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Toxicological study of nitrobenzene derivatives against tetrahymena pyriformis using topological parameters

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ABSTRACT

Eight topological descriptors namely molar refractivity, solvent accessible surface area (SASA), shape index (order-1), shape index (order-2), shape index (order-3), valence connectivity index (order-0), valence connectivity index (order-1) and valence connectivity index (order-2) of fifty four nitrobenzene derivatives have been calculated with the help of CAChe Pro of Fujitsu software. Observed toxicities of all compounds are in terms of -log (IGC_{50}), mM, which is the inverse logarithm of the concentration causing 50% growth inhibition of Tetrahymena pyriformis after 40 hours. These eight descriptors have been used in developing QSTR models with the help of multi linear regression (MLR) analysis. The quality of regression has been adjudged by correlation coefficient, cross validation coefficient and statistical parameters like standard error, standard error of estimate, degrees of freedom etc. The QSTR model developed from descriptors molar refractivity, solvent accessible surface area, shape index (order-1) and valence connectivity index (order-2) have very high predictive power and can be used to find out the toxicity of any new derivative of nitrobenzene. Reliable QSTR model has been obtained from single descriptor shape index (order-1) which is also present in all best four QSTR models. Therefore, shape index (order-1) appears an important descriptor for the toxicological study of nitrobenzene derivatives.

Keywords: Nitrobenzene derivatives, Tetrahymena pyriformis, topological descriptors, multi linear regression (MLR) analysis

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INTRODUCTION

Topological parameters gained much importance in recent years. Molar refractivity, solvent accessible surface area (SASA), shape indices and valence connectivity indices have been successfully applied in QSAR /QSTR study of various compounds [1-12]. Very recently quantum chemical descriptors have been successfully used for the development of QSTR models of fifty four nitrobenzene derivatives [13]. In this paper topological descriptors have been used for the QSTR study of fifty four nitrobenzene derivatives against *Tetrahymena pyriformis.*

Molar refractivity is calculated by the Lorenz-Lorentz formula [14]

$$MR = \frac{n^2-1}{n^2+2} \times \frac{M}{\rho}$$

where M is the molecular weight, n is the refractive index and ρ is the density. For a radiation of infinite wavelength, molar refractivity represents the real volume of the molecules.

The solvent accessibility surface area (SASA) is the surface area of a bimolecule that is accessible to a solvent and is usually quoted in square angstroms. Lee and Richards first described the solvent accessible surface area (SASA) of

a molecular surface [15]. SASA is typically calculated by using the 'rolling ball' algorithm developed by Sharke and Rupley [16] This algorithm uses a sphere of solvent of a particular radius to probe the surface of the molecule. The choice of the probe radius does have an effect on the observed surface area, as using a smaller probe radius detects more surface details and therefore reports a larger surface. A typical value is 1.4 Å, which approximates the radius of a water molecule.

Shape indices compare the molecule graph with "minimal" and "maximal" graphs, where the meaning of "minimal" and "maximal" depends on the order "n". This is intended to capture different aspects of the molecular shape. Kier was first to propose shape indices for molecular graphs, the so called kappa shape indices [17, 18]. The first order kappa shape index (1κ or κ_1) is given by,

$$^1K = \frac{A(A-1)^2}{(^1P)^2}$$

Where, iP = Length of paths of bond length i in the hydrogen suppressed molecule and A is the number of non hydrogen atoms in the molecule.

The second order kappa shape index (2κ or κ_2) is given by

$$^2K = \frac{(A-1)(A-2)^2}{(^2P)^2}$$

The third order kappa shape index (3κ or κ_3) is given by

$$^3K = \frac{(A-1)(A-3)^2}{(^3P)^2} \qquad \text{if "A" is odd}$$

$$^3K = \frac{(A-3)(A-2)^2}{(^3P)^2} \qquad \text{if "A" is even}$$

Valence connectivity indices, originally defined by Randic and subsequently refined by Kier and Hall, is a series of numbers designated by "order" and "subgraph type" [19, 20]. There are four subgraph types; path, cluster, path/cluster, and chain. These types emphasize different aspects of atom connectivity within a molecule, the amount of branching, ring structures present and flexibility. It is calculated from the hydrogen suppressed molecular graph and defined as follows,

$$^m\chi^v = \sum_{i=1}^{Ns} \prod_{k=1}^{m+1} \left[\frac{1}{\delta_k^v}\right]^{1/2}$$

Where, $\delta_k^v = \frac{(Z_k^v - H_k)}{(Z_k - Z_k^v - 1)}$ - valence connectivity for the k-th atom in the molecular graph, Z_k = the total number of electrons in the k-th atom, Z^v_k = the number of valence electrons in the k-th atom, H_k = the number of hydrogen atoms directly attached to the kth non-hydrogen atom, m = 0 - atomic valence connectivity indices (called order-0), m = 1 - one bond path valence connectivity indices (called order-1), m = 2 - two bond fragment valence connectivity indices (called order-2).

The above discussed descriptors have been calculated and used in QSTR study of fifty four nitrobenzene derivatives. The predicted toxicities obtained from developed QSTR models were found close to reported observed toxicities.

EXPERIMENTAL SECTION

The study material of this paper is fifty four nitrobenzene derivatives given in Table-1. The toxicity of these compounds was measured in terms of -log (IGC_{50}), mM, which is the inverse logarithm of the concentration causing 50% growth inhibition of *Tetrahymena pyriformis* after 40 hours. The 3D modeling and geometry optimization of all the compounds have been done with the help of CAChe Pro software of Fujitsu, using the DFT Methods [21-23]. Evaluation of values of descriptors has been done using semiemperical PM3 Hamiltonian [24]. The Project Leader program associated with CAChe Pro has been used for multi linear regression (MLR) analysis. The statistical parameters have been calculated by Smith's Statistical Package (version 2.80).

RESULTS AND DISCUSSION

Fifty four derivatives of nitrobenzene are given in Table-1 alongwith their observed toxicity in terms of -log (IGC_{50}). The values of eight descriptors of compounds, which have been calculated, are given in Table-2. For the development of QSTR models multi linear regression (MLR) analysis has been performed using different combinations of descriptors. The MLR analysis has indicated that the toxicity of nitrobenzene can be successfully modeled even in mono-parametric regression using descriptor shape index (order-1). This mono-parametric QSTR model obtained by using descriptor shape index (order-1) is given by following regression equation,

$$^{Mono-}PT = 0.434547*{}^{1}\kappa - 3.03887$$
$$r^2 = 0.825611,\ rCV^2 = 0.791353,\ Std.\ Error = 0.0637,\ SEE = 0.3185,$$
$$DOF = 0.8223,\ N = 54,\ VC = 1.$$

In the above regression equation, r^2 is correlation coefficient, rCV^2 is cross-validation coefficient, Std. Error is standard error, SEE is standard error of estimate, DOF is degrees of freedom, N is data points (compounds), and VC is variable count. Shape index (order-1) appears an important descriptor for the toxicological study of nitrobenzene derivatives of this set.

The addition of descriptor shape index (order-2) in the above mono-parametric model yields a model with improved predictability. The resulting bi-parametric QSTR model obtained by using descriptors shape index (order-1) and shape index (order-2) is given by following regression equation,

$$^{Bi-}PT = 0.537349*{}^{1}\kappa - 0.309119*{}^{2}\kappa - 2.83417$$
$$r^2 = 0.8406,\ rCV^2 = 0.771939,\ Std.\ Error = 0.0604,\ SEE = 0.3045,\ DOF = 0.8376,$$
$$N = 54,\ VC = 2.$$

Using combination of three descriptors, the best tri-parametric QSTR model is obtained with improved predictive power. This best tri-parametric QSTR model is given by following regression equation,

$$^{Tri-}PT = -0.0757781*MR + 0.474884*{}^{1}\kappa + 0.789414*{}^{2}\chi - 2.19889$$
$$r^2 = 0.869798,\ rCV^2 = 0.840194,\ Std.\ Error = 0.0537,\ SEE = 0.2753,\ DOF = 0.8672,$$
$$N = 54,\ VC = 3.$$

This QSTR model involves molar refractivity as first descriptor, shape index (order-1) as second descriptor and valence connectivity index (order-2) as third descriptor.

By the combination of four descriptors, the best tetra-parametric QSTR model is obtained with excellent predictive power. This best tetra-parametric QSTR model is given by following regression equation,

$$^{Tetra-}PT = -0.0615758*MR - 0.00786129*SASA + 0.509892*{}^{1}\kappa + 0.806314*{}^{2}\chi - 1.75936$$
$$r^2 = 0.872068,\ rCV^2 = 0.84377,\ Std.\ Error = 0.0531,\ SEE = 0.2728,\ DOF = 0.8697,$$
$$N = 54,\ VC = 4.$$

This QSTR model involves molar refractivity as first descriptor, solvent accessible surface area as second descriptor, shape index (order-1) as third descriptor and valence connectivity index (order-2) as fourth descriptor.

From the values of correlation coefficient (r^2), cross-validation coefficient (rCV^2) and other statistical parameters for the above four QSTR models, it is clear that the predictive power of all models is high. Among these four QSTR models the tetra-parametric model, i.e. $^{Tetra-}PT$, is the best which can be used to find the toxicity of any new

derivative of nitrobenzene. The predicted toxicity values, for nitrobenzene derivatives of this set, obtained from above four QSTR models are listed in Table-3 along with their observed toxicity. A correlation summary of the best four QSTR models is presented in Table-4.

Table-1: Fifty four nitrobenzene derivatives alongwith their observed toxicity

S. No.	Compounds	Observed Toxicity $-\log(IGC_{50})$
1	Nitrobenzene	0.14
2	2-Chloronitrobenzene	0.68
3	2-Bromonitrobenzene	0.75
4	3-Chloronitrobenzene	0.73
5	4-Ethylnitrobenzene	0.80
6	4-Chloronitrobenzene	0.43
7	4-Bromonitrobenzene	0.38
8	4-Fluoronitrobenzene	0.25
9	2,4,6-Trimethylnitrobenzene	0.86
10	2,4-Dichloronitrobenzene	0.99
11	3-Bromonitrobenzene	1.03
12	2,3-Dichloronitrobenzene	1.07
13	3-Methyl-4-bromonitrobenzene	1.16
14	3,4-Dichloronitrobenzene	1.16
15	1,2-Dinitrobenzene	1.25
16	1,4-Dinitrobenzene	1.30
17	2,5-Dibromonitrobenzene	1.37
18	4-Butoxynitrobenzene	1.42
19	2,4,6-Trichloronitrobenzene	1.43
20	2,3,4-Trichloronitrobenzene	1.51
21	5-methyl-1,2-dinitrobenzene	1.52
22	2,4,5-Trichloronitrobenzene	1.53
23	2,3,4,5-Tetrachloronitrobenzene	1.78
24	2,3,5,6-Tetrachloronitrobenzene	1.82
25	6-Iodo-1,3-dinitrobenzene	2.12
26	2,4,6-Trichloro-1,3-dinitrobenzene	2.19
27	1,2-Dinitro-4,5-dichlorobenzene	2.21
28	6-Bromo-1,3-dinitrobenzene	2.31
29	2,4,5-Trichloro-1,3-dinitrobenzene	2.59
30	4,6-Dichloro-1,2-dinitrobenzene	2.42
31	2,3,5,6-Tetrachloro-1,4-dinitrobenzene	2.74
32	1,3-Dimethyl-2-nitrobenzene	0.30
33	2,3-Dimethylnitrobenzene	0.56
34	3,5-Dichloronitrobenzene	1.13
35	3-Chloro-4-fluoronitrobenzene	0.80
36	2.5-Dichloronitrobenzene	1.13
37	1,2,3-Trifluoro-4-nitrobenzene	1.89
38	2,3,4,6-Tetrafluoronitrobenzene	1.87
39	1-Chloro-2,4-dinitrobenzene	2.16
40	2,4-Dinitro-1-fluorobenzene	1.71
41	Pentafluoronitrobenzene	2.43
42	1,5-Difluoro-2,4-dinitrobenzene	2.08
43	1,2-Dimethyl-4-nitrobenzene	0.59
44	1-Fluoro-3-iodo-5-nitrobenzene	1.09
45	1-Fluoro-2-nitrobenzene	0.23
46	1,2,3-Trichloro-5-nitrobenzene	1.55
47	1,3-Dichloro-4,6-dinitrobenzene	2.72
48	2,6-Dimethylnitrobenzene	0.30
49	2-Methyl-3-chloronitrobenzene	0.68
50	2-Methylnitrobenzene	0.05
51	2-Methyl-5-chloronitrobenzene	0.82
52	6-Chloro-1,3-dinitrobenzene	1.98
53	3-Methylnitrobenzene	0.05
54	4-Methylnitrobenzene	0.17

Table-2: Values of descriptors and observed toxicity of nitrobenzene derivatives

C. No.	MR	SASA	κ_1	κ_2	κ_3	$^0\chi$	$^1\chi$	$^2\chi$	$-\log(IGC_{50})$
1	33.383	148.015	7.111	3.240	2.000	4.650	2.499	1.593	0.14
2	38.188	162.417	8.100	3.408	1.991	5.707	2.983	2.116	0.68
3	41.006	166.457	8.100	3.408	1.991	6.537	3.398	2.563	0.75
4	38.188	166.960	8.100	3.408	2.286	5.707	2.977	2.173	0.73
5	43.025	184.088	9.091	4.133	2.500	6.280	3.471	2.277	0.80
6	38.188	167.195	8.100	3.408	2.286	5.707	2.977	2.170	0.43
7	41.006	171.912	8.100	3.408	2.286	6.537	3.392	2.649	0.38
8	33.599	156.028	8.100	3.408	2.286	4.951	2.599	1.734	0.25
9	48.506	195.586	10.083	3.806	2.250	7.418	3.743	3.004	0.86
10	42.992	181.561	9.091	3.600	2.215	6.764	3.461	2.696	0.99
11	41.006	171.883	8.100	3.408	2.286	6.537	3.392	2.653	1.03
12	42.992	178.418	9.091	3.600	1.975	6.764	3.467	2.615	1.07
13	46.047	187.118	9.091	3.600	2.215	7.460	3.809	3.045	1.16
14	42.992	183.147	9.091	3.600	2.215	6.764	3.461	2.670	1.16
15	40.707	170.949	10.083	4.297	2.493	5.837	3.005	2.003	1.25
16	40.707	177.412	10.083	4.297	2.778	5.837	2.999	2.031	1.30
17	48.628	190.659	9.091	3.600	2.215	8.424	4.291	3.623	1.37
18	53.719	237.488	12.071	6.478	4.388	8.103	4.610	2.953	1.42
19	47.797	196.352	10.083	3.806	2.250	7.820	3.944	3.226	1.43
20	47.797	194.607	10.083	3.806	2.041	7.820	3.950	3.115	1.51
21	45.749	189.864	11.077	4.481	2.721	6.759	3.416	2.506	1.52
22	47.797	197.939	10.083	3.806	2.250	7.820	3.944	3.196	1.53
23	52.602	210.917	11.077	4.022	2.083	8.877	4.434	3.618	1.78
24	52.602	208.545	11.077	4.022	2.083	8.877	4.434	3.644	1.82
25	53.116	201.637	11.077	4.481	2.721	8.295	4.183	3.312	2.12
26	55.122	215.772	13.067	4.888	2.571	9.006	4.456	3.558	2.19
27	50.317	206.186	12.071	4.680	2.750	7.950	3.966	3.084	2.21
28	48.330	195.690	11.077	4.481	2.721	7.723	3.898	3.004	2.31
29	55.122	217.236	13.067	4.888	2.571	9.006	4.456	3.531	2.59
30	50.317	204.214	12.071	4.680	2.750	7.950	3.966	3.113	2.42
31	59.927	228.742	14.063	5.104	2.488	10.063	4.945	3.979	2.74
32	43.465	176.417	9.091	3.600	1.975	6.496	3.333	2.498	0.30
33	43.465	178.13	9.091	3.600	1.975	6.496	3.333	2.476	0.56
34	43.465	185.513	9.091	3.600	2.500	6.496	3.321	2.603	1.13
35	38.404	173.521	9.091	3.600	2.215	6.008	3.083	2.263	0.80
36	42.992	181.824	9.091	3.600	2.215	6.764	3.461	2.696	1.13
37	34.032	168.489	10.083	3.806	2.041	5.552	2.816	1.952	1.89
38	34.248	174.183	11.077	4.022	2.083	5.853	2.922	2.074	1.87
39	45.512	191.741	11.077	4.481	2.721	6.893	3.482	2.557	2.16
40	40.924	183.07	11.077	4.481	2.721	6.137	3.105	2.150	1.71
41	34.465	180.483	12.071	4.245	2.020	6.154	3.034	2.176	2.43
42	41.140	188.606	12.071	4.680	2.750	6.438	3.210	2.269	2.08
43	43.465	182.629	9.091	3.600	2.215	6.496	3.327	2.526	0.59
44	46.007	186.102	9.091	3.600	2.500	7.409	3.778	3.130	1.09
45	33.599	153.96	8.100	3.408	1.991	4.951	2.605	1.709	0.23
46	47.797	199.473	10.083	3.806	2.250	7.820	3.944	3.173	1.55
47	50.317	206.474	12.071	4.680	2.750	7.950	3.966	3.084	2.72
48	43.465	176.742	9.091	3.600	1.975	6.496	3.333	2.498	0.30
49	43.229	177.71	9.091	3.600	1.975	6.630	3.400	2.548	0.68
50	38.424	162.369	8.100	3.408	1.991	5.573	2.916	2.044	0.05
51	43.229	181.639	9.091	3.600	2.215	6.630	3.394	2.624	0.82
52	45.512	192.104	11.077	4.481	2.721	6.893	3.482	2.557	1.98
53	38.424	166.658	8.100	3.408	2.286	5.573	2.910	2.096	0.05
54	38.424	166.721	8.100	3.408	2.286	5.573	2.910	2.093	0.17

Where; MR = Molar refractivity, SASA = Solvent accessible surface area, κ_1 = Shape index (order-1), κ_2 = Shape index (order-2), κ_3 = Shape index (order-3), $^0\chi$ = Valence connectivity index (order-0), $^1\chi$ = Valence connectivity index (order-1), $^2\chi$ = Valence connectivity index (order-2)

Table-3: Observed and predicted toxicity (in terms of −log(IGC_{50})) of fifty four nitrobenzene derivatives

Comp. No.	Observed Toxicity	Predicted Toxicity			
		$^{Mono-}$PT	$^{Bi-}$PT	$^{Tri-}$PT	$^{Tetra-}$PT
1	0.14	0.051	-0.015	-0.094	-0.068
2	0.68	0.481	0.465	0.424	0.449
3	0.75	0.481	0.465	0.564	0.604
4	0.73	0.481	0.465	0.469	0.459
5	0.80	0.912	0.773	0.655	0.616
6	0.43	0.481	0.465	0.467	0.455
7	0.38	0.481	0.465	0.631	0.630
8	0.25	0.481	0.465	0.470	0.473
9	0.86	1.343	1.407	1.285	1.280
10	0.99	0.912	0.938	0.989	0.975
11	1.03	0.481	0.465	0.635	0.634
12	1.07	0.912	0.938	0.925	0.935
13	1.16	0.912	0.938	1.033	1.025
14	1.16	0.912	0.938	0.968	0.942
15	1.25	1.343	1.256	1.086	1.146
16	1.30	1.343	1.256	1.108	1.118
17	1.37	0.912	0.938	1.293	1.304
18	1.42	2.207	1.650	1.794	1.602
19	1.43	1.343	1.407	1.514	1.496
20	1.51	1.343	1.407	1.426	1.421
21	1.52	1.775	1.733	1.573	1.600
22	1.53	1.343	1.407	1.490	1.46
23	1.78	1.775	1.875	1.931	1.909
24	1.82	1.775	1.875	1.952	1.948
25	2.12	1.775	1.733	1.651	1.703
26	2.19	2.639	2.676	2.638	2.682
27	2.21	2.207	2.206	2.155	2.163
28	2.31	1.775	1.733	1.770	1.797
29	2.59	2.639	2.676	2.617	2.649
30	2.42	2.207	2.206	2.178	2.202
31	2.74	3.072	3.145	3.079	3.131
32	0.30	0.912	0.938	0.797	0.827
33	0.56	0.912	0.938	0.779	0.796
34	1.13	0.912	0.938	0.879	0.84
35	0.80	0.912	0.938	0.995	0.972
36	1.13	0.912	0.938	0.989	0.973
37	1.89	1.343	1.407	1.551	1.536
38	1.87	1.775	1.875	2.103	2.083
39	2.16	1.775	1.733	1.631	1.641
40	1.71	1.775	1.733	1.658	1.663
41	2.43	2.207	2.340	2.64	2.609
42	2.08	2.207	2.206	2.207	2.209
43	0.59	0.912	0.938	0.819	0.801
44	1.09	0.912	0.938	1.103	1.104
45	0.23	0.481	0.465	0.451	0.470
46	1.55	1.343	1.407	1.472	1.429
47	2.72	2.207	2.206	2.155	2.161
48	0.30	0.912	0.938	0.797	0.824
49	0.68	0.912	0.938	0.854	0.872
50	0.05	0.481	0.465	0.350	0.376
51	0.82	0.912	0.938	0.914	0.902
52	1.98	1.775	1.733	1.631	1.638
53	0.05	0.481	0.465	0.391	0.385
54	0.17	0.481	0.465	0.388	0.382

Table-4: Correlation summary of the best four QSTR models for nitrobenzene derivatives.

QSAR Model	r^2	rCV^2	Std. Error	SEE	DOF	Variable Used
$^{Mono-}$PT	0.825611	0.791353	0.0637	0.3185	0.8223	κ_1
$^{Bi-}$PT	0.8406	0.771939	0.0604	0.3045	0.8376	κ_1, κ_2
$^{Tri-}$PT	0.869798	0.840194	0.0537	0.2753	0.8672	MR, κ_1, $^2\chi$
$^{Tetra-}$PT	0.872068	0.84377	0.0531	0.2728	0.8697	MR, SASA, κ_1, $^2\chi$

CONCLUSION

It is clear from the above study that, the best combination of topological descriptors is molar refractivity, solvent accessible surface area, shape index (order-1) and valence connectivity index (order-2) for the QSTR study of nitrobenzene derivatives against *Tetrahymena pyriformis.* Reliable QSTR model has been obtained from single

descriptor shape index (order-1) which is also present in all best four QSTR models. Therefore, shape index (order-1) appears an important descriptor for the toxicological study of nitrobenzene derivatives.

REFERENCES

[1]D Singh; MA Khan, *J. Chem. Pharm. Res.,* **2011**, 3(5), 1-14.
[2]RK Singh; MA Khan, *Res. Jour. of Chem. Sci.,* **2013**, 3(5), 47-56.
[3]RK Singh; MA Khan; PP Singh, *S. Afr. J. Chem.*, **2014**, 67, 12-20.
[4]MA Khan; SA Khan; PP Singh, *International Journal of ChemTec Research,* **2010**, 2(2), 996-1009.
[5]P Kumar; P Singh;, JP Singh, *J. Chem. Pharm. Res.,* **2011**, 3(4), 296-303.
[6]P Kumar; P Singh; JP Singh, J. *Chem. Pharm. Res.*, **2011**, 3(4), 296-303
[7]SA Khan; PK Verma; R Tewari, *Int. Jour. Of Life Sci. and Pharma Res*., **2011**, 1(1), 12-28.
[8]P Kumar; P Singh; JP Singh, *Int. Jour. of Res. in Pharma. and Biomed. Sci.,* **2012**, 1, 65-72
[9]A Thakur; M Thakur; N Kakani; A Joshi; S Thakur; A Gupta, *ARKIVOC,* **2004** (xiv), 36-43
[10]KA Hussain; WAH Radhi; SMH Ismael, *J. Chem. Pharm. Res.,* **2012**, 4(3), 1702-1707.
[11]SQ Nassab; Z Bayat; J Movaffagh, *J. Chem. Pharm. Res.,* **2011**, 3(1), 64-71.
[12]AB Padavala; VV Prasanth; SA Jayanthi; A Vadlamani; S Chitti, *J. Chem. Pharm. Res.,* **2010**, 2(2), 147-162.
[13]SK Mishra; V Mishra; PN Tripathi; MA Khan, *Res. Jour. of Chem. Sci.,* **2014**, 4(2), 29-37.
[14]RJ Padron; A Carrasco; RF Pellon, *J. Pharm. Pharmaceut. Sci.,* **2002**, 5, 258-266.
[15]B Lee; FM Richards, *J. Mol. Biol.*, **1971**, 55(3), 379-400.
[16]A Shrake; JA Rupley, *J. Mol. Biol.,* **1973**, 79(2), 351-371.
[17]LB Kier, *Quant. Struct.-Act. Relat.,* **1986**, 5, 1-7.
[18]LB Kier, *Med. Res. Rev.*, **1987**, 7, 417-440.
[19]AT Balaban, *Jour. of. Mol. Struc: THEOCHEM,* **1985**, 120, 117-142.
[20]M Petitjean; *J. Chem. Inf. Comput. Sci.,* **1992**, 32, 331-337.
[21]RG Parr and W Yang, Density-Functional Theory of Atoms and Molecules, Oxford University Press: New York, **1989**.
[22]JP Perdew; S Kurth, A Primer in Density Functional Theory, Springer: Berlin, **2003**.
[23]W Koch MC Holthausen, A Chemist's Guide to Density Functional Theory, Wiley-VCH: New York, **2000**.
[24]RC Bingham; MJS Dewar; DH Lo, *J. Am. Chem. Soc.,* **1975**, 97(6), 1285-1293.