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Distribution of the Adrenaline-forming Enzyme in the Adrenal Gland of a Snake, *Xenodon merremii*

CHROMAFFIN cells are present in two separate loci in the adrenal gland of the snake, *Xenodon merremii*. Most are distributed within a homogeneous ribbon on the periphery of the gland; these cells contain[1] and produce[2] large amounts of noradrenaline but little adrenaline. The remainder of the chromaffin cells are scattered among the adrenal cortical cells in the central portion of the gland. These cells contain large amounts of the methylated amine, adrenaline[1].

The conversion of noradrenaline to adrenaline is catalysed by an enzyme, phenylethanolamine-*N*-methyl transferase (PNMT)[3]. In mammals almost all of this enzyme is found in the adrenal medulla, where its activity[4] and synthesis[5] are stimulated by the adrenocortical steroids which perfuse this organ in high concentrations. The observation that, in *Xenodon*, there is a high concentration of adrenaline in the central chromaffin cells suggested that PNMT in this species might also be controlled by cortical hormones, so we examined the regional distribution of this enzyme in the adrenals of *Xenodon*.

Four snakes were captured in Argentina in March, 1966. The adrenal glands were dissected into central and peripheral portions, and parts of each were sent to the United States for biochemical analysis. PNMT was assayed by methods described previously[5], except that phenylethanolamine (3×10^{-3} molar) was used as substrate, and the ^{14}C-N-methyl-phenylethanolamine formed enzymatically was extracted into a mixture of toluene and isoamyl alcohol (97 : 3). The assay depends on the transfer of a ^{14}C-methyl group from ^{14}C-S-adenosylmethionine to the amine nitrogen. The resulting radioactive product (N-methyl-phenylethanolamine) is separated from the labelled cofactor by extraction into the organic solvent at *p*H 10.

An average of 9 ± 2 mμmoles of N-methyl-phenylethanolamine was formed by peripheral chromaffin tissue while central tissues were able to synthesize 104 ± 28 mμmoles of the methylated product. Because only a small percentage of the cells in the central zone are chromaffin cells (most are cortical cells which produce steroids), it is likely that the concentration of PNMT in the central chromaffin cells is even greater than ten times that of the cells in the periphery.

Synthesis of adrenaline in the frog, *Rana pipiens*, is catalysed by a variant of mammalian PNMT, which has the same substrate specificity but different physical properties[6]. This enzyme does not appear to be controlled by adrenocortical steroids: unlike the mammalian enzyme its activity does not decline after hypophysectomy[6]. It is possible that snake PNMT is also independent of hormonal control, and that its high degree of localization in the chromaffin cells which are surrounded by cortical tissue is simply a coincidence. This hypothesis could be tested by determining whether central adrenal PNMT declines when the snake is hypophysectomized, or if the peripheral enzyme increases following treatment with large doses of glucocorticoids. A more likely explanation for the distribution of PNMT within the adrenal would be that in reptiles (like mammals, but unlike amphibians) this enzyme is under hormonal control. We suggest that all chromaffin cells in the snake adrenal are potentially able to make adrenaline but that they actually do so only after they have been stimulated chronically with large amounts of adrenocortical steroids.

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