SOME OBSERVATIONS ON THE RELATIONSHIP BETWEEN VITAMIN B_{12} AND REPRODUCTION IN SWINE[1]

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ABSTRACT

Six female swine were maintained during one or more gestations on a vitamin B_{12}-deficient diet. Two that had been fed the deficient diet for more than 2 years prior to their first gestation aborted. Four that received the deficient diet for 7 months prior to their first gestation farrowed a total of 8 litters containing 68 live piglets; 27 died within 3 days. All piglets fed a synthetic milk diet, deficient in vitamin B_{12}, died within 14 days.

Five adult females fed the deficient diet supplemented with 400 μg. of vitamin B_{12} per animal per day farrowed a total of 8 litters containing 83 live piglets; none died within 3 days. Of 14 piglets fed a vitamin B_{12}-deficient synthetic milk diet, 2 died within 14 days.

The different dietary vitamin B_{12} intakes of the adults influenced the serum vitamin B_{12} levels of the adults and of their offspring.

Piglets fed vitamin B_{12} gained more, but feed conversion was not influenced by the vitamin B_{12} dietary intake of the piglets or by the intake of their dams during gestation.

It is concluded that vitamin B_{12} influences swine reproduction and neonatal survival.

INTRODUCTION

During the past decade a number of reports have dealt with the influence of vitamin B_{12} on the reproduction of various species of animals, including swine. Most, but not all of the evidence indicates that vitamin B_{12} does influence reproduction, and that the dietary intake of vitamin B_{12} might be important for swine maintained under practical husbandry conditions (4).

The work reported here was carried out over a period of several years to clarify various aspects of the relationship between vitamin B_{12}, reproduction and neonatal performance.

EXPERIMENTAL

Adults

Eleven female swine, approximately 7 months old, received a vitamin B_{12}-deficient, corn-soybean meal diet (7), with or without a 400 μg. supplement of vitamin B_{12} per animal per day, for different periods of time prior to their first mating and during subsequent gestation periods according to the following plan:

	Basal diet						Basal diet plus vitamin B_{12}				
Sow no.	1	2	3	4	5	6	7	8	9	10	11
Age at 1st mating (months)	36	36	15	15	21	15	36	36	15	15	15
No. gestations studied	1	1	2	3	2	1	1	1	3	2	1

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The basal diet was fed twice daily. It contained less than 1 μg. of vitamin B_{12} per pound, but not less than N.R.C. (9) recommended allowances of other nutrients. Animals 7 and 8 received no vitamin B_{12} supplement for the first 18 months and variable amounts until 2 months before conception. A supplement of 400 μg. of vitamin B_{12} daily was fed to animals 7 and 8 from 34 months of age and to animals 9-11 from 8 months of age. Animals 7-11 are designated hereafter as "treated" animals and animals 1-6 as "control" animals.

Animals were kept individually in pens on raised wooden floors covered with shavings. They were exercised twice weekly for 30 minutes on a mechanical exerciser that forced the pigs to walk at the rate of 3 m.p.h.

Blood samples were taken usually at 2-week intervals from the major blood-vessels in the region of the thoracic inlet. The 10 weeks before conception was considered the pre-gestation collection period. Reported values (Table 1) represent the mean serum vitamin B_{12} levels for the specified periods.

Piglets

Stillbirths, live births and neonatal deaths up to 3 days of age were recorded. Some of the piglets were removed from their dams on the third morning after birth. They were assigned to oral vitamin B_{12} supplementation tests as follows:

Piglet test 1

	Treated sows No.			
	7		8	
Oral vitamin B_{12} (μg./piglet/day)	0	20	0	20
Number of piglets	3	3	3	3
Observation period (days)	26	26	43	43

Piglet test 2

	Control sows					Treated sows			
	3		4		5	9		10	
Litter No.	1	2	1	2	1	1	2	1	2
Vitamin B_{12}/kg. dry matter intake (μg.)	No. piglets at each dosage level								
0	2	2	2	2	1	2	2	2	2
20	1	1	1	1	1	1	1	1	1
40	1	1	1	1	1	1	1	1	1
80	1	1	1	1	1	1	1	1	1

Piglets in test 1 were grouped together in metal cages according to treatments and dams. Those in test 2 were kept in individual cages for either 53 or 56 days. All test piglets were fed six times daily between 7 a.m. and 8 p.m. a vitamin B_{12}-deficient, synthetic milk diet similar to the medium fat diet of Cunningham and Brisson (2). Modifications of the diet included the use of vitamin-free casein, adjustment of the solids content from 10 to 15 per cent, and deletion of the vitamin B_{12} supplement. A solution of crystalline vitamin B_{12} was administered *per os* to the treated piglets in test 1, and it was added to the milk fed to the treated piglets in test 2. Feed efficiencies were calculated as kilograms synthetic milk dry matter consumed/kilogram body weight gained.

Of the piglets that remained with the control dams, one-half received intramuscular injections of vitamin B_{12} soon after birth.

Blood samples were obtained by the method described for sows or by cardiac puncture.

Vitamin B_{12} Assay

Vitamin B_{12} was assayed with *Euglena gracilis var. bacillaris* (5).

Data were analysed by common statistical methods.

RESULTS AND DISCUSSION

Blood serum vitamin B_{12} levels of control sows were distinctly lower than those of treated sows (Table 1). This was true both prior to and during the different gestations.

TABLE 1. — BLOOD SERUM VITAMIN B_{12} LEVELS OF SOWS

Sow No.	Gestation	Serum vitamin B_{12} ($\mu\mu$g./ml.)				
		Pre-gestation	Days of gestation			
			0–28	29–56	57–84	85–42
		Control sows				
1	1	151	117	89	113	155
2	1	123	143	97	114	137
3	1	64	11	56	10	98
	2	—	95	—	132	84
4	1	136	83	70	37	84
	2	—	90	—	159	129
5	1	77	115	—	127	134
	2	—	—	—	117	101
6	1	<20	—	—	—	<20
		Treated sows				
7	1	437	224	254	257	290
8	1	274	195	162	228	—
9	1	544	564	315	362	438
	2	—	592	438	374	405
10	1	470	531	383	383	551
	2	—	431	403	417	443
11	1	280	—	—	—	326

TABLE 2. — REPRODUCTIVE PERFORMANCE OF SOWS

Sow No.	Gestation	Farrowing history			Remarks
		Total born	Still-births	Neonatal deaths	
		Control sows			
3	1	12	0	1	
	2	12	2	3	
4	1	11	0	0	
	2	11	1	4	
	3	9	1	8	
5	1	11	2	3	
	2	9	3	6	
6	1	6	4	2	2 were deformed; 1 was born a day later.
Total		81	13	27	
		Treated sows			
7	1	13	7	0	Deaths were caused by anoxia resulting from prolonged delivery.
8	1	13	0	0	
9	1	15	2	0	
	2	15	1	0	
	3	10	2	0	The stillbirths were born a day later.
10	1	14	3	0	
	2	10	0	0	
11	1	11	3	0	
Total		101	18	0	

Sows 1 and 2, fed the deficient diet for the longest period prior to mating, aborted near term. Post-mortem examinations of their genital tracts revealed no evidence of infection.

The maternal dietary vitamin B_{12} status markedly influenced the survival of piglets born alive (Table 2). Clinical observations of successive litters born by basal-fed sows revealed progressively weaker newborn piglets. This is indicated only partially in Table 2; the effect was observed in offspring of sows maintained under practical conditions (6). Similar observations have been reported for rats (8) and mice (1). Piglets left with control dams survived only if they were given intramuscular injections of vitamin B_{12}.

Table 3 extends and corroborates the preceding observations on the effect of the different maternal and neonatal regimens on neonatal survival.

The original weight gain data revealed no apparent differences in the weight gains between piglets treated with 20, 40, or 80 μg. of vitamin B_{12} per kilogram of dry matter intake. Therefore the results for these treatments were grouped together. Variance analysis of mean litter weights of

TABLE 3. — EFFECT OF MATERNAL AND PIGLET VITAMIN B_{12} DIETARY INTAKES ON PIGLET SURVIVAL AND 8-WEEK WEIGHT GAINS (PIGLET TEST 2)

Vitamin B_{12} in piglet diet (μg./kg. D.M.)	Control sows			Treated sows		
	Piglet weight gain (kg.)		Survival ratio[1]	Piglet weight gain (kg.)		Survival ratio[1]
	Mean	S. E.		Mean	S. E.	
0	—	—	0/9	15.7	1.1	7/8
20	19.5	0.5	3/5	20.6	1.6	4/4
40	17.7	3.5	3/5	21.7	1.4	3/4
80	19.5	1.4	3/5	19.4	1.3	3/4

[1]Number of survivors/number initially allocated

treated piglets, either unweighted or weighted according to the number of piglets studied per litter, did not reveal any difference in piglet growth due to the maternal dietary vitamin B_{12} intake during gestation. Apparently, the deficiency acquired *in utero* was reversible in these treated piglets as far as weight gains were an indication, but this requires additional observations. It agrees with observations in mice (1) and rats (3, 8).

Comparison between final body weights of vitamin B_{12}-treated and untreated piglets born of treated sows were made on a within-litter basis, after adjustment for initial body weights. The mean difference for each litter was weighted inversely according to its variance, and the mean of these weighted differences was significant (P almost 0.001). Weight differences between treated and untreated piglets occurred in different litters at widely different times after allocation (2 to 8 weeks). The wide between-litter time differences may be part of the explanation for the widely different estimates of the vitamin B_{12} requirement of baby pigs reported in the literature.

In all groups there was a direct relationship between body weight gained and the amount of milk consumed. Average feed conversion efficiency for the different groups ranged between 7.2 and 7.7 kilograms dry matter consumed per kilogram body weight gained. Differences between groups were not apparent ($P > 0.05$).

The vitamin B_{12} dietary intake of dams influenced the blood serum vitamin B_{12} levels of piglets. Serum levels of three 3-day-old piglets born by control sows 3 and 4 ranged from 40 to 44 $\mu\mu$g./ml.; levels of four similar-aged piglets born by treated sows 9 and 10 ranged from 156 to 366 $\mu\mu$g./ml. Table 4 shows the effect of the neonatal dietary treatment of piglets born by treated sows 9 and 10 on serum levels at various intervals during the feeding trials. It also includes information from piglets born by treated sows 7 and 8. At each time interval the serum levels of untreated piglets were lower than those of treated littermates. Despite the different

TABLE 4. — EFFECT OF VITAMIN B_{12} SUPPLEMENTATION OF SYNTHETIC MILK DIET ON PIGLET BLOOD SERUM VITAMIN B_{12}($\mu\mu$g./ml.)[1]

Sow No.	Piglet age (weeks)	Piglet vitamin B_{12} supplement[2]			
		0	20	40	80
7	4	20-57 (3)[3]	239-724 (2)		
8	4	156-240 (2)	460-542 (2)		
	6	14-130 (2)	381-524 (2)		
9	8	162 (1)	265 (1)	231 (1)	258 (1)
10	8	134 (1)	260 (1)	258 (1)	191 (1)

[1]Data from piglet tests 1 and 2
[2]As μg./day for progeny of sows 7 and 8 and μg./kg. dry matter intake for progeny of sows 9 and 10
[3]Number of piglets in parentheses

TABLE 5. — BODY WEIGHT GAINS (KG.) OF PIGLETS BORN OF TREATED SOWS 7 AND 8 (PIGLET TEST 2)

Sow		Vitamin B_{12} dose (μg./day)	
		0	20
7		3.0	3.0
		3.2	3.7
		5.6	—
	Mean	3.9	3.4
8		8.7	18.0
		6.8	16.3
	Mean	7.8	17.2

4-week serum levels of treated and untreatd piglets born by sow 7, differences in weight gains were not apparent at that time (Table 5). This agrees with our belief that serum levels are a more sensitive criterion of vitamin B_{12} status in the growing animal than weight gains. Weight gain differences between treated and untreated offspring born by sow 8 occurred as early as 16 days after birth. The 16-day body weights of piglets born by treated sow 8 were compared with similar data from the litters born by treated sows 9 and 10. When the mean litter differences in the 16-day body weights were adjusted for the initial weights, the mean difference between the treated and untreated piglets born by sow 8 was significant ($P < 0.05$). Blood samples were taken from 4-week-old piglets born by sow 8. Differences in serum vitamin B_{12} levels between treated and untreated piglets were apparent and the levels of the same untreated piglets declined further during the following 2 weeks.

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