Influence of saline water on intake, digesta kinetics, and serum profiles of steers

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Abstract

Nine yearling Holstein steers (avg weight 234 kg) were used to evaluate the influence of water salinity on feed and water intake, as well as several ruminal and serum characteristics. The ruminally cannulated steers were individually fed low-quality mixed hay simulating a range diet. Steers were assigned randomly to receive either control (C) water containing 350 ppm total dissolved solids (TDS) or a treated water (HS) containing 2,300 ppm TDS. The experiment included a 14-day adjustment period and a 15-day measurement period. High-saline water did not affect (P = 0.18) feed or water intake, although there was a tendency for greater consumption of both feed and water in HS steers. The HS steers had slower (P = 0.10) particulate passage rates and longer (P = 0.06) rumen retention times on day 1 of the measurement period, indicating possible differences in particle density and (or) particle size. On day 1, undigested dry matter (DM) fill was greater (P = 0.05) in HS steers compared with C (80.7 vs 61.5 g/kg BW); similar trends occurred on day 8. The HS steers also had greater (P = 0.02) rumen fluid volumes, but similar (P = 0.45) fluid dilution rates compared with C steers. No in situ DM disappearance differences were detected ($P \ge 0.38$) at incubation times ranging from 12 to 72 hours. No clinical or sub-clinical toxicological symptoms were observed in HS compared with C steers. This study suggests that cattle can ingest saline water containing 2,300 ppm TDS on a short-term basis with no adverse effects.

Key Words: cattle nutrition, nutrient digestion, rumen kinetics, salt toxicity

Wells that penetrate salt deposits, lakes in arid areas, and irrigation runoff are sources of saline water that may exceed the salt tolerance of livestock (NRC 1974). Levels and types of minerals in water vary greatly, depending on soil structure and availability of soil minerals. Subtle differences in water quality may elicit subclinical responses in cattle. Limited exploration has been conducted to evaluate the short-term impact of water quality on feed and water intake, digestive kinetics, and hematological and serum chemistry profiles. Weeth and Capps (1972) found beef heifers discriminated against water with 1,462 ppm sulfate and rejected water containing 2,814 ppm sulfate. Cattle on increased production levels may suffer from saline water of lower salt content than those on a maintenance regimen (NRC 1980); however, Ray (1989) demonstrated that a high-energy diet reduced effects of heat stress and saline water ingestion. The present study used individually fed Holstein steers given a simulated range forage diet to compare the influence of drinking water containing 350 ppm total dissolved solids (TDS) to water containing 2,300 ppm TDS on intake, rumen kinetics, and hematological and serum chemistry profiles. We hypothesized that the greater water salinity would cause an increase in water intake sufficient to offset gross effects on feed intake and digestibility, but perhaps not sufficient to eliminate inconspicuous effects on certain ruminal and blood characteristics.

Materials and Methods

Nine ruminally cannulated yearling Holstein steers (234 kg \pm 5 kg) were assigned randomly to 2 treatment groups. All steers were fed a roughage diet consisting of 33% alfalfa hay (Medicago sativa L.; 18% crude protein, as-fed basis) and 67% mature blue grama grass hay (Bouteloua gracilis [H.B.K.] Lag Ex. Griffiths; 4.5% crude protein, as-fed basis). Both hays were ground with a Bearcat hammermill through a 2.54-cm screen, then hand-mixed for each individual feeding. Steers were housed in individual 1.8 m \times 4.9-m pens, offered feed and water ad libitum, and provided a free-choice block salt and mineral supplement. After a 14-day adjustment period, 4 steers were assigned randomly to receive high-saline water (HS) and the remaining 5 to control, low-saline domestic water (C: 18 February 1989 = day 1). All steers were maintained on the trial for an additional 15 days. The HS water consisted of 812 g sodium chloride, 385 g calcium chloride, 476 g calcium sulfate, and 840 g magnesium sulfate added to 1,000 liters domestic water. The C water contained 350 ppm TDS and the HS water contained 2,300 ppm TDS.¹ A 2-day supply of HS water was mixed and stored in 208-liter steel barrels; C water was stored in identical barrels at the same location to minimize any influence of temperature or other unidentified factors.

Steers were fed twice daily (0700 and 1600 hours) and refusals were collected and weighed before the a.m. feeding. Fresh water was offered free-choice after each feeding in individual containers, and consumption records were compiled daily.

Particulate passage rate (PPR) was measured by pulse dosing 250 g Ytterbium-labeled hay at 0700 hour on day 1 and day 8. The labeled hay was prepared by immersing 5 kg of the hay diet in a solution containing 25 g ytterbium chloride per liter of water for 24 hours (Teeter et al. 1984). Labeled hay was removed, rinsed for 15 min and dried at 50° C for 48 hours. Fecal grab samples were collected from each steer at 0, 4, 8, 12, 18, 24, 30, 36, 42, 48, 54, 60, 72, 84, and 96 hours post dosing. Fecal samples were dried immediately after collection at 50° C for 96 hours, and ground to pass a 2 mm screen. Ytterbium was extracted with diethylenetriaminepen-

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taacetic acid (DTPA) and potassium chloride, then double filtered through Whatman #1 filter paper (Karimi et al. 1986). Ytterbium concentration was measured by atomic absorption spectrophotometry with nitrous oxide plus acetylene flame. Particulate passage rate estimates were determined by fitting the data to an age-dependent, 1-compartment model (Krysl et al. 1988).

Fluid dilution rate (FDR) was determined by intraruminally dosing 200 ml Co-EDTA containing 2.354 mg Co/ml (Uden et al. 1980) on day 1 and day 8. Ruminal samples were collected 0, 4, 8, 12, 24, 36, and 48 hours after dosing. The samples were analyzed for pH with a combination electrode, strained through 4 layers of cheesecloth, acidified with 1 ml sulfuric acid per 100 ml of ruminal fluid, and frozen in plastic bags for storage. The Co-EDTA labeled ruminal fluid samples were thawed at room temperature and centrifuged at $10,000 \times g$ for 10 min. The supernatant was decanted and analyzed for cobalt concentration by atomic absorption spectrophotometry with an air plus acetylene flame (McCollum and Galyean 1985). Fluid dilutation rate estimates were calculated by regressing the natural logarithm of cobalt concentrations at 0, 4, 8, 12, 24, 36, and 48 hours after dosing on time. Ruminal fluid volume was calculated by dividing marker dose by marker concentration in the rumen at 0 hour.

Three g samples of the mixed hay diet, ground to pass a 2-mm Wiley mill screen, were placed in a 9×16 -cm nylon bag with a pore size of $27 \times 47 \ \mu$ m for determination of in situ dry matter disappearance (DMD). Beginning at 0700 hour on day 1 and day 8, duplicate bags of the diet plus a blank bag were suspended in the rumen of each steer for 12, 24, 48, and 72 hour incubation times. Following removal from the rumen, bags were rinsed with tap water until effluent was clear; bags were immediately placed in a -18° C freezer for storage. After drying the bags at 50° C for 48 hours and 100° C for 1 hour, DMD was estimated as weight loss of material at each incubation time.

Blood samples were taken at 0600 hour, before feeding on day 1, 2, 3, 4, 5, 8, and 15. Two jugular blood samples were drawn from each steer, one sample drawn in a heparin Vacutainer and the other sample drawn in a serum separation tube. The serum samples were allowed to clot for 30 min, then centrifuged at approximately 1,000 \times g for 20 min. Serum was decanted and analyzed for glucose, total cholesterol, triglycerides, blood urea nitrogen, creatinine, blood urea nitrogen-creatinine ratio, uric acid, albumin, globulin, total protein, albumin-globulin ratio, bilirubin, alkaline phosphatase, lactate dehydrogenase, serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), AST-ALT ratio, sodium, potassium, chloride, bicarbonate/carbon dioxide, anion gap, calcium, inorganic phosphorus, and calculated osmolality. Complete blood counts of whole blood were determined for white blood cells, red blood cells, hemoglobin, hematocrit, mean cell volume, mean cell hemoglobin, platelet count, and mean platelet volume. All blood analysis was conducted in a commercial laboratory² but blind standards were not submitted. These analyses were used to evaluate sub-clinical responses and possible renal and hepatic dysfunctions caused by ingestion of HS water. The steers were examined by a licensed veterinarian for general health on days 1, 8, and 15.

All statistical computations were made with the GLM procedures of SAS (1985). Feed and water intake data, blood parameters, and digesta kinetic measures were analyzed as a split plot design with treatment in the main plot and sampling day in the subplot. The in situ digestion data were analyzed as a split-split plot design with treatment in the main plot, day in the subplot, and incubation time in the sub-subplot. Since steers were individually penned, fed, and watered, they were considered as replications. Treatment effects were tested by the mean squares of steers within treatment; the remaining main effects and effects of interactions were tested against the residual mean square appropriate for the design used.

Results and Discussion

Feed and Water Intake

High-saline water did not affect (P=0.18) short-term feed or water intake, although consumption of feed and water by HS steers tended to be numerically greater than for C steers (Table 1). When

Table 1. Least squares means for feed and water consumption by steers given either control (C) or high-salinity (HS) drinking water.

	Water	source		OSL ³
Item	С	HS	SE ²	
Feed intake:				
Kg DM/d	5.47	5.51	.37	.94
Kg DM/d/kg BW	.023	0.25	.001	.25
Water intake:				
1/d	22.0	23.3	1.27	.47
ml/d/kg BW	91.0	105.0	.007	.18

¹Total dissolved solids for control and high-saline drinking water was 350 and 2,300 ppm, respectively. ²Standard error of the mean, $n \approx 9$.

³Observed significance level.

dry matter intake (DMI) was expressed as a percentage of body weight (BW), HS steers consumed 2.5% per day compared with 2.3% for C steers. Wilson and Dudzinski (1973) reported feed intakes increased as water intake increased. When diet moisture is low, increased water intake may stimulate feed intake. Peirce (1957) postulated that adaptation of ruminal microbes to increased ruminal salt concentration accounted for increased feed consumption associated with salt water ingestion by sheep. However, Rogers et al. (1979) reported DMI was reduced in steers fed a highroughage diet and given an intraruminal infusion of 1 kg of sodium chloride and 9 liters of water. Dry matter intake increased when 9 liters of water only were infused. This would indicate DMI response is possibly related to water intake, with a high level of salt ingestion being detrimental.

Water intake evaluated on a BW basis was similar (P = 0.18); however, a trend for greater consumption of HS water may have existed, as HS steers consumed 105 ml/kg BW compared with 91 ml/kg BW for the C steers (Table 1). Water intake by cattle has been described as a function of DMI and ambient temperature (Winchester and Morris 1956). Increased water consumption may suggest a response to increased salt intake; increased water consumption, in turn, appeared to stimulate feed consumption. It has been suggested increased water consumption is a physiological response to increased salt load. Squires (1988) indicated additional water is required to maintain systematic osmotic balance, with the excess salt flushed from the body via the urine. Potter (1961, 1963, 1966, and 1968) demonstrated that an adaptive mechanism, involving kidney function, allowed sheep to adapt to large quantities of ingested or infused salt. This mechanism also may be related to ruminal conditions; as the content of soluble salt in drinking water increased, water intake increased (NRC 1974).

Ray (1989) reported 4 consecutive feedlot experiments over a 2-year period (2 summers and 2 winters) using crossbred steers averaging 222.5 kg. Average maximum and minimum temperatures during winter were 25.1° C and 8.0° C compared with summer temperatures of 35.3° C and 17.5° C, respectively. Water consumption during the winter trials averaged 125.4 ml/kg BW compared with 144.6 ml/kg BW for summer. This demonstrated the effects of thermal conditions on water intake. Our study was conducted during moderate weather conditions; mean maximum

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Table 2. Effects of high-saline water ingestion on digesta kinetics of steers fed mixed hay diet¹ consuming either control (C) or high-saline (HS) water².

		Da	ay 1		Day 8		ıy 8	y 8		Combined		
	Trea	Treatment			Treatment				Treatment	tment		
	HS	С	SE ³	OSL⁴	HS	С	SE	OSL	HS	С	SE	OSL
]	Particulate	kinetics					
Particulate passage											······	
rate, % hour	2.4	3.0	0.24	.10	2.8	3.1	0.25	.52	2.6	3.0	0.23	.21
Ruminal retention												
time, hour	5.16	40.6	3.73	.06	43.0	39.8	3.33	.49	47.3	40.2	3.20	.14
Particulate gasto- intestinal retention												
time, hour	74.6	64.0	4.93	.15	65.3	63.8	3.18	.71	70.0	63.9	3.52	.24
Undigested dry matter												
fill, g kg BW	80.7	61.5	6.00	.05	75.2	65.0	7.24	.33	78.0	63.3	6.19	.12
						Fluid kin	netics					
Fluid dilution rate,												
% hour	7.0	7.7	0.54	.37	7.8	8.0	0.39	.70	7.4	7.9	0.43	.45
Fluid turnover time,											0.15	.45
hour	14.6	13.1	0.98	.29	12.8	12.5	0.63	.74	13.7	12.8	0.75	.40
Fluid volume												
ml/kg BW	138.5	106.4	11.04	.07	117.3	105.3	7.21	.25	127.9	105.9	5.42	.02

Mixed hay diet was 33% alfalfa hay (18% crude protein, as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis).

²Total dissolved solids for control and high-saline drinking water was 350 and 2,300 ppm, respectively. ³Standard error of the mean, n = 9.

⁴Observed significance level.

and minimum temperatures were 20.7° C and 3.7° C, respectively, with an average wind velocity of 3.4 km/h and an evaporation rate of 0.51 cm/d.

Digesta Kinetics

Particulate passage rate (PPR) was evaluated on day 1 and day 8 to assess short-term ruminal adaptation to saline water ingestion. No treatment by day interactions were detected (P > 0.15), and estimates for total period PPR were similar (P = 0.21) between treatments (Table 2). Rumen retention time was longer (P = 0.06) for HS steers than C on day 1; this trend also was evident for the total period (P = 0.14). Undigested dry matter fill was higher (P =0.05) for day 1 in HS than C steers; no differences were apparent for day 8 (P=0.52), but for the total period, a trend for greater (P=0.12) fill continued in HS steers. Total tract particulate retention time was similar ($P \ge 0.15$) for steers on the 2 water treatments. Results suggest a possible difference in particle density and (or) particle size. Longer rumen retention time of undigested forage particles depends not only larger particle size, but also on low specific gravity particles (Owens and Goetsch 1988).

Ruminal fluid kinetics also were evaluated on day 1 and day 8 to assess the influence of HS water on ruminal flow rate and rumen volume (Table 2). No treatment by day interactions were detected among the fluid kinetic parameters studied. Also, neither fluid dilution rate (P = 0.45) nor fluid turnover time (P = 0.40) differed between treatments. For the combined period, HS steers had a rumen fluid volume of 127.9 ml/kg BW compared to 105.9 ml/kg BW for C steers (P = 0.02). Other researchers have reported conflicting results. Rogers et al. (1979) reported that, when 9 liters of water were infused into the rumen of steers fed a high roughage diet, rumen liquid volume and fluid dilution rate were not altered. However, when 500 mg of sodium chloride were infused with the same amount of water, fluid dilution rate increased but rumen liquid volume was not affected. Potter et al. (1972), comparing fresh water with water containing 1.3% sodium chloride, reported greater voluntary intake of salt water and greater flows of fluid through the rumen of sheep. Both Potter et al. (1972) and Rogers et al. (1979) were infusing sodium chloride. In our experiment HS water contained calcium chloride, calcium sulfate, and magnesium sulfate in addition to sodium chloride. The combination of calcium, magnesium and sulfate ions in addition to sodium plus chloride ions could have altered fluid kinetics of the rumen. No differences or trends in ruminal pH (data not shown) were apparent in the HS steers compared with C steers.

In Situ Dry Matter Disappearance

No differences ($P \ge 0.38$) were detected for in situ dry matter disappearance (DMD) between HS and C steers for any of the 4 incubation times studied (Table 3). No treatment by day interactions (P>0.10) for any of the incubation times were detected; thus,

Table 3. Effects of high-saline water ingestion on in situ dry matter disappearance of steers fed a mixed hay diet' consuming either control (C) or high-saline (HS) water².

	Treat	tment		OSL⁴
Incubation time	HS	С	SE ³	
(Hour)				
12	37.2	36.1	1.47	.59
24	45.1	43.9	1.51	.56
48	53.9	51.6	1.88	.38
72	59.7	59.0	1.32	.71

¹Mixed hay diet was 33% alfalfa hay (18% crude protein as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis). ²Total dissolved solids for control and high-saline drinking water was 350 and 2,300

pm, respectively

ppm, respectively. ³Standard error of the pooled mean, n = 18. No treatment \times day interactions (*P*>0.10) were detected; observations represent means of day 1 and day 8 data. ⁴Observed significance level.

data shown in Table 3 represent an average of the results observed on days 1 and 8. In some studies, ruminal microflora changes have been observed in animals fed high-salt diets compared with more conventional diets. Thomson et al. (1978) reported changes in species of ruminal bacteria between sheep fed a high-salt diet compared with those fed a low-salt diet. Higher levels of dietary salt may also cause a reduction in size and number of protozoa (Hungate 1966). If such changes in microbial populations occurred, they had no discernible effect on rate of DMD in this experiment.

Blood and Serum Profiles

Serum profiles were evaluated at 0600 hours on day 1, 2, 3, 4, 5,

Table 4. Effects of high-saline water ingestion on serum electrolytes of steers fed a mixed hay diet1.

	Treatment ^{2,3}				
Item	HS	С	SE ⁴	OSL	
Sodium, mEq/liter	141.8	141.9	0.49	0.91	
Chloride, mEq/liter	105.5	105.1	0.39	0.43	
Potassium, mEq/liter	4.7	4.6	0.17	0.65	
Bicarbonate/carbon					
dioxide, mEq/liter	24.4	26.2	0.71	0.09	
Anion gap, mEq/liter	12.1	10.6	0.55	0.09	
Calculated osmolality,					
m oSM/liter	281.0	281.0	1.29	0.98	
Calcium, mEq/dl	9.5	9.5	.08	0.62	
Inorganic phosphorus,					
mEq/dl	4.7	4.6	0.17	0.65	

Steers were fed ad libitum, a diet consisting of 33% alfalfa hay (18% crude protein, as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis). ²High-saline (HS) and control (C) steers receive, ad libitum, water containing 2,300 and 350 ppm total dissolved solids, respectively. ³No treatment × day interactions (P>0.10) were detected, therefore, day means were

pooled across treatments. Standard error of the mean, n = 9.

⁵Observed significance level.

8, and 15. No treatment by day interactions (P > 0.10) were detected. Pooled across sampling days, HS steers had a wider (P= 0.09) anion gap compared with C steers (12.1 vs 10.6 mEq/liter; Table 4). This appeared to be the result of lower (P=0.09) bicarbonate/carbon dioxide concentration in HS steers compared with C steers (24.4 vs 26.2 mEq/liter, respectively). No other electrolytes differed ($P \ge 0.43$) between HS and C steers. No treatment by day interactions (P > 0.10) or treatment effects ($P \ge 0.21$) were evident for serum renal or hepatic parameters (Table 5), indicating no adverse short-term effects on kidney or liver functions for ingestion

Table 5. Effects of high-saline water ingestion on serum renal and hepatic parameters of steers fed a mixed hay diet1.

	Treatment ^{2,3}				
Item	HS	С	SE ⁴	OSL	
Urea nitrogen (BUN),	· · · · ·				
mg/dl	14.0	13.6	2.2	0.90	
Creatinine, mg/dl	1.1	1.2	0.07	0.21	
BUN/creatinine ratio	11.8	10.5	1.39	0.44	
Bilirubin, mg/dl	0.23	0.23	0.0026	0.99	
Uric acid, mg/dl	1.01	0.96	0.069	0.58	

¹Steers were fed ad libitum, a diet consisting of 33% alfalfa hay (18% crude protein, as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis).

²High-saline (HS) and control (C) steers received, ad libitum, water containing 2,300 and 350 ppm total dissolved solids, respectively. ³No treatment \times day interactions (*P*>0.10) were detected, therefore, day means were

pooled across treatments. *Standard error of the mean, n = 9.

⁵Observed significance level.

of HS water. No treatment by day interactions (P>0.10) or treatment effects ($P \ge 0.42$) were detected for serum energy metabolites and serum proteins (Table 6). Aspartate aminotransferase was lower (P = 0.07) for HS steers than C steers (65 vs 81 μ /liter, respectively; Table 7). All other enzyme values were similar ($P \ge 0.33$). Hematological profiles revealed no treatment by day interactions (P>0.10), but when pooled across sampling days, HS steers had a greater (P = 0.07) white blood cell count compared with C steers (Table 8). All serum constituents and hematological profiles evaluated were within normal ranges reported by Kaneko (1989) and Galycan and Hallford (1983). No clinical or sub-clinical symptom profiles were observed in the HS steers.

Management Implications

No differences were observed for feed intake, water intake or

Table 6. Effects of high-saline water ingestion on serum energy metabolites and proteins of steers fed a mixed hay diet¹.

	Treatment ^{2,3}				
Item	HS	С	SE ⁴	OSL	
Glucose, mg/dl	69.1	68.22	1.55	0.66	
Triglycerides, mg/dl	26.3	22.5	3.73	0.42	
Cholesterol, mg/dl	85.6	83.4	5.15	0.72	
Albumin, g/dl	3.4	3.3	0.12	0.49	
Globulin, g/dl	3.9	4.0	0.24	0.71	
Albumin/globulin ratio	0.88	0.81	0.080	0.46	
Total protein, g/dl	7.3	7.3	0.15	0.96	

'Steers were fed ad libitum, a diet consisting of 33% alfalfa hay (18% crude protein, as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis). ²High-saline (HS) and control (C) steers received, ad libitum, water containing 2,300 and 350 ppm total dissolved solids, respectively.

³No treatment \times day interactions (P>0.10) were detected, therefore, day means were pooled across treatments. ⁴Standard error of the mean, n = 9.

⁵Observed significance level.

Table 7. Effects of high-saline water ingestion on serum enzymes of steers fed a mixed hay diet¹.

	Treatment ^{2,3}					
Item	HS	С	SE ⁴	OSL		
Lactate dehydrogenase,						
μ/l	1320	1520	256.2	0.53		
Aspartate aminotrans-						
ferase (AST), $\mu/1$	65	81	6.6	0.07		
Alanine aminotrans-						
ferase (ALT), $\mu/1$	25	28	4.7	0.67		
AST/ALT ratio	2.6	3.2	0.43	0.33		
Alkaline phosphatase,						
μ/l	79	71	11.7	0.61		

¹Steers were fed ad libitum, a diet consisting of 33% alfalfa hay (18% crude protein, as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis). ²High-saline (HS) and control (C) steers received, ad libitum, water containing 2,300

and 350 ppm total dissolved solids, respectively. ³No treatment \times day interactions (P>0.10) were detected, therefore, day means were

pooled across treatments. Standard error of the mean, n = 9.

⁵Observed significance level.

Table 8. Effects of high-saline water ingestion on hematological profiles of steers fed a mixed hay diet1.

	Treatment ^{2,3}					
Item	HS	С	SE ⁴	OSL		
White blood cells						
(× 1000)	18.4	12.4	2.13	0.07		
Red blood cells						
(× 1,000,000)	5.4	6.0	0.43	0.35		
Hemoglobin, g/dl	11.9	12.3	0.59	0.65		
Hematocrit, %	24.0	26.8	2.01	0.33		
Mean cell volume, F/1	44.5	45.0	0.31	0.28		
Mean cell hemoglobin,						
picogms	22.9	20.8	1.28	0.26		
Mean cell hemoglobin						
conc., g/dl	51.4	46.1	2.97	0.22		

Steers were fed ad libitum, a diet consisting of 33% alfalfa hay (18% crude protein, as-fed basis) and 67% blue grama hay (4.5% crude protein, as-fed basis). ²High-saline (HS) and control (C) steers received, ad libitum, water containing 2,300

and 350 ppm total dissolved solids, respectively. ³No treatment \times day interactions (*P*>0.10) were detected, therefore, day means were

pooled across treatments. ⁴Standard error of the mean, n = 9.

⁵Observed significance level.

diet digestibility when HS steers were compared with C steers. Also, no clinical or sub-clinical effects of greater salt intake were detected in HS steers. This suggests that cattle may ingest water containing up to 2,300 ppm total dissolved solids on a short-term basis with no adverse effects.

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