[CONTRIBUTION FROM THE DEPT. OF CHEMISTRY, OREGON STATE COLLEGE]

PURINES V. THE PREPARATION OF CERTAIN 2,9-SUBSTITUTED PURINES AND AZAPURINES[1]

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Received September 6, 1954

Recently in connection with another study it was necessary to make a number of typical 9-substituted purines. The usual procedure for such preparations has been *via* alkylation of the corresponding purine. This method has the disadvantage in that it is limited in scope; furthermore the product may be a mixture of the 7 and 9 isomers or even a further alkylated product. Baddiley (1) avoided this problem in synthesizing a number of 9-substituted purines by refluxing 4-alkylamino-5-thioformamidopyrimidines in aqueous, pyridine or quinoline solutions in order to effect ring closure. Albert (2) has recently reported the synthesis of 9-methylpurine by cyclizing 4-methylamino-5-aminopyrimidine with formic acid. It was therefore of interest to this laboratory to study the general cyclization procedures employing both formamide (3) and nitrous acid to pyrimidines containing a secondary amino group in the 4 position. In order to determine the generality of this procedure, the 4-amino group was substituted with several different types of substituents. The treatment of the 4-substituted amino-5-aminopyrimidine intermediates with nitrous acid yields the previously unknown 3-substituted triazolopyrimidines.

The sequence of reactions for the preparation of the 2- and 9-substituted purines and their aza analogs is shown schematically in Figure 1. The reaction of 2,4-dichloro-5-nitropyrimidine with aniline yields 2,4-dianilino-5-nitropyrimidine (III R′, R = C_6H_5, Figure 1). Catalytic reduction of III gave 2,4-dianilino-5-aminopyrimidine (IV, R, R′ = C_6H_5). This compound was found to be extremely sensitive in alcoholic solutions to air oxidation; however, the diamine could be isolated as white needles by precipitating the compound out of aqueous alcohol. Recrystallization from carbon tetrachloride gave a white powder which gradually discolors upon standing in air. This diamine was readily formylated with formic acid to give 2,4-dianilino-5-formamidopyrimidine (V, R′, R = C_6H_5). The melting point of the product was difficult to reproduce due perhaps to the cyclization of the compound upon melting. On one determination a double melting point was obtained; the latter melting point was the same as the 2-anilino-9-phenylpurine (VI, R′, R = C_6H_5) obtained by cyclizing the formyl derivative with formamide.

3-Phenyl-5-anilino-*v*-triazolo[*d*]pyrimidine (VII, R′, R = C_6H_5, Figure 1) was prepared from 2,4-dianilino-5-aminopyrimidine by treatment with nitrous acid.

The reaction of 2,4-dichloro-5-nitropyrimidine with *n*-propylamine gave the expected 2,4-di-*n*-propylamino-5-nitropyrimidine (III, R′, R = *n*-C_3H_7 Fig. 1).

[1] This work was supported in part by grants from the Division of Research Grants and Fellowships, National Institutes of Health, Public Health Service. Published with the approval of the Monographs Publications Committee, Oregon State College, as Research Paper No. 263, School of Science, Department of Chemistry.

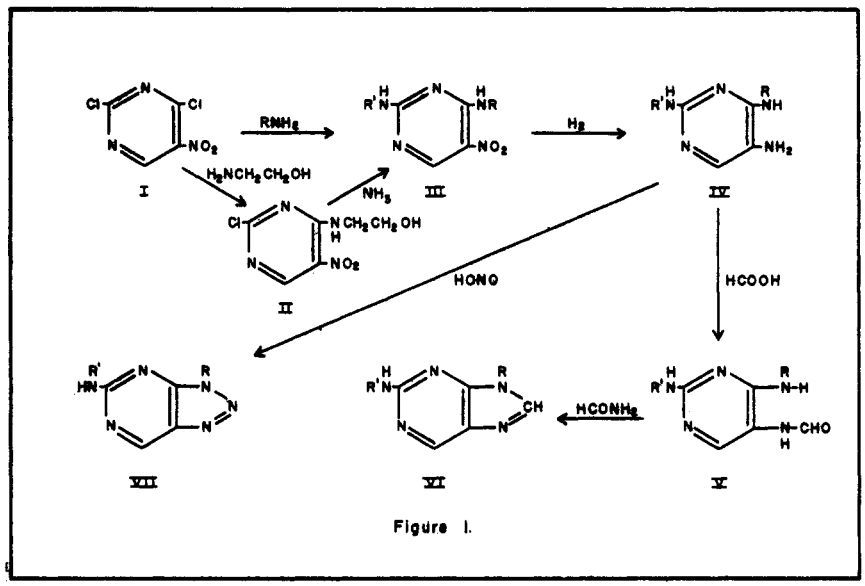

Figure 1.

Reduction of this compound gave a black gummy residue which failed to crystallize. This residue was converted to the sulfate salt and to the formyl derivative for identification. The formyl derivative (V) then was cyclized to the 2-*n*-propylamino-9-*n*-propylpurine (VI, R′, R = *n*-C_3H_7). The use of crude 2,4-di-*n*-propylamino-5-formamidopyrimidine in this reaction (in an attempt to improve the over-all yield) resulted in an oily product which could not be crystallized.

The corresponding aza compound, 3-*n*-propyl-5-*n*-propylamino-*v*-triazolo[*d*]pyrimidine (VII, R′, R = *n*-C_3H_7 Figure 1) was prepared by treating 2,4-di-*n*-propylamino-5-aminopyrimidine sulfate with nitrous acid.

The reaction of 2,4-dichloro-5-nitropyrimidine with ethanolamine according to the procedure of Ramappe (4) gave 2-chloro-4-(2′-hydroxyethylamino)-5-nitropyrimidine (II, Figure 1). Amination of this compound yielded 2-amino-4-(2′-hydroxyethylamino)-5-nitropyrimidine. Reduction of this compound gave the corresponding diamine which was labile in air. This diamine was converted to the sulfate salt which was shown by analysis to be impure; however, cyclization of this compound with nitrous acid gave a product from which pure 3-(2′-hydroxyethyl)-5-amino-*v*-triazolo[*d*]pyrimidine (VII, R′ = H, R = CH_2-CH_2OH) could be isolated.

The 2,5-diamino-4-(2′-hydroxyethylamino)pyrimidine was treated with formic acid which gave 2-amino-4-(2′-hydroxyethylamino)-5-formamidopyrimidine. Cyclization of this compound failed to give the desired purine. Analytical data showed it to be the formyl ester of the expected purine.

Since 6-mercaptopurine (5) was reported to have inhibitory properties, the preparation of the 2 isomer and of their aza analogs are of considerable biological interest. The preparation of 2-mercaptopurine has been previously

reported (3). The aza analogs on the other hand may easily be prepared as shown in Figure 2 from either 2,4-dichloro-5-nitropyrimidine (I, Figure 2) or 4,6-dichloro-5-nitropyrimidine. Since Bahner (6) and co-workers have recently prepared the 6-aza analog of 6-mercaptopurine from azahypoxanthine only the 2-aza analog was synthesized.

2,4-Dichloro-5-nitropyrimidine, prepared by the method of Whittaker (7), was mono aminated by a modification of Isay's method (8). This modification consisted essentially of the addition of alcoholic ammonium hydroxide to a cooled solution of the chloropyrimidine in ether.

The modification proposed by Albert (9) was also investigated; however, due to the difficult thermal control of the reaction some replacement of both the 2 and 4 chloro groups frequently occurs. Isay's method seemed to give better control over the reaction temperature—especially on larger runs.

Thionation and reduction of 2-chloro-4-amino-5-nitropyrimidine was accomplished by the method of Elion (10) yielding 2-mercapto-4,5-diaminopyrimidine (III, Figure 2). Treatment of this diamine with nitrous acid gave 5-mercapto-1*H*-*v*-triazolo[*d*]pyrimidine (IV, Figure 2). This compound was soluble in dilute ammonium hydroxide in contrast to the 5,7-dimercapto-1-*v*-triazolo-[*d*]pyrimidine (11) which was insoluble in cold base. Both compounds

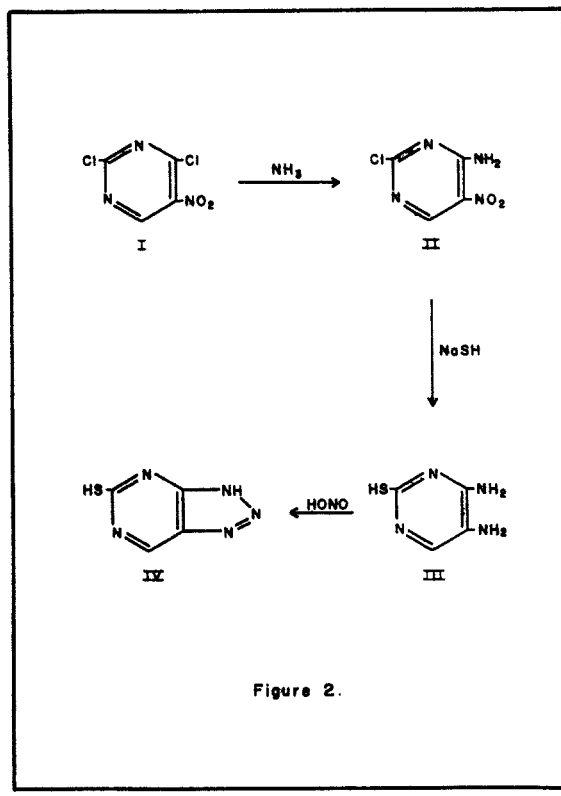

Figure 2.

exploded on heating which indicates this property may be characteristic of azapurines containing sulfur. This behavior made combustion analyses on a semi-micro basis difficult necessitating the use of micro analytical procedures.

EXPERIMENTAL

2,4-Dianilino-5-nitropyrimidine (III, R', R = C_6H_5, Figure 1). 2,4-Dichloro-5-nitropyrimidine (5.0 g.) was dissolved in 20 ml. of absolute ethanol and slowly added to a stirred solution of 10 g. of aniline dissolved in 200 ml. of absolute ethanol. A yellow precipitate formed immediately. The suspension was stirred under reflux for 45 minutes, cooled, filtered, and washed with alcohol and ether to yield 7.4 g. (94%) of yellow fluffy needles, m.p. 202–205°. Then 2.5 g. of this crude product was crystallized from 200 ml. of benzene to give 2.2 g. of light yellow, fluffy needles, m.p. 203–204°.

Anal. Calc'd for $C_{16}H_{13}N_5O_2$: N, 22.8. Found: N, 22.6.

2,4-Dianilino-5-aminopyrimidine (IV, R', R = C_6H_5, Figure 1). The crude 2,4-dianilino-5-nitropyrimidine (2 g.) was suspended in 150 ml. of absolute ethanol containing 2 g. of Raney nickel catalyst and hydrogenated (three to six hours). The catalyst was removed and the filtrate was evaporated to dryness; yield, 1.45 g. of purple crystals. These crystals were dissolved in 70 ml. of ethanol which gave a deep purple solution that yielded 1.25 g. (70%) of white crystals, m.p. 161–163° dec. upon dilution with 1 liter of water. A small amount recrystallized (for an analytical sample) from carbon tetrachloride gave a white fluffy powder, m.p. 165–168° dec.

Anal. Calc'd for $C_{16}H_{15}N_5$: C, 69.31; H, 5.41; N, 25.27.
Found: C, 69.0; H, 5.2; N, 25.0.

3-Phenyl-5-anilino-v-triazolo[d]pyrimidine (VII, R', R = C_6H_5, Figure 1). In 400 ml. of 5% acetic acid solution was placed 0.57 g. of crude 2,4-dianilino-5-aminopyrimidine. This solution was boiled, decolorized, and cooled to 10–20°. Upon the addition of 0.16 g. of sodium nitrite to the stirred solution an immediate white precipitate formed which gradually changes to a green to blue-green color. After stirring 15 minutes the mixture was brought to *p*H 8–9 with concentrated ammonium hydroxide. After cooling overnight 0.37 g. of (62%) green powder, (softened and melted 191–194° dec.) was obtained. This was decolorized and crystallized from 80 ml. of methanol twice to give 0.15 g. of light green needles, m.p. 195°.

Anal. Calc'd for $C_{16}H_{12}N_6$: C, 66.67; H, 4.17.
Found: C, 66.58; H, 4.28.

2,4-Dianilino-5-formamidopyrimidine (V, R', R = C_6H_5, Figure 1). Into a 30-ml. flask was placed 0.91 g. of crude 2,4-dianilino-5-aminopyrimidine and 10 ml. of (90%) formic acid; the solution was gently refluxed for 15 minutes. The excess formic acid then was removed by evaporation before a hot air fan and the resulting residue was dissolved in 5 ml. of water. After the solution was adjusted to *p*H 7–8 with concentrated ammonium hydroxide and cooled, the precipitate was removed by filtration, washed with water, and dried yielding 0.97 g. of grey powder, m.p. 183–185°. Crystallization from 50% alcohol-water gave white fluffy needles, m.p. 193.5–195°. The rate of heating had a marked influence on the melting point. A second determination gave m.p. 187–189°, resolidification and a second melting at 215°.

Anal. Calc'd for $C_{17}H_{15}N_5O$: C, 66.89; H, 4.92.
Found: C, 66.75; H, 4.90.

2-Anilino-9-phenylpurine (VI, R', R = C_6H_5, Figure 1). 2,4-Dianilino-5-formamidopyrimidine (1.0 g.) was gently boiled with 10 ml. of formamide for 15 minutes. At the end of this time a light brown solution was obtained which solidified on cooling. Then 10 ml. of water was added, and the solution was adjusted to *p*H 7–8 with ammonium hydroxide and cooled. The resultant precipitate was filtered, washed with water, and dried to give 0.86 g. (92%) pink needles, m.p. 214–215°. An analytical sample was prepared by crystallization and

decolorization from a 60% alcohol-water mixture. White fluffy needles, m.p. 215–216°, almost identical in appearance to the starting material were obtained.

Anal. Calc'd for $C_{17}H_{13}N_5$: C, 71.08; H, 4.53.

Found: C, 70.9; H, 4.46.

2,4-Di-n-propylamino-5-nitropyrimidine (III, R′, R = n-C_3-H_7, Figure 1). 2,4-Dichloro-5-nitropyrimidine (5.0 g.) dissolved in 20 ml. of ethanol was slowly added to a stirred solution of 8.5 ml. of n-propylamine in 100 ml. of ethanol and then refluxed 30 minutes. Upon cooling the resulting yellow solution, white needles slowly formed. After cooling, the needles were recovered by filtration, washed with water, and dried to yield 5.6 g. (91%), m.p. 120–121°. An analytical sample was prepared by crystallizing from an alcohol-water mixture which raised the melting point to 121–122°.

Anal. Calc'd for $C_{10}H_{17}N_5O_2$: C, 50.21; H, 7.12.

Found: C, 50.2; H, 7.26.

2,4-Di-n-propylamino-5-aminopyrimidine sulfate (IV, R′, R = n-C_3H_7, Figure 1). 2,4-Di-n-propylamino-5-nitropyrimidine (2.0 g.) was suspended with 2 g. of Raney nickel catalyst in 115 ml. of methanol and hydrogenated at 30 p.s.i. until the theoretical amount of hydrogen was taken up (2–3 hours). The catalyst was removed and the filtrate evaporated by means of a hot air fan leaving a black gummy residue which did not crystallize. This residue was decolorized, and converted to the sulfate salt by dissolving in 50 ml. of 5% sulfuric acid, boiling with charcoal, and filtering; on cooling the filtrate 1.84 g. (72%) of orange crystals was obtained. This crude sulfate salt was purified for analysis by recrystallization and repeated decolorizations from 5% sulfuric acid. The crystals were filtered, washed with water, and dried.

Anal. Calc'd for $C_{10}H_{19}N_5 \cdot H_2SO_4$: C, 39.09; H, 6.84.

Found: C, 39.26; H, 6.93.

3-n-Propyl-5-n-propylamino-v-triazolo [d] pyrimidine (VII, R′, R = n-C_3H_7, Figure 1). The crude 2,4-di-n-propylamino-5-aminopyrimidine sulfate (1.84 g.) was dissolved and decolorized from 200 ml. of water containing 2 drops of sulfuric acid. The solution was cooled to 10–20° and stirred while 0.55 g. of sodium nitrite was added. After stirring one minute a white precipitate formed. After 30 minutes the solution was adjusted to pH 7–8 with concentrated ammonium hydroxide after which the precipitate took on a pink cast. After cooling in a refrigerator the product was filtered and washed with water; yield 0.9 g., m.p. 97–98° (49% from the nitropyrimidine). An analytical sample was prepared by crystallizing and decolorizing from 50% methanol-water. Long white needles slowly formed, m.p. 97.5–98°.

Anal. Calc'd for $C_{10}H_{16}N_6$: C, 54.54; H, 7.27.

Found: C, 54.41; H, 7.26.

2,4-Di-n-propylamino-5-formamidopyrimidine (V, R′, R = n-C_3H_7, Figure 1). A suspension of 2.65 g. of 2,4-di-n-propylamino-5-nitropyrimidine and 3 g. of Raney nickel catalyst in 150 ml. of methanol was hydrogenated at 24 p.s.i. until the theoretical amount of hydrogen was absorbed (1–2 hours). The catalyst was removed and the resultant filtrate was evaporated yielding a black gummy residue. This was refluxed with 15 ml. of formic acid, 5 ml. of water was added and the solution was adjusted to pH 7–8 with concentrated ammonium hydroxide. An oil started to come out and finally oily crystals formed. After cooling, filtering, and washing with water, the brown precipitate was crystallized and decolorized from 100 ml. of an ethanol-water mixture (1:3) giving 1.5 g. of glistening brown platelets, m.p. 156–159°. Repeated recrystallization and decolorizations gave white shiny platelets, m.p. 159.5–160.5°.

Anal. Calc'd for $C_{11}H_{19}N_5O$: C, 55.70; H, 8.02.

Found: C, 56.0; H, 7.9.

2-n-Propylamino-9-n-propylpurine (VI, R′, R = n-C_3H_7, Figure 1). Into 10 ml. of formamide was placed 0.94 g. of purified 2,4-di-n-propylamino-5-formamidopyrimidine and the resulting solution was boiled gently for 15 minutes. The dark solution was filtered to

remove a little black insoluble material and then was cooled overnight. White crystals were filtered off and washed with water to yield 0.35 g. of white crystals, m.p. 82.5–83.5°. An analytical sample was prepared by recrystallization and decolorization from a 5:1 water-ethanol mixture. Shiny white crystals were filtered off, washed with water, and dried, m.p. 84–85°.

Anal. Calc'd for $C_{11}H_{17}N_5$: C, 60.27; H, 7.76.

Found: C, 60.12; H, 7.6.

2-Amino-4-(2'-hydroxyethylamino)-5-nitropyrimidine (III, R' = H, R = CH_2CH_2OH, Figure 1). 2-Chloro-4-(2'-hydroxyethylamino)-5-nitropyrimidine (4) (8.25 g.) was dissolved in 40 ml. of hot ethanol and added to 80 ml. of ethanol (95%) previously saturated with ammonia. The resulting solution was gently boiled under an atmosphere of ammonia. In a few minutes a white precipitate formed and after 30 minutes the mixture was cooled, filtered, washed with water to remove the ammonium chloride, and dried to give 6.95 g. (93%), m.p. 192–194°.

Anal. Calc'd for $C_6H_9N_5O_3$: C, 36.18; H, 4.52.

Found: C, 36.12; H, 4.41.

3-(2'-Hydroxyethyl)-5-amino-v-triazolo[d]pyrimidine (VII, R' = H, R = CH_2CH_2OH, Figure 1). 2-Amino-4-(2'-hydroxyethylamino)-5-nitropyrimidine (2.0 g.) and 3 g. of Raney nickel catalyst were suspended in 100 ml. of methanol and hydrogenated at 30 p.s.i. until the theoretical amount of hydrogen was absorbed (1–2 hours). The catalyst was removed yielding a grayish filtrate which gradually darkened on exposure to air. Concentrated sulfuric acid was added to the stirred solution until it was adjusted to *pH* 1. After cooling, white crystals were recovered by filtration, washed with a little methanol, and dried to give 1.46 g., m.p. 169–170°. A stirred solution containing 0.75 g. of this sulfate, 10 ml. of water, and 1 drop of concentrated sulfuric acid was cooled to 10–20° and then 0.3 g. of sodium nitrite was added. In a few seconds a fine white precipitate formed. After 30 minutes the solution was adjusted to *p*H 8–9 with concentrated ammonium hydroxide and then cooled. The white crystals were washed with water and dried; yield 0.33 g. (65%), m.p. 219.5–220.5°. Crystallization from water for analytical purposes raised the melting point to 220–221°.

Anal. Calc'd for $C_6H_8N_6O$: C, 40.00; H, 4.44.

Found: C, 39.8; H, 4.40.

2,5-Diamino-4-(2'-hydroxyethylamino)pyrimidine (IV R' = H, R = CH_2CH_2OH, Figure 1). 2-Amino-4-(2'-hydroxyethylamino)-5-nitropyrimidine (4.0 g.) and 5 g. of Raney nickel catalyst were suspended in 200 ml. methanol and hydrogenated until the theoretical amount of hydrogen was absorbed. The catalyst was recovered and the filtrate was concentrated to 30 ml. The crude diamine was precipitated by the addition of ether and cooling. The dried precipitate (2.3 g.) was dissolved in cold water, filtered, and repeatedly decolorized. Evaporation of this solution gave 1.2 g. of brown powder. This was further purified by dissolving the product in 20 ml. of ethanol, decolorizing, and then precipitating with 125 ml. of ether. The light brown precipitate was filtered, washed with ether and dried; yield, 1 g., m.p. 140–141.5°.

Anal. Calc'd for $C_6H_{11}N_5O$: C, 42.60; H, 6.51.

Found: C, 42.8; H, 6.22.

2-Amino-4-(2'-hydroxyethylamino)-5-formamidopyrimidine (V, R' = H, R = CH_2CH_2OH, Figure 1). Into 10 ml. of 90% formic acid was placed 0.68 g. of 2,5-diamino-4-(2'-hydroxyethylamino) pyrimidine and the mixture then was refluxed gently for 15 minutes. At the end of this time, the excess formic acid was evaporated by means of a hot air fan. The residue was dissolved in 3 ml. of water and adjusted to *p*H 8–9 with concentrated ammonium hydroxide. After cooling overnight 0.30 g. of the product was recovered. An analytical sample recrystallized from methanol-water (3:1) yielded white crystals, m.p. 165–166°.

Anal. Calc'd for $C_7H_{11}N_5O_2$: C, 42.64; H, 5.58.

Found: C, 42.4; H, 5.72.

2-Amino-9-(2'-formyloxyethyl)purine (VI, R' = H, R = CH_2-CH_2OCHO). 2-Amino-4-(2'-hydroxyethylamino)-5-formamidopyrimidine (0.55 g.) was gently boiled in 10 ml. of formamide for 15 minutes. The resulting solution was concentrated to 3 ml., 10 ml. of methanol was added, and the mixture was cooled overnight. The grey crystals were removed by filtration and washed with methanol; yield, 0.3 g. Recrystallization and decolorization from 5 ml. of water yielded white needles (0.2 g.) m.p. 172–173°.

Anal. Calc'd for $C_8H_9N_5O_2$: C, 46.37; H, 4.34.
Found: C, 46.27; H, 4.37.

5-Mercapto-1H-v-triazolo[d]pyrimidine (IV, Figure 2). 2-Mercapto-4,5-diaminopyrimidine (2.0 g.) was dissolved in 1200 ml. of water containing 2.0 g. of sodium nitrite; the solution was decolorized with Norit and filtered. The solution was cooled to 30° and acidified by the dropwise addition of acetic acid. It then was adjusted to *p*H 5–6 and set aside overnight at room temperature to precipitate; yield (1.6 g.). The crude product was dissolved in 50 ml. of dilute ammonium hydroxide and reprecipitated with acetic acid. The product was soluble in acetone in contrast to the starting material. It exploded on a melting point block.

Anal. Calc'd for $C_4H_3N_5S$: C, 31.37; H, 1.96.
Found: C, 31.1; H, 1.78.

SUMMARY

Cyclization of 5-aminopyrimidines containing a secondary amino-substituent in the 4-position gives a route to the preparation of the 9-substituted purines. The generality of this cyclization has been demonstrated with a phenyl, *n*-propyl, and an hydroxyethyl group substituted in the 4-amino group.

The ease of cyclization appears to be influenced by the basicity of the secondary amino group, since 2,4-dianilino-5-formamidopyrimidine cyclized on mere heating, whereas both 2-amino-4-(2'-hydroxyethylamino)-5-formamidopyrimidine and 2,4-di-*n*-propamido-5-formylaminopyrimidine cyclized only on refluxing in formamide. Further studies, however, would be needed to establish the significance of this observation.

Three new triazolo[*d*]pyrimidines have been prepared with a substituent in the 3 position. This class of compounds has not been previously reported.

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