Effects of continuous versus intermittent transport on plasma constituents and antibody response of lambs

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ABSTRACT: Recommendations for transportation of lambs, horses, calves, and pigs from a committee of the European Commission, which required rest stops of 6 or 24 h, every 8 h, were evaluated using Rambouillet × Suffolk lambs. The lambs of 17.6 ± 0.5 kg of BW were randomly assigned to 1 of 3 treatments: transported for 22 h (continuous; n = 15); transported for 8 h, unloaded and rested for 6 h, transported for 8 h, unloaded and rested for 24 h, transported for 6 h (rested, n = 15); or remained in the home pasture throughout the study (control, n = 16). Off-trailer rest with food and water occurred in novel pens. Food deprivation in the continuous lambs was reflected by a decrease (P < 0.001) in plasma concentrations of glucose and an increase (P < 0.02) in plasma concentrations of blood urea nitrogen, creatinine, and total bilirubin relative to rested or control lambs. Electrolytes varied within and among all 3 treatments (P < 0.05), but no distinct pattern indicating dehydration was evident. Serum concentrations of cortisol were elevated in continuous and rested lambs compared with control lambs at 22 h (P < 0.05). Plasma immunoglobulin G antibody response to ovalbumin was suppressed (P < 0.05) in the continuous and rested lambs relative to the control lambs. Differences (P < 0.05) between continuous and rested lambs indicated the rest stops were sufficient to maintain BW during transport; however, these results were confounded by the control lambs losing a similar (P = 0.50) percentage of their initial BW as the continuous lambs at 22 h. The rest stops eliminated the physiological indicators of food deprivation and maintained BW but did not alleviate evidence of immunosuppression, and 52 h was required to complete the otherwise 22-h-long trip.

Key words: immune function, lamb, rest, transport, welfare

INTRODUCTION

Concern for the welfare of livestock during long-distance transport in Europe led to a report prepared by the Scientific Committee on Animal Health and Animal Welfare that was published by the European Commission (2002). That report reviewed recent literature and proposed methods to improve the welfare of livestock during the various stages of transport. One recommendation for lambs, horses, calves, and pigs was to limit trip duration to 8 h with alternating rest stops of 6 or 24 h required until the final destination was reached. That recommendation, however, was not based on direct scientific evidence.

Specific aspects related to transport, and which affect the welfare of animals, such as loading and unloading stress (Parrott et al., 1998a), posttransport novel environments (Cockram et al., 2000), space requirements (Buchenauer, 1997), environmental conditions (Fisher et al., 2005), and the influence of the driver and road conditions (Cockram et al., 2004) have been studied. The effect on lambs (Cockram et al., 1997) or sheep (Parrott et al., 1998b) of a single rest stop during transport has also been determined; however, the implications of multiple rest stops have not been examined.

The aim of this study was to determine the efficacy of rest stops recommended to improve welfare during transport (European Commission, 2002) using lambs as a model. A multifaceted approach (Moberg, 1987), utilizing differences in plasma constituents, antibody response, and BW at major points during and after transport was used to compare continuous transport...
for 22 h, transport for 22 h with the prescribed rest stops, and remaining in the home pen.

MATERIALS AND METHODS

Animals

All procedures involving animals were approved by the Texas A&M University Agricultural Animal Care Committee.

Forty-six 14-wk-old Rambouillet × Suffolk lambs (23 males and 23 females) were randomly selected from the breeding population of the Physiology of Reproduction lab at Texas A&M University. Before use in this study, lambs were weaned at 8 wk of age and were then raised as a group on bermudagrass pasture. Water was available continuously in a 1-m-long water trough. For 6 wk before transport, all lambs were moved from the adjoining pasture into a covered home pen, where the group was fed 25 kg of a pelleted feedstuff (Producers Cooperative, Bryan, TX) containing 16% CP (DM basis) at 0900 and 1700. During these feeding sessions, lambs were also hand-fed grain and handled to habituate them to human contact. At the beginning of the transport experiments, the lambs averaged 17.6 ± 0.5 kg in BW and would readily approach the researchers.

Experimental Procedure

General. To ensure an equal distribution of sexes in all treatments, the lambs were blocked by sex and then randomly assigned to 1 of 3 treatments: continuous, rested, or control. Before the beginning of transport, lambs were moved from the pasture into the home pen. The schedule of data collection and other related activities is summarized in Table 1. Transport occurred mainly on straight, flat highways and with brief periods on rolling, paved county roads in the vicinity of College Station, TX. All drivers avoided abrupt accelerations or decelerations. Because most of the stress response during transportation occurs during the first few hours of transport, it was preferable to initiate transport treatments at the same time so heat stress, diurnal factors, and other factors were consistent. However, the time of sampling varied at the completion of transport.

Continuous Treatment. Lambs (n = 15) were transported for 22 h. The time spent on the trailer during blood sampling and while the rested lambs were loaded and unloaded was counted as transport.

Rested Treatment. Lambs (n = 15) were transported for 22 h but also had intermittent rest stops with access to food and water, as recommended by the European Commission (2002). After the initial 8 h of transport, the lambs were unloaded for 6 h, reloaded for an additional 8 h of transport, unloaded for 24 h, and then reloaded for the final 6 h of transport. During each rest stop, the lambs were unloaded into a novel pen (9.4 × 4.0 m) with wood shavings for bedding that mimicked the conditions the lambs might experience during a commercial shipment. Supplemental lighting was used at night to allow the observation of behaviors. A novel group of sheep was visible across an alleyway from the rest pens. While in the rest pen, approximately 11 kg of a 16% CP, pelleted feedstuff (Producers Cooperative) and ad libitum grass hay was provided in permanent feeders (6.5 m wide × 0.5 m tall) with adequate space for all lambs to access the feed at the same time. Water was provided in 4 buckets that were an average of 2 m from the feeders. Lambs had no prior experience with the feeding and watering arrangement before the first rest stop. No attempt was made during reloading to sort the rested lambs into the groups in which they were transported before a rest stop.

Control Treatment. Lambs (n = 16) remained in the home pasture throughout the study, except when sampled. Eleven kilograms of a 16% CP, pelleted feedstuff (Producers Cooperative) was provided at the customary times of 0900 and 1700.

Trailer Arrangement. The trailer (7.2 × 1.8 m; Goose-neck, Bryan, TX) had a deck height of 0.4 m, which allowed the lambs to load or unload without a ramp. Slots running the length of the trailer, beginning 0.4 m above the deck, provided ventilation. Each lamb was allowed the recommended space of 0.23 m² (0.19 m² plus 20% due to high ambient temperature; European Commission, 2002) for lambs less than 20 kg. The trailer was divided in half lengthwise and then into 4 pens on each side, each measuring 1.15 m². Three pens per side were used for transport of the lambs, and the remaining pen was used to provide the extra space needed for on-board blood collection and BW measurement. One treatment was located on each side, which allowed for one treatment to be loaded or unloaded without disrupting the other treatment. Pens could be opened to allow access by the researchers and to permit loading and unloading of the entire length of each half of the trailer.

Sample Collection and Analysis

Blood Collection. All blood samples were collected via jugular venipuncture using 20-g needles (VWR, West Chester, PA) and 10-mL Vacutainer tubes containing PST Gel and lithium heparin (VWR). An assistant straddled the lamb and held the head back and to the side to expose the jugular vein. Sample collection took an average of 1 min per lamb, and the lambs displayed only a slight reaction to the process. Treatments were sampled in a consistent order of rested, continuous, and control, and the lambs were randomly sampled within each treatment. Recovery from transport was the 24-h period after cessation of transport, which resulted in a reduced number of measures from continuous lambs. The samples were placed in an ice bath for no longer than 1 h. The samples were then centrifuged for 20 min at 1,000 × g. Plasma was harvested and frozen at −13°C until analyzed. Sampling occurred in
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Table 1. Schedule of major activities and location of sample collection relative to the initiation of transport and time of day

<table>
<thead>
<tr>
<th>Time relative to the beginning of transport</th>
<th>Time of day</th>
<th>Treatment</th>
<th>Activity</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>−2 d</td>
<td>1000</td>
<td>All</td>
<td>Prevaccination blood sample and vaccination with ovalbumin.</td>
<td>Home pen</td>
</tr>
<tr>
<td>0 h</td>
<td>0800</td>
<td>All</td>
<td>Blood sample and BW. Separation into treatments and continuous and rested loaded for transport.</td>
<td>Home pen</td>
</tr>
<tr>
<td>8 h</td>
<td>1600</td>
<td>Continuous</td>
<td>Blood sample.</td>
<td>Trailer</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Rested</td>
<td>Blood sample. Unloaded for 6-h rest period. Provided food and water.</td>
<td>Trailer</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>Blood sample.</td>
<td>Trailer</td>
</tr>
<tr>
<td>14 h</td>
<td>2200</td>
<td>Continuous</td>
<td>Blood sample.</td>
<td>Rest pen</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Rested</td>
<td>Blood sample. Loaded for transport.</td>
<td>Home pen</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>Blood sample.</td>
<td>Home pen</td>
</tr>
<tr>
<td>22 h</td>
<td>0600</td>
<td>Continuous</td>
<td>Blood sample and BW. Conclusion of transport. Start of 24-h recovery period. Provided food and water.</td>
<td>Trailer</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Rested</td>
<td>Blood sample. Unloaded for 24-h rest period. Provided food and water.</td>
<td>Trailer</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>Blood sample and BW.</td>
<td>Home pen</td>
</tr>
<tr>
<td>46 h</td>
<td>0600</td>
<td>Continuous</td>
<td>Blood sample and BW. Conclusion of 24-h recovery period.</td>
<td>Home pen</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Rested</td>
<td>Blood sample. Loaded for transport.</td>
<td>Rest pen</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>Blood sample and BW.</td>
<td>Home pen</td>
</tr>
<tr>
<td>52 h</td>
<td>1200</td>
<td>Rested</td>
<td>Blood sample and BW. Conclusion of transport. Start of 24-h recovery period. Provided food and water.</td>
<td>Trailer</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>Blood sample and BW.</td>
<td>Home pen</td>
</tr>
<tr>
<td>78 h</td>
<td>1200</td>
<td>Rested</td>
<td>Blood sample and BW. Conclusion of 24-h recovery period.</td>
<td>Rest pen</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>Blood sample and BW.</td>
<td>Home pen</td>
</tr>
<tr>
<td>8 d</td>
<td>1000</td>
<td>All</td>
<td>Posttransport immunoglobulin M and immunoglobulin G sample and BW.</td>
<td>Home pen</td>
</tr>
</tbody>
</table>

*Transport began on July 29, 2003, and ended on July 30, 2003, for the continuous lambs and on July 31, 2003, for the rested lambs.*

the home pen, on the trailer, in the rest pen (Table 1), or in a combination of these.

**Plasma Constituents.** Within 48 h of collection, the Texas Veterinary Medical Diagnostic Laboratory used a Hitachi 911 (Hitachi, Ltd., 6-6, Marunouchi 1-chome, Chiyoda-ku, Tokyo, Japan) to determine the total plasma protein, albumin, calcium, phosphorus, glucose, blood urea nitrogen, creatinine, total bilirubin, creatine kinase, aspartate aminotransferase, globulins, albumin/globulin ratio, gamma glutamyl transferase, magnesium, and plasma electrolytes (sodium, potassium, Na/K ratio, and chloride).

Within 6 mo of collection, the concentrations of cortisol were determined by RIA (Coat-A-Count Cortisol, Diagnostic Products Corporation, Los Angeles, CA) using the manufacturer’s specifications. Inter- and intraassay CV were 9.1%. Sensitivity was 0.2 μg/dL for cortisol.

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**Antibody Production.** The effect of treatment on the primary immune response to vaccination against ovalbumin was determined with a modified version of the indirect ELISA described by Ameiss et al. (2004), using 96-well plates coated with 5 μg of ovalbumin per well. The vaccination (Coppinger et al., 1991), a total of 0.5 mg of ovalbumin (Sigma Chemical Co., St. Louis, MO) suspended in 0.125 mL of PBS and 0.125 mL incomplete Freund's adjuvant (Sigma Chemical Co.) was given i.m. in 2 locations on the dorsal thigh 2 d before transport (Table 1).

Blood samples (10 mL), collected before vaccination and 8 d after transport (Table 1), were separated by centrifugation (1,000 × g), and plasma was frozen at −13°C until analyzed. The analysis was conducted within 6 mo of sample collection. Modifications to the indirect ELISA (Ameiss et al., 2004) used to evaluate the antibody response included substitution of ovalbumin for BSA, dilutions of the plasma evaluated of 1:2,560 for the immunoglobulin G (IgG) response and 1:1,280 for the immunoglobulin M (IgM) response, and secondary antibodies (FITC-conjugated, AntiSheep IgG and IgM, Bethyl Laboratories Inc., Montgomery, TX), as appropriate for the antigen and species used in this study.

One negative control sample (1 μL of plasma from an unvaccinated sheep in 10.24 mL of PBS) was included on each plate to ensure that the plate had been properly processed. Each sample (dilution) was analyzed in duplicate. Variations in readings among different ELISA plates were corrected by normalizing the readings from each sample within a plate to a positive control sample (1 μL of plasma from a vaccinated ewe in 159 mL of PBS) included in each plate. The mean of the corrected absorbance at 450 nm was used for analysis of treatment effects. Six plates were used to complete the assay for each antibody at each time of sampling.

**Weight Change.** The continuous, rested, and control lambs were weighed after the plasma sampling using a portable scale (Table 1). To minimize handling and
disturbance of the other lambs, a researcher held the lambs in his arms. The combined weight of the researcher and the lamb was recorded. The known weight of the researcher was used to check the accuracy of the scale.

**Statistical Analysis**

Data were plotted and visually checked for normal distribution. Due to the inherent confounding from the difference in duration of the transport treatments, control lambs were used to compare relative treatment differences at each sampling time. Treatment effects for plasma constituents, IgG and IgM response, and BW were determined using a repeated measures model appropriate for a split-plot design. Factors included in the model were treatment, time, the interaction of treatment with time, and lamb nested within treatment. Lamb nested within treatment was the error term used to test for treatment effects. Analyses were conducted using the GLM procedure (SAS Inst. Inc., Cary, NC). Mean separation was performed using the PDIFF option. When treatment x time interactions were detected, treatment effects were examined within each sample time.

**RESULTS**

The majority of the continuous and rested lambs were observed to be recumbent within the trailer upon returning from transport. Rested lambs readily unloaded and loaded at the start and conclusion of each rest stop, and no apparent injuries were observed. Continuous lambs unloaded at the conclusion of transport without difficulty or visible injury. Mean temperature was 28.4°C with a range of 18.2 to 39.6°C, and mean relative humidity was 59.9% with a range of 29.1 to 92.4% during transport. At the conclusion of transport, continuous and the rested lambs ignored the water and went immediately to the grain after unloading.

A treatment x time effect was evident (P < 0.001) for plasma concentrations of glucose, creatinine, total bilirubin, blood urea nitrogen, and cortisol. Plasma glucose decreased (P < 0.001) in the continuous lambs during transport (Figure 1A). Plasma concentrations of creatinine (Figure 1B), total bilirubin (Figure 1C), and blood urea nitrogen (Figure 2A) increased (P < 0.001) in the continuous lambs after 14 h of transport. Though sporadic differences (P < 0.05) between treatments for sodium, potassium, Na/K ratio, and chloride existed (data not shown), the variation relative to the control lambs suggested that no meaningful treatment effect existed. Concentrations of cortisol were influenced by treatments at 14 and 22 h. The concentration of cortisol was elevated in the continuous lambs (P < 0.05) relative to the control lambs at 14 h, and the concentration of cortisol was greater in the continuous and rested lambs (P < 0.001) relative to the control lambs at 22 h (Figure 2B). The remainder of the plasma constituents were not influenced (P > 0.05) by treatments.

A treatment x time effect was evident (P = 0.01) for the IgG response. Plasma IgG did not differ between treatment groups (Figure 3) before vaccination and initiation of the treatments. Posttransport, samples from the continuous and rested lambs had less absorbency at 450 nm (P < 0.05) than samples from the control lambs (Figure 3). A time effect was evident (P < 0.001) for IgM. The absorbency at 450 nm in the posttransport (8 d after the start of transport) samples was greater (P < 0.001) than the prevaccination (transport) samples (Figure 3).

A treatment x time effect was evident (P < 0.001) for percentage change in BW (Table 2). There was a difference between the continuous and rested lambs (P < 0.05) at the conclusion of transport for each group, which indicated that the rest stops helped the rested lambs maintain their BW. However, the nontransported control lambs had a decrease in BW during the data collection at 22 h that was similar (P = 0.50) to the continuous lambs. Even at 8 d after transport, the continuous lambs had not fully recovered their BW loss (P < 0.05).

**DISCUSSION**

This study was one of the first to quantify the implications of providing multiple rest stops during long distance transport of lambs or other species of livestock. The rest stops benefited the lambs by reducing the observable effects of food deprivation during hot conditions (mean temperature of 28.4°C). However, the elevated concentrations of cortisol at 22 h and suppression of IgG response to vaccination in both transported groups implied the rest stops failed to alleviate transport stress. Additionally, providing livestock with feed during transport to slaughter can be problematic due to the common recommendation that cattle be fasted for 12 to 24 h before slaughter to reduce the possibility of carcass contamination (Savell and Smith, 2004). It would be counterproductive to feed animals during transport and have to fast the animals after arrival at slaughter plants. Additionally, most slaughter plants will not have the space required to hold animals for fasting if they were fed during transport to the plant.

The stops required in this study for sampling and unloading of the rested lambs resulted in the continuous lambs experiencing stationary periods. However, during the commercial transport of lambs for 22 h the drivers will stop for food, water, traffic delays, and to inspect the animals. Also, hauling all transported lambs in the same trailer ensured they were subjected to the same roads, drivers, and environmental stressors to the greatest extent possible.

The report published by the European Commission (2002) contained a discussion of the potential positive and negative effects from on- and off-trailer rest. On-trailer rest was concluded to be preferable to reduce the spread of disease and risk of injury assuming sufficient
Figure 1. Mean concentrations (with SE) bars of (A) glucose, (B) creatinine, and (C) total bilirubin for continuous lambs (n = 15), transported for 22 h; rested lambs (n = 15), transported for 22 h with a 6-h rest after 8 h of transport and a 24-h rest after an additional 8 h of transport; and control lambs (n = 16), who remained in the home pasture. The initial sample (0 h) was collected immediately before the beginning of transport, and the time of each subsequent sample was relative to the initial sample. a–cMeans at each time without a common superscript letter differ (P < 0.05).
Figure 2. Mean concentrations (with SE bars) of (A) blood urea nitrogen and (B) cortisol for continuous lambs (n = 15), transported for 22 h; rested lambs (n = 15), transported for 22 h with a 6-h rest after 8 h of transport and a 24-h rest after an additional 8 h of transport; and control lambs (n = 16), who remained in the home pasture. The initial sample (0 h) was collected immediately before the beginning of transport, and the time of each subsequent sample was relative to the initial sample. a–cMeans at each time without a common superscript letter differ (P < 0.05).

space for eating, drinking, and rest were provided. Although the space provided may be sufficient for lambs to lay down during transport and assume a resting posture (Buchenauer, 1997, and many of the lambs in this study were lying at the conclusion of periods of transport), the report (European Commission, 2002) provided no specific methods on how to deliver food and water for livestock receiving 6 or 24 h of on-trailer rest. Ensuring adequate access to food and water is crucial to achieve the intended welfare gains of those recommendations. Because slaughter horses are required to be unloaded for 6 h after 28 h of transport in the United States (USDA Veterinary Services, 2002) and the lack of technical information on how to supply food and water during on-trailer rest stops, the rested lambs in this study were unloaded into pens to assess effects of rest stops. Previous work with cattle (Kenny and Tarrant, 1987) and calves (Kent and Ewbank, 1983; Grigor et al., 2004) showed that transport can potentially be a greater stressor than loading and unloading. However, crate-raised calves were found to be the most stressed from the loading and unloading process (Trunkfield and Broom, 1990). Grandin (1997) hypothesized that animals that have been intensively handled, as the lambs in this study were, are more likely to respond negatively to the trip rather than the loading and unloading. Lambs in the current study were habituated to handling to minimize effects of repeated handling that was necessary to obtain data. However, it is important, when interpreting these data, to consider that commercial truck drivers in the United States report that sheep can be very difficult to load and unload under the conditions commonly found in large-scale lamb production. Hence, this study needs to be replicated using condi-
Figure 3. Mean pre- and posttransport absorbency reading with SE bars determined from an indirect ELISA indicating antigen-specific immunoglobulin G (IgG) and immunoglobulin M (IgM) response to vaccination with ovalbumin in the continuous lambs (n = 15), transported for 22 h; rested lambs, transported for 22 h with a 6-h rest after 8 h transport and a 24-h rest stop after an additional 8 h of transport; and control lambs (n = 16), who remained in home pasture. The pretransport sample was collected 2 d before the start of transport and immediately before vaccination. The posttransport sample was collected 8 d after the start of transport. a,b Means within a variable at each time without a common superscript differ (P < 0.05). The IgM increased (P < 0.001) from pre- to posttransport.

The reduced concentrations of glucose (Horton et al., 1996) and elevated concentrations of creatinine (Li et al., 2000), total bilirubin (Levin et al., 1993), and blood urea nitrogen (Kannan et al., 2000) that occurred in the continuous lambs are indicators of food deprivation. Concentrations of glucose responded similarly to what was reported by Knowles et al. (1995), who found a decrease in concentration during 24 h without food followed by a return to the baseline concentrations after access to food and water. The response of blood urea nitrogen was also similar to the response reported by Knowles et al. (1995) for sheep transported for 24 h followed by a 24-h recovery period. The increased concentrations of creatinine are similar to the response found in goats during 2.5 h of transport (Kannan et al., 2000). Unexplainable differences in creatinine between the continuous and rested lambs at 0 h also resulted in the same difference resulting at 8 h. The effect of transport on concentrations of total bilirubin was not previously reported. The access to food during the periodic rest stops of this study is one of the main benefits for the rested lambs.

The lack of meaningful differences in concentrations of creatine kinase suggests that the 22-h trip was not physically strenuous. Previous work (Wilson et al., 1990) has shown physical exertion was a more important factor in changing concentrations of creatine kinase than transport or starvation. The relatively low density (0.23 m² per lamb) at which the lambs were transported is a likely explanation for the lack of physical exertion during transport. The majority of the continuous lambs were observed to be lying down upon arrival after each period of transport. Calves provided sufficient space to lie down during transport were metabolically indistinguishable from nontransported calves (Todd et al., 2000). Also, transport over mainly flat, straight roadways may have also decreased the potential stressful effect of the transport. The importance of straight roadways and avoiding sudden stops and starts on welfare of sheep during transport has been previously established (Cockram et al., 2004). Based on the similarity between concentrations of creatine kinase in continuous, rested, and control lambs and the recumbent position the majority of lambs were in at the conclusion of a transport period, the maximum density recommended by the European Commission report (2002) appears sufficient for rest and avoidance of injuries.

Table 2. The BW (kg) before loading (0 h) and the percentage change relative to the initial BW at each subsequent sample

<table>
<thead>
<tr>
<th>Time</th>
<th>Control</th>
<th>Continuous</th>
<th>Rested</th>
<th>Pooled SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before loading, 0 h</td>
<td>17</td>
<td>17.8</td>
<td>18.5</td>
<td>0.8</td>
</tr>
<tr>
<td>End of transport for continuous lambs, 22 h</td>
<td>−9.0a</td>
<td>−14.6a</td>
<td>18.8b</td>
<td>1.8</td>
</tr>
<tr>
<td>24 h recovery for continuous lambs, 46 h</td>
<td>6.0ab</td>
<td>−1.4a</td>
<td>9.9b</td>
<td>1.4</td>
</tr>
<tr>
<td>End of transport for rested lambs, 52</td>
<td>7.7</td>
<td>ND4</td>
<td>−0.6</td>
<td>2.3</td>
</tr>
<tr>
<td>24 h recovery for rested lambs, 76 h</td>
<td>9.8</td>
<td>ND</td>
<td>7.5</td>
<td>1.4</td>
</tr>
<tr>
<td>8 d after the beginning of transport</td>
<td>32.5a</td>
<td>17.8b</td>
<td>28.5b</td>
<td>2.7</td>
</tr>
</tbody>
</table>

a,b Means within a row without a common superscript differ (P < 0.05).

1Timing of each sample was relative to the initial sample (before transport).

2Continuous lambs (n = 15) were transported for 22 h. Rested lambs (n = 15) were transported for 22 h with a 6-h rest after 8 h transport and a 24-h rest stop after an additional 8 h of transport. Control lambs (n = 16) remained in the home pasture throughout the study.

3Data for this and remaining samples are the percentage of change from the initial BW. Negative values represent a loss of BW.

4ND = Data not collected for continuous lambs at these times.
While conducting this experiment (with peak temperature of 39.6°C), the researchers became concerned that the continuous lambs were suffering from water deprivation. The continuous lambs appeared to be much more lethargic than the rested lambs at the 14-h sampling. As a result, water was offered from buckets after the 14-h sampling. The majority of the continuous lambs, however, were not interested in the water offered on the trailer, and the few that showed interest appeared to only investigate the novelty of the bucket rather than consume appreciable amounts of water. Results of electrolyte analysis did not indicate dehydration had occurred during the 14 h of transport before the offering of water nor was there an indication that the continuous lambs were dehydrated at the conclusion of transport 8 h later. Continuous and rested lambs moved immediately to feed bunks and ignored water until all grain provided during rest stops or after their final unloading was consumed. There was no latency period between unloading and approaching feed after unloading of the rested lambs or the single unloading of the continuous lambs. This was consistent with Parrott et al. (1998b) and Cockram et al. (1999), who found sheep would not utilize water unless they had consumed food.

The elevated concentrations of plasma cortisol in continuous lambs at 14 h and the continuous and rested lambs at 22 h compared with the control lambs most likely resulted from the stress of transport (Odore et al., 2004) and food deprivation (Cockram et al., 1999). The similarity of cortisol concentrations at the conclusion of transport (52 h) in the rested and control lambs may indicate habituation to transport (De Boer et al., 1990), rather than a depletion of stored glucocorticoids typical of situations of chronic stress (Selye, 1955). De Boer et al. (1990) found the duration and intensity of the physiological stress response was diminished in rats after application of the same stressor at regular intervals, which was similar to the transport stress the rested lambs were subjected to in this study.

Both transported treatments had lower IgG responses to challenge against ovalbumin than control lambs, which indicate the transport periods had resulted in some immunosuppression. This was compounded by the lack of significant treatment effects in IgM response. However, there was a trend of reduced IgM response in the treatment groups with the continuous lambs having the lowest value. Transport has previously been found to reduce aspects of immune function of lambs (Horton et al., 1996) and goats (Kannan et al., 2000) and may have resulted from the elevated cortisol concentrations.

Access to food and water during the intermittent rest periods was sufficient to prevent rested lambs from experiencing the same decrease in BW as the continuous lambs during transport. The loss in BW by the continuous lambs was still significant 8 d after transport. The BW losses during this study were most likely caused by changes in fill. The loss in BW that occurred in control lambs at 22 h likely resulted from social disruption caused by removing two-thirds of the lambs from the original flock to conduct the continuous and rested treatments. Previous research with ewes (Sevi et al., 2001) found that disruption of social groupings had short-term, detrimental effects on production.

Off-trailer rest stops with feed and water during long-distance transport at high ambient temperatures eliminated signs of food deprivation and maintained BW but did not alleviate a reduced IgG response to vaccination against ovalbumin that was apparent in transported lambs. Neither continuous nor intermittent transport resulted in dehydration of the lambs. The benefits of eliminating food deprivation in the rested lambs could be negated for animals being transported to slaughter plants by the need to fast animals before slaughter. The economic cost of extending a 22-h transport to the 52 h needed to provide the recommended amount of rest, and replications using other breeds of lambs, other species, and a variety of environmental conditions should be considered before these results are widely applied.

**LITERATURE CITED**


