

Comparative Molecular Field Analysis of CXCR-2 Inhibitors

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Abstract

CXC chemokine receptor 2 (CXCR2) is a prominent chemokine receptor on neutrophils. The neutrophilic inflammation in the lung diseases is found to be largely regulated through CXCR2 receptor. Antagonist of CXCR2 may reduce the neutrophil chemotaxis and alter the inflammatory response. Hence, in the present study, ligand based Comparative molecular field analysis (CoMFA) was performed on a series of CXCR2 antagonist named pyrimidine-5-carbonitrile-6-alkyl derivatives. The optimum CoMFA model was obtained with statistically significant cross-validated coefficients (q^2) of 0.568 and conventional coefficients (r^2) of 0.975. The contour maps suggest the important structural modifications and this study can be used to guide the development of potent CXCR2 antagonist.

Keywords: Chemokines, CXCR-2, 3D-QSAR, CoMFA.

1. Introduction

Chemokines are small 8-10 kDa which have long been implicated in the initiation and amplification of inflammatory responses by their role in leukocyte chemotaxis^[1,2]. Chemokines act through G-protein-coupled receptors (GPCRs) to regulate a variety of effects, including cell migration and inflammatory events. They are currently seven known CXCR receptor found in mammals named CXCR1-CXCR7. CXCR2 (also called CD182, IL8) plays a critical role in the regulation of neutrophil homeostasis^[3] and is found on many cells including leukocytes, endothelial and epithelial cells^[4,5]. CXCR2 receptor can be released from a number of inflammatory and structural cell types and may have a broad functional role in number of acute and chronic diseases. It plays an important role in chronic obstructive pulmonary disease (COPD), asthma, fibrotic pulmonary disorders^[6-8]. It was found that neutrophilic inflammation in the lung diseases is found to be largely regulated through CXCR2^[2,9]. Therefore blockade of CXCR2 substantially reduces leukocyte recruitment, tissue damage and mortality. An antagonist of CXCR2 reduces neutrophilic chemotaxis and may alter the air-

way inflammation. To date, there are no CXCR2 receptor antagonists approved for use in humans. However, several pharmaceutical companies have disclosed CXCR2 antagonists and amongst these, navarixin and AZD-5069 are noteworthy.

Quantitative Structural Activity relationship (QSAR) models attempts to relate the chemical structure to biological activity computationally or mathematically and to discover new compounds with improved biological activity. When 3D structure of the macromolecular target is not available 3D-QSAR is the prominent computational means to support chemistry within drug design projects^[10] and is demonstrated in many studies^[11-14]. Hence, in the present 3D QSAR method, CoMFA model was generated utilizing pyrimidine-5-carbonitrile-6-alkyl derivatives as CXCR2 antagonist. The models obtained were all statistically significant with cross-validated coefficients (q^2) >0.5 and conventional coefficients (r^2) >0.5 and predictive r^2 (r^2_{pred}) \geq 0.5 indicating that reliability of the model. The CoMFA model was graphically interpreted by field a contribution map which provides guidelines for the design of new compounds with enhanced activity and specificity.

2. Materials and Methods

2.1. Data Set

The structure of the pyrimidine-5-carbonitrile-6-alkyl derivatives and their biological activities of 26 com-

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Table 1. Structures and biological activities (pIC₅₀) of CXCR2 inhibitors

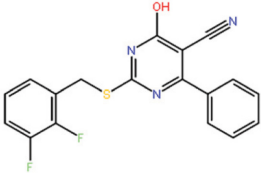
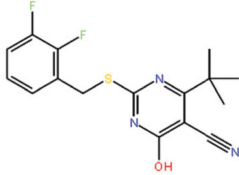
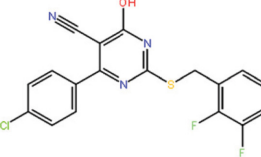
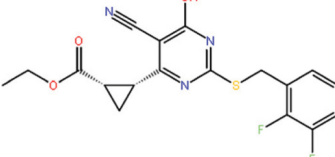
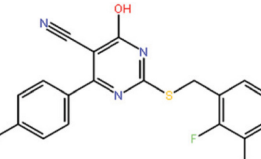
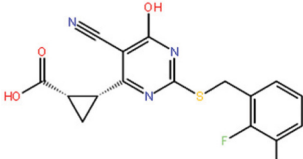
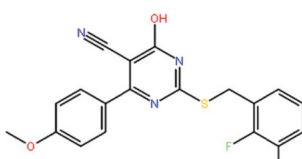
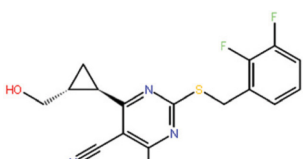
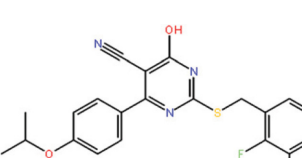
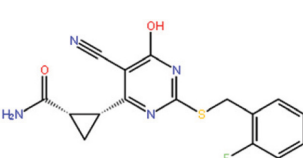
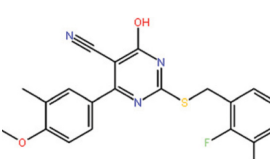
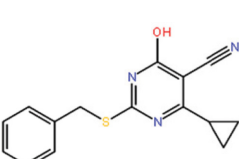
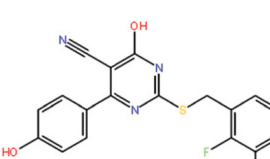
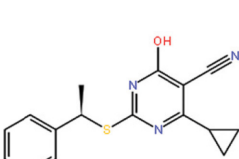
Cmpd no	Structure	pIC ₅₀ values	Cmpd no	Structure	pIC ₅₀ values
1		5.432	14		5.432
2		5.130	15		5.824
3		5.854	16		8.000
4		6.148	17		8.222
5		5.795	18		8.155
6		5.337	19		6.292
7		6.107	20		5.318

Table 1. Continued

Cmpd no	Structure	pIC ₅₀ values	Cmpd no	Structure	pIC ₅₀ values
8		5.327	21		5.193
9		5.366	22		4.522
10		6.045	23		5.000
11		5.309	24		6.495
12		5.769	25		5.854
13		6.853	26		5.495

pounds were taken from the literature^[15]. Biological activities i.e. IC₅₀ values of each inhibitor was converted into pIC₅₀ (-logIC₅₀) in order to use the data as dependent variable in CoMFA analysis. We segregated the dataset (26 compounds) into test (5 compounds) and

training set (21 compounds). The training and test sets were classified to ensure that both sets could completely cover the whole range of biological activity and structural diversity. The structures and their activity values are displayed in Table 1.

2.2. Structural Alignment

The structure of pyrimidine-5-carbonitrile-6-alkyl derivatives was drawn using sketch molecule function in SYBYL-X2.1^[16] version whose the partial atomic charges were assigned by utilizing Gasteiger-Huckel method. The highly active molecule (compound 17) among the dataset was selected as template and its bioactive conformation was searched through systematic search analysis. The bioactive conformation of highly active molecule was utilized and its lowest energy conformer was used to align all the compounds and it was subsequently used for CoMFA and CoMSIA analysis. The alignment of all molecules is represented in Fig. 1.

2.3. CoMFA Analysis

CoMFA studies helps in deriving a relation between the biological activities and three dimensional structures of the set of molecules. During CoMFA Analysis, ligand was placed in a 3D grid and the molecular field values of each conformation were calculated with 2 Å lattice spacing. A sp^3 carbon atom with a charge of +1.0 and a Van der Waals radius of 1.52 Å was used as a probe; this atom was placed at every lattice point to calculate various steric and electrostatic fields. The steric (Lennard-Jones) and electrostatic (Coulombic) potential energies of the Tripos force field implemented in SYBYL were evaluated by CoMFA. An energy cut off value of 30 kcal/mol was imposed on all CoMFA cal-

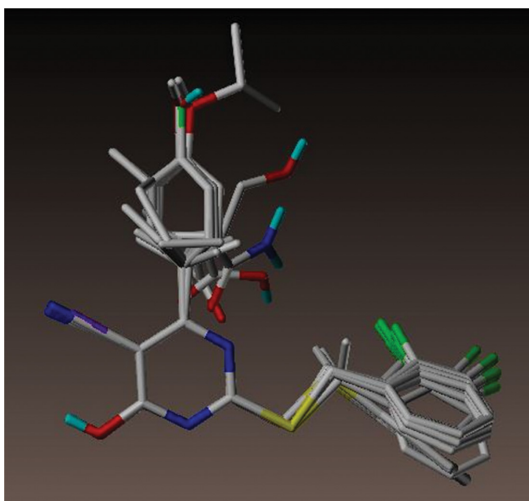


Fig. 1. Superimposed structure of all molecules based on atom by atom matching alignment.

culations to avoid excessively high and unrealistic energy values within the molecule.

2.4. Partial Least Square (PLS) Analysis

Partial least squares (PLS) algorithm^[17] quantifies the relationship between the structural parameters and the biological activities^[18]. CoMFA descriptors used as independent variables and pIC_{50} values used as dependent variables in PLS analysis for the generation of CoMFA model. Leave-one-out (LOO)^[19] cross-validation procedures were used to obtain the cross-validated correlation coefficient (q^2), non-cross-validated correlation coefficient (r^2), standard error estimate (SEE) and Fisher's values (F). A non-cross-validated analysis was carried out without column filtering was then followed. The cross-validated correlation coefficient (q^2) was calculated using the following equation:

$$q^2 = 1 - \frac{\sum (\gamma_{pred} - \gamma_{actual})^2}{\sum (\gamma_{actual} - \gamma_{mean})^2}$$

where γ_{pred} , γ_{actual} , and γ_{mean} are the predicted, actual, and mean values of the target property (pIC_{50}), respectively.

2.5. Predictive Correlation Coefficient (r^2_{pred})

The predictive power of CoMFA model was determined from five test molecules which were excluded during model development. The predictive correlation coefficient (r^2_{pred}) based on the test set molecules, is defined as:

$$r^2_{pred} = \frac{(SD - PRESS)}{SD}$$

where PRESS is the sum of the squared deviation between the predicted and actual activity of the test set molecules, and SD is defined as the sum of the square deviation between the biological activity of the test set compounds and the mean activity of the training set molecules^[20].

3. Results and Discussion

In the present work, we have generated CoMFA model for pyrimidine-5-carbonitrile-6-alkyl derivatives. Statistical values like q^2 , r^2 , SEE, F value and r^2_{pred} were calculated. Each model was validated with statistical cut

off values such as $q^2 > 0.5$, $r^2 > 0.5$ and $r^2_{\text{pred}} > 0.5$. The best predictions were obtained for CoMFA model ($q^2 = 0.568$, $r^2 = 0.975$).

3.1. CoMFA Statistical Analysis

CoMFA analysis was carried out to evaluate the steric and electrostatic properties. The cross-validated correlation coefficient, q^2 defines the goodness of prediction whereas the conventional correlation coefficient, r^2 indicates goodness of fit of a QSAR model. Many different CoMFA models were generated with different combinations of training and test set and the best model was selected based on statistical results. The statistical result of CoMFA model was given in Table 2. For the selected model leave one out analysis gave the cross-validated q^2 of 0.568 with three components and non-cross-validated PLS analysis resulted in a correlation coefficient r^2 of 0.975, $F = 196.628$, and an estimated standard error of 0.159. The steric contribution (0.420) of the model was lower than electrostatic field contribution (0.580). Predicted, experimental activities and their residual values of all inhibitors are shown in Table 3. The residual value of all compounds taken is less than 1 which shows the goodness of the model.

3.2. Mapping of CoMFA Contour Map

The CoMFA contour map was generated based on the ligand based (atom-by atom matching) alignment. The CoMFA result is usually represented as 3D contour map. It shows regions where variations of steric and electrostatic nature in the structural features of the dif-

ferent molecules contained in the training set leads to increase or decrease in the activity. In case of CoMFA the green contours denote favorable steric interactions and the yellow contours show the region where the steric group was not favored. The most active compound 17 was superimposed with steric contour plots and shown in Fig. 2(A). The green steric contour depicts the presence of bulky group in the cyclopropyl ring attached to pyrimidine ring makes the compound potent with higher activity. Hence the compounds 17, 18, 16 have higher activity compared to other molecules. The presence of green contour in 2nd position of benzyl attached to sulfur also indicates the bulky group at this position is favorable. The yellow contour indicates the presence of bulky group in 3rd, 4th and 5th posi-

Table 3. Predicted activities of CoMFA model compared with the experimental pIC_{50} values

S.No.	Actual pIC_{50}	Predicted pIC_{50}	Residual
1.	5.432	5.620	-0.188
2.	5.130	5.336	-0.206
3.	5.854	5.714	0.139
4.	6.148	5.986	0.162
5.	5.795	5.986	-0.191
6.	5.337	5.521	-0.183
7.	6.107	5.163	0.141
8.	5.327	5.163	0.164
9.	5.366	5.953	-0.058
10.	6.045	6.020	0.025
11.	5.309	5.258	0.051
12.	5.769	5.704	0.065
13.	6.853	6.299	0.554
14.	5.432	5.355	0.076
15.	5.824	5.586	0.237
16.	8.000	8.324	-0.324
17.	8.222	8.020	0.201
18.	8.155	8.111	0.043
19.	6.292	5.992	0.3
20.	5.318	5.676	-0.358
21.	5.193	5.382	-0.189
22.	4.522	4.720	-0.198
23.	5.000	4.953	0.049
24.	6.495	6.593	-0.098
25.	5.854	5.586	0.268
26.	5.495	5.453	0.042

Table 2. Statistical results of CoMFA model

PLS statistics	CoMFA model
q^2	0.568
N	3
r^2	0.975
SEE	0.159
F-value	196.628
r^2_{pred}	0.672
Field contribution	
Steric	0.420
Electrostatic	0.580

q^2 = cross-validated correlation coefficient; N= number of statistical components; r^2 = non-cross validated correlation coefficient; SEE=standard estimated error; F=Fisher value; $r^2_{\text{predictive}}$ = predictive correlation coefficient

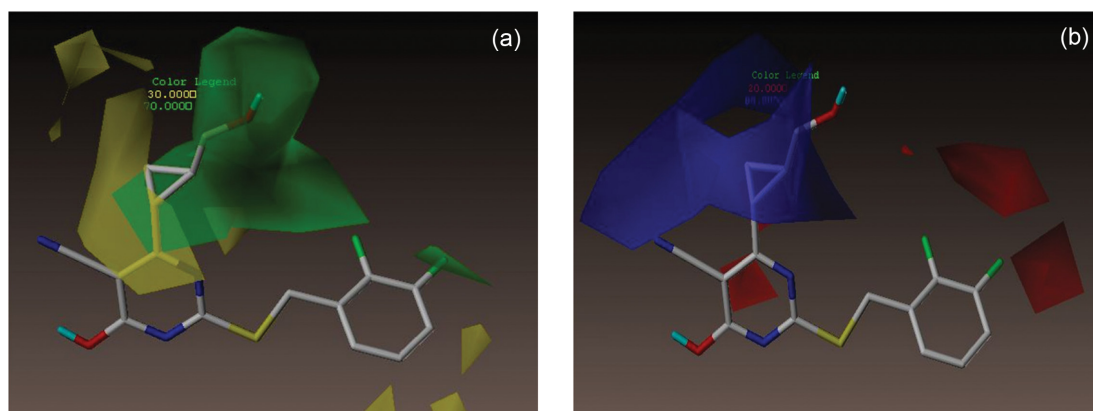


Fig. 2. CoMFA contour maps for compound 17. (a) In steric contour map, the green contour indicates regions where bulky groups increases the activity and yellow contours indicates bulky groups which decreases the activity. (b) In electrostatic contour map the blue contour indicates the regions where an electropositive group increases the activity and a red contour indicates regions where an electronegative group increases the activity.

tion of benzyl ring is not favorable. We have observed that the compounds which does not have bulky group in cyclopropyl ring have lower activity. The electrostatic interactions is represented by red and blue contours, among which blue colored regions show areas where more positively charged groups are favored, and red region highlight areas where groups with more negative charges are favored. The electrostatic contour plot of highly active compound 17 is depicted in Fig. 2(B). The blue colored region in electrostatic contour plot indicates the presence of electropositive groups in the cyclopropyl ring is most favorable and hence the electropositive group in these regions is very important for enhancing the biological activity. The red colored region indicates the electronegative atom in 2nd position of benzyl attached to sulfur is favorable in increasing the activity.

4. Conclusion

The CoMFA contour map gives valuable information to understand 3D-QSAR relationships between structures and their biological activity. The contour maps suggest the presence of bulky and electropositive group in the cyclopropyl ring attached to pyrimidine ring will help in improving the activity of the compound. The present results of the 3D-QSAR model can be useful for predicting the activities of new CXCR2 antagonists.

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