The potency was measured as the intravenous dose which produces 50% inhibition of brain cholinesterase activity in 150 s (the ID50). Table 1 shows for each compound the values for k_i, *Rm* and the ID50.

TABLE 1. *Bimolecular rate constants (k_i) for rat brain cholinesterase, the partitioning characteristics (Rm) and the doses of organophosphorus compounds which inhibit brain cholinesterase activity by 50% (ID50) in the anaesthetized rat at 150 s after intravenous injection*

Compound	k_i (in M^{-1} min^{-1})	ID50 (in μmol/kg)	*Rm*	log (k_i . ID50)
Monocrotophos	$1{\cdot}0\times10^4$	25·83	−0·79	5·41
Dichrotophos	$1{\cdot}3\times10^4$	8·76	−0·66	5·06
Mevinphos	$95{\cdot}6\times10^4$	0·51	−0·53	5·69
Dichlorvos	$9{\cdot}1\times10^4$	11·74	−0·29	6·03
Paraoxon	$758{\cdot}8\times10^4$	0·31	−0·27	6·37
Crotoxyphos	$166{\cdot}5\times10^4$	3·00	−0·18	6·70
WL 22864	$70{\cdot}9\times10^4$	2·94	−0·12	6·32
SD 14045	$48{\cdot}6\times10^4$	6·36	−0·09	6·49
Chlorfenvinphos	$37{\cdot}3\times10^4$	28·77	+0·16	7·03

The experimental data could be related by the equation:

$$\log (k_i \text{ ID50}) = 1{\cdot}94\ Rm + 6{\cdot}71$$
$$(\pm 0{\cdot}27) \quad (\pm 0{\cdot}12)$$

The correlation coefficient of the resulting straight line was 0·939 ($n=9$).

The relationship includes a rate constant which implies time dependence, demonstrating that the pharmacological responses *in vivo* during the pre-steady state are related to the physico-chemical characteristics of the organophosphorus compounds. This is in contrast to the steady-state relationships which have often been used as models to correlate chemical structure with biological activity (compare Hansch & Fujita, 1964; Fujita & Nakajima, 1969).

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The release of ^{14}C-glycine from electrically stimulated rat spinal cord slices

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Although there is now much electrophysiological evidence that glycine and γ-aminobutyric acid (GABA) are inhibitory transmitters in the central nervous system (Curtis, Hösli & Johnston, 1968; Krnjević & Schwartz, 1967), it has proved difficult

to demonstrate changes in the release of these amino-acids from the brain after nerve stimulation. This difficulty may be due to the efficient uptake mechanisms for glycine and GABA which are present in the spinal cord and brain (Neal & Pickles, 1969; Iversen & Neal, 1968). Electrical stimulation of cortical slices and the cerebral cortex *in vivo* has recently been shown to increase the release of GABA from the cortex (Srinivasan, Neal & Mitchell, 1969; Iversen, Mitchell, Neal & Srinivasan, 1970), but the release of glycine from the spinal cord has not yet been demonstrated. In the present experiments, the release of ^{14}C-glycine from slices of spinal cord has been studied.

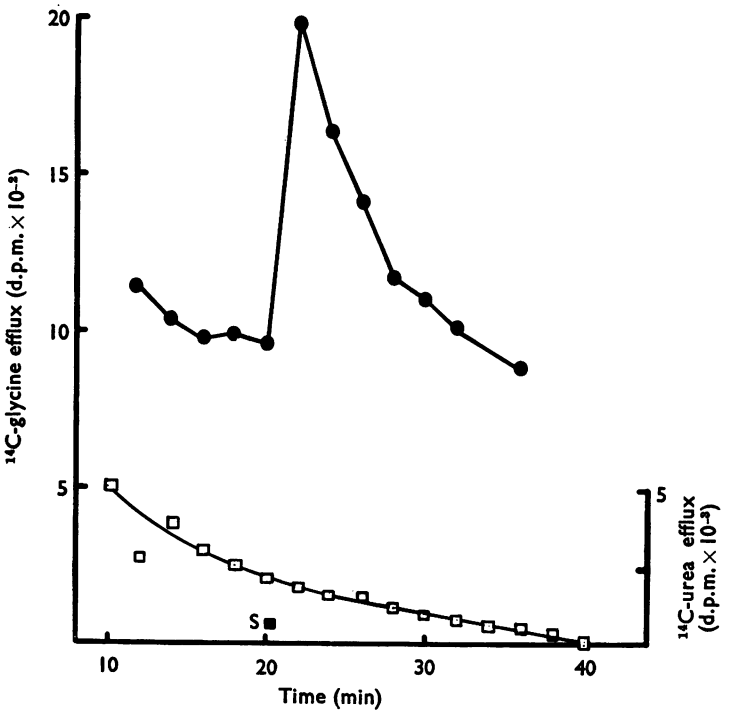

FIG. 1. Effect of electrical stimulation (S) on the efflux of ^{14}C-glycine (●) and ^{14}C-urea (□) from spinal cord slices. For glycine, each point is the mean of eight results; for urea, two results. Efflux shown as disintegrations per min × 10^{-2} (d.p.m. × 10^{-2}).

Slices of spinal cord (1 mm thick) were incubated with ^{14}C-glycine (specific activity 41·2 mCi/mmol, 6×10^{-7}M) at 37° C for 30 min in 10 ml of oxygenated Krebs-bicarbonate Ringer. The tissue was superfused in a small vessel (volume 0·75 ml) at a rate of 0·5 ml/min. The superfusate was collected into test tubes which were changed every 2 min. Aliquots (0·2 ml) were removed and the radio-activity was measured by liquid scintillation counting.

In two experiments in which spinal cord was incubated for 40 min, tissue extracts were subjected to paper chromatography, and it was found that 98% of the radioactivity present was unchanged ^{14}C-glycine. The efflux of radioactivity was therefore taken as a measure of glycine release.

After the first two or three collection periods, there followed a steady spontaneous efflux of ^{14}C-glycine from spinal cord. After 15–20 min superfusion, the tissue was

stimulated by rectangular pulses (60 Hz, 20 mA, 5 ms) for 30 s. This caused a significant increase in the efflux of radioactivity, as illustrated in Fig. 1. The maximum increase was 2·3 times the resting efflux (mean of eight experiments), and occurred during the period of stimulation. The increased efflux of glycine was not a non-specific effect on the cell membrane, as electrical stimulation did not cause an increased release of ^{14}C-urea from cord slices (Fig. 1). Although *in vitro* experiments in which the release of substances from nervous tissue after electrical stimulation must be interpreted with caution, these results are not inconsistent with a neurotransmitter role of glycine.

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The pharmacological effects of ouabain administered intracerebrally to conscious mice

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In certain species, cardiac glycosides may release endogenous catecholamines, which act on the myocardium to produce a positive inotropic effect (Tanz, 1967). Also, the lethal effects of ouabain in guinea-pigs are brought about partially by a liberation of catecholamines (Hermansen, 1970). The pharmacological effects of these glycosides on the central nervous system have not been examined fully; we have studied the pharmacological effects of ouabain administered by direct intracerebral injection.

Ouabain (0·1 to 0·4 μg in 10 μl) was injected into the cerebral ventricles of conscious male TO mice, using the method of Brittain & Handley (1967). These injections were followed by a central nervous depression lasting 2–3 h and characterized by a loss of locomotor activity, lowered body posture and lack of response to external stimuli. Ouabain was not anti-convulsant, but predisposed mice to electrically-induced convulsions. There was a whole-body hypothermia which reached a maximum fall of 11° C at 90 min. This hypothermia, together with the other effects, was dose dependent. The hypothermia was accompanied by peripheral vasodilatation associated with a transient rise in skin temperature. Higher doses of ouabain (1–100 μg) induced convulsions and death in at least 80% of the animals. The effects appeared to be centrally mediated, for doses of up to 20 μg subcutaneously produced no behavioural depression and only slight transient hypothermia of about 3° C fall at 30 min.