Duration of effect of atropine sulphate against the toxicity of oxotremorine in mice

The central actions of oxotremorine may be blocked with atropine sulphate but not with methylatropine (Lévy & Michel-Ber, 1966). Mice excrete 38% of a dose of [^{3}H]oxotremorine within 2 h of administration (Hammer, Karlén & others, 1968). Within 48 h of injection of [^{14}C]atropine sulphate, however, mice excrete 80–90% of the ^{14}C (Albanus, Sundwall & others, 1968). This information indicates the duration of residence of the radioactive atom in the body, but not that of the biologically active compound. To assess the duration of effect of atropine sulphate in mice, therefore, the duration of its protection against the lethal effects of oxotremorine, a compound which stimulates peripheral and central receptors sensitive to atropine blockade, was examined.

Female albino mice, CF No. 1 strain, 18–24 g, bred in these laboratories were divided into groups of 10. All drugs were dissolved in physiological saline and administered in a dose volume of 1 ml/100 g body weight. A dose-mortality curve to intravenously injected oxotremorine sesquifumarate (Aldrich Chemical Co., Inc.) was determined from which the LD99 (maximum lethal dose) was estimated to be 30 mg/kg. Further groups of mice were then injected intraperitoneally with graded doses of atropine sulphate, B.P. and challenged at measured time intervals thereafter with the intravenous LD99 of oxotremorine. The number of mice surviving at each dose level of atropine at the different time intervals was recorded after 24 h (Table 1), and was found to be inversely related to the time interval between atropine pretreatment and the oxotremorine challenge at each dose level of atropine. Plotting % survivors on the ordinate against time of pretreatment on the abscissa produces a series of parallel lines, which are displaced to the right as the dose of atropine is increased. The duration of protection of 50% of a group of mice (PT50) was calculated and found to be a sigmoid function of the $\log_{10}$ dose of atropine. Thus, the *duration* of protection of mice against the lethal effects of oxotremorine by atropine is a function of the dose.

Table 1. *Duration of protection of mice by atropine sulphate against the lethal effects of oxotremorine sesquifumarate* (30 mg/kg i.v.)

Atropine sulphate i.p. (mg/kg)	% surviving at different time intervals after treatment (min)										PT50 (min)
	30	60	90	120	150	180	210	240	270	300	
1·25	90	60	0	—	—	—	—	—	—	—	59·4
2·50	100	70	10	0	—	—	—	—	—	—	67·5
5·0	100	100	60	20	0	—	—	—	—	—	97·5
10·0	100	100	—	—	—	90	60	0	—	—	210·0
20·0	100	100	100	90	—	90	90	100	30	10	252·6

Kords, Lüllmann & others (1968) reported the degree of protection of mice by atropine sulphate injected 15 min before acutely toxic doses of di-isopropylfluorophosphonate to be unrelated to the dose of the cholinoceptor blocking agent. The results of the experiments now reported show that, even at the lowest doses examined, atropine sulphate may confer 100% protection against the LD99 of oxotremorine, providing the time interval between its injection and pretreatment with atropine is sufficiently short.

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REFERENCES

ALBANUS, L., SUNDWALL, A., VANGBO, B. & WINBLADH, B. (1968). *Acta pharmac. tox.*, **26**, 571–582.

HAMMER, W., KARLÉN, B., RANE, A. & SJÖQVIST, F. (1968). *Life Sci.*, **7**, 197–204.

KORDS, H., LÜLLMANN, H., OHNESORGE, F. K. & WASSERMANN, O. (1968). *Europ. J. Pharmac.*, **3**, 341–346.

LÉVY, J. & MICHEL-BER, E. (1966). *Bull. Acad. natn. Méd.*, **150**, 215–218.

When is a drug inactive? Concerning the uricosuric activity of some anti-inflammatory drugs

Many acidic drugs are extensively bound to plasma proteins and it has been generally accepted that the bound fraction has no pharmacological activity (Goldstein, 1949; Brodie, 1965). This concept certainly needs modifying in the light of recent reports that one drug may displace another from a common binding site on the albumin molecule; for example, salicylate and phenylbutazone can displace thyroxine and certain sulphonamides, sulphonylureas and oral anticoagulants (Solomon & Schrogie, 1967; Meyer & Guttman, 1968).

Such an interaction between two drugs *in vivo* can be formulated as follows:

Species A + Albumin–Species B combination $\rightleftharpoons$ Species B + Albumin–Species A. Thus one drug (A) may have an "adjuvant," or potentiating, action on the pharmacological activity of another drug (B) by either increasing its effective concentration (as unbound drug B) or otherwise making it more readily available to its responsive receptors. Drug A would, however, acquire and demonstrate this adjuvant activity only when it was itself bound to the albumin. We wish to extend and further illuminate this concept that when albumin-bound, a drug entity may "acquire" pharmacological activity, which need not necessarily be that of enhancing the activity of another drug or shortening its biological half-life.

Experiments in our clinic showed that the oral ingestion of certain drugs by healthy adult volunteers significantly lowered, in a reversible manner, the capacity of the plasma proteins to bind uric acid (Bluestone, Kippen & Klinenberg, 1969). Most of this urate-binding capacity (*ca* 70%) is associated with the albumin fraction. When the urate-binding capacity of human albumin preparations both in the presence and absence of added drugs *in vitro*, was measured (Table 1) it was found that a number of anti-inflammatory acids effectively inhibited urate binding to human albumin *in vitro*, and that in man, aspirin and phenylbutazone could probably displace uric acid from its albumin binding site(s) *in vivo*.

These observations suggested that phenylbutazone and salicylates are useful uricosuric drugs because they displace some of the albumin bound urate and so augment its renal clearance. When albumin bound, phenylbutazone and salicylate may well be inactive as analgesics, antipyretics or anti-inflammatory agents; but at the same time they could also have a uricosuric effect, quite apart from any other uricosuric activities they might have when not bound to albumin, e.g., acting directly on the kidneys. In fact it may now be helpful to subdivide drugs, currently classed as "uricosurics" according to whether they (*a*) can inhibit tubular reabsorption of urate, (*b*) can displace urate from its binding sites in plasma or other tissues, or