Eur J Cancer Clin Oncol, Vol. 19, No. 3, pp. 411–418, 1983.
Printed in Great Britain.

0277-5379/83/030411-08$03.00/0

Antitumor Activity in Mice of 4′-Deoxydoxorubicin in Comparison with Doxorubicin

ANNA MARIA CASAZZA,*† GIUSEPPINA SAVI,‡ GRAZIELLA PRATESI‡ and AURELIO DI MARCO*

Experimental Oncology, Centro Ricerche Farmitalia Carlo Erba, 20014 Nerviano, Milano and ‡Division of Experimental Oncology B, Istituto Nazionale per lo Studio e la Cura dei Tumori, 20133 Milano, Italy

Abstract—*4′-Deoxydoxorubicin has been compared with doxorubicin as regards potency, antitumor activity and toxicity in tumored and non-tumored mice treated i.v. according to different schedules. 4′-Deoxydoxorubicin was 1.5–3 times more toxic and more potent than doxorubicin. At equitoxic doses, 4′-deoxydoxorubicin was: as active as doxorubicin against Gross leukemia, mammary carcinoma and MS-2 sarcoma; slightly less active than doxorubicin against B16 melanoma; more active than doxorubicin against colon 38 adenocarcinoma. The best schedule of administration of 4′-deoxydoxorubicin in mice was the weekly treatment. The strong effectiveness against colon 38 adenocarcinoma makes 4′-deoxydoxorubicin a particularly interesting new anthracycline derivative that deserves clinical trials.*

INTRODUCTION

4′-DEOXYDOXORUBICIN (4′-deoxyDX, NSC-267469) is a new analog of doxorubicin (DX, NSC-123127) characterized by the lack of the hydroxyl group in position 4′ of the aminosugar [1]. Previous studies have shown that 4′-deoxyDX was definitely less cardiotoxic than DX in rabbits and mice treated chronically i.v. [2], but showed an antitumor activity equal to that of DX when given i.p. to mice bearing the P388 and L1210 ascitic leukemias and when given i.v. to mice bearing the i.v.-inoculated Gross leukemia [1, 2]. Furthermore, 4′-deoxyDX was significantly active against doxorubicin-resistant human colon tumors [3] and as active as DX against human breast and prostate tumors xenografted into nude mice [4]. These results prompted us to investigate more deeply the spectrum of antitumor activity of 4′-deoxyDX in comparison with DX in mice. We have compared the antitumor activity of 4′-deoxyDX with that of DX against Gross leukemia, mammary carcinoma of C3H mice, melanoma B16, MS-2 sarcoma, and colon 38 and 26 adenocarcinomas transplanted in mice. In all the experiments the drugs were injected i.v. As the antitumor activity of anthracyclines depends on the schedule of treatment [5], several schedules were tested. In one experiment against colon 26 adenocarcinoma, the activity of 4′-deoxyDX was compared to that of 5-fluorouracil (5-FU) because of the importance of the latter in the treatment of patients affected by colon cancer [6, 7]. The results show that 4′-deoxyDX has an antitumor activity different from that of DX, being slightly less active than DX against B16 melanoma and more active than DX against a colon tumor, in agreement with the findings obtained on human tumors in nude mice.

Accepted 14 September 1982.

†To whom requests for reprints should be addressed at: Centro Ricerche Farmitalia Carlo Erba, Via Giovanni XXIII, 20014 Nerviano, Milano, Italia.

MATERIALS AND METHODS

Drugs

DX hydrochloride and 4′-deoxyDX hydrochloride were supplied by Ricerca Chimica, Farmitalia Carlo Erba Research Laboratories, Milano, Italy. 5-Fluorouracil (5-FU) (Roche) was employed as a distilled water solution.

Animals

Inbred C3H/He, C57BL/6 and BALB/c mice, and hybrid (C57BL/6 × DBA/2) F_1 (BDF_1) mice of both sexes were supplied by the Charles River Breeding Laboratories, Calco, Italy. The animals were 2–3 months old, weighed 20–30 g and were maintained under standard laboratory conditions.

Tumors

Gross leukemia [8] was transplanted by i.v. inoculation of 2.0×10^6 cells of an homogenate of

spleen and lymph nodes of leukemic mice in C3H/He mice. In the case of mammary carcinoma, 20×10^6 cells from a tumor at the third generation were inoculated into the mammary tissue as previously described [9] in C3H/He female mice. The B16 melanoma was maintained by s.c. transplants in C57BL/6 mice, according to Geran *et al.* [10]. Colon 38 and 26 were serially transplanted in C57BL/6 mice and in BALB/c mice respectively by injecting s.c. fragments of tumor tissue of about 20 mg weight, as indicated by Corbett *et al.* [11]. The MS-2 sarcoma was transplanted i.m. into the right hind leg of BALB/c mice (10^5 cells/mouse). Each experimental group consisted of at least eight animals.

Drug administration

DX and 4'-deoxyDX were dissolved in distilled water and administered i.v. in a volume of 10 ml/kg of body weight. 5-FU was administered i.p. The treatment schedules are reported in the Results section.

Evaluation of antitumor activity and toxicity

In Gross leukemia the comparison of drug effectiveness was based on the increase in median survival time (MST) over the dose range used compared to untreated controls.

In solid tumors the tumor growth was assessed at least twice a week by caliper measurement. Tumor weight was calculated according to the following formula: $w = d^2 \times D/2$, where d is the minimum and D is the maximum diameter [10]. In experiments carried out for evaluation of effect against early tumors, the antitumor activity was established by the tumor weight inhibition on different days after tumor implantation. In the experiments on advanced solid tumors, the tumor weight of single mice was calculated by caliper measurement at the beginning of treatment and twice a week during and after treatment. Tumor growth was estimated for individual mice from the ratio between tumor weight at the indicated days and tumor weight at beginning of treatment. The data reported represent the average of individual tumor growths.

Toxicity was established on the basis of the macroscopic necropsy finding evaluation in all dead mice. The main findings were spleen and liver size reduction and hemorrhagic degeneration on the intestinal mucosa, as previously observed after daunorubicin or doxorubicin treatment [9]. In several experiments normal mice were treated in parallel at the same dose and schedule as the tumor-bearing mice for evaluation of toxicity: mice were observed for at least 3 months after beginning of treatment.

RESULTS

4'-DeoxyDX was given to C3H mice bearing Gross leukemia according to various schedules. The results are reported in Table 1. 4'-DeoxyDX was about twice as potent as DX, and at the optimal dose it was as active as DX. In fact, in mice that received one single treatment on day 1, the optimal non-toxic dose was 16.9 mg/kg for DX (T/C, % = 233) and 8.6 mg/kg for 4'-deoxyDX (T/C, % = 233); in mice treated for three consecutive days the optimal dose was 4.5 mg/kg for DX (T/C, % = 200 and 175) and 2.25 mg/kg for 4'-deoxyDX (T/C, % = 233 and 216); in mice treated once a week for three weeks the optimal dose was 7.7 mg/kg for DX (T/C, % = 200) and 3.3–3.8 mg/kg for 4'-deoxyDX (T/C, % = 308 and 230). For both drugs, the best results were obtained when treatment was performed once a week for 3 weeks.

This schedule was used for the treatment of mice bearing s.c. B16 melanoma and of normal BDF1 mice treated in parallel. The results of the two experiments are reported in Table 2. 4'-DeoxyDX was about 1.5 times more toxic than DX. At the tolerated doses of 6.6 and 10 mg/kg, DX produced a tumor weight inhibition ranging from 70 to 95%; 4'-deoxyDX at the tolerated doses of 4.4 and 6.6 mg/kg produced a tumor weight inhibition ranging from 52 to 79%, therefore showing a somewhat lower effect than DX in this experimental system. However, the optimal increase of life span given by the two compounds was of the same order of magnitude (T/C, % = 197 for DX and 182 for 4'-deoxyDX).

Several experiments were carried out against the mammary carcinoma in C3H female mice, treated according to different schedules. In one experiment, treatment started on day 1 after the tumor inoculation, and was carried out every other day for two cycles of five treatments each. The results, reported in Table 3, show that 4'-deoxyDX administered according to this schedule was less active than DX. Table 4 shows the data obtained in two experiments carried out on C3H mice bearing mammary carcinoma; treatment started when the tumor was palpable and was performed on a weekly schedule for four or five weeks. The results of the second experiment are also shown in Fig. 1. At the MxTD (maximal tolerated dose) of 7.5 mg/kg/day, DX caused a marked regression of tumor volume. Similar regression was observed after treatment with 4'-deoxyDX at the doses of 5 and 6.25 mg/kg/day, but in mice treated with the new derivative, tumor growth resumed earlier than in mice treated with DX (Fig. 1).

As a model of experimental sarcoma, we have investigated the MS-2 sarcoma transplanted i.m.

*Table 1. Activity of 4'-deoxyDX and DX against Gross leukemia. Dependence on the schedule of treatment**

Days of treatment†	Compound	Dose (mg/kg/day)	T/C‡ (%)	Toxic deaths§ Tumored mice	Toxic deaths§ Normal mice
1	DX	7.7	150	0/10	n.d.‖
		10.0	200, 175	0/18	n.d.
		12.0	200	0/8	n.d.
		13.0	200	0/10	n.d.
		14.4	225	0/8	n.d.
		16.9	233	0/10	n.d.
		22.0	266	1/10	n.d.
	4'-DeoxyDX	4.2	141	0/8	n.d.
		5.0	158, 200	0/18	n.d.
		6.0	175	0/8	n.d.
		6.6	216	0/10	n.d.
		7.2	191	0/8	n.d.
		8.6	233	0/10	n.d.
		11.3	241	2/10	n.d.
		14.6	125	8/10	n.d.
1, 2, 3	DX	3.5	183, 166	0/20	n.d.
		4.5	200, 175	0/20	n.d.
		5.4	216, 200	2/20	n.d.
		6.5	216	0/10	n.d.
	4'-DeoxyDX	1.75	200, 200	0/20	n.d.
		2.25	233, 216	1/20	n.d.
		2.75	233, 233	2/20	n.d.
		3.25	233	2/20	n.d.
1, 8, 15	DX	6.6	150	0/8	0/8
		7.7	200¶	0/10	n.d.
		10.0	316, 540¶	2/18	7/8
		13.0	390¶	6/10	n.d.
		15.0	350	7/8	8/8
	4'-DeoxyDX	3.3	308	0/8	1/8
		3.8	230	1/10	n.d.
		5.0	408, 320¶	0/18	2/8
		6.5	300°¶	5/10	n.d.
		7.5	391	3/8	2/8

*C3H/He mice received 2×10^6 leukemic cells i.v. on day 0; data of 6 experiments.
†Drugs were administered i.v. on the indicated days after tumor transplantation.
‡(Median survival time of treated mice/median survival time of controls) × 100. Median survival time of controls was 6 days, except when otherwise stated.
§No. of toxic deaths/No. of mice treated.
‖Not done.
¶In this experiment median survival time of control mice was 5 days.

in BALB/c mice. Mice were treated every 3 days 5 times, starting on day 1 after tumor inoculum. Groups of non-tumored mice were treated in parallel as a control of toxicity. The results reported in Table 5 show that 4'-deoxyDX, administered according to this schedule, was about 1.7 times as toxic as DX. In fact, the MxTD was 4.6 mg/kg for DX and 2.6 mg/kg for 4'-deoxyDX. The inhibition of tumor growth observed in three experiments is reported in Table 5; Fig. 2 shows the growth curve of the tumor of controls and treated mice of one representative experiment. It can be observed that at the MxTD the two compounds equally inhibited tumor growth; DX was more active than 4'-deoxyDX in one experiment in increasing the mice survival time.

In view of the previously reported [3] high effectiveness of 4'-deoxyDX against human colon carcinoma xenografts, a considerable number of experiments were carried out on two transplanted colon adenocarcinomas of mouse: colon 38 and colon 26. The results obtained on the colon 38 tumor are shown in Table 6 and in Fig. 3, in

*Table 2. Activity of 4'-deoxyDX and DX against B16 melanoma**

Compound	Dose† (mg/kg/day)	Tumor weight inhibition (%)‡ Exp. 1‖	Exp. 2¶	T/C (%)§ Exp. 1	Exp. 2	Toxic deaths Tumored mice	Normal mice
DX	6.6	88	70	180	96	0/18	n.d.**
	10.0	94	95	197	146	0/18	0/8
	15.0		99		110	6/10	6/8
4'-DeoxyDX	3.3	24		142		1/8	n.d.
	4.4		52		120	0/8	n.d.
	5.0	79		182		0/8	n.d.
	6.6		57		83	0/10	0/8
	10.0		toxic		31	10/10	3/8

*Mice were injected s.c. with B16 melanoma cell suspension. Data of 2 experiments.
†Treatment i.v. on days 1, 8, 15.
‡100 - [(tumor weight of treated mice/tumor weight of control mice) × 100]. Tumor weight was calculated from measurements of tumor diameter with calipers. Data refer to measurements on day 16 after tumor injection.
§See note to Table 1.
‖In Experiment 1, C57/BL female mice were used. Tumor weight in controls was 0.6 g on day 12 and 1.18 g on day 16.
¶In Experiment 2, BDF1 female mice were used. Tumor weight in controls was 1.4 g on day 12 and 2.9 g on day 16.
**Not done.

Table 3. *Activity of 4'-deoxyDX and DX against early mammary carcinoma of C3H mice**

Compound	Dose† (mg/kg/day)	Tumor weight inhibition (%)‡	LTS§	Toxic deaths‖
DX	2	100	7/10	0/10
	3	100	4/10	6/10
4'-DeoxyDX	1	84	0/10	0/10
	2	100	2/10	5/10

*C3H/He female mice were injected s.c. with 2×10^7 carcinoma cells on day 0.
†Treatment i.v. qd2 from day 1 to 9 and from day 15 to 23 after tumor inoculum.
‡Measured on day 42; see footnote to Table 2.
§No. of long-term survivors/total No. of mice treated.
‖Evaluated in tumored mice on the basis of necropsy findings.

which the growth curves of tumor in control mice and in mice treated once a week for 4 weeks are reported. Administered every 4 days for 3 times, starting on day 1 after tumor implant, 4'-deoxyDX was as active as DX at 1.5-fold lower doses. Administered once a week for 4 weeks, starting on day 1 after tumor implant, 4'-deoxyDX was more active than DX against this experimental tumor. In fact, DX at the tolerated dose of 6 mg/kg per day reduced tumor growth by 74 and 40% in two experiments and increased the life span of the mice only in one of the two experiments. 4'-DeoxyDX at the tolerated dose of 4 mg/kg per day reduced tumor growth by 90% or more, strongly increased the life span of mice in both experiments and caused a high number of long-term survivors (Table 6). The superiority of 4'-deoxyDX over DX in inhibiting colon 38 carcinoma is also evident by the Fig. 3.

The results of two experiments on the colon 26 adenocarcinoma are shown in Table 7. Mice were treated once a week for 4 weeks, starting on day 1 after tumor inoculation. This experimental tumor was less sensitive than the colon 38 tumor to anthracycline treatment. 4'-DeoxyDX was slightly more active than DX and inhibited tumor growth as much as 5-FU.

DISCUSSION

The effects that modifications in position 4' of the amino sugar of anthracyclines bring about in the biologic activity of these compounds have been previously investigated. In particular, 4'-epidoxorubicin was found to be as active as DX, but endowed with lower general and cardiac toxicity [12]. From a biochemical point of view, no substantial differences were observed between DX and 4'-epidoxorubicin [13], but pharmaco-

*Table 4. Activity of 4'-deoxyDX and DX against advanced mammary carcinoma of C3H mice**

Experiment No.	Compound	Dose (mg/kg/day)	Tumor growth (+) or regression (-)† (%)	T/C‡ (%)	Toxic deaths§
1‖	—	—	+ 521		n.d.¶
	DX	7.5	- 37	192	n.d.
		10.0	- 73	179	n.d.
	4'-DeoxyDX	3.5	+ 135	158	n.d.
		5.0	- 67	162	n.d.
2**	—	—	+ 866		0/10
	DX	6.0	- 21	100	0/10
		7.5	- 62	90	0/10
		9.3	- 82	64	11/20
	4'-DeoxyDX	4.0	+ 97	98	0/10
		5.0	- 31	99	0/10
		6.25	- 52	106	1/20
		7.75	n.d.		4/10
		9.3	n.d.		6/10

*See footnote to Table 3.

†(Tumor weight at end of treatment/tumor weight at beginning of treatment) × 100; average of individual variations -100.

‡(Median survival time of treated mice/median survival time of control mice) × 100.

§No. of toxic deaths/No. of mice treated; evaluated in non-tumored mice observed for 4 months.

‖Treatment i.v. once a week for 5 weeks, starting on day 28. Median survival time of control mice was 48.5 days.

¶Not done.

**Treatment i.v. once a week for 4 weeks, starting on day 41. Median survival time of control mice was 206 days.

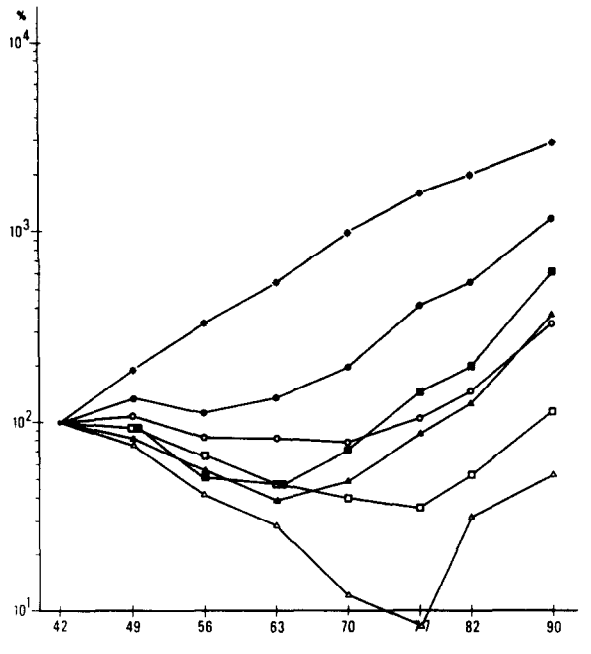

*Fig. 1. Effect of DX and 4'-deoxyDX on mammary carcinoma of C3H mice. *: controls; ○: DX 6 mg/kg; □: DX 7.5 mg/kg; △: DX 9.3 mg/kg; ●: 4'-deoxyDX 4 mg/kg; ■: 4'-deoxyDX 5 mg/kg; ▲: 4'-deoxyDX 7.5 mg/kg. Treatment i.v. q 7 days 4 times, starting on day 41 after tumor inoculum. Abscissa: days after tumor implant; ordinate: ratio between tumor weight at the indicated days/tumor weight at beginning of treatment (average of data of individual mice).*

kinetic properties both *in vivo* and *in vitro* were slightly modified [12, 13]. Similarly, 4'-deoxyDX was as active or slightly more active than DX against DNA and RNA polymerase in cell-free systems [14]. However, 4'-deoxyDX was taken up in much greater amounts than DX by L1210 cells, and was markedly more active than DX against HeLa cells *in vitro* [14]. The higher cytotoxic activity of 4'-deoxyDX in comparison with DX correlates with the higher potency and higher general toxicity observed *in vivo* and described in this paper: in the i.v. treatment schedules, 4'-deoxyDX was about 1.5–2 times more potent than DX.

The data regarding the antitumor activity of 4'-deoxyDX are of great interest. This new DX derivative, compared at the MxTD with the parent drug was: (1) as active as the parent compound against Gross leukemia, MS-2 sarcoma and against mammary carcinoma when administered on a weekly schedule; (2) slightly less active than DX against B16 melanoma; and (3) more active than DX against the transplanted colon 38 at the optimal treatment schedule. These data are in good agreement with the superiority of 4'-deoxyDX vs DX against human colon carcinomas transplanted in nude mice [3] and with

the data of antitumor activity against other human tumors transplanted in nude mice [4], in particular as regards the equal activity of DX and 4'-deoxyDX against mammary carcinomas and the lower activity of 4'-deoxyDX vs DX against melanomas. The good agreement between results obtained in antitumor activity tests carried out in such different systems as conventional mice transplanted with syngeneic tumors and nude mice transplanted with human tumors makes us confident about the reliability of the data and their possible predictivity of antitumor effectiveness in humans.

As regards the schedule of treatment, the most favorable for 4'-deoxyDX seems to be weekly administration, both against solid tumors (colon and mammary carcinomas) and against Gross leukemia. More frequent treatments give increased toxicity or reduction of antitumor activity at the MxTD. It has been shown that 4'-deoxyDX

*Table 5. Effect of 4'-deoxyDX and DX against MS-2 sarcoma**

Compound	Dose† (mg/kg/day)	Tumor growth inhibition‡(%)	T/C§ (%)	Toxic deaths‖
DX	2.0, 2.6	30, 29	103, 107	0/9
	3.0	70	98	n.d.¶
	4.0	93	>257 (6/10)	n.d.
	4.6	71	118	1/9
	5.0	94	231 (4/10)	4/10
4'-DeoxyDX	1.0	62	134	n.d.
	1.7, 2.0	29, 52, 80	103, 115, 170 (1/10)	0/19
	2.5, 2.6	77, 76	118, 165 (3/10)	1/19
	4.0	88	31	0/9

*BALB/c mice were injected i.m. with 10^5 MS-2 sarcoma cells; data of 3 experiments.
†Treatment i.v. every 3 days for 5 times, starting on day 1 after tumor inoculum.
‡100 − [(tumor weight of treated mice/tumor weight of controls) × 100]; data of 3 experiments; measured one week after the last treatment.
§(Median survival time of treated mice/median survival time of control mice) × 100; in parentheses = No. of long-term survivors/No. of mice treated.
‖No. of toxic deaths/No. of mice treated; evaluated in non-tumored mice observed for 108 days.
¶Not done.

*Table 6. Activity of 4'-deoxyDX and DX against colon 38 adenocarcinoma**

Days of treatment†	Compound	Dose (mg/kg/day)	Tumor weight inhibition (%)	T/C (%)	LTS	Toxic deaths‡
1, 5, 9§	DX	3.3	56	113	0/10	n.d.‖
		5.0	91	161	1/10	n.d.
		7.5	99	249	4/10	3/7
	4'-DeoxyDX	1.5	61	142	0/10	n.d.
		2.2	77	115	0/10	n.d.
		3.3	96	162	0/10	0/7
		5.0	98	211	2/10	1/7
1, 8, 15, 22¶	DX	6.0	74, 40	119, 186	3/20	0/6
		9.0	95, 99	175, 186	2/20	13/16
	4'-DeoxyDX	4.0	91, 99	>278, 273	8/20	0/6
		6.0	97, 99	>278, 171	8/20	10/16

*BDF1 mice were injected s.c. with colon 38 carcinoma fragments.
†Drugs were administered i.v. on the indicated days after tumor transplantation.
‡Determined in non-tumored mice treated in parallel, observed for about 4 months after the treatment.
§In this experiment tumor weight inhibition was determined on day 26 after tumor inoculum; tumor weight in controls was 1.3 g.
‖Not done.
¶Tumor weight inhibition was determined on day 31 (tumor weight in controls = 0.45 g), and on day 32 (tumor weight in controls = 1.3 g) in the two experiments.

*Table 7. Activity of 4'-deoxyDX and DX against colon 26 adenocarcinoma**

Compound	Dose† (mg/kg/day)	Tumor weight inhibition‡(%)	T/C (%)	Toxic deaths§
DX‖	6.0	50, 52	123, 164	0/8
	9.0	77, 85	173, 135	15/16
4'-DeoxyDX‖	4.0	59, 67	133, 169	0/8
	6.0	79, 94	175, 185	5/17
5-FU¶	50.0	62	137	n.d.
	75.0	toxic		9/9

*BALB/c mice were injected s.c. with colon 26 tumor fragments; data of 2 experiments.
†Treatment once a week for 4 weeks, starting on day 1 after tumor inoculum.
‡On day 22 after tumor inoculum.
§Determined in non-tumored mice treated in parallel, observed for 3–4 months after the treatment.
‖Administered i.v.
¶Administered i.p.

is eliminated more quickly than DX from mouse tissues, in particular from spleen and heart[15]. It is possible that in mice treated with 4'-deoxyDX the return of normal tissues to normal proliferation values, which follows the initial inhibition, takes place earlier than in mice treated with DX, since this drug remains in the tissues for a shorter time than DX. The treatment every 3–4 days with 4'-deoxyDX can therefore hit cells that are in active proliferation and can be highly sensitive to the toxic action of the drug. This hypothesis is presently under experimental investigation.

The data reported in this paper show that 4'-deoxyDX is slightly (1.5–2 times) more toxic than DX in mice treated according to various schedules. On the contrary, 4'-deoxyDX was

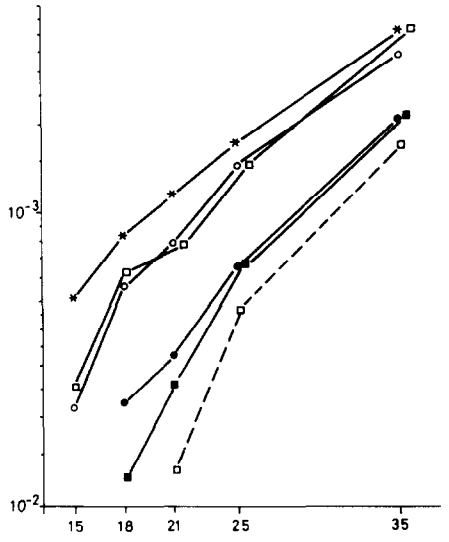

Fig. 2. Effect of DX and 4'-deoxyDX on MS-2 sarcoma in BALB/c mice. *: *controls;* O:*DX* 2.6 mg/kg; •: *DX* 4.6 mg/kg; □: 4'-*deoxyDX* 1.7 mg/kg; ■: 4'-*deoxyDX* 2.6 mg/kg; □ ----□: 4'-*deoxyDX* 4 mg/kg. *Treatment i.v. q* 3 days 5 *times, starting on day* 1 *after tumor inoculum. Abscissa: days after tumor implant; ordinate: tumor weight* (mg).

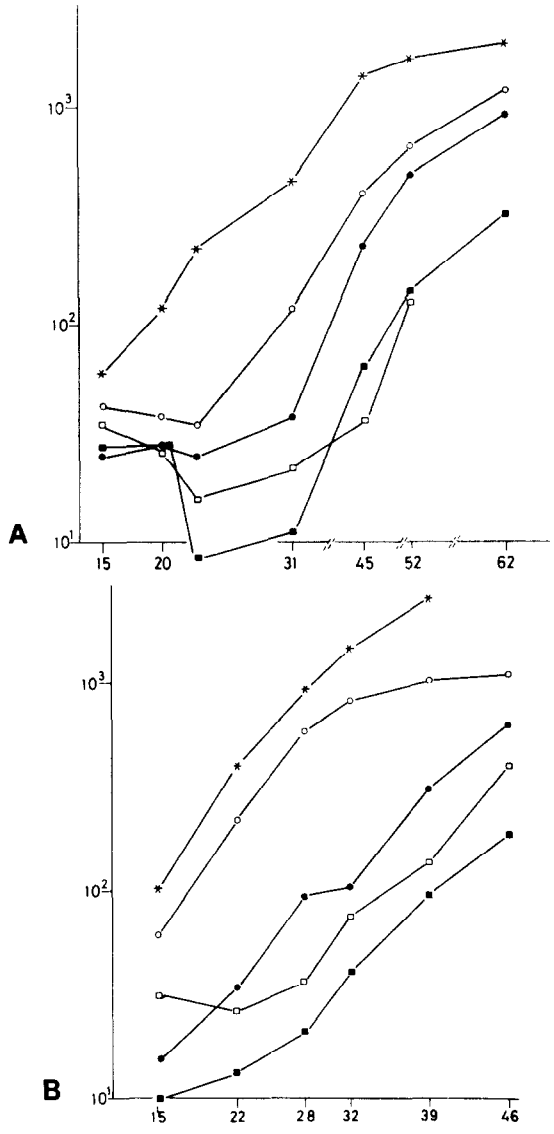

Fig. 3. *Effect of DX and 4'-deoxyDX on colon 38 adenocarcinoma.* * *controls;* O: *DX* 6 mg/kg; □: *DX* 9 mg/kg; •: 4'-*deoxyDX* 4 mg/kg; ■: 4'-*deoxyDX* 6 mg/kg. *A and B: data from two separate experiments. Treatment i.v. q* 7 *days* 4 *times, starting on day* 1 *after tumor inoculum. Abscissa: days after tumor implant; ordinate: tumor weight* (mg).

markedly less cardiotoxic than DX in rabbits treated 3 days/week for 6 weeks and in CD 1 mice treated twice a week for a total of 5 weeks: no heart lesions were seen in rabbits and scattered non-severe lesions were observed in some mice treated with the highest dose [2]. 4'-DeoxyDX therefore appears to be a peculiar new anthracycline derivative, which has in comparison with DX: (1) different antitumor activity against experimental tumors; (2) a higher potency; and (3) a markedly lower cardiac toxicity. The reasons for these selective differences in activity and toxicity are not yet known.

We are presently investigating the possibility that these profound differences may be partly related to the different pharmacokinetic properties of the two drugs [15, 16] and to the different effects on the immune system [17].

REFERENCES

1. ARCAMONE F, PENCO S, REDAELLI S, HANESSIAN S. Synthesis and antitumor activity of 4'-deoxydaunorubicin and 4'-deoxyadriamycin. *J Med Chem* 1976, **19**, 1424–1425.
2. CASAZZA AM, DI MARCO A, BONADONNA G *et al.* Effects of modifications in position 4 of the chromophore or in position 4' of the aminosugar, on the antitumor activity and toxicity of daunorubicin and doxorubicin. In: CROOKE ST, REICH SD, eds. *Anthracyclines: Current Status and New Developments*. New York, Academic Press, 1980, 403–430.
3. GIULIANI FC, KAPLAN NO. New doxorubicin analogs active against doxorubicin-resistant colon tumor xenografts in the nude mouse. *Cancer Res* 1980, **40**, 4682–4687.
4. GIULIANI FC, COIRIN AK, RICE MR, KAPLAN NO. Effect of 4'-doxorubicin analogs on heterotransplantation of human tumors in congenitally athymic mice. *Cancer Treat Rep* 1981, **65**, 1063–1075.
5. CASAZZA AM, PRATESI G, GIULIANI F, DI MARCO A. Antileukemic activity of 4-demethoxydaunorubicin in mice. *Tumori* 1980, **66**, 549–564.
6. CARTER SK, BAKOWSKI MT, HELLMANN K. Colorectal adenocarcinoma. In: CARTER SK, BAKOWSKI MT, eds. *Chemotherapy of Cancer*. New York, John Wiley, 1977, 116–122.
7. SMITH FP, BYRNE PJ, CAMBARERI RC, SCHEIN PS. Gastrointestinal cancer. In: PINEDO HM, ed. *Cancer Chemotherapy*. Amsterdam, Excerpta Medica, 1979, 292–316.
8. DI MARCO A, CASAZZA AM, DASDIA T *et al.* Cytotoxic, antiviral and antitumor activity of some derivatives of daunorubicin (NSC-82151). *Cancer Chemother Rep* 1973, **57**, 269–274.
9. DI MARCO A, LENAZ L, CASAZZA AM, SCARPINATO BM. Activity of adriamycin (NSC-123127) and daunomycin (NSC-82151) against mouse mammary carcinoma. *Cancer Chemother Rep* 1972, **56**, 153–161.
10. GERAN RI, GRENBERG NH, MACDONALD MM, SCHUMACHER AM, ABBOT BJ. Protocols for screening chemical agents and natural products against animal tumors and other biological systems (third edition). *Cancer Chemother Rep* (*Part* 3) 1972, **3**, 1–103.
11. CORBETT TH, GRISWOLD DP, ROBERTS BJ, PECKAM SC, SCHABEL FM. Single agents and combinations of chemotherapeutic agents in mouse colon carcinomas. *Cancer* 1977, **40**, 2660–2680.
12. CASAZZA AM, DI MARCO A, BERTAZZOLI C, FORMELLI F, GIULIANI F, PRATESI G. Antitumor activity, toxicity, and pharmacological properties of 4'-epiadriamycin. In: SIEGENTHALER W, LUTHY R, eds. *Current Chemotherapy*. Washington, DC, American Society of Microbiology, 1978, 1257–1260.
13. DI MARCO A, CASAZZA AM, GAMBETTA R, SUPINO R, ZUNINO F. Relationship between activity and amino sugar stereochemistry of daunorubicin and adriamycin derivatives. *Cancer Res* 1976, **36**, 1962–1966.
14. DI MARCO A, CASAZZA AM, DASDIA T *et al.* Changes of activity of daunorubicin, adriamycin and stereoisomers following the introduction or removal of hydroxyl groups in the amino sugar moiety. *Chem Biol Interact* 1977, **19**, 291–302.
15. FORMELLI F, POLLINI C, CASAZZA AM, DI MARCO A, MARIANI A. Fluorescence assays and pharmacokinetic studies of 4'-deoxydoxorubicin and doxorubicin in organs of mice bearing solid tumors. *Cancer Chemother Pharmacol* 1981, **5**, 139–144.
16. FORMELLI F, FUMAGALLI A, GIULIANI F, CASAZZA AM, KAPLAN NO. Tissue distribution of doxorubicin (DX) and 4'-deoxydoxorubicin (4'-deoxyDX) in nude and conventional mice bearing colon tumors. *Proc Am Assoc Cancer Res* 1981, **22** (Abstract no. 1060).
17. ISETTA AM, TRIZIO D. Effect of new anthracycline antibiotics on hormonal and cell mediated immune responses *in vivo*. In: *12th International Congress on Chemotherapy, Florence, Italy, 19–24 July 1981*. (Abstract No. 115.)