The negative results of Mueller and Ingvar[1] who found that destruction of the neural crests in the 48-hour chick would prevent the formation of the sympathetic trunks are explained on the ground that in using an electric cautery, the effects of which cannot be localized, these investigators injured more than the neural crests and so delayed or prevented the outgrowth of neuroblasts from the ventral portion of the neural tube. Also, the significance attached to the occurrence of both spinal and sympathetic ganglia in amyelous monsters without ventral roots is discounted by a 7-day embryo with an unclosed neural tube, but with ventral roots, spinal and sympathetic ganglia—an early stage of what would undoubtedly have become an amyelous embryo.

The weight of evidence afforded by these experiments and considerations, therefore, seems to favor the view that the source of most, if not all, of the neuroblasts that form the sympathetic trunks, is the neural tube.

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Production of Canine Blacktongue on Purified Diets.

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Goldberger and coworkers[1] produced the canine disease blacktongue, on diets consisting principally of natural foodstuffs (largely corn, cow peas, and casein). They believed that the ability of certain foodstuffs to prevent the disease was due to their content of a specific heat-stable component of the vitamin B complex identical with the human pellagra-preventive (P-P) factor. The P-P factor has been rather generally considered[2] to be identical with the rat factor, vitamin G (B_2).

Certain doubts have been cast on this view of the production of blacktongue. Zimmerman and Burack[3] maintained dogs on an "artificial, balanced ration adequate in all dietary essentials as far as is known except water-soluble, heat-stable vitamin B_2 (G)," and these dogs did not develop blacktongue but a different disease.

[1] Goldberger, J., Wheeler, G. A., Lillie, R. D., and Rogers, L. M., *U. S. Public Health Reports*, 1926, **41**, 297.

[2] Aykroyd, W. R., and Roscoe, M. H., *Biochem. J.*, 1929, **23**, 483.

[3] Zimmerman, H. M., and Burack, E., *J. Exp. Med.*, 1934, **59**, 21.

Rhoads and Miller[4] also reported that they were unable to produce blacktongue by feeding diets lacking in G (B_2). It has even been suggested[3] that blacktongue is due to multiple deficiencies of the Goldberger diet rather than to lack of a specific vitamin.

The demonstration of the multiple nature of the heat-stable fraction of the B complex has led to an explanation of some of these discrepancies. Birch, György, and Harris[5] fed 4 dogs on a "synthetic" basal ration, complete except for the B complex, supplemented with lactoflavin [now known to be the rat factor, vitamin G (B_2)[6]] and highly purified vitamin B (B_1). Typical blacktongue appeared in each dog, proving conclusively that blacktongue is due to lack of some member of the vitamin B complex other than B (B_1) or G (B_2).

At the time of the appearance of the above work I had nearly completed an independent investigation on the production of blacktongue on "synthetic" diets. Four dogs were fed the following basal ration which is complete except for B vitamins: purified casein 19 parts, glucose 66, salt mixture (Osborne-Mendel) 4, cottonseed oil 8, and cod liver oil 3 parts. This was supplemented by a vitamin B (B_1) concentrate prepared according to the method of Evans and Lepkovsky.[7] Typical oral symptoms of blacktongue ranging from slight to acute occurred in the 4 dogs in 21, 21, 32, and 45 days, respectively. Control dogs receiving the same diet, with 6% autoclaved yeast added, remained perfectly healthy. These results are in complete agreement with those of Birch, György, and Harris.

In view of the complexity and controversial nature of this problem of the etiology of canine blacktongue, and the belief on the part of many investigators that the solution of this question can throw considerable light on the cause of human pellagra, this independent confirmation of the results of Birch, György, and Harris seems significant. From the work accomplished thus far there is little question that blacktongue involves in its etiology a lack of some water-soluble dietary factor or factors found in the so-called vitamin B complex and distinct from vitamins B (B_1) and G (B_2).

[4] Rhoads, C. P., and Miller, D. K., *Science*, 1935, **81**, 159.

[5] Birch, T. W., György, P., and Harris, L. J., *Biochem. J.*, 1935, **29**, 2830.

[6] Booher, L. E., *J. Biol. Chem.*, 1934, **107**, 591.

[7] Evans, H. M., and Lepkovsky, S., *J. Nutrition*, 1931, **3**, 353.