THE ACTION OF ANTICHOLINESTERASES ON SPINAL REFLEXES FOLLOWING INTRA-ARTERIAL INJECTION

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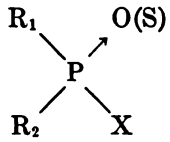
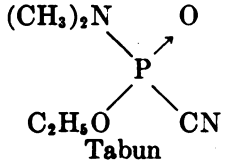
In investigations of the metabolism of acetylcholine (ACh) and the problems of transmission, various cholinesterase (ChE) inhibitors have played a major role. The arrival of the irreversible ChE inhibitors about ten years ago meant even better tools were available for elucidating biochemical and physiological problems in this respect. Every irreversible ChE inhibitor known until now belongs to the group of organo-phosphorus compounds. The following general formula can be assigned to them (11):



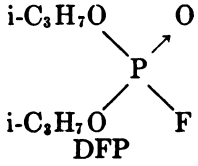
X = organic or inorganic acid residue.

Type A. R_1 = substituted amine.
R_2 = alkoxy group.



Tabun

Type B. $R_1 = R_2$ = alkoxy group.



DFP

The first of these compounds to be released from military secrecy was DFP, diisopropylfluorophosphate, with which many physiological and pharmacological investigations have been undertaken. Several other compounds, where R_1 and R_2 are alkoxy groups, have also been studied. These compounds, as a common feature, show a greater affinity for the non-specific ChE, although they are very powerful inactivators also of the specific type of enzyme.

Another group of organo-phosphorus compounds differs in containing N-P bonds and is substituted with amines, R_1 = substituted amine, R_2 = alkoxy. The best known member of this group is tabun which was produced on a technical scale to be used as a war gas (1). With respect to enzyme inhibition this group of ChE inhibitors generally shows about the same affinity for both groups of enzymes. This has been shown to occur in vitro (6) and it is also reflected in the recovery curves of the enzymatic activity in the blood, as could be demonstrated in dogs (5). In pharmacological experiments the compounds containing the N-P bond behaved like typical ChE inhibitors on all peripheral organs.

An investigation of the action of these compounds on central synapses then seemed pertinent, and a technique for intra-arterial injections into the spinal

cord was developed. The method has been described in detail by Holmstedt and Skoglund (7). Briefly, it consists in the injection of the drugs into the aorta, with all arteries excepting the lumbar arteries clamped off, so that the bloodstream carries the injected drug to the spinal cord. The ventral root potentials are recorded during stimulation of either the dorsal roots or afferent nerves. The spinal cord was used because of the accurate data available on the normal excitability characteristics of the different spinal systems, (10) and the fact that Koelle (8, 9) has localized specific ChE in cell bodies, axons and dendrites of the ventral and lateral horn cells in the cat's spinal cord. The high affinity of tabun for the latter enzyme (5), made us consider tabun as an adequate tool for investigating the synaptic processes of the cat's spinal cord.

The results obtained differed among various ChE inhibitors. A repetitive discharge of the motor cells could be demonstrated to occur immediately after an injection of tabun, as was previously assumed by Chennels, Floyd and Wright (2) with DFP. When mono- and multisynaptic responses were recorded after the injection of tabun, the often observed characteristic facilitation of the multisynaptic response was seen. In contrast to this, the monosynaptic spike showed successive depression. The depression was not a phenomenon secondary to the increased activity of the multisynaptic system; this was ruled out by studying the different types of reflexes separately by alternative stimulation of various afferent nerves.

When DFP was injected under similar circumstances, one could not obtain the primary inhibition of the monosynaptic reflex; instead, both mono- and multisynaptic reflexes increased simultaneously. Tabun and DFP consequently had identical actions on the multisynaptic flexor reflexes, whereas tabun caused an initial depression of the monosynaptic extensor reflex not seen after DFP. In higher doses both compounds abolished both types of reflexes. To explain these observations on the basis of the preference of the various groups of inhibitors for the two types of ChE is at present difficult to conceive, since the non-specific enzyme, according to Earl and Thompson (4), apparently plays some part in the maintenance of the myelin sheath rather than directly in conduction or transmission. The "moral" is that the common feature of the irreversible ChE inhibitors, to inhibit ChE in vitro, does not explain the differential action of various members of the group on the central synapses.

Further confirmation of this conclusion has recently been obtained in the experiments of Diamant (3), who injected various ChE inhibitors into the vestibular nuclei. Using among others tabun and DFP and injecting the compounds through the vertebral artery in guinea pigs, he studied circling behavior, nystagmus and other signs of disturbed vestibular function. Simultaneous determinations of the ChE content in various parts of the brain were performed. He found that various inhibitors provoked a vestibular syndrome at different degrees of ChE inhibition in the part of the brain where the vestibular nuclei are located.

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