



Euroopa Maaelu Arengu
Põllumajandusfond:
Euroopa investeeringud
maapiirkondadesse

Practical Aspects of the ET Programs

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Bova-Tech Ltd.



Embryo Transplants

www.bova-tech.com

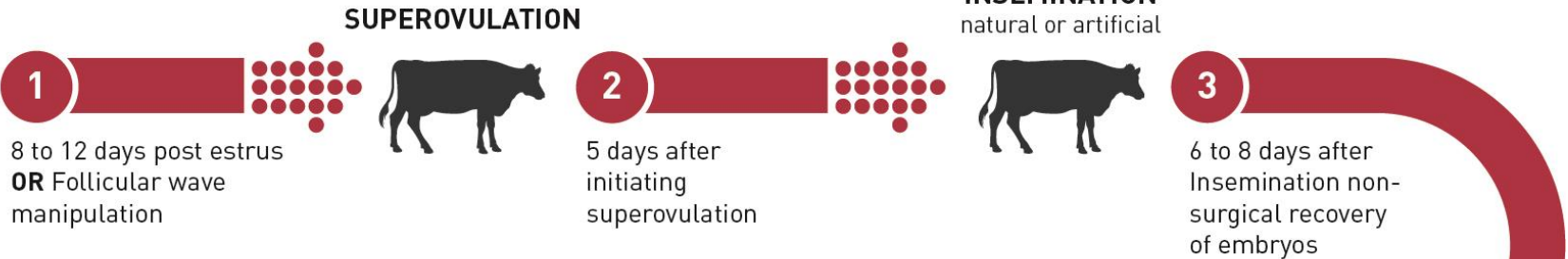


Introduction

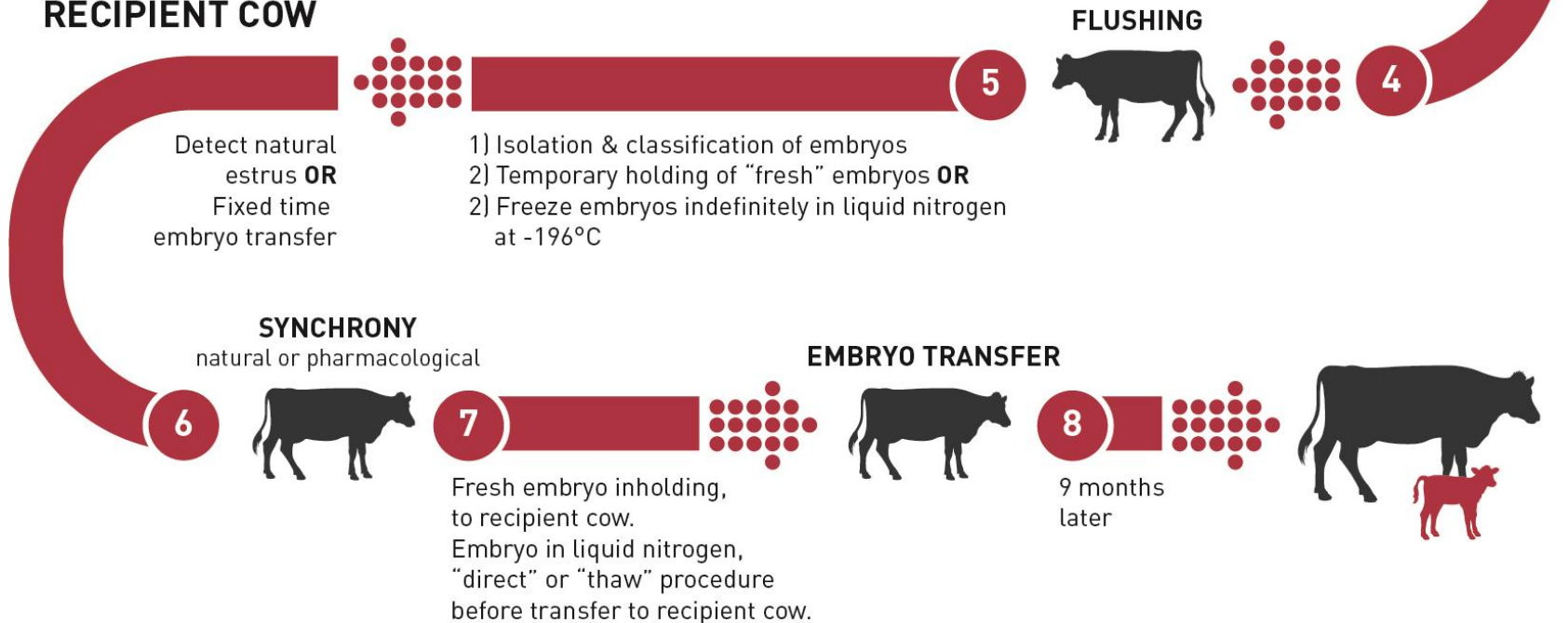
- To know how to raise cattle.
Production system in place (return on investment)
Direction
- Embryo Transfer is an expensive procedure, adding approximately \$300 for each calf born. Not including the cost of the semen.
- Understand that ET is a complicated procedure
Multifactorial and high difficulty level.
- ET is only to be performed by trained and licensed veterinarians in Canada.

What is ET

DONOR COW



RECIPIENT COW



Introduction cont..

- The impact that embryo transfer has on the livestock industry.

Dairy
Beef



Genomics



Tilda 530'15



Missy

Benefits of Embryo Transfer

- Traditionally, cows produce only one calf per year. ET allows the production of many offspring within a year from a single cow.
- ET can increase the genetic potential of a herd in a relatively short period of time.
- ET can increase several production traits in dairy and beef herds.
- Optimize use of expensive semen. Injured animals. Young heifers.
- ET allows other producers to take advantage of superior genetics because frozen embryos can be shipped almost anywhere.
- Disease free

What to expect?

- **GENETIC GAIN**

- **Donors**

- - 33 % rule
- 6.5 embryos per flush

- **Recipients**

- Pregnancy rates fresh embryos 55 – 60 %
- Pregnancy rates frozen embryos 50 – 55 %

Factors influencing ET programs

- Breed



Effect of cattle breed on superovulation

Beef^a	No. supered	Total ova	Good embryos	% good
Beefmaster	80	13.8	7.8	57
Brangus	1290	12.0	6.4	53
Brahma	144	8.9	5.6	63
Simmental	141	10.5	6.5	62
Dairy^b				
Holstein	6787	11.6	6.2	53
Ayrshire	332	14.8	7.4	50
Brown Sw.	84	10.1	5.5	54
Jersey	332	14.3	7.3	51

(^aLooney, 1986; ^bSauvé, 2003)

Factors influencing ET programs

- Parity & Age
 - Heifers
 - 1st calvers
 - Mature cows

Effect of age on superovulatory responses of 633 reproductively-healthy Holsteins

Age group	No. animals	Mean no. ova	Mean no. good embryos
Heifers	28	6.1	3.8
2 – 6 years	308	10.4	6.7
7 – 10 years	224	10.6	6.9
11 – 14 years	64	9.7	5.3
>15 years	9	5.6	2.6

(Hasler, et al., 1983)

Factors influencing ET programs cont...

- Lactational status
 - Dairy (Days in milk)
 - Beef cows (Postpartum to 1st flush)
- Reproductive history
 - Calving intervals
 - Calving history (normal vs complicated)

Factors influencing ET programs cont...

- Management
 - Nutrition
 - Body condition
 - Mineral supplementation
 - Post flushing care
 - Constant ultrasound
- Handling
 - Facilities
 - Keep stress level low



Superovulation

- Gonadotropin
- Dosage
- Synch. method



Superovulation protocol set-ups

1

Day 0*	1	2	3	4	10
FSH (AM & PM)	FSH (AM & PM)	FSH (AM & PM)	FSH (AM & PM)	Heat (AM)	Flush
		PG (AM & PM)	Heat (PM or D4)		

(* Day 8-13 of cycle)

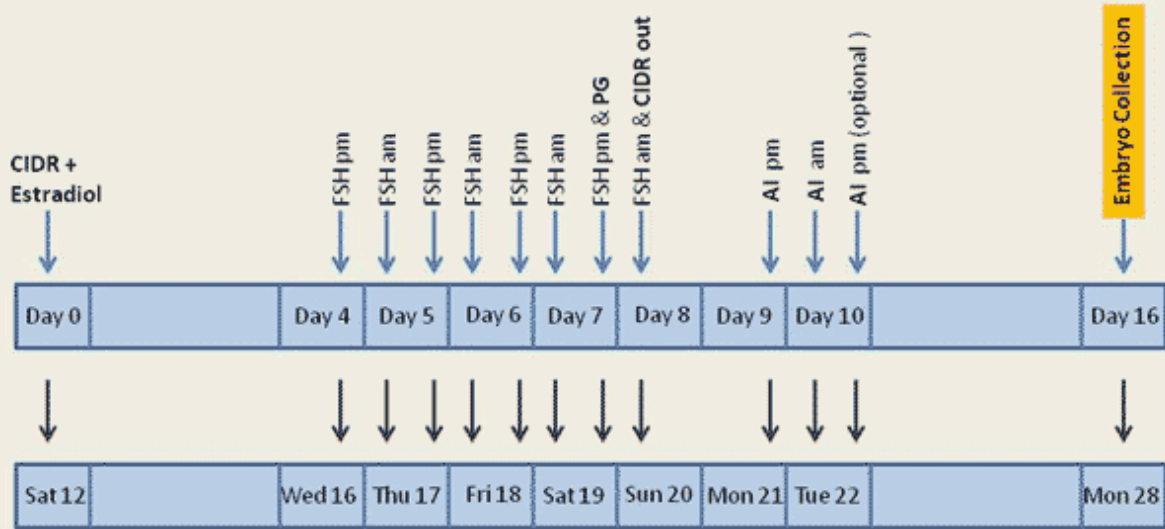
2

Day 0	4	5	6	7	9	16
CIDR	FSH (AM & PM)	FSH (AM & PM)	FSH (AM & PM)	FSH (AM & PM)	Heat	Flush
EB + P4				PG (AM & PM)		
				CIDR X (PM)		

3

Day 0	2	4	5	6	7	8	9	16
CIDR	GnRH (PM)	FSH (PM)	FSH (AM & PM)	FSH (AM & PM)	FSH (AM & PM)	FSH (AM)	Heat	Flush
					PG (PM)	PG (AM)	GnRH	
						CIDR X (AM)		

Typical Super-ovulation Protocol for Donor Cows



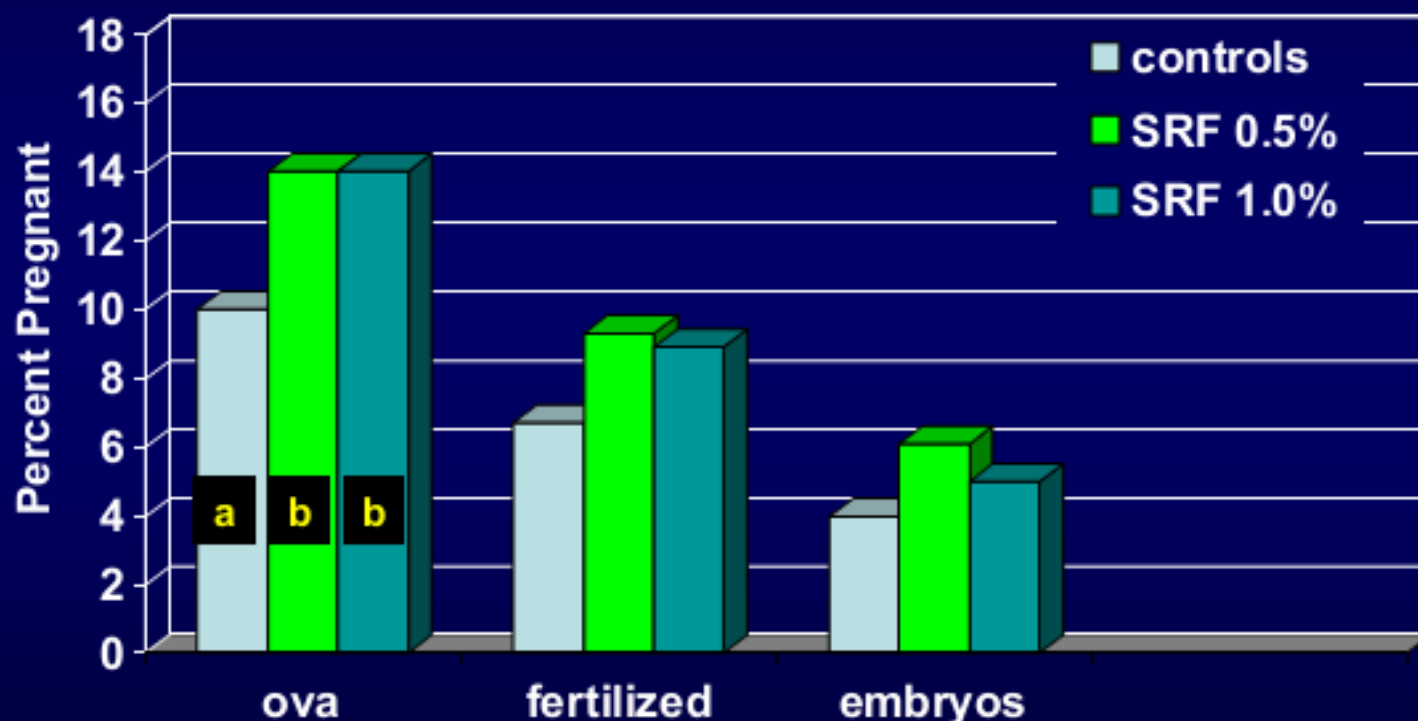
Example of days

Synch protocol at BTL

Donor cows lined up for shots at BTL



Superovulation of beef cattle with two injections of Folltropin® FSH dissolved in a slow release formula (SRF) diluent

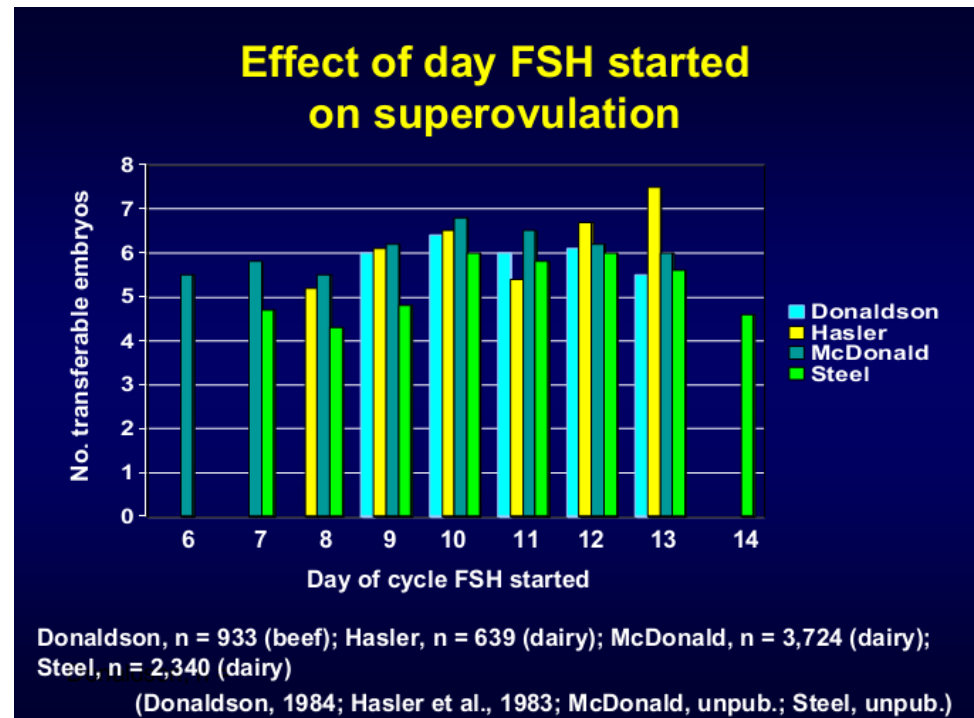


^{ab}P<0.05, 29 animals per group

(Bo et al., in Press)

Superovulation Protocol Alternatives

- Ovarian size
- # of Cls
- # Embryo-ova recovered
- Dosage adjustments
- Additional drugs
 - PMSG
 - GnRH
 - LH
- Natural heats



Superovulation Protocol Alternatives

- Breeding and semen
 - Timing of breeding

Effect of AI interval post estrus on fertilization and the number of good embryos recovered from superovulated cows

AI Treatment ^a	CL (Mean)	% Ova fertilized	% Transferable embryos (Mean)
12 h	10.0	72 ^b	61 ^b (4.7)
24 h	12.8	79 ^b	66 ^b (6.7)
36 h	13.0	54 ^b	30 ^c (3.0)
48 h	8.1	19 ^c	1.7 ^d (0.2)
12,24,36 & 48 h	10.0	77 ^b	61 ^b (3.7)

^a20 million sperm/straw, 2 straws per AI, 12 donors per group
^{b,c,d}p<0.05

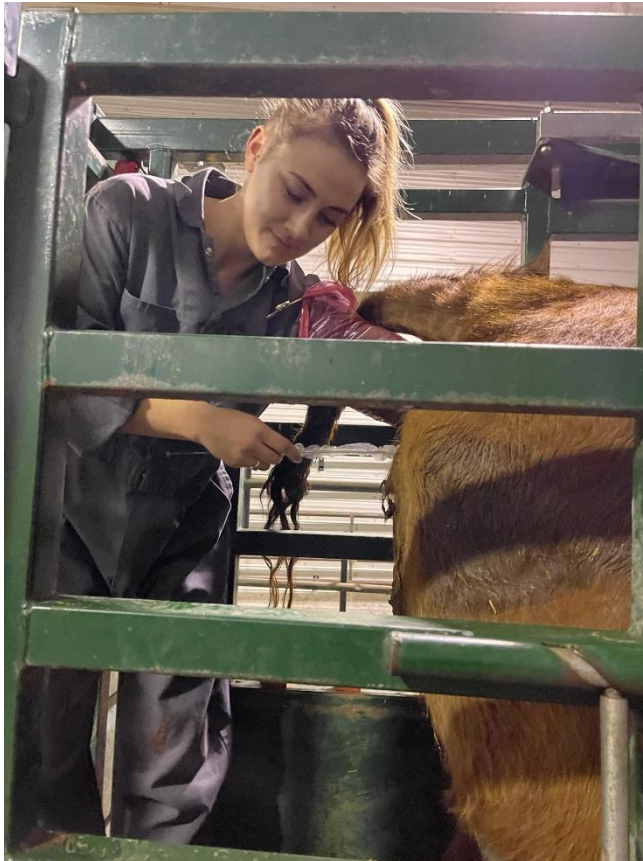
(Schiewe, et al., 1987)

Check Semen before breeding donors



Superovulation Protocol Alternatives

- Breeding and semen
 - Check semen



Effect of semen quality on fertilization rate and embryo quality in superovulated cattle (227 bulls used in 742 donors)

Semen Quality*	% Fertilized (n = 9,732)	% Excellent Embryos (n = 4,035)
Excellent	82.1 ^a	61.2 ^a
Good	67.7 ^b	55.7 ^b
Fair	58.3 ^c	53.9 ^c
Poor	51.8 ^d	33.7 ^d

abcdP<0.002

(Stroud and Hasler, 2006)

(*Semen quality combined the following factors: concentration, % motile, RFM, direction of movement and morphology)

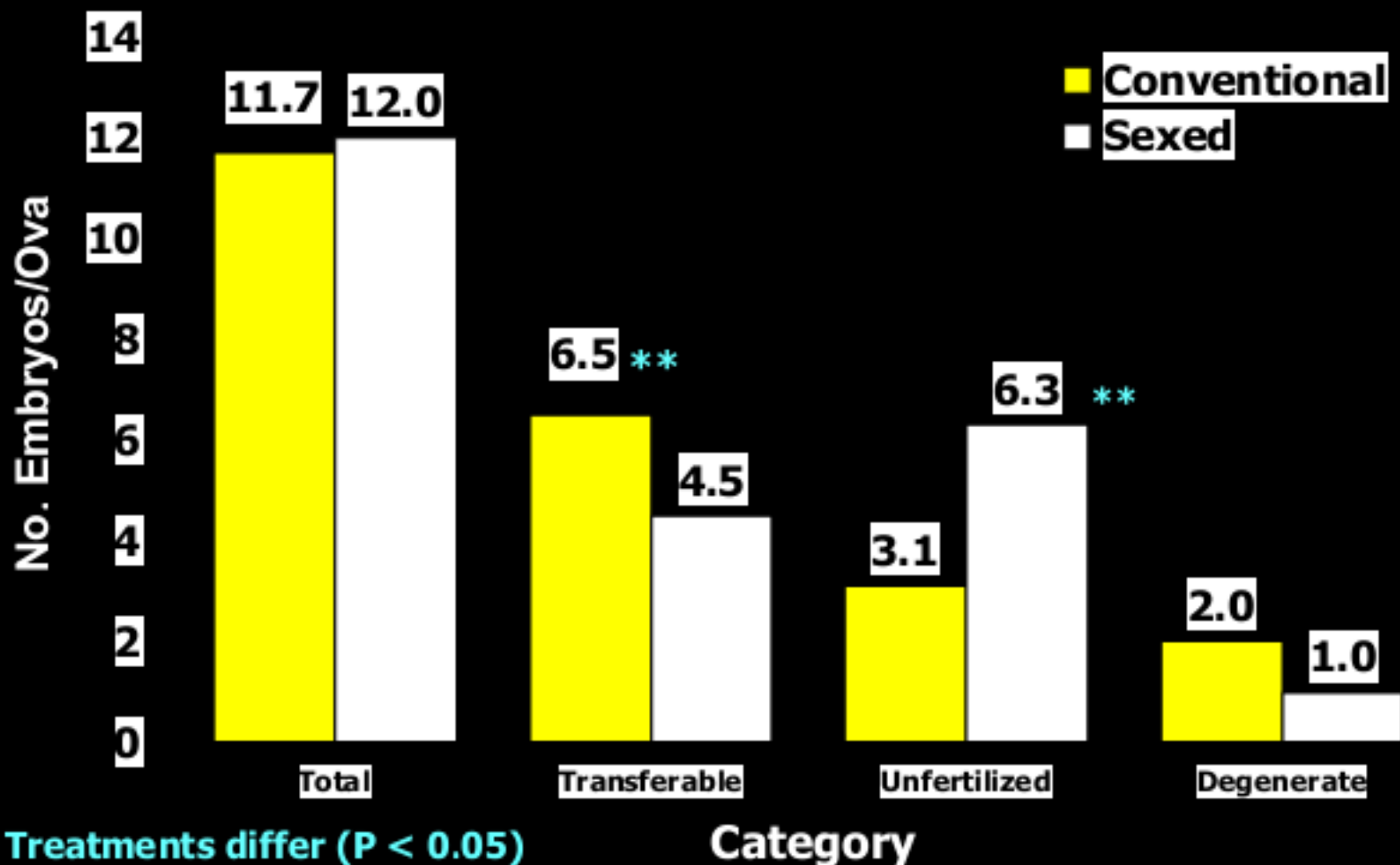
Comparison of semen quality when shipped to ET center directly from commercial AI unit compared to semen provided by breeder client

Semen quality at thawing	Source of semen	
	Direct from AI center	Provided by client
No. of shipments	1,278	981
Poor quality semen (%)	22 (1.7 ^a)	66 (6.7 ^b)
Semen with no motility (%)	0 (0 ^a)	24 (2.4 ^b)

^{a,b}Percentages in the same row differ significantly (P<0.001)

(Stroud and Hasler, 2006)

Embryo Production of Superovulated Angus Cows Inseminated with Sexed Semen



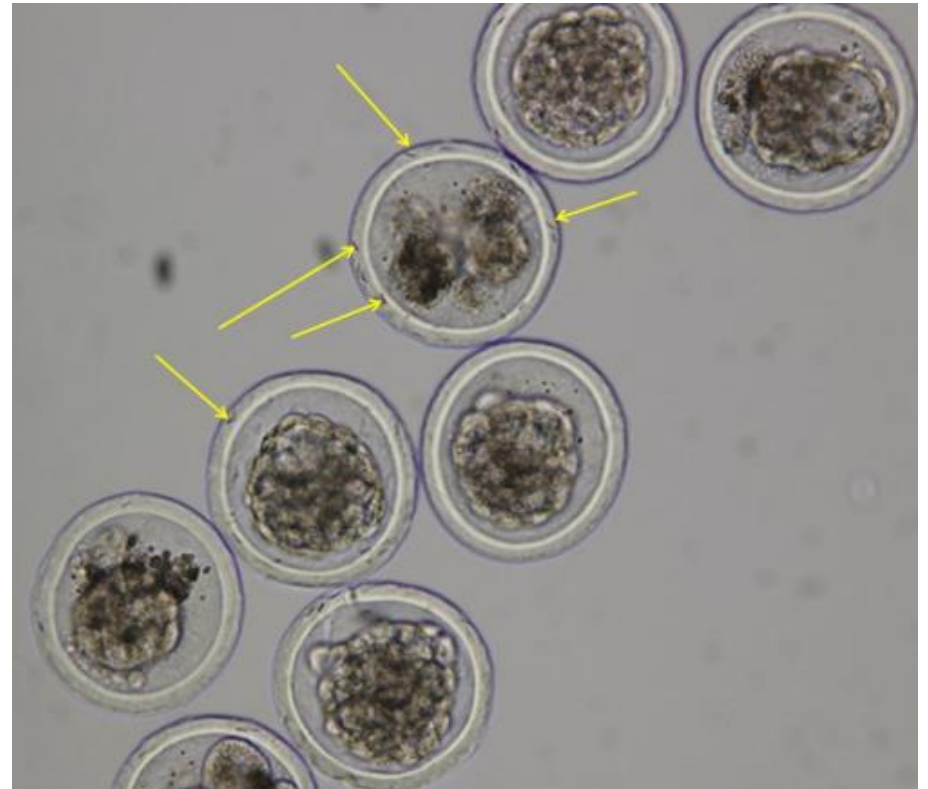
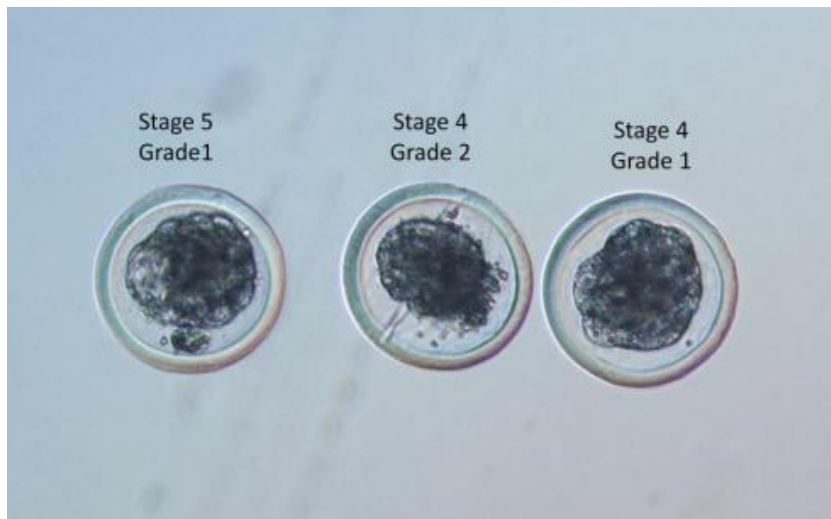
** Treatments differ ($P < 0.05$)

(32 cows per treatment inseminated at onset of heat and 12 (2X) & 24 h post. Control = 15M; Sexed = 2M)

(Lamb et al., 2007)

Embryo Factors

- Stage & grade



Embryo Factors

- **Fresh vs. Frozen**

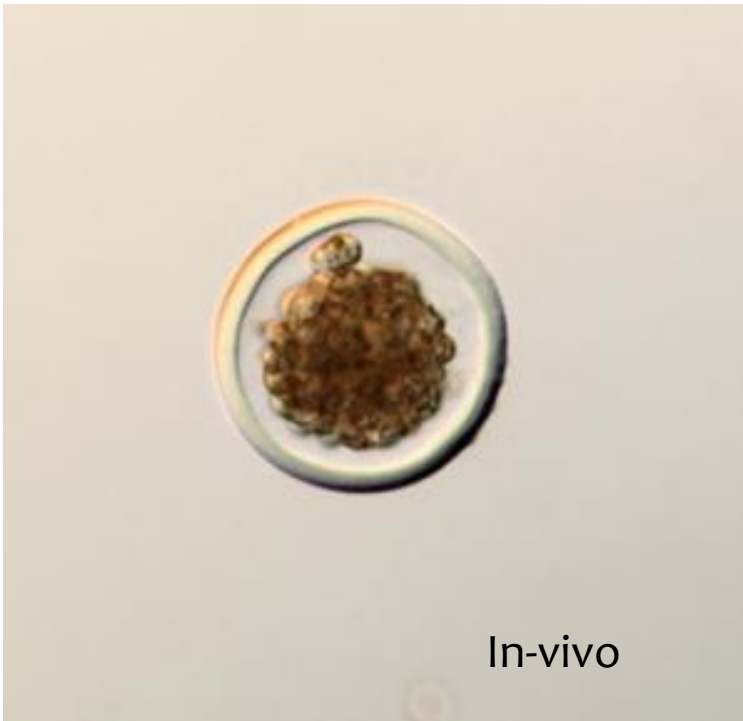
- **Fresh 60-65%**
- **Frozen 50-55%**



In small groups of well managed recipients is not difficult to achieve higher pregnancy rates that can be around 75-80%

Embryo Factors

- Biopsy, split, IVF



Embryo Factors

- Matting
- Size of the response
 - Overstimulated
- Type of semen
 - Conventional frozen
 - Sexed
 - Reversed sorted
- Length and conditions of storage

Recipient Factors

- Breed



Conception rates following embryo transfer in anestrus vs. cycling recipient beef cows

Cow status ¹	No. received CIDR	No. estrus detected (%)	No. received embryo (%) ²	No. Pregnant (%) ²
Anestrus	321	161 (50.1) ^a	140 (87.0)	56 (40.0) ^a
Cycling	376	338 (89.9) ^b	311 (92.0)	206 (66.2) ^b

¹Status determined by palpation per rectum and ultrasound at time of CIDR insertion; ²Percentages based on previous column.
a,bP<0.001

(Stroud and Hasler, 2006)

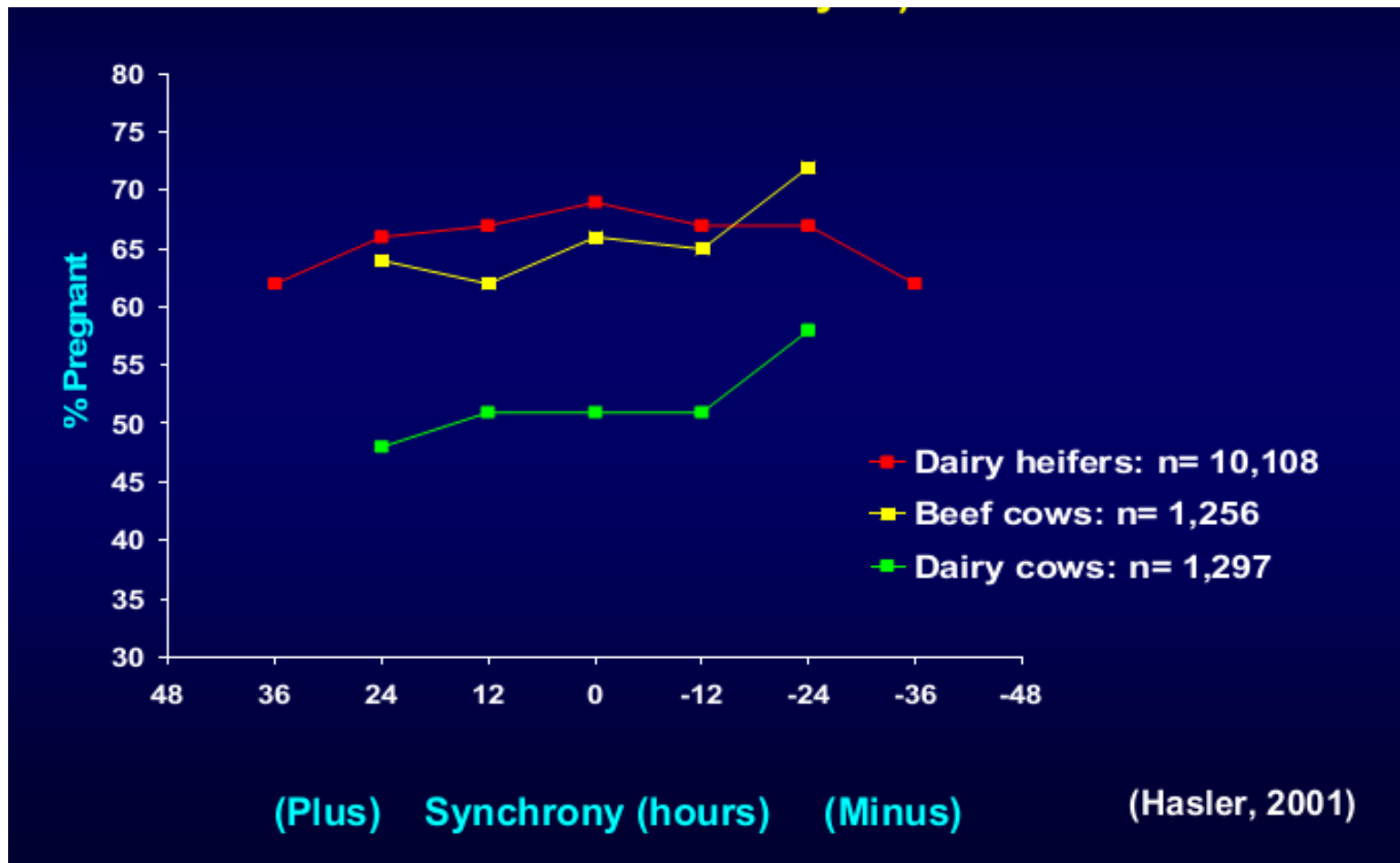
Recipient Factors

- Heifer
- 1st calvers
- Mature cows



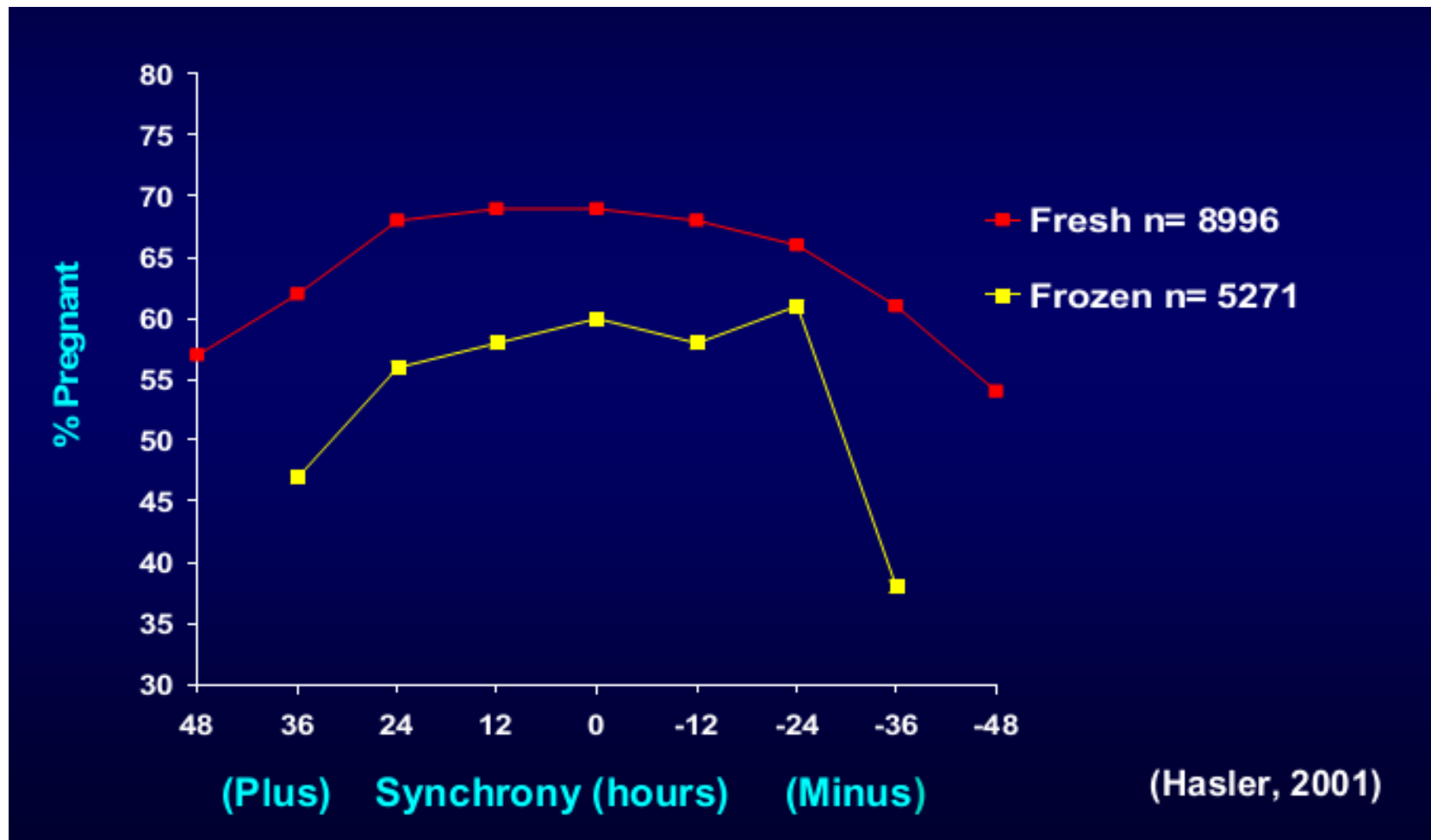
Recipient Factors

- Estrus synchrony with the donor
 - Breed & parity



Recipient Factors

- Estrus synchrony with the donor
 - Fresh vs. frozen embryos

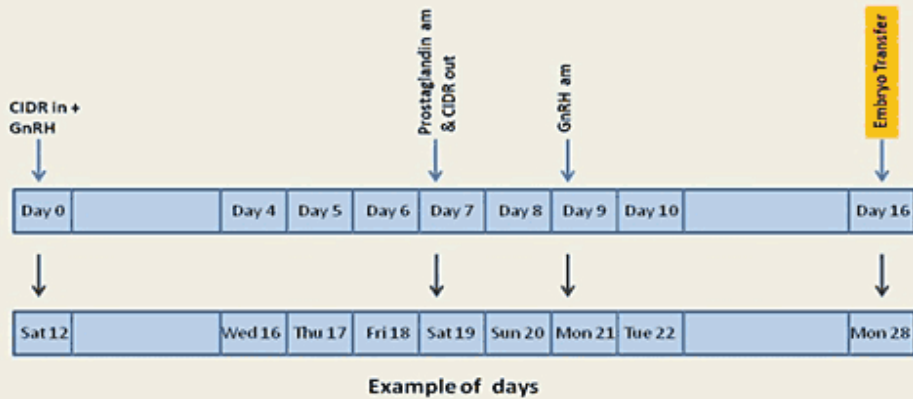


Recipient Factors

- Estrus induction
 - Synch vs. natural heats
 - CIDR GnRH
 - PG protocols

Recipient Factors

Typical Synchronization Protocol For Recipients



Recipient Factors

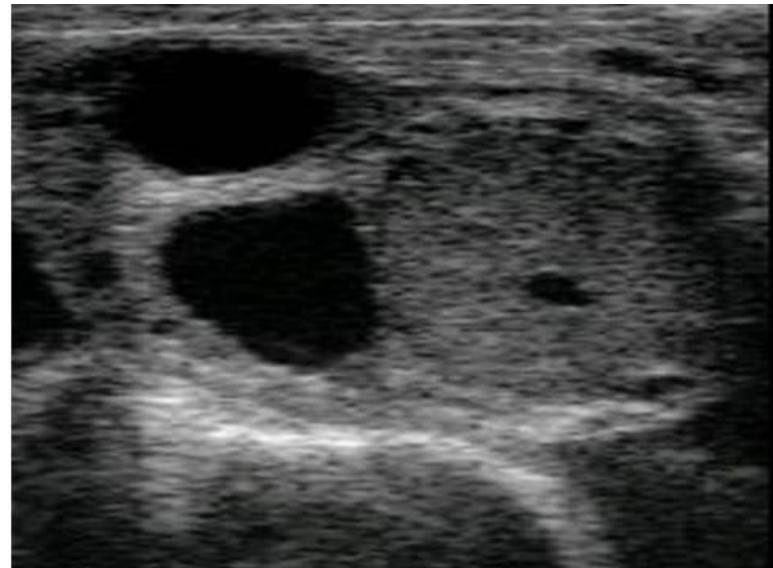
- Nutrition & Management
 - Body condition
 - Minerals
 - Vaccinations programs
 - Housing
- Climate & season
- Reproduction history

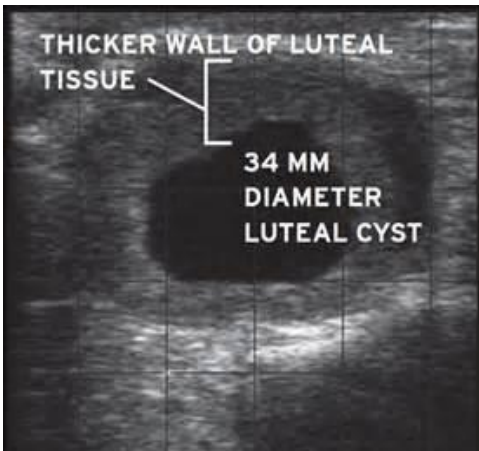
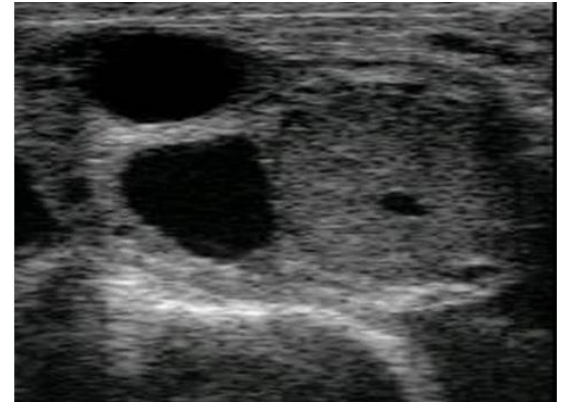
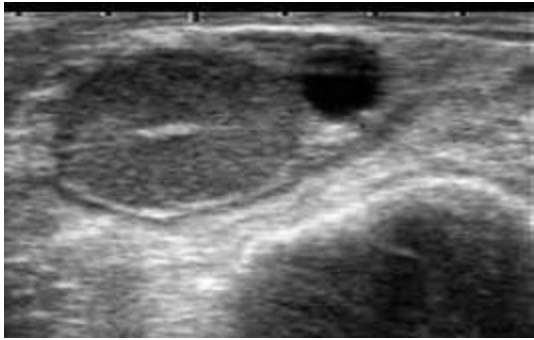
- **Recipients one of the most important factors and the one with less attention. “Take home message”**

Recipient Factors

Implanting embryos

- Corpus Luteum
 - Side
 - Size
 - Crown vs. no crown
 - Cavity

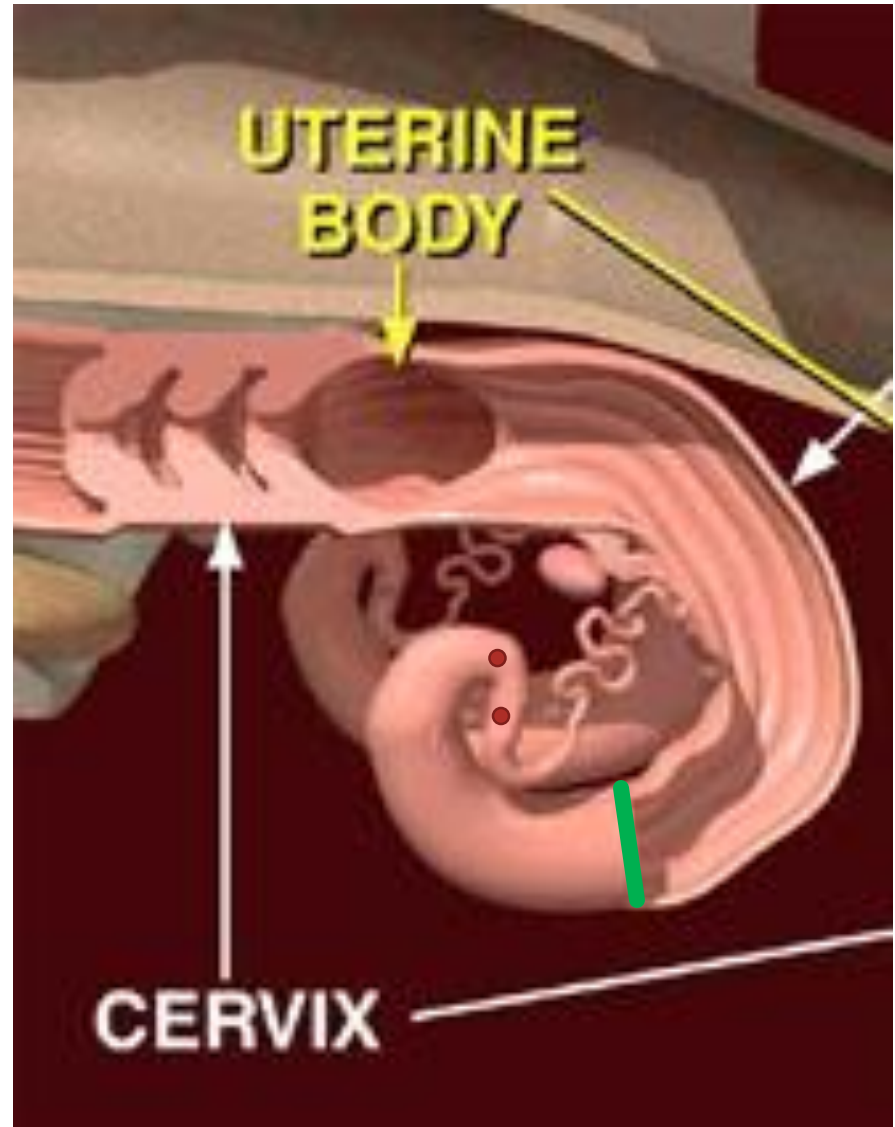




Recipient Factors

Implanting embryos

- Site of transfer



“The practitioner effect”

- Takes time to develop the skills for collection and implant techniques.
- Different ways to collect and implant embryos among practitioners.
 - Equipment
 - Embryo evaluation
 - Embryo handling, processing & thawing (Conv vs IVF)

“Luck”

Probabilities of success resulting from the transfer of four embryos if the true conception rate is 50%

No. Pregnant	Average frequency	Frequency %
0/4	1/16	6
1/4	1/4	25
2/4	3/8	38
3/4	1/4	25
4/4	1/16	6

Drugs & hormones used in attempts to improve pregnancy rates following AI and/or ET

- Progesterone
- LH
- hCG
- eCG (PMSG)
- GnRH
- Clenbuterol (smooth muscle relaxant)
- Ibuprofen (anti-inflammatory & anti-PG synthesis)
- Banamine (Flunixin Meglumine) (anti-inflammatory and anti-PG release)
- PG Receptor Competitive Antagonist (AL8810 or 'Embryo Armor')
- Growth Hormone (Posilac)
- Estradiol

Key to Success

If ET is conducted with reasonably Acceptable parameters involving Practitioner competence, media, embryo quality, hygiene, etc., almost certainly the Biggest factors affecting success are:

DONOR QUALITY
RECIPIENT QUALITY
MANAGEMENT



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Location:

Our office and embryo storage center is located in Airdrie, Alberta, Canada, only 10 minutes north of the Calgary International Airport, and only 5 minutes from Alta Genetics Inc., the biggest bovine semen collection center in Western Canada. This location facilitates the ability to ship embryos domestically or internationally.

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Location:



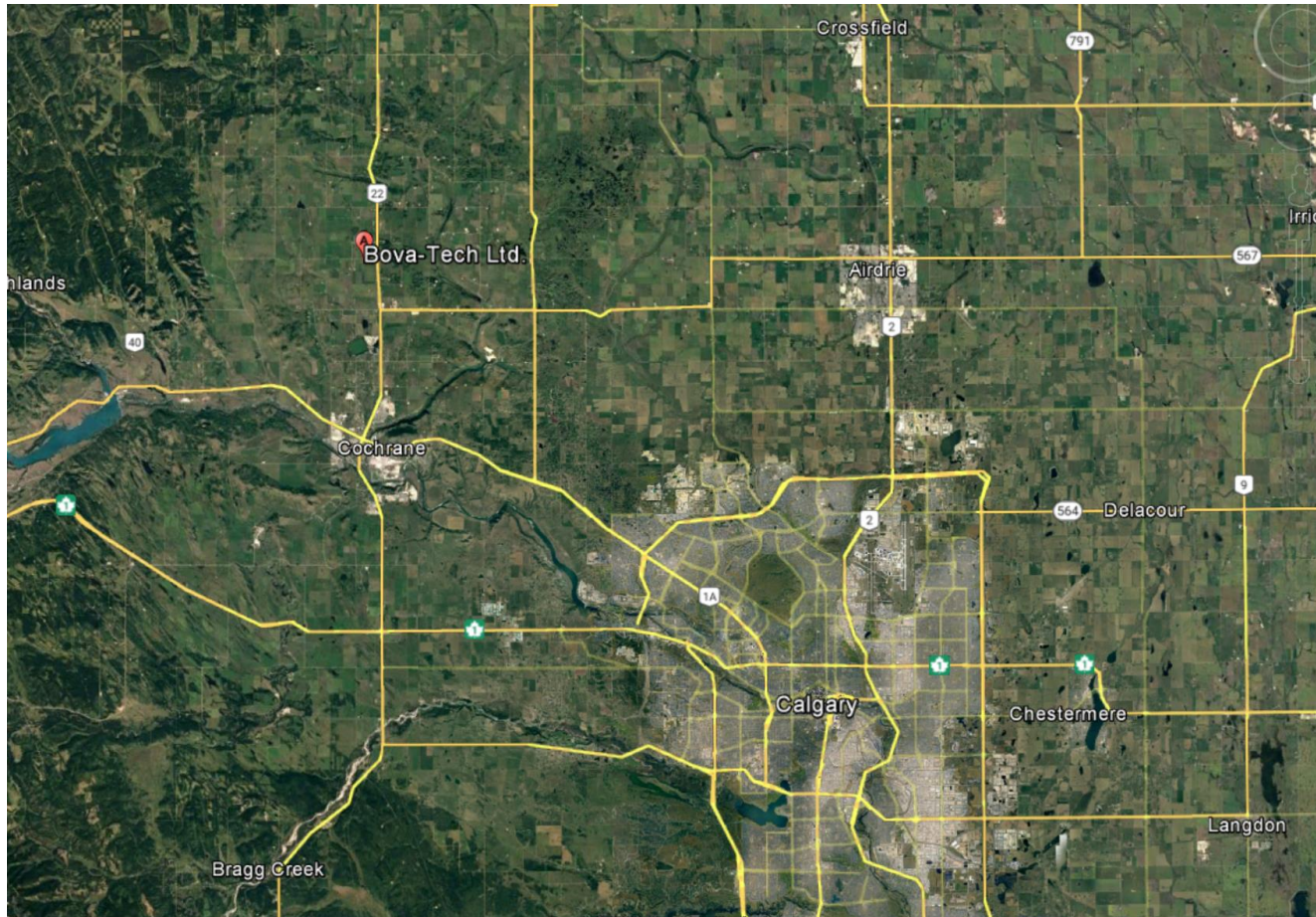
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Location:



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Location:



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Facilities



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Affiliations:

- Canadian Embryo Transfer association (CETA) Certified Practitioner
- International Embryo transfer Society (IETS)
- Canadian Food Inspection Agency (CFIA) Accredited facility for embryo exports.
- * Canadian Veterinary Medical Association.



<http://www.ceta.ca/>



<https://www.iets.org/>

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Embryo Transplants

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Services:

- In-clinic and on-farm services
- Embryo collection, freezing and transplants
- In-vitro embryo production
- Donor care facility
- Recipients programs
- Export certified
- International marketing



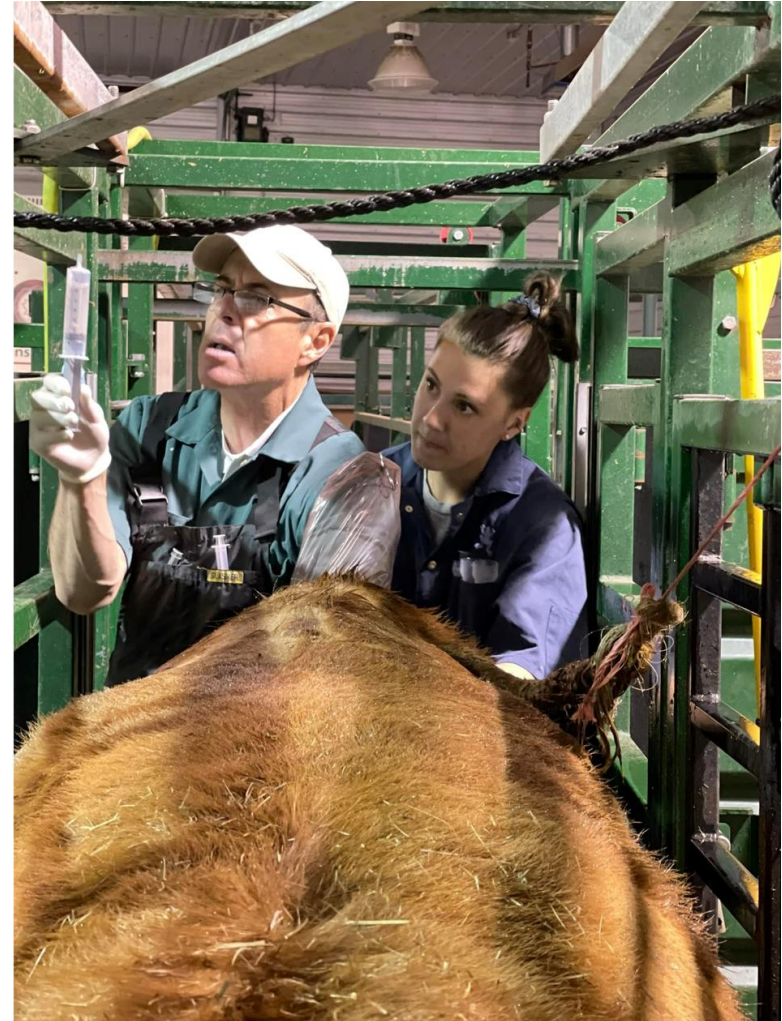
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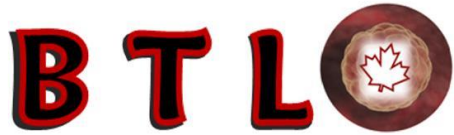
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In-clinic services



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Embryo Transplants

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On-farm services



Embryo collection, freezing and transplants



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In-vitro embryo production



IVF can occur as often
as every

two weeks

IVF can begin as young as

**seven
months**

One donor could
produce over

50

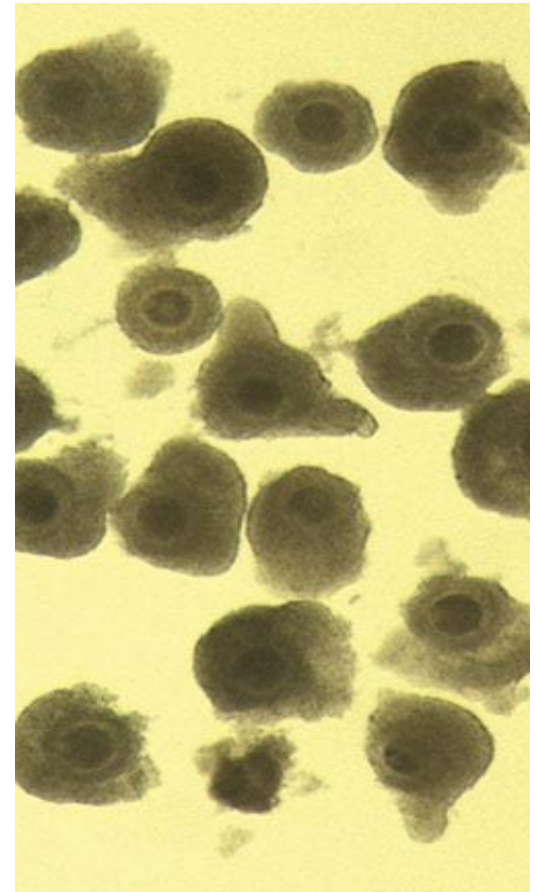
offspring each year

IVF embryos have shown

10-12%

increase in conception rates over
conventional semen

In-vitro embryo production



Oocytes (eggs)

In-vitro embryo production



Oocyte collection



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Recipients programs





Ian, this are the only recipients left for the Missy embryos, which one you want the yellow or the brown ones.

Read 2018-07-14

Brown all the way

Lo! what's with the camels?

International embryo marketing

Donor Cows



Angus



Red Angus



Charolais



Horned Hereford

Other breeds:

- Simmental
- Limousin
- Shorthorn
- Speckle Park
- Wagyu
- Gelbvieh
- Brahman
- Club calf
- Holstein
- Jersey

Embryo Export

- Thousands of embryos exported every year from Canada.
- Government programs
- Private sector
- Embryo sources:
 - Available Inventory
 - Specific breeding lines

Embryo Export

- Relatively straightforward process but require time.
- Embryos need to be produced and processed according the IETS guidelines.
- Export health certificate
- Certificate of embryo recovery and freezing
- Import permit
- insurance

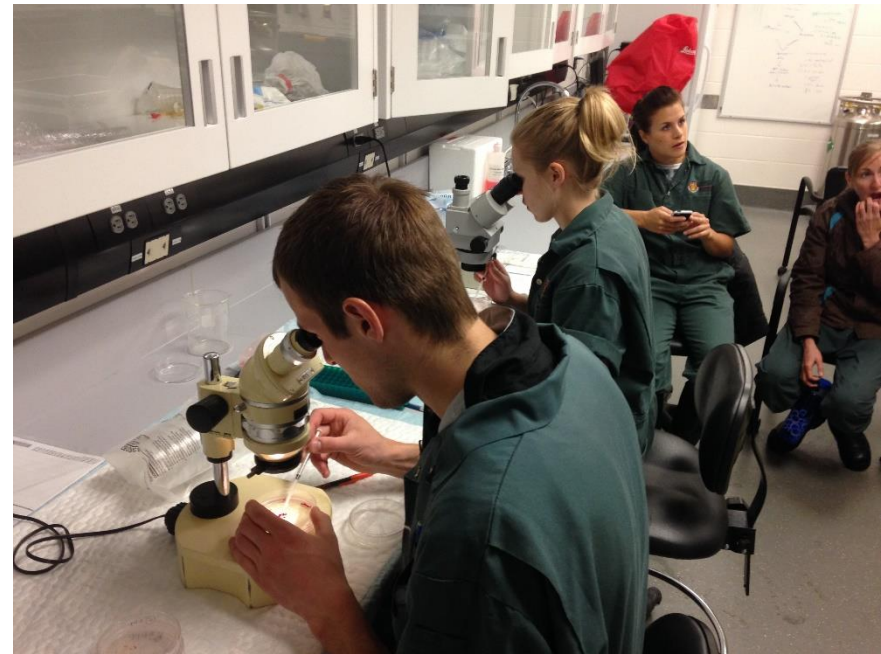
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Embryo Transplants

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Bnff Alberta Canada



Lake Louise Alberta Canada

<https://www.bing.com/videos/search?q=Amit+y+embryo+center+by+ben+wilson&&view=detail&mid=4F9ED777A54620AAFC0E4F9ED777A54620AAFC0E&&FORM=VRDGAR>