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Future Directions for Clinical Research With CPT-11 (Irinotecan)

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CPT-11 is a new agent with a unique mechanism of action, namely the inhibition of topoisomerase I. An examination of data from the laboratory reveals several leads which should be pursued in the clinic. A dose–response effect for CPT-11 activity has been noted in the human tumour cloning assay. CPT-11 has activity against breast and mesothelioma colony-forming units in a human tumour cloning assay, and has *in vivo* activity against a number of paediatric malignancies. Promising combinations in preclinical *in vivo* models include CPT-11/mitomycin C and CPT-11/cytosine arabinoside. There is incomplete cross-resistance among topoisomerase I inhibitors, suggesting that combinations of topoisomerase I inhibitors should be investigated. Several natural products have been identified which have potential to decrease CPT-11-induced diarrhoea. The level of carboxylesterase in a patient's tumour appears to be related to the *in vitro* activity of CPT-11, suggesting that measurement of carboxylesterase in a patient's tumour could be used to identify patients who are most likely to respond to treatment with CPT-11. These preclinical findings suggest substantial further clinical potential for CPT-11 in terms of decreased CPT-11-induced diarrhoea as well as increased antitumour activity, which should be explored in phase I and II studies. Copyright © 1996 Published by Elsevier Science Ltd

Key words: CPT-11, topoisomerase I, clinical trials, phase I, phase II, human tumour cloning assay, preclinical

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INTRODUCTION

CPT-11 is a promising new anticancer agent with a unique mechanism of action, namely inhibition of topoisomerase I [1, 2]. Levels of topoisomerase I are 14- to 16-fold higher in colon tumours than in normal colon mucosae [3], and this has led clinicians to investigate the suggestion that inhibitors of topoisomerase I may be of particular value in the treatment of patients with colon cancer. Indeed, following encouraging observations of substantial activity in colon cancer cell lines, clinical studies of CPT-11 monotherapy in both chemotherapy-naive and pretreated patients have produced response rates at least equivalent to those achieved with 5-fluorouracil (5-FU)/folinic acid combinations [4].

Our group in San Antonio has had extensive experience with clinical trials of a variety of topoisomerase I inhibitors and administration schedules, as listed in Table 1. Together with the results of preclinical investigations of the different topoisomerase I inhibitors, our clinical findings provide evidence that each of these inhibitors has a unique profile of pharmacological properties which result in activity in different clinical situations and against different types of tumours.

This review summarises some of the preclinical experience with one of the inhibitors of topoisomerase I, namely, CPT-11. The experimental observations described here provide leads for future clinical trials with CPT-11 which should be pursued.

NEW LEADS FOR PHASE I TRIALS WITH CPT-11

Monotherapy

Our group has extensively investigated the effects of CPT-11 against human tumour colony-forming units growing in soft agar [5]. Results of these studies indicate that there is a good concentration–response effect for CPT-11 in this human tumour cloning system (Table 2). These data suggest that if one can achieve higher concentrations of CPT-11 in patients, a higher level of antitumour activity than that already noted with CPT-11 in clinical trials should be achievable. Substantial efforts should, therefore, be made in the phase I setting, to escalate the dose of the drug.

To date, dose-limiting toxicities of CPT-11 have included neutropenia (slightly more prevalent with CPT-11 given as

Table 1. San Antonio experience with topoisomerase I inhibitors

Inhibitor	Schedule	Route
Topotecan	Single dose	i.v.
	120 h CI	i.v.
	72 h CI	i.v.
	Single dose	p.o.
	Daily × 21	p.o.
	Daily × 10	p.o.
	Daily × 5	p.o.
CPT-11	Weekly × 4 every 6 weeks	i.v.
	Every other week	i.v.
	Daily × 5	p.o.
Intoplicine	Single dose	i.v.
GT 147211	Daily × 5	i.v.

CI, continuous infusion; i.v., intravenous; p.o., oral.

a single dose every 3 weeks, compared with other schedules) and diarrhoea (slightly more prevalent with the weekly schedule of administration) [4, 7, 19]. Neutropenia is clearly a toxicity which can be dealt with by the use of colony-stimulating factors. However, the increasing incidence of diarrhoea with escalating doses of CPT-11 is a challenge to the medical oncologist. At a 'standard' dosage of CPT-11 (350 mg/m^2 as a 30-min infusion repeated every 3 weeks), the incidence of delayed diarrhoea of any grade is 87% [6].

Abigerges and colleagues [7] performed an important phase I trial in which CPT-11 was escalated to doses of 400–600 mg/m^2 as a 30-min infusion repeated every 3 weeks. Loperamide was initiated at a dosage of 2 mg every 2 h for any episode of diarrhoea noted 8 h after CPT-11, and was stopped only after a 12-h diarrhoea-free period. Of the 23 patients treated, 17 (74%) experienced delayed diarrhoea of any grade. However, only one patient was hospitalised. The lead provided by this promising trial should be pursued.

In addition to the use of loperamide to allow escalation of the dose of CPT-11, there is a suggestion by Sakata and colleagues that TJ-14, a Kampo (Chinese herb medicine) might also be helpful in reducing CPT-11-induced diarrhoea [8]. Of 23 patients treated with CPT-11 plus TJ-14, 9 patients experienced no diarrhoea and 9 patients had only grade 1 (ECOG scale) diarrhoea. 4 patients discontinued treatment with TJ-14 because they could not tolerate the odour or taste of the material [8].

The active component of TJ-14 is baicalin, an inhibitor of β-glucuronidase. The mechanism by which baicalin ameliorates CPT-11-induced diarrhoea is thought to be related to the excretion of SN-38, an active metabolite of CPT-11 which is generally accepted to be largely responsible for the clinical cytotoxic properties of CPT-11. Following conversion from CPT-11 by the enzyme carboxylesterase, cytotoxic SN-38 undergoes glucuronidation in the liver to form a non-cytotoxic derivative. The resulting SN-38 glucuronide is excreted in the bile and is transformed back into cytotoxic SN-38 in the gut by β-glucuronidase made by intestinal bacteria. As a result of enterohepatic circulation, the unconjugated SN-38 is taken up again (possibly in the terminal ileum) and re-excreted into the bile [9]. Inhibition of β-glucuronidase in the gut by baicalin could prevent breakdown of SN-38 glucuronide into SN-38 and thus prevent or ameliorate CPT-11-induced diarrhoea by decreasing exposure of the gastrointestinal mucosa to cytotoxic SN-38.

Other natural product inhibitors of β-glucuronidase, including wogonoside, luteolin-3′-glucuronide and glycyrrhizin, have been identified [8]. The use of these and TJ-14 should be investigated to attempt to escalate the dose of CPT-11 in future phase I trials with the agent.

Table 2. Dose–response effect for CPT-11 in the human tumour cloning assay

Concentration of CPT-11 (μg/ml)	Number of responses in evaluable specimens*
0.3	8/124 (6%)
1.5	20/135 (15%)
3.0	41/125 (33%)

*Response = ≤50% survival of tumour colony-forming units.

CPT-11-based combinations

In the field of colorectal cancer, much interest has been generated recently in the combination of CPT-11 with 5-fluorouracil (5-FU), the most active agent currently available for the treatment of this condition. The international experience with various schedules of this combination (with or without folinic acid) has been reviewed by Saltz and colleagues [10]. In addition, clinical trials are ongoing with combinations of CPT-11/cisplatin, CPT-11/etoposide, and others.

Data from *in vivo* animal studies indicate that combinations of CPT-11 with other agents should also be investigated. In particular, supra-additive (synergistic) activity has been demonstrated with the combinations CPT-11/mitomycin C and CPT-11/cytosine arabinoside [11]. Phase I trials with these combinations should be pursued.

Another regimen which should be considered for phase I study is the combination of one topoisomerase I inhibitor with another. Using a human tumour cloning assay, we have shown that there is not complete cross-resistance between topotecan and CPT-11 in terms of activity against human tumour colony-forming units. A total of 24 specimens taken directly from patients were exposed for 1 h to CPT-11 (1.0 μg/ml) and topotecan (0.1 μg/ml). As shown in Figure 1, non-cross-resistance was noted between the two topoisomerase I inhibitors in 9 specimens (38%). These data indicate that phase I/II investigations of the combination CPT-11/topotecan are warranted.

LEADS FOR PHASE II TRIALS WITH CPT-11

Solid tumours

The wide range of preclinical antitumour activity with CPT-11 in human tumour colony-forming units demonstrated by our group is shown in Table 3 [5]. Of those tumours in which *in vitro* activity has been noted, clinical antitumour activity with CPT-11 has already been documented in phase II studies in cervical cancer [12, 13], colon [4], non-small cell lung cancer [14, 15] and ovarian cancer [16].

Other tumour types against which one might also expect to see some antitumour activity in clinical practice include breast cancer, in which six *in vitro* responses were noted in 13 breast cancer specimens, and mesothelioma (responses

Topotecan/CPT-11	S	R	Total
S	3	5	8
R	1	15	16
Total	4	20	24

n = 24; non-cross-resistance = 9/24 = 38%; S, sensitive; R, resistant.

Figure 1. Sensitivity of 24 human tumour specimens exposed to equipotent concentrations of CPT-11 and topotecan.

in 4/12 specimens). Phase II trials in patients with breast cancer and with mesothelioma should be performed. In addition, although the number of sarcoma specimens tested was small, a phase II trial in patients with advanced sarcomas might also be considered.

Paediatric malignancies

A number of investigators have performed interesting studies of CPT-11 in human paediatric xenografts in nude mice [17, 18]. In these studies, significant antitumour activity was demonstrated against neuroblastoma, rhabdomyosarcoma and medulloblastoma xenografts. Phase II trials of CPT-11 should, therefore, be considered for paediatric patients with these tumours.

Chronic lymphocytic leukaemia

In the clinic, a frequently noted laboratory abnormality is that patients receiving CPT-11 develop significant lymphocytopenia on the weekly × 4 schedule of drug administration [19]. Although the clinical significance of this observation remains to be determined, it is possible that this effect of CPT-11 might translate into significant clinical activity against refractory chronic lymphocytic leukaemia (CLL). A phase II trial of CPT-11 should be initiated in patients with CLL who have progressed after treatment with fludarabine phosphate.

Identifying patients who will respond to CPT-11

It is logical to assume that tumour types in which topoisomerase I levels are elevated are likely to be excellent candidates for treatment with CPT-11. Indeed, good response rates have been demonstrated in clinical studies of CPT-11 in one such tumour, colorectal cancer [3, 4]. However, the relationship between elevated levels of topoisomerase I in a patient's tumour and the response of that patient's tumour to CPT-11 have not been explored prospectively.

Table 3. Tumour-specific activity of CPT-11 in a human tumour cloning assay

	Number of responses/number of evaluable specimens		
	Concentration of CPT-11 (μg/ml)		
Tumour type	0.3	1.5	3.0
Breast	1/13	3/15	6/13
Cervix	1/2	1/2	1/2
Colon	1/39	2/46	12/39
Lung (non-small cell)	1/12	4/12	6/12
Ovary	2/25	3/26	6/25
Pancreas	0/2	1/2	1/2
Mesothelioma	1/11	2/12	4/12
Sarcoma	0/2	1/2	2/2
Skin	0/1	1/1	1/1

*Response =⩽50% survival of tumour colony-forming units.

Another potential marker for sensitivity to CPT-11 therapy is carboxylesterase (CE). This enzyme catalyses the conversion of CPT-11 to SN-38, an active metabolite of CPT-11. Chen and colleagues [20] measured CE levels in 179 tumours taken directly from patients (18 different histological types). There was a wide range of CE activity among this cohort (0.009–1.274, median 0.125 μmol/min/mg protein). The tumour types containing the highest levels of CE activity included lymphoma, small cell lung cancer, thyroid cancer, bladder cancer, endometrial cancer and mesothelioma. Chen and colleagues also noted that those tumours with the highest levels of CE were the most sensitive to CPT-11 in a human tumour cloning assay. These observations suggest that the level of CE in a patient's tumour may be an important factor in determining the response of a patient's tumour to CPT-11. Phase II trials of CPT-11 should be considered in malignancies in which high levels of CE have been observed. These studies should investigate not only the efficacy and safety of CPT-11, but also the relationship between efficacy and level of CE in a patient's tumour.

CONCLUSION

There is no doubt that CPT-11 is an agent with substantial clinical activity against a variety of tumour types. However, a great deal of interesting preclinical information remains to be pursued in well-designed clinical trials. As our knowledge of the preclinical activity of CPT-11 continues to expand, there is no doubt there will be additional leads which should be tested in clinical trials.

1. Yokokura T, Sawada S, Nokata K, *et al.* Antileukemic activity of new camptothecin derivatives. *Proc Jap Cancer Assoc 40th Annual Meeting,* Sapporo, Japan 1981, 261.
2. Kunimoto T, Nitta K, Tanaka T, *et al.* Antitumour activity of 7-ethyl-10-[4-(1-piperidino)-1-piperidino]carbonyloxy-camptothecin, a novel water-soluble derivative of camptothecin, against murine tumors. *Cancer Res* 1987, **47**, 5944–5947.
3. Giovanella BC, Stehlin JS, Wall MB, *et al.* DNA topoisomerase I-targeted chemotherapy of human colon cancer xenografts. *Science* 1989, **246**, 1046–1048.
4. Shimada Y, Rougier P, Pitot H. Efficacy of CPT-11 (irinotecan) as a single agent in metastatic colorectal cancer. *Eur J Cancer* 1996, **32A**(Suppl. 3), S13–S17.
5. Shimada Y, Rothenberg M, Hilsenbeck SG, Burris HA III, Degen D, Von Hoff DD. Activity of CPT-11 (irinotecan hydrochloride), a topoisomerase I inhibitor, against human tumor colony-forming units. *Anticancer Drugs* 1994, **5**, 202–206.
6. Bugat R. CPT-11 in the treatment of colorectal cancer (CRC): safety profile. 5th International Congress of Anti-Cancer Chemotherapy. 31 Jan/3 Feb 1995, Paris, France (abstract S778).
7. Abigerges D, Armand JP, Chabot GG, *et al.* Irinotecan (CPT-11) high-dose escalation using intensive high-dose loperamide to control diarrhea. *J Natl Cancer Inst* 1994, **86**, 446–449.
8. Sakata Y, Suzuki H, Kamataki T. Preventive effect of TJ-14, a Kampo medicine, on diarrhea induced by irinotecan [abstract 1578]. *Proc Am Soc Clin Oncol* 1994, **13**, 456.
9. Chabot GG, Abigerges D, Catimel G, *et al.* Population pharmacokinetics and pharmacodynamics of irinotecan (CPT-11) and active metabolite SN-38 during phase I trials. *Ann Oncol* 1995, **6**, 141–151.

10. Saltz L, Shimada Y, Khayat D. CPT-11 (irinotecan) and 5-fluorouracil: a promising combination for therapy of colorectal cancer. *Eur J Cancer* 1996, **32A**(Suppl. 3), S24–S31.
11. Kano Y, Suzuka K, Akulsu M, *et al.* Effects of CPT-11 in combination with other anti-cancer agents in culture. *Int J Cancer* 1992, **50**, 604–610.
12. Chevallier B, Lhomme C, Dieras V, *et al.* A phase II study of CPT-11 (irinotecan) in chemotherapy naive patients with advanced cancer of the cervix uteri (C.C.U.) [abstract]. *8th European Conference on Clinical Oncology and Cancer Nursing (ECCO)*, Paris, 29 Oct–2 Nov 1995.
13. Takeuchi S, Noda K, Yakushiji M. Late phase II study of CPT-11, a topoisomerase I inhibitor, in advanced cervical carcinoma [abstract 708]. *Proc Am Soc Clin Oncol* 1992, **11**, 224.
14. Douillard JY, Riviere A, Spaeth D, Chomy P, Pujol JL. A phase II study of CPT-11 (irinotecan 350 mg/m^2 every 3 weeks) in untreated non-small cell lung carcinomas [abstract]. *8th European Conference on Clinical Oncology and Cancer Nursing (ECCO)*, Paris, 29 Oct–2 Nov 1995.
15. Fukuoka M, Niitani H, Suzuki A, *et al.* A phase II study of CPT-11, a new derivative of camptothecin, for previously untreated non-small-cell lung cancer. *J Clin Oncol* 1992, **10**, 16–20.
16. Takeuchi S, Dobashi K, Fujimoto S, *et al.* A late phase II study of CPT-11 on uterine cervical cancer and ovarian cancer. *Jpn J Cancer Chemother (Gan to Kagaku Ryoho)* 1991, **18**, 1681–1689.
17. Houghton PJ, Cheshire PJ, Hallman JC, Bissery MC, Mathieu-Boué A, Houghton JA. Therapeutic efficacy of the topoisomerase I inhibitor 7-ethyl-10-(4-[1-piperidino]-1-piperidino)-carbonyloxy-camptothecin against human tumor xenografts: lack of cross-resistance *in vivo* in tumors with acquired resistance to the topoisomerase I inhibitor 9-dimethylaminomethyl-10-hydroxycamptothecin. *Cancer Res* 1993, **53**, 2823–2829.
18. Vassal G, Morizet J, Bissery MC, *et al.* Activity of the camptothecin analog CPT-11 (irinotecan) against medulloblastoma xenografts [abstract 2813]. *Proc Am Assoc Cancer Res* 1994, **35**, 366.
19. Rothenberg ML, Kuhn JG, Burris HA III, *et al.* Phase I and pharmacokinetic trial of weekly CPT-11. *J Clin Oncol* 1993, **11**, 2194–2204.
20. Chen S-F, Rothenberg ML, Clark G, *et al.* Human tumor carboxylesterase activity correlates with CPT-11 cytotoxicity *in vitro*. *Proc Am Assoc Cancer Res* 1994, **35**, 365.