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A conceptual DFT approach towards analysing toxicity

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Abstract. The applicability of DFT-based descriptors for the development of toxicological structure–activity relationships is assessed. Emphasis in the present study is on the quality of DFT-based descriptors for the development of toxicological QSARs and, more specifically, on the potential of the electrophilicity concept in predicting toxicity of benzidine derivatives and the series of polyaromatic hydrocarbons (PAH) expressed in terms of their biological activity data (pIC_{50}). First, two benzidine derivatives, which act as electron-donating agents in their interactions with biomolecules are considered. Overall toxicity in general and the most probable site of reactivity in particular are effectively described by the global and local electrophilicity parameters respectively. Interaction of two benzidine derivatives with nucleic acid (NA) bases/selected base pairs is determined using Parr's charge transfer formula. The experimental biological activity data (pIC_{50}) for the family of PAH, namely polychlorinated dibenzofurans (PCDF), polyhalogenated dibenzo-*p*-dioxins (PHDD) and polychlorinated biphenyls (PCB) are taken as dependent variables and the HF energy (E), along with DFT-based global and local descriptors, viz., electrophilicity index (w) and local electrophilic power (w^+) respectively are taken as independent variables. Fairly good correlation is obtained showing the significance of the selected descriptors in the QSAR on toxins that act as electron acceptors in the presence of biomolecules. Effects of population analysis schemes in the calculation of Fukui functions as well as that of solvation are probed. Similarly, some electron-donor aliphatic amines are studied in the present work. We see that global and local electrophilicities along with the HF energy are adequate in explaining the toxicity of several substances, both electron donors or acceptors when they interact with biosystems, in gas as well as solution phases.

Keywords. Conceptual DFT approach; toxicity; structure–activity relationships; polyaromatic hydrocarbons.

1. Introduction

Quantitative structure–activity relationships (QSARs) are widely used to predict toxicity from chemical structure and corresponding physicochemical properties. In recent years, quantum chemical descriptors have been used in QSAR studies because the quantum chemical quantities are able to provide accurate quantitative description of the molecular structures and chemical properties. Atomic charges, molecular orbital energies, frontier orbital densities, atom-atom polarizabilites, molecular polarizabilities, dipole moments etc., have been used as descriptors within a QSAR parlance. Density functional theory (DFT)-based descriptors have found immense usefulness in the prediction of reactivity of atoms and molecules.[1–5] The importance of DFT descriptors in the development of QSAR has been recently reviewed.[3–5] Chemical hardness (h), chemical potential (m), polarizability (a) and softness (S) are known as global reactivity descriptors. Fukui function (FF) and local softness are called local reactivity descriptors. Recently, Parr *et al*[6] have defined a new descriptor to quantify the global electrophilic power of the molecule as electrophilicity index (w), which provided the direct relationship between the rates of reaction and the electrophilic power of the inhibitors.[7] Using the properties of FF, more powerful descriptors of reactivity and site selectivity have been proposed.[8] Subsequently, attempts have been made to probe the expediency of electrophilicity and other global quantities in the QSAR parlance.[9,10] The usefulness of electrophilicity index in unraveling the toxicity of polychlorinated biphenyls[11,12] and benzidine[13] has been analysed.

Many aromatic hydrocarbons, especially arylamines and amides, come under the class of potential mutagenic and carcinogenic environmental pollu-

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tants. The knowledge of biochemical mechanism of cancer induction by aromatic hydrocarbons especially arylamines and amides forms the basis of understanding tumour formation.[14–16] Some of the studies indicate that benzidine-based dyes can be metabolized to benzidine and human exposure to such dyes is associated with bladder cancer.[14–16] Benzidine must undergo metabolic activation to produce its deleterious effects, probably through binding of oxidized reactive intermediates to nucleic acids (DNA and RNA) and protein target molecules. Similar to benzidine, its derivatives viz., 3,3′-dimethoxybenzidine and 3,3′-dichlorobenzidine have also been identified as potential carcinogens. 3,3′-Dimethoxybenzidine is used as an intermediate in the manufacture of azo dyes and *o*-dianisidine diisocyanate. Various tests have shown that 3,3′-dimethoxybenzidine is moderately toxic after acute oral exposure. Animal studies have highlighted its adverse effects on the liver, kidney and bladder, as well as causing gastritis, intestinal hemorrhage and weight loss. Hence, 3,3′-dimethoxybenzidine is reasonably anticipated to be a human carcinogen.

On the other hand 3,3′-dichlorobenzidine degrades rapidly in sunlight. When heated to decomposition, these compounds emit toxic fumes of hydrochloric acid and other chlorinated compounds as well as nitrogen oxides. It is used mainly in the manufacture of pigments for printing ink, textiles, paper, paint, rubber, and plastic and as a curing agent for isocyanate-containing polymers and solid urethane plastics. 3,3′-Dichlorobenzidine is accordingly expected to be a human carcinogen based on sufficient evidence of carcinogenicity in experimental animals.[17]

Polychlorinated dibenzofurans (PCDFs), polyhalogenated dibenzo-*p*-dioxins (PHDDs) and polychlorinated biphenyls (PCBs) are chemicals of concern because of their elevated concentrations, wide distribution and toxicity. Biochemical and pathological studies on aquatic organisms have consistently reported that lateral substituted congeners are more potent than the non-lateral congeners.[18,19] Polychlorinated dibenzo-*p*-dioxins and polychlorinated dibenzofurans are ubiquitous contaminants, which are present in various environmental systems and biota.[20] PCDDs/DFs are released directly into the atmosphere from a variety of combustion sources and manufacturing processes, such as municipal solid waste incinerators,[21] automobile emissions[22] and chemical production processes.[20] They are mainly transported over long distances and deposited in terrestrial and aquatic ecosystems through dry or wet deposition. Therefore, atmospheric transport and deposition constitute the primary distribution pathway in moving PCDDs/DFs from numerous emission sources to environmental compartments.[23]

Toxicity of polychlorinated biphenyls has seen an upsurge of interest in recent years.[24–28] These compounds exhibit toxicity similar to that of polychlorinated dibenzo-*p*-dioxin. This information on PCB has prompted several investigators to understand the toxic nature of PCB and their interaction with cellular components.[29] The origin of toxicity of PCDDs has been attributed to the electron accepting nature in the charge transfer complex formation with a receptor in living cells.[30] Hence, electron affinity of PCDDs/PCBs is used as an important quantity in understanding their toxic effects. Due to their extreme toxicity and the existence of many isomers, experimental investigations on toxic PCDDs are difficult. It is well known that 2,3,7,8-TCDD is the most toxic of all the 75 PCDD isomers and causes dermal toxicity, immunotoxicity, reproductive ill-effects and carcinogenicity.[31]

In the present work, an attempt has been made to explore the uses of DFT-based reactivity descriptors to investigate the structure-activity relationship in the selected derivatives of benzidine. All the previously determined biological activity data[31], that is the negative of the log of molar concentration of chemical necessary to displace 50% of radiolabeled TCDD from the Ah receptor (pIC_{50}), are utilized for this purpose. Experimental biological activity (pIC_{50}) for PCDFs, PHDDs and PCBs are correlated with their corresponding calculated pIC_{50} values determined using three parameters multiple regression analysis in gas and solvent phases using MPA and HPA. A set of aliphatic amines which act as electron donors in their interaction with biomolecules has been studied for their $\log(IGC_{50}^{-1})$[32] activity using two parameter multiple regression analysis in gas phase using MPA and NPA schemes.

2. Theoretical background

Chemical hardness (h) has been shown to be a useful global index of reactivity in atoms, molecules and clusters.[2,33] The theoretical definition of chemical hardness has been provided by the density functional theory as the second order derivative of electronic energy with respect to the number of electrons N, for a constant external potential $V(\mathbf{r})$

$$h = \tfrac{1}{2}(\partial^2 E/\partial N^2)_{V(r)} = \tfrac{1}{2}(\partial m/\partial N)_{V(r)}, \quad (1)$$

where E is the total energy, N is the number of electrons of the chemical species and m is the chemical potential, which is identified as the negative of the electronegativity (c) as defined by Iczkowski and Margrave.[34] By applying finite difference approximation to (1) we get the operational definition for h as,

$$h = (IP - EA)/2. \quad (2)$$

The corresponding global softness is expressed as

$$S = 1/2h = (\partial^2 N/\partial E^2)_{V(r)} = (\partial N/\partial m)_{V(r)}. \quad (3)$$

Equation (2) can be rewritten using Koopmans'[2] theorem as

$$h = (e_{HOMO} - e_{LUMO})/2, \quad (4)$$

where e_{HOMO} and e_{LUMO} are the energies of highest occupied and lowest unoccupied molecular orbitals respectively.

The Fukui function, which measures the sensitivity of a system's chemical potential to an external perturbation at a particular site, is defined as[35,36]

$$f(\mathbf{r}) = (\partial r(\mathbf{r})/\partial N)_{n(\mathbf{r})} = (dm/dn(\mathbf{r}))_N. \quad (5)$$

Since the above derivatives are discontinuous, three different types of Fukui function have been defined[37–39]

$$f^+(\mathbf{r}) = r_{N+1}(\mathbf{r}) - r_N(\mathbf{r}), \quad \text{for nucleophilic attack,} \quad (6a)$$

$$f^-(\mathbf{r}) = r_N(\mathbf{r}) - r_{N-1}(\mathbf{r}), \quad \text{for electrophilic attack,} \quad (6b)$$

$$f^0(\mathbf{r}) = (r_{N+1}(\mathbf{r}) - r_{N-1}(\mathbf{r}))/2, \quad \text{for radical attack.} \quad (6c)$$

Parr *et al*[6] introduced the global electrophilicity index (w) in terms of chemical potential and hardness as

$$w = m^2/2h. \quad (7)$$

Recently, Chattaraj *et al*[8] have proposed a generalized concept of philicity containing electrophilic, nucleophilic and radical reactions. The condensed-to-atom variants for the atomic site k have been written as,

$$w_k^a = w f_k^a, \quad (8)$$

where a = +, – and 0 refer to nucleophilic, electrophilic and radical attacks respectively. The w_k^a vary from point to point in a molecule but the sum of any w_k^a over all atoms is conserved.

Biomolecules are the principal targets for the activated derivatives of aryl amines. In particular interaction of these molecules with constituent molecules of aryl hydrocarbon hydroxylase (AHH) receptor and DNA is of special interest. Global interactions between the constituents of AHH receptors and NA bases/base pairs have been determined using the parameter ΔN, which represents the fractional number of electrons, transferred from a system A to a system B, and is given by[40]

$$\Delta N = (m_B - m_A)/2(h_A + h_B). \quad (9)$$

3. Computational details

The general atom-numbering schemes of the 3,3′-dichlorobenzidine and 3,3′-dimethoxybenzidine are shown in figures S1a and S1b respectively. The geometries of 3,3′-dichlorobenzidine and 3,3′-dimethoxybenzidine are optimized by using Becke's three parameter hybrid density functional, B3LYP/6-31G*.[41–43] It is noted from the previous studies on similar molecular systems that B3LYP/6-31G* theory provides comparatively reliable results[11–13] and hence in the present investigation the same basis set has been used. All the calculations have been performed using G98W & G03W suites of programs.[44] The relative energies of these compounds are calculated as a function of torsional angle f (rotation through the C4–C7 bond). To calculate the relative energy, the geometry at various f values are optimized at B3LYP/6-31G* level. The relative energy values for the 3,3′-dimethoxybenzidine and 3,3′-dichlorobenzidine are calculated as $\Delta E(f) = [E(f) – E(f = 90{\cdot}0)]$ using the total energies of respective optimized conformations. To select a proper electronic descriptor based on DFT, for the possible toxicity of the 3,3′-dimethoxybenzidine and 3,3′-dichlorobenzidine, the various reactivity and selectivity descriptors such as chemical hardness (h), chemical potential (m), electrophilicity index (w) and the local electrophilic power (w_k^a) have been calculated using standard equations

and methodology described in the previous studies.[5–8] Since, the Hirshfeld[45] population scheme (stockholder partitioning scheme) is known to provide non-negative Fukui function (FF) values, it has been used to calculate FF values as implemented in the DMOL[46] package employing BLYP/DND method as followed in the earlier investigations. The electron density (r) at the bond critical point[47] of the active group/site for 3,3′-dimethoxybenzidine and 3,3′-dichlorobenzidine are calculated using AIM software package.[48] Further, the geometries of the nucleic acid bases/DNA base pairs viz., adenine, guanine, cytosine, thymine, uracil, ATH and GCWC are optimized using the B3LYP/6-31G* basis set. We have also calculated the amount of charge transfer[40] between 3,3′-dimethoxybenzidine and 3,3′-dichlorobenzidine and various bases, viz., adenine (A), guanine (G), thymine (T), cytosine (C), uracil (U) and DNA base pairs GCWC, ATH using Parr's formula given in (9).

The geometries for polychlorinated dibenzofurans (PCDFs), polyhalogenated dibenzo-*p*-dioxins (PHDDs), and polychlorinated biphenyls (PCBs) are minimised with the 6–31G* basis set in the framework of B3LYP theory comprising Becke's three-parameter hybrid exchange and LYP correlation functionals. Also a set of aliphatic amines is optimized using HF/6-311G** method only in gas phase. Solvent phase optimization has been carried out for other selected systems using polarizable continuum model (PCM) developed by Tomasi and coworkers.[49] The atomic charges for all the above molecules have been obtained in the framework of B3LYP theory using Mulliken population analysis (MPA).[50] Natural population analysis (NPA)[51,52] is also used to derive atomic charges for the set of aliphatic amines. Three- and two-parameter QSARs have been performed[53] using the least square error estimation method to predict the toxicity values.

4. Results and discussion

4.1 *Benzidine derivatives*

Earlier studies have shown that benzidine is supposed to be highly toxic due to their structural flexibility.[13] In the present study, we would like to analyse the possible toxicity of two derivatives of benzidine, namely 3,3′-dimethoxybenzidine and 3,3′-dichlorobenzidine using various chemical reactivity descriptors.

4.2 *Torsional rotation versus global reactivity descriptors*

Variation of relative energy, chemical hardness and electrophilicity index during rotation about the C4–C7 bond of 3,3′-dichlorobenzidine and 3,3′-dimethoxybenzidine are shown in figures 1a and 1b respectively. The most stable conformation corresponds to $f = 90°$ rotated conformation. It is seen from table 1 that the rotational energy barrier for the two compounds ranges from 0 to 2·63 kcal/mol and 0 to 2·73 kcal/mol respectively. These values clearly ensure that the compounds are highly flexible and as a consequence they can orient to any rotated conformations to exhibit toxicity. These results are comparable to the rotational energy barrier for benzidine[13] (0 to 2·66 kcal/mol). This rotational freedom allows benzidine and their selected derivatives to freely interact with various components in real life systems and is thus the possible reason for their toxicity.

4.3 *Local philicity versus torsional rotation*

Site selectivity profiles like condensed philicity (w_k^+) have been used in the earlier studies on polychlorinated biphenyls (PCB) to understand their toxic nature.[11] It can be seen from the previous study that local philicity provides the exact site of attack.[11] Since, PCB is an electron acceptor we used w_k^+ profiles,[11] whereas w_k^- profiles are used here owing to the electron-donating nature of the benzidines.

As in the case of benzidine,[13] a unified philicity concept has also been used to analyse the toxicity of these benzidine derivatives.[13] Due to the symmetry of the selected systems, the variations in the philicity (w_k^-) with rotational angle for symmetry atoms are presented in figures 2a to 2c. In the case of the 3,3′-dichlorobenzidine as seen from figure 2a, the C2 site corresponds to the most probable site and H14 is the least probable site for electrophilic attack. Figures 2b and 2c show that for 3,3′-dimethoxybenzidine, C1 is the most probable site and H34 is the least probable site for electrophilic attack.

4.4 *The topology of electron density*

Topology of the electron density (r) distribution is an important tool in structure–activity studies because, in principle, r contains all information that can be known about a molecule. Further r is an important entity that exists in 3-dimensional space and can be

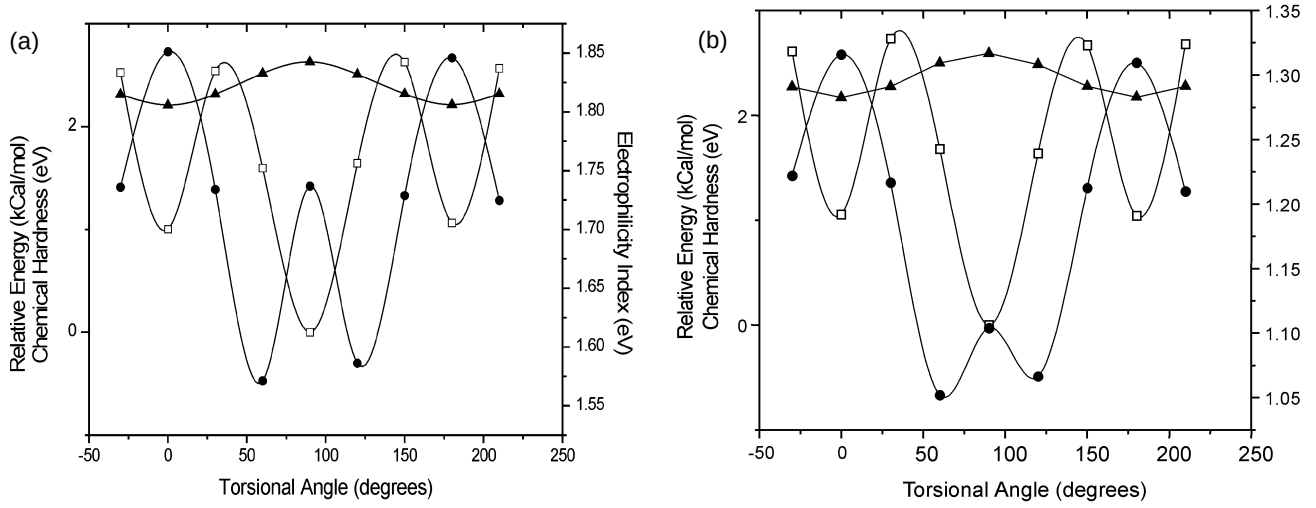

Figure 1. Variation of relative energy (kcal/mol), chemical hardness (eV) and electrophilicity index (eV) with torsional angles (degrees) for 3,3′-dichlorobenzidine (**a**) and for 3,3′-dimethoxybenzidine (**b**) (—□— relative energy (kcal/mol); —▲— chemical hardness (eV); —●— electrophilicity index (eV)).

Table 1. Calculated relative energy (ΔE), chemical hardness (h), chemical potential (m) and electrophilicity index (w) of 3,3′-dichlorobenzidine and 3,3′-dimethoxybenzidine for various torsional angles (f).

	3,3′-Dichlorobenzidine				3,3′-Dimethoxybenzidine			
$f^{\dagger}$	ΔE*	h**	m**	w**	ΔE*	h**	m**	w**
–30	2·524	2·312	–2·833	1·736	2·608	2·272	–2·356	1·222
0	1·002	2·210	–2·861	1·851	1·054	2·169	–2·389	1·316
30	2·540	2·314	–2·833	1·734	2·729	2·273	–2·352	1·217
60	1·598	2·517	–2·812	1·571	1·680	2·496	–2·292	1·052
90	0·000	2·628	–3·021	1·737	0·000	2·589	–2·391	1·104
120	1·643	2·507	–2·820	1·586	1·637	2·483	–2·301	1·067
150	2·625	2·318	–2·831	1·729	2·667	2·276	–2·349	1·213
180	1·064	2·214	–2·859	1·846	1·041	2·173	–2·386	1·309
210	2·566	2·319	–2·828	1·725	2·678	2·277	–2·347	1·210

†degrees; *kcal/mol; **eV

calculated using *ab initio* methods. The topology of the electron density is important in the theory of atoms-in-molecules (AIM),[47] that has been developed over the last three decades. In this study, variation in the electron density (r) at the bond critical points of the active group/site of the selected systems for various rotational angles has been probed. The electron density (r) distribution for the selected active groups in 3,3′-dichlorobenzidine, is shown in figures 3a–e. The variation in electron density (r) for C2–Cl13 group in one ring shows a maximum value at f = –30°, 30° and 60° and as per our observation Cl13 site is described as the most probable site for nucleophilic attack whereas for C9–Cl18 in the other ring it shows a maximum at f = 30° and 60° conformations. The variation in electron density (r) for $-CNH_2$ group in one ring shows a maximum value at f = 0° whereas for $-CNH_2$ in the second ring it shows a maximum at f = 180° conformations. The C4–C7 bond connecting the two rings also shows maximum variations in the electron density distributions at f = –30°, 30°, 150° and 180° conformations.

In the case of 3,3′-dimethoxybenzidine, the electron density (r) distribution for the selected groups is shown in figures S2 (a–f). The $-CNH_2$ group in one ring shows a maximum value in electron density (r) at f = 0°, whereas in the second ring it shows a maximum at f = 90° conformations. Similarly, the $-COCH_3$ group in one ring shows a maximum value in electron density (r) at around f = 90° and f = 210°,

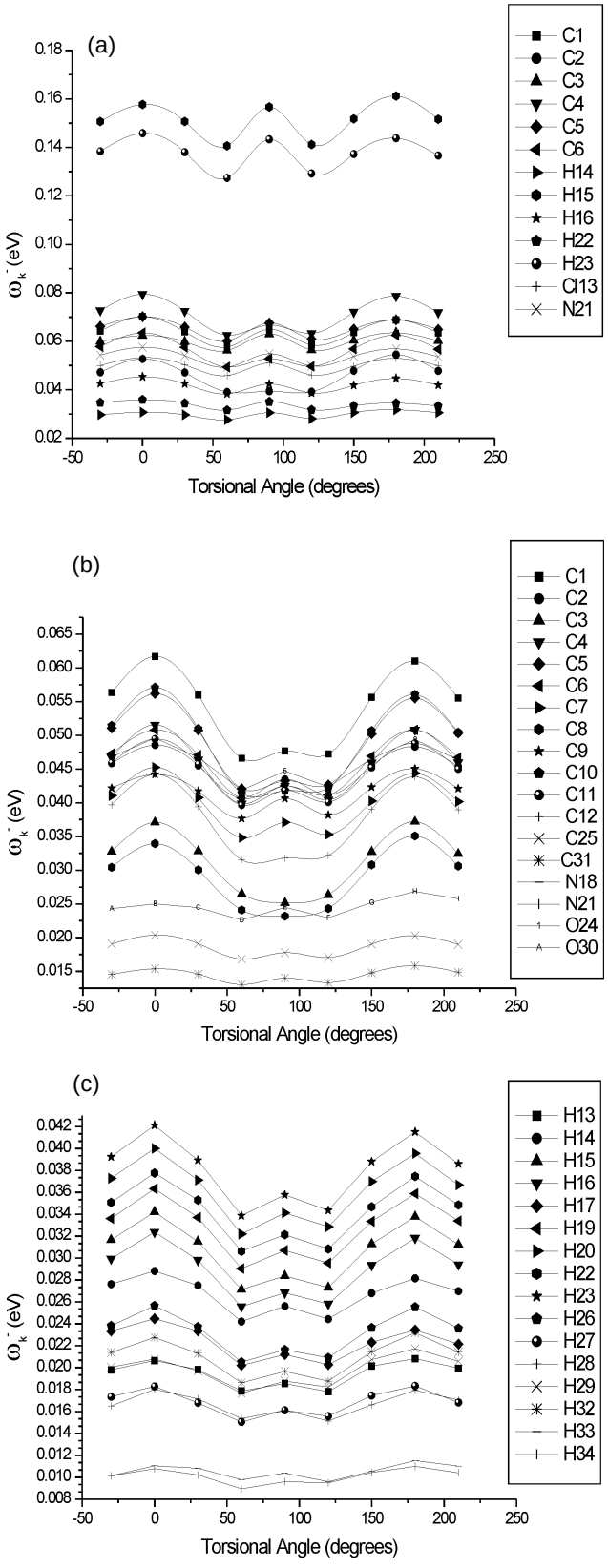

Figure 2. Variation of w_k^- (eV) with torsional angles (degrees) for 3,3′-dichlorobenzidine (**a**), carbon, nitrogen and oxygen atoms of 3,3′-dimethoxybenzidine (**b**) and hydrogen atoms of 3,3′-dimethoxybenzidine (**c**).

whereas in the second ring it shows a maximum peak only at $f = 90°$ conformation. As per our philicity analysis, C11 site is the most probable site for nucleophilic attack. So we have also done the electron density (r) distribution analysis at C11–H site and found that $f = 0°$ and $f = 180°$ conformations show maximum values for electron density (r). Also a close look at C4–C7 bond connecting the two rings shows maximum variations in the electron density distributions at $f = –30°$, 30°, 150° and 180° conformations. Above analysis gives us a clear picture of how various active sites in the selected systems exhibit their reactive nature while the molecule undergoes conformational changes.

5. Interaction studies based on charge transfer

The amounts of charge transfer between 3,3′-dichlorobenzidine (and also 3,3′-dimethoxybenzidine) and the nucleic acid bases/base pairs, viz. adenine, guanine, cytosine, thymine, uracil, ATH, GCWC have been computed using Parr's formula, because the toxicity of a compound is mainly understood through its interaction with a biological system via the possible charge transfer between them augmented by suitable *p*-stacking. The calculated values are presented in tables 2 and 3. It can be seen that the electron transfer for all the bases/base pairs, viz. adenine, guanine, cytosine, thymine, uracil, ATH, GCWC is minimum for the $f = 90°$ conformation for both the selected systems. Among the bases, uracil and guanine have the maximum and minimum values for ΔN respectively, whereas for the selected base pairs ATH has the maximum value for ΔN for all conformations for both the selected derivatives of benzidine. ΔN calculation reveals the electron-donating nature of the selected derivatives of benzidine with the exception of the 3,3′-dichlorobenzidine-guanine interaction, where 3,3′-dichlorobenzidine acts as an electron-accepting agent. Comparison of the charge transfer results of polychlorinated biphenyls (PCB), dioxins (DXN), and benzidines (BZN), including some of their derivatives indicate that in general PCB and DXN and their derivatives are electron acceptors whereas BZN and its derivatives are electron donors.

5.1 *QSAR analysis of polyaromatic hydrocarbons (PAHs) in gas phase*

The structural template of PCDFs, PHDDs and PCBs with required atom numbering is presented in figure

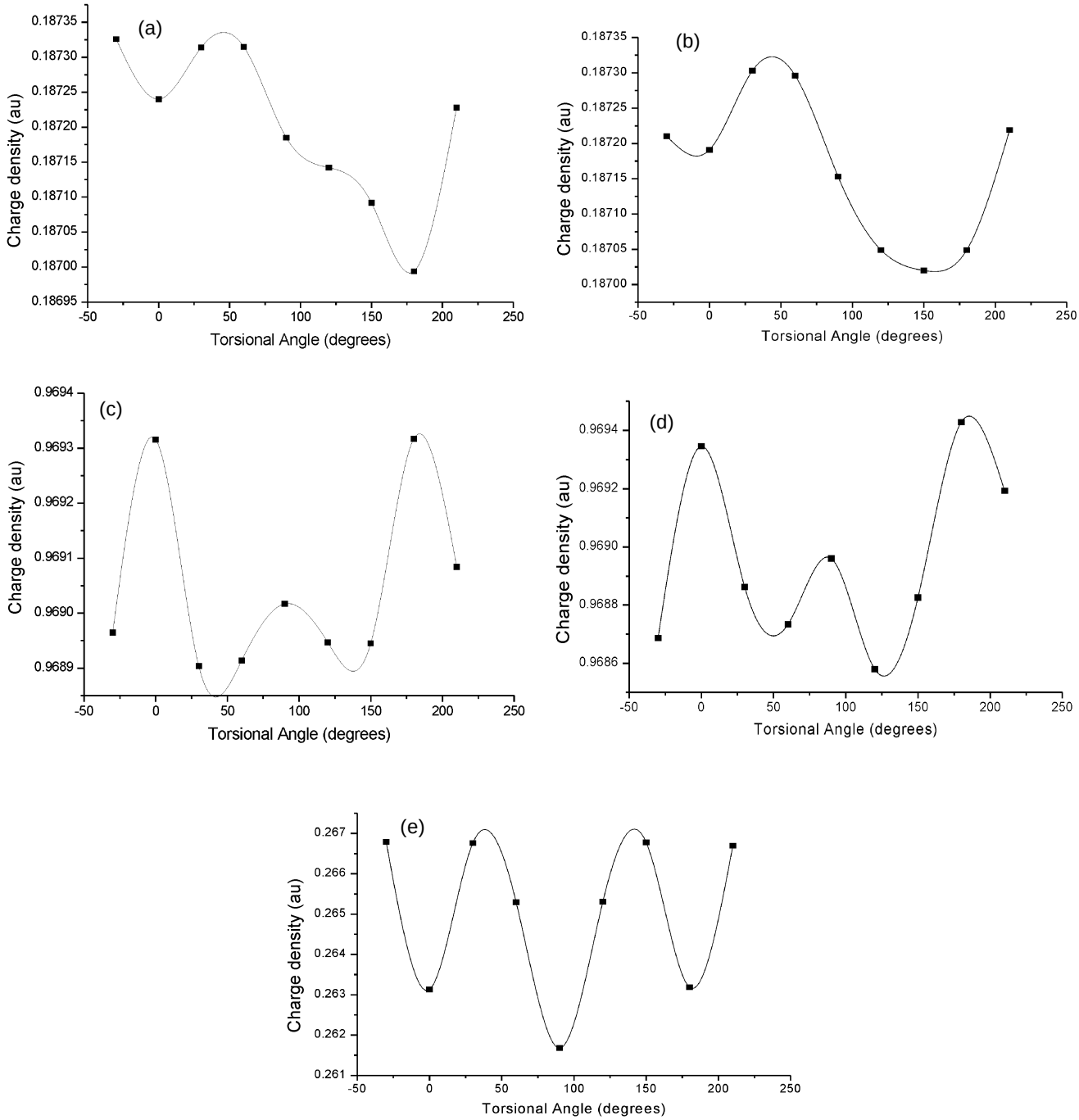

Figure 3. The charge density (r) distribution at the bond critical point of the selected group for 3,3′-dichlorobenzidine. (**a**) C_2Cl_{13}, (**b**) C_9Cl_{18}, (**c**) $C_1N_{24}H_{25}H_{26}$, (**d**) $C_{10}N_{21}H_{22}H_{23}$ and (**e**) C_4C_7.

S3 (a–c)$^+$. Tables S1–S3$^+$ provide the identity (ID) of the molecule with its substitution pattern. Previous studies[11–13] have revealed the fact that PCDFs, PHDDs and PCBs are electron acceptors in their interaction with biomolecules. Hence, for regression analysis, atoms with the maximum values of the local electrophilic power (w^+_{max}) in a molecule along with HF energy (E) and electrophilicity index (w) have been considered as independent variables.

The regression equations obtained by using parameters optimized in the gas phase have been presented in table 4.

$^+$See the end of this paper on the journal website: www.ias.ac.in/chemsci

Table 2. Calculated charge transfer (a.u.) between 3,3′-dichlorobenzidine and bases/base pairs.

Torsional angle (°)	Adenine	Thymine	Guanine	Cytosine	Uracil	GCWC	ATH
–30	0·026	0·082	–0·018	0·053	0·103	0·023	0·044
0	0·024	0·081	–0·021	0·051	0·102	0·020	0·042
30	0·026	0·082	–0·017	0·053	0·103	0·023	0·044
60	0·027	0·081	–0·015	0·053	0·101	0·024	0·044
90	0·007	0·061	–0·033	0·032	0·080	0·001	0·023
120	0·026	0·081	–0·016	0·052	0·101	0·023	0·044
150	0·026	0·082	–0·017	0·053	0·103	0·023	0·044
180	0·024	0·081	–0·020	0·051	0·103	0·020	0·042
210	0·026	0·083	–0·017	0·053	0·103	0·023	0·045

Table 3. Calculated charge transfer (a.u.) between 3,3′-dimethoxybenzidine and bases/base pairs.

Torsional angle (°)	Adenine	Thymine	Guanine	Cytosine	Uracil	GCWC	ATH
–30	0·073	0·129	0·028	0·100	0·149	0·078	0·094
0	0·071	0·129	0·026	0·099	0·149	0·076	0·093
30	0·073	0·130	0·029	0·101	0·150	0·079	0·095
60	0·076	0·130	0·033	0·102	0·149	0·082	0·096
90	0·065	0·119	0·024	0·091	0·138	0·069	0·085
120	0·075	0·129	0·032	0·101	0·149	0·081	0·096
150	0·073	0·130	0·029	0·101	0·150	0·079	0·095
180	0·071	0·129	0·026	0·099	0·149	0·077	0·093
210	0·073	0·130	0·029	0·101	0·150	0·079	0·095

Table 4. Regression models, coefficient of determinations and standard deviations for different groups of polyaromatic hydrocarbons (PAHs) in the gas phase.

System	Method	Regression equation	N	r^2	SD
PCDFs	MPA	$pIC_{50} = -13{\cdot}4794 + 0{\cdot}0020 \times \mathrm{E} + 3{\cdot}91050 \times w + 19{\cdot}2502 \times w^{+}_{max}$	27	0·821	0·701
	HPA	$pIC_{50} = -7{\cdot}1511 + 0{\cdot}0007 \times \mathrm{E} + 2{\cdot}7671 \times w + 15{\cdot}6018 \times w^{+}_{max}$	27	0·822	0·700
PHDDs	MPA	$pIC_{50} = 2{\cdot}5683 - 0{\cdot}0001 \times E - 5{\cdot}1179 \times w + 58{\cdot}6356 \times w^{+}_{max}$	19	0·835	0·753
	HPA	$pIC_{50} = 3{\cdot}2716 - 0{\cdot}0001 \times E - 5{\cdot}3056 \times w + 75{\cdot}7053 \times w^{+}_{max}$	19	0·874	0·660
PCBs	MPA	$pIC_{50} = -2{\cdot}3037 + 0{\cdot}0025 \times E + 8{\cdot}006 \times w - 28{\cdot}8824 \times w^{+}_{max}$	11	0·884	0·454
	HPA	$pIC_{50} = -3{\cdot}3700 + 0{\cdot}0025 \times E + 7{\cdot}6666 \times w - 30{\cdot}3092 \times w^{+}_{max}$	11	0·884	0·451

5.2 *Polychlorinated dibenzofurans (PCDFs)*

The gas phase data of the selected set of 27 PCDFs are given in table 5 including the observed and calculated values of pIC_{50} for both MPA and HPA methods. The HF energy, w and MPA (HPA) derived w^{+}_{max} are capable of providing 82·1% (82·2%) variation in data with standard deviation, SD of 0·701 (0·700) during a multiple regression with pIC_{50} as the dependent variable and HF energy, w and w^{+}_{max} as independent variables. A plot between observed and calculated pIC_{50} for PCDFs (figure 4a and b) shows a correlation of 0·906 (0·907) for MPA (HPA).

5.3 *Polyhalogenated dibenzo-p-dioxins (PHDDs)*

Table 6 presents the observed and calculated values of pIC_{50} for selected set of 19 PHDDs for MPA and HPA methods. HF energy, w and MPA (HPA) derived w^{+}_{max} provide the r^2 value of 0·835 (0·874) with standard deviation, SD of 0·753 (0·660). Figures 4c and d show plots between observed and calculated

Table 5. Energy, hardness, chemical potential, electrophilicity and local electrophilicities (MPA and HPA) for different polychlorinated dibenzofurans (gas phase).

Molecule	Energy (Hartree)	h (eV)	m (eV)	w (eV)	w^+_{max} (eV) MPA	w^+_{max} (eV) HPA	Observed pIC_{50}[a]	Calculated pIC_{50} MPA	Calculated pIC_{50} HPA
1	–996·9240	2·493	–3·712	2·763	0·429	0·224	4·061	3·612	3·337
2	–996·9220	2·528	–3·694	2·699	0·43	0·251	3·429	3·381	3·581
3	–1456·5160	2·452	–3·999	3·101	0·466	0·273	4·125	4·737	4·736
4	–1456·5180	2·453	–3·950	3·180	0·466	0·248	4·103	5·046	4·564
5	–1916·1100	2·451	–4·062	3·366	0·511	0·343	6·123	5·730	6·260
6	–1916·1120	2·399	–4·054	3·425	0·512	0·346	4·653	5·980	6·470
7	–1916·1000	2·439	–4.031	3·331	0·530	0·326	5·396	5·959	5·898
8	–1916·1080	2·414	–4·103	3·486	0·538	0·345	6·858	6·719	6·623
9	–2375·6910	2·412	–4·201	3·659	0·565	0·351	7·255	7·006	6·895
10	–2375·6940	2·392	–4·234	3·748	0·570	0·360	7·379	7·451	7·282
11	–2375·7020	2·400	–4·253	3·769	0·569	0·373	7·657	7·513	7·543
12	–2375·6970	2·347	–4·215	3·785	0·548	0·360	8·444	7·172	7·384
13	–2835·2850	2·403	–4·358	3·952	0·611	0·375	8·194	8·129	7·779
14	–2835·2820	2·34	–4·309	3·967	0·617	0·377	7·911	8·303	7·852
15	–2835·2850	2·329	–4·319	4·005	0·579	0·376	8·147	7·720	7·942
16	–2835·2830	2·343	–4·354	4·046	0·586	0·376	8·943	8·015	8·055
17	–3294·8710	2·309	–4·437	4·263	0·638	0·388	7·587	8·955	8·542
18	–3294·8680	2·342	–4·485	4·293	0·603	0·386	8·376	8·399	8·594
19	–2375·6980	2·402	–4·288	3·828	0·566	0·367	7·610	7·686	7·612
20	–2375·6940	2·414	–4·193	3·642	0·571	0·357	7·379	7·055	6·942
21	–2375·6970	2·356	–4·151	3·657	0·559	0·351	7·954	6·883	6·889
22	–2835·2880	2·325	–4·307	3·989	0·586	0·379	7·657	7·792	7·944
23	–2835·2870	2·349	–4·328	3·988	0·591	0·379	7·657	7·884	7·941
24	–2835·2780	2·330	–4·323	4·010	0·579	0·377	7·313	7·739	7·971
25	–2375·6970	2·356	–4·151	3·657	0·558	0·351	7·954	6·864	6·889
26	–2835·2880	2·325	–4·307	3·989	0·586	0·379	7·623	7·792	7·944
27	–2835·2870	2·349	–4·328	3·988	0·591	0·379	7·623	7·884	7·941

[a]Experimental data as given in ref. [31]

Table 6. Energy, hardness , chemical potential, electrophilicity and local electrophilicities (MPA and HPA) for different dibenzo-*p*-dioxins (PHDDs).

Molecule	Energy (Hartree)	h (eV)	m (eV)	w (eV)	w^+_{max} (eV) MPA	w^+_{max} (eV) HPA	Observed pIC_{50}[a]	Calculated pIC_{50} MPA	Calculated pIC_{50} HPA
28	–2910·4795	2.360	–3·796	3·053	0·348	0·269	8·118	7·513	7·601
29	–2450·8917	2.383	–3·647	2·79	0·315	0·243	7·768	6·914	7·003
30	–1991·3025	2·417	–3·496	2·529	0·298	0·230	7·610	7·234	7·405
31	–3370·0640	2·341	–3·931	3·300	0·363	0·281	7·490	7·153	7·217
32	–2450·8951	2·388	–3·689	2·849	0·331	0·251	6·975	7·503	7·294
33	–2910·4808	2·371	–3·831	3·095	0·350	0·263	6·811	7·398	6·954
34	–2450·8820	2·376	–3·606	2·737	0·326	0·252	6·728	7·796	7·972
35	–1991·3053	2·395	–3·499	2·555	0·302	0·233	8·171	7·281	7·447
36	–1531·7161	2·417	–3·324	2·286	0·272	0·203	6·281	6·905	6·644
37	–4289·2323	2·326	–4·170	3·737	0·381	0·295	5·715	6·033	6·072
38	–1072·1192	2·469	–3·099	1·944	0·206	0·154	4·572	4·762	4·654
39	–10896·934	2·347	–3·621	2·792	0·349	0·268	10·086	9·352	9·452
40	–6673·9141	2·361	–3·638	2·804	0·353	0·272	10·093	9·307	9·415
41	–6673·9136	2·361	–3·638	2·803	0·364	0·280	10·687	9·961	10·050
42	–4562·4040	2·367	–3·647	2·810	0·365	0·281	9·074	9·861	9·931
43	–10896·9340	2·365	–3·647	2·812	0·366	0·278	9·943	10·22	10·130
44	–13468·0310	2·348	–3·762	3·014	0·374	0·283	8·881	9·809	9·597
45	–8325·8338	2·374	–3·477	2·547	0·334	0·257	10·209	9·562	9·770
46	–5754·7340	2·406	–3·322	2·293	0·314	0·238	8·927	9·573	9·531

[a]Experimental data as given in ref. [31]

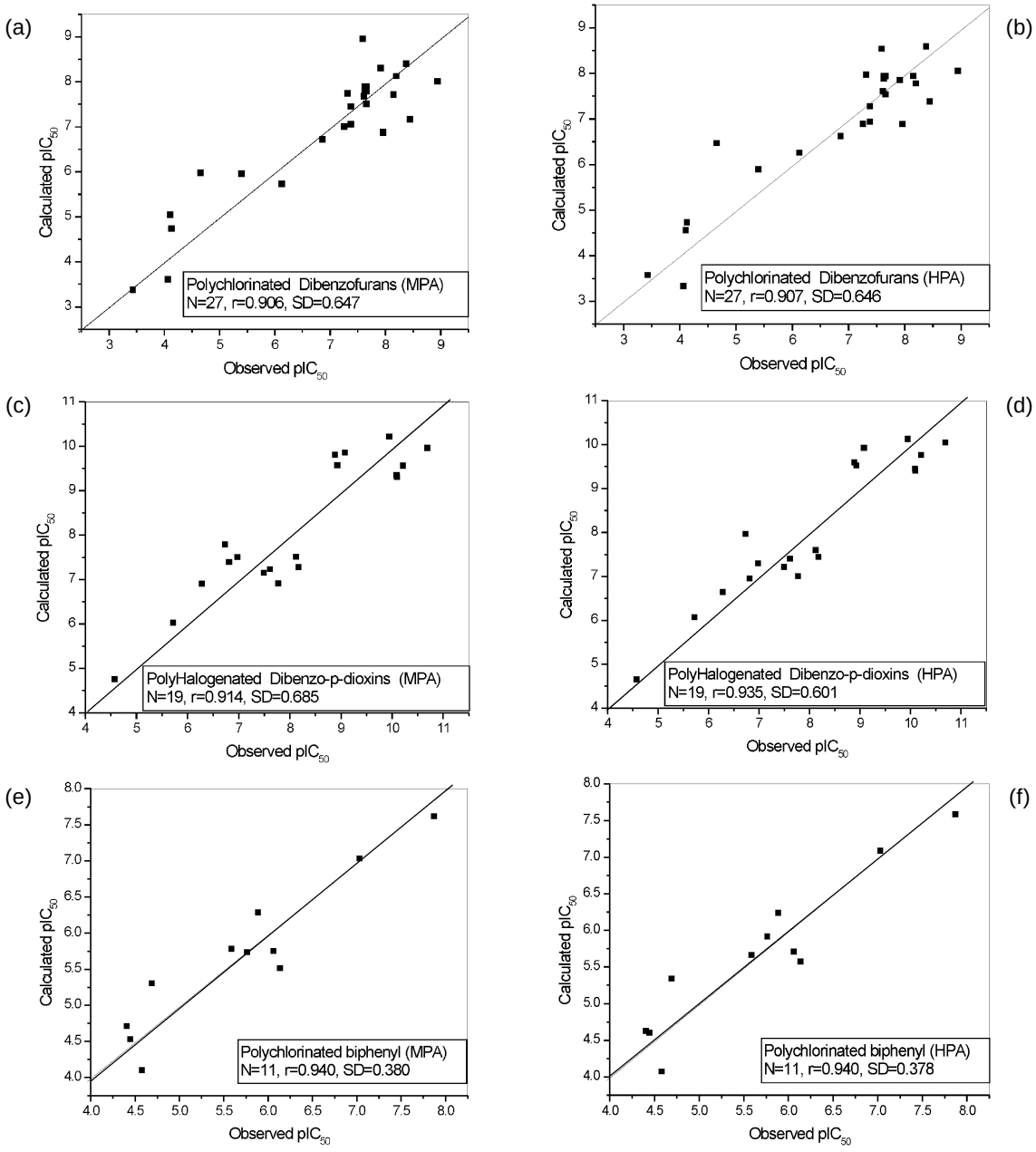

Figure 4. Observed versus calculated values of pIC_{50} using MPA and HPA methods for (**a**, **b**) PCDFs, (**c**, **d**) PHDDs and (**e**, **f**) PCBs.

pIC_{50} for PHDDs, giving a correlation of 0·914 (0·935) for MPA (HPA).

5.4 *Polychlorinated biphenyls (PCB)*

The selected set of 11 PCBs along with observed and calculated values of pIC_{50} for MPA and HPA methods are given in table 7. HF energy, w and MPA (HPA) derived w^{+}_{max} are capable of explaining 88·4% (88·4%) variation in data, with standard deviation, SD of 0·454 (0·451). Plots between observed and calculated pIC_{50} for PCDFs (figures 4e and f) give a correlation of 0·940 (0·940) for MPA (HPA).

This shows that HF energy, along with w and w^{+}_{max} can be effectively used in explaining the toxicity of the selected systems and two different population analysis schemes (MPA and HPA) provide identical trends.

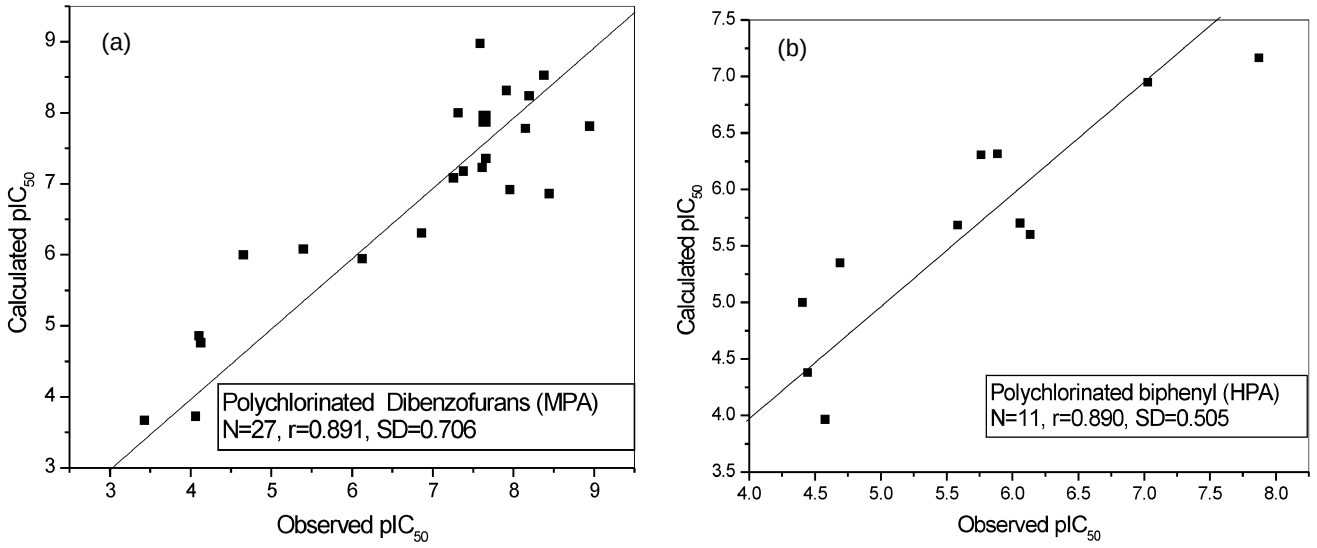

Figure 5. Observed versus calculated values of pIC_{50} in the solvent phase for (**a**) PCDFs using MPA, (**b**) PCBs using HPA method.

Table 7. Energy, hardness, chemical potential, electrophilicity and local electrophilicity (MPA and HPA) for different PCBs

Molecule	Energy (Hartree)	h (eV)	m (eV)	w (eV)	w^+_{max} (eV)		Observed pIC_{50}[a]	Calculated pIC_{50}	
					MPA	HPA		MPA	HPA
47	–2301·6789	2·495	–4·099	3·367	0·407	0·320	7·028	7·047	7·085
48	–2761·2677	2·483	–4·264	3·661	0·428	0·340	7·871	7·631	7·584
49	–2301·6730	2·667	–4·073	3·109	0·379	0·302	5·584	5·797	5·665
50	–2761·2625	2·607	–4·167	3·329	0·409	0·323	6·134	5·519	5·573
51	–2761·2675	2·578	–4·202	3·424	0·428	0·336	5·762	5·743	5·916
52	–3220·8507	2·582	–4·310	3·597	0·435	0·349	6·057	5·755	5·709
53	–3680·4392	2·587	–4·463	3·850	0·447	0·358	5·885	6·287	6·241
54	–2301·6787	2·802	–4·008	2·866	0·355	0·275	4·442	4·541	4·603
55	–3220·8572	2·715	–4·277	3·368	0·387	0·303	4·689	5·307	5·343
56	–2301·6655	2·664	–4·042	3·067	0·405	0·325	4·405	4·721	4·628
57	–3220·8559	2·915	–4·259	3·112	0·358	0·280	4·577	4·104	4·075

[a]Experimental data as given in ref. [31]

5.5 *QSAR analysis of polyaromatic hydrocarbons (PAHs) in solvent phase*

QSAR analysis in solvent phase has been carried out for PCDFs and PCBs as test cases to study the behaviour of the selected descriptors in solvent environment.

5.6 *Polychlorinated dibenzofurans (PCDFs)*

The regression equation for the selected set of 27 PCDFs using MPA method is given as,

$$pIC_{50} = -8{\cdot}2078 + 2{\cdot}4117w + 12{\cdot}0391w^+_{max}, \quad (10)$$
$$N = 27,\ r^2 = 0{\cdot}794,\ \mathrm{SD} = 0{\cdot}739.$$

The solvent phase output of the selected set of 27 PCDFs is given in table S4 along with observed and calculated values of pIC_{50} for MPA derived charges. The w and MPA derived w^+_{max} are capable of providing 79·4% variation in data with standard deviation, SD of 0·739. A plot between observed and calculated pIC_{50} for PCDFs (figure 5a) gives a correlation of 0·891 for MPA. Comparing with gas phase parameters, it is seen that there has been a de-

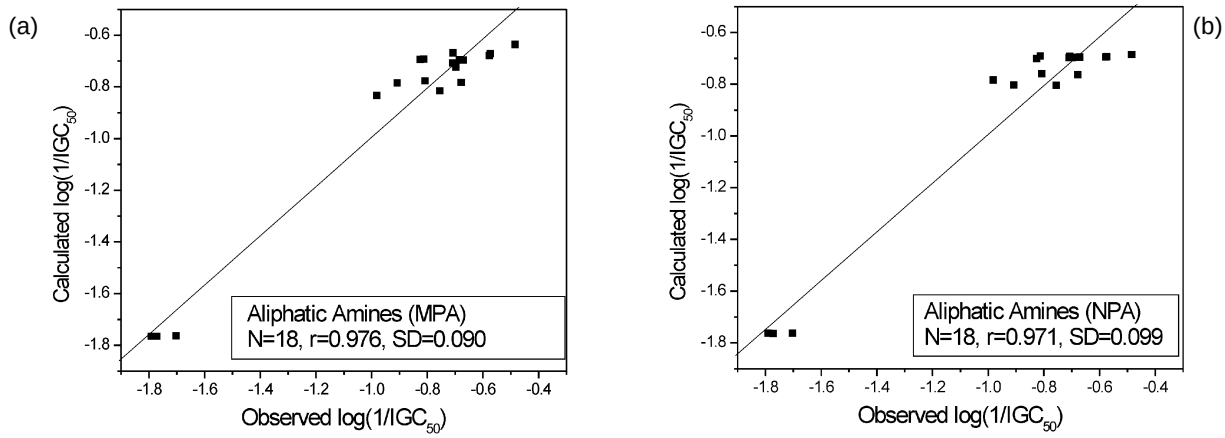

Figure 6. Observed versus calculated log (IGC_{50}^{-1}) values of aliphatic amines from (**a**) MPA and (**b**) NPA study.

Table 8. Experimental and calculated values of log (IGC_{50}^{-1}) for aliphatic amines with tetrahymena pyriformis in gas phase.

			log(IGC_{50}^{-1})				
			w_k^- (max)			Calculated	
Molecules	Energy (Hartree)	w (eV)	MPA (eV)	NPA (eV)	Observed[a]	MPA	NPA
Propylamine	–173·3338	0·6355	0·2981	0·4587	–0·7075	–0·6682	–0·6924
Butylamine	–212·3777	0·6337	0·2963	0·4570	–0·5735	–0·6707	–0·6933
N-Methylpropylamine	–212·3674	0·5458	0·2146	0·3634	–0·8087	–0·7762	–0·7601
Amylamine	–251·4186	0·6220	0·2993	0·4519	–0·4848	–0·6356	–0·6856
N-Methylbutylamine	–251·4131	0·5419	0·2104	0·3593	–0·6784	–0·783	–0·7629
N,N-Dimethylethylamine	–212·3565	0·4766	0·1714	0·2940	–0·9083	–0·7842	–0·8032
(±)-*s*-Butylamine	–212·3802	0·6629	0·3065	0·4775	–0·6708	–0·6958	–0·6948
Isoamylamine	–251·4203	0·6508	0·3042	0·4690	–0·5774	–0·6786	–0·6943
1-Methylbutylamine	–251·4241	0·6546	0·3019	0·4710	–0·6846	–0·6946	–0·6959
1-Ethylpropylamine	–251·4215	0·6305	0·2882	0·4558	–0·8129	–0·6928	–0·6907
N,N-Diethylmethylamine	–251·4034	0·4890	0·1701	0·3025	–0·7559	–0·8149	–0·8044
t-Amylamine	–251·4241	0·6998	0·3205	0·5033	–0·6978	–0·7235	–0·6973
(+/–)-1,2-Dimethylpropylamine	–251·4208	0·6369	0·2879	0·4577	–0·7095	–0·7071	–0·697
Propargylamine	–170·9325	0·6901	0·3234	0·4947	–0·8260	–0·6929	–0·7009
N-Methylpropargylamine	–209·9652	0·6358	0·2517	0·4187	–0·9818	–0·8329	–0·7833
2-Methoxyethylamine	–248·1944	0·6588	0·0020	0·0020	–1·7903	–1·7649	–1·762
3-Methoxypropylamine	–287·2359	0·6610	0·0033	0·0028	–1·7725	–1·765	–1·7638
3-Ethoxypropylamine	–326·2854	0·6595	0·0031	0·0027	–1·7027	–1·7626	–1·7616

[a]Experimental data as given in ref. [32]

crease in the value of all the selected descriptors in the solvent medium and also a decrease in correlation between the experimental and calculated pIC_{50} values.

5.7 *Polychlorinated biphenyls (PCB)*

The regression equation for the selected set of 11 PCBs using HPA method is given as,

$$pIC_{50} = -5{\cdot}7632 + 0{\cdot}0015 \times E + 5{\cdot}4326w - 7{\cdot}7886w^{+}_{\max}, \quad (11)$$
$$N = 11,\ r^2 = 0{\cdot}793,\ \mathrm{SD} = 0{\cdot}605.$$

The selected set of 11 PCBs in solvent phase has been given in table S5 along with observed and calculated values of pIC_{50} for HPA derived charges. HF energy, w and HPA derived $w^{+}_{\max}$ give a coefficient of determination of 0·793 with standard deviation,

SD of 0·605. A plot between observed and calculated pIC_{50} for PCBs (figure 5b) gives a correlation of 0·890 for HPA. This shows that HF energy along with electrophilicity index and local electrophilic power can be used as successful descriptors in the prediction of biological activity of molecules. It is important to note that MPA and HPA provide similar trends.

5.8 *QSAR analysis on aliphatic amines in gas phase*

Aliphatic amines are known[54] to be electron donors in their interaction with biomolecules. Hence for the regression analysis, the local philicity ($w_{\max}^{-}$) with the f^{-} value considered on the nitrogen (N) site along with electrophilicity index (w) have been considered as independent variables. In reference 54 respective charges were used. Corresponding population values change the most reactive (electrophilic/nucleophilic) centres in toxins with multiple reactive sites albeit with similar quality correlations between the toxicity and global and local electrophilicities.

The regression equations for MPA and NPA derived charges using gas phase optimized parameters are given as,

for MPA,

$$\log(IGC_{50}^{-1}) = -2{\cdot}0908w + 3{\cdot}5398w_{\max}^{-} - 0{\cdot}3947, \quad (12)$$

$$N = 18,\ r^2 = 0{\cdot}953,\ \mathrm{SD} = 0{\cdot}090,$$

for NPA,

$$\log(IGC_{50}^{-1}) = -1{\cdot}6429w + 2{\cdot}2581w_{\max}^{-} - 0{\cdot}6842, \quad (13)$$

$$N = 18,\ r^2 = 0{\cdot}943,\ \mathrm{SD} = 0{\cdot}099.$$

The gas phase outputs for the selected set of 18 aliphatic amines are given in table 8 along with observed and calculated values of $\log(IGC_{50}^{-1})$ for MPA and NPA methods. The w and MPA (NPA) derived $w_{\max}^{-}$ are capable of providing 95·3% (94·3%) variation in data with the standard deviation, SD of 0·090 (0·099). A plot between observed and calculated $\log(IGC_{50}^{-1})$ for aliphatic amines (figure 6a and b) gives a correlation of 0·976 (0·971) for MPA (NPA) which reveals the importance of the selected descriptors in structure–activity studies. Figures S1 to S3 and tables S1 to S5 are presented as the Supporting Information[a].

[a]See end of this paper on journal website: www.ias.ac.in/chemsci

Conclusions

Valuable information about the reactive sites for possible electrophilic attack on 3,3′-dichlorobenzidine and 3,3′-dimethoxybenzidine in different orientations is obtained in terms of various DFT-based chemical reactivity descriptors. It is possible to understand that relatively low energy barrier has provided greater flexibility to these selected systems, thereby allowing them to orient themselves in any desired conformation in the biological system leading to their toxic characteristics. Analysis of electron density values at the BCP for the selected active group/site gives an important clue regarding the behaviour of the selected systems while undergoing conformational changes. Further, the charge transfer between the selected derivatives of benzidine and nucleic acid bases/DNA base pairs has revealed the electron-donating nature of the 3,3′-dichlorobenzidine and 3,3′-dimethoxybenzidine.

Experimental biological activity (pIC_{50}) of different polyaromatic hydrocarbons (PAH) namely polychlorinated dibenzofurans (PCDF), polyhalogenated dibenzo-*p*-dioxins (PHDD) and polychlorinated biphenyls (PCB) has been shown to correlate well with their corresponding activity (pIC_{50}) calculated using the HF energy, the electrophilicity index and the local electrophilic power through regression analyses in gas and solvent phases. Also, in the case of aliphatic amines, local philicity ($w_{\max}^{-}$) and electrophilicity index (w) are shown to be capable of explaining the activity ($\log(IGC_{50}^{-1})$) in an elegant manner. While the PAHs behave as electron acceptors when they exhibit their toxic behaviour during their interactions with biosystems, the aliphatic amines act as electron donors. A reasonably good correlation has been obtained for all the systems showing the significance of the conceptual DFT-based selected descriptors in the prediction of toxicity in gas and solution phases with similar trends originating from different population analysis schemes. The strength of this approach lies in the fact that the other approaches with more descriptors, often disjoint, and with no apparent connection with toxicity, provide comparable or poorer correlations.

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