Advanced Reproductive Techniques in Small Ruminants

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Artificial Insemination

- Advantages:
  1. Improve genetics
  2. Improve herd management
  3. Inexpensive semen
  4. Eliminate bucks & rams
  5. Decrease venereal disease

Artificial Insemination

- Disadvantages:
  1. Increased labor
  2. Lack of standardization for packaging & freezing semen
  3. Costs of AI equipment & semen storage

Success of AI Programs

- Fresh vs. frozen semen
- Number of inseminations
- Insemination method
- Timed A-I, Inseminated to standing heat
- Quality & quantity of semen
- Semen handling practices
- Management; nutrition, health programs
Selection of does and ewes for AI

• Good health & free of disease, reproductively sound,
• “Flush” feeding for 2-5 weeks before breeding
• Body condition score of 2.5 to 3

Body Condition Scoring

• Simple, fast but need to put your hands on them due to wool and hair concealing the accurate BCS
• Monitor feeding & herd health programs
• 1.0 to 5.0 scale with 0.5 increments

Body Condition Scoring

• Evaluate 3 areas:
  1. Lumbar area
     - spinous & transverse processes
     - muscle & fat over vertebrae
  2. Sternum
     - size of the fat pad
  3. Ribs
     - fat cover over ribs

BCS 1
Selection of does/ewes for AI

- History
  - birth of live, healthy kids & lambs
  - raised those kids to weaning

- Preference
  - does & ewes that conceive early
  - raise multiple young

AI Programs

- There are no uniform standards for frozen semen
- Evaluation of semen before insemination
- 70% morphologically normal
- (not greater than 15% primary abnormalities, at least 30% progressively motile)
  - inseminate into uterus

Protocol for synchronization of ewe for artificial insemination

- Progestin: Implant, Vaginal sponge, CIDR
- Teasing by vasectomized ram
- GnRH 30-36 hrs
- Inseminate with fresh semen
- Inseminate with frozen semen

Recognition of Estrus

- Recognizing standing heat
  - Flagging tail, restless, urination
- Changes in cervical mucus
- ovulate late in estrus or shortly after the end of estrus
- beginning of standing heat = clear & thin
- middle to late heat = cloudy & stringy

JrJohnson: Theriogenology of Sheep and Goats, Sheep and Goat Medicine, DG Pugh 2nd ed.
Timing of Insemination

- Insemination
  - ≤ time mucus turns cloudy
  - us. 12-15 hours after onset of estrus
  - if doe or ewe continues to exhibit heat after insemination inseminate again after 12 hours, particularly if the program uses cooled or frozen semen

Vaginal Insemination

- Does
  - 3 x 10^9 fresh semen
- Ewes
  - 4 x 10^9 fresh semen
- Conception rates
  - 15-30%
  - higher with experience

Vaginal Insemination

- Equipment
  - Cassou gun
  - +/− speculum
  - 1. Clean vulva
  - 2. Advance pipette into cranial vagina
    - dorsal vaginal roof

Cervical Insemination

Equipment - speculum & light source

1. Elevate hindquarters & legs held over a table or bale of hay
2. Introduce a lubricated speculum ~12cm through cleaned vulva & into vagina
3. Visualize cervix and atraumatically pass pipette as far in to cervix as possible
Cervical Insemination

- More skill required
- Dose:
  - ewes-1 x 10^9 fresh semen
  - 1.5 x 10^9 cooled semen
  - 2 x 10^9 frozen semen
- Conception rates
  - 35-50% or higher

Transcervical Insemination

- Place semen directly into uterus

Transcervical AI
- Cervix of the does is easier to pass a pipette than a ewe
- 35-50% conception rate
- Sheep 50-100 million sperm cells
- Goats 150-200 million sperm cells

Transcervical Insemination

- In ewes- Guelph system
- Equipment
  - speculum
  - wand-type light source
  - 25cm Bozeman forceps
  - pipette
- 50-100million PM spermatozoa
- Conception rates
  - 40-70% lambing rates depending upon skill of operator & quality of semen

Transcervical Insemination

The Doe
1. Clean vulva & perineal area
2. Standing or “over the rail”
3. Insert lubricated speculum & light source
  - Visualize cervix & place pressure on spec to lock cervix into lumen
- 50-100 million PM spermatozoa
- Conception rates of 50-80%
- 150-200 million sperm
Artificial Insemination

- Laparoscopic AI
  - Used more commonly
  - Higher conception rates
    - Up to 90%
  - Lower breeding doses
    - Doe – 20 million sperm cells
    - Ewe – 50 million sperm cells

Transcervical Insemination

Laparoscopic Insemination

- Visualization of uterus
- Place semen directly into lumen
- Used more commonly
  - Higher conception rates
    - Up to 90%
  - Lower breeding doses
    - Doe – 20 million sperm cells
    - Ewe – 50 million sperm cells

Laparoscopic Insemination

- Equipment
  - laparoscope
    (6.5-10mm diameter, 0° telescope)
  - trocar/cannula
  - insemination needle
- IMV
Laparoscopic Insemination

• Conception rates
  - 20-90%
  - dependent upon quality & quantity of semen & of course operator

Laparoscopic Insemination

• Hold off feed/h20 24 hrs
• Sedate doe or ewe & place in dorsal recumbency, head tilted down at 45° angle
• Clip & prepare abdomen for aseptic sx

Laparoscopic Insemination

• Infiltrate local anesthetic into 2 sites
  - 5 cm cranial to udder,
  4 cm on either side of midline
Laparoscopic Insemination

Trocar placed & distend abdomen with 1-2 L of CO₂
Second trocar placed opposite the first
Laparoscope inserted into first trocar & uterus visualized
Insemination pipette inserted into second trocar
Each horn inseminated using a special needle on the end of the pipette

Laparoscopic Insemination

• Insemination
  - avascular area of anterior uterine horn
  - insert needle at right angle to the uterine wall
  - make sure needle is in the lumen
  - experienced AI technician = 3-8 minutes

Semen Collection and Storage

• Collection AV
  – Raw- 0.1mL dosage of good quality semen
  – Extended 1:1 to 1:4; can dilute at 30°C with extender and then cool to 4°C and kept 12-24 hours

Cryopreservation of Semen

• Methodology still changing
• Some basics
  – Concentration > 3mill/mL with motility of 70%
  – Minitube makes extenders
  – Place in incubator or water bath 30°C
  – Slowly cool to 5°C over 1-2hr period
  – Then add freeze buffer to achieve final desired concentration (usually add 1:1)
  – Into straws; over liquid vapor 10min then plunge
Embryo Transfer

- Estrus Synchronization
- Superovulation
- Artificial Insemination
- Embryo Collection – Laparoscopic or Surgical
- Embryo Transfer

Embryos Transferred Worldwide in Other Species

IETS Newsletter December 2006

- 25,000 Ovine
- 7,000 Caprine
- ≥ 300 Cervid
- 14,000 Equine
- 30,000 Swine

Protocol for superovulation and synchronization in the doe

- Progestin: Implant in vaginal sponge CIDR
- AM FSH PM FSH
- PMSG (eCG)
- Ram: Give 50 µg GnRH
- Ram remains or lap Al
- Remove progestin
- Teaser Ram: 50 µg GnRH
- Remove food/water
- Transfer embryos

JW Johnson. Sheep and Goat Medicine 2nd ed DGPugh
Protocol for ewe superovulation and synchronization

In vivo - Surgical Embryo Collection
- Ventral laparotomy, flank, paramedian incision cranial to the udder
- 35mL of fluid per horn (Emcare)
- Antegrade flush; uterine tube to body with a foley (9 FG) in the horn or body
- Fluid is collected into a petri dish

Embryo Recovery
- Recovery 5-7 days post breeding as late morula or early blastocyst
- Most use surgical methods for recovery

Recumbant Flank Approach
Potential Complications

- General anesthesia
- Adhesions
- Herniation
- Peritonitis
Questions