METABOLIC CHANGES ASSOCIATED WITH IRON DEXTRAN AND E. COLI AND THEIR EFFECT ON SUSCEPTIBILITY TO INFECTION

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Relationships with iron, microbial growth, and susceptibility to infection have been reported by many researchers (1). Since baby pigs raised in confinement routinely receive iron supplementation, often as iron dextran injections, we have investigated these relationships in young pigs. Previously we reported that the anti-coli effect of sow's milk could be removed by the addition of iron, both in vitro and in vivo in ligated intestinal segments (2). These observations, together with unpublished work in our laboratory, suggest that high-level iron supplementation may have adverse influences on colibacillary diarrhea and systemic infections in pigs. Bacterial infection is known to depress serum iron (3), thereby limiting the availability of iron to invading microbes. Therefore, we undertook experiments to determine if these effects happen in the pig, if the level of iron supplementation affects the ability of the pig to limit iron availability, and if bacterial toxins in a culture filtrate have the same effect as enteropathogenic E. coli. We further evaluated the effect of level of iron dextran injection on the ability of pig serum to support E. coli growth and on changes in glucose concentration.

In experiment 1, thirty-two pigs were used in a 2x2x2 factorial arrangement of treatments to determine the effects of the (1) level of iron dextran supplementation, (2) infusion of an E. coli 263 culture filtrate, and (3) presence of E. coli 263 in a ligated intestinal segment on the ability of the young pig to limit systemic iron availability. Iron dextran was administered IM at 3 days post partum. At 1 week of age pigs were removed from the sows and fed a milk replacer diet. Vena cava cannulae for obtaining blood samples were fitted at 12 days. The culture filtrate was prepared from an actively growing culture of E. coli 263 in tryptic soy broth and saline; controls were infused with sterile tryptic soy broth and saline. Pigs were laparotimized and a 40 cm ligated segment was made in the upper jejunum of each. Culture filtrate was infused IV and E. coli were injected into ligated intestines at 14 days post partum. Blood was sampled every 2 hours for 22 hours, following which pigs were killed and livers, spleens and kidneys were removed. Hemoglobin, hematocrit, serum iron and total iron binding capacity (TIBC) in blood were determined. Tissue iron content was analyzed using atomic absorption spectrophotometry.

Serum iron was depressed in all pigs within 6 hours, probably from the surgery to install ligated intestinal segments. However, pigs receiving 400 mg of iron (HiFe) exhibited greater serum iron and % saturation and lower TIBC than pigs injected with 100 mg (LoFe). The presence of E. coli in the intestine increased TIBC in LoFe but not HiFe pigs, which coincided with the time of maximal fluid secretion into the intestine. Interactions with iron level were also observed for organ iron content. Whether enteric infection causes an effect similar to systemic infection was not determined conclusively. Young pigs were shown to limit iron availability in a manner similar to other mammals when subjected to inflammation or infection, and this was influenced by the level of iron supplementation.

In experiment 2, the rapidity with which iron appeared in, and was cleared from the serum following IM injection of 0, 100 or 200 mg of iron as iron dextran was determined. Peak serum iron concentrations of 38 and 66 ppm occurred 8 and 16 hours post injection for the 100 and 200 mg treatments, respectively, and clearance of 90% of the peak values occured in about 48 to 56 hours.

Experiments 3 and 4 were conducted to determine the effects of iron dextran supplementation on the ability of pig serum to support E. coli growth (strains 263 and 026:B6). For each experiment, pigs were placed in separate cages at 1 week of age and fed milk replacer diets. One day before the start of each experiment pigs were fitted with vena cava cannulae for blood sampling. Blood samples to be assayed for E. coli growth were drawn following a 14-hour fast, on days 1, 3, 5, 7, 9 and 11 in experiment 3 and 1, 3 and 5 in experiment 4. Complement in serum was inhibited at 56 C and serum was diluted with saline (1:10 in experiment 3 and 1:5 in experiment 4), inoculated with E. coli and incubated. Bacterial counts were determined by serial dilutions and plate counts. Serum was also analyzed for iron and glucose concentrations. The treatments were 0, 100 mg and 200 mg iron as iron dextran in experiment 3, with the same treatments plus 215 and 430 mg of dextran (corresponding to the amounts of dextran in the iron dextran treatments) in experiment 4.

The most notable changes in E. coli growth were the increases associated with 200 mg as compared with 100 mg of supplemental iron. In both experiments this effect was significant for E. coli 263 on day 1, when serum iron levels were greatly elevated, and for E. coli 026:B6 on day 3, when serum iron levels had returned to more normal levels. Most treatment contrasts at other time periods showed no significant differences. Pigs receiving iron dextran had lower serum glucose values than controls in experiment 3. In experiment 4, there was a tendency for serum glucose to be lower in iron injected pigs than in dextran injected pigs, but not uninjected controls.

DISCUSSION: The shift in iron resulting in lowered serum iron concentrations associated with infection has been interpreted as a defensive mechanism by the host to inhibit microbial growth (1). This mechanism was shown here to be active in the pig, and its effectiveness to be influenced by the level of iron supplementation, high iron levels being removed from serum less completely. The iron status of the host has been shown in several test animals to affect the growth of bacteria in the serum (4). These studies showed a similar response in the pig; however, the effect in the serum of young pigs following IM iron injection was a transitory, time-dependent one, which was most clearly observed at high iron dosages (200 mg) 1 or 3 days after supplementation. Lower levels (100 mg) of supplemental iron did not consistently cause significant increases in E. coli growth in serum. Therefore, iron supplementation levels necessary for cellular mediated immunity (5) can be maintained without apparent risk of increasing susceptibility to infection. The importance of changes in serum glucose in response to iron dextran injection is uncertain.

CONCLUSIONS: Pigs respond to inflammation and infection by lowering serum iron levels, and this effect is influenced by the level of supplemental iron. Growth of E. coli in serum of pigs is transiently increased in pigs receiving 200 mg as compared with 100 mg iron as iron dextran.

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