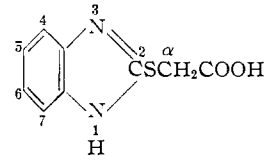
TABLE I

PROPERTIES OF 2-BENZIMIDAZOLYLTHIO ACIDS



Ring substitution	Acid side chain	M.p.,[a] °C.	Formula	Yield,[b] %	Neut. equiv. Calcd.	Neut. equiv. Found	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found	Nitrogen, % Calcd.	Nitrogen, % Found
	α-Propionic	181–182	$C_{10}H_{10}O_2N_2S$	41	222.3	218.8	54.04	53.87	4.54	4.48	12.60	12.72
5-Chloro-	Acetic	193–194	$C_9H_7O_2N_2SCl$	68	242.7	245.5	44.54	44.67	2.91	3.25	11.54	11.23
5-Chloro-	β-Propionic	103–108	$C_{10}H_9O_2N_2SCl$	60	256.7	259.0	46.78	46.58	3.53	3.73	10.91	10.88
5-Chloro-	α-Propionic	166–167	$C_{10}H_9O_2N_2SCl$	39	256.7	253.0	46.78	46.55	3.53	3.43	10.91	10.92
4,6-Dichloro-	Acetic	222–224	$C_9H_6O_2N_2SCl_2$	25	277.1	277.3	39.00	39.34	2.18	2.36	10.11	10.09
5,6-Dichloro-	Acetic	219–221	$C_9H_6O_2N_2SCl_2$	81	277.1	274.1	39.00	39.38	2.18	2.41	10.11	10.08
5,6-Dichloro-	α-Propionic	230–231	$C_{10}H_6O_2N_2SCl_2$	69	291.2	292.3	41.25	41.28	2.77	3.02	9.62	9.69
4,5,6-Trichloro-	Acetic	205–207	$C_9H_5O_2N_2SCl_3$	42	311.6	310.2	34.69	34.75	1.62	1.89	8.99	9.33
4,5,6-Trichloro-	α-Propionic	222–224	$C_{10}H_7O_2N_2SCl_3$	80	325.6	324.4	36.88	37.09	2.17	2.11	8.60	8.84
5-Bromo-	Acetic	194–196	$C_9H_7O_2N_2SBr$	70	287.1	288.1	37.64	37.86	2.46	2.68	9.76	9.84
5-Bromo-	α Propionic	180–181	$C_{10}H_9O_2N_2SBr$	43	301.2	302.4	39.88	39.43	3.01	2.89	9.30	9.45
5-Nitro-	Acetic	191–192	$C_9H_7O_4N_3S$	69	253.2	250.5	42.68	42.54	2.79	2.96	16.59	16.34
5-Nitro-	α-Propionic	186–188	$C_{10}H_7O_4N_3S$	52	267.3	271.0	44.94	45.03	3.39	3.15	15.72	15.99
5-Methoxyl-	Acetic	194–196	$C_{10}H_{10}O_3N_2S$	50	238.3	235.7	50.41	50.79	4.23	4.21	11.76	11.77
5-Methoxyl-	α-Propionic	151–152	$C_{11}H_{12}O_3N_2S$	71	252.3	255.1	52.36	52.63	4.79	4.85	11.10	11.07
5-Methyl-	Acetic	197–200	$C_{10}H_{10}O_2N_2S$	47	222.3	226.4	54.04	54.00	4.54	4.62	12.60	12.61
5-Methyl-	α-Propionic	160–162	$C_{11}H_{12}O_2N_2S$	55	236.3	240.3	55.91	56.09	5.12	5.47	11.86	11.71
4,6-Dimethyl-	Acetic	247–249	$C_{11}H_{12}O_2N_2S$	55	236.3	231.7	55.91	56.01	5.12	4.92	11.86	11.85
4,6-Dimethyl-	α-Propionic	160–161	$C_{12}H_{14}O_2N_2S$	64	250.3	248.2	57.58	57.68	5.64	5.99	11.19	10.98
5,6-Dimethyl-	Acetic	207–209	$C_{11}H_{12}O_2N_2S$	85	236.3	234.9	55.91	55.74	5.12	5.33	11.86	12.11
5,6-Dimethyl-	α-Propionic	208–210	$C_{12}H_{14}O_2N_2S$	64	250.3	253.7	57.58	57.97	5.64	5.91	11.19	11.32
5-Phenyl-	Acetic	215–216	$C_{15}H_{12}O_2N_2S$	74	284.3	282.8	63.36	63.40	4.25	4.21	9.85	9.79
5-Phenyl-	α-Propionic	200–202	$C_{16}H_{14}O_2N_2S$	87	298.4	299.4	64.41	64.16	4.73	4.52	9.39	9.35

[a] The melting points were determined on a Fisher–Johns melting point block and corrected for calibration of the instrument. [b] Yields are calculated on the basis of the once-recrystallized acid.

2-Nitro-4-methoxyaniline, 4-amino-3-nitrobiphenyl, 4-methyl-2-nitroaniline and 2,4-dimethyl-6-nitroaniline, respectively, by reduction with stannous chloride in concentrated hydrochloric acid. 1,3-Dichloro-2,5-diaminobenzene,[13] 4-bromo-1,2-diaminobenzene[14] and 4,5-dimethyl-1,2-diaminobenzene[15] were synthesized from 2,4-dichloroaniline, *m*-bromoaniline and 3,4-dimethylaniline, respectively, by nitrating the acetylated aniline derivative with concentrated nitric acid and reducing the nitro compound after hydrolysis with stannous chloride. 1,2-Dichloro-4,5-diaminobenzene[16] was made from 1,2-dichloro-2-nitrobenzene by nitration and reduction of the dinitro compound[17] with mossy tin in concentrated hydrochloric acid. 1,2,3-Trichloro-5,6-diaminobenzene was prepared by the reduction of 1,2,3-trichloro-5,6-dinitrobenzene[18] which in turn was synthesized by the nitration of 1,2,3-trichloro-5-nitrobenzene.[19]

EAST LANSING, MICHIGAN

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[A CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE UNIVERSITY OF PENNSYLVANIA]

Metabolite Analogs. VI. Preparation of Some Analogs of 4-Amino-5-imidazolecarboxamide[1]

BY JOHN R. E. HOOVER[2] AND ALLAN R. DAY

RECEIVED JULY 5, 1956

1,2,3-Triazole analogs of the imidazole intermediates of *de novo* purine synthesis have been prepared as potential modifiers of nucleic acid metabolism. The 1-benzyl-1,2,3-triazoles were formed readily by condensing benzyl azide with esters and nitriles containing an active methylene group and the benzyl group was removed by reduction with sodium in liquid ammonia.

The use of drugs which affect nucleic acid metabolism, especially certain analogs of the natural purines, has been one of the more fertile fields of cancer chemotherapy to date. In view of this, an investigation of the possibility of interfering with purine metabolism by the use of analogs of the recently elucidated natural purine precursors,[3] especially of

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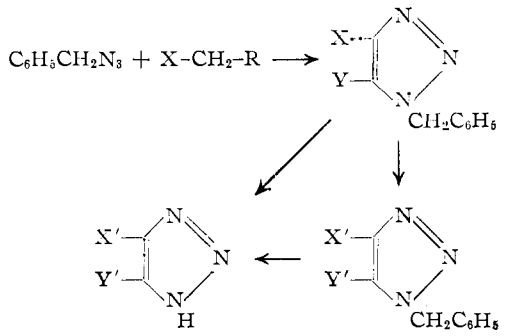
(3) W. Shive, W. W. Ackerman, M. Gordon, M. E. Getzendaner and R. E. Eakin, THIS JOURNAL, **69**, 725 (1947); S. C. Hartman, B. Levenberg and J. M. Buchanan, *ibid.*, **77**, 501 (1955); B. Levenberg and J. M. Buchanan, *ibid.*, **78**, 504 (1956); J. S. Gots, *Nature*, **172**, 256 (1953); G. R. Greenberg and E. L. Spilman, *J. Biol. Chem.*, **219**, 411 (1956).

5-amino-4-imidazolecarboxamide and 5-aminoimidazole, appearing interesting. Such analogs might display useful activity in one of several ways. The *in vivo* synthesis of purines might be blocked by direct competitive inhibition of the purine precursors; the analogs might be converted *in vivo* to unnatural purines or purine-like compounds which could compete with the natural purines in their incorporation into the nucleic acids; or, if incorporation of the modified purine occurred, this may block the ability of the nucleic acid to reproduce itself.

The initial approach to this study has been the preparation of a number of 1,2,3-triazoles with the 1-position free and with substituents on the 4- and 5-positions similar to those on the natural intermediates.

A large proportion of the simple vicinal triazoles with substituents of the type desired on the 4- and 5-positions which are reported in the literature also contain substituents on the 1-position in the form of an aromatic or similar grouping. The works of Dimroth and co-workers[4] describe the preparation of a number of triazoles from aromatic azides and active methylene compounds (esters, nitriles, etc.) in the presence of sodium alkoxide. The resulting 1-aryl derivatives cannot be converted, in most cases, to the corresponding parent homolog without disrupting other portions of the molecule. By using benzyl azide in place of the aromatic azides, 1-benzyl derivatives have been obtained which could be reductively debenzylated with sodium in liquid ammonia. Debenzylation by this method has been successfully applied to the imidazole system by Jones[5] and this technique was also recently reported by Wiley[6] for preparing 1H,1,2,3-triazole, starting with acetylenedicarboxylic acid and benzyl azide, while the work described here was nearing its completion.

Benzyl azide reacted in the presence of alkoxide in alcoholic solution with ethyl cyanoacetate, cyanoacetamide and cyanoacetic acid to give the corresponding 1-benzyl-4-substituted-5-amino-1,2,3-triazoles as indicated in the reaction scheme



R = (a) -CN, (b) $-COOC_2H_5$
X = C_2H_5OCO-, NH_2CO-, NC–, $C_2H_5O(C{=}NH)-$, HOCO–
X′ = X, NH_2CS-, NH_2NHCO-, H–
Y = (a) $-NH_2$, (b) -OH
Y′ = Y, Cl–, HS–

The placement of the benzyl group on the nitrogen atom is based on the analogous work of Dimroth and of others who used azides other than benzyl azide. In general, the yields were relatively low (20 to 81%) for all of the condensations. Malononitrile gave a mixture of the 4-cyano derivative along with the 4-carbiminoethoxy compound resulting from reaction of the nitrile with the solvent. Both of these derivatives gave 4-cyano-5-amino-1,2,3,-triazole on debenzylation under the conditions employed. The 4-cyano compounds reacted with hydrogen sulfide in ethanol to give the corresponding thioamides.

Ethyl malonate reacted with benzyl azide under somewhat more vigorous conditions than those employed for the cyano compounds to give the expected 1-benzyl-4-carbethoxy-5-hydroxy-1,2,3-triazole, again in rather low yields. The triazole esters served as routes for preparing the corresponding 4-carboxamides and the 4-acid hydrazides, although it proved to be more convenient to prepare the 5-amino-4-carboxamide from cyanoacetamide and benzyl azide and the 5-hydroxy-4-carboxamide from malononitrile and phenyl azide.[7] It is interesting that the 4-amino and 4-hydroxy esters were resistant to hydrolysis and ammonolysis, requiring 24 and 72 hours, respectively, in ethylene glycol at 100° for accomplishing the latter, while the 4-mercapto ester was hydrolyzed, even upon an attempted purification by solution in dilute sodium hydroxide and acidification.

1-Benzyl-5-amino-1,2,3-triazole-4-carboxylic acid readily decarboxylated on heating with dimethylaniline and served as a convenient route to 5(4)-amino-1,2,3-triazole hydrochloride.

A number of unsuccessful attempts were made to convert 1-benzyl-4-carbethoxy-5-hydroxy-1,2,3-triazole directly to the 5-mercapto compound by treatment with phosphorus pentasulfide under a variety of conditions. The mercapto group was finally introduced by treating the 5-hydroxy compound with phosphorus pentachloride and allowing the resulting 5-chloro derivative to react with sodium hydrosulfide in absolute alcohol. Ammonolysis of the ester grouping and debenzylation of the resulting amide were accomplished readily by the usual procedures.

All of the triazoles were white, or nearly white, crystalline compounds, the melting points and other physical properties being dependent upon the groupings attached to the ring. The 5-hydroxy derivatives were highly enolic, readily dissolving in dilute alkali and giving a color with ferric chloride solution. Although all of the 5-amino compounds gave colored products when treated with nitrous acid followed by a suitable coupling agent, they failed to form picrates and, with the exception of 5-amino-1,2,3-triazole and its 1-benzyl derivative, they did not give hydrochlorides.

The ultraviolet spectra of the 4-substituted-5-amino-1,2,3-triazoles show a fairly remarkable constancy regardless of the groupings attached to the 1- and 4-positions. All of the compounds stud-

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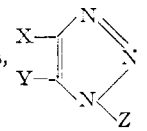
(7) O. Dimroth, E. Merckle and G. Fester, *Ann.*, **373**, 356 (1910). In this reaction, the 1-H compound was obtained directly although an aromatic azide was employed.

ied displayed two maxima, one around 230 mμ and the other around 265 mμ. The thioamides displayed a third strong band around 310 mμ. The absence of spectral data in the literature concerning this system makes it impossible to draw correlations for a group of this size. Attention can be directed to a few effects. All of the 1-benzyl compounds were unaffected by acid or alkaline media. The spectra of 1H compounds were unchanged by acid but, since this hydrogen can be replaced by metal ions, the spectra in alkaline media showed characteristic shifts. 5-Amino-1,2,3-triazole displayed only one maximum in the ultraviolet. The maximum was shifted in both acid and alkaline media, the compound being amphoteric. The 1-benzyl derivative, as would be expected, displayed a shift only under acidic conditions.

The three 5-hydroxy-1,2,3-triazoles studied also displayed two distinct maxima in the ultraviolet but, in contrast to the 5-amino compounds, the spectra were the same in neutral or alkaline media whether the substituent on the 1-position was hydrogen or benzyl, while in acid media a sharp shift occurred in both maxima. This might indicate high enolic character in media other than acidic. All of the spectral data are summarized in Table I.[8]

TABLE I

ULTRAVIOLET SPECTRA AND MELTING POINTS OF 5-AMINO- AND 5-HYDROXY-1,2,3-TRIAZOLES,



X	Y	Z	M.p., °C.	Ultraviolet max. (log ε)[a]	
EtOCO–	NH_2–	$C_6H_5CH_2$–	153	231 (3.83)	262 (3.91)
NH_2CO–	NH_2–	$C_6H_5CH_2$–	234	230 (3.89)	261 (3.93)
HOCO–	NH_2–	$C_6H_5CH_2$–	180	228 (3.83)	257 (3.87)
N≡C–	NH_2–	$C_6H_5CH_2$–	176	225 (3.93)	250 (3.86)
NH_2NHCO–	NH_2–	$C_6H_5CH_2$–	195	231 (3.92)	262 (3.91)
(acid soln.)				230s(3.86)	281 (4.10)
NH_2CO–	NH_2–	H–	226	226 (3.97)	260 (3.95)
(alkaline soln.)				223 (3.76)	264 (3.98)
NH_2NHCO–	NH_2–	H–	232	227 (3.89)	262 (3.83)
(alkaline soln.)				227i(3.6)	264 (3.87)
(acid soln.)				227 (3.81)	264 (3.83)
NH_2CS–	NH_2–	$C_6H_5CH_2$–[c]	228	244 (3.72)	278 (3.91)
NH_2CS–	NH_2–	H–[d,e]	210	234s(3.68)	272 (3.87)
(alkaline soln.)				252 (3.73)	277 (3.82)
H–	NH_2–	$C_6H_5CH_2$–	128	242 (3.60)	
(acid soln.)				254 (3.58)	
H–	NH_2–	H–[b]	136	239 (3.69)	
(acid soln.)				245 (3.50)	
(alkaline soln.)				228 (3.73)	
EtOCO–	HO–	$C_6H_5CH_2$–	113	228 (3.73)	274 (4.04)
(acid soln.)				230s(3.81)	293 (3.56)
NH_2CO–	HO–	$C_6H_5CH_2$–	175	227 (3.66)	272 (4.03)
(acid soln.)				224i(3.8)	290 (3.83)
NH_2CO–	HO–	H–	196	224 (3.68)	267 (3.84)
(acid soln.)				240 (3.66)	284s(2.7)

[a] 50% ethanol solution. [b] Hydrochloride in neutral and bicarbonate solution. [c] Also 312 (4.09). [d] Also 311 (4.00). [e] Also 321 (3.94).

Experimental[9]

Spectra.—The ultraviolet spectra were obtained using 1×10^{-4} or 5×10^{-5} molar solutions of the compound in 50% ethanol, the curves being obtained on a Beckman model DU spectrophotometer equipped with a Process and Instruments recording attachment. The curves in acid media were obtained by adding one drop of 2 *N* hydrochloric acid to 10 ml. of the above solutions; those in alkaline media by adding one drop of 0.1 *N* sodium hydroxide. The infrared spectra, when cited, were obtained on a Perkin–Elmer model 21 spectrophotometer using the potassium bromide disc technique.

1-Benzyl-4-carbethoxy-5-amino-1,2,3-triazole.—A solution of 13.3 g. (0.1 mole) of benzyl azide[10] and 11.3 g. (0.1 mole) of ethyl cyanoacetate in 250 ml. of absolute alcohol containing 0.1 mole of sodium ethoxide was heated under reflux for three hours. After cooling, the reaction mixture was poured into five volumes of water and cooled overnight. The resulting semi-crystalline mass was removed, washed with water and dried *in vacuo;* yield 6.15 g. (25%), m.p. 148–152°. This was extracted with 60 ml. of ether and the residue was purified by dissolving it in chloroform at room temperature and precipitating it with petroleum ether (30–60°) giving 3.98 g., m.p. 152–154°.

Anal. Calcd. for $C_{12}H_{14}N_4O_2$: C, 58.52; H, 5.73; N, 22.75. Found: C, 58.53; H, 6.00; N, 22.72.

1-Benzyl-4-carboxamido-5-amino-1,2,3-triazole. (a) **By Ammonolysis of 1-Benzyl-4-carbethoxy-5-amino-1,2,3-triazole.**—The ester was extremely resistant to the action of ammonia in alcoholic solution (no reaction in 16 hours at 100°). The use of aqueous alcoholic solutions resulted in evil smelling by-products. Preparation of the amide was accomplished by heating a solution of 4.92 g. (0.02 mole) of the carbethoxy compound for 24 hours in 40 ml. of ethylene glycol previously saturated with ammonia at 5°. The reaction was carried out at 100° in a micro autoclave. Three volumes of water were added to the reaction mixture and, after cooling, 2.91 g. (67%), m.p. 224–229°, of white crystals were removed. Recrystallization from alcohol gave 2.10 g. of crystals melting at 233–235°.

Anal. Calcd. for $C_{10}H_{11}N_5O$: C, 55.30; H, 5.10; N, 33.25. Found: C, 55.51; H, 5.28; N, 32.23.

(b) **From Cyanoacetamide.**—Equimolar portions of sodium (4.6 g.), cyanoacetamide (16.82 g.) and benzyl azide (26.6 g.) were dissolved in turn in 500 ml. of absolute alcohol and the mixture was heated under reflux for one hour. On cooling, 35.0 g. (81%) of nearly white crystals separated, m.p. 230–232°. According to mixed melting points and infrared spectra, the product was identical with that prepared by ammonolysis of the ester.

4(5)-Carboxamido-5(4)-amino-1,2,3-triazole.—1-Benzyl-4-carboxamido-5-amino-1,2,3-triazole (10.85 g., 0.05 mole) was suspended in liquid ammonia (100 ml.) and small pieces of sodium were added, with stirring, until a permanent blue color was produced. The color was discharged with a small amount of ammonium chloride and the solvent was allowed to evaporate. The toluene formed in the reaction was removed under vacuum. The white sodium salt which remained was dissolved in a minimal amount of water (very alkaline solution) and, after decolorization with a little charcoal, the solution was adjusted to *p*H 1 with concentrated hydrochloric acid. On cooling, white needles (4.20 g., 66%) separated, m.p., 222–224°. Repeated recrystallization from water, in which the compound is readily soluble, raised the melting point to 224–225°.

Anal. Calcd. for $C_3H_5N_5O$: C, 28.34; H, 3.97; N, 55.11. Found: C, 28.34; H, 3.87; N, 55.20.

1-Benzyl-5-amino-1,2,3-triazole-4-carboxylic Acid Hydrazide.—A suspension of 4.52 g. (0.02 mole) of 1-benzyl-4-carbethoxy-5-amino-1,2,3-triazole in 50 ml. of hydrazine hydrate was heated on the steam-bath for four hours. On cooling, 4.50 g. (97%) of product separated in the form of white plates, m.p. 193–195°. The material was washed with absolute alcohol, then with ether and dried. Recrystallization from water gave white plates melting at 194–195°; sparingly soluble in hot water, very sparingly soluble in cold water and alcohol, moderately soluble in warm 50% alcohol.

Anal. Calcd. for $C_{10}H_{12}N_6O$: C, 51.72; H, 5.21; N, 36.20. Found: C, 51.59; H, 5.01; N, 36.22.

(8) The ultraviolet spectra of the compounds discussed are available from the Chief, Photoduplication Service, ADI Auxiliary Publication Project, Library of Congress, Washington 25, D. C., by ordering the Document and remitting in advance $1.25 for microfilm or $1.25 for 8 × 10 photoprints, by check or money order payable to the Chief.

(9) The melting points are uncorrected. Most of the microanalyses were performed by Dr. Harry W. Galbraith, Knoxville, Tennessee.

(10) Reference 6, see also T. Curtius and G. Ehrhart, *Ber.* **55**, 1559 (1922).

5(4)-Amino-1,2,3-triazole-4(5)-carboxylic Acid Hydrazide.—Reduction of 9.28 g. (0.04 mole) of the 1-benzyl derivative was accomplished with sodium and liquid ammonia as described previously. Isolation of the product was accomplished by adjusting the aqueous solution of the sodium salt to *p*H 6 with hydrochloric acid whereupon the material separated; yield 4.95 g. (86%), m.p. 226° (with decomposition). Recrystallization from hot water, in which it is readily soluble, gave white needles, m.p. 232° (with decomposition). This compound was very soluble in dilute sodium hydroxide and dilute hydrochloric acid.

Anal. Calcd. for $C_3H_6N_6O$: C, 25.36; H, 4.26; N, 59.14. Found: C, 25.44; H, 4.25; N, 59.07.

1-Benzyl-5-amino-1,2,3-triazole-4-carboxylic Acid.—A mixture of 42.52 g. (0.5 mole) of cyanoacetic acid and 66.50 g. (0.5 mole) of benzyl azide in 1250 ml. of absolute alcohol containing 1.0 mole of sodium was heated under reflux for four hours (heating for 8 and 16 hours did not improve the yield). The mixture was poured into two liters of ice-water and hydrochloric acid was added to *p*H 1. On cooling, 21.25 g. (19.5%) of white needles separated (m.p., 179–180°, with decomposition), which were only slightly soluble in 2 *N* sodium hydroxide, nearly insoluble in 2 *N* hydrochloric acid, water, hot alcohol and hot dioxane. Purification was accomplished by dissolving the material in concentrated hydrochloric acid and precipitating it with water, followed by solution in dimethylformamide and precipitation by adding water.

Anal. Calcd. for $C_{10}H_{10}N_4O_2$: C, 55.04; H, 4.62; N, 25.68. Found: C, 55.22; H, 4.72; N, 25.60.

1-Benzyl-5-amino-1,2,3-triazole.—A suspension of 16.35 g. (0.075 mole) of 1-benzyl-5-amino-1,2,3-triazole-4-carboxylic acid in 75 ml. of dimethylaniline was heated under reflux for 15 minutes with copious evolution of carbon dioxide. Cooling the reaction mixture to 5° gave 7.87 g. (60%) of white needles melting at 127–128°. This material was very soluble in absolute alcohol, soluble in chloroform, slightly soluble in hot water. It did not form a picrate but was soluble in dilute hydrochloric acid. For analytical purposes, it was dissolved in chloroform and reprecipitated with petroleum ether (30–60°). The melting point remained unchanged.

Anal. Calcd. for $C_9H_{10}N_4$: C, 62.04; H, 5.79; N, 32.17. Found: C, 62.28; H, 5.64; N, 32.10.

4(5)-Amino-1,2,3-triazole Hydrochloride.—The residue from the reduction of 6.97 g. (0.04 mole) of the 1-benzyl derivative with sodium in liquid ammonia was dissolved in 10 ml. of water. A few drops of 2 *N* sodium hydroxide was added and 0.45 g. of starting material was removed (m.p. 127–129°). The filtrate was adjusted to *p*H 7, placed in a liquid–liquid extraction apparatus and extracted for six hours with ethyl acetate. The ethyl acetate solution, after drying over calcium chloride, was saturated with dry hydrogen chloride giving 1.78 g. (37%) of crystals, m.p. 134–136°. After solution in a small amount of absolute alcohol and precipitation with ether, 1.45 g. of white crystals, m.p. 139° (with decomposition), were obtained. The product did not form a picrate.

Anal. Calcd. for $C_2H_5N_4Cl$: C, 19.94; H, 4.27; N, 46.49; Cl, 29.43. Found: C, 20.27; H, 4.46; N, 46.39; Cl, 29.20.

1-Benzyl-4-cyano-5-amino-1,2,3-triazole and 1-Benzyl-4-carbiminoethoxy-5-amino-1,2,3-triazole.—A mixture of 19.83 g. (0.3 mole) of malononitrile, 39.9 g. of benzyl azide and 500 ml. of absolute alcohol containing 6.9 g. of sodium was allowed to stand at room temperature for 20 hours. This was poured into two liters of ice-water and stirred for approximately three hours until the oil which originally separated had solidified. The solid material was removed, washed with cold water and dried *in vacuo*. The dried material was extracted with 250 ml. of petroleum ether and again dried giving 30.46 g. of white solid which melted over a wide range.

This mixture was stirred for 30 minutes with 250 ml. of 2 *N* hydrochloric acid and 17.94 g. of solid residue was removed (m.p. 161–204°). Neutralization of the filtrate with solid sodium carbonate gave an oil which solidified on cooling overnight; yield of crude 1-benzyl-4-carbiminoethoxy-5-amino-1,2,3-triazole, 11.12 g. (15%), m.p. 100–109°. Recrystallization from chloroform–petroleum ether gave 9.57 g. of crystals, m.p. 115–117°. This material displayed three sharp bands in the N–H stretching region of the infrared spectrum at 2.90, 3.02 and 3.15 μ and none in the nitrile or carbonyl regions.

Anal. Calcd. for $C_{12}H_{15}N_5O$: C, 58.75; H, 6.16; N, 28.55. Found: C, 59.03; H, 5.95; N, 28.72.

The 17.94-g. fraction consisted of a mixture of the 4-cyano-5-amino derivative and a higher melting material. It was extracted at room temperature with 150 ml. of dioxane in two portions, leaving 4.94 g. of residue with a melting point of 205–218°. Addition of 600 ml. of petroleum ether to the dioxane solution gave 9.96 g. of white solid, the main portion melting from 170–175°, with contamination by a small amount of the higher melting material. The 1-benzyl-4-cyano-5-amino-1,2,3-triazole was isolated in a pure state by extracting the dioxane-soluble material with hot chloroform and adding petroleum ether to the cooled filtrate. After repeating this operation, the product melted at 175–176°. It displayed two bands in the N–H region of the infrared at 2.97 and 3.10 μ and a sharp band at 4.45 μ (nitrile).

Anal. Calcd. for $C_{10}H_9N_5$: C, 60.29; H, 4.55; N, 35.17. Found: C, 60.57; H, 4.37; N, 35.13.

4(5)-Cyano-5(4)-amino-1,2,3-triazole.—The debenzylation was carried out as in the previous cases. The residue, after evaporation of the ammonia, was dissolved in a small amount of 2 *N* hydrochloric acid and the solution was extracted repeatedly with ether. Evaporation of the ether gave tan crystals (yield 50%), m.p. 225–227°. Several recrystallizations by dissolving the material in absolute alcohol and adding petroleum ether resulted in a product which melted at 226–228° (with decomposition).

Anal. Calcd. for $C_3H_3N_5$: C, 33.02; H, 2.77; N, 64.21. Found: C, 33.07; H, 2.84; N, 64.14.

This material displayed sharp absorption bands in the infrared at 2.97, 3.12, 3.31, 3.50, 4.46, 6.03 and 6.18 μ (potassium bromide disc). The 1-benzyl-4-carbiminoethoxy-5-amino-1,2,3-triazole gave products with identical spectra and melting points when debenzylation was carried out according to the above procedure.

1-Benzyl-4-thiocarboxamido-5-amino-1,2,3-triazole.—To 50 ml. of absolute alcohol saturated first with ammonia and then with hydrogen sulfide, was added 3 g. (0.015 mole) of 1-benzyl-4-cyano-5-amino-1,2,3-triazole and the mixture was heated under reflux for 15 minutes. An additional 50 ml. of absolute alcohol, saturated as above, was then added and the heating was continued for one hour. The mixture was concentrated *in vacuo* to a volume of 50 ml. and the solution cooled giving 1.69 g. (48%) of white crystals, m.p. 221–224°. Recrystallization from 95% alcohol raised the melting point to 227–229°.

Anal. Calcd. for $C_{10}H_{11}N_5S$: C, 51.47; H, 4.75; N, 30.02; S, 13.75. Found: C, 51.45; H, 4.87; N, 30.02; S, 13.53.

4(5)-Thiocarboxamido-5(4)-amino-1,2,3-triazole.—More vigorous conditions than those used on the 1-benzyl derivative were required for the 1H compound. A mixture of 0.81 g. (0.0075 mole) of 4-cyano-5-amino-1,2,3-triazole in 20 ml. of cold absolute alcohol saturated as described previously was heated in a micro autoclave at 100° for three hours. The solvent was removed *in vacuo* and 15 ml. of water was added to the residue. The sirup crystallized giving 0.56 g. (52%) of product, m.p. 310° (with decomposition). This was recrystallized one time from approximately ten ml. of hot water and was then purified further by dissolving it in a minimal amount of hot absolute alcohol and adding petroleum ether to the cooled solution. Final m.p. 310–314° (with decomposition).

Anal. Calcd. for $C_3H_5N_5S$: C, 25.16; H, 3.52; N, 48.91. Found: C, 25.72; H, 3.47; N, 48.82.

1-Benzyl-4-carbethoxy-5-hydroxy-1,2,3-triazole.—The condensation was carried out under reflux for six hours using 32.04 g. of ethyl cyanoacetate, 26.6 g. of benzyl azide and 4.6 g. (0.2 mole each) of sodium methoxide in 500 ml. of absolute alcohol. The product was obtained by pouring the reaction mixture into 1.5 liters of ice-water and removing the organic layer which separated from the alkaline mixture. The aqueous solution was made strongly acid and cooled overnight. The white crystals so obtained were washed on the funnel, first with water and then with petroleum ether. The crude product, after drying at 50° (30.0 g.,

m.p. 110–112°, yield 48%), was dissolved in chloroform and crystallization was induced by adding petroleum ether; yield, after purification, 18.3 g., m.p. 111–112°. This material was readily soluble in dioxane and chloroform, moderately soluble in alcohol and hot water and gave a color with ferric chloride solution.

Anal. Calcd. for $C_{12}H_{13}N_3O_3$: C, 58.30; H, 5.30; N, 16.98. Found: C, 58.62; H, 5.22; N, 17.03.

1-Benzyl-4-carboxamido-5-hydroxyl-1,2,3-triazole.—The ammonolysis of the 4-carbethoxy-5-hydroxy derivative under the conditions employed for 1-benzyl-4-carbethoxy-5-amino-1,2,3-triazole required 72 hours for completion. From 24.72 g. (0.1 mole) of starting material in 400 ml. of ammonia saturated ethylene glycol, 21.0 g. of white crystals melting at 136–147° were obtained after acidification of the aqueous mixture. Recrystallization from 95% alcohol gave 16.8 g. of product (77%) melting at 165–170°. Several additional recrystallizations brought the melting point to 174–175° (with decomposition).

Anal. Calcd. for $C_{10}H_{10}N_4O_2$: C, 55.04; H, 4.62; N, 25.68. Found: C, 55.05; H, 4.61; N, 25.52.

The product from debenzylation of the above material with sodium and liquid ammonia proved difficult to purify. It was found to be more convenient to prepare the 4-carboxamido-5-hydroxy-1,2,3-triazole by the procedure of Dimroth[7] from malonamide and phenyl azide which gave the compound in a 51 per cent. yield, m.p. 196°. This reaction could not be accomplished using benzyl azide in place of the phenyl azide.

1-Benzyl-4-carbethoxy-5-chloro-1,2,3-triazole.—After heating 49.44 g. (0.2 mole) of 1-benzyl-4-carbethoxy-5-hydroxy-1,2,3-triazole, 41.66 g. of phosphorus pentachloride and 45 ml. of phosphorus oxychloride on the steam-bath for one hour, the phosphorus oxychloride was removed at 90° *in vacuo* (water aspirator). The oily residue was placed over potassium hydroxide in a vacuum desiccator overnight, then taken up in a little warm absolute alcohol, seeded and cooled in a freezer. Large white crystals (23.6 g., 44%), melting at 67–68°, were obtained after washing with cold alcohol, then with ether and drying. Further recrystallization from 95% alcohol did not alter the melting point.

Anal. Calcd. for $C_{12}H_{12}N_3O_2Cl$: C, 54.24; H, 4.55; N, 15.81; Cl, 13.35. Found: C, 54.28; H, 4.63; N, 15.66; Cl, 13.40.

1-Benzyl-4-carbethoxy-5-mercapto-1,2,3-triazole Sodium Salt.—Five-hundred ml. of absolute alcohol containing 5.40 g. (0.1 mole) of sodium methoxide was saturated with hydrogen sulfide. After adding 26.56 g. of 1-benzyl-4-carbethoxy-5-chloro-1,2,3-triazole, the solution was heated under reflux for 24 hours. The mixture was concentrated *in vacuo* to a volume of 200 ml. and the inorganic precipitate was removed. Concentration was continued to a final volume of 100 ml. and six volumes of ether were added giving 8.61 g. of white crystals, m.p. 241–242° (with decomposition). The filtrate was evaporated to dryness *in vacuo* and the residue was stirred with 400 ml. of ether. An additional 0.61 g. of product with the same melting point was removed; total yield 9.22 g. (32%). Recrystallization from isopropyl alcohol gave a product melting at 248–249° (with decomposition).

Anal. Calcd. for $C_{12}H_{12}N_3O_2SNa$: C, 50.51; H, 4.24; N, 14.73. Found: C, 50.74; H, 4.65; N, 14.69.

The ether filtrate was evaporated to dryness and the resulting sirup was dissolved in a small amount of warm 95% alcohol. The solution was cooled giving 12.1 g. (46%) of starting material, m.p. 62–64°. The above reaction, when run for four hours at 100° in a sealed tube, resulted in only about 25% conversion of the starting material. A reaction run at 100° for 12 hours gave only a 10% yield of the desired product along with a water and dilute alkali-insoluble material melting at 116–118°.

1-Benzyl-4-carboxy-5-mercapto-1,2,3-triazole.—The ester was converted in a nearly quantitative yield to the acid, m.p. 123° (with bubbling), by brief warming with dilute alkali followed by acidification.

Anal. Calcd. for $C_{10}H_9N_3O_2S$: C, 51.04; H, 3.85; N, 17.86. Found: C, 50.95; H, 3.68; N, 17.87.

1-Benzyl-4-carboxamido-5-mercapto-1,2,3-triazole.—The ammonolysis of 11.40 g. (0.04 mole) of 1-benzyl-4-carbethoxy-5-mercapto-1,2,3-triazole sodium salt was accomplished in nearly quantitative yield by heating at 100° for 15 hours in 50 ml. of ethylene glycol saturated with ammonia at 0°, yield 9.27 g. (99%), m.p. 205–206° (with bubbling, sintering at 199°). Recrystallization from approximately 1.5 liters of 95% alcohol gave 7.41 g. of cream colored crystals with the same melting point.

Anal. Calcd. for $C_{10}H_{10}N_4OS$: C, 51.26; H, 4.30; N, 23.92. Found: C, 51.33; H, 4.37; N, 23.98.

4(5)-Carboxamido-5(4)-mercapto-1,2,3-triazole.—The debenzylation of 7.03 g. (0.03 mole) of the 1-benzyl-5-mercapto derivative resulted in 2.81 g. (65%) of tan crystals melting at 194–195° (with decomposition). The decomposition point was somewhat dependent on the rate of heating. Recrystallization from hot water gave a white product with the same decomposition point.

Anal. Calcd. for $C_3H_4N_4OS$: C, 24.99; H, 2.80; N, 38.87. Found: C, 25.15; H, 2.78; N, 38.81.

PHILADELPHIA 1, PA.

[CONTRIBUTION FROM THE EASTERN REGIONAL RESEARCH LABORATORY[1]]

The Formation of 4-Carboxy-2-azetidinone from Asparagine in Phosphate Buffer

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The β-lactam, 4-carboxy-2-azetidinone, was synthesized from L- and DL-asparagine by cyclization in phosphate buffer of *p*H 6.7 at 100°. In addition to this compound, four compounds were formed which were ninhydrin positive.

While investigating analytical methods for use with the amides of potatoes, we had occasion to heat asparagine in phosphate buffer, as in the glutamine method described by Vickery and Pucher[3a] and Hamilton.[3b]

Although these investigators had found asparagine not to interfere to an appreciable extent in the glutamine determination, we found, by means of ion-exchange techniques, that definite reaction did occur in their time limit. From reaction mixtures resulting from longer periods of heating, we isolated a compound which was ninhydrin negative and Rydon–Smith[4] positive, indicating a secondary amide. This compound was similar in some properties to pyroglutamic acid produced from glutamine under the same conditions.

Efforts to identify the compound led to the conclusion that it was 4-carboxy-2-azetidinone (pyro-

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