

REST STOPS DURING ROAD TRANSPORT: IMPACTS ON PERFORMANCE AND ACUTE-PHASE PROTEIN RESPONSES OF FEEDER CATTLE

B. I. Cappellozza, R. F. Cooke, T. Guarnieri Filho, R. Almeida, J. M. Neto, D. McGuire, F. Cooke, and D. W. Bohnert
Oregon State University - Eastern Oregon Agricultural Research Center, Burns, OR

ABSTRACT: Angus × Hereford steers (n = 42) and heifers (n = 21) were ranked by gender and BW on d 0 of the experiment, and randomly assigned to 1 of 3 treatments: 1) no transport and full access to feed and water (**CON**); 2) continuous road transport for 1,290 km (**TRANS**), or 3) road transport for 1,290 km, with rest stops every 430 km (**STOP**; total of 2 rest stops). Treatments were applied from d 0 to 1 of the study. Cattle from **TRANS** and **STOP** treatments were transported in separate commercial livestock trailers, but through the exact same route. During each rest stop, **STOP** cattle were unloaded and offered alfalfa-grass hay and water for ad libitum consumption for 2 h. Upon arrival of **STOP** and **TRANS** on d 1, cattle were ranked by gender and BW within each treatment and assigned to 21 pens (7 pens/treatment; 2 steers and 1 heifer per pen). Full BW was recorded prior to (d -1 and 0) treatment application and at the end of experiment (d 28 and 29) for ADG calculation. Total DMI was evaluated daily from d 1 to 28. Blood samples were collected on d 0, 1, 4, 7, 10, 14, 21, and 28. Body weight shrink from d 0 to d 1 was reduced ($P < 0.01$) in **CON** vs. **TRANS**, **CON** vs. **STOP**, and **STOP** vs. **TRANS**. Mean ADG was greater ($P < 0.05$) in **CON** vs. **TRANS** and **STOP**, but similar ($P = 0.68$) between **TRANS** and **STOP**. Mean G:F was greater ($P = 0.05$) in **CON** vs. **STOP**, tended to be greater ($P = 0.08$) in **CON** vs. **TRANS**, and similar ($P = 0.85$) between **TRANS** and **STOP**. Plasma cortisol was greater ($P < 0.04$) in **TRANS** vs. **CON** and **STOP** on d 1, and greater ($P = 0.04$) in **TRANS** vs. **CON** on d 4. Serum NEFA was greater ($P < 0.01$) in **TRANS** vs. **CON** and **STOP** on d 1, and greater ($P \leq 0.05$) in **TRANS** vs. **CON** on d 4 and 7. Plasma haptoglobin was greater ($P < 0.04$) in **TRANS** vs. **CON** and **STOP** on d 1, and **STOP** vs. **CON** on d 1. In conclusion, inclusion of rest stops during a 1,290-km transport prevented the increase in circulating cortisol and alleviated the NEFA and haptoglobin response elicited by transport, but did not improve feedlot receiving performance of transported cattle.

Key Words: Acute-phase proteins, beef cattle, feedlot receiving, rest stops

Introduction

Cattle are exposed to several psychologic, physiologic, and physical stressors associated with management procedures currently practiced within beef production systems (Carroll and Forsberg, 2007). Transporting, for example, is one of the most stressful events experienced by feeder cattle (Swanson and Morrow-Tesch, 2001). Upon long transportation periods, cattle

experience inflammatory and acute-phase responses (Cooke et al., 2011) that may impair health and productivity during feedlot receiving (Araujo et al., 2010). Moreover, recent research from our group demonstrated that feed and water deprivation are major contributors to the acute-phase response and reduced feedlot receiving performance detected in transported cattle (Marques et al., 2012). Therefore, alternatives to prevent, or at least alleviate, prolonged periods of feed and water restriction during transport may modulate the acute-phase and performance responses during feedlot receiving.

A potential alternative would be adoption of rest stops during long transport for water and feed consumption, which are mandatory during 24-h or longer transports in Canada and European countries (Tarrant and Grandin, 2000; Council Regulation N° 1/2005, 2005). However, to our knowledge, no research studies have compared the acute-phase response between cattle transported for long distances and allowed, or not, to stop for water and feed intake. In addition, rest stops may further expose cattle to stressors, including handling for loading and unloading, known to stimulate the bovine acute-phase response (Carroll and Forsberg, 2007). Hence, the objective of this experiment was to evaluate the effects of rest stops during road transport on circulating concentrations of cortisol, NEFA, acute-phase proteins, and feedlot receiving performance of feeder cattle.

Materials and Methods

The experiment was conducted at the Oregon State University – Eastern Oregon Agricultural Research Center (Burns Station) from October to November 2012. All animals utilized were cared for in accordance with acceptable practices and experimental protocols reviewed and approved by the Oregon State University, Institutional Animal Care and Use Committee.

Animals and diets. Sixty-three Angus × Hereford steers (n = 42) and heifers (n = 21), which were weaned 45 d prior to the beginning of the experiment (d 0), were maintained in a single meadow foxtail (*Alopecurus pratensis* L.) pasture from d -15 to 0 for preconditioning. During this period, cattle were fed mixed alfalfa-grass hay ad libitum and 2.3 kg/animal daily (DM basis) of a concentrate containing (as-fed basis) 84% cracked corn, 14% soybean meal, and 2% mineral mix. On d 0, cattle were ranked by gender and initial BW (229 ± 2 kg; initial age = 211 ± 3 d) and assigned to 1 of 3 treatments: 1) no transport and full access to feed and water (**CON**); 2) continuous road transport for 1,290 km (**TRANS**), or 3) road transport for

1,290 km, with rest stops every 430 km (**STOP**; total of 2 rest stops). Cattle assigned to TRANS and STOP traveled from d 0 to d 1 in separate commercial livestock trailers, within a single 2.1 x 7.2 m compartment, but through the exact same route. However, STOP cattle were loaded and initiated transport 4 h before TRANS cohorts (0600 and 1000 h on d 0, respectively) so both treatment groups were unloaded at the same time on d 1 (1000 h). During each rest stop, STOP cattle had ad libitum access to mixed alfalfa-grass hay and water for 2 h before being re-loaded into the same commercial livestock trailer. Moreover, during the first rest stop (1300 to 1500 h), STOP cattle were individually allocated to drylot pens (8 x 20 m) for feed and water intake evaluation. During the second rest stop (2300 to 0100 h), STOP cattle were not individually penned due to the lack of visibility for proper animal handling, and were maintained in a single drylot pen (20 x 35 m). The CON treatment was included as a non-transport positive control for physiological and performance measurements, and remained in the same meadow foxtail pasture with ad libitum access to mixed alfalfa-grass hay and 2.3 kg/animal (DM basis) of the aforementioned concentrate while TRANS and STOP cattle were being transported.

Immediately upon arrival of STOP and TRANS groups on d 1, cattle were ranked by gender and BW within each treatment and assigned to 21 feedlot pens (7 pens/treatment; 2 steers and 1 heifer/pen; 8 x 20 m) for a 28-d feedlot receiving. During feedlot receiving, all pens were fed mixed alfalfa-grass hay ad libitum and 2.3 kg/animal daily (DM basis) of the aforementioned corn-based concentrate, which was offered separately from hay at 0800 h. Water was offered for ad libitum consumption from d -15 to 28, except to STOP and TRANS cattle when inside the livestock trailers.

All cattle were vaccinated against clostridial diseases (Clostrishield 7; Novartis Animal Health; Bucyrus, KS) and bovine virus diarrhea complex (Virashield 6 + Somnus; Novartis Animal Health) at approximately 30 d of age. At weaning (d -45), cattle were vaccinated against clostridial diseases and *Mannheimia haemolytica* (One Shot Ultra 7; Pfizer Animal Health; New York, NY), infectious bovine rhinotracheitis, bovine viral diarrhea complex, and pneumonia (Bovi-Shield Gold 5 and TSV-2; Pfizer Animal Health), and administered an anthelmintic (Dectomax; Pfizer Animal Health). No incidences of mortality or morbidity were observed during the entire experiment.

Sampling. Individual full BW was recorded and averaged over 2 consecutive days prior to treatment application (d -1 and 0) and at the end of experiment (d 28 and 29) for ADG calculation. Individual BW was also collected on d 1, immediately after treatment application, to evaluate BW shrink as percentage change from the average BW recorded on d -1 and 0. Concentrate, hay, and total DMI were evaluated daily from d 1 to 28 from each pen by collecting and weighingorts daily. Samples of the offered and non-consumed feed were collected daily from each pen and dried for 96 h at 50°C in forced-air ovens for DM calculation. Hay, concentrate, and total daily DMI of each pen were divided by the number of animals within each pen, and expressed as kg per animal/d. Total BW gain and

DMI of each pen from d 1 to 28 were used for feedlot receiving G:F calculation.

Blood analysis. Blood samples were collected on d 0 (prior to loading of TRANS and STOP cattle), 1 (immediately after unloading of TRANS and STOP cattle), and on d 4, 7, 10, 14, 21, and 28, via jugular venipuncture into commercial blood collection tubes with or without 158 USP units of freeze-dried sodium heparin for plasma and serum collection, respectively. Blood samples were collected prior to concentrate feeding, except for d 0 when STOP and TRANS cattle were transported immediately after blood collection. All blood samples were placed immediately on ice, centrifuged (2,500 x g for 30 min; 4°C) for plasma or serum harvest, and stored at -80°C on the same day of collection. Plasma concentrations of cortisol were determined in samples collected from d 0 to d 10 using a bovine-specific commercial ELISA kit (Endocrine Technologies Inc., Newark, CA). Plasma concentrations of ceruloplasmin and haptoglobin were determined in all samples according to colorimetric procedures previously described (Demetriou et al., 1974; Cooke and Arthington, 2012). Serum concentrations of NEFA were determined in samples collected from d 0 to d 10 using a colorimetric commercial kit (HR Series NEFA - 2; Wako Pure Chemical Industries Ltd. USA, Richmond, VA). The intra- and inter-assay CV were, respectively, 8.0 and 7.2% for cortisol, 3.7 and 5.8% for NEFA, 9.4 and 6.2% for ceruloplasmin, and 7.2 and 6.5% for haptoglobin.

Statistical analysis. Data were analyzed using the PROC MIXED procedure of SAS (SAS Inst., Inc., Cary, NC) and Satterthwaite approximation to determine the denominator df for the tests of fixed effects. The model statement used for BW shrink and ADG contained the effects of treatment, gender, and the treatment x gender interaction. Data were analyzed using animal(treatment x pen) as random variable. The model statement used for DMI and G:F contained the effects of treatment, as well as day and the treatment x day interaction for DMI only. Data were analyzed using pen(treatment) as the random variable. The model statement used for blood variables contained the effects of treatment, day, gender, all resultant interactions (treatment x gender, treatment x day, and treatment x gender x day), and values obtained on d 0 as covariate. Data were analyzed using animal(treatment x pen) as the random variable. The specified term for the repeated statements was day, pen(treatment) or animal(treatment x pen) as subject for DMI or blood variables, respectively, and the covariance structure utilized was based on the Akaike information criterion. Results are reported as least square means, as well as covariately adjusted least square means for blood variables, and were separated using PDIF. Significance was set at $P \leq 0.05$ and tendencies were determined if $P > 0.05$ and ≤ 0.10 . Results are reported according to main effects if no interactions were significant, or according to the highest-order interaction detected.

Results and Discussion

No interactions containing the effects of treatment and gender were detected ($P \geq 0.44$) for the variables

analyzed and reported herein; therefore, results are reported across steers and heifers. A treatment effect was detected ($P < 0.01$) for BW shrink from d 0 to 1. Shrink was greater ($P < 0.01$) for both TRANS and STOP compared with CON cattle, but also greater ($P < 0.01$) for TRANS compared with STOP cattle (Table 1). Previous research from our group reported equivalent BW shrink rates in feeder cattle exposed to the same transportation schedule as TRANS cattle were exposed to herein (Marques et al., 2012). Supporting our hypothesis, the rest stop schedule effectively allowed STOP cattle to consume water and feed, and consequently reduced the BW shrink resultant from the 1,290-km transport. During the first rest stop, all STOP cattle immediately consumed water whereas hay DMI was 1.49 ± 0.3 kg per animal. During the second rest stop, water consumption of STOP cattle was not monitored due to the lack of visibility, but average hay DMI was 1.64 kg per animal. However, the STOP treatment did not benefit feedlot receiving performance based on the treatment effects detected ($P = 0.05$) for ADG and G:F ($P = 0.10$; Table 1). Cattle assigned to CON had greater ADG compared with TRANS ($P = 0.05$) and STOP ($P = 0.02$) cohorts, whereas ADG was similar between ($P = 0.68$) TRANS and STOP cattle, corroborating that feedlot receiving ADG is reduced in transported cattle compared to non-transported cohorts (Marques et al., 2012). Still, treatment effects detected on ADG were not sufficient to impact ($P = 0.56$) cattle BW at the end of the experimental period (Table 1). No treatment effects were detected ($P \geq 0.18$) on hay, concentrate, and total DMI (Table 1), although other authors reported depressed feed intake in cattle upon road transport (Hutcheson and Cole, 1986) due to impaired ruminal function and altered endocrine or metabolic patterns (Cole, 2000). Nevertheless, CON had greater G:F compared with STOP ($P = 0.05$) and tended to have greater G:F compared with TRANS cattle ($P = 0.08$), whereas G:F was similar ($P = 0.85$) between TRANS and STOP cattle (Table 1). Hence, STOP cattle experienced a similar decrease in feedlot receiving performance compared with TRANS cohorts, indicating that 2-h rest stops failed to alleviate the performance losses caused by road transport.

Table 1. Feedlot receiving performance (28 d) of cattle assigned to continuous road transport for 1,290 km (TRANS), road transport for 1,290 km but with rest stops every 430 km (STOP), or no transport and full access to feed and water (CON)¹.

Item	CON	FM	TRANS	SEM	P =
BW, kg					
Initial	230	229	229	4	0.96
Final	268	261	262	5	0.56
Shrink, %	-1.25 ^a	5.82 ^b	10.17 ^c	0.47	< 0.01
ADG, kg/d	1.28 ^a	1.09 ^b	1.13 ^b	0.06	0.05
DMI, kg/d					
Hay	5.17	4.95	5.17	0.10	0.21
Concentrate	2.30	2.27	2.30	0.01	0.26
Total	7.48	7.21	7.47	0.11	0.18
G:F, g/kg	533 ^a	465 ^b	471 ^{ab}	24	0.10

¹Within rows, values with different superscripts differ ($P < 0.05$).

Plasma cortisol concentrations were greater ($P < 0.04$) in TRANS vs. CON and STOP on d 1, and greater ($P = 0.04$) in TRANS vs. CON on d 4 (Figure 1; treatment \times day interaction, $P = 0.09$), indicating that the STOP treatment prevented the transport-elicited increase in plasma cortisol during feedlot receiving (Cooke et al., 2011).

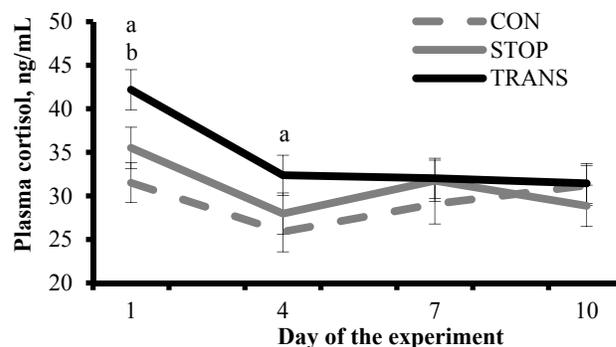


Figure 1. Plasma cortisol concentrations in cattle assigned to continuous road transport for 1,290 km (TRANS), road transport for 1,290 km but with 2-h rest stops every 430 km (STOP), or no transport and full access to feed and water (CON). A tendency for a treatment \times day interaction was detected ($P = 0.09$). Within days, letters indicate the following treatment differences; a = TRANS vs. CON ($P \leq 0.04$), b = TRANS vs. STOP ($P = 0.04$).

Serum NEFA concentrations were greater ($P < 0.01$) in TRANS vs. CON and STOP on d 1, and greater ($P \leq 0.05$) in TRANS vs. CON cattle on d 4 and 7 (Figure 2; treatment \times day interaction, $P < 0.01$). Serum NEFA concentrations directly reflect the amount of fat tissue mobilization caused by feed restriction during road transport (Earley and O'Riordan, 2006; Marques et al., 2012). Hence, the STOP treatment effectively reduced, but did not eliminate, fat tissue mobilization by allowing cattle to consume feed during the 2-h rest stops and preventing the transported-induced increase in serum cortisol.

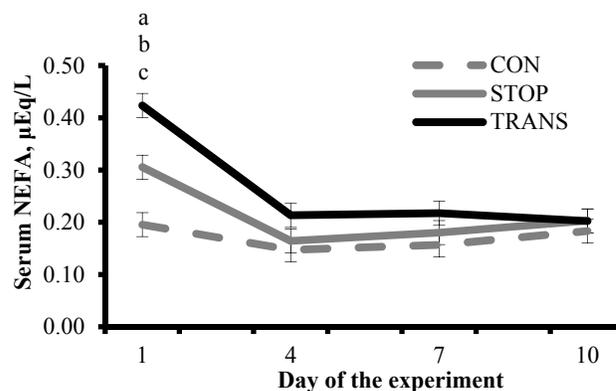


Figure 2. Serum NEFA concentrations in cattle assigned to continuous road transport for 1,290 km (TRANS), road transport for 1,290 km but with 2-h rest stops every 430 km (STOP), or no transport and full access to feed and water (CON). A treatment \times day interaction was detected ($P < 0.01$). Within days, letters indicate the following treatment differences; a = TRANS vs. CON ($P \leq 0.05$), b = TRANS vs. STOP ($P < 0.04$), c = STOP vs. CON ($P < 0.01$).

During feedlot receiving, mean plasma ceruloplasmin concentrations were similar ($P = 0.19$) between treatments (35.4, 37.8 and 36.2 mg/dL for CON, STOP, and TRANS, respectively; SEM = 0.95). Plasma haptoglobin concentrations were greater ($P < 0.04$) in TRANS vs. CON and STOP cattle on d 1, as well as greater ($P = 0.04$) in STOP vs. CON cattle on d 1 (Figure 3; treatment \times day interaction, $P = 0.08$). Previous research from our group also documented an increase in plasma haptoglobin concentrations in beef cattle upon a similar road transport (Araujo et al., 2010; Francisco et al., 2012) that impaired feedlot receiving ADG and G:F (Marques et al., 2012). Accordingly, circulating concentrations of haptoglobin in transported feeder cattle have been negatively associated with feedlot receiving performance (Araujo et al., 2010). However, the same treatment response was not detected for plasma ceruloplasmin concentrations, although plasma concentrations of ceruloplasmin and haptoglobin are typically correlated (Cooke et al., 2009).

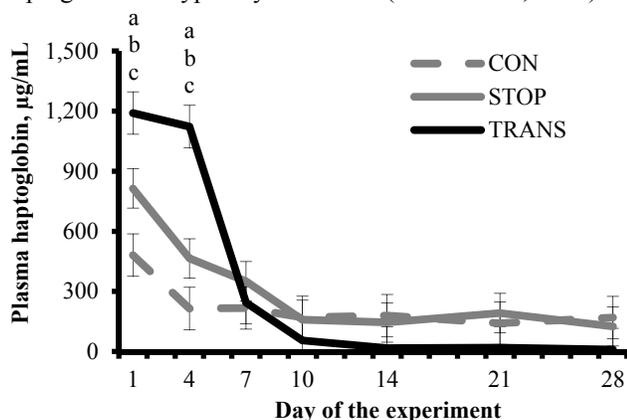


Figure 3. Plasma haptoglobin concentrations in cattle assigned to continuous road transport for 1,290 km (TRANS), road transport for 1,290 km but with 2-h rest stops every 430 km (STOP), or no transport and full access to feed and water (CON). A treatment \times day interaction was detected ($P < 0.01$). Within days, letters indicate the following treatment differences; a = TRANS vs. CON ($P \leq 0.05$), b = TRANS vs. STOP ($P < 0.01$), c = STOP vs. CON ($P < 0.01$).

Implications

In conclusion, providing 2-h rest stops during a 1,290-km transport prevented the increase in circulating cortisol and alleviated the NEFA and haptoglobin response elicited by transport, but did not improve feedlot receiving performance of transported cattle. Hence, rest stops appear to be an alternative to reduce neuroendocrine and acute-phase protein responses during transport and feedlot receiving.

Literature Cited

Araujo, D. B., R. F. Cooke, G. R. Hansen, C. R. Staples, and J. D. Arthington. 2010. Effects of rumen-protected polyunsaturated fatty acid supplementation on performance and physiological responses of growing cattle following transportation and feedlot entry. *J. Anim. Sci.* 87:4125-4132.

- Carroll, J. A., and N. E. Forsberg. 2007. Influence of stress and nutrition on cattle immunity. *Vet. Clin. Food. Anim.* 23:105-149.
- Cole, N. A. 2000. Changes in postprandial plasma and extracellular and ruminal fluid volumes in wethers fed or unfed for 72 hours. *J. Anim. Sci.* 2000. 78:216-223.
- Cooke, R. F. and Arthington, J. D. 2012. Concentrations of haptoglobin in bovine plasma determined by ELISA or a colorimetric method based on peroxidase activity. *J. Anim. Physiol. Anim. Nutr.* doi: 10.1111/j.1439-0396.2012.01298.
- Cooke, R. F., D. W. Bohnert, P. Moriel, B. W. Hess, and R. R. Mills. 2011. Effects of polyunsaturated fatty acid supplementation on ruminal in situ forage degradability, performance, and physiological responses of feeder cattle. *J. Anim. Sci.* 89:3677-3689.
- Cooke, R. F., J. D. Arthington, D. B. Araujo, and G. C. Lamb. 2009. Effects of acclimation to human interaction on performance, temperament, physiological responses, and pregnancy rates of Brahman-crossbred cows. *J. Anim. Sci.* 87:4125-4132.
- Council Regulation (EC) No 1/2005. 2005. On the protection of animals during transport and related operations and amending Directives 64/432/EEC and 93/119/EC and Regulation (EC) No 1255/97. *Off. J. L 3*, 0001-0044.
- Demetriou, J. A., P. A. Drewes, and J. B. Gin. 1974. Ceruloplasmin. Pages 857-846 in *Clinical Chemistry: Principles and Techniques*. 2nd ed. Harper and Row, Hagerstown, MD.
- Earley, B., and E. G. O'Riordan. 2006. Effects on transporting bulls at different space allowances on physiological, haematological and immunological responses to a 12-h journey by road. *Ir. J. Agric. Food Res.* 45:39-50.
- Francisco, C. L., R. F. Cooke, R. S. Marques, R. R. Mills, and D. W. Bohnert. 2012. Effects of temperament and acclimation to handling on feedlot performance of *Bos taurus* feeder cattle originated from a rangeland-based cow-calf system. *J. Anim. Sci.* 90:5067-5077.
- Hutcheson, D. P., and N. A. Cole. 1986. Management of transit-stress syndrome in cattle: nutritional and environmental effects. *J. Anim. Sci.* 62:555-560.
- Marques, R. S., R. F. Cooke, C. L. Francisco, and D. W. Bohnert. 2012. Effects of 24-h transport or 24-h feed and water deprivation on physiologic and performance responses of feeder cattle. *J. Anim. Sci.* 90:5040-5046.
- Swanson, J. C., and J. Morrow-Tesch. 2001. Cattle transport: Historical, research, and future perspectives. *J. Anim. Sci.* 79:E102-E109.
- Tarrant, P. V., and T. Grandin. 2000. Cattle transport. In: T. Grandin (ed.) *Livestock Handling and Transport*. 2nd ed. CAB International, Oxford, U.K.