dilution effect constant). A new band with a maximum at 3300 cm.$^{-1}$ appeared in the infrared spectra of these solutions. Individual solutions of THQ or (THQD) in CCl_4 and of pyridine in CCl_4 did not exhibit absorption at this wave length. Significantly, the 3300-cm.$^{-1}$ band was independent of the normal N–H stretching mode of THQ (or THQD) at 3420 cm.$^{-1}$. In this region of the infrared spectrum, the 3300-cm.$^{-1}$ band was assigned to an intermolecular H-bonded complex between THQ (or THQD) and pyridine.

The tetrahydroquinoline–pyridine interaction was investigated by adding increments of pyridine to fixed amounts of THQ. As additional H-bonded complex formation occurred, the absorbance at 3300 cm.$^{-1}$ increased to a constant value, as shown in Figure 4. From a pyridine–THQ mole ratio of 12:1, the absorbance value remained constant up to a pyridine–THQ mole ratio of 79:1. Measured absorbances were corrected where necessary for dilution with carbon tetrachloride.

For the THIQ–acetic acid system, the tendency for salt formation was demonstrated with synthetic blends of THIQ and acetic acid in carbon tetrachloride. An insoluble salt was precipitated and the filtrate was analyzed by infrared spectroscopy for uncombined THIQ and acetic acid. The stoichiometry of THIQ and acetic acid for salt formation was found to be in 1:1 molar proportions. This stoichiometry was confirmed with different blends in which one of the two reactants was present in excess, thus assuring complete removal of the other component from solution. Thus, when an excess of acid was present, as in the oxidation experiments, all of the THIQ was presumably converted to the acetate salt which would not react with oxygen.

The isolated THIQ–acetic acid salt was analyzed. Its infrared spectrum was identified as that of an acetate salt. New COO^- bands at 1640 and 1590 cm.$^{-1}$ replaced the acid carbonyl absorption at 1720 cm.$^{-1}$. (This feature is indicative of the ionic character of the salt.)

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The Production of 3-Benzylidene-6-isobutylidene-2,5-dioxopiperazine, 3,6-Dibenzylidene-2,5-dioxopiperazine, 3-Benzyl-6-benzylidene-2,5-dioxopiperazine, and 3,6-Dibenzyl-2,5-dioxopiperazine by a Variant of *Streptomyces noursei*[1]

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3-Benzylidene-6-isobutylidene-2,5-dioxopiperazine (I), 3,6-dibenzylidene-2,5-dioxopiperazine (II), 3-benzyl-6-benzylidene-2,5-dioxopiperazine (III), and 3,6-dibenzyl-2,5-dioxopiperazine (IV) were isolated from broth cultures of a streptomycete. II and IV are both known substances, and the isolated materials were identified by comparison with authentic samples. The hydrogenation sequence II → III → IV effectively establishes the structure of III, a new compound. Evidence is presented that I has the indicated structure.

Streptomyces noursei variant No. 5286 which yields the antibiotic phalamycin[3] produces also four dioxopiperazines to which the following structures are assigned: 3-benzylidene-6-isobutylidene-2,5-dioxopiperazine, 3,6-dibenzylidene-2,5-dioxopiperazine, 3-benzyl-6-benzylidene-2,5-dioxopiperazine, and 3,6-dibenzyl-2,5-dioxopiperazine. The organism was grown for 4 days with shaking in a yeast extract broth.[4,5] The whole culture at pH 7.5–8.0 was extracted with ethyl acetate. Colored matter and other impurities in the dry extractives were removed with small volumes of chloroform, 70% ethanol, and acetone in succession. Chloroform was then percolated through the remaining solids thereby taking out I and II. These products, after removal of the solvent by distillation *in vacuo*, were separated from each other by extraction of the first into glacial acetic acid from which it was precipitated by dilution with water. To separate III and IV that remained after the chloroform treatment, many fractional recrystallizations from 2-propanol, chloroform, and acetone in succession were employed. For the final recrystallizations acetone or methanol were the solvents of choice. The progress in separation was followed by analysis of the infrared spectra and in the later stages by melting point determinations. This paper presents the evidence for the identification of the four compounds.

Results

3-Benzylidene-6-isobutylidene-2,5-dioxopiperazine (I).—The structure of I is supported by the following data. Upon hydrogenation with 10% palladium–carbon as catalyst, I added 2 molar equiv. to form 3-benzyl-6-isobutyl-2,5-dioxopiperazine (V). This hydrogenated I had the infrared spectrum and melting point of authentic V (leucylphenylalanine anhydride). Acid hydrolysis of 1 mole of the hydrogenated natural product yielded 1 mole each of phenylalanine and leucine. Furthermore, in the degradation products of the acid hydrolysis of I itself, phenylpyruvic acid and ammonia (2 moles/mole) were observed; and, on basic hydrolysis, benzaldehyde, isobutyraldehyde, and ammonia were released. The ultraviolet spectrum with maxima at 234 and 318 mμ is consistent with the conjugated carbonyl system. Infrared data give evidence for N–H, monosubstituted benzene, ethylenic and lactam C=O groupings, and, as in the other compounds of this study, absence of OH. I had the same infrared spectrum as "albonoursin"[6] which is reported to be 3-benzylidene-6-isobutylidene-2,5-dioxopiperazine. The mixture of the two showed no depression of the melting point.

(1) Presented in part as a dissertation by C. Kelley in partial fulfillment of the Degree of Doctor of Philosophy, Rensselaer Polytechnic Institute, 1961.

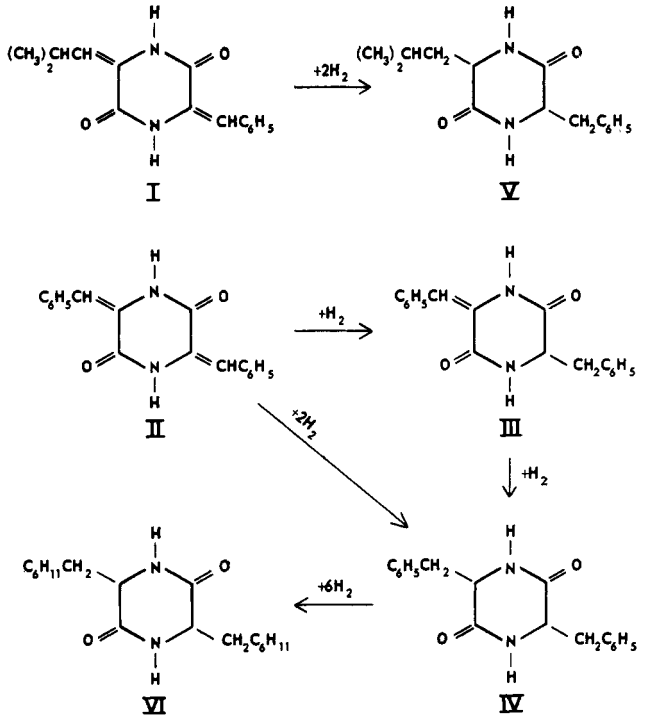
(2) Rensselaer Polytechnic Institute, Troy, N. Y.

(3) R. Brown and E. L. Hazen, *Antibiot. Chemotherapy*, **3**, 818 (1953).

(4) The cultures were kindly supplied by Mrs. Joan Brennan and Mr. Edward Lapa.

(5) No dioxopiperazine derivatives were isolated from the yeast extract used for the broth.

(6) A. S. Khokhlov and G. B. Lokshin, *Tetrahedron Letters*, **No. 27**, 1881 (1963); and personal communication. We are indebted to Dr. Khokhlov for a sample of "albonoursin."



3,6-Dibenzylidene-2,5-dioxopiperazine (II), 3-Benzyl-6-benzylidene-2,5-dioxopiperazine (III), and 3,6-Dibenzyl-2,5-dioxopiperazine (IV).—The structures II, III, and IV were derived from the following data. The natural product II has m.p. 298–300° and an infrared spectrum, both of which are considered identical with those of synthetic 3,6-dibenzylidene-2,5-dioxopiperazine. Addition of 1 mole of hydrogen to each mole of II yielded a compound having the ultraviolet and infrared spectra of III; addition of 2 moles of hydrogen to each mole of II gave IV; and addition of 1 mole to III also gave IV which was identified by comparison of its infrared spectrum and melting point with those of an authentic sample of *cis*-L-(−)-3,6-dibenzyl-2,5-dioxopiperazine.[7] Although both natural products III and IV displayed strong levorotation, III and IV prepared by hydrogenation of II displayed dextrorotation and were apparently mixtures of optical isomers. Upon acid hydrolysis, each mole of IV yielded 2 moles of phenylalanine. Moreover, when synthetic 3,6-dibenzylidene-2,5-dioxopiperazine, II, and III were hydrogenated under conditions which limited addition to olefinic bonds, and were then hydrolyzed, the degradation products were the same as that just described for IV. The ultraviolet absorption maxima of II and III at 338 and 296 mμ, respectively, are ascribable to conjugated carbonyl. Lack of conjugation in IV is reflected by absence of such absorption. The infrared spectra of the three compounds give evidence for N–H, monosubstituted benzene, and lactam C=O. Absorption due to olefinic stretching vibrations is present in II and III but absent in IV.

Discussion

Since the spectral properties of this type of compound have received little notice in the literature, they merit attention. The ultraviolet absorption of III at 296 mμ is only slightly higher than that observed for compounds having an α,β-unsaturated carbonyl in conjugation with a benzene ring,[8] but the large shift to 338 in II and to 318 mμ in I would be expected with considerable lengthening of the conjugated chain. In both I and II, two pairs of double-bonded groups are connected through nitrogen and, hence, these groups can conjugate each other through overlap of their π-electron orbitals with the p-orbitals of the nitrogen, and to the extent to which this occurs both cyclic and cross conjugation result[9]; to a lesser degree this phenomenon is applicable to III and IV. This interaction influences the infrared absorption also, and in general the disubstitution represented by I–IV leads to shifts in the absorption bands of unsubstituted dioxopiperazine.[10]

When examined as KBr disks these compounds show infrared absorption maxima characteristic of lactams: N–H stretching vibrations near 3180 and 3080; C=O absorption at 1680–1660; and absence of absorption in the 1550-cm.$^{-1}$ ("amide II") region. Furthermore a dilute chloroform solution of I (the only one so measured) showed a diminution of the 3180- and the appearance of a strong 3390-cm.$^{-1}$ band. The classical doublet at 1380 due to bending vibrations of the isopropyl group is not clearly apparent, possibly being obscured by the strong absorption bands at 1412 and 1358 cm.$^{-1}$. Weak absorption due to skeletal vibrations at 1176 and 1164 cm.$^{-1}$, however, as well as evidence from the n.m.r. spectrum[11] and the occurrence of isobutyraldehyde as a degradation product support the presence of an isopropyl group. In the spectra of tetrahydro I and decahydro I, a pair of weak bands at 1387 and 1365 cm.$^{-1}$ fits the isopropyl assignment. Comparison of the spectra of I, II, and III with those of their more highly hydrogenated forms points up certain correlations. Despite conjugation, the carbonyl frequency in both I and II is slightly higher than in the hydrogenated products.[12] Hydrogenation of the olefinic bonds results in loss or considerable diminution of the following strong bands: 1638, 1424, 1412, and 1358 in I; 1620, 1393, and 1355 in II; and 1626, 1437, 1402, and 1349 cm.$^{-1}$ in III. The 1638-, 1620-, and 1626-cm.$^{-1}$ bands in each, respectively, are attributed to the C=C stretching vibration influenced in intensity and location by conjugation and resonance. The strong aromatic band at 690 cm.$^{-1}$ in I and II is shifted 10 cm.$^{-1}$ higher when either one or two of the ethylenic bonds in II and two of the ethylenic bonds in I are hydrogenated. Bands in the 1500–1495-, 770–755-, and 700–690-cm.$^{-1}$ regions disappear upon hydrogenation of the monosubstituted benzene groups.

Although amino acid anhydrides have been isolated occasionally as metabolic products of streptomycetes

(7) J. H. Birkinshaw and Y. S. Mohammed, *Biochem. J.*, **85**, 523 (1962). We are indebted to Dr. Birkinshaw for this sample.

(8) A. L. Wilds, L. W. Beck, W. J. Close, C. Djerassi, J. A. Johnson, Jr., T. L. Johnson, and C. H. Shunk, *J. Am. Chem. Soc.*, **69**, 1985 (1947).

(9) For discussion of a related situation with oxo derivatives of 1,3,5-triazine, see M. Cignitti and L. Paoloni, *Spectrochim. Acta*, **20**, 211 (1964).

(10) K. Fukushima, Y. Ideguchi, and T. Miyazawa, *Bull. Chem. Soc. Japan*, **37**, 53 (1964).

(11) The n.m.r. spectrum, made on a deuterated chloroform solution of I with tetramethylsilane as internal reference and at 60 Mc. on a Varian A-60 spectrometer, showed that an isopropyl group must be present and this very probably as part of an isobutylidene grouping. Moreover, it gave evidence for two NH groups, probably both adjacent to carbonyl groups; an olefinic proton, probably β to a carbonyl and with no proton on the adjacent carbon; and a monosubstituted benzene ring. For this spectrum and its illuminating analysis the authors are indebted to Dr. James N. Shoolery of Varian Associates, Palo Alto, Calif.

(12) This uncommon occurrence has also been described by O. E. Edwards and T. Singh, *Can. J. Chem.*, **32**, 683 (1954).

and fungi, the simultaneous production of these four dioxopiperazines by a streptomycete has not been recorded; it appears that 3-benzyl-6-benzylidene-2,5-dioxopiperazine is a previously unreported natural product. As for the others, Khokhlov[6] isolated both I ("albonoursin") and II from cultures of *Streptomyces noursei* and *Streptomyces albus var. fungatus* and Birkinshaw and Mohammed[7] obtained *cis*-L-(−)-3,6-dibenzyl-2,5-dioxopiperazine from *Penicillium nigricans* (Bainier) Thom.

The presence of three of these amino acid anhydrides in dehydro forms is suggestive of significant dehydrogenase activity or of an interference with the normal biosynthesis of amino acids within the culture. Three stages are represented in the series presumed to be related to phenylalanine metabolism (II, III, and IV); the mixed anhydride, however, is found in only the didehydro form I. If other forms are present, they must be in such minute quantities as to escape detection in investigations on a laboratory scale. The amino acid metabolism of *Streptomyces noursei* variant No. 5286 is noteworthy, for in addition to these four dioxopiperazines another product, the antibiotic phalamycin, is composed of several amino acids, the major ones being leucine and phenylalanine.[13]

Experimental[14]

3-Benzylidene-6-isobutylidene-2,5-dioxopiperazine (I).[14a]—When recrystallized repeatedly from acetone, I gave soft, colorless needle-like crystals: m.p. 272°; λ_{max} 234, 318 mμ (log ε 3.9, 4.4); ν_{max} 3184, 3080–3030, 2935, 2857, 1680, 1638, 1496, 1455, 1424, 1412, 1358, 1176, 1164, 930, 851, 800–780, 748, 731, and 690 cm.$^{-1}$; no optical rotation in pyridine.

Anal. Calcd. for $C_{15}H_{16}N_2O_2$: C, 70.31; H, 6.25; N, 10.94; mol. wt., 256. Found: C, 70.14; H, 6.18; N, 10.81; mol. wt., 263; neut. equiv., 254.

Tetrahydro I (leucylphenylalanine anhydride) purified by vacuum sublimation had m.p. 260–262°; ν_{max} 3178, 3044, 2930, 1670, 1495, 1463, 1455, 1387, 1365, 1345, 1333, 1321, 1115, 1095, 1014, 911, 869, 838, 769, 753, and 700 cm.$^{-1}$. Acid hydrolysis yielded 1 molar equiv. each of leucine and phenylalanine.

Anal. Calcd. for $C_{15}H_{20}N_2O_2$: C, 69.18; H, 7.75; N, 10.77. Found: C, 69.18; H, 7.74; N, 10.59.

An authentic sample of leucylphenylalanine anhydride, prepared by cyclization of leucylphenylalanine[15] and purified by vacuum sublimation, melted at 262° and had the same infrared absorption spectrum.

Anal. Calcd. for $C_{15}H_{20}N_2O_2$: C, 69.18; H, 7.75; N, 10.77. Found: C, 69.45; H, 7.96; N, 10.65.

"Albonoursin" as well as an unidentified crystalline compound B-73[16] isolated from cultures of *Streptomyces albulus* and reported to have the formula $C_{15}H_{16}N_2O_2$, when compared with I, had identical infrared spectra, and when each was mixed with I there was no depression of the melting point. A mixture of "albonoursin" and B-73 also failed to show depression of the melting point.

3,6-Dibenzylidene-2,5-dioxopiperazine (II).—When repeatedly recrystallized from acetone, II gave soft greenish yellow platelets: m.p. 298–300°; λ_{max} 234, 338 mμ (log ε 3.9, 4.5); ν_{max} 3169, 3080–3020, 1674, 1620, 1495, 1456, 1428, 1393, 1355, 941, 921, 857, 814, 767, 747, 725, and 690 cm.$^{-1}$; no optical rotation in pyridine. This material had the same infrared spectrum and melting point as a sample prepared by the condensation of glycine anhydride with benzaldehyde.[17]

Anal. Calcd. for $C_{18}H_{14}N_2O_2$: C, 74.47; H, 4.86; N, 9.65; mol. wt., 290. Found: C, 74.82; H, 4.76; N, 9.51; neut. equiv., 281.

Dihydro II had m.p. 283–284° dec.; λ_{max} 296 mμ (log ε 4.1); infrared absorption spectrum of III; $[\alpha]^{24}D$ +42° (*c* 0.088, pyridine). Acid hydrolysis yielded 1 molar equiv. of phenylalanine.

Anal. Calcd. for $C_{18}H_{16}N_2O_2$: C, 73.95; H, 5.52; N, 9.58. Found: C, 73.50; H, 5.35; N, 9.54.

Tetrahydro II had m.p. 306–307° dec.; $[\alpha]^{26}D$ +72° (*c* 0.046, pyridine).

3-Benzyl-6-benzylidene-2,5-dioxopiperazine (III).—When recrystallized repeatedly from acetone III gave colorless fibrous crystals which had a strong tendency to become electrostatically charged: m.p. 288.5–290°; λ_{max} 296 mμ (log ε 4.1); ν_{max} 3189, 3059, 2908, 1661, 1626, 1500, 1455, 1437, 1402, 1349, 1323, 1312, 1206, 1194, 1093, 924, 910, 872, 859, 821, 766, 740, 700, and 638 cm.$^{-1}$; $[\alpha]^{24}D$ −490° (*c* 0.081, pyridine); $[\alpha]^{27}D$ −520° (*c* 0.028, glacial acetic acid). Acid hydrolysis yielded 1 molar equiv. of phenylalanine.

Anal. Calcd. for $C_{18}H_{16}N_2O_2$: C, 73.95; H, 5.52; N, 9.58; mol. wt., 292. Found: C, 73.71; H, 5.44; N, 9.34; neut. equiv., 284.

Dihydro III had m.p. 303–304°; $[\alpha]^{25}D$ −223° (*c* 0.051, pyridine).

3,6-Dibenzyl-2,5-dioxopiperazine (IV).—When recrystallized repeatedly from acetone and finally from methanol IV gave colorless fibrous crystals: m.p. 311–312°; ultraviolet absorption (end absorption only); ν_{max} 3187, 3080–3040, 2934, 2902, 2848, 1669, 1665, 1497, 1461, 1348, 1337, 1325, 1266, 1211, 1195, 1091, 1014, 921, 896, 870, 850, 801, 756, 699, and 660 cm.$^{-1}$; $[\alpha]^{25}D$ −242°; $[\alpha]^{25}_{5461}$ −270° (*c* 0.059, pyridine); $[\alpha]^{27}D$ −150° (*c* 0.025, glacial acetic acid). Acid hydrolysis yielded 2 molar equiv. of phenylalanine.

Anal. Calcd. for $C_{18}H_{18}N_2O_2$: C, 73.45; H, 6.16; N, 9.52. Found: C, 73.49; H, 5.81; N, 9.49.

Both *cis* and *trans* forms are known; the latter lacks optical activity and melts at 289–291°. Vejdelek[18] reports for the (+)-*cis*-isomeride $[\alpha]^{25}D$ +107° (2.20 mg. in 0.906 ml. of ethanol), m.p. 315–316°. Birkinshaw and Mohammed[7] report for the L-(−)-*cis*-isomeride $[\alpha]^{20}_{5461}$ −267 ± 3° (*c* 0.132, pyridine) and m.p. 326°. A sample provided by their laboratory melted at 311–312° and, when mixed with our product, m.p. 311–313°. The infrared spectrum of their sample is identical with that of IV and of the two products formed when II and III add, respectively, two and one molar proportions of hydrogen.

3,6-Dihexahydrobenzyl-2,5-dioxopiperazine (VI).—When IV was hydrogenated with platinum oxide as catalyst 6 molar equiv. were added to give VI. When recrystallized from ethanol VI decomposed at 320° without melting; ν_{max} 3183, 3080, 3037, 2899, 2838, 1682–1675, 1456, 1447, 1413, 1342, 1329, 1318, 1269, 1143, 1119, 1103, 964, 956, 924, 916, 852, 821, 786, 763, 752, and 665 cm.$^{-1}$; $[\alpha]^{24}D$ −37° (*c* 0.030, pyridine).

Anal. Calcd. for $C_{18}H_{30}N_2O_2$: C, 70.59; H, 9.80; N, 9.15. Found: C, 70.89; H, 9.99; N, 8.96.

Nonaqueous Titrations.—I, II, and III but not IV are weak monobasic acids as determined by potentiometric titration of their pyridine solutions with tetrabutylammonium hydroxide dissolved in benzene–methanol. None of these compounds showed

(13) Unpublished data.

(14) Analyses for C, H, and N and molecular weight determinations were done at the Carl Tiedcke Laboratory of Microchemistry, Teaneck, N. J. Molecular weights by the Rast method were satisfactory only in the case of I; II, III, and IV were not sufficiently soluble in camphor. The infrared spectra, generally, were measured using potassium bromide disks, 0.5% in 200 mg., on a Perkin-Elmer Model 21 spectrophotometer equipped with a sodium chloride prism and calibrated with polystyrene and air. A chloroform solution of I, 3.0 mg./ml. in 1-mm. matched cells, was also examined. The very low solubility of the compounds limits study of their solutions. Ultraviolet absorption was measured on a Beckman Model DU spectrophotometer with 95% ethanol solutions, and optical rotation on a Rudolph Model 80 photoelectric polarimeter. In the paper chromatography Whatman No. 1 paper was used. Melting points are uncorrected; those higher than 300° were determined on a Kofler micro hot stage, and those lower, generally, on a Fisher-Johns hot stage. These measurements were troublesome owing to the soft nature of the crystals, their strong tendency to sublime, the high temperatures required, and their proximity to decomposition points.

(14a) NOTE ADDED IN PROOF.—Since this paper was submitted for publication M. Vondráček and Z. Vaněk [*Chem. Ind.* (London), 1686 (1964)] have published evidence for the production of 3-benzylidene-6-isobutylidene-2,5-dioxopiperazine and 3,6-dibenzylidene-2,5-dioxopiperazine by *Streptomyces noursei*.

(15) E. Abderhalden and E. Rossner, *Z. Physiol. Chem.*, **163**, 149 (1927).

(16) K. V. Rao and W. P. Cullen, *J. Am. Chem. Soc.*, **82**, 1127 (1960). We are indebted to Dr. Rao for a sample of B-73.

(17) T. Sasaki, *Chem. Ber.*, **54B**, 163 (1921).

(18) Z. J. Vejdelek, *Collection Czech. Chem. Commun.*, **15**, 929 (1951). From the discussion in the paper, it appears that glacial acetic acid was the solvent used for optical rotation, not ethanol as recorded.

any basic groups when measured by titration of their acetic acid solutions with perchloric acid dissolved in glacial acetic acid.

Solubility.—All four compounds are very insoluble: in alcohol, acetone, ether, and benzene the solubility is less than 0.1 mg./ml.; chloroform dissolves I and II to a slightly greater extent; pyridine dissolves all to about 1–2%. Neither 6 *N* hydrochloric acid nor 10% sodium hydroxide dissolves any of them appreciably.

Hydrogenation.—Most experiments were performed with 15–30-mg. portions dissolved in 30 ml. of redistilled glacial acetic acid in an apparatus patterned after that of Ogg and Cooper.[19] As catalysts 150-mg. portions of 10% palladium–carbon or platinum oxide were used, the former for hydrogenation of olefinic bonds only, the latter for hydrogenation of aromatic groups as well. These catalysts were hydrogenated in 10 ml. of glacial acetic acid immediately prior to addition of the solution to be measured and account was taken of the hydrogen consumption by the solvent blank. In the preparation of III from II, only 20 mg. of 10% palladium–carbon was used with 60 mg. of II dissolved in 60 ml. glacial acetic acid; the reaction was stopped when 1 molar equiv. of hydrogen was taken up. For analysis, the product was purified by two extractions with chloroform and by five recrystallizations from acetone.

Hydrolyses. A. Hydrochloric Acid.[20]—The compounds were dissolved in a slight excess of glacial acetic acid and then one-fourth that volume of 25% hydrochloric acid (w./w.) was added; after flushing with nitrogen, the tubes were sealed *in vacuo* and heated at 95° for 48 hr. Generally the hydrolysates were evaporated to dryness, washed with ether (negligible extract except with I and III), and subjected to analysis for ammonia and amino acids on a Spackman–Stein–Moore column.[21] In those cases where a single degradation product was expected, the infrared absorption was also measured. In the total hydrolysate of I a 2 molar proportion of ammonia was measured on the column, and the identity was confirmed by the infrared spectrum which was that of ammonium chloride. Paper chromatography with the total hydrolysate and with its ether extract showed a product moving identically with phenylpyruvic acid: R_f 0.33, *n*-butyl alcohol saturated with 1.5 *N* ammonium hydroxide; R_f 0.39, *t*-butyl alcohol–ammonia (*d* 0.88)–water (20:1:4); R_f 0.88, butanol–acetic acid–water (12:3:5). Both gave uncommon reactions with these spotting reagents: ferric chloride, a juniper green; 2,6-dichloroindophenol, white on colored background. Also comparison of the infrared spectra of solids from the ether extract and of phenylpyruvic acid confirmed this identification.

B. Sodium Hydroxide.—A suspension of 400 mg. of I in 10 ml. of 1 *N* sodium hydroxide was heated at 100° for 4 hr. and the volatile products were swept through 2,4-dinitrophenylhydrazine traps which were changed frequently. From the early cuts, the 2,4-dinitrophenylhydrazone of isobutyraldehyde was separated; these yellow crystals melted at 185° with decomposition; an authentic sample melted at 182°, reported m.p. 182° and 187°; the behavior of both hydrazones on paper chromatography was identical: R_f 0.84, 5% ethyl ether in ligroin (b.p. 100–106°), and R_f 0.93, 30% tetrahydrofuran in ligroin. The 2,4-dinitrophenylhydrazone which formed in larger amounts throughout most of the run gave orange crystals, m.p. 243–245° (recrystallized from ethanol); the corresponding derivative of benzaldehyde had m.p. 243° and m.m.p. 242–243°; on paper chromatography both moved at similar rates: R_f 0.51, 5% ether in ligroin, and 0.88, 30% tetrahydrofuran in ligroin. Furthermore, after the mixture of these dinitrophenylhydrazones of the degradation products of I was resolved on a silicic acid–Celite column,[22] infrared absorption and melting point data proved the derivatives to be those of isobutyraldehyde and of benzaldehyde. In similar but smaller scale hydrolyses of I ammonia was identified by a positive Nessler reaction and by recognition of the infrared spectrum of ammonium chloride formed by passing the volatile degradation products through ether saturated with hydrogen chloride.

(19) C. L. Ogg and F. J. Cooper, *Anal. Chem.*, **21**, 1400 (1949).

(20) C. Kelley and R. Brown, *N. Y. State Dept. of Health Ann. Rept. Div. Lab. Res.*, 75 (1961).

(21) The operation of the Spackman–Stein–Moore column was by Mr. Edward F. Duchna and the data were analyzed by Mrs. Marijane A. McEwan.

(22) B. E. Gordon, F. Wopat, Jr., H. D. Burnham, and L. C. Jones, Jr., *Anal. Chem.*, **23**, 1754 (1951).

Sodium- and Potassium-Catalyzed Reactions of Toluene, Ethylbenzene, and Isopropylbenzene with Isoprene[1,2]

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In the sodium- and potassium-catalyzed alkenylation of alkylarenes with isoprene, the mode of addition seems to be determined by the relative stabilities of the resultant carbanions and also the "size effect" of alkali metal ions in the transition state. The ratios of monoadducts resulting from "head addition" and "tail addition" are 2.77, 1.88, and 1.98 for toluene, ethylbenzene, and isopropylbenzene, respectively, in the presence of sodium; and 3.04, 2.54, and 3.30 for toluene, ethylbenzene, and isopropylbenzene, respectively, in the case of potassium. The relative rates of alkenylation obtained from competitive reactions are as follows: in sodium-catalyzed reactions, 1.00 for toluene, 1.00 for ethylbenzene, and 0.058 for isopropylbenzene; and in potassium-catalyzed reactions, 1.00 for toluene, 1.12 for ethylbenzene, and 0.449 for isopropylbenzene.

The base-catalyzed side-chain alkylation of alkylbenzenes has been the subject of extensive study in our laboratory.[3] This study was recently extended to include the aralkylation of alkylbenzenes with α-methyl- and β-alkylstyrenes.[4,5] The purpose of the present paper is to investigate the addition of alkylbenzenes to the unsymmetrical diolefin isoprene in the presence of sodium and potassium as catalysts in order to gain a better understanding of the mechanism of the side-chain alkylation reaction.

Hoffman and Michael reported a 30% yield of 2-methyl-5-phenyl-2-pentene and 3-methyl-5-phenyl-2-pentene from the reaction of isoprene and toluene at 160° in the presence of sodium.[6] Robertson and Marion observed the presence of side-chain alkenylated products having the general formula $C_6H_5CH_2[CH_2—C(CH)_3{=}CH—CH_2]_nH$ (n = 1 to 5) in the sodium-initiated polymerization of isoprene at 90° in toluene solvent.[7] None of the authors gave detailed structures of the products obtained.

In the present study toluene, ethylbenzene, and isopropylbenzene were employed as aromatics, and the structures of the products were determined by means of chemical and physical methods.

(1) Paper XXVIII of the series "Base-Catalyzed Reactions." For paper XXVII, see J. Shabtai and H. Pines, *J. Org. Chem.*, **29**, 2408 (1964).

(2) This work was supported by the NSF Grant G14503.

(3) For the general review of the literature, see H. Pines and L. Schapp, *Advan. Catalysis*, **12**, 117 (1960).

(4) J. Shabtai and H. Pines, *J. Org. Chem.*, **26**, 4225 (1961).

(5) J. Shabtai, E. M. Lewicki, and H. Pines, *ibid.*, **27**, 2618 (1962).

(6) F. Hoffman and A. Michael, U. S. Patent 2,448,641 (1928).

(7) R. E. Robertston and L. Marion, *Can. J. Res.*, **26B**, 657 (1948).