Transferring Embryos

Clinical factors affecting pregnancy success

Recipient cows

• Selection
• Synchronization
• Embryo Transfer

Recipient Selection

• Selection criteria are very diverse

• Every situation is unique
  – Criteria should be established based on current needs
Recipient Selection

• There are a few essential criteria
  – Healthy, disease free recipients
  – Capable of giving birth to a calf from a different breed/mating
  – Can raise a calf better than 50% of herdmates

• Repeat breeder dairy cows

Recipient Selection

• In Beef Industry
  – Recips should be fertile cows
    • Some inherently more fertile than others
    • A lot is invested in the production of the embryo, give it the best chance possible

Recipient Selection

• In Beef Industry
  – Recips should be productive cows
    • Good milk production to feed a calf
      – Not true for dairy industry
    • Maintains her body condition for future reproductive success
Recipient Selection

• Recip cows should have shown evidence of a good standing heat
  – Aids in timing of transfer
  – Good expression of heat often results in quality CL (not always)

Recipient Selection

• Uterus should be healthy
  – Small, moderate tone
  – Do not use recipient with overly large, doughy uterus
  – Do not use recipient cow with abnormal vaginal or uterine discharge

Recipient Selection

• Additional thoughts
  – Recips are the carriers of a valuable calf
  – Uterus is site of development
    • Gene expression dictated by uterine environment?
Recipient Selection

– Recipient cow is providing nutrition for calf
  • Should have good maternal instincts
  • Should have ample milk to feed calf
– A poor recipient will produce a poor calf, regardless of that calf’s pedigree/genetics!
  • Some U.S. producers do not understand this concept…

CL Identification

• Ultrasound and/or palpation
  – Both?
    • Ultrasound more accurate
    • Sometimes scan is poor (reverberations, tissue folds, etc)

CL Identification

– Accurate identification of ovarian structures
– Accurate location of ovarian structures
  • Embryo must be placed in the ipsilateral uterine horn.
CL Qualification

• Quality of CL?
  – Current system at NCROC
    • Determine location of CL
    • Evaluate amount of visible luteal tissue
    • Evaluate texture of the CL

– Score based on these two criteria
  – R1 – good quality CL on Right ovary
  – L1 – good quality CL on Left ovary
  – R2 – Poor quality CL on Right ovary
  – L2 – Poor quality CL on Left ovary

– Very subjective and not precise
  • Better than nothing

CL Qualification

• Working on a new system
  – CL blood flow is better indicator of circulating progesterone than luteal tissue volume
  – CL blood flow measured by doppler
  – Requires US machine with doppler capability
    • Extra expense
CL Qualification

- Requires skilled US technician
  - Ovary must be free of tissue folds
  - Must hold probe/ovary very still for short time
  - Must not constrict ovarian artery during scan

- Classification system?

Embryo Selection

- Embryo Stage
  - Typically stage 4-7 (range of 3-8)
    - 4 = morula
    - 5 = early blastocyst
    - 6 = blastocyst
    - 7 = expanded blastocyst
    - 8 = hatched blastocyst

Embryo Selection

- Embryo Quality
  - 1 = Excellent
  - 2 = Fair
  - 3 = Poor
  - 4 = Degenerated
Transfer

• Embryo should be transferred into the uterine horn ipsilateral to the CL
• Placement can be shallow or deep

Transfer

• Shallow – superficial to the greater curvature of the uterus
• Deep – deep to the greater curvature of the uterus
• Deep transfer results in greater percentage of pregnancies
  – If there are no complications during transfer
  – Greatest benefit with poorer quality embryos

Transfer

• Procedure should involve putting the uterus over the transfer gun
  – Do not push the transfer gun in the uterus
• Key to a successful transfer is placement as close to the tip of the uterine horn as possible, without any irritation to the uterus
Transfer

- Irritation and damage to endometrium results in secretion of prostaglandin
  - Prostaglandin binds to the zona pelucida of the ovary, which inhibits hatching
  - No hatching → no interferon tau → no maternal recognition of pregnancy → CL lysis → return to estrus

Transfer

- Flunixin meglumine (cyclooxygenase inhibitor) can be beneficial
  - Must be administered prior to transfer
  - Causes significant uterine relaxation (increase difficulty of transfer)

Fresh Transfer

- Fresh
  - Often recips are set up with donor for fresh transfer
    - Identify qualified recipients before loading embryos
    - May need to freeze some embryos if the donor was highly productive
**Fresh Transfer**
- Identify eligible recipients
  - CL quality and location
- Load embryos in clear straws with holding media
- Apply epidural
  - 4.0-5.0ml of 2% lidocaine
- Load embryo into transfer gun
- Transfer

**Frozen Transfer**
- Direct Transfer
  - Apply epidural
  - Remove embryo from cane
    - Be quick and be careful
    - Duration above, and temperature reached above -130°C will cause cell damage to embryos (glass transition temperature of water)

**Frozen Transfer**
- Air thaw for 8-10 seconds
  - Gives plastic of straw a chance to warm up
- Water bath of 20⁰-30⁰ C (not to exceed 37⁰C) for 20 seconds
  - Allow embryo to rehydrate before resuming normal physiological activities
- Load into transfer gun
- Transfer
Frozen Transfer

• Vitrified
  – Apply epidural
  – Remove embryo from cane
    • Use caution
  – 8 seconds air thaw
  – 15 seconds water bath of 35°C water bath (not to exceed 37°C)
    • Straw thawed in horizontal position

Frozen Transfer

• Vitrified
  – Hold straw by cotton plug end and shake 4-7 times, briskly
  – Load into transfer gun
  – Transfer

Embryo Protection

• Protect embryo from extreme temperature changes
  – Cool is better than hot
    • If temperature of embryo exceeds normal body temperature of cow, embryonic death occurs rapidly
    • If traveling distance with embryos, be sure to keep them cool
      – If transferring fresh, embryos can be held at room temperature or refrigerated temperature for up to 16 hours prior to transfer
Embryo Protection

- If transferring frozen thawed embryos, transfer must occur within approximately 10 minutes after thaw
- Always protect embryos from direct sunlight
  - UV rays are great at killing stuff

Transferring Purchased Embryos

- Embryos often sold as packages
  - 3 embryos, guarantee 1 pregnancy
  - 4 embryos, guarantee 50% preg rate
- Caveat: Must be transferred by certified technician

Certification Standards

- Practitioner must pass certification exam
- 50 recoveries
- Handle 250 embryos
  - minimum of 50 frozen or transferred
- Business agreement with AETA
  - Renewed annually
Questions/Discussion

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References

- American Embryo Transfer Association. Website: www.aeta.org
- Canadian Embryo Transfer Association. Website: www.ceta.ca