Serum amyloid A concentration as an inflammatory marker in endotoxemic model calves

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Introduction

Serum amyloid A (SAA) is an acute phase protein that increases in concentration in the serum in the early stages of inflammation. SAA is used to diagnose inflammatory diseases in humans, cats, and horses. Such an inflammatory marker to diagnose inflammatory diseases needs to be investigated in cattle. Furthermore, the relationship between systemic inflammation and serum SAA concentrations needs to be examined in order to improve food animal health care. However, no studies have been reported on the sequential change in SAA in cattle with endotoxemia. Therefore, the aim of the present study was to examine serum SAA concentrations in cattle with systemic inflammation caused by endotoxin.

Materials and Methods

Five Holstein-Friesian breed calves weighing 416.0 ± 93.5 lb (188.7 ± 42.4 kg) were enrolled as a control group, and 4 Holstein-Friesian calves and 1 Jersey breed calf, weighing 371.9 ± 46.5 lb (168.7±21.1 kg), were enrolled as a LPS group in the present study. All calves were fit with an indwelling jugular catheter immediately before the endotoxin was infused, and received 2.5 μg/kg bolus doses of O111:B4 LPS (L4391, Sigma-Aldrich, St. Louis, MO, USA) intravenously in 10 mL of autologous serum via the jugular vein. Blood samples (10 mL) were withdrawn from the contralateral jugular vein before and 0.5, 1, 2, 4, 8, 12, and 24 hr after the endotoxin challenge, and stored in both serum separator and heparine-2K-coated tubes. Serum and plasma were harvested after centrifugation at 3,000 rpm at room temperature for 15 min, and stored at -112°F (-80°C) until analyzed. Then, the plasma endotoxin (ETX) activity was measured using the limulus amebocyte lysate kinetic turbidimetric assay (LAL-KTA, Endosafe KTA2, Charles River). SAA concentrations in serum were measured using an automated latex agglutination turbidimetric immunoassay on an automated clinical chemical analyzer (Hitachi 7170S, Hitachi Ltd., Tokyo, Japan). The data are shown as the means ± SD. Each dependent variable within the groups was compared with the baseline value using Dunnett's test after 2-way ANOVA. Measured dependent variables were compared between groups for each sample collection period using the Mann-Whitney U test after 2-way ANOVA. The significance level was $p<0.05$.

Results

Significant changes in plasma ETX activity were not observed in the control group. Plasma ETX activity in the LPS group peaked at 1.239±0.881 EU/mL at t=0.5 hr, which was significantly higher than the 0.057 ± 0.013 EU/mL at pre and 0.029 ± 0.004 in the control group at t=0.5 hr ($p<0.05$). The peak plasma ETX activity in the LPS group returned to pre-challenge values after t=4 hr.

Significant changes in serum SAA concentration were not observed in the control group. Serum SAA concentrations at t=24 hr in the LPS group was 114.26±23.55 μg/mL, which was significantly higher than the 16.72 ± 33.93 μg/mL at pre and 14.45 ± 11.69 in the control group at t=24 hr ($p<0.05$).

Significance

In cattle, C-reactive protein (CRP) cannot be used to diagnose inflammatory diseases because the blood CRP concentration does not change due to inflammation. Accordingly, SAA is suspected to be a better diagnostic marker for inflammatory diseases in cattle. In the present study, serum SAA concentrations in endotoxemic model calves increased due to systemic inflammation. In conclusion, SAA has the potential to be a diagnostic marker for inflammation in cattle.