produced only dilatation. Strong stimulation of the same nerve frequently produced only constriction. These results suggest that the nerves stimulated contained vasodilator and vasoconstrictor fibers, and that the vasodilators possessed a lower threshold. Furthermore, the area constricted was frequently only a portion of that originally dilated. As additional evidence of dual innervation, vasomotor nerves were found which produced only one type of response to all strengths of stimulation. Although our histological preparations show an anatomically continuous loosely-meshed non-myelinated nerve plexus continuous with the perivascular plexus, it is conceivable that dilator and constrictor fibers might occasionally become segregated.

Faradic stimulation of small nerves produced responses confined to limited vascular areas. Therefore, although the nerve plexus appears to be anatomically continuous, functional innervation is discontinuous. Stimulation of any one of several small nerves in the field produced a response in the same limited area. This fact suggests the concept of a smooth muscle motor-unit. Limited vascular areas were seen to beat rhythmically at times. No central reflex could be involved, and we have found no ganglion cells in the membrane.

Faradic stimulation of small nerves produced dilatation and contraction of the capillary only in the region of its origin. This region may respond independently of the supplying arteriole or precapillary, and function as a sphincter. Such sphincter-like regions sometimes show spontaneous rhythmic contractions, quite independent of the supplying vessel. Nuclei of contractile pericapillary cells were always seen in this region. In preparations vitally stained with methylene blue the capillary origins possessed a few modified smooth muscle cells with branched cytoplasmic processes. They are probably the type of cell originally described by Rouget,[9] redescribed by Vimtrup[10] in Amphibia, and reported by Field[11] in the rat. Furthermore, the perivascular nerve plexus was rich on the arterioles and precapillaries but sparse on the capillaries. Various pericapillary cells were found farther along the capillary. Except for an occasional cell, these did not respond to electrical stimulation. Further experiments are in progress, with drugs and with denervated preparations, to determine the nature of the independent activity and the rhythmic responses.

Both direct observation with a water immersion lens and careful study of the cinephotomicrographs of the active capillary origins failed to disclose the swelling of endothelial nuclei into the lumen, or the presence of endothelial valves. Endothelial contraction in response to stimulation of the nerves or to direct electrical and mechanical stimulation did not occur. It appears, therefore, that in the retrolingual membrane of the frog, the capillary origins are provided with modified smooth muscle cells and thus regulate capillary blood flow in a sphincter-like manner without the aid of the supplying vessel.

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[9] C. Rouget, *Arch. de Physiol. Norm. et Path.*, 5: 603–663, 1873.
[10] *Loc. cit.*
[11] *Loc. cit.*

EGG-WHITE INJURY IN CHICKS AND ITS RELATIONSHIP TO A DEFICIENCY OF VITAMIN H (BIOTIN)

THE action of Vitamin H in protecting against the injury caused by a diet containing egg white is somewhat unique in that the diet can not be considered to be deficient in an essential food constituent. Omission of the egg white from the injury-producing diet gives a ration which apparently does not lack any of the needed vitamins. This appears to be in contrast to the action of the various members of the vitamin B group in curing or preventing nutritional injuries, for the diets in these cases have always been found to be definitely deficient in the vitamin in question.

The recent observations of György, Melville, Burk and du Vigneaud have shown that vitamin H is probably identical with biotin (and co-enzyme R).[1, 2] In view of their results, it appeared that a study of the biotin intake and excretion and the biotin content in the tissues of chicks receiving egg-white injury diets might be helpful in throwing some light on the manner in which vitamin H functions.

Day-old chicks were placed on the following diet: yellow corn, 55 per cent.; wheat middlings, 20 per cent.; purified casein, 20 per cent.; bone meal, 1.5 per cent.; limestone, 2 per cent.; cod liver oil, 1 per cent.; and iodized salt, 0.5 per cent. When ten days old, the chicks were divided into two groups. One, the controls, was continued on this same diet, and the other group was given a ration in which the purified casein was replaced by dried egg white. Samples of the two diets, digested in 20 per cent. sulfuric acid for 18 hours at 100° C., gave the following assay values for biotin by the method of Snell, Eakin and Williams[3]: control diet, 0.39γ per gram; injury diet, 0.67γ per gram. Twenty-four-hour samples of the feces from the two groups were collected at intervals throughout a month, dried, weighed and carefully sampled. Aliquots were tested, both for free (extractable) biotin,

[1] Paul György, Donald B. Melville, Dean Burk and Vincent du Vigneaud, SCIENCE, 91: 243, 1940.
[2] Since this investigation was started, private information from Dr. du Vigneaud to one of us confirms the identity of vitamin H and biotin.
[3] E. E. Snell, Robert E. Eakin and Roger J. Williams, *Jour. Am. Chem. Soc.*, 62: 175, 1940.

and for "bound" biotin, *i.e.*, biotin which was liberated after 18 hours digestion of the feces at 100° C. in 20 per cent. sulfuric acid. Although there were some irregularities in the assays, it was found that both groups of chicks were excreting from 10 to 20 per cent. of their biotin intake as free biotin and approximately 15 to 25 per cent. additional as "bound" biotin. On an actual weight basis, the injured chicks were, of course, excreting more than the controls, since their intake was greater.

By the eighth week, the usual syndrome had become very pronounced in the injured group, so the tissues of two chicks from each group were then assayed for their biotin content; two weeks later tissues from an additional chick of each group were tested. These tissues were allowed to autolyze under toluene for three days at 37° C., after which they were thoroughly extracted with hot water. The tissues from the injured chicks were found to be consistently lower in their biotin content than were those from the control chicks, as can be seen from the tabulation of the assay values (Table 1).

These preliminary results indicate that the biotin which is present in the diet of the injured chicks (and which is more than sufficient in the absence of egg white) is not available to the tissues. Presumably it is destroyed by interaction with the egg white, and therefore an excess of biotin must be present in a diet containing egg white in order for the tissues to receive the necessary amount. It is probable that the injury caused by egg white is not due to any direct toxin, but rather is produced indirectly by the action of the egg white in making the biotin of the diet unavailable. If such is the case, it should be possible to produce similar syndrome by a diet which is actually deficient in biotin, but which contains no egg white.

TABLE 1

BIOTIN CONTENT OF TISSUES IN γ PER GRAM

Diet: / Age:	Injury 8 wks.	Injury 8 wks.	Injury 10 wks.	Control 8 wks.	Control 8 wks.	Control 10 wks.
Blood	0.0018	0.0021		0.0051	0.0067	0.0042
Liver	0.95	0.58	0.60	2.5	2.8	2.6
Kidney	0.45	1.3	1.0	1.9	1.8	2.5
Heart	0.018	0.041	0.036	0.11	0.033	0.11
Brain	0.025	0.029	0.044	0.067	0.018	0.065
Leg muscle.	0.008	0.016	0.015	0.027	0.018	0.033

We wish to thank Dr. T. H. Jukes for his kind cooperation in furnishing advice and certain materials for these experiments.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

MOLECULAR WEIGHT BY ISOTHERMIC DISTILLATION

OF all the solution methods for the determination of molecular weight of organic substances, the ingenious method of G. Barger[1] offers the widest applicability. This method is based upon the fact that a solution of higher molarity takes up solvent from a solution of lower molarity and *vice versa* until an equilibrium is reached. In a closed system, this produces changes in the volumes of a given standard solution of known molarity and the solution of the unknown substance but of known concentration. In practice, these changes in volume are determined by measuring at certain time intervals the diameter of several droplets contained in a sealed capillary and containing alternately the standard solution (*st*) and the solution of the unknown (*s*) in the same solvent which need not be pure (ethyl alcohol, pyridine, etc.). By appropriately choosing the molarity of the standard until the least changes are noted, the molecular weight of the unknown, the concentration of which, however, is known, may be readily calculated $\left(\frac{\%\ s}{M\ st} \times 10\right)$.

[1] G. Barger, *Jour. Chem. Soc.*, 85: 286, 1904; *Ber.*, 37: 1754, 1904.

The greatest drawback of this method as well as its various modifications[2] is that the droplets, either in the filling operation or subsequently on standing, frequently undergo mixing, thus invalidating the determination.

It has now been found that this objection, namely—the mixing—is readily overcome by having the two solutions, the standard and the unknown, in two separate capillaries of about 7–8 cm in length and 1–1.5 mm in diameter (Fig. 1, A and B). The capillaries

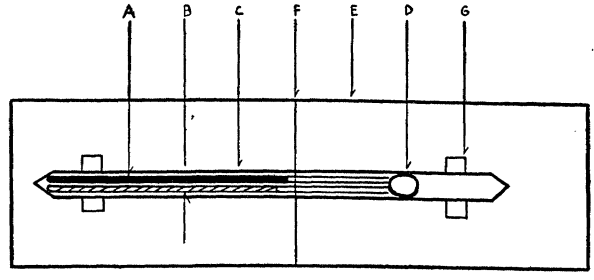

FIG. 1. Apparatus for isothermic distillation.

are filled by drawing up by suction the respective solutions, while they are open at both ends. Then the

[2] J. B. Niederl and V. Niederl, "Micromethods of Quantitative Organic Elementary Analysis," pp. 184–186. New York, N. Y.: J. Wiley and Sons. 1938.