Title
Heparin-Binding Proteins as an Indicator of Bull Potency

Authors
Ax, Roy L.
McCauley, Tod C.
Dawson, George R.
et al.

Publication Date
2004-04-15

HEPARIN-BINDING PROTEINS AS AN INDICATOR OF BULL POTENCY

Roy L. Ax,[1,2] Tod C. McCauley[1,2], George R. Dawson[1], Cynthia Daley[3] and David Daley[3]

[1]Department of Animal Sciences, University of Arizona
1200 E. South Campus Dr., Tucson, AZ 85721-0038
[2]TMI Laboratories International, LLC,
850 N. Kolb Rd., Tucson, AZ 85710
[3]College of Agriculture, California State University
Chico, CA 95929-0310

INTRODUCTION

Over 20 years ago, research was underway to develop methods for in vitro fertilization utilizing bovine sperm and eggs. Freshly ejaculated sperm cannot fertilize an egg. Those sperm must reside in the female reproductive tract for 6-8 h and become diluted from seminal fluid. That process is called capacitation because it allows sperm to acquire the "capacity" to fertilize an egg. The final change sperm cells undergo after capacitation involves a morphological remodeling with release of enzymes packaged in the tip of the sperm head's acrosome. This irreversible remodeling is known as the acrosome reaction. All of these events had to be controlled in the lab to successfully fertilize eggs from cows.

Proteins produced in the seminal vesicles, prostate, and Cowper's glands convey the capacitating effects of heparin, a carbohydrate, to bull sperm. Those proteins are collectively referred to a heparin-binding proteins because they function as "docking' molecules to allow heparin to physically attach to the sperm, causing capacitation. Heparin per se is not found in the female reproductive tract. However, several other heparin-like carbohydrates do exist, and heparin mimics their normal biological action.

One specific heparin binding protein has been named fertility-associated antigen (FAA). For the past 13 years, research has focused specifically on FAA, its identity, the ability to detect it in semen, and field trials comparing fertility of bulls classified as FAA-positive or FAA-negative. Trials included multiple-sire pastures with or without parentage of calves being confirmed by DNA testing. Herds have utilized A.I. in some instances, and serving capacity was also evaluated one year before bulls were allocated to pastures.

Field Trials Comparing Bulls Categorized as FAA-Positive or FAA-Negative

Since 1992, field trials have been conducted in Texas, Nebraska and California to compare prolificacy of bulls that produced semen classified as FAA-positive or FAA-negative.

Multiple-sire pastures: Table 1 contains data from 7 consecutive years of field trials at King Ranch. When bulls were 14-19 mo. of age, FAA status was determined after they passed a breeding soundness exam. All pastures contained 8-16 bulls for 60d at a constant ratio of 1 bull per 25 cows. Overall, FAA-positive bulls were 19 percentage points more fertile than their FAA-negative herdmates. FAA was quantified in the Ax lab at the University of Arizona.

Serving capacity and FAA: The ability of a bull to breed cows can be estimated as "serving capacity." This is ordinarily evaluated by placing a group of virgin bulls with heifers that were synchronized to be in heat. Mounts with penetration are scored for each bull over a period of 20 min. Bulls are then ranked as "high" or "low" in that social setting.

FAA-positive bulls with high serving capacity impregnated 87% of cows exposed to them for a 60d breeding season. FAA positive bulls with low serving capacity only impregnated 69% of the exposed cows. Bulls with semen lacking FAA but with high serving capacity impregnated 78% of the cows pastured with them. Therefore, their libido was able to compensate for the absence of FAA, but they were inferior to herdmates with high serving capacity possessing seminal FAA (Table 2). FAA was measured in the University of Arizona Lab.

A.I. outcomes: With A.I., serving capacity is not an issue because cows are inseminated when they are in estrus. Holstein heifers and range beef cows were inseminated once with semen from mixed breeds of beef bulls designated as FAA-positive (n=18) or FAA-negative (n=7). Overall, there was a 16% higher fertility in females inseminated with FAA-positive semen (66% pregnancy rate) compared to FAA-negative semen (50% pregnancy rate, Table 3). The University of Arizona Lab analyzed semen for FAA content.

Efficiency of the cow herd: What does selection for FAA-positive bulls do for the cow herd? Research obtained from 1992 through 1998 at King Ranch indicated that the distribution of calves born during the calving season shifted to births occurring earlier (Table 4). In the nucleus herd, cows were initially bred only to FAA-positive bulls. Their replacement daughters were also only bred to FAA-positive bulls in subsequent generations. By 1998, 22% more calves were born in the first 20 days of the calving season from this FAA selection management practice (Table 4). Clearly, efficiency in the cow herd had improved.

DNA parentage of calves: In a collaboration with Drs. Dave and Cindy Daley and Harris Ranches, FAA status of bulls was determined using a newly developed chute-side cassette. Those bulls were in multiple-sire pastures with cows for a 60-day breeding season in 3 consecutive breeding years (2000, 2001, 2002). The trial was conducted to relate parentage of calves by DNA fingerprinting to growth and carcass traits of individual sires. Analysis of FAA status became a retrospective comparison to evaluate utility of the cassettes to analyze semen for FAA within 20 minutes.

Results from this study are being analyzed. Overall, 12 out of 62 total bulls were found to be FAA-negative. This was close to the incidence found in a population of 914 bulls screened in 6 states in April, 2003. In those bulls, 26% were FAA-negative using the same test cassette to quantify FAA in semen.

With the Harris Ranch bulls, complete DNA profiles were achieved with 47 of the 62 bulls. Overall, as bulls got older, they sired more calves per bull (1.1 as yearlings to 22.2 as 5-year old breeding bulls). Irrespective of age, FAA-positive bulls produced 5.9 more calves in the 3 years (1.9 calves/year) compared to FAA-negative herdmates. That translated into a 19% higher calf production for FAA-positive bulls for the 3-year duration of the trial (Table 5). There was clearly an age influence in terms of calf production in relation to FAA status of bulls. As yearlings and 5-year olds, FAA status did not factor into calf yield. However, between the ages of 2 and 4, each FAA-positive bull averaged 35.4 total calves, whereas his FAA-negative herdmates produced 27.3 total calves in that period of time. Therefore, the FAA-negative bulls were 77% as prolific as their FAA-positive contemporaries based upon those numbers.

From ages 1 through 3 years, a higher proportion of FAA-negative bulls were more likely to not sire any calves compared to FAA-positive bulls. In other words, sterility of a bull in a given year corresponded to FAA status of bulls 3 years old or younger.

CONCLUSION

FAA is a good thing! Fertility data support that regardless of years, pasture, or breed, the FAA positive bulls resulted in a higher percentage of cows pregnant compared to FAA negative herdmates. A conservative estimate places pregnancy rates 15% higher in heifers or cows bred to FAA positive bulls.

The calving season should also tighten up if daughters are retained from FAA positive bulls and are bred to known FAA positive bulls. In tern, daughters in subsequent generations need to be bred to FAA positive bulls, and that practice should continue.

FAA testing only takes 20 minutes and is based upon visible detection of a reddish-purple line on a plastic cassette that contains all the necessary chemicals to detect FAA if it is in a semen sample. The projected payback per cow in a herd from testing for FAA in bulls will be 16 to -25 fold if net profit per calf is $50.00. Obviously, if profit per cow exceeds $50.00, then the value of testing for FAA increases substantially.

For more information, pricing, and to order testing kits, contact:
ReproTec, Inc.
(520) 888-0401 (520)888-0297 (FAX)
www.reprotec.us

Suggested Readings

1. Ax, R.L., H.E. Hawkins, S.K. DeNise, T.R. Holm, H.M. Zhang, J.N. Oyarzo and M.E. Bellin. 2002. New Developments in Managing the Bull. In: Factors Affecting Calf Crop. M.J. Fields, R.S. Sand, J.V. Yelich (eds.), CRC Press, Boca Raton, Chap. 21, pp. 287-296.

2. Bellin, M.E., H.E. Hawkins and R.L. Ax. 1994. Fertility of Range Beef Bulls Grouped According to Presence or Absence of Heparin-Binding Proteins in Sperm Membranes and Seminal Fluid. *J Anim Sci* 72: 2441-2448.

3. Bellin, M.E., H.E. Hawkins, J.N. Oyarzo, R.J. Vanderboom and R.L. Ax. 1996. Monoclonal Antibody Detection of Heparin-Binding Proteins on Sperm Corresponds to Increased Fertility of Bulls. *J Anim Sci* 74: 173-182.

4. Bellin, M.E., J.N. Oyarzo, H.E. Hawkins, H. Zhang, R.G. Smith, D.W. Forrest, L.R. Sprott and R.L. Ax. 1998. Fertility-Associated Antigen on Bull Sperm Indicates Fertility Potential. *J Anim Sci* 76: 2032-2039.

5. McCauley, T.C., H.M. Zhang, M.E. Bellin and R.L. Ax. 1999. Purification and Characterization of Fertility-Associated Antigen (FAA) in Bovine Seminal Fluid. *Mol Reprod Dev* 54: 145-153.

6. McCauley, T.C., G.R. Dawson, J.N. Oyarzo, J. McVicker, S.H.F. Marks and R.L. Ax. 2004. Development and Validation of a Lateral-flow Cassette for Fertility Diagnostics in Bulls. *In Vitro Diagnostic Technology*, In Press.

7. Miller, D.J. and R.L. Ax. 1990. Carbohydrates and Fertilization in Animals. *Mol Reprod Dev* 26:184-198.

8. Sprott, L.R., M.D. Harris, D.W. Forrest, et al. 2000. Artificial Insemination Outcomes in Beef Females Using Bovine Sperm with a Detectable Fertility-Associated Antigen. *J Anim Sci* 78: 795-798.

Table 1. Relationship of Sperm FAA Status to Fertility of Bulls Used for Natural Service.[a]

Sperm	No. Bulls	No. Cows Bred	No. Pregnant	Pregnant (%)
FAA Positive	242	5,317	4,497	85
FAA Negative	192	3,881	2,572	66
Total	434	9,198	7,069	19% Difference

[a]Adapted from Bellin et al., 1994;1996;1998.

Table 2. Impact of Serving Capacity and FAA Status on Fertility of Bulls.[a]

				20 d Interval			Fertility	
FAA	SC	Cows	BCS	1-20	21-40	41-60	%	Diff
Pos	High	270	4.2	50	19	18	87	-
Neg	High	143	4.8	45	13	20	78	9
Pos	Low	238	4.3	29	13	27	69	19

[a]Adapted from Bellin et al., 1998.

Table 3. Relationship between Sperm FAA Status and Actual First Service Pregnancy Outcomes From Artificial Insemination (AI). Adapted from Sprott et al., 2000.

Sperm	No. Bulls	No. Cows Bred	No. Pregnant	Pregnant (%)
FAA Positive	18	764	501	65.6
FAA Negative	7	386	192	49.7
Total	25	1,150	693	60.2

Table 4. Percentage of Calves Born During the Calving Season.[a]

Day of Calving Season	Births (%)		
	1991[b] 223 Head	1995 262 Head	1998 489 Head
1-30	61.4	66.0	83.8
1-45	83.4	85.1	93.4

[a]Daughters of FAA-positive bulls were retained and bred to FAA-positive bulls for the period of time indicated.
[b]Year Prior to FAA Testing.

Table 5. Calf production per year[a] in relation to age or FAA status of bulls used for multiple-sire breedings.[b]

Age (yrs)					FAA Status	
1	2	3	4	5	Positive	Negative
1.1	7.4	12.7	15.0	22.2	11.5	9.6

[a]Parentage determined by DNA fingerprinting
[b]1 bull per 25 cows for a 60-d breeding season