round-bottom flask equipped with a Dean-Stark trap, magnetic stirring bar, and heating mantle. After the benzene solution was refluxed for 0.5 hr. to remove all water from the system, 11.8 g. (0.0928 mole) of 3-chloroaniline was added dropwise to the reaction flask. White material precipitated. The reaction mixture was stirred at reflux temperature, overnight, then the reaction was interrupted and the hot solution was filtered immediately to remove the 3-chloroaniline hydrochloride. About 100 ml. of benzene was removed from the filtrate under reduced pressure causing a white precipitate to form. The solution was cooled in a refrigerator and 7.1 g. of crystals was collected by suction filtration. The solvent from the filtrate was evaporated to dryness under reduced pressure leaving a solid which was combined with the crystals previously collected and recrystallized from 2-propanol to give a total of 8 g. (57.5%) of the white crystalline 2-(3'-chloroanilinium)-5-methyl-5-nitro-1,3,2-dioxaphosphorinane 2-oxide, m.p. 171.5–173.5°.

Anal.—Calcd. for $C_{10}H_{12}ClN_2O_5P$: N, 9.14. Found: N, 9.30.

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(1) Billman, J. H., May, R. F., and Heard, J. E., *J. Pharm. Sci.*, 57, 1812 (1968).

Keyphrases

2-Alkylamino-5-alkyl-5-nitro-1,3,2-dioxaphosphorinane 2-oxides—synthesis
2-Arylamino-5-alkyl-5-nitro-1,3,2-dioxaphosphorinane 2-oxides—synthesis
Antitumor activity—1, 3, 2-dioxaphosphorinane 2-oxides
IR spectrophotometry—structure

1,3,2-Dioxaphosphorinane 2-Oxides III. Preparation and Antitumor Evaluation of Some 3,9-Disubstituted-2,4,8,10-tetraoxa-3,9-diphosphaspiro[5.5]-undecane 3,9-Dioxide

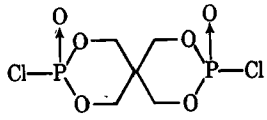
By JOHN H. BILLMAN and RALPH F. MAY

A series of 3,9-aminodisubstituted-2,4,8,10-tetraoxa-3,9-diphosphaspiro [5.5] undecane 3,9-dioxides has been prepared and screened for antitumor activity. Only one of these compounds showed activity toward Walker carcinosarcoma 256.

In view of the fact that some 2-alkylamino and 2-arylamino-5-alkyl-5-nitro-1,3,2-dioxaphosphorinane 2-oxides have been shown to possess some antitumor activity against Walker carcinosarcoma 256 (intramuscular) (1), one might expect that molecules containing two dioxaphosphorinane 2-oxide groupings would possess equal or greater activity than the single ring system.

Since Sweeting (2) had reported the synthesis of 3,9-dichloro-2,4,8,10-tetraoxa-3,9-diphosphaspiro [5.5] undecane 3,9-dioxide, I, it was decided that this dichloride would be an excellent starting material for making compounds that would fulfill the requirement of a double ring system. It was also felt that it would be of interest to see if the spirane moiety would act as a good carrier group and if in some way or other would have an effect on antitumor activities.

The desired amides were prepared by condensing the appropriate amines with the dichloride I in acetonitrile in the presence of triethylamine as outlined in Scheme I and as listed in Table I.



I

Biological Results—Only Compound 6 in Table I showed appreciable activity, and this was toward Walker carcinosarcoma 256. It showed an inhibition of 35% at a dose level of 25 mg./kg., 58% at 100 mg./kg., and 79% at 200 mg./kg. The activity of this compound is probably due to the alkylating groups in the molecule. Most of the other amides had an exceedingly high melting point and were very insoluble in most of the common organic solvents. The lack of antitumor activity for these diamides may be accounted for by their great insolubility.

Received April 8, 1968, from the Department of Chemistry, Indiana University, Bloomington, IN 47401

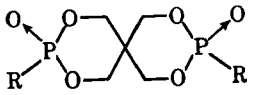
Accepted for publication June 14, 1968.

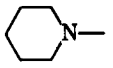
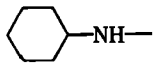
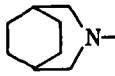
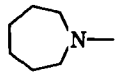
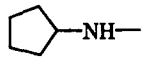
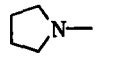
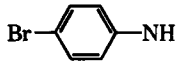
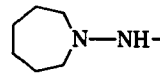
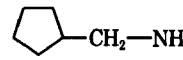
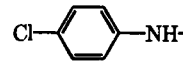
This investigation was supported by a Public Health Service grant (CA-06448-04) from the National Institutes of Health, U. S. Public Health Service, Bethesda, Md.

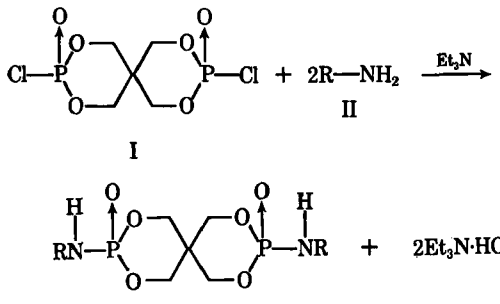
This paper represents part of a M.A. thesis submitted to the Graduate School, Indiana University, 1966.

The authors wish to thank Dr. O. J. Sweeting for supplying a large sample of 3,9-dichloro-2,4,8,10-tetraoxa-3,9-diphosphaspiro[5.5]undecane 3,9-dioxide (I) which was used in the early part of this work.

TABLE I—3,9-DISUBSTITUTED-2,4,8,10-TETRAOXA-3,9-DIPHOSPHASPIRO[5.5]UNDECANE 3,9-DIOXIDES



Compd. No.	R	Formula	M.p., °C.	% N Calcd.	% N Found	Infrared Assignment, μ		
1		$C_{15}H_{28}N_2O_6P_2$	315–315	7.11	7.16	8.05	9.75	
2		$C_{17}H_{30}N_2O_6P_2$	282–284	6.65	6.45	8.15	9.75	3.10
3		$C_{21}H_{36}N_2O_6P_2$	7360	5.91	5.76	8.00	9.75	
4		$C_{17}H_{32}N_2O_6P_2$	313–314	6.64	6.63	8.01	9.78	
5		$C_{15}H_{26}N_2O_6P_2$	272–273	7.15	7.02	8.15	9.80	3.10
6	$ClCH_2CH_2NH$—	$C_9H_{18}Cl_2N_2O_6P_2$	196–197	7.33	7.45	8.15	9.80	3.11
7		$C_{13}H_{24}N_2O_6P_2$	302–304	7.64	7.86	8.00	9.85	
8		$C_{17}H_{18}Br_2N_2O_6P_2$	286–287	4.95	4.89	7.95	9.75	3.11
9		$C_{17}H_{34}N_4O_6P_2$	159–160	12.41	12.06	8.11	9.70	3.11
10		$C_{15}H_{28}N_2O_6P_2$	204–205	6.56	6.34	8.15	9.70	3.08
11		$C_{17}H_{18}Cl_2N_2O_6P_2$	248–250	5.86	5.58	7.95	9.75	3.12



I II III

Scheme I

EXPERIMENTAL[1]

3,9-Disubstituted-2,4,8,10-tetraoxa-3,9-diphosphaspiro[5.5]undecane 3,9-Dioxide—The synthesis of these compounds is illustrated by the procedure for 3,9-dipyrolidino-2,4,8,10-tetraoxa-3,9-diphosphaspiro [5.5] undecane 3,9-dioxide. The spirane I, 5.00 g. (0.0168 mole), was dissolved in 125 ml. of hot acetonitrile, and the temperature was maintained at reflux. Dropwise addition of 4.77 g. (0.0672 mole) of the amine (II) resulted in an immediate precipitate. After 1 hr. of stirring, the solid was removed by filtration and washed with cold acetonitrile to afford 5.5 g. (89.5% yield) of a white solid. The material was recrystallized from a 20:1 mixture of acetonitrile and methanol to give 4.5 g. (73.4% yield) of pure product (m.p. 302–304°).

Anal.—Calcd. for $C_{13}H_{24}N_2O_6P_2$: N, 7.64. Found: N, 7.86.

[1] All melting points below 360° were taken on a Thomas-Hoover device and are corrected. The analyses were performed by Midwest Microlab, Inc., Indianapolis, Ind.

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(1) Billman, J. H., May, R. F., and Heard, J. E., *J. Pharm. Sci.*, **57**, 1809 (1968).

(2) Ratz, R., and Sweeting, O. J., *J. Org. Chem.*, **28**, 1608 (1963).

Keyphrases

3,9 - Disubstituted - 2,4,8,10 - tetraoxa - 3,9-diphosphaspiro[5.5]undecane-3,9-dioxides—synthesis

Antitumor activity—screening

IR spectrophotometry—structure

Centrally Acting Isosteric Mannich Bases

By RONALD E. ORTH*, J. W. BENNETT, ON-HOU MA†, and LARRY YOUNG

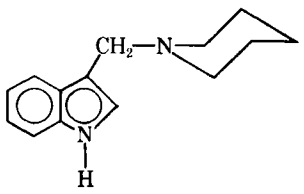
A number of *N*-heteroparaffinomethyl-1*H*-benzazoles is obtained by the Mannich and allied syntheses. Amine salts of all but the indoles are prepared. The degree of motricity and toxic levels are ascertained for the title compounds. Three major molecular modifications investigated cause variations in pharmacodynamic activity: substitution of the isosteric 1*H*-benzimidazole, 1*H*-indazole, and 1*H*-benzotriazole with 1*H*-indole; variation of the *N*-heteroparaffino-side chain moiety; and placement of the *N*-heteroparaffinomethyl substituents in different positions relative to the 1-nitrogen, depending on the particular benzazole. The 3-substituted indoles are most toxic and very active, while the 1-substituted benzimidazoles are best tolerated. Seven benzazoles (at least one from each chemical class) produce unusually high motility. Some general observations are reported. Methylpiperidino derivatives produce the greatest responses studied.

POWELL *et al.* (1) attribute hypertension and *in situ* uterine contraction to 3-(dimethylaminomethyl)indole (gramine). However, Bertaccini and Zamboni (2) report that the indolealkylamines (gramine included) with a single carbon atom lateral side chain have little smooth muscle activity. They find good 5-hydroxytryptamine-like activity exhibited by the indazolealkylamines. Studies by Walshe *et al.* (3) regarding factors influencing cerebral oxidation in relation to hepatic coma, using rat brain cortex slices, indicate gramine and 3-methylindole are active cerebral respiratory inhibitors at concentrations likely to be found in the body. Dubnik *et al.* (4), relating the effect of monoamine oxidase (MAO) inhibitors on brain serotonin of mice in addition to that resulting from inhibition of MAO, find gramine (a MAO-inhibitor) a cause of increased serotonin levels in the mouse brain.

Stimulated by inferences from the literature, this investigation is designed to (*a*) accommodate the synthesis of the title compounds and/or their water soluble salts (Table I) and (*b*) determine the degree of motor involvement and LD_{50}'s they produce in young white mice. Motricity data and subjective observations are noted in Table II.

Three major molecular modifications of gramine are responsible for producing the variations in bioactivity: interchange of the isosteric benzimidazole, 1*H*-indazole, and benzotriazole ring systems for indole; interchange of the *N*-heteroparaffino substituents on the side chain methyleno moiety; and placement of the *N*-heteroparaffinomethyl substituent in different positions relative to the 1-nitrogen, depending on the particular benzazole.

Gramine, synthesized by the Mannich reaction in 1935 (5), is the "open ended" tertiary amino model for the cyclized analogs prepared. Okuda (6), using a Mannich modification, has synthesized 3-(piperidinomethyl)indole (I). The product is found identical to Hellmann's (7) Mannich product, which is formed utilizing the acidic properties of benzenesulfonamide. Henry (8) has prepared the same compound from gramine *N*-oxide.



I

The Mannich reaction in methanol (9) is useful for preparing 1-(*N*-heteroparaffinomethyl)benzimidazole (II). 1-Hydroxymethylbenzimidazole (III) is also formed. The products of the normal Mannich condensation of benzimidazole with HCHO and *N*-heteroparaffins are stable and crystalline. Titration results using $(C_6H_5)_3C^-Na^+$ indicate a side chain attachment to the imidazole nitrogen (Scheme I).

The many attempts by Snyder *et al.* (10) at varying the Mannich reaction conditions to induce

Received April 28, 1967, from the Pharmaceutical Chemistry Department, College of Pharmacy, University of Kentucky, Lexington, KY 40506

Accepted for publication July 19, 1968.

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