Cancer Chemother Pharmacol (2000) 46: 235–240

ORIGINAL ARTICLE

Osama M. Ashour · Fardos N. M. Naguib
Naganna M. Goudgaon · Raymond F. Schinazi
Mahmoud H. el Kouni

Effect of 5-(phenylselenenyl)acyclouridine, an inhibitor of uridine phosphorylase, on plasma concentration of uridine released from 2′,3′,5′-tri-*O*-acetyluridine, a prodrug of uridine: relevance to uridine rescue in chemotherapy

Received: 10 November 1999 / Accepted: 14 March 2000

Abstract *Purpose*: The purpose of this investigation was to study the effects of combining oral 5-(phenylselenenyl)acyclouridine (PSAU) with 2′,3′,5′-tri-*O*-acetyluridine (TAU) on the levels of plasma uridine in mice. PSAU is a new lipophilic and potent inhibitor of uridine phosphorylase (UrdPase, EC 2.4.2.3), the enzyme responsible for uridine catabolism. PSAU has 100% oral bioavailability and is a powerful enhancer of the bioavailability of oral uridine. TAU is a prodrug of uridine and a far superior source of uridine than uridine itself. *Methods*: Oral TAU was administered to mice alone or with PSAU. The plasma levels of uridine and its catabolites as well as PSAU were measured using HPLC and pharmacokinetic analysis was performed. *Results*: Oral administration of 2000 mg/kg TAU increased plasma uridine by over 250-fold with an area under the curve (AUC) of 754 μmol · h/l. Coadministration of PSAU at 30 and 120 mg/kg with TAU further improved the bioavailability of plasma uridine resulting from the administration of TAU alone by 1.7- and 3.9-fold, respectively, and reduced the C_{max} and AUC of plasma uracil. *Conclusion*: The exceptional effectiveness of PSAU plus TAU in elevating and sustaining a high plasma uridine concentration could be useful in the management of medical disorders that are remedied by administration of uridine, as well as the rescue or protection from host toxicities of various chemotherapeutic pyrimidine analogues.

Key words 5-(Phenylselenenyl)acyclouridine · Uridine phosphorylase · Inhibitor · Triacetyluridine · Chemotherapy

Abbreviations *AUC* area under the curve · C_0 plasma concentration at zero time · C_{max} peak plasma concentration · Cl_T total plasma clearance · *HPLC* high performance liquid chromatography · *HPMC* hydroxypropylmethylcellulose · *MRT* mean residence time · *PSAU* 5-(phenylselenenyl)acyclouridine · $t_{1/2}$ elimination half-life · *TAU* 2′,3′,5′-tri-*O*-acetyluridine · T_{max} time to peak plasma concentration · *UrdPase* uridine phosphorylase, EC 2.4.2.3 · V_{dss} volume of distribution at steady state

Introduction

The pyrimidine nucleoside uridine has been used successfully as a ''protective'' and/or ''rescuing'' agent against host toxicity of various anticancer drugs (e.g. 5-fluorouracil) [20, 21, 25, 32, 35, 37] and anti-AIDS drugs (e.g. 3′-azido-3′-deoxythymidine and 2′,3′-dideoxycytidine) [19, 38, 39] without interfering with their chemotherapeutic efficacy. However, because of its rapid clearance [1, 2, 3, 23, 27, 39, 41, 42, 43, 44], it is necessary to administer substantial doses of uridine (10–12 g/m^2) [23] to attain and sustain the high plasma uridine concentrations (70 μ*M*) [26] required to achieve the protective or rescuing effect. Unfortunately, such large doses of uridine also produce dose-limiting side effects (e.g. phlebitis, pyrogenic reactions, high fever, cellulitis, diarrhea and superior vena cava syndrome) [4, 11, 33, 34, 41, 42, 43]. These side effects are not induced by uridine per se but by the accumulation of uridine catabolites [34]. The limited clinical utility of acute use of high-dose uridine regimens has led to the search for alternative approaches to increase uridine bioavailability.

O. M. Ashour · F. N. M. Naguib · M. H. el Kouni (✉)
Department of Pharmacology and Toxicology,
Comprehensive Cancer Center, Center for AIDS Research,
University of Alabama at Birmingham, Birmingham,
AL 35294, USA
e-mail: m.elkouni@ccc.uab.edu
Tel.: +1-205-9341132; Fax: +1-205-9348240

O. M. Ashour
Department of Pharmacology, Faculty of Medicine,
University of El Menia, El Menia, Egypt

N. M. Goudgaon · R. F. Schinazi
Veterans Affairs Medical Center (Atlanta), Decatur,
GA 30033, and Laboratory of Biochemical Pharmacology,
Department of Pediatrics, Emory University School of Medicine,
Atlanta, GA 30322, USA

Uridine is present at constant concentrations (1–5 μ*M*) in the plasma of various species [12, 27, 29]. However, the plasma half-life of uridine is approximately 2 min [12]. Hence, the turnover of the plasma uridine must be rapid and efficient. Indeed, more than 90% of the circulating uridine is catabolized in a single pass through the liver by the activity of hepatic uridine phosphorylase (UrdPase, EC 2.4.2.3), while constant amounts of uridine are synthesized de novo and released into the hepatic vein blood [7, 28]. Less than 2% of the uridine metabolized by the liver is salvaged and recovered in the uracil nucleotide pool in tissues of whole animals [5, 15, 16, 29], perfused rat liver [12, 27], and isolated liver cells [15]. The remainder is rapidly catabolized to products beyond uracil in the pyrimidine catabolic pathway [17, 28, 40].

Inhibition of uridine catabolism by UrdPase inhibitors has been used to increase the concentration and half-life of plasma uridine [1, 2, 6, 9, 26, 28, 34, 36, 40], and the salvage of uridine by various tissues [7, 8, 26, 34]. 2′,3′,5′-Tri-*O*-acetyluridine (TAU), a prodrug of uridine which is resistant to degradation by UrdPase [1], has also been shown to increase the concentration of plasma uridine in mice [1] and humans [18].

We have recently synthesized [13] and tested [2] 5-(phenylselenenyl)acyclouridine (PSAU) as a potent and specific inhibitor of UrdPase. PSAU was designed as a lipophilic inhibitor of UrdPase and as such its access to the liver and intestine, the main organs involved in uridine catabolism [12, 15, 16, 27, 28, 29], would be facilitated. PSAU is not metabolized, has 100% oral bioavailability, and is a powerful enhancer of the bioavailability of oral uridine [2]. In the present study we investigated the combined effect of PSAU and TAU on plasma uridine concentration.

Materials and methods

Chemicals

Heparinized Natelson pipettes, ammonium acetate, acetonitrile (HPLC grade), trichloroacetic acid, Gelman Acrodisc LC 13 PVDF 0.2-μm filters and ethyl ether (anesthetic grade) were obtained from Fisher Scientific (Pittsburgh, Pa.). TAU, tri-*n*-octylamine, freon (1,1,2-trichloro-trifluoroethane), hydroxypropylmethylcellulose (HPMC) and other chemicals were purchased from Sigma Chemical Company (St. Louis, Mo.). PSAU was synthesized as previously described [14].

Animals

Female CD-1 mice (18–20 g) were obtained from Charles River Laboratories (Wilmington, Mass.) and housed five per cage with water and food ad libitum under a normal light cycle (light 0600–1800 hours, dark 1800–0600 hours) according to an institutionally approved animal protocol.

Administration of drugs

TAU (alone or combined with PSAU) was mixed well with HPMC powder in hot water (80 °C) and homogenized thoroughly using a polytron homogenizer (Brinkmann Instruments, Westbury, N.Y.). The final concentration of HPMC was 0.75%. The drug solution was vortexed well before and periodically during dosing. HPMC was preferred over the commonly used methylcellulose because the latter must be cooled to 10 °C for complete hydration [1, 2]. Drugs were administered (0.1 ml/10 g) using 18G intubation needles (Popper and Sons, New Hyde Park, N.Y.). Drugs were administered to groups of female CD-1 mice (five mice per group, 20–25 g). PSAU was administered at 30 and 120 mg/kg. In an previous investigation, we studied the effect of different doses of TAU on the pharmacokinetics of plasma uridine [1]. We found that the administration of 2000 mg/kg TAU produced the best desirable effect on elevating plasma uridine concentration. Therefore, in the present study TAU was administered at 2000 mg/kg alone and in combination with PSAU (30 and 120 mg/kg) to determine the effect of PSAU on the modulation of plasma uridine released from TAU. Control mice received the carrier solution (0.75% HPMC). To avoid a possible circadian variation in UrdPase and dihydrouracil dehydrogenase (EC 1.3.1.2) activities [10, 31], TAU and/or PSAU were administered at the same time (between 8:30 and 9:00 a.m.).

Collection of samples

At various times (5, 10, 15 and 30 min, and 1, 2, 3, 4, 6, 8, 12 and 24 h) after drug administration, 250 μl of whole blood was collected from the orbital sinuses from each of five mice (lightly anesthetized with ethyl ether) into heparinized Natelson pipettes and placed on ice [1]. The whole blood was then centrifuged (Fisher Microcentrifuge Model 235 A) at 12,400 rpm for 5 min and the plasma recovered and immediately stored at −20 °C until analysis by HPLC.

Preparation of samples

Plasma was allowed to thaw on ice and then was deproteinized with two volumes of 15% trichloroacetic acid. After centrifugation (16,000 *g*, 4 °C) for 5 min, using a Brinkmann Eppendorf Microcentrifuge, the supernatant acid-soluble material was neutralized by extraction with a 1:2 mixture of tri-*n*-octylamine in freon. The neutralized supernatant was filtered through a Gelman Acrodisc LC 13 PVDF 0.2-μm filter prior to HPLC analysis [1, 2]. Under these conditions, the concentration of uridine released from TAU was not changed by acid treatment or by freezing during storage for up to 2 weeks (the longest duration of storage employed).

HPLC analysis

Samples were analyzed by HPLC using a computer-controlled Hewlett Packard model 1050 liquid chromatography apparatus equipped with an autosampler, a quaternary pump, and a multiple wavelength diode array base three channel UV detector as previously described [1, 2].

Uracil and uridine were identified by UV absorption at μ_{max}/254 nm, (μ_{max} 259.5 and 262 nm, respectively) and coelution with authentic samples. Uracil, uridine TAU, and PSAU eluted at 13, 27, 47, and 47.5 min, respectively. The recovery of uracil and uridine was more than 98% [1]. The areas under the curve (AUC) for uracil and uridine in the samples were calculated by the on-line computer. The concentrations of uracil and uridine in the samples were determined using standard curves prepared in double-distilled water. Plots of AUC vs uracil, uridine and PSAU concentrations were linear between 1 and 5000 μ*M*.

Pharmacokinetic analysis of plasma uridine, uracil and PSAU

The pharmacokinetic parameters of uridine, uracil and PSAU were estimated as previously described [1] by noncompartmental model-independent methods using the SIPHAR/BASE program [13]. The

Table 1 Effect of oral coadministration of different doses of PSAU with TAU at 2000 mg/kg on the pharmacokinetic parameters of plasma uridine and uracil in CD-1 mice. Values are mean ± SD from at least five mice at each time-point (C_{max} peak plasma concentration, C_0 baseline plasma concentration, T_{max} time to peak plasma concentration, *AUC* area under the curve)

	PSAU (mg/kg)	C_{max} (μ*M*)	Fold change (C_{max}/C_0)	T_{max} (h)	AUC (μmol · h/l)
Uridine	0	507 ± 298	252 ± 22.3	0.41 ± 0.22	754 ± 355
	30	1200 ± 329	484 ± 128	0.41 ± 0.00	1284 ± 422
	120	1183 ± 150	704 ± 128	0.21 ± 0.03	1217 ± 222
Uracil	0	665 ± 287	71.6 ± 13.9	1.00 ± 0.10	2115 ± 39
	30	261 ± 121	47.4 ± 22.1	0.52 ± 0.18	342 ± 180
	120	78.1 ± 29.2	19.4 ± 6.4	0.17 ± 0.21	111 ± 36

AUC was estimated by the trapezoidal rule with extrapolation to time infinity using the terminal disposition slope (K) generated by a weighted nonlinear least-squares regression of an exponential fit of the data [24], with the weighted square factor set as the reciprocal of the calculated concentration squared. Elimination half-life ($t_{1/2}$) values of uridine and PSAU were calculated from 0.693/K. The total plasma clearance (Cl_T) was calculated by dividing the administered dose by the AUC. The apparent volume of distribution at steady state (V_{dss}) was calculated as the product of Cl_T and the mean residence time (MRT) and normalized to the weight of the animals. The peak plasma concentration (C_{max}) and time of peak plasma concentration (T_{max}) values were estimated from the abscissa and ordinate, of the point with the highest ordinate on the computer-generated least squares curve depicting plasma concentration vs time. C_0 was the plasma concentration of endogenous uridine and uracil observed at zero time (0830 to 0900 hours). Relative bioavailability of uridine released from TAU was calculated from the AUC of plasma uridine resulting from oral administration of TAU expressed as a percentage the AUC of endogenous plasma uridine.

Results and discussion

The normal baseline concentrations (C_0) of plasma uridine and uracil at 8.30–9.00 a.m. in untreated CD-1 mice were relatively constant averaging 2.6 ± 0.7 and 7.4 ± 1.0 μ*M*, respectively. This is in agreement with previous studies [1, 2, 22]. Also, as reported previously [1], parenteral administration of 2000 mg/kg TAU increased the C_{max} value of plasma uridine by 250-fold and that of uracil by 10-fold (Table 1, Fig. 1). The data in Table 1 show that the AUC for plasma uridine released from TAU (754 μmol · h/l) was sevenfold higher than that reported for the administration of an equimolar concentration of oral uridine [1, 2]. The high efficiency of oral TAU in delivering uridine to the plasma is due to its structure. TAU, unlike uridine, is more lipophilic and hence absorbed from the gastrointestinal tract and reabsorbed from the renal tubules better than uridine. It is also resistant to catabolism by UrdPase [1]. Therefore, a large proportion of administered TAU is transported or diffused into the plasma unchanged and/or as mono- or diester uridine derivatives which act as depots. Thus, uridine is released by the action of plasma esterases over a longer period of time than when oral uridine is used [1]. Indeed, it has been shown that uridine released from oral TAU has a higher bioavailability (53%) than oral uridine (7%). Higher and sustained levels of plasma uridine have been also observed after administration of oral TAU than after uridine administration [1].

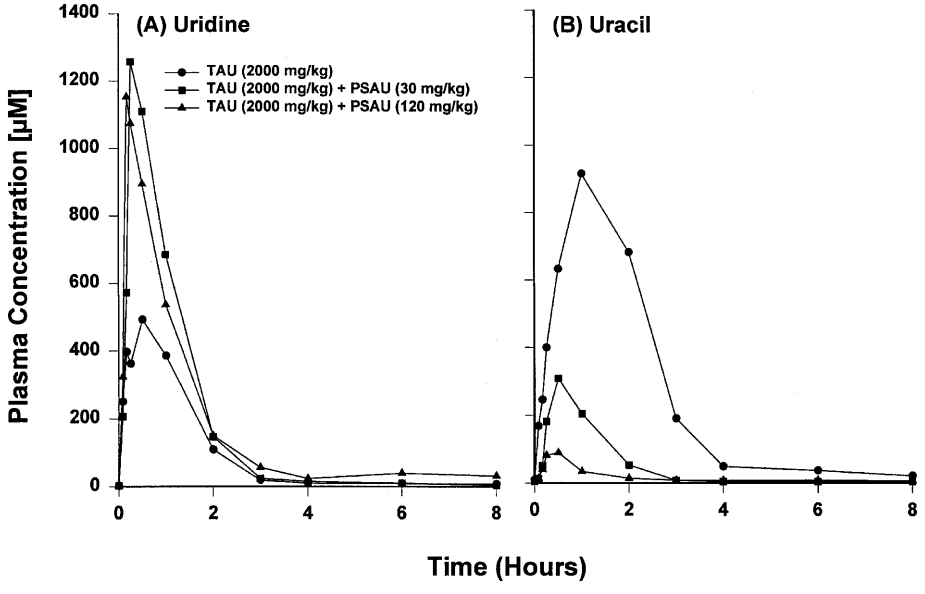

Fig. 1A,B Plasma concentration-time curves of (**A**) uridine and (**B**) uracil in CD-1 mice after oral administration of TAU (2000 mg/kg) alone and in combination with PSAU (30 and 120 mg/kg) mice at different time-points. Each point represents the mean from five mice

Table 2 The effect of coadministration triacetyluridine (TAU) on the pharmacokinetic parameters of plasma 5-(phenylselenenyl) acyclouridine (PSAU) in CD-1 mice. Values are means ± SD from at least five mice at each time-point (C_{max} peak plasma concentration, T_{max} time to peak plasma concentration, AUC area under the curve, Cl_T total plasma clearance, $t_{1/2}$ elimination half-life, V_{dss} volume of distribution at steady state, MRT mean residence time)

PSAU (mg/kg)	TAU (mg/kg)	C_{max} (μM)	T_{max} (h)	AUC (μmol · h/l)	V_{dss} (l/kg)	MRT (h)	Cl_T (l/h/kg)	$t_{1/2}$ (h)
30	0	64.0 ± 16.0	0.43 ± 0.05	134 ± 44.3	1.11 ± 0.23	1.82 ± 0.11	0.66 ± 0.14	1.17 ± 0.07
30	2000	37.0 ± 13.7	0.50 ± 0.21	73.0 ± 16.3	1.22 ± 0.13	1.40 ± 0.10	1.22 ± 0.25	0.78 ± 0.17
120	0	218 ± 60.2	0.76 ± 0.10	458 ± 204	0.61 ± 0.13	1.54 ± 0.21	0.83 ± 0.26	0.55 ± 0.08
120	2000	153 ± 48.4	0.28 ± 0.00	239 ± 75.2	1.80 ± 0.15	1.33 ± 0.00	1.52 ± 0.37	0.85 ± 0.01

The marked increase (72-fold) in plasma uracil concentration following the administration of TAU (Table 1) could have been due to the saturation of uracil catabolism. The first and rate-limiting enzyme of uracil catabolism in the liver, dihydrouracil dehydrogenase, is a saturable enzyme and is inhibited by high concentrations of its substrate, uracil [30]. The degradation of large amounts of uridine, released from TAU by UrdPase, would increase uracil concentrations. When the uracil concentration reaches the critical saturating limit (about 75 μM), it inhibits dihydrouracil dehydrogenase [30] resulting in the rise in plasma uracil concentration observed after administration of TAU.

Coadministration of PSAU at 30 and 120 mg/kg with 2000 mg/kg TAU decreased plasma uracil C_{max} and AUC by 2.5- and 8.5-, and 6.2- and 19.1-fold, respectively, compared to the values observed with TAU alone (Table 1, Fig. 1). Coadministration of 30 mg/kg with 2000 mg/kg TAU increased the C_{max} of plasma uridine resulting from the administration of TAU alone (507 μM) to over 1200 μM and expanded the AUC of plasma uridine (754 μmol · h/l) by 1.7-fold. Thus, coadministration of PSAU with TAU improved the relative bioavailability of uridine released from TAU (53%) [1] by 2-fold. The increase in the relative bioavailability of uridine released from TAU as a result of coadministration of PSAU was presumably due to inhibition of UrdPase as indicated by the increase in the AUC, C_{max} and C_{max}/C_0 of plasma uridine and the reduction in those of plasma uracil. However, increasing the dose of the coadministered PSAU from 30 to 120 mg/kg failed to improve the relative bioavailability of the uridine released from TAU above that achieved with the lower dose (30 mg/kg) of PSAU (Table 1). Similar results have been observed in monkeys [40] and humans [36].

The inability of higher doses of PSAU, when coadministered with TAU, to increase the bioavailability of uridine released from TAU is not due to a lack of available PSAU. The data shown in Table 2 and Fig. 2 demonstrate that increasing the dose of coadministered PSAU from 30 to 120 mg/kg still increased the C_{max} and AUC of plasma PSAU. Therefore, it is more likely that the effect of PSAU on increasing the bioavailability of uridine released from TAU may have reached a plateau at 30 mg/kg. Future studies using other combinations of lower doses of PSAU and TAU may prove beneficial in modulating plasma uridine bioavailability.

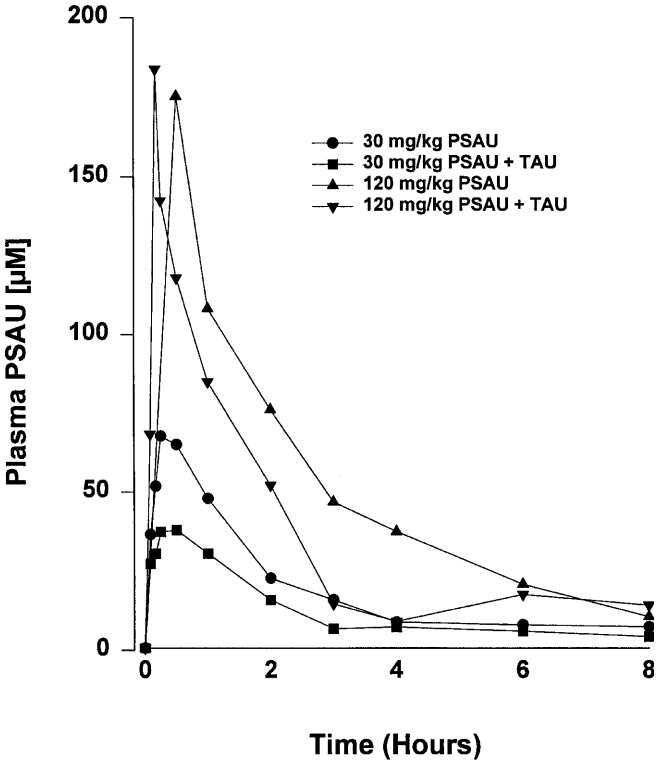

Fig. 2 Plasma concentration-time curves of PSAU after oral administration of PSAU at 30 and 120 mg/kg alone or in combination with TAU (2000 mg/kg) in CD-1 mice. Each point represents the mean from five mice

In conclusion, the combination of PSAU with TAU secured and maintained higher levels of plasma uridine than either alone. The high potency and excellent bioavailability (100%) make PSAU a promising and a convenient modulator of plasma uridine than the toxic massive doses of uridine used to rescue or protect from the host toxicities of certain anticancer and antiviral pyrimidine analogues.

References

1. Ashour OM, Naguib FNM, el Kouni MH (1996) 5-(m-Benzyloxybenzyl)barbituric acid acyclonucleoside, a uridine phosphorylase inhibitor and 2′,3′,5′-tri-o-acetyluridine, a prodrug of uridine as modulators of plasma uridine concentration, Implication for chemotherapy. Biochem Pharmacol 51: 1601

2. Ashour OM, Al Safarjalani ON, Naguib FNM, Goudgaon NM, Schinazi RF, el Kouni MH (2000) Modulation of plasma uridine concentration by 5-(phenylselenenyl)acyclouridine an inhibitor of uridine phosphorylase: relevance to chemotherapy. Cancer Chemother Pharmacol (in press)
3. Chan TCK, Markman M, Pfeifle CE, Taetle R, Abramson I, Howell SB (1988) Uridine pharmacokinetics in cancer patients. Cancer Chemother Pharmacol 22: 83
4. Cradock JC, Vishnuvajjala BR, Chin TF, Hochstein HD, Ackerman TK (1986) Uridine-induced hyperthermia in the rabbit. J Pharm Pharmacol 38: 226
5. Dahnke H-G, Mosebach K-O (1975) In-vivo-Untersuchungen zur Metabolisierung der Pyrimidinnucleoside. Hoppe-Seyler Z Physiol Chem 356: 565
6. Darnowski JW, Handschumacher RE (1985) Tissue-specific enhancement of uridine utilization and 5-fluorouracil therapy in mice by benzylacyclouridine. Cancer Res 45: 5364
7. Darnowski JW, Handschumacher RE (1989) Enhancement of fluorouracil therapy by the manipulation of tissue uridine pools. Pharmacol Ther 41: 381
8. Darnowski JW, Handschumacher RE, Wiegand RA, Goulette FA, Calabresi P (1991) Tissue-specific expansion of uridine pools in mice. Effects of benzylacyclouridine, dipyridamole and exogenous uridine. Biochem Pharmacol 41: 2031
9. Davis ST, Joyner SS, Chandrasurin P, Baccanari DP (1993) Species-dependent differences in the biochemical effects and metabolism of 5-benzylacyclouridine. Biochem Pharmacol 45: 173
10. el Kouni MH, Naguib FNM, Park KS, Cha S, Darnowski JW, Soong S-J (1990) Circadian rhythm of hepatic uridine phosphorylase activity and plasma concentration of uridine in mice. Biochem Pharmacol 40: 2479
11. Falcone A, Darnowski JW, Ruprecht RM, Chu SH, Burnetti I, Calabresi P (1990) Differential Effect of benzylacyclouridine on the toxic and therapeutic effects of azidothymidine in mice. Blood 76: 2216
12. Gasser T, Moyer JD, Handschumacher RE (1981) Novel single-pass exchange of circulating uridine in rat liver. Science 213: 777
13. Gomeni R, Gomeni C (1978) Interactive graphic package for pharmacokinetic analysis. Comput Biomed Res 11: 345
14. Goudgaon NM, Naguib FNM, el Kouni MH, Schinazi RF (1993) Phenylselenenyl and phenylthio substituted pyrimidines as potential inhibitors of dihydrouracil dehydrogenase and uridine phosphorylase. J Med Chem 36: 4250
15. Holstege A, Leser, H-G, Pausch J, Gerok W (1985) Uridine catabolism in Kupffer cells, endothelial cells, and hepatocytes, Eur J Biochem 149: 169
16. Holstege A, Pausch J, Gerok W (1986) Effect of 5-diazouracil on the catabolism of circulating pyrimidines in rat liver and kidneys. Cancer Res 46: 5576
17. Holstege A, Gengenbacher H-M, Jehle L, Gerok W (1992) Uridine catabolism by the isolated perfused rat liver. J Hepatol 14: 335
18. Kelsen DP, Martin D, O'Neil J, Schwartz G, Saltz L, Sung MT, Borstel R von, Bertino J (1997) Phase I trial of PN401, an oral prodrug of uridine, to prevent toxicity from fluorouracil in patients with avoidance responding. J Clin Oncol 15: 1511
19. Keilbauch SA, Hobbs GA, Simpson MV (1993) Anti-human immunodeficiency virus type 1 therapy and peripheral neuropathy: prevention of 2′,3′-dideoxycytidine toxicity in PC12 cells, a neuronal model by uridine and pyruvate. Mol Pharmacol 44: 702
20. Klubes P, Cerna I (1983) Use of uridine rescue to enhance the antitumor selectivity of 5-fluorouracil. Cancer Res 43: 3182
21. Klubes P, Cerna I, Meldon MA (1982) Uridine rescue from the lethal toxicity of 5-fluorouracil in mice. Cancer Chemother Pharmacol 8: 17
22. Klubes P, Geffen DB, Cysyk RL (1986) Comparison of the bioavailability of uridine in mice after either oral or parenteral administration. Cancer Chemother Pharmacol 17: 236
23. Leyva A, van Groeningen CJ, Kraal I, Peters GJ, Lankelma J, Pinedo HM (1984) Phase I and pharmacokinetic studies of high-dose uridine intended for rescue from 5-fluorouracil toxicity. Cancer Res 44: 5928
24. Marquardt DW (1963) An algorithm for least squares estimation of nonlinear parameters. J Soc Ind Appl Math 11: 431
25. Martin DS, Stolfi RL, Sawyer RC, Spiegelman S, Young CW (1982) High-dose 5-fluorouracil with delayed uridine "rescue" in mice. Cancer Res 42: 3964
26. Martin DS, Stolfi RL, Sawyer RC (1989) Use of oral uridine as a substitute for parenteral uridine rescue of 5-fluorouracil therapy, with and without the uridine phosphorylase inhibitor 5-benzylacyclouridine. Cancer Chemother Pharmacol 24: 9
27. Monks A, Cysyk RL (1982) Uridine regulation by the isolated rat liver: perfusion with an artificial oxygen carrier. Am J Physiol 242: R465
28. Monks A, Ayers O, Cysyk RL (1983) Effect of 5-benzylacyclouridine, a potent inhibitor of uridine phosphorylase, on the metabolism of circulating uridine by the isolated rat liver. Biochem Pharmacol 32: 2003
29. Moyer JD, Oliver JT, Handschumacher RE (1981) Salvage of circulating pyrimidine nucleosides in the rat. Cancer Res 41: 3010
30. Naguib FNM, el Kouni MH, Cha S (1985) Enzymes of uracil catabolism in normal and neoplastic human tissues. Cancer Res 45: 5405
31. Naguib FNM, Soong S-J, el Kouni MH (1993) Circadian rhythm of orotate phosphoribosyltransferase, pyrimidine nucleoside phosphorylases and dihydrouracil dehydrogenase in mouse liver: possible relevance to chemotherapy with 5-fluoropyrimidines. Biochem Pharmacol 45: 667
32. Peters GJ, van Groeningen CJ (1991) Clinical relevance of biochemical modulation of 5-fluorouracil. Ann Oncol 2: 469
33. Peters GJ, van Groeningen CJ, Laurensse EJ, Lankelma J, Kraal I, Leyva A, Lankelma J, Pinedo HM (1987) Effect of pyrimidine nucleosides on body temperatures of man and rabbit in relation to pharmacokinetic data. Pharm Res 4: 113
34. Peters GJ, van Groeningen CJ, Laurensse E, Levya A, Pinedo HM (1987) Uridine-induced hypothermia in mice and rats in relation to plasma and tissue levels of uridine and its metabolites. Cancer Chemother Pharmacol 20: 101
35. Peters GJ, van Dijk J, Laurensse E, van Groeningen CJ, Lankelman J, Levya A, Nadal JC, Pinedo HM (1988) In vitro biochemical and in vivo biological studies of the uridine "rescue" of 5-fluorouracil. Br J Cancer 57: 259
36. Pizzorno G, Yee L, Burtness BA, Marsh JC, Darnowski JW, Chu MYW, Chu HS, Chu E, Leffert JJ, Handschumacher RE, Calabresi P (1998) Phase I clinical and pharmacological studies of benzylacyclouridine, a uridine phosphorylase inhibitor. Clin Cancer Res 4: 1165
37. Seiter K, Kemeny N, Martin D, Schneider A, Williams L, Colofiore J, Sawyer R (1993) Uridine allows dose escalation of 5-fluorouracil when given with N-phosphoacetyl-l-aspartate, methotrexate, and leucovorin. Cancer 71: 1875
38. Sommadossi J-P, Carlisle R, Schinazi RF, Zhou Z (1988) Uridine reverses the toxicity of 3′-azido-3′-deoxythymidine in normal human granulocyte-macrophage progenitor cells in vitro without impairment of antiretroviral activity. Antimicrob Agents Chemother 32: 997
39. Sommadossi J-P, Zhu Z, Carlisle R, Xie MY, Weidner DA, el Kouni MH (1990) Novel pharmacologic approaches for the treatment of AIDS and potential use of uridine phosphorylase inhibitors. In: Diasio RB, Sommadossi J-P (eds) Advances in chemotherapy of AIDS. Pergamon Press, New York, p 63
40. Sommadossi J-P, Cretton EM, Kidd LB, McClure HM, Anderson DC, el Kouni MH (1995) Effects of 5-benzylacyclouridine, an inhibitor of uridine phosphorylase, on the pharmacokinetics of uridine in rhesus monkeys: implications for chemotherapy. Cancer Chemother Pharmacol 37: 14
41. van Groeningen CJ, Leyva A, Kraal I, Peters GJ, Pinedo HM (1986) Clinical and pharmacokinetic studies of prolonged

administration of high-dose uridine intended for rescue from 5-FU toxicity. Cancer Treat Rep 70: 745
42. van Groeningen CJ, Peters GJ, Leyva A, Laurensse E, Pinedo HM (1989) Reversal of 5-fluorouracil induced myelosuppression by prolonged administration of high-dose uridine. J Natl Cancer Inst 81: 157
43. van Groeningen CJ, Peters GJ, Nadal JC, Leyva A, Laurensse E, Pinedo HM (1991) Clinical and pharmacologic study of orally administration uridine. J Natl Cancer Inst 83: 437
44. van Groeningen CJ, Peters GJ, Pinedo HM (1992) Modulation of fluorouracil toxicity with uridine. Semin Oncol 19: 148