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# QSARs of some novel antibacterial benzimidazoles, benzoxazoles, and oxazolopyridines against an enteric gram-negative rod; *K. pneumoniae*

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## Summary

A set of previously synthesized 2,5-disubstituted benzimidazole (I), benzoxazole (II), and 2-substituted oxazolo(4,5-*b*)pyridine (III) derivatives were tested for in vitro growth inhibitory activity against *K. pneumoniae* and the quantitative structure-activity relationships (QSARs) were analyzed by a computer-assisted multiple regression procedure. The activity contributions for either heterocyclic ring systems or substituent effects were determined from the correlation equations and 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 favorable structure over the other heterocyclic nuclei against *K. pneumoniae*. The position R in the fused ring system was found to be important for improving the activity by substitution at this position by hydrogen accepting groups with the electronic effect of negative field interactions.

## Introduction

The development of new and different antimicrobial agents is a very important step and much research effort is directed toward to the design of available drugs that are resistant to inactivation by bacterial enzymes.

The abundant use of antimicrobial drugs, particularly in hospitalized patients, leads to the suppression of drug-susceptible organisms in the gut flora and favors the persistence and growth of drug resistant bacteria, including *Klebsiella* (Martin, 1982). The closed environment of hospitals favors transmission of these resistant organisms and such microorganisms constitute particularly difficult problems especially in granulopenic and immunocompromised patients (Boyd, 1984). *K. pneumoniae*, originally known as a respiratory pathogen, is now commonly encountered in hos-

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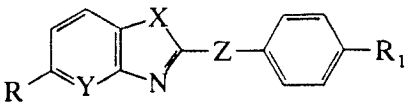
pital infections of the respiratory and urinary tracts. It produces extensive hemorrhagic necrotizing consolidation of the lung, which, if untreated, has a high mortality rate. Occasionally, it produces urinary tract infection or enteritis in children and bacteremia with focal lesions in debilitated patients (Joklik et al., 1980; Jawetz et al., 1984).

Nosocomial (hospital-acquired) infections caused by enteric gram-negative rods are often resistant to antibiotic therapy and have become a serious medical problem. Research has clearly established that multiple resistance among gram-negative organisms to a variety of antibiotics occurs and can be transmitted to previously nonresistant strains of the same species and, indeed, to different species of bacteria (Martin, 1982; Sande and Mandell, 1990).

In the search for new and different antimicrobial agents, a variety of benzimidazole derivatives have previously been investigated with respect to their antimicrobial activities (Islip, 1979; Pedini et al., 1987, 1990). Recent observations suggest that, besides the substituted benzimidazole derivatives, several analogs of this ring system, such as benzoxazole derivatives, also Indicate potential antibacterial activity with lower toxicity (Geigy, 1971; Brown et al., 1978; Haugwitz et al., 1982; Hisano et al., 1982; De Meo et al., 1989).

Consequently, in view of these phenomena and the fact that many effective antimicrobial agents bear a heterocyclic system in their molecule (Daidone, 1990), we have synthesized different derivatives of benzoxazoles, benzimidazoles, and oxazolo(4,5-*b*)pyridines during the last few years (Yalçin et al., 1985, 1990; Şener et al., 1987a,b). Our investigations suggest that they have a broad spectrum of antimicrobial activity, especially against gram-negative microorganisms (De Meo et al., 1989; Yalçin et al., 1990).

In this study, the in vitro growth inhibitory activity of the previously synthesized compounds, 2,5-disubstituted benzimidazoles (I), benzoxazoles (II) and 2-substituted oxazolo(4,5-*b*)pyridines (III) against *K. pneumoniae* was determined and quantitative structure-activity relationships (QSARs) of these derivatives were analyzed.




I, X: NH Y: CH Z: $CH_2$ or –
II, X: O Y: CH Z: $CH_2$ or –
III, X: O Y: N Z: –

The substituents at positions R and $R_1$, which are given in Table 1, are electron donating or withdrawing groups. The activity contributions for either the ring systems and/or the substituents have been calculated from the correlation equations and the predictions for the lead optimization have been described by the results obtained from QSAR analysis.

## Materials and Methods

*Data processing*

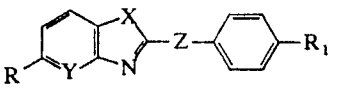
An extrathermodynamic approach in the quantitative analysis of structure-activity relationships (QSAR) has been most widely and effectively used for theoretical drug design. This method has also been called the Hansch approach and it assumes that the potency of a certain biological activity exerted by a series of congeneric compounds can be expressed in terms of a function of various physicochemical (electronic, steric, and hydrophobic) effects, with provision for structural or theoretical effects (Fujita, 1990). This assumption is summarized in Eqn 1:

$$f(\text{biological activity}) = f(\text{electronic}) + f(\text{steric}) + f(\text{hydrophobic}) + [f(\text{structural}) + f(\text{theoretical})] \quad (1)$$

If these functions could be formulated in a equation showing that certain effects are favorable for the activity, the structural modifications

TABLE 1

*Compounds and parameters used in Eqn 8*




| Compound | R | $R_1$ | X | Y | Z | $H_{ACCEPT,R}$ | $F_R$ | $I_X$ | $I_Y$ | $I_Z$ | MIC ($\mu$g/ml) | Observed [a] | Calculated[b] | Residual |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | H | H | O | CH | – | 0 | 0.00 | 0 | 0 | 0 | 12.5 | 4.1936 | 4.2445 | −0.0509 |
| 2 | H | $OCH_3$ | O | CH | – | 0 | 0.00 | 0 | 0 | 0 | 12.5 | 4.2557 | 4.2445 | 0.0112 |
| 3 | H | $C(CH_3)_3$ | O | CH | – | 0 | 0.00 | 0 | 0 | 0 | 12.5 | 4.3033 | 4.2445 | 0.0588 |
| 4 | H | $NH_2$ | O | CH | – | 0 | 0.00 | 0 | 0 | 0 | 12.5 | 4.2258 | 4.2445 | −0.0187 |
| 5 | H | $NHCH_3$ | O | CH | – | 0 | 0.00 | 0 | 0 | 0 | 12.5 | 4.2538 | 4.2445 | 0.0093 |
| 6 | Cl | $CH_3$ | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 3.9889 | 4.0491 | −0.0602 |
| 7 | Cl | $C_2H_5$ | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 4.0132 | 4.0491 | −0.0359 |
| 8 | Cl | $C(CH_3)_3$ | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 4.0580 | 4.0491 | 0.0089 |
| 9 | Cl | $NHCOCH_3$ | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 4.0595 | 4.0491 | 0.0104 |
| 10 | Cl | $NHCH_3$ | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 4.0148 | 4.0.191 | −0.0343 |
| 11 | Cl | Cl | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 4.0238 | 4.0.191 | −0.0253 |
| 12 | Cl | $NO_2$ | O | CH | – | 0 | 0.41 | 0 | 0 | 0 | 25 | 4.0624 | 4.0.191 | 0.0133 |
| 13 | $NO_2H$ | | O | CH | – | 1 | 0.67 | 0 | 0 | 0 | 12.5 | 4.2837 | 4.3253 | −0.0416 |
| 14 | $NO_2CH_3$ | | O | CH | – | 1 | 0.67 | 0 | 0 | 0 | 12.5 | 4.3083 | 4.3253 | −0.0170 |
| 15 | $NO_2C(CH_3)_3$ | | O | CH | – | 1 | 0.67 | 0 | 0 | 0 | 12.5 | 4.37.18 | 4.3253 | 0.0495 |
| 16 | $NO_2NH_2$ | | O | CH | – | 1 | 0.67 | 0 | 0 | 0 | 12.5 | 4.3100 | 4.3253 | −0.0153 |
| 17 | $NO_2Cl$ | | O | CH | – | 1 | 0.67 | 0 | 0 | 0 | 12.5 | 4.3418 | 4.3253 | 0.0165 |
| 18 | $NO_2Br$ | | O | CH | – | 1 | 0.67 | 0 | 0 | 0 | 12.5 | 4.4070 | 4.3253 | 0.0817 |
| 19 | $NH_2H$ | | O | CH | – | 1 | 0.02 | 0 | 0 | 0 | 6.25 | 4.5269 | 4.6351 | −0.1082 |
| 20 | $NH_2C_2H_5$ | | O | CH | – | 1 | 0.02 | 0 | 0 | 0 | 6.25 | 4.6060 | 4.6351 | −0.0291 |
| 21 | $NH_2NO_2$ | | O | CH | – | 1 | 0.02 | 0 | 0 | 0 | 6.25 | 4.6142 | 4.6351 | −0.0209 |
| 22 | $NH_2Br$ | | O | CH | – | 1 | 0.02 | 0 | 0 | 0 | 6.25 | 4.7197 | 4.6351 | 0.0846 |
| 23 | $NH_2F$ | | O | CH | – | 1 | 0.02 | 0 | 0 | 0 | 6.25 | 4.6296 | 4.6351 | −0.0055 |
| 24 | $NH_2N(CH_3)_2$ | | O | CH | – | 1 | 0.02 | 0 | 0 | 0 | 6.25 | 4.6313 | 4.6351 | −0.0038 |
| 25 | $CH_3CH_3$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.2519 | 4.2636 | −0.0117 |
| 26 | $CH_3C_2H_5$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.2783 | 4.2636 | 0.0147 |
| 27 | $CH_3OCH_3$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.2819 | 4.2636 | 0.0183 |
| 28 | $CH_3F$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.2595 | 4.2636 | −0.0041 |
| 29 | $CH_3NHCOCH_3$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.2874 | 4.2636 | 0.0238 |
| 30 | $CH_3NHCH_3$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.2801 | 4.2636 | 0.0165 |
| 31 | $CH_3N(CH_3)_2$ | | O | CH | – | 0 | −0.04 | 0 | 0 | 0 | 12.5 | 4.3050 | 4.2636 | 0.0414 |
| 32 | H | $CH_3$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5298 | 4.5763 | −0.0465 |
| 33 | H | $C_2H_5$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5584 | 4.5763 | −0.0179 |
| 34 | H | $C(CH_3)_3$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.6090 | 4.5763 | 0.0327 |
| 35 | H | $OCH_3$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5622 | 4.5763 | −0.0141 |
| 36 | H | $OC_2H_5$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5883 | 4.5763 | 0.0120 |
| 37 | H | $NH_2$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5319 | 4.5763 | −0.0444 |
| 38 | H | $NO_2$ | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5900 | 4.5763 | 0.0137 |
| 39 | H | Cl | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.5703 | 4.5763 | −0.0060 |
| 40 | H | Br | O | N | – | 0 | 0.00 | 0 | 1 | 0 | 6.25 | 4.6471 | 4.5763 | 0.0708 |
| 41 | H | H | O | CH | $CH_2$ | 0 | 0.00 | 0 | 0 | 1 | 6.25 | 4.5282 | 4.5918 | −0.0636 |
| 42 | H | $OCH_3$ | O | CH | $CH_2$ | 0 | 0.00 | 0 | 0 | 1 | 6.25 | 4.5865 | 4.5918 | −0.0053 |
| 43 | H | Br | O | CH | $CH_2$ | 0 | 0.00 | 0 | 0 | 1 | 6.25 | 4.6672 | 4.5918 | 0.0754 |
| 44 | H | Cl | O | CH | $CH_2$ | 0 | 0.00 | 0 | 0 | 1 | 6.25 | 4.5945 | 4.5918 | 0.0027 |
| 45 | H | $NO_2$ | O | CH | $CH_2$ | 0 | 0.00 | 0 | 0 | 1 | 6.25 | 4.6129 | 4.5918 | 0.0211 |
| 46 | $NO_2H$ | | O | CH | $CH_2$ | 1 | 0.67 | 0 | 0 | 1 | 6.25 | 4.6130 | 4.6725 | −0.0595 |
| 47 | $NO_2OCH_3$ | | O | CH | $CH_2$ | 1 | 0.67 | 0 | 0 | 1 | 6.25 | 4.6610 | 4.6725 | −0.0115 |
| 48 | $NO_2Br$ | | O | CH | $CH_2$ | 1 | 0.67 | 0 | 0 | 1 | 6.25 | 4.7300 | 4.625 | 0.05–5 |

TABLE 1 (continued)

| Compound | R | $R_1$ | X | Y | Z | $H_{ACCEPT,R}$ | $F_R$ | $I_X$ | $I_Y$ | $I_Z$ | MIC ($\mu$g/ml) | Observed [a] | Calculated[b] | Residual |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 49 | $NO_2$ | Cl | O | CH | $CH_2$ | 1 | 0.67 | 0 | 0 | 1 | 6.25 | 4.6680 | 4.6725 | −0.0045 |
| 50 | $NO_2$ | $NO_2$ | O | CH | $CH_2$ | 1 | 0.67 | 0 | 0 | 1 | 6.25 | 4.6840 | 4.6725 | 0.0115 |
| 51 | $NO_2$ | $NO_2$ | NH | CH | $CH_2$ | 1 | 0.67 | 1 | 0 | 1 | 12.5 | 4.373 | 4.3641 | 0.0132 |
| 52 | $NO_2$ | $OCH_3$ | NH | CH | $CH_2$ | 1 | 0.67 | 1 | 0 | 1 | 12.5 | 4.3549 | 4.3641 | −0.0092 |
| 53 | $NO_2$ | $OC_2H_5$ | NH | CH | $CH_2$ | 1 | 0.67 | 1 | 0 | 1 | 12.5 | 4.3i59 | 4.3641 | 0.0118 |
| 54 | $CH_3$ | $CH_3$ | NH | CH | $CH_2$ | 0 | −0.04 | 1 | 0 | 1 | 12.5 | 4.2760 | 4.3023 | −0.0263 |
| 55 | $CH_3$ | $OCH_3$ | NH | CH | $CH_2$ | 0 | −0.04 | 1 | 0 | 1 | 12.5 | 4.3045 | 4.3023 | 0.0022 |
| 56 | $CH_3$ | $OC_2H_5$ | NH | CH | $CH_2$ | 0 | −0.04 | 1 | 0 | 1 | 12.5 | 4.2869 | 4.3023 | −0.0154 |
| 57 | H | $CH_3$ | NH | CH | – | 0 | 0.00 | 1 | 0 | 0 | 25 | 3.9201 | 3.9361 | 0.0160 |
| 58 | H | $OCH_3$ | NH | CH | – | 0 | 0.00 | 1 | 0 | 0 | 25 | 3.9523 | 3.9361 | 0.0162 |
| 59 | $CH_3$ | $OCH_3$ | NH | CH | – | 0 | 0.00 | 1 | 0 | 0 | 25 | 3.9786 | 3.9551 | 0.0235 |

[a] Defined as log $1/C$.
[b] Using Eqn 8.

which enhance such properties would be expected to generate compounds of potent activity.

Multiple regression analysis which involves 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 (Chu, 1980). This is formally expressed as follows;

$$y = a_0 + a_1x_1 + a_2x_2 + \ldots + a_nx_n + \epsilon \qquad (2)$$

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

For the procedure of descriptor selection related to the activity among the candidate set of variables, forward step-wise multiple regression of elimination technique is applied. During the development of the best fit model of the correlation equation, the minimum $F$ value for entering and removing the variables in the step-wise multiple regression was taken as 8.0 which is statistically significant at the 1% level of probability (Şener et al., 1991).

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 and finally the resulting PRESS (predictive residual sum of squares) is calculated via the closed form given in Eqn 3 (Rawlings, 1988; Wold, 1991):

$$\text{PRESS} = \sum_i \left[ \left( y_i - \hat{y}_i \right)^2 / (1 - h_{ii})^2 \right] \qquad (3)$$

where $y_i$ and $\hat{y}_i$ are the response (activity) values of observation $i$ ($i = 1, 2, \ldots, n$), observed and calculated by the best equation, respectively. The diagonal elements of the 'hat' matrix are denoted by $h_ii$ in the equation and calculated by the computer program used in this QSAR study (Rawlings, 1988).

Regression analysis and calculations were run on an IBM 360/158 computer using the BMDP 2R statistical program package. Graphics were obtained from the STATGRAPHICS software. In equations, the figures in parentheses are the standard errors of the regression coefficients. For a given equation, $n$ is the number of compounds, $R^2$ denotes the square of the multiple correlation

coefficients, $F$ is the significance test and $s$ represents the residual standard deviation.

*Determination of parameters*

A congeneric set of 2,5-disubstituted benzimidazoles, benzoxazoles, and 2-substituted oxazolo (4,5-*b*)pyridine derivatives 1–59 were considered for this study.

The candidate set of variables used in this analysis are hydrophobic, electronic, steric and structural parameters. The structural variable $I_Y$ expresses the replacement of -CH = by the isosteric group of -N = in the six-membered ring of the fused ring system. $I_y$ is defined as 0 for type I and II compounds and 1 for type III compounds. The other structural variable $I_X$ expresses the exchange between -O- and -NH- groups in the five-membered ring and is represented as 1 for type I, and 0 for type II and III compounds. Additionally, $I_Z$ 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 hydrogen donating/accepting capabilities ($H_{DONOR}$/$H_{ACCEPT}$) of the substituents at R and $R_1$ are the indicator variables.

The screened physicochemical parameters in this QSAR study are $\pi$ for the hydrophobic effects, $F$ (field effect), $R$ (resonance effect) as the electronic influence and MR, Verloop's STERIMOL parameters ($L$, $B_1$, $B_4$) for the steric interactions of the substituents R and $R_1$. Values for all candidate physicochemical variables used in this QSAR study were taken from the table of Hansch and Leo (1979). The values of the descriptors related to the activity among the candidate set of variables of the best equation (Eqn 8) in the QSAR analysis are shown in Table 1.

*In vitro microbiological activity*

The antibacterial activities against the strain *K. pneumoniae* NTCC 52211 were determined as the minimum inhibitory concentration (MIC) values in vitro by a 2-fold serial dilution technique (Charles et al., 1979; Shadomy and Espinel, 1980). The test was performed using the compounds which were dissolved in absolute ethanol (0.4 mg/ml) and further control dilutions in the test medium were furnished at the required quantities of 200, 100, 50, 25, 12.5, 6.25, 3.12, and 1.56 $\mu$g/ml concentrations. In order to ensure that the solvent per se had no effect on bacterial growth, a control test was also performed containing inoculated broth supplemented with only ethanol at the same dilutions used in our experiments and found to be inactive in culture medium. For the antibacterial assay, the cultures were obtained in Mueller Hinton broth (Difco) after 24 h of incubation at 37 ± 1°C. Testing was carried out in Mueller Hinton broth at pH 7.4 and the 2-fold serial dilution technique was applied. A set of tubes containing only inoculated broth was kept as controls. After incubation for 24 h at 37 ± 1°C, the last tube with no growth of the

TABLE 2

*Stepwise Development of Eqn 8*

| Eqn no. | Equation | $n$ | $R^2$ | $s$ | $F$ |
|---|---|---|---|---|---|
| 4 | $\log 1/C = 0.196(\pm 0.058)H_{ACCEPT,R} + 4.315$ | 59 | 0.17 | 0.21 | 11 |
| 5 | $\log 1/C = 0.274(\pm 0.051)H_{ACCEPT,R} + 0.340(\pm 0.067)I_Y$ 4.237 + 59 | 0.43 | 0.18 | | |
| 6 | $\log 1/C = 0.245(\pm 0.043)H_{ACCEPT,R} + 0.398(\pm 0.0.058)I_Y +$ $0.219(\pm 0.046)I_Z + 4.178$ | 59 | 0.60 | 0.15 | 27 |
| 7 | $\log 1/C = 0.415(\pm 0.042)H_{ACCEPT,R} + 0.371(\pm 0.044)I_Y +$ $0.252(\pm 0.035)I_Z - 0.443(\pm 0.068)F_R + 4.205$ | 59 | 0.77 | 0.11 | 46 |
| 8 | $\log 1/C = 0.400(\pm 0.015)H_{ACCEPT,R} + 0.332(\pm 0.015)I_Y +$ $0.347(\pm 0.013)I_Z - 0.477(\pm 0.024)F_R -$ $0.308(\pm 0.015)I_X + 4.245$ | 59 | 0.97 | 0.04 | 393 $p < 0.001$ |

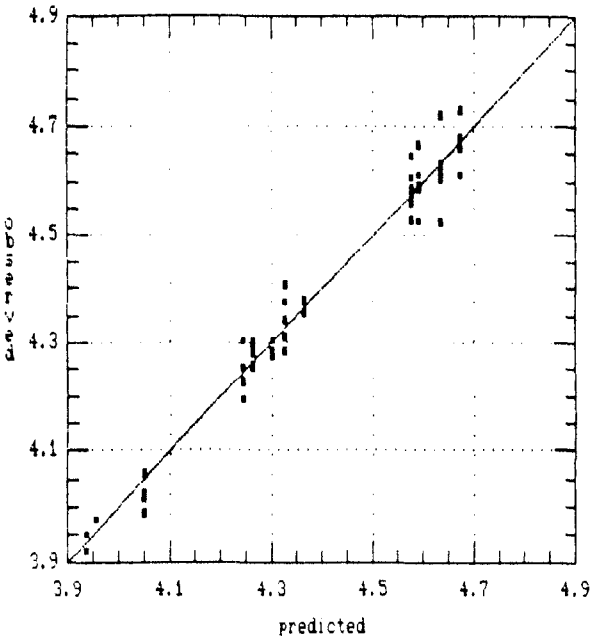



Fig. 1. Plot of the observed vs predicted values (log $1/C$) of the in vitro growth inhibitory activity of the compounds against *K. pneumoniae* using Eqn 8.

microorganism was recorded to represent MIC expressed in $\mu$g/ml. The potency has been defined as log $1/C$ in the QSAR analysis where $C$ is the molar MIC value of the compounds. MIC and the observed log $1/C$ values of the compounds are listed in Table 1.

## Results and Discussion

QSAR analysis reveals that Eqn 8, given in Table 2 represents the best fit equation for the predictions according to the applied regression analysis and validation test results.

As can be deduced from Fig. 1, the goodness of fit of Eqn 8 is significant, possessing a high $R^2$ (97%) and a small $s$ (0.04) with an overall $F$ test value of 393 at the significant level of $p < 0.001$. However, the complete analysis of the variance table of Eqn 8 is also given in Table 3 for further information.

In order to avoid the risk of chance correlation, some circumstances which were pointed out by Topliss and Edwards (1979) and Wold (1991) have been taken into consideration in the study. Thus, 59 observations (compounds) are used to screen the 22 variables for keeping the probability of encountering a chance correlation with $R^2 \geqq 0.9$ at the 1% level or less (Topliss and Edwards, 1979). The search of the simple correlation coefficients which are given in Table 4 also reveals that there is no colinearity between the independent variables in any case.

To prove the predictive power of Eqn 8, cross-validation is applied to the original data set and the resulting PRESS is calculated. The calculated overall PRESS is 0.0996, which is found to

TABLE 4

*Correlation matrix of variables used in Eqn 8*

| | Log $1/C$ | $H_{ACCEPT,R}$ | $I_X$ | $I_Y$ | $I_Z$ | $F_R$ |
|---|---|---|---|---|---|---|
| log $1/C$ | 1.00 | | | | | |
| $H_{ACCEPT,R}$ | 0.41 | 1.00 | | | | |
| $I_X$ | −0.33 | −0.01 | 1.00 | | | |
| $I_Y$ | 0.36 | −0.30 | −0.18 | 1.00 | | |
| $I_Z$ | 0.37 | 0.21 | 0.38 | −0.26 | 1.00 | |
| $F_R$ | −0.04 | 0.67 | −0.01 | −0.29 | 0.26 | 1.00 |

TABLE 3

*Complete analysis of variance table of Eqn 8*

| Source | Degrees of freedom | Sum of squares | Mean square | $F$ ratio | $p$ level |
|---|---|---|---|---|---|
| Total (corrected) | 58 | 3.0335 | | | |
| Regression | 5 | 2.9538 | 0.5908 | | |
| $H_{ACCEPT,R}$ | 1 | 1.2346 | 1.2346 | 821 | 0.000 |
| $I_Y/H_{ACCEPT,R}$ | 1 | 0.4031 | 0.4031 | 268 | 0.000 |
| $I_Z/H_{ACCEPT,R}$, $I_Y$ | 1 | 0.6957 | 0.6957 | 462 | 0.000 |
| $F_R/H_{ACCEPT,R}$, $I_Y$, $I_Z$ | 1 | 0.0156 | 0.0156 | 10 | 0.002 |
| $I_X/H_{ACCEPT,R}$, $I_Y$, $I_Z$, $F_R$ | 1 | 0.6049 | 0.6049 | 402 | 0.000 |
| Error | 53 | 0.0797 | 0.0015 | | |

be smaller than the value of SSY (sum of the squares of the response values of the total observations, SSY = 3.0335; see Table 3). This proves that the developed model (Eqn 8) predicts better than chance and can be considered statistically significant (Rawlings, 1988; Wold, 1991).

The ratio PRESS/SSY, which is the approximate confidence interval for predictions of new compounds, is 0.0328 and it also provides proof that the model is valid (Wold, 1991).

In QSAR analysis, Eqn 8 reveals that position R of the fused ring system is important for the antibacterial activity against *K. pneumoniae*. The hydrogen accepting property of a substituent at this position ($H_{ACCEPT,R}$) produces an additive contribution to the activity.

When the physicochemical properties of the substituents are compared, the fitted model indicates that electronic influences of negative field effects at R ($F_R$) enhance the potency.

In addition to this feature, Eqn 8 reveals that the structural parameters $I_X$, $I_Y$, and $I_Z$ are also significant for the activity. As can be deduced from Fig. 2, the compounds possessing a methylene group between the *p*-substituted phenyl moiety and the fused ring system in position 2 ($I_Z$) are important and provide an improvement in the activity (see compounds 41–50 in Fig. 2). Additionally, activity contributions of the other structural parameters $I_X$ and $I_Y$ indicate that the oxazolo(4,5-*b*)pyridine ring system is the preferred structure over the other heterocylic nuclei against *K. pneumoniae*.

On the other hand, it was observed that there was no statistical significant relationships between the activity and any parameters related to $R_1$ and the *para* substitution of the phenyl or benzyl moiety has a negligible effect on the activity.

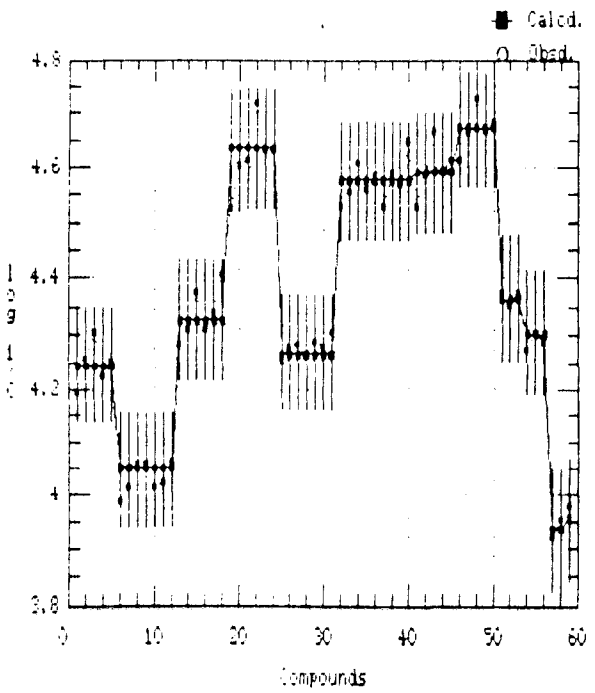



Fig. 2. Plot of the calculated and observed values (log 1/C) vs index of the compounds (1–59) with 99% intervals of the confidence limits for predictions using Eqn 8.

## Conclusion

According to the predictions obtained from QSAR analysis, the lead optimization in this set of compounds can be defined as follows.

The model indicates that the lead compound for the activity against *K. pneumoniae* has the heterocyclic structure of an oxazolo(4,5-*b*)pyridine ring system with the substitution of a benzyl moiety at position 2.

A substituent which possesses hydrogen accepting capability at R improves the activity via the physicochemical property of the electronic effect of negative field interactions.

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