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Anticachectic Activity of 5′-Deoxy-5-fluorouridine in a Murine Tumor Cachexia Model, Colon 26 Adenocarcinoma

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ABSTRACT

Murine colon 26 adenocarcinoma causes a progressive weight loss and physiological changes associated with cachexia when it grows to a certain size. By the use of this tumor model several types of cytostatics were examined for their ability to alleviate cachexia. Among them, 5′-deoxy-5-fluorouridine could reverse a progressive weight loss and improve hypoglycemia, hyperglucocorticism, and hepatic malfunctions, as well as inhibiting the tumor growth. Cyclophosphamide, nimustine, and 2′-deoxy-5-fluorouridine were only slightly effective in reversing the wasting, while 5-fluorouracil, tegafur, mitomycin C, *cis*-platinum, and doxorubicin were not active. Within 3 days after 5′-deoxy-5-fluorouridine was administered to cachectic mice with large tumor burdens, the wasting was immediately reversed even at doses in which there was increase or no significant reduction in tumor growth. These results indicate that the anticachectic activity of 5′-deoxy-5-fluorouridine is independent of its antiproliferative activity.

INTRODUCTION

Cachexia, which includes progressive wasting, weakness, anorexia, anemia, etc., is commonly seen in cancer patients (1). Although each of these symptoms is not so serious, this "tumor cachexia" has a negative impact on the prognosis of cancer patients. Cachexia is associated not only with the deterioration of the quality of life but also with shorter survival. DeWys *et al.* (2) have reported that the survival time of patients with most tumor types who had experienced substantial prechemotherapy weight loss, a typical symptom of cachexia, was much shorter than for those who had not experienced weight loss. His group reported that prechemotherapy wasting is also associated with a lower response rate to chemotherapy in breast cancer patients (3). Cachexia is one of the major obstacles in cancer chemotherapy and an appropriate treatment modality is necessary to counteract it.

We have tried to identify tumor lines for elucidating the mechanisms which cause cachexia and for discovering new treatment modalities for tumor cachexia. Among various murine transplantable tumor lines widely utilized for the assessment of cytostatics, we identified murine colon 26, which is an undifferentiated carcinoma induced by the carcinogen *N*-nitroso-*N*-methylurethan, as capable of causing cachexia (4). Colon 26 causes physiological changes associated with cachexia, such as adipose tissue and whole body wasting, hypoglycemia, hypercorticism, reduction of some liver functions, increase of acute phase proteins, etc., while tumor burdens are relatively small. Colon 26-bearing mice would be a suitable model for the identification of a treatment modality for cachexia like other cachexia models, such as MCG101 methylcholanthrene-induced sarcoma of mouse (5, 6), MCA sarcoma of rat (7, 8), MAC16 1,2-dimethylhydrazine-induced adenocarcinoma of mouse (9, 10), and Walker 256 carcinoma of rat (11, 12).

Although tumor cachexia is one of the major obstacles in cancer therapies, no specific modalities have been identified. Only nutritional controls, such as hyperalimentation, have partially counteracted the deterioration of the quality of life (1). Concerning a chemotherapeutic approach to the treatment of cachexia, hydrazine sulfate, which is an inhibitor of the key enzyme of gluconeogenesis phosphoenolpyruvate carboxykinase, has been reported to increase the survival of cancer patients (13). In addition, the cytostatic CPA[2] was reported to reverse the anorexia and cachexia in an experimental model (11). We have been interested in identifying anticachetic agents from various kinds of existing drugs by using the murine colon 26 carcinoma in mice and found that indomethacin can improve colon 26-induced cachexia (14). Here we studied the effectiveness of various cytostatics for alleviating cachectic symptoms in the above tumor model. Our studies revealed that the cachexia was improved by a new fluorinated pyrimidine nucleoside, 5′-dFUrd, and to some extent by CPA and ACNU. In addition, the anticachectic activity of some cytostatics, particularly that of 5′-dFUrd, was analyzed in relation to their antitumor activity.

MATERIALS AND METHODS

Mice. Four-week-old male BALB/c × DBA/2 F_1 (hereafter called $CD2F_1$) mice were obtained from SLC, Hamamatsu, Japan. The mice were used at the age of 5 weeks. They were fed on breeding diet F1 (Funabashi Farm, Funabashi, Japan) and water *ad libitum*.

Tumors. Murine colon 26 adenocarcinoma cells were cultured *in vitro* with RPMI 1640 containing 10% fetal calf serum. By treating the cells with trypsin, we obtained a single cell suspension, which was then inoculated s.c. into the right inguinal flank of $CD2F_1$ mice (10^6 cells/mouse).

Drugs and Treatment. CPA (Shionogi Co., Ltd., Osaka, Japan), ACNU (Sankyo Co., Ltd., Tokyo, Japan), mitomycin C (Kyowa Hakko Co., Ltd., Tokyo, Japan), doxorubicin (Kyowa Hakko Co., Ltd., Tokyo, Japan) or CDDP (Nippon Kayaku Co., Ltd., Tokyo, Japan) were dissolved in saline and administered i.p. to mice on the days indicated in the text. 5-FUra (Kyowa Hakko Co., Ltd., Tokyo, Japan), 2′-dFUrd (Hoffmann-La Roche Inc., Nutley, NJ), tegafur (Taiho Pharm. Co., Ltd., Tokushima, Japan), and 5′-dFUrd (Nippon Roche K. K., Tokyo, Japan) were dissolved in 0.5% carboxymethylcellulose and were administered p.o. to mice.

Measurement of Body Wasting. Body weight and the length (*a*) and width (*b*) of the tumors were measured 2 or 3 times a week. The tumor weight was sometimes estimated by first calculating the tumor volume ($ab^2/2$) and multiplying this by a correction factor. The correction factor was determined by comparing actual tumor weights with the tumor volume in separate experiments. Carcass weight, the difference in weight between the whole body and tumor tissue, was calculated. In order to determine the extent of tissue wasting, we measured the change in weight of the left epididymal adipose tissues.

Assays. Blood samples were collected from the heart via puncture for most assays and from the orbital veins for an assay of corticosterone level. The samples for the latter assay were collected between 9 and 10 a.m. to minimize the fluctuation of the hormone level based on a circadian rhythm. For the determination of the concentration of sub-

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[2] The abbreviations used are: CPA, cyclophosphamide; 5′-dFUrd, 5′-deoxy-5-fluorouridine; ACNU, nimustine; CDDP, *cis*-platinum; 5-FUra, 5-fluorouracil; 2′-dFUrd, 2′-deoxy-5-fluorouridine; IAP, immunosuppressive acidic protein.

stances in serum or plasma to be tested, the following methods and reagents were used: a color reaction method with *o*-toluidine for measuring glucose (15); a clotting time test with thrombin for measuring fibrinogen in plasma (16); an immunodiffusion assay with anti-mouse IAP antibody (Saikin Kagaku Institute, Sendai, Japan) for measuring IAP (17); sialic acid contents of plasma proteins with an assay kit (Kyowa Medix, Tokyo, Japan) (18); a radioimmunoassay method with anti-corticosterone antibody (Paesel GmbH & Co., Frankfurt, West Germany) and [^{3}H]corticosterone (New England Nuclear, Boston, MA) for measuring serum levels of corticosterone (19).

A liver homogenate was prepared with 0.05 M phosphate buffer, and its catalase activity and the amount of P-450 drug-metabolizing enzymes were determined by an H_2O_2 consumption assay (20) and by a spectrophotometric assay as described by Omura and Sato (21), respectively.

Statistics. Differences in tumor size, tissue weight, enzyme activities, and concentrations of substances were compared by using the Mann-Whitney *U* test. Differences were considered to be significant when the probability value was less than 0.05 ($P < 0.05$).

RESULTS

Recovery from Tumor-induced Wasting by Cytostatics. Two to 3 weeks after s.c. inoculation of the tumor, mice bearing colon 26 tumor became cachectic and lost weight when the tumor grew up to about 2 g as reported previously (4). On day 20 after the tumor inoculation, the difference in the carcass weight between the tumor-bearing mice and the age-matched controls reached 10 g, and the adipose tissue weight of the cachectic mice was 9 ± 3 (SD) mg compared with 304 ± 43 mg of the control mice. As Table 1 shows, CPA, ACNU, CDDP, and 5'-dFUrd given to the cachectic mice substantially suppressed the tumor growth. Particularly 5'-dFUrd at higher doses was able to reduce the tumor burdens. 5'-dFUrd was also capable of reversing the whole body and the adipose tissue wasting even at doses in which there was some increase (0.22 mmol/kg) or no significant reduction (0.44 mmol/kg) in the tumor size. CPA and ACNU reversed the wasting slightly, whereas CDDP did not. The other cytostatics, mytomycin C, doxorubicin, and 5-FUra, did not have either antitumor or anticachectic activity.

Effects of Fluorinated Pyrimidines on the Wasting and Physiological Changes Associated with Cachexia. Since 5'-dFUrd is a prodrug of 5-FUra, we compared the abilities of the four fluorinated pyrimidines 5-FUra, 2'-dFUrd, tegafur, and 5'-dFUrd to reverse carcass and adipose tissue wasting and to alleviate the physiological changes associated with cachexia, such as hypoglycemia and hypercorticism. As Table 2 shows, 5'-dFUrd, 2'-dFUrd, and tegafur at particular doses suppressed the tumor growth. 5'-dFUrd reversed the wastings and alleviated hypoglycemia and hypercorticism. Even at a dose (0.125 mmol/kg) allowing for progressive tumor growth, 5'-dFUrd improved the features of cachexia. In addition, 5'-dFUrd improved these parameters associated with cachexia only in tumor-bearing mice, whereas it did not show any significant effect on the parameters of age-matched normal mice at doses of 0.5 and 1.0 mmol/kg daily for 7 days (data not shown). On the other hand, 2'-dFUrd only at a dose of 0.25 mmol/kg slightly alleviated the all four features. Tegafur improved only hypoglycemia and hypercorticism at a dose of 1.5 mmol/kg, in which the tumor growth was inhibited, whereas it could not reverse the carcass and adipose tissue wasting at any doses tested. 5-FUra was least effective among the group in terms of both antitumor and anticachectic activities.

Kinetics of Reversing the Body Wasting by 5'-dFUrd. In an attempt to investigate whether the antitumor activity of 5'-dFUrd is related to its anticachectic activity, we precisely examined the improvement of the body wasting and the antitumor activity of this drug. Fig. 1 shows growth curves of the tumor and the fluctuation of the carcass weight in mice bearing large burdens of colon 26 carcinoma. 5'-dFUrd suppressed the tumor growth in a dose-dependent manner, while it immediately reversed the body wasting even when the tumor size was progressively increasing or not significantly reduced (0.125 and 0.25 mmol/kg). These results indicate that the recovery from cachexia by 5'-dFUrd is not the result of the reduction of tumor size.

Recovery by 5'-dFUrd from Other Physiological Changes Associated with Cachexia. 5'-dFUrd was then investigated to see whether it is capable of improving disorders of hepatic functions, which are possibly associated with cachexia in mice bearing colon 26 carcinoma (4). Mice bearing the tumor were given 5'-dFUrd when the weight loss was about the maximum. As Table 3 shows, the plasma concentration of the acute phase

Table 1 *Effect of various cytostatics on tumor, carcass, and adipose tissue weight of colon 26-bearing mice*

Groups of 6–8 mice were inoculated s.c. with colon 26 on day 0. The mice were given cytostatics either i.p. on days 20, 24, and 28 (experiment 1) or p.o. every day from day 20 for 10 days (experiment 2). The estimated tumor weight and carcass weight were determined on day 28. The weight of epididymal adipose tissue was measured on day 30. The tumor weight and carcass weight on day 20 (before treatment) were 1.82 ± 0.32 g and 17.22 ± 1.38 g, respectively.

Drug	Dose	Tumor wt on day 28 (g)	Carcass wt change (g)[a]	Adipose tissue wt (mg)
Experiment 1 (mg/kg)				
Vehicle		3.86 ± 0.41[b]	−0.98 ± 0.98	8.4 ± 2.8
CPA	25	3.87 ± 0.82	−0.55 ± 1.53	9.7 ± 4.6
	50	3.86 ± 0.85	0.92 ± 1.29[c]	12.4 ± 1.9[c]
	100	2.89 ± 0.43[c]	1.23 ± 1.04[c]	20.0 ± 3.1[c]
	200	1.93 ± 0.19[c]	0.97 ± 1.00[c]	39.6 ± 13.6[c]
	400	1.57 ± 0.41[c]	−1.71 ± 0.99	13.0 ± 5.0
ACNU	7.5	3.26 ± 0.51[c]	1.14 ± 0.74[c]	23.6 ± 5.8[c]
	15	2.17 ± 0.29[c]	0.94 ± 1.63[d]	26.0 ± 5.8[c]
	30	2.03 ± 0.29[c]	−0.86 ± 1.58	22.7 ± 9.3[c]
	60	(toxic)[e]		
Mitomycin C	1	3.83 ± 0.50	−0.01 ± 1.02	10.8 ± 3.9
	2	3.54 ± 0.76	−0.21 ± 0.82	15.2 ± 6.0[d]
	4	3.21 ± 0.75[d]	0.12 ± 1.30	7.8 ± 4.8
	8	(toxic)		
CDDP	1.25	3.75 ± 0.77	−1.07 ± 1.86	9.6 ± 3.8
	2.5	4.11 ± 0.45	−0.23 ± 1.30	9.2 ± 3.3
	5	3.26 ± 0.39[c]	−0.70 ± 2.27	12.0 ± 2.6[d]
	10	2.68 ± 0.33[c]	−1.52 ± 1.12	8.0 ± 2.1
Doxorubicin	2.5	3.85 ± 0.63	−1.48 ± 0.57	12.4 ± 3.1[d]
	5	3.22 ± 0.69[d]	−0.45 ± 0.87	12.5 ± 3.8[d]
	10	(toxic)		
Experiment 2 (mmol/kg)				
Vehicle		3.96 ± 0.66	−0.55 ± 1.07	7.6 ± 3.1
5-FUra	0.1	3.82 ± 0.33	−0.81 ± 0.67	6.5 ± 1.7
	0.2	3.51 ± 0.51	−0.13 ± 1.32	8.0 ± 2.8
	0.4	(toxic)		
5'-dFUrd	0.11	2.85 ± 0.33[c]	0.60 ± 0.77[d]	12.7 ± 6.2[d]
	0.22	2.53 ± 0.47[c]	1.56 ± 1.28[c]	30.5 ± 7.2[c]
	0.44	1.63 ± 0.51[c]	3.80 ± 1.71[c]	71.9 ± 44.7[c]
	0.88	1.50 ± 0.18[c,f]	2.86 ± 1.59[c]	44.3 ± 33.1[c]
	1.75	1.37 ± 0.29[c,f]	−1.58 ± 1.16	5.2 ± 2.1
	3.5	0.98 ± 0.31[c,f]	−1.98 ± 1.80	4.9 ± 1.9
Non-tumor-bearing mice			0.59 ± 0.58	250.3 ± 55.8[c]

[a] The difference of the carcass weight between days 20 and 28.
[b] Mean ± SD.
[c] $P < 0.01$, compared with vehicle-treated tumor-bearing mice.
[d] $P < 0.05$, compared with vehicle-treated tumor-bearing mice.
[e] The treatment group in which mortality rate was higher than 30% was judged as toxic.
[f] $P < 0.01$, compared with tumor-bearing mice of day 20.

Table 2 *Effects of fluorinated pyrimidines on tumor, carcass, and adipose tissue weight and the levels of serum glucose and glucocorticoid in mice bearing colon 26 adenocarcinoma*

Groups of 6 mice were inoculated s.c. with colon 26 on day 0. The mice were then given fluorinated pyrimidines p.o. every day from day 22 for 7 days. Assays and measurements were done on day 29, one day after the final administration. The tumor weight and the carcass weight on day 22 (before treatment) were 1.22 ± 0.18 g and 18.51 ± 1.17 g, respectively.

Drug	Dose (mmol/kg)	Tumor wt on day 29 (g)	Carcass wt change[a] (g)	Adipose tissue wt (mg)	Serum glucose (mg/dl)	Serum glucocorticoid (ng/ml)
Vehicle		2.21 ± 0.36[b]	−0.25 ± 0.84	12 ± 3	95 ± 26	290 ± 101
5-FUra	0.038	2.56 ± 0.57	0.24 ± 0.64	9 ± 5	98 ± 17	248 ± 88
	0.075	2.65 ± 0.62	0.28 ± 0.88	13 ± 5	84 ± 21	230 ± 146
	0.15	2.62 ± 0.60	0.19 ± 1.12	13 ± 7	96 ± 19	179 ± 48[c]
	0.3	2.15 ± 0.69	1.00 ± 0.47[c]	12 ± 2	111 ± 12	174 ± 60[c]
2'-dFUrd	0.063	2.39 ± 0.29	0.99 ± 0.59[c]	15 ± 5	90 ± 20	243 ± 119
	0.125	1.84 ± 0.32[c]	0.40 ± 0.84	14 ± 2	94 ± 17	210 ± 93
	0.25	1.59 ± 0.25[d]	1.53 ± 0.70[c]	22 ± 6[d]	121 ± 18[c]	119 ± 23[d]
	0.5	1.05 ± 0.19[d,e]	0.51 ± 1.64	16 ± 5	120 ± 5	130 ± 16[c]
Tegafur	0.19	2.55 ± 0.54	−0.42 ± 0.99	12 ± 6	100 ± 15	312 ± 86
	0.38	2.55 ± 0.96	0.18 ± 0.79	21 ± 8[d]	118 ± 25	269 ± 148
	0.75	1.92 ± 0.61	0.21 ± 1.02	14 ± 3	111 ± 28	179 ± 47[c]
	1.5	1.08 ± 0.45[d]	−0.62 ± 1.42	8 ± 2	137 ± 18[d]	133 ± 16[d]
5'-dFUrd	0.125	1.75 ± 0.35[c]	2.72 ± 1.07[d]	29 ± 9[d]	117 ± 21	152 ± 29[d]
	0.25	1.24 ± 0.18[d]	4.71 ± 1.20[d]	74 ± 15[d]	143 ± 7[d]	117 ± 15[d]
	0.5	1.16 ± 0.38[d,e]	5.70 ± 0.63[d]	104 ± 28[d]	147 ± 17[d]	130 ± 16[d]
	1.0	1.07 ± 0.24[d,e]	3.75 ± 2.00[d]	41 ± 20[d]	122 ± 24[c]	113 ± 15[d]
Tumor-free mice			0.08 ± 2.39	284 ± 73[d]	168 ± 8[d]	129 ± 33[d]

[a] The difference of the carcass weight between days 22 and 29.
[b] Mean ± SD.
[c] $P < 0.05$, compared with vehicle-treated tumor-bearing mice.
[d] $P < 0.01$, compared with vehicle-treated tumor-bearing mice.
[e] No significant reduction in tumor weight compared with the tumor weight on day 22.

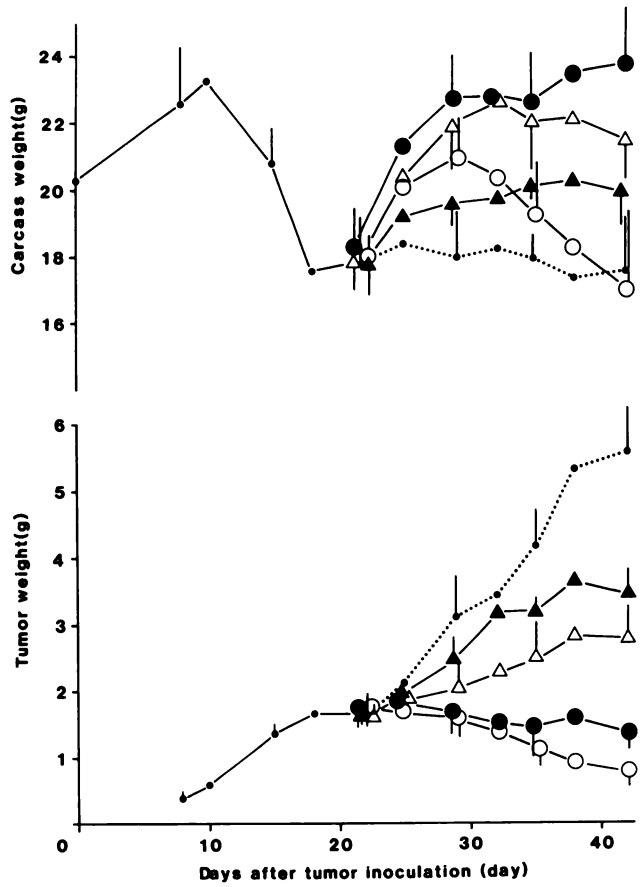

Fig. 1. Effect of 5'-dFUrd on the changes in carcass weight and tumor weight in mice bearing colon 26. Mice were implanted with colon 26 on day 0. Carcass and tumor weights were monitored every 3 or 4 days. From day 22, the tumor-bearing mice were given vehicle (·····), 5'-dFUrd 0.125 (▲), 0.25 (△), 0.5 (●), or 1.0 mmol/kg/day (○) every day for 3 weeks. *Bars*, SD.

proteins, fibrinogen, sialic acid contents in plasma glycoproteins, and IAP, was greatly increased in the cachectic mice with colon 26. 5'-dFUrd substantially reversed the increased production of these acute phase proteins at a dose (0.5 mmol/kg) showing no change of the tumor size. Table 4 indicates the activities of catalase, which is a typical indication of cachexia (20), and P-450 drug-metabolizing enzyme in the liver. The activities of these enzymes were less in the cachectic mice, but the activities began to increase again after 5'-dFUrd (0.5 mmol/kg) administration.

DISCUSSION

We have shown that murine colon 26 adenocarcinoma causes cachexia accompanying a substantial weight loss and physiological changes associated with cachexia (4). The present study showed that 5'-dFUrd, a prodrug of 5-FUra (22, 23), demonstrated the most potent anticachectic activity in terms of the recovery of the body wasting. Definitely, a progressive weight loss in mice bearing colon 26 carcinoma occurred only when the tumor reached an appreciable size, and excision of the tumor resulted in a cessation of weight loss with subsequent rapid weight gain (4). Regression of the tumor by cytostatics must result in recovery from cachexia. However, 5'-dFUrd significantly and immediately reversed cachexia even at doses allowing for the tumor growth in the present study (Tables 1 and 2, Fig. 1). Therefore, it is unlikely that the causal factors of the anticachectic activity of 5'-dFUrd are the same as those of its antiproliferative activity. 5'-dFUrd and other anticachectic cytostatics may inhibit the production of substance(s) triggering cachexia or the process by which the tumor induces cachexia in the host. By using sublines of colon 26 carcinoma resistant to 5'-dFUrd, we demonstrated that the anticachectic activity of 5'-dFUrd is clearly independent of its antitumor activity (24). 5'-dFUrd inhibited the *in vivo* growth of the sublines as well as that of the original colon 26 tumor, although

Table 3 *Concentration of acute phase proteins in the plasma of mice bearing colon 26 compared with the control*

Groups of 6 mice were inoculated s.c. with colon 26 on day 0. The mice were then given 5'-dFUrd p.o. (0.5 mmol/kg/day) every day from day 22 for 7 days. Plasma samples were collected on days 21 or 29 and concentration of acute phase reactants was determined.

Parameters	Mice	Concentrations in plasma (mean ± SD)		
		Before treatment (day 21)	After treatment with (day 29)	
			Vehicle	5'-dFUrd
Fibrinogen (mg/dl)	Non-tumor-bearing	170.1 ± 13.0	157.6 ± 11.6	ND
	Tumor-bearing	1181.4 ± 74.7[a]	844.3 ± 160.9[b]	429.5 ± 108.8[c,d]
Sialic acid (mg/dl)	Non-tumor-bearing	104.0 ± 5.4	94.6 ± 2.8	ND
	Tumor-bearing	195.7 ± 8.8[a]	170.2 ± 16.5[b]	128.6 ± 17.1[c,d]
IAP (μg/ml)	Non-tumor-bearing	89.2 ± 15.7	70.9 ± 35.7	ND
	Tumor-bearing	1012.7 ± 97.9[a]	1132.6 ± 91.1[b]	463.5 ± 117.8[c,d]

[a] $P < 0.01$, compared with non-tumor-bearing mice (day 21).
[b] $P < 0.01$, compared with non-tumor-bearing mice (day 29).
[c] $P < 0.01$, compared with vehicle-treated tumor-bearing mice (day 29).
[d] $P < 0.01$, compared with tumor-bearing mice of day 21.

Table 4 *Changes in activities of hepatic catalase and P-450 in mice bearing colon 26*

Groups of 6 mice were inoculated s.c. with colon 26 on day 0. The mice were then given 5'-dFUrd p.o. (0.5 mmol/kg/day) every day from day 22 for 7 days. The liver was resected out on day 21 or on day 29 and the activity of catalase and the amount on P-450 were determined.

Parameters	Mice	Concentrations in liver (mean ± SD)		
		Before treatment (day 21)	After treatment with (day 29)	
			Vehicle	5'-dFUrd
Catalase (K/g liver)	Non-tumor-bearing	17.4 ± 2.2	13.7 ± 2.3	ND
	Tumor-bearing	8.0 ± 2.2[a]	8.0 ± 2.1[b]	17.0 ± 4.0[c,d]
P-450 (nmol/mg protein in microsome)	Non-tumor-bearing	0.50 ± 0.04	0.54 ± 0.07	ND
	Tumor-bearing	0.11 ± 0.01	0.17 ± 0.06	0.37 ± 0.01

[a] $P < 0.01$, compared with non-tumor-bearing mice (day 21).
[b] $P < 0.01$, compared with non-tumor-bearing mice (day 29).
[c] $P < 0.01$, compared with vehicle-treated tumor-bearing mice (day 29).
[d] $P < 0.01$, compared with tumor-bearing mice of day 21.

it could not reverse cachexia caused by the sublines.

In the previous study, we have examined various murine transplantable tumor lines, widely utilized for the assessment of cytostatics, for their ability to cause cachexia while tumor burdens are still relatively small. This study identified colon 26 carcinoma as capable of causing cachexia (4). Colon 26 is an extraordinary line not only in causing cachexia but also in responding well to 5'-dFUrd. It was the most susceptible tumor line to the *in vivo* but not the *in vitro* antiproliferative action of 5'-dFUrd as compared to other tumor models, where the antitumor efficacy is not as remarkable as that observed in the present study with colon 26 carcinoma (22, 23). Cachexia is known to be clinically associated with the decrease in the clinical responses to chemotherapies. DeWys *et al.* reported that cancer patients who had experienced prechemotherapy weight loss showed lower responses to chemotherapy (3) and survived for shorter periods irrespective of types of subsequent therapies (2). Therefore it is likely that the improving cachexia of mice bearing colon 26 by 5'-dFUrd made the mice more susceptible to the antiproliferative activity of this drug.

In the present "colon 26 carcinoma-cachexia" model, 5'-dFUrd was far more effective in reversing weight loss and other changes associated with cachexia than 5-FUra and its derivatives 2'-dFUrd and tegafur (Table 2). It is likely that 5'-dFUrd exerts its anticachectic activity by a mechanism different from that of the antitumor mechanism reported for 5-FUra. 2'-dFUrd showed only marginal activity, while 5-FUra and tegafur slightly reversed changes in only the particular parameters hypoglycemia and hypercorticism. Only the reversal of these abnormalities may not contribute to that of the body wasting. Otherwise 5-FUra and tegafur may alleviate the abnormalities by a mechanism different from that of 5'-dFUrd.

5'-dFUrd reversed progressive wasting of the carcass, within 3 days after its administration to the cachectic mice (Fig. 1), indicating that 5'-dFUrd directly inhibits the tumor cells production of triggering factors for weight loss and other changes associated with cachexia or disturbs host-mediated process(es) for cachexia such as through inhibition of the production of possible cachexia mediators, tumor necrosis factor and interleukin 1. The acute phase reactions and the depression of hepatic drug metabolism observed in this study might be triggered by the production of tumor necrosis factor, interleukin 1, or interleukin 6 (25–30). Since we have identified colon 26 sublines resistant to the anticachectic action of 5'-dFUrd (24), the direct action to the tumor cells is likely. 5'-dFUrd is less toxic, particularly much less immunosuppressive, than other 5-FUra derivatives (31–33). Furthermore, 5'-dFUrd given daily at 0.5 mmol/kg did not cause any changes to normal mice in the parameters tested in the present study (data are not shown). 5'-dFUrd may suppress the production of particular molecules of either colon 26 or host cells, which trigger or mediate the cachexia process.

Nutritional therapy is presently being given to cancer patients in an attempt to improve cachexia and to alleviate some side effects of cancer therapy. Even though its efficacy is still controversial, nutritional therapy has benefited particular groups of patients (1). Therefore, new treatment modalities for cachexia, which could complement nutritional therapy, are required. The information obtained in the present study that 5'-dFUrd and some other cytostatics are capable of improving cachexia will give some insight into new treatment modalities.

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