We wish to thank Dr. D. A. Long for sending us a sample of his material to aid in the establishment of the biological assay. We are indebted to Mr. R. N. Boos and his associates for the microanalyses and to Mr. R. W. Walker for the determination and interpretation of the infrared absorption spectra.

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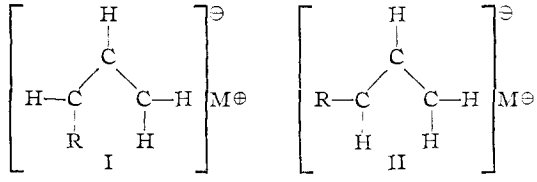
STRUCTURE OF ALLYLIC ANIONS

Sir:

The vibrational spectra of a large number of allylic organoalkali compounds are consistent with the proposition that these ions have a planar structure (C_{2v} symmetry) with angles of approximately 120° about the carbon atoms of the allylic system. That is, the ions have sp^2 hybridization as might be expected from a simple molecular orbital treatment if the double bond is considered to be completely delocalized.[1] On the basis of the observed spectra, the allylic ions of potassium, sodium and lithium have the same structure which may be represented approximately by I and II. Considerable evidence has been presented indicating Grignard reagents to be essentially covalent[2,3] (dicyclopentadienylmagnesium however, is reported to be ionic[4]) but the spectra of allylic magnesium compounds differ little from those of allylic organoalkali compounds.



Alkenylsodium and potassium compounds were prepared by metalation of olefins with amylsodium and amylpotassium.[5] Alkenyllithiums were prepared by interchange of sodium compounds with lithium chloride and by addition of butyllithium to butadiene. Magnesium compounds were prepared from reaction of allylic halides with magnesium in ether and by interchange with sodium compounds and subsequent extraction. Grignard reagents prepared in ether were heated at reduced pressure to free them of ether.[6] Solvent-free Nujol mulls were prepared under dry-box conditions and spectra were determined on a Perkin–Elmer infrared spectrophotometer. Except for the regions obscured by Nujol, excellent spectra were obtained with most of the allylic compounds studied.

Allylic ions should have a considerable energy barrier to rotation about the allylic carbon–carbon bonds, hence the existence of two distinct isomers, I and II, is possible. None of the compounds examined had bands in the normal double bond stretching region, but all the compounds investigated did show very strong bands in the 1500–1560 cm.$^{-1}$ range. Those ions where R = H should have only one isomer but where R is some alkyl substituent configurations I and II are possible. Symmetrical compounds such as allyl-, isobutenyl- or cyclohexenylsodium show only one very strong band at 1535, 1520 and 1522 cm.$^{-1}$, respectively. Compounds such as pentenylsodium have two very strong bands, one about 1525 cm.$^{-1}$ and the other about 1560 cm.$^{-1}$. No evidence has been found for structures of the type $(R—CH{=}CH{=}CHCH_3)^{\ominus}M^{\oplus}$ (III). Carbonation of allylic organoalkali compounds yields acids derivable from structures I and II.[5] Indeed, attempts to prepare a system like III by metalation of heptene-3 gave a spectrum identical with that from heptene-2. Although heptene-1 was not available, the spectra of heptenylsodium prepared from heptene-2 and -3 showed only minor differences when compared with compounds prepared from pentene-1 and -2, hexene-1 and -2 or octene-1 and -2. However, since olefins are easily isomerized by organoalkali compounds it is rather difficult to determine which isomeric olefin is actually metalated.[7]

From a fairly complete analysis of the vibrational spectra of allyl- and perdeuteroallylsodium, the band at 1535 cm.$^{-1}$ in allylsodium has been assigned to the carbon–carbon unsymmetrical stretching frequency.[8] Analogy with substituted olefins suggests that the band around 1525 cm.$^{-1}$ in the substituted allylic ions belongs to structure I and the band around 1560 cm.$^{-1}$ belongs to structure II.[9] This assignment is supported by the position of the band in cyclohexenylsodium at 1522 cm.$^{-1}$ since this ion can exist only in a form geometrically equivalent to structure I.

Acknowledgment.—This work was performed as a part of the research project sponsored by the National Science Foundation, Office of Synthetic Rubber.

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TRANSFER OF MOLECULAR OXYGEN BY PEROXIDASE

Sir:

The metabolic function of peroxidase is unknown. We wish to report observations, based upon the use of O^{18} as a tracer, which support the view that ferroperoxidase activates molecular oxygen.

The system consisting of horseradish peroxidase, dihydroxyfumarate and oxygen, catalyzes non-

specific hydroxylation of aromatic compounds.[1] When salicylic acid is hydroxylated by this system, 2,3-dihydroxybenzoic and gentisic acids are formed in equal amounts, together with smaller quantities of trihydroxybenzoic acids. We have carried out this reaction in an atmosphere of O^{18}_2, in H_2O solution free of heavy metal contaminants according to the method of Keilin and Hartree[2] and have separated the products on silica gel columns. Oxygen which was incorporated into salicylic acid as hydroxyl to form 2,3-dihydroxybenzoic and gentisic acids was found to arise entirely from the atmosphere. When the reaction was carried out in H_2O^{18} with an atmosphere of O_2, no excess of labelled oxygen was detected (Table I).

TABLE I

SOURCE OF OXYGEN INCORPORATED AS HYDROXYL INTO SALICYLIC ACID BY THE SYSTEM: HORSERADISH PEROXIDASE,[a] DIHYDROXYFUMARATE,[b] OXYGEN[c]

Experiment	Conditions[d]	Compd. isolated[d]	% Theor. O^{18} incorporation[e]
1, 2	$O^{18}_2 + H_2O^{16}$	2,3-Dihydroxybenzoic acid	106
		Gentisic acid	110
3	$O^{16}_2 + H_2O^{18}$	2,3-Dihydroxybenzoic acid	0
		Gentisic acid	0

[a] We gratefully acknowledge a gift of purified horseradish peroxidase (RZ = 1.9) from Professor D. Keilin. [b] Recrystallized from mixtures of acetone and deionized distilled water; $E_{mol.}$ = 8350, 308 mμ, in ether. [c] Prepared by electrolysis of H_2O containing 1.4 atom % O^{18} (Stewart Oxygen Company, San Francisco). [d] Hydroxylations were carried out in 20 ml. solutions containing phosphate (0.046 *M*), salicylic acid (300 μmoles), peroxidase (1.2 μmoles) and dihydroxyfumarate (0.81 mmole), brought to *p*H 6.0 with heavy-metal-free KOH. The mixture was acidified after two hours of reaction, and the products extracted with ethyl acetate. These were separated on silica gel columns, using chloroform and 4% ethanolic chloroform developers. Identities of the recovered gentisic acid (m.p. 202–203°) and 2,3-dihydroxybenzoic acid (m.p. 205–206°) were verified both spectroscopically and chromatographically. Under the conditions described, the combined yield (based upon a quantitative chromatographic procedure) was 16–21%. With heat-denatured peroxidase, a yield of 1.9% was observed. [e] We are grateful to Dr. C. C. Delwiche for performing the mass spectrometry upon samples which we obtained by Unterzaucher pyrolysis[3] of the hydroxylation products. An apparent isotope effect shown by our results is under study.

Since complex III of peroxidase predominates[4] in the dihydroxyfumarate–oxygen system[4] and the enzyme is inhibited by carbon monoxide, displaying the spectrum of carbon monoxide ferroperoxidase,[4,5] since dihydroxyfumarate (for which peroxidase is an oxidase) cannot be replaced by ascorbate in the hydroxylating system,[1] and since in this system molecular oxygen is activated toward non-specific hydroxylation of aromatic substances, oxyferroperoxidase appears to be involved.[6]

A complete description of these experiments will be published elsewhere.

(6) This study has been supported by a grant from the United States Public Health Service (A-971).

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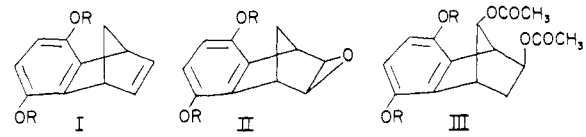
RECEIVED AUGUST 12, 1957

Ar_1-3 PARTICIPATION IN THE BICYCLO[2,2,1]HEPTANE SERIES

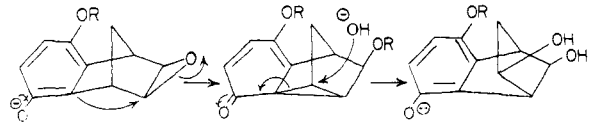
Sir:

The rearrangement of the bicyclo[2,2,1]heptane carbon skeleton, proceeding *via* carbonium ions, is a well known phenomenon.[1] We wish to report the observation of a novel, base-catalyzed analog of this type of transformation.



The key compound in this study is the epoxide IIA (m.p. 107.5–108.0°; calcd. for $C_{15}H_{14}O_5$: C, 65.69; H, 5.15. Found: C, 65.97; H, 5.16), prepared by treatment of IA[2] with peracetic or perfluoroperacetic acid. IIA reacted rapidly with hot 2 *N* sodium hydroxide to give an unstable phenolic product, the infrared spectrum of which lacked the strong 11.7 μ (epoxide) band of IIA. Acetylation of this material with pyridine–acetic anhydride gave a product from which the tetraacetate IIIA (m.p. 139–140°; calcd. for $C_{19}H_{20}O_8$; C, 60.63; H, 5.36; acetyl, 46.7. Found: C, 60.47; H, 5.27; acetyl, 45.64) could be isolated in 20% yield. An authentic sample of IIIA was readily obtained by *acid* hydrolysis of IIA, which on the basis of the behavior of norbornene epoxide[3] would be expected to give the rearranged product, followed by acetylation.

An attractive interpretation of this reaction involves nucleophilic opening of the epoxide ring by an internal phenoxide ion as shown below. Further attack by base on the dienone intermediate would regenerate the aromatic ring and give rise to a rearranged product.



A close analogy for this series of reactions is provided by the recent realization by Winstein and Baird of Ar_1-3 phenoxide participation in the

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