CURRENT & FUTURE ALTERNATIVE THERAPIES FOR REPEAT BREEDING IN CATTLE


Repeat breeder cow/buffalo is an animal which has failed to become pregnant when bred three cycles continuously, to a fertile bull or by AI.

The animal has apparently normal genitalia with clear discharge and having normal oestrous cycle (O. C.) length. The animal fails to become pregnant in spite of timely inseminations with proper technique of insemination. In case of cows, the age of the animal should not have exceeded beyond 14 years.

It is commonly noticed that, many people do not maintain proper records about the length of O.C. and consider the case as Repeat Breeder, though it may be falling into the category of Acute Endometritis or Early Embryonic Death (EED).

**Major causes of Repeat Breeding (RB) include:-**

1) Anovulatory Oestrous Cycle  
2) Delayed Ovulation  
3) Tubal blockage  
4) Failure of needation of embryo  
5) EED (within 16 days of AI – most commonly between 4th & 10th day)  
6) Deficiency of oxytocin  
7) Deficiency of energy  
8) Deficiency of progesterone  
9) Excess estrogen  
10) High ambient temperature & humidity  
11) Transport of cow/ buffalo in estrous.  
12) Uro-vagina  
13) Pneumo-vagina

Before discussing about treatment of RB, let us discuss the basic concepts regarding how stress, nutritional deficiencies etc. influence the process of reproduction. Let us also discuss the role of uterine defense mechanism (UDM) during O.C. and concepts of treatment of Endometritis or first degree Metritis.

- Morphine blocks ovulation. Opium has resemblance in action of morphine. Opium when derived endogenously is called ‘Opioid’. Endogenous opioid peptides (EOPs) are distributed throughout the brain and spinal cord in specific neuron tracks and have diverse physiological functions which regulates neuro-endocrine regulation of hypothalamus and pituitary hormones.

- The EOPs are derived from three different precursors, out of which important precursor is ‘PROPOPIO MELANOCORTIN (POMC)’.
POMC gives rise to Beta-Endorphin apart from other substances like Prolactin, MSH, ACTH and beta- Lipotrophin (another EOP). Beta-Endorphin is major opioid.

EOP may have important interaction with hypothalamic neurons, in control of Pituitary functions.

Stressors e.g. hypoglycemia, acute exercise and transport stimulate CRF, ACTH & concentration of B-endorphin which suppress GnRH secretion/ release, pre-ovulatory LH surge release/ tonic LH release or LH release response to GnRH, ACTH has also same supressive effects on tonic & pre-ovulatory LH release leads to suppression of ovulation.

Suckling leads to increase in EOPs in hypothalamus which inhibits LH secretion and after 72 hrs of weaning, EOPs decrease and LH release increases.

Sub maintenance feeding results in prolonged O.C., cessation of estrous with suppression of ovulation or ovulation without estrous.

Underfed animals also require more services per conception.

High nutritional status also leads to low conception rate due to EED caused by low level of progesterone level in plasma.

Nutrition plays an important role in initiation of post-partum ovarian activity. Many high yielder cows remain in –ve energy balance for first 70 days of lactation, but during first 2-3 weeks, the energy deficit is greatest. Ovulation is delayed by 0.7 days with each increase of one Megajoule of energy deficit during first 20 days of lactation. This occurs due to alteration in frequency of episodic release of LH for ovarian follicular development and ovulation. This leads to decreased Insulin concentration due to –ve energy balance in early lactation, which may render ovarian follicles less responsive to the gonadotrophin.

UDM during Oestrous Cycle:-

It is well known fact that uterus is more susceptible to infections during luteal phase and is highly resistant to infections during follicular phase of O.C. This may be due to;

- **During Follicular phase,**
  - I. uterine motility helps in physical clearance of bacteria.
  - II. Secretion of IgE is maximum and B-lymphocytes number is increased.
  - III. Increase in N-actyl-B-D-glucosaminidase which inhibits bacterial growth.

- **During Luteal phase,**
  - I. There is less infiltration of PMN in uterus
  - II. Bacterial activity of Myeloperoxidase enzyme of neutrophils decreases.
  - III. Secretion of lymphocyte proliferation inhibiting factor starts and hence less Ig are formed.
IV. Uterine motility is decreased.
V. Low intra-uterine pH provides more suitable environment for bacterial growth.
VI. Polysaccharide presence affects PMN surface receptors and inhibits phagocytosis.

CURRENT & FUTURE ALTERNATIVE THERAPIES FOR
BOVINE ENDOMETRITIS / 1st DEGREE METRITIS :-

Diagnosis of Endometritis / 1st degree Metritis on the basis of clinical manifestations and rectal examination is not reliable and hence we always treat the cow with various drugs to cure infection and avoid RB.

In general, the current therapies of Endometritis / 1st degree Metritis and uterine infections, can be classified into 4 major types;

I. **ANTIBIOTICS** :- these have been widely used as treatment of uterine infections, but success of these treatments varies from beneficial to no benefit.

   When **Penicillin** is used, it may be inactivated through bacterial production of the enzyme Penicillinase.

   In case of **Tetracyclines**, the need of large systemic dose to get effective tissue concentration against A. pyogenes could be toxic to the animal.

   The anaerobic environment of the uterus makes **Aminoglycoside** group (Gentamicin, Kanamicin, Streptomycin & Neomycin) ineffective because of the regular need of the oxygen for their activity.

   These antibiotics also inhibit phagocytosis. This suppression of leukocytic activity is further increased, if the bacteria are resistant to the antibiotic applied. In endometritis, the absorption of many drugs is diminished, due to which therapeutic levels in the deeper layers of the uterus and other parts of genital tract are not likely to be achieved.

II. **SULFONAMIDES** :- Bacteriocidal activity of sulfonamides, Aminoglycosides & Nitrofurazones is greatly reduced in an environment containing blood, pus, necrotic tissue and products of tissue damage & leukocytes.

III. **Antiseptics** :- Weak or dilute Lugol’s Iodine solution has been an effective treatment of endometritis.

IV. **Hormonal Therapy** :-
   A. **Estrogens** :- It increases the UDM of reproductive tract by increasing the blood circulation
   Leukocytic infiltration to the uterus, uterine contractions, mucus flow and phagocytosis. Dose of Estradiol benzoate given i/m is small i.e. 5 – 6 mg. However, long acting estrogens and stilbestrol have been associated with more severe infections of oviducts & myometrium and development of cystic ovaries.

   B. **Oxytocin** :- It increases phagocytosis. The effect can be seen upto 8 days post partum, if involution has been delayed.

   C. **Prostaglandin F2? analogues**: - It brings about luteolysis & decreases Progesteron
induced inhibition of UDM and increases estrogen induced UDM.

It has been observed that systemic use of antibiotics gives better results. Only a double dose of an antibiotic is required to achieve the optimum level of drug in the uterine lumen.

**DOSES RECOMMENDATION FOR ANTIBIOTICS:**

1. **Penicillins** :- It is rapidly absorbed following intra uterine infusion, reaching peak plasma concentration in 1 hr. and decline below MIC after 4 hrs. **Crystalline Penicillin G** (1 million IU) & **Procaine Penicillin** (1 million IU) maintained detectable levels 24 & 48 hrs, post-infusion respectively. **Benzyl Penicillin** following I/M route at 22000 IU / kg body wt. peak levels in blood & endometrial tissue were attained 15 & 60 minutes post-administration, respectively and remained for longer periods of time.

2. **Ampicillin** :- I/M @ 4-6 mg/kg b.w. – needs to be repeated at 24 hrs.
   Intra-uterine 3 gms- needs to be repeated at 24 hrs.

3. **Cephalexin** :- I/V 15 mg/kg b.w.- needs to be repeated at 9 hrs.

4. **Aminoglycosides** :- Dihydrostreptomycins attain peak 1-2 hrs following I/Ut.
   Intra-uterine dose is 1 gm.
   Detectable concentration remains for 24 hrs in uterine lumen.

4. **Gentamicin** :- I/M administration @ 5mg/kg is found in uterine lumen after 30 min.
   Needs to be repeated at 12 hrs.

5. **Tetracyclines** :- I/V twice daily@ 9-11 mg/kg b.w. It doesn’t give good results I/Ut.

6. **Chloramphenicol** :- Gives better results in uterine infection.
   I/M @ 20 mg/kg b.w. attains serum peak in 30 min. Needs to be repeated at 10 hrs in normal cycling & 12 hrs in dystokia suffering animals.

7. **Sulfonamides** :- Sulfonamides @ 80 mg/kg b.w. as loading dose and 65 mg/kg as maintenance dose provides antimicrobial cover to uterus upto 14 hrs.
   Repeat after 24 hrs.

**FUTURE THERAPY FOR BOVINE ENDOMETRITIS / 1st DEGREE METRITIS** :-

This is mainly based on using substances which cause activation of UDM. These substances are E. coli endotoxins, Serum plasma & hyperimmune serum, PMN extracts and Component Granululocyte-Macrophage colony stimulating factor and 1-10 % Oyster glycogen.

**RECENT IMPORTANT FINDINGS IN OESTROUS DETECTION:**

i. Average no. of mounts for a cow during oestrous period is **8.5**

ii. Average time from first to last mount during oestrous period is **7.1** hrs.
iii. Average oestrous mount lasts 4 seconds. This means that an average cow expressed oestrous for 34 seconds during 7 hrs period once every 21 days. I think you will understand the difficulty in detection of heat, from this information.

**EFFICACY OF UTERINE TREATMENTS IN COW (with ANTIMICROBIALS):**

- More effort is needed to improve sanitation and management practices in post partum uterine infections in individual herds/cows than using I/Ut antibiotics.
- Cows that are not severely affected will most likely resolve these infections with their own inherent defense mechanisms. Antimicrobial therapy may not offer any advantage.
- Cows severely affected by uterine infections may suffer some degree of impairment of fertility, whether or not treated, since, the value of intra-uterine antibiotics in cycling and RB cows is questionable.
- Fungi and yeast become prevalent due to excessive use of I/Ut broad spectrum antibiotics.
- Mild & localized uterine infection can be treated with antimicrobial therapy I/Ut except Aminoglycosides.

**ANOVULATORY HEAT and DELAYED OVULATION:**

In both the conditions, record the location of follicle on ovary. Examine the ovary for 2-3 or more days at 24 hrs interval as well as on 10th and 17th day of oestrous in cow.

In delayed ovulation, examine animal till you find ovulation and CL size on 10th and 17th day after heat. If ovulation occurs after 24 hrs. after the end of oestrous, it is delayed ovulation.

In anovulation, ovulation will not be there and hence no CL will be felt on above mentioned days of examination.

**TREATMENT FOR DELAYED OVULATION:**

1. Inject HCG or LH preparations 1500 to 3000 IU I/V on the day of AI. Give antihistaminic before this.
2. GnRH i.e. Receptal or Fertagyl 500 mcg I/M on the day of AI.
3. Inj. Bromocryptin 1 mg I/M on the day of AI.
4. Inj. Copper glycinate (as Copperglin of Omnapharma) 10 ml. I/M on the day of AI. Feed ?-Carotene daily 400-600 mg/day per cow.
5. Inj. Dienoestrol (an easily assimilable estrogen) by Ethinor Lab. 5 mg S/C in early oestrous in the next cycle.
6. Inj. Proluton depot 250 mg I/M at AI and 25% Dextrose 400 ml I/V + Inj. Vit. C 200 mg.
TREATMENT FOR ANOVULATORY HEAT :-

1. Remove stress as discussed.
2. Inject HCG or LH preparations 1500 to 3000 IU I/V in next O.C. in early estrous.
3. GnRH i.e. Receptal or Fertagyl 500 mcg I/M in early estrous.
4. Inj. 25% or 20% Dextrose 400 ml I/V + Inj. Vit. C 200 mg I/V at AI.
5. 10 ml Lugol’s Iodine drench for 15 days. Repeat after a month in non-responding cases or give I/V 5% Lugol’s Iodine, slowly.
6. Feed crushed Maize 1 kg. Daily for 45 days after proper soaking.
7. Feed 2 granules of Potassium iodide in concentrate mixture.
8. Homeopathy drugs:- Aurin Iodum 200 x Thyroidinum 30 x Aguns castus 200 x
   10 pills of each, 3 times a day for 10 days.
9. Feed mineral supplement daily especially containing Iodine 0.005 to 0.01 % Iodine and Manganese.
10. Feed ?-carotene in feed.

TUBAL BLOCKAGE :-

It is possible to diagnose the tubal blockage on careful P/R palpation. Accumulated fluids give a feeling of thickened zig-zag tube.

If the blockage is uni-lateral RB does occur.

Blockage can be diagnosed by injecting 10 ml of 0.1% Phenol red sol. intra-uterine in one horn preferably at the tip of the horn (otherwise the dye gets eliminated through external os and when get mixed with urine can result in a false +ve test). After injecting dye, collect urine after ½ an hour to within 3 hrs. by catheterization and record color of the urine.

How to prepare 0.1% Phenol red sol. :- Phenol red 100 mg + one small crystal of NaOH added in small quantity of D.W. to dissolve the powder. Add D.W. to make 100 ml. Autoclave.

If Fallopian tube is blocked, color of urine will not change. If it is not blocked, urine will turn Pink. This pink colour will remain for 12 hrs. It is preferred to carry out test for another horn during next O.C.

If you want to test both the horns during the same O.C., then
You have to catheterize & remove all the urine from bladder after 6 hrs. of testing the first horn.
Second horn can be tested by dissolving 1000 mg Indigocarmine in 100 ml D.W. and 10 ml dye in the horn at the tip and collect urine after 2-3 hrs by catheterization. If urine turns blue, there is no blockage.

If blockage is noticed Chromo-hydro-tubation is advised by using dial manometer. Fluid pressure should not be more than 180 mm/ cm². Use 5% DNS flexipack bag.
IV) FAILURE OF NIDATION OF FERTILISED OVUM:-

This is a common cause of RB and is due to endometrial infection or due to hypoluteal condition. It is advised to use antibiotic by systemic route on day 3rd of AI and on day 10th after AI. It is also advised to inject 1500 –3000 IU, HCG / LH on the day of AI to produce large size CL. Feeding of Vit. A and ?-carotene or greens daily is essential.

V) EED most common between 4th - 10th day after AI / NS :

This is a very common reason of RB. Under our managemental conditions, the main causes are Endometritis, too early or too late AI, High environmental temperature, Nutritional status of cow i.e. low energy diet, low protein diet, Mineral & vitamin deficiencies and Hormonal imbalance.

TREATMENT:- It is very difficult to diagnose 1st & 2nd degree Endometritis by P/R examination. In these conditions, uterus is atonic , tubularity may be normal for feeling but on milking out the uterus from tip to base during oestrous, uterus becomes flat. If mucus is examined under low power of microscope without staining for WBCs, they look like small dots and if WBCs are more than 8 no. in one field, you can suspect Endometritis.

In treatment, 2 principles are used:
1. Prostaglandin F2? treatment or
2. Shortening the O.C. by I/Ut infusion of Lugol’s Iodine.

In Prostaglandin F2? treatment 2 shots are given 10 to 14 days apart in case of cow, however more shots are required in buffaloes.

In Lugol’s Iodine, use 4% Lugol’s sol. On day 4 & 5 of the O.C., 5 ml quantity and estrous will follow after 5-7 days of infusion. However, in RB due to endometritis leave this estrous and breed at next estrous. If RB is not due to endometritis and if such estrous is induced with Lugol’s, breed the cow.

In order to correct the nutritional factor, crushed Maize ½ to 1 kg along with concentrate and mineral supplement 2-3 oz daily may be fed to animal along with green fodder min. 5-10 kg / day / animal. Feed ?-carotene 400-600 mg / day / cow.

Inj. HCG /LH 1500 – 3000 IU I/V or GnRH 100-500 mcg I/M at AI can be tried , when there is no endometritis.

Inseminate the cow at the end of heat and not too early or late i.e. follow AM / PM thumb rule.

Change the bull for AI and use recently frozen semen.

Byre should be well ventilated and better drainage for urine. Sprinkling of water on body of cows / buffaloes is advised to avoid ambient temperature as well as wallowing for buffaloes.

VI) Deficiency of Oxytocin:- Whenever uterine tonicity is lacking during estrous, there is possibility of uterine infection or deficiency of energy or deficiency of oxytocin. In oxytocin deficiency, animal passes large quantity of urine when examined P/R.
Give 50 IU of oxytocin I/M, after AI.

Atonicity may also be due to high progesterone content, since the CL may not have regressed totally

VII) Deficiency of Energy:- Atonicity is there during estrous however, clitoris reflex is present. In such cases, 20-25 ml of 25% Dextrose sol. may be given I/Ut 1-2 hrs before AI to enhance the fertility, however, if given I/Ut in endometritis, it precipitates infection and hence, it is better to give I/V 400 ml of 25% Dextrose.

Animal should be fed crushed Maize ½ kg well soaked daily for 60 days. Give 50 IU of Inj. Oxytocin I/M after AI, alongwith this treatment.

VIII) Deficiency of Progesterone:- This condition cannot be diagnosed in field condition except Examination of CL size on 8th to 10th day of cycle for hypoluteal condition, but not at the time of AI.

Inject 2000 mg of Vit. C, I/V alongwith 400 ml of 25% Dextrose, after AI. 1500 IU HCG / LH on the day of AI, I/V and on 14th to 16th day post estrous.

Feed ?-carotene 400-600 mg / day / cow and greens.

IX) Excess of Oestrogen:- Clinically difficult to diagnose. Rule out other causes, first. Sometimes, there occurs small prolapse of vagina during estrous.

Inject 2000 mg of Vit. C, I/V at AI.
Inject 250 mg Proluton depot I/M at AI.
Inject 1500 IU HCG / LH at AI, I/V.

X) High Ambient Temperature:- This leads to stress and EED. Well ventilated byres or keeping animals in tree shadows in hot times, sprinkling of water on body of animals, allowing buffaloes to wallow and availability of ample & fresh drinking water at all the times will take care of the problem.

XI) Transporting the estrous animal for AI:- This leads to stress due to which the cows / buffaloes around the onset of LH surge may release subnormal LH and leads to anovulatory infertility. Allow the animal to take rest for 30 minutes before AI. As far as possible, avoid transporting animal, instead call vet at home.

XII) Uro-Vagina:- In this condition, vulvar lips may look almost parallel to back bone. Aged cows may be having pendulus genitalia suspended into the abdominal cavity and at the time of urination the urine may get accumulated on the floor of the vaginal cavity. In such cases, at the time of P/R examination the urine gets expelled. At the time of AI, sheath alongwith sheath cover may be introduced upto the external os of uterus, then the sheath cover is torn by pulling it over and sheath with gun is introduced in cervix to carry out AI.

XII) Pneumo-Vagina:- At the time of P/R examination, you cannot get cervix, since it is displaced by Large quantity of air in vagina. The estrous mucus discharge comes out with froth. Perform Caslick’s operation.