
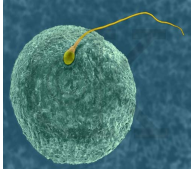


Insemination Related Factors Affecting Fertilization In Cattle

J.C. Dalton, S. Nadir,
M. Noftlinger, and R.G. Saacke

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Factors important to fertilization

- Semen quality and number of sperm
- Accessory sperm
- Bull effect and time of AI
- Semen handling
- Fertility associated antigen?

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Semen quality

“Compensable” seminal traits:

- Viability or morphology traits impairing sperm transport and ovum penetration; fertilization does not occur
- Severely misshapen sperm in an otherwise normal semen sample

Reduced fertility may be overcome or minimized by increasing sperm numbers

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(Saacke et al., 1994)

Semen quality

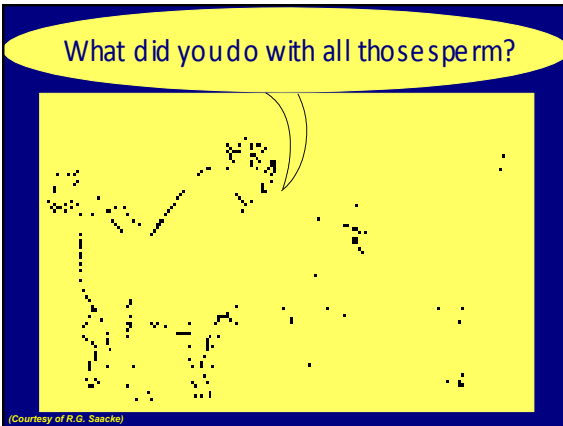
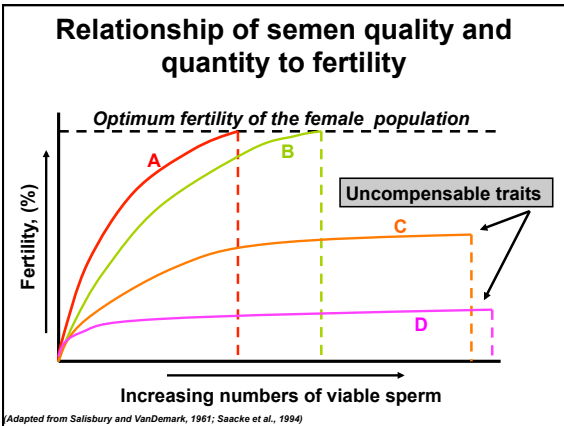
“Uncompensable” seminal traits:

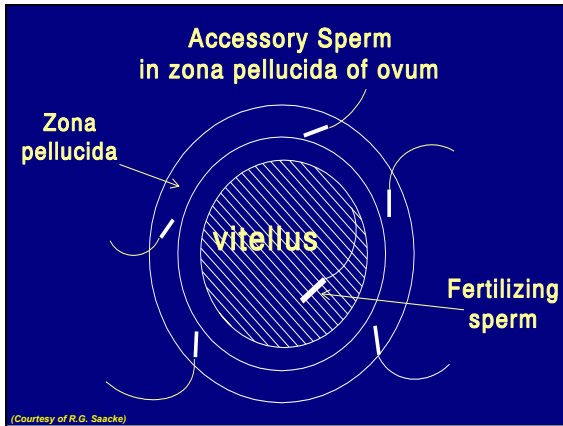
- Incompetence of the fertilizing sperm; completion of fertilization and maintenance of the embryo does not occur
 - Damaged DNA in otherwise normally shaped head of fertilizing sperm?

Reduced fertility regardless of sperm dosage

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(Saacke et al., 1994)





What is the significance of accessory sperm?

Increased accessory sperm number is associated with increased fertilization rate and embryo quality.

(DeJarnette et al., 1992; Nadir et al., 1993; Photo courtesy of R.G. Saacke)

Partial summary of efforts to increase accessory sperm

Positive effect:

- Specific bull
- Time of AI

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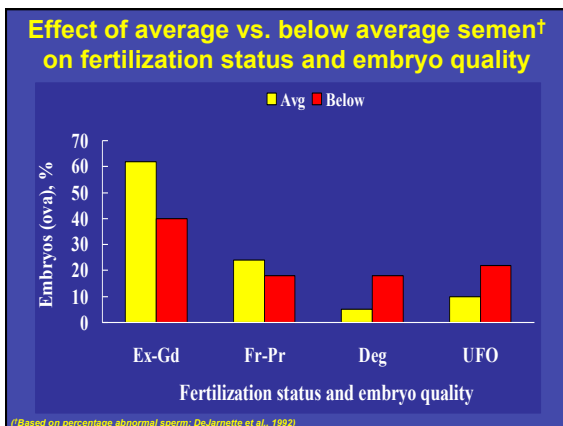
(Nadir et al., 1993; Dalton et al., 2001; Photo courtesy of R.G. Saacke)

Is there a bull effect on accessory sperm number?

Bull	n	Median	Mean ± SD
A	25	40	53 ± 61
B	37	8	15 ± 23
C	16	13	36 ± 65
D	20	2	11 ± 16

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(Adapted from Nadir et al., 1993)



Relationship between sperm morphology and chromatin structure

- Normal shaped sperm in abnormal ejaculates exhibit vulnerability to DNA denaturation.
- Denatured DNA does not always appear in abnormal shaped sperm.

Bull A: Day + 27 post scrotal insult

(Acevedo et al., 2002; Courtesy of R.G. Saacke)

Morphology: Tip of the iceberg?

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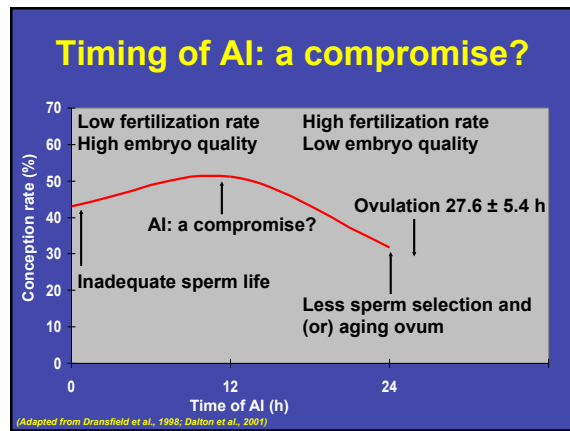
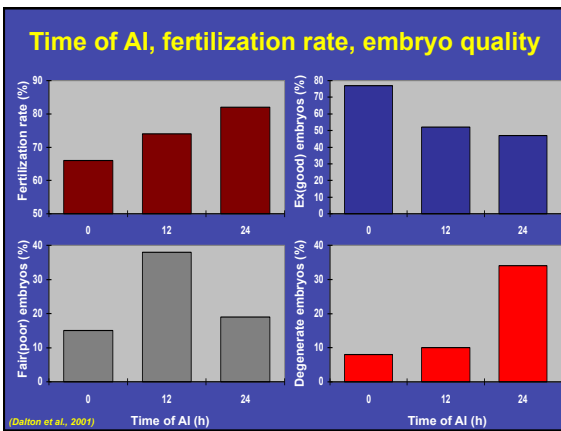
(GHSW Media Network)

How does time of AI affect fertilization rate, embryo quality and accessory sperm?

Time of AI	N	Accessory sperm		Fertilization rate, %
		Mean ± SD	Median	
0 h	39	9 ± 23	1	66
12 h	39	21 ± 46	2	74
24 h	39	33 ± 53	4	82

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(Dalton et al., 2001)



Sperm transport

Rapid phase

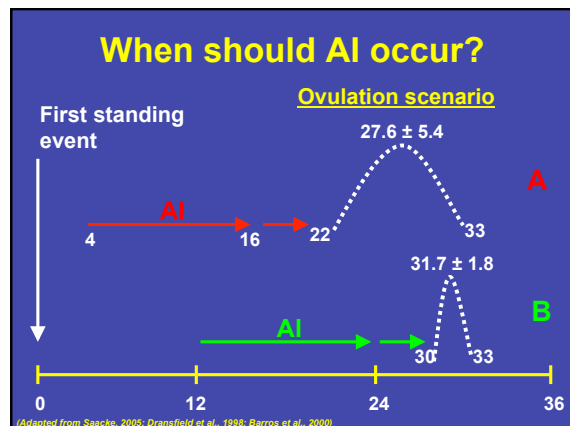
- Minutes
- Oviduct: Majority of sperm are dead

Sustained phase

- Hours (~6 to 12 h in cattle)
- Oviduct: Increased percentage
 - Motility
 - Morphologically normal sperm

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(Overstreet et al., 1978; Hunter and Wilmut, 1983; Wilmut and Hunter, 1984; Hawk, 1987)

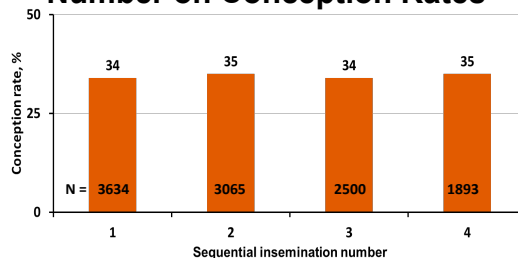


How many straws should be thawed at one time?



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Effect of Sequential Insemination Number on Conception Rates



(Adapted from DeJarnette et al., 2002)

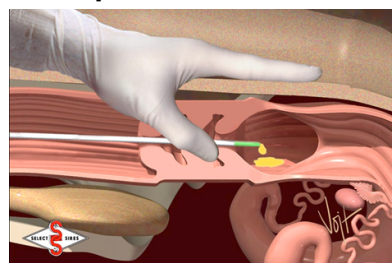
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How many straws should be thawed at one time?

- No more than can be used in 10 - 15 min.
- “Know your comfort zone.”
- Do not allow straws to touch when thawing.
- Use multiple thaw baths.
- Time, temperature, hygiene, skill.

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Inseminator skill: Deposition of semen



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How does deposition of semen into the cervix affect fertility?

- 10% decrease in fertility when compared to deposition in the uterine body.
- Cervical deposition of semen occurs in 20% of attempted uterine body depositions.

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(MacPherson, 1968; Williams et al., 1988; Peters et al., 1984)

Does fertility-associated antigen on sperm collected from Nelore (*Bos indicus*) bulls affect fertility at first-service timed AI?



(Dalton et al., 2012)

Fertility associated antigen

- During ejaculation, the seminal vesicles, prostate, and Cowper's glands secrete heparin-binding proteins (HBP) which coat the sperm.
- Bulls with sperm that exhibited a 31-kDa molecular weight HBP, called fertility-associated antigen (FAA), were 7 to 9 percentage points more fertile following AI than bulls producing sperm lacking FAA.

(Miller et al., 1990; Spratt et al., 2006)

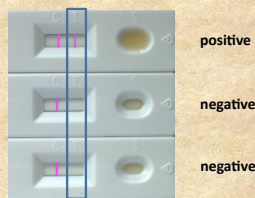
Materials and Methods

- Ejaculates from Nelore bulls (n = 49) were collected by artificial vagina.
- Immediately following semen collection, 2.0 mL of semen was transferred to a lateral flow cassette which facilitated rapid on-site determination of FAA in semen.

(ReproTest, ReproTec Inc., Tucson, AZ, USA)

Materials and Methods

- Presence of FAA in the sample results in a visible colored band at the test position.



(ReproTest, ReproTec Inc., Tucson, AZ, USA)

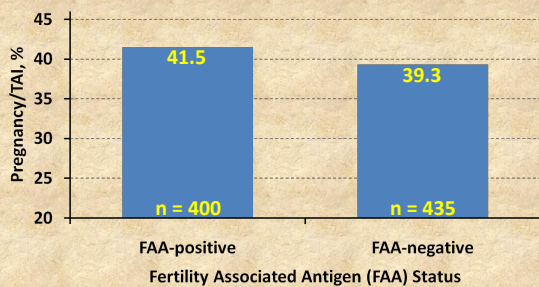
Materials and Methods

Six Nelore bulls:

- FAA status:
 - negative: n = 3
 - positive: n = 3
- Semen characteristics equal to or greater than:
 - 70% morphologically normal sperm
 - 60% estimated progressive motility before cryopreservation
- Extended semen was packaged and cryopreserved in 0.25-mL straws (30x10⁶ sperm).

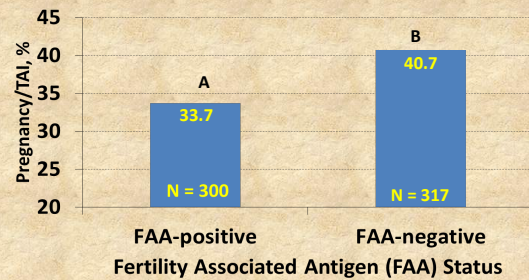
(Dalton et al., 2012)

Fertility (Pregnancy/TAI):Cows



(Dalton et al., 2012)

Fertility (Pregnancy/TAI):Heifers



(Dalton et al., 2012)

(A,B differ P = 0.04)

Conclusions

- There was no effect of FAA status on fertility at first-service TAI in lactating cows.
- Fertility, as measured by P/TAI, was different between FAA-positive and FAA-negative bulls (33.7% vs. 40.7%, respectively).

[Dalton et al., 2012]

Take home messages

- **Compensable** seminal traits: Ability of inseminated sperm to initiate fertilization?
 - Reduced fertility may be overcome or minimized by increasing sperm numbers.
- **Uncompensable** seminal traits: Competence of the fertilizing sperm to complete fertilization and sustain early embryonic development?
 - Reduced fertility regardless of sperm dosage.

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Take home messages

To reduce the risk of **uncompensable** seminal traits:

- Use semen from AI Studs where morphology is a routine part of the evaluation.
- Use multiple, proven high fertility bulls.
- Screen all natural service bulls with a complete breeding soundness evaluation, including sperm morphology.

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Take home messages

- Time of AI: Close enough to ovulation to maximize sperm access to the ovum, but not too late to have an aging ovum awaiting sperm arrival.
- Proper semen handling is critical to the success of AI.
- Presence of FAA on spermatozoal membranes does not guarantee higher fertility.

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Thank you.

