the height h of the repeating unit in a helical polymer of AzCOOH would be smaller by an amount approaching 1 Å than that of a corresponding polymer of proline. Azetidinecarboxylic acid, having a comparatively rigid four-membered ring and a value of ϕ some 20° to 30° higher than that of the average proline residue, must transmit most of this difference to any peptide into which it is incorporated. This implies that the growth-inhibitory properties of AzCOOH must result from its conformational differences with proline and that significant changes in tertiary structure are thus caused when AzCOOH is incorporated in place of proline in a polypeptide having a biological function. To confirm this, a model was constructed at a scale of 1.25 cm/Å using foam balls for the azetidine ring and gluing standard Corey–Pauling–Koltun model components[33] to the ring at C(1), O(1), and O(2), and substituting an amide carbon atom for the two hydrogen atoms on nitrogen. This model was compared with one of proline by alternately inserting them one residue short of the N-terminal residue in a model of an α helix. AzCOOH caused a change in direction at the N-terminal amide bond of 16° and an alteration of position of the succeeding amide carbons of 0.7 Å. However, the altered conformation resulted largely from features not expressible as ϕ or ψ angles; the square ring of AzCOOH differs from the pentagon of proline by about 18° in the direction of adjacent ring-to-out-of-ring bonds. Thus it was no surprise that the directions of successive amide bonds in the helix were changed by 16°.

We therefore conclude that the conformation of AzCOOH is sufficiently different from that of proline to cause the growth-modifying effects observed when it replaces proline in functional polypeptides.[34] Furthermore, the conformational features of AzCOOH and other amino acids cannot be fully described by reference to ϕ and ψ angles, even though the bond lengths have close to average values. We suggest that an examination of the detailed physical and chemical properties of polypeptides and proteins having azetidinecarboxylic acid in place of proline could be a fruitful line of investigation.

Acknowledgments. The authors gratefully acknowledge the technical assistance of Mrs. J. Klinger, Miss Rose Pantalone, and Mr. N. C. Seeman. Thanks are also due to Professor R. Shiono of the Crystallography Laboratory for programming assistance, and to Dr. Orrin Taulbee, Director of the Pitt Computing Center, for use of the IBM 7090. For reading the manuscript we express our appreciation to Professor G. A. Jeffrey.

(33) W. L. Koltun, *Biopolymers*, 3, 665 (1965).

(34) Studies dealing with the incorporation of azetidinecarboxylic acid into collagen have recently been published: T. Takeuchi and D. J. Prockop, *Biochim. Biophys. Acta*, 175, 142 (1969); T. Takeuchi, J. Rosenbloom, and D. J. Prockop, *ibid.*, 175, 156 (1969).

The Synthesis and Certain Pharmacological Properties of Deamino-oxytocinoic Acid Methylamide and Deamino-oxytocinoic Acid Dimethylamide[1,2]

Herbert Takashima, Wolfgang Fraefel, and Vincent du Vigneaud[3]

Contribution from the Department of Chemistry, Cornell University, Ithaca, New York 14850. *Received June 26, 1969*

Abstract: Deamino-oxytocinoic acid methylamide and deamino-oxytocinoic acid dimethylamide, analogs in which methyl groups have replaced one or both hydrogens, respectively, on the nitrogen of the 9-carboxamide group of deamino-oxytocin, have been synthesized by the Merrifield solid phase method and their oxytocic and avian vasodepressor potencies have been determined. Deamino-oxytocinoic acid methylamide possessed approximately 29 units/mg of oxytocic activity and the dimethylamide analog possessed approximately 16 units/mg. Neither analog exhibited any detectable amounts of avian vasodepressor activity. Deamino-oxytocin possesses 803 ± 36 units/mg of oxytocic activity and 975 ± 24 units/mg of avian vasodepressor activity.

In earlier investigations[4–6] it was found that oxytocinoic acid (9-deamido-oxytocin) possessed extremely low levels of oxytocic and avian vasodepressor activities in contrast to the high levels characteristic of oxytocin. Thus the amide group of the glycinamide residue at position 9 of oxytocin (Figure 1) is highly important for the manifestation of these activities. It has likewise been found that deamino-oxytocinoic acid[6] possesses an extremely low level of avian vasodepressor activity and no detectable oxytocic activity, in contrast to deamino-oxytocin[7–10] which is even more potent than oxytocin with respect to these activities. Deamino-oxy-

(1) This work was supported in part by Grant HE-11680 from the National Heart Institute, U. S. Public Health Service. Dr. Fraefel wishes to acknowledge a Geigy Chemical Corporation Fellowship.

(2) All optically active amino acid residues are of the L variety.

(3) To whom correspondence and reprint requests should be addressed.

(4) B. M. Ferrier and V. du Vigneaud, *J. Med. Chem.*, 9, 55 (1966).

(5) H. Klostermeyer, Ph.D. Thesis, Technisches Hochschule, Aachen, Germany, 1964.

(6) H. Takashima and V. du Vigneaud, unpublished data.

(7) V. du Vigneaud, G. Winestock, V. V. S. Murti, D. B. Hope, and R. D. Kimbrough, Jr., *J. Biol. Chem.*, 235, PC64 (1960).

(8) D. B. Hope, V. V. S. Murti, and V. du Vigneaud, *ibid.*, 237, 1563 (1962).

(9) D. Jarvis and V. du Vigneaud, *Science*, 143, 545 (1964).

(10) B. M. Ferrier, D. Jarvis, and V. du Vigneaud, *J. Biol. Chem.*, 240, 4264 (1965).

tocinoic acid methylamide and deamino-oxytocinoic acid dimethylamide have now been synthesized to see whether the presence of one or both of the hydrogens on the nitrogen of the carboxamide portion of the glycinamide residue is essential for the exhibition of these activities by deamino-oxytocin.

The desired precursor for both analogs, S-benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycyl nitrated resin, was prepared as in the solid phase synthesis of deamino-oxytocin.[11] Cleavage of the polypeptide chain from the nitrated resin with methylamine in one case and with dimethylamine in the other gave the methylamide and dimethylamide, respectively, of S-benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycine.[12] Debenzylation of the protected polypeptides with sodium and liquid ammonia by the method of Sifferd and du Vigneaud[13] followed by oxidative cyclization with potassium ferricyanide by the method of Hope, *et al.*,[8] yielded deamino-oxytocinoic acid methylamide and deamino-oxytocinoic acid dimethylamide. Both analogs were purified by partition chromatography[14] followed by gel filtration[15] on Sephadex G-25.

The crystalline deamino-oxytocinoic acid methylamide possessed 29 units/mg of oxytocic activity[16] and no detectable amount of avian vasodepressor activity.[16] Deamino-oxytocin itself possesses 975 ± 24 units/mg of avian vasodepressor activity and 803 ± 36 units/mg of oxytocic activity.[10] Thus the avian vasodepressor potency of deamino-oxytocin is eliminated and the oxytocic potency is drastically reduced by substitution of a methyl group for one of the hydrogen atoms of the 9-carboxamide group of the molecule.

Deamino-oxytocinoic acid dimethylamide was not obtained in crystalline form, but the highly purified compound exhibited 16 units/mg of oxytocic activity and no avian vasodepressor activity. Thus the substitution of a methyl group for the hydrogen of the methylamide moiety of the methylamide analog lowers still further the level of oxytocic activity.

Experimental Section

Boc-glycyl Nitrated Resin. The nitrated chloromethylcopolystyrene–2% divinylbenzene resin was prepared and esterified by the procedures reported earlier.[11] Amino acid analysis of an acid hydrolysate (dioxane–12 *N* HCl, 1:1) showed the product to contain 0.39 mmol of glycine/g of esterified resin.

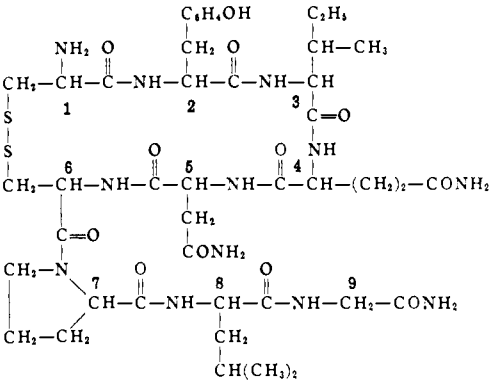


Figure 1. Structure of oxytocin with numbers indicating the position of the individual amino acid residues.

S-Benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycyl Nitrated Resin. Boc-glycyl nitrated resin (10 g) was placed in the reaction vessel. The procedure described in an earlier communication[11] for the introduction of each new amino acid residue was followed.

For the washing steps 50-ml portions of the appropriate solvent were used. In the deprotection step 50 ml of 1 *N* HCl in glacial acetic acid was used, and neutralization of the hydrochloride was accomplished by treatment with 7.5 ml of triethylamine in 42.5 ml of dimethylformamide. In the coupling steps 7.8 mmol of the appropriate Boc-amino acid in 35 ml of methylene chloride and 8.5 mmol of N,N′-dicyclohexylcarbodiimide in 15 ml of methylene chloride were used. The coupling reactions involving Boc-asparagine and Boc-glutamine were carried out *via* their *p*-nitrophenyl esters (7.8 mmol in 50 ml of distilled dimethylformamide) with a reaction time of 24 hr.

The incorporation of the Boc-isoleucine into the peptide chain was accomplished with the following modifications: (1) cleavage of the Boc group of the glutamine residue[11] by treatment with 50 ml of trifluoroacetic acid for 15 min at 25°; (2) neutralization of the trifluoroacetate with 4 ml of triethylamine in 46 ml of dimethylformamide for 5 min.

Following the incorporation of the S-benzyl-β-mercaptopropionic acid, the protected polypeptide-nitrated resin compound was further washed with 50-ml portions of glacial acetic acid (three times), absolute ethanol (three times), and methylene chloride (three times). The product was dried *in vacuo* over KOH pellets, yield 12.7 g.

S-Benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycine Methylamide. A suspension of the preceding protected polypeptide-nitrated resin compound (5.04 g) in 15 ml of anhydrous methanol was placed in a 100-ml round-bottomed flask equipped with a Dry Ice–acetone cold finger condenser and a sodium hydroxide drying tower. The slurry was cooled in an ice bath and methylamine (50 ml) was condensed dropwise into the reaction flask. The reaction mixture was stirred at 0–4° for 20 hr. The methanol and methylamine were removed under aspirator vacuum. Dimethylformamide (70 ml) was added to the dry residue and the suspension was stirred vigorously for 2 hr. The resin was filtered off and washed twice with 20-ml portions of dimethylformamide.

The solvent was removed from the combined filtrate and washings on a rotary evaporator. The residue was dissolved in dimethylformamide (25 ml), and water (15 ml) was added gradually with stirring. The turbid solution was allowed to stand in the refrigerator overnight. The precipitate was filtered off, washed with water (three times), and dried *in vacuo* over KOH pellets: yield 356 mg; mp 224–228°. This material was dissolved in dimethylformamide, and water was slowly added until the solution became slightly turbid. The solution was allowed to stand overnight in the refrigerator, and the precipitate was filtered off, washed three times with water, and dried *in vacuo* over KOH pellets: yield 196 mg; mp 230–232°; $[\alpha]^{22}D$ −41° (*c* 1, dimethylformamide).

Anal. Calcd for $C_{85}H_{37}O_{12}N_{11}S_2$: C, 61.0; H, 6.86; N, 12.0. Found: C, 60.6; H, 6.83; N, 12.0.

Deamino-oxytocinoic Acid Methylamide. S-Benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzyl-

(11) H. Takashima, V. du Vigneaud, and R. B. Merrifield, *J. Amer. Chem. Soc.*, 90, 1323 (1968).

(12) Cleavage of a protected polypeptide from the chloromethylcopolystyrene–2% divinylbenzene with methylamine has been successfully carried out by H. C. Beyerman and coworkers in the synthesis of N-benzyloxycarbonyl-S-benzylcysteinyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycine methylamide: H. C. Beyerman and H. Maassen Van Den Brink-Zimmermannová, *Recueil*, 87, 1 (1968).

(13) R. H. Sifferd and V. du Vigneaud, *J. Biol. Chem.*, **108**, 753 (1935).

(14) D. Yamashiro, *Nature*, **201**, 76 (1964); D. Yamashiro, D. Gillessen, and V. du Vigneaud, *J. Amer. Chem. Soc.*, **88**, 1310 (1966).

(15) J. Porath and P. Flodin, *Nature*, **183**, 1657 (1959).

(16) The four-point assay design of H. O. Schild [*J. Physiol.*, **101**, 115 (1942)] was used for measurement of pharmacological activities against the U.S.P. Posterior Pituitary Reference Standard. The assays for oxytocic activity were performed on isolated uteri from rats in natural estrus according to the method of P. Holton [*Brit. J. Pharmacol.*, **3**, 328 (1948)], as modified by R. A. Munsick [*Endocrinology*, **66**, 451 (1960)], with the use of magnesium-free van Dyke–Hastings solution. Avian vasodepressor assays were performed on conscious chickens according to the procedure employed by R. A. Munsick, W. H. Sawyer, and H. B. van Dyke, *Endocrinology*, **66**, 860 (1960).

cysteinylprolylleucylglycine methylamide (130 mg) was dissolved in 150 ml of stirred boiling liquid ammonia (distilled from sodium in an all-glass apparatus). A fresh sodium stick was momentarily introduced intermittently until the blue color persisted for 20 sec. The ammonia was evaporated at the water aspirator and the last 25 ml was lyophilized. The residue was dissolved in 270 ml of 0.05% aqueous trifluoroacetic acid. The pH of the solution was adjusted to 8.0 with dilute ammonium hydroxide and an excess of 0.1 *N* potassium ferricyanide (2.75 ml) was added to the stirred solution. After 15 min AG 3-X4 resin (Bio-Rad Laboratories) (trifluoroacetate form) was added and stirring was continued for 15 min to remove ferrocyanide and excess ferricyanide ions. The resin was removed by filtration and the solution was lyophilized. The residue was dissolved in 10 ml of the upper phase of the solvent system 1-butanol–benzene–3.5% aqueous acetic acid (containing 1.5% pyridine) (1:1:2) and applied to a Sephadex G-25 (100–200 mesh) column (2.80 × 60 cm) that had been equilibrated with the lower and upper phases of the solvent system. The column was eluted with the upper phase, and 48 fractions of 10.3 ml each were collected. The chromatogram obtained by plotting the Folin–Lowry color values[17] of the fractions showed a major peak with a maximum at fraction 29 (R_f 0.32). The fractions corresponding to this peak were pooled, twice the volume of water was added, and the resulting mixture was concentrated under reduced pressure to about 20 ml and lyophilized.

The lyophilized powder (53.6 mg) was dissolved in 5.5 ml of 0.2 *N* acetic acid and subjected to gel filtration on a Sephadex G-25 (200–270 mesh) column (2.82 × 68 cm) that had been equilibrated with 0.2 *N* acetic acid. The column was eluted with 0.2 *N* acetic acid, and 105 fractions of 6.2 ml each were collected. A plot of the Folin–Lowry color values of the various fractions showed a single symmetrical peak with a maximum at fraction 53. The fractions corresponding to this peak were pooled and lyophilized to give a white powder, yield 47.5 mg.

This lyophilized powder (47.5 mg) was dissolved in 0.8 ml of deionized water with warming in a water bath at 60–70°. The solution was filtered through a sintered-glass funnel. A 0.4-ml wash was also passed through the funnel. The combined filtrate and wash (1.2 ml) were allowed to stand in the refrigerator for 6 days. The deamino-oxytocinoic acid methylamide crystals were filtered off, washed twice with 0.2-ml portions of deionized water, and dried *in vacuo:* yield 31.2 mg; mp 182–183° uncor (Fisher-Johns melting point apparatus); $[\alpha]^{23}D$ −118.0° (*c* 0.5, 1 *N* acetic acid).

Anal. Calcd for $C_{44}H_{67}N_{11}O_{12}S_2$: C, 52.5; H, 6.71; N, 15.3. Found: C, 52.5; H, 7.01; N, 15.2.

A sample was hydrolyzed in 6 *N* HCl at 110° for 24 hr and analyzed[18] in the Beckman–Spinco amino acid analyzer. The following molar ratios were obtained: aspartic acid, 1.0; glutamic acid, 1.0; proline, 0.9; glycine, 1.0; half-cystine, 0.44; mixed disulfide of β-mercaptopropionic acid and cysteine,[8] 0.53; isoleucine, 1.0; leucine, 1.2; tyrosine, 1.0; ammonia, 2.0; and methylamine, 1.0.

S-Benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycine Dimethylamide. The peptide-nitrated resin (1.7 g) was suspended in freshly distilled dimethylamine (25 ml) and stirred for 24 hr at 4°. Further treatment of the reaction mixture was analogous to that described for the monomethyl analog: yield 351 mg of an off-white powder; mp 210–215°. Thin layer chromatography on silica gel G (E. Merck AG, Darmstadt) in the solvent system chloroform–methanol (8:2) showed a main spot accompanied by two faster moving spots.

For purification this protected peptide (245 mg) was dissolved in the upper phase of the solvent system toluene–glacial acetic acid–water (20:20:3.5) and put onto a Sephadex LH-20 column (2.83 × 56 cm) which had been equilibrated with the lower phase of the solvent system. Elution of the column was carried out with the upper phase, and 9.8-ml fractions were collected. Aliquots (0.1 ml) of every other fraction were dried *in vacuo* and 0.1 ml of dimethylformamide was added to each aliquot. Folin–Lowry analysis of these aliquots gave a major peak in the region of fractions 18–32 accompanied by a very small peak contained in fractions 10–16. Fractions 20–30 were pooled and the solvent was removed on a rotary evaporator. The residue was dissolved in 10 ml of glacial acetic acid and 50 ml of water was added. The white precipitate was filtered off, washed with two 5-ml portions of water, and dried *in vacuo* to give 208 mg of a white powder: mp 227–230°; $[\alpha]^{22}D$ −36.4° (*c* 0.85, dimethylformamide). Thin layer chromatography on silica gel G in the solvent system chloroform–methanol (2:8) showed a single spot.

Anal. Calcd for $C_{66}H_{89}O_{12}N_{11}S_2$: C, 61.3; H, 6.94; N, 11.9. Found: C, 61.4; H, 7.15; N, 11.7.

Deamino-oxytocinoic Acid Dimethylamide. S-Benzyl-β-mercaptopropionyl-O-benzyltyrosylisoleucylglutaminylasparaginyl-S-benzylcysteinylprolylleucylglycine dimethylamide (200 mg) was reduced and oxidized as described above for the preparation of deamino-oxytocinoic acid methylamide. After the removal of the ferrocyanide and ferricyanide ions with AG 3-X4 resin (trifluoroacetate form), the solution was concentrated to about 50 ml and lyophilized.

The lyophilized powder was dissolved in 10 ml of the upper phase of the solvent system 1-butanol–benzene–3.5% aqueous acetic acid containing 1.5% pyridine (1:2:3) and subjected to partition chromatography on a Sephadex G-25 (100–200 mesh) column (2.82 × 65.2 cm). In the resulting chromatogram deamino-oxytocinoic acid dimethylamide appeared as a symmetrical peak with R_f 0.19 accompanied by a fast moving smaller peak with R_f 0.60. Isolation of the material represented by the main peak gave 32 mg of the analog. Gel filtration on a Sephadex G-25 (200–270 mesh) column (2.82 × 65 cm) with 0.2 *N* acetic acid gave one peak with a maximum at effluent volume 322 ml. The fractions corresponding to this peak were cooled and lyophilized, yielding 26 mg of deamino-oxytocinoic acid dimethylamide, $[\alpha]^{22}D$ −98.3° (*c* 0.7, 1 *N* acetic acid). Thin layer chromatography on silica gel G in 1-butanol–glacial acetic acid–water (4:1:1) showed only one spot with R_f 0.33.

Anal. Calcd for $C_{45}H_{69}N_{11}O_{12}S_2$: C, 53.0; H, 6.81; N, 15.1. Found: C, 52.8; H, 6.84; N, 14.9.

Quantitative amino acid analysis after acid hydrolysis gave the following molar ratios: aspartic acid, 1.0; glutamic acid, 1.0; proline, 1.0; glycine, 1.0; half-cystine, 0.44; mixed disulfide of β-mercaptopropionic acid and cysteine, 0.49; isoleucine, 1.0; leucine, 1.1; tyrosine, 1.0; and ammonia, 2.1.

Since dimethylamine is ninhydrin negative, the quantitative determination of dinitrophenyldimethylamine was carried out on the hydrolysate after treatment of the hydrolysate with dinitrofluorobenzene under the conditions used by Sanger[19] for the dinitrophenylation of peptides.

The preparation of an authentic sample of dinitrophenyldimethylamine as a standard was based on the procedure of Levy and Chung[20] for preparing the dinitrophenyl derivative of amino acids. A mixture of dimethylamine hydrochloride (1.63 g), sodium carbonate (4 g), and 2,4-dinitrofluorobenzene (1.86 g) in 40 ml of water–methanol (1:1) was stirred overnight at 0°. The methanol was removed on a rotary evaporator. The solution was then cooled in an ice–water bath and acidified with 3 *N* HCl to pH 1. A yellow precipitate was filtered off, washed with three 5-ml portions of water, and dried *in vacuo* over KOH pellets: yield 1.89 g; mp 75–78°. The product was dissolved in ether (150 ml), and the solution was allowed to stand overnight at −20°. The yellow crystals were filtered off, washed with two 5-ml portions of ether, and dried *in vacuo:* yield 1.60 g; mp 76–78° (lit.[21] mp 74°).

The quantitative determination of the dinitrophenyldimethylamine in the hydrolysate after dinitrophenylation was performed by a modification of the method of Levy for the determination of amino acids.[22] Three spots (10, 20, and 30 μl) of an ethanolic solution of the dinitrophenylated hydrolysate corresponding to 0.02, 0.04, and 0.06 μmol of analog were put on a silica gel G plate (20 × 20 cm). After the development of the chromatogram in the solvent system toluene–pyridine–glacial acetic acid (80:10:1), the spots corresponding to dinitrophenyldimethylamine were scraped from the plate. The powder so obtained was extracted with 3.0 ml of 1% aqueous sodium bicarbonate solution. The suspended gel was then centrifuged off and the clear yellow supernatant was measured by photometry at 350 mμ. Comparison with a standard curve of the color values of authentic samples of dinitrophenyldimethylamine treated in the same manner gave 1.1 mol of dinitrophenyldimethylamine per mol of analog.

(17) O. H. Lowry, N. J. Rosebrough, A. L. Farr, and R. J. Randall, *J. Biol. Chem.*, **193**, 265 (1951).
(18) D. H. Spackman, W. H. Stein, and S. Moore, *Anal. Chem.*, **30**, 1190 (1958).
(19) F. Sanger and E. O. P. Thompson, *Biochem. J.*, **53**, 353 (1953).
(20) A. L. Levy and D. Chung, *J. Amer. Chem. Soc*, 77, 2899 (1955).
(21) A. M. Asatoor, *J. Chromatog.*, 4, 144 (1960).
(22) A. L. Levy, *Nature*, **174**, 126 (1954).

Acknowledgments. The authors are indebted to Miss Paula Glose for the bioassays done under the direction of Dr. L. Nangeroni, New York State Veterinary College, Cornell University.

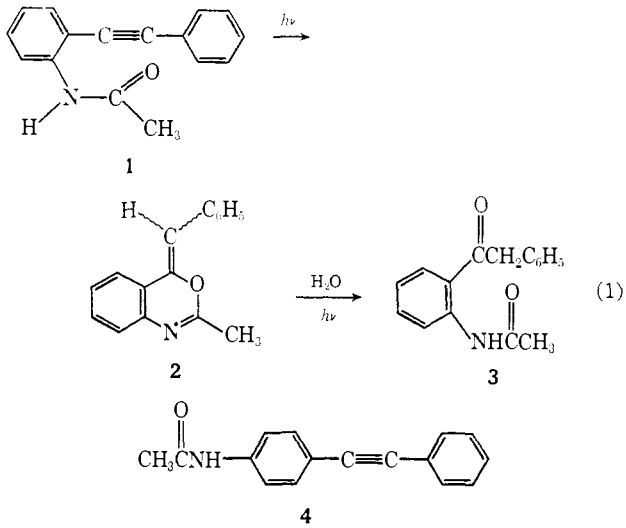
Communications to the Editor

New Photochemical Addition Reactions of Acetylenes. Neighboring Group Participation in Photolytic Hydration of Acetylenes

Sir:

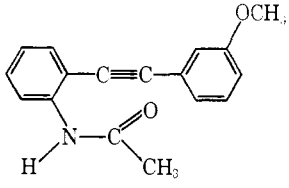
The photochemistry of diphenylacetylene has been previously limited to cycloaddition[1a,b] and to oxidation.[1c] We wish to describe three new photolytic reactions of diarylacetylenes: (1) hydration to arylcarbinyl aryl ketones involving accelerative intramolecular participation of amido groups, (2) addition of methanol to give methyl *cis*- and *trans*-1,2-diarylvinyl ethers, and (3) reduction by methanol resulting in *cis*- and *trans*-diarylethylenes.

It has been found that *o*-acetamidophenyl(phenyl)-acetylene (**1**) photolyzes[2] rapidly in wet hexane in the absence of oxygen to give *o*-acetamidophenyl benzyl ketone (**3**, mp 98.5–99.5°, lit.[3] 97–98°). Irradiation of **1** (0.005–0.01 *M*) under nitrogen in dry hexane in quartz for 8 hr, removal of solvent, and crystallization of the resulting yellow oil from acetone–water afforded **3** in 85% yield. Photolysis of *p*-acetamidophenyl phenyl ketone (**4**) under conditions identical with that for **1** did not result in detectable reaction. To account for the rapid photochemical response of **1** and the unidirectional hydration resulting in **3**, it is proposed that upon excitation (eq 1) there is neighboring group interaction of the *o*-acetamido and the acetylene groups, giving 2-methyl-4-benzylidene-4H-3,1-benzoxazines (**2**); hydrolysis of **2**, apparently photochemically accelerated, thus yields **3**.

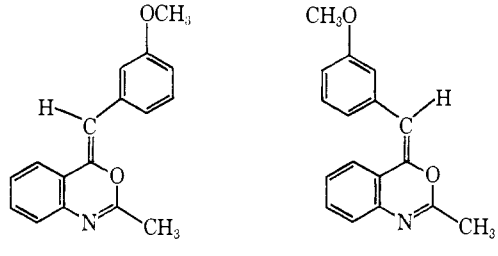


(1)

Inquiry as to whether benzoxazine intermediates of the type such as **2** are indeed formed photolytically and can be isolated was made by irradiating *o*-acetamidophenyl-*m*-methoxyphenylacetylene[4] (**5**, 0.005–0.01 *M*; mp 108–109°) in dry hexane containing Linde 3A Molecular Sieve. Photolysis[2] was effected for 25 hr to ensure complete conversion of **5**, the solvent was vacuum evaporated, and the yellow product was rapidly analyzed. The infrared spectrum did *not* show significant NH or C=O stretching. The nmr in $DCCl_3$ shows absorption at τ 7.92 (3 H, s), 6.4 and 6.31 (3 H, s, relative area *ca.* 2:3), 4.11 and 3.85 (1 H, s, relative area *ca.* 3:2), and 2.5–3.4 (8 H, m). Upon adding one drop

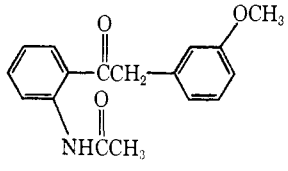


5



6a **6b**

of water and warming the nmr sample at 50° for several days, the spectrum gradually changed to that of *o*-acetamidophenyl *m*-methoxybenzyl ketone[5] (**7**, mp 100°);



7

(1) (a) G. Buchi, C. W. Perry, and E. W. Robb, *J. Org. Chem.*, 27, 4106 (1962); (b) O. L. Chapman and G. Lenz in "Organic Photochemistry," Vol. I, O. L. Chapman, Ed., Marcel Dekker, Inc., New York, N. Y., 1967, p 283 ff; (c) R. C. Henson, J. L. W. Jones, and E. D. Owen, *J. Chem. Soc., A*, 116 (1967), and references therein.

(2) In a typical run 16 General Electric G25T8 tubes mounted in a circular bank (shop fashioned) served as the light source for 500-ml solutions (0.01 *M*).

(3) D. W. Ockenden and K. Schofield, *J. Chem. Soc.*, 3440 (1953).

(4) This compound was chosen in order to more conveniently study intermediates of type **2** by nmr techniques.

(5) Mass spectral parent peak 283; nmr ($DCCl_3$) τ 7.96 (s, 3 H), 6.34 (s, 3 H), 5.86 (s, 2 H), 1.30–3.56 (m, 8 H), and −1.3 to −1.52 (s, 1 H); **7** gave proper analysis.