From these results, it was concluded that D-amino acid oxidase apo-protein can also be crystallized by combining with equimoles of *p*-nitrobenzoic acid and coenzyme in the same way as in the case of benzoate. This fact may be attributed to the low specificity of this enzyme.

Furthermore, it seems clear that the enzyme can be easily crystallized by combining with a suitable compound as 'substrate-substitute' which can combine with the protein at the same binding sites as the substrate, but not be oxidized by the enzyme. The crystal of the complex of the enzyme and such a 'substrate-substitute' may provide a valuable clue in elucidating the actual features of enzyme-substrate compounds.

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PHYSIOLOGY

Persistence of Sterols other than Cholesterol in Chicken Tissues

RECENT work by Davison and his collaborators[1-3] has shown that cholesterol may be incorporated in the nervous tissues of myelinating chickens and rabbits and that this cholesterol will persist in these tissues for considerable lengths of time. Our interests in the metabolism of cholesterol and other sterols prompted us to compare the distributions in chicken tissues of injected sitosterol and lanosterol with that of cholesterol. Emulsions of cholesterol-7-^{3}H (purchased from New England Nuclear Co., Boston, Mass.), sitosterol^3-H (randomly labelled, gift of Dr. Leon Swell), and lanosterol-^{3}H (randomly labelled, gift of Dr. George J. Alexander) in saline were prepared by the method used for preparation of cholesterol-^{14}C emulsions for experiments involving mitochondrial oxidation of this sterol[4]. Newly hatched White Leghorn chicks were used and the appropriate sterol emulsion (0·50 ml.) was injected directly into the yolk sac. After four days, radioactivity could be detected in the brain tissue of chickens from each group. After 9 weeks the one surviving chicken in each group was killed by exsanguination. The blood was collected in centrifuge tubes and the erythrocytes separated. All the brain, cord, liver, spleen and aorta was excised, weighed and dissolved in strong potassium hydroxide. The non-saponifiable fraction was assayed for tritium radioactivity in a liquid scintillation counter. Small pieces of sciatic nerve were taken from each chicken and all showed slight but consistently detectable radioactivity. Other findings are shown in Table 1.

It is of interest that all three sterols were incorporated into brain tissue. The rather large incorporation into erythrocytes is also noteworthy as is the small but persistent incorporation of radioactivity into the aortic tissue. That a small percentage of orally ingested cholesterol will be deposited in the aorta of adult rabbit[5] or man[6] has been shown. The relative persistence of lanosterol-^{3}H in the liver could indicate greatly reduced turnover as compared with the other two sterols. In view of reports that some insects and mammals may convert sitosterol and ergosterol to cholesterol or cholesterol-like compounds[7-10], it was necessary to prove the presence of the injected material. Gas chromatography of the non-saponifiable fractions from the brains and livers of all three chickens showed the presence of only the injected sterol and cholesterol. (We are greatly indebted to Dr. W. M. Holmes of Smith, Kline and French Laboratories, Philadelphia, for these analyses.)

Table 1. RECOVERIES OF TRITIUM RADIOACTIVITY FROM NON-SAPONIFIABLE FRACTIONS OF CHICK TISSUES NINE WEEKS AFTER INJECTION OF CERTAIN STEROLS

Sterol	Recovery (per cent)		
	Cholesterol-^{3}H	Sitosterol-^{3}H	Lanosterol-^{3}H
Input (c.p.m.)	$4·3 \times 10^5$	$8·2 \times 10^4$	$8·7 \times 10^5$
Brain	0·60	0·76	0·28
Cord	Lost	N.D.*	0·02
Erythrocytes	0·33	0·24	0·49
Liver	0·40	0·25	1·07
Spleen	0·14	N.D.*	0·35
Aorta	0·02	0·18	0·13

* N.D., not detectable.

Work with larger groups of chickens is in progress. Preliminary results are consistent with those presented here. Experiments involving more accurate characterization of the labelled sterols as well as autoradiography of specific tissues are under way.

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Effect of Sheep Pituitary Extract on Hypercholesterolæmia in Intact and Hypophysectomized Rats

THE prolonged administration of cortisone or adrenocorticotrophic hormone (ACTH) has sometimes been found to cause a change in the concentration of cholesterol in serum in human beings and in experimental animals. While some workers[1,4] report that they have found such an elevation, others have noted no effect[7]; still others have reported the opposite[8].

A study of endocrine factors influencing the cholesterol[9] content of serum has shown that, although a mild increase in the concentration of serum cholesterol occurred in rats from daily subcutaneous injection of 2·5 mgm. of cortisone acetate for two weeks, there was no significant change from corticotropin[5] in a dose of 0·3 mgm. (about 20 I.U.) suspended in peanut oil and given subcutaneously over the same period of time. However, in hypophysectomized rats there was an increase of 80 per cent or more in the cholesterol content of serum on treatment with 0·5 mgm. of cortisone acetate daily for a period of 4 days or with daily injection of 0·3 mgm. corticotropin in oil for two weeks. The effect of corticotropin was mediated through the adrenals as there was no such elevation in adrenalectomized hypophysectomized animals.

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