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187. Takanobu Itai,*1 Shigeru Sako,*1 and Genzo Okusa*2 : Potential Anticancer Agents. XIII.*3 Reaction of 3,6-Dimethylpyridazine 1-Oxide and Methylpyridazine 1-Oxides with Benzaldehyde.

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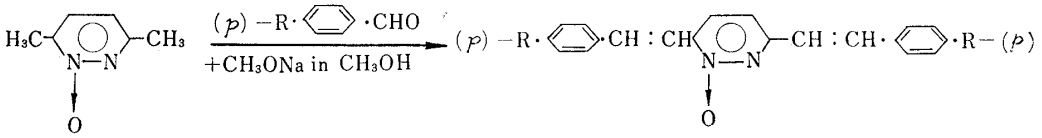
Sako previously examined the reactivity of chlorine atoms of 3,6-dichloropyridazine 1-oxide with sodium ethoxide and with ethylamine, and 3-chlorine atom was found more reactive than 6-chlorine atom.[1] He also reported that no difference in reactivity was found between 3- and 6-chlorine atoms[2] of monochloropyridazine 1-oxides but that 5-chlorine atom of 3,6-dimethylchloropyridazine 1-oxides was more active than the one in 4-position in the same reaction.[3]

It seemed of interesting to examine the reactivity of methyl groups at various positions in pyridazine 1-oxide, which seemed to be similarly influenced by the polar effect of the tertiary nitrogen and N-oxide group, in a reaction with benzaldehyde.

This reaction has been examined in picoline and its N-oxide with benzaldehyde,[4,5] and in methylpyridazine with anisaldehyde[6] etc., and in each case, corresponding styryl compounds were produced.

The present paper will describe the nucleophilic activity of the methyl groups in 3,6-dimethylpyridazine 1-oxide (I) and monomethylpyridazine 1-oxides (II) in their reaction with benzaldehyde.

When I was heated at 140° with benzaldehyde in the presence of piperidine, no condensation occurred, but with 2.5 molar equivalents of benzaldehyde in the presence of sodium methoxide in place of piperidine, 3,6-distyrylpyridazine 1-oxide (IIIa) was obtained in 80% yield heating on a water bath for one hour. With one molar equivalent of benzaldehyde under the same condition, I was converted to IIIa in 24% yield with a recovery of 44% of the starting material, and no monostyryl compound was formed.



I III

IIIa : R=H, IIIb : R=CH_3O
IIIc : R=$(CH_3)_2N$—

Chart 1.

The same reaction was examined with anisaldehyde or *p*-dimethylaminobenzaldehyde, which were thought less reactive and to prevent the second condensation due to the electron repelling effect of methoxyl or dimethylamino group in the *para*-position. However, mono-styryl compound was never produced. On heating on a water bath for one hour with one molar equivalent of anisaldehyde, 3,6-bis(*p*-methoxystyryl)

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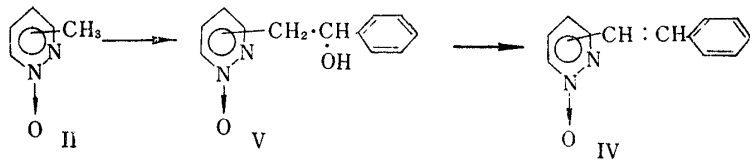
pyridazine 1-oxide (IIIb) was obtained in 18% yield with 40% recovery; and with one molar equivalent of *p*-dimethylaminobenzaldehyde on a water bath for three hours, 3,6-bis(*p*-dimethylaminostyryl)pyridazine 1-oxide (IIIc) was formed in 35% yield with 46% recovery of the starting material.

From these results, it was found that the reactivity of 3- and 6-methyl groups was almost the same, different from the case of 3,6-dichloropyridazine 1-oxide.

Next, 3-, 4-, 5-, and 6-methylpyridazine 1-oxides (IVa~d) were reacted with benzaldehyde at 100, 65, or 40° in the presence of sodium methoxide, and its results are summarized in Table I.



IIa : 3−, IIb : 4−
IIc : 5−, IId : 6−

Va : 3−, Vb : 4−
Vc : 5−, Vd : 6−

IVa : 3−, IVb : 4−
IVc : 5−, IVd : 6−

Chart 2.

Table I.

Starting compound	100°	65°		40°		
	IV (%)	IV (%)	II (%)	IV (%)	V (%)	II (%)
IIa	36	13.5	68	—	—	82
IIb	75	51	—	14.5	54	6
IIc	a)	58	—	32	28	26
IId	51	53	—	—	36	50

a) A deep brown viscous substance was produced.

From the result of these reactions at 100 and 65°, it was known, though rather qualitatively, that 4- and 6-methyl groups were similarly reactive, that 5-methyl was more active, and that 3-methyl less active than the 4- and 6-methyl groups.

Similarly, the reaction was carried out at 40° for one hour. In these cases, IIb and IIc gave corresponding styryl compounds in 14.5 and 32% yields, but IIa and IId did not. In addition, IIb, IIc, and IId produced corresponding (*β*-hydroxyphenethyl)-pyridazine 1-oxides (Vb~d), intermediates to styryl compounds, in 54, 28, and 36% yields, respectively. IIa was not changed and 82% of the starting material was recovered.

These (*β*-hydroxyphenethyl)pyridazine 1-oxides (Vb~d), were dehydrolyzed on heating in the presence of sodium methoxide to their corresponding styryl compounds. It

Table II.

Compound	Crystals	Solv. of recrystn.	m.p. (°C)
IVa	scales	benzene	134~136
IVb	〃	MeOH	186~188
IVc	needles	〃	158~159
IVd	〃	〃	159~161
Vb	prisms	EtOH	147~148
Vc	〃	benzene	146~147
Vd	microcrystals	benzene-EtOH	152~153
VIa	needles	Me_2CO	82~ 84
VIb	〃	petr. benzin-benzene	71~ 72
VIc	microcrystals	Me_2CO	81~ 82
VId	scales	petr. benzin-benzene	96~ 98
VII[a)]	needles	benzin-benzene	105~106

a) VII : 3, 6-Diphenethylpyridzine 1-oxide

seemed likely that hydroxy compounds (V) might be produced in reactions at 100 and 65°, but no effort was made to isolate these intermediates.

As these styryl compounds including bis-styryl compounds showed a medium to strong absorption at 960~980 cm^{-1} in their infrared spectra, it was considered that the compounds existed in *trans*-form.

These styryl compounds (IIIa, IVa~d) were catalytically hydrogenated over palladium-charcoal to the corresponding phenethylpyridazine 1-oxides (VII, VI a~b) physical data of which are tabulated in Table II.

From the results of the present experiments, it may be deduced, though rather qualitatively, that 3-methyl group is the most inactive one among the four in monomethylpyridazine 1-oxides, and the order of the reactivity is $5>4\geqq6$.

In order to determine exactly their reactivity further, it is necessary to examine kinetically. However, as the reaction proceeds in two steps and the first step is not separable from the second, this study will be left for future consideration.

Experimental

Reaction of 3,6-Dimethylpyridazine 1-Oxide (I) with Benzaldehyde; 3,6-Distyryl-pyridazine 1-Oxide (IIIa)——i) A mixture of 206 mg. of I and 500 mg. of benzaldehyde in 1 ml. of MeOH was heated with MeONa (prepared from 100 mg. of Na and 2 ml. of MeOH) in a sealed glass tube in a water bath for 1 hr. After cooling, yellow needles obtained were washed with MeOH and H_2O, and recrystallized from Me_2CO, yellow needles, m.p. 262~263° (decomp.). Yield, 398 mg. (80%). *Anal.* Calcd. for $C_{20}H_{16}ON_2$: C, 79.98; H, 5.37; N, 9.33. Found: C, 79.62; H, 5.52; N, 9.68. This was affected by sunlight.
ii) A mixture of 221 mg. of I and 200 mg. of benzaldehyde in 1 ml. of MeOH was heated with MeONa (prepared from 100 mg. of Na and 2 ml. of MeOH) in a sealed glass tube in a water bath for 1 hr. After cooling, precipitates were filtered and washed with MeOH and H_2O, m.p. 260~261° (decomp.). Yield 129 mg. (24%). No melting point depression was observed on admixture with product from i). Then, the filtrate was added H_2O, extracted with $CHCl_3$, and $CHCl_3$-extracts were dried over Na_2SO_4 and evaporated. The residue was recrystallized from benzene to give 97 mg. of colorless needles, m.p. 112~113°, which were identical with I by mixed melting point determination and comparison of IR spectra.

Reaction of 3,6-Dimethylpyridazine 1-Oxide (I) with Anisaldehyde; 3,6-Bis(*p*-methoxystyryl)pyridazine 1-Oxide (IIIb)——A mixture of 208 mg. of I and 220 mg. of anisaldehyde in 1 ml. of MeOH was heated with MeONa (prepared from 100 mg. of Na and 2 ml. of MeOH) in a sealed glass tube in a water bath for 1 hr. After cooling, yellow scales were filtered and washed with H_2O and MeOH, m.p. 244~246° (decomp.). Yield 97 mg. (18%). *Anal.* Calcd. for $C_{22}H_{20}O_3N_2$: N, 7.77. Found: N, 7.77. From the filtrate 76 mg. of I was obtained by the same method as mentioned above.

Reaction of 3,6-Dimethylpyridazine 1-Oxide (I) with *p*-Dimethylaminobenzaldehyde; 3,6-Bis(*p*-dimethylaminostyryl)pyridazine 1-Oxide (IIIc)——A mixture of 206 mg. of I and 246 mg. of *p*-dimethylaminobenzaldehyde in 1 ml. of MeOH was heated with MeONa (prepared from 100 mg. of Na and 2 ml. of MeOH) in a sealed glass tube in a water bath for 3 hr. After cooling, red-brown microcrystals were filtered and washed with MeOH and H_2O, m.p. 250° (decomp.). Yield, 216 mg. (35%). *Anal.* Calcd. for $C_{24}H_{26}ON_4$: C, 74.58; H, 6.78; N, 14.50. Found: C, 73.46; H, 6.88; N, 14.92. Then, the filtrate was evaporated and added H_2O, and extracted with $CHCl_3$ after precipitated *p*-dimethylaminobenzaldehyde had been filtered. The $CHCl_3$ layer was evaporated after drying over Na_2SO_4, and the starting material, 96 mg., m.p. 103~108°, was obtained.

Reaction of Monomethylpyridazine 1-Oxides (IIa~d) with Benzaldehyde; Monostyrylpyridazine 1-Oxides (IVa~d) and (β-Hydroxyphenethyl)pyridazine 1-Oxides (Vb~d)——i) At 100° and at 65°. A mixture of 200 mg. of a monomethylpyridazine 1-oxide (IIa~d) and 500 mg. of benzaldehyde in 1 ml. of MeOH was heated with MeONa (prepared from 100 mg. of Na and 2 ml. of MeOH) in a sealed glass

Table III.

Compound	Formula	Calcd. (%)			Found (%)		
		C	H	N	C	H	N
IVa	$C_{12}H_{10}ON_2$	72.71	5.09	14.13	73.35	5.52	13.86
IVb	〃	72.71	5.09	14.13	72.39	5.23	14.03
IVc	〃	72.71	5.09	14.13	72.50	5.09	
IVd	〃	72.71	5.09	14.13	72.82	5.94	13.64

tube in a water bath at about 100° or at 65° for 1 hr. After cooling, a crystalline residue was filtered, washed with MeOH and H_2O and recrystallized from MeOH or benzene. IVa was extracted with $CHCl_3$ from the reaction mixture. After evaporation of $CHCl_3$, the residue was recrystallized. The compounds (IVa~d) listed in Table II were obtained by treating in the same way. Their analytical data were shown in Table III.

ii) At 45°. At this temperature, the quantities of reagents were changed a little and each reaction was carried out as follows.

A mixture of 200 mg. of a monomethylpyridazine 1-oxide (IIa~d) and 200 mg. of benzaldehyde in 3 ml. of MeOH was warmed with MeONa (prepared from 50 mg. of Na in 1 ml. of MeOH) in a sealed glass tube at 40° for 1 hr., and MeOH was evaporated under reduced pressure.

The residue from IIa was mixed with H_2O and extracted with $CHCl_3$. Evaporation of the $CHCl_3$ from the extracts gave an oily substance, which was dissolved in $CHCl_3$ and chromatographed through Florisil, and was eluted with $CHCl_3$. After distilling the $CHCl_3$ off from the eluates to dryness, the residue was recrystallized from benzene to colorless plates, m.p. 68~69°, which were found identical with IIa by mixed melting point determination and by comparison of their IR spectra. Yield. 166 mg.

In the reactions with IIb~d, the residues, obtained on evaporation of MeOH from reacted mixtures under reduced pressure, became crystalline after adding H_2O to them and standing for 30 min. The crystals were filtered, washed with H_2O, and were recrystallized to pure samples (Vb-d). These were summarized in Table II, and their analytical data in Table IV.

Table IV.

Compound	Formula	Calcd. (%)		Found (%)	
		C	H	C	H
Vb	$C_{12}H_{12}O_2N_2$	66.65	5.59	66.84	5.61
Vc	〃	66.65	5.59	66.75	5.63
Vd	〃	66.65	5.59	66.57	5.68

The filtrates and the washings were extracted with $CHCl_3$, and the extracts were dried over Na_2SO_4, and evaporated. The residues were extracted with hot benzene, and the starting materials were recovered from the benzene extracts.

Conversion of (β-Hydroxyphenethyl)pyridazine 1-Oxides (Vb~d) to Styrylpyridazine 1-Oxides (IVb~d) ——i) A mixture of 12 mg. of Vb and methanolic MeONa (prepared from 50 mg. of Na and 2 ml. of MeOH) was heated in a sealed glass tube in a water bath for 1.5 hr. MeOH was evaporated, the residue was added to H_2O, crystals were filtered and washed with H_2O, and recrystallized from MeOH to give 5 mg. of light yellow needles, m.p. 186~188°, identical with IVb by comparison of IR spectra.

ii) A mixture of 40 mg. of Vc in methanolic MeONa (prepared from 50 mg. of Na and 2 ml. of MeOH) was heated in a sealed glass tube on a water bath for 1 hr. When the reaction mixture was treated in the same way as described above, 13 mg. of IVc was obtained as colorless needles, m.p. 158~159°.

iii) A mixture of 35 mg. of Vd in methanolic MeONa (prepared from 50 mg. of Na and 2 ml. of MeOH) was heated in a sealed glass tube in a water bath for 1 hr. When the reaction mixture was treated in the same way as described in i), 24 mg. of IVd was obtained as colorless scales, m.p. 159~161°.

Reduction of Styrylpyridazine 1-Oxides (IVa~d, IIIa) to Phenethylpyridazine 1-Oxides (VIa~d, VII) ——Compounds (IVa~d, IIIa), listed in Table II, were prepared by treating in the same way as mentioned below.

A mixture of 200 mg. of styrylpyridazine 1-oxide in 30 ml. of MeOH was hydrogenated over 100 mg. of 10% Pd-C. One molecular equivalent of H_2 was absorbed within 30 min. After removal of the catalyst by filtration, MeOH was evaporated to dryness. The residue was recrystallized from Me_2CO, petr. benzin-benzene or benzin-benzene.

These were summarized in Table II, and their analytical data in Table V.

Table V.

Compound	Formula	Calcd. (%)		Found (%)	
		C	H	C	H
VIa	$C_{12}H_{12}ON_2$	71.98	6.04	72.76	6.09
VIc	〃	71.98	6.04	72.36	5.96
VId	〃	71.98	6.04	71.94	6.21
VII	$C_{20}H_{20}ON_2$	78.92	6.62	79.01	6.69

The analysis of VIb has not yet been done. However, from the results of VIa, VIc, VId and VII, and its IR spectrum, it might not fail to consider VIb as 4-phenethylpyridazine 1-oxide.

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Summary

The nucleophilic activity of methyl group in 3,6-dimethylpyridazine 1-oxide (I) and monomethylpyridazine 1-oxides (II) with benzaldehyde was studied. Their corresponding (β-hydroxyphenethyl)pyridazine 1-oxides and styryl compounds were gained. From the results, it may be deduced as follows. (i) The reactivity of 3- and 6-methyl groups in I were almost same, different from the cases of 3,6-dichloropyridazine 1-oxide. (ii) 4- and 6-methyl groups in II were similarly reactive, but 5-methyl was more active, and 3-methyl less active than fore-mentioned two methyl groups. These styryl compounds that were produced respectively, were catalytically hydrogenated with Pd-C to corresponding phenethylpyridazine 1-oxides.

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188. Keizo Inoue, Yasuji Suhara, and Shoshichi Nojima : On the Cardiolipin Analogues. Syntheses of Dipalmitoyl-D,L-α-glycerylphosphoryl-propanol Sodium Salt and Bis(dipalmitoyl-D,L-α-glyceryl-phosphoryl)-1,3-propanediol Disodium Salt.

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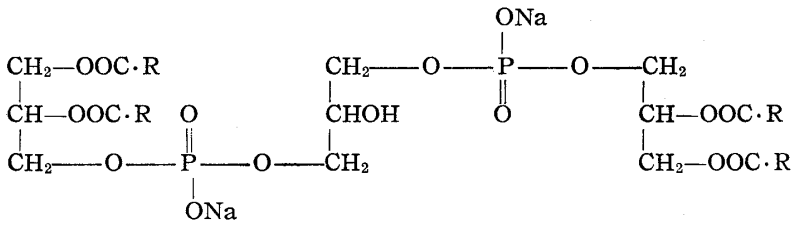
Cardiolipin, the phosphatide as a constituent of the Wasserman antigen in the diagnosis of syphilis, was first isolated from beef heart by Pangborn in 1941.[1] It was then characterized as a complex phosphatidic acid derivatives.[2] Similar derivatives with different fatty acids have been found in many animals and vegetable tissues and are, for the time being, recognized as one of the important components among the phosphatides in the tissues. The chemical composition and structure of this phosphatide were recently reinvestigated by several workers and the following new chemical structure was proposed.[3]



R·CO-=Unsaturated fatty acid residue

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