DRUG DISPOSITION

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Clinical Pharmacokinetics of Cytarabine Formulations

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Contents

Abstract

Cytarabine (cytosine arabinoside, Ara-C) is an effective chemotherapeutic agent for the treatment of acute myelogenous leukaemia and lymphocytic leukaemias. As cytarabine is an S-phase-specific drug, prolonged exposure of cells to cytotoxic concentrations is critical to achieve maximum cytotoxic activity. However, the activity of cytarabine is decreased by its rapid deamination to the biologically inactive metabolite uracil arabinoside. This rapid deamination is the reason for the ongoing search for effective formulations and derivatives of cytarabine that cannot be deaminated and exhibit better pharmacokinetic parameters.

Protection of cytarabine from fast degradation and elimination has been investigated by encapsulating the drug into pharmaceutically acceptable carriers. Cytarabine derivatives have shown promise *in vitro* and in animal models. For example, ancitabine (cyclocytidine), enocitabine and cytarabine ocfosfate have been used clinically in Japan. Cytarabine encapsulated into multivesicular liposomes has been approved in several countries for the intrathecal treatment of lymphomatous meningitis.

Although many compounds have been investigated, few cytarabine derivatives are currently available for clinical use. Further research is needed to improve the efficacy of cytarabine against haematological and solid tumours.

Cytarabine (1-β-D-arabinofuranosylcytosine, cytosine arabinoside, Ara-C; figure 1) is an effective chemotherapeutic agent for the treatment of acute myelogenous leukaemia and lymphocytic leukaemia.[1-3] It has often been utilised in combination chemotherapy, for example in combination with fluorouracil, methotrexate and dexamethasone, against solid tumours and leukaemias. Cytarabine is a nucleoside analogue of deoxycytidine in which the ribose sugar has been replaced with an arabinose sugar. Cytarabine has the same molecular weight as cytidine and differs in that the 2′ hydroxyl group is oriented in the *trans* position, resulting in metabolism similar to that of a deoxyribose sugar.[4] Cytarabine penetrates cells by a carrier-mediated process that is shared by other nucleosides.[5,6] As cytarabine is a cell cycle-dependent drug, prolonged exposure of cells to cytotoxic concentrations is critical to achieve maximum cytotoxic activity. *In vitro* studies suggest that maximum cytotoxic activity is achieved with administration of cytarabine to reach concentrations ≥0.1 mg/L that are maintained for at least 24 hours.[7]

Cytarabine must be phosphorylated intracellularly to a nucleotide (cytarabine 5′-triphosphate, Ara-CTP) before it can exert its cytotoxic effect (figure 2). Accumulation of cytarabine 5′-triphosphate appears to be saturated at plasma concentrations of cytarabine exceeding 8 to 10 μmol/L.[8] Cytarabine 5′-triphosphate inhibits DNA synthesis by inhibiting DNA polymerases and terminating DNA chain elongation.[9] Although the precise mechanism of cell death caused by cytarabine 5′-triphosphate is not completely understood, it is clear that both concentration and duration of exposure are critical for cytotoxicity.

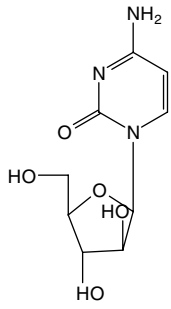
In clinical use, however, the activity of cytarabine is decreased by its rapid deamination by cytidine deaminase to a biologically inactive metabolite, 1-β-D-arabinofuranosyluracil (uracil arabinoside, Ara-U), and rapid elimination (figure 2).[8] In the systemic circulation, cytarabine is rapidly catabolised to uracil arabinoside, which is subsequently eliminated in the urine. Systemic elimination of cytarabine is biphasic, with an initial plasma half-life ($t_{½\alpha}$) of 7 to 20 minutes and a terminal half-life ($t_{½\beta}$) of 2 to 3 hours.[8]

In practical use in clinical chemotherapy, repetitive administration schedules or continuous intravenous infusion are considered to be essential. Typically, cytarabine is administered continuously for 5 days as a conventional standard dose schedule of 100 mg/m^2[10] or as a high dose regimen of 1 to 3 g/m^2.[11,12]

In contrast, following intrathecal administration of cytarabine, there is minimal conversion to uracil arabinoside in the CNS because of the low activity of cytidine deaminase in the brain and CSF. Elimination of cytarabine from the CSF is similar to CSF bulk flow (0.42 ml/min), and the $t_{½\beta}$ in the CSF is 3 to 4 hours.[13]

1. Approaches to Improving the Pharmacokinetics of Cytarabine

Cytarabine is poorly retained in blood and tissues, prompting the search for effective formulations and derivatives of cytarabine that cannot be deaminated and exhibit better pharmacokinetic parameters. Since the antitumour effect of cytarabine has been correlated with longer half-life, inhibition of cytidine deaminase activity might enhance the cytotoxic activity of cytarabine. Tetrahydrouridine (THU), a potent inhibitor of cytidine deaminase, has been shown to increase the antitumour activity of cytarabine both *in vivo* and *in vitro*.[14,15] Kreis et al.[15] treated patients with solid tumours with THU at 10 to 350 mg/m^2 with infusions of cytarabine at 25 to 100 mg/m^2. When cytarabine doses of 50, 75 and 100 mg/m^2 were coadministered with THU 250 or 350 mg/m^2, plasma cytarabine exposure [peak concentration (C_{max}) and area under



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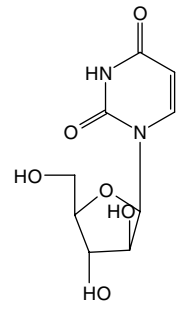

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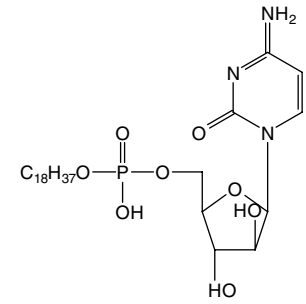

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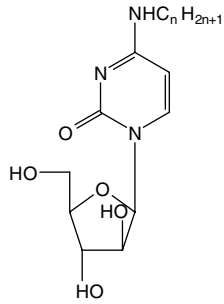

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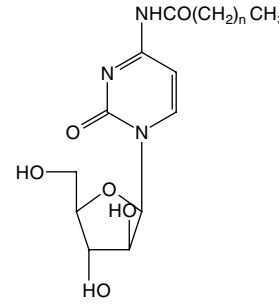

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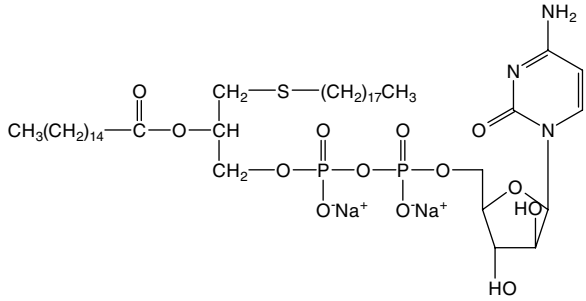

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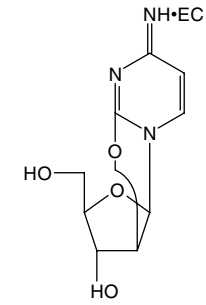

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Fig. 1. Chemical structures of cytarabine and its derivatives. **1** = cytarabine (1-β-D-arabinofuranosylcytosine, Ara-C); **2** = uracil arabinoside (1-β-D-arabinofuranosyluracil, Ara-U); **3** = cytarabine ocfosfate (1-β-D-arabinofuranosylcytosine-5′-stearylphosphate); **4** = NHAC (N^4-hexadecyl-1-β-D-arabinofuranosylcytosine, n = 16) and NOAC (N^4-octadecyl-1-β-D-arabinofuranosylcytosine, n = 18); **5** = PL-AC (N^4-palmitoyl-1-β-D-arabinofuranosylcytosine, n = 14) and enocitabine (N^4-behenoyl-1-β-D-arabinofuranosylcytosine, n = 20); **6** = Ara-CDP-L-dipalmitin (1-β-D-arabinofuranosylcytosine-5′-diphosphate-L-1, 2-dipalmitin); **7** = ancitabine (2,2′-anhydro-1-β-D-arabinofuranosylcytosine).

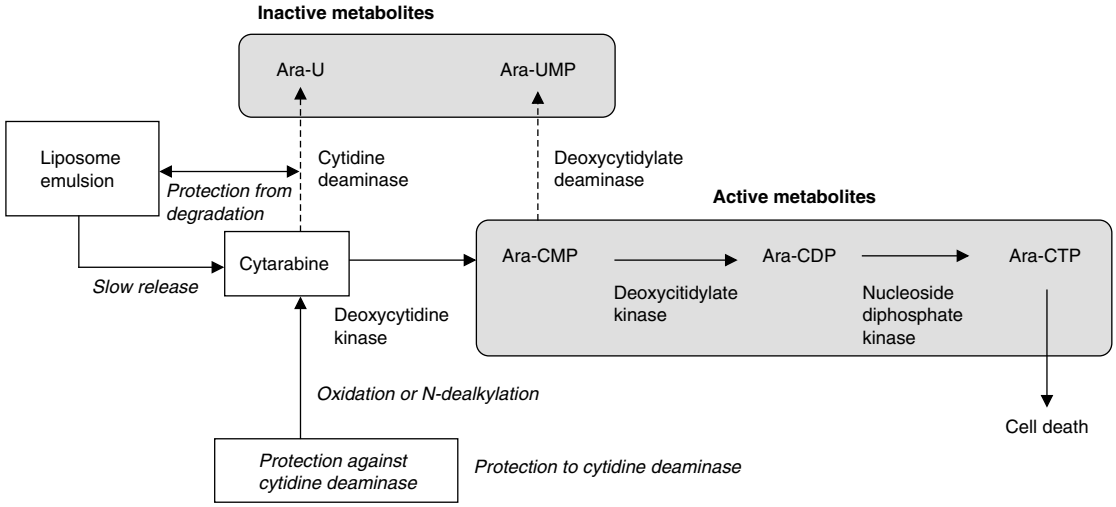

Fig. 2. Metabolic pathway of cytarabine and strategies for the development of sustained release formulations. **Ara-CDP** = cytarabine 5′-diphosphate; **Ara-CMP** = cytarabine 5′-monophosphate; **Ara-CTP** = cytarabine 5′-triphosphate; **Ara-U** = uracil arabinoside; **Ara-UMP** = uracil arabinoside 5′-monophosphate.

the concentration-time curve (AUC)] were significantly increased. In contrast, total body clearance and volume of distribution decreased significantly. This combination therapy provides plasma cytarabine concentrations (≥10 μmol/L) comparable to those after high dose cytarabine (1 g/m^2). The toxicity of this combination treatment was predominantly confined to bone marrow and gastrointestinal effects.

In other approaches, protection from rapid degradation and elimination was investigated by encapsulating the drug into pharmaceutically acceptable carriers, such as liposomes (figure 2 and table I).[16-20] However, significant protection from deamination and thus improvement of the antitumour activity of cytarabine could not be achieved, mainly due to leakage (initial burst) of cytarabine from liposomes, which is caused by the well-characterised instability of liposomes *in vivo*.[21] Modification of liposomes with polyethylene glycol (PEG)-containing lipids greatly enhances their stability in the circulation.[22] Allen et al.[23] demonstrated the cytotoxic activity of cytarabine entrapped in PEG-modified small unilamellar liposomes (termed 'stealth' liposomes) in mice. The resulting antitumour effect of cytarabine encapsulated within long-circulating PEG liposomes was superior to those of other liposome formulations or of the free drug. Kim et al.[24] also demonstrated prolongation of therapeutic cytarabine concentrations in cerebrospinal fluid (CSF) by encapsulation of the drug into multivesicular liposomes (DepoCyt™)**1** [see section 4].[25-27]

Improvement of the cytostatic efficacy of cytarabine can also be achieved by chemical modification (figure 1, table I). Although ancitabine (2, 2′- anhydro- 1-β-D-arabinofuranosylcytosine, cyclocytidine, cyclo-C) and enocitabine (N^4-behenoyl-1-β-D-arabinofuranosylcytosine, BH-AC) have been used clinically in Japan,[56-58,60,61] complicated parenteral procedures such as continuous infusion are still necessary. Hori et al.[54,55] reported that N^4-palmitoyl-1-β-D-arabinofuranosylcytosine (PL-AC), an analogue of cytarabine, exhibited marked activity against tumours when given orally. Kodama et al.[62,63] demonstrated the effi-

1 Use of tradenames is for product identification only and does not imply endorsement.

Table I. Summary of developmental status of cytarabine sustained-release formulations. A blank cell indicates no studies reported

Active agent	Formulation	Route	Animal studies	Clinical trials (phase)	Regulatory approval	References
Unmodified cytarabine						
Cytarabine	Emulsion	*In vitro* only				28-30
Cytarabine	Liposome	Intravenous	+			16-22,31,32
Cytarabine	Stealth liposome	Intravenous	+			23
Cytarabine	Polymer-coated liposome	Intravenous	+			33
Cytarabine	Liposome	Intravitreal	+			34
Cytarabine	Liposome	Intrathecal	+	I, II, III	Launch	25-27,35-39
Cytarabine derivatives						
NOAC, NHAC	Liposome	Intravenous	+			40-48
NOAC, NHAC	Liposome	Oral	+			49
Ocd-Grop-Ara-C	Liposome	Oral	+			50,51
Ara-CDP-L-dipalmitin	Liposome	Intravenous	+			31
Ara-CDP-DL-PTBA	Micelle	Intravenous	+			32,52,53
PL-AC	Suspension	Oral	+	I, II		54,55
Enocitabine	Lyophilisation	Intravenous	+	I, II, III	Launch	56-60
Ancitabine	Solution	Intravenous	+	I, II, III	Launch	61
Cytarabine ocfosfate	Capsule	Oral	+	I, II, III	Launch	62-70

Ara-CDP-DL-PTBA = 1-β-D-arabinofuranosylcytosine 5′-diphosphate-*rac*-1-*S*-octadecyl-2-*O*-palmitoyl-*1*-thioglycero*l;* **Ara-CDP-L-dipalmitin** = 1-β-D-arabinofuranosylcytosine-5′-diphosphate-L-1,2-dipalmitin; **NHAC** = N^4-hexadecyl-1-β-D-arabinofuranosylcytosine; **NOAC** = N^4-octadecyl-1-β-D-arabinofuranosylcytosine; **Ocd-Grop-Ara-C** = (1-octadecylglycero-3-phospho)-cytarabine; **PL-AC** = N^4-palmitoyl-1-β-D-arabinofuranosylcytosine; **+** indicates positive results.

cacy of cytarabine ocfosfate (1-β-D-arabinofuranosylcytosine-5′-stearylphosphate) against L1210 leukaemia in mice after oral administration.

A large number of other N^4-derivatives of cytarabine have been synthesised with the aim of increasing cytotoxic activity by protecting from deamination and altering pharmacokinetic properties.[71,72] Although short-chain modifications of cytarabine at the N^4-amino group generally result in weak enhancement of cytotoxicity, lipophilic derivatives with long-chain fatty acids showed strong antitumour activity in murine tumour models.[56,73,74] Rubas et al.[75] demonstrated that N^4-acyl derivatives incorporated into the membranes of small unilamellar liposomes were active against murine L1210 leukaemia and B16 cells. However, protection against enzymatic deamination was only partially achieved with the modification, and sufficient effect was not observed in a pilot phase I/II study.[76] To obtain a compound with higher stability, an N^4-alkyl cytarabine derivative, N^4-hexadecyl -1- β- D- arabinofuranosylcytosine (NHAC), was synthesised.[77] Because of the very low aqueous solubility of NHAC, this long-chain alkyl derivative was incorporated into lipid membranes of small unilamellar liposomes composed of soy phosphatidylcholine and cholesterol to allow its parenteral administration. These liposomal preparations exerted significantly higher cytotoxic activities in the L1210 leukaemia model at lower concentrations. Furthermore, the derivatives had a strong cytotoxic effect when administered as single doses, suggesting a long-lasting effect.

This article provides a review of the pharmacokinetics of the most promising sustained release formulations of cytarabine and its derivatives that have shown efficacy against blood and solid tumours.

2. Emulsions

Water-in-oil-in-water (w/o/w) double emulsions have been used to prolong drug release, to immobilise enzymes, and to treat overdosage. Hashida

et al.[78,79] stabilised a water-in-oil emulsion and a w/o/w double emulsion by gelling the inner aqueous phase, and Fukushima et al.[28] used the technique to prepare a stable w/o/w double emulsion containing cytarabine. Okochi and Nakano[29] showed that cytarabine release from an emulsion prepared by the membrane method was slower than that from an emulsion prepared by the stirring method. This membrane method was also found to be useful for preparation of w/o/w emulsions that are prepared by a two-step emulsification procedure.

The sustained release of cytarabine from w/o/w double emulsions was successful *in vitro*. Release was enhanced with increase either in the volume of the inner aqueous phase or in the concentration of the drug. Benoy et al.[30] demonstrated that the antitumour effect of cytarabine in mice could be enhanced by preparation of a w/o/w emulsion. However, w/o/w emulsions show poor stability (only 2 or 3 days), and clinical trials have not been carried out.

3. Liposomes

Liposomes are microscopic particles of lipid bilayer membrane that enclose an aqueous internal compartment. This approach offers great potential for improving cancer chemotherapy.[80] The lipid membranes are composed of amphipathic lipids identical to cell membranes. The encapsulated drugs partition between the aqueous internal chambers and lipid bilayers, hydrophilic molecules into the former and hydrophobic molecules into the latter. Encapsulation in liposomes modifies pharmacokinetics and tissue distribution, which may increase the therapeutic index of drugs.

Liposomal drugs such as amphotericin have proved significantly more effective than conventional drugs in therapy of certain parasitic and fungal diseases. Liposomal cytarabine has been shown to enhance significantly the survival time of mice bearing L1210 leukaemia.[16,17] This increased antitumour activity can be attributed to protection of cytarabine from rapid degradation in the blood and to its slow release from the liposomes.

3.1 Unmodified Cytarabine in Liposomes

Several investigators have demonstrated the efficacy of unmodified cytarabine in liposomes in animal model studies *in vivo*. Kobayashi et al.[16,17] showed that encapsulation of cytarabine into liposomes increased efficacy against tumours *in vivo*. In this study, cytarabine was encapsulated in four different types of liposomes to compare their antitumour effect on mouse leukaemia L1210 cells inoculated into CD2F mice with that of free cytarabine. The antitumour activity of cytarabine was markedly enhanced by encapsulation in the liposomes. Among the four types of liposomes, a physically stable and positively charged liposome consisting of sphingomyelin, stearylamine and cholesterol (20 : 2 : 15 molar ratio) most effectively enhanced the effect of cytarabine.

Hong and Mayhew[35] reported the therapeutic effects of liposomal cytarabine against intracranially inoculated L1210 leukaemia in mice. Intracranially administered liposomal cytarabine was more effective than intravenous liposomal cytarabine. A single intravenous dose of liposomal cytarabine was therapeutically superior to a 5-day intravenous infusion of free cytarabine. Intracranial or intravenous liposomal cytarabine at therapeutic doses resulted in less systemic toxicity than intravenous infusion of free cytarabine, suggesting the possible use of liposomal cytarabine as an adjunct to treatment of leukaemia cells in the CNS.

These observations indicated that liposomal cytarabine is protected from deamination in the liver, and is slowly released from liposomes in liver and spleen. Hunt et al.[81] showed in mice that the initial trapping of large multilamellar vesicles (MLV) during first passage in the lung was consistent with subsequent binding and retention. Release of cytarabine from large or small MLV in the lung is apparently slow relative to metabolism. Richardson et al.[82] showed that liposomal cytarabine was able to overcome drug resistance *in vitro*. Ganapathi et al.[83] showed that cytarabine could be encapsulated in anionic multilamellar liposomes prepared with different lecithin and cholesterol ratios, and that *in vivo* antitumour activity

in mice bearing L1210 leukaemia was influenced by the cholesterol content of the liposomal lipid bilayer. It was also found in vitrectomised and non-vitrectomised eyes that liposomal cytarabine decreased the cytotoxicity of cytarabine injected intravitreally.[34]

Delivery of liposomal cytarabine by direct injection into the CNS markedly increased its half-life[36] and efficacy[35] in animal models compared with unencapsulated cytarabine. Kim et al.[36] reported the use of multivesicular liposomes as a slow-release depot of cytarabine for intrathecal administration to rats. Following intrathecal administration of liposomal encapsulated drug, the intrathecal half-life of cytarabine was 148 hours, in contrast to 2.7 hours for unencapsulated drug. This extended half-life may be attributable to a sustained-release effect as well as protection from cytidine deaminase. The prolonged drug concentration may increase efficacy, and the limited C_{max} may decrease toxicity.

Prolonged therapeutic concentrations of cytarabine in human CSF are obtained after a single injection of liposomal cytarabine in multivesicular liposomes, and a high response rate was shown in patients with neoplastic meningitis.[25-27,37] This preparation is discussed further in section 4.

3.2 Polymer-Coated Liposomes

Sehgal and Rogers[33] reported that polymer coating improved liposome stability and the release profile of cytarabine. Liposomes of dimyristoylphosphatidylcholine or dipalmitoylphosphatidylcholine, cholesterol and dicetylphosphate containing cytarabine were prepared at pH 4.5 and subsequently coated with a chemically modified polysaccharide, *O*-palmitoylpullulan. Liposomes coated with *O*-palmitoylpullulan may increase the potential of usage of liposomes as a drug delivery system in harsh environments such as encountered after oral administration, although the magnitude of the effect was dependent on the liposome composition.

3.3 'Stealth' Liposomes

Encapsulation of cytarabine into 'stealth' liposomes increased its circulation time in an animal model. Allen et al.[23] reported that prolonged circulation time and dose-independent pharmacokinetics have been observed for cytarabine in mice bearing L1210 leukaemia.

3.4 Lipophilic Cytarabine Derivatives in Liposomes

Lipophilic derivatives of cytarabine have been shown to increase efficacy against animal tumours.[31,40-42,45-47,49-51,73,84-86] Koller-Lucae et al.[40] demonstrated in mice that N^4-octadecyl-1-β-D-arabinofuranosylcytosine (NOAC) behaved pharmacokinetically differently from cytarabine and suggested that its affinity for low-density lipoproteins might be advantageous in tumour therapy. N^4-Acyl derivatives of cytarabine, incorporated into the membranes of small unilamellar liposomes, were active against murine L1210 leukaemia and B16 melanoma at lower concentrations when compared with that of unmodified cytarabine. NOAC and NHAC show low aqueous solubility, so they can be incorporated into the lipid membranes of small unilamellar liposomes, allowing their parenteral administration. Liposomal preparations of NHAC and NOAC exerted significantly higher cytotoxic activities in murine L1210 leukaemia than cytarabine alone. Nishikawa et al.[86] reported a further comparison of the pharmacokinetic parameters of NOAC and cytarabine in mice, and revealed that NOAC is eliminated from the kidney and the liver, whereas cytarabine is excreted mainly in urine.

NHAC and NOAC are highly cytostatic when administered orally as liposome formulations.[49] Horber et al.[45-47] reported that the pharmacokinetic properties and interactions with blood components of NHAC incorporated into liposomes. When incubated with human blood, NHAC was transferred from the liposomes to plasma proteins, particularly albumin and high and low density lipoproteins, at about 47% efficiency. The remain-

ing part of NHAC was bound to erythrocytes (50%) and to leucocytes (3%). Compared with cytarabine, NHAC in liposome formulations was highly resistant to deamination, resulting in concentrations of uracil arabinoside 42 and 10 times lower in plasma and liver microsomes, respectively.

Schwendener et al.[50,51] reported the use of another lipophilic derivative, (1-octadecylglycero-3-phospho)-cytarabine, as a liposomal formulation in the treatment of mice bearing Lewis lung carcinoma. Rahman et al.[31] reported the activity of liposomal 1- β - D -arabinofuranosylcytosine -5′-diphosphate-L-1,2-dipalmitin (Ara-CDP-L-dipalmitin) when administered intravenously to mice bearing Lewis lung carcinoma. Ara-CDP-L-dipalmitin given as a single dose was more effective than the same dose divided into five injections; however, no cures have been obtained by treatment with Ara-CDP-L-dipalmitin alone.

4. Cytarabine Liposome

Intrathecal chemotherapy may be advantageous in the treatment of neoplastic meningitis.[87] The efficacy of systemic chemotherapeutic agents in the treatment of neoplastic meningitis is limited by the poor penetration of chemotherapeutic agents across the blood-brain barrier.[88] Direct intrathecal administration of cytarabine has become the standard treatment for neoplastic meningitis, but this approach is limited by the short half-life of cytarabine in the CSF.[89] Cytarabine encapsulated in multivesicular liposomes may offer a therapeutic advantage in the treatment of neoplastic meningitis. This formulation, DepoCyt™,[36,37] is an injectable suspension of cytarabine encapsulated into multivesicular lipid-based particles. DepoCyt™ consists of microscopic (3 to 30μm) spherical particles composed of numerous nonconcentric internal aqueous chambers containing cytarabine. Single intrathecal administration of DepoCyt™ maintains cytotoxic concentrations of cytarabine in the CSF for 2 weeks.

4.1 Preclinical Studies

In vivo studies using mice[36,90] revealed that, following either subcutaneous or intraperitoneal injection of DepoCyt™, the serum half-life of cytarabine is significantly prolonged when compared with that of unencapsulated cytarabine. Following subcutaneous administration of DepoCyt™, the serum half-life of cytarabine was 4 days compared with 10 minutes for unencapsulated cytarabine. The serum half-life after intraperitoneal administration of DepoCyt™ was 21 hours compared with 16 minutes for unencapsulated cytarabine. Unbound cytarabine concentrations in the CSF exceeded the minimal *in vitro* cytotoxic concentration of 0.1 mg/L for at least 14 days after intrathecal injection of DepoCyt™.[87]

A pharmacokinetic advantage of DepoCyt™ was also demonstrated following intrathecal administration.[24] Animals received a single injection of either DepoCyt™ 2mg or cytarabine 2mg as a control. The $t_{½\beta}$ of cytarabine was 156 hours following injection of DepoCyt™ compared with only 0.74 hour for cytarabine alone. A single intrathecal injection of DepoCyt™ can maintain a therapeutic drug concentration in the CSF for a prolonged period.

4.2 Clinical Pharmacokinetic Studies

In a dose-ranging pharmacokinetic study, patients with neoplastic meningitis received DepoCyt™ at doses ranging from 12.5 to 125mg via Ommaya reservoir or lumbar puncture. Following intrathecal administration of 50mg of DepoCyt™, the elimination of cytarabine from the ventricular CSF was biexponential, with $t_{½\alpha}$ of 5.5 hours and $t_{½\beta}$ of 80 hours. The $t_{½\beta}$ of cytarabine increased >20-fold following administration of DepoCyt™ compared with that of cytarabine.[26] The cytarabine concentrations in the ventricular CSF were maintained above the cytotoxic concentration of 0.1 mg/L for approximately 2 weeks after intraventricular administration of DepoCyt™ doses greater than 25mg.[25-27]

4.3 Efficacy and Adverse Effects

Some patients with positive CSF cytologies achieved a complete cytological response with DepoCyt™ therapy.[89,90] The median duration of complete cytological response was 111 days. A randomised trial compared the safety and efficacy of intrathecal DepoCyt™ 50mg once every 2 weeks with intrathecal free cytarabine 5mg twice weekly in 28 patients with neoplastic meningitis resulting from solid tumours.[38] Of the patients treated with intrathecal DepoCyt™, 71% had a complete response compared with 15% of patients treated with intrathecal cytarabine. Time to neurological progression (median 78.5 *vs* 42 days) and median survival (99.5 *vs* 63 days) tended to be improved in patients treated with DepoCyt™ compared with unencapsulated cytarabine.[39]

The efficacy of intrathecal DepoCyt™ versus methotrexate was investigated in patients with neoplastic meningitis secondary to solid tumours.[39] During the induction phase, patients received either DepoCyt™ (50mg once every 2 weeks) or methotrexate (10mg twice weekly). During the consolidation phase, DepoCyt™ was administered every 2 weeks for 1 month and then every 4 weeks for 2 months, whereas methotrexate was given every week for 1 month, and then every 2 weeks for 2 months. Complete responses were seen in 26% of DepoCyt™ recipients compared with 20% of methotrexate recipients. The median time to clinical progression was 58 days for patients receiving DepoCyt™ compared with 30 days for those receiving methotrexate.

Drug-induced chemical arachnoiditis, such as fever, headache, back and/or neck pain and nausea or vomiting was common in patients receiving DepoCyt™.[25] These adverse events were temporary and were manageable with dexamethasone 2 to 4mg twice daily for 5 days, starting on the day of DepoCyt™ administration.[39]

5. Cytarabine Derivatives

The usefulness of cytarabine is limited by its rapid deamination to the biologically inactive metabolite uracil arabinoside. The relatively high concentrations of 2′-deoxycytidine deaminase activity have been suggested as one of the reasons why cytarabine is not effective against solid tumours.[9] To protect the drug from deamination, a large number of chemical modifications of cytarabine have been tested.[54-58,60-63,71-74] Among these, ancitabine and enocitabine have been used clinically in Japan (see table I).[56,57,61]

In animal studies, Hori et al.[54,55] reported that PL-AC exhibited marked activity against tumours when given orally. After a single oral dose of a suspension of PL-AC at a therapeutic dose of 350 μmol/kg, a high concentration of the drug was found in the liver, lung and plasma of portal venous blood. The concentration of the drug in other tissues and peripheral plasma was rather low. The two main metabolites, cytarabine and uracil arabinoside, were found in plasma and various tissues. The plasma cytarabine concentration was maintained for at least 6 hours in the range of 2.3 to 5.1 μmol/L after oral administration of PL-AC 350 μmol/kg. On the other hand, when an equimolar amount of cytarabine was given, the plasma concentrations decreased rapidly; from 2 to 6 hours after administration, the concentration (1.0 to 4.1 μmol/L) was less than that obtained with PL-AC. These results suggest that PL-AC administered orally is absorbed intact from the gastrointestinal tract and that the absorbed compound is a depot form of cytarabine, releasing cytarabine over a prolonged period of time.

Conjugates of cytarabine with polysaccharides such as polygalacturonic acid and carboxymethylated yeast β-D-glucan have been tested for their antileukaemic activity.[48] 1-β-D-Arabinofuranosylcytosine 5′-diphosphate- *rac*-1-*S*-octadecyl-2-*O*-palmitoyl-1-thioglycerol (Ara-CDP-DL-PTBA) is stable cytarabine conjugate of thioether phospholipid that is effective against a variety of transplantable tumours in mice.[32,52,53] The conjugate is formulated in a micellar solution by sonication, in which the conjugate exists as micelles. Ara-CDP-DL-PTBA is active in the treatment of human

colorectal cancer xenografts at a non-toxic dose level.[53]

5.1 Cytarabine Ocfosfate

Cytarabine ocfosfate was designed and synthesised as a prodrug of cytarabine 5′-monophosphate by Kodama et al.[62] and Saneyoshi et al.,[63] demonstrated its activity against L1210 leukaemia in mice after oral administration. As a result of the introduction of a long-chain fatty acyl group at the 5′-position of cytarabine 5′-monophosphate, the drug shows higher lipophilicity. Koga et al.[65] suggested that cytarabine ocfosfate may be useful for induction and/or postoperative chemotherapy on the basis of studies with colorectal adenocarcinoma xenografts in nude mice.

Ueda et al.[66] reported a pharmacokinetic study of oral administration of cytarabine ocfosfate in six patients with haematological malignancies. After a single oral dose of cytarabine ocfosfate 500 mg/m^2, cytarabine ocfosfate, cytarabine and uracil arabinoside were detected in the plasma for 72 hours (table II). The plasma concentration-time curve of cytarabine ocfosfate could be fitted to a one-compartment model. The C_{max} of cytarabine ocfosfate was 322 nmol/L, and the time to reach C_{max} (t_{max}) was 6.5 hours; the half-life was 32.0 hours. The plasma concentration of cytarabine increased slowly to a C_{max} of 26.3 nmol/L (t_{max} 13.3 hours) after administration of cytarabine ocfosfate. For uracil arabinoside, C_{max}, t_{max} and terminal half-life were 483 nmol/L, 23.6 hours and 19.6 hours, respectively. No cytarabine ocfosfate was detected in the urine, and only 8% of the administered dose was excreted in the urine as cytarabine and uracil arabinoside within 72 hours. During clinical use of cytarabine ocfosfate, macrocytic anaemia was observed, and some patients also developed megaloblastic change of their erythroblasts, suggesting a mild and persistent cytostatic effect. Cytarabine ocfosfate has been available in Japan from 1992, and phase I and phase II trials are now also under way in Germany, including patients with relapsed or refractory acute myeloid leukaemia or low-grade non-Hodgkin's lymphoma.[67-70]

Table II here.

Table II. Pharmacokinetic parameters of cytarabine derivatives in humans

Derivatives	Dose	Route	Metabolite	AUC (μg • h/L)	C_{max} (μg/L)	t_{max} (h)	$t_{½}$ (h)	Reference
Cytarabine ocfosfate	500 mg/m^2	Oral	Cytarabine ocfosfate	15 100	322	6.5	32	66
			Cytarabine	1730	26.3	13.3	31.9	
			Ara-U	32 100	483	23.6	19.6	
Enocitabine	700 mg/m^2	Intravenous	Enocitabine	429 800	173 400	1	1 (α), 4.28 (β)	59
			Cytarabine	858	102	1	1.37 (α), 11.2 (β)	
			Ara-U	62 620	3500	7		
	200mg	Intravenous	Enocitabine	85 500	20 000	2	0.37 (α), 5.27 (β)	58
			Cytarabine		80	2		
			Ara-U		600	4		

Ara-U = uracil arabinoside; **AUC** = area under the plasma concentration-time curve; $\mathbf{C_{max}}$ = peak plasma concentration; $\mathbf{t_{max}}$ = predicted time of C_{max}; $\mathbf{t_{½}}$ = elimination half-life; $\mathbf{t_{½α}}$ = initial-phase half-life; $\mathbf{t_{½β}}$ = second-phase half-life.

5.2 Enocitabine

Enocitabine is resistant to deamination because it bears a highly lipophilic group at the 4-amino position of the cytosine moiety of cytarabine.[56,57] Ueda et al.[58] reported the pharmacokinetics of enocitabine in seven patients with acute leukaemia given enocitabine 200mg as a 90-minute intravenous infusion. The plasma disappearance curve of enocitabine was biphasic with $t_{½\alpha}$ and $t_{½\beta}$ of 0.37 and 5.27 hours, respectively (table II). The plasma concentration of uracil arabinoside increased to 603 μg/L at 4 hours after infusion, and it was over 129 μg/L for at least 22.5 hours after infusion. The plasma concentration of cytarabine was maintained over 0.08 mg/L for 8 hours after infusion.

Yoshida et al.[59] reported the pharmacokinetics of enocitabine after a 60-minute intravenous infusion of 700 mg/m^2 in patients with acute leukaemia. The plasma concentration of enocitabine reached a maximum (173.4 mg/L) at the end of the infusion and then declined in a biphasic pattern with $t_{½\alpha}$ of 1 hour and $t_{½\beta}$ of 4.28 hours. The plasma concentration of cytarabine similarly reached a maximum (102.2 μg/L) at the end of the infusion and then declined with $t_{½\alpha}$ of 1.37 hours and $t_{½\beta}$ of 11.2 hours.

5.3 Alkyl Derivatives

Structure-activity relationships among alkyl cytarabine derivatives have been reported.[91] Alkyl cytarabine analogues are less susceptible to hydrolysis and therefore more stable than the acyl analogues.[43] NOAC is poorly deaminated to uracil arabinoside after incubation with human serum or mouse liver microsomes.[44] The long-chain alkyl derivatives of cytarabine are highly lipophilic without amphiphilic properties and therefore exert no haemolytic toxicity in mice.[40]

Pharmacological studies with NHAC and NOAC suggest different mechanisms of action from that of cytarabine. Cell uptake may be independent of nucleoside transporters. The formation of cytarabine 5′-triphosphate from NHAC is very low and will contribute little to the cytotoxicity of NHAC, whereas apoptosis of tumour cells was induced by NHAC at a rate 20 times higher than that by cytarabine.[45-47] In human tumour xenografts in nude mice, NOAC showed a significantly better effect than cytarabine in various leukaemias, and an impressive antitumour activity against solid tumours.[44]

6. Conclusion

Many studies have demonstrated the potential of new cytarabine formulations, including liposomes, emulsions and derivatives. To develop new formulations for sustained release and improved drug delivery, studies are required in humans. Some of these drugs are currently in clinical trials, and have been shown to improve delivery to target tumours, reduce toxicity and allow convenient administration. It is highly probable that medically useful and commercially viable cytarabine derivatives in addition to DepoCyt™ will be available within a few years.

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