

PREPARATION AND BIOLOGICAL EVALUATION OF
4-O-DEMETHYLDAUNORUBICIN (CARMINOMYCIN I)
AND OF ITS 13-DIHYDRO DERIVATIVE

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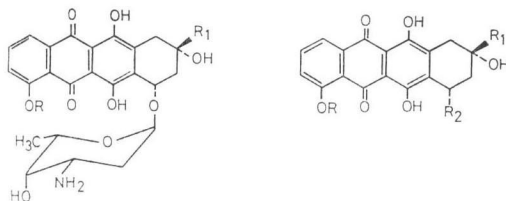
A mutant strain of *Streptomyces peucetius* produced an anthracycline antibiotic whose structure has been established to be 4-O-demethyl-13-dihydrodaunorubicin (**4**), by application of spectroscopic methods and chemical degradation. A new synthesis of 4-O-demethyl-daunorubicin (carminomycin I, **2**) starting from daunomycinone, together with the comparison of the antitumor activity of the anthracycline glycosides **2** and **4** are also reported.

During the course of our screening for new anthracycline antibiotics, 4-O-demethyl-13-dihydrodaunorubicin (13-dihydrocarminomycin, **4**) was found to be produced by a mutant strain of *Streptomyces peucetius*, the daunorubicin-producing microorganism. The structure of the glycoside was determined by spectroscopic method and chemical degradation including direct comparison of its aglycone with a sample of 13-dihydrocarminomycinone obtained by sodium borohydride reduction of semi-synthetic carminomycinone.

The present paper describes also the synthesis of 4-O-demethyl-daunomycinone (carminomycinone, **6**) and of 4-O-demethyl-daunorubicin (carminomycin I, **2**) starting from daunomycinone (**5**) together with the comparison of the antitumor activity of the glycosides **2** and **4** with those of daunorubicin (**1**) and of 13-dihydrodaunorubicin (**3**).

1. Biosynthesis and Isolation of 4-O-Demethyl-13-dihydrodaunorubicin (**4**)

The anthracycline glycoside **4** has been isolated from the mycelium of a mutant strain of *Strepto-*



- | | |
|--|---|
| 1, R = CH ₃ ; R ₁ = COCH ₃ | 5, R = CH ₃ ; R ₁ = COCH ₃ ; R ₂ = OH |
| 2, R = H ; R ₁ = COCH ₃ | 6, R = H ; R ₁ = COCH ₃ ; R ₂ = OH |
| 3, R = CH ₃ ; R ₁ = CHOCH ₃ | 7, R = H ; R ₁ = CHOCH ₃ ; R ₂ = OH |
| 4, R = H ; R ₁ = CHOCH ₃ | 8, R = H ; R ₁ = CHOCH ₃ ; R ₂ = H |

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myces peucetius, the daunorubicin-producing microorganism.^{1,2)} By extraction and chromatography on a silica gel column compound **4** was obtained in crystalline form as fine red needles, whose elementary analysis indicated a molecular formula $C_{26}H_{29}O_{10}N$.

Paper and thin-layer chromatographic data indicated that compound **4** could be differentiated from other known anthracycline antibiotics as shown in Table 1.

Table 1. Comparison with other anthracycline antibiotics.

Antibiotics	Rf	
	I	II
Adriamycin	0.52	0.25
13-Dihydrodaunorubicin (3)	0.56	0.48
Daunorubicin (1)	0.73	0.50
4-O-Demethyl-13-dihydrodaunorubicin (4)	0.63	0.58
4-O-Demethyl-daunorubicin (carminomycin I) (2)	0.77	0.60

I: Silica gel 60-F-254 thin-layer plates (E. Merck), solvent: $CHCl_3$ - methanol - acetic acid - water (80:20:14:6).

II: Whatman No. 1 paper, buffered at pH 5.4 with $M/15$ phosphate buffer, solvent: *n*-propanol - ethyl-acetate - water (7:1:2).

2. Structure Determination

The UV and visible spectra of compound **4** were similar to those of carminomycin (**2**)³⁾ and of daunorubicin (**1**)²⁾, suggesting the presence of a 1,4,5-trihydroxyanthraquinone chromophore. The IR spectrum (Fig. 1) indicated the presence of a strong band at $1,605\text{ cm}^{-1}$ and the absence of the carbonyl absorption at $1,715\text{ cm}^{-1}$ characteristic of the ketone function of the side chain of daunorubicin (**1**)²⁾ and of carminomycin (**2**).³⁾ The general appearance of the PMR spectrum of compound **4** both as free base and as hydrochloride was similar to that of 13-dihydrodaunorubicin (**3**, daunorubicinol)⁴⁻⁷⁾ the only difference being the absence of the methoxy signal and the presence of an additional phenolic OH signal. Mild acid hydrolysis afforded a red aglycone (**7**) and an aminotrioxysugar, $C_6H_{13}O_3N$, which was identified as daunosamine (3-amino-2,3,6-trideoxy-L-*yx*o-hexose)⁸⁾ by direct comparison with an authentic sample. The aglycone (**7**) showed IR (Fig. 2), UV and visible spectra very similar to those of the parent glycoside and the analytical data indicated a molecular formula $C_{20}H_{18}O_8$ (M.W. 386.36) confirmed by mass spectrometry. The attachment of the sugar moiety to the benzylic C-7 position was established by catalytic hydrogenolysis of the glycoside, under these conditions daunosamine and a deoxyaglycone (**8**) were obtained. Compound **8** had a molecular formula of $C_{20}H_{18}O_7$ (M.W. 370), confirmed by mass spectrometry. Catalytic hydrogenolysis of the aglycone (**7**), gave a compound undistinguishable from **8**, which was identified as 4-O-demethyl-7-deoxy-13-dihydrodaunomycinone. The IR and mass spectra of compound **8** were comparable to those of demethyl-deoxydaunorubicinol aglycone reported by TAKANASHI and BACHUR.⁹⁾ Thus the structural formula of glycoside **4**, corresponding to 4-O-demethyl-13-dihydrodaunorubicin (4-O-demethyl-daunorubicinol, 13-dihydrocarminomycin) may be proposed.

After completion of the present work preparations of 13-dihydrocarminomycin (**4**) by chemical and microbiological reduction of carminomycin (**2**) were reported.^{10,11)} Further evidence supporting the structure proposed for compound **4** was obtained by comparison of its aglycone (**7**) with a sample

Fig. 1. Infrared absorption spectrum of 4-O-demethyl-13-dihydrodaunorubicin (KBr)

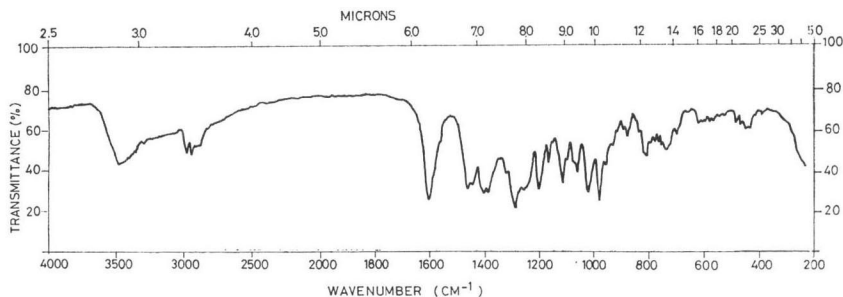
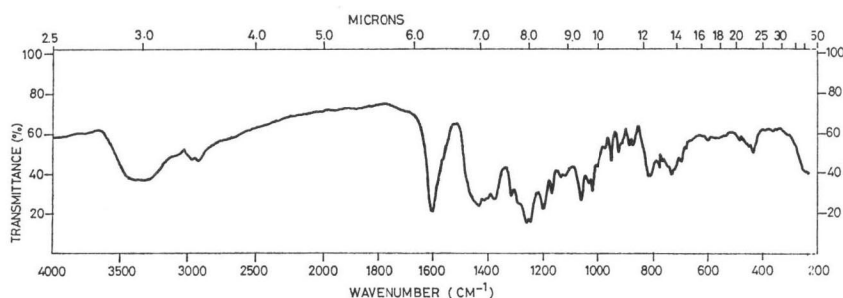


Fig. 2. Infrared absorption spectrum of 4-O-demethyl-13-dihydrodaunomycinone (KBr)



of 4-O-demethyl-13-dihydrodaunomycinone prepared from daunomycinone (**5**) via 4-O-demethyl-daunomycinone (carminomycinone, **6**) according to the method described below.

3. Synthesis of 4-O-Demethyl-daunorubicin (Carminomycin) from Daunomycinone

Treatment of daunomycinone (**5**) with anhydrous aluminum chloride in refluxing methylene chloride gave in high yield the corresponding de-O-methyl derivative, whose PMR spectrum showed three phenolic OH signals and UV, IR and mass spectra were comparable to those reported in the literature for carminomycinone (**6**).³¹ Reduction of compound **6** with sodium borohydride gave the corresponding 13-dihydro derivative (**7**). Following the procedure described for the synthesis of glycosides of daunomycinone^{12,131}, compound **6** was then condensed with N,O-di-trifluoroacetyl- α -daunosaminyl chloride¹²¹, in the presence of silver trifluoromethanesulfonate to give the corresponding protected glycoside. The subsequent mild alkaline treatment afforded as a major product glycoside **2**, which was isolated as the hydrochloride in an overall yield of 10% from daunomycinone (**5**). The configuration of the glycoside linkage was assigned on the basis of the anomeric proton PMR signal, which was a broad singlet ($W_H \sim 8$ Hz) at δ 5.83 in DMSO- d_6 , a characteristic value for the α -glycosides of anthracyclines.

A sample of our synthetic glycoside (**2**), as free base was identical by UV, IR, TLC and HPLC with an authentic sample of natural carminomycin I.*

4. Biological Activity

The antitumor activity of 4-O-demethyl-daunorubicin (carminomycin I, **2**) and of its 13-dihydro derivative (**4**) was compared with those of daunorubicin (**1**) and of 13-dihydrodaunorubicin (**3**) in experimental systems *in vitro* and *in vivo*. The results are reported in Tables 2 and 3.

* We are indebted to Dr. WILLIAM F. MINOR of BRISTOL Labs for a sample of natural carminomycin I.

Table 2. Activity on the cloning efficiency of HeLa cells *in vitro* (treatment for 24 hours.)

Compound	Dose (ng/ml)	No. of colonies (% of control)	ID ₅₀ (ng/ml)
Daunorubicin (1)	12.5	14	5.8
	6.25	38	
	3.12	90	
13-Dihydrodaunorubicin (3)	25	31	12.0
	12.5	41	
	6.25	107	
4-O-Demethyl-daunorubicin (2)	12.5	0	~ 3
	6.25	7	
	3.12	45	
4-O-Demethyl-13-dihydrodaunorubicin* (4)	12.5	3, 7	~ 4
	6.25	52, 25	
	3.12	52, 76	
	1.5	72	
	0.75	97	

* data of 2 experiments

Table 3. Activity against L1210 leukemia*

Compound	Dose ⁺ (mg/kg)	T/C ⁺⁺ %	No. of toxic deaths / No. of mice treated
Daunorubicin (1)	2.9	144, 150	1/20
	4.4	138, 162	4/20
	6.6	127	9/10
13-Dihydrodaunorubicin (3)	4.4	138	1/10
	6.6	138	
	10.0	111	
4-O-Demethyl-daunorubicin (2)	0.25	133	10/10
	0.4	133	
	0.6	138, 162 (1/20)	
	1.0	156	
	1.5	100	
4-O-Demethyl-13-dihydrodaunorubicin (4)	0.25	150	4/10
	0.4	162	
	0.6	168	
	0.85	150	

* BDF₁ mice received 10⁸ ascites cells on Day 0.⁺ Single treatment i. p. on Day 1 after tumor transplantation⁺⁺ Median survival time of treated mice/median survival time of control mice × 100.

In parenthesis: No. of long-term survivors/No. of mice treated.

As previously reported¹⁴, 13-dihydrodaunorubicin (daunorubicinol, 3) was less effective than daunorubicin (1) on HeLa cells *in vitro*. In L1210 leukemia-bearing mice, 13-dihydrodaunorubicin showed a range of active doses similar to that of daunorubicin.

4-O-Demethyl-daunorubicin (carminomycin, 2) was about two-times more active than daunorubicin on HeLa cells cloning efficiency while in L1210 leukemia it exerted an antitumor activity similar to that of daunorubicin at about five-times the lower dose.

4-O-Demethyl-13-dihydrodaunorubicin (4) was slightly less active than 4-O-demethyl-daunorubicin *in vitro*; in mice bearing L1210 leukemia, it was active at doses similar to those effective with 4-O-demethyl-daunorubicin.

Experimental Part

Melting points, taken on the Kofler hot-stage, were uncorrected. Rotations were recorded at 23°C

using a Perkin-Elmer 141 automatic polarimeter. ^1H -nmr spectra were measured on a Varian A-60A spectrometer, with Me_4Si as the internal reference, in the indicated solvents. The mass spectra were recorded with a Perkin-Elmer 270 spectrometer (direct inlet technique) at an electron ionizing voltage of 70 V. The IR spectra were recorded with a Perkin-Elmer 457 spectrometer. TLC on silica gel 60-F-254 plates (E. Merck) and PC were used for identification purposes and the homogeneity test.

Organism and culture conditions

The mutant strain* of *Streptomyces peucetius* designed B 441; F. I. was maintained on slants of a medium having the following composition (per liter, distilled water): sucrose, 20 g; brewer's dry yeast, 5 g; NaNO_3 , 2 g; K_2HPO_4 , 2 g; MgSO_4 , 1 g; agar, 20 g. A first stage seed was prepared by inoculating a loopful from the slant, grown for 14 days at 28°C , to 300 ml Erlenmeyer flasks containing 50 ml of the growth medium consisting of (per liter, distilled water): brewer's dry yeast, 3 g; peptone, 5 g; $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, 0.5 g. The flasks were allowed to incubate for 2 days on a rotary shaker, running at 220 rpm at $27 \sim 28^\circ\text{C}$. A 5% inoculum was then transferred to 300-ml Erlenmeyer flasks containing 40 ml of the following production medium (per liter, distilled water): glucose, 60 g; brewer's dry yeast, 25 g; NaCl , 2 g; $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, 0.1 g; $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, 0.01 g. Maximum production was reached after 10 days' incubation under the same conditions as described for the growth phase.

Isolation of 4-O-demethyl-13-dihydrodaunorubicin (4).

Preliminary detection of compound 4 was carried out with chloroform - methanol (9:1) extracts of the culture broths by means of the paper and silica gel chromatographic methods described in Table 1. The culture broth (pH 6.8) was brought to pH 4.5 and filtered. The antibiotic, present mainly in the mycelium was extracted at pH 8.5 with a chloroform - methanol mixture (9:1). The organic phase was concentrated and a crude mixture was precipitated by addition of diethyl ether. Chromatography of the crude red powder on a silica gel column, using as eluent a chloroform - methanol - water mixture (135:20:2) gave pure 4-O-demethyl-13-dihydrodaunorubicin (4) as fine red crystalline needles (from CHCl_3): m.p. $193 \sim 194^\circ\text{C}$; $[\alpha]_{\text{D}}^{25} +250^\circ \pm 10^\circ$ (c 0.05, CH_3OH); $\lambda_{\text{max}}^{\text{MeOH}}$ 236, 256, 294, 488 and 522 nm ($E_{1\text{cm}}^{1\%}$ 737, 554, 160, 302 and 203). IR (KBr): 1605 cm^{-1} (quinone and aromatic $\text{C}=\text{C}$).

Anal. Calcd. for $\text{C}_{20}\text{H}_{20}\text{O}_{10}\text{N}$: C, 60.58; H, 5.67; N, 2.72.

Found: C, 60.47; H, 5.72; N, 2.61.

Treatment of a chloroform solution of compound 4 with a molar equivalent of methanolic hydrogen chloride gave the corresponding hydrochloride as orange-red needles: m.p. $176 \sim 177^\circ\text{C}$; $[\alpha]_{\text{D}}^{25} +235^\circ \pm 10^\circ$ (c 0.05, CH_3OH); $\lambda_{\text{max}}^{\text{MeOH}}$ 235, 255, 294, 490 and 524 nm ($E_{1\text{cm}}^{1\%}$ 722, 543, 157, 293 and 193).

Anal. Calcd. for $\text{C}_{20}\text{H}_{20}\text{O}_{10}\text{N} \cdot \text{HCl}$: C, 56.55; H, 5.48; N, 2.53; Cl, 6.43.

Found: C, 56.40; H, 5.51; N, 2.42; Cl, 6.36.

Acid hydrolysis

A solution of 4 (100 mg) in 0.2 N aqueous HCl (10 ml) was heated at 100°C for 30 minutes to give a water-insoluble aglycone (7, 65 mg) that was recrystallized from chloroform - methanol as red needles: m.p. $185 \sim 186^\circ\text{C}$; $[\alpha]_{\text{D}}^{25} +170^\circ$ (c 0.05, dioxane); $\lambda_{\text{max}}^{\text{MeOH}}$ 235, 254, 295, 491 and 525 nm ($E_{1\text{cm}}^{1\%}$ 780, 673, 192, 376 and 269); IR (KBr): 1600 cm^{-1} (quinone and aromatic $\text{C}=\text{C}$).

Anal. Calcd. for $\text{C}_{20}\text{H}_{15}\text{O}_8$ (M. W. 386.36): C, 62.17; H, 4.70.

Found: C, 62.20; H, 4.74.

Mass spectrum: m/e 386 (M^+), 368 ($\text{M}-\text{H}_2\text{O}$), 350 ($\text{M}-2\text{H}_2\text{O}$), 325 ($\text{M}-\text{H}_2\text{O}-\text{CH}_3\text{CHOH}$).

The aqueous phase of the acid hydrolysis was brought to pH 5 with an anion-exchange resin (Dowex 1×2 , OH^- form), filtered and freeze-dried. Crystallization from methanol - acetone afforded an amino-sugar as the hydrochloride, $\text{C}_6\text{H}_{13}\text{O}_3\text{N} \cdot \text{HCl}$, which was identified as daunosamine by direct comparison with an authentic sample.

Catalytic hydrogenolyses

An aqueous solution (10 ml) of 4-O-demethyl-dihydrodaunorubicin HCl (50 mg) was hydrogenated in the presence of 5% Pd-BaSO₄ (30 mg) for 2 hours at 20°C , to give, after filtration, an almost colorless

* The mutant has been obtained by a mutagenic treatment of *Streptomyces peucetius* with N-methyl-N'-nitro-N-nitrosoguanidine, according to the method described by V. DELIC, D. A. HOPWOOD & E. J. FRIEND: Mutagenesis by N-methyl-N'-nitro-N-nitrosoguanidine (NTG) in *Streptomyces coelicolor*. Mutation Research 9: 167~182, 1970

solution containing daunosamine hydrochloride (13 mg) and a deoxy aglycone adsorbed on the catalyst. After removal from the latter with a CHCl_3 - MeOH mixture (1:1) and concentration, compound **8** was obtained as red needles: m.p. 269~270°C; $[\alpha]_D^{25} +25^\circ$ (c 0.1, dioxane); $\lambda_{\text{max}}^{\text{MeOH}}$ 235, 254, 295, 491 and 525 nm; IR (KBr): 1600 cm^{-1} .

Anal. Calcd. for $\text{C}_{20}\text{H}_{18}\text{O}_7$ (M. W. 370.36): C, 64.86; H, 4.90.

Found: C, 64.58; H, 4.92.

Mass spectrum: *m/e* 370 (M^+), 352 ($\text{M}-\text{H}_2\text{O}$), 325 ($\text{M}-\text{CH}_3-\text{CHOH}$), 307 ($\text{M}-\text{H}_2\text{O}-\text{CH}_2\text{CHOH}$).

A solution of compound **7** (25 mg) in dioxane (5 ml) was hydrogenated in the presence of Pd-BaSO₄ (125 mg) for 1 hour at 20°C. After filtration, solvent evaporation and crystallization from methanol, a red crystalline compound, m.p. 268~269°C, was obtained and identified as compound **8** obtained by catalytic hydrogenolysis of glycoside **4**.

4-O-Demethylation of daunomycinone

To a refluxed solution of daunomycinone (**5**, 10 g) in methylene dichloride (1 liter) anhydrous AlCl₃ (30 g) was added over a 2-hour period with stirring. After an additional 4-hour period, the reaction mixture was cooled and poured into ice-water containing oxalic acid (15 g). The organic layer was separated, washed with water and concentrated *in vacuo* to give crystalline compound **6** (6 g), as red needles: IR (KBr): 1610 and 1715 cm^{-1} (quinone and acetyl bands) mass spectrum: *m/e* 384 (M^+). The ¹H-nmr spectrum (1:1 CDCl₃-DMSO-d₆) showed "inter alia" three phenolic OH signals at δ 12.01, 12.78 and 13.35. By TLC compound **6** (Rf 0.70) could be differentiated from daunomycinone (**5**, Rf 0.60) by using 4:1 chloroform - acetone as eluent.

Reduction of 4-O-demethyl daunomycinone

A solution of compound **6** (0.2 g) in 0.1 N aqueous sodium hydroxide (20 ml) and methanol (5 ml) was treated under stirring with sodium borohydride (20 mg). After 5 minutes the reaction mixture was acidified, stirred for one half hour, and then extracted with chloroform. Chromatographic purification of the crude residue on a silica gel column, using 97:3 chloroform - methanol as eluent, gave a red crystalline compound (50 mg), m.p. 180~182°C, *m/e* 386 (M^+), undistinguishable by TLC, UV, IR and mass spectra from a sample of natural compound **7**.

Synthesis of 4-O-demethyl daunorubicin

A solution of compound **6** (1 g) and 2,3,6-trideoxy-3-trifluoroacetamido-4-O-trifluoroacetyl- α -L-lyxo-hexopyranosyl chloride²¹ (0.85 g) in 1:10 N,N-dimethylformamide - methylene dichloride (90 ml) was treated dropwise with a solution of silver trifluoromethanesulfonate (0.57 g) in anhydrous diethyl ether (15 ml) and stirred at room temperature. After 1 hour the reaction mixture was diluted with methylene dichloride, washed with an aqueous solution of sodium bicarbonate and finally with water. The solvent was removed *in vacuo* and the residue taken up in chloroform. The insoluble starting material was removed by filtration and the filtrate was evaporated to dryness. The residue was dissolved in 0.15 N aqueous sodium hydroxide (30 ml) and after 2 hours at room temperature, the reaction mixture was acidified with oxalic acid and extracted with chloroform in order to eliminate some impurities. The aqueous phase, adjusted to pH 7.5 with aqueous sodium bicarbonate, was extracted with methylene dichloride. The organic phase, washed with water, dried on anhydrous sodium sulfate, was concentrated to a small volume and acidified to pH 4.5 with 0.5 N methanolic hydrogen chloride. Addition of diethyl ether gave 4-O-demethyl daunorubicin (**2**) as the hydrochloride (0.3 g): IR (KBr) 1715 and 1600 cm^{-1} , UV: $\lambda_{\text{max}}^{\text{MeOH}}$ 235, 256, 290, 492 and 526 nm.

Acknowledgement

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References

- 1) GREIN, A.; C. SPALLA, A. DI MARCO & G. CANEVAZZI: Descrizione e classificazione di un Attinomicete (*Streptomyces peucetius* sp. nova) produttore di una sostanza ad alta attività antitumorale: la Daunomicina. *Giorn. Microbiol.* 11: 109~118, 1963
- 2) CASSINELLI, G. & P. OREZZI: Daunomicina: un nuovo antibiotico ad attività citostatica. Isolamento e proprietà. *Giorn. Microbiol.* 11: 167~174, 1963
- 3) BRAZHNIKOVA, M. G.; V. B. ZBARSKY, V. I. PONOMARENKO & N. P. POTAPOVA: Physical and chemical characteristics and structure of carminomycin, a new antitumor antibiotic. *J. Antibiotics* 27: 254~259, 1974
- 4) ARCAMONE, F.; G. CASSINELLI, S. PENCO & L. TOGNOLI: Dihydrodaunomycin antibiotic and derivative thereof. U. S. Patent 3,686,163, Aug. 22, 1972
- 5) FLORENT, J.; J. LUNEN & J. RENAUT: Neue Vorfahren zur Herstellung des Antibiotikum 20,789 RP. German Patent 2,456,139, May 28, 1975
- 6) BACHUR, N. R.: Adriamycin-daunorubicin cellular pharmacodynamics. *Biochem. Pharmacol. Suppl.* 2: 207~216, 1974
- 7) ASZALOS, A. A.; N. R. BACHUR, B. K. HAMILTON, A. F. LANGLYKKE, P. P. ROLLER, M. Y. SHEIKH, M. S. SUTPHIN, M. C. THOMAS, D. A. WAREHEIM & L. H. WRIGHT: Microbial reduction of the side-chain carbonyl of daunorubicin and N-acetyldaunorubicin. *J. Antibiotics* 30: 50~58, 1977
- 8) ARCAMONE, F.; G. CASSINELLI, P. OREZZI, G. FRANCESCHI & R. MONDELLI: Daunomycin. II. Structure and stereochemistry of daunosamine. *J. Am. Chem. Soc.* 86: 5335~5336, 1964
- 9) TAKANASHI, S. & N. R. BACHUR: Daunorubicin metabolites in human urine. *J. Pharmacol. Exp. Ther.* 195: 41~49, 1975
- 10) POVAROV, L. S.; V. A. SHORIN, V. S. BAZHANOV & N. G. SHEPEVTSEVA: Preparation of dihydrocarminomycin and a comparison of its antitumor activity with the activity of carminomycin. *Antibiotiki* 21: 1008~1011, 1976
- 11) Rhône-Poulenc: Antitumor naphthalene derivatives prepared by reduction of carminomycin or culture of *Streptomyces* microorganisms. Belgian Patent 839,540, Sept. 23, 1976
- 12) ARCAMONE, F.; S. PENCO & A. VIGEVANI: Adriamycin (NSC-123127): New chemical developments and analogues. *Cancer Chemoth. Repts. Part 3*, 6: 123~129, 1975
- 13) ARCAMONE, F.; S. PENCO, S. REDAELLI & S. HANESSIAN: Synthesis and antitumor activity of 4'-deoxydaunorubicin and 4'-deoxyadriamycin. *J. Med. Chem.* 19: 1424~1425, 1976
- 14) DI MARCO, A.; A. M. CASAZZA, T. DASDIA, F. GIULIANI, L. LENAZ, A. NECCO & C. SORANZO: Cytotoxic, antiviral and antitumor activity of some derivatives of daunomycin (NSC-82151). *Cancer Chemoth. Repts.* 57: 269~274, 1973