

Progestogen and prostaglandin-combined treatments for synchronization of oestrus in *post partum* crossbred (*Bos indicus* × *Bos taurus*) or zebu cows

S. M. LOKHANDE (1), D. R. INAMDAR (1), B. M. JOSHI (1),
M. R. BHOSREKAR (1), P. HUMBLLOT (2) and M. THIBIER (2, 3)

(1) Bharatiya Agro-Industries Foundation. Uruli-Kanchan, Pune, 412202, India.

(2) Union Nationale des Coopératives d'Élevage et d'Insémination Artificielle. Laboratoire d'Hormonologie, 13, rue Jouët, 94700 Maisons-Alfort, France.

(3) Institut National Agronomique Paris-Grignon. Département des Sciences Animales, 78850 Thiverval-Grignon, France. (To whom requests of reprints should be addressed).

RÉSUMÉ

LOKHANDE (S. M.), INAMDAR (D. R.), JOSHI (B. M.), BHOSREKAR (M. R.), HUMBLLOT (P.), THIBIER (M.). — Traitements combinés de prostaglandine et progestogène pour la synchronisation de l'oestrus chez des vaches métisses (*Bos indicus* × *Bos taurus*) ou zébu après gestation. *Rev. Elev. Méd. vét. Pays trop.*, 1984, 37 (1) : 73-78.

Cette étude a été réalisée dans l'Etat du Maharashtra (Inde) sur 248 vaches de deux génotypes distincts : Métisses (*Bos taurus* × *Bos indicus*, n = 146) et Zébu local (n = 102). Ces animaux ont été répartis au hasard dans deux groupes traités : l'un avec spirales vaginales-PMSG-PGF2 α (n = 91), l'autre avec implants sous-cutanés-PMSG-PGF2 α (n = 81). Un groupe témoin a reçu une injection de sérum physiologique (n = 76). Les vaches traitées ont été inséminées systématiquement 48 et 72 h après la fin du traitement et les témoins sur chaleurs observées. Celles-ci sont apparues chez tous les animaux traités lors des inséminations mais seulement chez 32 p. 100 des témoins dans les 21 jours suivant la période des traitements. Le taux de conception moyen à la première insémination artificielle a été de 50 p. 100 sans différence significative ni entre génotypes ni entre groupes malgré un pourcentage réduit de vaches Zébu gestantes dans le groupe témoin. Néanmoins, les pourcentages de vaches gestantes dans les 90 jours après les traitements diffèrent significativement entre génotypes (p < 0,02; 69 et 52 p. 100 respectivement pour les métisses et Zébu local). Cela est essentiellement dû à la fécondité médiocre des Zébu témoins.

De tels traitements combinés progestogènes-PMSG-PGF2 α peuvent donc être mis en oeuvre avec profit pour améliorer l'efficacité de la reproduction de ces populations bovines.

Mots clés : Maîtrise des cycles sexuels - Métisses laitières - Vache Zébu - Inde.

SUMMARY

LOKHANDE (S. M.), INAMDAR (D. R.), JOSHI (B. M.), BHOSREKAR (M. R.), HUMBLLOT (P.), THIBIER (M.). — Progestogen and prostaglandin-combined treatment for synchronization of oestrus in *post partum* crossbred (*Bos indicus* × *Bos taurus*) or zebu cows. *Rev. Elev. Méd. vét. Pays trop.*, 1984, 37 (1) : 73-78.

This trial was performed in Maharashtra (India) on 248 cows of two distinct genotypes : Crossbred (*Bos taurus* × *Bos indicus*, n = 146) and local zebu (n = 102). These animals were randomly allocated to one of the 3 following groups : (1) Silastic coils-Pregnant Mare Serum Gonadotropin (PMSG)-Prostaglandine F2 α (PGF2 α) combined treatment (n = 91), (2) Subcutaneous implant-PMSG-PGF2 α combined treatment (n = 81), and (3) controls (n = 76). All treated-cows were inseminated 48 and 72 hrs after end of treatment and controls were inseminated on observed heats. One hundred per cent of the treated animals were confirmed in oestrus at that time whereas only 32 p. 100 of the controls were seen in heat by 21 days following treatment. The conception rates at first A I were not significantly different between Crossbred and Zebu cows (mean : 50 p. 100) nor were they between any of the two treatments and control groups. However the percentages of cows bred 90 days after end of treatment, were significantly different between the two genotypes (p < 0.02; 69 and 52 p. 100 respectively for Crossbreds and non-descript Zebus) due to a low fecundity in the control zebu cows.

In conclusion, these progestogen-combined treatments seemed most promising in terms of breeding efficiency.

Key words : Oestrus control - Crossbred dairy cows - Zebu cows - India.

INTRODUCTION

Under temperate climate, anoestrus associated with ovarian inactivity is frequently encountered during the first two to three months after calving in nursing cows. In such groups of animals, an adequate treatment for the control of oestrous cycles should both synchronize and induce cycling activity. In European conditions, combined treatments associating progestogens, Pregnant Mare Serum Gonadotropin (PMSG) and prostaglandins were found to be the most efficient (3, 2, 13). Similarly under tropical environmental conditions, crossbred and zebu cows' reproductive efficiency is mainly limited by long periods of ovarian inactivity (14, 7, 4). Moreover heat detection is generally poorly accurate because of very discrete heats of these animals (15, 5), lack of training of the farmers and sometimes permanent tightened housing.

Consequently, oestrus synchronizing treatments may be most useful in order to artificially inseminate the local crossbred or zebu cows and hence both improve the mean breeding value of the offspring and the breeding efficiency of the dams.

The present trial was undertaken to compare the reproductive performances of crossbred or zebu cows under tropical conditions either submitted to oestrus synchronization treatment followed by pre-determined timing of insemination or just inseminated on naturally-occurring oestrus.

MATERIALS AND METHODS

Animals :

A total of 248 cows were involved in the present study undertaken in Maharashtra State (Pune; India; longitude 71°5 E; latitude 18°5 N) through the system of the BHARATIYA Agro-Industries Foundation (B.A.I.F.) cattle breeding centres. They were of two distinct genotypes (table 1) : (1) Crossbred (*Bos taurus* × *Bos indicus*) and (2) Local non-descript zebu (*Bos indicus*). The crossbred were from either Holstein or Jersey sires to the extent of 50 to 87,5 per cent of these genotypes. The dams were basically Gir or non-descript zebu cattle. Their lactation numbers were ranging from 1 to 4 and mean body weights were 475 and 300 kg

TABLE 1—Distribution of cows according to treatments and genotypes

	Silastic coils	Implants	Controls	Total
Crossbred	45	53	48	146
Local Zebu	46	28	28	102
Total	91	81	76	248

approximately respectively for crossbred and local cows at the time of treatment.

The trial was performed between October and January, i.e. the cooler season of the year (temperature ranges 22-32 °C). The crossed females were located in 6 different breeding areas.

All these crossbred cows were maintained in a zerograzing system and were fed with alfalfa (*Medicago sativa*) approximately 15-20 kg a day and green maize or sorghum about 20 kg complemented with adequate concentrates according to milk yield. Partial grazing was allowed for zebu, sorghum straw and sometimes concentrates were given additionally. Average milk productions (in 305 days) were about 3 000 kg for the crossbreds and 900 kg for the non-descript zebu cows.

The crossbred animals were selected as having calved for a range of days from 55 to 100 (mean 74 days). Intervals between calving to treatment could not be recorded for local cows but were estimated as in the magnitude of 6-7 months. In order to be included in the present trials, all cows were rectally palpated twice 10 days apart.

Animals with any reproductive abnormality or disease were excluded from the experiment.

Treatments :

The selected animals were randomly distributed (table 1) within the 2 genotypes and breeding areas in the 3 following groups :

- (1) Silastic coils-PMSG-PGF2 α
- (2) Subcutaneous implants-PMSG-PGF2 α
- (3) Controls.

A Silastic coil, so-called Progesterone Releasing Intravaginal Device (PRID-CEVA France) was inserted intravaginally for 10 days in group 1 females. The coils were having an attached capsule of oestradiol benzoate (10 mg). Two days before removal of coils, 25 mg PGF2 α (LUTALYSE-UPJOHN-France) were inject-

ted i.m. together with 400 i.u. Pregnant Mare Serum Gonadotropin (PMSG).

Group 2 was treated with a subcutaneous implant placed at the outer part of the ear, containing 6 mg of Norgestomet (17 α -acetoxyl, 11 β -methyl-19-nor-pregn-4-ene-3, 20 dione, INTERVET, France) and left for 10 days. At the time of insertion, an additional i.m. injection of oestradiol valerate (5 mg) + Norgestomet (3 mg) was given. Simultaneously to the previous group, PGF 2 α and PMSG were injected 2 days before removal of implant.

The control animals (group 3) were injected intramuscularly with 5 ml of saline on the day equivalent to day 1 of the treated group.

Heats were checked on the 2nd and 3rd days of the treated females and further confirmed by rectal palpation. In the control group, heats were observed by the herdsman twice a day.

Artificial Inseminations (A.I.) were performed in all these animals with frozen semen of European breeds bulls with known average fertility rates.

For control groups, A.I. was done on observed heats after the end of the 10 days treatment period. In treated groups, insemination time was pre-determined and performed at 48 and 72 hours after removal of coils or implants.

All dates of inseminations were carefully recorded and inseminated animals were confirmed for pregnancy by rectal palpation approximately 60 days after last breeding. Calving dates, and number of calves were also noted. No further treatment was performed in non pregnant animals after the first artificial insemination.

Statistics :

For analysis of these data, X² analysis and analysis of variance were performed according to SNEDECOR and COCHRAN (17). Multiple comparisons between groups were computed

according to Scheffe's method as reported in LELLOUCH and LAZAR (8). Probability values above 0.05 were not considered as significant.

RESULTS

Oestrus occurrence within treatment groups did not differ significantly between Crossbred and Zebu cows, hence results were pooled (table II). All treated cows were either seen or found in oestrus two to four days after end of treatment, by contrast only one third approximately ($p < 0.001$) of the controls was observed in oestrus in the 21 days following the end of the treatment period of the other two groups.

TABLE 2-Distribution (p.100) of detected heats according to groups

Type of oestrus	Silastic coils	Implants	Controls ⁽¹⁾
Standing oestrus ⁽²⁾	67 p.100	64 p.100	32 p.100
Silent oestrus ⁽³⁾	33 p.100	36 p.100	0 p.100
Absence of oestrus	0 p.100	0 p.100	68 p.100

(1) Cows seen in heat in the 21 days following the end of the treatment period of the other two groups ;

(2) Defined as a clearly seen oestrus by the farmer;

(3) Defined as a non reported-oestrus by the farmer but in fact confirmed after rectal and vaginal examinations on the expected days after treatment.

The mean conception rate on first A.I. was 50 p. 100 as illustrated in table III. The means in the crossbred and zebu cows although higher in the former group were not significantly different ($p > 0.05$). Moreover the rates observed in each of the treated groups did not differ significantly from those in controls although in the local zebu cows, this fertility rate was less than half of the silastic coils-treated females.

TABLE 3-Conception rates (p.100) at first oestrus following treatment or at first detected oestrus in the controls

Genotypes \ Groups	Silastic coils	Implants	Controls	Mean
Crossbred (N=146)	56 p.100 (N=45)	51 p.100 (N=53)	54 p.100 (N=48)	53 p.100
Zebu cows (N=84)	54 p.100 (N=46)	36 p.100 (N=28)	20 p.100 ⁽¹⁾ (N=10)	44 p.100
Mean (N=230)	55 p.100	46 p.100	48 p.100	50 p.100

(1) Only 10 of the 28 females of this group were seen in oestrus and hence inseminated.

As far as breeding efficiency is concerned, on the mean 69 p. 100 of the Crossbred and 52 p. 100 of the local Zebu cows were pregnant during the 90 days-period after treatment ($p < 0.02$). In the Crossbred population there was no significant difference of the mean percentages between the 3 groups (Table IV). By contrast, the mean zebu cows' conception rates were significantly different between groups ($p < 0.001$) as computed by the Scheffe's multiple comparison analysis. This is due to the much lower rate observed in the controls than in the other two groups. The latter have no significant different means ($p > 0.05$).

TABLE 4—Pregnant cows by 90 days after treatment and mean intervals (+ S.D.) from treatment to conception

Genotypes	Silastic coils	Implants	Controls
CROSSBRED			
Number	45	53	38
p. 100 pregnant	66 p.100	72 p.100	68 p.100
Days	18 ± 16	26 ± 25	31 ± 24
ZEBU			
Number	46	28	6
p. 100 pregnant	57 p.100	43 p.100	50 p.100
Days	13 ± 6	23 ± 22	87 ± 3

(a) Number of cows that were inseminated during this 90 days period.

As also shown in table IV, the mean intervals between treatment and conception of the Crossbred cows were in the range of 18-31 days with no mean difference ($p > 0.05$). Again by contrast, the control zebu cows had a much longer mean interval than the treated zebu cows ($p < 0.001$), yet only 6 were seen in heat out of which 3 got pregnant almost 90 days after the onset of the experiment.

Finally the twinning rates (including 3 triplets) were 25 p. 100 in both Crossbred or local treated cows vs 12 and 5 p. 100 respectively for control Crossbred and control Zebu cows.

DISCUSSION

The first striking feature of this study was the large difference observed in the heat detection rate between the treated and the control cows. This was excellent in the former and very poor

in the latter. Moreover this was not only so in the local zebu cows but also in the crossbred on which it is not unlikely that even better attention was paid because of more valuable females than the local ones. It is interesting to note that similarly to what has been recently reported in crossbred heifers by LOKHANDE *et al.*, (9), one hundred per cent of the treated cows came in heat within 2 to 4 days following coil or implant removal. This confirms that this type of combined treatments is very efficient in either inducing or synchronizing oestrus occurrence in herds where both cycling and non cycling females may be present together. These results are quite consistent to those reported under european (i.e. temperate climate) environmental conditions (10, 11, 2, 13).

The conception rates on first A.I. were over 50 p. 100 in each of the crossbred cows group which is very close to what is usually found in western temperate countries in cattle (6). The average conception rate in local zebu cows was lower than the latter due to the low rate found in the controls. However such a low conception rate does not seem exceptional as also reported by OYEDIPE *et al.* (12). It may of course be suggested that the difference found between crossbred and zebu cows are due to poorer management and feeding conditions in the local type of cattle herds but then it would stress even more the benefits of such treatments on fertility. It should be also noted that the results seen in the present study in the treated females are somewhat higher than those reported under tropical conditions after synchronization by a single progestogen treatment (7). Yet they are in the same order of magnitude than those found in Europe after progestogen alone (16, 18) or after a progestogen-PMSG combined-treatment (1, 13). It is not yet clear at this stage whether PGF₂ α administration two days before the end of treatment was of any necessity.

The high incidence of twins (or triplets) in treated-groups reflects a high sensitivity of these genotypes to PMSG treatment although doses of this gonadotropin-like hormone were here low on european standards (13). PETIT *et al.* have clearly shown in France some large variation in the response to a common dose of PMSG according to breed. They have also reported that even a small overdosage (100 or 200 i.u. over optimum) leads to a high increase of twins or triplets. As too high a percentage of twin births may be detrimental to further performances of the dams, it may be required to further

investigate lower doses of PMSG but yet achieving these satisfactory calving rates.

Finally, for crossbred cows, oestrus synchronization treatments did not hamper the breeding efficiency but did not improve it significantly either. However it must be stressed out that the oestrus detection procedure referred to here (twice a day) perhaps does not reflect accurately the common procedure used by the farmers in field conditions. Hence the potential income of such synchronization treatments may be here underestimated. In any event, this method keeps the advantage of better Artificial Insemination applicability and extension in the field. By contrast, reproductive performances of zebu cows were very much improved by these treatments both in terms of conception rates and intervals from treatment to conception. There-

fore for this type of cattle and/or farmers, such treatments seem to be most promising.

These encouraging results should now be confirmed by further trials on a larger scale to test both the feasibility and the average benefits.

ACKNOWLEDGEMENTS

The authors are indebted to the Senior staff members of the various Breeding Cattle Centers of the B.A.I.F. involved in this study for their kind and most efficient cooperation. They also wish to thank Drs F. DELETANG (CEVA-France), D. AGUER (INTERVET-France) and J. STEFAN (UPJOHN-France) for their generous gift of respectively the silastic coils (PRID), the Norgestomet implants and the Prostaglandins F₂α (Dinolytic).

RESUMEN

LOKHANDE (S. M.), INAMDAR (D. R.), JOSHI (B. M.), BHOSREKAR (M. R.), HUMBLLOT (P.), THIBIER (M.). — Sincronización del celo tras el parto en hembras del cruce *Bos indicus* × *Bos taurus* y en hembras de cebú, con distintas asociaciones de progestagenos y prostaglandinas. *Rev. Elev. Méd. vét. Pays trop.*, 1984, 37 (1) : 73-78.

Esta experiencia fue realizada en Maharashtra (India) sobre 248 vacas de 2 genotipos diferentes : cruce de *Bos taurus* y *Bos indicus* (n = 146) y raza local de cebú (n = 102). Estos animales fueron distribuidos al azar en 3 grupos : (1) vacas tratadas con una espiral vaginal-PMSG-PGF₂α combinados (n = 91); (2) vacas tratadas con un implante subcutáneo-PMSG-PGF₂α combinados (n = 81) y (3) grupo control (n = 76). Todas las vacas tratadas fueron inseminadas 48 y 72 horas tras el final del tratamiento, y las del grupo control lo fueron en el primer celo observado. El celo

en el momento de la inseminación fue confirmado sobre el 100 p. 100 de las vacas tratadas, mientras que sólo 32 p. 100 del grupo control fueron observadas en celo durante los 21 días posteriores al final del tratamiento de los otros animales. La proporción de gestaciones a la primera IA fue de 50 p. 100 aproximadamente, sin diferencias entre las dos razas ni entre ninguno de los grupos. En cambio, el porcentaje de vacas inseminadas 90 días después del tratamiento era significativamente mayor (69 p. 100) en el cruce que en el cebú local (52 p. 100) (p < 0,02), debido a la fecundidad mediocre del grupo control de esta raza.

En conclusión, estos tratamientos combinados de progestágenos ofrecen buenas perspectivas en cuanto a la eficacia de la reproducción por ellos inducida.

Palabras claves : Control del celo - Mestizas lecheras - Cebú - India.

REFERENCES

1. CHUPIN (D.), DELETANG (F.), PETIT (M.), PELOT (J.), LEPROVOST (F.), ORTAVANT (R.), PAREZ (M.) et MAULEON (P.). Utilisation de progestogènes ou implants sous-cutanés pour la maîtrise des cycles sexuels chez les bovins. *Annls. Biol. anim. Biochim. Biophys.*, 1974, 14 : 27-39.
2. CHUPIN (D.), PELOT (J.) et PETIT (M.). Induction et synchronisation de l'ovulation chez les femelles de races à viande. In *Physiologie et pathologie de la reproduction*. UNCEIA-ITEB Seminar, Paris, 1977, p. 43-48.
3. CHUPIN (D.), PELOT (J.), THIMONNIER (J.). The control of reproduction in the nursing cow with a progestogen short term treatment. *Annls. Biol. anim. Biochim. Biophys.*, 1975, 15 : 263-271.
4. CUQ (P.). Particularités du fonctionnement ovarien de la femelle zébu (*Bos Indicus*) dans la zone soudano-sahélienne de l'Afrique tropicale de l'Ouest. *Bull. Ass. Anatom.*, 1975, 59 : 139-144.
5. GALINA (C. S.), CALDERON (A.), Mc CLASKEY (M.). Detection of signs of estrus in the Charolais cow and its Brahman cross under continuous observation. *Theriogenology*, 1982, 17 : 484-498.
6. IRITANI (A.). Problems of freezing spermatozoa of different species. In : *Proc. 9th int. Congr. anim. Reprod. and A.I. Madrid*, 1980, 1 : 115-132.
7. JOCHLE (W.). The use of progestogens in cattle under tropical or subtropical conditions. *Annls. Biol. anim. Biochim. Biophys.*, 1975, 15 : 281-290.
8. LELLOUCH (J.) et LAZAR (P.). Méthodes statistiques en expérimentation biologique. Paris, Flammarion, 1974, 135 p.
9. LOKHANDE (S. M.), PATIL (V. H.), MAHAJAN (D. C.), PHADNIS (Y. P.), HUMBLLOT (P.), THIBIER (M.). Fertility on synchronized estrus in Crossbred (*Bos taurus* × *Bos Indicus*) heifers. *Theriogenology*, 1983, 20 : 391-406.
10. MAULEON (P.), CHUPIN (D.), PELOT (J.), AGUER (D.). Modifying factors of fertility after different estrus control treatment in beef cattle. In *Sreenan (J. N.), ed. Control of reproduction in the cow*. E.C.E. conference, Galway, 1977. Amsterdam, Martinus Nijhof, p. 531-545.
11. MULVEHILL (P.), SREENAN (J. N.). Oestrous cycle control and fertility in beef cattle following short term progestagen treatments. In *Sreenan (J. N.), ed. Control of reproduction in the cow*. E.C.E. Conference Galway, 1977. Amsterdam, Martinus Nijhof, p. 486-510.

12. OYEDIPE (E. O.), OSORI (D. I. K.), AKEREJOLA (O.), SAROR (O.). Effect of level of nutrition on onset of puberty and conception rates of zebu heifers. *Theriogenology*, 1982, **18** : 525-539.
13. PETIT (M.), DELETANG (F.), THIBIER (M.). Reproductive responses of beef heifers and cows to exogenous progesterone administered in silastic coils, oestradiol benzoate and pregnant mare serum gonadotropin. *Theriogenology*, 1978, **9** : 493-504.
14. PLASSE (D.), WARNICK (A. C.), ROGER (K.). Reproductive behaviour of *Bos indicus* females in a subtropical environment. I. Puberty and ovulation frequency in Brahman and Brahman × British females. *J. anim. Sci.*, 1968, **27** : 94-100.
15. RHODES (R. C.), RANDEL (R. D.). Reproductive studies of Brahman cattle. I. Behavioral effect of various doses of estradiol-17 β upon ovariectomized Brahman, Brahman × Hereford and Hereford cows. *Theriogenology*, 1978, **9** : 429-435.
16. ROCHE (J. F.). Calving rate of cows following insemination after a 12 day treatment with silastic coils impregnated with progesterone. *J. anim. Sci.*, 1976, **43** : 164-169.
17. SNEDECOR (G. W.), COCHRAN (W. G.). Statistical methods. Iowa State College Press, USA, 1956, 534 p.
18. WISHART (D.), YOUNG (I. M.), DREW (S. B.). Fertility of Norgestomet treated dairy heifers. *Vet. Rec.*, 1977, **100** : 417-420.