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Dec. 2.

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### Pseudo-Cholinesterase in the Central Nervous System of the Frog

A RECENT reference in the literature<sup>1</sup> suggests that there may be no pseudo-cholinesterase in the central nervous system of the frog. The conclusion is based on two kinds of evidence: (1) with Warburg manometric technique the tissue did not split significant quantities of benzoylcholine; and (2) there was evidently no splitting of butyrylthiocholine when the tissue was examined histochemically by Koelle's<sup>2</sup> modified technique. Our own evidence suggests that this conclusion is incorrect.

Many studies<sup>3</sup> have shown that benzoylcholine is for many species, notably the ruminants, not a suitable substrate for the demonstration of pseudo-cholinesterase. Butyrylcholine is for this reason the substrate to be preferred in comparative studies. Using butyrylcholine, one of us (D. C. H.) has found that frog tissue (homogenate of brain, spinal cord and sciatic nerve) can split butyrylcholine, and the results clearly suggest the presence of both true and pseudo-cholinesterase (see Table 1).

Table 1

Tissue	Substrate (10 mM)	μl./hr./mgm. tissue	
		No inhibitor	DFP 10 <sup>-6</sup> M
Brain	Acetylcholine	14.2	12.9
	Butyrylcholine	3.5	0
Spinal cord	Acetylcholine	18.4	14.6
	Butyrylcholine	7.3	0
Sciatic nerve	Acetylcholine	1.2	1.3
	Butyrylcholine	about 0.7	0
Mixed nervous tissue*	Acetylcholine	12.0	10.9
	Butyrylcholine	2.7	0

\* Mixed tissue = combined brain, spinal cord and sciatic nerve.

Moreover, as mentioned earlier<sup>4</sup>, it can be shown that sections of frog retina split butyrylthiocholine when the incubation system is that first described by Koelle<sup>5</sup>. Here, and in the spinal cord of the frog, the quantity of enzyme splitting butyrylcholine seems to be small; but it is demonstrable. An interesting finding is that both true and pseudo-cholinesterase may be present in the supporting cells of the optic nerve trunk.

These results underline the care required in attempting to characterize an enzyme by its activity towards one substrate only; this is particularly true of the use

of benzoylcholine as a test for pseudo-cholinesterases, the specificity pattern of which varies markedly from species to species.

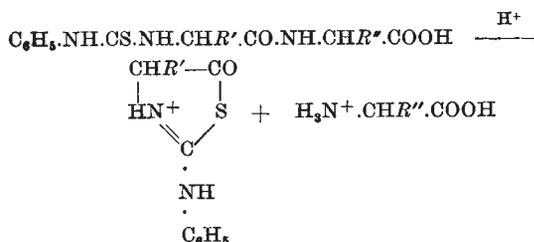
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### Mechanism of the Phenyl Isothiocyanate Degradation of Peptides

A STUDY of the mechanism of the phenyl isothiocyanate degradation of peptides<sup>1</sup> has made necessary a revision of our earlier formulation of this reaction. It has been found that the compound initially formed in the acid-catalysed cleavage of a phenyl thiocarbonyl peptide is not the expected 3-phenyl-2-thiohydantoin derivative of the N-terminal amino-acid, but instead the isomeric 2-anilino-5-thiazolinone.



This intermediate is clearly distinguishable from the thiohydantoin by its ultra-violet absorption curve (Fig. 1) and by its behaviour on paper chromatography.

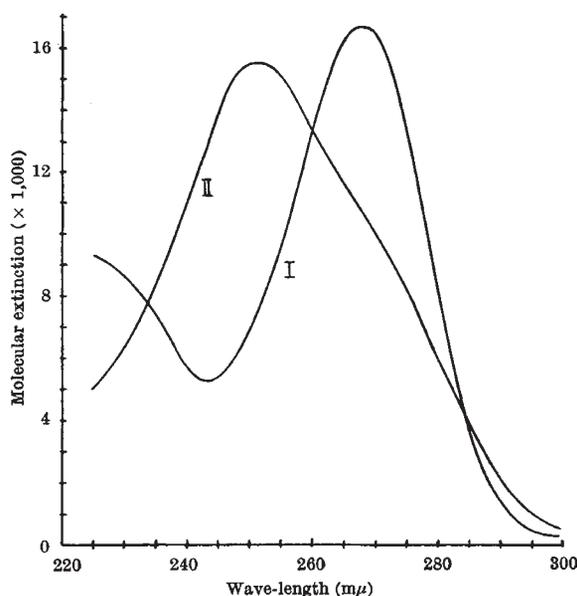


Fig. 1. Ultra-violet absorption curves of 5-isobutyl-3-phenyl-2-thiohydantoin (I) and 2-anilino-4-isobutyl-5-thiazolinone (II) in ethanol