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Eye white percentage as a predictor of temperament in beef cattle

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ABSTRACT: Accurately evaluating and selecting for calm temperament in beef cattle is important for economic and animal welfare reasons. Previous studies have shown that eye white (EW) can be a predictor of a multitude of emotions across different situations, but there is little research on the relationship between EW and temperament. The objective of this experiment was to assess the accuracy and reliability of using the percentage of exposed EW as a predictor of temperament in beef cattle. Forty-eight heifers (group 1), 39 bulls (group 2), and 60 steers (group 3) were video-recorded while in a squeeze chute, and 2 still digital images from each animal were selected for EW determination. Chute temperament scores were assigned: 1 (calm) to 5 (agitated). Flight speeds were measured blindly and independently during a subsequent test in which the amount of time it took a solitary animal to pass a handler and travel a specified distance was recorded. The EW area in each image was measured using Sigmascan Pro 5 and was expressed as the percentage of exposed eye area. Each image was analyzed twice to determine tracing repeatability. Pearson correlation coefficients were calculated among 2 images of the same animal, as well as among duplicate readings of the same image to determine animal and tracing repeatabilities. The mean percentages of EW were 30.14 ± 14.37, 31.43 ± 14.77, and 28.57 ± 12.38, and the average percentage accuracy for duplicate image EW measures was 96, 96, and 93 ($P < 0.0001$) for groups 1, 2, and 3, respectively. The Pearson correlation coefficients for EW percentage and chute temperament scores were 0.674 ($P < 0.0001$), 0.95 ($P < 0.0001$), and 0.696 ($P < 0.0001$), whereas the correlations between EW and flight speeds were 0.415 ($P < 0.0001$), 0.333 ($P < 0.05$), and 0.294 ($P < 0.01$) for groups 1, 2, and 3, respectively. Results from this study indicate that percentage EW in cattle could be used as a quantitative tool with minimal equipment to assess temperament in beef cattle, providing an objective method for temperament selection.

Key words: beef cattle, behavior, eye white, performance testing, selection program, temperament

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INTRODUCTION

There are many traits throughout the livestock industry that are selected for and are economically important to producers (Gauly et al., 2001; Petherick et al., 2002) and demanded by consumers. Temperament (a set of behavioral characteristics that are stable over time and across situations) in beef cattle is often overlooked because current measures are qualitative or deemed too invasive, expensive, or time consuming (Adeyemo and Heath, 1982; Curley et al., 2006; Bristow and Holmes, 2007). Many studies have shown direct [increasing workplace and animal safety (Vann, 2006)] and indirect [increased growth efficiency and meat quality (Fordyce et al., 1988; Burrow and Dillon, 1997; Brown et al., 2004)] advantages in including temperament within selection programs, the indirect often perceived as outweighing the direct financially.

An additional advantage of selecting for docility is increasing animal welfare through decreases in daily stress levels and rough handling during transport and processing (Grandin, 1993; Rushen et al., 1999). Selecting for docile beef cattle could be an important factor that could increase overall welfare.

A quantitative and inexpensive method is needed so temperament can become a part of more selection programs. Currently, chute temperament scores (**CTS**) and flight speeds (**FS**) are considered to be the gold standards of temperament evaluation (Voisinet et al., 1997; Curley et al., 2006; Müller and von Keyserlink, 2006). Sandem et al. (2002) found that the percentage of revealed eye white (**EW**) was highly repeatable and correlated with the startle response in dairy cattle, whereas Lanier et al. (2000) found that the magnitude of a startle response was positively correlated with temperament scoring. We therefore hypothesized that the amount of exposed EW would be a good predictor of

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Table 1. A summary of environmental, animal, and methodological differences between groups 1, 2, and 3

Item	Group 1	Group 2	Group 3
Sex	Heifers	Bulls	Steers
n	48	39	60
Housing	3.5 m × 15 m (n = 4/pen)	30 m × 30 m (n = 20/pen)	30 m × 30 m (n = 20/pen)
Restraint chute	Chute B	Chute A	Chute A
Treatment in chute	Infrared/ultrasound	Infrared/ultrasound	Infrared/ultrasound/blood sampling
Eye recorded	Left eye	Right eye	Right eye
Flight speed assessed	2 to 4 h after CTS[1]	1 wk after CTS	2 to 4 h after CTS
Flight speed environment	Alley near home pens (length: variable, width: 1 m)	Scrape alley (length: 13 m, width: 3 to 5 m)	Scrape alley (length: 13 m, width: 3 to 5 m)

[1]CTS = chute temperament score.

temperament in cattle. The objective was to assess the accuracy and reliability of using percentage EW as a predictor of temperament in beef cattle by comparing it with CTS and FS.

MATERIALS AND METHODS

All procedures that were used in this experiment involving animals were approved by the University of Guelph animal care committee.

Data were collected from 48 heifers (group 1), 39 bulls (group 2), and 60 steers (group 3) at the end of their respective feeding trials (84, 112, and 149 d, respectively). The 3 groups were a mix of British (predominantly Angus), Continental (predominantly Simmental), and Piedmontese breed crosses and were housed at the Elora Beef Research Centre in the township of Elora in southwestern Ontario (Canada). Animals that were used for this study were between 12 and 16 mo of age. A list of experimental and environmental differences that were present between the 3 groups of animals can be found in Table 1.

Animals were moved from their home pens into a holding pen (which held 4 to 8 animals) that was connected to a raceway leading to the squeeze chute. Each animal was moved through the raceway and restrained within the chute for 3 to 6 min on average. This was because of the amount of other data that were collected at the same time for other experiments. All of the images analyzed were taken within 2 to 4 min after the animal was restrained.

Five animals were restrained for approximately 10 min due to additional setup procedures. The EW video data and CTS were collected for each animal for this study while they were restrained in the chute. Collection of ultrasound measurements and infrared images occurred simultaneously for all animals while blood was collected simultaneously for group 3 animals. The ultrasound, infrared, and blood sampling measurements were collected for an alternative study. These measures were collected on a weekly basis so the animals were well habituated to these processes as well as being familiar with being restrained.

The video camera that was used to collect the EW video data was a color Panasonic CCTV camera (Panasonic WV-CP240, Panasonic Canada Inc., Mississauga, Ontario, Canada) that was positioned, on average, 1.5 m to the side of the head of the animal. An I3DVR International digital video recorder was used to store the video (Kodicom model i31808WM, i3DVR International Inc., Scarborough, Ontario, Canada), and the program SRX-Prolite (i3DVR International Inc.) was used for digital recording.

A single observer assigned CTS while standing at the side of the chute, behind the head of the animal, focusing on limb and body movements. The CTS criteria were based on a 1 to 5 scale as described by Voisinet et al. (1997): 1—calm, no movement; 2—restless shifting, 3—squirming, occasional shaking of device; 4—continuous vigorous movement and shaking of device; 5—rearing, twisting, or violently struggling. Vocalizations were also recorded while the animals were in the chute, but these data were not further analyzed because 95% of the animals received the same vocalization score. When all of the measurements were collected, the animal was released into a pen and from there herded back into its home pen.

The FS was measured independently during a subsequent test as an additional form of temperament assessment (Müller and von Keyserlink, 2006). The first handler stood at the far end of the alley, keeping the herd at that end. The second handler herded a segregated animal to the opposite end of the alley. When it reached the far wall, the handler continued to approach the animal with wide arms until it escaped back to the herd. An observer with a stopwatch was positioned at the finish line but was well out of the peripheral vision of the animal. The stopwatch was used to measure the amount of time it took each segregated animal to pass the handler and reach a specified point, closer to the herd. A FS was then determined in meters per second for each animal.

After all of the data were collected, the video was analyzed. Two still images were selected for each animal from the video and were used to calculate the percentage of exposed EW. The images were selected on the basis of being representative of the estimated average

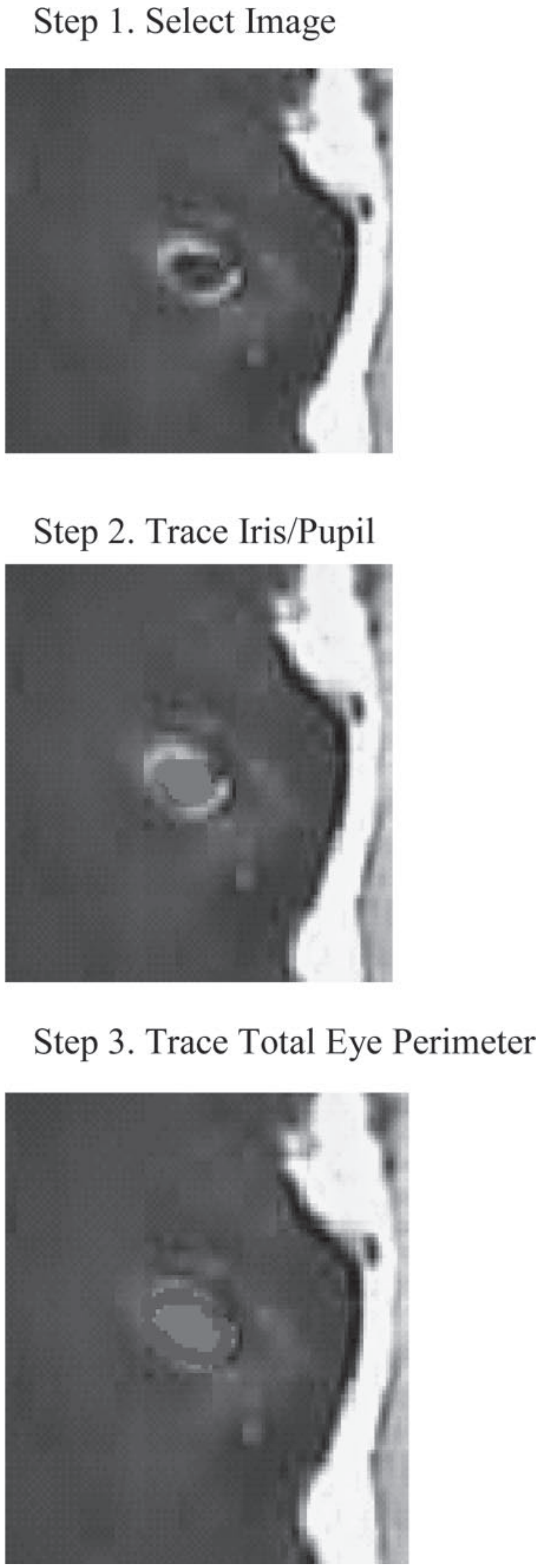

Figure 1. Eye diagram.

EW of the animal for the entire video clip. They were also selected when the head of the animal was perpendicular to the camera and when it was not performing head, neck, or eye movements. During image selection, the observer was blinded to any other temperament assessment results. Each picture was analyzed twice so that the accuracy of EW estimation could be calculated. The average EW estimation value was used in further analyses. The EW percentage was computed using the image analyses program Sigma Pro 5.0 (Aspire Software International, Ashburn, VA), which was programmed to evaluate the area of an image in pixels. The outer edge of the iris of the eye was traced and the area was calculated. Similarly, the total eye was also traced and an area was calculated. Figure 1 represents these steps pictorially.

The following formula was used to calculate EW percentage: [1 − (area of the iris/area of the total eye)] × 100%. This method ensured that the smallest areas of white at the top and bottom of the eye were most accurately estimated. This method of EW estimation differed from the techniques of Sandem et al. (2002); these authors used a ruler to measure the diameter of the iris and total eye diameter as they appeared on a computer screen. These values were then inputted into the formula for an ellipse, and an area was calculated using the same formula that was used in this experiment. The technique that was created for this experiment was believed to be more accurate because it did not assume the shape of the eye was a perfect ellipse.

The FS value as well as the CTS for each animal was compared with the percentage of exposed EW to determine correlation coefficients for each group of animals. Because of the differences in housing, handling experience, and testing procedures, each group was analyzed separately. The Mixed procedure (SAS Inst. Inc., Cary, NC) was used to determine the intraclass correlations and Pearson correlation coefficients among images from the same animal and among duplicate readings of the same image. The model that was used for the analysis included a random effects variable "image," nested within the variable "animal," and was compared with CTS and FS effects. The normality of the EW and FS distributions was assessed with the Univariate procedure in SAS (Figure 2).

RESULTS

The mean, SD, minimum and maximum for EW percentages, CTS, and FS are presented in Table 2. The pattern of results appeared to be similar for all 3 groups. Group 2 had the most variation in percentage EW; however, they had the least variation in CTS and had the slowest FS times. Group 3 had the greatest average CTS, whereas group 1 had the fastest FS. Direct comparisons of these results cannot be made due to the differences in test procedures that were used during the evaluations of each group.

Pearson correlation coefficients were calculated for both images of each animal as well as for the duplicate measures of each image to ensure that the method of testing and analyzing EW was reliable. The coefficients can be found in Table 3. Pearson correlation coefficients were also used to determine the correlation between percentage EW, FS, and CTS within each group (Table 4). The average of the 4 EW measurements was used for the following analyses because all 4 measures were highly correlated. Percentage EW was more strongly correlated with CTS than FS for groups, and values were highly significant. The correlation coefficients were consistent among groups with the exception of CTS in group 2. The CTS correlation with EW for group 2 was much greater than any of the other correlations that were obtained.

The results of the Proc Univariate analysis suggest that percentage EW had a more normal distribution than FS. The skewness values that were calculated for EW and FS distributions did not differ significantly; however, the kurtosis values were greater in all 3 animal groups for FS. Greater kurtosis values mean that more of the variance is due to infrequent extreme deviations. The skewness and kurtosis values are given in Table 5.

DISCUSSION

Research on EW percentage in cattle is still relatively new, and there are many questions and implications related to the topics of breeding programs, behavior, welfare, and meat science that are still left unanswered. Correlation coefficients for percentage of EW within and between images for each animal were high, ranging from 0.79 to 0.96, which shows that this method for quantifying percentage of EW was highly repeatable and that it was accurate at the level of the animal within a given test. It also implies a high level of accuracy for tracing ability, showing that the program Sigma 5.0 can be used as a reliable tool to estimate area. These increased correlations also suggest that selecting and tracing images did not provide a great source of error or noise within the rest of the analyses. Although the 3 animal groups had different experimental testing environments, the average, SD, and minimum and maximum values for all 3 temperament assessment techniques were similar. This suggests that all 3 methods of temperament assessment have similar properties in different situations.

Whereas the correlations between duplicates and images were high, the correlations were the least for group 3. This could be due to more reactive behavior in this group due to increased activity in the surrounding environment. During the data collection periods for groups 1 and 2, ultrasound data and infrared imaging were collected simultaneously and group 3 additionally underwent blood sampling. This may have caused more behavioral reactions to the environment rather than providing a pure sample of the true disposition of the animals, which may have altered the EW evaluations and CTS and resulted in overall decreased correlation coefficients.

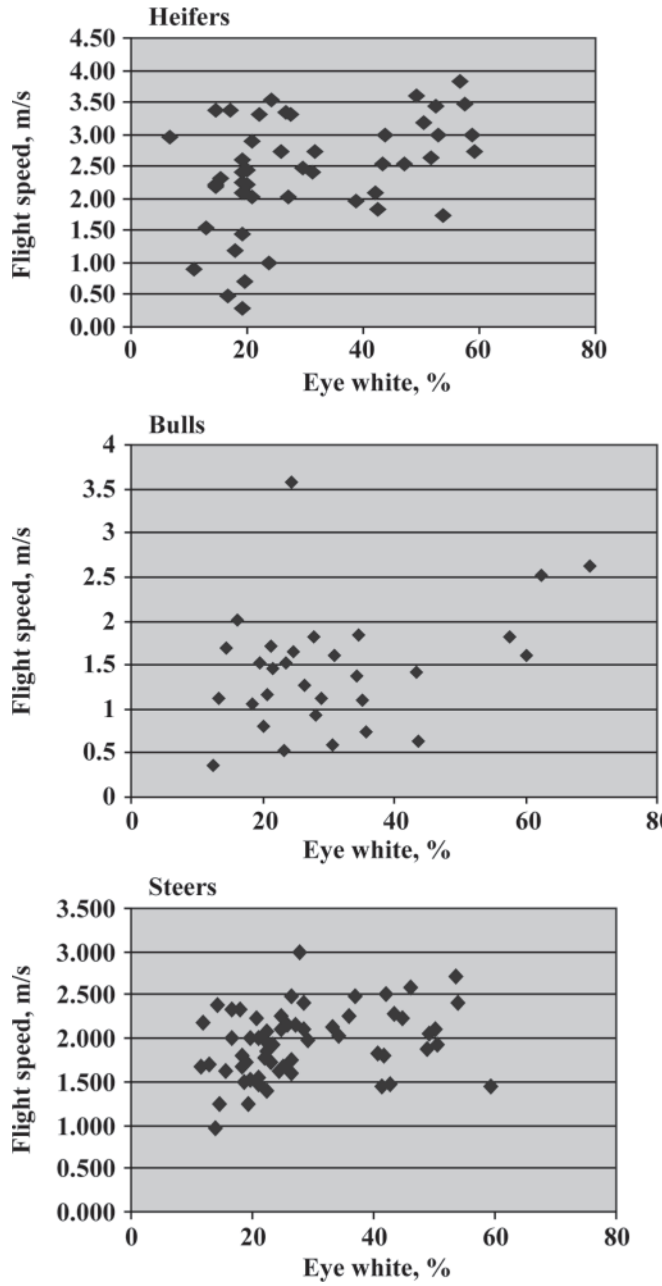

Figure 2. Flight speed and eye white distribution for heifers, bulls, and steers.

After determining that the method that was used to estimate EW was accurate at the level of the animal and within duplicates, correlations between EW percentage, CTS, and FS were calculated. Both chute scores (CTS) of reactivity and FS are measures of response to handling and are currently used to quantify temperament (Voisinet et al., 1997; Curley et al., 2006; Müller and von Keyserlink, 2006).

Assigning chute scores is relatively easy but because it is a subjective measure, the results can vary depending on the observer. Lanier et al. (2000) found the *P*-value of inter- and intrareliability tests of these types of scoring to be $P < 0.05$ with experienced observers; however, if the observers were not familiar with cat-

Table 2. Simple statistics for eye white average, flight time, and chute temperament score[1] for heifers, bulls, and steers

Variable	n	Mean	SD	Minimum	Maximum
Group 1					
Eye white average, %	48	30.14	15.37	6.8	59.2
Flight speed, m/s	48	2.41	0.85	0.29	3.83
Chute temperament score	48	2.15	0.87	1	4
Group 2[2]					
Eye white average, %	39	31.43	14.77	12.4	69.8
Flight speed, m/s	30	1.44	0.68	0.35	3.59
Chute temperament score	39	1.82	0.76	1	3
Group 3					
Eye white average, %	60	28.57	12.38	11.5	59.44
Flight speed, m/s	60	1.95	0.4	0.96	2.989
Chute temperament score	60	2.25	0.75	1	4

[1]The chute temperament score criteria were based on a 1 to 5 scale as described by Voisinet et al. (1997).

[2]Group 2: flight speed was only measured on 30 animals because 9 were slaughtered at market weight before this measure took place.

tle behavior or not practiced in scoring, the reliability dramatically decreased. The FS has also been shown to be a consistent measure within individuals and is correlated with other personality traits associated with fearfulness (Müller and von Keyserlink, 2006).

Although all of the measures were significant, the least correlation coefficient was between EW and FS for groups 1, 2, and 3 (0.415, 0.333, and 0.29, respectively). In most studies, FS is determined as animals exit a restraint chute (Curley et al., 2006), whereas in this study FS was determined at a different time and place from handling in the chute. This method was chosen because of the limitations of the facilities and provided a measure of temperament that was completely independent of all of the other evaluations. Peak speed for all 3 groups of animals was on average 3.5 m/s. The peak speed could be a limiting factor in assessing FS and explain the decreased correlation coefficients. The FS assessment took place in a scrape alley for groups 2 and 3 and an alley behind the home pens for group 1. Both locations were slippery due to fecal matter, and the average distance that the animals traveled was 4 m past the handler. In this short distance, on a slippery surface, it would have only physically been possible for the animals to reach a certain maximum speed, no matter how strong the desire to escape was. These maximum FS may have led to nonlinear relationships with EW percentage and decreased correlation coefficients. The kurtosis test results show that FS has a less normal distribution than percentage EW and that it has larger tails. This test suggests that EW percentage could be a more predictable measurement because it has a more normal distribution across all of the groups.

Although direct comparisons among the groups are not possible due to variation in the test procedures, there were some interesting differences in the results between the different groups. The correlation coefficients between EW and CTS were greater and more highly significant than those between FS and EW for all groups, but the correlation coefficient (0.95) was much greater for group 2 vs. groups 1 and 3, which were similar (0.674 and 0.696, respectively). This could

Table 3. Pearson correlation coefficients for heifers, bulls, and steers among 2 images and their duplicates[1]

	Picture 1	Picture 2	Picture 2
Item	Duplicate 2	Duplicate 1	Duplicate 2
Group 1			
Picture 1, duplicate 1	0.955*	0.842*	0.799*
Picture 1, duplicate 2		0.848*	0.822*
Picture 2, duplicate 1			0.966*
Group 2			
Picture 1, duplicate 1	0.947*	0.807*	0.843*
Picture 1, duplicate 2		0.793*	0.819*
Picture 2, duplicate 1			0.963*
Group 3			
Picture 1, duplicate 1	0.921*	0.803*	0.774*
Picture 1, duplicate 2		0.834*	0.802*
Picture 2, duplicate 1			0.938*

[1]Direct comparisons between groups were not made due to differences in environmental design.

*P-value < 0.0001.

Table 4. Pearson correlation coefficients for eye white average, flight speed, and chute temperament score[1] for heifers, bulls, and steers

Item	Flight speed	Chute temperament score
Group 1		
Eye white average	0.415	0.674
	$P < 0.0001$	$P < 0.0001$
Flight speed	1	0.378
		$P < 0.005$
Chute temperament score	0.378	1
	$P < 0.005$	
Group 2		
Eye white average	0.333	0.950
	$P < 0.05$	$P < 0.0001$
Flight speed	1	0.680
		$P < 0.001$
Chute temperament score	0.680	1
	$P < 0.001$	
Group 3		
Eye white average	0.294	0.696
	$P < 0.01$	$P < 0.0001$
Flight speed	1	0.460
		$P < 0.001$
Chute temperament score	0.460	1
	$P < 0.001$	

[1]The chute temperament score criteria were based on a 1 to 5 scale as described by Voisinet et al. (1997).

be due to group 2 having more experience in the chute and being handled more often. Increased amounts of handling experience and exposure to novel environments often results in animals having decreased fear responses to new environments and handlers, especially if the handling occurs at an early age (Boivin et al., 1994), although Müller and von Keyserlink (2006) found increases in FS in heifers over repeated tests. If the animals are more familiar with the handling procedure, a more accurate CTS could be assigned that is more indicative of the disposition of the animals because it may reflect a stable personality trait rather than response to novelty. The CTS have proved to be successful when the scoring criteria are clearly defined and is often included in Limousin breeding protocols in the United States (Le Neindre et al., 1995; Beckman et al., 2005). The correlation coefficient between FS and CTS was also the greatest for group 2 with a coefficient of 0.68 compared with 0.38 for group 1 and 0.46 for group 3. This result may also be due to less fear of handling in the chute and a better assessment of the chute behavior. Alternatively there may be sex differences in response to handling in the chute due to size and strength or differences in the physical ability of bulls (group 2) during the flight tests. Grandin (1993) found that steers were generally calmer in a chute test than bulls, which differs from our results. However, in the current study, steers were blood sampled during the test, whereas bulls were not, which could have been a significant stressor in our study. The current study was not designed to compare sexes directly.

Table 5. Skewness and kurtosis values calculated using the Proc Univariate[1] method for the temperament assessment distributions of eye white and flight speed in heifers, bulls, and steers

Group	Temperament assessment method	Skewness	Kurtosis
Group 1	Eye white	0.648	0.036
	Flight speed	0.618	−1.053
Group 2	Eye white	1.095	0.381
	Flight speed	1.087	2.370
Group 2	Eye white	0.067	−0.052
	Flight speed	0.790	−0.446

[1]SAS Inst. Inc., Cary, NC.

Further study is required to entirely understand the complex relationship between EW percent and temperament and to decipher any confounding factors such as age, sex, breed, environmental factors, and handling experience as well as possible differences between the left and right eye that could occur due to lateralization (differences in the magnitude of left and right brain outputs; De Boyer Des Roches et al., 2008). Additionally, more research could be focused on qualitative observations of EW that could be used in daily assessments. Cattle that reveal EW on the upper portion of the eye may have different temperaments that those that only reveal EW on the lower one-half of the eye. This type of research could be useful to provide producers with a quick and easy scoring system for temperament on breeding programs or to assess daily welfare conditions.

An additional area of future study could be focused on assessing EW as a tool for selection. Many more animals would need to be included in this type of study, and heritabilities should be calculated. Breed differences could also be studied more thoroughly as well as age and sex differences. The EW could be a very useful tool in aiding producers in selecting males for breeding and those for castrating at an early age. Because this method of temperament assessment is objective and reliable genetic progress could occur at a rapid rate if the heritability is moderate to high. Ideally, measures like EW would be recorded in a centralized database for the purposes of genetic evaluation and selection at the level of an industry-wide breeding program. Finally, EW selection could be beneficial for increasing product quality. Studies should be conducted on meat tenderness, marbling, and back fat qualities of animals that were scored based on an EW evaluation. In conclusion, the results of this preliminary study have provided strong evidence that percentage EW is a good indicator of temperament and that using a computer program to trace the EW area is reliable.

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