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Communication

QSARs of some novel isosteric heterocyclics with antifungal activity★✪

İsmail Yalçìn✉, İlkay Ören, Özlem Temiz and Esin Akì Şener

Ankara University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, TR-06100 Tandogan Ankara-Turkey

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QSAR analysis of a set of previously synthesized 2,5,6-trisubstituted benzoxazole, benzimidazole and 2-substituted oxazolo(4,5-b)pyridine derivatives tested for growth inhibitory activity against *Candida albicans,* was performed by using the computer-assisted multiple regression procedure. The activity contributions for either heterocyclic ring systems or substituent effects of these compounds were determined from the correlation equation and the predictions for the lead optimization were described. The resulting QSAR revealed that the oxazolo(4,5-b)pyridine ring system with the substitution of a benzyl moiety at position 2 was the most favourable structure among the heterocyclic nuclei. Moreover, the fifth position in the fused ring system is found more significant than the other positions in improving the activity.

Most fungi are completely resistant to the action of antimicrobial drugs. Only a few substances have been discovered which exert an inhibitory effect on the fungi pathogenic for man, and most of these are relatively toxic (Meyers *et al.*, 1976).

Consequently, spurred by the need of new antifungal agents and the fact that many effective antimicrobial drugs possess heterocyclic systems in their structure, we synthesized some novel derivatives of benzoxazoles, benzimidazoles, oxazolo(4,5-b)pyridines and

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✉To whom correspondence should be addressed.

Abbreviations: MIC, minimum inhibitory concentration; MVC, multi variable analysis.

benzothiazoles during the last few years (Yalçın *et al.*, 1990; 1992; Ören, 1995; Ören *et al.*, 1998; Mrozek *et al.*, 1999).

The synthesized compounds **1–61** used in this QSAR analysis indicated antifungal activity of MIC (minimum inhibitory concentration) values between 6.25–50 μg/ml against *Candida albicans*. Clotrimazole, oxiconazole and haloprogin were also used as standard drugs in order to compare the antifungal activity of these compounds. The observed data for the antifungal activity of the synthesized compounds and the standard drugs were found as given below (Ören, 1995):

Synthesized Compounds:	*Observed MIC values*
1–61	6.25–50 μg/ml
Standard Drugs:	*Observed MIC values*
Clotrimazole	6.25 μg/ml
Oxiconazole	6.25 μg/ml
Haloprogin	3.12 μg/ml

In this study, QSAR analysis of some novel antifungal benzoxazoles, benzimidazoles and oxazolo(4,5-b)pyridines **1–61** (Fig. 1) was performed by the MVA (multi variable analysis) technique.

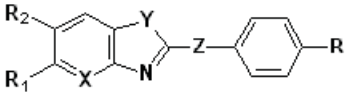


Figure 1.

X: =CH–, =N–

Y: –O–, –NH–

Z: –, $-CH_2-$, $-C_2H_4-$, $-CH_2O-$, $-CH_2S-$

R: –H, –Cl, –Br, –F, $-NO_2$, $-NH_2$, $-CH_3$, $-C(CH_3)_3$, $-C_2H_5$, $-OCH_3$, $-NHCH_3$, $-NHCOCH_3$,

R_1: –H, –Cl, $-NO_2$, $-NH_2$, $-COOCH_3$

R_2: –H, $-NO_2$

EXPERIMENTAL SECTION

The MVA approach in QSAR analysis has been most widely and effectively used for theoretical drug design due to various physicochemical (electronic, steric and hydrophobic) parameters and structural indicator parameters used together (Hansch & Fujita, 1964; 1969).

The assumption can be formulated as given in Eqn. 1:

$$\log 1/C = \Sigma a_i I_i + \Sigma b_j X_j + c \qquad \text{Eqn. 1}$$

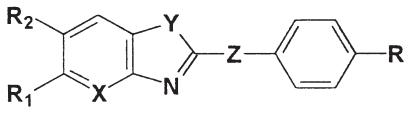
where I_i is the structural indicator parameter and X_j is the physicochemical variable.

In this study, the model is based on the *in vitro* activity of certain 2,5,6-trisubstituted benzoxazole, benzimidazole and 2-substituted oxazolo(4,5-b)pyridine derivatives **1–61** against *C. albicans*, where C is the MIC value expressed in molar concentration units (Table 1).

The variables used as descriptors in the analysis are electronic, steric and structural parameters. The structural indicator variable Ix expresses the replacement of –CH= by the isosteric group –N= in the six membered ring of the fused ring system. Ix is defined as 1 for –N= and 0 for –CH= in the compounds. The other indicator variable Iz has a value of 1 for the presence of a methylene group and 0 for its absence between the *p*-substituted phenyl moiety and the fused ring system in position 2. The indicator variable IR_2 has a value of 1 for a nitro group and 0 for a hydrogen atom at position 6 in the fused ring system (see Table 2).

Physicochemical parameters taken into consideration in QSAR study are *F* (field effect) as electronic influences and Verloop's STERIMOL parameters (L and B_4) for the steric interactions of the substituents R and R_1 (Hansch & Leo, 1979). L is defined as the length of a substituent along the axis of its substitution to the parent skeleton. B4 is defined as the maximum width of the substituent and might provide a better understanding of steric requirements in ligand-receptor interactions. Electronic effect of the

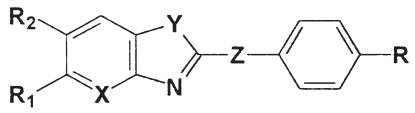
Table 1. The structure and *in vitro* antifungal activity of the analyzed compounds 1–61 against *C. albicans*



Comp. No:	X	Y	Z	R	R_1	R_2	MIC[a] µg/ml	C[b]
1	CH	O	-	H	H	H	25	$1.28x10^{-4}$
2	CH	O	-	$C(CH_3)_3$	H	H	25	$9.96x10^{-5}$
3	CH	O	-	NH_2	H	H	25	$1.19x10^{-4}$
4	CH	O	-	$NHCH_3$	H	H	25	$1.116x10^{-4}$
5	CH	O	-	C_2H_5	Cl	H	25	$9.71x10^{-5}$
6	CH	O	-	$NHCOCH_3$	Cl	H	25	$8.73x10^{-5}$
7	CH	O	-	$NHCH_3$	Cl	H	25	$9.67x10^{-5}$
8	CH	O	-	Cl	Cl	H	25	$9.47x10^{-5}$
9	CH	O	-	NO_2	Cl	H	25	$9.11x10^{-5}$
10	CH	O	-	H	NO_2	H	12.5	$5.23x10^{-5}$
11	CH	O	-	CH_3	NO_2	H	12.5	$4.92x10^{-5}$
12	CH	O	-	$C(CH_3)_3$	NO_2	H	12.5	$4.22x10^{-5}$
13	CH	O	-	NH_2	NO_2	H	12.5	$4.9x10^{-5}$
14	CH	O	-	Cl	NO_2	H	12.5	$4.55x10^{-5}$
15	CH	O	-	Br	NO_2	H	12.5	$3.93x10^{-5}$
16	CH	O	-	C_2H_5	NH_2	H	25	$1.05x10^{-4}$
17	CH	O	-	F	NH_2	H	25	$1.097x10^{-4}$
18	CH	O	-	$N(CH_3)_2$	NH_2	H	25	$9.88x10^{-5}$
19	CH	O	-	CH_3	CH_3	H	25	$1.12x10^{-4}$
20	CH	O	-	C_2H_5	CH_3	H	25	$1.055x10^{-4}$
21	CH	O	-	OCH_3	CH_3	H	25	$1.046x10^{-4}$
22	CH	O	-	F	CH_3	H	25	$1.101x10^{-4}$
23	CH	O	-	$NHCOCH_3$	CH_3	H	25	$9.398x10^{-5}$
24	CH	O	-	$NHCH_3$	CH_3	H	25	$1.05x10^{-4}$
25	CH	O	-	$N(CH_3)_2$	CH_3	H	25	$9.92x10^{-5}$
26	N	O	-	CH_3	H	H	12.5	$5.95x10^{-5}$
27	N	O	-	C_2H_5	H	H	12.5	$5.58x10^{-5}$
28	N	O	-	OCH_3	H	H	12.5	$5.53x10^{-5}$
29	N	O	-	OC_2H_5	H	H	12.5	$5.208x10^{-5}$
30	N	O	-	NH_2	H	H	12.5	$5.924x10^{-5}$
31	N	O	-	NO_2	H	H	12.5	$5.187x10^{-5}$
32	CH	O	CH_2	H	H	H	12.5	$5.98x10^{-5}$
33	CH	O	CH_2	OCH_3	H	H	12.5	$5.23x10^{-5}$
34	CH	O	CH_2	Cl	H	H	12.5	$5.133x10^{-5}$
35	CH	O	CH_2	NO_2	H	H	12.5	$4.92x10^{-5}$
36	CH	O	CH_2	H	Cl	H	12.5	$5.13x10^{-5}$
37	CH	O	CH_2	OCH_3	Cl	H	12.5	$4.57x10^{-5}$
38	CH	O	CH_2	Br	Cl	H	12.5	$3.89x10^{-5}$
39	CH	O	CH_2	NO_2	Cl	H	12.5	$4.33x10^{-5}$
40	CH	O	CH_2	H	NO_2	H	6.25	$2.46x10^{-5}$
41	CH	O	CH_2	OCH_3	NO_2	H	6.25	$2.2x10^{-5}$
42	CH	O	CH_2	Br	NO_2	H	6.25	$1.88x10^{-5}$
43	CH	O	CH_2	Cl	NO_2	H	6.25	$2.166x10^{-5}$
44	CH	O	CH_2	NO_2	NO_2	H	6.25	$2.09x10^{-5}$
45	CH	O	CH_2O	H	CH_3	H	25	$1.046x10^{-4}$
46	CH	O	CH_2O	H	H	NO_2	50	$1.85x10^{-4}$
47	CH	O	CH_2O	H	Cl	NO_2	50	$1.64x10^{-4}$
48	CH	O	CH_2O	Cl	Cl	NO_2	50	$1.475x10^{-4}$
49	CH	O	CH_2S	H	NO_2	H	12.5	$4.37x10^{-5}$
50	CH	O	CH_2S	H	CH_3	H	25	$9.795x10^{-5}$
51	N	O	CH_2O	H	H	H	12.5	$5.495x10^{-5}$
52	N	O	CH_2O	Cl	H	H	12.5	$4.797x10^{-5}$
53	CH	NH	CH_2O	Cl	CH_3	H	25	$9.18x10^{-5}$
54	CH	NH	CH_2S	H	NO_2	H	12.5	$4.385x10^{-5}$
55	CH	NH	CH_2S	H	CH_3	H	25	$9.795x10^{-5}$
56	CH	O	CH_2O	H	$COOCH_3$	H	25	$8.83x10^{-5}$
57	CH	O	CH_2O	Cl	$COOCH_3$	H	25	$7.87x10^{-5}$
58	CH	NH	CH_2O	Cl	$COOCH_3$	H	25	$7.907x10^{-5}$
59	CH	NH	CH_2S	H	$COOCH_3$	H	25	$8.39x10^{-5}$
60	CH	O	C_2H_4	H	NO_2	H	12.5	$4.667x10^{-5}$
61	N	O	C_2H_4	H	H	H	12.5	$5.585x10^{-5}$

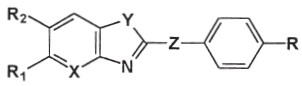
[a]minimum inhibition concentration, [b]MIC value expressed in molar concentration

Table 2. Parameters used in the best fitted equation



Comp. No:	Ix	Iz	I_{R2}	F_{R1}	$B4_{R1}$	L_{R1}	Observed log 1/C	Calculated log 1/C	Residuals
1	0	0	0	0	1	2.06	3.892	3.951	-0.059
2	0	0	0	0	1	2.06	4.001	3.951	0.050
3	0	0	0	0	1	2.06	3.924	3.951	-0.027
4	0	0	0	0	1	2.06	3.952	3.951	0.001
5	0	0	0	0.41	1.8	3.52	4.013	4.026	-0.013
6	0	0	0	0.41	1.8	3.52	4.059	4.026	0.033
7	0	0	0	0.41	1.8	3.52	4.015	4.026	-0.011
8	0	0	0	0.41	1.8	3.52	4.024	4.026	-0.002
9	0	0	0	0.41	1.8	3.52	4.040	4.026	0.014
10	0	0	0	0.67	2.44	3.44	4.282	4.341	-0.059
11	0	0	0	0.67	2.44	3.44	4.308	4.341	-0.033
12	0	0	0	0.67	2.44	3.44	4.375	4.341	0.034
13	0	0	0	0.67	2.44	3.44	4.310	4.341	-0.031
14	0	0	0	0.67	2.44	3.44	4.342	4.341	0.001
15	0	0	0	0.67	2.44	3.44	4.406	4.341	0.065
16	0	0	0	0.02	1.84	2.93	3.979	3.981	-0.002
17	0	0	0	0.02	1.84	2.93	3..960	3.981	-0.021
18	0	0	0	0.02	1.84	2.93	4.005	3.981	0.024
19	0	0	0	-0.04	2.04	3.0	3.950	3.988	-0.038
20	0	0	0	-0.04	2.04	3.0	3.977	3.988	-0.011
21	0	0	0	-0.04	2.04	3.0	3.980	3.988	-0.008
22	0	0	0	-0.04	2.04	3.0	3.958	3.988	-0.030
23	0	0	0	-0.04	2.04	3.0	4.027	3.988	0.039
24	0	0	0	-0.04	2.04	3.0	3.979	3.988	-0.009
25	0	0	0	-0.04	2.04	3.0	4.004	3.988	0.016
26	1	0	0	0	1	2.06	4.225	4.262	-0.037
27	1	0	0	0	1	2.06	4.253	4.262	-0.009
28	1	0	0	0	1	2.06	4.257	4.262	-0.005
29	1	0	0	0	1	2.06	4.283	4.262	0.021
30	1	0	0	0	1	2.06	4.227	4.262	-0.035
31	1	0	0	0	1	2.06	4.285	4.262	0.023
32	0	1	0	0	1	2.06	4.223	4.276	-0.053
33	0	1	0	0	1	2.06	4.282	4.276	0.006
34	0	1	0	0	1	2.06	4.290	4.276	0.014
35	0	1	0	0	1	2.06	4.308	4.276	0.032
36	0	1	0	0.41	1.8	3.52	4.290	4.351	-0.061
37	0	1	0	0.41	1.8	3.52	4.340	4.351	-0.011
38	0	1	0	0.41	1.8	3.52	4.410	4.351	0.059
39	0	1	0	0.41	1.8	3.52	4.363	4.351	0.012
40	0	1	0	0.67	2.44	3.44	4.609	4.667	-0.058
41	0	1	0	0.67	2.44	3.44	4.657	4.667	-0.010
42	0	1	0	0.67	2.44	3.44	4.725	4.667	0.058
43	0	1	0	0.67	2.44	3.44	4.664	4.667	-0.003
44	0	1	0	0.67	2.44	3.44	4.680	4.667	0.013
45	0	0	0	-0.04	2.04	3.0	3.980	3.988	-0.008
46	0	0	1	0	1	2.06	3.732	3.733	-0.001
47	0	0	1	0.41	1.8	3.52	3.785	3.808	-0.023
48	0	0	1	0.41	1.8	3.52	3.831	3.808	0.023
49	0	0	0	0.67	2.44	3.44	4.359	4.341	0.018
50	0	0	0	-0.04	2.04	3.0	4.009	3.988	0.021
51	1	0	0	0	1	2.06	4.260	4.262	-0.002
52	1	0	0	0	1	2.06	4.319	4.262	0.057
53	0	0	0	-0.04	2.04	3.0	4.037	3.988	0.049
54	0	0	0	0.67	2.44	3.44	4.358	4.341	0.017
55	0	0	0	-0.04	2.04	3.0	4.009	3.988	0.021
56	0	0	0	0.33	3.36	4.85	4.054	4.090	-0.036
57	0	0	0	0.33	3.36	4.85	4.104	4.090	0.014
58	0	0	0	0.33	3.36	4.85	4.102	4.090	0.012
59	0	0	0	0.33	3.36	4.85	4.076	4.090	-0.014
60	0	0	0	0.67	2.44	3.44	4.331	4.341	-0.010
61	1	0	0	0	1	2.06	4.253	4.262	-0.009

Table 3. Predicted log 1/C values of some additional analog compounds (62–68) compared with their observed values by using the Eqn. 2



Comp. No:	X	Y	Z	R	R_1	R_2	MIC μg/ml	Observed log1/C	Predicted Log1/C	Residuals
62	CH	O	-	Br	NH_2	H	25	4.110	3.972	0.138
63	CH	O	CH_2	Br	H	H	12.5	4.360	4.273	0.087
64	CH	O	CH_2O	Cl	H	H	25	4.016	3.953	0.063
65	CH	NH	CH_2O	H	NO_2	H	12.5	4.283	4.339	0.054
66	CH	NH	CH_2O	Cl	H	H	25	4.015	3.953	0.062
67	CH	NH	CH_2S	H	Cl	H	25	4.041	4.026	0.015
68	CH	NH	C_2H_4	H	H	H	25	4.078	4.087	0.089

substituents, expressed in term of *F*, is found to be important in determining the activity, as it is predictive in electrophilic reactions of the drugs with the nucleophilic functions of biomolecules.

The correlation equation was performed using the Stepwise Technique by entering and removing the F level as 4.0 for each variable in the regression.

The overall F test value for the calculated best equation (Eqn. 2) was found as 471.69 which is statistically significant at the level of probability $P < 0.001$ (Fig. 2), whereas the tabulated F value is 4.442.

On the other hand, the predictive power of the performed QSAR model was also examined by calculating the q^2 (predicted r^2) value of some additional synthesized analog compounds (**62–68**) and the Eqn. 2 was found significant for the lead optimization in this set of compounds.

RESULTS AND DISCUSSION

The best equation observed in this QSAR study is:

$$\log 1/C = 0.31(\pm 0.02)I_x + 0.33(\pm 0.01)I_z + 0.49(\pm 0.02)F_{R1} + 0.26(\pm 0.02)B4_{R1} - 0.23(\pm 0.02)L_{R1} - 0.22(\pm 0.02)I_{R2} + 4.16\ (\pm 0.03) \qquad \text{Eqn. 2}$$

$$n = 61\ R^2 = 0.98 \quad s = 0.03 \quad F = 471.69; \quad P < 0.001$$

For the observation of the predictive power of the developed QSAR model, predicted log 1/C values of some additional analog compounds (**62–68**) were also calculated by using the correlation Eqn. 2 and compared with the observed values (Table 3). Calculated q^2 and S_{press}/SSY values determine that the predictive power of this QSAR model is significant (Wold, 1991) as given below:

$$q^2 = 0.67; \quad S_{press} = 0.65; \quad S_{press}/SSY = 0.22 < 0.4$$

Predictions for the lead optimization in this set of compounds can be concluded as follows.

Due to the activity contribution of every positive 0.3 value of regression coefficients of the structural indicator parameters Ix, Iz and IR_2 in the developed QSAR model, Eqn. 2 increases the antifungal activity as two fold improved potency. Holding a pyridine ring in the

bicyclic system was found important for the heterocyclic fused system, providing two fold improved potency against *C. albicans*. Moreover, substituting position Z with a methylene group as a bridge element between the fused heterocyclic ring system and phenyl ring in this set of molecules also offered two-fold higher potency for the antifungal activity. However, having a nitro group at position R_2 in the bicyclic nucleus led to a nearly two-fold decrease in potency.

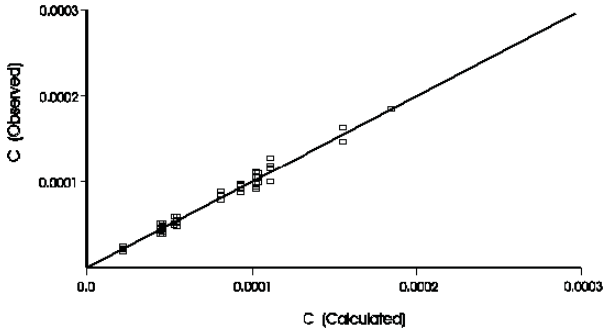


Figure 2. Plot of the observed and calculated C values of the growth inhibitory activity of compounds 1–61 against *C. albicans* using Eqn. 2.

On the other hand, position R_1 was found more significant than the positions R and R_2 for the screened antifungal activity. On the basis of Eqn. 2, electronic and steric properties of the substituents on position R_1 were found to be more indicative than the hydrophobic effects. The developed QSAR model revealed that substituting position R_1 with a group possessing a positive field effect (*F*), concerning minimum length (L) and maximum width (B4), increases the antifungal activity. Consequently, the interpretation of numerical values of weights of these physicochemical descriptors obtained in Eqn. 2 indicates that substituting position R_1 with such groups as NH_3^+ (F = 0.94, L = 2.93, B4 = 1.84) or NO_2 (F = 0.67, L = 3.44, B4 = 2.44) instead of CH_3 (F = –0.04, L = 3.00, B4 = 1.90), Cl (F = 0.41, L = 3.52, B4 = 1.80), Br (F = 0.44, L = 3.83, B4 = 1.95), $COOCH_3$ (F = 0.33, L = 4.85, B4 = 3.36), NH_2 (F = 0.02, L = 2.93, B4 = 1.84), CN (F = 0.51, L = 4.23, B4 = 1.60) and/or H (F = 0.00, L = 2.06, B4 = 1.00), enhances the antifungal activity against the screened microorganism.

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