data and HPLC assay results (>98% pure and free of MTX). Samples first dried in vacuo at 25–30 °C (over P_2O_5 and NaOH pellets) were allowed to attain constant weight by exposure to ambient conditions of the laboratory (relative humidity typically 50%) before analysis.

Methotrexate γ-methyl ester (15) was isolated as described for the 14 types. The α-methyl ester 16 was also isolated similarly, but it precipitated at a lower pH than was intended. Just after the reaction solution containing 16 was combined with cold H_2O, a yellow solid began separating, before the planned base treatment to raise the pH from 2.2 to 4.0 was started. The treatment with 1 N NaOH with ice-bath cooling was done after much solid had already separated, and then the mixture at pH 4.0 was stirred at 0–5 °C for 0.5 h. After centrifugation and decantation, the solid was collected by filtration from a small volume of H_2O. The dried and equilibrated product turned out to be a partial hydrobromide of 16 as listed in Table III.

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Adriamycin Analogues. Preparation and Biological Evaluation of Some *N*-Perfluoroacyl Analogues of Daunorubicin, Adriamycin, and *N*-(Trifluoroacetyl)adriamycin 14-Valerate and Their 9,10-Anhydro Derivatives[1]

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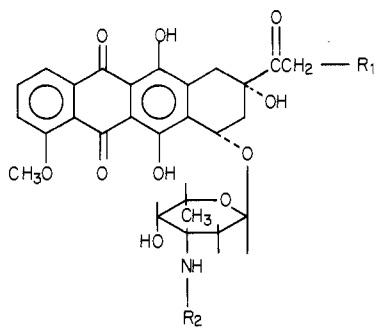
The experimental and clinical antitumor activity, as well as the low toxicity, of *N*-(trifluoroacetyl)adriamycin 14-valerate (AD 32), a non-DNA binding anthracycline analogue, has led us to prepare and evaluate several *N*-perfluoroacyl analogues of daunorubicin, adriamycin, and *N*-(trifluoroacetyl)adriamycin 14-valerate. Target compounds were prepared by reaction of the appropriate perfluoroacyl anhydride with daunorubicin in chloroform–ether, with adriamycin in cold pyridine, and with adriamycin 14-valerate in ethyl acetate. In connection with this work, it was found that reaction of perfluoroacyl anhydrides with N-acylated or N-unsubstituted anthracyclines in pyridine at room temperature afforded with ease and in good yield the corresponding 9,10-anhydro-N-acylated derivatives. A number of products showed good to highly significant antitumor activity in vivo against the murine P388 leukemia system. However, the lack of in vivo antitumor activity of the pentafluoropropionyl and heptafluorobutyryl analogues of *N*-(trifluoroacetyl)adriamycin 14-valerate is noteworthy. The results continue to show that non-DNA binding anthracycline analogues can exhibit in vivo antitumor activity. Loss of the anthracycline 9-carbinol function by dehydration leads to reduction of biological activity as compared to the parent compound.

The anthracycline antibiotic adriamycin (2) is widely used in the clinical management of leukemias and various solid tumors.[2-5] The value of this agent is compromised, however, by its toxic side effects, two of which (acute myelosuppression and total accumulated dose-dependent cardiac toxicity) are serious enough to be dose limiting. In addition, inadvertant paravenous extravasation can result in severe local tissue ulceration and necrosis, and gastrointestinal toxicity, manifested as nausea and vomiting, is almost universal.



1 (daunorubicin), $R_1 = R_2 = H$
2 (adriamycin), $R_1 = OH$; $R_2 = H$
3 (AD 32), $R_1 = OCO(CH_2)_3CH_3$; $R_2 = COCF_3$

N-(Trifluoroacetyl)adriamycin 14-valerate (AD 32; **3**)[6,7] is an adriamycin analogue conceived and developed in these laboratories and currently undergoing phase II clinical evaluation. In animal model systems, **3** is therapeutically superior, sometimes dramatically so, to adriamycin.[6,8,9] In addition, in animals **3** produces less toxicity in general and significantly less cardiotoxicity in particular compared to adriamycin.[8,10,11] Clinical antitumor activity and low toxicity have been documented for **3** in connection with phase I/II trials.[12,13] No patient has had evidence

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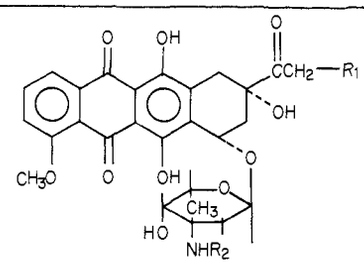
Table I. Biological Evaluation of Some *N*-Perfluoroacyl Analogues of Daunorubicin, Adriamycin, and *N*-(Trifluoroacetyl)adriamycin 14-Valerate



compd	R_1	R_2	in vitro:[a] ID_{50}, μM	in vivo vs. P388[b] optimal dose, (mg/kg)/day	in vivo vs. P388[b] % ILS[c]
1 (daunorubicin)	H	H	0.040	2.0	+91[d]
4	H	$COCF_3$	0.27	80.0	+83
6	H	$COCF_2CF_3$	1.01	50.0	+27
7	H	$COCF_2CF_2CF_3$	>1.35[e]	40.0	+45
2 (adriamycin)	OH	H	0.066	4.0	+132[d]
5	OH	$COCF_3$	0.23	20.0	+481
8	OH	$COCF_2CF_3$	0.41	50.0	+372
9	OH	$COCF_2CF_2CF_3$	1.49	50.0	+54
3 (AD 32)	*O*-valerate	$COCF_3$	0.24	40.0	+429[d]
10	*O*-valerate	$COCF_2CF_3$	1.18	50.0	0
11	*O*-valerate	$COCF_2CF_2CF_3$	>1.14[e]	50.0	0

[a] vs. CCRF-CEM cells in culture; 48 h continuous drug exposure. [b] Murine P388 leukemia; 10^6 tumor cell inoculum ip on day 0; treatment once daily on days 1–4 ip. [c] Percent increase in life span relative to untreated controls (median survival of control group 11.0 days). [d] Reference 6. [e] Highest dose tested.

of cardiac dysfunction with **3**, even if having received prior adriamycin chemotherapy; no local tissue damage has occurred in humans as a result of drug extravasation, and little is seen in the way of gastrointestinal toxicity.

The antitumor activity of **3** is remarkable in view of structure–activity arguments in the literature, summarized in ref 14, which predict that **3** should be inactive: the supposed mechanism of action for daunorubicin (**1**) and adriamycin is that of DNA intercalation, an event that requires an unsubstituted amino function on the anthracycline glycoside. Compound **3** bears a trifluoroacetamide substituent which interferes with and prevents DNA complexation.[15,16] Despite the structural relationship between adriamycin and **3**, a variety of pharmacological studies, including pharmacokinetic determinations and metabolism and disposition studies in mice, rats, rabbits, monkeys, and humans, have clearly established that **3** does not merely serve as an adriamycin prodrug.[17-26]

In connection with our ongoing program on the chemistry, biology, and pharmacology of the anthracyclines, we have undertaken a broad investigation of the structure–activity relationships among N-acylated anthracyclines, based in large measure on the unexpected biological properties of **3**. *N*-(Trifluoroacetyl)daunorubicin (**4**)[27] and *N*-(trifluoroacetyl)adriamycin (**5**)[7,27] have been previously prepared and evaluated here.[28] We have now extended our studies to include the corresponding *N*-pentafluoropropionyl and *N*-heptafluorobutyryl analogues of daunorubicin, adriamycin, and **3**. These products were prepared directly by reaction of daunorubicin, adriamycin, and adriamycin 14-valerate[7] with the corresponding perfluoroacyl anhydride. With daunorubicin and adriamycin 14-valerate, reactions were achieved at room temperature in chloroform–ether and ethyl acetate, respectively. Adriamycin could not be similarly perfluoroacylated because of its lack of solubility and poor chemical stability under similar reaction conditions; hence, acylation of adriamycin was accomplished in pyridine at 0 °C.

During initial attempts to acylate adriamycin in pyridine, when reactions were tried at room temperature, product formation was accompanied by the formation of the 9,10-anhydro derivative **15**. This reaction has been further studied and has been found to be a simple and convenient method for obtaining 9,10-anhydro-N-acylated anthracyclines. For 9,10-anhydro formation, reaction time and amount of anhydride vary, depending on the reactivity of starting material. The dehydration of adriamycin and

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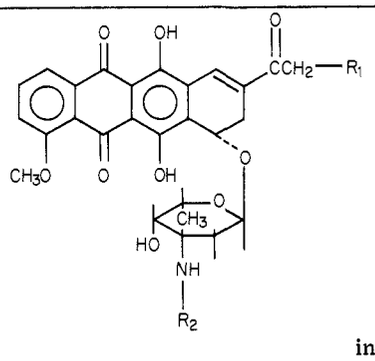
Table II. Biological Evaluation of Some 9,10-Anhydro Anthracycline Derivatives



compd	R_1	R_2	in vitro:[a] ID_{50}, μM	in vivo vs. P388[b] optimal dose, (mg/kg)/day	in vivo vs. P388[b] % ILS[c]
12	H	$COCF_3$	2.56	40.0	+45
13	H	$COCF_2CF_3$	0.50		
14	H	$COCF_2CF_2CF_3$	>13.8[d]		
15	OH	$COCF_3$	5.79	40.0	+63
16	OH	$COCF_2CF_3$	8.41		
17	OH	$COCF_2CF_2CF_3$	7.30		
18	*O*-valerate	$COCF_3$	1.19	40.0	+27
19	*O*-valerate	$COCF_2CF_3$	0.56		
20	*O*-valerate	$COCF_2CF_2CF_3$	2.31		

[a] vs. CCRF-CEM cells in culture; 48 h continuous drug exposure. [b] Murine P388 leukemia; 10^6 tumor cell inoculum ip on day 0; treatment once daily on days 1–4 ip. [c] Percent increase in life span relative to untreated controls (median survival of control group 11.0 days). [d] Highest dose tested.

3 occurs reasonably rapidly, whereas with daunorubicin and *N*-(trifluoroacetyl)daunorubicin the reaction is slow. In general, yields are quite good. Dehydration did not occur when trifluoroacyl anhydride was replaced by acetic anhydride or when the solvent was changed from pyridine to chloroform or ethyl acetate. The dehydration reaction occurred whether the glycosidic amine was unsubstituted or already acylated; if unsubstituted, N-acylation occurred simultaneously.

Target compounds were assayed for in vitro growth-inhibitory activity and, where sufficient amounts were available, for in vivo antitumor activity; results are summarized in Tables I and II. Against CCRF-CEM (human leukemic lymphoblastic) cells in cultures,[29] most of the products showed decreased activity relative to adriamycin and to 3. In most instances, the 9,10-anhydro derivative was significantly less active than the corresponding 9-carbinol compound.

When examined in the in vivo murine P388 leukemia system,[30] a number of compounds showed good to highly significant antitumor activity. Response data for daunorubicin, adriamycin, and 3 in this system are included in Table I for reference. *N*-(Trifluoroacetyl)adriamycin (5) has been shown to be the initial and major metabolite of 3 in plasma and tissues of animals and humans.[18,20,23,24,31,32]

We have previously reported that 5 is highly effective against murine L1210 leukemia, but its use is associated with a pattern of late deaths among long-term (>50 day) survivors.[28] In the P388 leukemia system, 5 again shows the pattern of high activity and late deaths. Interestingly, very significant antileukemia activity was exhibited by the pentafluoropropionamide of adriamycin (8), but no activity was seen with 10 and 11, the corresponding pentafluoropropionamide and heptafluorobutyramide analogues of 3. The lack of activity of 10 and 11 was not associated with any difference in the rate of enzyme-mediated deacylation, as measured by the rate of formation of 5 when the compounds were incubated with pH 7.0 phosphate buffer supplemented with 5% unfractionated mouse serum. Rather, we believe that the failure of 10 and 11 to show in vivo antitumor activity may be the result of the greatly increased lipophilicity of these agents (TLC evidence), which may result in plasma protein binding, with prevention of drug distribution into peripheral compartments. The in vivo activity of the three 9,10-anhydro compounds tested was measurably less than that of their 9-carbinol parents.

These studies continue to show that certain non-DNA binding analogues and derivatives of daunorubicin and adriamycin can retain significant in vivo antitumor activity but that loss of the 9-carbinol function and configurational changes in the A ring attendant to 9,10-dehydration lead to reduction of activity among this class of compounds.

Experimental Section

IR spectra were recorded as pellets on a Perkin-Elmer Model 137B Infracord. Proton NMR spectra were recorded on a Varian T60A System spectrometer with Me_4Si as internal standard. All compounds showed the expected signals for the anthracycline nucleus: δ 1.29 (d, J = 6.5 Hz, 5′-CH_3), 4.1 (s, OCH_3), 5.23 (br s, 7-H), 5.52 (br s, 1′-H), 7.43–8.18 (m, aromatic H), 13.2 (s, phenolic OH), 14.0 (s, phenolic OH), and in the case of perfluoroacylated compounds, 6.70 (d, J = 10 Hz, NHCOR). Rotational data were obtained using a Perkin-Elmer Model 144Mc spectropolarimeter. Elemental analyses were carried out by Galbraith Laboratories, Inc., Knoxville, TN, and are within ±0.4%, unless otherwise noted. TLC on silica gel G (Analtech), with either $CHCl_3$–CH_3OH–H_2O (120:20:1) or $CHCl_3$–CH_3OH–H_2O (80:30:3) as eluant, was used for identification and evaluation of homogeneity. Column chromatography was done on Bio-Sil A silicic acid (100–200 mesh, Bio-Rad Laboratories).

***N*-(Pentafluoropropionyl)daunorubicin (6).** To a solution of 250 mg (0.47 mmol) of daunorubicin in 15 mL of $CHCl_3$ and 10 mL of ether was added a solution of 0.8 mL (4 mmol) of pentafluoropropionic anhydride in 5 mL of ether dropwise for 10 min with stirring. After an additional 5 min of stirring, the reaction mixture was diluted with 200 mL of $CHCl_3$ and washed four times with 150-mL portions of distilled H_2O. The $CHCl_3$ layer was dried over anhydrous Na_2SO_4 for 2 h and then filtered and evaporated to dryness. To the residue was added 30 mL of methanol; the resulting solution was heated at reflux for 5 min and then cooled and reevaporated to dryness. The residue was dissolved in 10 mL of chloroform and reprecipitated by the addition of petroleum ether. The solid was filtered, washed with petroleum ether, and dried under high vacuum to give 223 mg (70% crude yield) of red-orange solid of greater than 90% purity. An analytical sample, obtained by crystallization from methanol–ether–petroleum ether, had mp 148–151 °C dec; $[\alpha]_D$ +333° (*c* 0.01, CH_3OH); IR (KBr) 3480, 3400 (OH, NH), 1710 (ketone, amide), 1615, 1580 (quinone C=O) cm^{-1}; UV-Vis λ_{max} (CH_3OH) 225 nm (ϵ 39 400), 243 (28 300), 279 (9440), 478 (13 300), 496 (13 000), 529 (7190). Anal. ($C_{30}H_{28}F_5NO_{11}$) C, H, F, N.

***N*-(Heptafluorobutyryl)daunorubicin (7).** A solution of 300 mg (0.57 mmol) of daunorubicin in 25 mL of $CHCl_3$ and 10 mL of ether was treated with a solution of 1.15 mL (4.5 mmol) of heptafluorobutyric anhydride in ether, and the reaction mixture was worked up in the same manner as for 6. After methanolysis, 288 mg (69% crude yield) of red-orange solid (95% pure) was

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obtained. An analytical sample was crystallized from methanol–ether–petroleum ether and had mp 155–158 °C dec; $[\alpha]_D$ +246° (c 0.027, CH_3OH); IR (KBr) 3460, 3400 (OH, NH), 1708, 1700 (ketone and amide C=O), 1620 (quinone) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 226 nm (ε 39 400), 243 (28 200), 281 (9430), 480 (13 100), 497 (13 000), 528 (7050). Anal. ($C_{31}H_{28}F_7NO_{11}\cdot 0.5H_2O$) C, H, F, N.

***N*-(Pentafluoropropionyl)adriamycin (8).** To a stirred, chilled (0 °C) solution of 200 mg (0.34 mmol) of adriamycin in dry pyridine was added, under N_2, a solution of 1.0 mL (5 mmol) of pentafluoropropionic anhydride in 5 mL of ether dropwise over a 10-min period. The reaction mixture was stirred at 0 °C for an additional 20 min before 2 mL of distilled H_2O was added. After another 5 min, the pyridine was diluted with 300 mL of distilled H_2O before drying over anhydrous Na_2SO_4 for 2 h. The filtered, dried $CHCl_3$ layer was evaporated to dryness, and the residue was dissolved and refluxed for 5 min in 25 mL of methanol. Removal of the solvent left an oily residue, which was chromatographed on an open column of silicic acid packed in $CHCl_3$. The product was eluted with 0.5% methanol in $CHCl_3$ and triturated from a concentrated solution in $CHCl_3$ with petroleum ether to provide 142 mg (59% yield) of red-orange solid, mp 165–169 °C dec; $[\alpha]_D$ +304.6 ° (c 0.016, CH_3OH); IR (KBr) 3410 (broad, OH, NH), 1710 (ketone and amide C=O), 1640 (quinone) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 224 nm (ε 40 300), 241 (28 600), 277 (11 400), 479 (14 000), 496 (13 800), 530 (7930). Anal. ($C_{30}H_{28}F_5NO_{12}$) C, H, F, N.

***N*-(Heptafluorobutyryl)adriamycin (9).** In the same fashion as for 8, 200 mg (0.34 mmol) of adriamycin was treated with 0.9 mL (3.5 mmol) of heptafluorobutyric anhydride. The same workup procedure, followed by column chromatography on silicic acid, provided 137 mg (54% yield) of red-orange solid, which was analyzed for percentage composition without further purification: mp 125–128 °C dec; $[\alpha]_D$ +242.8° (c 0.05, CH_3OH); IR (KCl) 3400 (broad, OH, NH), 1710, 1700 (sh, ketone, amide C=O), 1615 (quinone C=O) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 224 nm (ε 39 800), 240 (29 200), 278 (11 800), 478 (13 400), 495 (13 200), 529 (8100). Anal. ($C_{31}H_{28}F_7NO_{12}$) C, H, F, N.

***N*-(Pentafluoropropionyl)adriamycin 14-Valerate (10).** To a chilled (0 °C) solution of 300 mg (0.48 mmol) of adriamycin 14-valerate[7] in 25 mL of ethyl acetate was added, dropwise with stirring over a 10-min period, a solution of 1 mL (5 mmol) of pentafluoropropionic anhydride. After an additional 10 min of stirring, the reaction mixture was diluted with 200 mL of ethyl acetate, washed three times with 200-mL portions of distilled H_2O, and dried for 2 h over anhydrous Na_2SO_4. The dried ethyl acetate solution was filtered and evaporated to dryness; the residue was dissolved and refluxed in methanol for 5 min. The methanol was removed under reduced pressure, and the residue was dissolved in 2 mL of fresh methanol and precipitated by the addition of petroleum ether. The crude product was purified by column chromatography on silicic acid packed and eluted with $CHCl_3$. The product, precipitated from $CHCl_3$ by the addition of ether, weighed 202 mg (54% yield) and was analyzed without further purification, mp 215–217 °C dec; $[\alpha]_D$ +270° (c 0.033, CH_3OH); IR (KBr) 3385 (broad, OH, NH), 1720, 1690 (ketone, ester, and amide C=O), 1620 (quinone C=O) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 223 nm (ε 40 400), 241 (29 000), 278 (10 300), 478 (13 100), 495 (12 800), 530 (6150). Anal. ($C_{35}H_{36}F_5NO_{13}$) C, H, N; F: calcd, 12.27; found, 11.82.

***N*-(Heptafluorobutyryl)adriamycin 14-Valerate (11).** In a similar fashion to the preparation of 10, 300 mg (0.48 mmol) of adriamycin 14-valerate in cold ethyl acetate was treated with 1 mL (5 mmol) of heptafluorobutyric anhydride in ether. The reaction and workup procedure was the same. Column chromatography provided, after trituration, 230 mg (56%) of red-orange solid, which was analyzed without further purification: mp 218–220 °C dec; $[\alpha]_D$ +251° (c 0.038, CH_3OH); IR (KBr) 3460, 3420 (OH, NH), 1700 (ester, ketone, and amide C=O), 1618 (quinone C=O) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 223 nm (ε 42 700), 240 (29 500), 277 (9410), 478 (14 000), 494 (13 800), 529 (7320). Anal. ($C_{36}H_{36}F_7NO_{13}\cdot 0.5H_2O$) C, H, F, N.

***N*-(Trifluoroacetyl)-9,10-anhydroadriamycin 14-Valerate (18).** A solution of 0.4 mL (2.81 mmol) of trifluoroacetic anhydride in 5 mL of anhydrous ether was added dropwise over a 10-min period to a chilled (0 °C), stirred solution of 366 mg (0.5 mmol) of *N*-(trifluoroacetyl)adriamycin 14-valerate (3)[6,7] in pyridine. The reaction mixture was stirred at room temperature for 1.5 h before the addition of 5 mL of distilled H_2O. After an additional 5 min of stirring, the reaction mixture was evaporated to dryness and redissolved in 300 mL of $CHCl_3$. The $CHCl_3$ solution was washed three times with 200-mL portions of H_2O, dried over anhydrous Na_2SO_4, filtered, and evaporated to dryness. The residue was dissolved and refluxed for 5 min in 20 mL of methanol and then concentrated to dryness under reduced pressure. The residue was redissolved in $CHCl_3$ and washed with H_2O, as above. The crude product was purified by column chromatography on silicic acid packed and eluted with $CHCl_3$. A concentrated $CHCl_3$ solution of the eluted product was triturated with petroleum ether to provide 304 mg (85% yield) of red solid, mp 152–155 °C dec, $[\alpha]_D$ +296° (c 0.023, CH_3OH), which was analyzed without further purification: IR (KBr) 3400 (broad, OH, NH), 1730, 1710, 1680, 1612 (C=O and quinone) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 230 nm (ε 27 000), 280 (28 800), 480 (14 000), 495 (13 400), 528 (7200). Anal. ($C_{34}H_{34}F_3NO_{12}$) C, H, F, N.

In a like manner, when 100 mg (0.16 mmol) of adriamycin 14-valerate was dissolved in 10 mL of pyridine and treated with 0.25 mL (1.76 mmol) of trifluoroacetic anhydride for 2 h at room temperature, the reaction gave 73 mg (78%) of 18, identical with that obtained from 3 above.

***N*-(Trifluoroacetyl)-9,10-anhydrodaunorubicin (12).** In a similar fashion to the preparation of 18, 263 mg (0.5 mmol) of daunorubicin in pyridine solution was treated with 0.65 mL (4.58 mmol) of trifluoroacetic anhydride for 7 h at room temperature. Identical workup gave 192 mg (63% yield) of red solid: mp 162–165 °C; $[\alpha]_D$ +270.5° (c 0.011, CH_3OH); IR (KBr) 3450 (broad, OH, NH), 1710, 1700, 1651 cm^{-1}; UV–Vis λ_{max} (CH_3OH) 225 nm (ε 24 000), 245 (23 300), 270 (28 100), 500 (15 500), 512 (14 800), 551 (7120). Anal. ($C_{29}H_{26}F_3NO_{10}$) C, H, F, N.

When 312 mg (0.5 mmol) of *N*-(trifluoroacetyl)daunorubicin and 0.5 mL (3.5 mmol) of trifluoroacetic anhydride were used, reaction gave 224 mg (74% yield) of 12, identical with the material obtained directly from daunorubicin above.

Furthermore, 100 mg (0.16 mmol) of *N*-(trifluoroacetyl)daunorubicin and 0.5 mL of *heptafluorobutyric* anhydride gave in the same manner 59 mg (62% yield) of 12 identical with samples prepared as above.

***N*-(Pentafluoropropionyl)-9,10-anhydrodaunorubicin (13).** Reaction of 250 mg (0.47 mmol) of daunorubicin with 0.8 mL (5 mmol) of pentafluoropropionic anhydride in $CHCl_3$–ether solution afforded after workup 223 mg (70%) of red-orange solid of greater than 90% purity. An analytical sample was obtained by crystallization from methanol–ether–petroleum ether: mp 148–151 °C; $[\alpha]_D$ +333° (c 0.01, CH_3OH); IR (KCl) 3480 and 3400 (OH, NH), 1710 (ketone, amide), 1615 and 1580 (quinone C=O) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 225 nm (ε 39 400), 243 (28 300), 279 (9440), 478 (13 300), 496 (13 000), 529 (7190). Anal. ($C_{30}H_{28}F_5NO_{11}$) C, H, F, N.

***N*-(Heptafluorobutyryl)-9,10-anhydrodaunorubicin (14).** In a like manner, reaction of 300 mg (0.57 mmol) of daunorubicin and 1.15 mL (4.5 mmol) of heptafluorobutyric anhydride gave 288 mg of 14 (69%): mp 155–158 °C dec; $[\alpha]_D$ +246° (c 0.027, CH_3OH); IR (KCl) 3460 and 3400 (OH, NH), 1708 and 1700 (ketone and amide), 1620 (quinone) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 226 nm (ε 34 400), 243 (28 200), 281 (9430), 480 (13 100), 497 (13 000), 528 (7050). Anal. ($C_{31}H_{28}F_7NO_{11}\cdot 0.5H_2O$) C, H, F, N.

***N*-(Trifluoroacetyl)-9,10-anhydroadriamycin (15).** In a like manner, 50 mg (0.086 mmol) of adriamycin in 10 mL of dry pyridine was treated with 0.20 mL (1.48 mmol) of trifluoroacetic anhydride for 0.5 h at room temperature. The same workup and isolation procedure as for 12 afforded 21 mg (39% yield) of red solid: mp 167–171 °C; $[\alpha]_D$ +327.8° (c 0.02, CH_3OH); IR (KBr) 3410 (OH, NH), 1710, 1670, 1618 cm^{-1}; UV–Vis λ_{max} (CH_3OH) 221 nm (ε 9390), 234 (9830), 270 (12 400), 503 (7420), 517 (7200), 551 (3600). Anal. ($C_{29}H_{26}F_3NO_{11}\cdot H_2O$) C, H, F, N.

***N*-(Pentafluoropropionyl)-9,10-anhydroadriamycin (16).** A solution of 0.25 mL (1.25 mmol) of pentafluoropropionic anhydride in 3 mL of ether was added dropwise at 0 °C to a stirred solution of 58 mg (0.10 mmol) of adriamycin in 15 mL of dry pyridine. The same workup and isolation procedure as before gave 38 mg (56%) of red solid: mp 155–158 °C dec; IR (KBr) 3400 (broad, OH, NH), 1713, 1700 (sh, ketone, amide), 1670

(C=C), 1618 (quinone) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 222 nm (ϵ 21 000), 234 (22 200), 269 (27 800), 503 (15 400), 517 (15 700), 554 (7240). Anal. ($C_{30}H_{26}F_5NO_{11}$) C, H, F, N.

***N*-(Heptafluorobutyryl)-9,10-anhydroadriamycin (17).** A solution of 0.15 mL (0.58 mmol) of heptafluorobutyric anhydride in 3 mL of ether was added dropwise at 0 °C to a stirred solution of 29 mg (0.05 mmol) of adriamycin in 10 mL of dry pyridine. After stirring for 2 h at 10–15 °C, the reaction mixture was worked up in the same manner as before to provide 18 mg (50% yield) of red solid: mp 148–150 °C dec; $[\alpha]_D$ +498° (*c* 0.009, CH_3OH); IR (KBr) 3410 (broad, OH, NH), 1710 (ketone, amide), 1695 (C=C), 1668 (quinone) cm^{-1}; UV–Vis λ_{max} (CH_3OH) 220 nm (ϵ 21 700), 233 (24 500), 270 (30 200), 504 (17 000), 516 (16 000), 554 (8000). Anal. ($C_{31}H_{26}F_7NO_{11}\cdot H_2O$) C, H, F, N.

***N*-(Pentafluoropropionyl)-9,10-anhydroadriamycin 14-Valerate (19).** In a like manner, reaction of 300 mg (0.48 mmol) of adriamycin 14-valerate and 1 mL (5 mmol) of pentafluoropropionic anhydride in ethyl acetate–ether afforded 202 mg (54%) of product: mp 215–217 °C dec; $[\alpha]_D$ +270° (*c* 0.033, CH_3OH); IR (KCl) 3385 (broad, OH, NH), 1720, 1690, 1620 (carbonyls); UV–Vis λ_{max} (CH_3OH) 223 nm (ϵ 40 400), 241 (29 000), 278 (10 300), 478 (13 100), 495 (12 800), 530 (6150). Anal. ($C_{35}H_{36}F_5NO_{13}$) C, H, N; F: calcd, 12.27; found, 11.82.

***N*-(Heptafluorobutyryl)-9,10-anhydroadriamycin 14-Valerate (20).** As before, 300 mg (0.48 mmol) of adriamycin 14-valerate and 0.9 mL (3.5 mmol) of heptafluorobutyric anhydride in ethyl acetate–ether gave 230 mg (56%) of product: mp 218–220 °C dec; $[\alpha]_D$ +251° (*c* 0.038, CH_3OH); IR (KCl) 3460, 3420 (OH, NH), 1700, 1618 (carbonyls); UV–Vis λ_{max} (CH_3OH) 223 nm (ϵ 42 700), 240 (29 500), 277 (9410), 478 (14 000), 494 (13 800), 529 (7320). Anal. ($C_{36}H_{36}F_7NO_{13}\cdot 1.5H_2O$) C, H, F, N.

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Synthesis of *N*-(Carbonylamino)-1,2,3,6-tetrahydropyridines with Analgesic, Antiinflammatory, and Hyperglycemic Activity

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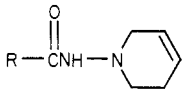
A group of *N*-(carbonylamino)-1,2,3,6-tetrahydropyridines was synthesized to investigate the effects that changes in functionality at the carbonyl group have on analgesic, antiinflammatory, and hyperglycemic activities. One of the most active analgesic compounds was *N*-[(ethoxycarbonyl)amino]-1,2,3,6-tetrahydropyridine (**5o**), which was comparable to that of morphine. Pretreatment with naloxone did not alter the activity of **5o** or **5q**. *N*-[(2-Furanylcarbonyl)amino]-1,2,3,6-tetrahydropyridine (**5q**) was the most potent hyperglycemic agent, elevating blood glucose 181% at 2 and 4 h after a 100 mg/kg po dose.

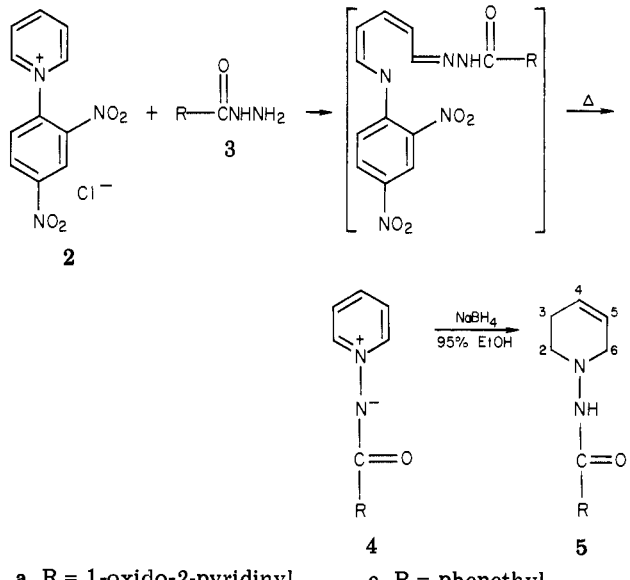
In an earlier study we described a facile method for the synthesis of *N*-(carbonylamino)-1,2,3,6-tetrahydropyridines **1**[1] via the sodium borohydride reduction of *N*-(carbonylimino)pyridinium ylides, which exhibited significant analgesic, antiinflammatory, and hyperglycemic activities.[2] It was therefore of interest to extend this series to determine what effect incorporation of other ring systems and functionality would have on pharmacological activity. We now describe the synthesis and analgesic, antiinflammatory, and hyperglycemic activities of structurally related *N*-(carbonylamino)-1,2,3,6-tetrahydropyridines **5**.



1a, R = phenyl
b, R = 2-pyridyl
1c, R = 3-pyridyl
d, R = 4-pyridyl

Chemistry. *N*-(Carbonylimino)pyridinium ylides **4** were prepared via two synthetic procedures. Reaction of 2,4-dinitrophenylpyridinium chloride (**2**) with carboxylic acid hydrazides **3a–h** yielded the respective 5-(2,4-dinitroanilino)penta-2,4-dienal carboxylic acid hydrazones, which on heating cyclized to *N*-(carbonylimino)pyridinium ylides **4a–h** (Scheme I). Alternatively, reaction of *N*-aminopyridinium iodide (**6**), obtained by amination of pyridine using hydroxylamine-*O*-sulfonic acid, with acid chlorides **7i–t** afforded ylides **4i–t** as illustrated in Scheme II and summarized in Table I. Reduction of ylides **4a–t** using sodium borohydride in ethanol at ice-bath temperature gave the title *N*-(carbonylamino)-1,2,3,6-tetra-

Scheme I



a, R = 1-oxido-2-pyridinyl
b, R = 3-pyridinylmethyl
c, R = 3-indolylmethyl
d, R = 3,4-methylenedioxyphenylmethyl
e, R = phenethyl
f, R = phenoxymethyl
g, R = cyclohexyl
h, R = *n*-hexyl

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