Acute feed intake and acute-phase protein responses following a lipopolysaccharide challenge in pigs from two dam lines

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Abstract

This study was conducted to evaluate the response of two dam lines of pigs to acute increases of LPS. Acute-phase proteins were also measured to determine their potential use as biological indicators of the immune response. Thirty-six pigs (initial body weight = 21.3 ± 0.48 kg) were allotted by dam line (Lines 1 and 2) and sex (castrates and gilts) to one of three LPS dose treatments and penned individually. Treatments were a single i.m. injection of 0 (LPS-0), 25 (LPS-25) or 50 μg LPS/kg body weight (BW) (LPS-50). Acute changes in feed intake were related to a pre-injection baseline intake. Feeders were weighed daily to establish baseline feed intake (average daily feed intake 0–48 h prior to injection). The acute feed intake response (AFIR) was computed as the average daily feed intake 0–48 h after injection divided by baseline intake. Serum was harvested at time 0 and 48 h after injection. LPS-0 pigs grew faster and consumed more feed than the LPS-25 or LPS-50 pigs (0.79 kg/d versus 0.51 and 0.50 kg/d; 1.15 kg/d versus 0.96 and 0.89 kg/d, respectively; P < 0.001). The AFIR of Line 1 castrates and Line 2 gilts was similar for LPS-25 and LPS-50 treatments, while Line 1 gilts and Line 2 castrates had decreased AFIR with increased LPS dose (sex × line × LPS, P < 0.05). Three of 18 castrates died but no gilts died following the LPS challenge (P < 0.10). Castrates had higher haptoglobin (Hpt) concentrations than gilts on d 0 (18.1 units of absorption/mg of protein versus 13.1 units of absorption/mg of protein; P < 0.03). Line 1 pigs had higher C-reactive protein (CRP) concentrations than Line 2 pigs (P < 0.05) on d 0. LPS treatment did not change serum concentrations of CRP, Hpt or ceruloplasmin (Cp). However, the change in serum amyloid A (SAA) concentration decreased quadratically (from 0 to 48 h) with increasing LPS dose (P < 0.02). This change in SAA was negatively correlated with the AFIR (r = −0.80; P < 0.001). In general, castrates...
appear to be more sensitive to endotoxin challenges than gilts. Serum amyloid A, but not the other acute-phase proteins evaluated, was a good biological indicator of immune system activation following an acute lipopolysaccharide challenge when compared to the acute change in feed intake.

Keywords: Pigs; Genotype; Sex; Lipopolysaccharide; Feed intake; Acute-phase protein

1. Introduction

The physiological response in the pig following a lipopolysaccharide (LPS) challenge has been well characterized in regards to the alterations in serum concentrations of various hormones and cytokines, changes in body temperature and reductions in feed intake (Warren et al., 1997; Webel et al., 1997; Wright et al., 2000). Reductions in intake are dose-dependent in intensity and duration (Johnson and von Borell, 1994). This response to LPS is typically alleviated after a 24–48 h period (Wright et al., 2000).

Acute-phase proteins (APP) are synthesized primarily in the liver in response to trauma or infection (Baumann and Gauldie, 1994). A number of hormones can influence the pattern of APP production and release, but this process is predominantly controlled by the proinflammatory cytokines (Steel and Whitehead, 1994). Some APP profiles have been characterized using a turpentine challenge in pigs and these include ceruloplasmin, haptoglobin, α1-acid glycoprotein and C-reactive protein (Eckersall et al., 1996; Lampreave et al., 1994). However, limited data are available evaluating APP in pigs following an LPS challenge.

Currently, it is believed that selection for high rates of lean growth potential has resulted in pigs that are more susceptible to immune stress, when based on disruption of food intake and livability. Boyd et al. (2000) and Leininger et al. (2000) have attempted to provide insight into this area of research with mixed results. Boyd et al. (2000) suggested that lean tissue accretion rate was decreased to a greater extent in pigs with a high genetic potential for lean growth compared to pigs with lower genetic potential for lean growth, while Leininger et al. (2000) was unable to demonstrate genotype differences in rectal temperature, cortisol or cytokine profiles.

This study was designed to evaluate the feed intake and APP response of two genders and dam lines of pigs to increasing doses of LPS. The APP responses were related to feed intake to determine if one or more could serve as a biological indicator of immune activation.

2. Materials and methods

2.1. Animals, experimental design and immune challenge

Thirty-six pigs (initial body weight = 21.3 ± 0.48 kg) were allotted by dam line and sex to receive one of three doses of LPS. This 2 × 3 factorial arrangement consisted of two dam lines (Lines 1 and 2) and sexes (castrates and gilts) and three levels of lipopolysaccharide (Sigma Chemical Co., St. Louis, MO Serotype 0111:B4 L2630). Pigs were penned individually and allowed a 3 d adjustment period. All pigs had ad libitum access to feed and water. The diet was corn-soybean meal based and provided 3.37 Mcal/kg of metabolizable energy and 22.5% crude protein, which met or exceeded minimum requirements for high lean growth genotypes (NRC, 1998). The lines used were the result of mating a common sire line to two different dam lines. The sire line was predominantly Duroc and the dam lines were Yorkshire × Landrace crosses. The immune challenge was administered as an i.m. injection of 0 (LPS-0), 25 (LPS-25) or 50 (LPS-50) μg LPS/kg body weight (BW). The LPS was dissolved in sterile saline at a concentration that would limit injection volume to approximately 2 ml per pig. Pigs receiving 0 μg LPS/kg BW were injected with an equivalent volume of the sterile saline. Feeders were weighed each 24 h to establish baseline feed intake (average daily feed intake –48 to 0 h relative to injection). Acute feed intake response (AFIR) was defined as average daily feed intake 0–48 h after injection divided by baseline intake (∼48 to 0 h). This value is then multiplied by
100 to express the value as a percent. Blood samples were collected by venapuncture at 0 and 48 h after LPS injection. Samples were placed on ice immediately and remained on ice for approximately 2 h. Serum was harvested after centrifugation and stored at −20 °C until later analysis. All procedures were approved by the University of Missouri Animal Care and Use Committee.

2.2. Acute-phase protein analysis

Serum samples (0 and 48 h) were analyzed in duplicate for acute-phase protein concentrations. C-reactive protein (CRP) was analyzed using a commercial kit and the methods described by the manufacturer (Tri-Delta Diagnostics Inc., Morris Plains, NJ). Serum amyloid A (SAA) was also analyzed using a commercial kit according to manufacturer specification (Tri-Delta Diagnostics Inc., Morris Plains, NJ). Plasma ceruloplasmin (Cp) oxidase activity was measured using colorimetric procedures described by Demetriou et al. (1974). Plasma haptoglobin (Hpt) concentrations were determined by measuring haptoglobin/hemoglobin complexing by the estimation of differences in peroxidase activity as described by Makimura and Suzuki (1982). Ceruloplasmin and Hpt were graciously analyzed in the laboratory of Dr. John Arthington (University of Florida).

2.3. Statistical analysis

Treatments were arranged as a 2 × 2 × 3 factorial and analyzed using the GLM procedures of SAS (SAS Inst. Inc., Cary, NC). The statistical model included the main effects of line, sex, LPS dose and all interactions. Pig was the experimental unit. Only those animals surviving the immune challenge were used for all measures of growth performance and the APP analysis (n = 33). Pre-planned orthogonal contrasts were used to test linear and quadratic responses to the level of LPS. Initial BW (d 0) was used as a covariate for growth data.

3. Results

3.1. Growth performance

Growth performance measures are presented in Table 1 for the 7 d trial period. Average daily gain (ADG), average daily feed intake (ADFI) and d 7 BW

<table>
<thead>
<tr>
<th>Line 1</th>
<th>Castrates</th>
<th>Gilts</th>
<th>Line 2</th>
<th>Castrates</th>
<th>Gilts</th>
<th>S.E.M.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>25</td>
<td>50</td>
<td>0</td>
<td>25</td>
<td>50</td>
<td></td>
</tr>
<tr>
<td>d 0 BW (kg)</td>
<td>21.0</td>
<td>20.5</td>
<td>20.1</td>
<td>21.1</td>
<td>20.7</td>
<td>21.2</td>
<td>21.2</td>
</tr>
<tr>
<td>d 7 BW (kg)</td>
<td>25.3</td>
<td>24.6</td>
<td>25.6</td>
<td>27.7</td>
<td>25.1</td>
<td>24.5</td>
<td>27.3</td>
</tr>
<tr>
<td>ADG (g/d)</td>
<td>562</td>
<td>467</td>
<td>608</td>
<td>898</td>
<td>540</td>
<td>445</td>
<td>853</td>
</tr>
<tr>
<td>ADFI (g/d)</td>
<td>980</td>
<td>925</td>
<td>930</td>
<td>1220</td>
<td>903</td>
<td>898</td>
<td>1170</td>
</tr>
<tr>
<td>G:F (g/kg)</td>
<td>554</td>
<td>490</td>
<td>654</td>
<td>743</td>
<td>594</td>
<td>468</td>
<td>711</td>
</tr>
<tr>
<td>AFIR (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0–24 h</td>
<td>87.1</td>
<td>9.26</td>
<td>13.5</td>
<td>76.1</td>
<td>47.5</td>
<td>3.2</td>
<td>89.9</td>
</tr>
<tr>
<td>24–48 h</td>
<td>118.0</td>
<td>57.2</td>
<td>58.9</td>
<td>94.7</td>
<td>67.4</td>
<td>29.1</td>
<td>99.6</td>
</tr>
<tr>
<td>0–48 h</td>
<td>102.6</td>
<td>33.2</td>
<td>36.1</td>
<td>85.4</td>
<td>56.1</td>
<td>16.1</td>
<td>94.8</td>
</tr>
<tr>
<td>Mortality (%)</td>
<td>0</td>
<td>33</td>
<td>33</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>33</td>
</tr>
</tbody>
</table>

* Three pigs per line, sex and LPS dose were individually housed and provided ad libitum access to feed and water. LPS dose units are µg/kg body weight (BW). ADG, average daily gain; ADFI, average daily feed intake; G:F, ADG/ADFI.

b D, dam line; S, sex; T, LPS dose; ×, an interaction among main effects. Significance: *P < 0.10, †P < 0.05, ‡P < 0.01, §P < 0.001.

c Day 0 BW used as a covariate for all data.

d Linear effect of increasing LPS dose.

e AFIR, acute feed intake response measured over 0–24, 24–48 and 0–48 h. Expressed as a percent of average daily feed intake 48 h immediately prior to LPS injection.

f Quadratic effect of increasing LPS dose.
decreased with increasing LPS dose (linear, \( P < 0.05 \)). A sex \( \times \) line \( \times \) LPS treatment interaction was observed for d 7 BW, ADG and gain:feed ratio (G:F) (\( P < 0.05 \)). Line 1 castrates and Line 2 gilts had the lowest d 7 BW, ADG and G:F when administered LPS-25 compared to LPS-0 and LPS-50. In contrast, Line 1 gilts and Line 2 castrates, when challenged with LPS-50, had the lowest d 7 BW, ADG and G:F versus the LPS-0 and LPS-25 treatments. However, the number of animals in each treatment cell is insufficient to place much confidence in this outcome.

The acute feed intake response measure indicates that LPS effectively suppressed feed intake following the single i.m. injection at either dose. From 0 to 24 h and 0 to 48 h, the AFIR decreased in a quadratic manner with increased LPS (\( P < 0.05 \)). The AFIR from 24 to 48 h declined linearly with increased LPS dose (\( P < 0.05 \)). The AFIR of Line 1 castrates and Line 2 gilts from 0 to 48 h decreased quadratically with increased LPS; however, the Line 2 castrates and Line 1 gilts AFIR decreased linearly with increased LPS (sex \( \times \) line \( \times \) LPS, \( P < 0.05 \)). Mortality was not affected by LPS dose or dam line, however, castrates tended to die to a greater extent than gilts (3 versus 0; \( P < 0.10 \)).

### 3.2. Acute-phase proteins

The acute-phase protein data are reported in Table 2. The change in APP concentration was calculated as the difference between the concentration at 48 h and the concentration at 0 h. Line 1 pigs had higher CRP concentrations at d 0 compared to Line 2 pigs (\( P < 0.05 \)). However, no differences were observed in d 2 concentrations. Although a 100 \( \mu \)g/ml decrease in CRP concentration was observed for Line 1 pigs following the LPS-25 and LPS-50 doses, there was no effect of LPS treatment, line or interactions.

### Table 2

Acute-phase protein concentrations in two dam lines of pigs following increasing levels of a lipopolysaccharide (LPS) challenge

<table>
<thead>
<tr>
<th></th>
<th>Line 1 Castrates</th>
<th>Line 1 Gilts</th>
<th>Line 2 Castrates</th>
<th>Line 2 Gilts</th>
<th>S.E.M.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>CRP (( \mu )g/ml)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>d 0</td>
<td>262</td>
<td>76</td>
<td>137</td>
<td>106</td>
<td>243</td>
<td>203</td>
</tr>
<tr>
<td>d 2</td>
<td>212</td>
<td>38</td>
<td>52</td>
<td>129</td>
<td>79</td>
<td>89</td>
</tr>
<tr>
<td>Change</td>
<td>-50</td>
<td>-38</td>
<td>-85</td>
<td>23</td>
<td>-164</td>
<td>-114</td>
</tr>
<tr>
<td>SAA (( \mu )g/ml)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>d 0</td>
<td>214</td>
<td>21</td>
<td>76</td>
<td>29</td>
<td>179</td>
<td>49</td>
</tr>
<tr>
<td>d 2</td>
<td>256</td>
<td>721</td>
<td>880</td>
<td>50</td>
<td>660</td>
<td>1172</td>
</tr>
<tr>
<td>Change</td>
<td>43</td>
<td>700</td>
<td>804</td>
<td>21</td>
<td>481</td>
<td>1123</td>
</tr>
<tr>
<td>Cp (mg/100 ml)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>d 0</td>
<td>778</td>
<td>770</td>
<td>619</td>
<td>552</td>
<td>781</td>
<td>724</td>
</tr>
<tr>
<td>d 2</td>
<td>822</td>
<td>895</td>
<td>611</td>
<td>548</td>
<td>659</td>
<td>662</td>
</tr>
<tr>
<td>Change</td>
<td>45</td>
<td>126</td>
<td>-8</td>
<td>-4</td>
<td>-134</td>
<td>-62</td>
</tr>
<tr>
<td>Hpt (uoa/mg of protein)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>d 0</td>
<td>19.3</td>
<td>25.8</td>
<td>11.8</td>
<td>13.7</td>
<td>17.7</td>
<td>17.4</td>
</tr>
<tr>
<td>d 2</td>
<td>31.8</td>
<td>42.7</td>
<td>19.5</td>
<td>15.3</td>
<td>16.4</td>
<td>16.4</td>
</tr>
<tr>
<td>Change</td>
<td>12.5</td>
<td>16.9</td>
<td>7.7</td>
<td>1.6</td>
<td>-1.3</td>
<td>-0.9</td>
</tr>
</tbody>
</table>

\(^a\) Three pigs per line, sex and LPS dose were individually housed and provided ad libitum access to feed and water. CRP, C-reactive protein; SAA, serum amyloid A; Cp, ceruloplasmin; Hpt, haptoglobin. Change: d 2 minus d 0 acute-phase protein concentration.

\(^b\) D, dam line; S, sex; T, LPS dose; \( \times \), an interaction among main effects. Significance: \(^* P < 0.10\), \(^* P < 0.05\), \(^* P < 0.01\), \(^* P < 0.001\).

\(^c\) Linear increase (\( P < 0.001 \)).

\(^d\) Quadratic increase (\( P < 0.02 \)).

\(^e\) uoa/mg of protein: units of absorption/mg of protein.
Interestingly, the change in SAA concentration and the AFIR from 0 to 48 h were highly correlated ($r = 0.80; P < 0.001$; Fig. 1). Ceruloplasmin concentrations were not different at d 0 or d 2 for any treatment group. However, differences were observed in the change in Cp concentration between the lines and sexes. Line 2 pigs had a net increase in Cp concentration, while Line 1 pigs showed a net decrease in Cp concentration ($P < 0.01$). In addition, castrates had greater changes in Cp concentration than gilts ($P < 0.05$).

Differences in Hpt concentration were not observed between line or LPS dose on d 0, d 2 or the change in concentration. Castrates had higher concentrations of Hpt on d 0 compared to gilts ($P < 0.03$). In addition, a sex × line interaction was observed for d 2 serum Hpt concentration ($P < 0.05$). This interaction was the result of Line 1 castrates having a higher concentration than Line 1 gilts, while Line 2 castrates had a lower concentration than Line 2 gilts. The net result was a similar pattern for the change in serum Hpt concentration. Line 1 castrates had a higher change in concentration than Line 1 gilts and Line 2 castrates had a lower change in concentration than Line 2 gilts ($P < 0.02$).

4. Discussion

The acute phase response (APR) following an acute immunological challenge with LPS includes transient fever, stimulation of the hypothalamic–pituitary–adrenal axis, increased serum concentrations of the proinflammatory cytokines and acute-phase proteins, as well as reduced feed intake (Baumann and Gauldie, 1994; Johnson, 1997; Kelley et al., 1994). Suppressions in feed intake following a single LPS challenge has been reported by Warren et al. (1997) and Wright et al. (2000) and this suppression appears to be dosedependent (Johnson and von Borell, 1994). Our results agree with the observations of Johnson and von Borell (1994). As the dose of LPS increased from 0 to 50 µg/kg BW, ADFI decreased in a linear manner. The AFIR from 0 to 24 h and 0 to 48 h decreased quadratically as the dose of LPS increased. While no increase in the serum concentrations of CRP, Cp or Hpt were detected following the administration of LPS, SAA concentration increased quadratically and was highly correlated with the AFIR from 0 to 48 h ($r = -0.80$). This is the first known report to evaluate SAA and Cp concentrations in pigs following an immunological challenge with LPS. It is also the first report of a highly correlated

Fig. 1. Association of the acute feed intake response 0–48 h following a lipopolysaccharide (LPS) challenge to the change in serum amyloid A (SAA) concentration. Feeders were weighed each 24 h to establish baseline feed intake (average daily feed intake averaged over the injection). Acute feed intake response (AFIR) was defined as average daily feed intake 0–48 h after injection divided by baseline intake. Pigs were challenged i.m. at time 0 h with 0, 25 or 50 µg/kg body weight (BW) LPS and serum samples were collected at time 0 and 48 h relative to LPS challenge ((●) 0 µg/kg BW LPS, (▲) 25 µg/kg BW LPS and (■) 50 µg/kg BW LPS).
response between an acute-phase protein and feed intake.

The two dam lines responded, on average, similarly with respect to AFIR and in growth performance when challenged with LPS. The lack of a line or line × LPS treatment interaction may be due to the relative similarities between Lines 1 and 2 progeny. Genotypic response to immune stress was studied earlier by Boyd et al. (2000), who used serial acute challenges over a period of 28 d. In their study, progeny were derived from two different sire lines mated to a common dam line. The LPS challenges reduced ADFI and tended to reduce ADG on both lines. However, G:F tended to improve for the fatter line, coincident with a decline for the high lean growth genotype, thereby eliminating the genetic advantage of the high lean deposition progeny. Leininger et al. (2000) evaluated the APR and the effects on energy metabolites to LPS in three different lines of pigs. The genotypes used in their trial were defined as high lean gain, moderate lean gain terminal cross and a moderate lean gain maternal cross. The three genotypes varied in composition of gain, with the high lean gain pigs having the lowest fat depth while the moderate lean gain maternal line having the greatest fat depth. They did not observe any genotype differences in serum tumor necrosis factor-α (TNF-α) or cortisol following an LPS challenge; however, genotype of the pig did influence serum glucose and insulin and mRNA levels of leptin 10 h post challenge. Physiological responses of the pigs in their trial appeared not to be related to fat deposition.

Our study was not designed to evaluate long-term differences in growth performance (ADG, ADFI and G:F) between the two dam lines or sexes. Nonetheless, a significant sex × line × LPS treatment interaction was observed for ADG and G:F. This effect may have been the result of the small number of observations. Additional research using a greater number of pigs is needed to specifically evaluate this phenomenon.

The linear decreases in the growth performance following the LPS challenge in our study agree with other published reports. Dritz et al. (1996) and Spurlock et al. (1997) documented decreases in ADG and ADFI during a period of serial LPS challenges. In the study by Dritz et al. (1996), non-challenged control pigs allowed ad libitum access to feed and water had similar G:F as LPS challenged pigs. However, pigs that were pair-fed to the same level of feed intake as the LPS challenged pigs had greater G:F than the challenge group. In our study, G:F decreased linearly with increasing dose of LPS. During an inflammatory challenge, nutrients are partitioned away from tissue accretion and utilized by an expanding immune system (Johnson, 1997; Spurlock, 1997). The substantial mobilization of body protein stores required to support the synthesis of APP was illustrated by Reeds et al. (1994).

Acute feed intake was measured over 24 and 48 h periods. Typically, the suppression in feed intake following an immune challenge with LPS subsides between 24 and 72 h (Warren et al., 1997; Wright et al., 2000; Frank et al., 2002). In the current study, the AFIR was quadratically depressed from 0 to 24 h and from 0 to 48 h with the increasing dose of LPS from 0 to 50 μg/kg BW. Feed intake had not returned to the level of the saline administered pigs between 24 and 48 h. Warren et al. (1997) reported that pigs challenged i.p. with 50 μg/kg BW returned to the feed intake levels of control pigs at 24 h following the challenge. Wright et al. (2000) indicated that pigs challenged i.p. with 100 μg/kg BW LPS returned to feed intake levels of control pigs at 24–48 h. The feed intake response of the pigs used in our study exhibited a longer period of intake suppression, even at a dose of 25 μg/kg BW administered i.m. We have also observed that pigs challenged i.v. with 15 or 45 μg/kg BW LPS returned to the intake levels of saline administered pigs between 24 and 48 h, while those pigs challenged with 75 μg/kg BW did not (Frank et al., unpublished data). The differences among these studies may be related to the route of administration, genotype of the pigs, strain (and serotype) of LPS, body weight and/or prior exposure to environmental pathogens.

The increased mortality following the LPS challenge of the castrates compared to the gilts is noteworthy. This response has not been reported previously in pigs in the scientific literature, but is typically observed in practice (D. Boyd, unpublished information). Castrates had greater concentrations of Hpt prior to challenge and had greater changes in serum Cp concentrations following the exposure to LPS. These responses suggest that castrates were either more immunologically stressed or sensitive than gilts. A recent report on causative agents or conditions of mortality in pigs following an immune challenge with LPS is needed as high lean gain, moderate lean gain terminal cross and a moderate lean gain maternal cross.
associated with mortality in nursery pigs found that castrates have a 23% higher mortality than gilts (Larriestra et al., 2002). Sexual dimorphisms in immune competence in human medicine are well documented. For example, females are more prone to develop autoimmune diseases compared to males (Feld and Heathcote, 2003; Soto et al., 2004). More detailed investigations are required to determine if castrates are more sensitive to an LPS challenge than their female counterparts.

Eckersall et al. (1996) evaluated the APP responses of $\alpha_1$-acid glycoprotein, Cp, Hpt and CRP in the pig following a turpentine challenge. They reported that Cp, Hpt and CRP were elevated 2 d following the turpentine challenge. However, $\alpha_1$-acid glycoprotein concentrations did not differ from the controls. This observation agrees with that of Lampreave et al. (1994), who reported no increase in serum $\alpha_1$-acid glycoprotein concentration following a turpentine challenge. Spurlock et al. (1997) using a serial LPS challenge reported an increase in serum $\alpha_1$-acid glycoprotein concentration in pigs. Elevated serum Cp and CRP concentrations by d 2 following a turpentine challenge have also been demonstrated in the rabbit (Giclas et al., 1985). Wright et al. (2000) reported a trend for increased serum Hpt at 24 and 72 h following a single LPS challenge. Taken as a whole, these observations suggest that turpentine and LPS challenge models elicit different APP responses. This would be expected because the cytokine pattern induced by systemic inflammation with LPS differs from the pattern induced by a local inflammatory response from turpentine injection (Moshage, 1997).

We collected serum samples at time 0 and 48 h after our LPS challenges to evaluate the response of APP to a single LPS challenge. Only SAA concentrations changed significantly following the LPS challenge. This was LPS dose-dependent and highly correlated with the 48 h AFIR. Serum amyloid A acts as an apolipoprotein and possibly modulates reverse cholesterol transport; however, this protein may also have a number of other physiological activities (see reviews by Steel and Whitehead, 1994; Jensen and Whitehead, 1998).

The synthesis and release of APP are stimulated by proinflammatory cytokines. These proteins have been classified as type I and type II in various species. Type I APP are stimulated by interleukin (IL)-1-like cytokines including IL-1$\alpha$, IL-1$\beta$ and TNF-$\alpha$. Type I APP include SAA, CRP, Hpt and others. Type II APP are stimulated by IL-6-like cytokines and include fibrinogen, Hpt and others (Baumann and Gauldie, 1994; Moshage, 1997). Webel et al. (1997) and Frank et al. (2003) have shown increases in serum TNF-$\alpha$, IL-1$\beta$ and IL-6 in pigs following an immune challenge with LPS. It was therefore unexpected to see increases in SAA and not the other APP measured in our study.

Ceruloplasmin concentrations were likely influenced by the diet fed to pigs in this study. Copper sulfate was included in the diet at a pharmacological level (250 ppm). This could have contributed to the high baseline Cp concentrations and less opportunity for elevated Cp concentrations following the LPS challenge beyond these levels. It is possible that Cp, CRP and Hpt concentrations were all elevated prior to challenge; however, this is unlikely considering the exceptional growth rate of control pigs. It is also possible that the sampling time did not allow for the detection of changes in Cp, CRP or Hpt. The apparent unresponsiveness of CRP and Hpt to the LPS challenge needs to be studied further. In addition, considerable variation was observed in the serum concentrations of the APP. Due to this variation, it is more difficult to detect differences among the treatments.

The differences in d 0 CRP concentrations between the lines suggest that there was some degree of immune stimulation prior to the LPS challenge, which was exhibited by Line 1 pigs to a greater extent than Line 2 pigs. No detectable differences in the prechallenge performance data were detected (not shown), therefore, the biological basis on which to explain this difference is unknown. This also applies to the sex effects observed with the APP analysis. No clearly defined pattern of response among the lines or sexes was observed.

5. Conclusion

This study indicates that an acute measurement of feed intake is a useful method of evaluating the APR of pigs to an immunological challenge with LPS. Acute feed intake decreased with increasing lipopolysaccharide dose and was highly correlated with
changes in serum concentration of SAA. Therefore, SAA may prove to be a valuable biological indicator of immune stress in growing pigs. The sexual dimorphism in mortality warrants more detailed investigation to evaluate the possible physiological mechanisms involved in this response. Although dam line differences to the acute LPS challenge were not observed, additional research on multiple pig genotypes is needed to provide insight into the variation in the immune response demonstrated by other trials.

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References


