**Topic : Artificial Insemination.  Unit III (2)**

**Artificial insemination (AI)**

is the manual placement of semen in the reproductive tract of the female by a method other than natural mating which is one of a group of technologies commonly known as "assisted reproduction technologies" (ART), whereby offspring are generated by facilitating the meeting of gametes (spermatozoa and oocytes). AI is by far the most common method of breeding of intensively kept dairy cattle. In developed countries, advances in artificial insemination have already had a major impact on livestock improvement programmes. AI speeds up genetic progress, reduces the risk of disease transmission and expands the number of animals that can be bred from a superior parent. The acceptance of AI technology worldwide provided the impetus for developing other technologies, such as cryopreservation and sexing of sperm, estrous cycle regulation, and embryo harvesting, freezing, culture and transfer, and cloning.

here.

**1).Method of semen collection**

Methods commonly used for semen collection

1 )Bull  :artificial-vagina, electroejaculation, massage, directly from vagina

2 )Ram and buck :directly from vagina, artificial vagina, electroejaculation

3 )Boar :using glove hand and artificial vagina

4 )Stallion :using artificial vagina

5 )Dog :using digital manipulation and using artificial vagina.

**2.Evaluation of semen**

1. Macroscopic evaluation

2. Microscopic evaluation

***2.1 Macroscopic evaluation***

The semen should be transferred to a water bath maintained at

35±1  °C.  Visual  evaluation  for  volume,  colour,

consistency/density,  odour  and  observation  for  presence  of

foreign material (blood,  pus cells,  dung, hair, etc.) shall be

made  and recorded. If dung  or hair  is found  in  the semen,

filtration with special semen filter is done.

***2.2 Microscopic examination***

Microscopic  evaluation  is  done  using  a  simple  or  phase

contrast  microscope  for  mass  activity  (wave  motion)  and

individual  motility.  Determination  of concentration  is done

with  a  hemocytometer  or  a  calibrated  photometer.  At  this

point  if  required  smears  can  be  made  for  morphological

studies  and  live/dead  count.  Nigrosin-eosin  stain  is

recommended. Buffered nigrosin-eosin solution is mixed with

a  drop  of  semen  and  smeared  on  a  glass  slide  for

morphological examination. It should be dried and examined

under oil  immersion. Automated computerised machines for

recording  motility  and  concentration  and  calculating  the

required extensions are normally used where ever affordable.

**Various biochemical tests used for asses the sperm fertility**

1. Methylene blue reduction test

2. Cold shock resistance test

3. Zone free hamster egg penetration test

4. Sperm mucus penetration test

5. Hypo-osmotic swelling test

6. Computer assisted semen evaluation

7. Fructolysis index

8. Oxygen utilization test

9. Millovanov’s resistance test

10. Hyaluronidase content.

. **Dilution of semen**

The first major improvement in the AI procedure was the development  of a  yolk-phosphate semen extender (Phillips and Lardy, 1940) Salisbury et al. (1941) improved the media by buffering the egg yolk with sodium citrate. Extender preserves fertilizing capacity of the spermatozoa for long  period and  increases  the  volume and thereby services to large number of females.

Dilutor provide viability and fertility of spermatozoa for prolong period. Ideal semen  dilutor  should  be  isotonic with  the  seminal  plasma, have a pH of 6.6 to 6.8 with high buffering capacity contain

lipoproteins  and  lecithin,  minerals  in  adequate  quality  and substances used for aerobic as well as anaerobic metabolism by the spermatozoa and glycerol. It should contain fructose to supply  energy  to  sperm and antibiotic control of microbes.

***The qualities of a good semen extender outlined by Samad (1985)***

1. )Osmotic pressure and electrolyte balance Equipment of seminal plasma(285 millimoles)

2. )Energy source Glucose,Fructose and Lactose in appreciable quantities.

3 )Buffering capacity Should maintain pH of diluted semen.

4. )Protection against cold shock Lecitten,Lipoprotein as casine in milk and egg yolk Protect against cold shock

5. )Cryoprotectant Glycerol

6. )Antibiotics

**4. Packing**

Packing of semen is done in medium straw (0.5 ml volume)

and mini straw (0.25 ml volume). Medium straw have a 3 mm size

**5)Semen preservation**

Semen is used either immediately after collection (“fresh”) for

example  turkeys,  human  beings; after  storage  at  a reduced

temperature (“stored”) for example horses, pigs, dogs; or after

freezing and thawing (“cryopreservation”) for example, bulls.

**Cryopreservation**

Semen is most useful for AI if it can be cryopreserved, since

this method  of preservation ideally enables the semen  to be

stored  for  an  unlimited time without losing quality.

**Thawing** semen in warm-water (35-38 °C) for 40 seconds is

the  most  commonly  used  thawing  procedure  reported  by

inseminators (80%). A significant increase in the conception

rate  (27%)  was  reported  when  thawing occurred  in  warm-

water (33-35°C) as opposed to air (Dejarnette and Marshall,

2005)6. Kaproth et al. (2005) [7] also reported a significant in-

crease (62.4%) in the  fertility rate  when thawing is  done in

warm-water 35 °C

7)Procedure for insemination

Firstly, it has to be ensured that the cow to be bred is truly in

heat. The cow should  be restrained  at first  and then  semen

should be thawed. The restraint area should be familiar to the

cow and free of stressful conditions. The tail is moved and the

cow is cleaned to remove any excess manure and debris from

the vulva. The gun is unwrapped and then inserted at a 30°-

40° angle into  the cow’s vulva.  The left hand is  inserted into

the rectum to check for the location of the end of the AI gun.

The cervix is grasped with the hand in the rectum of the cow

and is held steadily while the AI gun is thread into the cervix

of  the  cow.  When  the  AI  gun  is  all  the  way  through  the

cervix, the location is checked with index finger. The AI gun

should be only ½ to ¼ of an inch into the uterus. Slowly the

plunger is depressed at the end where the right hand is so that

½  of  the  straw’s  contents is deposited.

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Correct  timing  of  insemination  is  as  important  as  correct

placement of semen. Field experience has shown that the best

results are obtained when the insemination is performed at or

near the end of oestrus. The beginning and end of oestrus are

very difficult to determine. The simplest practical method of

timing inseminations is to use the a.m.- p.m. rule.

Cows first seen on heat Insemination time

Morning (a.m.) Same afternoon (p.m.)

Afternoon (p.m.) Next morning (a.m.)

Two-thirds of cows  commence oestrus at night and so will

first be seen  on heat in  the morning. With heifers and some

Bos indicus breeds, many authorities do not recommend the

a.m.– p.m. rule. They recommend  that these animals should

be inseminated soon after the first observed oestrus

**AI. Advantages and Disadvantages**

Advantages

1. Increased  efficiency:  During  natural  breeding,  a  male will  deposit  much  more  semen  than  is  theoretically needed  to  produce  a  pregnancy.  In  addition,  natural breeding  is  physically  stressful.  Both  of  these  factors limit  the  number of  natural matings a  male can  make. However, collected semen can be diluted and extended to create hundreds of  doses  from  a single  ejaculate. Also, semen  can  be  easily  transported,  allowing  multiple females  in  different  geographical  locations  to  be inseminated simultaneously, and semen can be stored for long  periods  of time,  meaning  that males  can produce offspring long after their natural reproductive live end.

2. Increased  potential  for  genetic  selection:  Because artificial  insemination  allows  males  to  produce  more offspring,  fewer  males  are needed.  Therefore,  one  can choose  only  the  few  best  males  for  use  as  parents, increasing the  selection intensity.  Furthermore,  because males  can have  more offspring,  their  offspring can  be used  in  a  progeny  test  program  to  more  accurately evaluate the genetic value of the male. Finally, individual farmers  can  use  artificial  insemination  to  increase  the genetic pool with which his or her animals can be mated, potentially decreasing effects of inbreeding.

3. Increased  safety  for  animals  and  farmers: As mentioned,  male  animals  can  become  large  and aggressive. These factors mean that maintaining a bull on a farm may be dangerous. Also, because of the relatively larger size of adult males than females, natural mating is more  likely to result  accidents and  injury to  either  the cow or the bull than is artificial insemination.

4. Reduced  disease  transmission: Exposure  of  sires  to infectious  genital  diseases  is  prevented  by  use  of  AI which  reduces  the  danger  of  spreading  such  diseases (Webb, 1992) [8]. In other way, if only males known to be free  from  disease  are  selected  for  semen  collection, artificial  insemination  can  play  an  important  part  in controlling  diseases  spread  through  sexual  contact. Among the diseases in this group are granular vaginitis, trichomoniasis, navel  ill, dourine, brucellosis and  coital exanthema.

5. Improving  animals’  productivity:  AI  plays  an important  role  in  enhancing  animal  productivity, especially milk yields, in developing countries that have a well-defined breeding strategy and a sound technical base to absorb  and adapt the  technology to  meet their needs (BBC,  2015)  [9].  Daughters  of  AI  sires  produce significantly more milk than those of herd bulls sires and the income from this extra milk may cover the extra costs resulting from extended calving intervals because of low heat detection.

6. Breeding can occur in the event of physical, physiological or behavioural abnormalities;

7. AI is a powerful tool when linked to other reproductive biotechnologies  such  as sperm  cryopreservation,  sperm sexing.

8. AI  can  be  used  in  conservation  of  rare  breeds  or endangered species.

9. The  use  of  semen extenders containing antibiotics also helped to prevent the transmission of bacterial diseases.

Disadvantage

1. Cost of  AI  compared  to  natural  service:  Despite the well-known advantages of artificial insemination, a large number  of  dairy  farmers  all  over  the  world  still  use natural service (NS) bulls to breed their cows. The main arguments allegedly justifying their choice are higher AI costs  compared  to  those  of  keeping  herd  bulls  and additional costs resulting from extended calving intervals because of low heat detection rates when AI is used.

2. Impact of  AI in  genetic  diversity: Even  though AI  is highly effective in improving animals’ productivity, there is  also  a  concern  that  its  inappropriate  or  unplanned usecan  lead  to  increased  rates  of  genetic  erosion  and breed extinction (Pilling et al., 2007) [10]. The heavy use of the best males results in a strong increase in inbreeding and a lose of genetic diversity.

3. Difficulty  of  heat  detection:  Among  different  factors that can affect conception rate per AI service, accuracy of heat (estrus) detection is the major one that determines AI program since  ova remains  viable for  only about  12-18 hours after  ovulation (Bekana,  1991) [11]. The  failure to detect heat is the most common and costly problem of AI programs and the major limiting factor  of reproductive performance  on  many  dairies

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