EFFECT OF STAGE OF THE ESTROUS CYCLE ON INTERVAL TO ESTRUS AND CONCEPTION RATE IN HEIFERS AND COWS TREATED WITH SYNCRO-MATE-B®

by

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Literature Review

Use of progestogens alone or in combination with estrogen for the synchronization of estrus in cattle has a history of nearly 40 years. Christian and Casida (1948) prevented estrus and ovulation in yearling heifers and ewes with daily injections of progesterone in corn oil. Willet (1950) effectively blocked estrus and ovulation in dairy heifers with 50 to 100 mg daily injections of progesterone, and estrus occurred an average of 5 d after cessation of treatment (range 4 to 7). A single injection of progesterone in starch suspension given to heifers on day 16 or 17 of the estrous cycle had an inhibitory effect on follicular growth and atresia was noted in many follicles (Nellor and Cole, 1957).

Ulberg et al. (1951b) administered varying doses of progesterone to heifers on different days of the estrous cycle. A daily dosage of 50 mg per head suppressed estrus and ovulation, but allowed some follicular growth during treatment. However, the follicle which ovulated at the synchronized estrus was in no case the follicle estimated to be the largest at the end of treatment. Heifers treated with 25 mg per head daily had more follicular growth and one silent ovulation occurred during treatment. As dosage was decreased below 25 mg, greater follicular growth occurred and a higher percentage of heifers ovulated during treatment. Estrus was delayed one or 2 d in some heifers treated with low doses of progesterone and expression of estrus was variable. Ovulation was blocked in heifers injected on either day 17 or 19 of the estrous cycle, whereas in pigs if progesterone was injected during the follicular phase ovulation was not blocked (Ulberg et al., 1951a). Ulberg and Lindley (1960) noted that as dosage levels of progesterone were increased in heifers, interval from the cessation of treatment to estrus increased.

In an attempt to reduce the number of treatments required for estrus regulation, Nellor and Cole (1956) injected beef heifers with crystalline progesterone in strarch emulsion. Estrus and ovulation were suppressed in all heifers regardless of stage of the cycle and estrus occurred in 89% of treated heifers 15 to 19 d after injection. Smith and Vincent (1973) gave 1 to 4 injections of norethandrolone to cows and found a linear increase in synchronization response with increasing numbers of injections.

Early reports concerning the effect of progesterone on early post-partum cows are conflicting. A large dose of progesterone 14 d after calving had no effect on interval to uterine involution but increased the interval to first ovulation and first estrus (Foote et al., 1960). Twenty-two injections of 17-alpha-hydroxyprogesterone-n-caproate (Delalutin) given on alternate days beginning with the day of calving increased the interval to uterine involution and estrus with a large number of treated animals having silent ovulations (Fosgate et al., 1962). Foote and Hunter (1964) later reported a decrease in interval to uterine involution, ovulation and estrus in cows treated with progesterone or progesterone plus estrogen 12 to 23 d after parturition.

Ulberg and Lindley (1960) noted lower conception rates in progesterone treated heifers at the synchronized estrus than in untreated controls. It was suspected that delayed embryo cleavage noted in progesterone treated heifers contributed to these lower conception rates. Since bovine ova travel down the oviducts at a greatly increased rate in the presence of progesterone an asynchrony between the embryo and the uterus may have developed (Rowson, 1951). Ray et al. (1961) looked at histological sections of endometrium and found superficial and glandular epithelial cells to be higher in control heifers than in heifers treated with progesterone. Presumably, this indicates that exogenous progesterone reduces the secretory activity of endometrial glands. Since progesterone and estrogen are both needed for normal uterine gland development, it is possible that an imbalance of these two hormones caused by exogenous progesterone leads to subfunction of the endometrium resulting in reduced pregnancy rates. Lowered conception rates at the adjusted estrus period had earlier been associated with progesterone treatment (Trimberger and Hansel, 1955; Nellor and Cole, 1956). Wishart and Young (1974) compared 9 and 18 d progestogen treatments and found fertility was normal after the short-term treatment. This finding was substantiated by other reports indicating normal fertility was associated with progestogen treatments of less than 10 d (Roche, 1974; Woody and Pierce, 1974; Sreenan and Mulvehill, 1975; Woody and Abenes, 1975). The conception rate at the second post-treatment estrus, regardless of length of treatment, appeared to be normal indicating lowered fertility associated with long-term progesterone or progestogen treatment was short-lived (Ulberg, 1955; Trimberger and Hansel, 1955).

Woody et al. (1967) treated cows and ewes with repeated progesterone injections early in the estrous cycle and effectively shortened cycle length in both species. It seemed likely that decreased corpus luteum weight (Loy et al., 1960) and reduced secretion of progesterone (Ray et al., 1961) were responsible for shortened estrous cycle lengths. In addition, exogenous progesterone caused a decrease in pituitary concentration of luteinizing hormone (LH), a hormone which is necessary for the proper formation of the corpus luteum (Ray et al., 1961). Estrogen injected from the second to the twelth day of the estrous cycle in cattle also caused early regression of the corpus luteum (Greenstein et al., 1958). Wiltbank et al. (1961) noted shortened cycles in heifers injected with estradiol valerate during the early and middle stage of the estrous cylcle, but estrogen on the 16th or 17th day of the cycle caused cystic ovaries in many cases. Loy et al.

(1960) postulated that injections of estrogens depleted pituitary stores of LH, so it seemed probable that subnormal luteal function was due to inadequate LH during corpus luteum formation. This theory was further supported by Wiltbank et al. (1961) who demonstrated that the estrogen induced early regression of the corpus luteum could be avoided by subsequent administration of follicle stimulating hormone (FSH) and LH.

Due to the impracticality associated with daily injections of progesterone and the limited success of single injection compounds, orally active progestogens which could be incorporated into feed seemed a more desirable method of hormone administration. A variety of these progestogens were tested during the 1960's.

Hansel et al. (1961) fed 6-methyl-17-acetoxyprogesterone (MAP) for 20 d to cows and inhibited estrus and ovulation in all cases. Estrus occurred in half of the cows 3 to 4 days after MAP withdrawl and a high percentage of the cows which did not show estrus ovulated without signs of estrus. Conception rates at the synchronized estrus were notably low (25%). A 15 d MAP feeding (Nelms and Combs, 1961) resulted in 90% synchronization of treated animals and conception rate at the synchronized breeding was comparable to controls. Anderson et al. (1962) demonstrated that a 20 d feeding of MAP could effectively synchronize heifers at all stages of the estrous cycle, and conception rates at the adjusted estrus were similar to controls. Dhinsda et al. (1967) fed MAP either once or twice daily and observed no difference in synchronization response due to number of daily feedings, however, heifers fed twice daily came into heat an average of 12 h earlier (36 h after last feeding). In this study, conception rates did not differ between heifers inseminated once (12 h after estrus was detected), twice (48 and 72 h after last feeding without regard to estrus) or three times (48, 60 and 72 h after last feeding).

In a study to determine the minimum effective dose of MAP, Zimbelman (1963) observed swelling of the external genitalia and clear mucus discharge during treatment near the time spontaneous estrus would have been expected. It was also noted that both size of follicles and percent of heifers with one or more follicles increased when treatment continued past the time of regression of the corpus luteum. A dosage of 135 and 120 mg per head daily synchronized estrus in 94% of dairy and beef cattle, respectively. First service conception rates averaged 51% but varied widely (26 to 75%).

Dihydroxyprogesterone acetophenide (DHPA) controlled estrus in 96% of heifers fed 500 mg daily for 20 d (Wiltbank et al., 1967). Periods of estrus in heifers treated with DHPA were an average of 4.5 h longer and more variable than estrual periods in untreated controls. Fertilization and conception rate were also significantly lower in treated heifers (56 vs 86%; 26 vs 54%). Wagner et al. (1968) also noted reduced fertilization rates in heifers synchronized with 6-chloro-d^6-dehydro-17-acetoxyprogesterone (CAP). Hansel et al. (1966) compared MAP to the more potent progestogen, CAP, and found MAP to be superior in both degree of synchronization and first service conception rate. Cows fed CAP had longer intervals to estrus after hormone withdrawal from the feed.

During the early 1960's, it was theorized that an injection of an estrogen following progestogen treatment might reduce the hormonal imbalance caused by exogenous progestogens. This endocrine inbalance was thought to be a factor in the reduced conception rates which had been observed following progestogen treatment. However, research data failed to substantiate this theory. An injection of 10 mg of estradiol benzoate (EB) after progesterone treatment hastened the onset of estrus and decreased variation in interval to estrus, but pregnancy rates were not improved over treatment with progesterone alone (Ulberg and Lindley, 1960). Hansel et al. (1961) reported that a 0.5 mg dose of estradiol given at the time of insemination had no effect on fertility. Wiltbank et al. (1965) used various dose combinations of progesterone and estradiol and found no evidence for increased fertility with the inclusion of the estrogen at the end of progestogen treatment. Average services per conception were increased in post-partum cows treated with progesterone and estradiol over untreated cows or cows treated with progesterone alone. However, conception occurred earlier in cows which received estrogen (Foote and Hunter, 1964; Saiduddin et al., 1968).

After Curl et al. (1968) demonstrated a high degree of synchrony of estrus in cows treated with 16-day subcutaneous implants of norethandrolone (17-ethyl-19-nortestosterone), use of implants containing progestogens nearly replaced oral progestogens for estrus synchronization of cattle. Treatment period could be more precisely controlled and dose per animal regulated more closely with implants. Variability of intake of treated individuals, which affected daily dosage with oral progestogens was not a problem with implants. Wiltbank et al. (1971) tested two types of hydron polymer implants and synchronized estrus with an implant containing 220 mg of norethandrolone in the implant wall. Little progestogen was eluded from an implant containing 200 mg of norethandrolone in the lumen and estrus was not controlled in any case.

Use of progestogen-impregnated vaginal inserts or pessaries is another method of hormone administration which proved useful for the synchronization of estrus in cattle. A progestogen-impregnated pessary in place for 20 d provided good synchronization (Wishart and Hoskin, 1968), but pessary loss during treatment and subnormal fertility at the synchronized estrus reduced the overall effectiveness of this long-term treatment. Short-term treatments resulted in higher retention rates (Sreenan and Mulvehill, 1975) and insertion of a progesterone releasing coil for 14 d (Breuer et al., 1977) resulted in the synchronization of 67% of treated heifers and conception rate was only slightly lower than controls (50 vs 60%).

Using the ability of estrogens to regress an early corpus luteum in combination with a progestogen treatment, Wiltbank et al. (1967) synchronized heifers by feeding 400 mg of DHPA per head daily for 9 d and injecting 5 mg estradiol valerate (EV) on the second day of feeding. Eighty-four of 100 heifers exhibited estrus within 96 h after last feeding. No difference was noted in the duration of estrus, interval from the onset of estrus to ovulation, ova transport or fertilization rate when treated animals were compared to controls. Reducing the DHPA feeding period to 7 d did not decrease degree of synchrony, but a high number of subsequent short estrous cycles were observed (Wiltbank and Kasson, 1968). Short cycles were attributed to EV carryover since results were similar in the 9-day treatment when the EV injection was increased to 10 mg. Roche (1974) detected a synchronized estrus in 73% of heifers treated for 10 d with progesterone-impregnated implants and 5 mg EB given at the time of implant insertion. Fertility in the synchronized heifers was equal to controls. Only 63% of heifers treated for 9 d with norethandrolone implants (250 mg) and 5 mg EV given at the time of implantation showed estrus in a 5 d period following implant removal (Woody and Peirce, 1974). Low response to synchronization was due to heifers early in the estrous cycle at the time of treatment as only 40% of these heifers were synchronized. In a series of trials, Fulton et al. (1978) synchronized estrus using a combination of CAP and EV. Excellent synchrony was obtained, but pregnancy rates were 24 to 43% lower than untreated controls. Whitman et al. (1972) and Burrell et al. (1972) demonstrated a 69 and 98% synchronization response in cows and heifers, repectively, that were treated with a norgestomet implant for 9 d and given a 5 mg EV injection at implant insertion. Conception rates in both studies were comparable to control cows and heifers.

Smith and Vincent (1973) reported that the combination of norgestomet and EV was ineffective in synchronizing estrus in heifers that were day 8 or earlier in the estrous cycle on the day of treatment. Only 58% of heifers on days 5, 6 or 7 of the estrous cycle showed estrus within 3 d following implant removal compared to 82% that were d 8 or later in the cycle. Additionally, 14 heifers that were less than day 5 were injected with 2 mg of norgestomet and 6 mg EV. Cycles were shortened to less than 15 d in only six of the 14 treated heifers, with the remaining eight having cycles 19 to 23 d in length. Lemon (1975) injected either EV or EB into heifers on day 3 or 6 of the estrous cycle. In some cases exogenous estrogens were luteolytic and in other cases, luteotrophic. The results of these two studies are not conclusive, as other workers have shown excellent synchronization responses in heifers and cows treated with norgestomet and EV at all stages of the estrous cycle (Miksch et al., 1978; Davenport et al., 1980; Burrell et al., 1972; Mares et al., 1977).

Wishart and Young (1974) synchronized heifers with a 9-day subcutaneous implant of norgestomet and an injection containing 3 mg of norgestomet and 5 mg of EV (Syncro-Mate-B®, Searle Co.). This treatment was purchased by Ceva Co. in 1981 and in 1982 it was approved and marketed under the name Syncro-Mate-B® (SMB). The SMB hormonal regime has been extensively tested in heifers and cows.

Kazmer et al. (1981) studied endogenous hormone response in dairy heifers treated with SMB. Concentration of estradiol-17B in serum remained essentially unchanged throughout the cycle in control heifers, but increased within 12 h after EV injection then gradually declined throughout the implant period in SMB treated heifers. Progesterone declined to less than 2 ng/ml within 24 h after the administration of the SMB treatment and remained low throughout treamtent. Concentration of FSH in serum declined in response to to SMB and peaked coincident with LH about 40 h after implant removal.

Estrus response in heifers treated with SMB have generally been greater than 80% (Spitzer et al., 1978; Davenport et al., 1980; Smith et al., 1979b). Reported conception rates at the synchronized estrus have been variable. Wiltbank and Mares (1978), Pexton (1980) and Smith et al. (1979b) reported first service conception rates among SMB treated heifers numerically higher than untreated controls, while Burrell et al. (1972), Miksch et al. (1978) and Spitzer et al. (1981) noted conception rates lower than controls. Pregnancy rates at 5, 25 and 45 d after the first day of breeding have been consistantly higher in SMB heifers (Mares, 1978; Spitzer et al., 1978; Smith et al., 1979b).

Induction of estrus in prepuberal heifers with SMB has been studied. Wiltbank and Gonzalez-Padilla (1975) treated 16 of 30 prepuberal heifers with SMB and within 4 d after implant removal, 94% of treated heifers had shown estrus. Only 50% of the remaining untreated heifers exhibited estrus in a subsequent 45 d period. Burrell et al. (1976) and Spitzer (1982) also successfully induced estrus in prepuberal heifers with SMB. Although, conception rates at the induced estrus have generally been lower (Short et al., 1976; Debenedetti, 1977; Hixon et al., 1981), but some studies have shown them to be comparable to conception rates observed in control heifers (Wiltbank and Gonzalez-Padilla, 1975; Wiltbank and Mares, 1977). Age made a difference in response to SMB (Short et al. 1976) with heifers 13 to 15 mo responding better than heifers less than 9 mo. No difference in estrus response was detected by Wiltbank and Mares (1977) between heifers greater than 250 kg or less than 250 kg, however, pregnancy rate after 21 d of breeding were 78 and 43% for the two groups, respectively. Burrell et al. (1976)

noted that weight did not affect response to treatment or conception when heifers were grouped in successive weights above 250 kg.

Data on cows treated with SMB is similar to that in heifers (Davenport et al., 1980). Henderson (1978a) reported 52% first service conception rate in 499 cows treated with SMB, and Mares (1978) detected estrus in 76% of 1341 treated cows with 45% becoming pregnant to a synchronized insemination. In general, conception rates in SMB cows have been lower than untreated controls (Davenport et al., 1980). Cows synchronized with SMB have consistantly shown higher pregnancy rates than controls during the first five and 25 d of the breeding season (Davenport et al., 1980; Mares, 1978). Spitzer et al. (1976) and Kiser et al. (1980) noted a linear relationship between days post-partum, estrus response and first service conception rate in SMB cows. Mares (1978) found no difference in response or conception rate between cows 30 to 60 d post-partum or more than 60 d post-partum. However, primiparous cows 60 d post-calving had fewer first service conceptions and lower 25 d pregnancy rates than either primiparous cows 90 d post-partum or mature cows 50 to 90 d post-partum.

A diffence in cyclicity was reported (Henderson, 1978b) between cows 42 or more days post-partum and cows less than 42 d (70 vs 21%). However, first service conception in cows less than 42 d was only slightly lower than cows 42 or more days post-partum (39 vs 44%), indicating SMB induces a fertile estrus in some anestrous cows. Estrus was induced in 8 of 10 non-cycling cows (Miksch et al., 1978), and 4 of 10 became pregnant to a synchronized breeding. Hixon et al. (1981) reported no difference in conception rate following SMB treatment of cycling and anestrous cows. These data are not conclusive, however, as Middleton (1985) treated 119 non-cycling cows with SMB and only 20% conceived at first service compared to 47% of 167 cycling cows. Walters et al. (1982) observed that although

10 of 19 thin anestrous cows showed estrus after SMB, only three ovulated. Hixon et al. (1981) noted short luteal phases after SMB in 17% of previously anestrous cows, with none conceiving at the synchronized estrus. It seems likely that fertility at the induced estrus in cows is depressed as seen in prepuberal heifers induced into estrus with SMB (Debenedetti, 1977).

Forty-eight hour calf removal (CR) starting at the time of implant removal increased the number of cows showing estrus and 4 and 21 d pregnancy rates in cows treated with SMB (Smith et al., 1979a; Mares, 1978; Kiser et al., 1981). Davenport et al. (1980) noted a similar increase in estrus response but no proportionally higher pregnancy rate. An increased estrus response was also noted in anestrous cows with CR after SMB (Walters et al., 1982). Mares (1978) and Pexton (1980) reported treated cows with CR had shorter intervals to estrus after implant removal and were more closely synchronized than those without CR. Circulating LH increased in response to CR and the LH surge was hastened but similar in magnitude (Walters et al., 1982). Hixon et al. (1981) noted lower first service conception rates if the LH surge occurred other than 33 to 36 h after implant removal in SMB cows inseminated 48 h after implant removal. Davenport et al. (1980) inseminated cows treated with SMB either at estrus (SMB-E) or by time (SMB-T) and found CR to be effective in increasing first service conception rate only in the SMB-T group.

The insemination of cattle by appointment after SMB has produced reasonable conception rates in many cases. Several studies suggest no difference between SMB-T and SMB-E in first service conception rate (Smith et al., 1979b; Spitzer et al., 1981) or 5 d pregnancy rate (Miksch et al., 1978; Smith et al., 1979b; Spitzer, 1982) in treated cows and heifers. Conceptions first service were higher in heifers inseminated either 45, 55 or 48 and 60 h after implant removal than heifers bred by estrus (Spitzer et al., 1981). Wiltbank and Mares (1977) inseminated heifers at different times from 44 to 60 h after implant removal and found 48 to 54 h to be optimal. Both first service conception rate and pregnancy rate were higher in SMB-T cows with CR than SMB-E cows with no CR (Mares et al., 1977). Some reports have indicated higher first service conception percentages when inseminating by estrus, but higher pregnancy rates when breeding by appointment (Mares, 1978; Davenport et al., 1980; Anderson et al., 1982). This is possible due to silent ovulations occurring in some females treated with SMB (Miksch et al., 1978).

Injections of gonadotropin releasing hormone (GnRH), pregnant mare serum gonadotropin (PMSG) or estradiol-17B (E_2) used in combination with SMB have generally failed to improve fertility in treated cows and heifers. Cows injected with 125 ug GnRH 30 h after implant removal and inseminated 16 h later had slightly lower first service conception rates than SMB-E cows with no GnRH (Zaied et al. 1976). Ferguson (1985) reported that GnRH at the time of insemination in SMB-T heifers was only effective in increasing pregnancy rates in heifers which did not show estrus after implant removal. In prepuberal heifers, 125 ug GnRH 30 h after implant removal and insemination 12 h later decreased conception rate and 5 d pregnancy rate compared to SMB-T or SMB-E heifers (Spitzer, 1982). Anderson et al. (1982) also treated heifers with GnRH after implant removal and found no increase in fertility. Twenty-one day pregnancy rates were not increased in anestrous cows treated with SMB and PMSG administered either at time of treatment initiation or implant removal (Smith et al., 1979a). Multiple ovulations were noted in 26 to 56% of PMSG treated cows. Interval to estrus was reduced in heifers receiving 0.5 or 1 mg E_2 at SMB implant removal. However, conception rate was lower than in SMB treated heifers and 6 of 14 heifers given E_2 had subsequent short estrous cycles (Anderson et al. 1982).

It is unclear how stage of the estrous cycle at the time of synchronization affects response to treatment and fertility at the synchronized estrus. Ninety-one percent of ova were fertilized in heifers started on CAP treatment during the last third of the estrous cycle, whereas only 52% were fertilized in heifers started on treatment during the first 14 d of the cycle (Wagner et al., 1968). Shorter intervals to estrus were noted in heifers treated after day 11 of the estrous cycle with norethandrolone implants and 5 mg EV than heifers treated day 8 or earlier (Woody and Pierce, 1974). Using the same treatment, it was observed that only 33% of heifers treated on day 2 of the cycle showed a synchronized estrus, while 100% of day 14 heifers were in estrus after treatment. Smith and Vincent (1973) failed to synchronize heifers in the first 5 d of the estrous cycle using 5 mg EV and 2 mg norgestomet with or without norgestomet implants. In the same study, only six of 14 treated heifers earlier than day 5 had subsequent cycles less than 15 days in length, though this treament is nearly the same as SMB. Greenstein et al. (1958) and Wiltbank et al. (1961) had reported estrogens to be luteolytic during the early stages of the estrous cycle. In contrast, Lemon (1975) treated heifers early in the cycle with either EV or EB and found exogenous estrogens to be either luteolytic or luteotrophic. A synergism may exist between norgestomet and EV when given together early in the estrous cycle since regression of the corpus luteum was greatly enhanced over administration of each hormone separately (Lemon, 1975; Miksch et al., 1978).

Introduction

Nearly 40 years ago, Christian and Casida (1948) prevented estrus and ovulation in yearling heifers with daily injections of progestrerone. Since that time, considerable research has been devoted to the use of progesterone or progestogens for the synchronization of estrus in cattle. Several workers reported successful synchronization with daily injections of progesterone (Ulberg et al., 1951b; Hansel et al., 1961; Anderson et al., 1962), but the intensive labor required for such a treatment was impractical. Single injections of progesterone or progestogens in starch suspension reduced the number of treatments required, but degree of synchrony was limited (Nellor and Cole, 1957; Ray et al., 1961; Smith and Vincent, 1973).

Oral progestational agents which could be incorporated into feed provided an alternative method of hormonal administration. During the 1960's, a number of oral progestogens were tested with various results. A 15 d feeding of 6-methyl-17-acetoxyprogesterone (MAP) resulted in the synchronization of 90% of treated animals (Nelms and Combs, 1961). Similar trials also produced excellent synchronization results (Zimbelman, 1963; Dhinsda et al., 1967) but conception rates were generally low. Reduced fertility at the synchronized estrus became associated with long-term (>10 d) progestogen treatment (Wishart and Young, 1974). This limited the use of oral progestogens because a treatment of 14 to 16 d was necessary for good synchronization (Woody and Pierce, 1974; Lemon, 1975).

An injection of estrogen prior to short-term progestogen treatment was effective in synchronizing estrus and conception rates were comparable to controls (Wiltbank et al., 1967). This hormonal combination evolved into Syncro-Mate-B® (SMB) which was approved for use in heifers by the Food and Drug Administration in 1982. The SMB treatment consists of a 6 mg norgestomet implant placed subcutaneously in the ear for 9 d and an injection containing 3 mg norgestomet and 5 mg estradiol valerate given at the time of implant insertion.

Researchers have shown SMB to be successful in synchronizing estrus in both heifers and cows (Mares, 1978; Miksch et al., 1978), however, synchronization response and conception rates have been variable. Smith and Vincent (1973) reported a treatment very similar to SMB to be ineffective in synchronizing heifers earlier than day 5 in the estrous cycle. Lemon (1975) reported that estradiol valerate had either luteolytic or in some cases, luteotrophic effects when administered to heifers on day 3 or 6 of the estrous cycle. Others have reported excellent results with heifers at all stages of the cycle (Burrell et al,. 1972; Davenport et al., 1980). Anderson et al. (1982) noted a higher conception rate in heifers treated with SMB on or before day 8 of the cycle compared to heifers treated on day 16.

One factor that might contribute greatly to the variability of response to SMB is day of the cycle at implantation. Therefore, this experiment was designed to determine if stage of the estrous cycle influences interval to the onset of estrus and first service conception rate in heifers and conception rate in cows treated with SMB.

Materials and Methods

In Trials 1 to 3, yearling beef and dairy heifers were used to study the influence of day of the estrous cycle at time of Syncro-Mate-B® (SMB) treatment on interval to estrus after implant removal and conception rate at the synchronized breeding. Heifers were maintained under dry-lot conditions, observed for estrus twice daily at 12 h intervals for 25 to 34 d and treated with SMB on selected days of the estrous cycle. The SMB treatment consisted of a 6 mg norgestomet implant placed subcutaneously on the convex surface of the ear, and an injection containing 3 mg norgestomet and 5 mg estradiol valerate in sesame oil with 10% benzyl alcohol given on the day of implantation. Implants were removed 9 d after insertion and heifers were observed for estrus at 4 to 6 h intervals for a period of 48 to 54 h following implant removal. A heifer was considered to be in estrus if she stood when mounted by another heifer. Interval from implant removal to the onset of estrus was determined for each heifer as the time between implant removal and when the heifer was first noted in estrus. Heifers were artificially inseminated 48 h after implant removal without regard to estrus, or in some cases, about 12 h after detection of estrus. Where more than one technician was used, inseminators were stratified according to day of the estrous cycle. Inseminator comments relative to each insemination were recorded at the time of breeding. Inseminations were classified as easier than normal (1) if they met one or more of the following criteria: mucus present at insemination, excellent tone of the reproducive tract noted, tube passed through the cervix with less than normal difficulty. Inseminations were classified as more difficult than normal (3) if they met one or more of the following criteria: dryness noted, reproductive tract had poor tone, tube passed through the cervix with more difficulty than normal. All other inseminations were considered normal (2). Heifers were rectally palpated for pregnancy 40 to 90 days post-insemination. A combination of palpation data, breeding records and in some cases calving dates was used to determine if heifers conceived to the synchronized insemination. Average interval to estrus and first service conception rate were compared between heifers at different stages of the estrous cycle using Chi-square analysis.

Trial 1. Thirty-three Angus, Hereford or Simmental heifers averaging 324 kg (range 284 to 382) and 12 to 15 mo of age were utilized for this experiment. Heifers were kept in a dry lot at the Kansas State University Purebred Beef Unit and the trial was conducted during April and May of 1984. Heifers received a ration consisting of 3.6 kg rolled milo per head daily and ad libitum alfalfa hay. Heifers were observed for estrus for 30 d, treated with SMB and inseminated by one of two technicians 46 to 50 h after implant removal without regard to estrus using semen from one of eight bulls. Following the synchronized insemination, heifers were moved to spring pasture. For 25 d heifers were checked for estrus daily with the aid of a bull with a surgically deviated penis and inseminated approximately 12 h after first detected in heat. Bulls of the respective breeds were then turned out and allowed to remain with the heifers for 30 d. Conceptions to the synchronized breeding were verified with calving dates the following spring.

Trial 2. Twenty-eight holstein heifers 13 to 17 mo of age with an average weight of 419 kg (range 337 to 535) from the Kansas State University Dairy Herd were utilized during August and September of 1984 for this experiment. Heifers were observed for estrus for 25 d, synchronized and observed for estrus at 6 h intervals for 54 h following implant removal. One technician inseminated all heifers 0 to 12 h after estrus was detected with semen from one of three Holstein bulls. Heifers which had not shown any sign of estrus by 52 h after implant removal were inseminated at this time.

Trial 3. During April and May of 1985, 43 Angus, Hereford or Simmental heifers 12 to 15 mo old and weighing an average of 357 kg (range 306 to 445) were used in this experiment. Heifers were located at the Kansas State University Purebred Beef Unit and received a ration similar to heifers in Trial 1. Thirty-one heifers were treated with SMB on selected days of the estrous cycle, and 12 heifers which had not been detected in estrus at the end of the 34 d estrus detection period received SMB at this time. Venous blood samples were taken from 17 heifers (Group 1) via jugular vein puncture 10 and 3 d prior to and on the day of SMB treatment. Blood was collected 18 and 11 d before and on the day of treatment from the 27 remaining heifers (Group 2). Additionally, blood was taken from heifers in Group 2 at the time of implant removal. All samples were assayed by radioimmunoassay for progesterone (Stevenson et al., 1984). Heifers which had >1 ng/ml of progesterone in any of the samples of serum were considered to be cycling and to have had a functional corpus luteum at the time the sample was taken. Heifers were inseminated 46 to 50 h after implant removal by two technicians with semen from one of four bulls. After the synchronized breeding, heifers were handled the same as in Trial 1, except conceptions to the synchronized service were not verified with calving dates.

In Trials 4 to 6, 140 primiparous and multiparous cows and 8 yearling heifers were used to study the effect of stage of the estrous cycle at time of SMB treatment and difficulty of insemination on conception rate at the synchronized breeding. Both spring and fall calving cows that were 15 to 88 d post-partum and had body condition scores ranging from 3 to 8 were used. Blood samples were taken from each female via jugular puncture 7 d before and on the day of SMB treatment. Serum was analyzed by radioimmunoassay for progesterone content as in Trial 3 and the stage of the estrous cycle estimated for each female using data from the two blood samples. On a predetermined date cows and heifers were treated with the standard SMB treatment, and implants remained in situ for 9 d. Calves were separated from cows at the time of implant removal and remained separated for 48 h. Cows and heifers were not observed for estrus and were time inseminated 44 to 54 h after implant removal. Inseminators were stratified according to cycling status and post-partum interval. Inseminations were scored by inseminator relative to difficulty at the time of breeding as described earlier. Cows and heifers were palpated per rectum for pregnancy 60 to 90 d after insemination and using palpation data, breeding records and subsequent calving dates conceptions to the synchronized breeding were determined. Conception rates between females having different insemination scores and at different stages of the estrous cycle were compared using Chi-square analysis.

Trial 4. Twenty-six crossbred cows and eight crossbred yearling heifers located at the Kansas State University Experiment Station, Hays, Kansas were used for this trial in the Fall of 1983. Cows were fall-calving and averaged 51 ± 17.9 d post-partum (range 17 to 88) at the time of SMB treatment. Four technicians inseminated the cows and heifers with semen from two bulls. Semen from a Simbrah bull was used on the cows as a genetic marker to aid in the identification of calves resulting from artificial insemination. Cows and heifers were exposed to clean-up bulls for a period of 64 d beginning 2 d after the synchronized insemination. A Red Angus bull was used on the heifers and a Simmental bull was used on the cows. Pregnancy diagnosis was performed 66 d after artificial insemination and conceptions were confirmed with calving dates.

Trial 5. Sixty-six Polled Hereford or Simmental cows located at the Kansas State University Cow-Calf Unit were used during the spring of 1984 for this experiment. Forty-three cows calved in March through May and averaged 55 ± 12.3 d post-partum (range 23 to 84) at the time of SMB treatment, while the remaining 23 cows were dry and open from the previous year. Body condition scores (Whitman,

1975) were given 72 and 34 d prior and on the day of SMB treamtent. The three scores were averaged for each cow and the average body condition score for all females was 5.0 on a 1 to 9 scale (range 4 to 8). Semen from six bulls was used by three technicians to inseminate the cows. For a period of 45 d following the synchronized insemination, cows were observed for estrus with the aid of androgenized cows equipped with chin-ball markers and artificially inseminated at estrus. Females were rectally palpated to determine pregnancy 60 to 90 d after the synchronized breeding and calving dates were used to verify which cows conceived during the synchronized period.

Trial 6. Forty-eight crossbred cows averaging 50.6 ± 16.6 d post-partum at the time of treatment (range 15 to 88) and located at the Kansas State University Experiment Station, Hays, Kansas were utilized for this trial during the spring of 1984. Cows were scored for body condition 30 d prior to SMB treatment and on the day of implant removal with the two scores being averaged for each individual. Average body condition score for all cows was 5.0 (range 4 to 8). Three technicians inseminated the cows with semen from two bulls. Two days later, two bulls were turned in and allowed to remain with the cows for 65 d. Cows were palpated for pregnancy 73 d after artificial insemination and pregnancies resulting from the synchronized breeding were verified by subsequent calving dates.

Results

Trial 1. Swelling of the external genitalia and clear mucus discharge were noted in some heifers while the implants were in place; this is a response which has been reported previously (Zimbelman, 1963). Thirty of the 33 heifers in this trial showed estrus within 54 h after implant removal. Two of the three which failed to show estrus had been treated with SMB on day 4 of the estrous cycle and the third was treated on day 7. These heifers apparently ovulated during the synchronized period since two of the three conceived to the timed insemination and the third returned to estrus 21 d after insemination. One heifer treated on day 6 of the cycle returned to estrus 16 d after insemination; the only shortened cycle noted. Two heifers treated on d 0 or 13 of the cycle were not detected in estrus until d 25 and 23 after insemination, respectively.

First service conception rate was 42% (14/33) and was significantly affected by stage of the estrous cycle. Twelve of 22 (54.5%) heifers treated with SMB on or before day 11 (early cycle) became pregnant to the timed insemination, while only two of 11 (18%) treated after day 11 (late cycle) conceived during the synchronized period (Table 1). Conception rate was different ($p<.01$) between the two groups.

Interval to estrus was shorter ($p=.09$, Table 2) in heifers treated early in the cycle (28.9 ± 8.0 h) compared to heifers treated late in the cycle (38.7 ± 9.7 h). Average interval to estrus for all heifers was 32.8 ± 8.8 h and was not different between heifers which conceived at first service (31.3 h) and those which did not conceive (34.3 h). A difference ($p<.01$) in conception rate was found between heifers which came into estrus ≤ 35 h after implant removal (7/16=44%) and those which came into estrus >35 h after implant removal (2/9=22%).

Nine of 21 (43%) heifers with nsemination scores of 2 conceived at the synchronized estrus, while only one of four (25%) heifers scored 1 in insemination difficulty conceived (Table 3). Only three heifers were scored 3, and one conceived to the timed breeding. There was no difference in conception rates among inseminators or among bulls in this trial.

Trial 2. Three heifers treated with SMB late in the estrous cycle and two treated early in the cycle failed to show any sign of estrus and were inseminated 52 h after implant removal. However, none of the five conceived to this breeding. Another late cycle heifer which demonstrated hyperactivity, and had been inseminated 52 h post-implant removal, subsequently showed estrus 66 h after implant removal and was reinseminated 4 h later. This heifer conceived and is included in conception results.

First service conception rate in this trial was 36% (10/28) and was not affected by stage of the estrous cycle. Five of 13 (38%) heifers started on the SMB treatment early in the estrous cycle conceived during the synchronized period compared with five of 15 (33%) started on treatment late in the cycle (Table 1).

Average interval to estrus was 35.4 ± 5.8 h for all heifers and there was no difference between heifers treated early in the cycle (34.6 ± 5.4 h) and heifers treated late in the cycle (34.5 ± 5.2 h, Table 2) or heifers which conceived to the timed insemination (34.5 h) and those which did not conceive (36.2 h). Conception rate was higher ($p<.05$) in heifers which came into estrus ≤ 35 h after implant removal (7/14=50%) than heifers with interals to estrus >35 h (3/9=33%).

First service conception rate was higher ($p<.01$) in heifers with insemination scores of 1 (8/11=73%) than in heifers scored 2 (1/11=9%) or 3 (1/16=16%, Table 3). Conception rates were not different between sires used in this trial.

Trial 3. Clear mucus discharge and swelling of the external genitalia were noted in some heifers as in Trial 1. Ninety-six percent (23/24) of the cycling heifers had shown estrus by 48 h after implant removal. The heifer which failed to show estrus had been treated with SMB on day 2 of the estrous cycle, was difficult to inseminate and returned to estrus 26 d after implant removal. First service conception rate in the cycling heifers was 50% (12/24) and conception rate was higher ($p<.05$) in heifers treated late in the estrous cycle (7/12=58%) than in heifers treated early in the cycle (5/12=42%, Table 1). Three of the 12 heifers which had not been detected in estrus prior to SMB treatment were cycling as indicated by concentrations of pogesterone in serum >1 ng/ml in one or more of the samples taken. Two of the three (67%) conceived to first service while 3/9 (33%) treated prepuberal heifers conceived. Estrous cycles were initiated in all six of the prepuberal heifers which did not conceive to the synchronized insemination as all heifers returned to estrus 18 to 23 d after implant removal.

Average interval to onset of estrus for all cycling heifers was 31.3 ± 7.2 h and did not vary according to stage of the estrous cycle (Table 2). Interval to estrus was 31.3 ± 8.2 h in heifers treated early in the cycle and 31.2 ± 6.3 h in heifers treated late in the cycle. There was no difference in interval to estrus between heifers which conceived to the synchronized insemination and those which did not conceive (33.3 vs 29.2 h). A higher ($p<.01$) conception rate was noted in heifers which came into estrus ≥ 35 h after implant removal (5/7=71%) than in heifers with intervals to estrus <35 h (7/16=44%). These results are do not agree with Trial 1 and 2.

Difficulty of insemination apparently affected conception rate only when inseminations were more difficult than normal. Conception rate was lower ($p<.01$) in heifers that were difficult (3) to inseminate (0/4=0%) than in heifers with normal

(2) or easier than normal (1) insemination scores (13/23=56.5%; 6/11=54.5%, Table 3). Conception rates did not vary between inseminators or bulls.

Average interval to estrus for all heifers in Trials 1 to 3 was 33.3±7.3 h (range 21 to 56 h), and ther was no difference between heifers treated early in the cycle (31.3±7.4 h) and late cycle heifers (35.2±7.2 h) or heifers which conceived to the synchronized insemination (33.1 h) and those which did not conceive (33.4 h). First service conception rate was not different between heifers which came into estrus <35 h after implant removal (41%) and heifers with ≥35 h intervals to estrus (47%). First service conception rate was slightly higher (p=.2) in heifers started on the SMB treatment early in the estrous cycle (47%) compared to heifers treated late in the cycle (37%, Table 1). Overall conception rate for the three trials was 42% (36/85).

Conception rates for heifers with insemination scores of 1 (15/26=58%) were somewhat higher (p=.08, Table 3) than heifers with scores of 2 (23/56=41%). Conception rate at the synchronized estrus was lower (p<.01) in heifers that were scored 3 at insemination (1/12=8.3%).

Trials 4-6. For cows and heifers in these trials, concentration of progesterone (ng/ml) in each of the two serum samples was used to estimate stage of the estrous cycle. Concentration of progesterone in serum is expected to be >1 ng/ml in cycling beef females on day 5 through 17 of the estrous cycle with day 4 and 18 being variable, either above or below 1 ng/ml (Henricks and Dickey, 1970; Adeymo and Heath, 1980). Therefore, in a 21-day estrous cycle progesterone is elevated above 1 ng/ml (H) for approximately 14 d and less than 1 ng/ml (L) for 7 d. In these three trials, samples were taken 7 d apart, so that the majority of cycling cows should have had H progesterone on at least one of the two sample days. Cows which had L serum progesterone at the first bleeding and H concentrations at the second bleeding (LH) were considered to be early ($\leq$day 11) in the estrous cycle at the time of SMB treatment. This is due to L progesterone in the first sample which indicates a young, not fully functional corpus luteum ($\leq$day 4) if one is present. Seven days later LH cows should each have a functional corpus luteum, indicating they were on or before day 11 in the cycle. Females which had H progesterone at both bleedings (HH) were considered late (>day 11) in the cycle because H concentrations on both sample days indicate the presence of a functioning corpus luteum that is at least 11 d old at the second bleeding. Animals with H progesterone on the first sample date and L progesterone in the second sample (HL) could be either very early or very late in the estrous cycle but it is not possible to accurately determine stage of the cycle in these females. Low progesterone readings on both sample dates (LL) indicated anestrous cows or prepuberal heifers.

Trial 4. Nineteen of 26 (73%) cows and seven of eight (87.5%) heifers were cycling at the initiation of SMB treatment, and 15 of the 34 females (44%) conceived to the timed insemination. Conception rate was lower ($p<.01$) in heifers (2/8=25%) than in cows (13/26=50%), but did not vary according to cycling status, stage of the estrous cycle, difficulty of insemination or post-partum interval. Five of 14 (38%) HH cows and heifers conceived to the synchronized breeding compared to three of six (50%) LH, three of six (50%) HL and four of eight (50%) LL females (Table 4).

First service conception rate was not different between cows with insemination scores of 1 or 2 (4/8=50% vs 11/25=44%, Table 5) Only one cow was given an insemination score of 3, and she did not conceive to the synchronized insemination. Six of 11 (54.5%) cows less than 50 d post-partum at insemination were pregnant to the artificial insemination compared to 5 of 13 (38.5%) cows 50 d or more post-partum (Table 6). Neither inseminators or bulls affected first service conception rate.

Trial 5. Thirty-three percent (22/66) of the females in this trial were anestrous at the time of SMB treatment. Cyclicity in the 23 open, dry cows and six cows which lost calves was 93 and 100%, respectively. A lower ($p<.01$) per cent cyclicity was noted in cows nursing calves (44%). Conception rates reflected these values and were higher ($p<.01$) in cows which lost calves (4/6=66.7%) than in lactating cows (14/38=36.8%) and dry cows (11/23=47.8%). Overall first service conception rate was 44% (29/66, Table 4) and was not affected by inseminator or bulls.

Seven of 22 (31.8%) anestrous cows became pregant as a result of the 48 h insemination which was a lower ($p=.07$) percentage than the overall conception rate. Conception rates were not different ($p>.1$) in either LH cows (7/16=43.8%) or HH cows (6/11=54.5%), but were lower ($p<.01$) in HL cows (2/8=25%, Table 4). Difficulty of insemination was a source of variation in first service conception rate after SMB in this experiment. Conception rate was higher ($p<.01$) in cows with insemination scores of 1 (5/7=71.4%) than cows scores 2 (24/52=46%) and lower ($p<.01$) in cows with insemination scores of 3 (0/7=0%, Table 5).

Neither body condition or post-partum interval significantly affected first service conception rate in this trial. However, numerically lower conception rate in cows with body condition scores less than 5.0 (8/25=32%) approached significance ($p=.07$) when compared to cows 5.0 or greater in body condition (21/41=51.2%, Table 8). This numerical difference reflected cyclicity in the two groups as only 48% (12/25) of cows less than 5.0 in body condition were cycling compared to 73% (30/41) of cows 5.0 or greater. No difference in conception rate was noted between cows less than 50 d post-partum at insemination (11/23=48%) and cows 50 d or more post-partum at SMB treatment (7/21=33%, Table 6).

Trial 6. Forty of 43 (83%) of the cows in this trial were cycling at the time SMB treatment was initiated. First service conception rate for the herd was 42% (20/48), and both stage of the estrous cycle and post-partum interval affected conception rate. A higher ($p<.01$) percentage of LH cows (10/13=76.9%) conceived during the synchronized period than HH cows (6/12=50%, Table 4). Conception rate was lower ($p<.05$) in LL cows (0/8=0%) and HL cows (4/15=26.5%) as seen in Trial 5.

Fewer cows less than 40 d post-partum were cycling compared to cows 40 d or more post-partum (73 vs 89%), and this was reflected in conception rates as only 25% (3/12, $p<.01$) of cows less than 40 d post-partum at insemination conceived to the timed insemination, whereas 47% (17/37) of cows 40 d or more conceived (Table 7). Body condition score had no effect on cyclicity or conception rate. Six of 17 (35.3%) cows less than 5.0 in body condition and 14/31 (45%) cows 5.0 or greater were pregnant to the first service. Conception rate at the synchronized estrus was not different between cows given insemination scores of 1 (7/17=41%) or cows with insemination scores of 2 (13/29=45%, Table 5). Only two cows were more difficult than normal to inseminate and neither of the two conceived to the artificial service. There was no difference in conception rate among inseminators or among bulls.

When data were pooled for Trial 4 to 6, first service conception rate was higher ($p<.01$) in cows and heifers treated with SMB early in the estrous cycle (20/35=57%) than anestrous females or cows and heifers at other stages of the estrous cycle (Table 4). Conception rate was lower ($p<.05$) in anestrous cows (11/38=30%) and numerically lower in HL (very early or very late cycle) cows

(9/29=31%) than in cows and heifers treated with SMB late in the estrous cycle (16/37=43.2%). Overall conception rate to the 48 h-insemination was 43% (64/148) for the three combined trials.

Conception rate at the synchronized estrus was influenced twice negatively and once positively by post-partum interval in all three trials when considered individually. However, when the results were pooled, no significant differences were found. When females were divided into groups of less than 40 d post-partum and 40 d or more post-partum at SMB treatment, conception rates were 33% (7/21) and 44% (42/95), respectively (Table 7). This difference approached significance ($p=.09$). When cows were divided into groups of either less than 50 d post-partum or 50 d or more post-partum, no difference in conception rate was found (43 vs 41%, Table 6)

There was no difference in combined conception rate between cows and heifers with easier than normal insemination scores (16/32=50%) and those with normal scores (48/106=45%). First service conception rate was lower ($p<.01$) in females that were more difficult than normal to inseminate (0/10=0%).

Table 1. FIRST SERVICE CONCEPTION RATE IN HEIFERS TREATED WITH SYNCRO-MATE-B ON DIFFERENT DAYS OF THE ESTROUS CYCLE

Trial	Treated d 0-11	Treated d 12-21	Overall
1	12/22(54.5%)[a]	2/11(18.2%)[b]	14/33(42.4%)
2	5/13(38.5%)	5/15(33.3%)	10/28(35.7%)
3	5/12(41.7%)[a]	7/12(58.3%)[b]	12/24(50%)
Total	22/47(46.8%)	14/38(36.8%)	36/85(42.3%)

[a,b]Conception percentages in the same row with different superscripts are different ($p<.01$).

Table 2. AVERAGE INTERVAL (hours) FROM IMPLANT REMOVAL TO THE ONSET OF ESTRUS IN HEIFERS TREATED WITH SYNCRO-MATE-B ON DIFFERENT DAYS OF THE ESTROUS CYCLE

Trial	Treated d 0-11	Treated d 12-21	Overall
1	28.9 ± 8.0[a]	38.7 ± 9.7	32.8 ± 8.8
2	34.6 ± 5.4	36.2 ± 5.2	35.4 ± 5.3
3	31.3 ± 8.2	31.2 ± 6.3	31.3 ± 7.2
Total	31.3 ± 7.4	35.2 ± 7.2	33.3 ± 7.3

a- Approached being different ($p=.09$) than heifers treated d 12-21 in Trail 1.

Table 3. FIRST SERVICE CONCEPTION RATE IN SYNCRO-MATE-B TREATED HEIFERS WITH DIFFERENT INSEMINATION SCORES

Trial	Easier Than Normal (1)	Normal (2)	More Difficult Than Normal (3)
1	1/4(25%)	9/21(43%)	1/3(33%)
2	8/11(73%)[a]	1/11(9%)[b]	1/6(16%)[b]
3	6/11(55%)[a]	13/23(57%)[a]	0/4(0%)[b]
Total	15/26(58%)[ac]	23/56(41%)[a]	2/12(17%)[b]

[a,b]Conception percentages in the same row with different superscripts are different ($p<.01$).

[c]Approached being different ($p=.08$) from total conception rate heifers with average insemination scores.

Table 4. CONCEPTION RATE IN ANESTROUS COWS AND COWS AT DIFFERENT STAGES OF THE ESTROUS CYCLE[a] AFTER SYNCRO-MATE-B AND 48-H INSEMINATION

Trial	Anestrous (LL)	Early (LH)	Late (HH)	Early Or Late (HL)	Overall
4	4/8(50%)	3/6(50%)	5/14(38%)	3/6(50%)	15/34(44%)
5	7/22(32%)[cd]	7/16(44%)[bc]	6/11(55%)[b]	2/8(25%)[c]	29/66(44%)
6	0/8(0%)[e]	10/13(77%)[b]	6/12(50%)[c]	4/15(27%)[d]	20/48(42%)
Total	11/38(30%)[d]	20/35(57%)[b]	16/37(43%)[c]	9/29(31%)[cd]	64/148(43%)

[a]Stage of the estrous cycle was determined by measuring serum progesterone in two blood samples taken, one 7 d before and one on the d of Syncro-Mate-B® treatment (L=<1 ng/ml; H=>1 ng/ml).

[b,c,d]Conception percentages in the same row with different superscripts are different ($p<.05$).

Table 5. CONCEPTION RATE IN COWS AND HEIFERS WITH DIFFERENT INSEMINATION SCORES AT 48-H INSEMINATION AFTER SYNCRO-MATE-B

Trial	Easier Than Normal (1)	Normal (2)	More Difficult Than Normal (3)
4	4/8(50%)[a]	11/25(44%)[a]	0/1(0%)[b]
5	5/7(71%)[a]	24/52(46%)[b]	0/7(0%)[c]
6	7/17(41%)[a]	13/29(45%)[a]	0/2(0%)[b]
Total	16/32(50%)[a]	48/106(45%)[a]	0/10(0%)[b]

[a,b,c]conception percentages in the same row with different superscripts are different ($p<.01$).

Table 6. CONCEPTION RATE IN COWS TREATED WITH SYNCRO-MATE-B AND 48-H INSEMINATION AT DIFFERENT POST-PARTUM INTERVALS[a]

Trial	<50 d	$\geq$50 d
4	6/11(55%)[b]	5/13(39%)[c]
5	11/23(48%)[b]	7/21(33%)[c]
6	8/24(33%)[b]	12/24(50%)[c]
Total	25/58(43%)	24/58(41%)

[a]Post-partum interval figured to the day of insemination.
[b,c]Conception percentages in the same row with different superscripts are different ($p<.05$).

Table 7. CONCEPTION RATES IN COWS TREATED WITH SYNCRO-MATE-B AND 48-H INSEMINATION AT DIFFERENT POST-PARTUM INTERVALS[a]

Trial	<40 d	$\geq$40 d
4	2/4(50%)	9/20(45%)
5	2/5(40%)	16/39(41%)
6	3/12(25%)[b]	17/36(47%)[c]
Total	7/21(33%)[d]	17/36(44%)

[a]Post-partum intervals figured to the day of insemination.
[b,c]Conception percentages in the same row with different superscripts are different ($p<.01$).
[d]Difference in conception rate between cows <40 d post-partum and cows $\geq$40 d post-partum approached significance ($p=.09$).

Table 8. CYCLICITY AND CONCEPTION RATE IN COWS WITH DIFFERENT BODY CONDITION SCORES[a] AFTER SYNCRO-MATE-B

Trial	Cycling[b]		Conception Rate	
	<5.0	$\geq$5.0	<5.0	$\geq$5.0
5	48%(12/25)[c]	73%(30/41)[d]	32%(8/25)[e]	51%(21/41)[f]
6	76%(13/17)	87%(27/31)	35%(6/17)	45%(14/31)
Total	59%(25/42)[c]	79%(57/72)[d]	33%(14/42)[g]	49%(35/72)[h]

[a]Body condition was scored according to Whitman (1975).

[b]A female was considered to be cycling if she had >1.0 ng/ml progesterone in at least one of two serum samples taken 7 d before and on the day of Syncro-Mate-B® treatment.

[c,d]Percent of cows cycling differ ($p<.01$).

[e,f]Conception rate in cows differ ($p<.01$).

[g,h]Conception rate in cows differ ($p<.05$).

Discussion

In Trials 1 to 3, no difference in interval to estrus was noted between heifers treated with SMB early in the estrous cycle compared to those treated late in the cycle. However, there was a slight trend for early treated heifers to come into estrus sooner, particularly in Trial 1. A greater difference might have been expected as heifers treated late in the cycle are under stimulation of endogenous progesterone from their own corpus luteum in addition to the norgestomet from the implant and as dose of progesterone increases, interval from the cessation of treatment to estrus also increases (Ulberg and Lindley, 1960). High doses of progesterone allow little follicle growth during treatment (Ulberg et al., 1951), so it seems reasonable that the folliculogenesis would take more time after treatment is stopped. Heifers treated early in the cycle are under very little endogenous progesterone stimulation from their own regressing corpus luteum (Kazmer et al., 1981).

Interval to estrus had no effect on conception rate at the synchronized insemination. There was a trend for higher conception rates in heifers which came into estrus on or before 35 h after implant removal compared to heifers with intervals to estrus greater than 35 h. Optimum conception rates were obtained after SMB when the luteinizing hormone surge occured 33 to 36 h after implant removal (Hixon et al., 1981). Heifers with intervals to estrus lees than or equal to 35 h would seem more likely to fit this optimum, but a difference was not apparent. Similar conception rates in heifers which came into estrus more than 35 h after implant removal and were time inseminated may indicate some cases of fertilization when ovulation occurs before insemination as has been reported by Hansel et al. (1961).

Only three of 47 (6.4%) heifers treated with SMB on or before day 11 of the estrous cycle failed to show a synchronized estrus which was not different from heifers treated after day 11 (3/38=7.9%). Thus, a high degree of luteolysis was obtained by the SMB treatment when administered during the early stages of the estrous cycle. These results disagree with Smith and Vincent (1973) and Lemon (1975) who reported poor responses in heifers treated with norgestomet and estradiol valerate on or before d 8 of the cycle.

In Trials 1 to 3, there was a tendency for higher conception rates in heifers which had been treated with SMB early (on or before day 11) in the estrous cycle as reported by Anderson et al. (1982). Dividing the days of the estrous cycle into four groups (day 0 to 5, day 6 to 11, day 12 to 16, day 17 to 20) and comparing conception rates in heifers treated on days within the four groups showed an advantage to the first half of the cycle. Conception rates were statistically higher ($p<.01$) in heifers treated day 0 to 5 (10/20=50%) and numerically higher in heifers treated day 6 to 11 (12/47=44%) than in heifers treated day 12 to 16 (9/24=37.5%) or day 17 to 21 (5/14=36%). A 10 percentage point advantage was seen in conception rate between heifers treated day 0 to 11 compared to heifers treated day 12 to 21 (22/47=47% vs 14/38=37%). When the estrous cycle was divided in thirds (day 0 to 8, day 9 to 15, day 16 to 21) no significant differences were found but the trend for higher conception rates in early cycle heifers was still evident. This data appears to agree with findings in Trials 4 to 6 where higher ($p<.05$) conception rates were seen in cows and heifers treated with SMB in the first half of the estrous cycle (20/35=57%) compared to females treated in the latter half of the cycle (16/37=43%). Although, lower conception rates were found in heifers and cows treated with SMB after day 11 of the cycle, it seems likely that this reduction in fertility is small and not a major source of the variation in conception rates seen in SMB use because results were not consistant in all six trials.

Reduced fertility in females treated late in the estrous cycle could be due to long-term progesterone-progestogen stimulation. As a heifer treated on day 15 of the estrous cycle has been under endogenous progesterone stimulation for 10 d prior to the 9-day norgestomet implant resulting in a longer period of progesterone-like dominance which has been shown to reduce fertility (Trimberger and Hansel, 1955; Nellor and Cole, 1956). Delayed embryo cleavage (Ulberg and Lindley, 1960) and decreased secretory function of the endometrial glands (Ray et al., 1961) seen after long-term progesterone treatment may be contributing to the lower conception rates seen in late cycle treated animals. It is also possible that a 48-h insemination used in five of the six trials may not be optimum for some late cycle individuals which tended to have longer intervals to estrus.

Pregnancies which resulted in some cows and heifers which were anestrous prior to treatment indicates the induction of a fertile estrus by the SMB treatment as reported by others (Miksch et al., 1978; Smith et al., 1979a; Spitzer, 1982). Reduced fertility, as indicated by lower conception rates at the induced estrus, has been noted in some previously non-cycling animals (Mares et al., 1977; Debenedetti, 1977; Hixon et al., 1981; Spitzer, 1982), but not in others (Miksch et al., 1978; Beal et al., 1984). Not all anestrous cows which show estrus after SMB ovulate (Walters et al., 1982) and a few short luteal phases occur after estrus induction with SMB (Hixon et al., 1981), so it seems likely that fertility in the induced animal is reduced in most cases. It is possible that the variation in conception between different groups of non-cycling animals is largely dependent on how far anestrous females are from cycling. This theory is supported by data from Short et al. (1976) who reported a lower conception rate in prepuberal heifers less than 9 mo old when treated with SMB compared to 13 to 15 mo prepuberal heifers.

Body condition, which was estimated in two trials, did not influence conception rate at the synchronized estrus. However, there was a trend for for cows which had lower body condition scores (<5.0) to have lower first service conception rates. This is likely due to lower cyclicity among cows less than 5.0 in body condition as seen in these trials and generally seen in thin compared to moderately conditioned post-partum cows (Jeanne Wright, personal communication). Perhaps the lack of effect of body condition in these experiments was due to the narrow range of body condition scores in the population of cows used (range 4 to 7).

In Trials 4 to 6, the difference in conception rate between cows less than 40 d post-partum and cows 40 d or more post-partum approached significance (33 vs 44%, $p=.09$). These results agree with Kiser et al. (1980) who found a linear relationship between post-partum interval and response to SMB. Mares (1978) reported no differences in estrus response or pregnancy rate in mature cows treated with SMB either 30 to 60 d post-partum or greater than 60 d post-partum. However, lower pregnancy rates were noted in primiparous cows treated 60 d post-partum compared to primiparous cows 90 d post-partum or mature cows 50 to 90 d post-partum. These reports support the contention that cows less than 40 d post-partum should not be treated if optimum results are to be obtained with SMB.

Difficulty of insemination was an indicator of conception rate after SMB only when inseminations were difficult. Only 2/22 (9%) females which were more difficult than normal to inseminate conceived at the synchronized estrus. No difference in conception rate was noted in cows and heifers which were normal or easier than normal to inseminate (71/162=44% vs. 28/58=48%). These data indicate that mucus at insemination, excellent tone of the reproductive tract and easy passage of the tube through the cervix are not indicators of above normal fertility after SMB treatment.

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EFFECT OF STAGE OF THE ESTROUS CYCLE ON INTERVAL TO ESTRUS AND CONCEPTION RATE IN HEIFERS AND COWS TREATED WITH SYNCRO-MATE-B®

by

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Six trials were conducted over two years (1983 to 1985) to determine the effect of stage of the estrous cycle at the time of Syncro-Mate-B® (SMB) treatment on interval to estrus after implant removal and on conception rate at the synchronized estrus.

In Trials 1 to 3, 57 beef and 28 dairy heifers were observed for estrus twice daily and treated with SMB on selected days of the estrous cycle. Interval to estrus was determined by observing for estrus every 6 h from the time of implant removal until insemination. Beef heifers were inseminated approximately 48 h after implant removal, while dairy heifers were inseminated 0 to 12 h after first detected in estrus. Inseminations were scored by inseminator relative to difficulty at the time of breeding and heifers were classified as either easier than normal, normal or more difficult than normal to inseminate.

Only three of 47 (6.4%) heifers treated with SMB on or before day 11 of the estrous cycle and three of 38 (7.9%) heifers treated after day 11 failed to show estrus within 66 h after implant removal. Interval to the onset of estrus was not different ($p>.1$) between heifers treated early ($\leq$day 11) in the cycle (31.3 ± 7.4 h) compared with heifers treated late (>day 11) in the cycle (35.2 ± 7.2 h). Conception rate at the synchronized estrus was numerically higher in early cycle heifers (22/47=46.8%) compared with late cycle heifers (14/38=36.8%), however, this difference was not significant ($p>.1$). Heifers that were more difficult than normal to inseminate had lower ($p<.01$) conception rates at the synchronized estrus (1/12=8%) than did heifers that were normal (23/56=41%) or easier than normal to inseminate (15/26=58%).

In Trials 4 to 6, 148 beef cows and 8 yearling heifers were used. Cows averaged 52 d post-partum over the three trials and in Trials 5 and 6 cows had average body condition scores of 5.0 on a scale of 1 through 9. Venous blood samples were taken from all cows and heifers 7 d before and on the day of SMB treatment. Progesterone in serum was quantified in each sample by radioimmunoassay and cyclicity and stage of the estrous cycle were estimated using concentration of progesterone in the two samples. Cows and heifers were treated with the standard SMB treatment and inseminated without regard to estrus 48-h after implant removal. Inseminations were scored as in the first three trials. Calves were removed from cows at the time of implant removal and returned 48 h later.

Overall conception rate was 43% (64/148) and was significantly higher in cows and heifers which were treated with SMB early ($\leq$day 11) in the estrous cycle (16/37=43%) compared to females treated late ($>$day 11) in the cycle (16/37=43%). Conception rate was lower ($p<.05$) in anestrous animals (11/38=30%) compared to cycling cows and heifers (45/101=44.6%). Cows and heifers which were difficult to inseminate had lower ($p<.01$) conception rates (0/10=0%) than females which were normal (48/106=45%) or easier than normal to inseminate (16/32=50%). Neither body condition or post-partum interval had any effect on conception rate after SMB.

Interval to estrus was not altered by the stage of the estrous cycle at time of SMB treatment, but concepton rate at the synchronized estrus was higher in heifers and cows treated on or before day 11 of the estrous cycle compared to females treated after day 11. Inseminator evaluation of difficulty of artificial insemination after SMB was an indicator of low conception if rated difficult, but conception rates between normal and easier than normal inseminations were not different.