Turk J Med Sci
31 (2001) 493-497

Özlem TEMİZ ARPACI

QSARs of Some 5- or 6-Methyl-2-Substituted Benzoxazoles/Benzimidazoles against *Candida albicans*

Received: November 07, 2000

Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Ankara University, Tandoğan 06100, Ankara - TURKEY

Abstract: QSAR analysis of some 5- or 6-methyl-2-substituted benzoxazoles/benzimidazoles was studied for the antifungal activity against *C. albicans* using Hansch analysis. Prediction for the lead optimization in this QSAR analysis was described by the description of various hydrophobic, electronic, steric and structural parameters related to positions R_1, R_2, R_5, R_6, X, Y. The cross-validation method was also applied to the data set in order to prove the predictive power by using the BILIN statistical software. The resulting QSAR revealed that substitution at position Y with the CH_2 group was significant for the improved antifungal activity. Moreover, hydrophobic properties of the substituents at position R_2 are indicative for the antifungal activity against *C. albicans*.

Key Words: Benzoxazole, Benzimidazole, Antifungal activity, QSAR, Hansch analysis

Introduction

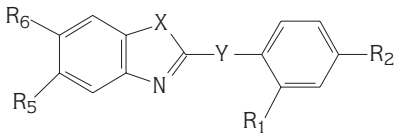
Mycotic illnesses in humans are divided into three groups: contagious skin and hair infections, noncontagious soilborne or airborne systemic infections and noncontagious foodborne toxemias. The responsible organisms and methods of prevention and treatment differ with each group. The prevalence of systemic fungal infections has increased significantly during the past decade. This increase is due to greater use of broad-spectrum antibiotics, immunosuppressive agents, central venous catheters, intensive care low birth weight infants, organ transplantation and the acquired immunodeficiency syndrome (AIDS) epidemic (1-13).

Significant antifungal chemotherapy began in 1903, with the successful use of potassium iodide for the treatment of sporotrichosis. Except for the development of flucytosine, there was little progress until the early 1970s and the development of the azole drugs. The current era, which is characterized largely by the modifications of azole drugs, began with miconazole and ketoconazole and brought the agents fluconazole and itraconazole and ravuconazole, which can be given orally and have increased potency, decreased toxicity and a broader spectrum of activity (14-19).

In the past 10 years there has been a major expansion in the development of antifungal drugs, but there are still weaknesses in the range and scope of current antifungal chemotherapy (5). New developments have included the modification of existing drug molecules to eliminate toxicity and improve activity. In short, antifungal therapy has advanced rapidly in the last few years compared to previous years and recommendations for treatment of fungal infections are likely to change in the near future as our understanding of fungal infections improves and new antifungal therapies are discovered.

Biologically active benzoxazoles have been known for a long time and it was seen that position 2 is decisive for the biological activity, whereas position 5 determines the intensity of their activity (20-22).

Recently, we reported the synthesis and in vitro antifungal activities of different 5- or 6-methyl-2-substitutedbenzoxazoles/benzimidazoles against the fungus *Candida albicans* (23-25) (Figure 1).



R_1=H, Cl, F, NO_2, CH_3, OCH_3
R_2= H, Cl, Br, NH_2, CH_3, OCH_3
R_5=H, CH_3
R_6= H, CH_3

Y= —, CH_2, CH_2O, CH_2S
X= O, NH

Figure 1. Antifungal active some benzoxazole and benzimidazole derivatives against *C. albicans*.

An extrathermodynamic approach in the analysis of quantitative structure activity relationships (QSAR) has been most widely and effectively used for theoretical drug design. This method has also been called the Hansch approach and it assumes that the potency of a certain biological activity exerted by a series of congeneric compounds can be expressed in terms of a function of various physicochemical (electronic, steric and hydrophobic) effects (26-29). This assumption is summarized in an equation as follows:

f (biological activity) =f (electronic) +f (steric) +f (hydrophobic) +[f (structural) +f (therotical)]

If these functions could be formulated in an equation showing certain effects favorable for the activity, structural modifications that enhance such properties would be expected to generate potent active compounds.

Multiple regression analysis involving finding the best fit of a dependent variable (microbiological activity) to a linear combination of independent variables (descriptors) by the least squares method was used (30). This is formally expressed as follows:

$y=a_0 +a_1x_1 +a_2x_2 +a_3x_3 + ... + a_nx_n$

where y is related to the microbiological activity of a compound, x_1, x_2, ... x_n are the descriptor values related to the activity and a_0, a_1, a_2, ... a_n are the regression coefficients determined by the least square analysis. This equation is developed for each compound in our QSAR study.

In this present study, QSAR analysis of some antifungal active 5- or 6-methyl-2-substitutedbenzoxazoles/benzimidazoles, 1-25 given in Figure 1, against *C. albicans* were determined using Hansch analysis.

Experimental

Multivariate Hansch analysis in QSAR has been most widely and effectively used for theoretical drug design due to various physicochemical (electronic, steric and hydrophobic) parameters and structural indicator parameters being used together. The assumption can be formulated as given in the equation below.

$log1/C= \Sigma a_iI_i + \Sigma b_jX_j + c$

where I_i is the structural indicator parameters and X_j is the physicochemical variables.

For the procedure of descriptor selection related to the activity among the candidate set of variables, forward step-wise multiple regression of elimination was applied. During the development of the best fit model of correlation equation, the minimum F value for entering and removing the variables in the step-wise multiple regression was taken to be 8.0, which is statistically significant at the 1% level of probability (31).

In order to judge the validity of the predictive power of the QSAR, the cross-validation method is also applied to the original data set by removing a group of compounds from the data in such a way that each observation (compound) is deleted once and once only. For each reduced data set, a model is developed and the response values of the deleted observations are predicted from this model. The cross-validation method was also applied to the data set in order to judge the validity of predictive power of the model and overall PRESS (Predictive Residual Sum of Square) values were calculated (32).

Regression analysis and calculations were run on a PC using the BILIN statistical program package (33,34). In the equations, the figures in parentheses are the standard errors of the regression coefficients. For a given equation, n is number of compounds, r^2 denotes square of the multiple correlation coefficients, F is the significance test and s represents the residual standard deviation.

A congeneric set of 5- or 6-methyl-2-substitutedbenzoxazoles/benzimidazoles 1-25 were considered in this study. The candidate set of variables used in this analysis includes hydrophobic, electronic, steric and structural parameters. For the structural variables I_{R6} was defined as 0 for possesing H, 1 for possesing CH_3 at position R_6; I_{R5} was defined as 0 for possesing H, 1 for possesing CH_3 at position R_5; IyO was defined as 1 for possesing O atom; $IyCH_2$ was defined as 1 for possesing CH_2 group; $IyCH_2O$ was defined as 1 for possesing CH_2O group; IyS was defined as 1 for possesing CH_2S group at position Y and Ix was defined as 1 for possesing O atom, 0 for NH atoms at position X. The hydrogen donating/accepting capabilities (H_{DONOR}/H_{ACCEPT}) of the substituents at R_1, R_2, R_5, R_6 were the indicator variables.

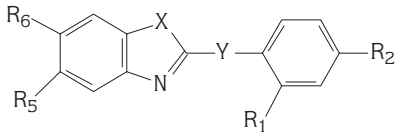
The screened physicochemical parameters in this QSAR study were π for the hydrophobic effects, F (field

effect), R (resonance effect) and σ as the electronic influence and MR, MW, Es, Es-V, Verloop's STERIMOL parameters (L, B_1, B_4) for the steric interactions of the substituents R_1, R_2, R_5 and R_6. Values for all candidate physicochemical variables used in this QSAR study were taken from the table of Hansch et al. (14). The values of the descriptors related to the activity among the candidate test set of variables of the best results in the QSAR analysis are shown in the Table.

Results and Discussion

Predictions for the lead optimization in this QSAR analysis of antifungal active 5- or 6-methyl benzoxazoles/benzimidazoles were described by the description of various hydrophobic, electronic, steric and structural parameters related to positions R_1, R_2, R_5, R_6, X, Y. One leave out cross-validation method was also applied to the data set in order to prove the predictive power by using the BILIN statistical software. Compounds

Table. Compounds and the parameters used in the best equation.



Com. No:	R_1	R_2	R_5	R_6	X	Y	πR_2	$IyCH_2$	MIC µg/mL	Obs. log1/C	Cal. log1/C	Residual
1	Cl	H	CH_3	H	O	—	0	0	25	3.989	3.996	-0.007
2	OCH_3	H	CH_3	H	O	—	0	0	25	3.980	3.996	-0.015
3	NO_2	H	CH_3	H	O	—	0	0	25	4.007	3.996	0.011
4	Cl	Cl	CH_3	H	O	—	0.71	0	25	4.046	4.024	0.023
5	CHH_3	CH_3	CH_3	H	O	—	0.56	0	25	3.977	4.018	-0.041
6	OCH_3	OCH_3	CH_3	H	O	—	-0.02	0	25	4.032	3.995	0.037
7	Cl	H	H	CH_3	O	—	0	0	25	3.989	3.996	-0.007
8	OCH_3	H	H	CH_3	O	—	0	0	25	3.980	3.996	-0.015
9	F	H	H	CH_3	O	—	0	0	25	3.958	3.996	-0.038
10	NO_2	H	H	CH_3	O	—	0	0	25	4.007	3.996	0.011
11	Cl	Cl	H	CH_3	O	—	0.71	0	25	4.046	4.024	0.023
12	CH_3	CH_3	H	CH_3	O	—	0.56	0	25	3.977	4.018	-0.041
13	OCH_3	OCH_3	H	CH_3	O	—	-0.02	0	25	4.032	3.995	0.037
14	H	H	CH_3	H	O	CH_2	0	1	12.5	4.251	4.298	-0.047
15	H	Br	CH_3	H	O	CH_2	0.86	1	12.5	4.383	4.332	0.051
16	H	NH2	CH_3	H	O	CH_2	-1.23	1	12.5	4.280	4.250	0.030
17	H	H	H	CH_3	O	CH_2	0	1	12.5	4.251	4.298	-0.047
18	H	H	CH_3	H	NH	CH_2	0	1	12.5	4.249	4.298	-0.049
19	H	Cl	CH_3	H	NH	CH_2	0.71	1	12.5	4.312	4.326	-0.014
20	H	Br	CH_3	H	NH	CH_2	0.86	1	12.5	4.382	4.332	0.049
21	H	NH2	CH_3	H	NH	CH_2	-1.23	1	12.5	4.278	4.250	0.028
22	H	H	CH_3	H	O	CH_2O	0	0	25	3.980	3.996	-0.015
23	H	H	CH_3	H	O	CH_2S	0	0	25	4.009	3.996	0.013
24	H	H	CH_3	H	NH	CH_2S	0	0	25	4.007	3.996	0.011
25	H	Cl	CH_3	H	NH	CH_2O	0.71	0	25	4.037	4.024	0.014

and parameters used in the best equation and observed, calculated and residual values are given in the Table. The plot of the observed and calculated C values of the growth inhibitory activity of compounds 1-25 against *C. albicans* using the best equation is shown in Figure 2.

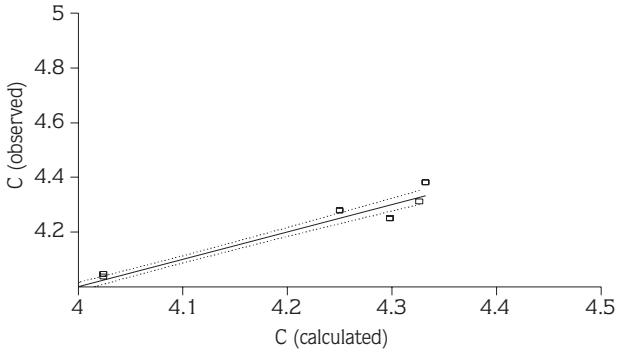


Figure 2. Plot of the observed and calculated C values of the growth inhibitory activity of compounds 1-25 against *C. albicans*.

The best equation and cross-validation results observed in this QSAR study are:

log1/C=+0.0393 (±0.027) πR_2 +0.303 (±0.030) $IyCH_2$ +3.996 (±0.017)

n=25; r=0.97; s=0.03; F=224.38;

p<0.001; Q2=0.93; s-PRESS = 0.03

n: number of compounds

r: correlation coefficient

s: standard deviation

F: Fischer value (measure of the statistical significance)

Q^2: squared cross-validation regression coefficient

s-PRESS: standard deviation of cross-validation predictions

Calculated Q^2 and s-PRESS values show that the predictive power of this QSAR model is significant. Predictions for the lead optimization in this set of compounds can be summarized as follows:

- The activity contributions obtained in this QSAR model show that the indicator structural parameter $IyCH_2$ is significant for the activity. So substitution at position Y with the CH_2 group is more important than with CH_2O and CH_2S groups for the antifungal activity against *C. albicans*.

- The substitution at R_2 is found to be more significant than R_1, R_5, R_6 and X in improving the antifungal activity.

- Hydrophobic properties of the substituents at position R_2 are more indicative than electronic and steric properties of the antifungal activity against *C. albicans*.

Correspondence author:

Özlem TEMİZ ARPACI

Ankara University, Faculty of Pharmacy,

Department of Pharmaceutical Chemistry,

Tandoğan 06100, Ankara - TURKEY

References

1. T.C. Wu, "On the development of antifungal agents: Perspective of the U.S. Food and Drug Administration" Clin. Infect. Dis., 19 (1), 54-58, 1994.
2. P.A. Pizzo, L.S. Young, "Limitations of current antimicrobial therapy in the immunosuppressed host" Am. J. Med., 76, 101-110, 1984.
3. J.R. Graybill, "Future directions of antifungal chemotherapy" Clin. Infect. Dis., 14 (1), 170-181, 1992.
4. M.G. Rinaldi, "Laboratory evaluation of antifungal agents: a brief overview" Clin. Infect. Dis., 14 (1), 130-133, 1992.
5. J.R. Graybill, "The future of antifungal therapy" Clin. Infect. Dis., 22 (2), 166-168, 1996.
6. C.E. Gonzalez, D. Venzon, S. Lee, B.U. Müeller, P.A., Pizzo, T.J. Walsh, "Risk factors for fungemia in children infected with HIV: a case control study" Clin. Infect. Dis., 23, 515-521, 1996.
7. F. Meunier, "Targetting fungi: a challenge" Am. J. Med., 29, 60-67, 1995.
8. D.W. Warnock, "Fungal complications of transplantation: diagnosis, treatment and prevention" J. Antimic. Chemother, 36, 73-90, 1995.
9. J.N. Anker, N.L. Popele, P.J. Sauer, "Antifungal agents in neonatal systemic candidiasis" J. Antimic. Chemother, (B), 1391-1397, 1995.
10. G.D. Gilfillan, D.J. Sullivan, K. Haynes, T. Parkinson, D.C. Colemann, N.A. Gow, "*Candida dubliniensis*: phylogency and putative virulence factors" Microbiology, 144, 829-838, 1998.

11. M.A. Pfaller, S.A., Messer, S. Gee, S. Joly, C. Pujol, D.J. Sullivan, D.C. Colemann, D.R. Soll, "*In vitro* susceptibilities of *Candida dubliniensis* isolates tested against the new triazole and echinocandin antifungal agents" J. Clin. Microbiol., 37, 870-872, 1999.

12. I. Polachek, J. Strahilevitz, D. Sullivan, S. Donnelly, S. Salkin, D.C. Colemann, "Recovery of *Candida dubliniensis* from non-human immunodeficiency cirus-infected patient in Israel" J. Clin. Microbiol., 38, 170-174, 2000.

13. A. Marot, L. Grimaud, S. Bouterige, V. Apaire, D.J. Sullivan, R. Robert, "New monoclonocal antibody specific for *Candida albicans* germ tube" J. Clin. Microbiol., 38, 61-67, 2000.

14. D.A. Stevens, "Therapy for opportunistic fungal infections: past, present and future" Indian J. Cancer, 32, 1-9, 1995.

15. A. Espinel-Ingroff, S. Shadomy, "In vivo and in vitro evaluation of antifungal agents" Eur. J. Clin. Microbiol. Infect. Dis., 8, 352-361, 1989.

16. J.W. Hiemenz, T.J. Walsh, "Lipid formulations of amphotericin B: recent progress and future direction" Clin. Infect. Dis., 22(2), 133-144, 1996.

17. K. Hata, H. Kimura, T. Toyosawa, T. Nakamura, K. Katsu, "In vitro and in vivo antifungal activities of ER-30346, A novel oral triazole with a broad antifungal spectrum" Antimicrob. Agents Chemother, 40, 2237-2242, 1996.

18. J.C. Fung, E. Huczko, B. Minassian, D.P. Bonner, "In vitro activity of a new oral triazole, ER-30306" Antimicrob. Agents Chemother, 42, 313-318, 1998.

19. D.J. Sheehan, C.A. Hitchcock, C.M. Sibley, "Current and emerging azole antifungal agents" Clin. Microbiol. Rev., 12, 40-70, 1999.

20. W. G. Bywater, W. R. Colemann, O. Kamm, H. H. Merritt, "Synthetic anticonvulsants, the preparation and properties of some benzoxazoles" J. Ame. Chem. Soc., 67, 905-907, 1945.

21. D. W. Dunwell, D. Evans, "Synthesis and antiinflammatory activity of some 2-aryl-6-benzoxazoleacetic acid derivatives" J. Med. Chem., 20, 797-801, 1977.

22. M. Pedini, G. De Meo, A. Ricci, L. Bastianin, P. Jaquignon, "New heterocyclic derivatives of benzimidazole with germicidal activity: II 2-(5'-nitro-2'furyl or 2'-thienyl) benzimidazoles with different substituents in the 5-position" Il Farmaco, 45(3), 303-312, 1990.

23. İ. Yalçın, E. Şener, T. Özden, S. Özden, A. Akın, "Synthesis and microbiological activity of 5-methyl-2-[p-substituted phenyl]benzoxazoles" Eur. J. Med. Chem., 25, 705-708, 1990.

24. Ö. Temiz, İ. Ören, E. Şener, İ. Yalçın, N. Uçartürk, "Synthesis and microbiological activity of some novel 5- or 6-methyl-2-(2,4-disubstitutedphenyl)benzoxazole derivatives" Il Farmaco, 53, 337-341, 1998.

25. İ. Ören, Ö. Temiz, İ. Yalçın, E. Şener, A. Akın, N. Uçartürk, "Synthesis and microbiological activity of 5(or 6) –methyl-2-substitutedbenzoxazole and benzimidazole derivatives" Arzneim. Forsch., 47, 1393-1397, 1997.

26. C. Hansch, "On the structure of Medicinal chemistry" J. Med. Chem., 19(1), 1-6, 1976.

27. T. Fujita, "The extrathermodynamic approach to drug design" Comprehensive Medicinal Chemistry, Pergamon, Oxford, Vol. 4, pp. 497-561, 1990.

28. C. Hansch, S. D. Rockwell, P.Y. Jow, A. Leo, E. Steller, C. Hansch, "Substituent constants for correlation analysis" J. Med. Chem., 20 (2), 304-306, 1977.

29. R. Franke, "Theoretical Drug Design Methods", "Theoretical Drug Design Methods", Elseiver, Amsterdam, pp. 256, 1984.

30. K. C. Chu, "The quantitative analysis of structure-activity relationships" "Burger's Medicinal Chemistry", Wiley, New York, pp. 393-419, 1980.

31. E. Şener, İ. Yalçın, E. Sungur, "QSARs of some novel antibacterial benzimidazoles, benzoxazoles and oxazolopyridines against an enteric gram-negative rod: *K. pneumoniae*" QSAR, 10, 223-228, 1991.

32. S. Wold, "Validation of QSARs" QSAR, 10, 191-193, 1991.

33. H. Kubinyi, BASF AG ZHF/G-A30 67045 Ludwigshafen, Germany.

34. H. Kubinyi, O.-H. Kehrhahn, "Quantitative activity relationships, 6. Nonlinear dependence of biological activity on hydrophobic character, calculation procedures for the bilinear model" Arzneim.-Forsch (Drug Res.), 28, 598-601, 1978.