because the enzyme may be localized in a small fraction of the remaining volume. The figures found for the activity of the total giant fiber are, as could be expected, intermediate between those for the sheath and those for the axoplasm.

The experiments bring evidence for the high concentration of the enzyme at or near the surface of the nerve cell. They suggest that the activity of nerve cells is connected everywhere at or near the surface with the metabolism of acetylcholine and that the phenomenon is only quantitatively more important at synapses. This would explain the findings of Lorente de Nó[6] that acetylcholine can be liberated from fibers as well as at synapses and is compatible with the conclusion of Gasser and Erlanger[7,8] that conduction of nerve impulses along fibers and across synapses differs only quantitatively.

We are greatly obliged to Dr. H. B. Steinbach for the dissection of the fibers.

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FELINE PELLAGRA[1]

IN recent years, a large number of investigators have shown that nicotinic acid is essential for the health and well being of dogs,[2,3,4] pigs,[5] monkeys[6] and human beings,[7,8] but so far as we have ascertained no studies have been reported of its role in the nutrition of cats.

The present communication describes typical symptoms of nutritional deficiency in cats which are relieved dramatically following a daily oral dose of 80 to 100 milligrams of nicotinic acid.* Each of the six cats studied had lost weight and refused food. They appeared weak, sluggish, apathetic, and usually the head hung much lower than the rest of the body. They made no effort to move even when poked, and offered no resistance to hyperextension of the mouth and other physical manipulation. Examination of the oral cavity presented a peculiar yet typical appearance, characterized by an ulcerated, reddish margin in the upper part of the palate close to the midline, and a tongue that was very red over the terminal portion. Thick saliva of an extremely foul odor drooled from the mouth. Each animal had an elevation in temperature of around 3 to 5 degrees.

Within 48 hours following the administration of nicotinic acid, there was a return of appetite, subsidence of symptoms and disappearance of the oral lesions, and the temperature became normal.

The following representative case history describes the symptoms of feline pellagra and illustrates the dramatic response of an affected cat to nicotinic acid therapy.

A fourteen-month old male cat was brought to the veterinarian on August 10, 1940. For two days he had refused all food, had slept most of the time, and had remained in one place unless forced to move. His temperature was 105.8° F. The tongue was fiery red in color. The anterior border of the tongue showed an area about ¼ inch wide of ulceration and congestion. There was slight congestion and a few small ulcers in the throat. A small amount of thick saliva drooled from the mouth.

This animal was given orally 80 milligrams of nicotinic acid for two days, at the end of which time the tongue began to clear, his throat became normal and the drooling ceased. He began moving around in the cage voluntarily. By the third day, the tongue had resumed its normal color, and the cat appeared normal in every way. He drank milk and was able to eat canned food. He was dismissed from the hospital at this time, and a daily oral dose of 30 milligrams of nicotinic acid was prescribed for the next four days.

Summary. The present communication describes a deficiency disease in cats which responds promptly and dramatically to the administration of nicotinic acid.

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STRUCTURE–PROTEINS

IT is generally believed that fibrous protein molecules are found only in tissues having a mechanical function, like hair (keratin), muscle (myosin), tendon (elastin). Most of the other proteins studied have been found to be globular.

The globular shape entails a certain mobility; the rod shape an ability to form solid structures. Accord-

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9 This work was made possible by a grant of the Dazian Foundation.
1 From the Department of Internal Medicine, University of Cincinnati, Cincinnati, Ohio; the Department of Medicine, University of Alabama, and the Hillman Hospital, Birmingham, Alabama: these studies were aided by a grant from Anheuser-Busch, Inc.
2 H. R. Street and G. R. Cowgill, *Proc. Soc. Exper. Biol. and Med.*, 37: 547, 1937.
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* Supplied through the courtesy of Merck and Company, Rahway, New Jersey.

ingly, we think that wherever nature needs a mobile protein (serum albumin and globulin, secretions like milk-proteins, hormones like insulin, different enzymes, etc.) it applies the globular shape, and wherever it wants to build a solid structure it applies the rod shape. Proteins have been found to be mostly globular because unconsciously the mobile and more easily accessible proteins have been selected for study. The fibrous nature of proteins, having a mechanical function, has been recognized not because these are the only fibrous proteins, but because the mechanical function conditions a coaxial arrangement of the molecules, which arrangement makes the rod shape easily detectable.

If an animal tissue, like kidney, is extracted with weak saline, the mobile, globular proteins go into solution making about one third of the total protein. The remainder is insoluble in weak saline and represents the morphological structure, the solid edifice of the cell. This part of the protein is fibrous, as can be demonstrated by the intense double refraction of flow. In most tissues about half of this structure-protein can be brought into solution by extraction with Edsall's fluid (0,6 m KCl, 0,01 m Na_2CO_3, 0,04 $NaHCO_3$) containing 30 per cent. urea. The viscous, thixotropic protein can be precipitated by diluting (1:5) and neutralizing the solution. The protein, extracted in this way, seems to form a definite fraction analogous to myosin. The protein also contains P in quantities comparable to the P of vegetable viruses. The rod shape and P content of these proteins invalidates two of the arguments brought forward for the oxogenous nature of certain viruses. Chloroplasts also contain fibrous proteins. The remainder of the protein, representing about one third of the original, can be brought into solution by treatment with 2 per cent. NaOH containing 30 per cent. urea at 60°. This fraction also shows a marked double refraction of flow.

By squirting the urea-salt solution into water the proteins can be pulled into threads, which, after drying, can be studied röntgenoscopically. The first measurements indicate that they are built on the same pattern as myosin or keratin.

Globular proteins, if treated with urea-salt or urea-NaOH give no double refraction of flow (serum albumin and globulin, casein, ovalbumin, lactalbumin, gelatin, edestin).

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

DOUBLE-STAINING IN TOTO WITH HEMATOXYLIN AND EOSIN

A METHOD of double-staining *in toto* with hematoxylin and eosin has been devised primarily for vertebrate embryos, but has also been applied to various vertebrate and invertebrate tissues with equal success. It is simple, reliable and a great time-saver when large numbers of sections, especially serial sections, are required. Furthermore, it eliminates the danger of losing any of the sections, which might easily occur if these were stained on the slide.

By using a modification of Ehrlich's hematoxylin as a toto-stain I have obtained excellent results with both embryological and histological materials of sizes up to 1 cm in diameter. The formula is as follows:

Ehrlich's hematoxylin, stock solution	8 cc
50 per cent. alcohol	30 cc
Glacial acetic acid	2 cc

This staining solution is highly penetrating, does not overstain and gives a practically pure nuclear stain if it is followed by a proper washing with very weak acetic acid. The entire procedure involved in this double-staining method is outlined below:

(1) Fix embryos or pieces of tissue in Bouin's fluid for 1 to 3 days, according to size. Wash and preserve in 70 per cent. alcohol.

(2) The yellow color in the tissue is removed by soaking in several changes of 70 per cent. alcohol saturated with $NaHCO_3$ or $KHCO_3$ (allowing about one hour for each millimeter of tissue). Then wash out the bicarbonate in 50 per cent. alcohol for one or more hours.

(3) Stain in 20 vols. of modified Ehrlich's hematoxylin for 2 to 5 days (about one day for 2 mm of tissue). Shake from time to time to insure uniform penetration of stain from all sides of tissue.

(4) After rinsing in water, soak tissue in several changes of 30 per cent. alcohol containing 0.5 per cent. acetic acid to extract the excess stain (about one hour for each millimeter of tissue). The last change of acetic alcohol should remain practically colorless after the tissue has been in it for half an hour.

(5) Slow running tap water overnight.

(6) Dehydrate through alcohols of 30, 50, 70, 85 and 95 per cent., 6 to 24 hours in each grade.

(7) 95 per cent. alcohol with 0.3 per cent. eosin (alcohol-soluble), 12 to 24 hours.

(8) Absolute alcohol with 0.3 per cent. eosin, 12 to 24 hours.

(9a) For small embryos and soft tissues, clear in chloroform by the sinking method as follows: