These results indicate that amantadine HCl has a significant stimulant effect on locomotor activity in rats. In mice, amantadine appears to increase spontaneous locomotor activity only after reserpine pretreatment (Svensson & Stromberg, 1970; Stromberg, Svensson & Waldeck, 1970). The mechanism by which amantadine exerts this effect is not clearly understood. It is interesting, however, that this drug has recently been shown to resemble (+)-amphetamine in some of its actions on dopamine and noradrenaline metabolism in brain (Scatton, Cheramy & others, 1970; Stromberg & others, 1970). α-Methyl-*p*-tyrosine, a drug which is known to block amphetamine-mediated locomotor stimulation (Weissman, Koe & Tenen, 1966) also appears to antagonize amantadine-mediated excitation in rats (Fibiger, Fox, McGeer and McGeer, unpublished observations) and in reserpine pretreated mice (Stromberg & others, 1970).

We were impressed by the variability of the response to amantadine, a point also discussed by Abuzzahab (1971) both in locomotor activity and in brain dopamine concentrations.

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The Kinsmen Laboratory of Neurological Research,
Department of Psychiatry,
The University of British Columbia,
Vancouver 8, B.C., Canada.

H. C. FIBIGER
M. FOX
E. G. MCGEER
P. L. MCGEER

May 13, 1971

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An automated method for the determination of dissolution rate and urinary concentration of sulphonamides

Several methods of determining dissolution rate have been automated (Schroeter & Wagner, 1962; Niebergall & Goyan, 1963; Michaels, Greely & others, 1965). We have developed a method similar to that described by Barzilay & Hersey (1968) except that our method uses an AutoAnalyzer dialyser.

Determination of sulphonamides in blood by means of automated methods has been described by Falk & Kelly (1965) and Probst, Rehm & others (1965), the sulphonamide being diazotized and coupled according to the procedure of Bratton & Marshall (1939) on which our automated procedure is also based. We have compared manual and automated procedures in the assessment of dissolution rates and urine concentrations of sulphonamides.

Sulphathiazole was dissolved, with heat if necessary, in fresh, protein-free urine to give concentrations of 5, 10, 25, 50, 75 and 100 mg%. Also, the urine collected under normal urine conditions (Goossens & van Oudtshoorn, 1970) from subjects who had ingested 500 mg sulphathiazole (tablet) was analysed both for free and total sulphathiazole by both methods.

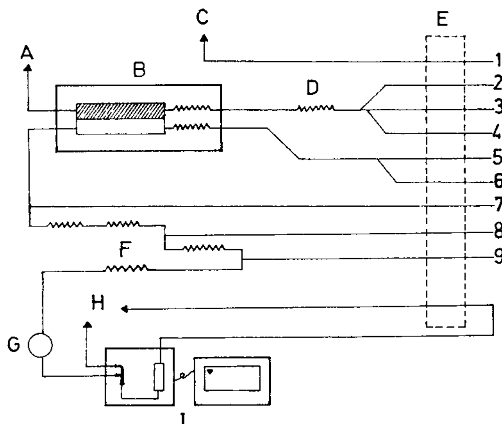

FIG. 1. Flow diagram for automated determination of sulphonamides in urine. A, Waste. B, dialyser. C, Fluid receptacle. D, Mixing coil. E, Proportioning pump. F, Mixing coils. G, 5 min time delay coil. H, Waste. I, Spectrophotometer-recorder. 1, 1% Tween 2·0 ml/min. 2, Sample 0·1 ml/min. 3,5, 0·2N HCl 1·6 and 2·9 ml/min. 4,6, Air 0·8 ml/min. 7, 0·1% $NaNO_3$ 0·6 ml/min. 8, 0·5% ammonium sulphamate 0·6 ml/min. 9, 0·1% *N*-(1-naphthyl)ethylenediamine dihydrochloride 0·6 ml/min.

Standard solutions were prepared by dissolving the sulphonamide (at about 60°) in 0·1N hydrochloric acid. For the manual procedure the Bratton and Marshall method was used, except that the coupling reagent was dissolved in 0·1N hydrochloric acid. In all cases a urine blank was read. All solutions were read in a 10 mm cell.

For the automated method a Technicon AutoAnalyzer (flow diagram Fig. 1) was connected to a Beckman DBG spectrophotometer and 10 inch recorder. The sampler was set to withdraw 20 samples/h. In every run, samples of both standard and urine solutions were pumped through, the standard samples being separated by one and the urine samples by 2 cups of distilled water. The absorption of the coloured solutions were recorded at 545 nm in a 10 mm flow cell using a dialyser temperature of 20° to obviate any hydrolysis of conjugated sulphonamides. A standard curve was plotted for every run of samples and the sulphonamide concentration of each urine sample read from this curve.

For the evaluation of the two methods, a dilution factor of 250 was used throughout with the manual method. For the excreted sulphathiazole, dilution factors were chosen to obtain absorptions between 0·2 and 0·5. Total sulphathiazole was determined after diluting the urine sample and hydrolysing with an equal volume of N hydrochloric acid.

The urinary excretion data correlate well, the biggest difference between the methods being 2·89 mg for the amount of sulphathiazole excreted in a specific period of time.

With the automated method, it is important to keep the fluid receptacle of the sampler filled with a surface-active reagent such as a 1% solution of Tween 80. This prevents high recovery of sulphathiazole from the urine, presumably because of the difference between the surface activity of the urine and that of the hydrochloric acid used in the standard solutions. Sulphamethizole and sulphamethoxazole in urine, but not sulphafurazole, could also be determined by the method.

The dissolution apparatus used resembled that of Ganderton, Hadgraft & others (1967). One 500 mg tablet in a 2 litre beaker containing 1000 ml of 0·1N hydrochloric acid, at 37°, was stirred at 100 rev/min.

For the manual method, 1 or 2 ml samples were withdrawn at 5 min intervals through a sintered glass filter (porosity 3) and the beaker volume was immediately

made up with 0·1N hydrochloric acid at 37°. After a suitable dilution the samples were analysed.

The automated procedure described was used for all but one of the sulphonamides examined, sulphafurazole, for which the volume of 0·2N hydrochloric acid fed into the lower half of the dialyser was reduced to 1·2 ml/min. After the standard solutions were run, the sampling probe was dipped into the dissolution medium, and the dissolution process started. Dissolution medium withdrawn was replaced at the same rate by 0·1N hydrochloric acid. The recording continued until the concentration of sulphonamide dissolved remained constant.

The results of 7 sets of determinations by the manual method and 6 by the automated procedure revealed that for samples from 5–100 mg% of sulphathiazole the mean $\pm$ standard deviation was at 5 mg% 5·19 $\pm$ 0·57 for the manual method and 4·80 $\pm$ 0·13 for the automated procedure, and at 100 mg% 95·44 $\pm$ 1·09 and 95·40 $\pm$ 0·78 respectively.

In dissolution rate studies good correlation of the manual and automated methods was found with the total amount of sulphonamide dissolved and with the time required for half of the total amount of sulphonamide to be released from the tablet. The automated method is superior to the manual method in that the results must be regarded as more accurate since a continuous recording of the dissolution process takes place, providing a true dissolution profile.

The automated procedure was successfully used in dissolution rate studies of tablets of sulphafurazole, sulphathiazole, sulphamethoxazole, sulphadimidine, sulphadiazine, and sulphamethizole, including a sustained release sulphamethizole tablet in a wax matrix.

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Department of Pharmaceutics,
Potchefstroom University for C.H.E.,
Potchefstroom, South Africa.

A. P. GOOSSENS.
M. C. B. VAN OUDTSHOORN

May 11, 1971

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