Assessment of Novel Semen Evaluation Technologies Between Two Breeds of Yearling Beef Bulls

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Introduction

- Current semen evaluation techniques include evaluation of motility and morphology, and although these are insightful, they are highly subjective and not always a definitive test of fertility.
- New fertility markers and technologies have been identified that provide an objective analysis of spermatozoa:
  - Flow cytometry – a method that aids in the detection of fertility markers such as reactive oxygen species (ROS) and the effects they have on characteristics of spermatozoa
    - Between-breed differences in motility and morphology have been observed, but differences in fertility markers have yet to be investigated.
  - The iSperm is a portable computerized semen analysis device that measures motility and concentration.

Objective

- During yearling bull breeding soundness exams (BSEs) evaluate:
  - Correlations of fertility markers
  - Breed comparisons
  - Validate use of the iSperm for on farm use

Experimental Procedures

- Ejaculates collected via electroejaculation on one of three consecutive days from Angus and Charolais yearling bulls (403 ± 11 d of age; n=46) as part of a BSE
- One veterinarian conducted all BSEs, and ejaculates were evaluated by one technician
- Each ejaculate were assessed with the iSperm for progressive and gross motility
- Ejaculates meeting minimum thresholds for passing a BSE underwent flow cytometry evaluation
- Flow cytometry assays included live/dead, acrosome and cell membrane integrity, mitochondrial energy potential, and oxidation status
- Correlations were assessed using Pearson's correlation coefficients in SAS
- The GLIMMIX procedure of SAS was used to determine breed differences
  - Experimental unit = bull
  - Main effect = bull breed
  - Random = collection date

Conclusion

- Technician and iSperm sperm motility data were positively correlated, offering producers an on-farm evaluation tool. Bull breed had little influence on sperm quality assessments, negative ROS status in sperm appears to impair sperm health and function.

Results

Sperm quality assessments on ejaculates from Angus and Charolais breeds of yearling bulls meeting BSE threshold requirements

	Least squares mean ± Standard error of mean		
FACTOR	Angus n=23	Charolais n=23	P-value of factor
Bull age, days	402.9 ± 2.36	403.3 ± 2.36	0.90
SEMEN CHARACTERISTICS			
Technician progressive motility[1] (%)	43.7 ± 1.69	47.39 ± 1.69	0.26
iSperm progressive motility[2] (%)	50.1 ± 2.26	47.8 ± 3.18	0.66
iSperm gross motility[2] (%)	71.6 ± 2.78	70.5 ± 2.78	0.82
Cells live and viable[3] (%)	42.3 ± 3.95	43.6 ± 3.95	0.83
Cells live with intact acrosome[4] (%)	41.5 ± 3.40	42.6 ± 3.40	0.83
Cells viable with positive ROS[5] (%)	29.1 ± 3.52	28.5 ± 3.52	0.92
Active mitochondrial potential[6] (%)	17.6 ± 3.35	31.1 ± 3.35	0.10

[1]Percentage of spermatozoa from ejaculate analyzed by a single technician for progressive motility; [2]Progressive and gross motility of each ejaculate were analyzed using the iSperm software and manufacturer recommendations; [3]Live and viable cells; [4]Live with intact cell membrane and acrosome; [5]Live with a positive ROS status; [6]Polarized active mitochondrial membranes

Pearson's correlation coefficients of technician progressive motility assessment and iSperm motility assessment

	iSperm progressive motility[2] (%)	iSperm gross motility[2] (%)
	r (P-value)	
Technician progressive motility[1] (%)	0.39 **(<0.001)**	0.30 **(<0.001)**

[1]Percentage of spermatozoa from ejaculate analyzed by a single technician for progressive motility; [2]Progressive and gross motility of each ejaculate were analyzed using the iSperm software and manufacturer recommendations

Pearson's correlation coefficients of sperm attributes from ejaculates meeting BSE threshold requirements in yearling bulls

	% Live Negative ROS Spermatozoa[1]	% Live Positive ROS Spermatozoa[2]
SEMEN CHARACTERISTICS	r (P-value)	
Primary abnormalities[3] (%)	0.28 (0.06)	-0.15 (0.33)
Secondary abnormalities[4] (%)	**0.33 (0.02)**	-0.23 (0.12)
iSperm progressive motility[5] (%)	-0.27 (0.10)	**0.53 (<0.001)**
Cells live with intact acrosome[6] (%)	-0.16 (0.29)	**0.92 (<0.001)**
Cells live with disrupted acrosome[7] (%)	**0.66 (<0.001)**	**-0.31 (0.04)**
Cells live and viable[8] (%)	-0.19 (0.22)	**0.94 (<0.001)**
Active mitochondrial potential[9] (%)	0.03 (0.84)	**0.58 (<0.001)**

[1]Percentage of spermatozoa from ejaculate live with a negative ROS status; [2]Live with a positive ROS status; [3]Exhibiting primary abnormalities; [4]Exhibiting secondary abnormalities; [5]Progressive motility of each ejaculate were analyzed using the iSperm software and manufacturer recommendations; [6]Intact cell membrane and acrosome; [7]Intact cell membrane and disrupted acrosome; [8]Live and viable ; [9]Polarized active mitochondrial membranes

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