I, CH_2 should have an angle larger than 160°. Extrapolating from θ_{IV} to θ_I to θ_{CH_2}, a linear geometry for CH_2 is indicated. Although all the derivatives which have been observed by e.s.r. are bent, nonconjugated alkylmethylenes may be linear as phenylmethylmethylene has an E/D ratio[11] close to that of phenylmethylene.

A white chemiluminescence is observed after irradiation of a solution of bis(trifluoromethyl)diazomethane in perfluorodimethylhexane. At 77°K. the light is emitted for more than an hour after irradiation ceases and appears only if the sample contains oxygen or is exposed to oxygen after irradiation. On warming, the emission is sufficiently bright to be observed under ordinary room illumination. The spectrum is identical with the phosphorescence of hexafluoroacetone measured at 77°K. Presumably IV reacts with oxygen and eventually yields the lowest triplet state of hexafluoroacetone.[12]

Acknowledgment. We wish to thank Mr. R. M. R. Cramer for his assistance in obtaining the e.s.r. spectra.

the values for phenyltrifluoromethylmethylene and those of I were said to lead to $D_{CH_2} = 0.74$ cm.$^{-1}$. This conclusion involved a correction of +0.02 cm.$^{-1}$ on going from a bent to a linear configuration with I, following the dependence of D on angle predicted in ref. 10. However, such an angular dependence of D, if it is correct, is overshadowed in I and IV by the effect of the CF_3 groups. With I and IV a decrease in angle accompanies an increase in D. In any case, 0.74 cm.$^{-1}$ may be viewed as an upper limit to our extrapolated values, but should be given less weight than the lower values mentioned.

(11) $D = 0.4957$ cm.$^{-1}$, $E = 0.0265$ cm.$^{-1}$ (R. W. Murray, to be published).

(12) The phosphorescence of benzophenone has been observed from the reaction of oxygen with diphenylmethylene [A. M. Trozzolo, R. W. Murray, and E. Wasserman, *J. Am. Chem. Soc.*, **84**, 4490 (1962)].

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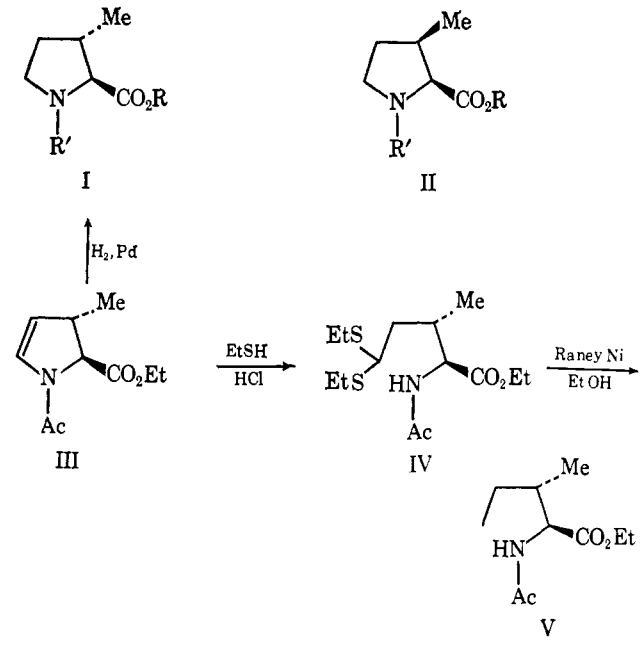
Correlation of *cis*- and *trans*-3-Methylproline with (Allo) Isoleucine

Sir:

The biosynthesis of actinomycin from *Streptomyces antibioticus* is strongly inhibited by 3-methylproline.[1] As little as 0.1 μg./ml. of the mixture of racemic *cis*- and *trans*-3-methylprolines causes a 50% inhibition. In order to extend and detail these observations the diastereoisomers were separated and their configuration elucidated as follows.

The racemates of *cis*- and *trans*-3-methylproline have recently been synthesized.[2] One isomer, m.p. 218–219°, had a n.m.r. coupling constant $J_{23} = 4.6$ c.p.s. while the other, m.p. 210–211°, had $J_{23} = 7.2$ c.p.s. Although the question of relative configuration was left open, it seemed probable, by analogy with 3-hydroxyproline,[3,4] that the isomer having the larger coupling constant J_{23} would be *cis* (II; R = R′ = H: L forms are shown throughout to represent racemates).

The *cis* and *trans* isomers were originally separated by fractional crystallization. For small-scale separations



we have found ion-exchange chromatography (Amberlite IR-120) more convenient. For large-scale preparations the method of choice is preferential saponification. A study of the relative rates of saponification (4.5-fold excess of 0.24 *N* methanolic sodium hydroxide at 32°) of a mixture of I and II (R = Me, R′ = Tos) by gas–liquid partition chromatography showed that one of the isomers was saponified to the extent of 96% after 55 min. Only 5% of the other ester had hydrolyzed during this time as shown by g.l.p.c. of the acid fraction after re-esterification. The more resistant ester must be the sterically hindered *cis* form II (R = Me); the saponified acid is therefore the *trans* form I (R = H). This confirms the initial assignments based on n.m.r. data.

Final proof was obtained by correlation of I (R = Et, R′ = Ac) with DL-isoleucine *via* III, IV, and V.

N-Acetyl-3-methyl-4,5-dehydro-DL-proline ethyl ester, the synthesis of which will be described elsewhere, was separated into *cis* and *trans* isomers by preferential saponification. The *trans* ester III, m.p. 49–51°,[5] was hydrogenated to N-acetyl-3-methyl-DL-proline ethyl ester, identified by g.l.p.c. (Table I) with the corresponding derivative of I (R = R′ = H).

Reaction of III with ethyl mercaptan and hydrogen chloride in dioxane at room temperature gave the mercaptal IV. Desulfurization of IV with Raney nickel in boiling ethanol afforded N-acetyl-DL-isoleucine ethyl ester (V), which was compared and identified with the corresponding derivative of L-isoleucine by g.l.p.c. and n.m.r. (the α-proton in N-acetyl-L- and -D-alloisoleucine ethyl ester appeared as multiplets at δ 4.68 and 4.78 p.p.m., respectively). The other diastereoisomer of III was similarly converted both into N-acetyl-DL-alloisoleucine ethyl ester and into II (R = Et, R′ = Ac).

These interconversions establish unambiguously the stereochemistry of the isomeric 3-methylprolines. Additional interest is provided by the recent claim that *cis*-3-methyl-L-proline occurs in the peptide antibiotic

(1) T. Yoshida, A. B. Mauger, B. Witkop, and E. Katz, 148th National Meeting of the American Chemical Society, Chicago, Ill., Sept. 1964, Abstracts, p. 40C. Also in press.

(2) D. A. Cox, A. W. Johnson, and A. B. Mauger, *J. Chem. Soc.*, 5024 (1964).

(3) F. Irreverre, K. Morita, A. V. Robertson, and B. Witkop, *J. Am. Chem. Soc.*, **86**, 8293 (1964).

(4) J. Blake, C. D. Willson, and H. Rapoport, *ibid.*, **86**, 5293 (1964).

(5) Most of the 3-methylproline derivatives described were low melting solids or liquids. They all gave satisfactory analytical results for C, H, and N and were characterized by n.m.r., infrared, and g.l.p.c. techniques.

Table I. Characterization of N-Acetyl Esters of the Diastereoisomeric Pairs of 3-Methylprolines and Isoleucines by G.l.p.c.

Compound	Column[a]	Temp., °C.	Retention, min.
N-Acetyl-*trans*-3-methyl-DL-proline ethyl ester (I, R′ = Ac, R = Et)	A	138	5.9
	B	189	7.1
N-Acetyl-*cis*-3-methyl-DL-proline ethyl ester (II, R′ = Ac, R = Et)	A	138	6.8
	B	189	8.1
N-Acetyl-L- (and -DL-) isoleucine ethyl ester (V)	B	158	11.7
N-Acetyl-D- (and -DL-) alloisoleucine ethyl ester	B	158	11.0

[a] A: 3% SE52 on 6-ft. Gaschrom A; B: 3% neopentyl glycol succinate on 6-ft. Gaschrom Z.

bottromycin A.[6] This is the first reported instance of the occurrence of this amino acid in a natural product, although *trans*-4-methyl-L-proline occurs in apples[7] and *cis*-4-methyl-L-proline was isolated from hydrolysates of antibiotic I.C.I. 13,959 from a strain of *Paecilomyces*.[8]

The optical resolution of the *cis*- and *trans*-3-methyl-DL-prolines and their inhibitory effects on the biosynthesis of actinomycin are under study.

(6) S. Nakamura, T. Chikaike, K. Karasawa, H. Yonehara, and H. Umezawa, *J. Antibiot.* (Tokyo), **A18**, 47 (1965); S. Nakamura, T. Chikaike, and H. Umezawa, *ibid.*, **A18**, 60 (1965); S. Nakamura, T. Chikaike, H. Yonehara, and H. Umezawa, *Chem. Pharm. Bull. Japan*, **13**, 599 (1965).

(7) A. C. Hulme and W. Arthington, *Nature*, **170**, 659 (1952); **173**, 588 (1954).

(8) G. W. Kenner and R. C. Sheppard, *ibid.*, **181**, 48 (1958).

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Received September 3, 1965

Aminomalononitrile and 4-Amino-5-cyanoimidazole in Hydrogen Cyanide Polymerization and Adenine Synthesis[1]

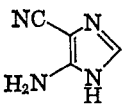
Sir:

The formation of adenine spontaneously in ammoniacal cyanide solutions[2a,b,g] or during the irradiation of dilute aqueous solutions of hydrogen cyanide[2d,e,h,k] has led to much speculation concerning the role of these reactions in the prebiological synthesis of adenine.[2] Several reaction pathways have been considered, but for the most part the evidence remains fragmentary (see particularly ref. 2j, which claims the isolation of aminomalononitrile but gives no details).

We wish to report the preparation of two new "polymers" of hydrogen cyanide, aminomalononitrile (I) and 4-amino-5-cyanoimidazole (II), and to demonstrate their use in the synthesis of heterocyclic compounds[3] and in the study of the mechanism of HCN polymerization[4] and adenine synthesis.[2]

Reduction of oximinomalononitrile[5] with aluminum amalgam in ether–tetrahydrofuran gave a 45–50% yield of I isolated as the *p*-toluenesulfonate, m.p. 180–181°. *Anal.* Calcd. for $C_{10}H_{11}N_3O_3S$: C, 47.41; H, 4.38; N, 16.59. Found: C, 47.20; H, 4.39; N 16.52.[6]

Treatment of I with acid anhydrides yielded the corresponding oxazoles. Thus acetic anhydride in formic acid yielded III (R = H), m.p. 184–186°. *Anal.* Calcd for $C_4H_3N_3O$: C, 44.04; H, 2.77; N, 38.52. Found: C, 43.99; H, 2.93; N, 38.58. Acetic anhydride gave III (R = CH_3), m.p. 153–155°. *Anal.* Calcd. for $C_5H_5N_3O$: C, 48.78; H, 4.09; N, 34.13. Found: C, 48.77; H, 4.35; N, 33.91. Propionic anhydride gave III (R = C_2H_5), m.p. 148–149°. *Anal.* Calcd. for $C_6H_7N_3O$: C, 52.55; H, 5.14; N, 30.64. Found: C, 52.32; H, 5.29; N, 30.55. Benzoic anhydride gave III (R = C_6H_5), m.p. 241–243°. The oxazole structure was proved by direct comparison of III (R = C_6H_5) with a sample prepared by a published procedure.[7]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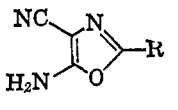
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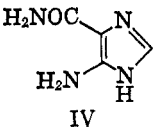
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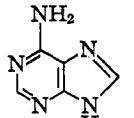
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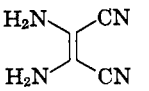
VI

The imidazole ring system could be formed by the condensation of I with formamidine acetate in ethanol to give a 35% yield of II as the *p*-toluenesulfonate, m.p. 168–169° (*Anal.* Calcd. for $C_{11}H_{12}N_4O_3S$: C, 47.13; H, 4.31; N, 20.00. Found: C, 46.90; H, 4.54; N, 19.62), which was also obtained in 15% yield by dehydration of 4-aminoimidazole-5-carboxamide (IV)[8] with thionyl chloride in pyridine. Treatment of II with formamidine acetate in boiling methoxyethanol[9] yielded adenine (V) (68%), m.p. 357–360°.

A brown polymer and diaminomaleonitrile (VI), m.p. 183–185°, result from the treatment of I with aqueous potassium cyanide at pH 9–10. Compound VI is the

(1) This work was supported by Grant GB-3152 from the National Science Foundation.

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(5) G. Ponzio, *Gazz. chim. ital.*, **61**, 561 (1931).

(6) All new compounds prepared in this work had infrared, ultraviolet, and nuclear magnetic resonance spectra in agreement with the proposed structures. The spectra of known compounds were identical with spectra of authentic samples or with literature spectra.

(7) H. T. Clarke, J. R. Johnson, and R. Robinson, "The Chemistry of Penicillin," Princeton University Press, Princeton, N. J., 1949, p. 729.

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(9) This approach to the synthesis of purines was developed by E. C. Taylor; *e.g.*, see E. C. Taylor and R. W. Hendess, *J. Am. Chem. Soc.*, **87**, 1995 (1965), and references therein.