[CONTRIBUTION FROM THE DEPARTMENT OF PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS, THE JOHNS HOPKINS UNIVERSITY AND THE DEPARTMENT OF BIOLOGY, NEW YORK UNIVERSITY]

Properties of the Nitrogen–Carbon–Nitrogen System in N^1-Heterocyclic Sulfanilamides[1]

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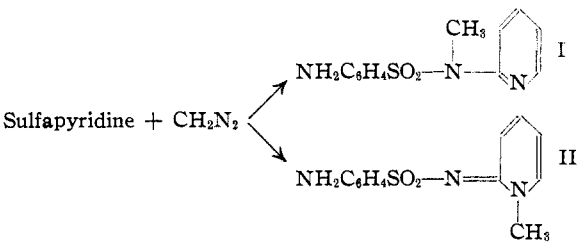
Our chemotherapeutic studies led to the preparation of two types of isomeric derivatives of certain sulfanilamido heterocycles. The structures of these derivatives were determined[3] and direct evidence for the constitution of the parent sulfonamides was obtained by means of ultraviolet absorption measurements.

2-Sulfanilamidopyridine and 2-sulfanilamidothiazole[4] in absolute ether suspension both react rapidly with diazomethane at room temperature to give two methylated isomers. Although 2-(N^4-acetylsulfanilamido)-pyridine[5] under the same conditions reacts much more slowly, a mixture of two products likewise results. This reaction and others throughout this paper will be illustrated by sulfapyridine to represent the behavior of any of the three sulfonamides mentioned above. The



yield-ratio of I:II was 70:30 with the first two substances and 60:40 with the acetyl compound.

The structures of types I and II were determined by three methods. Hydrolysis of both types with 12 *N* hydrochloric acid gave sulfanilic acid and bases whose picrates or hydrochlorides were identified by mixed melting points with the same salts of bases of established structures. The preparation of the N^1-methyl[5] derivative (I) from N^1-methyl-N^4-acetylsulfanilamide and 2-bromopyridine[6] served as a check on the degradation procedure. Structures of type II were also obtained by synthesis from acetylsulfanilyl chloride and the corresponding base of known structure. The products of these two syntheses as well as the amines prepared by de-acetylation were compared by mixed melting points with the corresponding products from the diazomethane reaction.

Alkylation of the sodium salts of sulfapyridine, sulfathiazole and N^4-acetylsulfapyridine with dimethyl sulfate or various alkyl halides was found to produce compounds of type II. The structures of all the products were determined as described above in order to see if the product varied with the alkylating agent. Although our work does not eliminate the possibility of some N^1-alkylation, the main product in each case had structure II. Many of these substances have been erroneously assigned[7] structure I. Our structural conclusions are in direct opposition to the work of Ewins and Phillips, which presumably showed that alkylation of sodium 2-sulfanilamidopyridine produced N^1-alkyl derivatives. They reported the formation of N^1-methyl-2-(N^4-acetylsulfanilamido)-pyridine from condensation of N^1-methyl-N^4-acetylsulfanilamide and 2-bromopyridine by the Ullmann method and from coupling acetylsulfanilyl chloride with 2-methylaminopyridine. The products from both reactions melted at 231° and were de-acetylated to an amine melting at 225°. The corresponding substances from sodium salt methylation were found to have the same melting points and were therefore designated as N^1-methyl derivatives. The acetyl compound which we obtained from the Ullmann synthesis melted at 119.5–120.0° and gave on de-acetylation an amine melting at 86.5–87.0°. We were unable by exhaustive purification to raise these melting points to the values reported. Moreover, our results obtained from acid degradation and from unequivocal syntheses prove conclusively that these products are isomers of

(1) This investigation had been aided by a grant from the John and Mary R. Markle Foundation.

(2) Lalor Foundation Fellow.

(3) After completion of the present work, the proof of structure of certain of the 2-sulfanilamidothiazole derivatives described herein was reported by Druey, *Helv. Chim. Acta*, **24**, 226E (1941), and Jensen, *ibid.*, **24**, 1249 (1941).

(4) The usual names for these substances have been retained although our results indicate that 2-sulfanilimido-1,2-dihydropyridine and 2-sulfanilimido-2,3-dihydrothiazole are perhaps just as correct. They are hereafter referred to as sulfapyridine and sulfathiazole.

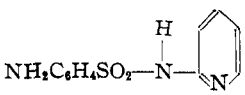
(5) Nomenclature of Crossley, *et al.*, THIS JOURNAL, **60**, 2217 (1938).

(6) Phillips, *J. Chem. Soc.*, 9 (1941).

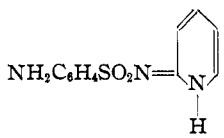
(7) Ewins and Phillips, British Patents 512,145 and 517,272; Phillips, *Nature*, **148**, 409 (1941); Marshall, Bratton, White and Litchfield, *Bull. Johns Hopkins Hosp.*, **47**, 163 (1940); Sprague and Kissinger, THIS JOURNAL, **63**, 578 (1941).

those resulting from sodium salt methylation. Contrary to the evidence of Phillips,[7] we have shown that reaction of sodium sulfapyridine with ethyl chloroacetate or chloroacetamide produces ring nitrogen substitution.

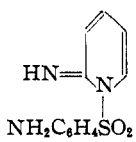
The sulfanilamido heterocycles prepared by coupling acetylsulfanilyl chloride with a heterocyclic amine may have the following three structures (illustrated by sulfapyridine)



III



IV



V

Structure V is eliminated by the preparation from sulfapyridine and sulfathiazole of *two* isomeric methyl derivatives as well as by their proof of structure. Likewise, Phillips' synthesis[6] of sulfapyridine from sulfanilamide and 2-bromopyridine excludes V. Structure IV cannot be eliminated on the basis of the evidence proposed by Crossley, *et al.*[8] The same sodium salt would be expected from III and IV since the negative ion of the salt is capable of resonance. Furthermore, the stability of these sulfonamides to alkaline cleavage is not valid evidence against IV since substituted compounds of that structure (such as II) are very stable compared to the 1-alkyl-2-pyridone imines. A more fundamental objection to the proposed analogy between IV and the 1-alkyl-2-pyridone imines is the fact that a compound having structure IV would first be changed in alkaline solution to a salt of unknown bond structure. The sodium salt alkylation cannot be used as suggested by Druey[3] as evidence of IV in the sulfanilamido heterocycles since this reaction involves a salt whose properties need have no simple relation to the parent.

The results of the diazomethane alkylation may indicate the presence of both III and IV. However, diazomethane cannot be considered a reliable diagnostic reagent for tautomeric equilibria if the reaction mechanism is ionic. In the case of compounds such as the sulfanilamido heterocycles which are capable of yielding a resonating negative ion, an ionic mechanism would invalidate its diagnostic use since the products would be determined by the electronic nature of this ion, obtainable by dissociation of H^+ from both tautomers. Moreover, even if the reaction is non-ionic, the rates of reaction of the tautomers with diazomethane must be approximately equal and be very great compared to the rate of tautomeric rearrangement in order to get a mixture of methyl derivatives comparable to the original tautomeric mixture.

These considerations necessitate a more direct approach to the constitution of such sulfonamides. A distinct difference was observed in the ultraviolet absorption spectra of the methyl derivatives represented by structures I and II, and comparison was made with the spectra of the parent compounds.

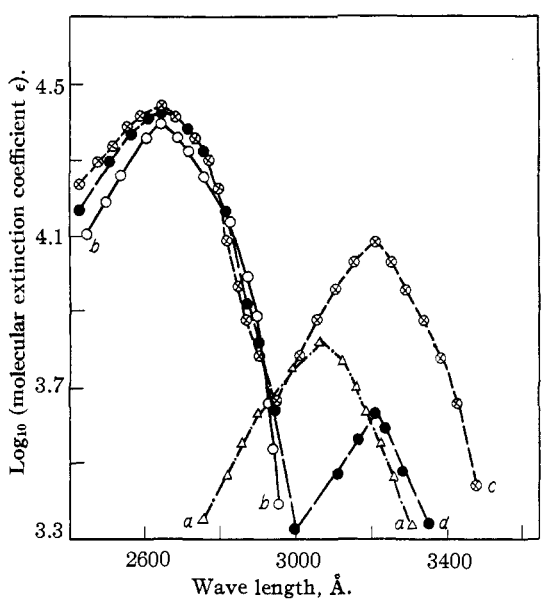


Fig. 1.—(a) Δ, 1-(β-Hydroxyethyl)-2-pyridone imine (Hilger-1 mg. %); (b) ○, N^1-methyl-2-(N^4-acetylsulfanilamido)-pyridine (Hilger-0.5 mg. %); (c) ⊗, 1-methyl-2-(N^4-acetylsulfanilimido)-1,2-dihydropyridine (Hilger-0.5 mg. %); (d) ●, 2-(N^4-acetylsulfanilamido)-pyridine (Hilger-1 mg. %). The peaks at 3215 Å. in (c) and (d) had log ε = 3.98 (5.2 mg. %) and 3.7 (8.1 mg. %), respectively, when determined with the Beckman instrument.

The peak at 3215 Å. in Fig. 1 is assigned to the pyridone imine structure since it appears in (a) and (c) and not in (b). Likewise, the maximum at 2600 Å. in Fig. 2 is exhibited only by (a) and (d) which are known to contain the thiazolone imine structure and is not shown by the isomeric constitution (c). Qualitatively, the presence of structure IV in N^4-acetylsulfapyridine, sulfapyri-

(8) Crossley, *et al.*, THIS JOURNAL, **62**, 372 (1940).

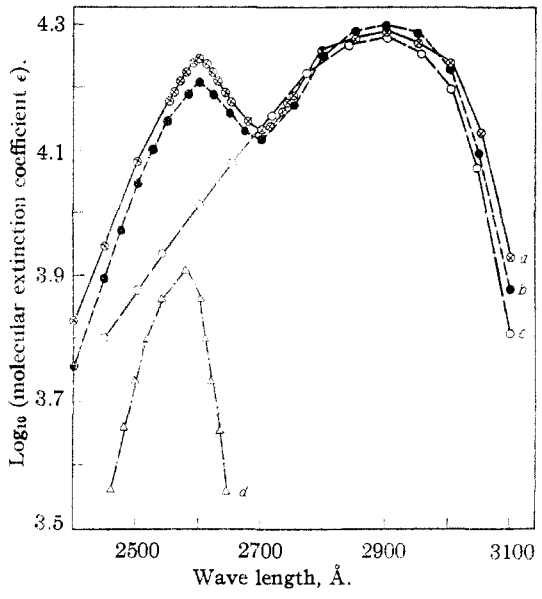


Fig. 2.—(a) ⊗, 3-Methyl-2-sulfanilimido-2,3-dihydrothiazole (Beckman-4.1 mg.%); (b) ●, 2-sulfanilamidothiazole (Beckman-2.6 mg. %); (c) O, N^1-methyl-2-sulfanilamidothiazole (Beckman-4.2 mg. %); (d) Δ, 3-methyl-2-thiazolone imine (Hilger-1 mg. %)—whole curve moved up 0.5 along ordinate axis.

dine and sulfathiazole is demonstrated by the observation of these maxima. The quantitative use of these results to determine the amount of this structure is based on the reasonable assumption that the extinction coefficients of the methyl derivatives are approximately the same as those of the corresponding hydrogen compounds. Therefore, the quantitative conclusions are not rigorous but are estimations subject to the limitations of this assumption.

On the basis of data obtained with the Beckman spectrophotometer, N^4-acetylsulfapyridine contains about 60% of structure IV. The spectra of sulfapyridine and its methyl derivatives show that this sulfonamide is similarly constituted. The maximum at 3215 Å. in 1-methyl-2-sulfanilimido-1,2-dihydropyridine has a slightly lower extinction coefficient ($\log_{10} \epsilon = 3.93$) than in its acetyl derivative. Although these measurements were made on absolute ethanol solutions, we observed the pyridone imine absorption in a few results on 95% ethanol or aqueous solutions. This is confirmed by Scudi's curve[9] for sulfapyridine in water which showed the characteristic pyridone imine maximum at about 3200 Å. The amount of this absorption indicates that a considerable proportion of tautomer IV is also present in aqueous solution. The absorption of sulfathiazole at 2600 Å. corresponds to the presence of about 90% of structure IV. The curve for this substance is the same as that found by Bergeim, *et al.*,[10] who also observed the 2600 Å. maximum in the 4-methyl and ethyl derivatives.

The region of absorption of the pyridine and thiazole rings has not been investigated at present. Observation of the absorptions of these rings in sulfapyridine and sulfathiazole would be necessary to prove the presence of structure III. The available data prove the presence of IV but do not permit one to decide whether these substances have that structure entirely or are tautomeric mixtures of both III and IV.

(9) Scudi, *Science*, **91**, 486 (1940).

TABLE I

In Vitro ACTIVITY RATIOS, USING THE MACLEOD STRAIN OF *E. coli*

Compound		Activity ratio[a]
2-Sulfanilamidopyridine		1[b]
N^1-Methyl-2-sulfanilamidopyridine		1/128
( )-2-sulfanilimido-1,2-dihydropyridine	1-Methyl-	1/4
	1-(β-Hydroxyethyl)-	1/128
	1-Carboxymethyl-	1/256
2-Sulfanilamidothiazole		1[c]
N^1-Methyl-2-sulfanilamidothiazole		1/1024
( )-2-sulfanilimido-2,3-dihydrothiazole	3-Methyl-	1/16
	3-(β-Hydroxyethyl)-	1/128

[a] Based on the relative minimal inhibitory concentrations using an end-point reading after forty-eight hours incubation at 37°. [b] Taken as unity; actual min. inhib. concn. = 0.16 mg.%. [c] Taken as unity; actual min. inhib. concn. = 0.04 mg.%.

Chemotherapeutic Activity.—Table I shows the *in vitro* activity of these sulfonamides tested against *E. coli* in a synthetic medium by the method described in detail elsewhere.[11]

The *in vivo* activity of the ring N-methyl derivatives of sulfapyridine and sulfathiazole was determined in mice infected with β-hemolytic streptococcus (strain C 203) using the drug-diet method.[12] On the basis of blood concentrations, both were about equal to sulfanilamide in activity. Since it has been shown[13] that sulfapyridine and sulfathiazole are about equal to sulfanilamide in this infection, the two ring N-methyl compounds are as active as the parents. The ring N-(β-hydroxyethyl) derivatives appear to be as active as the

(10) Bergeim, *et al.*, THIS JOURNAL, **62**, 1873 (1940).

(11) White, Litchfield and Marshall, *J. Pharmacol.*, **73**, 104 (1941).

(12) Litchfield, White and Marshall, *ibid.*, **67**, 437 (1939); **69**, 89 (1940).

(13) Marshall, Litchfield, White, Bratton and Shepherd, *ibid.*, in press.

TABLE II

Compound	M. p., °C.	Empirical formula	Carbon, % Calcd.	Carbon, % Obs.	Hydrogen, % Calcd.	Hydrogen, % Obs.	Nitrogen, % Calcd.	Nitrogen, % Obs.	Colorimetric factor[a] Calcd.	Colorimetric factor[a] Obs.	Water soly. at 37° mg. %
N^1-Methyl-2-sulfanilamidopyridine	86.5–87.0[b]	$C_{12}H_{13}N_3O_2S$	54.74	54.76	4.98	4.82	15.96	15.94	0.654	0.658	136
( )-2-sulfanilimido-1,2-dihydropyridine											
1-Methyl-	232–3	$C_{12}H_{13}N_3O_2S$	54.74	54.68	4.98	4.85	15.96	15.90	.654	.670	112
1-Benzyl-	235[c]	$C_{18}H_{17}N_3O_2S$	63.70	64.00	5.05	5.04	12.38	12.18	.511	.511	
1-Carboxymethyl-	165 (dec.)	$C_{13}H_{13}N_3O_4S$	50.81	50.75	4.26	4.37	13.67	13.59	.560	.570	754
1-(β-Hydroxyethyl)-	184–5	$C_{13}H_{15}N_3O_3S$	53.23	53.41	5.15	5.18	14.33	14.35	.587	.630	440
N^1-Methyl-2-sulfanilamidothiazole	111–2	$C_{10}H_{11}N_3O_2S_2$	44.60	45.00	4.12	4.19	15.60	15.36	.639	.618	57
( )-2-sulfanilimido-2,3-dihydrothiazole											
3-Methyl-	250–1	$C_{10}H_{11}N_3O_2S_2$	44.60	44.49	4.12	4.13	15.60	15.54	.639	.656	22
3-(β-Hydroxyethyl)-	159–60	$C_{11}H_{13}N_3O_3S_2$	44.13	44.42	4.38	4.35	14.04	14.36	.575	.600	169

[a] Ratio of color produced by a compound to the color from an equal weight of sulfanilamide on diazotization and coupling according to Bratton and Marshall, *J. Biol. Chem.*, **128**, 537 (1939). [b] Ewins and Phillips (ref. 7) reported 225°. [c] Ewins and Phillips (ref. 7) found 179°.

unsubstituted sulfonamides against this streptococcus infection; the quantitative data have been reported elsewhere.[13]

When tested in a pneumococcus infection (Neufeld type I) of mice, both N^1-methyl derivatives (Table I) appeared to be practically inactive and quite toxic. On the other hand, the ring N-methyl isomers were active, less toxic than the N^1-methyl compounds and apparently less active than the parents. The inactivity of N^1-methyl-2-sulfanilamidothiazole with respect to its ring N isomer was also observed in duck malaria.[14]

Experimental Section[15]

Methylation of Sulfonamides with Diazomethane.—Sulfapyridine and sulfathiazole react rapidly enough with diazomethane to permit distillation of this reagent into a stirred mixture of the sulfonamide and ether. This continuous procedure avoids the danger of handling 10-g. quantities of diazomethane.

Diazomethane (7.5 g.) was distilled[16] into a stirred mixture of 0.25 mole of sulfonamide (200 mesh) and 75 cc. of absolute ether at 15–20°. The evolution of nitrogen proceeded during the addition of this reactant and the mixture was subsequently stirred until this evolution ceased. After warming to the boiling point, the reaction mixture was filtered and the insoluble material extracted twice with hot ether containing 3% ethanol. The residue from this extraction was shaken with 2 *N* sodium hydroxide to remove starting material, and the residual insoluble ring-nitrogen derivative was recrystallized from 6 *N* acetic acid. The original ether filtrate and the ether extracts were combined and evaporated to a sirup which crystallized after cooling and seeding. This was suction-filtered and carefully washed dropwise with methanol and absolute ether. Methanol was used for recrystallization of the N^1-methyl derivative thus obtained. The yield of methylated products was 50–80% of the sulfonamide, depending, primarily, on its state of subdivision. The yield-ratio of N^1-methyl to ring N-methyl derivative was about 70:30. This ratio was changed only slightly by methylation of 2-sulfanilamidopyridine immediately after liberation from its sodium salt at about −100°.

The lower order of reactivity of 2-(N^4-acetylsulfanilamido)-pyridine with diazomethane under these conditions necessitates using smaller quantities than above. Since the spontaneous inflammability of the vapors was regularly observed, the use of large quantities is somewhat dangerous. Isolation of the two products was carried out in essentially the same way as above. The yield-ratio of N^1-methyl to ring N-methyl derivative was about 60:40. N^1-methyl-2-(N^4-acetylsulfanilamido)-pyridine: m. p. 119.5–120.0°; *colorimetric factor*, calcd., 0.564, obs., 0.557. 1-Methyl-2-(N^4-acetylsulfanilimido)-1,2-dihydropyridine: m. p. 239–40°; *colorimetric factor*, calcd., 0.564, obs., 0.555.

The N^1-methyl derivatives of sulfapyridine, acetylsulfapyridine and sulfathiazole are all quite soluble in methanol, ethanol and ether containing 3% ethanol; their isomers are only slightly soluble.

Methylation of Sodium Sulfonamides with Dimethyl Sulfate.—Four-tenths of a mole of sulfonamide was alkylated in alkaline solution as described by Ewins and Phillips.[7] The product was recrystallized from 300–400 cc. of 6 *N* acetic acid with charcoal decolorization, washed with 20% ethanol and dried at 120°; yield 140–50%. Recrystallization from ethanol resulted in further purification.

Increasing the amount of dimethyl sulfate used in one step or re-treating the reaction filtrate did not increase the yield of desired product but increased the production of an alkali-insoluble gum. An important constituent of this gum was shown to be the result of methylation of the arylamine group as well as the heterocyclic nitrogen.

Preliminary tests demonstrated that these conditions do not give satisfactory methylation of certain sulfanilamido heterocycles derived from imidazole, pyrimidine and pyrazine.

Sodium Sulfonamide Benzylation.—The procedure of Ewins and Phillips[7] was modified slightly by the use of a 75% ethanol reaction mixture which resulted in an increased yield. 1-Benzyl-2-(N^4-acetylsulfanilimido)-1,2-dihydropyridine; m. p. 213–4°; *colorimetric factor*, calcd., 0.452, obs., 0.458.

Acetic Acid Derivatives of 2-Sulfanilamidopyridine.—To 48 g. of sodium 2-sulfanilamidopyridine in 55 cc. of 50% ethanol was added a solution of 40 cc. of ethyl chloroacetate in 95 cc. of 95% ethanol and the mixture refluxed for one hour, then cooled and precipitated with ice and water.

(14) Marshall, Litchfield and White, *J. Pharmacol.*, **75**, 89 (1942).
(15) All the melting points recorded are corrected.
(16) Hellerman and Newman, THIS JOURNAL, **54**, 2864 (1932).

After filtration the solid was shaken thoroughly with ether, then with 2 *N* sodium hydroxide and finally washed well with water. The yield of 1-carbethoxymethyl-2-sulfanilimido-1,2-dihydropyridine was 18 g; m. p. 200.5–201.0° after recrystallization from absolute ethanol or methanol. *Colorimetric factor:* calcd., 0.514; obs., 0.534. 1-Carbethoxymethyl - 2 - (N^4 - acetylsulfanilimido) - 1,2 - dihydropyridine obtained from a similar reaction melted at 212–3°; *colorimetric factor:* calcd., 0.457; obs., 0.463.

Five grams of the first ethyl ester and 8.9 cc. of 11% potassium hydroxide in 98% methanol were refluxed together for twenty minutes. After acidification with 1.1 cc. of 12 *N* hydrochloric acid, the potassium chloride was separated from the hot solution. The crystals of 1-carboxymethyl-2-sulfanilimido-1,2-dihydropyridine resulting from the cooled filtrate were suction-filtered and washed free of chloride ion with water. A monohydrate melting at 97–98° (sealed tube) was formed on recrystallization from water; *colorimetric factor:* calcd., 0.529; obs., 0.529.

1 - Carbamidomethyl - 2 - sulfanilimido - 1,2 - dihydropyridine was prepared by reaction of the sulfonamide in alkaline solution with chloroacetamide; m. p. 230° (dec.); *colorimetric factor:* calcd., 0.562; obs., 0.555. Alkaline hydrolysis of the amide group produced an acid identical to that obtained by the same treatment of the ester described above.

Reaction of Sodium Sulfonamides with Ethylene Chlorohydrin.—The anhydrous sodium salt of N^4-acetylsulfapyridine or N^4-acetylsulfathiazole was refluxed for thirty minutes in an oil-bath at 130° with five moles of ethylene chlorohydrin. The mass was then cooled, powdered, washed with ether, dried and washed with dilute sodium hydroxide, followed by water. The crude products were recrystallized from 6 *N* acetic acid with the addition of charcoal. 1-(β-Hydroxyethyl)-2-(N^4-acetylsulfanilimido)-1,2-dihydropyridine; m. p. 217–8°. *Anal.* Calcd. for $C_{15}H_{17}N_3O_4S$: C, 53.72; H, 5.11; N, 12.53. Obs.: C, 53.67; H, 5.26; N, 12.64. 3-(β-Hydroxyethyl)-2-(N^4-acetylsulfanilimido)-2,3-dihydrothiazole; m. p. 231–2° (dec).

The former substance was de-acetylated by refluxing for two hours with 2 moles of 1 *N* alcoholic sodium hydroxide. After concentrating on the steam-bath, the crystalline mass was washed with a little water and recrystallized from water. The latter compound was de-acetylated by boiling thirty minutes with 6 *N* hydrochloric acid (3 cc. per g.), precipitated by neutralization and recrystallized from alcohol with charcoal decolorization. The yield of both hydrolysis products was about 50%, based on the original sodium salts.

Structure Determination by Acid Degradation.—All of these sulfanilamide derivatives were hydrolyzed by heating one to two hours in a steam-bath with 10 moles of 12 *N* hydrochloric acid. After cooling, the precipitated sulfanilic acid was collected and recrystallized from dilute hydrochloric acid. It was identified by its decomposition point, strong acidity in the absence of chloride ion and quantitative diazotization. The acid filtrate from the hydrolysis was concentrated and the heterocyclic base was either allowed to crystallize as the hydrochloride or sodium acetate was added to *p*H 4 and the base precipitated as a picrate in 80–90% yield. The picrates were recrystallized from water or ethanol and then compared with the salts of bases whose structures had been established in the literature indicated. The melting points of the latter salts are recorded in Table III. These values are the same as those found for the picrates from hydrolysis and for the mixed melting points of the two groups.

TABLE III

DETERMINATION OF STRUCTURE

For	By degradation Picrate, m. p., °C.	By synthesis NH_2R,[a] m. p., °C.	By synthesis CH_3CONHR,[b] m. p., °C.
N^1 - Methyl - 2 - sulfanilamidopyridine	194–5[c]	86.5–87	119.5–120[d]
N^1 - Methyl - 2 - sulfanilamidothiazole	207–8[e]		
( )-2-Sulfanilimido-1,2-dihydropyridine			
1-Methyl-	205–6[f]	232–233	239–240[g]
1-Benzyl-	153–4[h,c]	235	213–214[g]
1-Carboxymethyl-	213[i]		
1-Carbamidomethyl-	213[i]		
1-(β-Hydroxyethyl)-	172–3[j]		
( )-2-Sulfanilimido-2,3-dihydrothiazole			
3-Methyl-	199–200[k]	250–251	272–273[g]
3-(β-Hydroxyethyl)-	160–162		

[a] NH_2R = the free amine of the sulfonamides. [b] CH_3CONHR = the N^4-acetyl derivatives of the sulfonamides. [c] Chichibabin, *Ber.*, **54,** 814 (1921). [d] Prepared according to Ewins and Phillips, British Patent 512,145. [e] Näf, *Ann.*, **265,** 113 (1891). [f] Chichibabin, *et al.*, *Ber.*, **54B,** 814–822 (1921). [g] Prepared by coupling acetylsulfanilyl chloride with the heterocyclic base. [h] The hydrochlorides were also compared: m. p. and mixed m. p. 207–208°. [i] Melts with decomposition; Chichibabin, *ibid.*, **57,** 2092 (1924); Reindel and Rauch, *ibid.*, **58,** 393 (1925); 59, 2921 (1926). [j] Knunyantz, *ibid.*, **68B,** 397 (1935); Gautier, *Compt. rend.*, **196,** 1124 (1933). [k] This base is more conveniently prepared using dimethyl sulfate rather than methyl iodide; *cf.* Näf, ref. *e*.

The bases necessary for this comparison were synthesized by available methods, with the exception of 2-imino-3-(β-hydroxyethyl)-2,3-dihydrothiazole which was prepared as follows: In a large test-tube, a mixture of 4 g. of 2-aminothiazole and 8.6 g. of iodoethyl acetate was heated gradually in an oil-bath to about 130°, stirring the mixture constantly with a thermometer. Suddenly the temperature began to rise and the tube was placed in a cold oil-bath to prevent the temperature from exceeding 180°. After the reaction had subsided, the mixture was heated for thirty minutes at 150°, cooled and taken up in 15 cc. of hot absolute ethanol. The crystalline 2-imino-3-acetoxyethyl-2,3-dihydrothiazole hydroiodide, obtained on chilling, was recrystallized several times from glacial acetic acid; yield, 1.9 g.; m. p. 153.5–154.5°. *Anal.* Calcd. for $C_7H_{11}N_2O_2SI$: C, 26.76; H, 3.53. Found: C, 26.91, H, 3.41; m. p. of picrate 164–165° (recryst. from glacial acetic acid).

After hydrolyzing the ester by refluxing for thirty minutes with 1 *N* hydrochloric acid, the picrate was formed by adding picric acid to the concentrated and buffered hydrolyzate; m. p. 159.5–161.0° (recryst. from methanol). *Anal.* Calcd. for $C_{11}H_{11}N_5O_9S$: C, 35.39, H, 2.97. Found: C, 35.66, H, 3.04

Structure Determination by Alkaline Degradation.—When 1 - (β - hydroxyethyl) - 2 - sulfanilimido - 1,2 - dihydropyridine was hydrolyzed by boiling for five hours with 6 *N* sodium hydroxide (10 cc. per g.), sulfanilamide and 1-(β-hydroxyethyl)-2-pyridone were obtained. It was found that the latter is dimorphic: m. p. 84–85° by rapid crystallization of the fused substance; m. p. 93.5–95° by slow crystallization from dilute solution. This accounts for the discrepancies in melting point which have been reported.[17]

It was impossible to decompose sulfapyridine under similar alkaline hydrolytic conditions. In this connection, it should be pointed out that in the short time required to completely decompose the alkyl pyridone imines with alkali, no appreciable cleavage of the ring N derivatives (II) of sulfapyridine was observed.

Structure Determination by Synthesis.—Coupling acetylsulfanilyl chloride with the 1-alkyl-2-pyridone imines[18] and 3-alkyl-2-thiazolone imines yields sulfonamides whose point of substitution is definitely established. This is also true for the product obtained by reacting N^1-methyl-N^4-acetylsulfanilamide with 2-bromopyridine according to the method of Ewins and Phillips.[7] The acetyl compounds prepared by these routes and the corresponding amines resulting from de-acetylation were proved to be identical with those prepared by alkylation of the acetylsulfanilamido and sulfanilamido heterocycles. The melting points are recorded in Table III; the mixed melting points were identical.

1-Carboxymethyl-2-sulfanilimido-1,2-dihydropyridine.—Having proved the position of substitution, there still remain two possible structures differing in the position of a hydrogen atom. One would be a carboxymethylpyridone imine derivative; the other, an aminopyridine betaine derivative. On methylation, the former would presumably give a methyl ester; the latter, an N-methyl derivative. On treating the compound in question with diazomethane, there was obtained a neutral substance which without purification contained 99.2% of an alkali-labile ester. Repeated treatment of the acid with dimethyl sulfate in alkaline solution gave no evidence of N^1-methylation. Although these results are not entirely conclusive, they indicate that the carboxymethyl-pyridone imine structure is probably correct.

Ultraviolet Absorption Measurements.—These data were obtained on absolute ethanol solutions with the aid of two instruments: a medium Hilger "Spekker" spectrograph employing a tungsten-steel arc as the light source and a 4-cm. cell; a Beckman quartz spectrophotometer with a hydrogen-discharge tube as the light source and a 1-cm. cell.

(17) Knunyantz, *Ber.*, **68B**, 397 (1935); *cf.* Gautier, *Compt. rend.*, **196**, 1124 (1933).

(18) Polyakova and Kirsanov, *cf. C. A.*, **35**, 2146 (1941).

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Summary

1. 2-Sulfanilamidopyridine, 2-sulfanilamidothiazole and 2-(N^4-acetylsulfanilamido)-pyridine react with diazomethane to give both N^1-methyl and ring N-methyl derivatives, the ratio of products being 70:30 for the first two compounds and 60:40 for the last.

2. Alkylation of the sodium salts of these sulfonamides is shown to produce ring N-alkyl derivatives in all the cases examined.

3. The structures of the products from these two reactions have been conclusively determined by both degradative and synthetic methods.

4. The dimorphism of 1-(β-hydroxyethyl)-2-pyridone is demonstrated.

5. A method is proposed for determination of the constitution of the sulfanilamido heterocycles based on the comparison of the ultraviolet absorption spectra of the parent sulfonamide with the spectra of its two isomeric N-methyl derivatives. The available data indicate the presence of large amounts of the pyridone imine structure in sulfapyridine and N^4-acetylsulfapyridine solutions and of the thiazolone imine structure in sulfathiazole solution.

6. Chemotherapeutic results show that the ring nitrogen derivatives of sulfapyridine and sulfathiazole are approximately as active as the parent sulfonamides, while the isomeric N^1 compounds are practically inactive.

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