

ACETYLCHOLINESTERASE INHIBITORS ARE USED AS DRUGS AND POISONS

The therapeutic and toxic properties of acetylcholinesterase inhibitors are of considerable practical importance. *Physostigmine* (also called eserine) is an alkaloid derived from the Calabar bean, which was once used as an ordeal poison in witchcraft trials. Physostigmine and related inhibitors such as neostigmine are *carbamoyl esters* (Figure 39-30). They inhibit acetylcholinesterase by forming a covalent intermediate that is hydrolyzed very slowly. The serine residue at the active site becomes carbamoylated. *This carbamoyl-enzyme intermediate is subsequently hydrolyzed at a very slow rate, in contrast with the acetyl-enzyme intermediate normally formed when acetylcholine is the substrate.* Thus, the active site of the enzyme is effectively blocked. Neostigmine is used to treat glaucoma, an eye disease characterized by abnormally high intraocular pressure. The therapeutic rationale is that neostigmine inhibits acetylcholinesterase and thereby enhances the effects of acetylcholine.

"BIOCHEMISTRY"
3RD EDITION
LUBERT STRYER
W.H. FREEMAN
p. 1023-4

See p. 196
4th Edition
and p 324

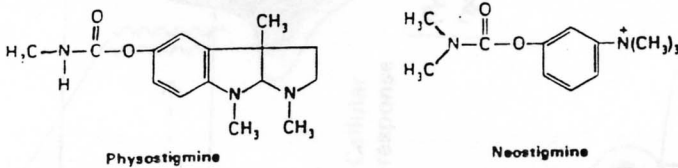


Figure 39-30
Physostigmine and neostigmine inhibit acetylcholinesterase by carbamoylating the serine in the active site.

Even more potent inhibitors are the *organic fluorophosphates*, such as diisopropyl phosphofluoridate (DIPF). These compounds react with acetylcholinesterase to form *very stable covalent phosphoryl-enzyme complexes* (Figure 39-31). The phosphoryl group becomes bonded to the active-site serine, as in serine proteases that have reacted with DIPF (p. 223). Many organic phosphate compounds have been synthesized for use as *agricultural insecticides* or as *nerve gases* for chemical warfare (Figure 39-32). These compounds kill by causing respiratory paralysis. Tabun and sarin are among the most toxic of fluorophosphates. Parathion has been widely used as an agricultural insecticide.

The number of acetylcholinesterase molecules in the end plate of mouse-diaphragm muscle has been counted using radioactive DIPF as a label. The density is $12,000 \mu\text{m}^{-2}$, only a factor of 2 less than the density of acetylcholine receptors. Thus, *the postsynaptic membrane is very densely packed with both acetylcholinesterase and the acetylcholine receptor.* Only a small fraction of the acetylcholinesterase is required for the transmission of nerve impulses at low frequencies. However, most of the enzyme molecules must be active to sustain transmission at high rates of firing.

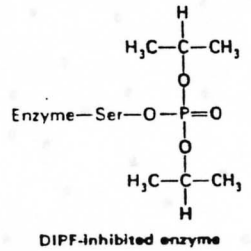


Figure 39-31
DIFP-inhibited acetylcholinesterase.

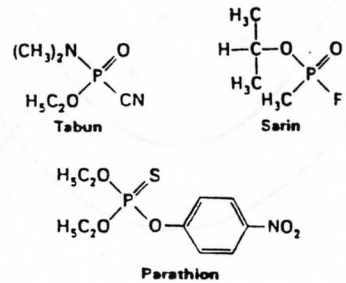


Figure 39-32
Organic phosphate inhibitors of acetylcholinesterase.

INHIBITORS OF THE ACETYLCHOLINE RECEPTOR

Neuromuscular transmission can also be impaired by *compounds that act directly on the acetylcholine receptor.* Curare has been used for centuries by South American Indians. Soon after Columbus returned, d'Anghera wrote in his *De Orbe Novo* that "the natives poisoned their arrows with the juice of a death-dealing herb. . . ." One of the active components of curare is *d-tubocurarine* (Figure 39-33). *Tubocurarine inhibits the depolarization of the end plate by competing with acetylcholine for binding to the acetylcholine receptor.* α -Bungarotoxin and cobra toxin have a similar action.

"Sarin" used
in Tokyo
subways

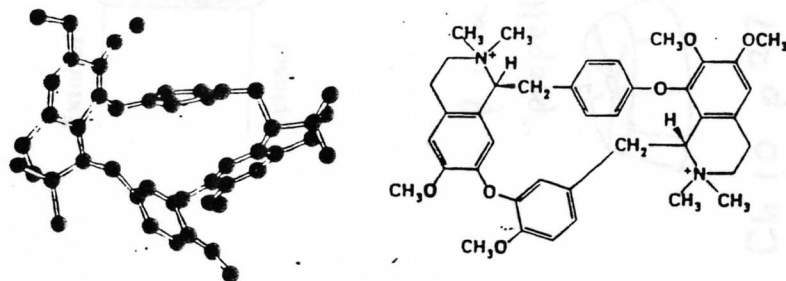


Figure 39-33
Formula and model of *d-tubocurarine*.