Quantitative Structure−Activity Relationship Studies of *O,O*-Bisaryl Alkyl Phosphonate Fungicides by Hansch Approach and Principal Component Analysis

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Quantitative structure−activity relationship studies of *O,O*-bisaryl alkyl phosphonate fungicides were conducted by the Hansch approach. The variation in fungicidal activity among members of the series against all the four test fungi depended mainly on changes in hydrophobicity of the phenyl substituents and molar refractivity of the alkyl substituent. Molar refractivity of the substituents at the *para* position of the phenyl ring and electronic factors also played significant roles in two cases each. Principal component analysis of the fungicidal activity data revealed that the first principal component had 79.2% of the information content. Physicochemical interpretation of the scores corresponding to the first principal component gave results consistent with those obtained by Hansch analysis. It appears that the fungicidal molecule interacts with the bioreceptor through the two *para* positions of the phenyl rings and the alkyl substituent, and that the steric bulk of the substituents play a dominant role during the interaction.

Keywords: *Quantitative structure−activity relationships; O,O-bisaryl alkyl phosphonate fungicides; principal component analysis*

INTRODUCTION

O,O-Bisaryl alkyl phosphonates are a group of compounds that exhibit broad-spectrum fungicidal activity. We previously reported the synthesis and fungicidal activity of *O,O*-bisaryl methyl phosphonates and *O,O*-bisaryl 1,1-dichloroethyl phosphonates (Roy *et al*., 1980; Vasu and Roy, 1983). Synthesis, fungitoxicity, and quantitative structure−activity relationship (QSAR) studies of *O,O*-bisaryl isopropyl phosphonates were subsequently reported (Nidiry and Roy, 1988; Nidiry and Roy, 1989). A detailed study of the various steric effects influencing the fungicidal activity of *O,O*-bisaryl isopropyl phosphonates was also reported (Nidiry and Roy, 1994). This report is of a comprehensive study of the QSAR of a set of 62 *O,O*-bisaryl alkyl phosphonate fungicides that consists of 18 *O,O*-bisaryl methyl phosphonates, 20 *O,O*-bisaryl isopropyl phosphonates, and 24 *O,O*-bisaryl 1,1-dichloroethyl phosphonates. The fungicidal activity data of this set of fungicides against *Helminthosporium oryzae*, *Pyricularia oryzae*, *Alternaria alternata*, and *Rhizoctonia bataticola* is correlated with the physicochemical parameters of the set as determined by Hansch analysis.

Hansch analysis is widely used in quantitatively determining the effect of physicochemical parameters, especially from the point of view of structure−selectivity relationships, but the method of principal component analysis (PCA) is supposed to give more vital information from the point of view of structure−broad spectrum relationships (Schaper and Kaliszan, 1986; Nendza and Seydel, 1988). In principle, PCA is a mathematical tool to reduce the variables of a data matrix) [e.g., 50% effective concentration (EC_{50}) values of a congeneric series against different fungi] to a fewer variables (namely, the principal components) and to thereby recognize the collinearities. The PCA of the fungicidal activity data of *O,O*-bisaryl alkyl phosphonates is also reported in this paper, and the results of PCA are compared with the results obtained by Hansch analysis.

Table 1. Descriptor Variables of Substituents

	descriptor variable					
substituent	π	σ_m	σ_p	F'	E_s	MR
H	0.00	0.00	0.00	0.00	0.00	1.03
CH_3	0.56	−0.07	−0.17	−0.04	−1.24	5.65
C1	0.71	0.37	0.23	0.41	−0.97	6.03
NO_2	−0.28	0.71	0.78	0.67	−2.52	7.36
OCH_3	−0.02	0.12	−0.27	−0.26	−0.55	7.87
SCH_3	0.61	0.15	0.00	0.20	−1.07	13.82
$C(CH_3)_3$	1.98	−0.10	−0.20	−0.07	−2.78	19.62
$CH(CH_3)_2$	−[a]	−[a]	−[a]	−[a]	−[a]	14.96
CH_3CCl_2	−[a]	−[a]	−[a]	−[a]	−[a]	20.96

[a] Not relevant to the present study.

MATERIALS AND METHODS

Toxicity Data and Biological Materials. Fungicidal activity data of the compounds were taken as the dependent variables and were expressed as $-\log EC_{50}$, where EC_{50} is the median effective molar concentration for mycelial growth inhibition on potato dextrose−agar medium against the following phytopathogenic fungi: *Helminthosporium oryzae* Anct.; *Pyricularia oryzae* Cav.; *Alternaria alternata* Breda de Haan.; and *Rhizoctonia bataticola* (Tassi) Goid. These data are presented in Table 2 along with the activity of ediphenphos (*O*-ethyl *S,S*-diphenyl phosphorodithioate), a commercial standard (Roy and Taneja, 1992) that was included for reference purposes.

Chemical Descriptors. The hydrophobocity parameter, $\Sigma\pi$, for each compound was calculated by simple addition of Hansch−Fujita's substituent parameters, π, characterizing hydrophobicity (Hansch and Leo, 1979). The electronic pa-

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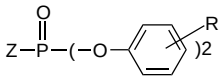
Table 2. Experimental and Calculated Fungicidal Activity Data of *O,O*-Bisaryl Alkyl Phosphonates



			−log EC_{50} (mol/L) against test species							
			H. oryzae		*P. oryzae*		*A. alternata*		*R. bataticola*	
compound no.	Z	R	exptl	calcd[a]	exptl	calcd[b]	exptl	calcd[c]	exptl	calcd[d]
I	CH_3	H	3.10	3.22	3.22	3.41	3.42	3.29	3.34	3.40
II	CH_3	2-Cl	3.27	3.50	3.63	3.73	3.58	3.72	3.41	3.63
III	CH_3	4-Cl	3.41	3.43	3.69	3.66	3.64	3.50	3.58	3.63
IV	CH_3	2-CH_3	3.42	3.44	3.53	3.52	3.46	3.43	3.50	3.58
V	CH_3	3-CH_3	3.38	3.44	3.63	3.52	3.49	3.46	3.49	3.58
VI	CH_3	4-CH_3	3.41	3.38	3.21	3.46	3.58	3.46	3.50	3.58
VII	CH_3	2-OCH_3	3.22	3.21	3.13	3.29	3.48	3.43	3.52	3.39
VIII	CH_3	4-OCH_3	3.46	3.09	3.36	3.17	3.42	3.28	3.52	3.39
IX	CH_3	4-SCH_3	3.26	3.08	3.33	3.22	3.48	3.47	3.15	3.59
X	CH_3	2,4-Cl_2	3.84	3.72	3.61	3.98	3.60	3.93	3.57	3.55
XI	CH_3	3-CH_3,4-Cl	3.84	3.66	3.72	3.83	3.66	3.67	3.66	3.80
XII	CH_3	2,3-$(CH_3)_2$	3.82	3.67	3.67	3.67	3.65	3.60	3.93	3.76
XIII	CH_3	2,4-$(CH_3)_2$	3.40	3.61	3.40	3.57	3.51	3.60	3.60	3.76
XIV	CH_3	2,6-$(CH_3)_2$	3.38	3.67	3.54	3.63	3.45	3.58	3.58	3.76
XV	CH_3	3,4-$(CH_3)_2$	3.49	3.61	3.54	3.61	3.46	3.62	4.05	3.76
XVI	CH_3	3,5-$(CH_3)_2$	3.41	3.67	3.54	3.71	3.49	3.62	4.14	3.76
XVII	CH_3	2,4,5-Cl_3	4.51	4.00	4.88	4.37	4.58	4.14	4.88	4.08
XVIII	CH_3	Cl_5	4.39	4.57	4.45	5.07	4.63	4.79	4.34	4.53
XIX	$(CH_3)_2CH$	H	3.42	3.54	3.30	3.10	3.35	2.85	3.48	3.81
XX	$(CH_3)_2CH$	2-Cl	4.53	3.83	3.28	3.43	3.17	3.29	4.21	4.04
XXI	$(CH_3)_2CH$	4-Cl	3.52	3.76	2.87	3.35	2.84	3.06	4.09	4.04
XXII	$(CH_3)_2CH$	2-OCH_3	3.41	3.54	3.09	2.98	3.08	2.99	3.73	3.80
XXIII	$(CH_3)_2CH$	4-OCH_3	3.59	3.41	3.27	2.86	3.17	2.85	3.63	3.80
XXIV	$(CH_3)_2CH$	2-CH_3	4.39	3.77	3.97	3.21	3.46	3.00	3.99	3.99
XXV	$(CH_3)_2CH$	3-CH_3	3.81	3.77	3.92	3.25	3.66	3.02	3.95	3.99
XXVI	$(CH_3)_2CH$	4-CH_3	4.05	3.71	3.63	3.15	3.40	3.02	3.95	3.99
XXVII	$(CH_3)_2CH$	2-NO_2	3.55	3.43	3.35	3.34	3.13	3.13	3.97	3.72
XXVIII	$(CH_3)_2CH$	4-NO_2	2.56	3.33	2.75	3.23	2.02	2.77	3.76	3.72
XXIX	$(CH_3)_2CH$	4-$C(CH_3)_3$	<2.59*	3.57	<2.59*	2.89	<2.59*	3.44	<2.59*	4.44
XXX	$(CH_3)_2CH$	4-SCH_3	2.69	3.41	2.14	3.27	2.24	3.03	3.92	4.00
XXXI	$(CH_3)_2CH$	2,3-$(CH_3)_2$	4.18	3.99	3.56	3.36	3.18	3.16	4.20	4.17
XXXII	$(CH_3)_2CH$	2,4-$(CH_3)_2$	4.27	3.93	3.07	3.26	2.90	3.16	4.16	4.17
XXXIII	$(CH_3)_2CH$	2,5-$(CH_3)_2$	4.16	3.45	3.45	3.36	3.20	3.16	4.23	4.17
XXXIV	$(CH_3)_2CH$	3,4-$(CH_3)_2$	3.88	3.93	2.93	3.30	2.90	3.18	4.27	4.17
XXXV	$(CH_3)_2CH$	3,5-$(CH_3)_2$	4.25	3.99	3.82	3.40	3.34	3.18	4.37	4.17
XXXVI	$(CH_3)_2CH$	2,3,5-$(CH_3)_3$	4.22	4.22	3.61	3.51	<2.56*	3.34	4.45	4.34
XXXVII	$(CH_3)_2CH$	2,4,5-Cl_3	4.71	4.33	4.76	4.06	4.14	3.71	4.58	4.49
XXXVIII	$(CH_3)_2CH$	Cl_5	4.64	4.90	4.54	4.76	4.15	4.35	4.33	4.94
XXXIX	CH_3CCl_2	H	2.67	2.56	2.74	2.76	2.57	2.57	<2.52*	2.73
XL	CH_3CCl_2	4-Cl	2.60	2.78	2.60	3.01	2.60	2.78	<2.60*	2.95
XLI	CH_3CCl_2	2-Cl	2.60	2.85	2.60	3.08	2.60	3.00	<2.60*	2.95
XLII	CH_3CCl_2	4-NO_2	2.72	2.34	2.93	2.89	2.72	2.49	2.72	2.64
XLIII	CH_3CCl_2	3-NO_2	2.67	2.45	2.78	2.96	2.62	2.49	2.67	2.64
XLIV	CH_3CCl_2	2-NO_2	2.72	2.45	2.93	2.99	2.78	2.92	2.62	2.64
XLV	CH_3CCl_2	4-CH_3	2.86	2.72	2.56	2.80	2.65	2.74	2.55	2.90
XLVI	CH_3CCl_2	3-CH_3	2.86	2.79	2.60	2.91	2.56	2.74	2.56	2.90
XLVII	CH_3CCl_2	2-CH_3	2.86	2.79	2.78	2.87	2.65	2.72	2.65	2.90
XLVIII	CH_3CCl_2	4-OCH_3	<2.59*	2.59	2.59	2.51	2.59	2.74	<2.59*	2.72
XLIX	CH_3CCl_2	2-OCH_3	2.59	2.56	2.59	2.64	2.59	2.71	2.59	2.72
L	CH_3CCl_2	4-$C(CH_3)_3$	<2.65*	2.59	<2.65*	2.54	<2.65*	3.16	<2.65*	3.35
LI	CH_3CCl_2	4-SCH_3	2.63	2.43	2.63	2.57	2.63	2.75	2.63	2.92
LII	CH_3CCl_2	2-NO_2,5-CH_3	2.65	2.67	2.65	3.14	2.87	3.02	<2.65*	2.81
LIII	CH_3CCl_2	3-CH_3,4-Cl	3.03	3.00	2.73	3.16	3.23	2.95	2.61	3.13
LIX	CH_3CCl_2	2-Cl,4-NO_2	3.79	2.62	3.85	3.21	3.39	2.92	3.80	2.86
LV	CH_3CCl_2	2,4-Cl_2	3.67	3.06	3.67	3.33	3.50	3.92	3.50	3.18
LVI	CH_3CCl_2	3-CH_3,4-SCH_3	<2.65*	2.65	<2.65*	2.72	<2.65*	2.92	<2.65*	3.10
LVII	CH_3CCl_2	2,4,5-Cl_3	3.83	3.35	4.43	3.71	3.95	3.42	4.13	3.40
LVIII	CH_3CCl_2	2,4-$(CH_3)_2$	2.74	2.95	2.68	2.91	2.89	2.88	2.59	3.08
LIX	CH_3CCl_2	2,6-$(CH_3)_2$	2.59	3.01	2.59	2.98	2.63	2.86	<2.59*	3.08
LX	CH_3CCl_2	3,4-$(CH_3)_2$	2.81	2.95	<2.59*	2.96	2.81	2.90	<2.59*	3.08
LXI	CH_3CCl_2	3,5-$(CH_3)_2$	<2.59*	3.01	<2.59*	3.06	<2.59*	2.90	<2.59*	3.08
LXII	CH_3CCl_2	2,3-$(CH_3)_2$	2.68	3.01	<2.59*	3.02	2.63	2.88	<2.59*	3.08
Ediphenphos	−	−	4.19	−	4.59	−	3.80	−	3.86	−

[a] Calculated by eq 1. [b] Calculated by eq 2. [c] Calculated by eq 3. [d] Calculated by eq 4. * These values correspond to EC_{50} > 1 mg/mL.

rameter, $\Sigma\sigma$, for each compound was calculated by addition of σ_p and σ_m values, which were also taken from the literature (Hansch and Leo, 1979). For the *ortho* substituent, σ_p along with F', the Swain−Lupton constant parameter for proximity polar effect (Fujita and Nishioka, 1976), was considered. From our detailed study on the steric effect of substituents on the fungicidal activity of *O,O*-bisarayl isopropyl phosphonates, it was clear that the steric effect of the *para* substituent is best modeled by molar refractivity (MR) and the steric effect of the *meta* and *ortho* substituents, which are directional in nature,

Table 3. Development of Equation 1

a	*b*	*c*	*d*	*e*	*n*	*R*	*s*	*F*
$-\log EC_{50} = a + b\sum\pi + c[MR(Z)]^2 + dMR(Z) + e[MR(p)]^2$								
3.055	0.457				57	0.580	0.519	27.93
(±0.193)	(±0.169)							
3.443	0.404	−0.0014			57	0.693	0.464	24.92
(±0.262)	(±0.154)	(±0.0007)						
1.085	0.398	−0.0128	0.304		57	0.843	0.349	43.41
(±0.517)	(±0.116)	(±0.0033)	(±0.092)					
1.923	0.402	−0.0133	0.303	−0.002	57	0.857	0.338	35.89
(±0.506)	(±0.112)	(±0.0033)	(±0.089)	(±0.002)				

Table 4. Development of Equation 2

a	*b*	*c*	*d*	*e*	*n*	*R*	*s*	*F*
$-\log EC_{50} = a + b\sum\pi + c[MR(Z)]^2 + d\sum\sigma + e[MR(p)]^2$								
2.936	0.472				56	0.638	0.468	37.00
(±0.172)	(±0.152)							
3.307	0.411	−0.0014			56	0.738	0.414	31.72
(±0.237)	(±0.137)	(±0.0007)						
3.387	0.320	−0.0016	0.398		56	0.790	0.380	28.82
(±0.221)	(±0.137)	(±0.0006)	(±0.235)					
3.463	0.320	−0.0016	0.415	−0.002	56	0.813	0.364	24.93
(±0.222)	(±0.131)	(±0.0006)	(±0.225)	(±0.002)				

is better modeled by Taft's parameter, E_s (Nidiry and Roy, 1994). Therefore, Taft's parameters were taken into consideration for *ortho* and *meta* substituents, which were designated as E_s(o) and E_s(m), respectively. The MR of the *para* substituent, designated as MR(p), was also taken into consideration. For the alkyl substituent directly attached to the phosphorus atom, the MR, designated as MR(Z), was considered. The square terms of $\sum\pi$, MR(p), and MR(Z) were also taken into consideration. The additive nature of all substituent parameters were presumed. The various parameters were considered for only one phenyl ring because the substitution pattern on both the rings is exactly the same for all the compounds.

Multiple Regression Analysis. The regression analysis was done with a computer software package known as "Statistical Package for Social Sciences", SPSS/PC Varian 2.0 (Anonymous, 1990) that is used at Indian Agricultural Statistics Research Institute, New Delhi, India. The regression was done in a stepwise manner, with the parameter minimizing the sum of squared deviations being introduced step by step and the analysis being stopped when the introduction of the new parameter was no longer statistically significant as evidenced by the Student's *t* test.

Principal Component Analysis. Because PCA starts with a correlation matrix for the fungicidal activity data against the test organisms, it is necessary that all the compounds considered in the analysis have defined EC_{50} values against all the test fungi under consideration. Therefore **13** compounds (i.e., **XXIX**, **XXXVI**, **XXXIX**, **XL**, **XLI**, **XLVIII**, **L**, **LII**, **LVI**, **LIX**, **LX**, **LXI**, and **LXII**) were excluded from PCA. The correlation matrix for the remaining 49 compounds is presented in Table 7. The eigen values and the eigen vectors were calculated by the computer software package SPSS/PC Varian 2.0 (Anonymous, 1990). The loading parameters were obtained by multiplying the eigen vectors by the square roots of the corresponding eigen values. The PC scores (principal properties) of the compounds were calculated in the following way. First, the data matrix (49 × 4) containing the fungicidal activity of the 49 compounds against the four fungi was normalized columnwise. Normalization involves first subtracting the mean activity from every value in the column and then dividing throughout by the standard deviation. This procedure resulted in a mean of 0 and a standard deviation of ±1 for each column. This normalized matrix was post-multiplied by the loading parameters (4 × 1) of PC1 to get the matrix (49 × 1) with the first principal properties (PC1 scores) of the compounds. In a similar way, the second principal properties (PC2 scores) were obtained when the normalized matrix was post-multiplied by the loading parameters of PC2. Multiple regression of the principal properties was done in a stepwise manner exactly the same way as it was done in the case of Hansch analysis.

RESULTS

The results of the multiple regression analysis for the four fungi are given along with the statistical values (n = number of compounds; R = correlation coefficient; s = standard deviation; F = significance index with respect to the equation). The figures in parenthesis are for 95% confidence interval. All terms included in eqs 1−5 are significant at the 95% level on the basis of the Student's *t* test.

Helminthosporium oryzae. Five compounds, **XXIX**, **XLVIII**, **L**, **LVI**, and **LXI**, did not give measurable EC_{50} values (≤1 mg/mL), so these compounds were eliminated from the regression analysis. Equation 1 gives the relationship between the variation in the fungicidal activity of the remaining 57 compounds of the series against *H. oryzae* and their physicochemical parameters:

$$-\log EC_{50} = \underset{(\pm 0.506)}{1.923} + \underset{(\pm 0.112)}{0.402}\sum\pi - \underset{(\pm 0.003)}{0.013}[MR(Z)]^2 + \underset{(\pm 0.089)}{0.303}MR(Z) - \underset{(\pm 0.002)}{0.002}[MR(p)]^2 \quad (1)$$

$$n = 57, \quad R = 0.857, \quad s = 0.338, \quad F = 35.89$$

The stepwise development of eq 1 is presented in Table 3. Experimental values and values calculated by eq 1 are given in Table 2 for comparison. Equation 1 explains 73.4% of the variation in fungicidal activity of the members of the series against *H. oryzae*; 71.1% of the variation is explained by changes in hydrophobicity of the phenyl substituents and steric bulk of the alkyl group modeled by molar refractivity [MR(Z)]. Apart from giving satisfactory statistical values, eq 1 also explains the very low activity of compounds **XLVIII** and **L**, which were excluded from the regression analysis. Equation 1 allows us to calculate the optimum value of MR(Z) as 11.65.

Pyricularia oryzae. Compounds **XXIX**, **L**, **LVI**, **LX**, **LXI**, and **LXII** were excluded from the regression analysis because these compounds did not give measurable EC_{50} values (≤1 mg/mL). Equation 2 gives the relationship between variation in fungicidal activity of the remaining 56 compounds of the series against *P. oryzae*:

$$-\log EC_{50} = \underset{(\pm 0.222)}{3.463} + \underset{(\pm 0.131)}{0.320}\sum\pi - \underset{(\pm 0.0006)}{0.0016}[MR(Z)]^2 + \underset{(\pm 0.225)}{0.415}\sum\sigma - \underset{(\pm 0.002)}{0.002}[MR(p)]^2 \quad (2)$$

$$n = 57, \quad R = 0.813, \quad s = 0.364, \quad F = 24.93$$

Development of eq 2 is presented in Table 4. Equation 2 explains 66.1% of the variaion in activity exhibited by the compounds; 54.5% of the variation is explained by changes in hydrophobicity of the phenyl substituents and steric bulk of the alkyl groups alone. Experimental values and those calculated by eq 2 are presented in Table 2 for comparison.

Alternaria alternata. Compounds **XXIX**, **XXXVI**, **L**, **LVI**, and **LXI**, which did not give measurable EC_{50} values (≤1 mg/mL), were excluded from the regression analysis. Equation 3 gives the relationship between the variation in activity of the remaining 57 compounds and their physicochemical parameters:

$$-\log EC_{50} = \underset{(\pm 0.215)}{3.556} + \underset{(\pm 0.124)}{0.296}\sum\pi - \underset{(\pm 0.012)}{0.047}MR(Z) + \underset{(\pm 0.331)}{0.543}\sum F' \quad (3)$$

$$n = 57, \quad R = 0.851, \quad s = 0.290, \quad F = 46.32$$

Development of eq 3 is presented in Table 5 Equation 3 explains 72.4% of the variation in the activity exhibited by the compounds; 66.9% of the variation is explained by changes in hydrophobicity of the phenyl substituents and steric bulk of the alkyl groups alone. Experimental values and values calculated by eq 3 are given in Table 2 for comparison.

Rhizoctonia bataticola. Compounds **XXIX**, **XXXIX**, **XL**, **XLI**, **XLVIII**, **L**, **LII**, **LVI**, **LIX**, **LX**, **LXI**, and **LXII** were excluded from the regression analysis because they did not give measurable EC_{50} values (≤1 mg/mL). Equation 4 gives the relationship between variation in activity of the remaining 50 compounds and their physicochemical parameters:

$$-\log EC_{50} = \underset{(\pm 0.474)}{1.910} + \underset{(\pm 0.106)}{0.317}\sum\pi - \underset{(\pm 0.0033)}{0.0147}[MR(Z)]^2 + \underset{(\pm 0.084)}{0.347}MR(Z) \quad (4)$$

$$n = 50, \quad R = 0.871, \quad s = 0.314, \quad F = 47.96$$

Development of eq 4 is presented in Table 6. Equation 4 expains 75.8% of the variation exhibited by the compounds, which is entirely accounted for by changes in hydrophobicity of the phenyl substituents and steric bulk of the alkyl groups. Experimental values and values calculated by eq 4 are given in Table 2 for comparison.

Details of the PCA of the fungicidal activity data are presented in Tables 7 and 8. Eigen values of the correlation matrix show that the PC1 has 79.8% of the information content of the data set. PC1 is heavily loaded with all the test organisms and is basically the average of activities over all four species. PC2 is loaded mainly with *A. alternata* and *R. bataticola*. A documentation of the vectors of loadings of the toxicity data of *O,O*-bisaryl alkyl phosphonates is given in Figure 1. The scores (principal properties) corresponding to PC1 and PC2 were calculated and are presented in Table 9.

Table 5. Development of Equation 3

a	*b*	*c*	*d*	*n*	*R*	*s*	*F*
$-\log EC_{50} = a + b\sum\pi + cMR(Z) + 0.543\sum F'$							
2.863 (±0.153)	0.426 (±0.137)			57	0.636	0.418	37.37
3.453 (±0.233)	0.359 (±0.104)	−0.044 (±0.013)		57	0.818	0.314	54.79
3.556 (±0.215)	0.296 (±0.104)	−0.047 (±0.012)	0.543 (±0.331)	57	0.851	0.290	46.32

Table 6. Development of Equation 4

a	*b*	*c*	*d*	*n*	*R*	*s*	*F*
$-\log EC_{50} = a + b\sum\pi + c[MR(Z)]^2 + dMR(Z)$							
3.317 (±0.208)	0.381 (±0.178)			50	0.519	0.534	17.68
3.687 (±0.276)	0.329 (±0.161)	−0.0016 (±0.0008)		50	0.653	0.478	17.44
1.910 (±0.474)	0.317 (±0.106)	−0.0147 (±0.0033)	0.347 (±0.084)	50	0.871	0.314	47.96

Table 7. Correlation Matrix for the Fungicidal Activity of *O,O*-Bisaryl Alkyl Phosphonates

fungi	*H. oryzae*	*P. oryzae*	*A. alternata*	*R. bataticola*
H. oryzae	1.00			
P. oryzae	0.80	1.00		
A. alternata	0.71	0.91	1.00	
R. bataticola	0.84	0.69	0.41	1.00

Table 8. PCA of Fungicidal Activity of *O,O*-Bisaryl Alkyl Phosphonates

Percentage of Variance Explained

PC	eigen values	percent variance explained	sum (%) variance explained
1	3.19	79.8	79.8
2	0.66	16.4	96.2
3	0.14	3.5	99.7
4	0.01	0.3	100.0

PC Loadings

parameter	PC1	PC2
H. oryzae	0.94	−0.20
P. oryzae	0.95	0.21
A. alternata	0.85	0.52
R. bataticola	0.82	−0.55

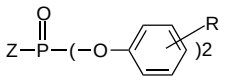
As expected, the scores are clearly orthogonal ($r = -0.01$). To find the physicochemical meaning of PC1 scores, they were subjected to multiple regression analysis in the same way as in the cases of individual organisms. This analysis resulted in the following regression equation:

$$PC1_{sc} = \underset{(\pm 2.640)}{-4.370} + \underset{(\pm 0.651)}{2.066}\sum\pi - \underset{(\pm 0.018)}{0.040}[MR(Z)]^2 + \underset{(\pm 0.470)}{0.849}MR(Z) + \underset{(\pm 2.197)}{2.498}\sum F' - \underset{(\pm 0.010)}{0.012}[MR(p)]^2 \quad (5)$$

$$n = 49, \quad R = 0.866, \quad s = 1.701, \quad F = 25.70$$

Development of eq 5 is presented in Table 10. Equation 5 explains 75% of the variation in the PC1 scores of the compounds; 68.2% of the variation is explained by changes in hydrophobicity of the phenyl substituents and steric bulk of the alkyl group. The scores obtained directly by PCA and those predicted by eq 5 are given in Table 9 for comparison. Equation 5 is quite consistent with the regression equations obtained for individual organisms (eqs 1−4). Equation 5 allows us to calculate the optimum value of MR(Z) as 10.6 and of MR(p) as 0.

Table 9. PC Scores of *O,O*-Bisaryl Alkyl Phosphonates



serial no.	compound no.	Z	R	$PC1_{sc}$	$PC1_{sc}^{Pred}$[a]	$PC2_{sc}$
1	**I**	CH_3	H	−1.10	−0.86	0.47
2	**II**	CH_3	2-Cl	0.18	1.63	0.65
3	**III**	CH_3	4-Cl	0.82	0.18	0.53
4	**IV**	CH_3	2-CH_3	0.18	0.19	0.36
5	**V**	CH_3	3-CH_3	0.32	0.29	0.45
6	**VI**	CH_3	4-CH_3	−0.16	−0.08	0.37
7	**VII**	CH_3	2-OCH_3	−0.71	−0.25	0.28
8	**VIII**	CH_3	4-OCH_3	−0.06	−1.63	0.23
9	**IX**	CH_3	4-SCH_3	−0.82	−1.88	0.68
10	**X**	CH_3	2,4-Cl_2	1.28	2.67	0.32
11	**XI**	CH_3	3-CH_3,4-Cl	1.68	1.37	0.34
12	**XII**	CH_3	2,3-$(CH_3)_2$	1.91	1.35	0.07
13	**XIII**	CH_3	2,4-$(CH_3)_2$	0.16	0.98	0.28
14	**XIV**	CH_3	2,6-$(CH_3)_2$	0.22	1.25	0.29
15	**XV**	CH_3	3,4-$(CH_3)_2$	1.04	1.08	−0.16
16	**XVI**	CH_3	3,5-$(CH_3)_2$	1.09	1.45	−0.19
17	**XVII**	CH_3	2,4,5-Cl_3	7.72	4.12	0.31
18	**XVIII**	CH_3	Cl_5	6.19	8.10	0.74
19	**XIX**	$(CH_3)_2CH$	H	−0.40	−0.63	0.19
20	**XX**	$(CH_3)_2CH$	2-Cl	2.00	1.86	−1.04
21	**XXI**	$(CH_3)_2CH$	4-Cl	−0.93	0.41	−1.06
22	**XXII**	$(CH_3)_2CH$	2-OCH_3	−0.85	−0.02	−0.38
23	**XXIII**	$(CH_3)_2CH$	4-OCH_3	−0.26	−1.41	−0.20
24	**XXIV**	$(CH_3)_2CH$	2-CH_3	3.07	0.42	−0.26
25	**XXV**	$(CH_3)_2CH$	3-CH_3	2.34	0.52	0.15
26	**XXVI**	$(CH_3)_2CH$	4-CH_3	1.84	0.15	−0.28
27	**XXVII**	$(CH_3)_2CH$	2-NO_2	0.19	0.46	−0.50
28	**XXVIII**	$(CH_3)_2CH$	4-NO_2	−4.38	−1.85	−1.28
29	**XXX**	$(CH_3)_2CH$	4-SCH_3	−4.59	0.53	−1.47
30	**XXXI**	$(CH_3)_2CH$	2,3-$(CH_3)_2$	1.91	1.58	−0.80
31	**XXXII**	$(CH_3)_2CH$	2,4-$(CH_3)_2$	0.76	1.21	−1.25
32	**XXXIII**	$(CH_3)_2CH$	2,5-$(CH_3)_2$	1.78	1.58	−0.84
33	**XXXIV**	$(CH_3)_2CH$	3,4-$(CH_3)_2$	0.07	1.31	−1.27
34	**XXXV**	$(CH_3)_2CH$	3,5-$(CH_3)_2$	2.93	1.68	−0.73
35	**XXXVII**	$(CH_3)_2CH$	2,4,5-Cl_3	6.71	4.37	0.04
36	**XXXVIII**	$(CH_3)_2CH$	Cl_5	5.94	8.33	0.23
37	**XLII**	CH_3CCl_2	4-NO_2	−4.12	−5.38	0.37
38	**XLIII**	CH_3CCl_2	3-NO_2	−4.67	−4.77	0.28
39	**XLIV**	CH_3CCl_2	2-NO_2	−4.15	−3.07	0.52
40	**XLV**	CH_3CCl_2	4-CH_3	−4.84	−3.37	0.27
41	**XLVI**	CH_3CCl_2	3-CH_3	−4.91	−3.00	0.20
42	**XLVII**	CH_3CCl_2	2-CH_3	−4.34	−3.10	0.26
43	**XLIX**	CH_3CCl_2	2-OCH_3	−5.26	−3.55	0.28
44	**LI**	CH_3CCl_2	4-SCH_3	−5.01	−5.18	0.28
45	**LIII**	CH_3CCl_2	3-CH_3,4-Cl	−3.29	−1.96	0.80
46	**LIV**	CH_3CCl_2	2-Cl,4-NO_2	1.57	−2.89	0.00
47	**LV**	CH_3CCl_2	2,4-Cl_2	0.86	−0.62	0.37
48	**LVII**	CH_3CCl_2	2,4,5-Cl_3	3.91	0.84	0.45
49	**LVIII**	CH_3CCl_2	2,4-$(CH_3)_2$	−4.39	−2.32	0.56

[a] Predicted by eq 5.

The *F* values of eqs 1−5 show significance at the 1% level (Snedecor and Cochran, 1968).

DISCUSSION

Equation 5 clearly gives the strategies to be adopted in the development of broad-spectrum fungicides of the series. These strategies are: (1) optimum bulk of the alkyl substituent attached to phosophorus; (2) high hydrophobicity of the phenyl substituents; (3) inductive electron withdrawing group at the *ortho* position; and (4) absence of any substitution at the *para* position of the phenyl rings.

In a recent study of the steric effects of phenyl substituents of *O,O*-bisaryl isopropyl phosphonates, we concluded that the molecules interact with the receptor through the *para* position of the phenyl ring (Nidiry and Roy, 1994). The present study shows that the steric bulk of alkyl groups attached to phosphorus has a significant role in the mode of action. In view of this observation, the receptor diagram depicted earlier in the case of *O,O*-bisaryl isopropyl phosphonates was modified

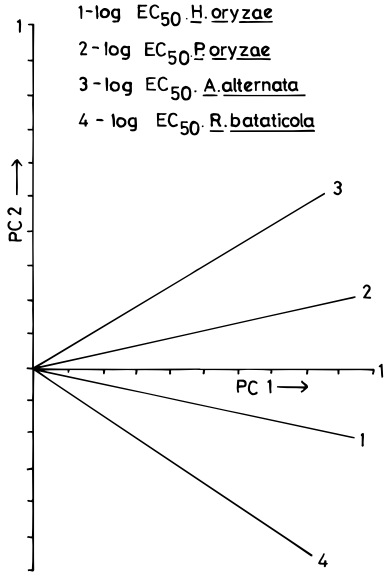


Figure 1. Documentation of the vectors of loadings of the toxicity data of *O,O*-bisaryl alkyl phosphonates to the four phytopathogenic fungi.

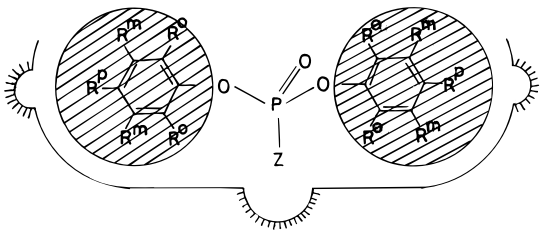


Figure 2. Receptor diagram of *O,O*-bisaryl alkyl phosphonates. Substituents at the *ortho, meta,* and *para* positions of the phenyl rings are designated as R^o, R^m, and R^p, respectively, and the alkyl substituent is designated as Z. The shaded area shows hydrophobic sites that facilitate binding and transport to the bioreceptor. Lines with fringes represent three-dimensional steric interaction sites of the receptor to which the molecules must fit.

Table 10. Development of Equation 5

a	*b*	*c*	*d*	*e*	*f*	*n*	*R*	*s*	*F*
$PC1_{sc} = a + b\Sigma\pi + c[MR(Z)]^2 + d MR(Z) + e\Sigma F' + f[MR(p)]^2$									
−2.131	2.617					49	0.685	2.366	41.64
(±0.925)	(±0.796)								
−0.358	2.359	−0.007				49	0.779	2.060	35.46
(±1.188)	(±0.704)	(±0.004)							
−4.824	2.360	−0.040	0.869			49	0.826	1.810	32.28
(±2.873)	(±0.639)	(±0.020)	(±0.517)						
−4.792	1.989	−0.041	0.874	2.836		49	0.849	1.774	28.39
(±2.726)	(±0.674)	(±0.019)	(±0.492)	(±2.272)					
−4.370	2.066	−0.040	0.849	2.498	−0.012	49	0.866	1.701	25.70
(±2.640)	(±0.651)	(±0.018)	(±0.470)	(±2.197)	(±0.010)				

as depicted in Figure 2. Our earlier conclusion that the permeation and transport to the site of action is largely governed by hydrophobicity of the phenyl substituents remains valid in light of the present study also. However, once the molecules reach the site of action, it appears that the interaction of the molecules with the bioreceptor is a three-point one as opposed to the two-point attachment depicted earlier. Phosphonates possess a tetrahedral structure corresponding to the sp^3 hybridization of the central phosphorus atom (Fest and Schmidt, 1982). This structure means that the carbon atom of the alkyl group and the three oxygen atoms attached to the phosphorus atom should be occupying the four corners of a tetrahedron. The molecules interact with the receptor not only through the *para* position of the phenyl ring but also through the alkyl group (Figure 2). The fitting of the molecules into the bioreceptor through the alkyl group and the *para* substituents appears to be the crucial step in the mode of action.

Preliminary phytotoxicity studies on two selected compounds, **XVII** and **XXXVII**, that showed remarkably high fungicidal activity indicated that the compounds are not toxic to the germination of rice seeds at a concentration of 0.5 mg/mL (Vasu and Roy, 1985; Nidiry and Roy, 1993). Residue and metabolism studies on a closely related compound, namely *O*,*O*-diphenyl dichloromethyl phosphonate, on rice plant indicated that the compound is degradable and is unlikely to leave toxic metabolites (Bedi and Roy, 1978; Bedi and Roy, 1980). However, more extensive studies on the toxicity of the compounds to animals and environment are required before commercialization of the compounds is possible.

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