INTRODUCTION

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Plant protection chemicals play an important role towards achieving self-sufficiency in the production of food, fodder and fibre. Pesticides with their higher cost-benefit ratio than that of fertilizers have become one of the essential inputs in modern agriculture and so the quality of pesticides is of utmost importance for not only ensuring better efficacy but also assuring safety to the users and environment. As a result various types of pesticides are being prepared, and their pesticidal, toxicological and other properties are being studied everyday. Of these, organophosphorus compounds is a class in which quite a large number of compounds have been synthesized and examined as effective pesticides (1). Owing to the wide diversity in their pesticidal activities, these organophosphorus compounds have won epoch-making popularity for use in field as potential pest controlling agents. Discovery of Saligenin cyclic phosphate as a biologically active metabolite of tri-o-tolyl phosphate (2, 3) has led to extensive studies on synthesis and chemical, biochemical and biological properties of many related compounds (4, 5). Of them Salithion (2-methoxy-4H-1, 3, 2-benzodioxaphosphorin-2-sulphide) has been produced in a large quantity and practically used as an insecticide in Japan (1, 5).

Various cases of poisoning due to O-cresyl phosphate (TOCP) led to an intensive research in this field (6, 7). The phosphate triesters of cresols have been widely used in industries as plasticizers, lubricants, solvents, oil additives etc. Later, Aldridge and Barnes observed that all neurotoxic aryl phosphate, except tri-p-ethyl-phenyl phosphate, had at least one alkyal group carrying the α-hydrogen atom on the ortho position (6). This structure-neurotoxicity relationship of triaryl phosphate became clearly understable by the isolation and characterization of the active metabolite (M) was O-tocyl saligenin cyclic phosphate. This compound (M) was about 100 times by the dose of 20 mg/kg in mice. TOCP potentiated the toxicity of Malathion 4 times by the dose 100 mg/kg; (M) was ten million times more active than TOCP in the <u>in vitro</u> inhibition of plasma Cholinesterase (4). The conversion of TOCP into active metabolite (M) proceeds in two steps; the first step, which is catalysed by mixed-function oxidases (mfo), involves the hydroxylation of a ring methyl group; the second step involves the cyclization of this intermediate product to (M) through intramolecular transphosphorylation; the latter reaction is catalyzed by plasma albumin (7).

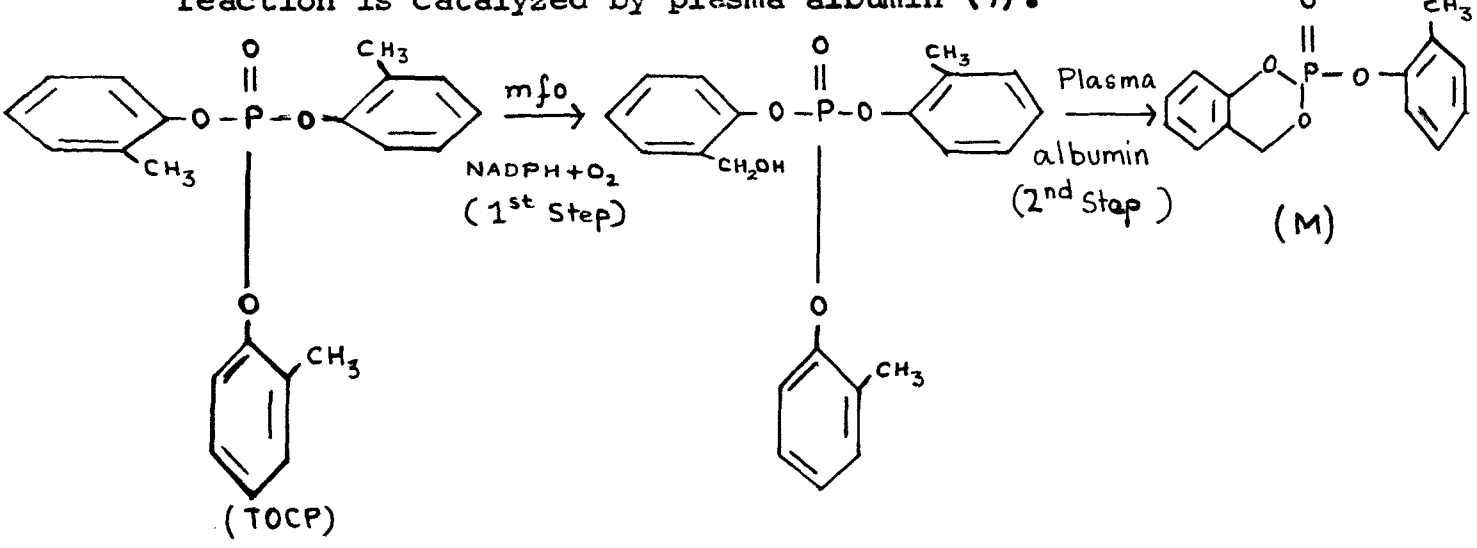


Fig. 1 Metabolic Activation of TOCP

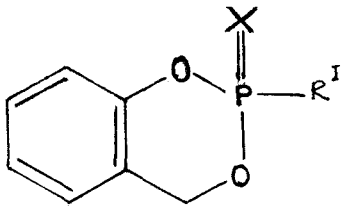
In the cyclisation reaction, no alkyl ester group participated as the leaving group (4). Actually alkyl not aryl saligenin cyclic phosphate was formed *in vitro* from alkyl di-o-tolyl phosphate (8). Such metabolic conversion of TOCP or its analogs was observed in hens (3), rats (3), cats (10), and insects (9).

Aryl saligenin cyclic phosphates have shown no insecticidal activity but manifested a high delayed neurotoxicity to cause ataxia in hens; surprisingly the corresponding cyclic esters (both P=O and P=S compounds) having a small alkyl group on phosphorus revealed high insecticidal activity (2, 10). As a result of this research salithion was discovered by the pesticide research group of Kyushu University in Japan in 1963 and was developed into a commercial insecticide in 1968 by Sumitomo Chemical Co.

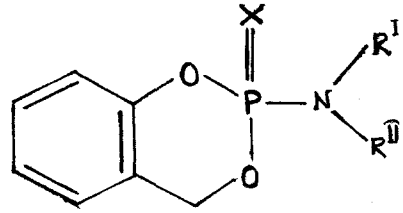
The cyclic phosphate and phosphonate esters of saligenin are readily synthesized by condensation of saligenin and substituted phosphoryldichlorides in the presence of a dehydrochlorinating agent such as tertiary amine in a dry solvent like chloroform or toluene at low temperature (10). In some cases where the reaction is affected difficulty by using tertiary amine, the reaction has been made to proceed by heating the mixture for 10 to 20 hours in the presence

of anhydrous potassium carbonate together with copper powder (12) instead of tertiary amine.

Such compounds which are produced by the method employing a tertiary amine, include the compounds having X=S and R^1= methoxy in formula (I) and X = S, R^I= H and R^2= alkyl containing more than one carbon atom or R^I = R^{II}= alkyl in the formula (II).



I



II

The process using potassium carbonate is made to proceed by a reaction between liquid and solid phases. Therefore, even if potassium carbonate is employed as finely divided powder form, it often causes a remarkable lowering and fluctuation of the yield (12). Thus Salithion was first prepared with inconsistent, and often, very low yield by heating (90°C) saligenin and methyl phosphorodichloridothionate in toluene for a long period (more than 15 hours) in the presence of anhydrous potassium carbonate together with copper powder as catalyst (13).

This difficulty, was, overcome later by applying the well-known Schotten-Baumann acylation procedure using an aqueous solution of sodium hydroxide (Fig.2).

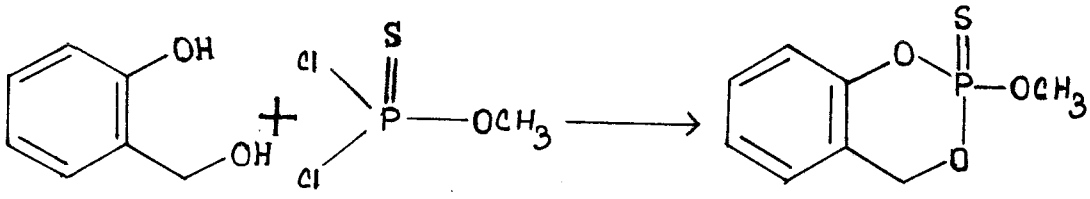


Fig. 2. Synthesis of Salithion

A careful observation of literatures furnishes (12, 13, 14, 15, 17, 18, 19) a variety of Saligenin cyclic phosphorus esters in good numbers, which have been prepared and examined for insecticidal activity as well as other biological properties. They involve phosphates, phosphorothiolates, phosphoramidates, phosphonates and their thionoanalogs. A comprehensive but not a complete list of saligenin and ring substituted saligenin cyclic phosphorus esters is given in Table 1.

Insecticidal Activity : The insecticidal activities of various saligenin cyclic phosphorus compounds including phosphates (15, 13) phosphoramidates (20), phosphorothiolates (18), phosphonates (21) and their thiono analogs (16, 14), have been tabulated (Table I). The saligenin cyclic compounds in any series having an aryl group as an exocyclic substituent on the phosphorus atom have either poor or no insecticidal activity, but small alkyl derivatives have high insecticidal activity. The insecticidal activity appears to relate with the size of the exocyclic substituent on the phosphorus atom. Methyl derivatives are much more active than higher alkyl and aryl derivatives, except for phosphonothionate series in which ethyl derivative is more

active than methyl derivative. N,N-dialkyl phosphoramidates are much less active than NH-alkyl derivatives. It is interesting to note that the exocyclic substituent of the most active compound in each series (OCH_3, SCH_3, $NHCH_3$, CH_2CH_3) differs from each other in electronic characteristics, but is similar in size and resembles in steric property such as the distance (about 2.9 A°) between P and C atoms in the P-X-C function, if the bond angle of bivalent S is near 90° rather than 109.5°. The phosphorothionothionates are not enough in insecticidal activity (Table I). The phosphates, phosphorothiolates, and phosphonates are too unstable to be used practically as insecticides. The phosphoramidates are several times more toxic to mammals than phosphorothionates. Introduction of any substituents on the benzene ring, the hetero ring or the exocyclic ester group decreases the insecticidal activity (Table I). Thus, the simplest phosphorothionate, Salithion, was the most useful compound as the whole series of saligenin cyclic phosphorus compounds. Salithion is a broad spectrum insecticide for use in orchards and vegetable gardens. It is particularly effective for controlling aphids, mites, mealybugs and lepidopteran larvae. It manifests the insecticidal action not only as contact and stomach poisons but also as a fumigant.

The systemic activity of some saligenin cyclic phosphoramidates against rice stem-borers and green rice

leafhoppers on rice plants are observed. Methylphosphoramidate is more active than Schradan but less active than Thimet against rice leafhoppers. Against rice stem-borers cyclic N-methylphosphoramidothionate is superior to Lindane and Diazinon. Salithion also showed more or less systemic activity against armyworm and mite. No systemic activity was observed in other compounds.

Nematocidal Activity : Some saligenin cyclic phosphoramidates and phosphoramidothionates are very effective in killing nematodes (Table II). N-methylphosphoramidate is most active, but N,N-dimethyl-phosphoramidate is inactive against Rhabditis sp. suspended in water. Phosphates and phosphonates are almost inactive, but their thiono analogs are effective. Saligenin cyclic phenyl phosphonothionate exhibit high activity.

Fungicidal Activity : Salithion has no fungicidal activity (Table III). Some of them are effective to protect rice plants from rice blast disease (due to infection by Pyricularia oryzae). Ethyl and n-butyl phosphorothiolates are most promising as fungicides.

Synergistic Activity : Saligenin cyclic aryl phosphates and phosphorates have synergistic activity with Malathion against insects and mites, particularly their resistant strain (24). 7-methyl-2-phenyl-4H-1,3,2, benzodioxaphosphorin-2 oxide is the

most active synergist against resistant houseflies and green rice leafhoppers. For the resistant strains of red citrus mite, Panonychys citwi (McGregor), saligenin cyclic phenylphosphonate showed good synergistic action with Malathion. Saligenin cyclic alkyl phosphates and phosphorothionates are only active as synergists of malathion against houseflies and green rice leafhoppers.

Antiesterase activity : The insecticidal saligenin cyclic methyl phosphate (Salixon) is very active as an inhibitor of cholinesterase. However, the highly neurotoxic aryl phosphate is a poor inhibitor of cholinesterase, but is a very specific inhibitor of aliesterase (1,22). The less nemotoxic arylphospharate occupied an intermediate position. In any series, when the size of the exocyclic substituent increases, the compound becomes a more selective inhibitor of aliesterase; in contrast, the compound carrying a small substituent is a more selective inhibitor of cholinesterase (Table IV). Arylphosphonates are more specific inhibitors of pseudocholinesterase; alkyl phosphates are less specific and aryl phosphates are intermediate.

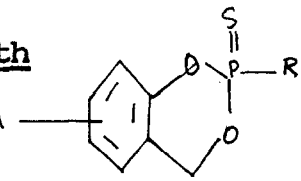
Anti-SH Enzyme Activity : The saligenin cyclic phosphorothiolates (Table I) have high activity to salicylate (alkylate) mercaptans and to inhibit "SH enzymes" such as alcohol dehydrogenase(12,19).

The activities seem to be related with fungicidal property but not with insecticidal activity. Saligenin cyclic phosphorothiolates are partially hydrolyzed by opening of the heterocyclic POC (aryl) bond, more easily than phosphate esters. It was demonstrated that the hydrolyzate of saligenin cyclic methyl phosphorothiolate i.e. O-ortho-hydroxy benzyl s-methyl hydrogen phosphorothiolate, reacted with mercaptans to give salicyl thioethers (19). These observations indicate that the saligenin cyclic phosphorothiolates may react with cholinesterase to phosphorylate its serine hydroxyl group and may, on the other hand, be hydrolyzed to the O-hydroxybenzyl esters which react with "SH enzymes" to alkylate their thiol group. "SH enzymes" may be essential for the life of fungi.

The high biological activities of saligenin cyclic phosphorus compounds may be attributed to the heteroring involving erol and benzyl ester linkages. The alkylation reaction may be responsible for "SH-enzyme" inhibition and fungicidal activity. The phosphorylation reaction is responsible for esterase inhibition, and insecticidal activity and animal toxicity. An exocyclic substituent group affects physical and biological properties by virtue of its electronic and steric characteristics. Thus, methyl phosphorothionate is useful as an insecticide, alkylamidates have systemic activity, alkyl phosphorothiolates have fungicidal activity, phenyl phosphonates have antifilarial activity, and aryl phosphates are nemotoxic and have synergistic activity.

Table 1

Substituted Saligenin Cyclic Phosphorus Esters with Physical Properties and Insecticidal Activity.



R	A	X	* Procedure	b.p.°C/mm Hg(m.p.°C)	LD_{50} (μg/housefly)
OCH_3	H	S	(S)	55-56°C	0.05
OCH_3	H	O	(P)	110-2°/0.05	0.035
O-n-C_3H_7	H	O	(P)	129-32°/0.05	7.1
O-n-C_4H_9	H	O	(P)	150-4°/0.05	10(40%)
O-C_2H_5	H	S	(P)	Liquid(not distilled)-	0.33
OC_6H_5	H	S	(P)	(36°C)	2.0
CH_5	H	S	(P)	(37°)	-
CH_3	H	O	(P)	140°/0.5 (35°)	-
C_2H_5	H	O	(P)	143-9°/0.3(25°)	0.17
i-C_3H_7	H	O	(P)	(80°)	0.33
Sec-C_4H_9	H	O	(P)	110°/0.5	7.0
CH-CH_2	H	O	(P)	(74°)	-
CH-CH_2	H	O	(P)	155°/2.5	0.68
CH Cl_2	H	O	(P)	160°/0.8(51°)	10(60%)
CH_2CH_2Cl	H	O	(P)	139-141°/0.1	0.99
CH_3	H	O	(P)	130°/0.6	0.31
C_2H_5	H	S	(P)	120°/0.6	0.08
i-C_3H_7	H	S	(P)	108°/0.6	0.09
CH_2Cl	H	S	(P)	146-155/0.4	1.14

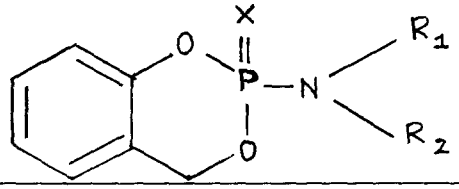
R	A	X	*	Procedure	b.p.°C/mm Hg(m.p.°C)	LD_{50}(µg/housefly)
OCH_3	6-CH_3	O		(P)	139-140/0.3	0.1
OC_2H_5	6-CH_3	O		(P)	152-156/0.3	0.4
OCH_3	7-CH_3	O		(P)	109/0.05	0.43
O-n-C_3H_7	7-CH_3	O		(P)	141-147/.01	7.2
C_6H_5	7-CH_3	O		(P)	(93-95)	10.0
$NHCH_3$	7-CH_3	O		(P)	(145-146)	0.14
OCH_3	8-CH_3	O		(P)	118-120/0.5	2.0
OC_2H_5	8-CH_3	O		(P)	165/0.6	2.1
OC_6H_5	8-CH_3	O		(P)	135-140/0.6	10.0
OCH_3	6-CL	O		(P)	145-152/0.2	0.09
OC_2H_5	6-CL	O		(P)	160/0.2	0.13
O-n-C_3H_7	6-CL	O		(P)	167-169/0.15	0.70
O-n-C_4H_9	6-CL	O		(P)	187/0.18	2.5
OC_6H_5	6-CL	O		(P)	(890)	10.0
$NHCH_3$	6-CL	O		(P)	(148°)	0.09
OCH_3	8-CL	O		(P)	170-171/0.15	0.23
OC_2H_5	8-CL	O		(P)	151/0.18	0.15
O-n-C_3H_7	8-CL	O		(P)	183/0.18	0.30
O-i-C_3H_7	8-CL	O		(P)	137/0.04	-
OC_6H_5	8-CL	O		(P)	203/0.52(540)	10.0
$NHCH_3$	8-CL	O		(P)	(128-1290)	0.30
OCH_3	6-CH_3	S		(S)	(34-35°)	2.0

R	A	X	*	Procedure	b.p.°C/mm Hg(m.p.°C)	LD_{50} (μg/housefly)
OC_2H_5	6-CH_3	S		(S)	(71-72°)	10.0
O-n-C_3H_7	6-CH_3	S		(S)	158-160/0.2	10.0
OCH_3	7-CH_3	S		(S)	110-115/0.65	0.23
O-n-C_3H_7	7-CH_3	S		(S)	140-142/0.65	7.5
OCH_3	8-CH_3	S		(S)	68-70/0.15	1.3
OC_2H_5	8-CH_3	S		(S)	108-109/0.15	3.0
O-n-C_3H_7	8-CH_3	S		(S)	120-124/0.15	7.5
$NHCH_3$	8-CH_3	S		(S)	(30°)	3.6
OCH_3	6-C_6H_5	S		(S)	Oil **	0.4
OC_2H_5	6-C_6H_5	S		(S)	Oil **	0.5
O-n-C_3H_7	6-C_6H_5	S		(S)	Oil **	1.0
OCH_3	6-OCH	S		(S)	Paste**	0.55
OCH_3	6-$COCH_3$	S		(S)	Paste*	2.50
OCH_3	6-CL	S		(P)	170-178/0.2	1.75
$NHCH_3$	6-CL	S		(P)	175-180/0.25	0.06
SCH_3	6-CL	S		(P)	160-170/0.2	-
OCH_3	8-CL	S		(S)	(72-730)	0.13
$NHCH_3$	8-CL	S		(P)	(46-47°)	0.09
SCH_3	H	S		(S)	(69-70°)	0.18
SC_2H_5	H	S		(S)	145-147/0.2	9.0
S-n-C_3H_7	H	S		(S)	145-150/0.25	2.2
S-i-C_3H_7	H	S		(S)	140-143/0.1	5.0

R	A	X	*	Procedure	b.p.°C/mm Hg(m.p.°C)	LD_{50} (μg/housefly)
$S-C_3H_5$	H	S		(S)	140-147/0.3	1.7
$S-n-C_4H_9$	H	S		(S)	160-167/0.25	10.0
$S-C_6H_5$	H	S		(S)	(79-80°)	10(0%)
SCH_3(MTBO)	H	O		(P)	144-145/0.1(44)	3.0
SC_2H_5	H	O		(P)	140-145/0.04	11.21
$S-n-C_3H_7$	H	O		(P)	145-147/0.07	94.50
$S-i-C_3H_7$	H	O		(P)	155-158/0.1	17.23
$S-n-C_4H_9$	H	O		(P)	157-160/0.02	211.8
SC_6H_5	H	O		(P)	(88-890)	73.61

Table II

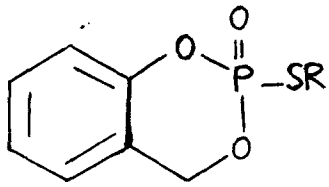
Saligenin Cyclic Phosphoramidates and Phosphoramidothionol Insecticidal Activity toxicity and nematocidal activity.



X	$\begin{matrix} R_1 \\ \diagup \\ N \\ \diagdown \\ R_2 \end{matrix}$	LD_{50} (μg/gm) (Rice stem borer)	LD_{50} (μg/gm) green rice-leaf hopper	LD_{50} mg/kg Mouse	Minimum Conc(ppm) to kill 100% nematodes-Rhabditis
O	NHMe	2.84	0.4	5.0-7.5	10
O	NHEt	22.29	3.5	30-50	10-25
O	NHnPr	33.60	33.0	50	20-50
O	NHiPr	103.34	350	50	50-100
O	NhnBu	214	400	50	25-50
O	$N(Mo)_2$	13-80	4.0	-	200(10%)
O	$N(Et)_2$	167.61	34.1	50	100-150
S	NHMe	4.84	4.1	20.30	25-50
S	NHiEt	Liquid (undistillable)	36.25	-	-
S	$N(Me)_2$	-	-	-	50-100
S	$N(Et)_2$	-	-	-	200(30%)
S	OMe (Salithion)	1.13	30.6	88	-
O	DMe (Salioxon)	2.16	1.8	52	-
	Parathion	3.43	3.6	5-7	-
	Malathion	-	0.8	347	-
	D-D mixture (Mixture of 1,3 dichloro propene and 1-2 dichloroprane)	-	-	-	800(85%)

Table III

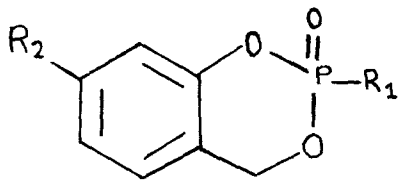
Saligenin Cyclic Phosphorothiolates : Fungicidal activity



R	Protective value(%) against *P.oryzae*				Therapeutic Value(%) to *P. oryzae*
	250 ppm	100 ppm	50 ppm	25 ppm	at 200 ppm
CH_3 (MTBO)	100	100	100	84.8	7.1
C_2H_5	100	93.7	92.5	81.5	100
n-C_3H_7	100	57.1	34	-	-
i-C_3H_7	-	68.7	34.4	-	-
n-C_4H_9	100	91.7	93.3	75.6	97.6
C_6H_5	50.2	-	-	-	-
Salithion	52 (at 500 ppm)	-	--	-	-
Hinosan	100	-	86.2	-	95.2(at 250 ppm)
Blasticidine-S	98.5	-	86.3	-	97.6
Pentachloro-benzyl alcohol	-	98.8	93.5	-	0

Table IV

The Inhibition of Housefly, Human Blood, Horse-Serum-Esterases by some Saligenin Cyclic Phosphorus Compounds.



R_1	R_2	House-fly I_{50} (M) X 10^8		Human Blood I_{50} (M) X 10^8		Horse plasma I_{50} (M) X 10^8	
		ChE	Ali E	p-ChE	t-ChE	Ali E	Malathionase
OCH_3	H (Salioxon)	7.6	8.4	1.8	17.0	280	620
OC_2H_5	H	13.2	2.1	1.6	25.0	240	-
O-n-C_3H_7	H	50.7	3.0	-	-	-	-
O-n-C_4H_9	H	37.5	2.3	-	-	-	-
OC_6H_5	H	155	1.4	0.5	12.0	120	120
C_6H_5	H	-	-	0.65	72.0	180	470
C_6H_5	CH_3	-	-	1.6	68.0	230	-
OPh-2-CH_3	H	-	-	1.3	39.0	200	-

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