Potential Long-Acting Anticonvulsants. 1. Synthesis and Activity of Succinimides Containing an Alkylating Group at the 2 Position

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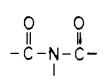
The synthesis of succinimide derivatives in which alkylating groups have been attached to the 2 position of the ring or to the para position of the 2-phenyl substituent is described. The alkylating groups used were (a) α-haloacetyl, (b) α-haloacetamido, (c) maleimido, and (d) maleamyl. These compounds were prepared as potential long-acting anticonvulsants. Several of these derivatives exhibited activity against metrazole-induced seizures comparable to phensuximide. The maleimide 16 and the bromoacetamido derivative 23 exhibited a duration of action of at least 3.5 h.

The anticonvulsant activity of compounds containing the imide structure (1) is well recognized.[2a] Among the succinimides, phensuximide (**2a**), methsuximide (**2b**), and ethosuximide (**2c**) are used in the treatment of petit mal epilepsy.[2b]



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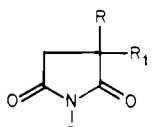


2a, R = C_6H_5; R_1 = H; R_2 = CH_3
b, R = C_6H_5; R_1 = CH_3; R_2 = CH_3
c, R = C_2H_5; R_1 = CH_3; R_2 = H

The fact that epilepsy is found in 0.5–1.0% of the general population constitutes a major public health problem. Epileptics frequently have a feeling of inferiority and self-consciousness with total withdrawal from society.[3] A further complication for the epileptic is that many of the anticonvulsant agents have some degree of undesirable side effects. Toman[4] has stated that due to individual variation in response to antiepileptic agents, there is need for new and better drugs.

Our present investigation was undertaken in an attempt to prepare potential long-acting anticonvulsants. Two types of compounds were synthesized in which alkylating groups were attached to either the 2 position of the ring or to the para position of the 2-phenyl substituent.

The use of alkylating agents in chemotherapy or as enzyme inhibitors is well documented. However, very few attempts at exploiting alkylating agents as pharmacodynamic agents have been reported. Ehrenpreis, Stubbins, and co-workers reported that a chloroethyl derivative of lidocaine produced a long-acting local anesthetic action.[5,6] Portoghese and co-workers have reported the synthesis and biological activity of a series of *N*-acylanileridines having alkylating capacity in an attempt to demonstrate analgetic receptor blockade.[7,8] Miyadera, Kosower, and Kosower[9,10] have investigated probing receptor sites by the use of alkylating agents. These workers attached alkylating groups to amphetamine and barbital, but none of the derivatives had an appreciably long duration of action.

If a succinimide derivative can first reversibly bind to its active site followed by alkylation of a nucleophile at or near the active site, then a long duration of action should result. Baker described this phenomena as "active site-directed irreversible inhibition".[11,12] In an effort to fully investigate this approach (a) the alkylating group was varied in order to offer a greater selection to a potential nucleophile at or near the receptor site; (b) the distance between the alkylating group and that part of the molecule thought to be responsible for biological activity, the imide structure (1), has been varied; and (c) the alkylating group has been attached at two different positions on the succinimide nucleus in order to search for an appropriate nucleophile.

Chemistry. Succinimide derivatives in which the alkylating group is attached to the 2 position of the ring are shown in Table I. Phensuximide (**2a**) was first converted to its sodium enolate with sodium ethoxide followed by reaction with the appropriate acid halide to give compounds **3–5**. Catalytic reduction of **5** with palladium on carbon yielded the *p*-aminobenzoyl derivative **6** which was allowed to react with bromoacetyl bromide in the presence of triethylamine to afford **7**. Other potential alkylating derivatives were synthesized by reaction of **6** with maleic anhydride to yield the maleamic acid **8**. Subsequent cyclization of **8** with acetic anhydride and sodium acetate utilizing the procedure of Cava and co-workers[13] gave the maleimide **9**.

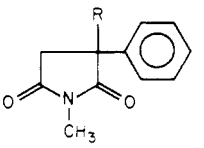
Alkyl bridges of two and three carbon atoms were inserted into the 2 position of **2a** in an effort to vary the distance between the alkylating group and the succinimide ring. Alkylation of **2a** with chloroacetonitrile using sodium hydride in tetrahydrofuran gave the cyanomethyl derivative **10**. Cyanoethylation of **2a** with acrylonitrile in the presence of Triton B readily afforded **12**. Catalytic reduction of the nitriles **10** and **12** with platinum oxide in ethanolic hydrochloric acid gave the amine hydrochlorides **11** and **13** which were allowed to react with chloroacetic anhydride in dimethylformamide using triethylamine as the base to yield the α-chloroamides **14** and **17**. Conversion of **11** and **13** into the corresponding maleimidoethyl **16** and maleimidopropyl **19** succinimides was carried out in the usual manner via intermediate maleamic acids **15** and **18**.

Nitration of **2a** with fuming nitric acid produced the nitro derivative **20**. Evidence that para substitution had occurred was provided by the NMR spectrum. The presence of two doublets at δ 8.27 and 7.49 with an ortho coupling constant of J = 9 Hz indicated that the para isomer had been formed. Catalytic reduction of **20** with palladium on carbon gave the arylamine **21** which was smoothly converted to the α-chloroamide **22** and the α-bromoamide **23**. Reaction of 2-(*p*-aminophenyl)-*N*-methylsuccinimide (**21**) with iodoacetic acid using dicyclohexylcarbodiimide yielded the α-iodoamide **24**. In the normal manner, the maleimide **26** was obtained by cyclization of the maleamic acid **25**. The physical properties of the compounds in which the alkylating group is attached to the para position of the 2-phenyl substituent of *N*-methyl-2-phenylsuccinimide and their precursors are shown in Table II.

Results and Discussion

All of the alkylating succinimide derivatives were tested

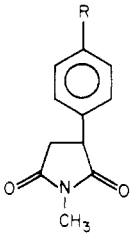
Table I



Compd	R	Mp, °C	Recrystn solvent	Yield, %	Method	Formula (analyses)[a]
3	-$COCH_2Cl$	122-125	EtOH	33	A	$C_{13}H_{12}ClNO_3$ (C, H, N, Cl)
4	-$COCH_2Br$	119-121	*s*-BuOH	38	A	$C_{13}H_{12}BrNO_3$ (C, H, N, Br)
5	-COC_6H_4-*p*-NO_2	151-153	EtOH-$CHCl_3$	37	A	$C_{18}H_{14}N_2O_5$ (C, H, N)
6	-COC_6H_4-*p*-NH_2	139-141	EtOH-H_2O	56	H	$C_{18}H_{16}N_2O_3$ (C, H, N)
7	-COC_6H_4-*p*-$NHCOCH_2Br$	224-225	EtOH	53	B	$C_{20}H_{17}BrN_2O_4$ (C, H, N, Br)
8	-COC_6H_4-*p*-NHCOCH=CHCOOH (cis)	202-204.5	EtOH-$CHCl_3$	76	D	$C_{22}H_{18}N_2O_6$ (C, H, N)
9	-COC_6H_4-*p*-NCOCH=CHCO	204-206.5	EtOH-$CHCl_3$	69	F	$C_{22}H_{16}N_2O_5$ (C, H, N)
10	-CH_2CN	73-75	EtOH	48		$C_{13}H_{12}N_2O_2$ (C, H, N)
11	-$(CH_2)_2NH_3^+Cl^-$	221-223	EtOH	57	G	$C_{13}H_{17}ClN_2O_2$ (C, H, N)
12	-CH_2CH_2CN	92-94	EtOH	80		$C_{14}H_{14}N_2O_2$ (C, H, N)
13	-$(CH_2)_3NH_3^+Cl^-$	183.5-185	EtOH-Et_2O	68	G	$C_{14}H_{19}ClN_2O_2$ (C, H, N)
14	-$(CH_2)_2NHCOCH_2Cl$	61-65[b]	*c*	31	C	$C_{15}H_{17}ClN_2O_3$ (C, H, N)
15	-$(CH_2)_2$NHCOCH=CHCOOH (cis)	161.5-162.5	EtOH	54	E	$C_{17}H_{18}N_2O_5$ (C, H, N)
16	-$(CH_2)_2$NCOCH=CHCO	98-99	*d*	38	F	$C_{17}H_{16}N_2O_4$ (C, H, N)
17	-$(CH_2)_3NHCOCH_2Cl$	101-101.5	C_6H_6	35	C	$C_{16}H_{19}ClN_2O_3$ (C, H, N)
18	-$(CH_2)_3$NHCOCH=CHCOOH (cis)	151-152.5	EtOH	57	E	$C_{18}H_{20}N_2O_5$ (C, H, N)
19	-$(CH_2)_3$NCOCH=CHCO	110.5-112	EtOH	30	F	$C_{18}H_{18}N_2O_4$ (C, H, N)

[a] Compounds were analyzed for the elements shown in parentheses and are within 0.4% of the calculated value. [b] Solidified upon standing for several weeks. [c] Purified by column chromatography using silica gel as the absorbent and a C_6H_6-$CHCl_3$ mixture as the eluent and analyzed directly without recrystallization. [d] Purified by column chromatography employing silica gel as the adsorbent and a $CHCl_3$-EtOH-C_6H_6 mixture as the solvent system and analyzed directly without recrystallization.

Table II



Compd	R	Mp, °C	Recrystn solvent	Yield, %	Method	Formula (analyses)[a]
20	-NO_2	121-125	EtOH	56		$C_{11}H_{10}N_2O_4$ (C, H, N)
21	-NH_2	163-165	EtOH	46	H	$C_{11}H_{12}N_2O_2$ (C, H, N)
22	-$NHCOCH_2Cl$	167.5-168.5	EtOH	61	B	$C_{13}H_{13}ClN_2O_3$ (C, H, N)
23	-$NHCOCH_2Br$	148-149.5	C_6H_6[b]	26	B	$C_{13}H_{13}BrN_2O_3$ (C, H, N)
24	-$NHCOCH_2I$	183-185	EtOH	49		$C_{13}H_{13}IN_2O_3$ (C, H, N)
25	-NHCOCH=CHCOOH (cis)	181-181.5	EtOH	81	D	$C_{15}H_{14}N_2O_5$ (C, H, N)
26	-NCOCH=CHCO	154.5-157.5	EtOH-$CHCl_3$	49	F	$C_{15}H_{12}N_2O_4$ (C, H, N)

[a] Compounds were analyzed for the elements shown in parentheses and were within 0.4% of the calculated value. [b] Recrystallized first from *s*-BuOH.

for anticonvulsant activity by the methods described in the Experimental Section using **2a–c** as reference compounds. The results are shown in Table III.

Of the sixteen compounds tested, seven exhibited an-

Table III. Anticonvulsant Effect

	MES			sc Met			
	Activity[a]			Activity[a]			
Compd	0.5 h	4 h	ED_{50}	0.5 h	4 h	ED_{50}	TD_{50}[b]
2a	++	+	112 (104-131)	+++	+++	50 (21-65)	232 (187-267)
2b	+	+	76 (63-89)	+++	–	35 (27-47)	188 (160-236)
2c	–	–	>1000	+	–	130 (111-150)	441 (383-485)
3	+	+	225 (206-245)[c]	+++	–	100-120[c]	*d*
4	–	–	*d*	+++	–	*d*	*d*
7	–	–	*d*	–	–	*d*	*d*
8	–	–	*d*	–	–	*d*	*d*
9	–	–	*d*	–	–	*d*	*d*
14	–	–	*d*	–	–	*d*	*d*
15	–	–	*d*	–	–	*d*	*d*
16	–	+++	*d*	+++	+++	*d*	*d, e*
17	–	–	*d*	–	–	*d*	*d*
18	–	–	*d*	–	–	*d*	*d*
19	–	–	*d*	–	++	*d*	*d, e*
22	–	–	*d*	–	–	*d*	*d*
23	–	–	*d*	+++	+++	35 (29-42)[f]	54 (47-62), *e*
24	–	++	*d*	+++	–	*d*	*d, e*
25	–	–	*d*	–	–	*d*	*d*
26	–	–	*d*	+++	–	*d*	*d, e*

[a] +++, ++, and + signify activity at 30, 100, or 300 mg/kg, respectively; – denotes no activity observed at 300 mg/kg. [b] TD_{50} = median toxic dose in the rotorod test. [c] Determined at time of peak effect (2 h). [d] Not determined. [e] Animals displayed CNS depression. [f] Determined at time of peak effect (0.5 h).

ticonvulsant activity. Compounds **3**, **16**, and **24** showed activity against maximal electroshock (MES), whereas **3**, **4**, **16**, **19**, **23**, **24**, and **26** provided some degree of protection against metrazole-induced seizures. Duration of effect is evidenced in **3** against MES and in **16** and **23** against metrazole-induced seizures.

In the active compounds, the alkylating function is either α-haloacyl or maleimido; the maleamic acid derivatives **8**, **15**, **18**, and **25** were uniformly inactive. The most bulky compounds **7**–**9** were also completely devoid of activity which may indicate a steric effect in the complexing of the drug to the active site.

The preliminary testing of the potential alkylating succinimide derivatives indicates that several of the compounds have a duration of action of at least 3.5 h. However, further testing is necessary in order to determine the actual duration of action.

Experimental Section

Spectra and Analyses. Melting points were determined on a Thomas-Hoover melting point apparatus and are uncorrected. The IR spectra were taken on a Beckman IR-8 and a Perkin-Elmer 700 spectrometer as either liquid films or as KBr pellets. The NMR spectra were recorded on a Varian A-60A spectrometer using tetramethylsilane as the internal standard, and the chemical shift data are on file with the editors. Elemental analyses were performed by Micro-Analysis, Wilmington, Del.; PCR Inc., Gainesville, Fla.; and Dr. Kurt Eder, Geneva, Switzerland.

***N*-Methyl-2-phenylsuccinimide (2a).** Compound **2a** was prepared according to the method of Miller and Long[14] in 70% yield: mp 71–73 °C (lit. mp 71–73 °C).

Method A. 2-Chloroacetyl-*N*-methyl-2-phenylsuccinimide (3). This general method is an adaptation of the method of Magarian and co-workers.[15] Freshly cut Na metal in the amount of 1.30 g (0.055 g-atom) was charged into a round-bottomed flask and 35 ml of dry ethanol[16] was added at such a rate so as to maintain vigorous but controlled refluxing. After all the Na had reacted a Dean-Stark trap was fitted and 9.5 g (0.05 mol) of *N*-methyl-2-phenylsuccinimide (**2a**) in 200 ml of anhydrous $C_6H_5CH_3$ was added. Heating was discontinued after 55 ml of distillate had been collected and the vapor temperature had reached 110 °C. The reaction mixture was cooled to 0–5 °C and the thick purple slurry was treated dropwise with 6.20 g (0.055 mol) of $ClCH_2COCl$ in 40 ml of anhydrous $C_6H_5CH_3$. After the addition had been completed, the reaction mixture was allowed to come to room temperature and stir for 1 h. The yellow solution was treated with 100 ml of saturated $NaHCO_3$ solution and two layers formed. The $C_6H_5CH_3$ layer was separated, washed with H_2O (3 × 50 ml), dried ($MgSO_4$), filtered, and evaporated under reduced pressure. The yellow oil which remained solidified upon cooling. Two recrystallizations from 95% EtOH yielded 4.2 g of a white powder.

2-Bromoacetyl-*N*-methyl-2-phenylsuccinimide (4). Compound **4** was obtained in a similar manner as in the preparation of **3** using the quantities: 1.73 g (0.075 g-atom) of Na metal, 13.3 g (0.070 mol) of **2a**, and 15.5 g (0.077 mol) of $BrCH_2COBr$. After the addition of the $BrCH_2COBr$, the mixture was allowed to react for 17 h.

2-(*p*-Nitrobenzoyl)-*N*-methyl-2-phenylsuccinimide (5). The sodium enolate of **2a** was generated as in the synthesis of **3** from 5.10 g (0.22 g-atom) of Na metal and 37.8 g (0.20 mol) of the imide. The purple slurry was cooled to 10 °C and 37.1 g (0.20 mol) of *p*-nitrobenzoyl chloride in 100 ml of $C_6H_5CH_3$ was added dropwise. The temperature of the reaction mixture increased to 25 °C during the addition. After stirring for 2 h at room temperature, the reaction mixture was filtered to remove the precipitated solid. The solid was washed repeatedly with H_2O and the washings were extracted with $C_6H_5CH_3$ (3 × 50 ml). The toluene extracts were combined with the original toluene filtrate, dried ($MgSO_4$), and evaporated under reduced pressure. A yellow oil remained which solidified into a yellow mass upon cooling. The yellow solid was combined with the solid which precipitated from the original reaction mixture and recrystallized from an EtOH–$CHCl_3$ mixture to yield 25.1 g of yellow needles.

Method B. 2-[*p*-(Chloroacetamido)phenyl]-*N*-methylsuccinimide (22). A mixture of 0.70 g (0.0034 mol) of **21**, 0.42 g (0.0041 mol) of Et_3N, and 50 ml of dry THF was treated dropwise with a solution of 0.46 g (0.0041 mol) of $ClCH_2COCl$ in 5 ml of dry THF at room temperature. Immediately, a white precipitate formed and the reaction mixture was stirred at room temperature for 17 h. The reaction mixture was filtered to remove the Et_3N·HCl, and the filtrate was concentrated under reduced pressure leaving a white solid residue. The residue was washed with 100 ml of H_2O and suction filtered. The H_2O-insoluble material was recrystallized from 95% EtOH to yield 0.585 g of **22**.

2-[*p*-(Bromoacetamido)phenyl]-*N*-methylsuccinimide (23). This compound was prepared from **21**, 2.30 g (0.011 mol), 1.21 g (0.012 mol) of Et_3N, and 2.22 g (0.011 mol) of $BrCH_2COBr$ in the same manner as described for the synthesis of **22**.

2-[*p*-Bromoacetamido)benzoyl]-*N*-methyl-2-phenylsuccinimide (7). A solution of 1.0 g (0.0032 mol) of **6** in 50 ml of anhydrous $C_6H_5CH_3$ was heated to 70 °C and treated with 0.32 g (0.0032 mol) of Et_3N. To the stirred solution was added 0.65

g (0.0032 mol) of $BrCH_2COBr$ in 5 ml of anhydrous $C_6H_5CH_3$. Immediately, a white solid precipitated from solution. After heating for 5 h at 70 °C, the reaction mixture was cooled to room temperature and suction filtered to remove the precipitated solid. The precipitate was washed thoroughly with several portions of H_2O and recrystallized to yield 0.73 g of yellow powder.

Method C. 2-(2-Chloroacetamidoethyl)-*N*-methyl-2-phenylsuccinimide (14). To a mixture of 3.0 g (0.0112 mol) of **11**, 1.92 g (0.0112 mol) of $(ClCH_2CO)_2O$, and 25 ml of dry DMF was added 1.14 g (0.0112 mol) of Et_3N. The temperature of the reaction mixture increased to 40 °C and a considerable amount of white solid precipitated from solution. After the exothermic reaction had subsided, the reaction mixture was heated at 75 °C for 4 h. The yellow solution was cooled to room temperature and poured into 150 ml of an ice–H_2O mixture. The aqueous solution was extracted several times with a total volume of 250 ml of $CHCl_3$. The combined $CHCl_3$ extracts were washed successively with 25-ml portions of saturated $NaHCO_3$ solution and H_2O. The $CHCl_3$ layer was dried ($MgSO_4$), and the solvent evaporated under reduced pressure. The yellow oil which remained was dissolved in a minimum amount of C_6H_6 and chromatographed on 50 g of silica gel using a C_6H_6–$CHCl_3$ solvent system. The chromatography yielded 1.07 g of a colorless oil.

2-(3-Chloroacetamidopropyl)-*N*-methyl-2-phenylsuccinimide (17). See synthesis of **14** and Table I.

Method D. 2-[*p*-Maleamyl(benzoyl)]-*N*-methyl-2-phenylsuccinimide (8). A solution of **6**, 1.0 g (0.003 mol), and 0.30 g (0.003 mol) of maleic anhydride in 80 ml of anhydrous Et_2O and 15 ml of dry THF was stirred at room temperature for 2 h followed by 21 h of refluxing. The reaction mixture was cooled and filtered to remove the precipitated solid. Evaporation of the filtrate produced a light green solid. The solids were combined and recrystallized to yield 1.0 g of a light green powder.

2-(*p*-Maleamylphenyl)-*N*-methylsuccinimide (25). A suspension of 4.1 g (0.020 mol) of **21** and 1.97 g (0.020 mol) of maleic anhydride in 100 ml of THF was refluxed for 2 h. The reaction mixture was cooled and filtered to yield 4.5 g of a white powder, mp 178–179.5 °C. The filtrate was concentrated to yield an additional 0.4 g of a white powder, mp 177.5–179 °C. The combined crude yield was 4.9 g. An analytical sample was obtained by recrystallization of a small sample of crude **25** from 95% EtOH to yield analytically pure **25**, mp 180–181.5 °C.

Method E. 2-(2-Maleamylethyl)-*N*-methyl-2-phenylsuccinimide (15). To a stirred solution of 2.40 g (0.0089 mol) of **11** and 0.878 g (0.0089 mol) of maleic anhydride in 20 ml of dry DMF was added in one portion 0.90 g (0.0089 mol) of Et_3N. Immediately, a white solid precipitated from solution. The reaction mixture was heated at 70 °C for 4.5 h, cooled, and concentrated until only a few milliliters of solvent remained. The yellow solution was poured in 150 ml of an ice–H_2O mixture and a sticky white oil separated. The aqueous layer was decanted off and the oil was triturated with benzene to produce a fluffy white solid. Recrystallization of the solid yielded 1.6 g of **15**.

2-(3-Maleamylpropyl)-*N*-methyl-2-phenylsuccinimide (18). See synthesis of **15** and Table I.

Method F. 2-(*p*-Maleimidobenzoyl)-*N*-methyl-2-phenylsuccinimide (9). A mixture of 1.7 g (0.0042 mol) of **8**, 0.3 g of anhydrous NaOAc, and 6.7 g (0.065 mol) of $(CH_3CO)_2O$ was heated on a steam bath for 0.5 h and poured into 50 ml of an ice-H_2O mixture. The precipitated product was filtered and washed several times with portions of cold H_2O. Recrystallization from an EtOH–$CHCl_3$ mixture produced 0.9 g of a pink powder, mp 204–206.5 °C. The filtrate was concentrated to yield a second crop of 0.2 g, mp 196.5–200.5 °C.

2-(2-Maleimidoethyl)-*N*-methyl-2-phenylsuccinimide (16). See synthesis of **9** and Table I.

2-(3-Maleimidopropyl)-*N*-methyl-2-phenylsuccinimide (19). See synthesis of **9** and Table I.

2-(*p*-Maleimidophenyl)-*N*-methyl-2-phenylsuccinimide Hydrochloride (13). See synthesis of **9** and Table II.

Method G. 2-(3-Aminopropyl)-*N*-methyl-2-phenylsuccinimide Hydrochloride (13). The general method for the catalytic hydrogenation of nitriles is an adaptation of the methods of Hendrickson[17] and Harley-Mason.[18] A mixture of 19.3 g (0.080 mol) of **12**, 32 ml of concentrated HCl, 0.23 g of PtO_2, and 250 ml of 95% EtOH was shaken for 19.5 h on a Parr hydrogenator. The reaction mixture was diluted with 80 ml of H_2O, filtered, and evaporated until the total volume was about 80 ml. The aqueous solution was extracted with $CHCl_3$ (3 × 50 ml) and then concentrated under reduced pressure. The yellow oil which remained solidified upon standing in the refrigerator overnight. Recrystallization of the crude hydrochloride yielded 15.4 g of pure **13**.

2-(2-Aminoethyl)-*N*-methyl-2-phenylsuccinimide Hydrochloride (11). See preparation of **13** and Table I.

Method H. 2-(*p*-Aminobenzoyl)-*N*-methyl-2-phenylsuccinimide (6). A suspension of 4.0 g (0.012 mol) of **5** and 0.5 g of 5% Pd/C in 150 ml of 95% EtOH was shaken on the Parr hydrogenator for 10 min. The reaction mixture was filtered and the filtrate evaporated under reduced pressure. The orange oil which remained was dissolved in 95% EtOH and diluted with H_2O just to the point of cloudiness. Filtering and drying of the crystallized solid yielded 2.0 g of a light yellow powder.

2-(*p*-Aminophenyl)-*N*-methylsuccinimide (21). See synthesis of **6** and Table II.

2-Cyanomethyl-*N*-methyl-2-phenylsuccinimide (10). A 57% mineral oil dispersion of NaH in the amount of 4.63 g (0.11 mol) was suspended in 25 ml of dry THF. The stirred suspension was heated to reflux and 18.9 g (0.10 mol) of **2a** in 80 ml of THF was added dropwise. Slow hydrogen evolution was observed after a few milliliters of the imide solution had been added. The reaction mixture was cooled and the thick pink suspension was treated dropwise with 8.30 g (0.11 mol) of $ClCH_2CN$ in 25 ml of THF. External cooling was applied in order to keep the temperature below 45 °C. After the exothermic reaction had subsided, the reaction mixture was heated at reflux for 17 h. The mixture was cooled and decomposed with 50 ml of saturated NH_4Cl solution. The organic layer was decanted off and the inorganic sludge extracted with THF (3 × 25 ml). The combined THF extracts were evaporated under reduced pressure and the residue was taken up into 150 ml of $CHCl_3$. The $CHCl_3$ was washed with H_2O (2 × 50 ml), dried ($MgSO_4$), filtered, and evaporated. The brown oil which remained solidified into a yellow mass upon stirring. Recrystallization from 95% EtOH gave 10.9 g of a white powder.

2-(2-Cyanoethyl)-*N*-methyl-2-phenylsuccinimide (12). This synthesis is a modification of the method of Bruson.[19] To a solution of 12.0 g (0.063 mol) of **2a** in 30 ml of dioxane was added 0.7 ml of a 40% methanolic solution of Triton B. Immediately, the reaction mixture became dark red in color and 3.38 g (0.063 mol) of $CH_2{=}CHCN$ in 10 ml of dioxane was added at such a rate that the temperature did not exceed 30 °C. After stirring for 17 h at room temperature, the reaction mixture was neutralized with 10% HCl solution and poured into 200 ml of an ice–H_2O mixture. An oil separated which solidified into a white mass upon stirring. Recrystallization of the solid gave 12.3 g of a white powder.

2-[*p*-Iodoacetamido)phenyl]-*N*-methylsuccinimide (24). To 1.6 g (0.0078 mol) of **21** in 80 ml of dry CH_3CN under a nitrogen atmosphere was added 1.45 g (0.0078 mol) of ICH_2COOH followed by 1.60 g (0.0078 mol) of DCC. Immediately, a white solid precipitated from solution. After stirring for 6 h at room temperature, the mixture was filtered to remove the precipitated dicyclohexylurea. The filtrate was evaporated under reduced pressure to give a yellow solid. Recrystallization of the solid from 95% EtOH yielded 1.41 g of **24**.

2-(*p*-Nitrophenyl)-*N*-methylsuccinimide (20). A total of 70 ml of fuming HNO_3 was cooled to 0 °C and 20.6 g (0.109 mol) of **2a** was added at such a rate that the temperature of the reaction mixture remained below 5 °C. After the addition had been completed, the reaction mixture was stirred for an additional 30 min and then poured into 300 ml of an ice–H_2O mixture. The light green solid which precipitated was taken up into 300 ml of $CHCl_3$. The $CHCl_3$ was washed with H_2O (2 × 100 ml) and dried ($MgSO_4$), and the solvent was evaporated under reduced pressure. Recrystallization of the residue from 95% EtOH gave 14.3 g of a light yellow powder.

Pharmacological Testing. All compounds were tested for anticonvulsant activity by the Antiepileptic Drug Development Program administered by the Section on Epilepsy, National Institute of Health, Bethesda, Md. 20014. The compounds were evaluated using the Anticonvulsant Screening Project Test Systems.[20] Three tests were performed: MES (maximal elec-

troshock seizure test), sc Met (subcutaneous pentylenetetrazole seizure threshold test), and the rotorod test to evaluate neurotoxicity.

All tests were performed on male Carworth Farms No. 1 mice. All compounds were tested at three dosage levels (30, 100, and 300 mg/kg) at 30 min and 4 h after their intraperitoneal administration. Four animals are injected with each dose. Thirty minutes later, each animal is examined for toxicity in the rotorod test. Immediately thereafter, anticonvulsant activity is evaluated by subjecting one mouse to the MES test and another to the subcutaneous metrazole test. The same tests are repeated 4 h later on the two remaining mice. All compounds are solubilized in either 0.9% sodium chloride or 30% polyethylene glycol 400 and administered intraperitoneally in a volume of 0.01 ml/g. The ED_{50} and TD_{50} values and their confidence limits were determined by the method of Litchfield and Wilcoxin.[21] MES activity is defined as abolition of the hind limb tonic extensor component of the maximal electroshock seizure elicited in mice with a 60-Hz alternating current of 50 mA delivered for 0.2 s via corneal electrodes. sc Met activity is defined as failure to observe even a threshold seizure (a single episode of clonic spasms of at least 5 s in duration).

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Alkaloids of *Vinca rosea* L. (*Catharanthus roseus* G. Don). 38. 4′-Dehydrated Derivatives[1]

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A series of 4′-dehydrated derivatives of various dimeric Vinca alkaloids has been synthesized to further define the structure–activity relationships of Vinca alkaloids with oncolytic potency. The concentrated sulfuric acid dehydration in most cases gave mixtures of the 3′,4′- and two isomeric 4′,20′-alkenes, which were isolated and characterized primarily by proton and ^{13}C NMR. Compounds tested for antitumor activity include the three dehydro isomers of 4-deacetylvinblastine, 4-deacetylvincristine, and 4-deacetylvinblastine-23-amide and some 4′-dehydrated derivatives epimeric at C-18′. Generally, the decrease in toxicity imparted by the new double bond was accompanied by a decrease in potency. An exception was 3′,4′-dehydro-4-deacetylvincristine, which showed a decrease in toxicity and increase in potency against at least one tumor in which vincristine itself has little effect.

Vinblastine (VLB, **1**)[2] and vincristine (VCR, **2**)[3] are dimeric indole alkaloids from *Catharanthus rosea* (*Vinca rosea*) used clinically in the treatment of various types of cancer. Despite their widely differing spectra of clinical activity and toxicities, VLB and VCR differ structurally only in the functional group on the dihydroindole nitrogen. Previous structure modification studies have shown that changes in the dihydroindole (vindoline) moiety have unpredictable results, causing either an increase or decrease in the oncolytic potency.[4,5] The effect of functional variation in the indole moiety (velbanamine) is evidenced in the activity of two other dimeric alkaloids isolated from *Catharanthus rosea*: leurosidine (**3**),[6] the 4′ epimer of VLB, and leurosine (**4**),[6] a 3′,4′-epoxide derivative. Although these compounds are somewhat lower in oncolytic potency as compared to VLB, their concomitant decrease in toxicity encouraged us to probe the effects of other functional changes in the velbanamine moiety. We have therefore prepared both the *exo*- and *endo*-4′-dehydro derivatives of several Vinca alkaloids. These serve to further define the structure–activity relationships at that position and provide intermediates to additional derivatives. Subsequent to the completion of our work, articles describing the synthesis of dehydrovinblastine have appeared[7,8] without reference to its biological activity. This paper reports the synthesis, isolation, and activities of a