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Original article

Effect of injected vitamin A and level of dietary vitamin E on α-tocopherol status in gestating swine

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Summary — A 2 × 2 trial was conducted to determine the effects of injected vitamin A and dietary level of vitamin E on blood serum and tissue concentrations of α-tocopherol during early gestation of gilts. Thirty-two crossbred gilts were fed a corn soybean meal basal diet supplemented with DL-alpha tocopheryl acetate to provide either 25 or 500 IU of vitamin E/kg of diet. Gilts were fed daily 1.9 kg/gilt beginning 7 days before breeding until day 25 of gestation. Sixteen gilts were injected (im) with 350 000 IU of retinol palmitate 7 days before breeding, at the time of breeding (d0), and 7 days after breeding. Blood samples were collected on day –7, 0, 7, and 24, and all gilts were slaughtered on day 25 of gestation. Supplemental vitamin E at 500 IU/kg of diet increased α-tocopherol concentrations ($P < 0.01$) in blood serum in all tissues examined, including reproductive and embryonic, except fat. Vitamin A injections had no effect ($P > 0.10$) on blood serum α-tocopherol concentrations except on day 7 when a small increase ($P < 0.06$) was noted. Vitamin A injections had no effect ($P > 0.10$) on tissue α-tocopherol concentrations. Increasing dietary level of vitamin E increased blood serum and tissue α-tocopherol concentrations, and vitamin A injections had little or no effect on these concentrations during the early gestation of gilts.

pig / vitamin A / vitamin E / reproduction

Résumé — Effet de la vitamine A injectée et de l'apport alimentaire de vitamine E sur le statut en α-tocophérol chez la truie gestante. Un essai de type 2 × 2 a été conduit pour déterminer les effets de l'injection de vitamine A et du niveau alimentaire de vitamine E sur les concentrations sériques et tissulaires d'α-tocophérol chez les truies en début de gestation. Trente-deux truies de race croisée ont reçu un régime à base de maïs et tourteau de soja supplémenté en DL-alpha tocophéryl acétate pour fournir 25 ou 500 UI de vitamine E par kg de régime. Chaque truie recevait 1,9 kg d'aliment par jour, de 7 jours avant l'insémination jusqu'à 25 jours de gestation. Seize truies ont reçu par injection im 350 000 UI de palmitate de rétinol 7 jours avant l'insémination, le jour de

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l'insémination (d0) et 7 jours après. Des échantillons de sang ont été prélevés aux jours –7, 0, 7 et 24 et toutes les truies ont été abattues au 25e jour de gestation. Un supplément de 500 UI/kg de vitamine E a augmenté les concentrations d'α-tocophérol ($p < 0,01$) dans le sérum sanguin et dans tous les tissus examinés, incluant les tissus de reproduction et embryonnaires, sauf la graisse. Les injections de vitamine A n'ont pas eu d'effet ($p > 0,10$) sur le tocophérol sérique sauf au jour 7 où une légère augmentation ($p < 0,06$) a été observée. Aucun effet ($> 0,10$) n'a été constaté sur les concentrations tissulaires d'α-tocophérol. Un supplément alimentaire de vitamine E a augmenté les concentrations sériques et tissulaires d'α-tocophérol mais les injections de vitamine A n'ont pas (ou peu) d'effet sur les concentrations au début de la gestation chez la truie.

porc / vitamine A / vitamine E / reproduction

INTRODUCTION

Vitamin A and(or) β-carotene injected just before and(or) shortly after breeding seems to enhance the reproductive performance of gilts and sows (Brief and Chew, 1985; Coffey and Britt, 1993). There is evidence from studies with chicks and laboratory rats that very high dietary vitamin A (eg, 100 × requirement) may interfere with both vitamin E absorption and blood α-tocopherol concentration (Abawi and Sullivan, 1989; Blakely et al, 1991). The effect of injected vitamin A on the vitamin E status of gestating pigs is not known. The objective of this study was to evaluate the effects of injecting vitamin A just before, at and shortly after breeding, and of dietary supplementation of vitamin E on blood and tissue concentrations of α-tocopherol and retinol in gestating gilts.

MATERIALS AND METHODS

Animals

The trial involved 32, 7- to 8-month-old Yorkshire × Hampshire crossbred gilts. Gilts were divided into groups of eight each. Gilts were from a previous trial that had involved the feeding of diets supplemented with either 2 000 or 20 000 IU of vitamin A/kg of diet. Previous treatment was taken into consideration when allotting the gilts to a treatment so that each group of eight consisted of four that previously received the low treatment and four the high treatment. The previous trial ended 40 days before the start of the present trial. The gilts were fed daily approximately 2 kg/head of a standard gestation diet containing 5 000 IU and 28 IU/kg of vitamin A and vitamin E, respectively, during the interim. For the present study, each group was randomly assigned to one of four treatments in a 2 × 2 factorial design. The basal diet was a corn soybean meal-based, non-vitamin E supplemented diet (table I) calculated to meet NRC (1988) guidelines for nutrients other than vitamin E. Treatments consisted of supplementing the basal diet with DL-α-tocopheryl acetate (Hoffmann-La Roche, Nutley, NJ, USA) at either 25 or 500 IU/kg of diet. Two groups were given

Table I. Composition of basal diet fed to gestating gilts, as fed basis.

Ingredients	*Percentage*
Ground yellow corn	84.66
Soybean meal (48% CP)	12.40
Dicalcium phosphate	1.54
Limestone	0.65
Salt	0.50
Trace mineral[a]	0.10
Vitamin premix[b]	0.10
Se premix[c]	0.05

[a]Provided 200 ppm of zinc, 100 ppm of iron, 55 ppm of manganese, 11 ppm of copper, 1.5 ppm of iodine, and 1 ppm of cobalt; [b]provided 4.4 mg of riboflavin, 22 mg of niacin, 18 mg of d-pantaothenic acid, 300 mg of choline chloride, 20 μg of vitamin B_{12}, 3 mg of vitamin K, 880 IU of vitamin D_3, and 4 000 IU of vitamin A per kg of diet; [c]provided 0.1 ppm of selenium.

the low vitamin E diet and the other two the high diet. Sixteen gilts (two of the four groups; one low E, one high E) were given three injections (im in the neck) of 350 000 IU/injection of vitamin A (vitamin A palmitate, Hoffmann-La Roche) and 16 were injected with vehicle only (corn oil). Gilts were injected 7 days before breeding (d–7; mean), at breeding (d0), and 7 days after breeding (d7).The gilts were fed the experimental diets beginning 7 days before breeding through day 25 of gestation, and were fed individually 1.9 kg of feed once daily. Water was available ad libitum. Gilts were housed in an open-sided building in pens with solid concrete floors. Each group was housed together in a pen equipped with individual feeding stalls. The gilts were checked twice daily for estrus. Upon their second or third observed estrus, gilts were mated twice to Duroc × Hampshire × Yorkshire boars.

Blood samples were collected by jugular vein puncture using vacuum blood collecting tubes from each gilt 7 days before mating (–7; mean) and on day 0, 7, and 24 of gestation to monitor blood α-tocopherol and retinol concentrations. The –7, 0–, and 7–d samples were taken just before the vitamin A injection given the same day. Blood samples were centrifuged and serum harvested. Serum samples were stored at –2 °C until they were analyzed. Precautions were taken during sampling, storage and analysis to avoid exposure to light.

All gilts that conceived (not returning to estrus) were slaughtered on day 25 of gestation at the University of Florida Meat Science laboratory. The reproductive tracts were immediately removed and placed in a refrigerator until further examination. The tracts were examined for number of corpora lutea (CL) and dissected to recover embryos. Tissue samples were collected from the endometrium, embryo, ovary, uterus, liver, leaf and back fat, and muscle (*semimembranosus* and *rhomboideus*). Tissue samples were stored at –20 °C until they were analyzed.

The trial was carried out during the summer and early fall (July through October). Pigs were managed according to practices approved by the University Animal Use committee. Data collection included serum and tissue concentrations of α-tocopherol, serum and liver concentrations and of retinol, and reproductive performance data. Three gilts, each from a different treatment group, were eliminated from the study due to factors unrelated to treatment (death, lameness, and failure to conceive).

Analytical methods

Serum and tissue vitamin E (α-tocopherol) concentrations were determined by high performance liquid chromatography (HPLC) (Anderson et al, 1995a). Alpha-tocopherol was extracted from the serum in propanol instead of ethanol which was a modification of the procedure of McMurray and Blanchflower (1979a). Extraction of α-tocopherol from tissues was done using a procedure outlined by Chung et al (1992). This procedure was a modification to that of McMurray and Blanchflower (1979b) and Hatam and Kayden (1979).

Alpha-tocopherol was determined by injecting 10 μL of the reconstituted extracted sample (serum or tissue) into the HPLC system. The HPLC system consisted of a Perkin Elmer 550 terminal (Perkin-Elmer, Analytical Instruments, Norwalk, CT, USA), a Perkin Elmer ISS-100 auto sampler (Perkin-Elmer), a Perkin Elmer Series 4 Liquid chromatograph pump (Perkin-Elmer), and a 250 mm × 4 mm Lichrosorb SI-60 column (E Merck, Darmstadt, Germany). The mobile phase consisted of 900:99:1 HPLC-grade iso-octane, tetrahydrofuran, and acetic acid.The detector was a Perkin Elmer LS-4 Fluorescence Spectrometer (Perkin-Elmer) with an excitation wavelength of 290 nm and an emission wavelength of 320 nm. Flow rate was 1 mL/min. The retention time of α-tocopherol was 5.2 min. The standard used was DL-α-tocopherol (Eastman Kodak Company, Rochester, NY, USA).

Vitamin A (retinol) was extracted from serum and liver tissue and assayed by methods described previously (Mooney, 1992; Anderson et al, 1995b). Retinol was extracted from serum and tissue using a modification of that described by Chew et al (1991). Tissue was homogenized in acetone using a Polytron Homogenizer (Brinkmann Instruments, Westbury, NY, USA). Before extraction, isoproponal was used instead of ethanol to precipitate the protein. Extraction procedures were performed under dark conditions with either yellow or subdued light. Retinol concentration was determined by HPLC analysis of 20 μL aliquots. The HPLC system was the same as used for α-tocopherol analysis except for the mobile phase, an 800:199:1 mixture of iso-octane, tetrahydrofuran and acetic acid, and

the mode of detection. Retinol was detected by absorbance spectroscopy at a wavelength of 254 nm (Spectroflow 757 Absorbance Detector, ABI Analytical, Ramsey, NJ, USA). The retention time was 10.5 min. The standard was all trans retinol (Sigma Chemical Co, St Louis, MO, USA).

Statistics

Data were analyzed using PROC GLM of SAS (1988) as a 2 × 2 factorial arrangement of treatments within a completely randomized design with the factors being dietary vitamin E level and whether or not gilts were injected with vitamin A. Tissue data were log transformed before analysis to improve homogeneity of variance. The experimental unit was the individual gilt. A univariate repeated measures ANOVA was performed on the blood serum data. A time × treatment interaction was present and separate analysis for treatment effects were conducted for each sampling date.

RESULTS

Previous exposure to elevated dietary vitamin A had no influence ($P > 0.10$) on any criteria measured in the gilts except liver retinol. Initial (d–7) serum α-tocopherol concentrations in the gilts were similar ($P > 0.10$; table II) between treatment groups. When dietary level of vitamin E was increased from 25 to 500 IU/kg of diet, average serum α-tocopherol concentration increased ($P < 0.01$) by day 0 and this new concentration was maintained throughout the duration of the study.

Serum concentration of α-tocopherol was not affected ($P > 0.10$) by injecting gilts three times with vitamin A (retinyl palmitate) at 350 000 IU each injection, except on day 7 (table II). On day 7, the gilts fed low (25 IU/kg) dietary vitamin E had similar serum α-tocopherol concentration regardless of vitamin A treatment, whereas gilts fed the high (500 IU/kg) vitamin E level had higher serum α-tocopherol concentration when injected with vitamin A than gilts not injected with vitamin A (E × A, $P < 0.08$; table II, footnote g). However, the difference noted was small.

In general, there were no consistent effects on serum retinol concentration due to dietary supplementation of vitamin E or injection of vitamin A (table III). However, differences ($P < 0.08$) in serum retinol were observed on day 0 in that the average con-

Table II. Main mean serum α-tocopherol concentrations in gestating gilts given dietary additions of vitamin E and injected with vitamin A[a].

Sampling day [b]	*Vitamin E, IU/kg* [c, d]		*Vitamin A injection* [e]		*SE* [f]
	25	*500*	*No*	*Yes*	
	μg/mL				
–7	0.8	0.9	0.9	0.8	0.12
0	1.2	3.6	2.4	2.4	0.13
7[g]	1.1	3.7	2.3	2.6	0.11
24	1.3	3.7	2.5	2.5	0.11

[a]Each mean is based on information from 14, 15 or 16 gilts; [b]days relative to breeding (d0). Day–7 differed ($P < 0.01$) from other days upon addition of 500 IU vitamin E (repeated measures ANOVA, MSE = 0.14); [c]amount added to diet; [d]effect of vitamin E ($P < 0.01$), d0, 7 and 24; [e]three injections of 350 000 IU each; [f]n = 14; [g]effect of vitamin A ($P < 0.06$); effect of E × A ($P < 0.08$; 1.1, 1.1, 3.4 and 4.0 μg/mL for 25 IU vit E/no vit A inj, 25 E/A inj, 500 E/no A inj and 500 E/A inj treatments, respectively).

centration was higher ($P < 0.08$) in gilts injected with vitamin A than in the non-injected gilts, and was also higher ($P < 0.02$) in gilts fed the high vitamin E diet than in those fed the lower level. The differences were small and within the normal range for serum retinol concentration usually found in the pig.

Tissue α-tocopherol concentration in gestating gilts increased ($P < 0.01$) in all tissues, except adipose, as the dietary supplementation of vitamin E increased (table IV). Vitamin A injections had no effect ($P > 0.10$) on tissue α-tocopherol concentrations except in the endometrium. There was a vitamin E × vitamin A inter-

Table III. Main mean serum retinol concentrations in gestating gilts given dietary additions of vitamin E and injected with vitamin A[a].

Sampling day[b]	*Vitamin E, IU/kg*[c, d]		*Vitamin A injection*[e, f]		
	25	*500*	*No*	*Yes*	*SE*[g]
	μg/mL				
−7	0.53	0.52	0.52	0.53	0.02
0	0.46	0.55	0.47	0.54	0.02
7	0.41	0.44	0.43	0.42	0.02
24	0.46	0.47	0.47	0.46	0.01

[a]Each mean is based on information from 14, 15 or 16 gilts; [b]days relative to the breeding (d0); [c]amount added to diet; [d]vitamin E effect ($P < 0.02$), d0 only; [e]three injections of 350 000 IU each; [f]vitamin A effect ($P < 0.08$), d0 only; [g]n = 14 (repeated measures ANOVA MSE = 0.01).

Table IV. Main mean tissue α-tocopherol concentrations in gestating gilts given dietary additions of vitamin E and injected with vitamin A[a].

Tissues[b]	*Vitamin E, IU/kg*[c]		*Vitamin A injection*[d]		
	25	*500*	*No*	*Yes*	*SE*[f]
	μg/g[e]				
Liver	4	24	14	14	1.1
Back fat	7	9	7	9	1.0
Leaf fat	9	12	10	11	1.5
Semimembranosus	3	4	3	3	0.2
Rhomboideus	3	7	5	5	0.4
Endometrium[g]	1	5	3	3	0.2
Embryo	0.4	0.8	0.6	0.6	0.03
Oviduct	1	4	3	2	0.2
Uterus	1	4	3	3	0.1
Ovary	19	97	61	55	3.8

[a]Each mean is based on information from 14, 15, or 16 gilts; [b]effect of vitamin E ($P < 0.01$) for all tissues except back fat and leaf fat; [c]amount added to diet; [d]three injections of 350 000 IU each; [e]wet tissue basis; [f]n = 14; [g]effect of vitamin A ($P < 0.08$); effect of E × A ($P < 0.04$; 2, 1, 4, and 5 μg/g for 25 IU vit E/no vit. A inj, 25 E/A inj, 500 E/no A inj and 500 E/A inj treatments, respectively).

action ($P < 0.04$) in the endometrium. Gilts fed the low vitamin E diet and injected with vitamin A had slightly lower α-tocopherol concentration in their endometrial tissue, whereas gilts fed the highest vitamin E and injected with vitamin A had higher α-tocopherol concentration than the non-injected gilts (table IV, footnote g). Alpha-tocopherol concentration in the embryos was not influenced ($P > 0.10$) by vitamin A injections.

Among the tissues sampled, the highest average concentration of α-tocopherol due to high vitamin E supplementation was found in the ovary followed by liver, adipose, *rhomboideus,* endometrium, and three tissues (*semimembranosus,* oviduct, uterus) with similar concentrations (table IV). Embryonic tissue had the lowest α-tocopherol concentration (units per wet tissue basis).

Injecting vitamin A had no effect ($P > 0.10$) on retinol concentration in the liver. Previous dietary vitamin A treatment resulted in increased ($P < 0.01$) liver retinol concentration. There was no evidence, however, of previous and present treatments influencing liver concentration as no interactions were noted ($P > 0.10$). Concentration in the liver averaged 381 μg/g (wet tissue basis).The ester form of retinol was not quantified in the liver.

The reproductive data are presented in table V. Evaluation of reproductive performance is preliminary due to the small number of gilts involved. However, even with the small numbers, embryonic survival tended to be improved ($P = 0.10$) in gilts injected with vitamin A and fed either the low or high vitamin E diet compared with gilts not given vitamin A injections.

DISCUSSION

Brief and Chew (1985) reported higher embryonic survival and larger litter size in gilts injected weekly with vitamin A (12 300 IU) and β-carotene (33 mg) than those in gilts fed vitamin A and β-carotene at the same level. These gilts, however, were depleted of vitamin A and β-carotene for 5 weeks before the initiation of the study. Coffey and Britt (1993) observed an average one-half pig

Table V. Mean reproduction response criteria of gestating gilts given dietary additions of vitamin E and injected with vitamin A[a].

Items	*Treatment*				*SE*[d]
	25 IU vitamin E[b]		*500 IU vitamin E*		
	– vitamin A	*+ vitamin A*[c]	*– vitamin A*	*+ vitamin A*	
No of CL	14.2	14.0	14.8	15.9	0.8
No of embryo[e]	11.9	12.5	12.4	15.2	1.1
Embryonic survival[f] (%)	81	90	84	95	5.8
Embryo weight (g)	8.5	9.3	7.2	7.7	1.0
Ovary weight (g)	13	13	14	15	0.9
Uterus weight (kg)	2.5	2.4	2.0	2.4	0.2

[a]Each mean represents data from seven gilts (eight gilts treatment 2, 25 IU E + A); [b]amount added to diet; [c]three injections of 350 000 IU each; [d]n = 7; [e]effect of vitamin E ($P = 0.14$), effect of vitamin A ($P = 0.13$); [f]effect of vitamin A ($P = 0.10$).

increase in the number of pigs born alive in sows given im injections of vitamin A palmitate (50 000 IU) compared with sows given vehicle only (corn oil), on day of weaning, mating, and 7 days after mating. All sows in that study were also supplemented with 11 000 IU of vitamin A acetate per kilogram of diet. Schoenbeck et al (1994), however, noted inconsistent results on injection of vitamin A or β-carotene at weaning on subsequent litter size of sows in a large on-farm trial.

The injected levels for vitamin A utilized in the present study were higher than those utilized in the above previous studies. The injected level was based on previous research and had been found to be a safe upper limit for eliciting a response, but was not toxic to the gilts (Mooney, 1992).The gilts in the present study did not display visual symptoms of vitamin A toxicity. Preliminary results from a large, long term regional study have also noted no evidence of toxicity upon injection of one million IU vitamin A to gilts and sows (unpublished results, S-145 Committee, American Society of Animal Science, Savoy, IL, USA). Levels of added dietary vitamin E used were chosen to reflect NRC (1988) requirements and to give an elevated level (20 to 25 × requirement).

The increases in serum and tissue α-tocopherol concentrations noted in our study upon increased dietary vitamin E supplementation are in agreement with previous studies (Jensen et al, 1988; Asghar et al, 1991; Mahan, 1991; Anderson et al, 1995a, b). Average α-tocopherol concentration increased twofold in the embryos upon high dietary supplementation indicating that α-tocopherol is transferred from the dam to the embryo following high dietary supplementation of the dam.

The lack of increase in serum and liver retinol concentrations in our study after vitamin A injection agrees with Mooney (1992). In contrast, Brief and Chew (1985) noted increased plasma vitamin A concentrations following injections of vitamin A; however, the gilts used in their research were fed vitamin A-deficient diets before the study, and the gilts were also injected with β-carotene. Serum retinol may have been elevated upon injection of vitamin A in our study, but may have been missed since blood samples were not taken until 7 days after injection. In addition, the alcohol form of vitamin A, retinol, was measured in our study, which may also explain the lack of increased serum concentration as well as the lack of an increased liver concentration after injection. Most of the vitamin A in blood, however, is as retinol, whereas most in the liver is in the ester form (Blomhoff et al, 1990). However, we have previously noted a ninefold increase in liver retinol concentration at slaughter as a result of a tenfold increase in dietary level of vitamin A in a study with growing and finishing pigs (Anderson et al, 1995b).

There was no consistent evidence found in this study that injecting vitamin A (3 × 350 000 IU/injection) interfered with serum or tissue concentrations of α-tocopherol in gestating gilts fed diets supplemented with 25 or 500 IU of vitamin E. However, α-tocopherol concentration was increased further in the endometrial tissue when vitamin A was given along with high dietary vitamin E. No evidence was found that injections of vitamin A interfered with the transfer of α-tocopherol to the developing embryo. In contrast, Weaver et al observed a decrease in plasma tocopherol concentration upon feeding higher levels of vitamin A. The research of Weaver et al (1989), however, was carried out with young, growing pigs and the vitamin A was administered in the feed. Also there was no consistent evidence in our study that dietary vitamin E interfered with serum or liver concentration of retinol.

In conclusion, the vitamin E status of gilts during early gestation was not detrimentally influenced by three 350 000 IU

injections of vitamin A shortly before, at and shortly after breeding.

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REFERENCES

Abawi FG, Sullivan TW (1989) Interaction of vitamins A, D_3, E, and K in the diet of broiler chicks. *Poult Sci* 68, 1490-1498

Anderson LE Sr, Myer RO, Brendemuhl JH, McDowell LR (1995a) Bioavailability of various vitamin E compounds for finishing swine. *J Anim* Sci 73, 490-495

Anderson LE Sr, Myer RO, Brendemuhl JH, McDowell LR (1995b) The effect of excessive dietary vitamin A on performance and vitamin E status in swine fed diets varying in dietary vitamin E. *J Anim Sci* 73, 1093-1098

Asghar A, Gray JI, Miller ER, Ku PK, Booren AM, Buckley DJ (1991) Influence of supranutritional vitamin E supplementation in the feed on swine growth performance and deposition in different tissues. *J Sci Food Agric* 57, 19-29

Blakely SR, Mitchell GV, Jenkins MY, Grundel E, Whittaker P (1991) Canthaxanthin and excess vitamin A alter α-tocopherol, carotenoid and iron status in adult rats. *J Nutr* 121, 1649-1655

Blomhoff R, Green MH, Berg T, Norum KR (1990) Transport and storage of vitamin A. *Science* 250, 399-404

Brief S, Chew BP (1985) Effects of vitamin E and β-carotene on reproductive performance in gilts. *J Anim Sci* 60, 998-1004

Chew BP, Wong TS, Michal JJ, Standaert FE, Heirman LR (1991) Kinetic characteristics of β-carotene uptake after an injection of β-carotene in pigs. *J Anim Sci* 69, 4883-4891

Chung YK, Mahan DC, Lepine AJ (1992) Efficacy of dietary d-α-tocopherol and dl-α-tocopheryl acetate for weaning pigs. *J Anim Sci* 70, 2485-2492

Coffey MT, Britt JH (1993) Enhancement of sow reproductive performance by β-carotene or vitamin A. *J Anim Sci* 71, 1198-1202

Hatam LJ, Kayden HJ (1979) A high-performance liquid chromatographic method for the determination of tocopherol in plasma and cellular elements of the blood. *J Lipid Res* 20, 639-645

Jensen M, Hakkarainen J, Lindholm A, Jonsson L (1988) Vitamin E requirement of growing swine. *J Anim Sci* 66, 3101-3111

Mahan DC (1991) Assessment of the influence of dietary vitamin E on sows and offspring in three parities: reproductive performance, tissue tocopherol, and effects on progeny. *J Anim Sci* 69, 2904-2917

McMurray CH, Blanchflower WJ (1979a) Application of a high-performance liquid chromatographic fluorescence method for the rapid determination of α-tocopherol in the plasma of cattle and pigs and its comparison with direct fluorescence and high-performance liquid chromatography-ultraviolet detection methods. *J Chromatogr* 178, 525-531

McMurray CH, Blanchflower WJ (1979b) Determination of α-tocopherol in animal feedstuffs using high-performance liquid chromatography with spectrofluorescence detection. *J Chromatogr* 176, 488-492

Mooney K (1992) The effects of supplemental vitamin A or β-carotene on reproduction and the localization of β-carotene during gestation in gilts. MS Thesis, Univ of Florida, USA

NRC (1988) Nutrients requirements of swine, 9th ed, National Academy Press, Washington, DC, USA

SAS (1988) SAS User's Guide, Statistics SAS Inst Inc, Cary, NC, USA

Schoenbeck RA, Thompson J, Didion BA (1994) A comparison of vitamin A supplements on reproductive performance of weaned sows. *J Anim Sci* 77, suppl 1, 375

Weaver EM, Libal GW, Hamilton CR, Parker IS (1989) Relationship between dietary vitamin A and E on performance and vitamin E status of the weaned pig. *J Anim Sci* 67, suppl 2, 113