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Theoretical Study Related to the Carcinogenic Activity of Polycyclic Aromatic Hydrocarbon Derivatives

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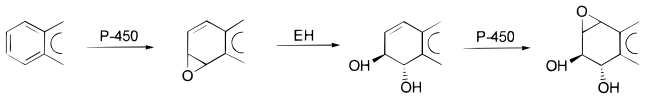
Ab initio, density functional, and semiempirical calculations concerning the reactivity of polycyclic aromatic hydrocarbons (PAHs) were performed. The reactions considered were those related to the carcinogenic activity of PAHs. The increased reactivity of the diol epoxide derivatives is explained by the greater propensity of benzene diol epoxide to undergo acid-catalyzed opening of the epoxide ring, as well as its thermodynamic tendency to react with nucleophiles in substitution reactions. However, rearrangement to phenol was the favored path for benzene oxide. Bay-region compounds were found to open spontaneously upon protonation, thus revealing a greater reactivity in the rate-determining step of the mechanism of carcinogenesis. A correlation was observed between the exothermicity of the process and the charge delocalization in the resulting carbocation. The enhanced activity of bay-region methyl-substituted compounds was attributed to the instability of the closed structures due to steric interactions rather than stabilization of the carbocations by hyperconjugation.

Introduction

Chemical carcinogens exert their activity through reaction with cellular macromolecules, generally DNA. Formation of carcinogen−DNA adducts can result in mutations that lead to the initiation of tumorigenesis.[1] Among these compounds, polycyclic aromatic hydrocarbons (PAHs) constitute a relevant group because of their widespread environmental prevalence[2] and their relatively high tumorigenic potency.[3]

As electrophilicity is required for binding to the amino active sites of DNA, the ultimate carcinogenic forms of most chemical carcinogens are electron-deficient reactants. Activation of PAHs to ultimate carcinogens requires three steps, as illustrated below: initial epoxidation by the cytochrome P-450 monooxygenases,[4] followed by epoxide hydrase enzyme-mediated hydrolysis to the trans diol, and a second epoxidation at the adjacent double bond. This metabolic process yields a diol epoxide



that can interact with tissue nucleophiles, giving rise to the alkylation of DNA.[5,3e]

A critical step in the mechanism involved in carcinogenesis for this type of compounds is considered to be the epoxide ring opening to yield a carbocation at the benzylic position of the epoxide function.[6] It is likely that electrophilic attack of DNA by hydrocarbon epoxides is S_N1-like and proceeds through proton-stabilized transition states in which the hydrocarbon exhibits significant carbonium ion character.[7] Arene oxides are also involved in the formation of phenols, diols, ketones, glutathione conjugates, and other important metabolites of PAHs.[8]

PAHs that contain a bay-region angular benzo ring dihydrodiol epoxide with one carbon atom of the epoxide ring in the bay region present an increased mutagenic and carcinogenic activity,[9] and these observations have led to the development of the so-called "bay-region

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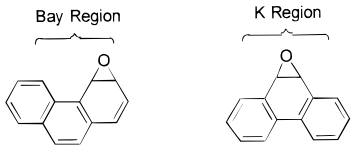
theory".[10] By this approach, both mutagenicity and



carcinogenicity of PAHs have been correlated with the ease of the epoxide ring opening and consequent carbonium ion formation from the diol epoxide metabolites, calculated by the PMO method.[11] Bay region diol epoxides were predicted to be more active than their isomers, in agreement with the experimental findings.[12]

Methyl substitution in appropriate molecular regions of polyarenes frequently results in substantial enhancement of carcinogenic activity. Thus, substitution by a methyl group in the non-benzo ring of a bay region tends to markedly increase the activity.[3e] For example, 7,12-dimethylbenzo[*a*]anthracene and 5-methylchrysene are among the most potent known carcinogens, whereas benzo[*a*]anthracene and chrysene exhibit only weak borderline activity.[13]

Many theoretical studies concerning PAHs have been carried out by several authors.[14] They include principally semiempirical calculations (Hückel,[15] INDO,[16] MINDO/3,[17] MNDO,[18] and AM1[19]), although ab initio (mostly with small basis sets) and some density functional theory[20] (DFT) calculations have also been performed. These studies are related to electronic and structural properties, substituent effects, and mechanistic features associated with the carcinogenic activity. The large number of them, in conjunction with the enormous amount of experimental work performed, accounts for the importance of this kind of compounds in cancer research.

The aim of this paper was to apply quantum mechanical methods to a comprehensive study of the main factors determining the reactivity of arene epoxides and diol epoxides. As previous publications generally focused on one or two specific aspects, this work intends to give a wider view about geometrical, electronic, and methyl-substitution effects on the mechanism of carcinogenesis and detoxification reactions, as well as the role of the solvent. Semiempirical calculations were compared with more rigorous ab initio and DFT methods, employing a good quality basis set.

Computational Methods

Geometries were fully optimized and stationary points were characterized as minima (no imaginary frequencies) or transition states (one imaginary frequency) by calculation of the harmonic vibrational frequencies. The semiempirical methods AM1[19] and PM3[21] were used as implemented in the AMPAC 5.0 package of programs.[22] Ab initio and DFT/B3LYP[23] calculations were carried out with the Gaussian 94[24] and GAMESS[25] programs, employing the 6-31G* split-valence shell basis set.[26] The ab initio geometries optimized at the Hartree−Fock (HF) level were used for single-point calculations at the third-order Møller−Plesset perturbation correction (MP3)[27] for treatment of electron correlation effects. Optimizations

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Table 1. Ab Initio and DFT Total Energies (hartree), Zero Point Energies (kcal/mol), and Semiempirical Heats of Formation (kcal/mol) for the Protonated Species

	benzene oxide			benzene diol epoxide		
method	closed minimum	transition state	open carbocation	closed minimum	transition state	open carbocation
RHF/6-31G*	−305.81637	−305.81468	−305.84756	−456.69300		−456.72191
MP2/6-31G*//6-31G*	−306.73989	−306.73696	−306.75629	−457.97834		−457.98882
MP3/6-31G*//6-31G*	−306.77417	−306.77051	−306.79296	−458.01783		−458.03173
ZPE (RHF/6-31G*)	77.07	78.41	77.49	100.92		99.98
MP2/6-31G*	−306.74563	−306.73944	−306.76095	−457.98455		−457.99435
B3LYP/6-31G*	−307.72450	−307.72402	−307.74814			−459.37527
ZPE (B3LYP/6-31G*)	73.13	72.85	72.14			92.57
PM3	213.69	213.69	182.98			89.02
AM1	205.16	205.27	173.78	94.03	94.03	70.75
AM1-SM2.1	151.94	152.86	121.35	34.23	34.86	12.13

Table 2. Calculations for the Epoxide Opening Reaction of Protonated Benzene Oxide and Diol Epoxide (kcal/mol)

	benzene oxide		benzene diol epoxide	
method	$\Delta H^{\ddagger}$	ΔH_r	$\Delta H^{\ddagger}$	ΔH_r
RHF/6-31G*	1.06	−19.57	<0.01	−18.14
MP2/6-31G*//6-31G*	1.84	−10.29		−6.58
MP3/6-31G*//6-31G*	2.29	−11.79		−8.72
RHF/6-31G* [a]	0.40	−21.15	<0.01	−19.08
MP2/6-31G*	3.88	−9.61		−6.15
B3LYP/6-31G*	0.30	−14.84		
B3LYP/6-31G*[a]	0.02	−15.83		
PM3	<0.001	−30.71		
AM1	0.11	−31.38	<0.01	−23.28
AM1-SM2.1	0.92	−30.59	0.63	−22.10

[a] Including zero-point vibrational contribution.

at the MP2[28] level were also performed. Natural bond orbital population analysis was evaluated by means of the NBO program.[29] The solvent effect was taken into account by means of aqueous-phase calculations with the AM1-SM2.1 model.[30]

Results and Discussion

As mentioned previously, the ultimate carcinogenic metabolites derived from PAHs are considered to be diol epoxides.[5,3e] The electrophilicity of both epoxides and diol epoxides is thought to arise from their propensity to undergo acid-catalyzed ring opening, forming carbocations in the rate-determining step.[6] Calculations of this opening step were performed for O-protonated 1β,2β-epoxydihydrobenzene (benzene oxide) and 1α,2β-dihydroxy-3β,4β-epoxytetrahydrobenzene (benzene diol epoxide) as model systems, for the sake of comparison of the behavior of epoxides and diol epoxides derived from PAHs. Results are presented in Tables 1−4. Structures are shown in Figures 1 and 2.

O-protonated benzene oxide presented C_S symmetry, the six-membered ring being almost planar. It should be noted that this structure is a minimum at ab initio, AM1, and B3LYP levels and not a transition state as it had been established by George et al.[31] However, it is a first-order saddle point according to the PM3 results. The structure with the O−H bond pointing toward the ring in a cis disposition was located by the present calculations, being lower in energy than the trans one reported by George et al. The corresponding species for benzene diol epoxide was a minimum by the HF, MP2, and AM1 calculations. A hydrogen-bond interaction was observed between the hydrogen atom attached to the epoxide and the oxygen of the closest hydroxy group. This fact determined the lack of symmetry of the protonated epoxide ring, whose C−O bond distances (ca. 1.5 and 1.6 Å) were not identical as in the nonprotonated species. However, this species could not be located with B3LYP and PM3, as these methods afforded the open carbocation.

Activation energies for the epoxide ring opening of both compounds were very small with all methods, being negligible with the semiempirical ones. Inclusion of the solvent raised the barriers, although the values remained very low. The transition state for protonated benzene oxide presented differences between their C−O bond distances of ca. 0.1−0.3 Å, according to the method. The transition state for diol epoxide was very similar in structure to the corresponding protonated minimum, as they were very close in energy. This feature prevented the characterization of the saddle point by the ab initio calculations. The activation energy for benzene diol epoxide was lower than for benzene oxide by AM1 and AM1-SM2.1. The fact that the O-protonated species of the diol epoxide could not be isolated with B3LYP and PM3, because these methods collapsed to the open carbocation, reinforces this result. These calculations reflect the experimental observations concerning the increased carcinogenic activity of the diol epoxides, even when the present calculated barriers are very low to account for the rate-determining nature of this step. The more facile opening of the epoxide ring in the diol epoxide can be ascribed to the hydrogen bonding between the hydrogen of the epoxide and its closest hydroxy group. This interaction induces the asymmetry of the epoxide ring, the C−O bond that will break becoming longer, and therefore, diminishing the barrier. This finding can account for the experimentally observed higher activity of the anti isomers (which present the benzylic hydroxyl group and the epoxide oxygen atom on opposite faces of the molecule) of trans diol epoxides,[5c] compared to the

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Table 3. Some Selected Geometrical Parameters Calculated for Structures in Figure 1[a]

structure	parameter	AM1	PM3	RHF/ 6-31G*	MP2/ 6-31G*	B3LYP/ 6-31G*
a	C1−C2	1.4958	1.4915	1.4572	1.4862	1.4853
	C1−C3, C2−C4	1.4582	1.4611	1.4724	1.4545	1.4603
	C3−C5, C4−C6	1.3525	1.3500	1.3309	1.3616	1.3555
	C5−C6	1.4518	1.4532	1.4720	1.4535	1.4592
	C1−O13, C2−O13	1.5584	1.5793	1.5366	1.5766	1.5900
	C1−H11	1.1135	1.1104	1.0716	1.0855	1.0845
	O13−H14	0.9864	0.9619	0.9606	0.9937	0.9847
	∠C1C2O13, ∠C2C1O13	61.3	61.9	58.6	61.9	62.2
	∠C3C1O13, ∠C4C2O13	114.4	114.4	117.0	115.5	115.9
b	C1−C2	1.4932	1.4901	1.4608	1.4769	1.4827
	C1−O13	1.5180	1.5779	1.4356	1.4978	1.4615
	C2−O13	1.6451	1.5876	1.4713	1.5732	1.5259
c	C1−C2	1.4850	1.4862	1.4882	1.4697	1.4820
	C3−C5	1.3704	1.3688	1.3526	1.3799	1.3738
	C5−C6	1.4113	1.4087	1.4115	1.4084	1.4126
	C1−O13	1.4068	1.4023	1.3779	1.3950	1.3926
	O13−H14	0.9693	0.9497	0.9502	0.9754	0.9720

[a] Bond lengths in angstroms, bond angles in degrees.

Table 4. Some Selected Geometrical Parameters Calculated for Structures in Figure 2[a]

structure	parameter	AM1	PM3	RHF/ 6-31G*	MP2/ 6-31G*	B3LYP/ 6-31G*
a	C1−C2	1.4828		1.4489	1.4635	
	C1−O15	1.6164		1.5801	1.6089	
	C2−O15	1.4967		1.4842	1.5257	
	O15−H18	0.9916		0.9593	0.9910	
	O7−H18	2.1116		2.9947	3.0479	
	∠C1C2O15	65.7		65.2	65.2	
	∠C3C1O15	115.4		116.0	116.0	
b	C1−C2	1.4820				
	C1−O15	1.6346				
	C2−O15	1.4926				
	O15−H18	0.9675				
	O7−H18	2.1077				
c	C1−C2	1.4870	1.4871	1.5038	1.4926	1.4967
	C2−O15	1.4054	1.3993	1.3784	1.4039	1.3986
	O15−H18	0.9715	0.9504	0.9516	0.9787	0.9758
	O7−H18	2.2788	2.4982	2.2633	2.1970	2.1791

[a] Bond lengths in angstroms, bond angles in degrees.

syn isomers (benzylic hydroxyl and epoxide oxygen on the same face), where this interaction cannot be achieved.

Both opening reactions were calculated as exothermic with all methods, the semiempirical ones yielding the most exothermic values. The process was more favored thermodynamically for the epoxide. The solvent did not produce a significant effect.

It should be noted that DFT, ab initio, and semiempirical methods afforded the same trend in the comparison of the reactivity of both systems. This can be considered as an evidence of the reliability of these semiempirical methods for the study of the behavior of this type of compounds.

The carbocations resulting from the epoxide ring opening can suffer nucleophilic attack to yield DNA adducts, or they can rearrange via hydrogen migration to form a ketone or an alcohol, the second being the thermodynamically preferred of the rearranged products. As another way to explain the higher carcinogenic activity of diol epoxides, the enthalpy change for the following reactions was calculated for benzene oxide (**1a**) and benzene-1,2-diol-3,4-epoxide (**1b**): alcohol formation by rearrangement of the carbocation, and nucleophilic substitutions by NH_3 (to model DNA adduct formation), H_2O (to consider solvent attack), and H_2S (to take into account detoxification reactions that yield glutathione

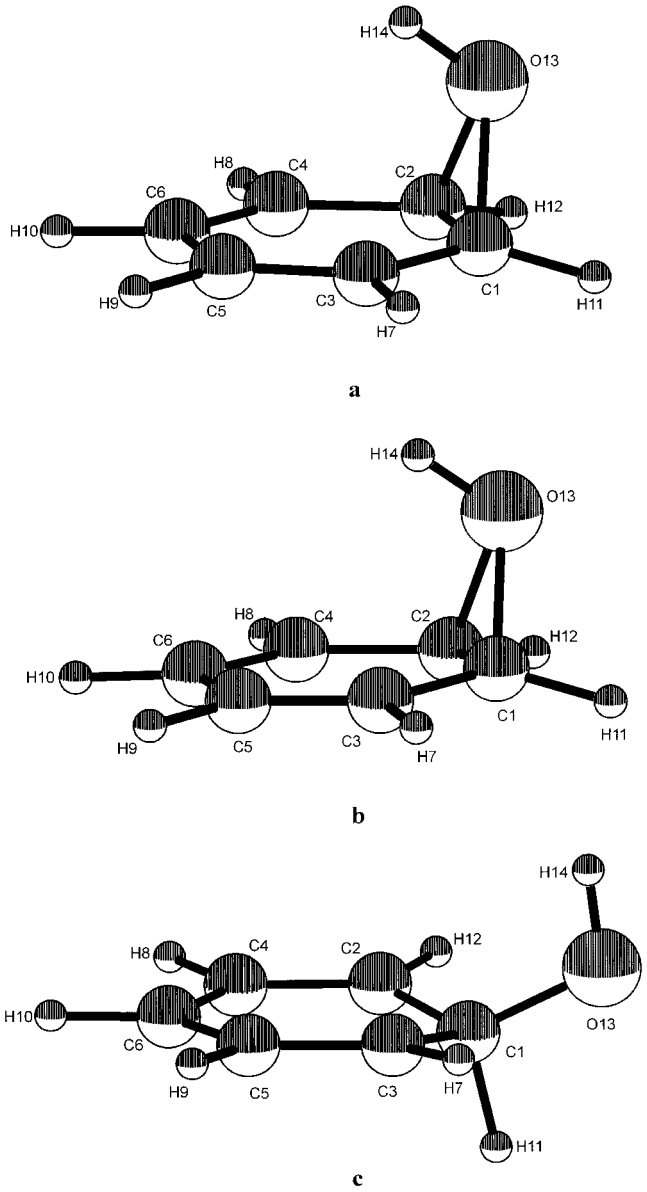


Figure 1. Calculated structures for O-protonated benzene oxide: (a) closed minimum, (b) transition state for the epoxide opening, and (c) open carbocation.

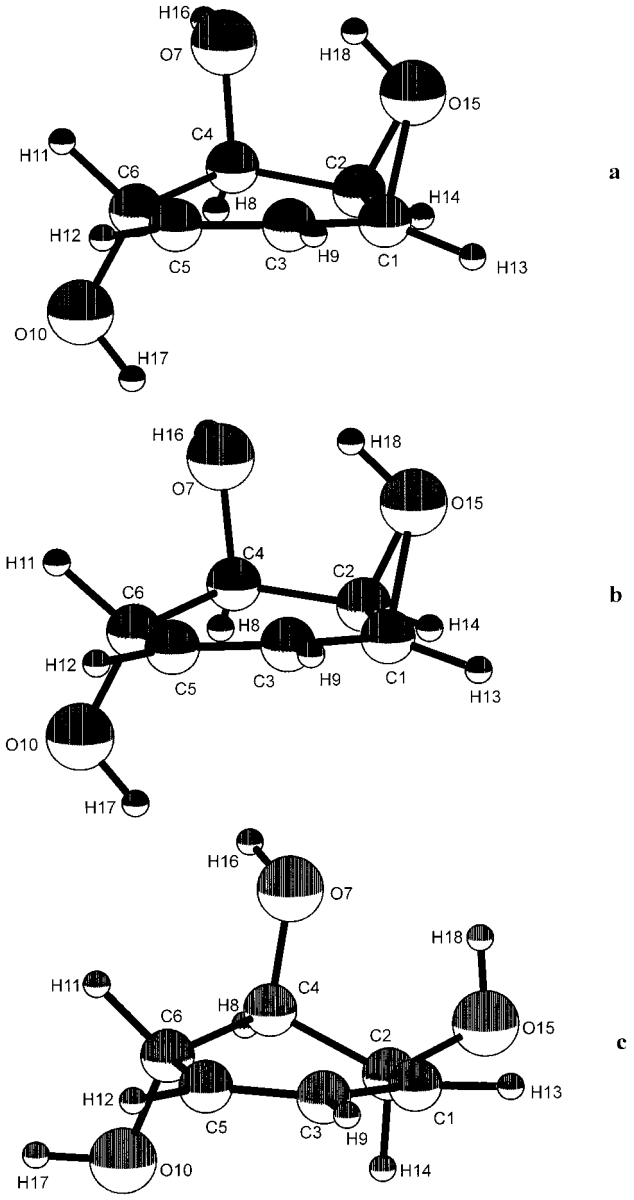


Figure 2. Calculated structures for O-protonated benzene diol epoxide: (a) closed minimum, (b) transition state for the epoxide opening, and (c) open carbocation.

conjugates). The reactions are shown in Scheme 1 , and the calculated results are presented in Table 5. The products resulting from a trans addition were considered for the substitution reactions because an S_N2 mechanism had been previously reported.[32] In that study an unsymmetrical transition state in which the epoxide C−O bond breaking is more advanced than the formation of the C-nucleophile bond is suggested, so that a partial positive charge is developed at the position of attack. Trans opening of the epoxide by the amino active sites of DNA has been reported.[33]

For benzene oxide, calculations showed that phenol formation is thermodynamically preferred over substitution by nucleophilic attack. This preference was more markedly evidenced by the ab initio results. In contrast, for benzene diol epoxide, substitution was the favored path, as the driving force of aromatization is not present in the rearrangement process of this compound. In its three substitution products, a hydrogen-bond interaction was observed between the new hydroxy group and the vicinal one. According to the methods of calculation employed, there was no clear distinction of a more favored reaction among the three substitution paths, although their enthalpy changes were not very different with each method. Inclusion of the solvent in the AM1 results did not produce significant modifications. These calculations coincide with the increased carcinogenic activity of diol epoxides over epoxides and agree with MINDO/3 results that had compared the enthalpy change for hydrolysis and for ring opening and subsequent alcohol formation for the same compounds.[14n]

The bay-region theory suggests a relationship between the ease of formation of a carbonium ion at the benzylic carbon atom of a bay-region and the carcinogenicity of the hydrocarbon.[10] Taking this into account, the enthalpy

Scheme 1. Calculated Reactions for Benzene Oxide and Benzene Diol Epoxide

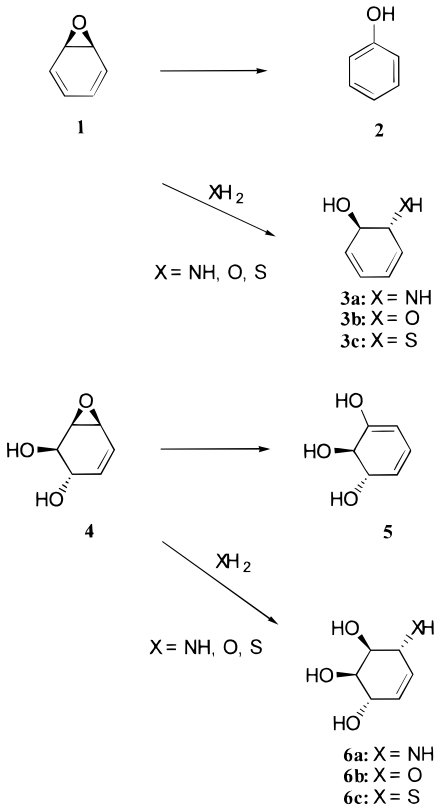


Table 5. Calculated Δ*H* for Reactions in Scheme 1 (kcal/mol)

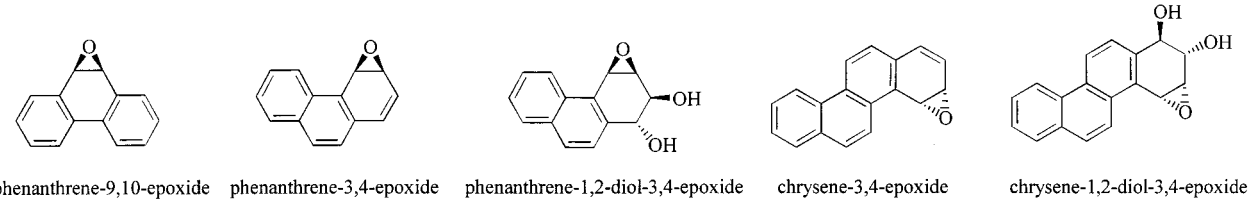
	method				
reaction	RHF/ 6-31G*	RHF/ 6-31G*[a]	PM3	AM1	AM1/SM2.1
1 → 2	−46.60	−46.93	−47.32	−50.16	−51.89
1 → 3a	−22.12	−18.35	−36.86	−42.69	−42.72
1 → 3b	−25.60	−21.52	−32.60	−38.47	−36.51
1 → 3c	−24.64	−19.73	−34.98	−47.04	−47.73
4 → 5	−16.11	−16.71	−26.04	−30.20	−30.99
4 → 6a	−26.94	-−22.76	−36.83	−42.63	−43.17
4 → 6b	−29.48	−25.02	−32.46	−40.54	−38.49
4 → 6c	−29.40	−24.29	−34.26	−48.58	−48.90

[a] Including zero-point vibrational contribution.

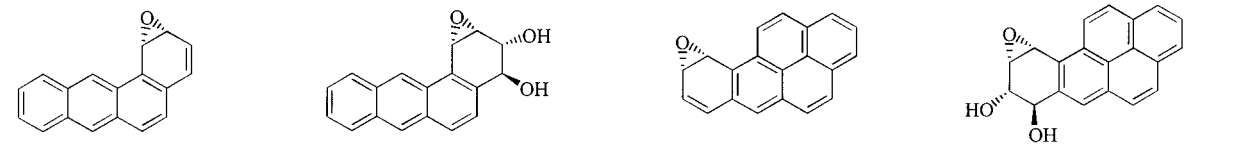
(32) Nashed, N. T.; Bax, A.; Loncharich, R. J.; Sayer, J. M.; Jerina, D. M. *J. Am. Chem. Soc.* **1993**, *115*, 1711−1722.

(33) (a) Thomson, C. *Carcinogenesis* **1989**, *10*, 317. (b) Venegas, R. E.; Reggio, P. H.; Rabinowitz, J. R. *Int. J. Quantum Chem.* **1992**, *41*, 497. (c) Venegas, R. E.; Reggio, P. H.; Rabinowitz, J. R. In *Polycyclic Aromatic Compounds PAH XIII*; Garrigues, P., Lamotte, M., Eds.; Gordon and Breach: Yverdon, Switzerland, 1993; pp 841−848.

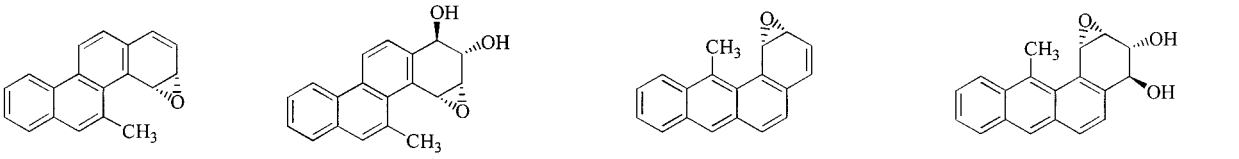
Scheme 2. Epoxides and Diol Epoxides Considered in Table 6



phenanthrene-9,10-epoxide **7** | phenanthrene-3,4-epoxide **8** | phenanthrene-1,2-diol-3,4-epoxide **9** | chrysene-3,4-epoxide **10** | chrysene-1,2-diol-3,4-epoxide **11**



benzo[*a*]anthracene-1,2-epoxide **12** | benzo[*a*]anthracene-3,4-diol-1,2-epoxide **13** | benzo[*a*]pyrene-9,10-epoxide **14** | benzo[*a*]pyrene-7,8-diol-9,10-epoxide **15**



5-methylchrysene-3,4-epoxide **16** | 5-methylchrysene-1,2-diol-3,4-epoxide **17** | 12-methylbenzo[*a*]anthracene-1,2-epoxide **18** | 12-methylbenzo[*a*]anthracene-3,4-diol-1,2-epoxide **19**

Table 6. Calculations for the Epoxide Opening Reaction of the Protonated Compounds

compound	ΔH_r (kcal/mol) AM1	ΔH_r (kcal/mol) PM3	AM1 charge at carbocationic center
1	−31.38	−30.71	0.028
7	−26.93	−25.77[a]	0.100
8	−34.20[a]	−33.66[a]	0.026
10	−33.24[a]	−32.84[a]	0.039
12	−36.89[a]	−37.16[a]	0.038
14	−37.16[a]	−36.85[a]	0.009
16	−33.45[a]	−32.66[a]	0.023
18	−38.47[a]	−38.15[a]	−0.032
4	−23.28	−22.05[a]	0.200
9	−33.49[a]	−31.14[a]	0.109
11	−33.13[a]	−32.72[a]	0.080
13	−38.34[a]	−35.86[a]	−0.004
15	−42.98[a]	−41.95[a]	−0.013
17	−34.46[a]	−33.40[a]	0.056
19	−42.77[a]	−41.48[a]	−0.036

[a] Value estimated by keeping fixed the O−C−C angle of the epoxide ring, as the full-optimized protonated species collapsed into the open carbocation.

change involved in the ring opening process for several protonated epoxides and the anti isomers of their *trans*-diol was compared. Anti isomers were selected considering their higher activity in relation to the syn isomers. Structures are shown in Scheme 2 , and results are summarized in Table 6. Considering the size of the compounds under study, semiempirical methods were employed. AM1 has proved to afford molecular geometries that are in good agreement with experimentally measured values for PAHs[14a,34] and has been found to be well-suited to study specific effects of the methyl-group substitution on them.[14l]

It was found that ΔH of the reaction becomes more exothermic on going from a K-region to a bay-region epoxide, as can be seen by comparing the values for phenanthrene-9,10-epoxide (**7**) and phenanthrene-3,4-epoxide (**8**). Moreover, the protonated bay-region compound is not a minimum on the potential energy surface by both AM1 and PM3 but opens spontaneously upon protonation. The same instability of the protonated species was observed for all of the calculated bay-region structures. In this way, the open carbocations were obtained by protonation of the neutral epoxides. The corresponding ΔH's for opening were estimated by keeping fixed the O−C−C angle of the epoxide ring in the protonated form. Spontaneous opening upon protonation had been reported by an AM1 study of PAH epoxides.[14a] This behavior has been explained in a previous MNDO study by the suggestion that the little conformational flexibility of the fused aromatic ring systems cannot relieve sterically strained nonbonded interactions (bay-region hydrogen and bay-region epoxide hydrogen at a distance of less than twice the van der Waals radius).[14b]

Diol epoxides opened spontaneously upon protonation, with the exception of benzene-1,2-diol-3,4-epoxide, which was determined to be a minimum by AM1 and presented a less exothermic ΔH for opening than benzene oxide. MNDO calculations had yielded this structure to be stable, in addition to protonated phenanthrene-1,2-diol-3,4-epoxide (**9**).[14b] Experimental results had shown that benzo[*a*]pyrene-7,8-diol-9,10-epoxide (BDPE) (**15**), which is highly mutagenic, is difficult to prepare because of its high reactivity.[35] For BDPE hydrolysis, protonation and ring opening occur in a concerted manner for general acids with pK_a's < 8.[36] The present calculated ΔH's for opening roughly correlate with experimental kinetic rate constants (M^{-1} s^{-1}) for hydrolysis of the PAH bay-region

(34) Yagi, H.; Thakker, D. R.; Hernandez, O.; Koreeda, M.; Jerina, D. M. *J. Am. Chem. Soc.* **1977**, *99*, 1604−1611.

anti-diol epoxides: 160 for phenanthrene, 127 for chrysene, and 1400 for benzo[*a*]pyrene.[37] Moreover, as the epoxide ring of the diol epoxide becomes increasingly unstable to protonation and the ΔH for ring opening is more exothermic, carcinogenicity increases: benzene (no information available about its carcinogenic activity), phenanthrene (noncarcinogenic),[38] chrysene (weakly carcinogenic),[38,39] and benzo[*a*]pyrene (strongly carcinogenic)[38,40] In this way, these calculations correlate very well with experimental observations.

The ease of carbonium ion formation, expressed by a more negative ΔH, correlated with the stabilization by delocalization of the open structures, seen as a decreasing net positive charge at the benzylic carbon, in both the epoxide and diol epoxide series. Moreover, negative charges were observed at the benzylic carbon in the compounds for which the epoxide ring opening afforded the more negative values. Thus, the observed trend was that formation of the open carbocation was more favored as the positive charge at the benzylic carbocation diminished by delocalization into the π system. In this way, the positive charge at the benzylic carbon decreases as the compound becomes more conjugated by the presence of fused aromatic rings. The same increment in stability as the positive charge is more diffuse had been reported.[14a] AM1 charges had been successfully matched in comparison with NMR studies of charge distribution in this type of carbocations.[14h−j]

According to their more exothermic ΔH's, bay-region methyl compounds (5-methylchrysene and 12-methylbenzo[*a*]anthracene derivatives) exhibited a larger tendency to open the protonated epoxide ring than did the corresponding nonmethyl structures (Table 6). This fact agrees with the greatest carcinogenic potency of the methylated compounds. Following the general trend, the charge was less positive at the benzylic position for the open methylated carbocations. As no depletion of the C−H bonds of the methyl group was observed by NBO analysis, stabilization of the open structures due to hyperconjugative effects was precluded. Instead, distortion of the methylated molecules by nonplanarity of the aromatic system was observed in the closed (fixed) and open structures, as the rings do not remain in the same plane but present a twisted conformation. Angular and torsional distortion had been observed by X-ray diffraction in the very potent carcinogen dibenzo[*def*,*p*]chrysene and in various PAHs containing fjord- or bay-region methyl groups.[32,41] It follows from these calculations that the increased activity of these overcrowded molecules arises from the greater instability of their epoxide ring upon protonation rather than from formation of a more stabilized carbocation. Similar MNDO results concerning the destabilization of 5-methylchrysene-1,2-diol-3,4-epoxide (**17**) had been reported.[14b]

Conclusions

Comparisons in reactivity of the model compounds show the thermodynamic preference of benzene oxide to rearrange to phenol after carbocation formation, aromatization acting as the driving force of this detoxification process. However, substitution reactions achieved by attack from nucleophiles are favored over a triol formation path for benzene diol epoxide. Although the calculated activation energies for both compounds are very low, the diol epoxide exhibits a greater tendency for opening of the epoxide ring upon protonation despite its lower exothermic enthalpy change. These observations explain the higher carcinogenic activity of diol epoxides over that of epoxides derived from PAHs, as carbocation formation is considered the rate-determining step in the mechanism of bonding to DNA. The decrease in the barrier for opening of the diol epoxide is ascribed to a hydrogen-bond interaction between the hydrogen atom of the protonated epoxide and the oxygen of the closest hydroxy group, an observation that agrees with the increased activity of anti over syn isomers of *trans*-diol epoxides.

Bay-region structures open more readily, in accordance with their higher carcinogenic potency. The increase in the exothermicity of the opening step correlates with a greater charge delocalization in the carbocation formed, which decreases the net positive charge at the benzylic carbon as the structure becomes more conjugated. Methyl substitution at the bay region favors the opening of the epoxide ring, in agreement with the enhanced biological activity of these derivatives. This fact arises from the instability of the closed species, which presents a distorted nonplanar structure, and not by stabilization of the open carbonium ion due to hyperconjugative effects of the methyl group.

The present calculations are in good agreement with experimental evidences concerning the reactivity of the compounds under study, which accounts for their reliability. According to this, the carcinogenic potency of new PAH derivatives could be theoretically predicted by the methods employed in this work. The same trend observed between the semiempirical methods and the more rigorous density functional and ab initio results supports the validity of the former in the study of this type of molecules and encourages its applicability for larger systems. Aqueous-phase calculations did not reveal an important solvent effect.

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