Vitamin and Mineral Transfer During Fetal Development and the Early Postnatal Period in Pigs1,2,3,4

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ABSTRACT: There are periods during pregnancy when sows may have a temporally high requirement for certain vitamins and minerals. Proteins transferring retinol and Fe to the developing pig fetus have been discovered, whereas transport mechanisms for other vitamins and minerals are probably present but have not yet been identified. Sow body tissues can serve as a reservoir for many micronutrients, but it is not known whether these reserves can supply an adequate quantity during critical fetal developmental periods. There is a low placental transfer of vitamin E to the fetus even if the dietary concentration fed to a gestating animal is high, but colostrum and milk concentrations can be increased when the nutrient is fed to sows. If the dam’s diet contains inadequate Ca or P, the concentration of these elements in the developing fetus and milk will not be affected. Consequently, sow bone demineralization will occur under conditions of dietary inadequacy of Ca and P. Other nutrients can be depleted from sow tissue reservoirs over several parities (e.g., Se), resulting in low quantities being provided in the milk for nursing pigs. Scientific information involving adequate vitamin and mineral nutrition for female pigs to improve conception rate and embryonal survival that will result in optimum fetal and postnatal pig development can be considered to be in its infancy.

Key Words: Reproduction, Fetus, Vitamins, Minerals, Pigs


Introduction

In adult swine, vitamins and minerals are required for a variety of biochemical functions. These same nutrients are likely required by fetal pigs, but the timing and mechanism for delivery for conceptus tissue formation are not completely understood. Early pregnancy is often considered a relatively safe period when nutrient demands on females are low and not particularly critical. We now recognize that the initial half of pregnancy establishes micronutrient transport mechanisms for the conceptus, whereas during the latter half of pregnancy these and possibly other mechanisms transport larger quantities of these nutrients to fetal and mammary tissue.

Brambel (1933) noted that early uterine secretions were likely an important nutrient source for early conceptus formation. Murray et al. (1972) demonstrated an array of uterine proteins during pregnancy, but their rate of secretion changed as gestation proceeded. Subsequent research revealed that these uterine secretions were induced principally by progesterone and that their release into the lumen was possibly stimulated by estrogen. Although passive transfer of some nutrients undoubtedly occurs between the maternal-fetal blood barrier, most micronutrients are probably more dependent on an active transport mechanism. A tremendous amount of metabolic activity takes place in placental and fetal tissues, requiring an ample supply of most vitamins and minerals. The antioxidant vitamins not only may have a role in the attainment and maintenance of pregnancy, but also in fetal development.

This review will evaluate the current status of vitamin and mineral transfer to the developing pig
fetus and the nutrient status of neonatal pigs as they enter postnatal life. Because of an inadequate amount of available research, not all of the vitamins and minerals will be addressed, and nutrients of common biological function will be discussed jointly.

Discussion

**Vitamin A and β-Carotene.** Vitamin A is the generic name given to compounds having the biological activities of retinol; the activities include paracrine, protein modifying, and antioxidant functions. Dietary retinyl esters are hydrolyzed by the pancreatic esterases in the digestive tract, becoming re-esterified in the mucosal cells of the small intestine. These retinyl esters are incorporated into chylomicrons as they pass through the mucosal cell and are thus transported associated with hydrophobic lipoproteins in the lymphatic circulation system. These lipoproteins are transported to and stored in the parenchymal and satellite cells of the liver, where retinol is stored as retinyl esters (Blomhoff et al., 1992). Retinol subsequently released from liver storage depots becomes bound to retinol binding protein (RBP), which is also secreted by the liver (Rask, 1974). The resulting complex is the major transport form of vitamin A. Retinol bound to RBP is not subject to oxidation, so RBP concentrations will regulate plasma retinol concentration. The secretion of RBP is regulated by estrogen and liver stores of retinol (Combs, 1992).

Delivery of retinol to the developing conceptus is accomplished through endometrial synthesis of RBP (Adams et al., 1981; Clawitter et al., 1990; Harney et al., 1993). Secretion of endometrial RBP seems to be largely controlled by progesterone (Adams et al., 1981; Trout et al., 1992) and may be influenced by estrogen (Trout et al., 1992). The synthesis and secretion of endometrial RBP has been shown to increase 390-fold between d 10 and 13 postcoitum; with a further twofold increase between d 19 and 30 of pregnancy (Harney et al., 1993, 1994a; Vallet et al., 1996). Endometrial RBP may be taken up by conceptus/placenta tissue by fluid phase pinocytosis and delivered into fetal blood, similar to mechanisms described for uteroferrin (Raub et al., 1985). The correlation between size of the embryo and the retinol content in uterine flushings is high (r = .97; Trout et al., 1992; Seijas, 1995). Retinol in the uterine secretions may function by protecting the conceptus against the oxidizing activity of uteroferrin (Vallet, 1995). Consequently, RBP may have several functions within the reproductive tract of pigs: 1) transfer of retinol to the fetus, 2) protection of tissues against oxidative reactions, and 3) provision of a substrate to generate retinoic acid and other biologically active metabolites of retinol. Retinoic acid and other metabolites of retinol have important functions in cellular differentiation processes (Mangelsdorf et al., 1993). Messenger RNA for cellular RBP and retinoic acid receptors α and γ have been detected in pig conceptus and endometrial tissues collected on d 15 of pregnancy (Harney et al., 1994b).

Increasing the dietary level of vitamin A has not resulted in increased blood concentrations of retinol (Hoppe et al., 1992), suggesting that either it is stored or cleared by the liver. Delivery of large quantities of retinol can be easily achieved via injection (Chew, 1993; Coffey and Britt, 1993). The injection 10,000 IU retinyl palmitate, which is five times the daily NRC (1988) requirement for this vitamin, into neonatal gilts has raised total plasma retinol (retinol plus retinyl esters) concentration by approximately 20-fold, but there was only a 28% increase in plasma RBP (Vallet et al., 1995). The effect of injecting retinyl palmitate on the secretion rate of endometrial RBP and/or delivery to the fetus in the mature female pig has not been reported.

The effect of administering vitamin A on subsequent reproductive performance has had mixed responses. Coffey and Britt (1993) demonstrated that after the injection of 200 mg of provitamin A (β-carotene) at weaning, litter size was increased in multiparous but not in primiparous sows. In contrast, Tokach et al. (1994) conducted a study on a commercial swine farm with 956 sows and demonstrated that a single injection of β-carotene, vitamin A, or the combination of the two at weaning did not affect subsequent reproductive performance. Feeding diets without supplemental vitamin A has not lowered the sow's reproductive performance for at least two reproductive cycles, suggesting that liver vitamin A stores are in most cases adequate to maintain pregnancy and to prevent congenital defects in the progeny (Hjarde et al., 1961; Selke et al., 1967). Various factors such as dietary nitrates or nitrates can, however, reduce liver vitamin A stores (Seerley et al., 1965). A dietary concentration of 7,000 IU of vitamin A per day was adequate to maintain serum and liver vitamin A concentrations in reproducing female pigs (Parrish et al., 1951).

**Vitamin E and Selenium.** Severe vitamin E and Se deficiencies in reproducing sows reportedly cause fetal resorption (Adamstone et al., 1949) and a reduction in litter size (Mahan et al., 1974). How these nutrients function in swine reproduction is not known, but the antioxidant properties of both nutrients may be their principal mode of action. The most biologically active form of vitamin E (α-tocopherol) crosses the placental barrier of pigs at a relatively low rate. Consequently, neonatal pigs are born with a low body α-tocopherol content (Mahan, 1991). Loudenslager et al. (1986) demonstrated that an Fe injection (200 mg) into neonatal pigs resulted in a lowered serum α-tocopherol concentration, suggesting that the antioxidant status of young pigs can be challenged upon the administration of the prooxidant element.
In contrast to vitamin E, Se can effectively cross the placental barrier of the dam, but the retention by developing fetuses is affected by the dietary concentration and source of Se fed to the dams (Mahan and Kim, 1996). The relative concentration of Se in developing fetuses declines as pregnancy progresses; the total Se content in neonates is approximately 0.07 mg (Mahan, unpublished data). Neonatal pigs from sows fed sodium selenite have a lower liver Se concentration than pigs from sows fed an organic Se source (Mahan and Kim, 1996). This may help to explain why sodium selenite fed to pregnant females may result in progeny more prone to the deficiency.

Because neonatal pigs are born with low body concentrations of α-tocopherol and a relatively low Se content, they must rely on lactating dams to provide a source of these nutrients. α-Tocopherol is effectively transported across mammary tissue. Colostrum α-tocopherol concentration can be elevated by increasing the gestation dietary level of vitamin E or via injection during the last 14 d of pregnancy (Chung and Mahan, 1995). Colostrum α-tocopherol concentration is approximately fivefold higher, with Se concentrations approximately three times higher than later milks (Mahan and Moxon, 1978; Mahan, 1991, 1994). Sow serum α-tocopherol concentration has been shown to decline during the last 10 d of pregnancy (Mahan, 1996), suggesting its diversion to the mammary tissue.

Neonatal progeny from Se/vitamin E-deficient sows are subject to Fe toxicity upon Fe administration (Lannek et al., 1962; Tollerz, 1973). Neonates must therefore consume colostrum that has a high Se and α-tocopherol content or receive an injection of these nutrients if the Se/vitamin E deficiency is to be prevented. The α-tocopherol and Se concentrations in sow’s milk declines as sows become older (Mahan, 1991, 1994), supporting the observation that the occurrence of the deficiency is encountered more frequently in the progeny of older sows.

Riboflavin. Riboflavin is absorbed by active transport mechanisms in the free form in the proximal region of the small intestine, where much of it is quickly phosphorylated to a flavomononucleotide (FMN). Consequently, the vitamin is present in plasma either as free riboflavin or as FMN; both are bound to plasma proteins by weak hydrogen bonds (Combs, 1992). Estrogen stimulates the secretion of a riboflavin-binding protein in the liver of several species. Riboflavin-binding protein has been found in the uterine secretions of mature reproducing females (laying hens, pregnant cows, pregnant mice, and rats) and is believed to be involved in the transplacental/transovarian movement of riboflavin (Combs, 1992).

Murray et al. (1980) identified a yellowish secretion from the uterus of gilts on d 6 to 8 postcoitum that was later found to be riboflavin. Its secretion was stimulated by estrogen and/or progesterone. Bazer and Zavy (1988) later demonstrated that feeding 100 mg of riboflavin to gilts from d 4 to 7 postcoitum increased the number and percentage of live embryos. These workers concurrently reported that 100 mg of riboflavin fed from d 4 to 7 postcoitum resulted in an increased farrowing rate and number of live pigs born in a group of 99 gilts. Pettigrew et al. (1996) reported that high riboflavin supplementation to sow diets during the initial 21 d of pregnancy increased the percentage of sows farrowing, but it did not improve litter size. Frank et al. (1984) evaluated various dietary levels of riboflavin throughout the entire gestation period and demonstrated that 6.4 to 6.6 mg of riboflavin/d was adequate to meet the needs of reproducing sows.

These results suggest that although the mechanism of riboflavin transfer into the intrauterine environment is not known, a high intake of riboflavin during early pregnancy may increase riboflavin transfer to the conceptus, improving conception rate and/or embryonal survival. Although there is clearly a metabolic need for dietary riboflavin, these results suggest that there may also be specific periods when the riboflavin requirement for reproduction exceeds that of daily metabolic needs.

Folic Acid. Folate is present in most feedstuffs fed to swine, but reported grain values are quite variable. It is generally present in feed grains in the reduced foly polyglutamate form, but it must be cleaved and converted to the mono- or di-glutamate form before absorption from the intestinal tract. Dietary folic acid (foly monoglutamate) and the cleaved foly polyglutamates from grains are absorbed across the intestinal mucosa by an active folate-binding protein transport system that is mediated by sodium. There are three main types of folate-binding receptors (α, β, and γ) in various body tissues including the placenta, choroid plexus, and mammary gland (Combs, 1992).

Folate derivatives function in the body as acceptors or donors of single C-units in amino acid and nucleotide metabolism. Folate is required for thymidine synthesis, which is the rate-limiting step in DNA synthesis (Shane and Stokstad, 1985). Consequently, during the early stages of conceptus development, when DNA, RNA, and protein synthesis is high, the need for folic acid would be expected to be higher than at other periods of pregnancy.

Lindemann (1993) recently reviewed the folic acid requirement of gestating and lactating sows. He reported from a Romanian study by Otel et al. (1972) that when “folycysteine A” (a folic acid-cysteine complex) was injected on d 1 and d 9 postcoitum litter size was increased by 1.5 pigs per litter. Matte et al. (1984a) demonstrated that serum folate declined by approximately 30% from breeding to 7 d postcoitum, and declined another 30% from 30 to 60 d postcoitum. Supplementing the diet with folic acid during early gestation has often increased litter size, particularly with multiparous sows (Lindemann and Kornegay, 1989; Thaler et al., 1989). Matte et al. (1984b) and
Tremblay et al. (1989) demonstrated that folic acid addition improved embryonal survival, particularly when ovulation rates were increased due either to flushing or from hormone stimulation. The supplementation of folic acid during late gestation or lactation has generally not improved subsequent sow reproductive performance (Pharazyn and Aherne, 1987).

Folic acid may have an important role in maintaining the reproductive state of the dam and in early fetal development. The mechanism of action, the identification of the active transport mechanisms in the placenta and(or) conceptus, and the minimum dietary level to achieve these results are not known. From the existing evidence, embryonal survival seems to be improved with added folate, the most critical period being the initial 60 d of pregnancy.

Vitamin C. Petroff (1996) demonstrated that developing ovarian follicles of gilts contain increasing concentrations of ascorbic acid as they reach maturation, and it is further increased in the corpus luteum. Concentrations of ascorbic acid are approximately eightfold higher in a corpus luteum at 40 d of pregnancy than those in a mature follicle.

Zavy et al. (1982) demonstrated that uterine secretions collected during the estrous cycle and early pregnancy contained substantial quantities of vitamin C. Vitamin C combines with uteroferrin and catalyzes lipid oxidation (Vallet, 1995).

During pregnancy, fetal pigs do not synthesize vitamin C and they are dependent on a maternal source of this vitamin (Brown et al., 1972). Plasma ascorbate rises in developing fetuses; its highest concentration is detected by 80 to 100 d of gestation (Wegger and Palludan, 1984). Supplementing the dam's diet with vitamin C has not increased the placental transfer of the vitamin or maternal tissue stores (Wegger and Palludan, 1984). The ascorbic acid gradient transfer across the placenta to the fetus is high, achieving a fetal to sow plasma concentration ratio of 14:1 by 112 d of gestation (Brown, 1984). This suggests an active transport mechanism to the developing fetus, where it is most likely used for connective tissue synthesis. The vitamin may also serve as an antioxidant for the many metabolic reactions taking place in rapidly growing fetal tissue.

Fortification of sow diets with vitamin C has had no effect on fetal or neonatal pig ascorbate concentrations (Wegger and Palludan, 1984). When plasma was collected at the time of birth, Yen and Pond (1983) demonstrated an approximate 3:1 ratio in neonatal to sow plasma ascorbate levels. The stress of parturition and resulting maternal hypoxia has resulted in a lower vitamin C content in the dam's adrenal (Brown et al., 1972).

Sow colostrum ascorbic acid content is high, and milk ascorbate levels decline as lactation progresses (Wegger and Palludan, 1984). Supplementing sow diets with vitamin C has not increased colostrum or milk ascorbic acid contents. Consequently, the plasma ascorbate concentrations of nursing progeny are similar regardless of whether sows' diets have been supplemented with ascorbic acid.

Liver is the site of ascorbic acid synthesis in postnatal pigs, in which synthesis begins within 1 wk of age (Braude et al., 1950). Others have suggested that the rate of synthesis in young pigs may be insufficient to meet their daily requirement, particularly if pigs are weaned early (Wegger and Palludan, 1984). Plasma ascorbate concentrations have been shown to decline as pigs age (Yen and Pond, 1983).

Ascorbic acid synthesis by reproducing sows seems adequate to meet the needs of reproduction and that of the developing fetus. No benefit has been demonstrated when diets for reproducing sows have been supplemented with additional vitamin C.

Other Vitamins. Although other vitamins, particularly the B vitamins, are involved in fetal tissue development and metabolism, the mechanism of how they transfer and whether there is a critical period when they traverse the maternoplacental barrier is largely unknown. Passive transport of water-soluble vitamins is unlikely, because they would not easily traverse the endometrial and placental epithelial plasma membranes. Consequently, receptor sites for these nutrients on endometrial and conceptus tissues and their subsequent transport to the developing fetus are most likely.

Iron. Iron is an integral component of many proteins having a wide array of functions. Upon absorption, the ferric form of Fe is bound by transferrin and transported in the bloodstream to various tissues (Laurell, 1952; Aisen, 1980). The Fe saturation level of transferrin is frequently used as an indicator of iron adequacy. Liver takes up much of the Fe from plasma and subsequently stores it as ferritin.

The regulation of Fe absorption occurs largely in the mucosal cells of the intestinal tract. Intestinal absorption mechanisms present in mature female pigs provide a relatively constant replenishment of body Fe from the diet. Developing fetuses must, however, depend on Fe crossing the maternoplacental barrier. Because neonatal pigs are born with relatively low body Fe contents (e.g., 35 to 50 mg), and the postnatal need for this element is relatively high (e.g., 7 to 16 mg/d; NRC, 1988), there have been several attempts to increase the Fe status of pigs at birth and(or) to increase the amount of Fe transferred to the milk by mammary tissue.

Iron transport to developing conceptuses in swine is accomplished via endometrial secretion of uteroferrin, a purple protein with acid phosphatase activity (Renegar et al., 1982; Roberts and Bazer, 1988). Uteroferrin secreted by the endometrium is taken up by the areolae of the placenta and released to fetal circulation (Raub et al., 1985). Receptors that bind the carbohydrate moiety of uteroferrin have been described for fetal liver (Saunders et al., 1985; Michel
et al., 1992), and these receptors may bind uteroferrin. The Fe of the uteroferrin molecule is used for hemoglobin formation (Ducsay et al., 1982) and liver is the primary site for hematopoiesis activity in developing fetuses. Endometrial secretion of uteroferrin is controlled primarily by progesterone (Schlosnagle et al., 1974; Simmen et al., 1989) and possibly by estrogen (Geisert et al., 1982). The secretion rate of this protein increases 41-fold from d 10 to d 13. This coincides with the onset of synthesis of blood by the yolk sacs of developing conceptuses and an additional 23-fold protein increase from d 19 to 40 of pregnancy. This also coincides with the onset of blood synthesis by fetal liver. Possibly as a result of uteroferrin, the relative concentration of Fe in developing fetuses does not decline as pregnancy proceeds, as with most trace elements (Mahan, unpublished data).

Because of the low saturation rate of plasma transferrin, dietary and injectable Fe have increased the amount of Fe bound in this transport molecule. The transferrin molecule can bind additional Fe only until it becomes saturated by Fe, whereupon the excess Fe is excreted. The capacity of this molecule to bind an exogenous Fe supply helps protect body tissues from Fe toxicity by inhibiting its ability to engage in oxidative reactions (Aisen, 1980; Vallet, 1995). However, increasing the delivery of Fe to fetuses by this route has been only marginally effective, probably because the transfer mechanisms in the endometrium (i.e., uteroferrin) are not increased when supplemental Fe is administered (Ducsay et al., 1984; O'Connor et al., 1989; Guise and Penny, 1990).

**Calcium and Phosphorus.** Many factors influence the absorption, utilization, and metabolism of Ca and P. Control of blood Ca is within relatively narrow limits, ultimately controlling the rate of Ca absorption and consequently bone metabolism. This homeostatic system involves two hormones: calcitonin and parathyroid hormone (PTH). These hormones influence the formation of 1,25-dihydroxycholecalciferol (1,25-[OH]2D) in kidney. When the diet is low or marginal in Ca, the active transport absorption mechanism is stimulated. Intestinal mucosal cells under the influence of PTH and 1,25-(OH)2D produce a protein that binds Ca and facilitates the transfer of this element through the intestinal cell. Passive transport of Ca occurs, but it is the major route of absorption only when the dietary concentrations of Ca are high (Gilbert, 1983). Phosphorus absorption is affected by several factors, but it largely seems to be indirectly absorbed in conjunction with the Ca-protein complex.

Calcium-binding proteins are present in mammary tissue, which helps to regulate the constancy of Ca concentration in the milk supply. The results of Harmon et al. (1975) and Maxson and Mahan (1986) demonstrated the constancy of milk Ca and P concentrations even when sow diets contained wide ranges but constant ratios of both minerals.

The transfer of Ca and P through the placenta for developing pig fetuses has received relatively little attention. Hansard and Itoh (1968) demonstrated that the tissue of fetal pigs at 35 d of development had a higher Ca concentration than during the later (i.e., 70 and 114 d) portion of the gestation period. The transfer mechanism for either Ca or P to the developing fetus has not been determined, but because there are binding proteins in the intestinal tract and mammary tissue, they probably also exist in placental tissue.

Itoh et al. (1967) determined that fetal deposition of Ca was not greatly influenced by maternal dietary levels of this mineral. The bone ash content of neonatal pigs was not affected when sows were fed dietary levels below and above those normally provided to gestating sows (Mahan and Fetter, 1982). This suggests that mineral reserves are diverted from maternal bone tissue to meet fetal development needs when the sow diet provides inadequate levels of these minerals. The prevalence of posterior paralysis in reproducing sows is most common during periods of high Ca and P demand (i.e., late gestation or lactation). Bones with a higher trabecular content (i.e., vertebrae and ribs) have a greater amount of demineralization in reproducing sows than cortical bones (Mahan and Fetter, 1982).

Fetal Ca deposition increases quadratically as pregnancy progresses, the largest increase occurring during the latter part of gestation (Hansard et al., 1966; Itoh et al., 1967). Pregnant sows must meet these needs either from the diet or by mobilizing bone mineral reserves. Nimmo et al. (1981) demonstrated a high incidence of leg problems at the end of the first parity when gilts were fed diets low in Ca and P during their grower and gestation periods.

Milk contains a relatively large and constant supply of Ca and P, and its composition seems to be independent of the dietary content of these minerals. Sow body reserves must therefore be used to maintain this constancy when the dietary supply is inadequate. When lactating sows nurse larger litters, a greater amount of bone demineralization occurs in the dams (Maxson and Mahan, 1986). The bones of reproducing sows had substantially lower mineral contents after three parities than those of females a similar age that remained nongravid (Mahan and Newton, 1995), suggesting substantial metabolism and mobilization of Ca and P from the bones of mature, high-reproducing sows.

**Other Minerals.** Other macro- and microminerals are essential for fetal development and to maintain pregnancy in adult female pigs. Potassium, Na, and Cl are involved in nutrient transport and electrolyte balance in developing fetuses as well as in reproducing females. Adequate dietary levels of I, Zn, and Mg in the diet of pregnant dams are clearly necessary to
prevent congenital defects in the developing fetus (NRC, 1988). Recent evidence indicates that sows fed 250 ppm Cu gave birth to larger litters with pigs that were heavier, suggesting a need for this element perhaps beyond its nutritional role (Cromwell et al., 1993). Even though these and other minerals are all recognized as dietary essentials for reproducing swine, the mechanism of transfer and whether the dietary levels required are higher than currently recommended (NRC, 1988) at specific periods during fetal development is not known.

Implications

For fetal development to proceed normally, high concentrations of specific nutrients are often required during specific periods of pregnancy. Transfer proteins are needed to actively transport nutrients across the placental membranes of the gravid female. A few of the transfer proteins have been identified in pigs. In some cases, the transfer of nutrients across the maternal-placental barrier is clearly insufficient. Consequently, in these cases, the neonates are dependent on the consumption of the nutrients from colostrum and milk. Although sows can buffer against many nutrient deficiencies during gestation and lactation, they may not be able to provide an adequate supply of some nutrients during critical time periods to ensure maximum reproductive performance. Various factors can cause the depletion of certain vitamins and minerals over several parities and can affect a sow’s longevity in the herd.

Literature Cited


synthesis and secretion of retinol-binding protein by the uterus of the pig. Endocrinology 130:2557-2564.