that the occupation of the π^* orbital shortens the single and lengthens the double bonds. The C-N triple bond length is 1.14 Å. We have chosen the symmetrical arrangement shown in Chart I (C_s symmetry). The planes of TCNE and the Cp rings are assumed to be parallel and separated by a distance of 3.51 Å. The resulting resonance and overlap integrals between the π^* orbital of TCNE and the e_{1g} and e_{1g}^* orbitals of ferrocenium are given in Table I. The integrals involving the orbitals b and b* vanish for symmetry reasons.

The H_{ii}'s and the Slater exponents of the extended Hückel calculation are given in Table II. The coefficients for the double-ζ expansion of the Fe d orbitals are given in parantheses.

Registry No. DMFcTCNE, 105399-77-7; ferrocenium, 12125-80-3.

Chemical Model Studies on the Mutagenesis of Benzofuran Dioxetanes in the Ames Test: Evidence for the Benzofuran Epoxide as Ultimate Mutagen

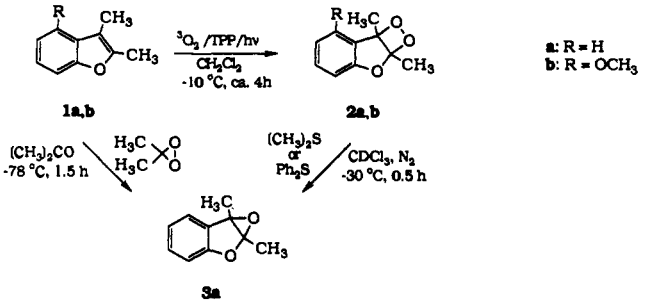
W. Adam,*,† L. Hadjiarapoglou,† T. Mosandl,† C. R. Saha-Möller,† and D. Wild‡

Contribution from the Institute of Organic Chemistry, University of Würzburg, Am Hubland, D-8700 Würzburg, FRG, and Institute of Toxicology and Pharmacology, University of Würzburg, Versbacher Strasse 9, D-8700 Würzburg, FRG. *Received February 22, 1991*

Abstract: The synthesis of the first benzofuran epoxide **3a** was achieved by epoxidation of the benzofuran **1a** with dimethyldioxirane and alternatively by deoxygenation of the benzofuran dioxetane **2a** with sulfides. This labile epoxide formed with nucleophiles such as water, methanol, thiophenol, and imidazole the corresponding adducts **13a–16a**. In contrast to epoxide **3a**, the dioxetanes **2** required acid catalysis (CF_3CO_2H) for the addition of water, methanol, and azide ion to give the corresponding adducts **9–11**; in the absence of nucleophiles the allylic hydroperoxides **8** were formed. The decomposition of benzofuran dioxetanes **2** in the polar, protic solvents water and methanol afforded not only the expected cleavage products **4** but also the 1,3-dioxols **5**, the spiroepoxide dimer **6a**, and the 1,4-dioxines **7**. An intramolecular electron-transfer mechanism is postulated for the formation of the spiroepoxide, which subsequently dimerizes to **6a** or rearranges into **5** and **7**. Only the benzofuran epoxide **3a**, besides the benzofuran dioxetanes **2**, was mutagenic in the *Salmonella typhimurium* strain TA100. Therefore, we implicate the epoxide **3a** as the ultimate mutagen responsible for the high mutagenic activity observed with dioxetane **2a** in the Ames test. We postulate that in the oxidative metabolism of polycyclic arenes and heteroarenes the corresponding epoxides are generated from the intermediary dioxetanes by deoxygenation with sulfides.

Extensive investigations on the photogenotoxicity of 1,2-dioxetanes—efficient sources of triplet excited carbonyl compounds—revealed that indeed DNA can be damaged when treated with dioxetanes under physiological conditions. For isolated calf thymus DNA pyrimidine dimers were detected[1] and for superhelical PM2 DNA pyrimidine dimer-specific repair endonucleases revealed a correspondence between the amount of dimer formation and the triplet excitation flux of alkyl-substituted dioxetanes.[2] Furthermore, additionally single strand breaks and apyrimidinic and apyrinic (AP) sites were observed, but these results suggest[1c,2,3] that such toxicological damage does not correspond to that caused by direct UV (260 nm) irradiation and appears to be more similar to that produced by singlet oxygen and radical species. In bacteria and mammalian cells the main damage was single strand breaks, which are presumably derived from active oxygen species.[1c,4] For example, in *Escherichia coli* bacteria, dioxetanes induce dose-dependent SOS function sfiA and in mammalian cells (HL-60, SHE) they generate micronuclei (damage at the chromosomal level).

Scheme I



It was, therefore, surprising that alkyl-substituted dioxetanes are not mutagenic in several *Salmonella typhimurium* strains (Ames test). Nevertheless, recently it was observed[2,5] that benzofuran dioxetanes **2** (Scheme I) are highly mutagenic in the *S. typhimurium* TA100 strain. Thus, these derivatives are the first known dioxetanes with potent mutagenicity. Since the mutagenic damage elicited by dioxetanes **2** in the *S. typhimurium* TA100 strain could not be photoreactivated,[2] the mutations seem not to be of typical photochemical origin such as pyrimidine dimers. Moreover, recent toxicological results imply[2] that an alkylating

(1) (a) Lamola, A. A. *Photochem. Photobiol.* **1969**, *9*, 291. (b) Lamola, A. A. *Biochem. Biophys. Res. Commun.* **1971**, *43*, 893. (c) Adam, W.; Beinhauer, A.; Epe, B.; Fuchs, R.; Griesbeck, A.; Hauer, H.; Mützel, P.; Nassi, L.; Schiffmann, D.; Wild, D. In *Primary Changes and Control Factors in Carcinogenesis*; Friedberg, T., Oesch, R., Eds.; Deutscher Fachschriften-Verlag: Wiesbaden, 1986; p 64. (d) Lown, J. W.; Koganty, R. R.; Kopecky, K. R. *Photobiochem. Photobiophys.* **1986**, *12*, 295.

(2) (a) Adam, W.; Beinhauer, A.; Mosandl, T.; Saha-Möller, C. R.; Vargas, F.; Epe, B.; Müller, E.; Schiffmann, D.; Wild, D. *EHP, Environ. Health Perspect.* **1990**, *88*, 89. (b) Adam, W.; Hadjiarapoglou, L.; Mosandl, T.; Saha-Möller, C. R.; Wild, D. *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 200.

(3) Epe, B.; Mützel, P.; Adam, W. *Chem.-Biol. Interact.* **1988**, *67*, 149.

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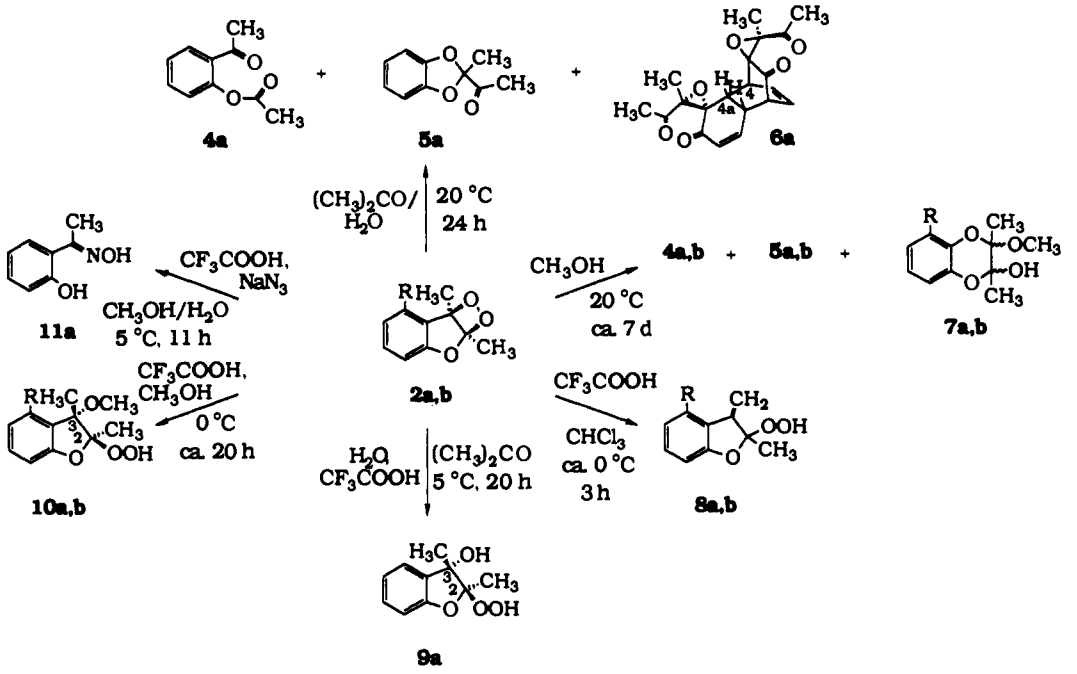
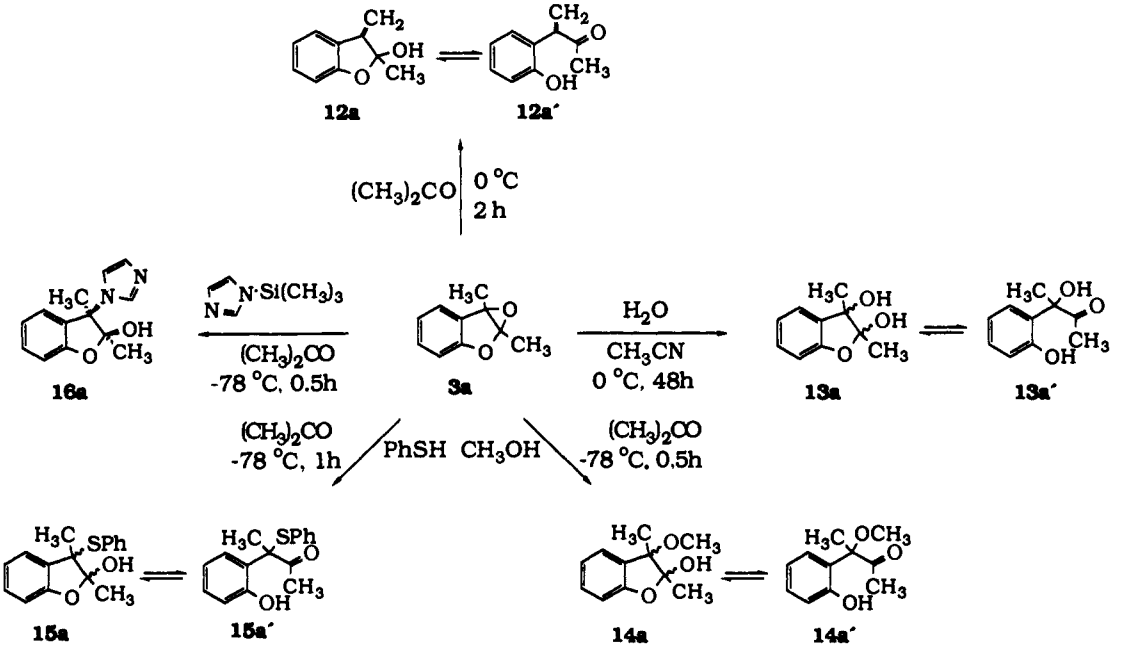
Scheme II



Scheme III



intermediate is responsible for the DNA damage. We suggest that the highly reactive benzofuran epoxides **3** (Scheme I) serve as efficient alkylating agents and constitute the ultimate mutagens derived from dioxetanes **2**.[2b]

Presently we report on the synthesis of epoxides **3** by two independent routes (Scheme I), of which one involves epoxidation of the benzofurans **1** with dimethyldioxirane[6] and the other deoxygenation of dioxetanes **2** by sulfides.[7] As chemical model studies for elucidating the molecular nature of the DNA damage, we have investigated the reaction of the novel benzofuran dioxetanes **2** (Scheme II) and their epoxides **3** (Scheme III) with nucleophiles under a variety of conditions. On the basis of our present chemical and toxicological results, we postulate that the in situ deoxygenation of polycyclic arene and heteroarene dioxetanes, produced through enzymatic oxygenation by dioxygenases directly in cells, into the highly reactive epoxides, may constitute a significant pathway (Scheme VI) for cellular DNA damage.[8]

Results

Synthesis of Epoxide 3a. On treatment of benzofuran dioxetane **2a** in deuteriochloroform with stoichiometric amounts of dimethyl or diphenyl sulfide at −30 °C,[7] ^{1}H and ^{13}C NMR monitoring of the reaction mixture at this temperature revealed after 15 min quantitative formation of benzofuran epoxide **3a** and dimethyl or diphenyl sulfoxide (Scheme I). An independent and quantitative synthesis of this epoxide, postulated as extremely labile inter-

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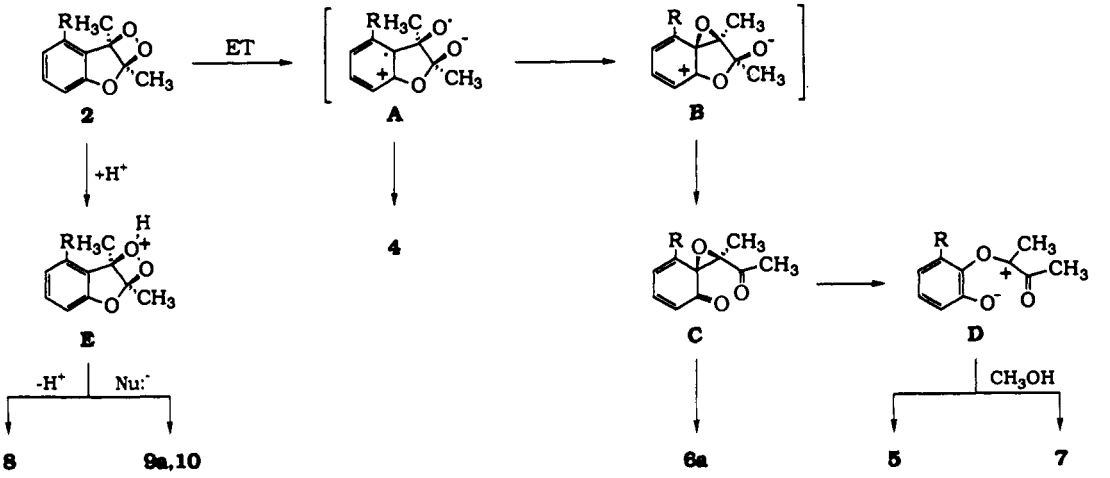
Scheme IV



mediates,[9,10] was achieved by epoxidation of 2,3-dimethylbenzo[*b*]furan (**1a**) with dimethyldioxirane[6] at −78 °C (Scheme I). The structure of the epoxide **3a** was unequivocally assigned on the basis of its NMR spectral data.

Chemical Transformations of the Benzofuran Dioxetanes 2. Whereas benzofuran dioxetane **2a** decomposes in aprotic organic solvents predominantly (>96%) into the cleavage product **4a**,[5a,11] its thermolysis in water at 20 °C afforded within 24 h three products. Column chromatography yielded, besides cleavage product **4a** (49%), the known[12] 1,3-dioxole **5a** (26%) and the unknown spiroepoxide dimer **6a** (10%) (Scheme II).

The chemical constitution of dimer **6a** was confirmed by comparison of its ^{1}H NMR spectral data with those of similar known compounds formed in the Diels–Alder dimerization of cyclohexa-2,4-dien-1-one derivatives.[13a] Additionally, satisfactory elemental analysis and spectral data (cf. Experimental Section) corroborate the assigned structure of **6a**. To assess the configuration of dimer **6a**, NOE experiments were performed. Irradiation of the methyl protons bound to the spiroepoxy rings caused ca. 14% enhancement of the **4-H** and **4a-H** protons and 4% vice versa, whereas irradiation of the acetyl protons gave no effect. Consequently, an endo and *R*,S** configuration was assigned to dimer **6a**. The high diastereoselectivity of this Diels–Alder dimerization of epoxide **C** (Scheme IV) is analogous to that found for the unsubstituted spiroepoxycyclohexadienone due to symmetry control and steric effects in the transition state.[13b,c]

In methanol, benzofuran dioxetane **2a** gave at 20 °C after 5 days, besides the cleavage product **4a**, the 1,3-dioxole **5a**, and the known[14] 1,4-dioxine **7a** (as 67:33 mixture of diastereomers) in the relative proportions 93:3:4, as established by ^{1}H NMR (Scheme II). Similarly, dioxetane **2b** afforded at 20 °C after 7 days in methanol the cleavage product **4b**, the 1,3-dioxole **5b**, and the 1,4-dioxine **7b** (as 77:23 mixture of diastereomers) in the relative proportions 52:27:21, which were isolated as analytically pure samples (Scheme II). Additional structure proof rests on the observed spectral properties (IR, ^{1}H and ^{13}C NMR, and MS).

Ring-opening of the benzofuran dioxetanes **2a,b** by rupture of the carbon—oxygen bond was observed in chloroform at subambient temperatures. In particular, acid catalysis by trifluoroacetic acid produced the hydroperoxides **8a,b** in almost quantitative yield (Scheme II). By means of NMR analysis of the reaction mixture the regioisomeric 2-methylidene-3-hydroperoxybenzofurans were not detected. The structure assignment is based on a strong peroxide test (KI and AcOH) and the expected spectral data (IR, ^{1}H and ^{13}C NMR); satisfactory elemental analyses were achieved.

Treatment of the benzofuran dioxetanes **2a,b** with catalytic amounts of trifluoroacetic acid in the presence of methanol at 0–5 °C led to the methanol adducts **10a,b** (**10a**, 61%; **10b**, 55%). The known[15] acetophenone oxime **11a** was isolated in 61% when sodium azide was used as the nucleophile. With water the extremely labile adduct **9a** was obtained in 78% (Scheme II), which eliminates water on heating at 40 °C for 2 h to give the cleavage product **4a** in quantitative yield by Hock-type cleavage.

The hydroperoxides **9a** and **10a,b** exhibit a strong peroxide test (KI in AcOH), and their structure assignment rests on spectral data (IR, ^{1}H, and ^{13}C NMR, and MS); satisfactory elemental analyses were obtained. The configuration of the diastereomerically pure adducts **9a** and **10a,b** was ascertained by NOE experiments. Irradiation of the 3-methyl protons left the vicinal 2-methyl protons unaffected. On the other hand, irradiation of the latter protons showed no effect as well on the 3-methyl protons but an ca. 2% enhancement of the 3-methoxy protons of methanol adduct **10a**. These NOE results establish the trans configuration for the hydroperoxides **9a** and **10a**. The trans configuration was also assumed for the hydroperoxide **10b** in view of the similar NMR spectral data with those of **10a**.

Chemical Transformations of the Benzofuran Epoxide 3a. Warming an acetone solution of epoxide **3a** up to 0 °C resulted in complete decomposition into the known[10] tautomeric alcohols **12a/12a′**, as revealed by ^{1}H NMR spectroscopy (Scheme III). After column chromatography, 53% of the tautomers **12a/12a′** were isolated.

Since benzofuran dioxetane **2a** can be deoxygenated by sulfides to the epoxide **3a** (Scheme I), an attractive chemical pathway is available for in situ generation of this reactive intermediate. Indeed, a chemical model experiment showed that an aqueous solution of the biologically pertinent sulfide L-methionine effected the deoxygenation of dioxetane **2a** at 0 °C within 48 h into the epoxide **3a**. The latter suffered prompt in situ hydrolysis to the tautomeric diols **13a/13a′**, isolated as a 66:34 mixture in 90% yield; the ring tautomer **13a** consisted of two diastereomers in a ratio of 68:32. The L-methionine was oxidized to its sulfoxide and isolated in 96% yield (Scheme III).

With regard to the chemical reactivity, addition of nucleophiles such as methanol, thiophenol, and trimethylsilylimidazole to in situ generated epoxide **3a**, as described above, proceeded smoothly even at −78 °C. For methanol, the tautomeric adducts **14a/14a′** were isolated in a ratio of 89:11 in 90% yield, of which the ring tautomer **14a** consisted of two diastereomers in a 84:16 ratio. The thiophenol adducts **15a/15a′** were obtained in 71% yield as tau-

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(15) Coulthard, C. E.; Marshall, J.; Pyaman, F. L. *J. Chem. Soc.* **1930**, 280.

tomers in a ratio of 95:5 and a diastereomeric ratio of 79:21 for the ring tautomer **15a**. The silyl group of the imidazole adduct **16a** (90%) was lost during silica gel chromatography (Scheme III). Surprisingly, the N-substituted imidazole adduct was isolated exclusively as the cis diastereomeric ring tautomer **16a**. The cis configuration of the ring tautomer **16a** is stabilized by an intramolecular hydrogen bridge between the alcohol proton and the imidazole nitrogen. All adducts **13a**/**13a'**–**16a** are hitherto unknown compounds and were fully characterized (IR, ^1H and ^{13}C NMR, and MS); satisfactory elemental analyses were obtained.

Mutagenicity in the Ames Test. The transformation products **3a**–**6a** and **8a**,**9a** of the benzofuran dioxetane **2a** were assayed for mutagenicity by employing the *S. typhimurium* strain TA100 (reverted mainly by alkylating agents and UV^{260} radiation) in the concentration range of 5–100 μg per plate. The preincubation technique, as described by Ames[16] and without exogenous metabolic activation (S9), was employed. Only in the case of benzofuran epoxide **3a** did incubation of the *Salmonella* bacteria produce a linear, dose-dependent increase of revertants; the specific mutagenicity was calculated to be ca. 1800 revertants per μmol. For comparison, dioxetane **2a** possesses a specific mutagenicity of 117000 revertants per μmol in this test.[5a]

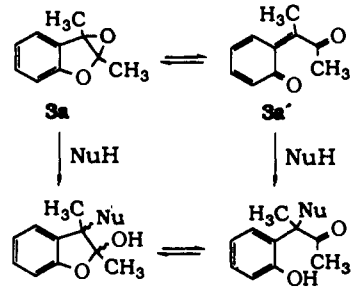
Discussion

The benzofuran dioxetanes **2** exhibited a variety of novel reactions for four-membered ring peroxides, which have until recently[5b] been unprecedented. Thus, besides the well-known thermal cleavage into the carbonyl products **4**,[5a] dioxetanes **2** afforded in polar, protic solvents such as water and methanol the 1,3-dioxol derivatives **5**, the spiroepoxide dimer **6a**, and the 1,4-dioxine derivatives **7** (Scheme II). Analogous to our recent work[5b] on the naphthofuran dioxetanes, we propose (Scheme IV) that the spiroepoxide **C**, observed here in the form of its dimer **6a** (Scheme II), is also the precursor to the rearrangement products **5** and **7**. Intramolecular electron transfer (ET)[17] leads to the "ion pair **A**", which on fragmentation affords the normal carbonyl product **4**. Alternatively, cyclization to the 1,4-dipole **B** and subsequent cleavage generates the spiroepoxide **C**. The latter dimerizes into **6a** or opens up in the polar medium (H_2O, CH_3OH) to produce the 1,5-dipole **D**, which serves as the precursor to the benzo-1,3-dioxols **5** and the benzo-1,4-dioxines **7** (methanol trapping).

The rearrangement of the benzofuran dioxetanes **2** to the hydroperoxides **8** was observed here for the first time (Scheme II). This rare reaction of 1,2-dioxetanes,[18] i.e., lateral carbon–oxygen bond cleavage, was previously established.[19] Since we have observed a highly diastereoselective addition of water and methanol to the benzofuran dioxetanes **2** in the presence of traces of acid (Scheme II), we favor a protonated intermediate **E** instead of a ring-opened cation as precursor both to the hydroperoxides **8** and the trans adducts **9a** and **10**; the latter are formed by S_N2 attack of the nucleophiles at the C-3 carbon of the protonated dioxetane **E**.

The formation of oxime **11a** in the CF_3CO_2H-catalyzed reaction of sodium azide with benzofuran dioxetane **2a** (Scheme II) is also explained in terms of nucleophilic attack by N_3^- on the protonated dioxetane intermediate **E**. The resulting azido hydroperoxide is, however, too labile and fragments into the acetylene oxime, which on hydrolysis in aqueous medium generates finally the observed oxime **11a**. As precedent for our mechanism we cite that in aqueous solutions β-hydroperoxy azides yield oximes by immediate evolution of dinitrogen with concomitant cleavage of the peroxide bond.[20]

Scheme V



A most relevant reaction of the benzofuran dioxetane **2a** was its deoxygenation with sulfides to give the benzofuran epoxide **3a** (Scheme I). This has led to first synthesis and spectral characterization of the extremely labile furan epoxides. Such deoxygenations are known reactions,[7] however, have never been before observed in a quantitative yield because catalytic decomposition of the 1,2-dioxetane prevails. More conveniently these labile epoxides were prepared by direct epoxidation of benzofurans with isolated dimethyldioxirane (as acetone solution) at −78 °C.[21]

The labile nature of epoxide **3a** is manifested in its high chemical reactivity with nucleophiles such as methanol, thiophenol, and imidazole (Scheme III). The corresponding adducts **14a**–**16a** were formed essentially quantitatively at −78 °C within less than 1 h and without acid catalysis. In contrast, and biologically significant, the corresponding dioxetane **2a** required activation by acids to achieve nucleophilic addition. At elevated temperatures the dioxetane **2a** afforded the carbonyl product **4a** rather than the peroxide **8a** by lateral carbon–oxygen bond cleavage.

In view of the electrophilic nature of the benzofuran epoxides, their potential as effective alkylants also in biological systems, e.g., DNA, was expected. Indeed, epoxide **3a** induced mutations (1800 rev/μmol) in *S. typhimurium* strain TA100, a strain mutating especially in response to alkylation of DNA. In this context, it is known[22] that furan epoxides, which arise from the oxidative metabolism of furans with mixed function oxidases, are responsible for cellular damage; however, until now their direct detection in cellular and even chemical systems was not possible. A prominent related example is the potent mutagen aflatoxin B_1, a 2,3-dihydrofuran derivative, whose ultimate DNA-binding metabolite is the corresponding epoxide.[23] Moreover, the regiochemistry of the nucleophile attack on epoxide **3a** (Scheme III) is consistent with that found for the intermediary epoxides of the cytotoxic methylfurans, important terpenoids,[22c] The ultimate mutagens in these cases were established to be the ene dicarbonyl products, which have been suggested to arise from the furan epoxides by rearrangement.

In view of the qualitative similarity and despite the appreciable quantitative difference in the mutagenic action of the benzofuran dioxetane **2a** (117000 rev/μmol)[2] and the benzofuran epoxide **3a** (1800 rev/μmol) in *S. typhimurium* strain TA100, we propose that the latter is the ultimate mutagen of the former.[24] The much lower activity of the epoxide **3a** must arise from its substantial hydrolysis so that only a small fraction reaches the DNA target, while deoxygenation of dioxetane **2a** into epoxide **3a** takes place presumably in the vicinity of the DNA target and thus is more readily available for alkylation.

Although the dioxetanes **2** exhibit an unusually high reactivity toward nucleophiles, it is unlikely that they are directly alkylating the DNA, because, as already stated above, the chemical model

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(21) Sauter, M. Diplomarbeit, University of Würzburg, March 1991.
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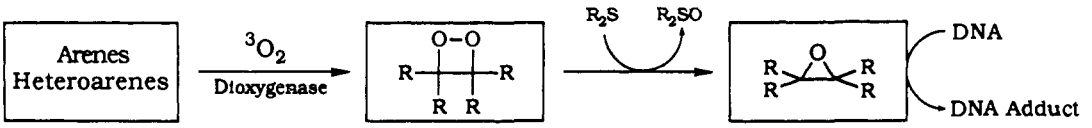
(23) Baertschi, S. W.; Raney, K. D.; Stone, M. P.; Harris, T. M. *J. Am. Chem. Soc.* **1988**, *110*, 7929.
(24) Chemical and toxicological studies on the molecular level are in progress to demonstrate the structures of DNA adducts formed by dioxetane **2a** and epoxide **3a**.

Scheme VI



reactions (Scheme II) require acid catalysis and are also relatively slow compared to those for the epoxide **3a**. Since dioxetane **2a** can be efficiently deoxygenated by sulfides to the corresponding epoxide **3a**, an attractive molecular pathway is available for in situ generation of such reactive heteroarene oxides. Moreover, the quantitative deoxygenation of dioxetane **2a** was also realized by the amino acid L-methionine, as confirmed by the isolation of the hydrolysis product **13a** and L-methionine sulfoxide (Scheme III). Consequently, such deoxygenations of dioxetanes into epoxides by biologically pertinent sulfides are feasible in cellular systems.

Analogous to the rearrangement of furan epoxides into ene dicarbonyl products,[9a,22c] such chemistry obtains also for the benzofuran epoxide **3a**. Trapping experiments with ethyl vinyl ether and tetracyanoethylene suggest that a tautomeric equilibrium between the epoxide **3a** and the quinomethide **3a'** operates (Scheme V).[21] Since **3a** and **3a'** afford on nucleophilic attack the same products, e.g., in the case of methanol the adducts **14a** and **14a'**, in principle both **3a** and its tautomer **3a'** may serve as alkylating agents of DNA.

The conclusion, that dioxetanes may serve as precursors to mutagenic epoxides in cellular systems, represents a challenging hypothesis, which has not been recognized so far as possible mode for the induction of mutations. The following experimental key facts speak in support for this novel mutagenesis pathway: (*i*) the labile benzofuran epoxide **3a**, isolated and characterized for the first time, is an excellent alkylant, (ii) benzofuran dioxetane **2a** was deoxygenated with sulfides (e.g., methionine) to this epoxide **3a**, and finally (iii) mutagenic activity of similar quality was demonstrated for dioxetane **2a** and epoxide **3a** in *S. typhimurium* strain TA100.

We propose that this unusual mutagenesis route is of general biological significance, in that polycyclic arenes such as benzo[*a*]pyrene or polycyclic heteroarenes such as furocoumarins may be transformed into their reactive epoxides by deoxygenation of the corresponding dioxetanes (Scheme VI). In fact, in the cytotoxic and mutagenic activity of benzo[*a*]pyrene[8a,b] and furocoumarins,[8c] the corresponding dioxetanes have been implicated previously as intermediates. However, the role of such dioxetanes as precursors to highly reactive epoxides has not been considered so far.

Experimental Section

Materials. Commercial reagents were purchased from standard chemical suppliers and purified to match the reported physical and spectral data. All solvents for the preparative work were distilled from EDTA. The known compounds **1a**[25] and **2a**,**b**[5a] were prepared according to literature procedures.

Apparatus. Boiling and melting points are uncorrected; the latter are taken on a Reichert Thermovar Kofler apparatus; infrared spectra, Perkin-Elmer 1420 infrared spectrometer; ^{1}H NMR spectra, Bruker AW 80 (80 MHz), Bruker AC 200 (200 MHz), Bruker AC 250 (250 MHz), or Bruker WM 400 (400 MHz), tetramethylsilane as internal standard; ^{13}C NMR spectra, Bruker AC 200 (50 MHz), or Bruker AC 250 (63 MHz), chloroform-*d*, acetone-d_6, or tetramethylsilane as internal standard; mass spectra, Variant MAT CH 7; combustion analyses for elemental composition, Carlo Erba 1106; thin-layer chromatography (TLC), Polygram SIL/G/UV (40 × 80 mm) from Machery & Nagel Co was employed; column chromatography, silica gel (60–230 mesh) from Woelm or silylated silica gel 60 from Merck (adsorbent/substrate ratio was 80–100:1).

Decomposition of Dioxetane 2a in Water. A sample of 98.2 mg (0.551 mmol) of dioxetane **2a** was dissolved in 70 mL of a 1:6 acetone/water solvent mixture. The solution was stirred for 24 h at 20 °C, and the aqueous layer was extracted with methylene chloride (3 × 50 mL). The combined organic layers were dried over $MgSO_4$. Removal of the solvent by roto-evaporation (ca. 20 °C, 17 Torr) and purification of the crude product by column chromatography (silica gel, CH_2Cl_2) yielded 25.2 mg (26%) of 2-acetyl-2-methyl-1,3-benzo[*b*]dioxole (**5a**, lit.[12]) as colorless oil [TLC (CH_2Cl_2) R_f 0.55], 48.1 mg (49%) of 2-acetylphenyl acetate (**4a**) as colorless prisms, mp 89–90 °C (CCl_4) (lit.[11] mp 89 °C) [TLC (CH_2Cl_2) R_f 0.17], and 10.1 mg (10%) of dimer **6a** as colorless powder, mp 231–232 °C (CH_2Cl_2) [TLC (CH_2Cl_2) R_f 0.04].

4a: ^{1}H NMR ($CDCl_3$, 250 MHz) δ 2.35 [s, 3 H, O(CO)CH_3], 2.56 [s, 3 H, (CO)CH_3], 7.12 (dd, J = 8.1 Hz, 1.1 Hz, 1 H, aryl H), 7.33 (m_c, 1 H, aryl H), 7.55 (m_c, 1 H, aryl H), 7.82 (dd, J = 7.8 Hz, 1.7 Hz, 1 H, aryl H).

5a: ^{1}H NMR ($CDCl_3$, 200 MHz) δ 1.73 (s, 3 H, CH_3), 2.27 [s, 3 H, (CO)CH_3], 6.85 (s, 4 H, aryl H).

6a: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 1.49 (s, 3 H, CH_3), 1.52 (s, 3 H, CH_3), 2.31 [s, 3 H, (CO)CH_3], 2.34 [s, 3 H, (CO)CH_3], 2.88 (dd, $J_{4a,8a}$ = 8.9 Hz, $J_{4a,4}$ = 1.9 Hz, 1 H, 4a-H), 2.96 (ddd, $J_{4,9}$ = 6.7 Hz, $J_{4,4a}$ = 1.9 Hz, $J_{4,10}$ = 1.7 Hz, 1 H, 4-H), 3.50 (ddd, $J_{1,10}$ = 6.3 Hz, $J_{1,8a}$ = 2.3 Hz, $J_{1,9}$ = 1.4 Hz, 1-H), 3.56 (dddd, $J_{8a,4a}$ = 8.9 Hz, $J_{8a,8}$ = 4.3 Hz, $J_{8a,1}$ = 2.3 Hz, $J_{8a,7}$ = 1.7 Hz, 1 H, 8a-H), 6.15 (ddd, $J_{10,9}$ = 8.0 Hz, $J_{10,1}$ = 6.3 H, $J_{10,4}$ = 1.7 Hz, 1 H, 10-H), 6.16 (dd, $J_{7,8}$ = 10.1 Hz, $J_{7,8a}$ = 1.7 Hz, 1 H, 7-H) 6.62 (dd, $J_{8,7}$ = 10.1 Hz, $J_{8,8a}$ = 4.3 Hz, 1 H, 8-H), 6.63 (ddd, $J_{9,10}$ = 8.0 Hz, $J_{9,4}$ = 6.7 Hz, $J_{9,1}$ = 1.4 Hz, 1 H, 9-H); ^{13}C NMR ($CDCl_3$, 63 MHz) δ 15.0 (q), 15.2 (q), 26.3 (q), 27.1 (q), 34,8 (d), 39.2 (d), 39.3 (d), 52.9 (d), 67.2 (s), 67.3 (s), 71.9 (s), 73.1 (s), 129.9 (d), 131.9 (d), 132.9 (d), 146.2 (d), 191.3 (s), 202.0 (s), 205.1 (s), 206.1 (s); IR ($CDCl_3$) 3000, 2980, 2930, 1740, 1710, 1690, 1600, 1440, 1410, 1350, 1280, 1250, 1230, 1200, 1150, 1130, 1100, 1080, 1000 cm^{-1}; MS (70 eV) m/z (%) = 356 (<1) [M^+], 313 (1) [M^+ – (CO)CH_3], 285 (1), 271 (1), 243 (1), 229 (2), 211 (1), 201 (1), 187 (2), 179 (1), 161 (2), 155 (2), 135 (82), 121 (21), 115 (4), 99 (4), 93 (3), 77 (4), 65 (5), 55 (4), 43 (100). Anal. Calcd for $C_{20}H_{20}O_6$: C, 67.40; H, 5.67. Found: C, 67.58; H, 5.70.

Decomposition of Dioxetane 2a in Methanol. A solution of 59.0 mg (0.331 mmol) of dioxetane **2a** in 10 mL of methanol was stirred for 7 days at ca. 20 °C. After removal of the solvent by roto-evaporation (ca. 20 °C, 17 Torr), the residue was dissolved in chloroform-*d* and ^{1}H NMR analysis showed that 100% conversion and a 93:3:4 product ratio for **4a**:**5a**:**7a** were obtained (spectra of **4a** and **5a** see above).

2,3-Dimethyl-3-methoxy-1,4-benzo[*b*]dioxen-2-ol (**7a**, lit.[14]) (as diastereomers in a ratio of 67:33); ^{1}H NMR ($CDCl_3$, 250 MHz, main isomer) δ 1.63 (s, 3 H, CH_3), 1.64 (s, 3 H, CH_3), 3.28 (s, 3 H, OCH_3), 3.29 (s, 1 H, OH), 6.88–6.93 (m, 4 H, aryl H); ^{1}H NMR ($CDCl_3$, 250 MHz, minor isomer) δ 1.47 (s, 3 H, CH_3), 1.57 (s, 3 H, CH_3), 3.30 (s, 1 H, OH), 3.31 (s, 3 H, OCH_3), 6.88–6.93 (m, 4 H, aryl H),

Decomposition of Dioxetane 2b in Methanol. A solution of 580 mg (2,79 mmol) of dioxetane **2b** in 40 mL of methanol was stirred for 5 days at ca. 20 °C, and the solvent was removed by roto-evaporation (20 °C, 17 Torr). Purification of the residue by column chromatography (silica gel, CH_2Cl_2) yielded 141 mg (24%) of 2-acetyl-4-methoxy-2-methyl-1,3-benzo[*b*]dioxole (**5b**) as pale yellow oil [TLC (CH_2Cl_2) R_f 0.60], 273 mg (47%) of 2-acetyl-3-methoxyphenyl acetate (**4b**) as colorless prisms, mp 47–48 °C (CH_2Cl_2, pentane) [TLC (CH_2Cl_2) R_f 0.18], and 130 mg (19%) of 3,5-dimethoxy-2,3-dimethyl-1,4-benzo[*b*]dioxen-2-ol (**7b**) as colorless needles, mp 161–165 °C (CH_2Cl_2, pentane) [TLC (CH_2Cl_2) R_f 0,14].

4b: ^{1}H NMR ($CDCl_3$, 250 MHz) δ 2.26 [s, 3 H, O(CO)CH_3], 2.43 [s, 3 H, (CO)CH_3], 3.73 (s, 3 H, OCH_3), 7.03 (dd, J = 8.2 H, 1.6 Hz, 1 H, aryl H), 7.15 (m_c, 1 H, aryl H), 7.26 (dd, J = 7.8 Hz, 1.6 H, 1 H, aryl H); ^{13}C NMR ($CDCl_3$, 63 MHz) δ 20.7 (q), 29.8 (q), 56.2 (q), 116,0 (d), 121,1 (d), 126.4 (d), 132.1 (s), 138.6 (s), 151.9 (s), 168.7 (s), 197.5 (s); IR (CCl_4) 3040, 2980, 2950, 2850, 1790, 1710, 1590, 1490, 1470, 1450, 1380, 1370, 1330, 1290, 1220, 1210, 1190, 1150, 1070, 1020, 980, 930 cm^{-1}; MS (70 eV) m/z (%) = 208 (5) [M^+], 166 (84), 151 (100), 133 (5), 119 (10), 108 (7), 93 (3), 77 (6), 65 (4), 51 (6), 43 (45). Anal. Calcd for $C_{11}H_{12}O_4$: C, 63.45; H, 5.81, Found; C, 63.52; H, 5,68,

5b: ^{1}H NMR ($CDCl_3$, 250 MHz) δ 1.72 (s, 3 H, CH_3), 2.27 [s, 3 H, (CO)CH_3], 3.88 (s, 3 H, OCH_3), 6.51 (dd, J = 7.9 Hz, 0.9 Hz, 1 H, aryl H), 6.52 (dd, J = 8.5 Hz, 0,9 Hz, 1 H, aryl H), 6,79 (m_c, 1 H, aryl H); ^{13}C NMR ($CDCl_3$, 63 MHz) δ 20,2 (q), 24.1 (q), 56.5 (q), 102,5 (d), 107.5 (d), 114.3 (s), 122.4 (d), 134.6 (s), 144.0 (s), 148.0 (s), 201,2 (s); IR (CCl_4) 3020, 2950, 2840, 1750, 1650, 1510, 1470, 1440, 1380, 1370,

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1310, 1280, 1260, 1220, 1200, 1180, 1130, 1070, 1060, 990, 920, 890, 880 cm^{-1}; MS (70 eV) m/z (%) = 208 (5) [M$^+$], 165 (100) [M$^+$ - (CO)CH$_3$], 150 (11), 123 (1), 108 (7), 95 (2), 79 (1), 65 (1), 51 (3), 43 (27). Anal. Calcd for $C_{11}H_{12}O_4$: C, 63.45; H, 5.81. Found: C, 63.56; H, 5.53.

7b (as diastereomers in a ratio of 77:23): ^{1}H NMR (CDCl$_3$, 250 MHz, main isomer) 1.64 (s, 3 H, CH$_3$), 1.70 (s, 3 H, CH$_3$), 3.19 (s, 1 H, OH), 3.27 (s, 3 H, OCH$_3$), 3.86 (s, 3 H, OCH$_3$), 6.54–6.61 (m, 2 H, aryl H), 6.83 (m$_c$, 1 H, aryl H); ^{13}C NMR (CDCl$_3$, 63 MHz, main isomer) δ 17.4 (q), 22.3 (q), 49.4 (q), 56.0 (q), 96.0 (s), 98.4 (s), 105.1 (d), 109.9 (d), 120.8 (d), 130.8 (s), 140.5 (s), 148.9 (s); ^{1}H NMR (CDCl$_3$, 250 MHz, minor isomer) δ 1.50 (s, 3 H, CH$_3$), 1.57 (s, 3 H, CH$_3$), 3.30 (s, 3 H, OCH$_3$), 3.79 (s, 1 H, OH), 3.87 (s, 3 H, OCH$_3$), 6.54–6.61 (m, 2 H, aryl H), 6.83 (m$_c$, 1 H, aryl H); ^{13}C NMR (CDCl$_3$, 63 MHz, minor isomer) δ 16.8 (q), 20,3 (q), 49.3 (q), 56.0 (q), 97.2 (s), 97.9 (s), 105.1 (d), 109.5 (d), 120.4 (d), 132.2 (s), 139.6 (s), 148.7 (s); IR (KBr) 3520–3480, 3130, 3080, 3020, 2980, 2960, 2840, 1620, 1510, 1490, 1470, 1440, 1410, 1390, 1380, 1340, 1300, 1260, 1240, 1220, 1200, 1180, 1120, 1110, 1090, 1080, 1050, 980, 950, 930, 910, 890, 830 cm^{-1}; MS (70 eV) m/z (%) = 240 (19) [M$^+$], 208 (15), 193 (12), 181 (4), 165 (91), 151 (36), 140 (100), 137 (6), 125 (13), 108 (9), 101 (73), 95 (7), 87 (19), 79 (6), 73 (18), 65 (6), 59 (6), 51 (10), 43 (82). Anal. Calcd for $C_{12}H_{16}O_5$: C, 59.98; H, 6.71. Found: C, 59.98; H, 6.77.

(2,3-Dihydro-3-methylidene-2-methylbenzo[*b*]furan-2-yl)hydroperoxide (8a). To a solution of 600 mg (3.37 mmol) of dioxetane **2a** in 6 mL of chloroform was added 106 mg (1.12 mmol) of trifluoroacetic acid at −10 °C. The solution was stirred at −10 °C for 3 h, and the solvent was removed by roto-evaporation (0 °C, 17 Torr). Column chromatography [silica gel, 1:5 Et$_2$O/petroleum ether (30–50), −10 °C] of the crude product afforded 310 mg (52%) of hydroperoxide **8a** as a colorless oil [TLC (1:5 Et$_2$O/petroleum ether (30–50)) R_f 0.29]: ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.70 (s, 3 H, CH$_3$), 5.32 (s, 1 H, =CH), 5.72 (s, 1 H, =CH), 6.93 (m, 2 H, aryl H), 7.25 (m, 1 H, aryl H), 7.42 (m, 1 H, aryl H), 8.55 (br s, 1 H, OOH); ^{13}C NMR (CDCl$_3$, 50 MHz) δ 22.2 (q), 106.2 (t), 110.2 (d), 114.4 (s), 121.0 (d), 121.3 (d), 124.4 (s), 130.8 (d), 144.8 (s), 159.5 (s); IR (CCl$_4$) 3520, 3410, 3000, 1620, 1590, 1460, 1340, 1330, 1260, 1150, 1090, 890 cm^{-1}; peroxide content 90% by iodometry.

(2,3-Dihydro-3-methylidene-4-methoxy-2-methylbenzo[*b*]furan-2-yl)-hydroperoxide (8b). To a solution of 312 mg (1.50 mmol) of dioxetane **2b** in 4 mL of chloroform was added 57.0 mg (0.500 mmol) of trifluoroacetic acid at 0 °C. The solution was stirred at 0 °C for 3 h, and the solvent was removed by roto-evaporation (0 °C, 17 Torr). Column chromatography [silica gel, 1;5 Et$_2$O/petroleum ether (30–50), −10 °C] of the crude product gave 180 mg (58%) of hydroperoxide **8b** as a colorless oil [TLC (1:5 Et$_2$O/petroleum ether (30–50)) R_f 0.09]: ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.70 (s, 3 H, CH$_3$), 3.87 (s, 3 H, OCH$_3$), 5.34 (s, 1 H, =CH), 5.73 (s, 1 H, =CH), 6.83 (dd, J = 8.0 Hz, 1.4 Hz, 1 H, aryl H), 6.92 (dd, J = 8.0 Hz, 7.4 Hz, 1 H, aryl H), 7.04 (dd, J = 7.4 Hz, 1.4 Hz, 1 H, aryl H), 8.76 (s, 1 H, OOH); ^{13}C NMR (CDCl$_3$, 50 MHz) δ 22,4 (q), 55.6 (q), 107,1 (t), 112.1 (d), 113.0 (d), 115.0 (s), 122.0 (d), 125.6 (s), 144.0 (s), 147.8 (s), 157.6 (s), IR (CCl$_4$) 3540, 3450, 2950, 1630, 1600, 1515, 1460, 1335, 1290, 1240, 1100, 1070, 900 cm^{-1}. Anal. Calcd for $C_{11}H_{12}O_4$: C, 63.45; H, 5.81. Found: C, 63.21; H, 5.63.

(2*R,3*S**)-(±)-2,3-Dihydro-2,3-dimethyl-2-hydroperoxybenzo[*b*]-furan-3-ol (9a).** A sample of 250 mg (1.40 mmol) of dioxetane **2a** was dissolved in 20 mL of a 1:1 acetone/water solvent mixture and treated with three drops of trifluoroacetic acid. The reaction mixture was stirred for 20 h at 5 °C and extracted with methylenechloride (2 × 20 mL). The combined organic layers were washed with saturated aqueous NaHCO$_3$ (2 × 10 mL) and dried over MgSO$_4$. After removal of the solvent by roto-evaporation (0 °C, 17 Torr), purification of the crude product by column chromatography (silylated silica gel, CH$_2$Cl$_2$) yielded 215 mg (78%) of hydroperoxide **9a** as colorless powder, mp 97–99 °C dec (CH$_2$Cl$_2$) [TLC (CH$_2$Cl$_2$) R_f 0.00]: ^{1}H NMR (CD$_3$CN, 200 MHz) δ 1.51 (s, 3 H, CH$_3$), 1.65 (s, 3 H, CH$_3$), 3.53 (br s, 1 H, OH), 6.89 (dd, J = 8.0 Hz, 0.7 Hz, 1 H, aryl H), 6.99 (m$_c$, 1 H, aryl H), 7.25–7.35 (m, 2 H, aryl H), 9.63 (br s, 1 H, OOH); ^{13}C NMR (CD$_3$CN, 50 MHz) δ 15.7 (q), 79.7 (s), 111.4 (d), 118.9 (s), 122.3 (d), 124.2 (d), 130.9 (d), 134.0 (s), 159.0 (s); IR (KBr) 3530–3490, 3440–3250, 3030, 3000, 2940, 1610, 1480, 1470, 1450, 1390, 1350, 1260, 1130, 1100, 1080, 1060, 940, 900, 850 cm^{-1}; MS (70 eV) m/z (%) = 196 (2) [M$^+$], 163 (4), 136 (41), 121 (79), 107 (3), 91 (10), 77 (6), 65 (14), 51 (4), 43 (100). Anal. Calcd for $C_{10}H_{12}O_4$: C, 61.21; H, 6.17. Found; C, 61.48; H, 6.03.

((2*R,3*S**)-(±)-2,3-Dihydro-2,3-dimethyl-3-methoxybenzo[*b*]furan-2-yl)hydroperoxide (10a).** A solution of 354 mg (1.98 mmol) of dioxetane **2a** and 63.0 mg (0.660 mmol) of trifluoroacetic acid in 5 mL of methanol was stirred at 0 °C for 6 h. The excess methanol was removed by roto-evaporation (5 °C, 17 Torr) and column chromatography [silica gel, 1:5 Et$_2$O/petroleum ether (30–50 °C), −5 °C] of the crude product afforded 255 mg (61%) of hydroperoxide **10a** as a colorless oil, which solidified at −20 °C [TLC (1:5 Et$_2$O/petroleum ether (30–50)) R_f 0.29]: ^{1}H NMR (CDCl$_3$), 200 MHz) δ 1.52 (s, 3 H, CH$_3$), 1.74 (s, 3 H, CH$_3$), 3.00 (s, 3 H, OCH$_3$), 6.90 (d, J = 7.6 Hz, 1 H, aryl H), 7.00 (m, 1 H, aryl H), 7.30 (m, 2 H, aryl H), 8.26 (s, 1 H, OOH); ^{13}C NMR (CDCl$_3$, 50 MHz) δ 14.9 (q), 16.4 (q), 51.0 (q), 83.7 (s), 110.7 (d), 116.1 (s), 121.3 (d), 124.6 (d), 128.1 (s), 130.5 (d), 158,4 (s); IR (CCl$_4$) 3530, 3400, 2950, 1600, 1475, 1380, 1250, 1090, 900 cm^{-1}, Anal. Calcd for $C_{11}H_{14}O_4$: C, 62.85; H, 6.71. Found: C, 62.78; H, 6.91.

((2*R,3*S**)-(±)-2,3-Dihydro-2,3-dimethyl-3,4-dimethoxybenzo[*b*]-furan-2-yl)hydroperoxide (10b).** A solution of 407 mg (1.95 mmol) of dioxetane **2b** and 62.0 mg (0.650 mmol) of trifluoroacetic acid in 12 mL of a 1:1 methanol/chloroform mixture was stirred at 0 °C for 20 h. The solvent was removed by roto-evaporation (5 °C, 17 Torr) and column chromatography [silica gel, 1:5 Et$_2$O/petroleum ether (30–50), −5 °C] of the crude product afforded 257 mg (55%) of hydroperoxide **10b** as a colorless oil [TLC (1:5 Et$_2$O/petroleum ether (30–50)) R_f 0.13]: ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.52 (s, 3 H, CH$_3$), 1.76 (s, 3 H, CH$_3$), 2.96 (s, 3 H, OCH$_3$), 3.67 (s, 3 H, OCH$_3$), 6.75–7.00 (m, 3 H, aryl H), 9,16 (s, 1 H, OOH); ^{13}C NMR (CDCl$_3$, 50 MHz) δ 14.8 (q), 16.7 (q), 51.2 (q), 55.5 (q), 84.1 (s), 112.9 (d), 116.5 (d), 118.6 (s), 121.9 (d), 129.3 (s), 144.8 (s), 146.8 (s); IR (CCl$_4$) 3530, 3450, 2960, 1600, 1500, 1460, 1300, 1270, 1205, 1100, 900 cm^{-1}. Anal. Calcd for $C_{12}H_{16}O_5$: C, 59.99; H, 6.71. Found: C, 60.56; H, 6.33.

2-Hydroxyacetophenone Oxime (11a). To a solution of 390 mg (6.00 mmol) of sodium azide in 6 mL of a 3;2 methanol/water solvent mixture was added 178 mg (1.00 mmol) of dioxetane **2a**, followed by 66.0 mg (0.700 mmol) of trifluoroacetic acid at 5 °C. The solution was stirred at 5 °C for 11 h, and the solvent was removed by distillation (5 °C, 0.1 Torr), Column chromatography [silica gel, 1:4 Et$_2$O/petroleum ether (30–50), −10 °C] and crystallization from chloroform yielded 92.0 mg (61%) of oxime **11a** as colorless needles, mp 113–115 °C (lit.[15] 112 °C): ^{1}H NMR (CDCl$_3$, 250 MHz) δ 2.30 (s, 3 H, CH$_3$), 6.88 (m, 2 H, aryl H), 7.21 (m, 1 H, aryl H), 7.37 (dd, J = 7.9 Hz, 1.6 Hz, 1 H, aryl H), 7.62 (br s, 1 H, OH), 11.36 (br s, H, N–OH); ^{13}C NMR (CDCl$_3$, 63 MHz) δ 10.7 (q), 117.2 (d), 118.6 (s), 119.3 (d), 127.6 (d), 130.7 (d), 157.3 (s), 159.4 (s),

2,3-Dihydro-2,3-dimethyl-2,3-epoxybenzo[*b*]furan (3a). To a stirred solution of 109 mg (0.750 mmol) of 2,3-dimethylbenzo[*b*]furan (**1a**) in 10 mL of acetone was added rapidly at −78 °C 15 mL of a solution of dimethyldioxirane in acetone (0.079 M, 1.19 mmol), which had been dried over molecular sieves 4 Å. Stirring was continued for 1.5 h at −78 °C and 1 h at −30 °C, and removal of the solvent by distillation (−15 °C, 0.03 Torr) afforded quantitatively epoxide **3a** as a colorless solid, which decomposed on warmup to room temperature: ^{1}H NMR (CD$_3$COCD$_3$, 200 MHz, −20 °C) δ 1.85 (s, 3 H, CH$_3$), 1,97 (s, 3 H, CH$_3$), 6.90–7.50 (m, 4 H, aryl H); ^{13}C NMR (CD$_3$COCD$_3$, 50 MHz) δ 12.2 (q), 14.2 (q), 67.3 (s), 94.8 (s), 111.1 (d), 120.9 (d), 123.8 (d), 129.2 (s), 129.8 (d), 158.9 (s).

2,3-Dihydro-3-methenyl-2-methylbenzo[*b*]furan-2-ol (12a) and 3-(2′-Hydroxyphenyl)-3-buten-2-one (12a′). A sample of 107 mg (0.601 mmol) of dioxetane **2a** in 50 mL of acetone was treated at −78 °C with 37.3 mg (0.601 mmol) of dimethyl sulfide in 10 mL of acetone, The reaction mixture was stirred for 2 h at −78 °C and stored for 24 h at 0 °C. After removal of the solvent by roto-evaporation (ca. 20 °C, 17 Torr), the residue was purified by column chromatography (silica gel, CH$_2$Cl$_2$, 0 °C) to yield 51.3 mg (53%) of the 74:26 tautomeric mixture of **12a**/**12a′** (lit.[10]) as a yellowish oil [TLC (CH$_2$Cl$_2$) R_f 0.15].

12a: ^{1}H NMR (CDCl$_3$, 250 MH) δ 1.70 (s, 3 H, CH$_3$), 3.37 (s, 1 H, OH), 5.31 (s, 1 H, =CH), 5.59 (s, 1 H, =CH), 6.85 (d, J = 8,1 Hz, 1 H, aryl H), 6.94 (m$_c$, 1 H, aryl H), 7.25 (m$_c$, 1 H, aryl H), 7.40 (dd, J = 7.6 Hz, 0.9 Hz, 1 H, aryl H).

12a′: ^{1}H NMR (CDCl$_3$, 250 MHz) δ 2.48 (s, 3 H, CH$_3$), 5.97 (s, 1 H, =CH), 6,28 (s, 1 H, =CH), 6,82–6,95 (m, 1 H, aryl H), 7.10 (dd, J = 8.0 Hz, 1.8 Hz, 1 H, aryl H), 7.25 (m$_c$, 1 H, aryl H), 7.58 (s, 1 H, OH).

2,3-Dihydro-2,3-dimethylbenzo[*b*]furan-2,3-diol (13a) and 3-Hydroxy-3-(2′-hydroxyphenyl)butan-2-one (13a′). A sample of 100 mg (0.561 mmol) of dioxetane **2a** was dissolved in 5 mL of acetonitrile and treated at −20 °C with 83.7 mg (0.561 mmol) of L-methionine in 20 mL of water. The reaction mixture was stored for 2 days at 0 °C and extracted with a 7:1 methylene chloride/acetone solvent mixture (3 × 30 mL). The combined organic layers were dried over MgSO$_4$, and the solvent was removed by roto-evaporation (ca. 20 °C, 17 Torr). Column chromatography (silica gel, CH$_2$Cl$_2$) of the crude product yielded 91.0 mg (90%) of the 66;34 tautomeric mixture of **13a**/**13a′** as a colorless powder, mp 101–105 °C (CH$_2$Cl$_2$, pentane) [TLC (CH$_2$Cl$_2$) R_f 0.05].

13a (as diastereomers in a ratio of 68:32): ^{1}H NMR (CD$_3$CN, 250 MH, main isomer) δ 1.44 (s, 3 H, CH$_3$), 1.47 (s, 3 H, CH$_3$), 3.80 (s, 1 H, OH), 5.15 (s, 1 H, OH), 6.70–7.35 (m, 4 H, aryl H); ^{1}H NMR (CD$_3$CN, 250 MHz, minor isomer) δ 1.50 (s, 3 H, CH$_3$), 1.57 (s, 3 H,

CH_3), 3.30 (s, 1 H, OH), 4.52 (s, 1 H, OH), 6.70–7.35 (m, 4 H, aryl H).

13a': ^{1}H NMR (CD_3CN, 250 MHz) δ 1.63 (s, 3 H, 4-H), 2.05 (s, 3 H, 1-H), 4.69 (br s, 1 H, OH), 6.70–7.35 (m, 4 H, aryl H), 8.09 (br s, 1 H, OH).

13a and **13a'**: ^{13}C NMR (CD_3CN, 63 MHz) δ 20.7 (q), 21.0 (q), 22.1 (q), 22.7 (q), 24.7 (q), 24.8 (q), 78.6 (s), 80.3 (s), 81.6 (s), 111.1 (d), 111.3 (d), 112.0 (s), 113.4 (s), 117.6 (d), 121.1 (d), 121.8 (d), 121.9 (d), 124.9 (d), 125.1 (d), 128.2 (d), 128.5 (s), 130.7 (d), 130.9 (d), 131.1 (d), 133.5 (s), 134.5 (s), 156.3 (s), 157.9 (s), 159.0 (s), 206.3 (s); IR (KBr) 3480–3440, 3400, 3000, 2960, 2920, 1590, 1470, 1460, 1410, 1380, 1350, 1320, 1280, 1230, 1220, 1180, 1120, 1100, 1050, 1010, 950, 930, 920, 860, 850, 830, 800 cm^{-1}; MS (70 eV) m/z (%) = 180 (7) [M$^+$], 163 (1) [M$^+$ – OH], 147 (4), 137 (87), 119 (27), 105 (18), 91 (28), 77 (9), 65 (11), 51 (5), 43 (100). Anal. Calcd for $C_{10}H_{12}O_3$: C, 66.65; H, 6.71. Found: C, 66.65; H, 7.02.

Workup of the aqueous layer by removal of the solvent at 60 °C/17 Torr yielded 89.1 mg (96%) of L-methionine sulfoxide as colorless powder, mp 231–233 °C (lit.[26] 238 °C).

Reaction of Epoxide 3a with Nucleophiles. Samples (1.1–3.2 mmol) of dioxetane **2a** in 30–60 mL of acetone were treated at −78 °C with equimolar amounts of dimethyl- or diphenyl sulfide in 20 mL of acetone. The reaction mixture was stirred for 1 h at this temperature and a 2- to 3-fold excess of the nucleophile (methanol, thiophenol, 1-(trimethylsilyl)imidazole) in 20 mL of acetone was added. The solution was allowed to warm up to ca. 20 °C within 3 h, and the solvent was removed by roto-evaporation (ca. 20 °C, 17 Torr). The residue was purified by column chromatography (silica gel, CH_2Cl_2). The details of the individual cases are summarized below.

2,3-Dihydro-2,3-dimethyl-3-methoxybenzo[*b*]furan-2-ol (14a) and 3-(2'-Hydroxyphenyl)-3-methoxybutan-2-one (14a'). From 223 mg (1.25 mmol) of dioxetane **2a**, 233 mg (1.25 mmol) of diphenyl sulfide, and 120 mg (3.75 mmol) of methanol was obtained 219 mg (90%) of the 89:11 tautomeric mixture of **14a**/**14a'** as colorless oil [TLC (CH_2Cl_2) R_f 0.25].

14a (as diastereomers in a ratio of 84:16): ^{1}H NMR ($CDCl_3$, 250 MHz, main isomer) δ 1.48 (s, 3 H, CH_3), 1.51 (s, 3 H, CH_3), 3.15 (s, 3 H, OCH_3), 5.22 (s, 1 H, OH), 6.82 (dd, J = 8.5 Hz, 1.0 Hz, 1 H, aryl H), 6.91 (m$_c$, 1 H, aryl H), 7.22–7.30 (m, 2 H, aryl H); ^{1}H NMR ($CDCl_3$, 250 MHz, minor isomer) δ 1.57 (s, 3 H, CH_3), 1.68 (s, 3 H, CH_3), 3.01 (s, 3 H, OCH_3), 3.39 (br s, 1 H, OH), 6.80–7.30 (m, 4 H, aryl H).

14a': ^{1}H NMR ($CDCl_3$, 250 MHz) δ 1.73 (s, 3 H, CH_3), 2.13 [s, 3 H, (CO)CH_3], 3.34 (s, 3 H, OCH_3), 6.80–7.30 (m, 4 H, aryl H), 8.28 (s, 1 H, OH).

14a and **14a'**: ^{13}C NMR ($CDCl_3$, 63 MHz) δ 15.7 (q), 16.1 (q), 19.1 (q), 20.2 (q), 21.9 (q), 25.0 (q), 50.8 (q), 50.9 (q), 51.7 (q), 80.9 (s), 84.2 (s), 88.1 (s), 110.8 (d), 111.1 (d), 112.0 (s), 112.7 (s), 117.8 (d), 119.9 (d), 120.3 (d), 120.8 (d), 123.1 (s), 125.2 (d), 125.4 (d), 127.1 (s), 127.3 (d), 128.2 (s), 130.1 (d), 130.5 (d), 130.9 (d), 155.9 (s), 157.6 (s), 158.4 (s), 206.8 (s); IR (CCl_4) 3520–3480, 3360, 3060, 3020, 2980, 2920, 2840, 2820, 1710, 1600, 1590, 1540, 1470, 1450, 1400, 1370, 1350, 1320, 1240, 1230, 1190, 1150, 1090, 1060, 1010, 970, 950, 900, 860 cm^{-1}; MS (70 eV) m/z (%) = 194 (7) [M$^+$], 163 (3) [M$^+$ – OCH_3], 151 (100), 135 (11), 119 (46), 104 (4), 91 (29), 77 (4), 65 (7), 51 (3), 43 (25), 39 (4). Anal. Calcd for $C_{11}H_{14}O_3$: C, 68.02; H, 7.27. Found: C, 68.00; H, 7.38.

2,3-Dihydro-2,3-dimethyl-3-(phenylthio)benzo[*b*]furan-2-ol (15a) and 3-(2'-Hydroxyphenyl)-3-(phenylthio)benzo[*b*]furan-2-one (15a'). From 561 mg (3.15 mmol) of dioxetane **2a**, 196 mg (3.16 mmol) of dimethyl sulfide, and 694 mg (6.30 mmol) of thiophenol was obtained 611 mg (71%) of the 95:5 tautomeric mixture of **15a**/**15a'** as colorless powder, mp 69–76 °C (CH_2Cl_2, pentane) [TLC (CH_2Cl_2) R_f 0.25].

15a (as diastereomers in a ratio of 79:21): ^{1}H NMR ($CDCl_3$, 250 MHz, main isomer) δ 1.46 (s, 3 H, CH_3), 1.64 (s, 3 H, CH_3), 5.50 (s, 1 H, OH), 6.45–7.35 (m, 9 H, aryl H); ^{1}H NMR ($DCDl_3$, 250 MHz, minor isomer) δ 1.71 (s, 3 H, CH_3), 1.93 (s, 3 H, CH_3), 3.36 (s, 1 H, OH), 6.45–7.35 (m, 9 H, aryl H).

15a': ^{1}H NMR ($CDCl_3$, 250 MHz) δ 1.67 (s, 3 H, CH_3), 2.09 [s, 3 H, (CO)CH_3], 6.45–7.35 (m, 10 H, OH, aryl H).

15a and **15a'**: ^{13}C NMR ($CDCl_3$, 63 MHz) δ 19.6 (q), 22.0 (q), 22.3 (q), 23.4 (q), 25.1 (q), 29.7 (q), 62.6 (s), 65.2 (s), 66.2 (s), 110.2 (d), 110.6 (d), 111.4 (s), 112.0 (s), 117.4 (d), 120.7 (d), 120.8 (d), 124.1 (d), 127.0 (s), 127.3 (d), 128.1 (d), 128.3 (d), 128.5 (d), 128.9 (d), 129.0 (d), 129.1 (s), 129.4 (d), 129.5 (d), 129.6 (d), 129.8 (s), 130.1 (s), 131.1 (s), 131.7 (s), 136.9 (d), 137.2 (d), 137.4 (d), 153.9 (s), 155.7 (s), 156.4 (s), 205.4 (s); IR (CCl_4) 3400–3360, 3080, 3060, 3000, 2940, 2860, 1710, 1600, 1480, 1460, 1440, 1390, 1380, 1350, 1330, 1280, 1240, 1180, 1140, 1070, 960, 870, 860 cm^{-1}; MS (70 eV) m/z (%) = 272 (2) [M$^+$], 218 (51), 185 (11), 163 (100) [M$^+$ – SPh], 154 (15), 147 (10), 145 (31), 137 (16), 135 (14), 121 (16), 119 (26), 109 (38), 107 (14), 91 (28), 77 (9), 65 (19), 43 (26). Anal. Calcd for $C_{16}H_{16}O_2S$: C, 70.56; H, 5.91. Found: C, 70.33; H, 5.80.

1-(2',3'-Dihydro-2',3'-dimethyl-2'-hydroxybenzo[*b*]furan-3'-yl)-imidazole (16a). From 190 mg (1.07 mmol) of dioxetane **2a**, 66.5 mg (1.07 mmol) of dimethyl sulfide, and 402 mg (2.86 mmol) of 1-(trimethylsilyl)imidazole was obtained 221 mg (90%) of **16a** as colorless powder, mp 169–170 °C (CH_2Cl_2, pentane) [TLC (CH_2Cl_2) R_f 0.04]: ^{1}H NMR (CD_3SOCD_3, 250 MHz) δ 1.01 (s, 3 H, CH_3), 1.90 (s, 3 H, CH_3), 6.70 (s, 1 H, imidazole H), 6.86 (s, 1 H, imidazole H), 6.90–7.10 (m, 2 H, aryl H), 7.20–7.40 (m, 4 H, imidazole H, aryl H, OH); ^{13}C NMR (CD_3SOCD_3, 63 MHz) δ 18.7 (q), 20.4 (q), 68.2 (s), 110.3 (d), 112.0 (s), 118.2 (d), 121.3 (d), 124.5 (d), 128.1 (d), 130.2 (s), 130.7 (d), 135.5 (d), 157.6 (s); IR (KBr) 3200–2400, 3180, 3140, 3000, 1600, 1520, 1500, 1480, 1460, 1390, 1370, 1350, 1330, 1290, 1260, 1240, 1230, 1190, 1130, 1120, 1110, 1080, 1070, 1050, 1020, 1000, 970, 960, 930, 870, 840, 820, 800 cm^{-1}; MS (70 eV) m/z (%) = 232 (2) [M$^+$ + 2], 231 (16) [M$^+$ + 1], 230 (77) [M$^+$], 229 (3), 187 (26), 163 (56), 147 (43), 135 (18), 120 (14), 119 (16), 115 (10), 107 (21), 91 (37), 77 (11), 69 (100), 65 (13), 43 (34). Anal. Calcd for $C_{13}H_{14}N_2O_2$: C, 67.81; H, 6.13; N, 12.17. Found: C, 68.03; H, 6.17; N, 12.42.

Mutagenic Assays (Reversion of *his*$^-$ *Salmonella*). *S. typhimurium* mutagen tester strain TA100 was cultured in Oxoid no. 2 medium at 37 °c for 10 h. The handling of the strain, the experimental cultures, and the Vogel–Bonner minimal medium used were as described by Maron and Ames.[16] The bacteria were treated with the test compounds (or the solvent as control) by using the preincubation technique. For this purpose a mixture of 500 μL of 0.1 M sodium phosphate buffer (pH = 7.4), 100 μL of bacterial culture (equivalent to 2–4 × 10^8 bacterial cells), and 20 μL of a solution of the test compounds in acetone (or 20 μL of acetone as control) was shaken at 37 °C. After 30 min the incubates were mixed with top agar, which contained histidine and biotine, and plated onto minimal agar plates. After incubating the plates for 2 days, the revertant colonies were counted, and the raw counts were corrected as described previously.[27] All plates were set up in duplicate; all experiments included five doses of mutagen and were reproduced independently at least once. The specific mutagenic activities were obtained from the slope of the linear section of the dose versus response curves, their standard deviations are ca. 10%. The spontaneous revertant frequencies per plate were 150–200 in the strain TA100.

Acknowledgments. We thank the Deutsche Forschungsgemeinschaft (Sonderforschungsbereich No. 172 "Molekulare Mechanismen kanzerogener Primärveränderungen") and the Wilhelm-Sander-Stiftung for generous financial support.

Registry No. 1a, 3782-00-1; **2a**, 135831-47-9; **2b**, 135831-48-0; **3a**, 135831-49-1; **4a**, 7250-94-4; **4b**, 135831-50-4; **5a**, 91057-65-7; **5b**, 135831-51-5; **6a**, 135831-52-6; **7a** (isomer 1), 135831-53-7; **7a** (isomer 2), 135831-54-8; **7b** (isomer 1), 135831-55-9; **7b** (isomer 2), 135831-56-0; **8a**, 135831-57-1; **8b**, 135831-58-2; **9a**, 135831-59-3; **10a**, 135831-60-6; **10b**, 135831-61-7; **11a**, 1196-29-8; **12a**, 135831-62-8; **12a'**, 135831-63-9; **13a** (isomer 1), 135831-64-0; **13a** (isomer 2), 135831-65-1; **13a'**, 135831-66-2; **14a** (isomer 1), 135831-67-3; **14a** (isomer 2), 135831-68-4; **14a'**, 135831-69-5; **15a** (isomer 1), 135831-70-8; **15a** (isomer 2), 135831-71-9; **15a'**, 135831-72-0; **16a**, 135831-73-1; thiophenol, 108-98-5; 1-(trimethylsilyl)imidazole, 18156-74-6.

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