Antitumor activity of the novel derivatives of fluoropyrimidine with organosilicone compounds

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The new fluoropyrimidine-derivatives with organosilicone compounds were synthesized and their antitumor effects were evaluated against mouse leukemia. N-{2-[[2-(n-Butyldimethylsilyl)ethyl] thio] ethyl}-5-fluoro-3,4-dihydro-2,4-dioxo-1(2 H)-pyrimidinecarboxyamide (SDK-12 B-8), N-{3-[[2-(Isopropyl-dimethylsilyl)ethyl]thio]propyl}-5-fluoro-3,4-dihydro-2,4-dioxo-1(2 H)-pyrimidinecarboxamide (SDK-12 B-13) and N-{3-[[3-(Trimethylsilyl)-propyl]thio]propyl}-5-fluoro-3,4-dihydro-2,4-dioxo-1(2 H)-pyrimidine-carboxamide (SDK-12 B-14) showed potent antitumor effect by oral administration against L-1210 and P-388 leukemia. The therapeutic ratios were 7.7, 5.1 and 2.7 for SDK-12 B-8, SDK-12 B-13 and SDK-12 B-14 against L-1210, on the other hand, their ratios against P-388 were 5.5, 4.0 and 4.7 for SDK-12 B-8, SDK-12 B-13 and SDK-12 B-14, respectively.

Key words: organosilicone compounds, 5-FU derivatives, antitumor activity

INTRODUCTION

5-Fluorouracil (5-FU) is an anticancer agent synthesized in 1957[1)] 5-FU is widely used for many types of cancer because it shows suppressive effect on a wide spectrum of cancer, and is even effective against carcinomas of the stomach, intestine, breast and other adenocarcinoma which are relatively resistant to chemotherapy. It is well known that a metabolite of 5-FU, 5-fluorodeoxyuridine monophosphate (5-FdUMP), acts its antitumor activity mainly throgh competitive antimetabolic action against thymidylate synthetase in DNA synthesis[2)], although another metabolite, 5-fluorouridine triphosphate (5-FUTP) shows the cytotoxic action by being incorporated into RNA or inhibiting the synthesis of uridine[3~5)]. It is also known that tumor cells are most sensitive to 5-FU from G 1 phase to the S phase of the cell proliferation cycle. Thus, a better therapeutic effect would be expected by maintaining its effective plasma concentration as long as possible. From this point of view, 1-(2-tetrahydrofuryl-5-fluorouracil (FT-207, ftoraful) was developed as a drug which could be metabolized into 5-FU gradually in liver to give long lasting effective plasma concentration level of 5-FU[6)]. After that, numerous attempts were made to develop new derivatives of 5-FU. At present, 1-hexylcarbamoyl-5-fluorouracil (HCFU, carmoful) has been clinically used[7,8)], and 5'-deoxy-5-fluorouridine (5'-DFUR, doxifluridine) was recently developed that is metabolized into 5-FU by pyrimidine nucleoside phosphorylase which shows higher activity in tumors than in normal tissues, resulting in a high concentration of 5-FU in tumor cells and less side-effects[9,10)] Although development of the derivatives of 5-FU has been forcused on enhancing the anticancer potency and lessening the side-effects, the recent trials have been investigated on drugs with actions of immune response and others[11,12)]

Previously we reported antitumor activity of newly synthesized organosilicone compounds, among them, 2-(2-trimethylsilylethyl)-thioethylamine (SDK-12 A) was most effective against several mouse transplantable tumors and less side-effects[13~16]. In this series of our studies on development of new antitumor drugs, new derivatives of 5-FU with organosilicones were synthesized by our group and their antitumor effects were screened. In this report, the antitumor activity of them against murine leukemia was evaluated.

MATERIALS AND METHODS

1. Compounds

The new derivatives of 5-FU complex with organosilicone compounds examined in this study were named SDK-12 B compounds and were listed in Fig. 1. These compounds were synthesized in the Institute of Sanwa Kagaku Kenkyusyo, Ltd. (Mie, Japan). The tested compounds were suspended in 0.1% carboxymethylcellulose sodium salt (CMC) before oral administration.

2. Animals

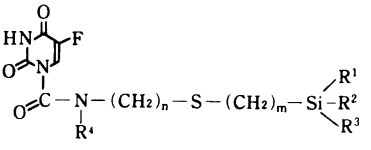
Female BDF 1 mice (Charlus River Atugi, Japan) were used at 6 weeks old. They were maintained with free access to pellet food and water in filtered laminar air flow isolation cages at 21± 1 °C temperature and 60% humidity.

3. Antitumor effect *in vivo*

L-1210 and P-388 leukemia were maintained by weekly passage of the cavity of the C 57 BL/6 mice as an ascites form. To examine the antitumor effect of the tested compounds, 1×10^5 cells of leukemia were inoculated intraperitonealy in BDF 1 mice and the compounds were given orally at day 1, day 5 and day 9 after tumor inoculation. The anti-tumor evaluation was determined survival time of mice and expressed as therapeutic index (ILS_{max}/ILS_{30})

4. Statistics

Student's t-test was employed to analyze the statistical difference between treated and non treated group. The value of $p<0.05$ was considered to be significantly difference.



Compound	m	n	R 1	R 2	R 3	R 4
SDK-12B	2	2	Me	Me	Me	H
SDK-12B-2	2	2	Me	Me	Me	Me
SDK-12B-4	2	2	Et	Et	Et	H
SDK-12B-5	2	2	Me	Me	Ph	H
SDK-12B-6	2	2	Me	Me	n-Oct	H
SDK-12B-7	2	2	Me	Me	i-Pro	H
SDK-12B-8	2	2	Me	Me	n-Bu	H
SDK-12B-9	2	2	Me	Ph	Ph	H
SDK-12B-11	2	2	Me	Me	Me	Et
SDK-12B-10	2	3	Me	Me	Me	H
SDK-12B-13	2	3	Me	Me	i-Pro	H
SDK-12B-1	3	2	Me	Me	Me	H
SDK-12B-16	3	2	Me	Me	i-Pro	H
SDK-12B-17	3	2	Me	Me	t-Bu	H
SDK-12B-14	3	3	Me	Me	Me	H
SDK-12B-15	3	3	Me	Me	i-Pro	H
SDK-12B-18	3	3	Me	Me	t-Bu	H
SDK-12B-19	3	3	Me	Me	n-Bu	H
SDK-12B-20	3	3	Me	Me	Ph	H
HCFU						

Fig. 1. Chemical structure of SDK-12 B compounds.

Table 1. Antitumor activity of SDK-12B compounds against L-1210 leukemia by oral administration

Compound	LD_{50}[a] (mg/kg)	Dose[b] (mg/kg/day)	ILS over controls (%)	Antitumor activity[c]	Compound	LD_{50} (mg/kg)	Dose (mg/kg/day)	ILS over controls (%)	Antitumor activity
SDK-12B	1,096	1,000	70	++	SDK-12B-13	2,250	1,000	101	+++
		750	85	+++			750	87	+++
		500	105	+++			500	80	+++
		250	44	+			250	45	+
SDK-12B-4	1,545	1,000	77	+++	SDK-12B-1	1,575	1,000	70	++
		750	91	+++			750	85	+++
		500	95	+++			500	105	+++
		250	19	±			250	44	+
SDK-12B-5	1,500	1,000	84	+++	SDK-12B-16	1,820	500	69	++
		750	101	+++			250	24	±
		500	82	+++	SDK-12B-17	1,380	500	66	++
		250	35	+			250	−3	−
SDK-12B-6	1,580	1,000	95	+++	SDK-12B-14	2,170	750	68	++
		750	76	+++			500	111	+++
		500	38	+			250	49	+
SDK-12B-7	1,380	1,000	71	++	SDK-12B-15	2,120	750	61	++
		750	105	+++			500	81	+++
		500	107	+++			250	18	±
		250	51	++	SDK-12B-18	1,800	750	76	+++
SDK-12B-8	1,385	1,000	105	+++			500	68	++
		750	89	+++			250	19	±
		500	73	++	SDK-12B-19	1,850	750	44	+
		250	35	+			500	62	++
SDK-12B-9	1,450	750	15	±			250	26	+
		500	11	±	SDK-12B-20	1,800	750	8	toxic
SDK-12B-11	1,380	750	10	±			500	86	+++
		500	10	±			250	23	±
SDK-12B-10	1,355	1,000	63	++	HCFU	1,845	750	81	+++
		750	89	+++			500	84	+++
		500	70	++			250	20	±

[a]By single oral administration. [b]Treatment schedule; day 1, day 5 and day 9 by oral administration.

[c]Antitumor activity was graded as − 0−9, ± 10−24, + 25−50, ++ 51−74 and +++ 75 or more of ILS %.

RESULTS

1. Antitumor effect of SDK-12 B compounds against L-1210 leukemia

SDK-12 B compounds tested in this study were synthesized approximately 20 and their chemical structures were shown in Fig. 1.

The toxicities of them when given once oral administration were determined before beginning of the therapeutical examination and their results were shown in Table 1. Their lethal doses (LD_{50}) showed widely range from 1,096 mg/kg for SDK-12 B to 2,250 mg/kg for SDK-12 B-13.

The antitumor activity was examined by oral administration at doses of 1,000 mg/kg, 750 mg/kg, 500 mg/kg and 250 mg/kg of each compound, beginning on day 1, day 5 and day 9 after tumor inoculation. As seen in Table 1, almost all of the tested compounds showed antitumor effect against L-1210 leukemia. Of them, the most effective antileukemic activity was obtained when given 500 mg/kg/day of SDK-12 B-14.

2. Antitumor effect of SDK-12 B compounds against P-388 leukemia

In the therapeutical examination in L-1210 system, each therapeutical ratio of SDK-12 B compounds was calculated. The high ratios over 2.5 were obtained in the compounds as follows; 7.7, 5.1, 3.5, 2.9, 2.7 and 2.6 for SDK-12 B-8, SDK-12 B-13, SDK-12 B-5, SDK-12 B-10, SDK-12 B and SDK-12 B-14, and SDK-12 B-7, respectively. That of the reference drug, HCFU was 1.8. The antitumor effect against P-388 leukemia were examined even in these seven compounds expressing the high therapeutical reatio against L-1210. The results were shown in Table 2.

The maximum % of ILS was obtained in SDK-12 B-14 when given 800 mg/kg/day and its therapeutical ratio was 4.7, The other therapeutical ratios were as follows; 5.5, 4.0, 2.7 and 2.2 for SDK-12 B-8, SDK-12 B-13, SDK-12 B-7 and SDK-12 B-5, respectively. That of the reference drug was 2.1.

DISCUSSION

The derivatives of 5-fluorinated complex with organosilicone compounds were synthesized to develop new antitumor agents showing different action mechanism from that of available antitumor drugs and less side-effects. The organosilicone compounds exhibited antitumor effects against some strains of murine transplantable tumor except leukemia and less toxicities[13~16]. Though the 50% lethal doses of SDK-12 B compounds by once oral administration distributed widely range, they expressed generally less toxicities. Their toxicities might depend on the number of alkyl moiety in the chemical structure of SDK-12 B compounds such as m=n=2>m=n=3. Among them, SDK-12 B-5, SDK-12 B-8, SDK-12 B-13 and SDK-12 B-14 were

Table 2. Therapeutic evaluation in mouse leukemia systems by oral administration

Compound	L-1210 leukemia				P-388 leukemia			
	ILS_{30} (mg/kg/day)	ILS_{max} (mg/kg/day)	Maximum ILS (%)	Therapeutic ratio[a)]	ILS_{30} (mg/kg/day)	ILS_{max} (mg/kg/day)	Maximum ILS (%)	Therapeutic ratio
SDK-12B	188	500	105	2.7	ND[b)]			
SDK-12B-5	214	750	101	3.5	275	600	78	2.2
SDK-12B-7	192	500	107	2.6	220	600	83	2.7
SDK-12B-8	130	1,000	105	7.7	110	600	71	5.5
SDK-12B-10	260	750	89	2.9	ND			
SDK-12B-13	195	1,000	101	5.1	200	800	61	4.0
SDK-12B-14	184	500	111	2.7	170	800	89	4.7
HCFU	272	500	84	1.8	190	400	57	2.1

[a)] Therapeutic ratio = ILS_{max}/ILS_{30}

[b)] ND = not determined

selected and the doses of 1/10 and 1/5 of LD_{50} of them were administered orally once a day for five consecutive days in mice and then side-effects such as cytopenia, body weight-loss, histopathological abnormality were examined for three weeks after administration. The side-effects such as cytopenia and body weight loss appeared for 9 days duration beginning from day 5 to day 14 in mice treated with 5-FU and HCFU at dose of $1/5 \times LD_{50}$, however, adverse effects were not observed in mice treated with SDK-12 B compounds. In the 1st screening test of SDK-12 B compounds, the antitumor effects by several treatment schedules against L-1210 were investigated. The experimental results were obtained as day 1, 5, 9>day 1, 3, 6, 9>day 1, 5> day 1—5>day 1—9 (unpublished observation). As seen in the results, the antitumor activity of SDK-12 B compounds was more potent by interval-treatment than by consecutive-treatment. In this study, organosilicone moiety did not show any antitumor effect against leukemia[13], however, SDK-12 B compounds expressed potent antileukemic effects. Among them, SDK-12 B-8, SDK-12 B-13 and SDK -12 B-14 showed extremely high therapeutic ratios. They are thought to be a masked form of 5-FU and may release 5-FU after oral administration. Their pharmacokinetics including cellular incorporation, mode of cytocidal action, oral adsorption and metabolism may depend on their antitumor activity and less side-effects. Further studies to explain the detailes of the antitumor effects against other solid tumors and the pharmacokinetics of SDK-12 B compounds are in progress.

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有機シリコーン化合物をもった新しいフッ化ピリミジン誘導体の抗腫瘍効果

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有機シリコーン化合物をもった新しいフッ化ピリミヂン誘導体が合成されマウス白血病に対する抗腫瘍効果が検討された。N-{2-[[2-(n-Buthyldimethylsilyl]ethyl]thio]ethyl}-5-fluoro-3,4-dihydro-2,4-dioxo-1(2 H)-pyrimidinecarboxyamide (SDK-12 B-8), N-{3-[[2-(Isopropyl-dimethylsilyl)-ethyl]thio]propyl}-5-fluoro-3,4-dihydro-2,4-dioxo-1(2 H)-pyrimidinecarboxyamide (SDK-12 B-13), および N-{3-[[3-(Trimethylsilyl)-propyl]-thio]propyl}-5-fluoro-3,4-dihydro-2,4-dioxo-1(2 H)-pyrimidine-carboxyamide (SDK-12 B-14) が L-1210 および P-388 白血病に対して経口投与で強い抗腫瘍効果を示した。L 1210 白血病に対する化学療法係数は SDK-12 B-8 が 7.7, SDK-12 B-13 が 5.1, および SDK-12 B-14 が 2.7 であった。一方, P-388 に対しては SDK-12 B-8 が 5.5, SDK-12 B-13 が 4.0, SDK-12 B-14 が 4.7 のそれぞれ化学療法係数を示した。

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