Anti-Cancer Drugs 2001, **12,** pp. 619–625

Case report

Toxicity of irinotecan (CPT-11) and hepato-renal dysfunction

Simon YK Ong,[1] Stephen J Clarke,[1] James Bishop,[1] Helen M Dodds[2] and Laurent P Rivory[1,2]

[1]Medical Oncology, Sydney Cancer Centre, Missenden Road, Camperdown, NSW 2050, Australia.
[2]Department of Pharmacology, University of Sydney, NSW 2006, Australia.

Various clinical and laboratory parameters have been investigated for their ability to predict toxicity arising from the use of the anticancer drug, irinotecan (CPT-11). In particular, patients deficient in the conjugation of SN-38, a metabolite of CPT-11, are known to be at greater risk. We describe one case of a patient with metastatic colorectal cancer treated with a single dose of CPT-11 at 125 mg/m^2. Although this patient lacked any known predictive factors for toxicity, he experienced severe side-effects several days later. We hypothesized that the toxicity in this patient was due to compromised SN-38 conjugation. Plasma samples were analyzed by reversed-phase high-performance liquid chromatography assay for CPT-11 and its metabolites at 96, 144, 168, 192 and 288 h post-administration. We observed that the concentrations of both the parent drug and its metabolites were markedly raised (11- to 60-fold expected). Additionally the estimated terminal half-lives were 1.5–7 times those expected (29.5, 101, 39.6 and 41.8 h for CPT-11, APC, SN-38G and SN-38, respectively). We conclude that the toxicity in this patient was not caused by deficient SN-38 conjugation, but by decreased drug excretion through both hepatic and renal routes. [© 2001 Lippincott Williams & Wilkins.]

Key words: Conjugation, drug excretion, irinotecan, pharmacokinetic, toxicity.

Introduction

Irinotecan (CPT-11) is a topoisomerase I poison which has significant activity in colorectal cancer.[1–4] The combination of CPT-11 and fluorouracil is now regarded as standard first-line therapy for patients with advanced or recurrent disease.[5,6] CPT-11 has also been shown to be active against a number of other tumor types.[7–9]

Correspondence to LP Rivory, Medical Oncology, Sydney Cancer Centre, Missenden Road, Camperdown, NSW 2050, Australia.
Tel: (+61) 2 9515 7376; Fax: (+61) 2 9519 1546;
E-mail: lrivory@canc.rpa.cs.nsw.gov.au

The principal toxicities of irinotecan are neutropenia and diarrhea, which in some cases may cause significant morbidity and occasional mortality.[10–14] There is significant inter-patient variability in toxicity, which may be in part due to variability in drug metabolism. CPT-11 is a prodrug of SN-38, the active metabolite, the conversion of which is mediated by carboxylesterases.[15–18] SN-38 undergoes inactivation by hepatic uridine diphosphate glucuronosyltransferase (UGT) 1A1 to its glucuronide ester SN-38G.[19] The latter is subsequently excreted in both bile and urine.[20–22] CPT-11 also undergoes oxidation by cytochrome P-450 (CYP) 3A to yield a number of mostly inactive metabolites.[23–26]

It is important to be able to identify patients who are at greater than normal risk of toxicity from CPT-11, particularly given that its use is likely to be expanded to the adjuvant setting. For example, recent work has demonstrated that some patients deficient in SN-38 conjugation are more at risk of severe toxicity.[19,27–29] We recently treated a patient with advanced colorectal cancer who experienced severe toxicity several days after the first dose of CPT-11. We hypothesized that the toxicity in this patient was due to compromised drug handling because of significant co-morbidity and to confirm this the CPT-11 pharmacokinetics were analyzed.

Case history

The patient was a 55-year-old man of Mediterranean origin with intercurrent medical problems of uncomplicated α-thalassemia trait, left ureteric calculus complicated by hydronephrosis treated 8 months previously by ureteric stenting and cholecystitis secondary to cholelithiasis. The latter condition had been treated by laparoscopic cholecystectomy 1

month prior to treatment with CPT-11. This procedure was complicated 2 weeks later by an episode of cholangitis.

The patient first presented with a moderately differentiated stage II adenocarcinoma of the cecum in March 1999 and was treated with a right hemicolectomy. He declined adjuvant chemotherapy. Inoperable hepatic secondaries were diagnosed in December 1999. The patient was enrolled in a phase II trial of oxaliplatin and fluorouracil to which he achieved a partial response with disease progression occurring in September 2000, 3 months after completion of trial chemotherapy.

Weekly CPT-11 at 125 mg/m^2 for 4 weeks followed by a 2-week rest was commenced in October 2000. The patient developed grade III diarrhea a few days after the first injection, which was not controlled by the recommended loperamide regimen. He was hospitalized on day 5 post-treatment with worsening abdominal pain and moderate dehydration. Baseline and admission laboratory values are depicted graphically in Figures 1–3. The patient was found to have grade IV neutropenia that lasted from day 7 to day 11 post-treatment before recovering on day 12. He became briefly febrile on day 8. Other hematological toxicities at presentation included grade II anemia (8.5 g/dl), grade II thrombocytopenia (55 000/mm^3) and grade I coagulopathy (PT=21.4 s and INR=1.9). There was no laboratory evidence of hemolysis. Non-hematological toxicities included grade II renal impairment (urea 23 mmol/l and creatinine 141 μmol/l) and oral thrush. Grade III liver dysfunction was also evident with elevated bilirubin (81 μmol/l, predominantly conjugated) and transaminases. Computerized tomography of the abdomen and pelvis was suggestive of pancolitis and showed no biliary tract pathology. Cultures of blood, urine and stool revealed no significant bacterial pathogens. He was treated with i.v. rehydration, blood transfusion of 2 U, combination antibiotics and colony stimulating factors, which resulted in a satisfactory improvement in his condition as well as the hematological and biochemistry profiles (Figures 1 and 2). After 10 days in the hospital, the patient's condition improved sufficiently for him to be discharged. At this time the abnormal blood counts and biochemistry had essentially returned to baseline levels with the exception of the liver indices. These values, though improved, remained elevated up to 2 weeks later. At discharge, bilirubin was 36 μmol/l, ALP was 373 U/l and GGT was 439 U/l.

Materials and methods

Plasma samples were obtained at 96, 144, 168, 192 and 288 h following the infusion of CPT-11. The plasma analysis was initially performed using an ultra-sensitive method initially developed for the measurement of trough concentrations of SN-38.[30] However, it soon became apparent that the concentrations were much higher than anticipated, and the method of Rivory and Robert for the quantitation of CPT-11, APC, SN-38 and SN-38G[31] was used instead.

Briefly, each sample was prepared by mixing 50 μl of plasma with 100 μl of ice-cold methanol:acetonitrile (50:50, v/v) containing 5 ng 20(*S*)-camptothecin as the internal standard. The mixture was vortexed briefly followed by centrifugation at 8000 *g* for 5 min. The supernatant was separated and mixed with 2.5 μl of 1N HCl and centrifuged again. The final supernatant was transferred into an insert and placed in the auto sampler at 4°C for analysis.

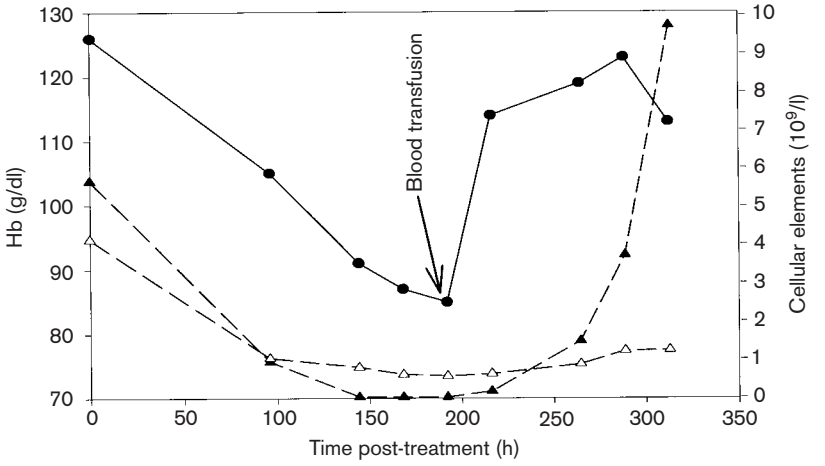

Figure 1. Hemoglobin level (●), absolute neutrophil count (▲) and platelet count/100 (△) following a single dose of CPT-11.

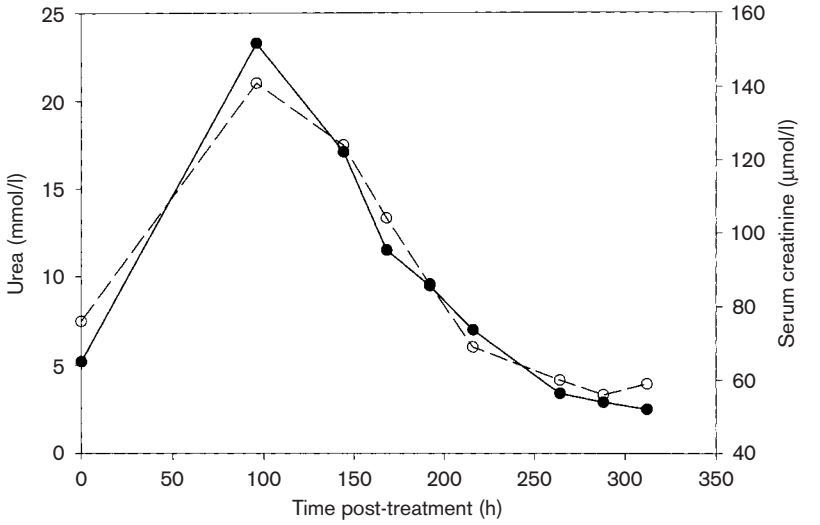

Figure 2. Urea (●) and creatinine (○) values following a single dose of CPT-11.

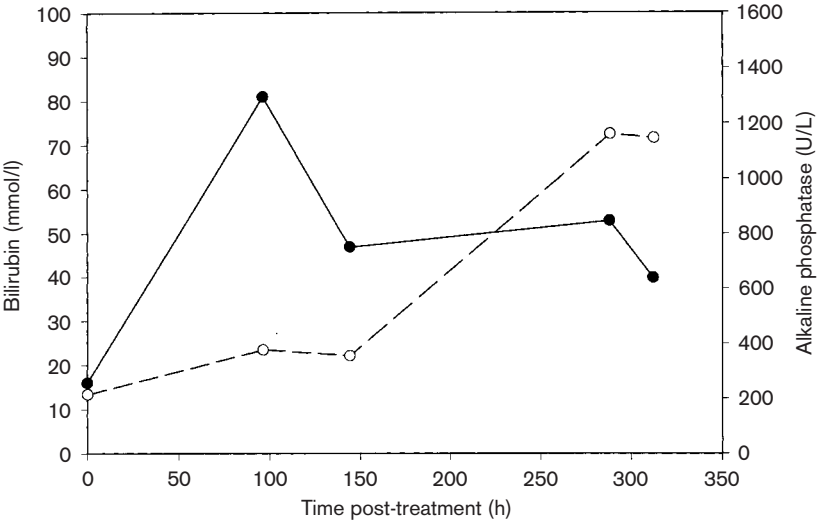

Figure 3. Bilirubin (●) and alkaline phosphatase (○) values following a single dose of CPT-11.

For SN-38G quantification, 50 μl of plasma was incubated at 37°C with 25 U of glucuronidase for 1 h. The incubated plasma was then treated in the same manner as described above.

Plasma standards were prepared by adding 5 μl of standard solution to 45 μl of blank plasma and treated as per the samples. Standards were prepared to give the following ranges of concentration: CPT-11, 10–5000 ng/ml, APC, 5–2500 ng/ml, and SN-38, 0.5–250 ng/ml. Separation was performed at ambient temperature using a Waters Nova-Pak Radial-Pak C_{18} reversed-phase column (5 × 250 mm, 4 μm) using 0.075 M ammonium acetate buffer (pH 4.5):acetonitrile (78:22, v/v) as the mobile phase at a flow rate of 1 ml/min. Fluorescence detection (RF-10AXL; Shimadzu, Sydney, Australia) was optimized for detection of SN-38 with excitation and emission wavelengths set at 380 and 530 nm, respectively. Ten microlitre aliquots were injected and concentrations calculated using the area ratio method.

Pharmacokinetic analysis

The terminal half-lives were calculated from the semi-log concentration versus time curves. The average SN-38G:SN-38 ratio was calculated as the mean of the ratio of concentrations at the five time points. The average

pharmacokinetic data obtained from four patients previously treated with CPT-11 at 125 mg/m^2 in another study[32] were used to act as comparators. The concentrations of CPT-11, APC and SN-38 generally become undetectable within 50 h post-treatment using the analytical method described above. Therefore, the concentration–time profiles of the four comparator patients were extrapolated out to 250 h using the terminal half-lives reported by Sparreboom *et al.*[21]

Previously, we have described the relationship between pre-treatment 'trough' concentrations of SN-38 and SN-38G and toxicity experienced by patients treated with the same weekly schedule of CPT-11.[33] These data, available for 10 patients on day 7 of treatment, were also used for comparison.

Results

The concentrations of CPT-11, APC, SN-38 and SN-38G are depicted in Figure 4 along with the comparator pharmacokinetic profiles. Levels of both the parent drug and its metabolites were markedly raised (11- to 60-fold predicted) in our patient. The day 7 concentrations of SN-38 (2.96 ng/ml) and SN-38G (19.5 ng/ml) observed in this patient are much higher than those found in our 'trough' study (*cf.* 0.20 and 0.69 ng/ml, respectively). Concentrations of CPT-11 and its metabolites remained detectable even as late as 288 h post-administration. The estimated terminal half-life ($t_{1/2}$) values of CPT-11, APC, SN-38G and SN-38 were 29.5, 101, 39.6 and 41.8 h, respectively. These values are approximately 1.5–7 times those expected. The SN-38G:SN-38 serum concentration ratio averaged 7.9 over the sampling interval.

Discussion

The two most common dose-limiting toxicities of irinotecan are neutropenia and delayed diarrhea (onset greater than 24 h). Severe (grade III/IV) neutropenia occurs in about 20–30% of patients and severe (grade III/IV) diarrhea in up to 40% of patients was initially reported.[10–12] The incidence of the latter has been reduced to around 4–6% with aggressive use of loperamide. Other common difficult toxicities that have been observed are nausea/emesis, asthenia and anemia.[34] Less frequent drug-related events include thrombocytopenia,[35] toxic death,[13,14,36] major hepatic dysfunction,[28,36,37] cardiac dysrrhythmia,[38] peumonitis[39–41] and tumor lysis syndrome.[36,42]

Our patient presented with major hematological, gastrointestinal and hepatic toxicity accompanied by mild coagulopathy and transient renal impairment. The myelotoxicity was particularly impressive as it involved suppression of all three cell lineages with the granulocytes being most profoundly affected. Some authors have attempted to determine the predictive parameters for toxicity, amongst other things, and have identified baseline bilirubin, hemoglobin, number of organs involved and time from diagnosis as predictive factors for neutropenia. Also, performance

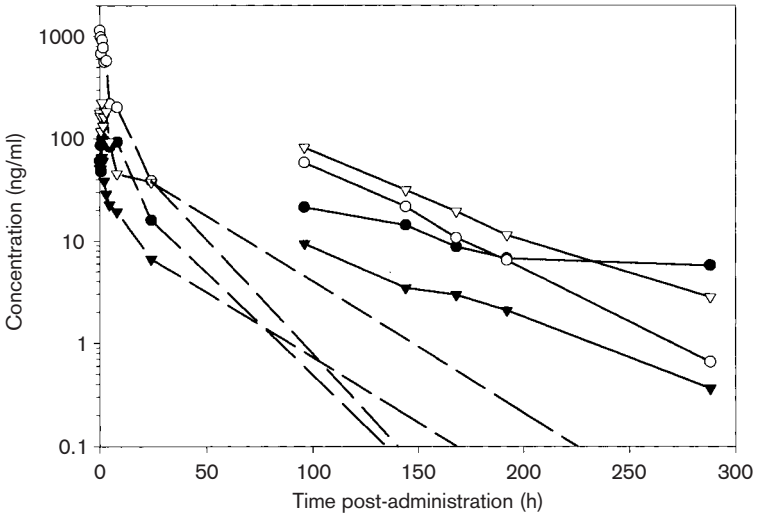

Figure 4. Concentrations of CPT-11 (○), APC (●), SN-38 (▼) and SN-38G (▽) observed in the case of a patient with severe toxicity following treatment with one dose of CPT-11 at 125 mg/m^2 (solid lines). Historical data from a previous study[26] and extrapolation based on data from Sparreboom[21] are shown here for comparison (broken lines).

status, serum creatinine, leukocyte count, time from fluorouracil progression and prior abdominopelvic irradiation are predictive factors for delayed diarrhea.[43]

Patients with Gilbert's syndrome are particularly prone to CPT-11-related toxicity because of a deficiency in UGT1A1 activity.[28] The deficit in conjugation of SN-38 is usually reflected by an altered SN-38G:SN-38 concentration ratio. The genotyping of patients for the most common polymorphism associated with Gilbert's syndrome, an additional TA repeat in the regulatory region of UGT1A1, may represent a useful screening test for patients prior to treatment with CPT-11.[44] Because bilirubin is conjugated by the same UGT enzyme, we suspected that increased heme turn-over (from thalassemia) might have compromised SN-38 glucuronidation in our patient. However, the clinical history did not support a diagnosis of Gilbert's syndrome. Also, the SN-38G:SN-38 ratio did not suggest any perturbation with the UGT 1A1 activity. Typically, the plasma SN-38G:SN-38 ratio is of the order of 2–10,[45] although it may decrease at very late time points.[46] At day 7, we previously observed a mean SN-38G:SN-38 concentration ratio of 3.84 ± 1.71 at 144 h.[33] Therefore, the ratio observed in our case was, if anything, higher than expected. Finally, the predominance of conjugated to unconjugated bilirubin provided additional support for an intact UGT1A1 activity in this patient.

Rather, the evidence gathered in this case points to a multifactorial impairment of drug elimination. High and persisting concentrations of APC were also observed in this patient which would argue against diminished drug metabolism through cytochrome P450 3A, which is recognized as an important elimination pathway.[24,25] Concomitant liver disease is known to have an impact on the pharmacokinetics of CPT-11, sometimes with dire consequences.[37] In a recent study, patients with elevated alkaline phosphatase and bilirubin were found to have reduced clearance of CPT-11.[47] The latter declined in a non-linear fashion with increasing hepatic dysfunction reaching an asymptotic value of approximately 4 l/h/m^2 (as compared to around 15 l/h/m^2 in normal patients). This would suggest that renal excretion, which accounts for 20–25% of the elimination of unchanged CPT-11,[22] represents a major route of elimination in patients with extensive liver involvement.

The canalicular transporter cMOAT (also known as MRP2) is thought to be largely responsible for the biliary excretion of CPT-11, SN-38 and SN-38G,[48–51] although P-glycoprotein may be responsible for the high-affinity component of the transport of the carboxylate form of CPT-11.[48–51] These transporters have been shown to be down-regulated in response to inflammation, sepsis and biliary obstruction.[52,53] In view of the recent history of cholangitis and in the presence of hydronephrosis, it is likely that a combination of renal and disease-related hepatic dysfunction (reduction in functional parenchyma plus reduced expression of transporters) led to grossly compromised drug elimination in this patient.

However, we cannot rule out a possible direct contribution of the drug on this multi-organ dysfunction. Four patients recently described by Nikolic-Tomasevic had a combination of neutropenia, diarrhea, renal impairment and liver dysfunction of varying severity after exposure to CPT-11 culminating in death.[36] Because all four patients had normal pre-treatment parameters, the authors suggested that CPT-11 might interfere with renal and hepatic function in a subset of patients who did not fulfil the usual predictive criteria for toxicity.

Conclusion

We conclude that the profound toxicities experienced by this patient were correlated to the abnormal pharmacokinetic findings. We believe that multiple endogenous and exogenous factors exerting their effects on the two key organs, the liver and the kidneys, have resulted in markedly decreased drug excretion in this case. We recommend that close monitoring of compromised renal function should be performed in patients with liver involvement receiving CPT-11. Also, prudence should be exercised when treating patients with a recent history of surgery (particularly hepatobiliary) and infection.

References

1. Cunningham D, Pyrhonen S, James RD, *et al*. Randomised trial of irinotecan plus supportive care versus supportive care alone after fluorouracil failure for patients with metastatic colorectal cancer [see Comments]. *Lancet* 1998; **352**: 1413–8.
2. Cunningham D, Glimelius B. A phase III study of irinotecan (CPT-11) versus best supportive care in patients with metastatic colorectal cancer who have failed 5-fluorouracil therapy. V301 Study Group. *Semin Oncol* 1999; **26**(1 suppl 5): 6–12.
3. Van Cutsem E, Blijham GH. Irinotecan versus infusional 5-fluorouracil: a phase III study in metastatic colorectal cancer following failure on first-line 5-fluorouracil. V302 Study Group. *Semin Oncol* 1999; **26**(1 suppl 5): 13–20.
4. Rougier P, Van Cutsem E, Bajetta E, *et al*. Randomised trial of irinotecan versus fluorouracil by continuous infusion after fluorouracil failure in patients with metastatic colorectal cancer [see Comments] [published erratum appears in *Lancet* 1998; **352**(9140): 1634]. *Lancet,* 1998; **352**: 1407–12.

5. Saltz LB, Cox JV, Blanke C, *et al*. Irinotecan plus fluorouracil and leucovorin for metastatic colorectal cancer. Irinotecan Study Group [see Comments]. *N Eng J Med* 2000; **343**: 905–14.
6. Douillard JY, Cunningham D, Roth AD, *et al*. Irinotecan combined with fluorouracil compared with fluorouracil alone as firstline treatment for metastatic colorectal cancer: a multicentre randomised trial [published erratum appears in *Lancet* 2000; **355**(9212): 1372]. *Lancet* 2000; **355**: 1041–7.
7. Sandler A, van Oosterom AT. Irinotecan in cancers of the lung and cervix. *Anti-Cancer Drugs* 1999; **10**(suppl 1): S13–7.
8. Rosen LS. Irinotecan in lymphoma, leukemia, and breast, pancreatic, ovarian, and small-cell lung cancers. *Oncology* 1998; **12**(8 suppl 6): 103–9.
9. Bleiberg H. CPT-11 in gastrointestinal cancer. *Eur J Cancer* 1999; **35**: 371–9.
10. Wiseman LR, Markham A. Irinotecan. A review of its pharmacological properties and clinical efficacy in the management of advanced colorectal cancer. *Drugs* 1996; **52**: 606–23.
11. Rothenberg ML, Eckardt JR, Kuhn JG, *et al*. Phase II trial of irinotecan in patients with progressive or rapidly recurrent colorectal cancer. *J Clin Oncol* 1996; **14**: 1128–35.
12. Pitot HC, Wender DB, O'Connell MJ, *et al*. Phase II trial of irinotecan in patients with metastatic colorectal carcinoma. *J Clin Oncol* 1997; **15**: 2910–9.
13. Negoro S, Fukuoka M, Masuda N, *et al*. Phase I study of weekly intravenous infusions of CPT-11, a new derivative of camptothecin, in the treatment of advanced non-small-cell lung cancer. *J Natl Cancer Inst,* 1991; **83**: 1164–8.
14. Lhomme C, Fumoleau P, Fargeot P, *et al*. Results of a European Organization for Research and Treatment of Cancer/Early Clinical Studies Group phase II trial of first-line irinotecan in patients with advanced or recurrent squamous cell carcinoma of the cervix. *J Clin Oncol* 1999; **17**: 3136–42.
15. Slatter JG, Su P, Sam JP, Schaaf LJ, Wienkers LC. Bioactivation of the anticancer agent CPT-11 to SN-38 by human hepatic microsomal carboxylesterases and the *in vitro* assessment of potential drug interactions. *Drug Metab Disp* 1997; **25**: 1157–64.
16. Rivory LP, Bowles MR, Robert J, Pond SM. Conversion of irinotecan (CPT-11) to its active metabolite, 7-ethyl-10-hydroxycamptothecin (SN-38), by human liver carboxylesterase. *Biochem Pharmacol* 1996; **52**: 1103–11.
17. Haaz MC, Rivory LP, Riche C, Robert J. The transformation of irinotecan (CPT-11) to its active metabolite SN-38 by human liver microsomes. Differential hydrolysis for the lactone and carboxylate forms. *Naunyn-Schmiedebergs Arch Pharmacol* 1997; **356**: 257–62.
18. Humerickhouse R, Lohrbach K, Li L, Bosron WF, Dolan ME. Characterization of CPT-11 hydrolysis by human liver carboxylesterase isoforms hCE-1 and hCE-2. *Cancer Res* 2000; **60**: 1189–92.
19. Iyer L, King CD, Whitington PF, *et al*. Genetic predisposition to the metabolism of irinotecan (CPT-11). Role of uridine diphosphate glucuronosyltransferase isoform 1A1 in the glucuronidation of its active metabolite (SN-38) in human liver microsomes. *J Clin Invest* 1998; **101**: 847–54.
20. Lokiec F, du Sorbier BM, Sanderink GJ. Irinotecan (CPT-11) metabolites in human bile and urine. *Clinl Cancer Res* 1996; **2**: 1943–9.
21. Sparreboom A, de Jonge MJ, de Bruijn P, *et al*. Irinotecan (CPT-11) metabolism and disposition in cancer patients. *Clin Cancer Res* 1998; **4**: 2747–54.
22. Slatter JG, Schaaf LJ, Sams JP, *et al*. Pharmacokinetics, metabolism, and excretion of irinotecan (CPT-11) following I.V. infusion of [(14)C]CPT-11 in cancer patients. *Drug Metab Disp* 2000; **28**: 423–33.
23. Rivory LP, Riou JF, Haaz MC, *et al*. Identification and properties of a major plasma metabolite of irinotecan (CPT-11) isolated from the plasma of patients. *Cancer Res* 1996; **56**: 3689–94.
24. Haaz MC, Riche C, Rivory LP, Robert J, *et al*. Biosynthesis of an aminopiperidino metabolite of irinotecan [7-ethyl-10-[4-(1-piperidino)-1-piperidino]carbonyloxycamptothecine] by human hepatic microsomes. *Drug Metab Disp* 1998; **26**: 769–74.
25. Haaz MC, Rivory L, Riche C, Vernillet L, Robert J. Metabolism of irinotecan (CPT-11) by human hepatic microsomes: participation of cytochrome P-450 3A and drug interactions. *Cancer Res* 1998; **58**: 468–72.
26. Dodds HM, Haaz MC, Riou JF, Robert J, Rivory LP. Identification of a new metabolite of CPT-11 (irinotecan): pharmacological properties and activation to SN-38. *J Pharmacol Exp Ther* 1998; **286**: 578–83.
27. Iyer L, Hall D, Das S, *et al*. Phenotype-genotype correlation of *in vitro* SN-38 (active metabolite of irinotecan) and bilirubin glucuronidation in human liver tissue with UGT1A1 promoter polymorphism. *Clin Pharmacol Ther* 1999; **65**: 576–82.
28. Wasserman E, Myara A, Lokiec F, *et al*. Severe CPT-11 toxicity in patients with Gilbert's syndrome: two case reports. *Ann Oncol* 1997; **8**: 1049–51.
29. Ando Y, Saka H, Asai G, *et al*. UGT1A1 genotypes and glucuronidation of SN-38, the active metabolite of irinotecan. *Ann Oncol* 1998; **9**: 845–7.
30. Rivory LP, Findlay M, Clarke S, Bishop J. Trace analysis of SN-38 in human plasma by high-performance liquid chromatography. *J Chromatogr B: Biomed Sci Appl* 1998; **714**: 355–9.
31. Rivory LP, Robert J. Reversed-phase high-performance liquid chromatographic method for the simultaneous quantitation of the carboxylate and lactone forms of the camptothecin derivative irinotecan, CPT-11, and its metabolite SN-38 in plasma. *J Chromatogr B: Biomed Sci Appl* 1994; **661**: 133–41.
32. Dodds HM, Clarke SJ, Findlay M, *et al*. Clinical pharmacokinetics of the irinotecan metabolite 4-piperidinopiperidine and its possible clinical importance. *Cancer Chemother Pharmacol* 2000; **45**: 9–14.
33. Rivory LP, Clarke S, Bishop JF, *et al*. Irinotecan (CPT-11): A prelimary investigation of the relationship between trough concentrations of the active metabolite, SN-38, and toxicity. *Ann Oncol* 1998; **9**(suppl 2): abstr 222.
34. Verschraegen CF, Levy T, Kudelka AP, *et al*. Phase II study of irinotecan in prior chemotherapy-treated squamous cell carcinoma of the cervix. *J Clin Oncol* 1997; **15**: 625–31.
35. Takimoto CH, Morrison G, Harold N, *et al*. Phase I and pharmacologic study of irinotecan administered as a 96-hour infusion weekly to adult cancer patients. *J Clin Oncol* 2000; **18**: 659–67.

36. Nikolic-Tomasevic Z, Jelic S, Popov I, Radosavljevic D. Colorectal cancer: dilemmas regarding patient selection and toxicity prediction. *J Chemother* 2000; **12**: 244–51.
37. van Groeningen CJ, Van der Vijgh WJ, Baars JJ, *et al*. Altered pharmacokinetics and metabolism of CPT-11 in liver dysfunction: a need for guidelines. *Clin Cancer Res* 2000; **6**: 1342–6.
38. Miya T, Fujikawa R, Fukushima J, *et al*. Bradycardia induced by irinotecan: a case report. *Jpn J Clin Oncol* 1998; **28**: 709–11.
39. 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 [see Comments]. *J Clin Oncol* 1992; **10**: 16–20.
40. Yamada M, Kudoh S, Hirata K, Nakajima T, Yoshikawa J. Risk factors of pneumonitis following chemoradiotherapy for lung cancer. *Eur J Cancer* 1998; **34**: 71–5.
41. Masuda N, Fukuoka M, Kusunoka Y, *et al*. CPT-11: a new derivative of camptothecin for the treatment of refractory or relapsed small-cell lung cancer. *J Clin Oncol* 1992; **10**: 1225–9.
42. Boisseau M, Bugat R, Mahjoubi M. Rapid tumour lysis syndrome in a metastatic colorectal cancer increased by treatment (CPT-11) [Letter]. *Eur J Cancer* 1996; **32A**: 737–8.
43. Freyer G, Rougier P, Bugat R, *et al*. Prognostic factors for tumour response, progression-free survival and toxicity in metastatic colorectal cancer patients given irinotecan (CPT-11) as second-line chemotherapy after 5FU failure. CPT-11 F205, F220, F221 and V222 study groups. *Br J Cancer* 2000; **83**: 431–7.
44. Iyer L, Hall D, Das S, *et al*. Phenotype–genotype correlation of *in vitro* SN-38 (active metabolite of irinotecan) and bilirubin glucuronidation in human liver tissue with UGT1A1 promoter polymorphism. *Clin Pharmacol Ther* 1999; **65**: 576–82.
45. Rivory LP, Haaz MC, Canal P, *et al*. Pharmacokinetic interrelationships of irinotecan (CPT-11) and its three major plasma metabolites in patients enrolled in phase I/II trials. *Clin Cancer Res* 1997; **3**: 1261–6.
46. Kehrer DF, Yamamoto W, Verweij J, *et al*. Factors involved in prolongation of the terminal disposition phase of SN-38: clinical and experimental studies. *Clin Cancer Res* 2000; **6**: 3451–8.
47. Boige V, Vernillet L, Hua A, *et al*. Total plasma clearance of irinotecan (CPT-11) correlates with both alkaline phosphatase and bilirubin levels in patients with tumoral hepatic involvement. *Proc Am Ass Cancer Res* 2000; **41**: abstr 3873.
48. Chu XY, Kato Y, Ueda K, *et al*. Biliary excretion mechanism of CPT-11 and its metabolites in humans: involvement of primary active transporters. *Cancer Res* 1998; **58**: 5137–43.
49. Chu XY, Kato Y, Sugiyama Y. Possible involvement of P-glycoprotein in biliary excretion of CPT-11 in rats. *Drug Metab Disp* 1999; **27**: 440–1.
50. Chu XY, Suzuki H, Ueda K, *et al*. Active efflux of CPT-11 and its metabolites in human KB-derived cell lines. *J Pharmacol Exp Ther* 1999; **288**: 735–41.
51. Sugiyama Y, Kato Y, Chu X. Multiplicity of biliary excretion mechanisms for the camptothecin derivative irinotecan (CPT-11), its metabolite SN-38, and its glucuronide: role of canalicular multispecific organic anion transporter and P-glycoprotein. *Cancer Chemother Pharmacol* 1998; **42**(suppl): S44–9.
52. Paulusma CC, Kothe MJ, Bakker CT, *et al*. Zonal down-regulation and redistribution of the multidrug resistance protein 2 during bile duct ligation in rat liver. *Hepatology* 2000; **31**: 684–93.
53. Tang W, Yi C, Kalitsky J, Piquette-Miller M. Endotoxin downregulates hepatic expression of P-glycoprotein and MRP2 in 2-acetylaminofluorene-treated rats. *Mol Cell Biol Res Commun* 2000; **4**: 90–97.

(Received 19 April 2001; accepted 10 May 2001)