The aim of the present study was to evaluate the response of postpartum anestrus cow to progesterone implant and PGF$_{2\alpha}$ on oestrus cyclicity. None of the animals were divided in six groups having six animals in each group. Animals of group I received normal saline solution (2ml, I/M) on day 5 and day 15 and they served as control. The animals of groups II were received 1ml Duraprogen TM (250 mg progesterone) I/M after prostaglanding injection. Animals of group III received 1st first dose of 1ml Duraprogen TM on day 5 and second dose of 1ml Duraprogen TM on day 15. Animals of group IV were received 2ml Duraprogen TM (500 mg) progesterone on day 5 after prostaglandin injection while the animals of group V were received 1st dose of 2 ml Duraprogen TM on day 5 and second dose of 2ml Duraprogen TM on 15 respectively. Animals of group VI received 3 ml Duraprogen TM (750 mg progesterone) I/M on day 15 after Fenbendazole administration. For the animals of group of VI Prostaglandin (Lutalyse, 25 mg) was administered 8th day after Duraprogen TM administration. Blood samples were collected from each animal on day 0 just before PGF$_{2\alpha}$ administration and on day 5, day 15 and day of oestrus in group I to V and from the animals of group VI on the day of progesterone and prostaglandin administration and on day of oestrus. None of the animals in control group was detected in oestrus while one animal of group II, six animal of group V and group VI on the day of progesterone and prostaglandin administration. All the animals were divided in six groups having six animals in each group. Animals of group I received normal saline solution (2ml, I/M) on day 5 and day 15 and they served as control. The animals of groups II were received 1ml Duraprogen TM (250 mg progesterone) I/M after prostaglanding injection. Animals of group III received 1st first dose of 1ml Duraprogen TM on day 5 and second dose of 1ml Duraprogen TM on day 15. Animals of group IV were received 2ml Duraprogen TM (500 mg) progesterone on day 5 after prostaglandin injection while the animals of group V were received 1st dose of 2 ml Duraprogen TM on day 5 and second dose of 2ml Duraprogen TM on 15 respectively. Animals of group VI received 3 ml Duraprogen TM (750 mg progesterone) I/M on day 15 after Fenbendazole administration. For the animals of group of VI Prostaglandin (Lutalyse, 25 mg) was administered 8th day after Duraprogen TM administration. Blood samples were collected from each animal on day 0 just before PGF$_{2\alpha}$ administration and on day 5, day 15 and day of oestrus in group I to V and from the animals of group VI on the day of progesterone and prostaglandin administration and on day of oestrus. None of the animals in control group was detected in oestrus while one animal of group II, six animal of group V and two animal of group VI were detected in oestrus 2 to 16 days after the last duraprogen injection. It was concluded that the crossbred cows having normal genitalia and sound health can be brought in to fertile oestrus during summer months by a combined treatment with 25 mg progostaglan and 500 mg to 750 mg progesterone.

Keywords: Cow, postpartum anestrus, cyclicity, progesterone implant and PGF$_{2\alpha}$ cyclicity.

Introduction

In India delayed puberty, acyclic after attaining puberty, and post-partum anestrus which lead to prolong inter calving period are major causes of poor reproductive efficiency in cattle and buffaloes. Real anestrus with inactive, smooth, small and round and flat ovarian condition is a major limiting factor in greater utilization of artificial insemination for rapid improvement of livestock productivity. This also results in loss of production and increased cost of maintenance. The problem of anestrus has been recognized as having moderate to high incidence affecting the fertility of the animal vis-à-vis economy of the farmer (Kurien and Madhavan, 1985; Kumar et al., 1986 and Sinha et al., 1987) [1, 2, 3]. Anestrus is one of the most commonly occurring reproductive problems in cattle and buffalo in India, affecting livestock productivity and enterprise to a great extent. Anestrus is observed in post pubertal heifers, during pregnancy, lactation and in early postpartum period in adult animals. The condition may be associated with uterine pathology such as pyometra, fetal resorption, maceration and mummification. Expression of estrus is also influenced by seasonal changes, stress and aging. In heifers, it poses a herd problem possibly due to low plane of nutrition, stress of seasonal transition or extremes of climatic conditions. Expression of overt signs of estrus is greatly affected by heat stress in buffaloes. Besides breed and climate, management and nutrition also play vital role in determining the reproductive disorder in cattle and buffalo. Reproductive failures such as anestrus, repeat breeding and pathological condition of the genital tract suggest the nutritional deficiencies, hormonal imbalance and deranged enzymatic activity affect the normal reproductive behavior of the animal, causing serious morphological and physiological alterations (Roberts, 1971) [4].
Nutritional deficiencies and excesses may cause infertility. They may act via the hypothalamus and anterior pituitary thus influencing the production of gonadotropins or directly on the ovaries, thus influencing oogenesis and endocrine function. To augment the milk production in indigenous cows, cross breeding with exotic germ plasm has become the general practice of the day. Progressive farmers are largely concerned as higher milk yield is dependent on normal reproduction. Any deviation or prolongation in breeding rhythm results in a progressive economic loss due to decreased production maintenance of non-productive animals for long periods, decrease number of off-springs and increased depreciation cost (Zemjanis, 1970) [5]. Progesterone has of late been identified as an important component of the reproductive hormone complex, bringing about cyclicity. Induction of oestrus and ovulation by progesterone has been studied by (Roche, 1976) [6]. Use of this treatment for oestrus and ovulation induction is based on the fact that exogenous progesterone administered in physiological doses (20-50 mg) during normal breeding permits time for corpus luteum (CL) at various stages of development to complete their natural life span and inhibit follicular growth and estrogen production. Thus when progesterone is withdrawn, follicles grows rapidly producing an estrogen peak initiating LH receptors and enhancing LH response for ovulation. Prostaglandin F2α (PGF2α) is an unsaturated 20-carbon fatty acid containing a cyclopentane ring and two aliphatic side chains. It is widely distributed in animal tissues ad may act as a local hormone. One of its physiological effects in its lytic action on the CL. PGF2α induces luteal regression in many species including bovine, ovine and equine. Its luteolytic effects in the bovine have been well documented (Chenault et al., 1976) [7]. As reported by (Chenault et al., 1976) [7], injection of PGF2α (33.50 mg than salt) is followed by a sequence of hormonal changes very similar to those observed during spontaneous CL regression, follicular maturation and ovulation. Following a single administration of PGF2α, plasma progesterone decline rapidly reaching oestrus concentrations by 24 hours post-treatment. Considering above mentioned point in view, it is proposed to investigate the response of postpartum anestrous cows to the administration of progesterone implant and PGF2α on oestrous cyclicity.

2. Materials and Methods
2.1. Experimental animals
The present experiment was conducted in summer months (April to June) on thirty six crossbreed cows which level of exotic genotype was not known and were maintained under rural management system located from the villages of Vaishali and Patna district. Only those crossbred lactating cows were considered for the preset experiment that had not been detected in oestrus 90 days after parturition. Their previous parturitions were at full term and normal. The genitalia of all the cows were examined per rectum. The cows between 1st to 6th lactation that had not been detected in oestrus 6 to 24 months after parturition but having normal genital with smooth ovaries and sound health were included in present experiment.

2.2. Grouping of animals and their treatment
Forty crossbreed cows with normal physical appearance and genital condition were dewormed once with broad spectrum anthelmintic (Panacur, Fenbendazole-3 gm). The stool examination of all the animals were done on three consecutive days from day 13 to 15 after the treatment with anthelmintics. The stools of all the animals were found negative for parasitic infestation. Out of 40, 36 healthy crossbred cows with normal genitalia were selected were divided in 6 groups having 6 animals in each. The animals were first treated with PGF2α at the dose rate of (25 mg) intramuscularly on day ‘0’. Animals of group I received normal saline solution (2ml, I/M) on day 5 and day 15 and they served as control. The animals of groups II were received 1ml Duraprogen™ (250 mg prostegosterone) I/M after prostaglandin injection. Animals of group III received 1st first dose of 1ml Duraprogen™ on day 5 and second dose of 1ml Duraprogen™ on day 15. Animals of group IV were received 2ml Duraprogen™ (500 mg) progesterone) on day 5 after prostaglandin injection while the animals of group V were received 1st dose of 2 ml Duraprogen™ on day 5 and second dose of 2ml Duraprogen™ on 15 respectively. Animals of group VI received 3 ml Duraprogen™ (750 mg progesterone) I/M on day 15 after Fenbendazole administration. For the animals of group VI Prostaglandin (Lutalyse, 25 mg) was administered 8th day after Duraprogen™ administration. Blood samples were collected from each animal on day 0 just before PGF2α administration and on day 5, day 15 and day of oestrus in group I to V. The samples were also collected from the animals of group VI on the day of progesterone and prostaglandin administration and on day of oestrus.

2.3. Oestrus detection and ovarian response
The examination of genitalia and detection of oestrus in the treatment and control group animals were done under village management system by approaching to each animal at farmer’s door step. The animal owners where also made acquainted with the behavioural oestrus symptoms apparently visible. The intensity of oestrus was classified into three viz. strong, moderate and weak. The animals with behavioural symptoms of oestrus i.e. bellowing, mounting behavior either on animal or on owner, restlessness, off feeding, reduction in milk production, vaginal mucus discharge, swelling of vulva and vagina, increased uterine tone, hyperamia in vaginal mucus membrane and presence of follicle on the ovary was considered as strong symptoms of oestrus. Each cow was observed closely by the animal owners and trained animal attendant of the state government veterinary hospital and artificial insemination (AI) centers of Baidyanathpur, Hajipur and Danapur hospitals. All the cows detected in oestrus were allowed for AI by frozen semen. The ovulation in each cow was confirmed through rectal palpation of the ovary between 9th and 11th day of oestrus. Pregnancy was confirmed through per rectal examination after 60 days of AI.

3. Results and Discussion
One animal of group II responded to the treatment of progaglandin and progesterone combination and detected in oestrus on day 5 after the progesterone administration. The remaining 5 animals could not show any response to the treatment in terms of oestrus cyclicity and genital changes. The animals of Groups III and IV did not have any response to the treatment of prostaglandin and progesterone combination. The animals of Group V receiving PGF2α and two injections each of 500mg progesterone on day 5 and 15 responded to the second injection of progesterone. Six out of six animals detected in oestrus between day 2 and day 8 after second Duraprogen™ injection. They were allowed for AI.
Four out of six animals were declared pregnant by per rectal examination. Two out of six animals of group VI were detected in oestrus one each on day 8 and day 16 after Duraprogen™ injection. Both the animals detected in oestrus and inseminated artificially were declared pregnant. During the course of sexual development from birth to puberty certain changes in anatomical, physiological and biochemical configuration are taking place in hypothalamus, pituitary, gonads and tubular genitalia leading to the maturation of hypothalamo-hypophysial and gonadal system resulting in to bringing the animals to pubertal oestrus in all known domestic large animals. Establishment of these systems requires the stepwise increase in the level of progesterone from birth to the pubertal stage. That serves as a primer sensitizer for the establishment of these systems and the development of feedback mechanism for the functioning of these systems. Once these systems mature, the mechanisms responsible for exhibition of reproductive potential in the female are triggered and the sequence of reproductive events are maintained subsequently and remain operative throughout the reproductive life (Hafez, 1982) [8]. Any deviation from the normal events depict either deficiency of certain factors or in-coordination between the functioning of hypothalamo-hypophysial and gonadal system (Mc Donald, 1989) [9]. Several investigators in past have conducted trials in both temperate (Williams and Ray, 1980; Rhodes, 2002) [10, 11] and tropical (Singh et al., 2010 and 2003) [12, 13] climate by using ovarian steroids to shorten the oestrus cycle lengths and calving intervals with beneficial effect. The animals of groups V and VI responded to the treatment of progesterone and exhibited oestrus cyclicity similar to the observation recorded in the cows in temperate (Williams and Ray, 1980; Rhodes, 2002) [10, 11] and tropical (Singh et al., 2010 and 2003) [12, 13] after progesterone administration. The previous investigators have reported that the injection of 50 mg progesterone for 10 to 15 days on different days after 60 days of parturition induced the ovulatory oestrus. It also directly or indirectly increased the fertility of the oestrus animals by decreasing the variations of the intervals of oestrus, ovulation and conception (Saiduddin et al., 1968) [14] and significantly reduced the period of uterine involution, interval from calving to first oestrus and ovulation with and without combination of estradiol (Foote and Hunter, 1964) [15]. The response of the double injection of 500 mg progesterone at the interval of 10 days in terms of exhibition of oestrus among 6 out of 6 animals and confirmation of conception in 4 out of 6 inseminated animals, reveals that the two doses of 500 mg of progesterone at 10 days interval can be tried in the rural anestrous animals to bring them in fertile oestrus. The administration of 750 mg progesterone as a single dose brought 2 out of 6 animals in oestrus. Such higher single dose of progesterone did not induce the desired fertility in cows of group VI. Through the present intravaginal or parenteral administration either in form of intramuscular injection or subcutaneous ear implant, even at low doses of 50 and 25 mg progesterone (Roche, 1976, Rao and Rao, 1984) [6, 16], Though the present investigation reveals that anestrous cows in rural management system can be brought to the oestrus by administering 500 mg progesterone at interval of 10 days beyond 60 days of postpartum anestrus period. However, detailed studies to standardize the schedule of steroid treatment to bring the anestrous animal in to oestrous cyclicity are required to develop suitable bio-technique to bring the animal at oestrus throughout the year without loss of time to make the dairy industry economically viable.

4. References