, N.M., Gabriel, G.J., Kattan, M.W., Wang, R.Y., Hagemeister, F.B., Cabanillas, F.F. & Fuller, L.M. (1993) A new chemotherapy regimen for treatment of Hodgkin's disease associated with minimal genotoxicity. *Leuk. Lymphoma*, **9**, 503–508

LINFO Läkemedelsinformation AB (1998) *FASS 1998 Läkemedel i Sverige*, Stockholm, p. 894

Llesuy, S.F. & Arnaiz, S.L. (1990) Hepatotoxicity of mitoxantrone and doxorubicin. *Toxicology*, **63**, 187–198

Lu, K., Savaraj, N. & Loo, T.L. (1984) Pharmacological disposition of 1,4-dihydroxy-5-8-bis[{2 [(2-hydroxyethyl)amino]ethyl}amino]-9,10-anthracenedione dihydrochloride in the dog. *Cancer Chemother. Pharmacol*., **13**, 63–66

Manandhar, M., Cheng, M., Iatropoulos, M.J. & Noble, J.F. (1986) Genetic toxicology profile of the new antineoplastic drug mitoxantrone in the mammalian test systems. *Arzneimittelforschung*, **36**, 1375–1379

Markman, M., Alberts, D., Rubin, S., Hakes, T., Lewis, J.L., Jr, Reichman, B., Jones, W., Curtin, J., Barakat, R., Brodar, F., Peng, Y.-M., Pennie, K., Almadrones, L. & Hoskins, W. (1993) Evidence for persistence of mitoxantrone within the peritoneal cavity following intraperitoneal delivery. *Gynecol. Oncol.*, **48**, 185–188

Matney, T.S., Nguyen, T.V., Connor, T.H., Dana, W.J. & Theiss, J.C. (1985) Genotoxic classification of anticancer drugs. *Teratog. Carcinog. Mutag.*, **5**, 319–328

Medeiros, M. das G. & Takahashi, C.S. (1994) Effects of treatment with mitoxantrone in combination with novobiocin, caffeine and ara-C on human lymphocytes in culture. *Mutat. Res*., **307**, 285–292

Melillo, L.M.A., Sajeva, M.R., Musto, P., Perla, G., Cascavilla, N., Minervi, M.M., D'Arena, G. & Carotenuto, M. (1997) Acute myeloid leukemia following 3M (mitoxantrone, mitomycin and methotrexate) chemotherapy for advanced breast cancer (Letter to the Editor). *Leukemia*, **11**, 2211–2213

Mewes, K., Blanz, J., Ehninger, G., Gebhardt, R. & Zeller, K.-P. (1993) Cytochrome P-450-induced cytotoxicity of mitoxantrone by formation of electrophilic intermediates. *Cancer Res*., **53**, 5135–5142

Millefiorini, E., Gasperini, C., Pozzilli, C., D'Andrea, F., Bastianello, S., Trojano, M., Morino, S., Morra, V.B., Bozzao, A., Cao, A., Bernini, M.L., Gambi, D. & Prencipe, M. (1997) Randomized placebo-controlled trial of mitoxantrone in relapsing–remitting multiple sclerosis: 24-month clinical and MRI outcome. *J. Neurol*., **244**, 153–159

Nagel, J.D., Varossieau, F.J., Dubbelman, R., ten Bokkel Huinink, W.W. & McVie, J.G. (1992) Clinical pharmacokinetics of mitoxantrone after intraperitoneal administration. *Cancer Chemother. Pharmacol*., **29**, 480–484

Nishio, A., DeFeo, F., Cheng, C.C. & Uyeki, E.M. (1982) Sister-chromatid exchange and chromosomal aberrations by DHAQ and related anthraquinone derivatives in Chinese hamster ovary cells. *Mutat. Res*., **101**, 77–86

Ogilvie, G.K., Moore, A.S., Obradovich, J.E., Elmslie, R.E., Vail, D.M., Straw, R.C., Salmon, M.D., Klein, M.K., Atwater, S.W. & Ciekot, P.E. (1993) Toxicoses and efficacy associated with administration of mitoxantrone to cats with malignant tumors. *J. Am. vet. Med. Assoc*., **202**, 1839–1844

Panousis, C., Kettle, A.J. & Phillips, D.R. (1995) Myeloperoxidase oxidizes mitoxantrone to metabolites which bind covalently to DNA and RNA. *Anticancer Drug Des*., **10**, 593–605

Parkin, D.M., Whelan, S.L., Ferlay, J., Raymond, L. & Young, J., eds (1997) *Cancer Incidence in Five Continents, Vol. VII* (IARC Scientific Publications No. 143), Lyon, IARC*Press*

Partridge, S.E. & Lowdell, C.P. (1999) Myelodysplastic syndrome as a complication of neo-adjuvant triple M chemotherapy and radiotherapy. *Clin. Oncol.*, **11**, 187–189

Pedersen-Bjergaard, J. & Philip, P. (1991) Balanced translocations involving chromosome bands 11q23 and 21q22 are highly characteristic of myelodysplasia and leukemia following therapy with cytostatic agents targeting at DNA-topoisomerase II (Letter to the Editor). *Blood*, **78**, 1147–1148

Philpott, N.J., Bevan, D.H. & Gordon-Smith, E.C. (1993) Secondary leukaemia after MMM combined modality therapy for breast carcinoma (Letter to the Editor). *Lancet*, **341**, 1289

Powles, T.J., Jones, A.L., Judson, I.R., Hardy, J.R. & Ashley, S.E. (1991) A randomized trial comparing combination chemotherapy using mitomycin C, mitozantrone and methotrexate (3M) with vincristine, anthracycline and cyclophosphamide (VAC) in advanced breast cancer. *Br. J. Cancer*, **64**, 406–410

Quesnel, B., Kantarjian, H., Pedersen Bjergaard, J., Brault, P., Estey, E., Lai, J.L., Tilly, H., Stoppa, A.M., Archimbaud, E., Harousseau, J.L., Bauters, F. & Fenaux, P. (1993) Therapy-related acute myeloid leukemia with t(8;21), inv(16), and t(8;16): A report on 25 cases and review of the literature. *J. clin. Oncol.*, **11**, 2370–2379

Rentsch, K.M., Horber, D.H., Schwendener, R.A., Wunderli-Allenspach, H. & Hänseler, E. (1997) Comparative pharmacokinetic and cytotoxic analysis of three different formulations of mitoxantrone in mice. *Br. J. Cancer*, **75**, 986–992

Rentsch, K.M., Schwendener, R.A., Pestalozzi, B.C., Sauter, C., Wunderli-Allenspach, H. & Hänseler, E. (1998) Pharmacokinetic studies of mitoxantrone and one of its metabolites in serum and urine in patients with advanced breast cancer. *Eur. J. clin. Pharmacol*., **54**, 83–89

Reynoso, E.E. & Huerta, F. (1994) Acute leukemia and pregnancy—Fatal fetal outcome after exposure to idarubicin during the second trimester. *Acta oncol*., **33**, 709–710

Richard, B., Launay-Iliadis, M.-C., Iliadis, A., Just-Landi, S., Blaise, D., Stoppa, A.-M., Viens, P., Gaspard, M.-H., Maraninchi, D., Cano, J.P. & Carcassone, Y. (1992) Pharmacokinetics of mitoxantrone in cancer patients treated by high-dose chemotherapy and autologous bone marrow transplantation. *Br. J. Cancer*, **65**, 399–404

Rosenberg, L.J. & Hittelman, W.N. (1983) Direct and indirect clastogenic activity of anthracenedione in Chinese hamster ovary cells. *Cancer Res*., **43**, 3270–3275

Rote Liste Sekretariat (1998) *Rote Liste 1998,* Frankfurt, Rote Liste Service GmbH, pp. 86-111–86-112

Royal Pharmaceutical Society of Great Britain (1999) *Martindale, The Extra Pharmacopoeia*, 13th Ed., London, The Pharmaceutical Press [MicroMedex Online: Health Care Series]

Russell, L.B., Hunsicker, P.R., Johnson, D.K. & Shelby, M.D. (1998) Unlike other chemicals, etoposide (a topoisomerase-II inhibitor) produces peak mutagenicity in primary spermatocytes of the mouse. *Mutat. Res.*, **400**, 279–286

Savaraj, N., Lu, K., Manuel, V. & Loo, T.L. (1982) Pharmacology of mitoxantrone in cancer patients. *Cancer Chemother. Pharmacol.*, **8**, 113–117

Schnittger, S., Wormann, B., Hiddemann, W. & Griesinger, F. (1998) Partial tandem duplications of the *MLL* gene are detectable in peripheral blood and bone marrow of nearly all healthy donors. *Blood*, **92**, 1728–1734

Senkal, M., Tonn, J.C., Schönmayr, R., Schachenmayr, W., Eickhoff, U., Kemen, M. & Kollig, E. (1997) Mitoxantrone-induced DNA strand breaks in cell-cultures of malignant human astrocytoma and glioblastoma tumors. *J. Neuro-oncol.*, **32**, 203–208

Shenkenberg, T.D. & Von Hoff, D.D. (1986) Possible mitoxantrone-induced amenorrhea. *Cancer Treat. Rep.*, **70**, 659–661

Shipp, N.G. & Dorr, R.T. (1991) Biochemical effects and toxicity of mitoxantrone in cultured heart cells. *Adv. exp. Med. Biol.*, **283**, 821–825

Shipp, N.G., Dorr, R.T., Alberts, D.S., Dawson, B.V. & Hendrix, M. (1993) Characterization of experimental mitoxantrone cardiotoxicity and its partial inhibition by ICRF-187 in cultured neonatal rat heart cells. *Cancer Res.*, **53**, 550–556

Smith, I.E. (1983) Mitoxantrone (novantrone): A review of experimental and early clinical studies. *Cancer Treat. Rev.*, **10**, 103–115

Smith, I.E. & Powles, T.J. (1993) MMM (mitomycin/mitoxantrone/methotrexate): An effective new regimen in the treatment of metastatic breast cancer. *Oncology*, **50** (Suppl. 1), 9–15

Smith, P.J., Morgan, S.A., Fox, M.E. & Watson, J.V. (1990) Mitoxantrone–DNA binding and the induction of topoisomerase II associated DNA damage in multi-drug resistant small cell lung cancer cells. *Biochem. Pharmacol.*, **40**, 2069–2078

Smyth, J.F., Macpherson, J.S., Warrington, P.S., Leonard, R.C.F. & Wolf, C.R. (1986) The clinical pharmacology of mitozantrone. *Cancer Chemother. Pharmacol.*, **17**, 149–152

Stein, R.C., Bower, M., Law, M., Bliss, J.M., Barton, C., Gaset, J.-C., Ford, H.T. & Coombes, R.C. (1992) Mitozantrone and methotrexate chemotherapy with and without mytomycin C in the treatment of advanced breast cancer: A randomized clinical trial. *Eur. J. Cancer*, **28A**, 1963–1965

Štetina, R. & Veselá, D. (1991) The influence of DNA-topoisomerase II inhibitors novobiocin and fostriecin on the induction and repair of DNA damage in Chinese hamster ovary (CHO) cells treated with mitoxantrone. *Neoplasma*, **38**, 109–117

Stewart, D.J., Green, R.M., Mikhael, N.Z., Montpetit, V., Thibault, M. & Maroun, J.A. (1986) Human autopsy tissue concentrations of mitoxantrone. *Cancer Treat. Rep.*, **70**, 1255–1261

Sumner, A.T. (1995) Inhibitors of topoisomerase II delay progress through mitosis and induce a doubling of the DNA content in CHO cells. *Exp. Cell Res.*, **217**, 440–447

Sundman-Engberg, B., Tidefelt, U., Gruber, A. & Paul, C. (1993) Intracellular concentrations of mitoxantrone in leukemic cells *in vitro* vs *in vivo*. *Leuk. Res.*, **17**, 347–352

Suzuki, H. & Nakane, S. (1994) Differential induction of chromosomal aberrations by topoisomerase inhibitors in cultured Chinese hamster cells. *Biol. pharm. Bull.*, **17**, 222–226

Swiss Pharmaceutical Society, ed. (1999) *Index Nominum, International Drug Directory*, 16th Ed., Stuttgart, Medpharm Scientific Publishers [MicroMedex Online: Health Care Series]

Thomas, J., ed. (1998) *Australian Prescription Products Guide*, 27th Ed., Victoria, Australian Pharmaceutical Publishing, Vol. 1, pp. 2060–2063

US Pharmacopeial Convention (1994) *The 1995 US Pharmacopeia*, 23rd Rev./*The National Formulary*, 18th Rev., Rockville, MD, pp. 1034–1036

US Pharmacopeial Convention (1998a) *Drug Information for the Health Care Professional, USP Dispensing Information*, 18th Ed., Rockville, MD, Vol. I, pp. 2050–2053

US Pharmacopeial Convention (1998b) *The 1995 US Pharmacopeia*, 23rd Rev./*The National Formulary*, 18th Rev., Supplement 8, Rockville, MD, pp. 4230–4232

Van Belle, S.J.P., de Planque, M.M., Smith, I.E., van Oosterom, A.T., Schoemaker, T.J., Deneve, W. & McVie, J.G. (1986) Pharmacokinetics of mitoxantrone in humans following single-agent infusion or intra-arterial injection therapy or combined-agent infusion therapy. *Cancer Chemother. Pharmacol.*, **18**, 27–32

Vicari, A.M., Ciceri, F., Folli, F., Lanzi, R., Colombo, B., Comi, G. & Camba, L. (1998) Acute promyelocytic leukemia following mitoxantrone as single agent for the treatment of multiple sclerosis (Letter to the Editor). *Leukemia*, **12**, 441–442

Zee-Cheng, R.K.-Y. & Cheng, C.C. (1978) Antineoplastic agents. Structure–activity relationship study of bis(substituted aminoalkylamino)anthraquinones. *J. med. Chem.*, **21**, 291–294