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Low Dose 1-Hexylcarbamoyl-5-fluorouracil (HCFU) Recommended for Cirrhotic Patients with Hepatocellular Carcinoma

KENJI TAKENAKA, TAKASHI KANEMATSU, MITSUO SHIMADA and KEIZO SUGIMACHI

Second Department of Surgery, Kyushu University, 3-1-1 *Maidashi, Higashi-ku, Fukuoka* 812, *Japan*

Abstract—*The metabolism of* 1-*hexylcarbamoyl-5-fluorouracil* (*HCFU*), *a drug prescribed for treating patients with hepatocellular carcinoma* (*HCC*), *was studied in relation to liver function, with the objective of clarifying the occurrence of any adverse side-effects on the central nervous system. Twenty-five HCC patients were administered* 3.4 mg/kg *HCFU once orally, after which the blood levels of HCFU and its derivatives* (5-*FU, CPEFU, CPRFU, HHCFU, OHCFU and F-β-alanine*) *were serially measured using high performance liquid chromatography. The area under the concentration curve* (*AUC*) *of HCFU in the group of ICG R*15 ⩾ 30% (*group* 2) *was* 5.35 ± 1.73 h.μg/ml, *a value which was significantly higher than the* 2.60 ± 1.19 h.μg/ml *recorded for the group of ICG R*15 < 30% (*group* 1) (P < 0.001). *The AUC of HCFU had a significant positive correlation with the value of ICG R*15 (P = 0.002) *or the serum total bilirubin* (P = 0.0005). *The AUC of* 5-*FU showed no difference between the two groups. The AUC of CPRFU in group* 2 *was* 0.16 ± 0.25 h.μg/ml, *a value significantly lower than the* 0.48 ± 0.39 h.μg/ml *in group* 1 (P = 0.023). *There was no correlation between the AUC of other derivatives and the markers of liver function.*

These data suggest that, in patients with advanced cirrhosis, the accumulation of HCFU is related to the occurrence of side-effects from the administered drug, ingested over a long-term period. Therefore, when HCFU is given to cirrhotic patients with both HCC and 30% *or more ICG R*15, *a careful monitoring for side-effects is required.*

INTRODUCTION

THE anticancer drug 1-hexylcarbamoyl-5-fluorouracil (HCFU) is a derivative of 5-fluorouracil (5-FU) [1]. Because the activation of this drug is due to non-enzymatic natural decomposition as well as enzymatic oxidation in the liver [2, 3], the blood level of 5-FU can be maintained even in cirrhotic patients [4]. This activation mechanism differs considerably from the metabolic pathway of another masked compound of 5-FU, 1-(2-tetrahydrofuryl)-5-fluorouracil (Tegafur), which is converted enzymatically to 5-FU by cytochrome P-450 in liver microsomes [5]. Therefore, HCFU is considered to be an effective drug for the treatment of patients with solid tumors [6] including hepatocellular carcinoma (HCC) [7, 8] associated with impaired liver function or liver cirrhosis.

However, these compounds tend to have adverse effects on the central nervous system [9] and the incidence is particularly high in cirrhotic patients [7]. In an attempt to determine the optimal dosage of HCFU for the most effective administration to cirrhotic patients with HCC, we studied HCFU metabolism in these patients.

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All correspondence should be sent to: Kenji Takenaka, M.D., Second Department of Surgery, Kyushu University, 3-1-1 Maidashi, Higashi-ku, Fukuoka 812, Japan.

PATIENTS AND METHODS

Twenty-five patients suffering from HCC either with or without liver cirrhosis were evaluated. The patient's informed consent for the study was obtained in each case. The patients were divided into two groups according to their indocyanine green retention rate at 15 min (ICG R15): group 1 had a rate of ICG R15 <30% (13 patients), while group 2 had a rate of ICG R15 ⩾30% (12 patients). All related background factors are shown in Table 1. All the patients in group 2 had associated liver cirrhosis which had been proven either clinically or histologically from tissue specimens obtained at the time of surgery.

HCFU was administered once orally at a dose of 3.4 mg/kg body wt before a meal, at a dosage corresponding to about 600 mg/day. Blood samples

Table 1. Background of patients

		Group 1 (n = 13)	Group 2 (n = 12)	P†
Age		62.4 ± 8.0*	61.8 ± 6.6	n.s.
Sex (M:F)		10:3	12:0	—
Body wt (kg)		55.8 ± 9.0	57.6 ± 7.3	n.s.
Biohemical findings				
ICG R15	(%)	12.5 ± 7.7	42.2 ± 8.8	$P < 0.001$
PT	(%)	92 ± 13	60 ± 18	$P < 0.001$
LCAT	(μg/ml/h)	30.0 ± 12.3	18.5 ± 9.2	$P < 0.05$
γ-glob	(%)	20.7 ± 8.4	29.0 ± 9.0	$P < 0.05$
Alb	(g/dl)	3.68 ± 0.63	3.42 ± 0.44	n.s.
T.bil	(mg/dl)	0.64 ± 0.29	1.73 ± 0.74	$P < 0.001$
Chol	(mg/dl)	181 ± 37	145 ± 30	$P < 0.05$
γ-GTP	(U/l)	63.9 ± 58.8	76.6 ± 47.8	n.s.
BUN	(mg/dl)	13.3 ± 4.6	13.7 ± 2.0	n.s.
S.Cr.	(mg/dl)	0.97 ± 0.19	0.97 ± 0.11	n.s.

*Mean ± S.D.

†t-test.

Abbreviations: ICG R15, indocyanine green retention rate at 15 min; PT, prothrombin time; LCAT, lecithin cholesterol acyltransferase; S.Cr., serum creatinine; n.s., not significant.

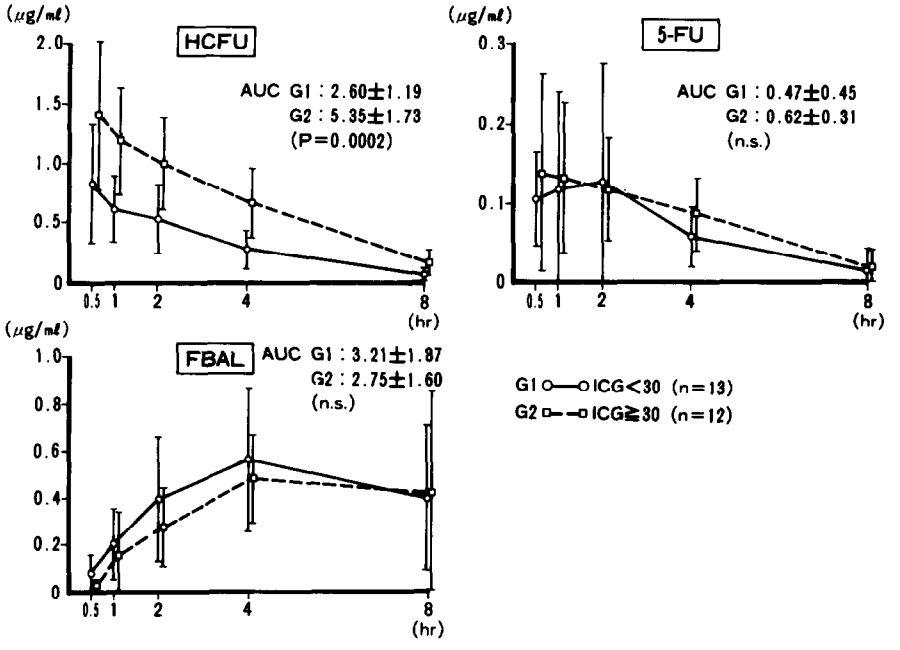

Fig. 1. Serum levels of HCFU, 5-FU and FBAL after oral administration of HCFU. HCFU was administered once orally at a dosage of 3.4 mg/kg *body wt. Blood samples were collected at indicated times and levels of metabolites were measured. Abbreviations: G1, group 1; G2, group 2; AUC, area under concentration curve.*

for analysis were collected before and at 0.5, 1, 2, 4 and 8 h after administration. The patient was given food after the blood collection at 2 h. After the serum was immediately separated and two drops of 6 N HCl were added, then the samples were stored at −20°C until analyzed.

Analysis of 5-FU

One millilitre of serum was put into a test tube containing 5-bromouracil, as an internal standard. After adding 0.5 ml of 0.5 N HCl and 6 ml of $CHCl_3$, the aqueous layer was separated and then evaporated completely to a state of dryness. After dissolving the residue in 0.5 ml of 1 M phosphate buffer solution (pH 7.0), 10 ml of ethyl acetate was added, then the preparation was centrifuged to separate the ethyl acetate layer and finally it was again completely evaporated. The level of 5-FU in the residue was measured using high performance liquid chromatography (HPLC).

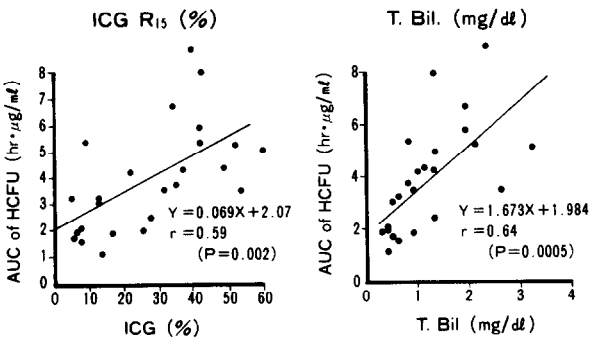

Fig. 2. Correlation between AUC (0–8 h) of HCFU and ICG R15, serum total bilirubin. HCFU was administered once orally at a dosage of 3.4 mg/kg *body wt. AUC (area under concentration curve) was calculated within* 8 h *after administration.*

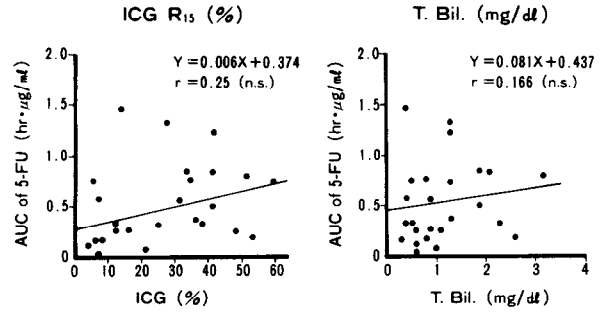

Fig. 3. Correlation between AUC (0–8 h) of 5-FU and ICG R15, serum total bilirubin. See also Fig. 2.

Analyses of HCFU and its metabolites

After adding 0.5 ml of 0.6 N HCl and 8 ml of ethyl acetate to 1.0 ml serum, the sample was extracted by vigorous shaking. The organic layer was separated by centrifugation and evaporated to dryness. The levels of HCFU, 1-carboxypentylcarbamoyl-5-fluorouracil (CPEFU), 1-carboxypropylcarbamoyl-5-fluorouracil (CPRFU), 1-(5-hydroxyhexylcarbamoyl)-5-fluorouracil (HHCFU) and 1-(5-oxohexylcarbamoyl)-5-fluorouracil (OHCFU) in the residue were respectively measured, using HPLC.

Analysis of α-fluoro-β-alanine (FBAL)

Serum (0.2 ml) was put into a test tube containing *o*-fluoro-DL-phenylalanine (O-FPAL), as an internal standard. After adding 2 ml of 2.0 N HCl and 2 ml of $CHCl_3$, the aqueous layer was separated and evaporated to dryness. The obtained residue was treated as described [10, 11] and then the level of FBAL was measured using gas chromatographic–mass spectrometry.

The statistical analysis was performed using Student's *t*-test.

RESULTS

The serum levels of HCFU, 5-FU and FBAL after the oral administration of HCFU are shown in Fig. 1. The area under the concentration curve (AUC) from 0.5 to 8 h after the administration of HCFU in group 2 patients was about twice of that of group 1 patients. However, no difference was found in the AUC of 5-FU or FBAL between the two groups. The correlations between the AUC of the HCFU and the liver function tests in all 25 patients are shown in Fig. 2. The AUC of the HCFU increased with a significant positive correlation to the ICG R15 and to serum total bilirubin. On the other hand, no significant correlation was found between either the AUC of 5-FU and the ICG R15 or the AUC of 5-FU and serum total bilirubin in these 25 patients (Fig. 3). The serum levels of HCFU oxidative metabolites after the oral administration of HCFU are shown in Fig. 4. No difference was found in the AUC of the CPEFU, HHCFU, or OHCFU between the two groups. The AUC of the CPRFU, however, was significantly lower in group 2 patients, being only one-third of the AUC in group 1 patients.

DISCUSSION

In our study, both the accumulation of HCFU and the decreased production of CPRFU were found in advanced cirrhotic patients. This is mainly caused by decreased enzymatic oxidation activity of HCFU during impaired liver function, because CPRFU is produced from CPEFU by β-oxidation in the liver [3]. On the contrary, Onji *et al.* [12] and Okamoto *et al.* [13] reported that 5-FU itself accumulated after oral administration to cirrhotic patients and also in experimental animals with impaired liver function, due to the lower activity of dihydrouracil dehydrogenase, a catalytic enzyme of 5-FU, present in the liver. In our experiment, the 5-FU tended to accumulate in those with advanced cirrhosis, as the value of AUC of 5-FU was still higher in group 2 patients, albeit with no statistical significance.

Concerning the problem of side-effects on the central nervous system and HCFU metabolism in advanced cirrhotic patients, firstly, HCFU itself correlates to the occurrence of these side-effects. HCFU is a lipophilic drug and has been shown in mice to cross the blood–brain barrier [14]. Secondly, 5-FU causes localized changes in blood–brain barrier permeability [15] and also neurotoxicity [16, 17]. Thirdly, Koenig and Patel [18] reported on the inhibition of the Krebs cycle by fluoroacetate metabolized from FBAL. Okeda *et al.* [19] also found that FBAL played a crucial role in neurotoxic actions in dogs which were administered HCFU orally for 6 months. However, in our study, no differences were found in the AUC of the FBAL between the two groups.

We usually administer HCFU orally at a dosage of 600 mg/day to non-cirrhotic patients. However,

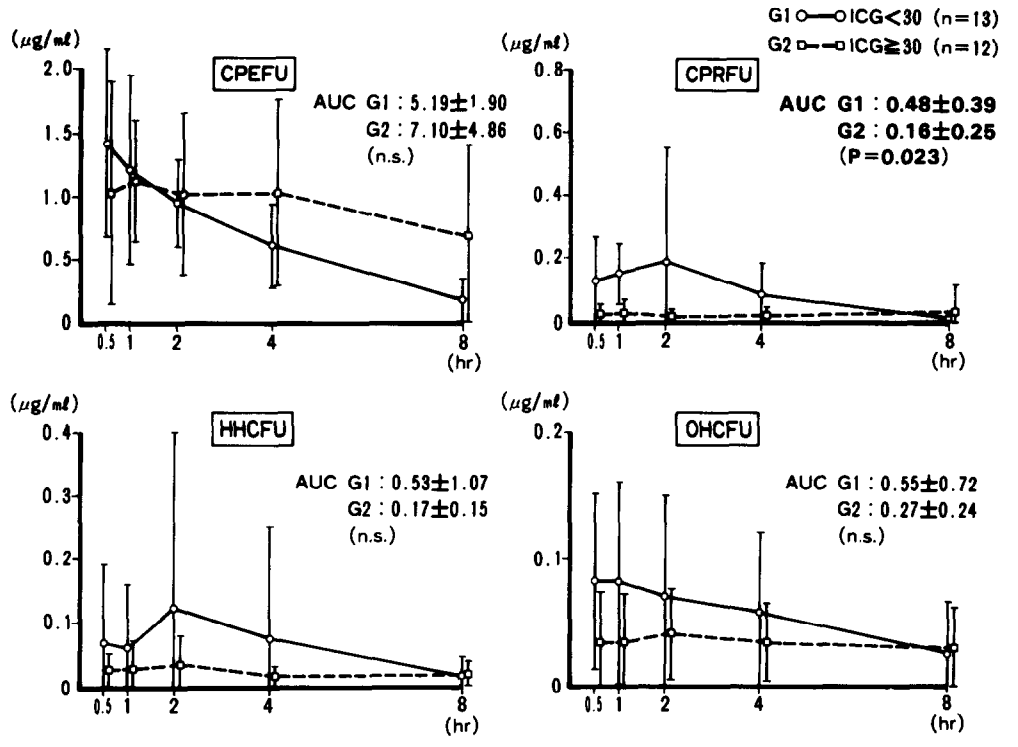

Fig. 4. Serum levels of HCFU oxidative metabolites after oral administration of HCFU. See also Fig. 1.

HCFU at a dosage of 300–400 mg/day may be sufficient and preferable for those with an advanced cirrhosis of 30% or more of the ICG R15 in order to avoid any potentially adverse side-effects, since we were able to obtain an effective 5-FU blood level of 0.05 μg/ml [20], even when the dosage itself was reduced by half.

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