

For purposes of comparison a second assay was made using etherized cats, precaution being taken that the anesthetic be very light throughout the entire procedure. Table II is a summary of these results.

#### DISCUSSION AND CONCLUSIONS.

Judging by our experiments we believe that the method of decerebration as elaborated by Pollock and Davis is free from many of the objections which embarrass methods heretofore described.

A comparison of the tables shows a difference of 8.2 mgm. leaf per kilogram cat in the two assays: this might be taken to indicate an unfavorable influence exerted by the procedure of decerebration. In our experience, however, it is rarely possible to have different series of etherized cats give results on the same preparation of digitalis which check more closely than this. So far as the number of experiments justify, it may be assumed that the light ether anesthesia necessitated in the Hatcher-Brody cat method of digitalis assay does not materially influence the resulting minimal lethal dose.

We owe our thanks to Parke, Davis and Co. for their generosity in furnishing the digitalis leaf used in these experiments.

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### STUDIES OF THE VITAMIN POTENCY OF COD-LIVER OILS—XXI—THE STIMULATION OF REPRODUCTION BY FAT-SOLUBLE VITAMINS.\*

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In a previous paper<sup>3</sup> it was reported that the addition of a vitamin-rich cod-liver oil to the ration of laying birds at the rate of one pint per 100 birds per week caused a material increase in the number of chicks hatched. These data regarding the influence of cod-liver oil on reproduction were obtained in a study conducted with three different breeds of domestic fowl maintained under a variety of conditions as regards ration and management.

The present investigation was undertaken to secure additional data concerning the effect of fat-soluble vitamins on reproduction, particularly with respect to the effect of various amounts of supplementary vitamin feeding.

#### EXPERIMENTAL PROCEDURE.

The experimental birds in this instance were eight months old Rhode Island Red pullets. The experimental period was of thirty-two weeks' duration, which

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<sup>3</sup> *Poultry Science*, Vol. V, No. 3 (February-March 1926).

was judged to be adequate for determining the reproductive performance of birds during their first laying season. The birds were kept indoors throughout the experimental period. The house, which was unshaded, was divided into five longitudinal pens, each of which had a north and south exposure. Accordingly, all the birds received like amounts of sunshine. Seventy-five birds were placed in experimental pens, 12' x 24', which provided approximately four square feet floor space per bird.

RATION.

The ration for both the experimental and control birds consisted of dry mash, hard grain and green feed. Oyster shells, grit, charcoal and water were before the birds constantly. The dry mash was fed in open hoppers of sufficient size so that all birds could eat at one time. The analysis of the dry mash showed it to have the following composition:

Moisture	9.0%	Crude protein	21.2%
Fat	5.3%	Ash	5.5%
Crude fiber	4.7%	N. F. E.	54.3%

The hard grain was fed twice daily in amounts which the hens would completely consume in about 20 minutes. The 8:00 A.M. feeding consisted of two parts wheat and one part oats; the afternoon feeding consisted of whole yellow corn.

It was necessary to change the type of succulent feed during the experiment. From November 25th to December 2nd all birds were fed cabbage; December 2nd to March 1st mangel beets, and for the remainder of the experiment sprouted oats. For ten days preceding the experimental period all birds received a tonic consisting of the following ingredients:

Gentian	1 lb.
Ginger	1/4 lb.
Salt petre	1/4 lb.
Sulphate of Iron	1/2 lb.

The tonic was given in the dry mash at the rate of two teaspoonsful per 100 birds per day. On the 20th of each month the birds were given magnesium sulphate in the drinking water at the rate of one pound per 100 birds.

NATURE OF COD-LIVER OIL.

A crude cod-liver oil produced by the steam process was used for this investigation. It was blended from oils made along the Massachusetts and Nova Scotia coasts; the proportion being 18% Massachusetts and 82% Nova Scotia oil. Its chemical and physical characteristics were determined in accordance with the methods of the United States Pharmacopœia<sup>1</sup> and were found to be:

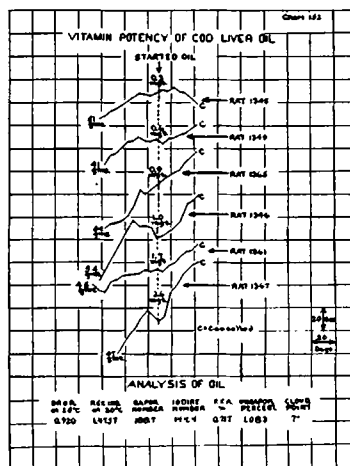


Chart 152—Fig. 1.

<sup>1</sup> U. S. Pharmacopœia X, J. P. Lippincott Co.

Sp. gr.	Ref. ind.	Sapon. value.	Iodine number.	F. F. A. per cent.	Unsapon.
0.920	1.4757	188.7	144.4	0.717	1.083

The vitamin A potency of the oil was determined by the usual laboratory procedure described in detail in earlier papers.<sup>1</sup> The results obtained are reported in Chart No. 152.

From these results it is evident that the oil in question had a vitamin content of approximately 1660 units per Gm.

The oil was fed to the experimental birds at the rate of  $\frac{1}{4}$  cc.,  $\frac{1}{2}$  cc., 1 cc. and 2 cc. per day for Pens Nos. 2, 3, 4 and 5 respectively. Pen No. 1 was reserved as a control pen. The oil was incorporated in the hard grain ration on Mondays, Wednesdays and Fridays. The amount of oil allotted to the respective pens was poured over three quarts of cracked corn, the night preceding the feeding of the oil. The oil and corn were thoroughly stirred when first mixed and again the

following morning before being fed to the birds. The corn-oil mixture was spread on top of the dry mash in the open-dry mash hoppers. The birds of Pens 3, 4 and 5 consumed all the corn and a considerable amount of mash within a half hour or so after feeding. For some unexplained reason, Pen No. 2 was much longer consuming the oil-soaked corn.

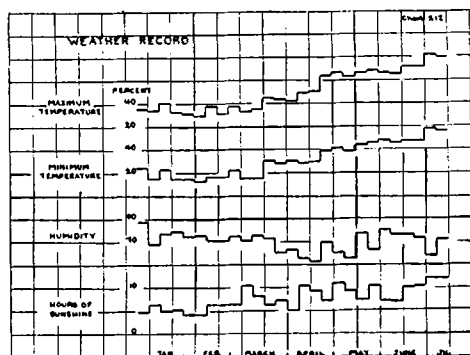


Chart 212—Fig. 2.

reported on a percentage basis; *i. e.*, the weekly egg production of a pen divided by 7 (days)  $\times$  the number of birds in the pen.

The average egg production (Chart No. 213) for the experimental period of thirty-two weeks was 38.3% for Pen No. 1; 47.0% for Pen No. 2; 51.2% for Pen No. 3; 49.2% for Pen No. 4; and 56.6% for Pen No. 5. Expressing these results in another manner, the returns for the eggs produced by the five different pens considered on the basis of 75 birds for each pen throughout the experimental period were \$308.34, \$372.29, \$399.49, \$391.06 and \$445.39 for Pens Nos. 1, 2, 3, 4 and 5 respectively.

#### WEIGHT OF EGGS.

In this study of the effect of a variation in the vitamin content of the ration on reproductive functions, data were collected concerning the size of eggs produced by the different pens. For this purpose, the eggs of each seventh day were weighed. The average weight of eggs for Pen No. 1 was 57.1 Gm.; for Pen No. 2, 58.6 Gm.; for Pen No. 3, 57.9 Gm.; for Pen No. 4, 58.8 Gm. and for Pen No. 5, 58.0 Gm.

<sup>1</sup> *J. Metabolic Research*, 2, No. 3 (Sept. 1922); *Ind. Eng. Chem.*, Vol. 16, No. 11, p. 1181 (Nov. 1924). *Ind. Eng. Chem.*, Vol. 17, No. 1, p. 75 (Jan. 1925).

While the difference in weight of eggs produced by the different pens is not large, the pens receiving cod-liver oil produced larger eggs than those of the control pen.

#### BLOOD CLOTS.

It has been often observed that stimulation of egg production is accompanied by an increase in the number of eggs containing blood clots. The occurrence of blood clots is supposed to indicate faulty functioning of reproductive organs. Accordingly, the effect of varying the vitamin content of the ration on the occurrence of blood clots was noted. The number of eggs containing blood clots was determined by candling the eggs each day. A further check on the number of blood clots was made when the eggs were consumed. The eggs produced by the pens under observation were used at the nearby Essex County Tuberculosis Sanatorium. The eggs were cooked in the patient's room and were opened at the bedside where

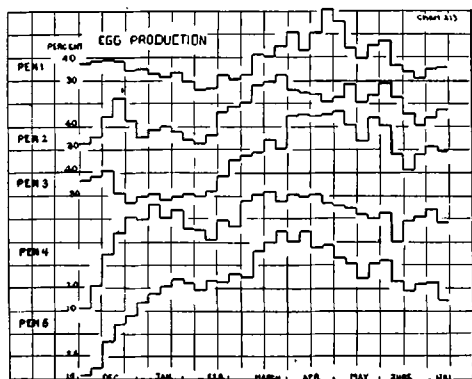


Chart 213—Fig. 3.

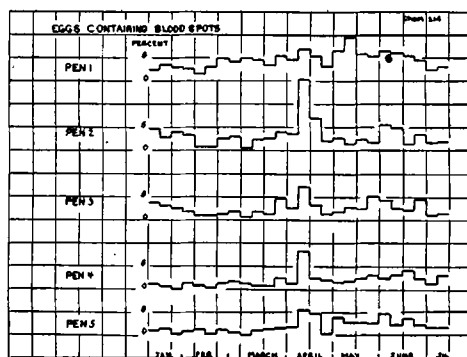


Chart 214—Fig. 4.

both patient and nurse had ample opportunity for observing the condition of the egg.

On referring to Chart No. 214, attention should be directed to the blood clots for the second week in April. The unusually large number reported was due to the overzealousness of a new assistant to make sure that no eggs containing blood clots reached the sanatorium. His error was not detected until too late to be corrected, and his record for blood clots for the second week of April is included, although it was known to be inaccurate.

The number of eggs containing blood clots was found to be 4.4%, 3.2%, 2.6%, 2.0% and 2.0% for Pens Nos. 1, 2, 3, 4 and 5 respectively. From these results, it is evident that under the condition of these experiments cod-liver oil materially decreased the occurrence of blood clots in hens' eggs.

#### WEATHER.

It has been assumed that weather conditions more or less affect egg yield, particularly during the winter season. On this account weather conditions were observed daily. Chart No. 212 reports the maximum and minimum temperature, the morning humidity and the number of hours of sunshine per day. For the sake of convenience, these data have been reduced to a weekly basis by averaging each

of these factors for the same seven-day periods as are reported for egg production and blood clots.

Comparing weather conditions with egg production and occurrence of blood clots, it appears that other factors such as ration, reproductive capacity of the birds, etc., overshadow any effect that weather may have on the reproductive performance of the domestic fowl. It is of interest to note that during March and April, the egg production of all pens increased while the average number of hours of sunshine daily remained more or less constant. In this instance, increased production was not primarily due to additional stimulation by sunshine, but rather to the birds inherent tendency for reproduction. The amount of data at hand are too limited to permit of generalization concerning the relationship between weather conditions and reproductive activity.

#### FERTILITY.

This study of the influence of fat-soluble vitamins on reproduction obviously included collecting data concerning the fertility of eggs produced by birds receiving varying amounts of cod-liver oil. Male birds were added on December 2nd to each of the five pens at the rate of one male to fifteen females. The first incubation was started January 12th. When the males were added, the question arose as to whether the males should be permanently assigned to the respective pens or whether they should be rotated from pen to pen weekly. In the latter case the fertility of the males would be more or less uniform for all pens for the period of the experiment. It was felt that for the purpose of this study, it was preferable to assign the males permanently to the different pens for then the effect of the fat-soluble vitamins on fertility applied to both male and female. Accordingly, record was made of the number of eggs discarded after seven and after fourteen days incubation either on account of the eggs being infertile or containing a weak germ. The data collected for the incubation of 4906 eggs are summarized in the following table:

FERTILITY RECORD.

Pen number.	Number of eggs incubated.	Eggs discarded after 7 days.	Eggs discarded after 14 days.	Average fertility.
1	1008	7.3%	3.6%	94.6%
2	923	9.8%	2.1%	94.1%
3	887	7.7%	2.2%	95.1%
4	1025	7.2%	2.3%	95.3%
5	1063	6.1%	1.6%	96.2%

While the number of eggs discarded on the first and second tests does not vary consistently with the amount of oil fed to the birds in the various pens, it is quite evident that oil-fed birds produced more fertile eggs than those of the control pen.

#### HATCHABILITY.

Inasmuch as some chicks which develop to apparent maturity are unable to get out of the shell, data concerning the hatchability of eggs are of more material importance to one interested in reproduction than data regarding fertility. Accordingly, the detailed data concerning the chicks obtained from the 4906 eggs which were incubated under like conditions are summarized:

## HATCHABILITY RECORD.

Date set.	Pen No. 1.		Pen No. 2.		Pen No. 3.		Pen No. 4.		Pen No. 5.	
	No. eggs.	Per cent.	No. eggs.	Per cent.	No. eggs.	Per cent.	No. eggs.	Per cent.	No. eggs.	Per cent.
Jan. 12	120	56.5	93	64.5	82	69.5	133	76.0	120	80.6
Feb. 3	120	59.2	120	59.8	118	60.2	120	70.0	120	65.0
Feb. 28	217	47.5	160	51.2	152	55.3	251	48.6	246	53.7
Feb. 28	120	50.0	120	65.8	120	58.3	120	64.2	120	74.2
Mar. 9	283	63.6	276	70.0	265	60.3	251	76.5	300	78.0
Apr. 24	148	47.8	154	70.7	150	76.6	150	77.9	157	81.4
Total eggs set	1008		923		887		1025		1063	
Average hatchability		53.9		63.6		63.3		68.8		72.1

These tests show the hatchability of the eggs produced by the different pens to be 53.9% for Pen No. 1; 63.6% for Pen No. 2; 63.3% for Pen No. 3; 68.8% for Pen No. 4 and 72.1% for Pen No. 5. These data show quite conclusively the effect of supplementary fat-soluble vitamin feeding on hatchability.

## VIABILITY OF CHICKS.

A study of reproduction involves first obtaining information concerning the number of young that may be produced and second, concerning the capacity of young to live and grow. Accordingly, information was collected concerning the viability of chicks produced from the different pens. In order to make the data more or less general in character, the day-old chicks from each pen of birds were distributed equally in five different brooder houses. Also each house contained, as controls, 200 day-old chicks (Rhode Island Reds, Barred Rocks and White Leghorns) produced under normal flock conditions by birds receiving one pint of cod-liver oil per 100 birds per week.

The houses were so distributed in an open field as to be somewhat differently effected by atmospheric conditions. The first night the chicks were in the brooder houses the temperature suddenly fell to 4° F., and coincidentally the caretaker contracted influenza and was removed to the hospital. As a consequence, the chicks were subjected to extremely unfavorable environmental conditions. From the standpoint of practical poultry management, this situation was little short of a catastrophe, but from the standpoint of securing information relative to the chicks' capacity to withstand adversity, the situation was extremely favorable.

## VIABILITY OF CHICKS.

## Mortality during First Five Weeks.

Experimental pen.	Per cent mortality.	Control.	Per cent mortality.
1	58%	Reds	11%
2	23%	Rocks	11%
3	8%	Leghorns	9%
4	15%		
5	9%		

At the end of five weeks, the number of chicks remaining was 42% for Pen No. 1; 77% for Pen No. 2; 92% for Pen No. 3; 85% for Pen No. 4; 91% for Pen No. 5 and 89%-91% for the controls. These data offer quite definite evidence

that the viability of chicks produced by birds receiving a diet rich in the fat-soluble vitamins is greater than that of chicks produced by birds maintained on a diet containing no supplementary source of fat-soluble vitamins.

#### BROODY BIRDS.

As data accumulated showing that supplementary fat-soluble vitamin feeding increased the reproductive activity of the birds, it was questioned whether it would also increase the tendency of birds to become broody. Records kept concerning the total number of broody birds showed 105 birds for Pen No. 1; 84 for Pen No. 2; 77 for Pen No. 3; 105 for Pen No. 4 and 78 for Pen No. 5. In each instance the number of broody birds exceeds the total number of birds in the pens. This is due to some birds being broody twice and occasionally even three or four times. A portion did not exhibit signs of broodiness at any time. Considering the data as a whole, it does not appear that the supplementary use of fat-soluble vitamin influence broodiness.

#### BODY WEIGHT.

All birds included in this investigation were weighed five times during the experimental period. The average body weight of the different pens is reported below:

Pen no.	AVERAGE WEIGHT OF PULLETS.				
	Average wt. 11/11/25	Average wt. 1/29/26	Average wt. 4/6/26	Average wt. 5/14/26	Average wt. 7/6/26
1	5.6 lbs.	6.2 lbs.	6.3 lbs.	6.2 lbs.	6.2 lbs.
2	5.9 lbs.	6.5 lbs.	6.5 lbs.	6.5 lbs.	6.3 lbs.
3	6.1 lbs.	6.5 lbs.	6.4 lbs.	6.4 lbs.	6.2 lbs.
4	4.9 lbs.	6.6 lbs.	6.6 lbs.	6.6 lbs.	6.4 lbs.
5	4.6 lbs.	6.6 lbs.	6.6 lbs.	6.4 lbs.	6.5 lbs.

Of particular interest is the average body weight of the birds of different pens at the termination of the experiment. It is a matter of repeated observation by poultrymen that birds which are relatively high egg producers ordinarily suffer a reduction in body weight as the period of high egg production progresses. In this instance, however, it will be noted that the birds of Pen No. 5 averaged a higher body weight at the termination of the experiment than that of the birds of any of the other pens, even though their egg production exceeded that of the other birds.

#### MORTALITY.

Inasmuch as stimulated reproduction is often accompanied by a lowered body resistance to disease, it was of interest to collect data concerning the mortality for the different pens of birds under observation. The following table supplies

MORTALITY OF PULLETS.				
Pen 1.	Pen 2.	Pen 3.	Pen 4.	Pen 5.
9 birds	12 birds	5 birds	3 birds	2 birds
12%	16%	6%	4%	2%

*Cause of Death.*—Cancer, 1; Over Fat, 1; Roup, 1; Tuberculosis, 1; Bronchitis, 1; Heart Rupture, 2; Undetermined, 3; Prolapsis, 21.

information concerning the total number of deaths and concerning the cause of the deaths.

Inasmuch as nine of the control birds and twelve birds in the pen receiving only  $\frac{1}{4}$  cc. of cod-liver oil a day died as compared with 5, 3 and 2 birds respectively for Pens Nos. 3, 4 and 5, it is evident that the addition of fat-soluble vitamins as a supplement to the ration materially increased body resistance to disease. On referring to the various causes of deaths, it is of interest to note that by far the greatest number of deaths was occasioned by prolapsis of the oviduct. Of striking importance was the apparent immunity of these birds from the common poultry diseases that were usually prevalent in the immediate locality during the period of this experiment. The mortality rate for common poultry diseases was 2.2%, and the deaths from such diseases was practically confined to Pen No. 1 and Pen No. 2.

#### FLAVOR OF EGGS AND FLESH OF BIRDS.

As stated above, the eggs produced by the cod-liver oil-fed birds, except those reserved for incubation, were eaten by patients of the Essex County Tuberculosis Sanatorium. Inasmuch as at no time during the thirty-two weeks' experiment was any comment made by any of the patients concerning the eggs possessing any unusual flavor it was concluded that the cod-liver oil used in this test did not impart any flavor to the eggs.

To test whether long continued feeding of cod-liver oil imparts flavor to the flesh, a number of birds from Pen No. 5, which had received 2 cc. of oil daily for thirty-two weeks, were killed. Some of the birds were roasted under normal household conditions and eaten in private homes by persons uninformed concerning the bird's ration. Some were stewed and eaten by poultrymen and their families. Also a number of birds were sampled by instructors and investigators in poultry husbandry. None of the sixty persons who partook of the birds made any comments indicating the presence of any unusual flavor. From this it is evident that the cod-liver oil under consideration did not impart any detectable flavor to the flesh of the birds.

#### VITAMIN CONTENT OF EGGS.

The vitamin A content of eggs produced by the experimental birds was determined early in the experiment and at its termination. Albino rats were used as test animals and the usual laboratory procedure was followed. Each day during the thirty-five day vitamin test, an egg was collected from each of the five pens. Five groups of rats were fed 0.03, 0.06, 0.12, 0.50 and 1.00 cc. of egg yolk daily. In the first test, which was started two months after the birds began to receive cod-liver oil, the eggs produced by the cod-liver oil fed birds were found to possess a higher vitamin content than among those produced by the control birds. In the test conducted after the birds had received oil for six months, it was found that the difference in the vitamin A potency of eggs from the control and experimental birds was materially greater than in the earlier test. The vitamin A potency of eggs from the control birds had decreased as the experimental period advanced. On the other hand, the vitamin content of eggs from the experimental birds increased during the experimental period. From this it appears that supplementary feeding of cod-liver oil increases the vitamin content of hens' eggs.



## SUMMARY.

In order to make it possible to easily summarize the results of the studies here discussed, the data essential for this purpose has been brought together in the table which follows:

	SUMMARY OF RESULTS.				
	Pen 1.	Pen 2.	Pen 3.	Pen 4.	Pen 5.
<i>Egg Record.</i>					
Production	38.3%	47.0%	51.2%	49.2%	56.6%
Blood spots	4.4%	3.2%	2.6%	2.0%	2.0%
Weight of eggs	57.1 Gm.	58.6 Gm.	57.9 Gm.	58.8 Gm.	58.0 Gm.
Fertility	94.6%	94.1%	95.1%	95.3%	96.2%
Hatchability	53.9%	63.6%	63.3%	68.8%	72.1%
Viability of chicks	42.0%	77.0%	92.0%	85.0%	91.0%
<i>Pullet Record.</i>					
No. of broody birds	105	84	77	105	78
Body wt. at end of expt.	6.2 lb.	6.3 lb.	6.2 lb.	6.4 lb.	6.5 lb.
Mortality	9	12	5	3	2

To determine the influence of fat-soluble vitamins on the reproductive performance of domestic fowl, five pens of birds, one control and four receiving  $\frac{1}{4}$ ,  $\frac{1}{2}$ , 1 and 2 cc. of cod-liver oil daily were under observation for a period of thirty-two weeks.

The egg production for the different pens shows that the addition of fat-soluble vitamins to the poultry ration employed very definitely stimulated egg production.

The average weight of eggs produced by the oil-fed birds was slightly greater than that of the eggs produced by the control birds.

The appearance of blood clots in hens' eggs indicates the inability of the reproductive organs to function normally. In these tests the percentage of eggs containing blood clots decreased consistently with the increase of cod-liver oil in the experimental ration.

From the limited data available, it does not appear that the weather conditions materially influenced the reproductive activities of birds under observation.

The number of eggs discarded during incubation either from being infertile or on account of containing a weak germ was less for the oil-fed than the control birds; Pen No. 5 showing the highest fertility.

The number of chicks obtained from the eggs produced by the oil-fed hens was greater than that obtained from the eggs from the control birds; approximately 18 chicks per 100 eggs more for Pen No. 5 than for Pen No. 1.

The viability of chicks from the oil-fed hens exceeded that of the control birds; this was particularly noticeable for the birds receiving the larger amounts of oil.

The number of birds in the different pens becoming broody during the course of the experiment does not show that supplementary use of cod-liver oil influences this factor.

Contrary to commonly observed conditions, the body weight of the high producing birds did not decrease, but their weight at the end of the thirty-two weeks' experiment exceeded that of the control birds.

The mortality of the high producing birds was lower than that of the control birds. The data obtained for body weight and mortality showed that the physical conditions of the high producing birds was equal to and probably better than that of the birds which received no supplementary vitamin feeding.

The cod-liver oil used in this test did not impart any detectable flavor to either the eggs or the flesh of the birds.

The vitamin A content of the eggs was determined twice during the experiment. In both cases the potency of eggs from the experimental birds exceeded that of the controls. The potency of eggs from the control birds decreased, and the potency of eggs from the experimental birds increased as the experiment progressed.

Considering the results as a whole, the evidence is quite conclusive that supplementary fat-soluble vitamin feeding increases the reproductive performance of the domestic fowl.

#### ABSTRACT OF DISCUSSION.

**Arno Viehoever** inquired whether the growth of the chicks was stimulated by regular feedings of cod-liver oil.

The author replied that it depends on conditions—an adequate diet need not be bolstered.

**L. F. Kebler** inquired whether cod-liver oil would increase egg production.

The author replied that he would not have any one draw the conclusion that cod-liver oil always increases egg production, but if hens are receiving a low vitamin diet, their egg production can often be materially increased by using cod-liver oil.

**H. C. Wood** inquired relative to the diet of the control chickens.

The author explained the feeding and stated that the rations were those usually given by the commercial poultrymen.

**L. F. Kebler** inquired of the author whether he had made observations on increased egg production by the addition to feed of vitamins, directly or indirectly.

The author referred to the results reported in this paper and to work being done in Ohio, Rhode Island, etc.

**F. W. Nitardy** referred to work being done in Connecticut. His recollection was that cod-liver oil produced better results than other feedings.

The question was asked whether the author had experimented with rice in the feed.

**A. R. L. Dohme** asked whether there were statistics available along related lines in reproduction of other animals than pullets.

The author replied in the affirmative and referred to studies of the effect of adding calcium and mineral to the ration and to the investigations in Texas, Idaho and Massachusetts.

**A. R. L. Dohme** commended the work of the author; he thought Mr. Holmes was over-cautious in his statements; if pharmacists can, in confidence, recommend cod-liver oil for speeding the growth of poultry and improving its condition they will be rendering a service.

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#### CONVENTION EXPENSES HELD DEDUCTIBLE FROM INCOME.

Expenses incurred in attending meetings of the American Chemical Society and other scientific gatherings are ordinary and necessary business expenses in the case of Alexander Silverman, professor of Chemistry at the University of Pittsburgh, according to a ruling by the Board of Tax Appeals.

Prof. Silverman deducted \$558.75 from his

gross income in 1921 as expenses in attending meetings of the American Chemical Society and the American Ceramic Society for the purpose, as he contended, of keeping thoroughly informed in his field of work and in touch with other scientists, and in order to advance the interests of the University.

The Board held that expenditures of this character and made under such circumstances are deductible as an ordinary and necessary business expense.