theophylline (see Table 2). Since these agents are known to act by increasing the level of 3′,5′-cyclic AMP in the adipose tissue although not by the same means (Butcher, 1966), it may be possible that an elevation of 3,5-cyclic AMP in the adipose tissue is the cause of the resistance.

One of us (A.B.) has found that 3′,5′-cyclic AMP (dibutyryl ester) also antagonizes *in vitro* the effect of 5CA3MP and other pyrazole derivatives.

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Interaction of aspirin with urea in water

SIR,—Previously it has been shown that urea increases the aqueous solubility of benzoic and salicylic acids (Bolton, 1963). As an extension of this, we have observed the effect of urea on aspirin solubility and stability.

Excess aspirin in water was shaken at 30° in the presence of varying amounts of urea for 5 hr. Clear aliquots were then analysed spectrophotometrically for aspirin content (Bolton, 1960). At pH 2·0, hydrochloric acid was used as a buffer and, at pH 3·5, formic acid-sodium formate. The pH of all solutions was carefully checked before and after equilibration.

Kinetic studies were made at pH values of 2·0, 2·5, 2·75, 3·0 and 3·5 at 30° ±0·2°. Formate buffers were used at pH values above 2·0 and hydrochloric acid was the buffer at pH 2·0.

The effect of urea on aspirin solubility at pH 2·0 and 3·5 is shown in Fig. 1. Although the increased solubility observed may be due to other than complexing effects, e.g. solvent effects, the solubility curve may well be described by two constants, K_1 and K_2, corresponding to the formation of 1:1 and 2:1 urea-aspirin complex species.

K_1 and K_2 can be determined graphically (Higuchi & Bolton, 1959). The values of the constants are in Table 1.

It is surprising that the values of K_1 and K_2 for the unionized and ionized complexes are of the same order of magnitude. However the calculations involved approximations as well as the neglect of other factors which may be responsible for the solubilization. The stronger solubilization of the unionized species is to be expected because of the weak basic nature of the urea molecule.

The first order rate constants from the kinetic studies are in Table 2.

Urea increases the rate of hydrolysis below pH 2·75 and decreases the rate at pH values greater than 2·75. It is interesting to note that this "crossover"

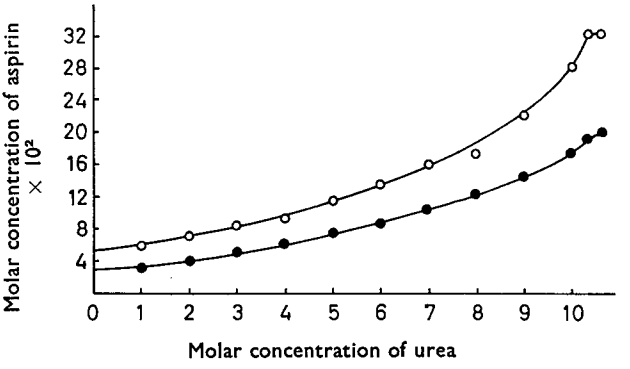

FIG. 1. Effect of urea on aspirin solubility in water at 3·0°. ○, pH 3·5; ●, pH 2·0.

TABLE 1. APPARENT EQUILIBRIUM CONSTANTS FOR UNIONIZED ASPIRIN-UREA AND IONIZED ASPIRIN-UREA INTERACTIONS

Complex		K
Unionized aspirin urea	(1:1)	0·186
,, ,, ,,	(1:2)	0·188
Ionized aspirin urea	(1:1)	0·072
,, ,, ,,	(1:2)	0·114

TABLE 2. RATE CONSTANTS FOR HYDROLYSIS OF ASPIRIN IN THE PRESENCE OF UREA AT 30° ($k = hr^{-1} \times 10^{-3}$)

Urea concentration M	pH				
	2·0	2·5	2·75	3·0	3·5
0	3·2	4·8	6·4	8·8	14·5
2	3·6	4·8	5·6	7·9	13·3
4	3·7	5·2	5·6	7·0	11·9
8	4·1	6·0	5·8	6·6	10·0

occurs at a pH corresponding to the pH of maximum stability as reported by Edwards (Edwards, 1950). This pH may thus represent a point where the hydrolysis mechanism changes, and could provide an explanation for the change in the effect of urea. Since the sites and mechanism of the interaction are not defined, any presentation of possible reasons for this effect on the basis of the present data would be highly speculative.

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