Brit. J. Pharmacol. (1947), **2**, 271.

THE PERSISTENCE IN THE BLOOD STREAM OF SOME ANALOGUES OF SULPHADIMETHOXYPYRIMIDINE

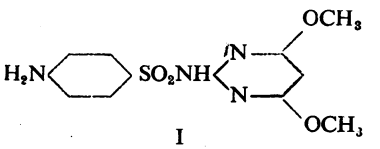
BY

A. SPINKS

From the Research Laboratories, Imperial Chemical Industries, Ltd., Hexagon House, Manchester, 9

(Received May 14, 1947)

An account has already been given of the properties of sulphadimethoxypyrimidine (I), which displays marked persistence in the blood stream after oral or parenteral administration (Gage *et al.*, 1947).



I

The availability of some of its homologues (Rose and Tuey, 1946) prompted a comparative examination of their persistence; further compounds were specially prepared by Drs. F. L. Rose, E. H. Hoggarth, and E. H. P. Young. Twenty compounds in all were examined in mice for absorption and persistence. The majority were also tested as antibacterial agents; an account of their tuberculostatic activity *in vitro* and *in vivo* will be published separately (Hoggarth, Young, and Martin, 1948). No compound appeared likely to have marked therapeutic value. Three (III, VII, and XVI), as well as sulphadimethoxypyrimidine itself, had previously been examined by van Dyke *et al.* (1945); their findings in general parallel those described here.

Experimental Section

The standard techniques used have been described in previous publications (Rose and Spinks, 1946, 1947; Gage *et al.*, 1947). Each compound was administered orally to a group of three mice, as a 1 per cent (w/v) solution of the sodium salt, or as a 1 per cent (w/v) dispersion, in doses of 250 mg./kg. It was then estimated in pooled tail blood, at standard intervals after dosing, by the micro-method of Rose and Bevan (1944). At least six groups of three mice were used for each compound (except XXI). No statistical comparison was attempted, because many compounds were so highly persistent that the maximum concentration was difficult to determine accurately in individual experiments. Values of maximum

concentration (Max.), the time after dosing at which this was attained (t max.), and the persistence in the blood (expressed as the time (C7) taken for the concentration at 7 hours to fall to two-thirds of that value), were read from the mean blood concentration-time curves.

TABLE

COMPARISON OF ANALOGUES OF SULPHADIMETHOXYPYRIMIDINE

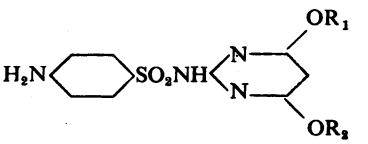
Group	Compound No.	Max. blood concentration		C7 (hours)	Number of mice
		mg./100 ml.	Time (min.)		
A	I	14.4	210	16.5	66
	II	13.8	100	15.3	18
	III	11.8	90	13.0	18
	IV	5.8	540 (?)	>17.0	24
	V	2.2	180	7.8	21
B	VI	5.7	180	10.4	21
	VII	2.1	210	17.0	18
C	VIII	4.6	150	11.0	39
	IX	2.3	210	10.5	18
	X	5.7	270	9.0	30
D	XI	12.6	90	4.7	18
E	XII	24.7	90	7.7	30
	XIII	5.7	120	8.6	27
	XIV	5.9	40	6.0	18
F	XV	13.8	60	7.5	18
	XVI	18.6	65	5.5	18
	XVII	13.7	45	7.6	18
G	XVIII	7.0	40	7.1	18
	XIX	3.5	40	*	18
	XX	0.9	40	*	21
	XXI	5.1	150	†	6

* Disappears very rapidly from the blood. † Disappears rapidly from the blood.

Results and Discussion

Compounds have been classified on the basis of their chemical structure into seven groups, each of which is considered separately; the characteristic values obtained from the mean concentration-time curves are given in the Table.

Group A. 2-*Sulphanilamido*-4 : 6-*di*-n-*alkoxypyrimidines*



Compound No.	I	II	III	IV	V
R_1	CH_3	CH_3	C_2H_5	n-C_3H_7	n-C_4H_9
R_2	CH_3	C_2H_5	C_2H_5	n-C_3H_7	n-C_4H_9

The mean concentrations of these compounds (except II) found in the blood of mice at intervals after the oral administration of 250 mg./kg., are recorded in Fig. 1 ; the characteristic values obtained from the mean curves are compared in the Table. Compounds I–IV are highly persistent ; the di-*n*-butoxy homologue (V) is fairly persistent. The other main difference between the five compounds is in maximum blood concentration, which falls with increasing molecular weight ; this effect can probably be ascribed mainly to reduced solu-

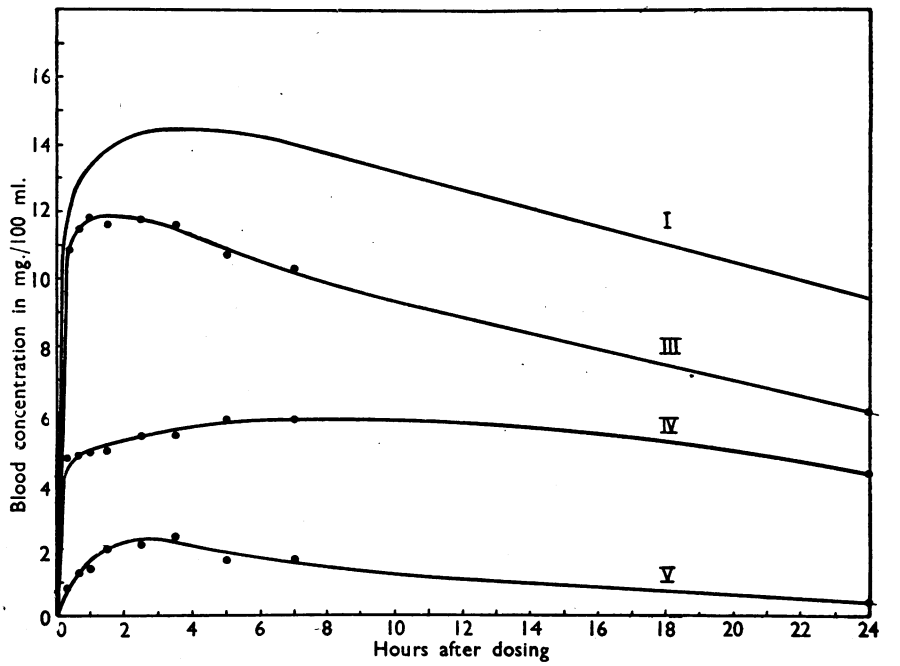
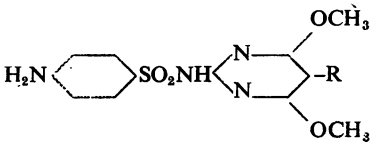


FIG. 1.—Blood concentrations in mice of sulphadimethoxypyrimidine (I), sulphadiethoxypyrimidine (III), sulphadi-*n*-propoxypyrimidine (IV) and sulphadi-*n*-butoxypyrimidine (V).

bility of the higher homologues, which would result in a decreased concentration gradient between lumen and blood stream, and therefore in decreased speed and extent of absorption. The solubility data reported by van Dyke *et al.* (1945) for I and III, and an extensive series of related compounds, support this view. The practical importance of the effect is pointed out by Hoggarth *et al.* (1948), who show that *in vitro* tuberculostatic activity increases with increasing molecular weight. It is probably the accompanying decrease in blood concentration which prevents the higher homologues from showing marked therapeutic action *in vivo*.

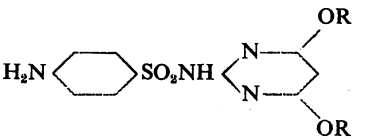
Group B. 2-*Sulphanilamido*-5-*alkyl*-4 : 6-*dimethoxypyrimidines*



Compound No.	VI	VII
R	CH_3	C_2H_5

These two compounds are both highly persistent (see Table), but attain lower maximum blood concentrations than the parent compound. The reduction in maximum concentration is greater than when the additional carbon atoms are substituted in the alkoxy group—i.e., VI and VII give lower concentrations than their isomers II and III. This was unexpected, since in other groups of compounds it has been found that the distribution of a given number of methylene groups between three, instead of two, normal alkyl radicals leads to increased blood concentrations. The effect may be due to the exceptionally low solubility of this type (van Dyke *et al.*, 1945 ; Rose and Tuey, 1946).

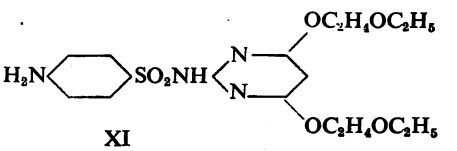
Group C. Branched chain sulphadialkoxy derivatives



Compound No.	VIII	IX	X
R	*iso*-C_3H_7	*iso*-C_4H_9	*sec*-C_4H_9

The characteristic values of the three compounds are summarized in the Table. They are all highly persistent, the butoxy compounds somewhat less so than the *iso*propoxy compound (cf. Group A). VIII and IX resemble their *normal* isomers (IV and V) in maximum blood concentration ; X gives a much higher maximum concentration than its isomers V and IX. In some other homologous series, e.g., of sulphones, it has been observed that compounds containing branched alkyl chains give much higher blood concentrations than their isomers containing straight chains. It is hoped to describe examples in future publications.

Group D. 2-*Sulphanilamido*-4 : 6-*diethoxyethoxypyrimidine*

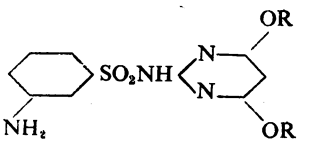


XI

The ethoxyethoxy compound (see Table) has lost much of the persistence characteristic of the other dialkoxypyrimidines, and gives much higher blood

concentrations. It should, of course, be compared with its closest analogue the di-*n*-butoxy derivative (V). The methoxyethoxy homologue was examined by van Dyke *et al.* They found that it was well absorbed, but of low persistence. Its solubility (4 mM./l. at *p*H 6.5 and 37° C.) was five times that of the dimethoxy compound (0.8 mM./l.), and about thirty times that of the diethoxy compound (0.12 mM./l.). It may be assumed that the facile absorption of these alkoxyalkoxy compounds is related to their relatively high solubility, but a different effect must cause the low persistence. The relatively low persistence of the di-*n*-butoxy compound (Group A) suggests that this effect might be connected with steric hindrance; possibly, the bulky alkoxy groups of these compounds hinder their access to some other molecule with which they must be associated to exhibit marked persistence.

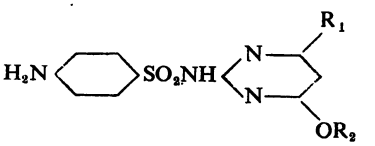
Group E. 2-*Metanilamido*-4 : 6-*dialkoxypyrimidines*



Compound No.	XII	XIII	XIV
R	CH_3	*n*-C_3H_7	*iso*-C_3H_7

The metanilamides are compared in the Table. Reference should also be made to the properties of the corresponding *para* isomers. Each metanilamide differs from its *para* isomer in giving higher maximum blood concentrations, and in disappearing more rapidly from the blood stream. Nevertheless, all three compounds are fairly persistent, approximately of the same order as sulphamerazine (Rose and Spinks, 1946).

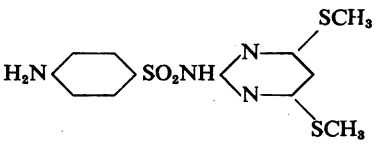
Group F. 2-*Sulphanilamido*-4-*alkoxypyrimidines*



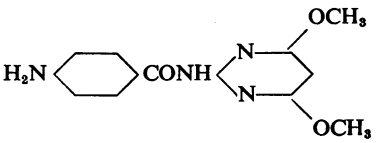
Compound No.	XV	XVI	XVII
R_1	CH_3	CH_3	*iso*-C_3H_7
R_2	H	CH_3	CH_3

The results with these three compounds are given in the Table. They are all fairly persistent, approximately of the same order as the metanilamides (Group E) and sulphamerazine, but less so than the dialkoxy derivatives. The mean curve for XVII is given in Fig. 2.

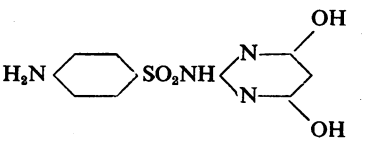
Group G. Miscellaneous compounds



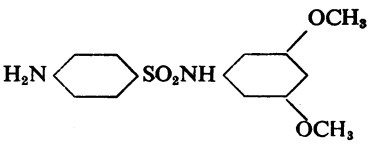
XVIII



XIX



XX



XXI

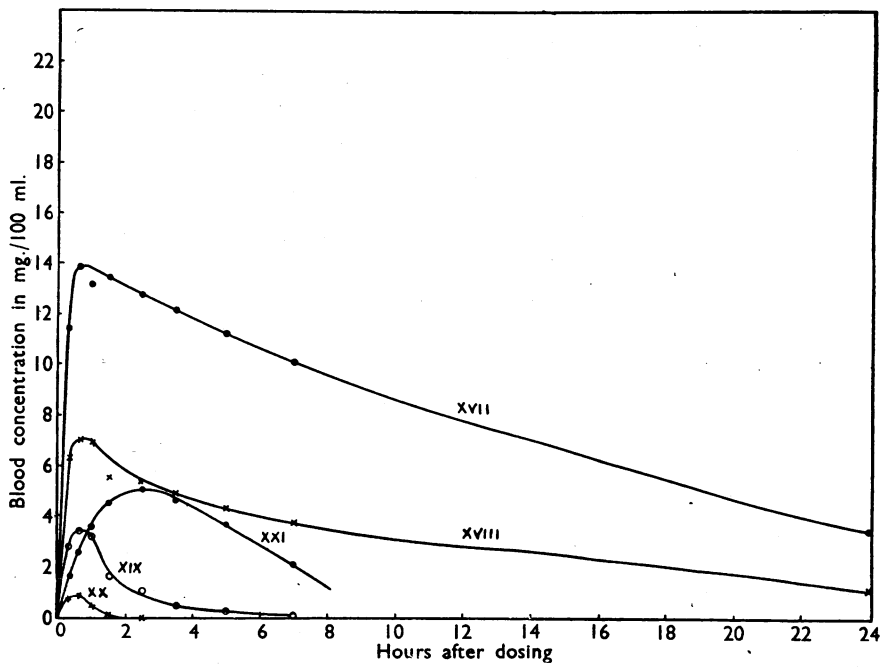


FIG. 2.—Blood concentrations in mice of sulphamethyl*iso*propoxypyrimidine (XVII), sulphadimethylmercaptopyrimidine (XVIII), 2-*p*-aminobenzamido-4: 6-dimethoxypyrimidine (XIX), sulphadihydroxypyrimidine (XX), and 3: 5-dimethoxy-1-sulphanilanilide (XXI).

These compounds show striking differences from those previously described (Fig. 2; Table). The dimethylmercapto analogue of sulphadimethoxypyrimidine is less persistent than the latter, and gives a much lower maximum concentration. The other three compounds disappear very rapidly from the blood stream. The low concentrations they attain may be associated with this rapid disappearance

from the blood rather than with poor absorption. They clearly show no resemblance whatsoever to sulphadimethoxypyrimidine, although each retains certain structural features of the latter.

Considering the results as a whole, it is clear that marked persistence in the blood stream is conferred by the presence in the sulphapyrimidine molecule of two alkoxy groups in positions 4 and 6 of the pyrimidine ring. All modifications of this structure result in reduction of persistence, including removal of one alkoxy group, transfer of the *para* amino group to the *meta* position, substitution of a benzene ring for the pyrimidine ring, or substitution of a carboxamide group for the sulphonamide group. The persistence of the dialkoxy compounds appears to fall with increasing molecular weight, although one compound (IV) is a marked exception to this rule.

Although the precise nature of the physical and physiological factors which confer persistence on a compound is at present unknown it seems probable that they include a high degree of binding to the plasma proteins and a high degree of tubular reabsorption (Fisher *et al.*, 1943 ; Beyer *et al.*, 1944 ; Earle, 1944 ; van Dyke *et al.*, 1945). The strength of the bond uniting drug to protein may also be of importance (Gregerson and Rawson, 1943 ; Rawson, 1943). Sulphadimethoxypyrimidine and some related compounds have been shown to be extensively bound to plasma proteins (van Dyke *et al.*, 1945 ; Gage *et al.*, 1947), but no information is available on the other important factors. A high degree of protein binding alone would not necessarily result in high persistence, absorption by the tubules being of equal or greater importance (Fisher *et al.*, 1943 ; Lundquist, 1945). Besides these factors, which influence the rate of excretion of a compound by the kidney, and others, less adequately investigated, which influence the rate of excretion into the intestine (*cf.* Silber and Clark, 1946), diazotizable amines are also removed from the blood stream by conversion to non-diazotizable or rapidly excreted metabolites, such as acetyl derivatives, sulphates or glucuronides. It is improbable that the persistence, even of closely related compounds, is uniformly affected by such metabolic processes.

The complexity of all these mechanisms, which influence persistence, is such that speculation on the nature of the correlation between structure and persistence is hardly justifiable. However, it is clear that the correlation is a very delicate one. It had been hoped that 4 : 6-dimethoxypyrimidine, or even *m*-dimethoxybenzene, might behave as a " conductophoric " group, and confer persistence on any molecule containing it, in the same manner as the dialkylamino-alkylamino chain of mepacrine, pamaquin, and 3349 (Magidson *et al.*, 1934, 1936 ; Spinks and Tottey, 1946), or the biguanide chain of paludrine and related drugs (Spinks, 1946, 1947), has been presumed to confer favourable pharmacological properties on the antimalarial containing it. The reduced persistence of compounds XII, XIII, XIV, XIX, and XXI strongly suggests that the introduction of 1 : 3-dimethoxy groups into a nucleus other than pyrimidine, or the combination of

4 : 6-dimethoxypyrimidine as a " conductophoric " group with a " toxicophoric " group other than sulphanilamide would be unlikely to confer high persistence on the resulting molecule.

SUMMARY

The absorption and persistence in mice of twenty compounds related to sulphadimethoxypyrimidine have been described. High persistence is a property of 2-sulphanilamidopyrimidines carrying dialkoxy groups in positions 4 and 6. Removal of one alkoxy group, transfer of the *p*-amino group to the *meta* position, substitution of a benzene ring for the pyrimidine ring, or substitution of a carboxamide group for the sulphonamide group, results in reduced persistence. In each homologous series examined, maximum blood concentration fell with increasing molecular weight.

The author wishes to thank Miss E. France for technical assistance.

REFERENCES

Beyer, K. H., Peters, L., Patch, E. A., and Russo, H. F. (1944). *J. Pharmacol.*, **82**, 239.
Dyke, H. B. van, Tupikova, N. A., Chow, B. F., and Walker, H. A. (1945). *J. Pharmacol.*, **83**, 203.
Earle, D. P. (1944). *J. clin. Invest.*, **23**, 914.
Fisher, S. H., Troast, L., Waterhouse, A., and Shannon, J. A. (1943). *J. Pharmacol.*, **79**, 373.
Gage, J. C., Martin, A. R., Rose, F. L., Spinks, A., and Tuey, G. A. P. (1947). *Brit. J. Pharmacol.*, **2**, 149.
Gregerson, M. I., and Rawson, R. A. (1943). *Amer. J. Physiol.*, **138**, 698.
Hoggarth, E., Young, E. H. P., and Martin, A. R. (1948). *Brit. J. Pharmacol.*, in the press.
Lundquist, F. (1945). *Acta pharmacol. toxicol.*, **1**, 307.
Magidson, O. J., Delektorskaya, N. M., and Lipowitsch, I. M. (1934). *Arch. Pharm.*, **272**, 74.
Magidson, O. J., and Grigorowsky, A. M. (1936). *Ber. dtsch. chem. Ges.*, **69**, 396.
Rawson, R. A. (1943). *Amer. J. Physiol.*, **138**, 708.
Rose, F. L., and Bevan, H. G. L. (1944). *Biochem. J.*, **38**, 116.
Rose, F. L., and Spinks, A. (1946). *J. Pharmacol.*, **86**, 264.
Rose, F. L., and Spinks, A. (1947). *Brit. J. Pharmacol.*, **2**, 65.
Rose, F. L., and Tuey, G. A. P. (1946). *J. chem. Soc.*, 81.
Silber, R. H., and Clark, I. (1946). *Arch. Biochem.*, **10**, 9.
Spinks, A. (1946). *Ann. trop. Med.*, **40**, 153.
Spinks, A. (1947). *Ann. trop. Med.*, **41**, 30.
Spinks, A., and Tottey, M. M. (1946). *Ann. trop. Med.*, **40**, 145.