

Anoestrus and Repeat Breeding in Dairy Cattle: Causes and Methods of Improvement

Amanuel B^{1*} and Tekalign T¹

¹*Department of Animal Science, Mettu University, P.O.Box 318, Mettu, Ethiopia*

***Corresponding author:** Amanuel Bekuma, Department of Animal Science, Mettu University, P.O.Box 318, Mettu, Ethiopia. E-mail: amanielbekuma11@gmail.com

Received: July 27, 2020

Published: August 17, 2020

Abstract

This review was aimed to summarize and synthesize the fragmented information on anoestrus and repeat breeding in dairy cows along with its causes and available methods of improvement. The goal of reproductive management in dairy cattle is to have cows become pregnant at a biologically optimal time and at an economically profitable interval after calving. However, reproductive inefficiency of cattle due to repeat breeding syndrome (RBS) and anoestrus is an expensive hitch in profitable dairy production as the insemination cost is increased, age at first calving in heifers is delayed; the inter-calving interval is extended, thus leading to lowering of calf crop. The causes of RBS were shown to be often very intricate, but can nevertheless be management factors (bad timing of insemination, missing heat detection and unprofessional insemination); semen factors (low quality of semen or infertile bulls); cow factors (endometritis, pyometra, mucometra, hydrometra, ovulatory defect, follicular cyst, adhesions, antibodies to seminal antigen, inappropriate uterine involution, infectious diseases, uterine tumours, anovulatory heats); and environmental factors (heat stress, nutritional factors). Whereas managerial, physiological, pathological, and nutritional factors are among the most important factors responsible for incidence of anoestrus in dairy cattle. Induction of ovulation by administration of gonadotrophin releasing hormone (GnRH), intra-uterine infusion of antibiotic and AI using double doses of semen are available methods for improvement of RBS. While for the purpose of anoestrus treatment, ultrasonography, prostaglandins and gonadotrophin-releasing hormone or progestagens in proper doses can be used.

Key words: Anoestrus; Causes; Dairy Cattle; Repeat Breeding; Treatment

Introduction

The most important objective of any species on this planet earth is self-perpetuation. Reproduction is the tendency of a species to produce individuals of its own kind. Reproductive performance, which depends upon a number of factors, is one of the most important factors affecting dairy farm profitability and the development of national economy, as well as the living standard of rural and urban societies; because, it directly or indirectly influences the yield of milk, reproductive culling rate and the cost for breeding and calf sales [1]. The goal of reproductive management in dairy cattle is to have cows become pregnant at a biologically optimal time and at an economically profitable interval after calving [2]. So, high reproductive efficiency is necessary for a successful dairy operation and requires a calving interval that maximizes milk production within the herd [3].

Dairy cows should calve one time every year to maximize economic efficiency. However, in contrast with this, the prevalence of reproductive problems resulted in poor reproductive performance which brings considerable economic losses to small holder dairy farms and the dairy industry [4]. Cows that have been highly selected for milk production in recent decades have suffered a decline in cow fertility, fertility is a

multi-factorial trait and its deterioration has been caused by a network of genetic, environmental and managerial factors and their complex interactions make it difficult to determine the exact reason for this decline [5]. Moreover, among the major problems those have a direct impact on reproductive performance of dairy cows, retained fetal membrane and the subsequent metritis and endometritis have been reported to be the most common clinical and economic problems [6]. Besides these, infertility or anovulatory and repeat breeding, abortion, dystocia, uterine infection and management practices are among the most and critical factors influencing reproductive performance of dairy cows (Hoojer et al., 1999 and ILCA, 1994 cited by [7]). The ultimate manifestation of infertility is failures to cycle, aberration to estrus cycle, failure to conceive and failure to produce offspring.

Anestrus and repeat breeding are two important reproductive problems leading to infertility in the dairy cows. It results into delay in puberty and lengthening of calving interval which squarely affect the economy by way of reducing the calf crops and life-time milk production. Anestrus is not a disease but symptom of different conditions like period before puberty, period of pregnancy, incomplete uterine involution and symptoms of infertility [8]. The pathogenesis of repeat breeding involves either failure of fertilization or early embryonic death

[9]. Even though some authors [10; Bartlett et al., 1986 cited by [11] and [12] have indicated as anoestrus and repeat breeding is an important reproductive disorder which causes great economic losses in farm animals and a major source of economic waste and poor reproductive performance in dairy herds, well thought-out information is not available and not yet compiled. Hence, this review was aimed to summarize and synthesize the fragmented information on anoestrus and repeat breeding in dairy cows along with their causes and available methods of improvement.

Definition and Concept of Anoestrus and Repeat Breeding in Dairy Cattle Repeat Breeder Cow

Repeat-breeder cows are commonly referred to sub-fertile animals without any anatomical or infectious abnormality that do not become pregnant until the third or subsequent breeding or remain infertile after numerous services [13, 14]. A cow is called as repeat breeder when it has failed to conceive even after three or more number of services [15]; has normal estrus cycle length, no abnormality in the vaginal discharge, no palpable abnormality in the reproductive tract, has calved at least once before and less than ten year of age. According to [16,17], a cow with repeat breeding syndrome (RBS) is a cow that looks healthy and has regular estrous cycle, however she will not conceive after three or more sequential inseminations, without any clinically obvious pathological disease. Similarly, a repeat breeder cow is a cow that has not conceived after three or more services associated with true oestrus; has normal or nearly normal oestrus cycles and genital tract, and even though it had been bred three or more times by a fertile bull semen had failed to conceive [18]. Moreover, a repeat breeder cow is a cow that looks apparently healthy and has regular oestrous cycle every 18 to 24 days [19], with no clinical detectable reproductive disorders or without anatomical abnormalities and infections by exhibiting a variety of reproductive disturbances [14]; however, it does not conceive when bred three times continuously, either with a bull of well-known fertility or inseminated artificially with excellent semen quality [20].

Repeat breeding is one of the most important infertility problem faced by field veterinarians. The cows look ostensibly normal and it is difficult to diagnose the cause. Repeat breeder females return to service repeatedly after being bred with a fertile male. The factors which are responsible for these abnormalities are multiple, but the major ones are anatomical, hormonal, management and infectious and vary from herd to herd and animal to animals [21]. Repeat breeder cows exhibit normal signs of estrus every 18 to 24 days but require more than 3 services to become pregnant [22,23].

Anoestrus in Dairy Cattle

Anoestrus is defined as failure of cows to exhibit overt estrus but is more commonly a problem with estrus detection. It is a functional disorder of the reproductive cycle which is characterized by absence of overt signs of estrus manifested either due to lack of expression of estrus or failure of its detection [24]. It is absence of periodic manifestation of oestrus without pregnancy [25]. It is a state of complete sexual inactivity with no manifestation of estrus for more than two months [26]. The anoestrus is usually associated with the presence of inactive ovaries even in the presence of follicular development where none of the growing follicles become mature enough to ovulate [27]. Some cows exhibit overt estrus for a very short time,

have few mounts or show signs of estrus in the middle of the night making estrus detection in these animals difficult. The estrus detection rate on many farms is less than 50%, being a very limiting factor to reproductive efficiency. In this condition there is insufficient stimulus from pituitary for secretion of Follicle-Stimulating Hormone (FSH) or Luteinizing Hormone (LH) resulting in failure of maturation and rupture of Graafian follicle. Many factors, such as footing, management and milk production level, will affect demonstration of estrus. While these cows are not observed in estrus, they have normal estrous cycles and will respond well to ovulation synchronization programs [28].

True Anoestrus:

It can be defined as the ovaries may be quiescent and inactive; this is referred to as a true anoestrus. The cow is not observed in oestrus either because she has not come into oestrus (not cycling) or because oestrus was not detected (cycling). The reasons for the failure of normal activity may be insufficient release or production of gonadotropins to cause folliculogenesis, or it may reflect the failure of the ovaries to respond [29]. Clinical rectal palpation will reveal small ovaries which are usually flat and smooth, absence of corpus luteum, either developing, mature or regressing cow in true anestrus will have virtually unchanged ovaries whilst a cow in late dioestrus or early dioestrus (metaestrus) will have a distinctly palpable corpus luteum [30]. Equine chorionic gonadotropin hormone can be used to stimulate ovarian activity, it can induce follicular growth and estrus; at a dose rate of 3000-4500 IU. GnRH, Progesterone and estrogen has been used successfully to treat anoestrus in dairy cows [31].

Sub-estrus or Silent Heat:

There may be normal cyclic ovarian activity but the cow is not showing the normal behavioral signs; this is described as sub-estrus or silent heat. The first and second ovulations post-partum is frequently not preceded by behavioral signs of estrus and is thus truly 'silent heat'. After the second estrus it is unlikely, that may result in true 'silent heats' to occur. When ovulation occurs in the absence of observed estrus it is more likely to be result of a failure of observation due to the short duration of behavior than to poor detection [32]. The cow has normal cyclic activity, but is not observed in oestrus due to weak or absent oestrous behaviour, or insufficient observation. If oestrus has not been observed in a dairy cow by 60 days post-partum the condition is defined as Post-Partum Anoestrus (PPA), whether she is cycling or not.

Classification of Anoestrus in Dairy Cattle

Anoestrus is categorized in to four clinical forms: silent heat; cystic ovarian disease; ovarian function and corpus luteum pseudo-graviditis [33, 34]. Benefiting from the use of ultrasonography and growing knowledge of follicular dynamics in cattle, the anovulatory status can be classified as: anovulation with follicle growth up to the emergence stage and anovulation with follicle growth up to deviation phase.

- Anovulation with follicle growth up to the emergence stage
- Cows exhibit very small follicles that are growing only to the emergence phase and do not proceed further. Fairly common in cattle exposed to poor feeding conditions such as extensive pasture management in tropical zones. The underlying factor is inadequate FSH stimulation.
- Anovulation with follicle growth up to deviation phase

Follicular growth takes place and proceeds through emergence and deviation but does not lead to ovulation; and frequently reported form of anoestrus. Commonly occurs in the postpartum period in lactating dairy and suckled beef cows. The characteristic signs of this condition are small ovaries, with the absence of a corpus luteum or ovulatory size follicles. The ovaries, however, show continuing follicular growth in a dynamic wave pattern up to the deviation phase. The underlying physiological problem is an inhibitory effect of oestradiol on GnRH/LH pulses that does not allow the final growth or oestradiol production by the post deviation dominant follicle.

Causes of Repeat Breeding and Anoestrus

Cause of Repeat Breeding

Repeat breeding can be caused by a number of factors, including:

- Sub-fertile bulls, endocrine problems, malnutrition, reproductive tract infections and poor management [35].
- Due to increment of no of insemination, longer calving interval and increased culling rates.
- Due to genetic, hormonal and nutritional imbalance, sub-clinical infection of uterus, and early embryonic mortality [36].
- Inadequate and inaccurate estrus detection is frequently a cause of cows becoming repeat breeders [37].
- Further, many risk factors such as breed of cows, herd body condition score (BCS), number of breedable cows in each farm may influence the occurrence of repeat breeding in population. Nevertheless, age, parity, BCS and milk yield of cows may affect the occurrence of repeat breeding in individual cows.
- Fertilization failure and early embryonic death are the major causes of repeat breeding those are influenced by uterine infection, genetics, ovulatory failure, error in estrus detection, improper timing of service [38].
- As the conclusion of [39] repeat breeding syndrome (RBS) in dairy cows is a multifactorial crisis involving the parturition, peri-partum, and post-partum factors. Age, BCS, parity number, milk production, lactation length, age of puberty and calving age, number of services, gestation length may influence the occurrence of RBS. The causes of RBS were shown to be often very intricate, but can nevertheless be summarized in short as follows:

- a) Management factors: bad timing of insemination, missing heat detection and unprofessional insemination;
- b) Semen factors: low quality of semen or infertile bulls;
- c) Cow factors (undetected): endometritis, pyometra, mucometra, hydrometra, ovulatory defect, follicular cyst, adhesions, antibodies to seminal antigen, inappropriate uterine involution, infectious diseases, uterine tumours, anovulatory heats;
- d) Environmental factors: heat stress, nutritional factor, etc [40].

Causes of Anoestrus

The anoestrus is usually associated with the presence of inactive ovaries even in the presence of follicular development where none of the growing follicles become mature enough to ovulate ([27]. It is also a result of managerial, physiological, pathological and nutritional factors. These factors include age, breed, pre- and postpartum nutrition, body condition at calving, milk yield, suckling, calving season, presence or absence

of the bull, delayed uterine involution, dystocia and general health status influence duration of postpartum anoestrus [41-45,27].

Methods of Treatment

Methods of Treatment of Repeat Breeding

Usually repeat breeder cows are diagnosed and treated by the veterinarians on the basis of history of the previous services and clinical examination of the cows. However, accurate treatment for repeat breeder cows has not been possible in most cases and the success of such treatment is not evaluated. Induction of ovulation by administration of gonadotrophin releasing hormone (GnRH) is commonly practiced in cattle to treat repeat breeders [46]. GnRH has been used at the time of AI in repeat breeding cows for improvement of conception [47]. It is likely that two times AI in a single heat may improve the conception rate in repeat breeding cows. Moreover, intra-uterine infusion of antibiotic may be beneficial for treatment of repeat breeding cows with subclinical uterine infection. Accordingly, intra uterine infusion of penicillin improved the recovery rate in repeat breeding cows with uterine infection [48]. Moreover, AI using double doses of semen has been practiced for improvement of conception in repeat breeding cows elsewhere [49].

Specific treatments for conditions like endometritis, delayed ovulation may be carried out whenever suspected them as the cause. If specific cause was not identified the following guidelines may be followed.

- Bring the animal into positive nutritive balance
- Use good quality semen having more than 50 per cent progressive forward motility.
- Inseminate the cow at right time of the estrum. Do AI twice at 12 to 24 hour interval.
- Follow proper AI technique.
- After AI, Clitoral massage or 100 micro grams of GnRH or 1500 IU of luteinizing hormone may be administered to stimulate ovulation.
- Skip the AI; administer 1 million units of penicillin in saline twice at 12 hours interval during estrum.
- Flushing the uterus with normal saline with moderate pressure to remove cellular debris/ mild blocks in oviducts.
- Sexual rest for two consecutive cycles and breeding [50].

Methods of Treatment of Anoestrus/Anovulatory

Treatment is based on:

- a) Improvement in energy status- optimal nutrition during the transition period and during early lactation.
- b) Hormonal treatments- combined with increased energy supplementation or reduced suckling stimulus may also help to stimulate oestrus [50].
- c) Cause, diagnostic facility, availability and efficiency of drugs, response of the animal to the drug, dose of administration and health status of the animal

The control of oestrus and ovulation by the use of prostaglandins, gonadotrophin-releasing hormone or progestagens may ameliorate some of the problems of oestrus detection by helping the farmer to detect oestrus within a defined period. The syndrome can be treated successfully with intra-uterine infusion of 1% Lugol's iodine [10]. In prolonged estrus exhibiting repeat breeder cattle, the use of single insemination along with administration of buserelin acetate, a GnRH analogue, is sufficient; however, in the absence of hormonal treatment, the use

of double insemination at 24 h interval also gives optimal results [51]. Anoestrus is treated with PMSG at a dose of 3000 to 4500 IU to stimulate the ovarian activities and induce follicular growth and ovulation, GnRH at a dose of 0.5 mg to stimulate LH release. But in suckled cows second injection after 10 days is important after transient raise in progesterone to initiate normal cycle [52]. For the purpose of treatment of anoestrus and to improve breeding efficiency Cloprostenol is one of the most effective drugs [53]. Ultrasonography is becoming the first choice for sustainable improvement in the reproductive management program of dairy farms [54].

Conclusion and Recommendation

Dairy cows should calve one time every year to maximize economic efficiency. However, in contrast with this, the prevalence of reproductive problems (anoestrus and repeat breeding) resulted in poor reproductive performance which brings considerable economic losses to small holder dairy farms and the dairy industry; resulting into delay in puberty and lengthening of calving interval which squarely affect the economy by way of reducing the calf crops and life-time milk production. These reproductive problems in dairy cows are a multifactorial crisis involving a network of genetic (hereditary), environmental and managerial factors (malnutrition, lack of intensive follow ups and stress) and their complex interactions. Therefore, based on the above conclusive statements, the following recommendations are forwarded:

- Routine and periodical examination of cows during postpartum period is essential; since most cows acquire uterine infection during this period.
- Prevention of anoestrus is preferable over treatment and can be achieved by maintaining the healthy status of the animals by adopting efficient farm managemental practices.
- AI with double dose of semen and single AI with GnRH administration may improve the conception rate in repeat breeding cows.

References

1. Plaizier JC, Lissemore KD, Kelton D, King GJ. Evaluation of overall reproductive performance of dairy herds. *J Dairy Sci.* 1998;1848-1854.
2. Shamsuddin, M, Goodger, WJ, Hossein MS, Azizunnesa T. and N.K. Bennett, A survey to identify economic opportunities for smallholder dairy farms in Bangladesh, *Tropical Animal Health and Production.* 2006;8:131-140.
3. Ferguson, J. and Galligan, D. (2000): Assessment of reproductive efficiency in dairy herds. *Compend Contin Ed. Prac. Vet.* 2000;22:159-158
4. Bekana, M, Ekman, T and Kindahl, H. Intrauterine bacterial findings in postpartum cows with retained fetal membrane. *J. Vet. Med. A.* 1999;41:663-670.
5. Edwell SM, Slawomir Z, Tomasz J. Comparative study on the efficacy of hormonal and non-hormonal treatment methods in ovarian function affected dairy cows. *Vet Inst Pulawy* 2004;48:265-267.
6. Mukasa-Mugerwa E, Azage T, Tafese M, et al. Reproductive efficiency of *Bos indicus* (zebu) cows under artificial insemination. *Animal Reproduction Science.* 1991;24(1-2):63-72.
7. Regassa T, Ashebir G. Major factors influencing the reproductive performance of dairy farms in Mekelle City, Tigray, Ethiopia. *J Dairy Vet Anim Res.* 2016;3(4):145-149. DOI: 10.15406/jdvar.2016.03.00088
8. Noakes D, Parkinson T, England G. *Arthur's Veterinary Reproduction and Obstetrics.* 8th edn, China, Saunders. 2001;384-408.
9. Ayisheshim A, Abegaz S, Mohammed A. Study on the Major Dairy Cows Reproductive Problems in and Around Gondar Town, Northwest Ethiopia. *J Vet Sci Technol.* 2017;8:484. doi:10.4172/21577579.1000484
10. Faisal O and Adil S. Treatment of Repeat Breeding in Dairy Cows with Lugol's Iodine. *IOSR Journal of Agriculture and Veterinary Science (IOSR-JAVS).* 2014;7(4):22-26
11. Khan MA, Mushtaq MH, Munibullah, Khan A, Ahmad N, Nawaz M. . Incidence of repeat breeding in cattle and buffaloes of Pakistan. *Veterinaria* 2016;4:18-20.
12. Jayaganthan P, Vijayarajan A, Prabakaran V, Sivakumar A and Raja S. Synchronization of Ovulation in Repeat Breeding Crossbred Jersey Cows Using GnRH and PGF2 α . *International Journal of Science, Environment and Technology.* 2016;5(4):2377-2381
13. Canu S, Boland M, Lloyd GM, Newman M, Christie MF, May PJ, et al. Predisposition to repeat breeding in UK cattle and success of artificial insemination alone or in combination with embryo transfer. *Veterinary Record.* 2010;167:44-51.
14. Yusuf M, Nakao T, Ranasinghe RB, Gautam G, Long ST, Yoshida C, et al. Reproductive performance of repeat breeders in dairy herds. *Theriogenology* 2010;73:1220-1229.
15. Purohit, G. N., Kumar, P., Solanki, K., Shekher C and Yadav SP. Perspectives of fetal dystocia in cattle and buffalo. *Veterinary Science Development.* 2012;2(8):31-42.
16. Gustafsson H, Emanuelson U. Characterisation of the repeat breeding syndrome in Swedish dairy cattle. *Acta Vet. Scand.* 2002;43:115-125.
17. Moss N, Lean IJ, Reid SWJ, Hodgson DR. Risk factors for repeat-breeder syndrome in New South Wales dairy cows. *Preventive Veterinary Medicine.* 2002;54(2):91-103.
18. Srijit, T. Phylogenetics to Manage Reproductive Disorders in Ruminants. *International Animal Health Journal.* 2015;2(4):50-53.
19. Grooms D. Reproductive losses caused by bovine viral diarrhoea virus and leptospirosis. *Theriogenology.* 2006;66(3):624-628.
20. Ahmadi MR, Dehghan SA. Evaluation of the treatment of repeat breeder dairy cows with uterine lavage plus PGF2 α , with and without Cephapirin. *Turkish Journal of Veterinary and Animal Sciences.* 2007;31(2):125-129.
21. Singh J, Dadarwal D, Honparkhe M, Kumar A. Incidences of various etiological factors responsible for repeat breeding syndrome in cattle and buffaloes. *The Inter. J. Vet. Med.* 2008;6(1):1-6.
22. Parkinson T. Infertility. In: D. Noakes T. Parkinson and G. England, (Eds) *Arthur's Veterinary Reproduction and Obstetrics.* 8 Edition, Saunders Company, USA. 2001:463-464.
23. Ferraz ML, Araújo AB, Rodrigues CA, Watanabe YF, Vireque AA, Joaquim DC, Smith LC, Meirelles F V, Baruselli PS. The low fertility of repeat-breeder cows during summer heat stress is related to a low oocyte competence to develop into blastocysts, *Journal of Dairy Science.* 2011;94(5):2383-2392.
24. Kumar PR, Singh SK, Kharche SD, Chethan Sharma G, Behera BK, et al. Anoestrus in cattle and buffalo: Indian perspective. *Advances in Animal Veterinary Sciences.* 2014;2(3):124-138.
25. Chakurkar E, Barbudhe S and Sundaram R. Infertility in farm animals: causes and remedies. *Technical Bulletin No: 15, ICAR Research Complex for Goa (Indian Council of Agricultural Research), Ela, Old Goa- 403402, Goa, India.* 2008;1-14.
26. Hafez E. Sequential sterile Autolysis in cattle, *Vet. Med.* 2004;4:5423.
27. Montiel F and Ahuja C. Body condition and suckling as fac-

- tors influencing the duration of postpartum anestrus in cattle: a review. *Animal Reproduction Science*. 2005;85:1-26.
28. Wiltbank MC, Gümen A, Sartori R. Physiological classification of anovulatory conditions in cattle. *Theriogenology*. 2002;57:21-52.
 29. Dransfield MBG, Nebel RL, Pearson RE, Warnick LD. Current therapy in theriogenology. 1998;81:1874.
 30. Peters AR, Ball HJH. *Reproduction in dairy cattle*. (2nd ed), Blackwell science ltd, London, UK. 1995;61:89-105.
 31. Cuppus PT. *Current veterinary therapy*, Philadelphia, WB Saunders. 1991;692-695.
 32. Arthur GH, Noakes DE, Pearson H. *Arthur's veterinary reproduction and obstetrics*. 1992;366.
 33. Mwaanga, E and Janowski, T. Anoestrus in dairy cows: causes, prevalence and clinical forms. *Reproduction in Domestic Animals*. 2000;35:193-200.
 34. Zdunczyk, S., Mwaanga, E.S., Malecki-Tepicht, J., Baranski, W. and Janowski T. (2002): Plasma progesterone levels and clinical findings in dairy cows with post-partum anoestrus. *Bulletin of the Veterinary Institute in Pulawy*, 2002;46:79-86.
 35. Defra L. Organic dairy cows: milk yield and lactation characteristics in thirteen established herds and development of a herd simulation model for organic milk production. Project Report 0170. 2000.
 36. Keskin Abdulkadir, Gumen Ahmet, Yilmazbaş-Mecitoglu Gülnaz, Karakaya Ebru, Tasdemir Umut, Celik Yakup, et al. The effect of Progesterone Based Ovsynch Protocol and GnRH Treatment after Artificial Insemination on Conception Rate in Repeat Breeder Cows. *Uludag Univ. J. Fac. Vet. Med.* 2010;29(2):65-70.
 37. Rorie RW, Bilby TR, and Lester TD. Application of electronic estrus detection technologies to reproductive management of cattle. *Theriogenology*. 2002;57:137-148.
 38. Shamsuddin M, Bhuiyan MMU, Sikder TK, Sugulle AH, Chanda PK, Alam MGS, et al. Constraints limiting the efficiency of artificial insemination of cattle in Bangladesh. *IAEA Tecdoc*. 2001;1220:9-27.
 39. Hasan MMI, Hassan MM, Mohanta RC, Miah MAH, Harun-Or-Rashid M, et al. A comparative study on productive, reproductive and ovarian features of repeat breeder and normal cyclic cows in the selected areas of Bangladesh. *Journal of Advanced Veterinary and Animal Research*. 2018;5(3):324-331.
 40. Mandefro M and Negash G. Repeat breeder syndrome in dairy cows: influence of breed and age on its prevalence. *World Journal of Agricultural Sciences* 2014;10:200-203.
 41. Short RE, Bellows RA, Staigmiller RB, Berardinelli JG and Custer EE. Physiological mechanisms controlling anestrus and infertility in postpartum beef cattle. *Journal of Animal Science*. 1990;68:799-816.
 42. Yavas Y and Walton JS. Postpartum acyclicity in suckled beef cows: A review. *Theriogenology*. 2000;54:25-55
 43. Webb R, Garnsworthy PC, Gong JG and Armstrong DG. Control of follicular growth: Local interactions and nutritional influences. *Journal of Animal Science*, 2004;82(1):63-74.
 44. Hess BW, Lake SL, Scholljegerdes EJ, Weston TR, Nayigihugu V, Molle JDC, et al. Nutritional controls of beef cow reproduction. *Journal of Animal Science*. 2005;83(1) 90-106.
 45. Peter AT, Levine H, Drost M, Bergfelt DR. Compilation of classical and contemporary terminology used to describe morphological aspects of ovarian dynamics in cattle. *Theriogenology*. 2009;71:1343-1357.
 46. Taponen J, Katila T and Rodriguez-Martinez H. Induction of ovulation with gonadotrophin releasing hormone during pro-estrous in cattle: Influence of subsequent follicular growth and luteal function. *Animal Reproduction Science*. 1999;55:91-105.
 47. Hossain MM. Improvement of conception in repeat breeding cows using synthetic gonadotrophin releasing hormone. MS in Theriogenology Thesis. Department of Surgery and Obstetrics, Faculty of Veterinary Science, Bangladesh agricultural University, Mymensingh. 2002.
 48. Khair A. Hormonal intervention to increase the fertility in anoestrus and repeat breeding cows at Bangladesh Agricultural University veterinary clinic. MS in Theriogenology Thesis. Department of Surgery and Obstetrics, Faculty of Veterinary Science, Bangladesh agricultural University, Mymensingh, Bangladesh. 2005.
 49. Islam N. Efficacy studies of drugs used for the treatment of repeat breeding cows at Bangladesh Agricultural University Veterinary Clinic. MS in Theriogenology Thesis. Department of Surgery and Obstetrics, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh, Bangladesh. 2011.
 50. Indian Council of Agricultural Research, Extension Folder No. 49/2012): Anoestrus and Repeat Breeding in Dairy Cows: Causes and Management
 51. Sharma A, Singh M and Vasishta NK. Effect of Gonadotrophin releasing hormone administration on conception rate following artificial insemination in repeat breeder cattle. *Indian J. Anim.* 2006.
 52. Vadakkadath Meethal S, Atwood CS. The role of hypothalamic-pituitarygonadal hormones in the normal structure and functioning of the brain. *Cell Mol Life Sci* 62: 257-261. *Sci.* 2005;76(4):330-332.
 53. Dudhatra GB, Mody SK, Patel HB, Modi CM, Chukewar AB, Kumar A, Awale MM. Prostaglandins and its Analogues : An approach for Treatment of Anoestrus and to enhance Breeding Efficiency, *Vet. World*. 2012;5(6):378-384. doi: 10.5455/vetworld.2012.378-384.
 54. Hauque M, Akhter M, Shamsuddin M. and Bari F. Ultrasonography for the management of reproductive disorders in dairy cows, *IOSR Journal of Agriculture and Veterinary Science (IOSR-JAVS)*. 2013;5(5):61-66.