STUDIES IN SYNTHETIC ANTIMALARIALS

Part XX. Some Suiphabiguanide Derivatives

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Summary

Nineteen N^1-sulphanilamido-N^5-alkyl biguanide derivatives have been synthesised with a view to studying their antimalarial and antibacteria properties.

The discovery of N^1-*p*-chlorophenyl-N^5-isopropyl biguanide[1] and its unique antimalarial property led to a vast amount of research in the fields of biguanide and guanidine derivatives. As a part of the extensive programme of research in the chemotherapy of malaria that has been undertaken in this laboratory to study the effects of different substitutions at either ends of the tautomeric biguanide structure, a new line of approach had been opened by Bami, *et al.*,[2,3,4] who have synthesised a number of N^5-sulphonamido-N^1-aryl biguanides. Some of these compounds were found to possess slight suppressive antimalarial activity[5] when screened against *P. gallinaceum* in chicks.

Considering the general standard of activity of the sulphonamide group of drugs, as also the very encouraging pharmacological data obtained in this laboratory with alkyl and aryl guanidine derivatives of sulphonamides,[6] it was thought worthwhile to extend this line of work in the search for further possibilities of a potential antimalarial in the same field. This line of work was further supported by previous findings that sulphadiazine, sulphathiazole, sulphaguanidine and sulphanilamide are in descending order of activity against malarial infections[7,8,9]; and of these sulphadiazine is the most outstanding due to the fact that it had slight but definite causal prophylactic action which is not demonstrated by other antimalarial drugs.

The replacement of the isopropyl group of the paludrine molecule by various sulphonamide residues did not give any fruitful results, though some slight suppressive activity was noticed in a few cases. It was felt that a comparison of the antimalarial activity of the new compounds like N^1-sulphonamido-N^5-alkyl biguanides of the type A, with those of the other sulphabiguanide compounds prepared previously in this laboratory[2,3,4] would

be very interesting. Accordingly a number of new compounds of the type A have been synthesised and their antimalarial properties are being studied.

$$R.NH.SO_2.C_6H_4.NH.\underset{\underset{NH}{\|}}{C}.NH.\underset{\underset{NH}{\|}}{C}.NHR'$$ Type A

where R = H, 2-thiazolyl, 2-pyrimidyl, 2-pyrimidyl-4-methyl, pyridyl, guanyl, etc,

R′ = alkyl radical.

Compounds of the type A could be synthesised either by (1) reaction of an alkyl cyanoguanidine with the appropriate sulphonamide or (2) by the reaction between a sulphonamido-cyanamide and an alkyl guanidine. Of these two methods the first one was not found to be very convenient owing to the poor yields in the preparation of dicyanimide which is necessary for the preparation of alkyl cyano guanidines. The second method was quite suitable and fairly satisfactory yields were obtained when the reactions were carried out in pyridine medium. All the compounds were isolated either as their hydrochlorides or their sulphates.

EXPERIMENTAL

Ethyl-cyanoguanidine.—The dicyanimide required for the preparation of alkyl cyanoguanidines was prepared according to the method of Medelung and Kern.[10]

Ethylamine hydrochloride (12 g.), sodium salt of dicyanimide (13·5 g.) and *n*-butyl alcohol (70 ml.) were refluxed with stirring during four hours. The cooled suspension was filtered and the filtrate evaporated on the water-bath. The residual semi-solid mass solidified on treatment with dioxane and the compound was recrystallised from the same solvent. Colourless crystals, m.p. 108°.

N^1-Sulphonamidophenyl-N^5-ethyl biguanide hydrochloride (*I*).—Sulphanilamide (8·6 g.), ethyl cyanoguanidine (7·7 g.), concentrated hydrochloric acid (5 ml.) and water (50 ml.) were refluxed together for six hours. The mixture was again refluxed for half an hour after the addition of norite and finally filtered hot. The clear filtrate was concentrated to a small bulk and allowed to cool, when the biguanide hydrochloride precipitated out. The product was filtered and recrystallised twice from water, and was obtained in the form of white crystalline powder, m.p. 192° (Found: N, 26·34; $C_{10}H_{17}O_2N_6SCl$ requires, N, 26·25 per cent.).

Isopropyl cyanoguanidine.—It was prepared from isopropylamine hydrochloride (7·2 g.), and sodium dicyanimide (6·7 g.). The compound melts at 112°.

N^1-Sulphonamidophenyl-N^5-isopropyl biguanide hydrochloride (II).—Sulphanilamide (8·6 g.), isopropylcyanoguanidine (8·5 g.), concentrated hydrochloric acid (5 ml.) and water (50 ml.) were refluxed during four hours and filtered hot. The filtrate on cooling deposited the product. The compound was recrystallised from water. Shining crystals m.p. 238°, (Found: N, 25·02, Cl, 10·55, $C_{11}H_{19}O_2N_6SCl$ requires N, 25·14; and Cl, 10·61 per cent.).

The free base of the hydrochloride was obtained by adding dilute caustic soda solution to a hot saturated solution of the biguanide salt. On cooling the biguanide base separated out and was crystallised from water. M.P. 192–93° (Found: N, 27·84; $C_{11}H_{18}O_2N_6S$ requires N, 28·19 per cent.).

Diethyl-cyanoguanidine.—It was prepared in the same way as ethyl cyanoguanidine, from diethyl amine hydrochloride (11 g.) and sodium dicyanimide (8·9 g.). The product was crystallised from toluene. The compound melts at 151–52°.

N^1-Sulphonamido phenyl-N^5-diethyl biguanide hydrochloride (III).—Sulphanilamide (8·6 g.) diethyl cyanoguanide (7 g.) hydrochloric acid (5 ml.) and water (35 ml.) were heated under reflux during three hours. The product was isolated and purified as under I. Shining white crystals, m.p. 232–34° (Found: N, 24·29, $C_{12}H_{21}O_2N_6SCl$ requires N, 24·10 per cent.).

Methyl cyanoguanidine was prepared from methyl amine hydrochloride (7 g.) and sodium dicyanimide (8·9 g.). The product was crystallised from dioxane, m.p. 104-05°.

N^1-Sulphonamidophenyl-N^5-methyl biguanide hydrochloride (IV).—Sulphanilamide (8·6 g.), methyl cyanoguanidine (7 g.), hydrochloric acid (5 ml.) and pyridine (30 ml.) were refluxed for six hours. The pyridine was removed by means of steam distillation. After removal of pyridine, the aqueous solution was boiled with norite for about half an hour and filtered. The filtrate was chilled in an ice-bath when the biguanide crystallised. The product was recrystallised from water and the compound obtained as white crystals, m.p. 151° (Found: N, 27·31, $C_9H_{15}O_2N_6SCl$ requires N, 27·45 per cent.).

Methylguanidine sulphate was prepared from methyl isothiourea sulphate and aqueous solution of methyl amine[4] by the method due to Phillips and Clarke.[11]

Isopropyl guanidine sulphate and Butyl guanidine sulphate were similarly prepared.

2-Pyrimidyl-sulphonamido phenyl-cyanamide was prepared from the thiourea of sulphadiazine. Sulphadiazine (15 g.) was added to water (1 litre) and slightly excess of hydrochloric acid added to it. The whole was heated on a water-bath and potassium thiocyanate (15 g.) added to it, and the solution was evaporated to dryness on a steam-bath. The thiourea thus formed was dissolved in sodium hydroxide solution (30 per cent., 100 ml.), and the solution was diluted by the addition of water (500 ml.) and the alkaline solution heated on a water-bath for another one hour. To the hot alkaline solution was added a solution of basic lead acetate (50 g.) in water (150 ml.) and the whole was heated on the steam-bath for ten minutes with occasional stirring. The lead sulphide formed in the reaction was filtered off, the filtrate cooled with ice and made slightly acidic with acetic acid, when the cyanamide precipitated. The precipitate was filtered and dissolved in aqueous ammonia; after filtration the filtrate was acidified with acetic acid under cooling when the pure cyanamide precipitated. The product was filtered and washed well with water and dried. The product is a light pink coloured solid. M.P. 192–94° (with decomposition) (Found: N, 23·64, $C_{12}H_9O_2N_5S$ requires N, 24·38 per cent.).

N^1-2-pyrimidyl-sulphonamido-phenyl-N^5-methyl biguanide sulphate (V).— 2-Pyrimidyl-sulphonamido-phenyl cyanamide (4 g.), methyl guanidine sulphate (2 g.) and pyridine (25 ml.) were refluxed together over a small flame during eight hours. The product was isolated and purified as under compound (IV); yellow powder, m.p. 156–58° [Found: N, 28·35, $(C_{13}H_{16}O_2N_8S)_2 \cdot H_2SO_4$ requires N, 28·21 per cent.].

N^1-2-pyrimidyl-sulphonamido phenyl-N^5-isopropyl biguanide sulphate (VI).— 2-Pyrimidyl sulphonamido phenyl cyanamide (3 g.) and isopropyl guanidine sulphate (1·6 g.) and pyridine (25 ml.) were refluxed together over a small flame during eight hours. The product was isolated and purified as under compound IV. White crystalline powder. Yield 2 g. M.P. 159° [Found: N, 26·43 $(C_{15}H_{20}O_2N_8S)_2.H_2SO_4$ requires N, 26·35 per cent.].

N^1-2-pyrimidyl-sulphonamido phenyl-N^5-butyl biguanide sulphate (VII).— 2-Pyrimidyl-sulphonamido phenyl cyanamide (3 g.) and butyl guanidine sulphate (1·7 g.) and pyridine (25 ml.) were refluxed together for six hours. The product was isolated and purified as under compound IV. White crystals. Yield 2 g. M.P. 251° [Found: N, 25·64 $(C_{16}H_{22}O_2N_8S)_2.H_2SO_4$ requires N, 25·51 per cent.].

Sulphanilamido phenyl guanidine cyanamide was prepared from the thiourea of sulphaguanidine. Sulphaguanidine (12 g.) was taken in 400 ml. of water and a slight excess of hydrochloric acid was added to it. To the hot solution 7·5 g. of potassium thiocyanate was added. The cyanamide was obtained by following the same procedure as was adopted in the preparation of 2-pyrimidyl-sulphonamido phenyl cyanamide, using 25 g. of basic lead acetate in 125 ml. of water for desulphurising the thiourea formed. The cyanamide melts at 138° with decomposition (Found: N, 28·91; $C_8H_9O_2N_2S$ requires N, 29·29 per cent.).

N^1-sulphanil guanidino-N^5-methyl biguanide sulphate (*VIII*).—Sulphanilamido phenyl guanidine cyanamide (3 g.), methyl guanidine sulphate (1·5 g.) and pyridine (30 ml.) were refluxed together for eight hours. The product was isolated and purified as under compound IV. White crystals. Yield 2 g. M.P. 104° [Found: N, 31·40 $(C_{10}H_{16}O_2N_8S)_2.H_2SO_4$ requires N, 31·02 per cent.].

N^1-sulphanil guanidino-N^5-isopropyl biguanide sulphate (*IX*).—Sulphanil amido phenyl guanidine cyanamide (2·4 g.), isopropyl guanidine sulphate (1·5 g.) and pyridine (25 ml.) were refluxed together for six hours. The product was isolated and purified as under compound IV. White crystals. Yield 1·5 g. M.P. 235° with decomposition [Found: N, 28·69 $(C_{12}H_{20}O_2N_8S)_2.H_2SO_4$ requires N, 28·78 per cent.].

N^1-sulphanil guanidino-N^5-butyl biguanide sulphate (*X*).—Sulphanilamido phenyl guanidine cyanamide (3 g.) and butyl guanidine sulphate (2 g.) and pyridine (30 ml.) were refluxed together for six hours. The product was isolated and purified as under compound IV. Light yellow coloured crystals. Yield 2·5 g. M.P. 280° *d.* [Found: N, 27·84, $(C_{13}H_{22}O_2N_8S)_2.H_2SO_4$ requires N, 27·79 per cent.].

N^1-2 (4-methyl pyrimidyl) sulphonamido phenyl cyanamide was prepared from the thiourea of sulphamerazine. Sulphamerazine (20 g.) was taken in 450 ml. of water and a slight excess of hydrochloric acid was added to it. To the hot solution 10·5 g. of potassium thiocyanate was added. The cyanamide was obtained by following the same procedure as was adopted in the preparation of 2-pyrimidyl-sulphonamido phenyl cyanamide, using 33 g. of basic lead acetate in 150 ml. of water for desulphurising the thiourea formed. The cyanamide melts at 232–33° (Found: N, 24·46, $C_{12}H_{11}O_2N_5S$ requires N, 24·22 per cent.).

N^1-N^1-2 (4-methyl pyrimidyl) sulphonamido phenyl-N^5-Methyl biguanide sulphate (*XI*).—N^1-2-(4-methyl pyrimidyl) sulphonamido phenyl cyanamide

(2·5 g.), methyl guanidine sulphate (1 g.) and pyridine (15 ml.) were refluxed together for ten hours. The product was isolated and purified as under compound IV. Yellow crystals. Yield 1·5 g. M.P. 226° [Found: N, 27·12 $(C_{14}H_{18}O_2N_8S)_2.H_2SO_4$ requires N, 27·25 per cent.].

N^1-N^1-2-(4-methyl pyrimidyl) sulphonamido phenyl-N^5-isopropyl biguanide sulphate (*XII*).—N^1-2-(4-methyl pyrimidyl) sulphonamido phenyl cyanamide (3·5 g.), isopropyl guanidine sulphate (1·8 g.) and pyridine (20 ml.) were refluxed together for eight hours. The product was isolated and purified as under compound IV. White crystals. Yield 2 g. M.P. 221° [Found: N, 25·80 $(C_{13}H_{22}O_2N_8S)_2.H_2O_4$ requires N, 25·51 per cent.].

N^1-N^1-2-(4-methyl pyrimidyl) sulphonamido phenyl-N^5-butyl biguanide sulphate (*XIII*).—N^1-2-(4-methyl pyrimidyl) sulphonamido phenyl cyanamide (4 g.), butyl guanidine sulphate (2·3 g.) and pyridine (25 ml.) were refluxed together for six hours. The product was isolated and purified as under compound IV. Yellow crystals. Yield 2 g. M.P. 320° *d*. [Found: N, 24·92; $(C_{17}H_{24}O_2N_8S)_2 \cdot H_2SO_4$ requires N, 24·73 per cent.].

Sulphanilamido phenyl thiazole cyanamide was prepared from the thiourea of sulphathiazole. Sulphathiazole (15 g.) was taken in 500 ml. of water and a slight excess of hydrochloric acid was added to it. To the hot solution 9 g. of potassium thiocyanate was added. The cyanamide was obtained by following the same procedure as was adopted in the preparation of 2-pyrimidyl-sulphonamido phenyl cyanamide, using 26 g. of basic lead acetate in 120 ml. of water for desulphurising the thiourea formed. The cyanamide melts at 189° (Found: N, 19·89, $C_{10}H_8O_2N_4S_2$ requires N, 20·00 per cent.).

N^1-sulphanilamido phenyl thiazolyl-N^5-methyl biguanide sulphate (*XIV*).—Sulphanilamido phenyl thiazole cyanamide (3 g.), methyl guanidine sulphate (1·3 g.) and pyridine (25 ml.) were refluxed together for six hours. The product was isolated and purified as under compound IV. White crystals. Yield 1·5 g. M.P. 212° [Found: N, 24·53 $(C_{12}H_{15}O_2N_7S_2)_2.H_2SO_4$ requires N, 24·38 per cent.].

N^1-sulphanilamido phenyl thiazolyl-N^5-isopropyl-biguanide sulphate (*XV*).—Sulphanilamido phenyl thiazole cyanamide (4·5 g.). isopropyl guanidine sulphate (2·4 g.), pyridine (25 ml.) were refluxed together for eight hours. The product was isolated and purified as under compound IV. Yellow crystals. Yield: 2·5 g. M.P. 179° [Found: N, 22·87 $(C_{14}H_{19}O_2N_7S_2)_2.H_2SO_4$ requires N, 22·79 per cent.].

N^1-sulphanilamido-phenyl thiazolyl-N^5-butyl biguanide sulphate (*XVI*).—Sulphanilamido phenyl thiazole cyanamide (2·5 g.), butyl guanidine sulphat

(2·2 g.) and pyridine (25 ml.) were refluxed together for eight hours. The product was isolated and purified as under compound IV. Yellow crystals. Yield: 1·5 g. M.P. 189° [Found: N, 22·39; $(C_{15}H_{21}O_2N_7S_2)_2.H_2SO_4$ requires N, 22·08 per cent.].

N^2-pyridyl-sulphonamido phenyl cyanamide.—Sulphapyridine (10 g.) was taken in 400 ml. of water and a slight excess of hydrochloric acid was added to it. To the hot solution 6 g. of potassium thiocyanate was added. The cyanamide was obtained by following the same procedure as was adopted in the preparation of 2 pyrimidyl-sulphonamido phenyl cyanamide, using 18 g. of basic lead acetate in 85 ml. of water, for desulphurising the thiourea formed. The cyanamide melts at 152–53° (Found: N, 19·73; $C_{12}H_{10}O_2N_4S$ requires N, 19·97 per cent.).

N^1-(N^2-pyridyl-sulphonamido phenyl)-N^5-methyl biguanide sulphate (XVII).—N^2-pyridyl sulphonamido phenyl cyanamide (3·5 g.), methyl guanidine sulphate (2·8 g.) and pyridine (25 ml.) were refluxed together for eight hours. The product was isolated and purified as under compound IV. Yellow crystals. Yield: 2 g. M.P. 180° [Found: N, 24·67 $(C_{14}H_{17}O_2N_7S)_2.H_2SO_4$ requires N, 24·75 per cent.].

N^1-(N^2-pyridyl-sulphonamido phenyl)-N^5-isopropyl biguanide sulphate (XVIII).—N^2-pyridyl sulphonamido phenyl cyanamide (4 g.) and isopropyl guanidine sulphate (2·1 g,) and pyridine (30 ml.) were refluxed for six hours. The product was isolated and purified as under compound IV. White crystals. Yield: 2·5 g. M.P. 230° *d.* [Found: N, 23·29; $(C_{16}H_{21}O_2N_7S)_2.H_2SO_4$ requires N, 23·12 per cent.].

N^1-(N^2-pyridyl-sulphonamido phenyl)-N^5-butyl biguanide sulphate (XIX).—N^2-pyridyl sulphonamido phenyl cyanamide (3·5 g.), butyl guanidine sulphate (2·0 g.) and pyridine (20 ml.) were refluxed together for eight hours. The product was isolated and purified as under compound IV. Colourless crystals. Yield: 2·5 g. M.P. 210° [Found: N, 22·44; $(C_{17}H_{23}O_2N_7S)_2.H_2SO_4$ requires N, 22·38 per cent.].

Pharmacological Investigation

Of the nineteen compounds described above, only the following five compounds (IV, VI, IX, XI and XVIII) have been screened against blood induced *Plasmodium gallinaceum* infection in young chicks. The dose followed is 10 mg. per 100 g. body-weight in aqueous solution. Of these two compounds, *i.e.*, N^1-sulphonamidophenyl-N^5-methyl biguanide hydrochloride (IV) and N^1-N^1-2-(4-methyl pyrimidyl) sulphonamido phenyl-N^5-methyl

biguanide sulphate (XI) have been found to possess marked suppressive activity.

Further pharmacological investigations are in progress and details will be published later.

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