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## The Effect of Method of Collection and Number of Sequential Ejaculates on Semen Characteristics of Beef Bulls

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## **The Effect of Method of Collection and Number of Sequential Ejaculates on Semen Characteristics of Beef Bulls**

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We would like to thank Kansas Artificial Breeding Services Unit's employees and customers for providing their data and insight to make this project possible.

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## The Effect of Method of Collection and Number of Sequential Ejaculates on Semen Characteristics of Beef Bulls

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### Abstract

As more genomic information becomes available for young beef bulls, age at time of semen collection has decreased. Factors affecting collection characteristics include collection method, electro-ejaculate or artificial vagina, and the number of ejaculates collected. The objective of this study was to determine the effect of managerial factors on collection characteristics. From 2008 to 2018, 11,642 individual ejaculates were analyzed by a single technician at the Kansas Artificial Breeding Service Unit. Bulls not receptive to the artificial vagina after 3 or 4 attempts were subject to electro-ejaculation. Collection characteristics were evaluated using multiple regression models; fixed effects included collection method and sequence of ejaculates collected/day and were evaluated for their impact on collection characteristics. Progressive motility before freezing was greater ( $P < 0.0001$ ) for bulls collected with electro-ejaculate compared to artificial vagina. Ejaculate volume for electro-ejaculate collections was greater ( $P < 0.0001$ ) than those collected with artificial vagina. The percent of spermatozoa with secondary abnormalities was greater ( $P < 0.05$ ) for bulls collected with electro-ejaculate compared to artificial vagina. Concentration of spermatozoa/mL was less ( $P < 0.0001$ ) for bulls collected with an electro-ejaculate ( $514 \times 10^6$ ) compared to artificial vagina ( $617 \times 10^6$ ). Total number of straws frozen/ejaculate were less ( $P < 0.001$ ) for bulls collected with electro-ejaculate (94) compared to artificial vagina (108). Bulls that were collected more than once/day produced a greater percentage of secondary spermatozoa abnormalities ( $P < 0.001$ ). As ejaculate number/day increased, the concentration of spermatozoa decreased ( $713, 580, 535, \text{ and } 434 \times 10^6/\text{mL}$ , respectively;  $P < 0.0001$ ), and the number of straws frozen/ejaculate decreased (123, 107, 93, and 82, respectively;  $P < 0.0001$ ). In conclusion, artificial vagina collections resulted in a higher number of straws frozen. The method of collection could cause a significant impact when collecting young high-demand bulls.

### Introduction

As age of bulls admitted to collection facilities decreases, many management challenges arise. These young bulls are often identified as genetically superior prior to puberty. This creates an issue for producers and bull studs to manage young bulls in such a way that semen collection can begin as soon as possible, without compromising quality. The

average age of bulls at the time of collection has decreased from 4.5 years to 1.5 years of age at most major bull studs (Hartstine, 2018). Artificial vagina is the preferred method of collection for ejaculates, however; inexperienced bulls are often more hesitant to mount and serve an artificial vagina. When bull studs are unable to collect bulls with an artificial vagina, they must use an electro-ejaculator. Electro-ejaculators are believed to have similar effectiveness as artificial vaginas, but this has not been recently investigated. To our knowledge, literature also lacks information on the total sequential ejaculates that can be collected/day by either method before impairing semen quality.

## Experimental Procedures

Data were provided from Kansas Artificial Breeding Services Unit and were collected from January 2008 to December 2018. A total of 11,642 ejaculates from 906 bulls were provided for analysis. Bulls were collected twice weekly on Mondays and Thursdays, with the preferred collection method, artificial vagina. Bulls at this facility not receptive to the mount steers or the artificial vagina after 3 or 4 attempts, were subject to electro-ejaculation to ensure ejaculates were collected.

Once an ejaculate was collected, a single technician at Kansas Artificial Breeding Services Unit was responsible for all pre-freeze and post-thaw semen analysis. Ejaculates were required to meet quality standards which included a pre-freeze progressive motility of greater than 50% and post-thaw progressive motility of greater than 30% at initial evaluation and two-hour evaluation. The ejaculates could not contain greater than 30% abnormal spermatozoa post-thaw to pass quality standards. All ejaculates that passed initial assessment were extended and frozen in half cubic centimeter straws. The descriptive information provided for each ejaculate was volume, concentration of spermatozoa/mL, progressive motility prior to freezing, progressive motility initially post-thaw, two-hour post-thaw progressive motility, primary and secondary sperm abnormalities, and straws frozen. Collection characteristics were evaluated using multiple regression models in Statistical Analysis System (SAS v. 9.4 (SAS Inst. Inc., Cary, NC)); fixed effects included collection method and sequence of ejaculates collected/day and were evaluated for their impact on collection characteristics.

## Results and Discussion

The age of bulls ranged from 10 months to 13 years, with a median age of 25 months. Average motility prior to freezing was 40%, the average volume was 4.6 mL, and the average units of straws frozen were 121. The average percentage of primary spermatozoa abnormalities was 40%, while the average for secondary spermatozoa abnormalities was 16%.

Table 1 displays the least square means for the effect of the collection method. Progressive motility before freezing was greater ( $P < 0.0001$ ) for bulls collected with electro-ejaculate (44%) compared to artificial vagina (43%). Ejaculate volume for electro-ejaculate (4.8 mL) collections was greater ( $P < 0.0001$ ) than those collected with artificial vagina (4.8 mL). Percent of spermatozoa with secondary abnormalities was greater ( $P < 0.05$ ) for bulls collected with electro-ejaculate (16%) compared to artificial vagina (15%). Concentration of spermatozoa/mL was less ( $P < 0.0001$ ) for bulls collected with an electro-ejaculate ( $514 \times 10^6$ ) compared to artificial vagina

( $617 \times 10^6$ ). Bulls that were collected with electro-ejaculate had a greater volume yet a lower concentration, resulting in a lower total number of straws of frozen/ejaculate ( $P < 0.001$ ). The method of collection did not have a significant impact on primary spermatozoa abnormalities, initial post-thaw motility, or two-hour post-thaw motility.

Bulls that were collected more than once/day had a decreasing percentage of secondary spermatozoa abnormalities, as ejaculate frequency increased (17, 16, 14, and 15%, respectively;  $P < 0.001$ ). As ejaculate number/day increased, the concentration of spermatozoa decreased ( $713, 580, 535, \text{ and } 434 \times 10^6/\text{mL}$ , respectively;  $P < 0.0001$ ; Figure 1). The number of straws frozen/ejaculate also decreased as ejaculate number/day increased (123, 107, 93, and 82, respectively;  $P < 0.0001$ ; Figure 2). Conversely, initial post-thaw motility increased as ejaculate frequency increased (36, 39, 40, and 40%, respectively;  $P < 0.0001$ ).

In conclusion, artificial vagina collections resulted in a higher number of straws frozen. Understanding the impacts of collection method on production could help producers better understand the difficulties of the collection process. While artificial vagina is the preferred collection method, bulls can still be collected with electro-ejaculate; this will result in fewer frozen straws of semen. Producers and bull studs should choose the collection method that best fits each bull and is most economically beneficial. Collecting more than one ejaculate/day will help increase straws frozen over time, and potentially have the greatest economic impact for producers. It should be noted that semen quality does decrease with increased ejaculates/day, and the optimum ejaculate/day may be individually dependent.

## Implications

Producers and collection facilities should work together to balance the collection method and number of ejaculates collected/day to maximize production while maintaining semen quality.

## Acknowledgments

We would like to thank Kansas Artificial Breeding Services Unit's employees and customers for providing their data and insight to make this project possible.

## References

Harstine, B.R., 2018. Invited Review: Focusing on bull management and puberty attainment in the genomic era. *The Professional Animal Scientist*, 34(6), pp. 523-532.

**Table 1. Effects of method of collection, artificial vagina or electro-ejaculate, on collection characteristics in beef bulls\***

	Number of ejaculates	Collection method		Standard error of the mean	<i>P</i> -value
		Least square means of artificial vagina	Least square means of electro-ejaculate		
Progressive motility prior to freezing, % <sup>1</sup>	11,642	43	44	0.32	< 0.0001
Volume, mL <sup>2</sup>	11,520	4.0	4.8	0.06	< 0.0001
Concentration, × 10 <sup>6</sup> /mL <sup>3</sup>	11,315	617	514	10.35	< 0.0001
Secondary abnormalities, % <sup>4</sup>	3,699	14.7	16.3	0.56	< 0.01
Straws frozen/ejaculate <sup>5</sup>	2409	108	94	4.13	< 0.001

\*From 2008 to 2018, individual ejaculates were analyzed by a single technician at the Kansas Artificial Breeding Service Unit, Manhattan, KS. Bulls that were not receptive to the artificial vagina after 3 or 4 attempts, were subject to electro-ejaculation.

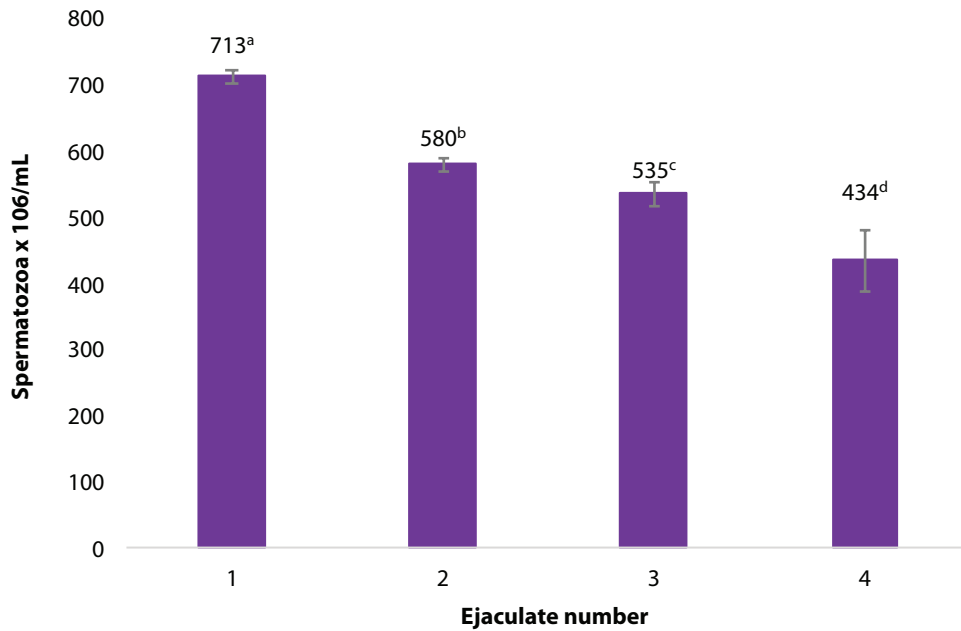
<sup>1</sup>Percent progressive motility prior to freezing was determined at time of collection. Based on the regression model other significant factors included: breed, sequential ejaculate number, age, Julian date of collection, and temperature humidity index 75 days before collection.

<sup>2</sup>Volume of semen collected/ejaculate. Based on the regression model other significant factors included: breed, sequential ejaculate number, age, and temperature humidity index 75 days before collection.

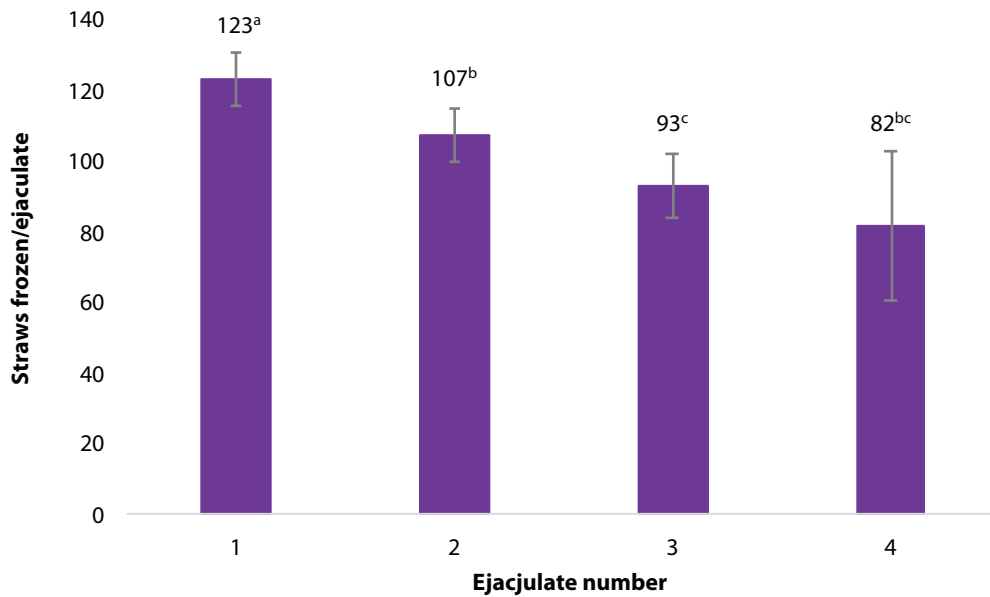
<sup>3</sup>Concentration of spermatozoa/mL in each ejaculate. Based on the regression model other significant factors included: sequential ejaculate number, age, and temperature humidity index 75 days before collection.

<sup>4</sup>Percentage of secondary abnormalities/ejaculate. Based on the regression model other significant factors included: sequential ejaculate number, age, and temperature humidity index 75 days before collection.

<sup>5</sup>Units of 0.5 cm<sup>3</sup> straws frozen/ejaculate. Based on the regression model other significant factors included: breed, sequential ejaculate number, age, and temperature humidity index 75 days before collection.



**Figure 1. Effect of sequential ejaculates on the concentration of spermatozoa in beef bulls.**  
<sup>a,b,c,d</sup>Values within a factor without a common superscript differ ( $P < 0.05$ ).



**Figure 2. Effect of sequential ejaculate collection on number of straws frozen/ejaculate in beef bulls.**  
<sup>a,b,c,d</sup>Values within a factor without a common superscript differ ( $P < 0.05$ ).