EXHIBIT 23

AR0000257

Responses of Horses to Trailer Design, Duration, and Floor Area During Commercial Transportation to Slaughter[1]

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ABSTRACT: Nine trailer loads of horses (n = 306) transported to slaughter facilities with distances ranging 596 to 2,496 km were studied to characterize the type of horses used in commercial markets and the physiological responses and number of injuries due to transportation under summer environmental conditions. Slaughter horse candidates were middle-aged (11.4 ± .4 yr), possessed moderately fleshy body condition, weighed 432 ± 3.3 kg, and were of Quarter Horse or Thoroughbred breeding. The mean weight loss during commercial transport was 4%. The percentage of injured horses was greater ($P < .05$) for two-tiered "potbelly" (29.2%) compared with straight-deck (8.0%) trailers; however, the stress indicators of cortisol and neutrophil:lymphocyte ratio and rectal temperature showed greater ($P < .05$) responses following transport in straight-deck trailers. As trip duration increased from 5 h 45 min to 30 h, muscle fatigue (lactate concentration) and dehydration (hematocrit and total protein concentration) were the major physiological considerations, especially in durations over 27 h. The percentage of horses injured was less ($P < .05$) in trailers with 1.14 to 1.31 m^2 of floor area per horse than in trailers with 1.40 to 1.54 m^2 of floor area per horse. However, most physiological responses (white blood cell count, total protein concentration, and neutrophil:lymphocyte ratio) to transportation were less ($P < .05$) in horses provided with the greater floor area.

Key Words: Horses, Transport, Handling, Stress, Animal Welfare

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Introduction

Approximately 87,200 horses (USDA, 1998) were processed for human consumption at U.S. slaughter facilities in 1997 and the resultant meat products were marketed for export. Horses tend to travel longer distances to slaughter than other livestock, because there is a limited number of equine slaughterhouses. The 1996 Farm Bill (Sec. 901–905) authorized the Secretary of Agriculture to issue guidelines for the regulation of commercial transportation of equines for slaughter within the United States. However, there are limited scientific data available to support appropriate transportation regulations for slaughter horses.

Previous equine transport studies have examined behavioral and autonomic responses of orientation during travel (Smith et al., 1994; Waran et al., 1996), neuroendocrine responses (Baucus et al., 1990; Ferlazzo et al., 1993), or immunological changes (Anderson et al., 1985; Crisman et al., 1992) primarily in race or show horses. One recent study examined water deprivation during long-term transportation in university-owned, tame horses traveling in an open-type trailer (Friend et al., 1998). Horses may respond differently to transportation stress depending on their previous experience, health status, social interactions, environmental conditions, breed, age, and gender.

Although it is difficult to obtain knowledge about the previous handling and management of individual horses procured for slaughter, an applied study under commercial conditions will provide information to characterize the type of horses used in the system. Additionally, data collected on selected physiological responses and number of injuries incurred during transport for comparison of trailer designs, floor space per horse, and trip duration will assist in drafting science-based guidelines. Thus, our objectives for the study were to characterize slaughter horse candidates and to examine physiological responses and number of injuries due to commercial transport under summer environmental conditions.

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Materials and Methods

Horses and Basic Study Design

All phases of the study were approved by the University of California Animal Care and Use Committee. Horses (n = 306) were identified as slaughter horses by cooperating brokers and transport drivers. A total of nine tractor-trailer loads of horses were assembled in the states of Pennsylvania, California, Kentucky, and Texas. All loads were transported to a USDA inspected processing plant in Fort Worth, Texas. Horses were assembled in groups prior to loading for a minimum of 1 h, but the interval varied up to several days depending on the procurement procedure. The commercial trailers used were of two basic designs, either a single, straight-deck (**SD**, n = 5) or two-tiered "pot-belly" livestock trailer (**PB**, n = 4). The length and width of the SD trailers was approximately 15.2 m and 2.6 m, respectively, with ceiling heights between 2.1 and 2.7 m. Three or four compartments with swinging gates were available to segregate groups of horses. Rubber padding lined the sides of the trailers from the floor to an approximate height of 1.2 m. The length and width of PB trailers was 14.4 and 2.5 m, respectively. The PB trailers were divided into four compartments with the two middle sections stacked to form two tiers, each with a ceiling height of 1.7 to 1.8 m. The front and back sections were placed over the two front and two back wheel axles and the ceiling height was 2.1 m. There was an attempt to match the two trailer designs over routes of similar duration. The number of horses per load, the available floor area, distance traveled, and time between loading and unloading were recorded. No feed or water was available during transit, but they were available before loading.

Temperature and relative humidity inside the trailer during transport were monitored continually with microcomputer data logger units (Model WTA32 and SRHA32, respectively; Onset Instruments, Pocasset, ME) placed near the center of the trailer. The concentrations of ammonia and carbon monoxide gas were quantified using dosimeter gas detection tubes (Gastec, Yokohama, Japan) placed near the center of the trailer during transit.

Prior to loading into trailers, numbered tags were adhered to the hip of each horse as identification. The breed, gender, height at withers, and age as estimated by examination of teeth were documented. Body condition scores (**BCS**) were assigned using a scale (1 to 9) with maximum score of 9 (obese) (Henneke et al., 1983). Prior to loading, the location of any abrasions or lacerations were documented on a diagram profiling an outline of a horse. Upon arrival, any additional injuries were documented; thus, injuries sustained during loading, transportation, and unloading were deduced.

Collection of Physiological Data

Similar data collection procedures were used to collect data within 4 h (pre-samples) before transport and after arrival (post-samples) at the slaughter facility. Horses were either gently restrained with a halter or slowly processed through a chute to collect samples prior to transport. All horses were processed through a chute at the slaughter facility. Rectal temperature and body weight (if scales were available) were recorded. Blood samples (20 mL) were collected in evacuated glass tubes via needle puncture of the jugular vein. Blood samples to be analyzed for concentrations of cortisol, lactate, and total protein were immediately placed on ice and allowed to clot. Serum was obtained and frozen at −70°C. Samples intended for complete blood cell counts were collected in tubes containing EDTA.

Laboratory Analyses. Cortisol was determined in duplication with the microplate enzyme immunoassay technique (Munro and Stabenfedlt, 1984). Intra- and interassay CV were less than 10 and 15%, respectively. Lactate concentrations were determined using an autoanalyzer (YSI 2300 STAT Plus, Yellow Springs, OH). White blood cell counts (**WBC**) and hematocrit (**hct**) were determined using an automated cell counter (System 9000, Serono-Baker Diagnostics, Allentown, PA). Whole blood was used to determine WBC differential counts using standard laboratory staining techniques (Heckner et al., 1988); the neutrophil:lymphocyte ratio (**N:L**) was calculated. Total serum protein concentration was determined with commercially available enzymatic colorimetric assays (Procedure 541, Sigma Diagnostics, St. Louis, MO).

Statistical Analyses

Data were analyzed with ANOVA and analysis of covariance (ANOCOVA) methods using the GLM procedure (SAS, 1989). The factors considered in these analyses were a horse's age, gender, and breed, and the load in which it was transported. The analyses were run alternately as main effects models and as factorial models, to examine possible interactions among these factors. Whenever load, or an interaction involving load, was significant, contrast statistics were constructed to compare loads with respect to the trailer type (SD or PB), the duration of the trips (short, < 6 h; medium, 16 to 23 h; or long, 27 to 30 h), and the loading density represented by the available floor area per horse (high, 1.40 to 1.54 m^2/horse [**HFA**], or low, 1.14 to 1.31 m^2/horse [**LFA**]. Data were analyzed in raw form or following log transformation, as appropriate to satisfy the model assumptions of normality and constant residual variance. Significance is claimed whenever $P < .05$.

Results

The specific characteristics of each of the loads are given in Table 1. Distances from the point of loading to the slaughter plant ranged from 596 to 2,496 km, and duration of the trips ranged from 5 h 45 min to 30 h. The floor space per horse ranged from 1.14 to 1.54 m^2/horse. All trips to the slaughter plant occurred dur-

Table 1. Load characteristics including trailer design, length of travel, duration of trip, number of horses per load, floor area per horse, minimum and maximum temperature, minimum and maximum relative humidity inside the trailer, and time of the initiation of blood collection for the pre- and post-transport samples

Load	Trailer type[a]	km	Duration h:min	No. of horses per load	Floor area, m^2/horse	Temperature, °C		Humidity, %		Sampling time[b]	
						Min.	Max.	Min.	Max.	Pre	Post
1	SD	596	5:45	38	1.14	23	34	40	74	0500	1200
2	PB	596	5:45	39	1.54	22	31	32	85	1800	0300
3	SD	1,433	16:15	29	1.40	19	29	38	83	1000	0600
4	SD	1,433	17:30	31	1.31	20	25	NR[d]	NR	1300	0900
5	PB	2,496	28:00	44	1.23	3	31	14	71	1900	0100
6	PB	2,496	27:00	44	1.31	−2	33	8	88	1200	1800
7[c]	SD	2,335	30:00	15	1.24	13	39	21	100	0100	1000
8	SD	2,225	23:00	25	1.44	21	41	34	83	1800	2200
9	PB	2,415	30:00	41	1.43	15	38	31	100	1800	0300

[a]SD = straight-deck trailer; PB = two-tiered "pot-belly" trailer.
[b]Central Standard Time.
[c]Study horses constituted only a portion of the entire load.
[d]Data were not recorded due to equipment failure.

ing the months of July, August, and September. The maximum environmental temperature range for all loads was 25 to 41°C, and minimum temperatures ranged between −2 and 22°C. The range of minimum and maximum relative humidity inside all trailers was 8 to 40% and 71 to 100%, respectively. Eight of the nine loads were successful in recording readings on the gas diffusion sticks. The ammonia and carbon monoxide gas concentrations inside the trailer were 3.1 ± 2.0 and 2.4 ± 1.5 ppm, respectively. The ammonia gas concentration (5.2 ± 1.5 ppm) recorded in the three trailers bedded with either sawdust or straw on the floor was higher ($P < .05$) than that in the five trailers without bedding (1.8 ± .9 ppm).

Characteristics of Slaughter Horses

The mean age recorded (n = 285) was 11.4 ± .4 yr, with a range of 1 to 30 yr (Table 2). The percentage of horses with ages from 1 to 5, 6 to 15, 16 to 20, and 20 to 30 yr was 28, 39, 23, and 10%, respectively. Only 8% of the horses were yearlings or 2-yr of age. The breed of horse was recorded for 279 horses; 48% were classified as Quarter Horses, 18% as Thoroughbreds, 14% as having Morgan lineage, which included the Saddlebred and Standardbred breeds, 9% as Arabians, 6% as Appaloosas, and the remaining 5% as pony or draft breeds, mustangs, or mules. Mares constituted 50% of the slaughter horses, and geldings 47%. Fewer than 3% of the horses (n = 9) were stallions.

Table 2. Characteristics of slaughter horse candidates

Characteristic	n	Mean	SE	Range
Age, yr	285	11.4	.4	1–30
Weight, kg	301	432	3.3	284–807
BCS[a]	295	5.6	.08	2.0–9.0
Height, m	286	1.50	.01	1.12–1.68

[a]Body condition score (Henneke et al., 1983).

The BCS ranged from 2 to 9 with a mean score of 5.6 ± .08 for all horses. The mean BCS of the horses in the nine loads ranged from 5.3 to 6.4; horses with higher mean BCS originated from an equine feedlot. No difference ($P > .05$) in BCS was found between the trailer designs, trip duration, or floor area. Body weight (n = 301) documented at the slaughter facility following transport was 432 ± 3.3 kg, with a range of 284 to 806 kg. Height at the horse's withers ranged from 1.11 to 1.68 m, with a mean of 1.50 ± .01 m.

Physiological Comparisons

Age effects were not shown for any of the physiological variables evaluated, so this effect was suppressed in additional analyses. Breed effects were shown for body weight; breed and gender effects were shown for the hct values as described below in detail. Tables 3, 4, and 5 present pre- and post-transport data of the physiological indices for the comparisons of trailer design, trip duration, and floor area per horse, respectively.

Body Weight. Scales were only available at six (Loads 1, 2, 5, 6, 7, and 8) of the nine loading sites to obtain preshipment body weights. All horses were weighed upon arrival at the slaughter facility. The mean body weight for horses (n = 205) weighed prior to loading and at arrival was 441 ± 4 and 424 ± 4 kg, respectively, showing a 4% loss in body weight. The largest weight loss of 8% was recorded for Load 7, which was transported the longest (30 h). Breed effects ($P = .0003$) were shown for body weight prior to loading; Arabian horses (395 ± 10 kg) had lower body weights than Thoroughbreds (462 ± 8 kg), Quarter Horses (448 ± 5 kg), and Standardbreds (453 ± 7 kg). No difference ($P < .05$) in weight loss was demonstrated between SD and PB trailers, or LFA and HFA. Weight loss during short duration trips (12 ± 6 kg) was less ($P < .05$) than that during medium (24 ± 10 kg) or long (20 ± 6 kg) duration trips.

Table 3. Comparison of pre- and post-transit physiological data collected from horses transported on straight-deck or two-tiered "pot-belly" trailers; response values are the difference between the pre- and post-transit values

	Straight-deck trailer				Pot-belly trailer			
Variable	Pre	Post	Response	SE	Pre	Post	Response	SE
Temperature, °C	37.4	38.5	1.1[a]	.1	37.8	38.1	.3[b]	.1
WBC, $\times 10^3/\mu L$	11.2	13.4	2.1[a]	.3	11.5	12.4	.9[b]	.3
Neutrophil:lymphocyte ratio	2.9	4.7	1.8[a]	.3	3.2	4.2	.9[b]	.3
Cortisol, ng/mL	48	67	18[a]	2	44	50	6[b]	2
Hematocrit, %	43	45	2[a]	.6	43	47	4[a]	.5
Total protein, g/dL	8.5	8.9	.4[a]	.1	8.4	8.8	.4[a]	.1
Lactate, mg/dL	37	53	16[a]	2	33	49	16[a]	1
Weight, kg	447	425	22[a]	6	438	422	16[a]	5

[a,b]Means within a row with different superscripts differ ($P < .05$).

Rectal Temperature. Rectal temperature of 282 of the 306 horses shipped was measured prior to loading and upon arrival. Prior to shipping, rectal temperature was 37.7 ± .1°C, it increased to 38.2 ± .1°C following transport, but it remained in the normal range of 37.5 to 38.5°C for resting horses (Fraser, 1986). This increase was greater ($P < .05$) in the SD (.5 ± .2°C) than in the PB (.25 ± .05°C) group. Rectal temperature of horses shipped an intermediate duration (1.1 ± .1°C) increased ($P < .05$) more than rectal temperature of horses shipped for a short (.5 ± .1°C) or long (.3 ± .1°C) duration. There was no difference ($P > .05$) in rectal temperature response due to floor area per horse.

White Blood Cell Counts. The WBC counts increased slightly (11.3 ± .2 to 14.3 ± .3 × $10^3/\mu L$) with transportation, but these remained within the reference range (5.5 to 14.3 × $10^3/\mu L$) for healthy horses (Jain, 1993). The WBC counts of horses traveling in SD trailers were greater ($P < .05$) than those of horses shipped in PB trailers (2.1 ± .3 and .9 ± .3 × $10^3/\mu L$, respectively). Horses traveling longer than 27 h had greater ($P < .05$) elevations in WBC (1.8 ± .3 × $10^3/\mu L$) than those shipped for short (.7 ± .4 × $10^3/\mu L$) durations. White blood cell count elevations were greater ($P < .05$) in horses in trailers with LFA (2.0 ± .3 × $10^3/\mu L$) than in those in trailers with HFA (.6 ± .3 × $10^3/\mu L$).

Neutrophil:Lymphocyte Ratio. Transportation increased the mean N:L (n = 296) from 3.1 ± .1 to 4.4 ± .2, which was elevated over the normal range established for healthy horses of .8 to 2.8 (Morris and Large, 1990). Following transportation, horses traveling in SD (1.8 ± .3) showed ($P < .05$) a greater increase in N:L than horses shipped in PB (.9 ± .3). Horses shipped for a medium duration had a smaller increase ($P < .05$) in N:L (.6 ± .3) than horses shipped for short (1.6 ± .4) or long (1.6 ± .3) durations. Following transport, horses in trailers with LFA showed greater increases ($P < .05$) in N:L values than horses in trailers with HFA (1.9 ± .2 and .6 ± .3, respectively).

Cortisol. Using venous blood samples collected from indwelling jugular catheters, cortisol concentration in resting horses has been documented at 45 ± 1 ng/mL, with a circadian rhythm exhibiting peak concentrations (65 ± 2 ng/mL) in the morning hours (0600 to 1200) and low concentrations (25 ± 2 ng/mL) in the evening (1300 to 1600) (Stull and Rodiek, 1988). Transportation elevated cortisol concentrations in all horses from 46 ± 1 to 58 ± 2 ng/mL. Horses traveling in SD trailers showed greater elevation ($P < .05$) in cortisol concentration than horses in PB trailers (18 ± 2 and 6 ± 2 ng/mL, respectively). No difference ($P > .05$) in cortisol response was

Table 4. Comparison of pre- and post-transit physiological data collected from horses in trailers with trips of short, medium, or long duration; response values are the difference between the pre- and post-transit values

	Short, < 6 h				Medium, 16–23 h				Long, 27–30 h			
Variable	Pre	Post	Response	SE	Pre	Post	Response	SE	Pre	Post	Response	SE
Temperature, °C	38.0	38.6	.6[a]	.1	37.3	38.4	1.1[b]	.1	37.7	38.1	.3[a]	.1
White blood cells, $\times 10^3/\mu L$	12.7	13.5	.8[a]	.4	10.5	11.9	1.5[ab]	.4	11.1	13.0	1.8[b]	.3
Neutrophil:lymphocyte ratio	2.4	4.0	1.6[a]	.4	3.3	3.8	.6[b]	.3	3.3	4.9	1.6[a]	.3
Cortisol, ng/mL	51	65	14[a]	3	45	62	17[a]	3	44	51	7[a]	2
Hematocrit, %	44	45	3[a]	.5	42	45	4[b]	.8	43	47	5[b]	.6
Total protein, g/dL	8.4	8.1	–.3[a]	.1	8.9	9.4	.5[b]	.1	8.2	8.9	.7[c]	.1
Lactate, mg/dL	42	49	6[a]	2	36	54	18[b]	2	30	49	18[b]	1
Weight, kg	434	422	12[a]	6	455	431	24[b]	10	443	423	20[b]	6

[a,b,c]Means within a row with different superscripts differ ($P < .05$).

Table 5. Comparison of pre- and post-transit physiological data collected from horses in trailers with low (1.14 to 1.31 m^2/horse) and high floor area (1.40 to 1.54 m^2/horse); response values are the difference between the pre- and post-transit values

Variable	Low floor area				High floor area			
	Pre	Post	Response	SE	Pre	Post	Response	SE
Temperature, °C	37.8	38.4	.6^a	.1	37.5	38.1	.6^a	.1
White blood cells, × $10^3/\mu L$	11.4	13.6	2.1^a	.3	11.2	11.8	.6^b	.3
Neutrophil:lymphocyte ratio	2.8	4.7	1.9^a	.2	3.4	3.9	.6^b	.3
Cortisol, ng/mL	50	62	12^a	2	40	51	11^a	2
Hematocrit, %	44	46	2^a	.5	42	46	4^b	.6
Total protein, g/dL	8.2	8.7	.6^a	.1	8.8	9.1	.2^b	.1
Lactate, mg/dL	34	49	15^a	1	36	52	16^a	2
Weight, kg	441	423	18^a	5	440	424	16^a	8

^{a,b}Means within a row with different superscripts differ ($P < .05$).

shown for duration of trip or the floor area allowed per horse.

Hematocrit. Hematocrit values in all horses prior to loading and upon arrival at the slaughter facility were 43.1 ± .4 and 46.2 ± .4%, respectively, which were within the normal range of 32 to 53% (Jain, 1993). Breed effects ($P = .0003$) were shown for the pre-transport hct values; Appaloosa (38 ± 2%) exhibited lower values than the Thoroughbred (46 ± 1%), Quarter Horse (43 ± 1%), Standardbred (41 ± 1%), or Arabian breeds (46 ± 1%). A gender effect ($P = .0001$) was shown in hct values in samples collected prior to loading. The hct values in mares (45 ± 1%) and stallions (45 ± 2%) were greater than in geldings (41 ± 1%), but all three gender categories were within the normal reference range. No difference ($P > .05$) was shown between SD and PB trailers. The increase in hct values of horses experiencing the trips of shorter duration (3 ± .5%) were smaller ($P < .05$) than those in horses that were shipped for medium (3.9 ± .8%) or long (4.5 ± .6%) durations. The increase during transportation in hct of 2.5 ± .5% in LFA horses was less than ($P < .05$) that in HFA horses (4.2 ± .6%).

Total Protein. The total protein concentration (8.4 ± .1 g/dL) of all horses prior to transport was slightly elevated over reference range values of 5.2 to 7.9 g/dL (Kaneko et al., 1997). Following transportation, total protein concentration increased to 8.9 ± .1 g/dL. No difference ($P > .05$) was found between horses transported in different types of trailers. The difference between pre- and post-transportation total protein levels was significant ($P < .05$) for all three durations; horses transported for long or medium durations showed an increase in protein values (.7 ± .1 and .5 ± .1 g/dL, respectively), and those transported short durations showed a slight decrease (−.3 ± .1 g/dL) during transit. Floor area also affected total protein change during transportation, with a greater increase in LFA (.6 ± .1 g/dL) horses than in HFA ($P < .05$) horses (.2 ± .1 g/dL).

Lactate. The mean pre-transit concentration of lactate (34 ± 1 mg/dL) for all horses was elevated over established resting values of approximately 5 to 10 mg/dL (Stull and Rodiek, 1995). Transportation further elevated the concentration to 50.3 ± 1.0 mg/dL. There were no differences ($P > .05$) when trailer design or floor area per horse were compared. Trips of short duration increased lactate concentration (6 ± 2 mg/dL) less than ($P < .05$) trips of medium (18 ± 2 mg/dL) or long (18 ± 1 mg/dL) duration.

Injury Data

A total of 60 horses (19.6%, n = 306) sustained 81 injuries during transport (Table 6). The injuries consisted of either single or multiple abrasions and(or) lacerations. The percentage of horses traveling in PB (n = 168) that sustained injuries was 29.2%, as compared ($P < .05$) to 8.0% of the horses in SD (n = 138). The face/head was the most likely area for injury; 47 (58%) of the injuries reported occurred in this area. The other areas of the body with injuries were the legs, hips, and neck/shoulder/back, with 18 (22%), 12 (15%), and 4 (5%) injuries, respectively. Of the 47 face/head injuries, 38 occurred in 49 injured horses (78%) traveling in PB, and 9 occurred in 11 injured horses (77%) in SD. Of the 168 horses transported in PB trailers, the percentage of horses injured in the front, middle two tiers, or back compartments was 28.6, 31.0, and 16.7%, respectively. The percentage of horses injured in the HFA group (29%) was greater ($P < .05$) than in the LFA (12%) group. Additionally, 33% of the horses were injured in trips of long duration, and this was greater than ($P < .05$) the 9 and 8% of horses injured in trips of medium or short duration, respectively.

No horse died while in transit to the slaughter plant. However, one mare (Load 7) was removed from the load approximately 12 h after embarking. The 20-yr-old Paint mare died 2 d later. Prior to transport, she had a normal rectal temperature (37.9°C) and WBC count (8.8 × $10^3/\mu L$) and a BCS of 4, but an observable facial paralysis was noted prior to loading.

Discussion

Cold and heat stress can affect younger and sick animals much more severely than mature, healthy horses.

Table 6. The number of horses injured per load and the location of the injury

	Load								
Body area	1	2	3	4	5	6	7	8	9
Legs	—	1	—	1	2	—	1	1	12
Face/head	1	4	2	3	11	2	1	2	21
Hips/loin	—	—	—	5	—	—	—	—	7
Neck/shoulder/back	—	—	—	3	—	—	—	—	1
No. of horses injured[a]	1	5	2	3	13	2	2	3	29

[a]Injuries may have occurred in more than one area per horse.

The thermoneutral zone of horses is estimated to be between −1 and 24°C (Young and Coote, 1973). This optimal thermal environment promotes maximum performance and the least amount of stress in the animal (Curtis, 1983). As air temperature falls below −1°C, known as the lower critical temperature (**LCT**), horses must divert food energy from production and growth to produce additional metabolic heat and maintain body temperature. Several of the loads experienced temperatures near the LCT as the vehicles passed through desert conditions at night, but the duration was short and probably did not affect the physiology of the horses. The upper critical temperature (**UCT**) is approximately 24 to 32°C and is reached when horses cannot dissipate enough metabolic heat to the environment to maintain homeothermy (Curtis, 1983). This heat is then dissipated through sweating and respiration mechanisms. Relative humidity above 50% further exacerbates the ability of horses to dissipate heat. All loads exceeded the UCT, and humidity was recorded at values greater than 50%. Thus, horses experienced heat stress conditions while in transit, as would be expected under summer conditions.

Long-term exposure to toxic gas can predispose a horse to respiratory and eye irritations. No permissible levels of these gases have been established for horses. The permissible limit for an 8-h exposure for humans to ammonia and carbon monoxide is 25 and 50 ppm, respectively (ACGIH, 1989). The ammonia gas ranged from .7 to 6.7 ppm. Interestingly, the use of absorbent floor bedding with either sawdust or straw seems to increase the concentration of ammonia gas. This increase in ammonia gas concentration may partially be due to the accumulation of excreta resulting from the longer duration of the trip. None of the loads recorded gas levels for carbon monoxide near the permissible level (50 ppm) for humans, indicating that the exhaust fumes of the truck did not present a health hazard for the horses on board.

These slaughter horses reflected the general population of horses in the United States. Life expectancy in domestic horses is 20 yr or more, and maturity is considered to be attained between 3 and 4 yr. Over 90% of the study horses were mature and fewer than 10% were over 20 yr of age. The average horse was 11.4 yr old. Body condition scoring systems are used to evaluate the stored body fat in horses by visual appraisal and palpable fat cover in six areas of the body (Henneke et al., 1983). A BCS of 1 indicates extreme emaciation and 9 extreme fatness. The mean BCS of 5.6 ± .08 in this study indicated that the horses were neither emaciated nor extremely fat, but were considered moderately fleshy.

Two-thirds of the horses were either Quarter Horse or Thoroughbred breeds, and the mean weight of the horses was 432 ± 3.3 kg. The prevalence of these two breeds may represent the relative size of the breed registries, because the Quarter Horse and Thoroughbred breeds account for approximately 50% of the horses in the United States (NAHMS, 1998), or may be due to the buyers' selection of horses with similar conformation and weight to supply retail meat products of similar portions. No preference was shown for either mares (50%) or geldings (46%), and the study population contained fewer than 3% stallions. Stallions were managed during transportation by being tied by a halter and lead rope, placed unrestrained in compartments with only geldings, or integrated with a group of mares prior to loading and hauled with the same herdmates.

Physiological Responses to and Injuries from Transport

Rectal temperature and WBC were assessed as general indicators of morbidity and the ability of the horses to respond to heat stress during transit. The body weight, hct, and total protein indices were examined as dehydration measurements, becasue water (and feed) were not available during transit and environmental conditions were considered hot and humid. The stress variables of cortisol and N:L ratio have been previously documented as indicator of stress in other agricultural species (Friend et al., 1985; McFarlane et al., 1988; Stull and McDonough, 1994); N:L may be a more reliable indicator of stress than cortisol concentrations (Gross and Siegel, 1983). This may be partially attributed to the short half-life of cortisol of 1.5 h in resting horses that decreases during exercise to approximately 1 h (Lassourd et al., 1996). Additionally, cortisol concentrations in resting horses exhibit a daily circadian rhythm (Stull and Rodiek, 1988). This factor was not considered in the analyses because the distance of travel and time of departure and arrival was determined by the commercial transporters. Lactate produced during anaerobic metabolism was evaluated as an indicator of muscle fatigue.

Rectal temperature, WBC, and hct mean values were within reference range for a normal horse both prior to and following transport. The stress indicators of cortisol and N:L were slightly elevated above the reference range prior to transport and may have been due to the novel environment at the point of assembly, herd interactions, or other influences. The response to transportation further elevated stress variables, as expected. Total protein concentration prior to transportation showed a slight elevation over reference values, indicating mild dehydration, whereas hct was within normal range prior to and following transport. Elevated lactate concentration above established resting levels prior to transport may have been due to ambulatory activity at the point of assembly. Lactate concentration increased slightly following transport. However, compared to lactate concentrations greater than 200 mg/dL in horses following racing (Snow et al., 1983), this increase following transport is indicative of minimal anaerobic muscular fatigue.

Trailer Design. Two major types of trailers with capacity of over 15 horses are currently used to transport horses to slaughter. The PB trailers in this study carried up to 44 mature horses, and the SD hauled 38 horses, showing the economic advantage of increased capacity with the PB trailer. No difference was shown between PB and SD trailers for weight loss or increases in hct, total protein, or lactate responses to transportation. Thus, neither trailer design is advantageous in minimizing dehydration. The elevation in lactate was minimal, so muscle fatigue by anaerobic metabolism was not a limiting factor. Rectal temperature, WBC, cortisol, and N:L showed greater responses in horses shipped via SD trailers than in those shipped in PB trailers. Rectal temperature may reflect environmental heat stress, which ventilation assists in alleviating. The rubber padding lining the interior walls of the SD trailers limited the ventilation capacity and may have added to the heat stress. The WBC, cortisol, and N:L data indicate that the horses traveling in PB, as compared to SD, trailers experienced less stress and fewer immunological changes, supporting the use of PB trailers. However, there is the possibility, although it was not measured, that a specific design feature other than decking may have contributed to the difference.

Sustained injures approximately 3.5 times greater in PB (29.2%) than in SD (8.0%) trailers may have occurred during loading, transport, or unloading. The door width of SD trailers is the same as entire width of the trailer, whereas doors in the PB trailer are only approximately one-third the width of the trailer. Because the head and face were the most prevalent area for injury, the width of the door opening may be a factor in the greater injury rate in the PB horses. Horses loading into PB trailers used ramps from the door located at the back of the trailer to enter the appropriate compartment, and this may have contributed to the increase in injury rate of horses in the front (28.6%) and middle (31.0%) as compared to the rear (16.7%) compartment. However, head injuries do not represent an economic loss to meat quality or quantity from the carcass.

Duration of Trip. The duration of travel ranged from 5 h 45 min to 30 h. The cooperating slaughter facility was located in Fort Worth, Texas, and most travel from any region of the United States to Texas can be completed in less than 30 h. Because horses were not fed or watered during the journeys, dehydration and muscle fatigue were considered probable. Lactate, an indicator of fatigue, showed greater elevation over pre-transit values in horses following the long (27 to 20 h) and medium (16 to 23 h) trips compared to short (< 6 h) trips. Greater weight loss in horses shipped for long and medium durations than in those shipped short durations may be due to dehydration and fecal and urine losses. Hematocrit and total protein concentration showed similar results, with incremental increases as duration increased. The mean post-transport values for hct of horses transported long, medium, or short distances did not exceed the normal reference range (Jain, 1993). However, the mean pre-transit concentration for total protein of 8.4 g/dL in all horses was slightly elevated above the reference range (Kaneko et al., 1997), and all three duration groups continued to exceed the reference range following transport, indicating that the horses were mildly dehydrated prior to loading and that this was exacerbated during transport. All horses had access to water in the interval between sample collection and loading, and this may account for the small decrease in total protein in the short duration group. These trends in dehydration indices support a previous study using tame horses undergoing road transport that showed hct values remained within reference range but total plasma protein concentration continued to rise during the 24-h transport period (Friend et al., 1998).

The mean WBC numbers were within normal reference range both before and after transit in horses shipped for long, medium, or short durations; however, the response for the long duration group was greater ($P < .05$) than that for the medium or short duration groups, indicating an immunological impact of transport over 27 h. The cortisol increase showed no difference due to the duration of transport, perhaps due to its short half-life of 1 to 1.5 h in horses (Lassourd et al., 1996). The response of the N:L ratio was greatest in long and short duration groups. This elevation may be due to the stress of loading in the horses shipped a short distance and the fatigue in those shipped the longest distance. Rectal temperature increased in the medium duration group compared to the short or long duration groups. However, the pre and post-transit rectal temperature for all three groups was within the normal range of 37.5 to 38.5°C.

Fatigue, dehydration, and stress may contribute to the threefold increase in injuries in horses traveling 27 to 30 h compared with those that traveled for a shorter duration. Additionally, the horses of the long duration group had an extended period of time to interact with other horses in the confined area, thus sustaining additional injuries.

Floor Area. The amount of space per animal has been studied for cattle (Eldridge et al., 1988; Tarrant et al., 1992) to identify the stocking density that provided maximum comfort and minimum injuries (Grandin, 1981, 1997). These data are limited for horses, because many recreational horses are transported individually in trailers or vans. Generally, an individual stall in a trailer or van is .76 to .91 m in width, 1.8 to 2.1 m in length, and provides 1.40 to 1.95 m^2/horse. The trailers with HFA (1.40 to 1.54 m^2/horse) provided floor areas similar to those in trailers with individual stalls, whereas the LFA (1.14 to 1.31 m^2/horse) trailers provided less floor area. No difference due to floor area was shown for weight loss or increase in rectal temperature, cortisol, or lactate following transport. The increase due to transport of WBC, N:L, and total protein values was less in the HFA than in the LFA group. The dehydration indicators, hct and total protein, showed conflicting responses. The LFA group showed less increase in hct change than the HFA, but all mean values were within the reference range (32 to 53%) (Jain, 1993). With the exception of the hct data, the physiologic indices exhibited significantly smaller responses in HFA than in LFA in pre- and post-transit samples. These data support the recommendation of providing not less than 1.40 to 1.54 m^2/horse during travel. This may depend on the weight, conformation, and size of the horse. However, there was a twofold increase in the percentage injured in the HFA compared to the LFA groups. Thus, providing more space per horse may increase the percentage of injured horses, but the physiological responses to transportation stress are generally less.

Implications

Slaughter candidates are middle-aged, mature horses, usually of Quarter Horse or Thoroughbred breeding, in moderately fleshy body condition. The design of trailers for transporting the horses unequivocally affects their physiological responses and frequency of injuries. Thus trailer designs should be improved. As the duration of the transport incrases, especially beyond 27 h in the summer conditions, muscle fatigue and dehydration become major physiological concerns, and the physiological responses should clearly be evaluated during each season of the year. Physiological data indicate the floor area should be approximately 1.40 to 1.54 m^2/horse), but the number of injuries seems considerably less in trailers with floor areas from 1.14 to 1.31 m^2/horse. This information is important for drafting regulations appropriate for transporting horses in commercial markets in the United States.

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