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# Metabolic Activation of Polycyclic Aromatic Carcinogens: A Theoretical Study

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SUMMARY

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Hückel molecular orbital calculations have been performed on a series of polycyclic aromatic hydrocarbons (PAH) to obtain bond localization energies and $\pi$-electron stabilization energies for the reaction sequence through which the "bay region" diol epoxide is formed. These calculations suggest that carcinogenic PAH and non-carcinogenic PAH may exhibit different behavior at three points in the sequence. The highest localization energies for formation of the initial epoxide are exhibited exclusively by a block of non-carcinogenic PAH, suggesting that they may form only minimal amounts of the initial dihydrodiol. Of the remaining PAH, the carcinogens generally exhibit greater $\pi$-electron stabilization than do non-carcinogens following opening of both the initial epoxide and the diol epoxide rings, possibly indicating more facile production of the initial dihydrodiol and of the final biomolecule adduct. The relative potencies of the twelve carcinogenic PAH considered, as measured by the Iball Index, can be satisfactorily reproduced (correlation coefficient = 0.90) through an equation combining indices for formation of the initial epoxide and for $\pi$-electron stabilization of its ring-opened cation.

## INTRODUCTION

The relative structural simplicity of polycyclic aromatic hydrocarbons (PAH) as unsubstituted, planar, polyaromatic systems, has elicited considerable theoretical effort to relate their structures and reactivities to their mode of action in chemical carcinogenesis (1-10). The known degrees of carcinogenic potency of a large number of related PAH have provided an experimental measure of the relevance of various theoretical models, with the most extensively elaborated attempt to correlate calculated molecular properties with carcinogenic strength being the work of the Pullmans (1, 2). Applying the concept of localization energies (11, 12) to predict reactivities in specific regions of PAH, they found that, with a few exceptions, carcinogenic PAH displayed low localization energies (i.e., high reactivity) in the mesophenanthrenic "K-region" double bond, combined with high localization energies (i.e., low reactivity) in the meso-anthracenic "L-region." On this basis they proposed that reaction through the K-region, probably to link PAH with cellular receptors, was necessary for carcinogenesis.



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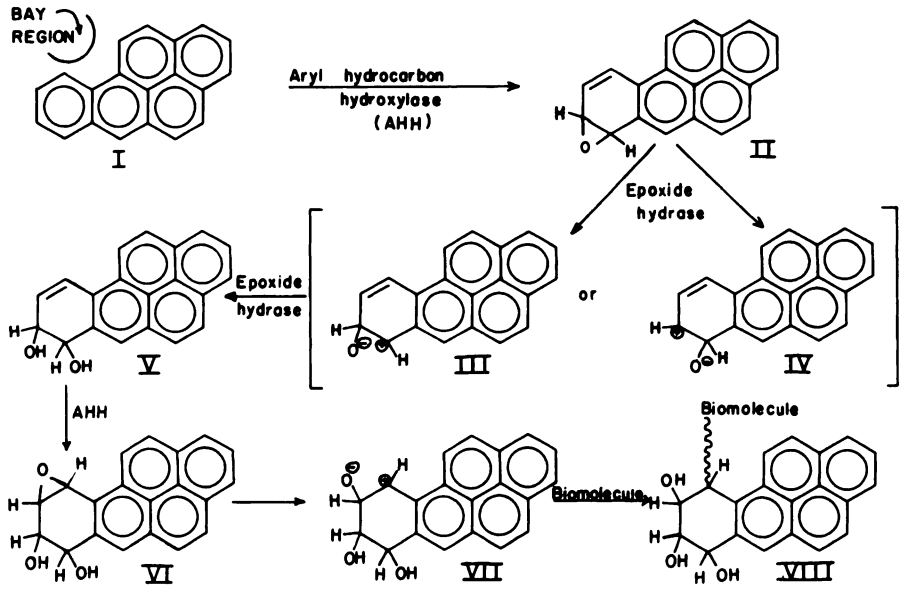
Recent work, however, has cast doubt upon the role of the K-region in carcinogenesis, implicating instead reaction at an angular benzo ring in what Jerina and Daly have called the "bay region" (7, 8, 13–18). Metabolism in this region appears to provide a major route, though not necessarily the sole route, for metabolically-induced binding of PAH to DNA *in vivo* (14, 15, 19), and generates metabolites whose carcinogenic and mutagenic properties are strongly consistent with involvement in carcinogenesis (7, 8, 16–18). In the case of the potent carcinogen benzo[a]pyrene (BaP), (I), for example, the bay region metabolic reactions involved in nucleic acid binding appear to be as follows (17, 18):




SCHEME 1

In this reaction sequence, the "ultimate carcinogen" is seen to be the diol epoxide VI.

Accordingly, we felt it would be fruitful to apply molecular orbital methods to reexamine the whole question of PAH carcinogenicity within the context of the bay region hypothesis. It is an important precondition for this work that, although the reactions involved are enzymatic in nature, calculations on isolated molecules should be able to predict trends in reactivities among sterically equivalent sites in different molecules of similar geometries. While it is not possible to rigorously test this assumption, our calculations agree closely with known product distributions of PAH metabolism, and exhibit suggestive parallels between calculated reactivities and carcinogenic potencies of PAH. While parallels do not require causation, the observed consistency with experiment is encouraging and suggests possible avenues for experimentation.

## RESULTS

Since the presence of a bay region has been postulated as a necessary (although not sufficient) condition for carcinogenicity (16), we performed simple Hückel molecular orbital calculations[1] on an extended series of bay-region-containing PAH and their metabolites as they progress through a series of transformations like that shown in Scheme I. The results of these calculations are presented in Table 1. The calculations involved the following energetic parameters for PAH reactions analogous to those shown for BaP in Scheme I: 1) *ortho-*

[1] The Hückel calculations were non-iterative and employed uniform Coulomb terms ($\alpha$) for each carbon atom and uniform bond integrals ($\beta$) for each bond.

TABLE 1

*Calculated values of PAH energy parameters corresponding to mechanism shown in Scheme I*

| Name | Loc. E (I → II) (−β) | E(III) − E(II) (β) | E(IV) − E(II) (β) | Loc. E (V → VI) (−β) | E(VII) − E(VI) (β) |
|---|---|---|---|---|---|
| Dibenzo[a,c]naphthacene (−) | 3.394 | 0.931 | 1.024 | 2.450 | 0.850 |
| Dibenz[a,c]anthracene (−) | 3.392 | 0.923 | 1.014 | 2.448 | 0.833 |
| Benzo[e]pyrene (−) | 3.383 | 0.915 | 1.011 | 2.447 | 0.826 |
| Dibenzo[a,l]pyrene (−) | 3.382 | 0.910 | 1.007 | 2.446 | 0.821 |
| Triphenylene (−) | 3.380 | 0.903 | 1.002 | 2.446 | 0.813 |
| Tetrabenzo[a,c,f,h]naphthalene (−) | 3.348 | 0.864 | 1.011 | 2.448 | 0.830 |
| Benzo[a]pentacene (−) | 3.345 | 0.873 | 1.042 | 2.452 | 0.878 |
| Benzo[a]naphthacene (−) | 3.343 | 0.867 | 1.035 | 2.452 | 0.867 |
| Dibenzo[a,j]naphthacene (−) | 3.340 | 0.860 | 1.031 | 2.451 | 0.861 |
| Dibenzo[a,l]naphthacene (−) | 3.340 | 0.862 | 1.029 | 2.451 | 0.858 |
| Benz[a]anthracene (±) | 3.337 | 0.850 | 1.022 | 2.450 | 0.848 |
| Dibenz[a,j]anthracene (+) | 3.329 | 0.844 | 1.014 | 2.449 | 0.833 |
| Dibenz[a,h]anthracene (++) | 3.329 | 0.839 | 1.017 | 2.450 | 0.840 |
| Benzo[c]pentaphene (−) | 3.326 | 0.837 | 1.016 | 2.434 | 0.838 |
| Tribenz[a,c,h]anthracene (−) | 3.324 | 0.835 | 1.011 | 2.447 | 0.828 |
| Phenanthrene (−) | 3.319 | 0.822 | 1.001 | 2.446 | 0.812 |
| Dibenzo[c,g]phenanthrene (−) | 3.306 | 0.823 | 0.995 | 2.442 | 0.798 |
| Picene (−) | 3.305 | 0.812 | 1.001 | 2.445 | 0.811 |
| Benzo[c]chrysene (+) | 3.305 | 0.827 | 0.992 | 2.445 | 0.795 |
| Benzo[c]phenanthrene (+) | 3.299 | 0.822 | 0.987 | 2.440 | 0.784 |
| Chrysene (−) | 3.299 | 0.807 | 0.995 | 2.446 | 0.803 |
| Benzo[b]chrysene (−) | 3.293 | 0.808 | 0.998 | 2.444 | 0.805 |
| Dibenzo[b,g]phenanthrene (−) | 3.292 | 0.826 | 0.986 | 2.440 | 0.780 |
| Tribenzo[a,e,i]pyrene (++) | 3.286 | 0.794 | 1.029 | 2.450 | 0.861 |
| Benzo[g]chrysene (++) | 3.285 | 0.808 | 0.984 | 2.439 | 0.777 |
| Dibenzo[a,e]pyrene (+++) | 3.276 | 0.788 | 1.019 | 2.449 | 0.846 |
| Dibenzo[a,i]pyrene (++++) | 3.274 | 0.782 | 1.042 | 2.458 | 0.891 |
| Benzo[a]pyrene (++++) | 3.261 | 0.775 | 1.032 | 2.455 | 0.867 |
| Dibenzo[a,h]pyrene (++++) | 3.249 | 0.773 | 1.041 | 2.456 | 0.882 |
| Dibenzo[a,l]pyrene (++++) | 3.246 | 0.776 | 1.012 | 2.441 | 0.830 |

localization energy[2] (11, 12) for formation of the first epoxide II (Column 2, Table 1); 2) the difference in total π-electron energies between II and each of the two potential ring-opened intermediates, III and IV (Columns 3 and 4, respectively, Table 1); 3) *ortho*-localization energy for formation of the diol epoxide VI, from the dihydrodiol V (Column 5, Table 1); 4) the difference in total π-electron energies between VI and the ring-opened intermediate VII (Column 6, Table 1).

Before entering into a detailed discussion of the values contained in Table 1, several general features of the table should be mentioned. First, the compounds are listed in order of decreasing localization energies

[2] *Ortho*-localization energies were calculated by subtracting the total π-electron energy for the reacting species from the total π-electron energy of the fragment obtained by removal from the π-system of the two carbons and two π-electrons involved in the epoxidation. The latter fragment represents the conjugated portion of the σ-complex formed by localization of two π-electrons on the reacting carbons as they interact with the attacking species. As the σ-complex is thought to closely resemble the transition state, the difference in energy between its π-system and that of the reacting species should, for a series of related compounds undergoing the same reaction, be proportional to the activation energy. Our computation of the *ortho*-localization energy differs slightly from that of Pullman (reference 1.) in that we have considered the two "localized" π-electrons in the transition state to occupy non-interacting p orbitals (sum of electron energies = 2α) rather than an ethylene-type π-bond (sum of electron energies = 2α + 2β). This does not affect the relative ordering of the localization energies through the series of PAH studied, but results in a uniform increase of 2 in the absolute value of the coefficient of β.

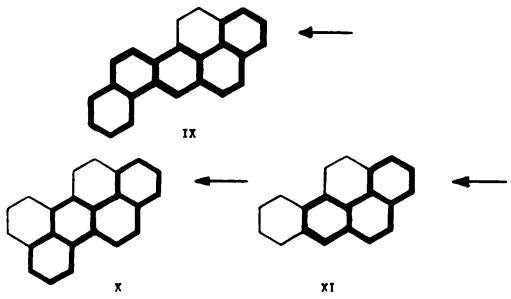
(for a series of closely related molecules, this should correspond to increasing reaction rates) for formation of the first epoxide. Second, only $\pi$-electron energies are considered here. The implications of this will be discussed in a later section. Third, when more than one nonequivalent bay region is present in a molecule, the values given in Table 1 are those for the reaction sequence at the bay region which displays the lowest localization energy for the initial reaction: formation of the first epoxide.[3] Fourth, the parenthetical values beside the name of each compound refer to carcinogenic strength (4, 20).

## DISCUSSION

*Ortho-localization energy for formation of first epoxide II.* Column 2 in Table 1 presents the calculated *ortho*-localization energy values for the first step in the postulated series of PAH metabolic activation reactions: formation of the initial epoxide. The top ten compounds, for which initial epoxidations are calculated to occur at the slowest rates, are seen to be carcinogenically inactive. A "borderline" compound follows, benz[a]anthracene, which has been shown to be weakly active in certain systems (21). The compounds falling below the top eleven on the list are almost evenly divided between carcinogens and noncarcinogens, although the bottom seven compounds are, without exception, carcinogens.

Predictions based on these values agree substantially with the results of product distribution studies carried out in several laboratories (22–24). In the cases of dibenz[a,c]anthracene and benzo[e]pyrene, two molecules appearing in the top ten of Table 1, no metabolic products (dihydrodiols or phenols) were found in the regions of the molecules analogous to the position of initial epoxidation in BaP (Scheme I). In the case of the "borderline" benz[a]anthracene, phenols were formed in this region (presumably through spontaneous rearrangement of the epoxide), although only as minor products (phenols at carbons 3 and 4 forming 17% of the total products arising from rat liver homogenates). Of the four molecules investigated which fell below the top eleven in Table 1, three have significant amounts of products (dihydrodiols and phenols) arising in the region of "first epoxidation" (dibenz[a,h]anthracene (carbons 3 and 4): 75% of total products and 77% of total products for microsomal and liver homogenate oxidation, respectively; phenanthrene (carbons 1 and 2): 47% of total products and 48% of total products for microsomal and liver homogenate oxidation, respectively; chrysene (carbons 1 and 2): 44% of total products and 33% of total products for microsomal and liver homogenate oxidation, respectively).

In the case of a fourth molecule, BaP, which falls in the bottom section of Table 1, the product in the region of first epoxidation (the 7,8-dihydrodiol) forms only 5–15% of total metabolic products in the several systems studied (23–26). The predominant metabolite is found to be the 3-OH derivative, although the 2,3 bond is predicted to be considerably less reactive than the 7,8 bond (1). That BaP deviates from the behavior toward initial epoxidation exhibited by the other six compounds may be due to a difference in binding orientation within the active site of the monooxygenase enzyme. If one asumes that the enzyme recognizes a common feature of PAH structure, one can seek possible common binding orientations for the PAH by superimposing their molecular skeletons so as to maximize overlap. One such set of orientations, shown below for BaP (normal lines) superimposed upon which are structures (heavy lines) of dibenz[a,h]anthracene (IX), chrysene (X), and phenanthrene (XI), results in the "first epoxidation" bond of the latter three compounds (identified by arrows) corresponding exactly in position to the 2,3 bond of BaP:

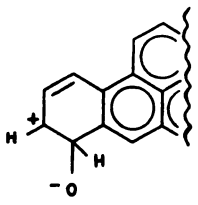



[3] Since nonequivalent bay-regions may not be isosteric, this assumption may fail. However, lacking additional data, we have chosen to make it.

While this behavior illustrates that isolated-molecule calculations may not succeed in all cases in predicting reactivity trends in these enzyme-catalyzed reactions, it is encouraging that in only one molecule out of the seven which we could study was such an exception noted. Further, as both carcinogens and noncarcinogens are present in the bottom two-thirds of Table 1, it is clear that for these molecules carcinogenicity cannot be correlated in any simple way with extent of initial epoxidation.

The above experimental results do, however, suggest a rough correlation between the proportion of a PAH's metabolism occurring at the "first epoxidation" bond, and the predictions of the localization energy calculations. Thus, compounds for which formation of the first epoxide is predicted to be difficult (high *ortho*-localization energy) tend experimentally to undergo little or no metabolism at that bond.

*Stabilization of cationic π-system (III or IV) from ring opening of first epoxide II.* Columns 3 and 4 of Table 1 indicate that the cationic π-system corresponding to IV in Scheme I is, for all PAH studied, predicted to be more stable relative to the parent epoxide than the cationic π-system corresponding to III. This prediction can be compared with experiment through consideration of the reaction by which the phenol is formed from the epoxide. The reaction proceeds through a mechanism, involving the "NIH shift" (16), in which the rate-determining step involves opening of the oxirane ring to form a cation bearing a positive charge on the carbon adjacent to the carbon which will ultimately bear the hydroxyl group (27). Our calculations of the relative stabilities of cations III and IV (Table 1) predict the mode of ring opening in every case to be




leading to a phenol bearing the hydroxyl group at a position corresponding to carbon 7 of BaP (i.e., at the carbon atom more remote from the bay region within the "first epoxide" bond). These predictions are in excellent agreement with product distribution studies (22, 26), as well as with the theoretical results of Harvey *et al.* (10), based on the perturbational molecular orbital (PMO) method (28). In the case of BaP itself, rearrangement of the 7,8-epoxide was found by Waterfall and Sims to yield exclusively the predicted 7-OH product (26). In the cases of three other PAH studied by Sims (22), the phenol formed at the "first epoxide" bond had the hydroxyl group predominantly in the predicted position: Chrysene metabolized by a rat liver homogenate system yielded the predicted 1-OH isomer over the 2-OH isomer in a ratio of 4.8:1. Phenanthrene, under analogous conditions, yielded the predicted 1-OH isomer over the 2-OH in a ratio of 1.9:1. Benz[a]anthracene favored formation of the predicted 4-OH isomer over the 3-OH isomer by ratios of 1.4 and 1.67 for liver microsomal and homogenate metabolism, respectively. In addition, Hecht *et al.* (29) have recently reported that metabolism of 5-methylchrysene likewise affords the predicted 1-OH and 7-OH isomers, but no detectable 2-OH or 8-OH compounds. Dibenz[a,h]anthracene proved an exception, in producing the alternative (3-OH) isomer at a ratio (compared with the predicted 4-OH isomer) of 1.4:1 for the microsomal system and 1.5:1 for the homogenate system. Interestingly, for the four compounds exhibiting preferential production of the phenol at the predicted position,[4] the relative ratios by which production of the predicted phenol exceeds that for the alternative phenol (BaP > chrysene > phenanthrene > benz[a]anthracene) parallel the differences in calculated stabilization energy (Table 1) between the preferred and the alternative cations (BaP = $0.257\beta$; chrysene = $0.188\beta$; phenanthrene = $0.179\beta$; benz[a]anthracene = $0.172\beta$).

Although the cation stabilization energies in column 4, Table 1 exhibit a wide variation, seemingly unrelated to carcinogenicity, elimination of those PAH whose

[4] 5-Methylchrysene is not included here since our calculations do not make allowance for the methyl group.

high *ortho*-localization energies make formation of the initial epoxide unlikely (i.e., the top ten—all noncarcinogens) discloses a pattern among the remaining twenty compounds. With four exceptions, the extent of cation stabilization for the carcinogens (lying between 1.012$\beta$ and 1.042$\beta$) exceeds that for the noncarcinogens (lying between 0.986$\beta$ and 1.011$\beta$). The presence of exceptions (benzo[c]phenanthrene, benzo[c]chrysene, benzo[g]chrysene, and benzo[c]pentaphene) at this point need not be surprising, however, as differing reactivities at a single step in the activation sequence are unlikely to be sufficient to distinguish carcinogens from noncarcinogens. It is more probable that the cumulative effects of differing reactivities at several steps would be a significant determinant of a compound's carcinogenicity, and a later section of this paper discusses improved results using a compound index of carcinogenicity.

Nevertheless, the cation stabilization energy does discriminate roughly between carcinogens and noncarcinogens, a point that has been noted previously by others (7, 8). While gratifying, this energy parameter is unable to distinguish levels of carcinogenic potencies among the carcinogens. For example, a plot of cation stabilization energy versus Iball Index for the bottom twenty compounds in Table 1 (Fig. 1, Table 2) exhibits considerable scatter. However, since the Iball Index, given its most optimistic interpretation, must reflect minimally the ease of progression of the PAH through both the epoxidation and hydration steps, with possibly significant contributions from the remaining metabolic steps, the expectation of a simple relationship must, as mentioned above, be naive. We will return to this point.

*Ortho-localization energy for formation of second epoxide VI from dihydrodiol V.* The values shown in Column 5, Table 1, show relatively little significant variation through the PAH studied in the rates of formation of the second epoxide VI from the dihydrodiol V. Since there are virtually no published comparative experimental data on formation of diol epoxides (or their metabolic products, bis-(dihydrodiols) or dihydrodiol-phenols), little detailed analysis of our calculated values is possible. However, Berger *et al.* have recently reported

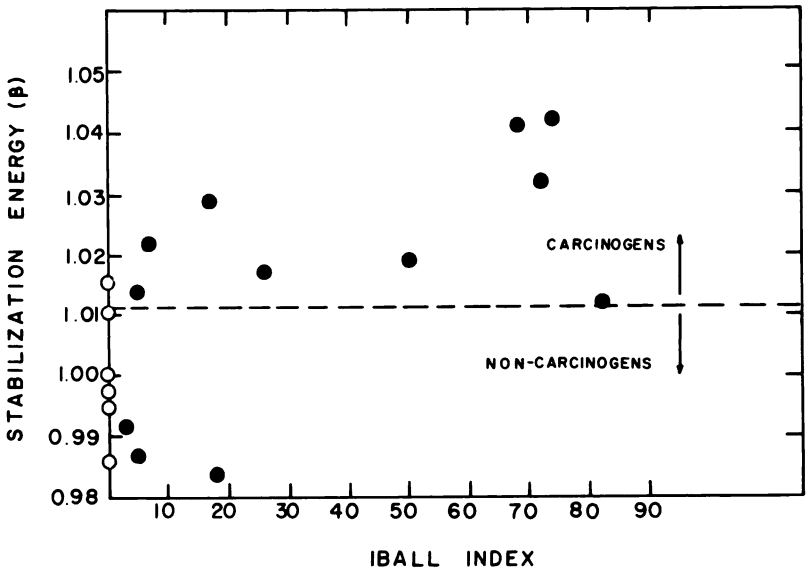



FIG. 1. *Stabilization energy of cation of type IV (Scheme I) vs Iball Index of parent compound*

Compounds plotted are carcinogens (●) and non-carcinogens (○) falling below the top ten compounds in Table 1. Values of cation stabilization energies for non-carcinogens are taken from Table 1, column 4, and for carcinogens from Table 1, column 4, or Table 2, column 2. Iball Indices are taken from Table 2, column 4. The dashed line represents a rough division in cation stabilization energies between carcinogens and non-carcinogens.

TABLE 2

*Cation stabilization energies[a] and Iball indices[b] for carcinogens in Table 1*

| Name | E(IV) − E(II) (β) | E(VII) − E(VI) (β) | Iball index |
|---|---|---|---|
| Dibenzo[a,l]pyrene | 1.012 | 0.830 | 82 |
| Dibenzo[a,i]pyrene | 1.042 | 0.891 | 74 |
| Benzo[a]pyrene | 1.032 | 0.867 | 72 |
| Dibenzo[a,h]pyrene | 1.041 | 0.882 | 68 |
| Dibenzo[a,e]pyrene | 1.019 | 0.846 | 50 |
| Dibenz[a,h]anthracene | 1.017 | 0.840 | 26 |
| Benzo[g]chrysene | 0.984 | 0.777 | 18 |
| Tribenzo[a,e,i]pyrene | 1.029 | 0.861 | 17 |
| Benz[a]anthracene | 1.022 | 0.848 | 7 |
| Dibenz[a,j]anthracene | 1.014 | 0.833 | 5 |
| Benzo[c]phenanthrene | 0.987 | 0.784 | 5 |
| Benzo[c]chrysene | 0.992 | 0.795 | 3 |

[a] Values taken from Table 1, columns 4 and 6.

[b] Values taken from text references 4 and 20.

that carcinogenic PAH tend to have high superdelocalizability indices at the bond of second epoxidation (9).

*Stabilization of cationic π-system VII from ring opening of diol epoxide VI.* Cation stabilization energies derived from oxirane ring-opening of dihydrodiol epoxides VI (Column 6 of Table 1) display a pattern which parallels closely the corresponding relation found for the stabilization of IV. In the present case, the extent of stabilization for the carcinogens varies between 0.830β and 0.891β, exceeding that for the noncarcinogens, which varies between 0.780β and 0.828β. As before, the cation stabilization energy is incapable of accounting for variations in carcinogenicity among the carcinogens. A plot of Iball Index of parent compounds versus extent of cation VII stabilization (not shown) presents an almost exact replica of the relation found in the case of cation IV.

These results are in general agreement with those of Jerina *et al.* (7, 8), who have used a PMO approach (28) to demonstrate that: (1) benzylic carbonium ion formation in the bay region of a given PAH (corresponding to the cationic π-system in compound VII) produces the greater energetic stabilization compared with formation of other possible benzylic carbonium ions, and (2) there is a very rough relation between carcinogenic strength of PAH and extent of stabilization of their bay region benzylic carbonium ions.

As indicated above, while *ortho*-localization energy for formation of the first epoxide and cation stabilization energies for oxirane ring-opening of epoxide and dihydrodiol epoxide can serve as indicators of noncarcinogenicity, neither reactivity index alone satisfactorily mirrors the order of carcinogenic potency, as measured by the Iball Index. Recognizing the inadequacy of characterizing such a complex biological response as tumor induction by a single number, we nevertheless felt it would be instructive to investigate whether the experimental data could be satisfactorily reproduced by a two-parameter fit involving both *ortho*-localization energy and cation stabilization energy. Multiple linear regression analysis of the relevant data in Table 2 yielded the equation

$$\mathrm{ICI} = -722.9\ \mathrm{OLE} + 515.6\ \mathrm{CSE} + 1891.2$$

with a correlation coefficient of 0.90. Here OLE and CSE are the *ortho*-localization energy and cation stabilization energy for formation and ring-opening of the initial epoxide. ICI is the appropriate Iball Index. Only one compound out of twelve, tribenzo[a,e,i]pyrene, gave a calculated Iball Index statistically significantly different from the observed value (although it is correctly predicted to be in the carcinogenic range). Significantly, the deviations noted earlier no longer appear. This suggests that both indices are significant determinants of carcinogenicity, a point worth emphasizing in view of the almost exclusive preoccupation with carbonium ions in recent theoretical studies (7, 8, 10). In addition, application of the relation to the first ten PAHs in Table 1 leads to small (i.e., less than 10) or negative calculated Iball Indices, consistent with their noncarcinogenicity. It may be that *ortho*-localization energies and cation stabilization energies calculated by more reliable methods (e.g., self-consistent field (SCF) theory or PMO theory) might give a superior correlation, but given the questionable quantitative significance of the Iball Index, we have not felt it worth pur-

suing, but content ourselves with pointing out the importance of both epoxidation *and* hydration steps.

*Approximations involved in the calculations.* Both the Hückel molecular orbital method and the localization energy concept involve simplifying assumptions which reduce their effectiveness if one seeks absolute numerical values of selected parameters for individual molecules (30). More sophisticated molecular orbital methods have been applied to large, polyaromatic systems (5), as have alternative methods of calculation (4). Nevertheless, results obtained from the Hückel and localization energy approximations are valuable when used to compare relative values of a carefully defined parameter within a family of related molecules (30).

A more specific approximation involves the use of $\pi$-electron energies to represent the total energies of the molecules. The destabilizing influences of the strained epoxide ring and of the dipolar character of III, IV, and VII, are not explicitly taken into account in our calculations. Nor are the effects of possible internal hydrogen bonding within the angular benzo ring of the *syn* stereoisomer (31) of diol epoxide VI, which would be expected to assist epoxide ring opening (32). Nevertheless, to a first approximation, the effects exerted by these factors should represent relatively uniform perturbations through the series of PAH studied, and we have thus considered them to have little effect on the relative ordering of the values presented here.

Caution is required when applying molecular orbital reactivity indices to enzymatic reactions, as mentioned earlier. In a given PAH, differing enzyme binding affinities for different regions of the PAH molecule may predominate over the effects of differing reactivities of those regions. For example, while localization energy calculations predict the K region double bond to be the most reactive within a given PAH, product distribution studies show that in fact the K region is usually not the most extensively metabolized bond in the molecule.

On the other hand, little experimental information is available to permit comparison among a number of PAH of extent of reaction at a single site. Possibly in this case, as reaction at a common site in a group of related molecules is being compared, the effects of differing reactivities from one molecule to another could predominate over differences in binding to the enzyme. In the limited number of cases in which our calculated results were compared with the results of product distribution studies, the extent of agreement was gratifying, suggesting that here the calculated local reactivity may be the dominant factor in determining extent of reaction among these structurally related molecules.

*The model.* Our results present several suggestive relations concerning carcinogenic potency and PAH metabolism. These should be prefaced by emphasizing that our calculations, and thus our interpretations, are based on the mechanism of activation of carcinogenic PAH occurring through dihydrodiol epoxide formation adjacent to and in the bay region, and it must be recalled that direct experimental evidence for the generality of the dihydrodiol epoxide-bay region mechanism is at present limited (7, 8, 13–19).

On the basis of this mechanism, however, our calculations suggest three possible points along the pathway of formation and reaction of the diol epoxide at which carcinogenic PAH may differ from noncarcinogenic PAH. First, carcinogenic PAH exhibit sufficient reactivity at the bond of "first epoxidation" to produce at least some threshold amount of the initial dihydrodiol. In terms of the values reported in Column 2, Table 1, carcinogenic PAH exhibit an *ortho*-localization energy of $-3.337\beta$ or less.

Second, among those PAH which exhibit the above property, carcinogens undergo ring-opening reactions of the two epoxides which pass through intermediates of sufficiently high $\pi$-electron stabilization energy. Presumably, increased ease of formation of ring-opened intermediates IV and VII would provide for more facile production of the dihydrodiol V and the biomolecule adduct VIII. Additionally, the combined effects of cation stabilization energy and *ortho*-localization energy may be an improved measure of carcinogenic strength.

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