METOESTRUAL BLEEDING AND ITS EFFECT ON FERTILITY IN NATURAL AND INDUCED OESTRUS IN CATTLE

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Thesis submitted in partial fulfilment of the requirement for the degree of

Master of Veterinary Science

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University, Thrissur

2003

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DECLARATION

I hereby declare that this thesis entitled **"METOESTRUAL BLEEDING AND ITS EFFECT ON FERTILITY IN NATURAL AND INDUCED OESTRUS IN CATTLE"** is a bonafide record of research work done by me during the course of research and that this has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other university or society.

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Certified that the thesis, entitled **"METOESTRUAL BLEEDING AND ITS EFFECT ON FERTILITY IN NATURAL AND INDUCED OESTRUS IN CATTLE"** is a record of research work done independently by **Dr. LEEBA CHACKO** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.

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ACKNOWLEDGEMENT

It gives me inexplicably immense pleasure to place on record my sincere and deep sense of gratitude and indebtedness to my outstanding mentor Dr. K N Aravinda Ghosh,, Major Advisor and Chairman of the Advisory Committee, for his able guidance, creative suggestions, sustained encouragement and critical comments during the period of the entire academic programme especially in the ship shaping of the thesis. It is a matter of great fortune and pride, I deem it, to get an opportunity to work under the potential guidance of an affectionate, dynamic and versatile academician and clinician of his caliber and esprit.

My unreserved gratitude goes to Dr.T Sreekumaran, Associate Professor and Head, Department of Animal Reproduction and Member of Advisory Committee for his constant supervision, creative criticisms, timely suggestions, and unreserved regard rendered to me during the entire course of study.

I am greatly indebted to Dr. G Krishnan Nair, Associate professor, Department of Microbiology and Dr. Joseph Mathew, Associate Professor and Head, University College Hospital, Mannuthy for their valuable help and guidance proffered to me in their respectful and respective positions as the Members of Advisory Committee.

My native thanks are due to Dr.P.P.Balakrishnan, Associate Dean, College of Veterinary and Animal Sciences, Pookkode, for his valuable suggestions and encouragement throughout the research work.

I extend my heart felt thanks to Dr.K.V.Athman, Dr.V.Vijayakumaran, Dr.Metilda Joseph, Dr.Ajitkumar and Dr.C. Ibraheemkutty for their fruitful discussions and professional guidance.

My unreserved gratitude goes to Dr.A.M.Vahida for her constant support, valuable guidance, lively suggestions and motivations, generous encouragement and cordial company through out the course of study.

At one point or the other all the staff of the Department of Animal Reproduction were involved in this endeavour, I thank all of them.

The invaluable help rendered by the Department of Statistics for statistical analysis is gratefully acknowledged.

I wish to express my thanks to Dr. E. Nanu, Dean i/c, College of Veterinary and Animal Sciences, Mannuthy for providing the facilities for the research work.

I warmly remember the help and support rendered by Dr.E.Mathai and family throughout the course of study.

My sincere thanks are due to Dr.Antony, Dr.Smithamol, Dr.Sunitha, and Smt. Jayalakshmi of the Dept of Microbiology who imparted selfless service and technical guidance without which the work would not have been successful.

I am in short of words to express my gratitude and love to my colleague Dr.Afsal for his frank friendship extended to me making every moment momentous.

I wish to place on record the valuable help and fruitful company rendered to me by Dr.Lydia Priscilla, Dr.R.Velayudhakumar, Dr.Rejitha, Dr.Gopikrishnan, Dr.Raji Rose, Dr.Shiny John, Dr.Thushara and Dr. Durga Rani.

My heartfelt appreciation goes to the staff of M/s Peagles, Mannuthy for their efforts in formatting the manuscript.

I thank my mother-in-law for her divine wisdom, discipline and counsel; my sisters, spouses and their children for their support and care and my papa and mummy for their unstinted love and encouragement. I remember the support given by my husband, Eby David, to pursue this work even in the midst of the most calamitous situations and his endless co-operation in this endeavour. To these, my family, I dedicate this book.

Above all, I thank God for his guidance and inspiration.

LEEBA CHACKO

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Introduction

1. INTRODUCTION

Domestication has gradually transformed the reproductive process of farm animals from free range, seasonal mating system to an intensive production year round mating system. As a result the efficiency of reproduction of farm animals are often subjected to a multitude of environmental and managemental factors. Reproductive efficiency in a dairy herd has a marked effect on profitability.

Bovine oestrous cycle is a complex series of neurohormonal, endocrine and biochemical events. It is regulated by genetic, physiologic, hormonal, behavioural, environmental and psychosocial factors. An understanding of the mechanisms involved in the control of oestrous cycle has been influenced by a number of major discoveries made since 1950.

Reproductive efficiency in dairy cattle is affected adversely by aberrations in the oestrous cycle. Metoestrum is a poorly defined period following oestrus in cattle. Metoestrum is characterized by a phenomenon called metoestrual bleeding or post oestrual bleeding 12-24 hours after the cessation of oestrus. During prooestrum and oestrum, high oestrogen concentration increases the vascularity of endometrium, its vacsularity reaching a peak about one day after the end of oestrum. When ovulation occurs the oestrogen level drops rapidly. Soon the congested vessels begin to break down, and small quantity of blood is discharged into the uterus. This blood becomes mixed with mucus and passes from the reproductive tract and is observed as postoestrual bleeding. Muller (1999) studied the etiology of post oestrual bleeding and opined that it could be due to non synchrony of hormonal changes around oestrus. Majority of cows, which showed visible metoestrual bleeding, failed to conceive and showed signs of bleeding in concurrent heat also.

Metoestrual bleeding and its influence on subsequent conception seems to be one of the prime aspects in the widely speculated sphere of animal reproduction. There exist different opinions regarding the importance of metoestrual bleeding. Some think that the bleeding is a normal event in the cycle and it does not have any importance as some cows stay pregnant. Quayam and Austin (1983) reported significant difference in conception rate between cattle which showed metoestrual bleeding and which did not.

There is consensus that temporary infertility rather than permanent sterility poses greater threat to livestock population,warranting greater vigil and care. It is well established that the overall production efficiency of any bovine population has to be built upon a strong and sound foundation of scientific management of breeding, failure of which will result in low reproductive efficiency. Practical methods of oestrus control in cattle are based on either preventing ovulation or by causing regression of corpus luteum with luteolytic substances like prostaglandin. Reproductive management protocols for the regulation of oestrous cycle using prostaglandin or its synthetic analogues have been used successfully to increase oestrus detection over specified days.

In view of differing opinion, an attempt has been made to elucidate the relationship between metoestrus bleeding and conception in cows. The objective of the present study is to find out the occurrence, onset and intensity of metoestrual bleeding and its effect on fertility in natural and induced oestrus in cattle.

Review of Literature

2. REVIEW OF LITERATURE

Metoestrual bleeding and its subsequent effect on fertility is a matter of perpetual debate in the field of veterinary practice. There exist conflicting opinions concerning the importance of metoestrual bleeding with reference to fertility in dairy cattle. Perusal of literature reveals paucity of studies and references related to the pathopyhsiological aspects of metoestrual bleeding.

2.1 OCCURRENCE OF METOESTRUAL BLEEDING

Trimberger (1941) reported that 61 per cent of cows and 81 per cent of heifers showed metoestrual bleeding on the second day following oestrus.

Weber *et al.* (1948) stated that although clinical evidence of bleeding occurred in only 55 of 68 heifers, but all heifers showed presence of red blood cells on examination of vaginal swabbing at this time.

Hansel and Asdell (1951) reported that 50 to 60 per cent of the cows and 75 to 85 per cent of heifers showed metoestrual / postoestrual bleeding.

Mathai and Raja (1973) observed two cases of metoestrual bleeding (2.77 per cent) in 72 oestrous cycles in nine Jersey- Sindhi F1 heifers.

Salisbury *et al.* (1978) stated that the primary cause for metoestrual bleeding was the sudden withdrawal of oestrogen following ovulation. This may result in congested endometrial vessels beginning to break down and small quantity of blood is discharged into the uterine lumen, exhibited as metoestrual bleeding.

Sobhanan (1978) reported that out of the 1568 animals brought for insemination at ICDP centre, Anthikkad, 15 (0.96 per cent) showed postoestrual bleeding. Out of the 867 heifers and 701 cows, nine heifers (1.03 per cent) and six

cows (0.9 per cent) showed post oestrual bleeding and the difference was non-significant.

On contrary to the earlier reports, Neduncheralathan *et al.* (1981) opined that the incidence of metoestrual bleeding was more frequent in cows than in heifers.

Quayam and Austin (1983) reported the incidence of metoestrual bleeding as 3.69 per cent in heifers and 0.93 per cent in cows. The overall incidence in both cows and heifers was 1.35 per cent.

Janardhanan and Nair (1990) screened 690 animals out of which 23 (3.33 per cent) showed signs of metoestrual bleeding of varying degree. The incidence was found to be higher in heifers (4.3 per cent) than in cows (2.76 per cent). They also stated that the incidence of metoestrual bleeding in crossbred cattle was much lower than that in continental breeds, but slightly higher than that in Sindhi and Sahiwal.

2.2 ONSET OF METOESTRUAL BLEEDING

Trimberger (1941) noticed that about 75 per cent of cows and heifers showed metoestrual bleeding on the second day following oestrus.

Weber *et al.* (1948) reported that the average occurrence time of metoestrual bleeding was 50 to 60 h after the onset of oestrus.

Out of the 74 animals showing metoestrual bleeding, 15 (20.25 per cent) showed bleeding on the day of oestrus (0 day), 30 (40.5 per cent) on day 1 of oestrus, 15 (20.25 per cent) on day 2 of oestrus and 14 (19 per cent) on day 3 of oestrus (Janardhanan and Nair, 1990).

Deshpande (1994) stated that post oestrual bleeding in cows occurred 12 to 24 h after the cessation of oestrus.

2.3 DURATION AND INTENSITY OF METOESTRUAL BLEEDING

Thirteen animals (17.6 per cent) showed scanty bleeding, 41 animals (55.4 per cent) showed moderate bleeding, and 20 animals (27 per cent) showed profuse bleeding. (Janardhanan and Nair, 1990). However duration of metoestrual bleeding was not defined by any authors.

2.4 CONSISTENCY AND CYTOLOGY OF METOESTRUAL DISCHARGE

Sharma *et al.* (1968) studied the oestrous cycle characteristics in Hariana cows and examined the cervico-vaginal mucus smear for fern pattern and cellular infiltration. Branches of fern pattern lay scattered in disintegrated form and smears presented heavy infiltration of leucocytes in the sample collected 24 h after the onset of oestrus. While fern like pattern was not observed, only leucocytic infiltration and epithelial casts were seen in samples collected 28 h after the onset of oestrus.

Hussain and Khan (1978) collected vaginal scrapings from apparently normal and healthy animals in different stages of oestrous cycle and found that during metoestrum the smears had plenty of cellular debris, leucocytes and parabasal cells.

Kurade *et al.* (1993) prepared vaginal smears using samples taken from the dorsolateral part of anterior vagina and studied the exfoliative vaginal cytology in normal and repeat breeder animals. Parabasal cells and intermediate cells were the predominant cells in metoestrus period. Most of the smears were characterised by the presence of R.B.Cs. The presence of R.B.Cs was attributed to metoestrual bleeding due to diapedesis or capillary bleeding from the endometrium.

2.5 ANTIBIOGRAM OF METOESTRUAL DISCHARGE

Kharade and Kulharni (1983) surveyed the intensity of occurrence of genital microflora of normal and repeat breeding cows. Samples of cervico vaginal mucus were collected from 40 repeat breeder and 10 normal breeder cows of which nine

samples of the former yielded 4-5 isolates and one of the normal 10 cows showed microbial growth. It was observed that the genital microflora of repeat breeders were highly resistant to most of the antimicrobial agents, compared to normal breeders, due to antibiotic therapy resulting in the formation of drug resistant mutants.

Krishna *et al.* (1994) subjected the cervico vaginal mucus sample of 44 repeat breeding animals and 84 normal breeding cows for antibiotic sensitivity tests. Organisms were isolated from 45 (35.1 per cent) out of 128 samples, while the remaining 83 (64.9 per cent) samples were bacteriologicaly sterile. Highest sensitivity was obtained to gentamicin (92 per cent), followed by chloramphenicol (84 per cent), furazolidone (72 per cent), ampicillin (48 per cent) and penicillin (44 per cent).

2.6 INFLUENCE OF METOESTRUAL BLEEDING ON CONCEPTION RATE

Out of 293 cows inseminated in the post oestrus haemorrhagic period (about 48 h after the end of oestrus), Antrup and Rasbech (1951) reported a conception rate of 29.69 per cent. They opined that delayed ovulation might have occurred; or rather, the ova might have survived for a longer period.

Larson and Bayley (1955) recorded a conception rate of 21 per cent in animals with metoestrual bleeding.

Asdell (1968) propounded that there was a little more likelihood that bleeding might occur during early pregnancy, but was not enough to use it as a diagnostic sign.

Sobhanan (1978) recorded a conception rate of 20 per cent in animals that had metoestrual bleeding and the bleeding continued in those that failed to conceive.

Study on the efficacy of progestational preparations in controlling metoestrual bleeding and enhancing conception rate in cattle was undertaken by

Neduncheralathan *et al.* (1981). Out of the 13 animals treated 11 conceived to first insemination with a conception rate of 84.6 per cent.

Quayam and Austin (1983) reported that three out of 17 cows and three out of 12 heifers that showed bleeding conceived in first insemination, with a conception rate of 17.64 per cent and 25 per cent, respectively. While the overall conception rate was 20.68 per cent in animals that showed bleeding, it was 36.96 per cent in those that did not and the difference was highly significant ($P<0.01$). There was also a difference in the conception rate, which was higher in heifers than in cows that showed bleeding. This difference was also statistically significant ($P< 0.05$).

In a study on 80 cows showing metoestrual bleeding, only 15 animals conceived, with a conception rate of 18.75 per cent (Sandhu, 1992). Clinical study was conducted on 21 cows with metoestrual bleeding, treated with two ml of Proluton depot (500 mg) through intramuscular route, when presented for insemination and found that bleeding stopped in 19 animals within 24 hours and 14 animals conceived to first insemination with a conception rate of 66.66 per cent.

Muller (1999) propounded that post oestrual bleeding was common in young cows and heifers and more common in Holstein Friesian breed of cattle. Progesterone level of milk was determined by milk progesterone assay on day Zero (day of insemination), on the day of bleeding and on day seven. The average milk progesterone level on the day of bleeding was 0.5-0.6 ng/ml, while in cows without bleeding it amounts to 0.5-1.0 ng/ml. The milk progesterone level on day seven were approximately equal in all the cows. He further opined that post oestrual bleeding could be regarded as a hormonal dysfunction of progesterone deficiency.

2.7 PGF_2 ALPHA TREATMENT

PGF_2 alpha is a potent luteolytic agent in cattle and is probably the natural uterine luteolytic factor. Luteolytic dose of prostaglandin or one of its synthetic analogues will cause regression of mature bovine corpus luteum (Lauderdale, 1972

and Cooper, 1974). However, exogenous prostaglandin is not effective in altering the bovine oestrous cycle if administered before day 5 or beyond day 16 of the cycle (Gordon, 1996). Recently, research has shown that a higher percentage of cattle treated with prostaglandin during late luteal phase (Day 10 to 15) exhibited oestrus than those treated during early luteal phase (Day 5 to 9) (Larson *et al.* 1996).

Fertility of heifers after oestrus synchronisation with prostaglandin was found to be normal or even higher than controls, especially after single prostaglandin treatment (Macmillan and Day, 1982 and Mc Intosh *et al.* 1984). However, results on fertility following prostaglandin treatment have been inconsistent in lactating cows (Macmillan *et al*, 1977; Macmillan, 1983 and Larson and Ball, 1992).

2.7.1 Oestrus Response after PGF_2 alpha Treatment

Oxender *et al.* (1974) observed that the mean intervals to onset of oestrus and ovulation after administration of 30 mg of PGF_2 alpha intramuscular were 74 ± 3 h and 104 ± 6 h, respectively.

Cumming *et al.* (1977) reported that 90 per cent of the fluprostenol treated cows ovulated within 92 h of treatment.

Vivanco and Delgado, (1980) administered two injections of PGF_2 alpha at an interval of 10 to 11 days to Brown swiss, Santa Getrudis and Nellore heifers and found that the interval from second injection to standing oestrus averaged 58.63, 57.70 and 69.66 h, respectively.

Nair and Madhavan (1984) observed that 98.15 per cent of suboestrus cows exhibited oestrus within 48 to 72 h after the administration of 500µg of Estrumate. The average duration of oestrus in these animals was within the range of 16 to 24 h.

Plunkett *et al.* (1984) induced oestrus using single injection of 25 mg PGF_2 alpha (lutalyse) after detection of a corpus luteum by ovarian palpation. Cows were inseminated artificially at 72 and 96 h after PGF_2 alpha injection. Fifty four per cent

of cows treated with PGF_2 alpha were detected in oestrus. The time taken for induction of oestrus was 65.7 ± 2.1 h after PGF_2 alpha injection.

El-Menoufy and Abdou (1989) recorded 82 and 90 per cent of cows in oestrus after double dose of 25 mg dinoprost or 500µg cloprostenol administered 11 days apart.

Studies on oestrus induction in crossbred cows using PGF_2alpha analogue *dinofertin administered 25 mg intramuscularly at 10 days interval, revealed that 75* per cent of the animals responded to treatment. The average time required for the onset of oestrus was 71.87 ± 1.25 h and the duration of oestrus was 18.25 ± 0.04 h. (Pawshe *et al.* 1991).

Archbald *et al.* (1994) studied effect of PGF2 alpha treatment on oestrus and pregnancy of lactating dairy cows by administering 25mg PGF_2 alpha intramuscularly. Fifty five per cent of cows were observed in oestrus after the administration of prostaglandin.

Oestrus was induced in normally cyclic cows by single intramuscular injection of 25 mg PGF_2 alpha during mid luteal phase of oestrous cycle. Eighty per cent of animals exhibited oestrus with in 74.64 ± 5.51 h after single injection of PGF_2 alpha (Chauhan *et al.* 1994).

Jacob *et al.* (1995) reported an average of 61.81 h for the induction of oestrus in cows after intramuscular injection of PGF_2 alpha (Lutalyse). The duration of oestrus recorded after induction of oestrus was 28.36 h. The percentage of intense, medium and weak oestrus recorded was 66.66. 19.04 and 14.28 per cent , respectively.

Momcilovic *et al.* (1998) determined the reproductive performance of lactating dairy cows treated with gonadotropin releasing hormone and PGF_2 alpha for synchronisation of oestrus and ovulation. The percentage of cows observed in oestrus within 7 days were 52 per cent and 50 per cent with two luteolytic doses of PGF_2 alpha given14 days apart and single dose of PGF_2 alpha respectively. There

was no significant difference in oestrus response between single and double dose of PGF_2 alpha.

Pinheriro *et al.* (1998) studied on the oestrus behaviour of Nelore breed (*Bos indicus*) induced with PGF_2 alpha in heifers and lactating cows. Heifers exhibited oestrus 82.5 ± 4.7 h after the second injection of PGF_2 alpha, while cows manifested oestrus at 70.5 ± 4.8 h.

Gopikrishnan (2001) observed that the time taken for induction of oestrus after two injections of PGF_2 alpha (Prosolvin) given 11 days apart was 69.50 ± 1.34 h. The duration of oestrus ranged from 24 to 36 h with a mean of 28 ± 1.07 h. The percentage of heifers showing high, medium and low oestrus was 64.08, 28.57 and 7.35 per cent, respectively.

Madhav *et al.* (2001) treated 25 suboestrus cows with 25 mg PGF_2 alpha intramuscularly of which 80 per cent of cows responded to treatment. The average time taken for the onset of oestrus was 83.10 ± 7.71 h. The average duration of oestrus was 21.20 ± 0.68 h. The percentage of cows showing intense, moderate and weak oestrus was 30, 35 and 35 per cent, respectively.

Rao *et al.* (2001) reported that the average time taken for induction of oestrus after single intramuscular injection of 25 mg PGF_2 alpha analogue dinoprost was 72.44 h.

2.7.2 Fertility after PGF_2 alpha Treatment

Oxender *et al.* (1974) in his experiment to assess the fertility of cows, after administration of 30 mg PGF_2 alpha intramuscularly and inseminated at 70 and 88 h after treatment, obtained a conception rate of 78.94 per cent.

Leaver (1975) used prostaglandin analogue (ICI 80996) to induce luteolysis in Friesian heifers and reported that a greater proportion of heifers conceived to insemination at 72 h than at 96 h, suggesting that the optimum time for single AI was nearer to 72 h than 96 h after the prostaglandin injection.

Carter and Parsonson (1976) reported a conception rate of 63 per cent after two injections of cloprostenol in Hereford heifers.

Lopez-Barbella *et al.* (1980) obtained a conception rate of 37.51 per cent after two injections of PGF_2 alpha at an interval of 12 days to cows at 45 days post partum.

Deutscher *et al.* (1982) studied oestrus synchronization and pregnancy rates in beef cattle using single injection of PGF_2 alpha. Out of 85 per cent lactating cows which exhibited oestrus, a conception rate of 59 per cent was obtained. Heifers exhibiting oestrus and being pregnant during synchronized breeding tended to be higher for the one injection of PGF_2 alpha than the two injection program (89 vs 77 per cent in oestrus and 49 vs 40 per cent pregnant, respectively).

Plunkett *et al.* (1984) induced oestrus using single injection of 25 mg PGF_2 alpha (lutalyse) after detection of a corpus luteum by ovarian palpation. The cows were inseminated artificially at 72 and 96 h after PGF_2 alpha injection. First service conception rate was 43 per cent, with an overall conception rate of 86 per cent.

In a study conducted by Stevenson *et al.* (1987) 55 per cent of conception rate was obtained in twice inseminated cows (72 and 96 h) and 38 per cent of conception rate in once inseminated cows (80 h), after two injections of PGF_2 alpha.

Mukasa-Mugerwa and Mesfin (1988) administered two doses of 25 mg PGF_2 alpha (Lutalyse), 11 days apart with artificial inseminations at 48, 72 and 80 h after the second injection. None of the animals conceived on insemination at 48 h after treatment. Conception rate with double insemination at 48 and 72 h was 18 per cent and with single insemination at 72 hour was 16 per cent. The highest conception rate obtained was 20 per cent following single insemination at 80 h.

El-Menoufy and Abdou (1989) studied the fertility after double dose of 25 mg dinoprost or 500µg cloprostenol administered 11 days apart. The first service conception rate after double insemination was 52.9 and 53.3 per cent in dinoprost and cloprostenol treated groups respectively.

Mukasa-Mugerwa *et al.* (1989) in his study on Boran cows using single injection of 25 mg of PGF_2 alpha, and inseminated at six, 12 and 18 h after the onset of oestrus, obtained a conception rate of 56, 33 and 33 per cent respectively.

Young (1989) got a conception rate of 68 per cent in cows treated with single injection of 25 mg dinoprost, administered during 14 to 28 days post partum.

Archbald *et al.* (1994) studied effect of PGF_2 alpha treatment on oestrus and pregnancy of lactating dairy cows, by administering 25mg PGF_2 alpha intramuscularly and recorded a pregnancy rate of 24 per cent.

Gaines (1994) treated 129 heifers with 25mg of prostaglandin (Lutalyse) and obtained a conception rate of 70 per cent.

Jacob (1993) reported a conception rate of 52.27 per cent in crossbred cows, induced by 25 mg of PGF_2 alpha (Lutalyse) administered intramuscularly and inseminated twice at an interval of 12-14 h in standing heat.

Stevenson *et al.* (1997) recorded a conception rate of 59.2 per cent in beef cows and heifers after double injection of PGF_2 alpha given 14 days apart.

The conception rates in cows treated with single and double luteolytic dose of PGF_2 alpha were 22 and 23 per cent, respectively, while in untreated control group conception rate at natural oestrus was 24 per cent (Momcilovic *et al.* 1998)

Mughal *et al.* (1998) reported a conception rate of 54.57 and 50 per cent respectively after double and single injections of cloprostenol in Sahiwal cows.

Patterson *et al.* (1998) compared oestrus response and fertility of post partum suckled beef cows after treatment with two injections of PGF_2 alpha 14 days apart and found that 55 per cent of cows exhibited oestrus with a conception rate of 51 per cent.

Britt and Gaska (1998) compared the ovsynch (GnRH + PGF_2 alpha + GnRH) and PGF_2 alpha synchronisation programmes. Fifty eight per cent of cows

responded to PGF_2 alpha treatment with an overall conception rate of 32 per cent, while 100 per cent of cows responded to ovsynch programme with a conception rate of 47 per cent.

Rekwot *et al.* (1999) studied the oestrus synchronisation programme in Zebu cattle of three farms using single injection of PGF_2 alpha. The animals treated were inseminated at 72 and 96 h after PGF_2 alpha injection and obtained a conception rate of 61.4, 45.7 and 46.9 per cent.

Stevenson *et al.* (2000) conducted experiments to induce oestrus in suckled beef cows using two injections of 25mg of PGF_2 alpha (Lutalyse) given 14 days apart. Out of 47.2 per cent of cows detected in oestrus a conception rate of 60.6 per cent was recorded.

The overall postpartum conception rate in suboestrus cows with palpable *corpus luteum* treated with 25 mg of PGF_2 alpha (Lutalyse) intra muscularly was 64 per cent (Gacche *et al.* 2001).

Gopikrishnan (2001) reported a conception rate of 78.57 per cent in crossbred heifers after two doses of 15mg of PGF_2 alpha (Luprostiol) given 11 days apart.

Giving a single intramuscular injection of 25 mg of dinoprost on day 10 of oestrous cycle to crossbred cows with endometritis, Rao *et al.* (2001) reported conception rate of 77.78 per cent.

In a comparative study of two oestrus synchronization programmes using ovysynch (100 µg of GnRH + 500 µg of PGF_2 alpha +100 µg of GnRH) and PGF_2 alpha (500µg of Estrumate), Tallam *et al.* (2001), got a conception rate of 40.3 per cent in ovysynch and 36.2 per cent in PGF_2 alpha treated groups.

Materials and Methods

3. MATERIALS AND METHODS

The material for the present study consisted of crossbred cows and heifers belonging to University Livestock Farm, Mannuthy and those presented at Artificial Insemination Centre and Bull Station, attached to the Department of Animal Reproduction, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, during the period from July 2002 to June 2003. Detailed data regarding the occurrence, onset, duration and intensity of metoestrual bleeding was collected using a suitable proforma. From among these, 32 animals (8 cows and 8 heifers in each group) with the history of metoestrual bleeding were selected randomly and grouped into group I and II. Another 16 animals (8 cows and 8 heifers) without the history of metoestrual bleeding was grouped as group III.

3.1 OCCURRENCE OF METOESTRUAL BLEEDING

History of metoestrual bleeding in the previous cycles of all cows and heifers were collected to determine the occurrence of metoestrual bleeding. The relationship of metoestrual bleeding with age, breed and parity of the animal was studied.

3.2 ONSET OF METOESTRUAL BLEEDING

The time of onset of metoestrual bleeding was noted when the discharge was first noticed from the vulva. The onset of metoestrual bleeding was recorded as on day 0 (day of oestrus), day 1 (24-48 h after onset of oestrus), day 2 (48-72 h after the onset of oestrus) and day 3 (72-96 h after the onset of oestrus).

3.3 DURATION AND INTENSITY OF METOESTRUAL BLEEDING

The period from beginning to the end of metoestrual discharge was recorded as the duration of metoestrual bleeding. The intensity of metoestrual bleeding was graded as scanty, moderate and profuse, based on the quantity of

metoestrual discharge observed after rectal examination and from the history narrated by the owner.

3.4 *CONSISTENCY AND CYTOLOGY OF METOESTRUAL DISCHARGE*

The metoestrual discharge was collected using a sterile pipette to determine the consistency and cytology of the discharge. Smears were prepared with metoestrual discharge on a clean glass slide and stained using Wright's stain (Benjamin, 1985).

3.5 ANTIBIOGRAM OF METOESTRUAL DISCHARGE

A portion of sample collected was streaked on Tryptone Soya Agar and Mc Conkey's Agar by streak plate method in order to get well isolated colonies of organisms present in the sample and incubated at 37 °C for 18 to 24 h.

3.6 EXPERIMENTAL GROUP

Group I

Sixteen animals comprising of 8 cows and 8 heifers with the history of metoestrual bleeding were inseminated at natural oestrus.

Group II

Sixteen animals comprising of 8 cows and 8 heifers with the history of metoestrual bleeding were subjected to induction of oestrum on tenth day using 25 mg PGF_2 alpha intramuscularly (Lutalyse*), when they had a functional corpus luteum as determined by rectal palpation. These animals were inseminated at 72 and 96 h after the administration of Lutalyse.

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* *Lutalyse (Injection): 10 ml (UpJohn). Each ml contains Dinoprost Tromethamine equivalent to Dinoprost 5 mg.*

Group III

Sixteen animals (8 cows and 8 heifers) which did not exhibit metoestrual bleeding were watched for natural oestrus and inseminated.

3.7 OESTRUS RESPONSE

Number of animals responded with exhibition of oestrus after PGF_2 alpha administration.

3.8 TIME TAKEN FOR INDUCTION OF OESTRUS

Group II animals were closely observed for the signs of oestrus every six hours after PGF_2 alpha injection and those found in oestrus were confirmed by rectal palpation. The interval from the administration of PGF_2 alpha to the onset of oestrus was recorded as the time taken for the induction of oestrus.

3.9 DURATION OF OESTRUS

Duration of oestrus was determined by close observation of clinical signs like cervical mucus discharge, vulval oedema, hyperemia of vaginal mucous membrane, bellowing and mounting. The period from the beginning to the end of clinical signs was considered as the duration of oestrus.

3.10 INTENSITY OF OESTRUS

Intensity of oestrus was graded as high, medium and low from clinical and behavoiural signs (Sharma *et al.*, 1968).

3.11 OCCURRENCE AND NATURE OF METOESTRUAL BLEEDING IN OESTRUS INDUCED GROUP

The group II animals were observed for the occurrence of metoestrual bleeding after induction of oestrus using PGF_2 alpha.

3.12 CONCEPTION RATE

Pregnancy was confirmed by per rectal examination on 45-60 days after insemination and overall conception rate was calculated.

3.13 STATISTICAL ANALYSIS

The data obtained were compiled and subjected to statistical analysis as per Snedecor and Cochran (1980).

Results

4. RESULT

Study was carried out to find out the occurrence, onset, duration and intensity of metoestrual bleeding and its effect on fertility in natural and induced oestrus in cattle. Results of the investigation are presented in Tables 1 to 7 and Fig. 1 to 9.

4.1 OCCURRENCE OF METOESTRUAL BLEEDING

Out of 1626 animals (476 heifers and 1150 cows) screened, 99 (6.09 per cent) animals exhibited signs of metoestrual bleeding in one or more oestrous cycles. The incidence was found to be higher in heifers (9.87 per cent) than in cows (4.52 per cent) (Table 1 and Fig. 1 to 3). Higher percentage of Brown Swiss crossbred cattle (9.88 per cent) exhibited signs of metoestrual bleeding followed by Holstein-Friesian crossbred (7.66 per cent) and Jersey crossbreds (7.47 percent).

4.2 ONSET OF METOESTRUAL BLEEDING

Out of 99 animals with metoestrual bleeding, eight (8.08 per cent) animals showed bleeding on the day of oestrus (day 0), 44 (44.44 per cent) on day 1 of oestrus, 26 (26.26 per cent) on day 2 of oestrus and 21 (21.21) on day 3 of oestrus (Table 2 and Fig.4).

4.3 DURATION AND INTENSITY OF METOESTRUAL BLEEDING

Duration of metoestrual bleeding ranged from 6-36 hours with a mean of 13.38 ± 2.64 h. Out of 99 animals with metoestrual bleeding, 13(13.13 per cent) exhibited scanty bleeding, 57 (57.58 per cent) animals showed moderate bleeding and 29 (29.30 per cent) animals evinced profuse bleeding (Table 3 and Fig.5).

4.4 CONSISTENCY AND CYTOLOGY OF METOESTRUAL DISCHARGE

The consistency of metoestrual discharge was graded as thin mucus blood tinged discharge and thick mucus blood tinged discharge. Thin mucus blood tinged discharge was observed in 40 per cent of cases. Thick mucus blood tinged discharge was observed in 60 per cent of cases.

Thick mucus discharge on examination under the low power of the microscope revealed fern pattern, which lay scattered in disintegrated form with presence of RBCs, in stained smears. In thin mucus discharge, fern pattern was not observed, however, plenty of RBCs could be noticed.

4.5 ANTIBIOGRAM OF METOESTRUAL DISCHARGE

Microbial growth was not detected when metoestrual discharge was inoculated on the culture media and incubated at 37°C for 24 h. Gram positive bacilli were detected as contaminants, after 48 h.

4.6 OESTRUS RESPONSE AFTER ADMINISTRATION OF PGF_2ALPHA

Out of eight cows in group II, seven (87.5 per cent) evinced oestrus and one (12.5 per cent) did not show any response to treatment.

In group II heifers all the eight (100 per cent) animals exhibited oestrus symptoms. . The overall oestrus response was 93.75 per cent (Table 4 and Fig.6).

4.7 TIME TAKEN FOR INDUCTION OF OESTRUS

The time taken for the induction of oestrus in group II cows was 66.14 ± 1.55 h as against 59.88 ± 1.66 h in group II heifers (Table 4).

On statistical analysis, there was significant difference ($P<0.01$) in the time of induction of oestrus between cows and heifers in group II.

4.8 DURATION OF OESTRUS

The duration of oestrus in group I cows and heifers were 25.13 ± 0.99 and 18.75 ± 0.53 h, respectively. The duration of oestrus in group II cows was 31.43 ± 1.13 h, while in heifers it was 28.75 ± 2.00 h. Group III cows and heifers had a duration of 23.63 ± 2.27 and 19.38 ± 0.65 h, respectively (Table 5 and Fig.7). Analysis of data revealed significant difference in the duration of oestrus between cows in group II from that of cows in groups I and III ($P<0.01$). Duration of oestrus in heifers in group II were found to be significantly different from that of heifers in groups I and III. Duration of oestrus in induced group (group II) was longer than that of groups I and III.

4.9 INTENSITY OF OESTRUS

In group I out of eight cows, two (25 per cent) cows showed high intensity of oestrus, three (37.5 per cent) showed medium intensity and three (37.5 per cent) showed low intensity of oestrus. Out of eight heifers, one (12.5 per cent) showed high intensity of oestrus, three (37.5 per cent) showed medium intensity and four (50 per cent) showed low intensity of oestrus.

In group II out of seven cows responded, four (57.14 per cent) cows exhibited high intensity of oestrus, two (28.57 per cent) cows exhibited medium intensity and one (14.28 per cent) exhibited low intensity of oestrus. Out of the eight heifers responded, five (62.5 per cent) heifers exhibited high intensity of oestrus, two (25 per cent) exhibited medium intensity and one (12.5 per cent) exhibited low intensity of oestrus.

Out of eight cows in group III, one (12.5 per cent) animal showed high intensity of oestrus, three (37.5 per cent) showed medium intensity and four (50 per cent) had low intensity of oestrus. Out of eight heifers, two (25 per cent) showed high intensity of oestrus, three (37.5 per cent) had medium intensity and three (37.5 per cent) showed low intensity of oestrus (Table 6 and Fig.8).

4.10 OCCURRENCE AND NATURE OF METOESTRUAL BLEEDING IN OESTRUS INDUCED GROUP

Out of eight cows and eight heifers induced, none exhibited metoestrual bleeding in the induced heat.

4.11 CONCEPTION RATE

Conception rate of group I, II and III animals are presented in Table 7 and Fig.9).

In group I, out of eight cows and eight heifers, one (12.5 per cent) cow conceived to first insemination and one (12.5 per cent) to second insemination while one (12.5 per cent) heifer conceived to second insemination. The overall conception rate in group I was 18.75 per cent.

In group II animals, out of 7 cows responded, three (42.86 per cent) cows conceived at first insemination and one (14.29 per cent) with second insemination. Out of 8 heifers, four (50 per cent), one (12.5 per cent) and one (12.5 per cent) heifers conceived at first, second and third inseminations, respectively. The overall conception rate was 66.66 per cent.

In group III, out of 8 cows two (25 per cent) required single insemination and two (25 per cent) required three inseminations while in heifers two (25 per cent) required single insemination and one (12.5 per cent) required two inseminations with an overall conception rate of 43.75 per cent.

Table 1. Occurrence of metoestrual bleeding among crossbred cattle

	Cow	Heifer	Total
Number of animals screened	1150	476	1626
Number of animals with metoestrual bleeding	52	47	99
Percentage of animals exhibited metoestrual bleeding	4.52%	9.87%	6.09%

Table 2. Onset of metoestrual bleeding

	Number	Per cent
Bleeding on the day of oestrus (0-<24 hours)	8	8.08
Bleeding on day 1 of oestrus (24-<48 hours)	44	44.44
Bleeding on day 2 of oestrus (48-<72 hours)	26	26.26
Bleeding on day 3 of oestrus (>72 hours)	21	21.21

Table 3. Duration and intensity of metoestrual bleeding

Duration of metoestrual bleeding		Intensity of metoestrual bleeding					
Range (hours)	Mean ± SE (hours)	Scanty		Moderate		Profuse	
		Number	Per cent	Number	Per cent	Number	Per cent
6-36	13.38 ± 2.64	13	13.13	57	57.58	29	29.30

Table 4. Response to oestrus after administration of PGF_2 alpha in group II animals

Group II	Number of animals examined	Response to treatment			Time taken for induction of oestrus (hours)	Duration of oestrus (hours)
		Number	Per cent	Overall per cent		
Cows	8	7	87.5	93.75	66.14 ± 1.55	31.43 ± 1.13
Heifers	8	8	100		59.88 ± 1.66	28.75 ± 2.00

Table 5. Duration of oestrus in group I, II and III animals

Treatment groups		Number of animals	Duration of oestrus (hours)
Group I	Cow	7	25.13 ± 0.99
	Heifer	8	18.75 ± 0.53
Group II	Cow	8	31.43 ± 1.13
	Heifer	8	28.75 ± 2.00
Group III	Cow	8	23.63 ± 2.27
	Heifer	8	19.38 ± 0.65

Table 6. Intensity of oestrus in group I, II and III animals

Treatment groups		Number of animals	Intensity of oestrus					
			High		Medium		Low	
			Number	Per cent	Number	Per cent	Number	Per cent
Group I	Cow	8	2	25	3	37.5	3	37.5
	Heifer	8	1	12.5	3	37.5	4	50
Group II	Cow	7	4	57.14	2	28.57	1	14.28
	Heifer	8	5	62.5	2	25	1	12.5
Group III	Cow	8	1	12.5	3	37.5	4	50
	Heifer	8	2	25	3	37.5	3	37.5

Table 7. Conception rate in I, II and III groups

Treatment Groups		No. & per cent of animals conceived to first insemination	No. & per cent of animals conceived to second insemination	No. & per cent of animals conceived to third insemination	Conception rate (per cent)	Overall conception rate (per cent)
Group I (with metoestrual bleeding)	Cow	1 (12.5)	1 (12.5)	Nil	25	18.75
	Heifer	Nil	1 (12.5)	Nil	12.5	
Group II (induced group)	Cow	3 (42.86)	1 (14.29)	Nil	57.14	66.66
	Heifer	4 (50)	1 (12.5)	1 (12.5)	75	
Group III (control group)	Cow	2 (25)	Nil	2 (25)	50	43.75
	Heifer	2 (25)	1 (12.5)	Nil	37.5	

Figures in parenthesis indicate per cent

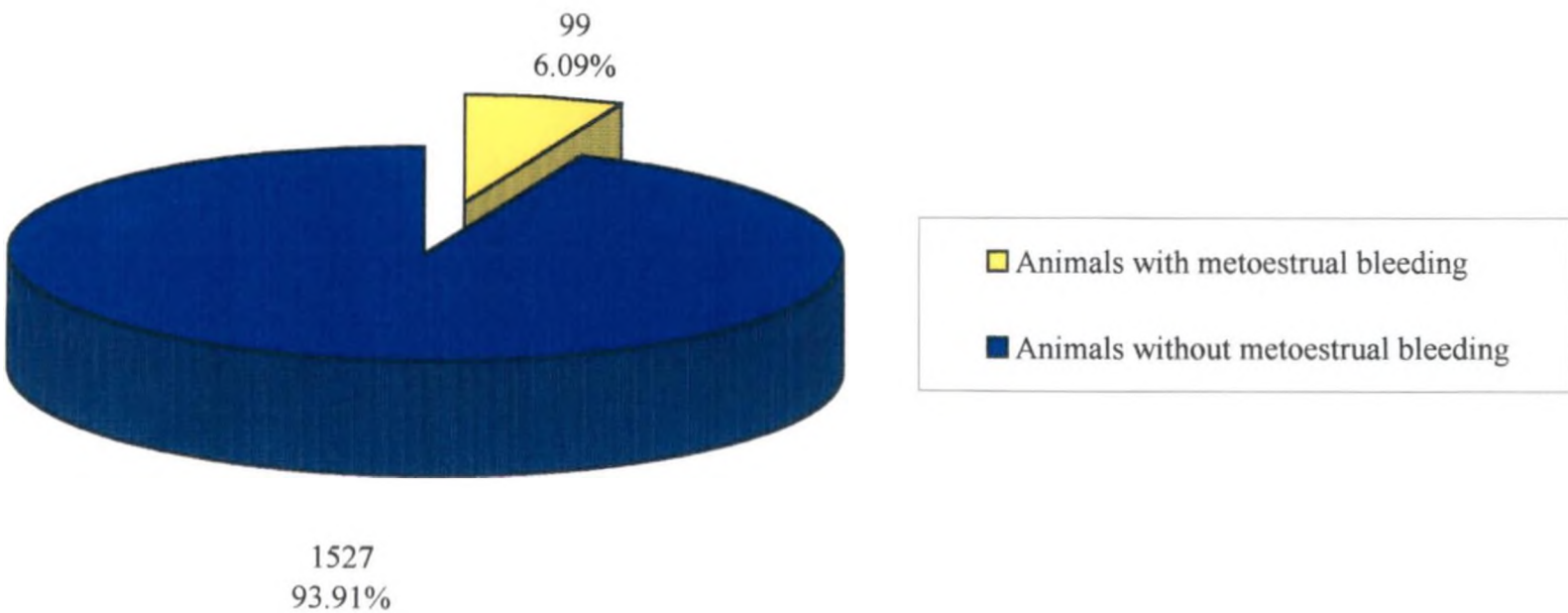

Fig.1 Occurrence of metoestrual bleeding in cows and heifers

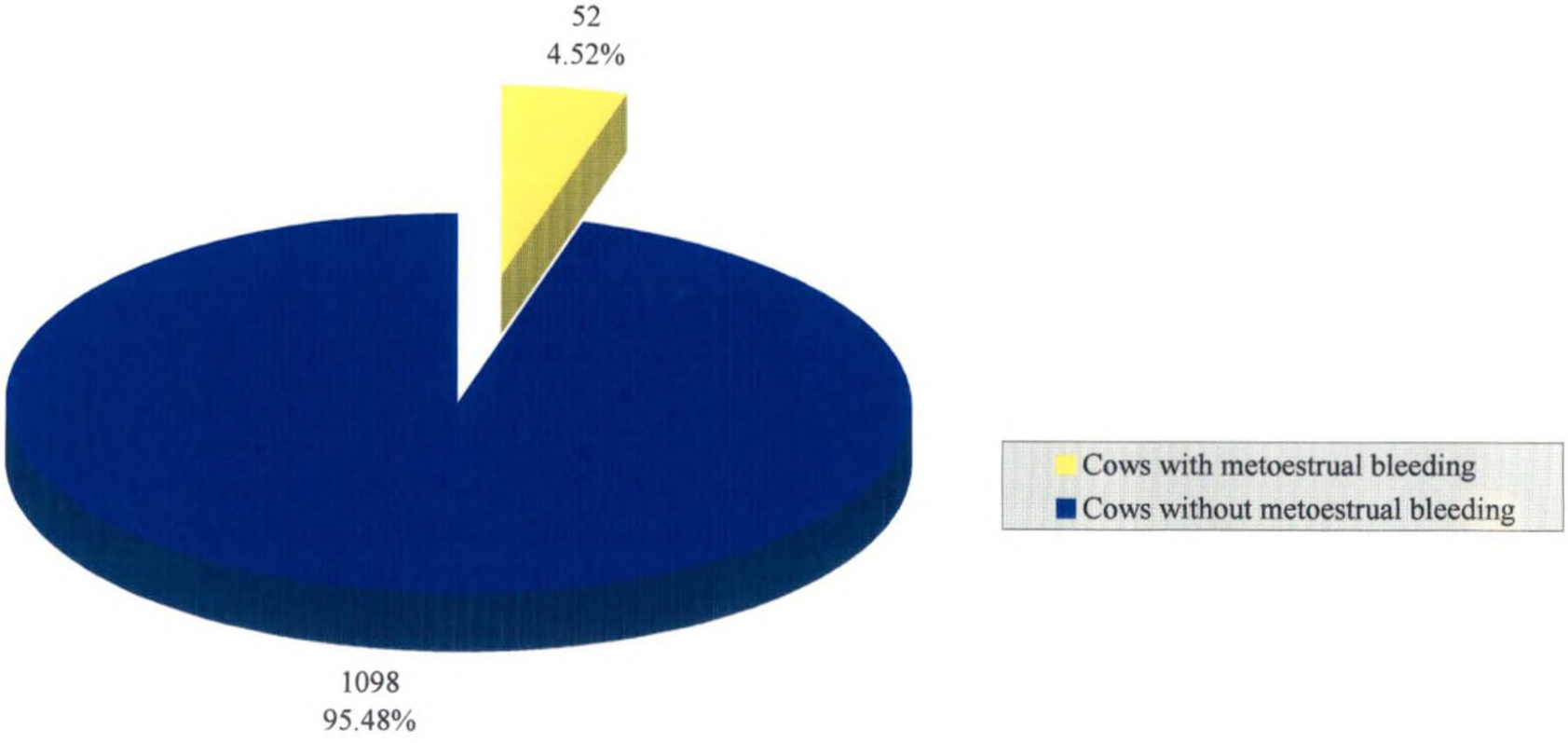

Fig. 2 Occurrence of metoestrual bleeding in cows

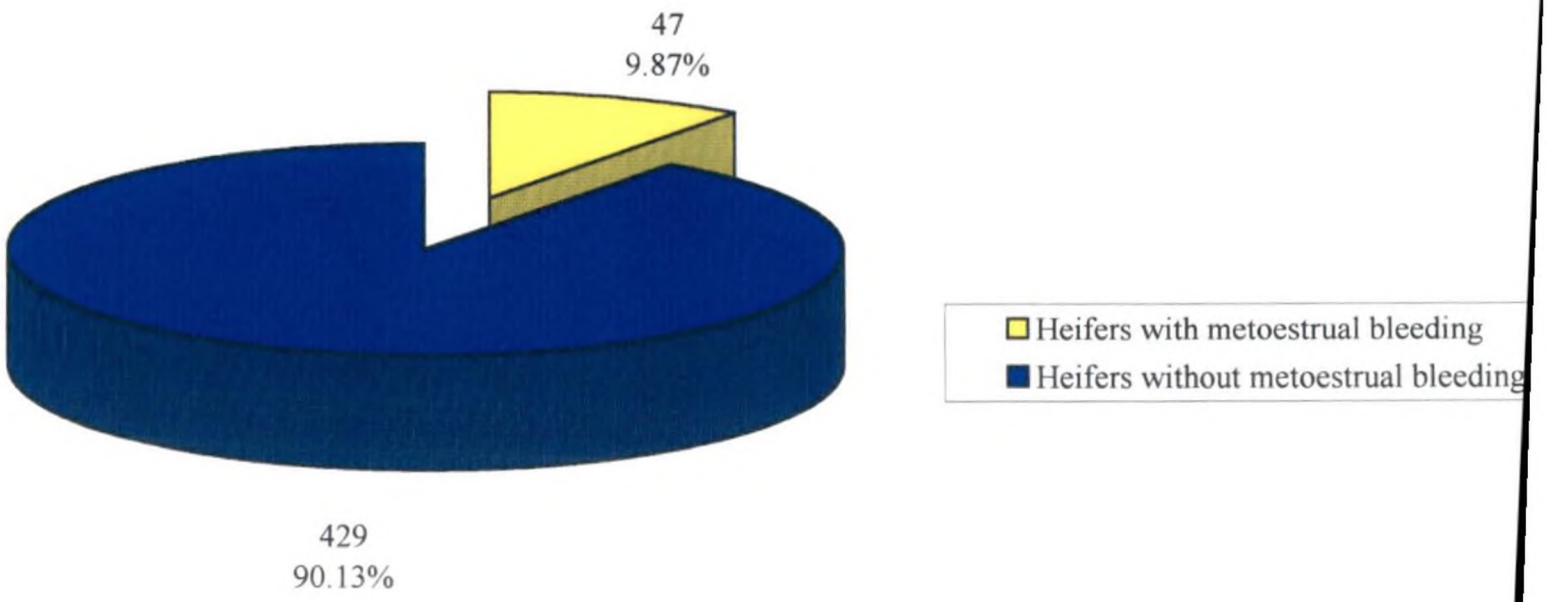

Fig.3 Occurrence of metoestrual bleeding in heifers

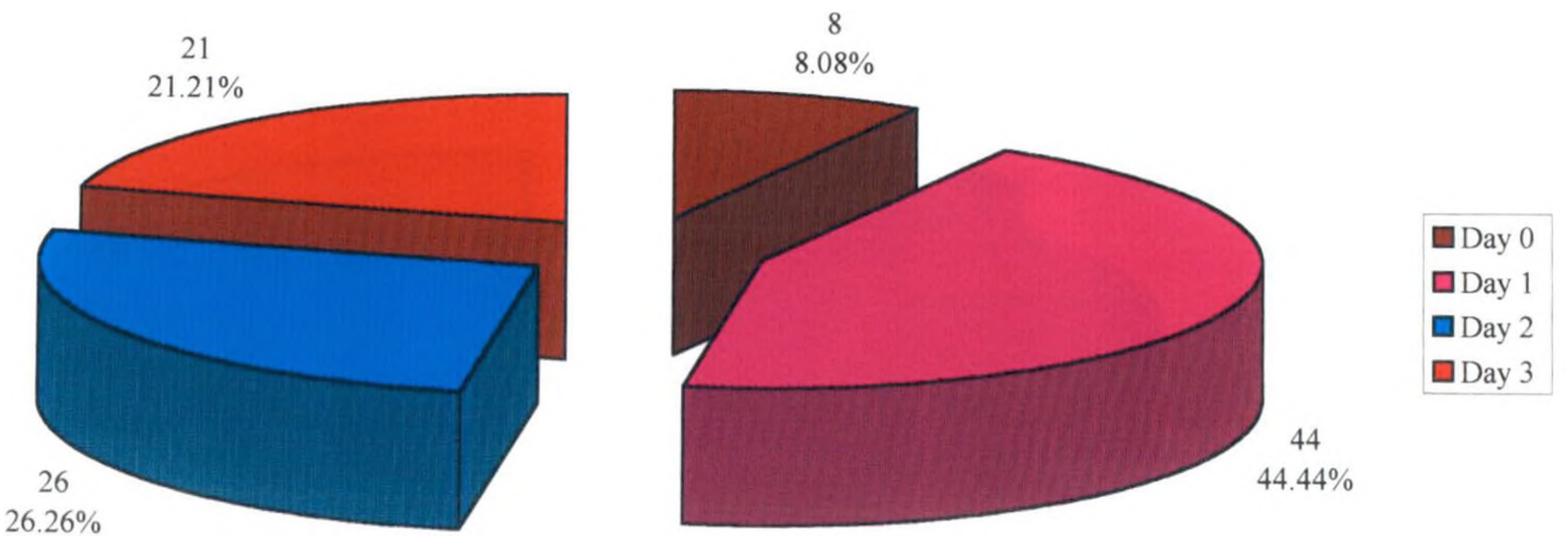

Fig.4 Onset of metoestrual bleeding

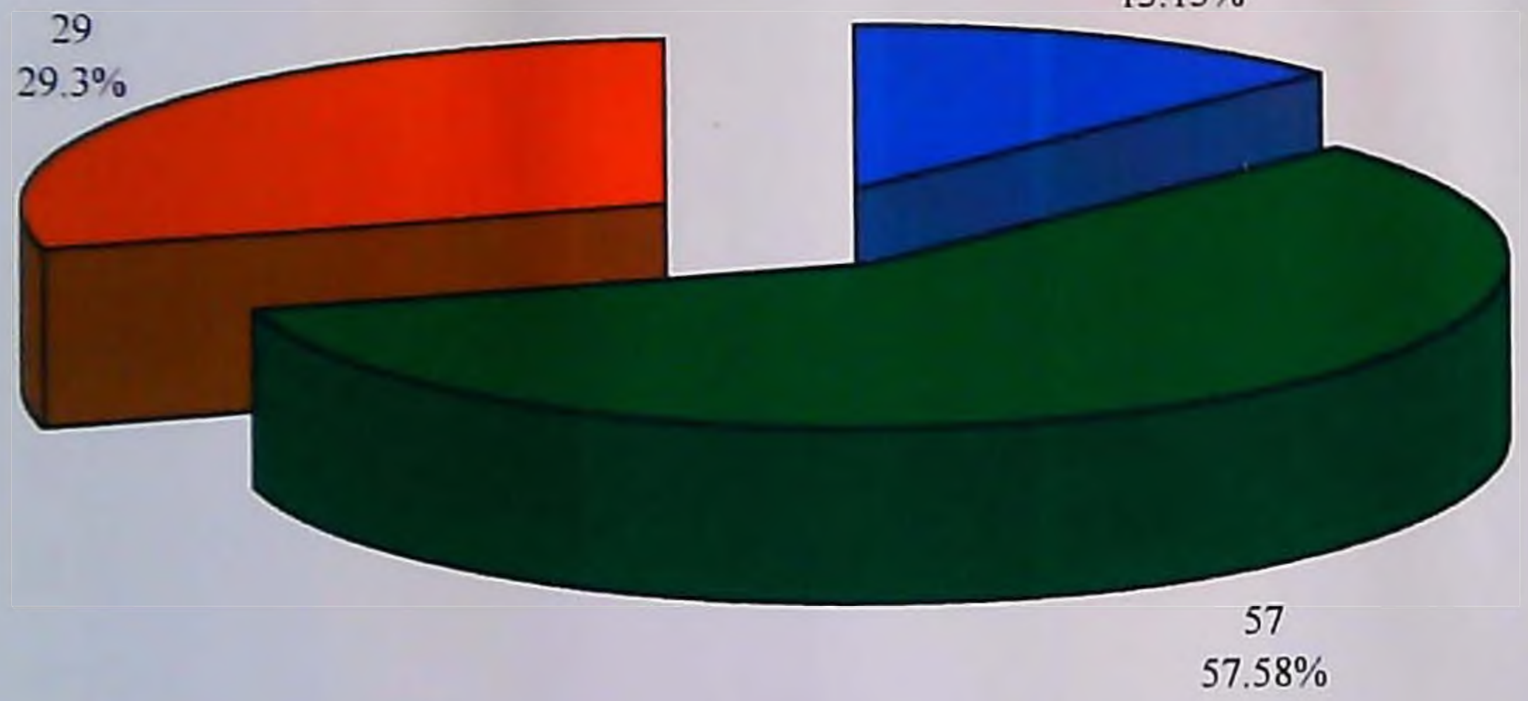

Fig.5 Intensity of metoestrual bleeding

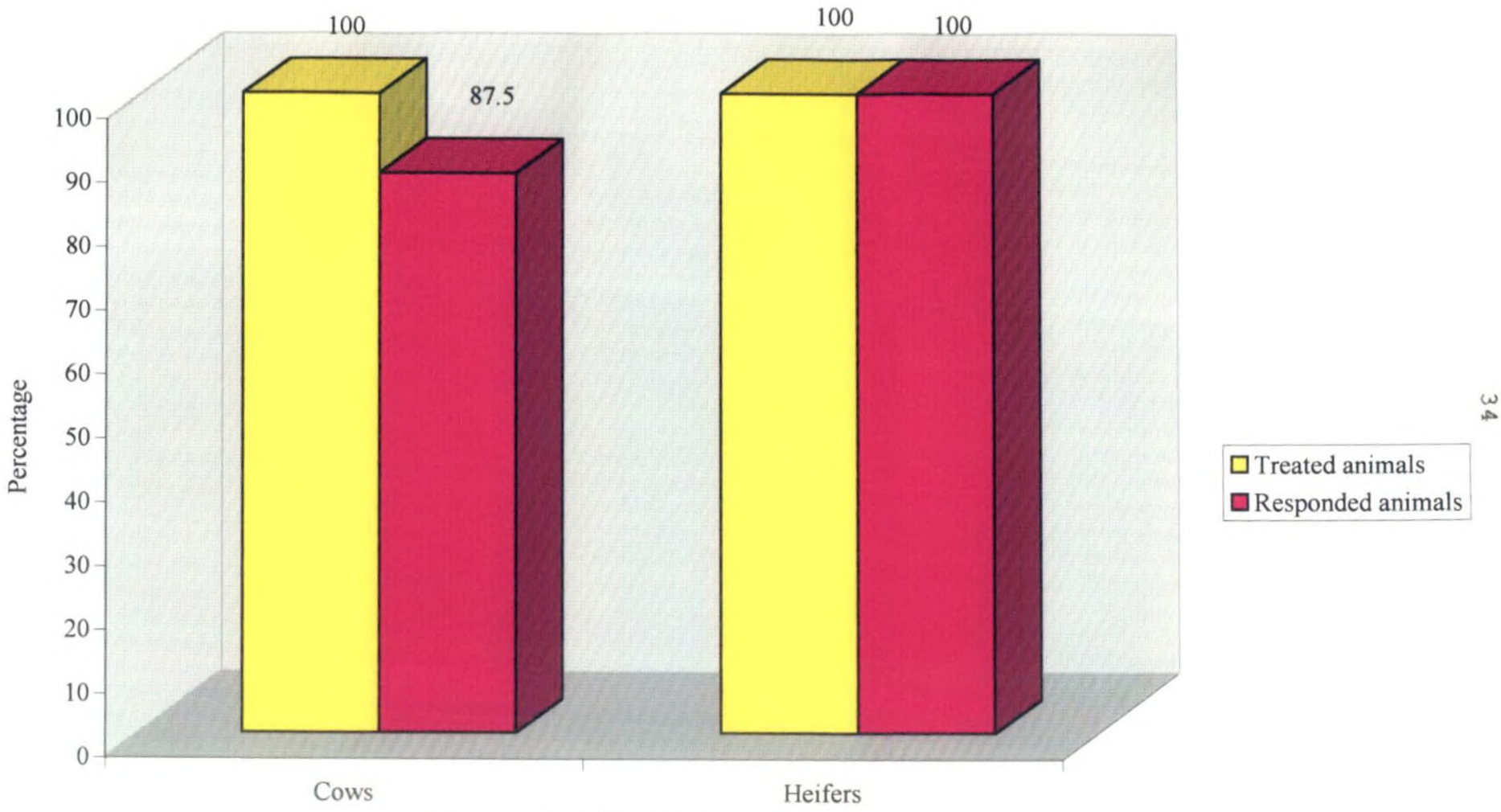

Fig. 6 Response to PGF2alpha treatment

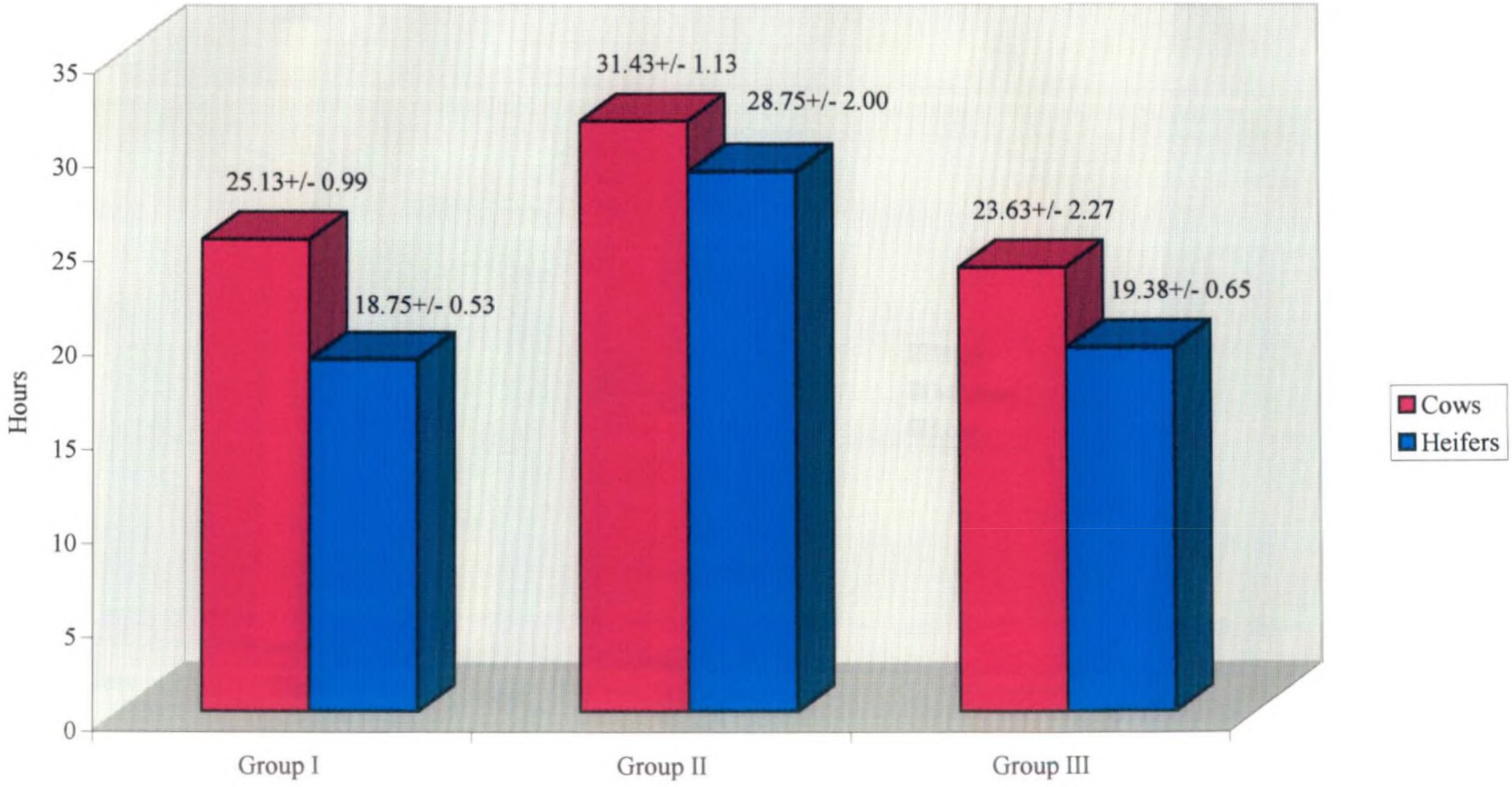

Fig.7 Duration of oestrus in group I,II and III animals

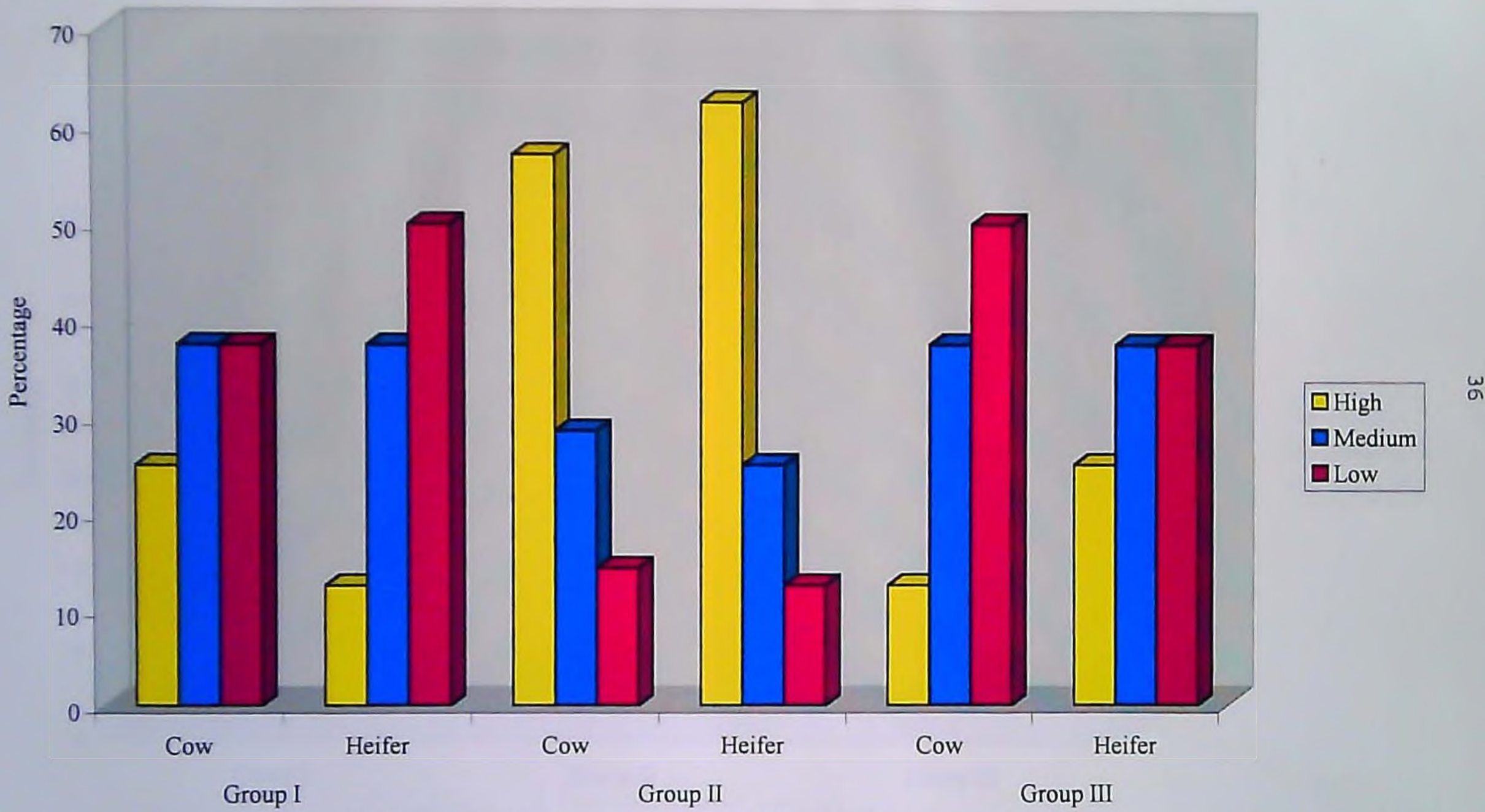

Fig.8 Intensity of oestrus in group I, II and III animals

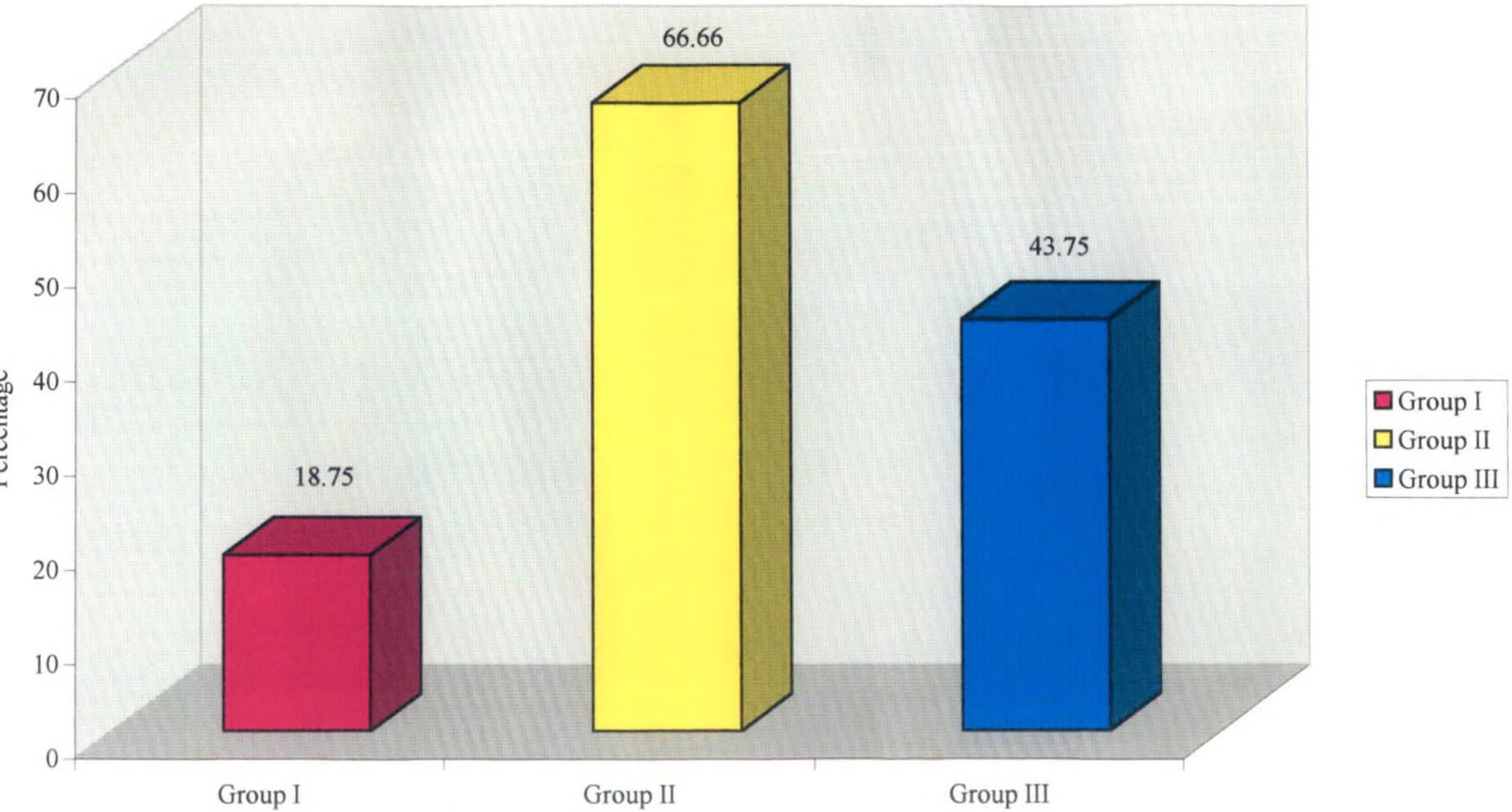

Fig.9 Conception rate in various groups

Discussion

5. DISCUSSION

The present investigation was undertaken with the object of studying the occurrence, onset, duration and intensity of metoestrual bleeding and its effect on fertility in natural and induced oestrus in cattle.The study was aimed to find the beneficial effects of prostaglandin and its structural analogues in the management of oestrous cycle in cycling cows and heifers with the history of metoestrual bleeding.

5.1 OCCURRENCE OF METOESTRUAL BLEEDING

Perusal of data presented in table 1 revealed that 6.09 per cent of cows and heifers exhibited signs of metoestrual bleeding in one or more oestrous cycles. Occurrence was found to be higher in heifers (9.87 per cent) than in cows (4.52 per cent). The overall incidence reported here was higher than that reported by Mathai and Raja, 1973; Sobhanan, 1978; Quayam and Austin, 1983 and Janardhanan and Nair, 1990. Higher occurrence of metoestrual bleeding in the present study was probably due to the increased level of exotic inheritance due to intensive cross breeding programme. However, a lower level was observed as compared to those reported in exotic animals by Trimberger, 1941.

In the present study higher percentage of Brown Swiss crossbred cattle (9.88 per cent) exhibited signs of metoestrual bleeding, followed by Holstein Friesian crossbred (7.66 per cent) and Jersey cross bred (7.47 per cent). Muller (1999) reported higher incidence of metoestrual bleeding in Holstein Friesian crossbred animals. However, variation in the occurrence of metoestrual bleeding in various breeds was not significant.

5.2 ONSET OF METOESTRUAL BLEEDING

Eight (8.08 per cent) animals showed bleeding on the day of oestrus (day 0), 44 (44.44 per cent) on day 1 of oestrus, 26 (26.26 per cent) on day 2 of oestrus and

21 (21.21) on day 3 of oestrus. More number of animals evinced bleeding on day 1 of oestrus (24 to 48 hours after the onset of oestrus). In a study by Janardhanan and Nair (1990) out of the 74 animals showing metoestrual bleeding, 15 (20.25 per cent) showed bleeding on the day of oestrus (0 day), 30 (40.5 per cent) on day 1 of oestrus, 15 (20.25 per cent) on day 2 of oestrus and 14 (19 per cent) on day 3 of oestrus. Higher percentage of animals showed bleeding on day 1 of oestrus which is in agreement with the findings of the present study. Observations here are in agreement with Deshpande (1994) who stated that post oestrual bleeding in cows occurs 12 to 24 hours after the cessation of oestrus.

5.3 DURATION AND INTENSITY OF METOESTRUAL BLEEDING

Duration of metoestrual bleeding ranged from 6-36 hours with a mean of 13.38 ± 2.64 hours. Scanty, moderate and profuse bleeding was exhibited by 13(13.13 per cent), 57 (57.58 per cent) and 29 (29.30 per cent) animals, respectively. There is paucity of information regarding the duration and intensity of metoestrual bleeding. In a clinical note on metoestrual bleeding, Janardhanan and Nair (1990) stated the intensity of metoestrual bleeding as scanty (17.6 per cent), moderate (55.4 per cent) and profuse (27 per cent). The data in the present study are in close agreement with that of Janardhanan and Nair (1990).

5.4 CONSISTENCY AND CYTOLOGY OF METOESTRUAL DISCHARGE

Thick mucus blood tinged discharge was observed in 60 per cent of cases whereas thin mucus blood tinged discharge was observed in 40 per cent of cases.

Thick mucus discharge on microscopic examination revealed fern pattern which lay scattered in disintegrated form with presence of RBCs in stained smears. While in thin mucus discharge, fern like pattern was not observed, plenty of RBCs could be noticed. Hussain and Khan (1978) observed cellular debris; leucocytes and parabasal cells in the vaginal scrapings of cattle collected during metoestrum. Kurade *et al.*, (1993) reported that most of the smears of cervicovaginal mucus collected during metoestrum were characterised by the

presence of RBCs. The present study is in agreement with Kurade *et al.*, (1993) who described that the presence of RBCs in the mucus collected during metoestrum was due to diapedesis or capillary bleeding from the endometrium.

5.5 ANTIBIOGRAM OF METOESTRUAL DISCHARGE

Microbial growth could not be detected for 24 h when metoestrual discharge was inoculated to culture media. Gram positive bacilli were detected as contaminants after 48 h of incubation. Literature on culture and sensitivity of cervical mucus during oestrus of normal and repeat breeder animals are available, but data regarding the microbiological studies of metoestrual discharge are scarce.

5.6 OESTRUS RESPONSE AFTER PGF_2ALPHA TREATMENT

Out of eight cows and eight heifers in group II subjected to PGF_2alpha administration, seven (87.5 per cent) and eight (100 per cent), respectively, responded to treatment. Nair and Madhavan (1984) observed that 92 per cent of cows evinced oestrus response after administration of 500 µg of Estrumate. In a study conducted by El-Menoufy and Abdou (1989) 82 and 90 per cent of cows were detected in oestrus after double dose of 25 mg dinoprost or 500µg cloprostenol administered 11 days apart. Similar results were obtained by Jacob *et al.*, 1995; Chauhan *et al.*, 1994 and Madhav *et al.*, 2001. A lowered oestrus response of 55 per cent and 50 per cent were reported by Archbald *et al.*, 1994 and Momcilovic *et al.*, 1998, respectively. In the present study an overall oestrus response of 93.75 per cent was obtained. This data shows that the PGF_2 alpha analogue, dinoprost is effective in inducing oestrus in cross bred cows and heifers with the history of metoestrual bleeding.

5.7 TIME TAKEN FOR INDUCTION OF OESTRUS

In the present study, the time taken for induction of oestrus after treatment in group II cows and heifers were 66.14 ± 1.55 h and 59.88 ± 1.66 h, respectively. On statistical analysis, significant difference was observed ($P<0.01$) in the time of

induction of oestrus between cows and heifers. Nair and Madhavan (1984) reported an interval of 48 to 72 h for induction of oestrus after administration of Estrumate, a PGF_2 alpha analogue in crossbred cows. Pawshe *et al.*, (1991) stated that the average time taken for the onset of oestrus in crossbred cows was 71.87 ± 1.25 h. Jacob *et al.*, (1995) reported an average of 61.81 hours for the induction of oestrus after PGF_2 alpha injection. Pinheriro *et al.*, (1998) observed that the time taken for induction of oestrus in heifers was 82.5 ± 4.7 h after the second injection of PGF_2 alpha while cows manifested oestrus at 70.5 ± 4.8 h. Rao *et al.*, (2001) reported that the average time taken for induction of oestrus after single intramuscular injection of 25 mg PGF_2 alpha analogue dinoprost was 72.44 h. Variation in the time taken for the induction of oestrus reported by different workers could be due to difference in the stage of corpora lutea at the time of administration of prostaglandin and difference in dose and potency of PGF_2 alpha analogue.

5.8 DURATION OF OESTRUS

Average duration of oestrus in cows and heifers was 25.13 ± 0.99 and 18.75 ± 0.53; 31.43 ± 1.13 and 28.75 ± 2.00, and 23.63 ± 2.27 and 19.38 ± 0.65 h for groups I, II and III, respectively. Analysis of data revealed significant difference in the duration of oestrus in cows and heifers in group II from that of cows and heifers in groups I and III ($P<0.01$). Duration of oestrus in induced group (group II) was longer than that of groups I and III. In a similar study, Jacob *et al.*, (1995) observed a longer duration of oestrus in prostaglandin induced animals, compared to normal animals. However, Nair and Madhavan (1984) observed that the duration of oestrus induced by PGF_2 alpha did not show marked variation from normal oestrus in crossbred cows. The response to prostaglandin treatment in terms of onset of oestrus and duration of oestrus was reported to be variable, especially in lactating cows, irrespective of the schedule of administration.

5.9 INTENSITY OF OESTRUS

Data presented in table 6 reveal that 25, 37.5 and 37.5 per cent of cows and 12.5, 37.5 and 50 per cent heifers in group I showed high, medium and low

intensity of oestrus, respectively. In group II the corresponding values were 57.14, 28.57 and 14.28 per cent in cows and 62.5, 25 and 12.5 per cent in heifers. In group III 12.5, 37.5 and 50 per cent cows and 25, 37.5 and 37.5 per cent heifers exhibited high, medium and low intensity of oestrus, respectively. Majority of animals in group II induced with prostaglandin showed medium to high intensity of oestrus than the other two groups. Jacob *et al.*, (1995) reported intense, medium and weak oestrus as 66.66. 19.04 and 14.28 per cent , respectively, after induction with prostaglandin. Majority of animals in group II showed intense signs of oestrus after prostaglandin treatment probably due to complete luteal regression and better folliculogenesis.

5.10 OCCURRENCE OF METOESTRUAL BLEEDING IN OESTRUS INDUCED GROUP

In prostaglandin treated group II animals, none of the animals exhibited signs of metoestrual bleeding. Salisbury *et al.*, (1978) stated that the primary cause for metoestrual bleeding was the sudden withdrawal of oestrogen following ovulation. This may result in congested endometrial vessels beginning to break down and small quantity of blood is discharged into the uterine lumen, exhibited as metoestrual bleeding. In the present study, prostaglandin treated animals showed intense heat signs with increased duration of oestrus, probably due to prolonged oestrogen stimulation. In these animals, a sharp decline in oestrogen level might not have occurred as in normal animals, which might explain the reason for absence of metoestrual bleeding in induced group.

5.11 CONCEPTION RATE

In group I, two (25 per cent) cow and one heifer (12.5 per cent) conceived, with an overall conception rate of 18.75 per cent. Sobahnan (1978) reported an overall conception rate of 20 per cent in animals with metoestrual bleeding. Quayam and Austin (1983) observed a conception rate of 17.64 per cent in heifers and 25 per cent in cows with metoestrual bleeding, while the overall conception rate was 20.68 per cent. Sandhu (1992) reported a lower conception rate of 18.75

per cent in animals with metoestrual bleeding. It was observed that the conception rate was much lower in animals that showed metoestrual bleeding than in those which did not show metoestrual bleeding, probably due to exhibition of mild oestrus and single day insemination was not enough to bring about adequate conception rate. The observations in the present study are in agreement with Sobahnan, 1978; Quayam and Austin, 1983 and Sandhu 1992. Muller (1999) opined that post oestrual bleeding could be regarded as hormonal dysfunction due to progesterone deficiency, as confirmed by milk progesterone assay in animals with metoestrual bleeding.

In group II oestrus induced animals, out of seven cows and eight heifers responded, four (57.14 per cent) cows and six (75 per cent) heifers conceived with an overall conception rate of 66.66 per cent. Oxender *et al.*, (1974) obtained a conception rate of 78.94 per cent in cows, after administration of 30 mg PGF_2alpha intramuscularly and inseminated at 70 and 88 h. Carter and Parsonson (1976) reported a conception rate of 63 per cent after induction of oestrus with prostaglandin in Hereford heifers. An enhanced conception rate of 84.6 per cent was obtained by using progestational preparations for enhancing conception rate in animals with metoestrual bleeding (Neduncheralathan *et al.*, 1981). Young (1989) recorded a conception rate of 68 per cent in cows treated with single injection prostaglandin. Gaines (1994) reported 70 per cent conception rate in heifers after oestrus induction with Prostaglandin analogue. An increase in conception rate with prostaglandin might be attributed to high intensity of induced oestrus and fixed time insemination at 24 h interval. Fertility of heifers after oestrus synchronisation with prostaglandin has been found to be normal or even higher (Macmillan and Day, 1982 and Mc Intosh *et al.*, 1984). However the results on fertility following prostaglandin treatment have been inconsistent in dairy cows (Macmillan, 1983).

In group III animals, four (50 per cent) cows and three (37.5 per cent) heifers conceived with an overall conception rate of 43.75 per cent. An increased conception rate in group III animals as compared to animals with the history of metoestrual bleeding (group I) is in agreement with the findings of Sobahnan,

1978; Quayam and Austin, 1983 and Sandhu 1992. Higher conception rate in group II animals is due to high intensity of oestrus and fixed time insemination at 24 h interval.

From the present study it can be inferred that the treatment with prostaglandin was highly effective in inducing oestrus in animals with the history of metoestrual bleeding and reasonable conception rate could be obtained. However, more studies are required to establish the relationship between the hormonal profile and incidence of metoestrual bleeding. Further detailed microbial studies could also help to find a suitable remedial measure to control metoestrual bleeding so as to improve the fertility in these animals.

Summary

6. SUMMARY

The objective of the present study was to find out the occurrence, onset, duration and intensity of metoestrual bleeding and its effect on fertility in natural and induced oestrus in cattle.

The material for the present study consisted of crossbred cows and heifers belonging to University Livestock Farm, Mannuthy and those presented at Artificial Insemination Centre and Bull Station, attached to the Department of Animal Reproduction, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, during the period from July 2002 to June 2003. Detailed data regarding the occurrence, onset, duration and intensity of metoestrual bleeding were collected using a suitable proforma. Animals with the history of metoestrual bleeding were randomly selected and allotted to the following groups. Group I consisted of 16 animals, comprising of eight cows and eight heifers, with the history of metoestrual bleeding which were inseminated at natural oestrus. Sixteen animals in group II (eight cows and eight heifers) with the history of metoestrual bleeding were subjected to induction of oestrus on tenth day using 25 mg PGF_2 alpha (Lutalyse), when they had a functional corpus luteum as determined by rectal palpation. These animals were inseminated at 72 and 96 h after the administration of Lutalyse. Sixteen animals (eight cows and eight heifers) which did not show metoestrual bleeding formed the group III.

Out of 1626 animals screened, 99 (6.09 per cent) animals exhibited signs of metoestrual bleeding in one or more oestrous cycles. The occurrence was found to be higher in heifers (9.87 per cent) than in cows (4.52 per cent). Higher percentage of Brown Swiss cross bred cattle (9.88 per cent) exhibited signs of metoestrual bleeding, followed by Holstein-Friesian cross bred (7.66 per cent) and Jersey cross breds (7.47 per cent).

Out of 99 animals with metoestrual bleeding, eight (8.08 per cent) animals showed bleeding on the day of oestrus (day 0), 44 (44.44 per cent) on day 1 of oestrus, 26 (26.26 per cent) on day 2 of oestrus and 21 (21.21) on day 3 of oestrus.

Duration of metoestrual bleeding ranged from 6-36 h with a mean of 13.38 ± 2.64 h. Out of 99 animals with metoestrual bleeding, 13(13.13 per cent) exhibited scanty bleeding, 57 (57.58 per cent) animals showed moderate bleeding and 29 (29.30 per cent) animals evinced profuse bleeding.

Thick mucus blood tinged discharge was observed in 60 per cent of cases whereas thin mucus blood tinged discharge was observed in 40 per cent of cases. Thick mucus discharge on microscopic examination revealed fern pattern, which lay scattered in disintegrated form with presence of RBC's in stained smears. In thin mucus discharge, fern like pattern was not observed, but plenty of RBCs could be noticed.

Microbial growth was not detected when metoestrual discharge was inoculated on the culture media and incubated at 37°C for 24 h. Gram positive bacilli were detected as the contaminants after 48 h.

Out of eight cows and eight heifers in group II subjected to PGF_2alpha administration, seven (87.5 per cent) and eight (100 per cent) responded to treatment, respectively. The overall oestrus response was 93.75 per cent. The time taken for the induction of oestrus in cows was 66.14 ± 1.55 h as against 59.88 ± 1.66 in group II heifers. Average duration of oestrus in cows and heifers was 25.13 ± 0.99 and 18.75 ± 0.53; 31.43 ± 1.13 and 28.75 ± 2.00, and 23.63 ± 2.27 and 19.38 ± 0.65 h for groups I, II and III, respectively. Analysis of data revealed significant difference in the duration of oestrus in cows in group II from that of cows in groups I and III ($P<0.01$). Duration of oestrus in heifers in group II were found to be significantly different from that of heifers in groups I and III. In group I, 25, 37.5 and 37.5 per cent of cows and 12.5, 37.5 and 50 per cent heifers showed high, medium and low intensity of oestrus, respectively. In group II the corresponding values were 57.14, 28.57 and 14.28 per cent in cows and 62.5, 25

and 12.5 per cent in heifers. In group III 12.5, 37.5 and 50 per cent cows and 25, 37.5 and 37.5 per cent heifers exhibited high, medium and low intensity of oestrus, respectively. Out of eight cows and eight heifers induced, none exhibited metoestrual bleeding in the induced heat.

In group I, two (25 per cent) cows and one heifer (12.5 per cent) conceived with an overall conception rate of 18.75 per cent. Out of seven cows and eight heifers responded in group II, four (57.14 per cent) cows and six (75 per cent) heifers conceived with an overall conception rate of 66.66 per cent. In group III animals, four (50 per cent) cows and three (37.5 per cent) heifers conceived with an overall conception rate of 43.75 per cent.

In the present study, occurrence of metoestrual bleeding was 6.09 per cent in cows and heifers and their conception rate was only 18.75 per cent. Treatment with prostaglandin was highly effective in inducing oestrus in these animals and the conception rate was enhanced to 66.66 per cent. Hence it can be recommended that induction of oestrus using prostaglandin can be employed for enhancing the conception rate in animals with the history of metoestrual bleeding.

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METOESTRUAL BLEEDING AND ITS EFFECT ON FERTILITY IN NATURAL AND INDUCED OESTRUS IN CATTLE

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Abstract of a thesis submitted in partial fulfilment of the requirement for the degree of

Master of Veterinary Science

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ABSTRACT

With the objective of studying the occurrence, onset, duration and intensity of metoestrual bleeding and its effect on fertility in natural and induced oestrus in cattle, detailed data of animals belonging to University Livestock Farm and those presented at Artificial Insemination Centre and Bull Station, attached to the Department of Animal Reproduction, College of Veterinary and Animal Sciences, Mannuthy, were collected using a suitable proforma. Animals with the history of metoestrual bleeding were randomly selected and allotted to the following groups. Group I consisted of 16 animals, comprising of eight cows and eight heifers, with the history of metoestrual bleeding that were inseminated at natural oestrus. Sixteen animals in group II (eight cows and eight heifers) with the history of metoestrual bleeding were subjected to induction of oestrus on tenth day using 25 mg PGF_2 alpha (Lutalyse), which were inseminated at 72 and 96 h after the administration of Lutalyse. Sixteen animals (eight cows and eight heifers) which did not show metoestrual bleeding formed the group III.

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The overall conception rate in groups I, II and III were 18.75, 66.66 and 43.75 per cent, respectively. It can be recommended that induction of oestrus using prostaglandin can be employed for enhancing the conception rate in animals with the history of metoestrual bleeding.