

CURRENT THERAPY OF REPEAT BREEDING IN CROSS-BRED DIARY COWS WITH DILUTED POVIDONE IODINE

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المستخلص

أجريت هذه التجربة لدراسة إمكانية استخدام محلول أيودين البوفيدون المخفف (2%) في علاج ظاهرة إبقار الألبان مرتدة الشبق . استخدمت 60 بقرة حليب هجين مرتدة الشبق لدراسة هذه الآثار التي يحدثها أيودين البوفيدون المخفف. تم تقسيم الأبقار عشوائياً إلى ثلاثة مجموعات (أ) , (ب) , (ج) في كل واحدة 20 بقرة ثم أخذت مسحات رحميه من هذه الأبقار للتزريع البكتيري فوجد أن جميع الأبقار تعاني من إصابات رحم بكتيرية خفيفه. المجموعه (أ) تم علاجها بتسريب الرحم بأيودين البوفيدون المخفف قبل التلقيح ب 6 ساعات والمجموعه (ب) تم علاجها بتسريب الرحم بنفس المحلول بعد التلقيح ب 6 ساعات والمجموعه (ج) كمجموعه شاهدة. قيمت فعالية هذا المحلول في علاج ظاهرة البقرة المرتدة الشبق من خلال تأثيره على الفتره المفتوحه, معدلات التلقيحات اللازمه للحمل والفتره بين الولادات . أظهرت نتائج هذه التجربة أن تسريب الرحم بأيودين البوفيدون المخفف قبل أو بعد التلقيح يقصر معنويًا ($P < 0.001$) الفتره المفتوحه و الفتره بين الولادتين كما يقلل من معدل التلقيحات اللازمه لإحداث الحمل إذا ما قورنت بمجموعه الشاهد ($P < 0.001$). خلصت هذه الدراسة الي أن تدني الكفاءة التناسلية لأبقار الهجين يعزى بشكل أساسي إلى إصابات الرحم البكتيرية الخفيفه ونقص عنصر اليود الذين يؤديان في مجملها لظاهرة البقرة مرتدة الشبق بانتظام. وأن تسريب الرحم بهذا المحلول يساهم بفعالية ونجاح في تحسين الكفاءه التناسليه لهذه الابقار .

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Abstract

This study was carried out to examine treatment of repeat breeder (RB) dairy cows with diluted Povidone iodine (2%). Sixty cross-bred dairy cows suffer repeat breeding were used to investigate the efficacy of intra-uterine infusion of diluted Povidone iodine for treatment of repeat breeding in dairy cows. Uterine swabs were collected from uteri of the candidate cows prior treatment. After bacterial culture the uteri of the selected cows were found mildly infected (subclinical endometritis). They were then divided randomly into three equal groups. Group A (n = 20 cows) was intra-uterine infused with 2% Povidone iodine 6 hours pre-insemination. Group B (n = 20 cows) was infused with the same solution 6 hours post-insemination. Group C (n = 20 cows) was left without treatment to serve as control. The potency and efficacy of diluted Povidone iodine to cure repeat breeding in dairy cows was evaluated by studying three parameters, the days open (DO), rate of service per conception and the calving interval (CI). The results of this study revealed that, the DO, rate of service per conception and CI were significantly ($P < 0.001$) reduced by intra-uterine infusion of diluted Povidone solution 6 hours pre and post insemination as compared to the control.

It is concluded that, the repeat breeding (RB) syndrome in cross-bred dairy cows is most likely to be due to sub-clinical endometritis of the reproductive tract as well as deficiency of Iodine. Furthermore, intra-uterine infusion of 2% Povidone iodine can be used successfully to improve the reproductive efficiency of RB dairy cows.

Introduction

A repeat breeder cow (RB) is an apparently healthy animal with normal genitalia, young, free from puerulent vaginal discharge, multiparous, without anatomical abnormalities and has regular oestrous cycle ((Warelach, et al, 2008; Katagiri, 2011). However, this cow did not conceive when bred three times continuously, either with a well known fertility bull or inseminated artificially with excellent semen quality (Ahmad and Dehghan, 2007). Repeat breeding is the major problem involves in the herds infertility (Bartlett, et al, 1986). The incidence of repeat breeding world- wide in dairy cows range between 3 to 10 % (Bartlett, et al, 1986, Kimura, et al, 1987). Potential causes of repeat breeding mainly include subclinical endometritis (Rao, 1982, Ahmed, 2009, Ahmed and Elshiekh, 2013), nutritional deficiency specially , trace minerals and vitamin A (Francos, et al 1977, Peters, 1996, Ahmed, 2009), age of the dam (Bartlett, et al, 1986), improper heat detection (Dekruif, 1978), endocrine dysfunction (Gustafsson, et al 1986, Bage, et al 1997).

Concerning situations In the Sudan, there is a very high incidence (35%) of repeat breeding in cross-bred dairy cows which could be mainly as a sequel of postpartum (PP) subclinical endometritis, nutritional insufficiency, improper management, natural service by sub-clinically infected bulls or heat stress (Ahmed, 2009). The repeat breeding is the major source of economic loss in dairy herds (Bartlett, et al, 1986, Ahmad and Dehghan, 2007), the costs are increased through artificial insemination (AI) expenses, increased days open (DO), culling and replacement. Also treatment of repeat breeding with antibiotics and hormones is extremely expensive, hazardous and with inconsistent results. Recently There has been some interest in finding new, effective protocols for repeat breeding syndrome therapy in dairy cows such as intra-uterine infusion with diluted iodine compounds (Ahmed, 2009) or injection of insulin (Selvarajo, et al, 2002).

The objectives of the present study are to investigate the effect of intra-uterine infusion of diluted Povidone iodine on days open (DO), rate of service per conception and calving interval (CI) in repeat breeder (RB) cross-bred dairy cows in the Sudan.

Materials and Methods

Study area: This study was carried out at Khartoum University Dairy Farm about four Km North of the Faculty of Veterinary Medicine at Shambat, North Khartoum.

Animals: The study was conducted on 60 cross-bred (Friesian × Kenana) Sudanese dairy cow 4 to 7 years old. Their body condition score (BCS) was varied between 3.00 to 3.50 according to the five-scale point system outlined by Wildman *et al.* (1982). Emaciated cows were scored 1.00; thin cows 2.00, average cows 3.00, fat

cows 4.00 and obese cows were scored 5.00. These cows were diagnosed as suffering sub-clinical endometritis as described by Barrow and Feltham (1993).

Heat detection: All cows in the herd were checked for oestrus signs by visual observations by well trained sheperd thrice a day: early in the morning at 7:00 am, in the mid day at 12:00 and at 7:00 pm for at least 30 minuits for each (Elsheikh and Ahmed, 2004). The cows were recorded on heat when it becomes restless, licks the perineum of other cows, jumps on other cows, allow other cows or bull to mount her, it bellows and there is a transparent clear vaginal mucus discharge. The cow was considered in a full response when it stands to be mounted by the bull and mating was completed (Arthur *et al.*, 2001; Elsheikh and Ahmed, 2004).

Number of service per conception: It was calculated according to Elsheikh and Ahmed, (2005). It is the number of services given to the animals that display oestrus signs and resulted in a confirmed pregnancy not less than 42 days after service.

Days open (DO): It was done by calculating the intervals in days from calving to the subsequent effective service date of those cows that conceived (Arthur *et al.*, 2001; Elsheikh and Ahmed, 2004).

Calving intervals (CI): this parameter was carried out according to Arthur *et al.*, (2001), Bath *et al.*, (1985), Elsheikh and Ahmed (2004,) and Amed and Elshiekh (2013) where the CI is the duration between two consecutive calving.

Uterine swabs collection and bacteriology: A transcevicul guarded sterile disposable swabs were collected from the endometrium of the candidate cows that were diagnosed as repeat breeders before intra-uterine infusion with diluted Povidone iodine (Noakes *et al.*, 1989). The swabs were transferred to sterile test tube and were cultured within 2 hour of collection. The swabs were cultured aerobically in pre-equilibrated sheep blood agar and on MacConkey agar. Identification of bacteria was based on the characteristic of colony, Gram-stain and morphology (Barrow and Feltham, 1993). Bacterial growth on the cultured plates was scored semi –quantitatively depending on the number of bacterial colonies detected on the plate 0 no growth; 1 < 10 colonies 2: 10 to 100 colonies 3: 100 to 500 colonies and 4 > 500 colonies (Noakes *et al.*, 1991).

Intra-uterine infusion of diluted Povidone iodine: A volume of 150 ml of 2% Povidone iodine was infused into the uteri of the repeat breeder dairy cows at 6 hours pre-insemination or post-insemination (Geiser *et al.*, 1995). The apparatus used for uterine infusion consisted of a 2 liter stainless steel jug with a projected nozzle at the bottom. A rubber tube of 100 cm long connected with 30 cm catheter was fitted to the nozzle of the apparatus (Elsanousi and El tayeb, 1979).

Pregnancy diagnosis: It was carried out for none retain cows by rectal palpation at 60 days after last cow service (Elsheikh and Ahmed, 2005).

Experimental design: The animals were divided randomly into 3 equal groups A, B and C (n = 20 cows each). Group A and B were infused with 150 ml diluted Povidone iodine 6 hours pre/post insemination respectively according to Mutiga, (1978) and Edwell *et al.*, (2004). Group C (n = 20 cows) were considered untreated

control. The DO, rates of service per conception and CI were assessed as described in the materials and methods.

Statistical analysis: Data generated were subjected to analysis of variance (ANOVA) followed by Fisher's protect least significant difference (PLSD) in a one factorial design using Stat View Analytical Computer Package version 4.01 . Significant differences at $P < 0.001$ were considered.

Results

Days open (DO): As shown in figure (1) repeat breeder dairy cows that were infused pre/post insemination with Povidone iodine, the DO were significantly ($P < 0.001$) reduced compared to the control. The mean length of the DO for the repeat breeder dairy cows that were infused with 2% Povidone iodine pre/post insemination were 134.30 ± 5.70 days and 151.40 ± 7.10 days respectively. While the DO of the control was 227.70 ± 6.40 days.

The rate of service per conception: Figure (2) showed that, in repeat breeder dairy cows the rate of service per conception was significantly ($P < 0.001$) improved by infusion of diluted Povidone iodine pre/post insemination (1.30 ± 0.10 and 1.90 ± 0.20 respectively). Seventy percent of the candidate cows that were infused with diluted Povidone iodine pre-insemination conceive from the first insemination, but only 25% of the repeat breeder dairy cows that were infused post-insemination conceived from the first insemination as compared to the control where most of the cows conceived after more than three inseminations (5.10 ± 0.20).

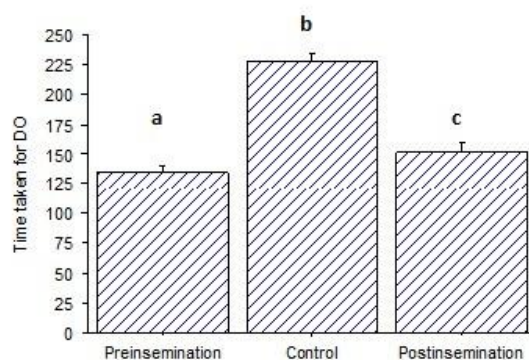


Fig1. The effects of intra-uterine infusion of diluted Povidone iodine pre/post insemination on the length of the DO in repeat breeder dairy cows (^{a,b,c} $P < 0.001$).

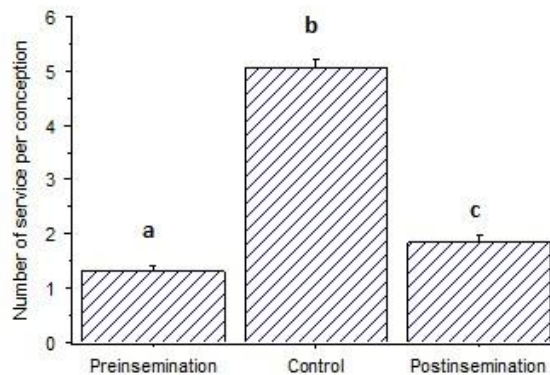


Fig2. The effects of intra-uterine infusion of diluted Povidone iodine pre/post insemination on the rate of service per conception in repeat breeder dairy cows (^{a,b,c} $P < 0.001$).

Calving interval (CI): Figure (3) indicates that, the repeat breeder dairy cows that were infused with diluted Povidone iodine pre/post insemination had a shorter CI as compared to the control ($P < 0.001$). Pre/post insemination, infusion of diluted Povidone iodine in repeat breeder dairy cows decreased the CI by 92 and 75 days respectively. The mean length of the CI of the repeat breeder dairy cows that were infused with diluted Povidone iodine at pre/post insemination were 407.90 ± 5.80 and 424.70 ± 7.50 day respectively and the CI of the control was 499.90 ± 5.90 days.

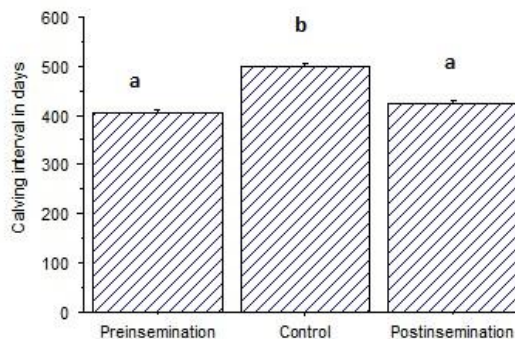


Fig.3. The effects of intra-uterine infusion of diluted Povidone iodine pre/post insemination on the length of CI in repeat breeder dairy cows (^{a,b} $P < 0.001$).

Discussion

This study confirmed that, the reduced reproductive efficiency with reference to repeat breeder cow syndrome in cross-bred Sudanese dairy cows is mainly due to subclinical endometritis and trace minerals deficiency specially iodine. Furthermore, this study indicated that, this problem could be solved radically by intra-uterine infusion of 2% Povidone solution pre/post insemination.

Subclinical endometritis is mainly due to contamination of the genital tract of the cows via the descending bacterial infection (Ahmed 2009, Ahmed and Elshiekh 2013). Bacterial products or the associated inflammation reduce pituitary FSH, inhibits LH release and perturb ovarian follicular growth and function, which delays the ovulation in the dairy cows (Opsomer, *et al*, 2000). It also perturbs embryo survival and implantation (Huszenicza, *et al.*, 2005). Also this subclinical endometritis altered the pH of the genital tract of the subclinically infected cows and consequently led to failure of conception in the subsequent breeding. This result is in agree with the result of Grohan and Saoloemi (1990) who reported that subclinical endometritis leads to silent heat, failure of inseminations and repeat breeding in dairy cows. The intra- uterine infusion of diluted Povidone iodine into the uteri of the repeat breeder dairy cows pre-insemination had improved conception rate. This improvement could be due to the potent bactericidal effects of the solution, adjustment of the reproductive tract tissues pH before insemination, slight hyperemia of the endometrium which enhanced uterine blood circulation, increased defense mechanisms of the reproductive system that become reasonably efficient (Sarkar, 2006). Consequently, the rate of service per conception and CI were reduced in the repeat breeder dairy cows. However, the infusion of diluted Povidone iodine into the uteri of the repeat breeder dairy cows post-insemination had less improvement on the rate of service per conception and CI. This difference definitely is due to sub-clinical endometritis that occurred in the genital tract of the repeat breeder dairy cows, which disturbs the uterine pH and decreases the liveliness of the embryo growth (Edwell, *et al.*, 2004). This result is in consistent with that of Koujan, *et al* (1996) who reported that infusion of diluted Povidone iodine post-insemination in repeat breeder dairy cows improve the CI and reduce the rate of service per conception. It is well known that iodine is one of an essential trace element for initiation of follicular wave dynamic, ovarian activities, release of the best quality mature oocytes and improvement of rate of service per conception and consequently CI in dairy cows (Bailey, *et al.*, 1999; Sarkar, 2006). Povidone iodine 2% is known to increase blood supply of the genital tract tissues and the absorbed iodine contained in diluted Povidone solution transferred to the thyroid gland and associated with thyroid binding globulin (TBG) in the thyroid gland to release thyroxine under control of hypothalamus (TRH) and pituitary gland (TSH) which is important for internal cellular metabolism, improvement of follicular development,

ovulation of high quality oocyte and maintenance of pregnancy (Yu and Tak-yin, 1993). Furthermore, Povidone iodine has a broad spectrum bactericidal, fungicidal anti protozoa effect (Sheldon, *et al.*, 2006). These activities of iodine will enhance the healing of the endometrium and restoration of its activities as well as the improvement of activity of pituitary and thyroid glands. Also, the internal cellular metabolism of the reproductive system including the ovaries will also be improved by infusion of diluted Povidone iodine (Sarkar, 2006). Thus, in this study the DO and CI were reduced. Moreover, the rate of service per conception at subsequent breeding was obviously improved.

Iodine deficiency is mainly exaggerated by heat stress that is common in the Sudan. The reduced iodine in the body of the dairy cows will lead to reduction in thyroxin release that is essential for improvement of herd fertility (Sarkar, 2006). Thus, from the results of this study it is clear that intra-uterine infusion of diluted Povidone iodine could be the most popular therapy improves the reproductive efficiency of repeat breeder dairy cows.

Reference

- Ahmadi, M. R. and Dehghan, S. A. (2007). Evaluation of the treatment of repeat breeder dairy cows with uterine lavage plus PGF₂, with and without cephalixin. *Turk. J. of Anim. Sci.* 31(2):125 – 129.
- Ahmed, F. O. (2009). The efficacy of intra-uterine infusion of Iodine compounds on the reproductive efficiency of postpartum and repeat breeder dairy cows in the Sudan. PhD thesis, university of Khartoum, Sudan
- Ahmed, F.O and Elshiekh, A.S. (2013). Uterine bacterial infection during postpartum delays recrudescence of reproductive traits in cross-bred dairy cows. *The J. of American Sci.* 9 (6): 593 – 593.
- Arthur, G. H. Noaks, D. E., Harold, P. and Parkinson, T. J. (2001). *Veterinary reproduction and obstetrics* 8th ed. W.B. Saunders company Limited Pp 402 – 464
- Bage, P., Gustafsson, H., Forsberg, H., Larsson, B. (1997). Progesterone levels in repeat breeder heifers during pro and oestrous period. *Theriogenology* 47: 141 – 142.
- Bailey, T. L., Dasonio, J., and Murphy, J. (1999). Analysis of reproductive records to improve dairy herd production. *J. Vet. Med.* 94: 269.
- Barrow, G. I. and Feltham, R. K. (1993). *Manual for the identification of medical Bacteria*. 3rd. Cambridge university press: 50 - 150.
- Bartlett, P. C., Kirk, J. and Mather, E. (1986). Repeated insemination in Michigan Holstein Friesian cattle : incidence descriptive epidemiology and estimated economic impact. *Theriogenology* 26: 309 – 322.
- Bath, D. R. Dickinson, F. and Appleman, R. (1985). *Dairy cattle principles, practices, problems and profits*. 3rd ed lea and Febiger philadeiphia pp 284 - 258.
- Dekriuf, A. (1978). Factors influencing the fertility of a cattle population. *J. Reprod. Fert.* 57: 507- 518.

- Francos, G. , Davidson, M. and Mayer, E. **(1977)**. The influence of some nutritional factors on the incidence of the repeat breeder syndrome in high producing herds. *Theriogenology* 7: 105 – 111.
- Edwell, M. S., Zduncz, K. and Janowski, T. **(2004)**. Comparative study on the efficacy of hormonal and non hormonal treatment method in ovarian function affected dairy cows. *Bull Vet. Inst pulaway* 48: 265 - 267.
- Elsanosi, S. M. and Eltyeb, A. B. **(1979)**. Bacteria isolated from uterine washings from mares in the Sudan. *Equine Vet. J.* 11(4): 219 - 222
- Elsheikh, A. S. and Ahmed, F. O **(2004)**. Reproductive performance of cross-bred dairy cows treated with GnRH early postpartum. *J. Anim and Vet. Adv.* 3(5): 329 - 334.
- Elsheikh. A. S and Ahmed, F. O. **(2005)**. Backing up postpartum dairy cows with PGF₂ α . *J. Anim. And vet Adv.* 4(5): 506 - 509.
- Geiser, R. L., Siegenthaler, H. and Martig. J. **(1995)**. Effectiveness of local treatment using a PVP. Iodine compound for retained placenta in cows. *Ber/munch tierarztl wochenschr.* 108(7): 264 - 268.
- Grohan, Y. B. Erb, H. and Saoloemi, H. **(1990)**. Epidemiology of reproductive disorders in dairy cattle. *J. Prev. Med.* 8: 25 -39.
- Gustafsson,H., Larsson, K., and Madej, A. **(1986)**. Sequential endocrine changes and behavior during oestrus and metoestrus in repeat breeder heifers. *Anim. Reprod. Sc.* 10: 261 – 273.
- Huszenicza, G. F, Janosi, S. and Peters, A., R (**2005**). Effects of clinical mastitis on ovarian function in postpartum dairy cows. *Reprod. in Domest. Anim.* 40 (3): 199 – 204.
- Katagiri, S. (2011). Repeat breeding in dairy cows. *J. of Vet. Med.* 41: 51 – 53.
- Kimura, M., Nakao, T. and Kawala, K. **(1987)**. Luteal phase difficiency as a possible cause of repeat breeding in dairy cows. *British Vet. J.* 143: 560 - 566.
- Koujan, A. D., Elssah, Hussein, M. Ayoub, M. and Afiefy, M. **(1996)**. Therapeutic efficacy of Povidone iodine (Bectadine and Dichloroxylonol) (Septocid) in Holstein cows affected with endometritis and/or cervicitis. *Acta. Vet. Hung.* 44 (1): 111- 119.
- Noakes, D. E., Till, D., and Smith, G. R.. **(1989)**. Bovine uterine flora postpartum, a comparison of swapping and biopsy. *Vet Rec.* 124: 563 - 564.
- Noakes, D. E. Wallace, L., and Smith, G. R **(1991)**. Bacterial flora of the uterus of cows after calving on two hygienically contrasting farm. *Vet. Rec.* 128: 440 – 442
- Opsomer, G. N., Grohn, Y. T. and Kruif, A. **(2000)**. Risk factors for postpartum ovarian dysfunction in high producing dairy cows in Belgium. *Theriogenology* 53: 841 - 857.
- Peters, A. R. **(1996)**,. Embryonic mortality in the cow. *Anim. Breed abstr.* 64: 587 – 598.
- Rao, A. V. N. **(1982)**. Causes and incidence of reproduction disorders among Zebra x Taurus cross-bred cows. *Theriogenology* 17; 189 – 191.
- Sarkar, A., K **(2006)**. Therapeutic management of anoestrus cows with diluted Lugol's iodine and massage on reproductive organs. *J. of Anim. Vet. Sci.* 1(1): 30 - 32.
- Selvarajo, S. Agarwa, S. K. and Shanker, U. **(2002)**.Fertility responses and hormonal profile in repeat breeding cows treated with insulin. *Anim. Reprod.Sci.* 73: 141 – 149.
- Sheldon, I. M., Lewis G. S., Le Blanc S., and Gilbert R. O. **(2006)**. Defining postpartum uterine disease in cattle. *Theriogenology* 65 (8): 1516 - 1530

- Warriach, H. M. , Ahmad, G. and Ahmad, I. **(2008)**. Effect of antibiotic treatment on pregnancy rate of repeat breeding dairy cross-bred cows with sub-clinical uterine infection. *Pakistan Vet. J.* 28 (1): 40 - 42
- Wildman, G. M., Kotwiga, J., Slinger, W. and Johnson, K. **(1982)**. Effects of suckling on pituitary responsiveness to GnRH hormones throughout the early postpartum of beef cows. *J. Anim. Sci.* 54: 594 - 603.
- Yu, H., O. and Tak-yin, M. H., **(1993)**. The efficiency of Povidone iodine pessaries in a start, low-dose treatment regime on candidal, trichomonal and non specific vaginitis. *Posgrad. Med. J.* 69(spp13): 58 – 61.