extract of vitelline membrane has not as yet been investigated, but it seems probable that some of the vitelline substance was dissolved at the temperature of the extraction.

The eggs were then exposed to ethylene dibromide, the supernatant fluid from this procedure being thereafter drawn off and placed in a dish and evaporated to dryness. A waxlike substance remained on the bottom of the crucible. Neither the acetonic nor the ethylene dibromide extractions gave positive ninhydrin or biuret reactions for protein, whereas the eggs after the ethylene dibromide extraction gave strong positive reactions.

The waxlike substance produced by the ethylene dibromide extraction is the material which formed the vitelline membrane in the original living egg. This substance was tested for the presence of an ester (R_1COOR_2) in the following manner. A drop of the ethyl ethereal solution of the vitelline membrane was placed in a microcrucible. To this were added a drop of an alcoholic solution of hydroxylamine hydrochloride and a drop of alcoholic sodium hydroxide. The crucible was heated over a flame until there was a slight effervescence. The solution was acidified with 0.5*N* hydrochloric acid and a drop of 1% ferric chloride was added. Reddish-yellow lumps formed. Upon evaporation pyramidal crystals (with a square base) were formed. Testing under a microscope with polarized light showed that these crystals were nonbirefringent, but on their surface a few minute, elongate, birefringent crystals were observed. We are not entirely satisfied with this test.

A control of the above test was run with a known ester, myricyl palmitate (beeswax), and an identical reaction was obtained. Crystal formation was not observed, but the presence of impurities was shown by the low melting point (65° C). U.S.P. beeswax (chiefly myricyl palmitate) was obtained for comparison and further refined by dissolving in warm acetone, chilling, and centrifuging. The melting point of the product was found to be 66.2° C. The melting point of pure myricyl palmitate is given as 72° C. The vitelline membrane wax produced by ethylene dibromide extraction was tested for melting point; it began to melt at 69° C and ran slowly at 72° C. In the living egg the vitelline membrane does not begin to melt until the temperature 70° C is reached and it melts completely at or below 73° C. Hence our product is not as pure as that produced by the nematode. Equal amounts of the vitelline membrane wax and the refined beeswax were mixed together, the mixture was fused at 75° C and cooled to 58° C, and thereafter the temperature was raised 1° C at 5-min intervals. This mixture began to melt at 68° C and ran slowly at 70° C. Such evidence is usually accepted as conclusive proof of the identity of the two principal ingredients of the mixture. If the principal ingredients had not been identical, the mixture should have had a melting point below either of the original ingredients. Instead, the melting point was higher than one ingredient and lower than the other. One would judge from these facts that the refined commercial U.S.P. beeswax contained impurities not present in the nemic vitelline membrane wax. Upon the basis of all of this evidence, we feel justified in concluding that the vitelline membrane in the living egg of *Ascaris lumbricoides* var. *suis* is myricyl palmitate.

The myricyl palmitate as produced by our ethylene dibromide extraction was softer than commercial beeswax and took longer to harden. There may be some stratification on cooling. The latter point will require further investigation. The myricyl palmitate produced by precipitating the hot acetonic extract of either the commercial grade or the U.S.P. grade of beeswax is of a higher degree of purity than the original material. It seems possible that one might be able to produce a completely pure compound by repeating this process an adequate number of times.

This is the first paper presenting proof of the nature of the vitelline membrane in the Nematoda. Since the melting point, solubility, and other characteristics of the vitelline membrane as previously determined in *Meloidogyne hapla, M. javanica, Parascaris equorum, Rhabditis strongyloides, Ditylenchus dipsaci,* and *Strongyloides canis* do not differ materially, it seems probable that the vitelline membrane in these organisms is the same or a similar compound. This may be the first paper presenting evidence of a wax in unsegmented animals.

Effect of Trace Minerals on Growth and Fattening of Swine [1]

Earle W. Klosterman, W. E. Dinusson, Earl L. Lasley, and M. L. Buchanan

Department of Animal Husbandry, North Dakota Agricultural College, Fargo

The beneficial effects of cobalt for ruminants in areas of cobalt deficiency have been demonstrated. Knowledge is limited as to the value of trace minerals, particularly cobalt, for swine. Willman and Noland (*5*) reported increased weight gains for swine receiving four minerals—cobalt, copper, iron, and manganese—when added to a corn-soybean oil meal ration in dry lot.

Rickes *et al.* (*4*) showed that vitamin B_{12} contains cobalt. Recently, Abelson and Darby (*1*) demonstrated that cobalt is used in the synthesis of vitamin B_{12} by rumen bacteria. Becker, Smith, and Loosli (*2*) reported, however, that on the basis of preliminary observations, administration of vitamin B_{12} did not altogether relieve symptoms of cobalt deficiency in sheep, and that cobalt was probably required for other body processes in the ruminant.

For the initial study of the effects of cobaltized and trace-mineralized salt on pigs, a ration was formulated using ground yellow corn, ground barley, linseed oil meal, tankage, ground alfalfa, steamed bone meal, and salt. These pigs were kept in concrete pens, which were washed daily. Treatments and results are presented in Table 1.

Two more experiments were conducted to study the effect of cobalt on the growth and fattening of swine

[1] North Dakota Agricultural Experiment Station Progress Report Project R&M 10, published with approval of director.

TABLE 1

EFFECT OF TRACE MINERALS ON WEIGHT GAIN AND FEED EFFICIENCY OF PIGS

Lot No.	I	II	III
Treatment	Iodized salt	Cobaltized salt*	Trace-mineral salt†
No. of pigs per lot	5	5	5
Initial wt, avg lb	52.6	53.2	51.4
Final wt, avg lb	163.8	184.2	192.6
Days on feed	98	98	98
Daily gain, avg lb	1.13	1.34‡	1.44‡
Feed in lb per 100-lb gain	431.3	389.0	394.9

* Cobalt chloride added to iodized salt to supply 0.026% cobalt.

† Trace-mineralized salt used contained the following amounts of minerals: manganese, 0.45%; iodine, 0.019%; cobalt, 0.026%; iron, 0.48%; copper, 0.048%; and salt, 96.0%.

‡ Differences from control lot significant at 5% level.

and its possible relationship to vitamin B_{12} (APF).[1] These pigs were self-fed on a basal ration of ground yellow corn, soybean oil meal, ground alfalfa, steamed bone meal, and salt. Lot IV received 8% meat scraps at the expense of soybean oil meal, and the corn was increased. Each lot was weighed at the end of the experiment when the lot average was approximately 180 lb. The pigs in these lots were fed in concrete pens, which were washed out daily. Table 2 summarizes these two experiments. The differences in gain are significant at the 5% level.

These results demonstrate that additions of cobalt to these rations stimulated weight gains more than additions of 8% of meat scraps and almost as much as the APF supplement. The differences in gains between lots I, II, and IV are not large enough to be significant but these differences were of about the same magnitude in both experiments. The results on feed per 100-lb gain were more erratic, with lots II and IV requiring slightly less feed per lb of gain.

TABLE 2

EFFECT OF COBALT, APF, AND MEAT SCRAPS ADDED TO A CORN, SOYBEAN MEAL, ALFALFA MEAL RATION FOR PIGS*

Lot No.	I	II	III	IV
Ration	Basal + cobalt†	Basal + APF‡	Basal	Meat scraps 8%
No. of pigs per lot	10	10	10	10
Initial wt, avg lb	28.7	28.7	28.7	28.8
Final wt, avg lb	180.5	180.8	176.8	181.4
Avg days to reach 180 lb	108	106	119	111
Avg daily gain, lb	1.40§	1.43§	1.24	1.37§
Feed in lb per 100-lb gain	377	365	375	358

* Average of two experiments.

† Salt (0.75 lb) containing 0.026% cobalt added per 100 lb of ration.

‡ Merck and Company supplied APF Supplement No. 3 that furnished about 0.42 μg vitamin B_{12} per lb of ration.

§ Differences from basal lot significant at 5% level.

[1] APF is the animal protein factor.

It is not known how cobalt functions. It is possible that cobalt is used in intestinal synthesis of vitamin B_{12}. Briggs (*3*) suggested, on the basis of preliminary studies, that under certain conditions cobalt is at least partially effective in counteracting a vitamin B_{12} deficiency in the chick. This action of cobalt is perhaps due to intestinal synthesis. It is less likely that the beneficial results of cobalt feeding in the present experiments were due to coprophagy, because the pens were washed daily and the pigs were never observed to be consuming feces. Neither is it probable that there was some synthesis in the feed mixture, although the ration was mixed in a spiral mixer in amounts to last 5–6 days.

Further research is under way to obtain additional information and to ascertain whether a combination of cobalt and APF, or of cobalt, APF, and meat scraps would further increase gains above that of any ingredient alone.

References

1. ABELSON, P. H., and DARBY, H. H. *Science,* 1949, **110,** 566.
2. BECKER, D. E., *et al.* *Science,* 1949, **110,** 71.
3. BRIGGS, G. M. *Feedstuffs,* 1949, **21,** 58.
4. RICKES, E. L., *et al.* *Science,* 1948, **108,** 134.
5. WILLMAN, J. P., and NOLAND, P. R. *Farm Res.,* 1949, **15,**

Evidence of a Homing Instinct in the Bermuda Spiny Lobster[1]

Edwin P. Creaser[2] and Dorothy Travis[3]

During the summer of 1949 the authors,[4] in connection with various other investigations at the Bermuda Biological Station on the spiny lobster, tested local migration in this species by the use of tags or by telson and uropod markings. In the original studies, various combinations of uropod and telson punches were employed for identification, but subsequent use of plastic tags was undertaken for individual recognition. The plastic tags were made with a barb which penetrated into the muscle when thrust through the intersegmental membane between abnominal segments. The tag used was of the same general type developed by Smith (*1*) in the Florida and Caribbean spiny lobster investigation.

Intensive trapping operations were conducted through special arrangements with a Bermuda commercial fisherman, Egbert Spurling. The gear employed was a grappling hook and 20 fish traps, the latter of typical Bermuda design, made of chicken wire and supported by spicewood. The trapping operations were conducted from motorboats, and all work was conducted in the

[1] Contribution from Bermuda Biological Station No. 164.

[2] Hofstra College, Hempstead, New York, and Bermuda Biological Station, Bermuda.

[3] Radcliffe College of Harvard University, Cambridge, Massachusetts.

[4] The cooperation of all persons at the Bermuda Biological Station is acknowledged, especially that of Tommy Gleeson, Brunell Spurling, and Egbert Spurling.