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A Quantitative Structure–Activity Relationship Study on *Clostridium histolyticum* Collagenase Inhibitors: Roles of Electrotopological State Indices

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Abstract—A quantitative structure–activity relationship (QSAR) study has been made on eight different series of *Clostridium histolyticum* collegenase (ChC) inhibitors. These series are comprised of four different groups of sulfonylated amino acids and their corresponding hydroxamates. In each series, the inhibition potency of the compounds has been found to be significantly correlated with the electrotopological state (E-state) indices of nitrogen and sulfur atoms of the sulfonylated amino group in the molecules, showing the importance of the electronic characterstics of these atoms in controlling the inhibition potency of the compounds.

Introduction

Recently, we have made a number of quantitative structure–activity relationship (QSAR) studies[1–4] on the inhibitors of matrix metalloproteinases (MMPs), which are a family of zinc containing enzymes that degrade and remodel structural proteins in the extracellular matrix (ECM), such as membrane collagenase, aggrecan, fibronectin, and liminin.[5,6] These MMPs have been recently found to be interesting targets in the search of novel anticancer, antiarthritis, and other pharmacological agents useful in the management of inflammatory process.[5,7–10] Like MMPs, there are also other enzymes, such as bacterial collagenase, that degrade ECM. As narrated by Scozzafava and Supuran,[9,11–15] *Clostridium histolyticum* collagenase (EC 3.4.24.3) is one of the bacterial collagenases, isolated from *C. histolyticum*. It is a 116 kDa zinc-protein, belonging to M-31 metalloproteinase family,[16,17] that is able to hydrolyze triple helical regions of collagen under physiological conditions as well as an entire range of synthetic peptide substrates.[18–24] In fact, the crude homogenate of *C. histolyticum*, which contains several distinct collagenase isozymes,[16,17] is the most efficient system known for the degradation of connective tissue, being also involved in the pathogenicity of this and related clostridia, such as *Clostridium perfringens*, which causes human gas gangrene and food poisoning among others.[25]

Like MMPs, the *C. histolyticum* collagenase (abbreviated as ChC) is also a multiunit protein. Both MMPs and ChC are considered to have similar mechanism of action for the hydrolysis of proteins and synthetic substrates.[16,17,21,22,25,26] In our recent QSAR studies on MMP inhibitions,[1–4] we found that Kier's first-order valence molecular connectivity index ($^1\chi^v$) of substituents or molecules and the electrotopical state (E-state) indices of some atoms could be good descriptors of the inhibition potencies of the compounds, unraveling the mechanism of inhibition of the enzymes. In the present communication, an overdominance is shown of the E-state indices to describe the inhibition of ChC by different groups of sulfonylated amino acids and their hydroxamates.

Materials and Methods

Scozzafava and Supuran[11–14] reported the ChC inhibition potencies of the following series of sulfonylated amino acids and their hydroxamates:

(a) Sulfonylated *N*-(4-nitrobenzyl)-L-alanine derivatives (**1A**) and corresponding hydroxamates (**1B**).[11]

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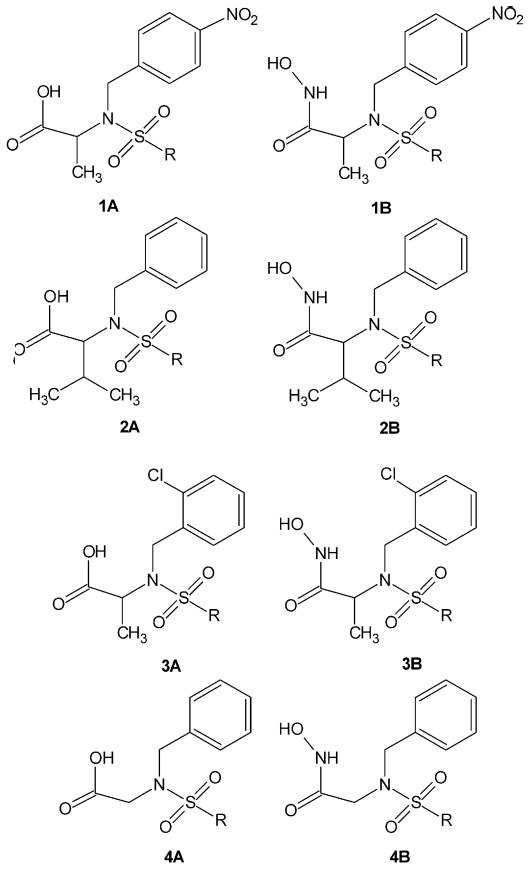
(b) Sulfonylated *N*-benzyl-L-valine derivatives (**2A**) and corresponding hydroxamates (**2B**).[12]
(c) Sulfonylated *N*-(2-chlorobenzyl)-L-alanine derivatives (**3A**) and corresponding hydroxamates (**3B**).[13]
(d) Sulfonylated *N*-benzyl-glycine derivatives (**4A**) and corresponding hydroxamates (**4B**).[14]



All these series of compounds are listed in Tables 1–8 along with the values of their enzyme inhibition constant K_i obtained from Dixon plot using a linear regression programme, from three different assays.[11–14] Along with the K_i values are the E-state indices of the sulfur and nitrogen atoms of sulfonylated amino group, S_S and S_N, respectively. In our very comprehensive analysis we found these two parameters to be most important to govern the inhibition potency of the compounds. The E-state indices are calculated as follows.[27,28]

To calculate S_i of an atom i, one first defines the intrinsic state of the atom, I_i, as

$$I_i = \left(\delta_i^v + 1\right)/\delta_i \tag{1}$$

where δ_i is the σ electron count of the atom and δ_i^v is the valence vertex connectivity index of the atom, which is calculated for the second and third rows of atoms as[29]

$$\delta_i^v = \left(Z_i^v - h_i\right)/\left(Z_i - Z_i^v - 1\right) \tag{2}$$

In eq 2, Z_i^v is the number of valence electrons of the atom i, h_i is the number of hydrogen atoms attached to it in a molecule or group, and Z_i is its atomic number. After calculating I_i, one calculates a factor ΔI_i for the atom using the equation,

$$\Delta I_i = \Sigma_{j=1}\left(I_i - I_j\right)/n^2 \tag{3}$$

where n refers to the number of the atoms in the path i to j, including both i and j. I_i and ΔI_i are then used to find the E-state index S_i of the atom i according to the equation:

$$S_i = I_i + \Delta I_i \tag{4}$$

Results and Discussion

For all the series of compounds, the inhibition constants were found to be significantly correlated with the E-state indices of the sulfur and nitrogen and an indicator variable I used for an R-substituent like C_6F_5 or 3-CF_3–C_6H_4. The correlations obtained were as follows:

1A (Table 1)

$$\begin{aligned}\log(1/K_i) = {} & 3.933(\pm1.385)S_N - 2.271(\pm0.767)S_S \\ & + 1.061(\pm0.372)I - 11.385(\pm5.703) \\ n = 31,\ & r = 0.858,\ r_{cv}^2 = 0.65,\ R_A^2 = 0.707, \\ s = 0.24,\ & F_{3,27} = 25.03(4.60)\end{aligned} \tag{5}$$

1B (Table 2)

$$\begin{aligned}\log(1/K_i) = {} & 4.060(\pm0.946)S_N - 2.312(\pm0.524)S_S \\ & + 0.510(\pm0.263)I - 9.633(\pm3.962) \\ n = 31,\ & r = 0.886,\ r_{cv}^2 = 0.60,\ R_A^2 = 0.761, \\ s = 0.17,\ & F_{3,27} = 32.92(4.60)\end{aligned} \tag{6}$$

2A (Table 3)

$$\begin{aligned}\log(1/K_i) = {} & 2.980(\pm1.250)S_N - 1.781(\pm0.689)S_S \\ & + 0.996(\pm0.330)I - 8.293(\pm5.331) \\ n = 29,\ & r = 0.875,\ r_{cv}^2 = 0.68,\ R_A^2 = 0.734, \\ s = 0.22,\ & F_{3,25} = 27.22(4.68)\end{aligned} \tag{7}$$

2B (Table 4)

$$\begin{aligned}\log(1/K_i) = {} & 4.171(\pm1.007)S_N - 2.374(\pm0.554)S_S \\ & + 0.480(\pm0.261)I - 10.763(\pm4.358) \\ n = 29,\ & r = 0.891,\ r_{cv}^2 = 0.75,\ R_A^2 = 0.770, \\ s = 0.17,\ & F_{3,25} = 32.06(4.68)\end{aligned} \tag{8}$$

Table 1. Sulfonylated *N*-(4-nitrobenzyl)-L-alanine derivatives (**1A**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 5
1	CH_3	−5.058	0.941	4.74[b]	3.80
2	CF_3	−7.398	−0.101	5.33	5.02
3	CCl_3	−5.324	1.066	5.29	4.90
4	*n*-C_4F_9	−8.920	−0.809	5.64	5.69
5	*n*-C_8F_{17}	−9.734	−1.247	5.74	5.82
6	Me_2N	−5.410	0.941	4.44	4.60
7	C_6H_5	−5.587	0.978	4.68[b]	5.15
8	$PhCH_2$	−5.440	1.017	4.82	4.97
9	4-F–C_6H_4	−5.785	0.865	5.00	5.16
10	4-Cl–C_6H_4	−5.612	0.992	5.05	5.26
11	4-Br–C_6H_4	−5.598	1.002	5.00	5.27
12	4-I–C_6H_4	−5.595	1.004	4.92	5.27
13	4-CH_3–C_6H_4	−5.618	0.987	4.89	5.27
14	4-O_2N–C_6H_4	−5.887	0.831	5.28	5.25
15	3-O_2N–C_6H_4	−5.996	0.787	5.26	5.33
16	2-O_2N-C_6H_4	−6.176	0.721	5.32	5.48
17	3-Cl–4-O_2N–C_6H_3	−5.920	0.850	5.48	5.40
18	4-AcNH–C_6H_4	−5.785	0.921	5.51	5.38
19	4-BocNH–C_6H_4	−5.866	0.910	5.62	5.52
20	3-BocNH–C_6H_4	−5.954	0.892	5.59	5.65
21	4-Ac–C_6H_4	−5.776	0.916	5.68	5.34
22	C_6F_5	−7.224	0.120	6.40	6.55
23	3-CF_3–C_6H_4	−6.232	0.645	6.52	6.37
24	2,5-Cl_2–C_6H_3	−5.669	1.027	5.44	5.53
25	4-CH_3O–C_6H_4	−5.692	0.957	5.28	5.31
26	2,4,6-$(CH_3)_3$–C_6H_2	−5.742	1.029	5.22	5.70
27	4-CH_3O-3-BocNH–C_6H_3	−6.055	0.869	5.68	5.78
28	2-OH–3,5-Cl_2–C_6H_2	−5.967	0.878	5.60	5.62
29	3-HOOC–C_6H_4	−5.955	0.817	5.70	5.35
30	4-HOOC–C_6H_4	−5.857	0.853	5.85[b]	5.27
31	1-Naphthyl	−5.799	1.012	5.92	5.77
32	2-Naphthyl	−5.737	1.001	5.89	5.58
33	5-Me_2N-1-Naphthyl	−5.782	1.011	5.96	5.72
34	2-Thienyl	−5.395	1.051	5.68[b]	5.00
35	Quinoline-8-yl	−5.725	1.006	5.92	5.57

[a]Taken from ref 11.
[b]Not used in the derivation of eq 5.

Table 2. Sulfonylated *N*-(4-nitrobenzyl)-L-alanine hydroxamates (**1B**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 6
1	CH_3	−5.073	1.000	7.12[b]	6.16
2	CF_3	−7.413	−0.042	7.12	7.34
3	CCl_3	−5.339	−1.125	7.28	7.28
4	*n*-C_4F_9	−8.935	−0.750	7.96	7.98
5	*n*-C_8F_{17}	−9.749	−1.188	8.15	8.09
6	Me_2N	−5.425	1.000	7.16	6.97
7	C_6H_5	−5.603	1.037	7.27	7.53
8	$PhCH_2$	−5.455	1.076	7.30	7.35
9	4-F–C_6H_4	−5.800	0.924	7.46	7.53
10	4-Cl–C_6H_4	−5.627	1.051	7.49	7.64
11	4-Br–C_6H_4	−5.613	1.061	7.52	7.65
12	4-I–C_6H_4	−5.610	1.063	7.43	7.65
13	4-CH_3-C_6H_4	−5.633	1.046	7.44	7.64
14	4-O_2N-C_6H_4	−5.902	0.890	8.00[b]	7.63
15	3-O_2N-C_6H_4	−6.011	0.846	7.92	7.70
16	2-O_2N-C_6H_4	−6.191	0.780	7.89	7.85
17	3-Cl–4-O_2N–C_6H_3	−5.936	0.989	8.05	8.11
18	4-AcNH–C_6H_4	−5.800	0.979	7.96	7.75
19	4-BocNH–C_6H_4	−5.881	0.968	8.00	7.90
20	3-BocNH–C_6H_4	−5.969	0.951	8.10	8.03
21	4-Ac–C_6H_4	−5.791	0.975	8.05	7.72
22	C_6F_5	−7.239	0.179	8.30	8.34
23	3-CF_3–C_6H_4	−6.247	0.704	8.22	8.18
24	2,5-Cl_2–C_6H_3	−5.684	1.086	7.89	7.92
25	4-CH_3O–C_6H_4	−5.707	1.015	7.68	7.68
26	2,4,6-$(CH_3)_3$–C_6H_2	−5.757	1.088	7.77	8.10
27	4-CH_3O–3-BocNH–C_6H_3	−6.070	0.927	8.10	8.17
28	2-OH–3,5-Cl_2–C_6H_2	−5.982	0.937	7.92	8.00
29	3-HONHOC–C_6H_4	−5.986	0.885	8.05	7.70
30	4-HONHOC–C_6H_4	−5.888	0.917	8.22[b]	7.70
31	1-Naphthyl	−5.814	1.071	8.15	8.16
32	2-Naphthyl	−5.752	1.059	8.10	7.97
33	5-Me_2N-1-Naphthyl	−5.797	1.070	8.15	8.12
34	2-Thienyl	−5.410	1.110	7.96[b]	7.38
35	Quinoline-8-yl	−5.740	1.064	8.10	7.96

[a]Taken from ref 11.
[b]Not used in the derivation of eq 6.

3A (Table 5)

$$\log(1/K_i) = 1.676(\pm0.808)S_N - 1.067(\pm0.443)S_S + 0.988(\pm0.375)I - 2.588(\pm3.360)$$
$$n = 30,\ r = 0.849,\ r^2_{cv} = 0.64,\ R^2_A = 0.692,$$
$$s = 0.25,\ F_{3,26} = 22.30(4.64) \quad (9)$$

3B (Table 6)

$$\log(1/K_i) = 4.218(\pm0.947)S_N - 2.404(\pm0.523)S_S + 0.539(\pm0.257)I - 10.718(\pm4.041)$$
$$n = 30,\ r = 0.899,\ r^2_{cv} = 0.77,\ R^2_A = 0.784,$$
$$s = 0.17,\ F_{3,26} = 36.69(4.64) \quad (10)$$

4A (Table 7)

$$\log(1/K_i) = 3.329(\pm1.170)S_N - 1.998(\pm0.649)S_S + 0.920(\pm0.301)I - 9.313(\pm4.778)$$
$$n = 29,\ r = 0.893,\ r^2_{cv} = 0.72,\ R^2_A = 0.773,$$
$$s = 0.20,\ F_{3,25} = 32.96(4.60) \quad (11)$$

4B (Table 8)

$$\log(1/K_i) = 3.655(\pm0.831)S_N - 2.102(\pm0.459)S_S + 0.522(\pm0.219)I - 7.960(\pm3.432)$$
$$n = 27,\ r = 0.918,\ r^2_{cv} = 0.81,\ R^2_A = 0.826,$$
$$s = 0.14,\ F_{3,23} = 40.85(4.76) \quad (12)$$

Table 3. Sulfonylated *N*-benzyl-L-valine derivatives (**2A**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 7
1	CH_3	−4.962	1.162	4.74[b]	4.01
2	CF_3	−7.302	0.120	5.48	5.07
3	CCl_3	−5.228	1.287	5.25	4.85
4	*n*-C_4F_9	−8.824	−0.588	5.49	5.67
5	*n*-C_8F_{17}	−9.638	−1.026	5.72	5.81
6	Me_2N	−5.313	1.162	4.39	4.63
7	C_6H_5	−5.491	1.199	4.70	5.06
8	$PhCH_2$	−5.344	1.238	4.82	4.91
9	4-F–C_6H_4	−5.688	1.086	4.92	5.07
10	4-Cl–C_6H_4	−5.516	1.213	4.96	5.14
11	4-Br–C_6H_4	−5.501	1.223	5.00	5.15
12	4-I–C_6H_4	−5.498	1.226	5.00	5.15
13	4-CH_3–C_6H_4	−5.522	1.208	4.82	5.14
14	4-O_2N–C_6H_4	−5.791	1.052	5.30	5.15
15	3-O_2N–C_6H_4	−5.899	1.008	5.29	5.22
16	2-O_2N–C_6H_4	−6.080	0.942	5.28	5.34
17	3-Cl-4-O_2N–C_6H_3	−5.824	1.072	5.52	5.27
18	4-AcNH–C_6H_4	−5.689	1.142	5.46	5.24
19	4-BocNH–C_6H_4	−5.770	1.131	5.62	5.35
20	3-BocNH–C_6H_4	−5.857	1.113	5.64	5.45
21	C_6F_5	−7.127	0.341	6.40	6.41
22	3-CF_3–C_6H_4	−6.136	0.866	6.22	6.21
23	2,5-Cl_2–C_6H_3	−5.573	1.248	5.40	5.35
24	4-CH_3O–C_6H_4	−5.596	1.178	5.20	5.18
25	2,4,6-$(CH_3)_3$–C_6H_2	−5.646	1.250	5.21	5.49
26	4-CH_3O-3-BocNH–C_6H_3	−5.959	1.090	5.48	5.57
27	2-OH–3,5-Cl_2–C_6H_2	−5.870	1.099	5.49	5.44
28	3-HOOC–C_6H_4	−5.858	1.038	5.35	5.23
29	4-HOOC–C_6H_4	−5.761	1.074	5.62[b]	5.17
30	1-Naphthyl	−5.703	1.233	5.48	5.54
31	2-Naphthyl	−5.641	1.222	5.85[b]	5.40
32	5-Me_2N-1-Naphthyl	−5.685	1.232	5.70	5.50
33	2-Thienyl	−5.298	1.272	5.68[b]	4.93

[a]Taken from ref 12.
[b]Not used in the derivation of eq 7.

Table 4. Sulfonylated *N*-benzyl-L-valine hydroxamates (**2B**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 8
1	CH_3	−4.977	1.221	7.00[b]	6.15
2	CF_3	−7.317	0.179	7.33	7.36
3	CCl_3	−5.243	1.346	7.28	7.30
4	*n*-C_4F_9	−8.839	−0.529	8.00	8.02
5	*n*-C_8F_{17}	−9.653	−0.967	8.10	8.12
6	Me_2N	−5.328	1.221	7.19	6.98
7	C_6H_5	−5.506	1.258	7.27	7.56
8	$PhCH_2$	−5.359	1.297	7.40	7.37
9	4-F–C_6H_4	−5.704	1.145	7.38	7.56
10	4-Cl–C_6H_4	−5.531	1.272	7.36	7.67
11	4-Br–C_6H_4	−5.516	1.282	7.44	7.68
12	4-I–C_6H_4	−5.513	1.284	7.52	7.68
13	4-CH_3–C_6H_4	−5.537	1.267	7.48	7.67
14	4-O_2N–C_6H_4	−5.806	1.111	7.82	7.66
15	3-O_2N–C_6H_4	−5.914	1.067	7.96	7.73
16	2-O_2N–C_6H_4	−6.095	1.001	7.74	7.88
17	3-Cl–4-O_2N–C_6H_3	−5.839	1.130	7.96	7.81
18	4-AcNH–C_6H_4	−5.704	1.200	8.00	7.78
19	4-BocNH–C_6H_4	−5.785	1.189	8.10	7.93
20	3-BocNH–C_6H_4	−5.872	1.172	8.15	8.07
21	C_6F_5	−7.142	0.400	8.30	8.34
22	3-CF_3–C_6H_4	−6.151	0.925	8.22	8.18
23	2,5-Cl_2–C_6H_3	−5.588	1.307	7.96	7.96
24	4-CH_3O–C_6H_4	−5.611	1.236	7.77	7.71
25	2,4,6-$(CH_3)_3$–C_6H_2	−5.661	1.309	7.75[b]	5.14
26	4-CH_3O–3-BocNH–C_6H_3	−5.974	1.149	8.15	8.21
27	2-OH–3,5-Cl_2–C_6H_2	−5.885	1.158	8.15	8.04
28	3-HONHOC–C_6H_4	−5.890	1.106	8.05	7.83
29	4-HONHOC–C_6H_4	−5.792	1.139	8.10[b]	7.74
30	1-Naphthyl	−5.718	1.292	8.05	8.20
31	2-Naphthyl	−5.656	1.280	8.22	8.00
32	5-Me_2N-1-Naphthyl	−5.701	1.291	8.10	8.16
33	2-Thienyl	−5.313	1.331	8.00[b]	7.40

[a]Taken from ref 12.
[b]Not used in the derivation of eq 8.

In all the above equations, n is the number of data points, r is the correlation coefficient, r^2_{cv} is the square of cross-validated correlation coefficient obtained from leave-one-out jackknife procedure, R^2_A is the adjusted r^2, given as $R^2_A = r^2(1-1/F)$, s is the standard deviation, and F is the F-ratio between the variances of calculated and observed activities. The data given within the parenthesis following the F-valve is the standard statistical value of F at 99% level and the data given with (±) sign within the parentheses following the coefficients of the variables are 95% confidence intervals. The value of R^2_A (also called explained variance, EV) accounts for the variance in activity in percentage when multiplied by 100.

Now all the above equations exhibit parallel correlations, indicating that in all the cases the inhibition potency of the compounds will increase with the increase in the E-state index of the nitrogen and will decrease with the increase in the E-state index of the sulfur. Since E-state indices are the measure of the availability of the π and/or lone pair electrons on the atoms, it is certain that both nitrogen and sulfur might be playing some electronic roles in the interaction of the compounds with the receptors. Nitrogen can be assumed to be involved in some charge-transfer phenomenon with the enzymes where the stability of the charge-transfer complex formed will depend upon the value of S_N, that is, on the availability of π or the lone pair electrons on the nitrogen. Similarly, the sulfur can be assumed to be involved in some charge–charge repulsion interaction with the receptors, where increase in the availability of π or lone pair electrons will destabilize the bonding due to the increase in the repulsion. Such type of roles of nitrogen and sulfur have been described by us in the inhibition of MMPs also.[1–4]

Similarly, as some of our previous studies,[1,2] this study also points out a specific role of C_6F_5- and 3-CF_3–C_6H_4-like R-substituents, for which an indicator parameter I has been used with a value of unity. 'I' is equal to 1 for R = C_6F_5 or 3-CF_3–C_6H_4 and zero for R being any other substituent. There are only two such compounds in each series which have R = C_6F_5 or 3-CF_3–C_6H_4 (compds **22** and **23** in Tables 1 and 2 and compds **21** and **22** in Tables 3–8). A positive coefficient of I in all the equations suggests that these two types of R-substituents would be beneficial to the inhibition potency for all the series of compounds discussed here. However, the point to be noted is that in all acid series (**1A**–**4A**) the coefficient of I is essentially the same and so is the case

Table 5. Sulfonylated *N*-(2-chlorobenzyl)-L-alanine derivatives (**3A**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 9
1	CH_3	−4.901	1.118	4.68	4.52
2	CF_3	−7.241	0.076	5.48	5.27
3	CCl_3	−5.167	1.243	5.28	5.01
4	*n*-C_4F_9	−8.763	−0.632	5.52	5.70
5	*n*-C_8F_{17}	−9.577	−1.070	5.82	5.84
6	Me_2N	−5.253	1.118	4.40	4.89
7	C_6H_5	−5.431	1.155	4.62[b]	5.14
8	$PhCH_2$	−5.283	1.194	4.80	5.05
9	4-F–C_6H_4	−5.628	1.042	4.92	5.16
10	4-Cl–C_6H_4	−5.455	1.168	4.89	5.19
11	4-Br–C_6H_4	−5.441	1.179	5.00	5.19
12	4-I–C_6H_4	−5.438	1.181	5.00	5.19
13	4-CH_3–C_6H_4	−5.461	1.164	4.74	5.19
14	4-O_2N–C_6H_4	−5.730	1.008	5.29	5.22
15	3-O_2N–C_6H_4	−5.839	0.964	5.30	5.26
16	2-O_2N–C_6H_4	−6.019	0.898	5.31	5.34
17	3-Cl-4-O_2N-C_6H_3	−5.764	1.027	5.51	5.28
18	4-AcNH–C_6H_4	−5.628	1.097	5.52	5.26
19	4-BocNH–C_6H_4	−5.710	1.086	5.57	5.33
20	3-BocNH–C_6H_4	−5.797	1.369	5.62	5.89
21	C_6F_5	−7.067	0.297	6.40	6.44
22	3-CF_3–C_6H_4	−6.075	0.822	6.30	6.26
23	2,5-Cl_2–C_6H_3	−5.512	1.204	5.39	5.31
24	4-CH_3O–C_6H_4	−5.535	1.133	5.22	5.22
25	2,4,6-$(CH_3)_3$-C_6H_2	−5.585	1.206	5.17	5.39
26	4-CH_3O–3-BocNH–C_6H_3	−5.898	1.045	5.52	5.46
27	2-OH–3,5-Cl_2–C_6H_2	−5.810	1.055	5.62	5.38
28	3-HOOC–C_6H_4	−5.798	0.994	5.39	5.27
29	4-HOOC–C_6H_4	−5.700	1.030	5.42	5.22
30	1-Naphthyl	−5.642	1.189	5.80	5.43
31	2-Naphthyl	−5.580	1.177	5.82[b]	5.34
32	5-Me_2N-1-Naphthyl	−5.625	1.188	5.68	5.41
33	2-Thienyl	−5.238	1.228	5.62[b]	5.06
34	Quinoline-8-yl	−5.568	1.182	5.70[b]	5.33

[a]Taken from ref 13.
[b]Not used in the derivation of eq 9.

Table 6. Sulfonylated *N*-(2-chlorobenzyl)-L-alanine hydroxamates (**3B**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 10
1	CH_3	−4.916	1.176	7.04[b]	6.06
2	CF_3	−7.256	0.135	7.12	7.30
3	CCl_3	−5.182	1.301	7.15	7.23
4	*n*-C_4F_9	−8.778	−0.573	7.96	7.97
5	*n*-C_8F_{17}	−9.592	−1.011	8.10	8.08
6	Me_2N	−5.268	1.176	7.11	6.91
7	C_6H_5	−5.446	1.213	7.24	7.49
8	$PhCH_2$	−5.298	1.253	7.27	7.31
9	4-F–C_6H_4	−5.643	1.100	7.44	7.49
10	4-Cl–C_6H_4	−5.470	1.227	7.46	7.61
11	4-Br–C_6H_4	−5.456	1.238	7.48	7.62
12	4-I–C_6H_4	−5.453	1.240	7.52	7.62
13	4-CH_3–C_6H_4	−5.476	1.223	7.35	7.61
14	4-O_2N–C_6H_4	−5.745	1.066	7.82	7.59
15	3-O_2N–C_6H_4	−5.854	1.023	7.96	7.67
16	2-O_2N–C_6H_4	−6.034	0.956	7.80	7.82
17	3-Cl–4-O_2N–C_6H_3	−5.779	1.086	8.00	7.76
18	4-AcNH–C_6H_4	−5.643	1.156	7.96	7.73
19	4-BocNH–C_6H_4	−5.725	1.145	8.05	7.88
20	3-BocNH–C_6H_4	−5.812	1.127	8.10	8.01
21	C_6F_5	−7.082	0.355	8.30	8.35
22	3-CF_3–C_6H_4	−6.090	0.880	8.22	8.18
23	2,5-Cl_2–C_6H_3	−5.527	1.263	7.82	7.90
24	4-CH_3O–C_6H_4	−5.550	1.192	7.66	7.65
25	2,4,6-$(CH_3)_3$–C_6H_2	−5.600	1.264	7.77	8.08
26	4-CH_3O–3-BocNH–C_6H_3	−5.913	1.104	8.10	8.16
27	2-OH–3,5-Cl_2–C_6H_2	−5.825	1.113	8.05	7.98
28	3-HONHOC–C_6H_4	−5.830	1.061	8.10[b]	7.77
29	4-HONHOC–C_6H_4	−5.731	1.094	8.15[b]	7.67
30	1-Naphthyl	−5.657	1.248	8.10	8.15
31	2-Naphthyl	−5.595	1.236	8.05	7.95
32	5-Me_2N-1-Naphthyl	−5.640	1.246	8.00	8.10
33	2-Thienyl	−5.253	1.286	7.96[b]	7.33
34	Quinoline-8-yl	−5.583	1.241	8.15	7.94

[a]Taken from ref 13.
[b]Not used in the derivation of eq 10.

in all the hydroxamate series (**1B–4B**). Another point, which is more important, is that the coefficient of I in any **A** series is just the double of that in corresponding **B** series. Thus, in all **A** series the C_6F_5 and 3-CF_3–C_6H_4 substituents are equally 2-fold more effective (in log unit) than in the **B** series.

Since the ChC enzyme catalyzes the cleavage of the Xaa-Gly (Xaa: amino acid residue) peptide bond of the repeating sequence of the collagen:-Gly-Pro-Xaa-Gly-Pro-Xaa-, it appears that the $S_{3'}$, $S_{2'}$, and $S_{1'}$ subsites[30] of the enzyme are occupied by Gly, Pro, and Xaa, respectively.[16,18–22,31] In the design of the ChC inhibitors mentioned here, the following structural elements were opted by Scozzafava and Supuran, which were based on strong MMP inhibitory properties of some arylsulphonyl-aminohydroxamic acids studied by Jeng et al.[32] and Hanessian et al.[33]

1. A strong zinc-binding function (of the carboxylic acid or better hydroxamic acid type).
2. A relatively compact spacer between this function and the rest of the molecule, that is, any amino acid moiety.
3. A variant of the already optimized benzyl group to interact with $S_{2'}$ site.
4. An arylsulfonyl moiety to interact with $S_{3'}$ site.

All these structural elements for a hydroxamate and their interactions with ChC can be schematically shown as in Figure 1.

Now, the nitrogen and sulfur that are indicated in this study to play dominant roles in the binding appear to seek some electronic sites in the enzyme to interact with, for which a typical conformation of the molecule may be required.

In general, an R-substituent has been indicated by Scozzafava and Supuran[11–14] to have a hydrophobic interaction with $S_{3'}$ site of the enzyme. Our study, however, has shown that there can be a more advantageous effect if R = C_6F_5 or 3-CF_3–C_6H_4. In these substituents, the fluorine(s) present specifically at the 3-position might have either hydrogen bond interactions or charge-charge interactions, or undergo the charge-transfer complex formation phenomenon with the

Table 7. Sulfonylated *N*-benzyl-glycine derivatives (**4A**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 11
1	CH_3	−4.796	1.046	4.60[b]	3.75
2	CF_3	−7.136	0.005	5.30	4.96
3	CCl_3	−5.062	1.171	5.22[b]	4.70
4	*n*-C_4F_9	−8.658	−0.703	5.52	5.65
5	*n*-C_8F_{17}	−9.472	−1.142	5.70	5.81
6	Me_2N	−5.148	1.046	4.33	4.46
7	C_6H_5	−5.326	1.083	4.62	4.93
8	$PhCH_2$	−5.178	1.123	4.72	4.77
9	4-F–C_6H_4	−5.523	0.970	4.82	4.95
10	4-Cl–C_6H_4	−5.350	1.097	4.85	5.03
11	4-Br–C_6H_4	−5.336	1.108	4.85	5.04
12	4-I–C_6H_4	−5.333	1.110	4.89	5.04
13	4-CH_3–C_6H_4	−5.356	1.093	4.77	5.03
14	4-O_2N–C_6H_4	−5.625	0.936	5.22	5.04
15	3-O_2N–C_6H_4	−5.734	0.893	5.30	5.12
16	2-O_2N–C_6H_4	−5.914	0.826	5.15	5.25
17	3-Cl-4-O_2N–C_6H_3	−5.659	0.956	5.52[b]	5.18
18	4-AcNH–C_6H_4	−5.523	1.026	5.40	5.14
19	4-BocNH–C_6H_4	−5.605	1.015	5.52	5.27
20	3-BocNH–C_6H_4	−5.692	0.997	5.69	5.38
21	C_6F_5	−6.962	0.225	6.15	6.27
22	3-CF_3–C_6H_4	−5.970	0.750	6.15	6.03
23	2,5-Cl_2–C_6H_3	−5.407	1.133	5.30	5.26
24	4-CH_3O–C_6H_4	−5.430	1.062	5.15	5.07
25	2,4,6-$(CH_3)_3$–C_6H_2	−5.480	1.134	5.15	5.41
26	4-CH_3O–3-BocNH–C_6H_3	−5.793	0.974	5.52	5.51
27	2-OH–3,5-Cl_2–C_6H_2	−5.705	0.983	5.40	5.36
28	3-HOOC–C_6H_4	−5.693	0.922	5.30	5.13
29	4-HOOC–C_6H_4	−5.595	0.959	5.30	5.06
30	1-Naphthyl	−5.537	1.117	5.40	5.47
31	2-Naphthyl	−5.475	1.106	5.40	5.31
32	5-Me_2N-1-Naphthyl	−5.520	1.116	5.30	5.43
33	2-Thienyl	−5.133	1.156	5.40[b]	4.79

[a]Taken from ref 14.
[b]Not used in the derivation of eq 11.

Table 8. Sulfonylated *N*-benzyl-glycine hydroxamates (**4B**) with their E-state indices and observed and calculated ChC inhibition potencies

Compd	R	S_S	S_N	log $(1/K_i)$	
				Obsd[a]	Calcd eq 12
1	CH_3	−4.811	1.105	6.95[b]	6.19
2	CF_3	−7.151	0.063	7.28	7.31
3	CCl_3	−5.077	1.230	7.29	7.21
4	*n*-C_4F_9	−8.673	−0.645	7.88	7.92
5	*n*-C_8F_{17}	−9.487	−1.083	8.04	8.03
6	Me_2N	−5.163	1.105	7.02	6.93
7	C_6H_5	−5.341	1.142	7.27	7.44
8	$PhCH_2$	−5.194	1.181	7.32	7.28
9	4-F-C_6H_4	−5.538	1.029	7.29	7.44
10	4-Cl-C_6H_4	−5.365	1.156	7.35	7.54
11	4-Br-C_6H_4	−5.351	1.166	7.41	7.55
12	4-I-C_6H_4	−5.348	1.169	7.52	7.56
13	4-CH_3-C_6H_4	−5.363	1.151	7.36	7.52
14	4-O_2N-C_6H_4	−5.640	0.995	7.80[b]	7.53
15	3-O_2N-C_6H_4	−5.749	0.951	7.82	7.60
16	2-O_2N-C_6H_4	−5.929	0.885	7.62	7.74
17	3-Cl-4-O_2N-C_6H_3	−5.674	1.015	7.92	7.68
18	4-AcNH-C_6H_4	−5.539	1.085	7.89	7.65
19	4-BocNH-C_6H_4	−5.620	1.074	8.00	7.78
20	3-BocNH-C_6H_4	−5.707	1.056	8.05	7.90
21	C_6F_5	−6.977	0.284	8.22	8.27
22	3-CF_3-C_6H_4	−5.985	0.809	8.15	8.10
23	2,5-Cl_2-C_6H_3	−5.423	1.191	7.67	7.79
24	4-CH_3O-C_6H_4	−5.445	1.121	7.57	7.59
25	2,4,6-$(CH_3)_3$-C_6H_2	−5.495	1.193	7.82	7.95
26	4-CH_3O-3-BocNH-C_6H_3	−5.808	1.033	8.10	8.03
27	2-OH-3,5-Cl_2-C_6H_2	−5.720	1.042	8.05[b]	7.87
28	1-Naphthyl	−5.553	1.176	7.96	8.01
29	2-Naphthyl	−5.490	1.165	7.92	7.84
30	5-Me_2N-1-Naphthyl	−5.535	1.175	7.89	7.97
31	2-Thienyl	−5.148	1.215	7.80[b]	7.30

[a]Taken from ref 14.
[b]Not used in the derivation of eq 12.

receptor. Here, the receptor site involved may be $S_{3'}$ itself, for which we can now assume that it may not be fully hydrophobic but a part of it may be polar also to provide the opportunity to some specific substituents, such as C_6F_5 and 3-CF_3–C_6H_4, for polar interactions, too. However, one can also assume that the advantageous role of C_6F_5 and 3-CF_3–C_6H_4 substituents may be because of their steric effects, rather than any kind of polar effects, due to their proper orientation to the active site.

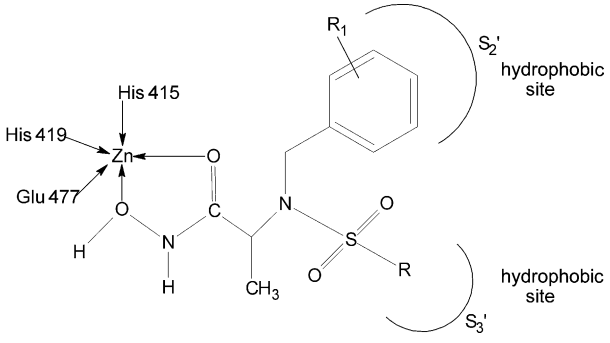


Figure 1. A model showing the binding features of a hydroxamate within the ChC.

In the derivation of all the equations, some compounds as indicated in each table, were excluded since they were exhibiting aberrant behaviors. Since all the outliers are not common in all the tables, it is difficult to assign the reasons for the aberrant behavior of each and every compound. However, there are two compounds, first and last, in each table, with R = CH_3 and 2-thienyl, respectively, that are exhibiting aberrant behavior in all the series (the exception is only Table 5, where compound **1** is not an outlier). These two compounds in all the series have much higher observed potency than predicted for them. The only explanation that can be given for this discrepancy is that the R-substituents in them might be having a good steric fit with the active site ($S_{3'}$) of the receptor.

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References and Notes

1. Gupta, S. P.; Maheswaran, V.; Pande, V.; Kumar, D. J. *Enzyme Inhib. Med. Chem.* **2003**, *18*, 7.

2. Kumar, D.; Gupta, S. P. *Bioorg. Med. Chem.* **2003**, *11*, 421.
3. Gupta, S. P.; Kumar, D.; Kumaran, S. *Bioorg. Med. Chem.* **2003**, *11*, 1975.
4. Kumar, D.; Gupta, S. P. *Eur. J. Med. Chem.* Submitted for publication.
5. Babine, R. E.; Bender, S. L. *Chem. Rev.* **1997**, *97*, 1359.
6. Leung, D.; Abbenanate, G.; Fairlie, D. P. *J. Med. Chem.* **2000**, *43*, 305.
7. Whittaker, M.; Floyd, C. D.; Brown, P.; Gearing, A. J. H. *Chem. Rev.* **1999**, *99*, 2735.
8. Bottomley, K. M.; Johnson, W. H.; Walter, D. S. *J. Enzyme Inhib.* **1998**, *13*, 79.
9. Supuran, C. T.; Scozzafava, A. In *Proteinase and Peptidase Inhibition: Recent Potential Targets for Drug Development*; Smith, H. J., Simons, C., Eds.; Taylor and Francis: London and New York, 2002; p 35.
10. Nagese, H.; Woessner, J. F.Jr *J. Biol. Chem.* **1999**, *274*, 21491.
11. Scozzafava, A.; Supuran, C. T. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 499.
12. Supuran, C. T.; Scozzafava, C. T. *Eur. J. Pharm. Sci.* **2000**, *10*, 67.
13. Scozzafava, A.; Supuran, C. T. *Bioorg. Med. Chem.* **2000**, *8*, 637.
14. Scozzafava, A.; Supuran, C. T. *Eur. J. Med. Chem.* **2000**, *35*, 299.
15. Scozzafava, A.; Supuran, C. T. *J. Med. Chem.* **2001**, *44*, 2253.
16. Van Wart, H. E. In *Handbook of Proteolytic Enzymes*; Barret, A. J., Rawlings, N. D., Woessner, J. F., Eds.; Academic: Oxford, 1998; p 368.
17. Rawling, N. D.; Barret, A. J. *Methods Enzymol.* **1995**, *248*, 183.
18. Bond, M. D.; Van Wart, H. E. *Biochemistry* **1984**, *23*, 3077.
19. Bond, M. D.; Van Wart, H. E. *Biochemistry* **1984**, *23*, 3085.
20. Van Wart, H. E.; Stienbrink, D. R. *Anal. Biochem.* **1981**, *113*, 356.
21. Matsushita, O.; Jung, C. M.; Minami, J.; Katayama, S.; Nishi, N.; Okabe, A. *J. Biol. Chem.* **1998**, *273*, 3643.
22. Jung, C. M.; Matsushita, O.; Katayama, S.; Minami, J.; Sakurai, J.; Okabe, A. J. *J. Bacteriol.* **1999**, *181*, 2816.
23. Schiff, W. M.; Speaker, M. G.; McCormick, S. A. *CLAO J.* **1992**, *18*, 59.
24. Groos, E.B. In *Cornea: Fundamentals of Cornea and External Disease*; Krachmer, J. H., Mannis, M. J., Holland, E. J., Eds.; Mosby-Year Book: St. Louis, MO, 1997; Vol. 6, p 105.
25. Rood, J. I. *Ann. Rev. Microbiol.* **1998**, *52*, 333.
26. Matsushita, O.; Jung, C. M.; Minami, J.; Katayama, S.; Minami, J.; Takashi, Y.; Okabe, A. *J. Bacteriol.* **1999**, *181*, 923.
27. Kier, L. B.; Hall, L. H. *Adv. Drug Res.* **1992**, *22*, 1.
28. Kier, L. B.; Hall, L. H. *Molecular Structure Descriptors: Electrotopological State Indices*; Academic: San Diego, 1999.
29. Kier, L. B.; Hall, L. H. *J. Pharm. Sci.* **1983**, *72*, 1170.
30. In a standard nomenclature, S_n is used to represent a binding site of the enzyme and a P_n to represent the corresponding binding group in substrate.[5] If the enzyme has structural symmetry, then the symmetrical binding sites are represented by $S_{n'}$ and the corresponding binding groups in the substrate by $P_{n'}$.[5]
31. Grobelny, D.; Galardy, R. E. *Biochemistry* **1985**, *24*, 6145.
32. Jeng, A. Y.; Chou, M.; Parker, D. T. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 897.
33. Hanessian, D. L.; Bouzbouz, S.; Boudon, A.; Tucker, G. C.; Peyroulan, D. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1691.