

**VOL 9, No. 2 :
DECEMBER, 1988**

ISSN 0970-2997



Indian Journal of Animal Reproduction

**JOURNAL OF THE
INDIAN SOCIETY FOR THE
STUDY OF
ANIMAL REPRODUCTION**

(Regd. No. Bom. 253/78)

Raymond's

EMBRYO RESEARCH CENTRE

The Raymond's Embryo Research Centre adopts Embryo Transfer Technology with unique distinction and dexterity. The centre had already produced 38 calves out of 71 pregnancies established by Embryo Transfer till May 1988, which itself is a living example of its dedication and expertise.



E. T. CALVES

If you are really in search of "Top class Holstein F Crossbred Breeding Bulls born out of Embryo Transfer please contact—



Dr. S. G. Zawar

Manager

Raymond's Embryo Research Centre

GOPALNAGAR-495 663

Dist. Bilaspur (M. P.)

Tel. No. 5389 & 5481

Telex No. 0273-215 RCW IN

THE INDIAN JOURNAL OF ANIMAL REPRODUCTION

Official Organ of the Indian Society for the Study of Animal Reproduction

(Regd. No. Bom. 253/78)

Vol. 9

No. 2

December 1988

EDITORIAL BOARD

A. Ramamohana Rao

Ph.D., F.R.V.C.S. (Sweden)

B. R. Deshpande

Ph.D., F.R.V.C.S. (Sweden)

M.L. Madan

Ph.D. (USA)

K. Singh

Ph.D. (Vienna)

Velhankar

Ph.D., F.R.V.C.S. (Sweden)

Benjamin

Ph.D. (Canada)

Prakiraman

Ph.D.

Hukeri

Ph.D.

Dr. C.R. Sane

D.Sc., F.R.V.C.S. (Sweden)

Dr. S. N. Luktuke

G.B.V.C., F.R.V.C.S. (Sweden)

Dr. H.C. Pant

Ph.D. (U.K.)

Dr. S.K. Gupta

Ph.D. (U.K.)

Dr. Y.G. Dugwekar

Ph.D.

Dr. I.S. Kohli

G.B.V.C., F.R.V.C.S. (Sweden)

Dr. K. Prabhakaran Nair

Ph.D.

Dr. S.B. Kodagali

Ph.D., F.R.V.C.S. (Sweden)

EDITOR

Dr. A. S. Kaikini

Ph.D., F.R.V.C.S. (Sweden)

Emeritus Scientist (ICAR) PKV,

Ex-Dean, Faculty of Veterinary Science, PKV

Annual Subscription Payable in Advance:-

Rs. 100/- to all Public/ Government and University Institutions/ Libraries. Foreign : U. S. \$ 50 by surface mail. Free to Life Members/ Regular Members/ Student Members of the Society (ISSAR)

Correspondence Address :

Editor, The Indian Journal of Animal Reproduction

B-306, Ujwal Flats, Rahate Colony, Jail Road,

Nagpur - 440 022

The Indian Society for the Study of Animal Reproduction
(Regd. No. Bom. 253/78)

Patron : Dr. C. R. Sane
D.Sc., F.R.V.C.S. (Sweden)

President :	Dr. A. Ramamohana Rao Ph.D., F.R.V.C.S. (Sweden)	Jt. Hon. Secretary :	Dr. B. N. De Ph.D., F.R.V.C.S. (Sweden)
Vice-President :	Dr. C. K. Rajkonwar Ph.D.	Hon. Treasurer :	Dr. S. R. Pattabiraman Ph.D.
Hon. Secretary :	Dr. D. R. Pargaonkar Ph.D., F.R.V.C.S. (Sweden)	Publication Editor :	Dr. A. S. Kaikini Ph.D., F.R.V.C.S. (Sweden)

EXECUTIVE COMMITTEE MEMBERS

Dr. G. M. Wani
Ph.D. (Jammu & Kashmir)

Dr. S. K. Verma
Ph.D. (Haryana)

Dr. B. L. Bishnoi
M. V. Sc. (Rajasthan)

Dr. R. Roy Choudhary
Ph.D. (West Bengal)

Dr. V. R. Jani
M. V. Sc. (Gujarat)

Dr. C. V. Dindorkar
Ph.D. (Maharashtra)

Dr. R. V. Patil
M. V. Sc., F.R.V.C.S. (Sweden) (Karnataka)

Dr. D. Kathiresan
M. V. Sc. (Tamilnadu)

Dr. M. K. Tamuli
M. V. Sc. (Assam)

Dr. R. D. Sharma
Ph.D. (Punjab)

Dr. S. V. Vadnere
Ph.D. (Uttar Pradesh)

Dr. K. G. Kharche
M. S. (USA) (Madhya Pradesh)

Dr. A. V. N. Rao
M. V. Sc., Dip. R. B. (Vienna)
T.C.T. (Canada) (Andhra Pradesh)

Dr. L.D. Mohanty
Ph.D. (Orissa)

Dr. S. A. Chaubal
M. V. Sc. (Bombay)

Dr. C. P. N. Iyer
Ph.D. (Kerala)

Dr. S. K. Ghosh
M.V.Sc. (West Bengal)

Correspondence Address :

Hon. Secretary, Indian Society for the Study of Animal Reproduction
C/o Department of Gynaecology and Obstetrics, College of Veterinary
and Animal Sciences, Marathwada Agricultural University,
PARBHANI—431402 (Maharashtra State)

EDITORIAL

We are happy to record that our earnest appeal (IJAR,8 (i) : June 1987) for strengthening the financial base of the Journal had a favourable response. We are thankful to all the office bearers and Members of ISSAR, State Directors of Animal Husbandry/Veterinary Services and the Advertisers for their ready response and request them for their continued and more sustained vigorous support.

We are extremely thankful for the annual financial assistance of Rs. 7,000/- received from the Indian Council of Agricultural Research (ICAR), New Delhi towards publication of this Journal (IJAR). We request the Council to enhance the assistance, hereafter, in view of the escalating cost of publication.

Rapid technological advances in all spheres are going on at the National level and Printing technology is no exception to it. The change for better will be evident from the current issue of IJAR, which is now published using modern techniques like Phototypesetting and Offset printing. We Sincerely thank Shri. G. Lawate, Printer and Shri. Shyam Bhoskar, Director Comp-Comp, Nagpur for their devotion in bringing out this issue in record time. The help rendered by Shri Ajit Gokarn, Chartered Accountant and Mrs. Shreelekha Kaikini is gratefully acknowledged.

We hope that in years to come, IJAR will be on a much sound financial base with the sustained active interest of all its readers, advertisers and supporters.

We wish you all a very Happy and Prosperous New Year - 1989.

-EDITORIAL BOARD.

THE INDIAN JOURNAL OF ANIMAL REPRODUCTION

CONTENTS

Vol. 9

No.2

DECEMBER 1988

EDITORIAL

1. Gynaecology and Obstetrics :

1. Integrated approach to enhance reproductive efficiency of a Herd.
- B. R. Deshpande
2. Pulsatile secretion of Luteinizing Hormone in jugular venous, carotid and ovarian arterial blood samples following Oestrus Synchronization in Ewes.
- P. V. Sarma and T. J. Reimers
3. Factors affecting Gonadotrophin Releasing Hormone induced LH and FSH Release in anoestrous cows.
- A. V. Narasimha Rao, W. M. Palmer and M. A. Sheikeldin
4. Effect of Gn RH Analogue (Buserelin) on some blood constituents in postpartum crossbred cows.
- S. B. Bagal and M. S. Kadu
5. Effects of various concentrations of Lugol's Iodine and Oxytetracycline on the Cytomorphology of the endometrium of Repeat Breeding cows
- Umed Singh, R. C. Gupta, D. N. Sharma and A. K. Sinha.
6. Studies on factors influencing calving interval in Nondescript Rural Cattle.
- S. Kumar, S. K. Agarwal, L.N. Purbey and R. Prakash
7. Studies on repeat breeder Friesian cows in Libya.
- K. M. Ben Haj and S. Abdulmola
8. Blood-serum Prolactin levels in Red Dane, Swedish Red and Jersey cross-breds during various phases of reproduction.
- C. V. Dindorkar, A. S. Kaikini and A. R. Sheth
9. Pattern of conception and calving in a crossbred herd.
- J. M. Patel, S. V. Shah, M. R. Dhangar and M. M. Trivedi
10. Incidence of certain types of oestrus and pathological termination of pregnancy.
- S. N. Luktuke and L. N. Purbey.
11. Leptospirosis abortion in cattle and buffaloes.
- R. C. Rajasundaram and S. Nedunchellian.

73

100

104

- 12 Bacteriological studies on the cervical mucus of repeat breeding cross-bred cattle, their treatment and conception rate.
- R. N. Sharma, B. K. Singh and M. P. Sinha 105
- 13 Service behaviour of bulls of different breeds under uniform management conditions.
- M. R. Bhosrekar, J. R. Purohit, A. B. Pande and B. R. Mangurkar 109
- 14 Different types of Scroti and preputial sheaths in crossbred bulls.
- K. Babu Rao and A. Ramamohan Rao 114
- 15 Types of preputial sheath in Murrah buffalo bulls.
- A. R. Rao, G. Somasekharam and K. Mouli Krishna 117
- 16 A Note on training of boars for collection of semen.
- M.K. Tamuli and C. K. Rajkonwar 118
- 17 Efficacy of tris-citric acid and sodium citrate buffers against cold shock to buffalo spermatozoa.
- R. K. Tuli, S. P. Singal and M. N. Razdan 122
- 18 Leakage of phosphatases from Buffalo bulls spermatozoa during freezing : Effect of dilutors, seasons, bulls and stages of freezing.
- A. J. Dhami and S. B. Kodagali 124
- Leakage of transaminases and lactic dehydrogenase in chilled semen of bucks of different breeds.
- A. K. Sinha, B. K. Singh and A. K. Sinha 131
- Effect of different concentrations of protective agents in extender and their influence on the quality of buck semen
- S. Prasad, B. K. Singh and M. P. Singh 134
- Protective effect of different concentrations of Dimethyl Sulfoxide (DMSO) and glycerol on freezing of goat embryos.
- S. V. Vadnere and S. V. Mani 136

Communications

- Spermine : Absence in goat semen.
- N. M. Markandeya, D. R. Pargaonkar and S. A. Bakshi. 138
- 23 A field method for collection of bovine cervical mucus for microbiological studies.
- Y. P. S. Dabas and S. N. Maurya. 138
- 24 Post-partum changes of the uterus and ovaries in relation to uterine microflora in cows
- K. C. Dekka, K. C. Nath and C. K. Rajkonwar. 139
- 25 Incidence of mycotoxic abortions in a dairy herd.
- B. Rameshkumar, R. C. Rajasundaram and A. C. Subramanian 141
- 26 A genital abnormality in vaginal passage in a Bannur ewe.
- S. R. Chinchkar, D. P. Velhankar, B. R. Deshpande and V. L. Deopurkar 142
- 27 Agenesis of mullerian duct system in a gilt.
- P. K. Sriraman, S. V. Ramachandraiah, K. Subramanyam Naidu and K. Krishna Reddy 143

3. Farm News

- 28 Pregnancy duration in Jersey Kankrej crosses.
- A. M. Patel 145

Induction of New Life Members**AN APPEAL**

The new Office Bearers of the ISSAR are striving their best to invigorate the Association from all angles. This is possible only if senior members take interest and play an active role in the affairs of the Association. We have about 200 life members on the rolls of our Society and I consider this number as too small. I therefore solicit earnest co-operation of all members to induct new life members. The life membership fee is Rs. 500/-. For further details you may kindly contact our Treasurer, Dr. S.R. Pattabiraman, Professor and Head, Department of Clinics, Madras Veterinary College, Vepery, Madras — 600007

A. Ramamohana R.
President ISSAR

With Best Compliments From :

**MEHSANA DISTRICT CO-OP. MILK PRODUCERS
UNION LTD. (DUDHSAGAR DAIRY)**

MEHSANA - 384 002

DUDHSAGAR

- * Second largest milk products manufacturing complex in the country.
- * Whole Milk Powder, Skim Milk Powder and Baby Food by Spray dried process under ISI Specifications and Table Butter and Ghee as per 'AGMARK' Std.
- * The only Co-operative enterprise with membership of 2,00,000 farmers in Mehsana District, providing input facilities such as :
Balanced Cattle Food, Veterinary services, A. I. with frozen semen, extension training programme, member's group insurance scheme and subsidy for cattle group insurance for construction societies buildings and common fodder farms for the benefit of landless marginal farmers and widows and distribution of Subabul for forestry.

V. M. PATEL
Managing Director

M. R. CHAUDHARI, (M.P.)
Chairman

Integrated Approach to Enhance Reproductive Efficiency of a Herd*

B.R. DESHPANDE

Ex-Head, Dept. of Animal Reproduction & Surgery, Bombay Veterinary College, Parel, Bombay-400012

The trend in the articles on Animal Reproduction published, appears to be less towards clinical research observations, which is evident from the table given below:

with all other species accounting for only 12.5% to 22.95%.

The success of breeding depends largely on the reproductive ability and prolificacy of the

Subject	Percent (over all)	Clinical observation	Lab. observation
aimed articles	23.65	63.93	27.87
of LH of Reproduction	26.36	48.53	50.00
simultaneous	52.72		
artery and def Reproduction		61.76	35.30
origin of ovarology		50.82	44.26
Mous	4.26		

Fig. 2: *Size of articles devoted to physiology and phase of reproduction (Gynaecology) on Obstetrics, A.I. and Andrology other half. Regarding clinical research, more than 60% articles are on etrics and pathology of reproduction, otherwise it is almost equal in other subjects. species-wise, cattle and buffaloes form the bulk of articles in all the subjects (77% to 87%)*

animals and the viability of their progeny. To achieve this, the animals should be maintained in good health and reproductive soundness. This is possible by resorting to good managerial practices in the herd- giving special attention to housing, nutrition, health care, good breeding practices and sexual health control including systematic record keeping. Fundamentally, the nature of occurrence and symptomatology of infertility

Species	Obstetrics	Gynaecology	A.I. & Andrology
Cattle & Buffaloes	77.05%	87.5%	80.33%
Sheep, Goats etc.	22.95%	12.5%	19.67%

*C.R. SANE ORATION delivered at the National Symposium of ISSAR on Animal Reproduction, held at Mannuthy (Trichur) from 22nd to 24th August, 1988.

or low reproductive efficiency is connected with hereditary factors and effect of environment. These two basic factors are particularly apparent in high yielding cows and large scale animal breeding.

Subfertility and infertility have been described as the largest sources of economic loss in livestock industry. About 30% of the economic loss from diseases in livestock is attributable to infertility. Problems of reproduction generally encountered in a herd are:

1. Pubertal anoestrous condition and postpartum anoestrous conditions. These are most important causes of infertility accounting for upto 43% in some herds.
2. Weak or silent oestrus which may be as high as 35% of oestruses in a herd.
3. Anovulatory heats, prolonged heat (delayed ovulation) and split oestrus cases can only be diagnosed by continuous rectal examination for 3 to 4 days as well as at an interval of 8-10 days for the presence of GF and CL.
4. Repeat breeding problem varies from 8 to 20 per cent in the herds.
5. Genital infections: Endometritis varies from 9 to 14%. Puerperal metritis is observed generally 4th day after parturition. Cervicitis 9 to 27%, Salpingitis 4 to 17%, Brucellosis 4% and may reach 25-30% during abortion storm. In bulls the following problems are encountered
 1. Lack or loss of libido
 2. Serving inability
 3. Seminal defects
 4. Morphological abnormalities of sperm
 5. Genital infections.

To enhance reproductive efficiency of the herd, following measures are suggested:

1. *Anoestrous condition*: It is advisable to weigh heifers and cows every 15 days or one month and record the weights. Those not gaining weights should be put on higher plane of nutrition.
2. *Detection of Nutritional deficiencies*: Extra concentrate feeding during postpartum period results in earlier weight gains and improved conception. Cows which gain weight, conceive better. Since the cow has a negative energy balance after parturition, addition of feed stuffs to maintain a positive balance, improves conception.
3. *Various indigenous drugs*: Intrusions to a (Indian Herbs), Aloes and return to (Alarsin) and Fervivet (Aurs (Zolman *et al*)) have been found effective in inducing oestrus, simulating the above heat that occurs fulfilled and there is no gap. The ability of response to
4. *Hormonal assay of progesterone*: reveals the imbalance and by several remedial measures can be undulating. The progesterone content has been found to be 1 to 3 ng/100 ml. in anoestrous condition which rise to 6 ng/100 ml. when animal returns to normal.
5. *Proper detection of oestrus*: The best remedy for weak or silent oestrus is to use teaser bulls 3 to 4 times a day to detect heats. In addition, a regular sexual health control programme should be followed.
6. *Diagnostic aids to detect genital infections* (specific/non-specific) including sexually transmitted diseases should be provided. Timely diagnosis and treatment with antibiotics after sensitivity

test of the vaginal discharge, is essential to prevent further complications.

7. To have an immediate idea of the nature of the infertility problem, the following records be studied.

8. Effect of climatic elements on reproduction have been investigated and the impact should be taken into account.

9. Other important components include the breeding bulls. The semen of proven bull

High Fertility Herd

Total 1st	No. Pregnant to					Pregnant %	
Services	1st	2nd	3rd	4th	Multiple	1st	2nd
50	38	9	2	1	—	76%	94%

The Interval between repeat services in days

aimed abort of LH simultaneous	17—25	26—33	34—50	Over 50
	15	—	—	1

artery and delivery figures are read after 3 months. The fact that all but one cow repeated at a 3 week origin of ova (within 17 to 25 days) is an indication of genital health.

Low Fertility Herd

Fig. 2: Pulsatile 1st spr	No. Pregnant to					Pregnant %	
pulsatile 1st spr	1st	2nd	3rd	4th	Multiple	1st	2nd
phase v 58	36	15	2	3	2	62%	71%

The Interval between repeat services in days

Short	17—25	26—33	34—50	Over 50
1	22	1	8	5

The number of service required per conception in this herd trailed on into multiple category after a moderate first service success rate of 62%. The high proportion of intervals between repeat services falling into 6 weeks or more categories, indicate irregular oestrous cycles or early abortions.

is always advocated. The importance of Andrological investigation of the breeding bull for reproductive soundness and certification cannot be minimised. At present, it is lamentable that there are hardly any bulls whose dam's lactational milk yield is at least 5000 lts. leave aside proven bulls. An expert committee

consisting of Geneticist, Andrologist, Gynaecologist, A.I. and Animal Husbandry Scientists should sit together and give a serious thought to this.

10. It is necessary to inspect the Deep Frozen semen at regular intervals for the bacterial flora; higher percentage would affect the fertility.
11. Due attention to the protective clothing of Veterinarians involved in the activities of animal reproduction, has not yet been paid. Veterinarian may become the source of infection to animals. As such, it will be of immense importance that, each Polyclinic, A.I. Centre, Department of

Gynaecology etc. should use protective clothing and have a separate cleaning and washing room for protective clothing used by the staff.

12. Lastly, record keeping of the cases pertaining to Obstetrics, Gynaecology, A.I. and Andrology by the staff in Veterinary hospitals, polyclinics, milk federations, A.I. Centres is not uniform. In this regard, the Director of Animal Husbandry, M.S. Pune has agreed to the suggestion to have a joint meeting of staff in this field to work out formats to maintain the records. Hope the other States will also follow this example.

IJAR 9:2: 76-80:1988

Pulsatile Secretion Of Luteinizing Hormone In Jugular Carotid And Ovarian Arterial Blood Samples Following Synchronization In Ewe*

P.V. SARMA** and T.J. REIMERS

Endocrinology Laboratory, New York State College of Veterinary Medicine Cornell University, Ithaca, NY 14853

ABSTRACT

Oestrus cycle of a non-pregnant ewe with polyvinyl catheters implanted surgically into external jugular vein, carotid artery and near ovarian artery, was synchronized with progesterone impregnated vaginal sponge. Concentration of progesterone in serum was monitored to identify the occurrence of oestrus after removal of vaginal sponge kept for 16 days. Blood samples for quantifying luteinizing hormone were collected simultaneously from all the three sampling sites every 15 min for 8 hrs on day of oestrus (day 0), early diestrus (day 3) and late diestrus

(day 8) of the post synchronization. Response to LH pulses were studied for the several appearance, frequency and amplitude at the three sites. There was a remarkable resemblance in LH pulses observed from the three sites during any period of study. During the day of oestrus (day 0), the LH pulse frequency was high (11-12 pulses/8 hr) but of low amplitude (1.75 ng/ml), compared to low frequency (3-4 pulses and 5-6 pulses/8 hr) but high amplitude (2.9 ng/ml and 5.1 ng/ml) pulses observed during early (day 3) and late (day 8) diestrus periods of post synchronization oestrus respectively. ***

*Study conducted as a part of training in radioimmunoassay of reproductive hormones under SIDA/IAEA fellowship by the first author.

**Present address: Scientist S-2, Southern Regional Station, N.D.R.I., Bangalore- 560 030.

A pulsatile pattern of LH release in peripheral circulation has been reported in ewes (Hauger *et al.*, 1977) and in cows from birth to puberty and during oestrus cycles (Rahe *et al.*, 1980; Schams *et al.*, 1981). The basic characteristic mode of LH secretion in farm and laboratory animals has also been reviewed (Weick, 1981). However, it is not yet clear whether this pulsatile nature of LH observed in peripheral circulation such as in jugular venous samples will remain as discrete pulses, when reach and impinge upon ovaries after passing through heart and lungs and being diluted by a large volume of blood from the lower body. The present investigation was aimed at characterization of secretory pattern of LH in blood samples collected simultaneously from jugular vein, carotid artery and descending aorta at a site near the origin of ovarian arteries in ewe.

Materials and Methods

Fig. 2: us cycle of a non-pregnant ewe was
Pulsati- sized with progesterone impregnated
sponge. While sponge is in place,
catheters were implanted surgically
phase al jugular vein, a carotid artery and
al., 10 (via a branch of femoral artery) of
ewe. The tip of the aortic catheter was
located rostral to the origin of ovarian artery
so that the blood samples drawn through this
catheter represent the blood supply to the
ovaries. The jugular venous and carotid
arterial catheters were passed subcutaneously
and exteriorized on the top of the neck of the
animal while the aortic arterial catheter was
exteriorized on the back of the animal. All the
catheters were connected to a syringe pump
with a 3 way stopcock located at the rear end
of the cage where the animal has been
restrained. This facilitated the infusion of
heparinised saline to maintain the patency of
catheters and withdrawal of blood samples for

different sites simultaneously without disturbing the animal.

Intravaginal sponge was removed after 16 days. Concentration of progesterone in serum samples was determined by radioimmunoassay (RIA) technique (Reimers *et al.*, 1983) for monitoring the occurrence of post synchronization oestrus. Blood samples (1.5 ml each) for quantifying LH were collected every 15 min. for 8 hr. from all 3 sampling sites simultaneously during oestrus (day 0), early diestrus (day 3) and late diestrus (day 8) of post synchronization oestrus as indicated by the progesterone concentration reaching a nadir. Serum obtained from the blood samples were stored at -20°C until subjected to radioimmunoassay for LH (Reimers *et al.*, 1983). Pulses of LH in serum samples from all the three sampling sites during different periods were examined for general appearance, amplitude and frequency.

Results and Discussion

Patterns of LH secretion in serum samples on day 0, day 3 and day 8 of post synchronization oestrus from all the three sampling sites are presented in Fig. 1 to 3.

It was astonishing to note that the pattern of LH pulses in jugular venous samples bore a remarkable resemblance to that observed in carotid or ovarian artery in spite of its passage through heart and lungs and being diluted by a large volume of blood from the lower part of the body to be detected as discrete pulses, irrespective of sampling day and time. The approach of Mc Neilly *et al.* (1982) to induce ovulation and luteal function by pulsatile infusion of LH into the proximal blood supply and by Paluso *et al.* (1983) into organ culture of whole ovaries to study folliculogenesis seems to be quite justified, in that the administration of exogenous LH in

pulsatile manner could be perceived by the ovary.

During the day of oestrus (day 0, fig. 1), the LH pulse frequency was about 12 pulses/8 hr and the amplitude of the pulses ranged from 0.75 to 1.75 ng/ml from a basal level of about 0.5 ng/ml. Insignificant variation either

and Convey, 1983), invariably resulted in a characteristic pattern of high frequency but low amplitude pulses in cows (Rahe *et al.* 1980) and in ewes (Baird, 1978) on close sampling. It has also been pointed out that the progesterone plays 'a key role in the establishment of a pulsatile pattern of

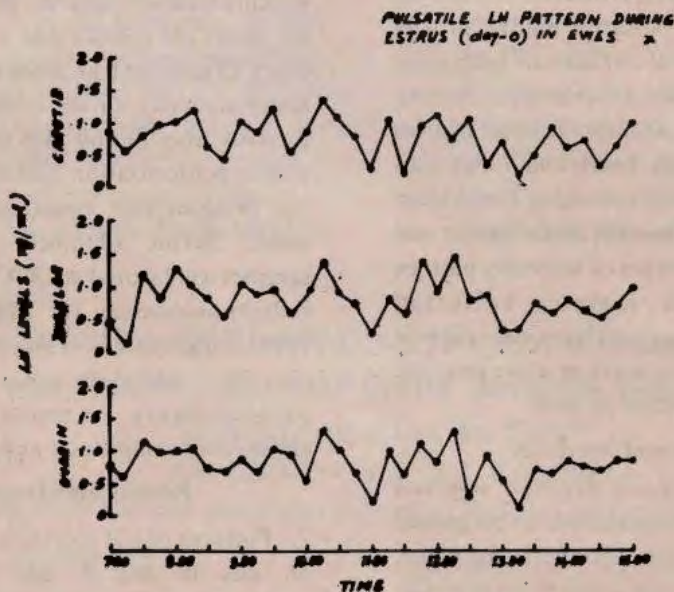


Fig. 1: Pulsatile LH pattern during Estrus (day-0) in ewes

in pulse frequency or pulse amplitude, between the three sampling sites at any given period was noticed. Generally, the nature of the pulse was that of high frequency but low magnitude. Similar increase in LH pulse frequencies were also reported by Goodman and Karsch (1980, 1981). However, high LH peaks at or shortly before beginning of oestrus cycles as observed in cows (Hensel and Snook, 1970) could not be detected in the present study. Increase in LH concentration in the peripheral blood following natural or prostaglandin induced luteal regression or withdrawal of progesterone treatment (Hansel

gonadotrophic secretion, appropriate development of an ovulatory surge (Schäperclaus *et al.*, 1981) and increased secretion of oestradiol from the preovulatory graafian follicle due to the repeated stimulation of episodic pulses of LH which occur with increasing frequency (Baird, 1978). At least in ewes, the increase in the number of LH pulses of small physiological amount are sufficient to induce follicular maturation, ovulation and formation of functional corpus luteum (Mc Neilly *et al.*, 1982) in anoestrus conditions and a failure of normal preovulatory follicular development was invariably associated with inadequate frequency of pulsatile LH release.

During early diestrus stage (day 3, fig. 2) the pulse frequency was about 3 to 4 pulses/8 hr with a pulse amplitude between 0.75 to 2.9 ng/ml as compared to 5 to 6 pulses of 0.75 ng/ml to 5.1 ng/ml amplitude observed during the late diestrus period (day 8, fig. 3). A

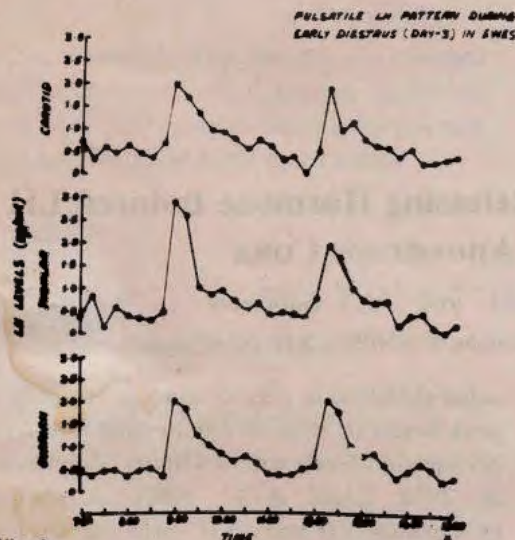


Fig. 2:

Pulsatile LH pattern during early Diestrus (day-3) in ewes

pulsatile pattern of LH secretion during luteal phase was also observed in cows (Rahe *et al.*, 1980) besides in ewes (Hauger *et al.*, 1977). Increase in LH pulse frequency during the luteal phase nadir of one pulse every 3 to 4 hr to a maximum of about one pulse every 30 min. just prior to the onset of preovulatory surge (Baird, 1978, Goodman and Marsch, 1981) has also been reported. Our findings, in

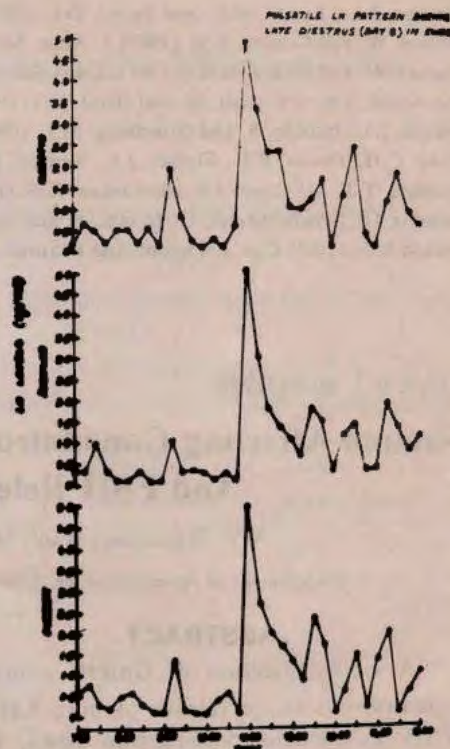


Fig. 3:

Pulsatile LH pattern during late Diestrus (day-8) in ewes

the present study also revealed similar pattern. Thus the frequency and magnitude of LH pulses change with changes of steroid hormone secretion. Low frequency, high amplitude pulse was associated when progesterone is dominant and high frequency, low amplitude pulses under oestrogen dominance (Rahe *et al.*, 1980), both hormone playing a role in a negative feed back control of gonadotrophic secretion.

REFERENCES

- Baird, D.T. (1978) *Biol. Reprod.* **18**, 359.
- Goodman, L.R. and Karsch, T.J. (1980) *Endocrinology* **107**, 1286.
- Goodman, R.L. and Karsch, F.D. (1981) In *Biological clocks and seasonal Reproductive cycles* (B.K. Follette and D.E. Follette, eds.)
- Wright, Bristol, p. 223.

- Hauger, R.L., Karsch, R.D. and Foster, D.L. (1977) *Endocrinology*, **101**: 807.
 Hansel, W. and Convey, E.M. (1983) *J. Anim. Sci. (Suppl.2)* **57**: 404.
 Hansel, W. and Snook, R.B. (1970) *J. Dairy Sci.* **53**: 945.
 Mc Neilly, A.S., O'Connel, M. and Baird, D.T. (1982) *Endocrinology*, **110**: 1292.
 Peluso, J.J., Juttmer, S. and Gruenberg, M.L. (1983) *proc. soc. Study—Reprod. (Abstr.)* 54.
 Rahe, C.H., Owens, R.E., Fleeger, J.L., Newton, H.J. and Harms, P.G. (1980) *Engocrinology*, **107**: 498.
 Reimers, T.J., Mc Cann, J.P. and Cowan, R.G. (1983) *J. Anim. Sci.* **57**: 683.
 Schams, D., Schallenberger, E., Gombe, S. and Karg, H. (1981) *J. Reprod. Fert. (Suppl. 30)*, 103.
 Weick, R.F. (1981) *Can. J. Physiol. and Pharmac.* **59**: 779.

IJAR 9:2: 80-84:1988

Factors Affecting Gonadotrophin Releasing Hormone Induced LH And FSH Release In Anoestrous Cows

A.V. NARASIMHA RAO¹, W.M. PALMER and M.A. SHEIKELDIN

Department of Animal Science, University of Manitoba, WINNIPEG, R3T 2N2 (Canada)

ABSTRACT

A single injection of GnRH induced a synchronous surge release of both LH and FSH in suckled post-partum cows. High ambient temperature ($>22.1^{\circ}\text{C}$), early post-partum (38-50 days), low body weight (<600 Kg) and high body condition score (>7) were associated with diminished pituitary sensitivity to release LH in response to GnRH. Exogenous oestradiol- 17β given 6 h earlier to GnRH, selectively augmented LH response and the E_2 modulation on LH release was affected by post-partum interval and body condition. The FSH release, however, was not limited by any of these factors studied.

* * *

Systemic administration of Gonadotrophin Releasing Hormone (GnRH) in cattle induces a surge release of both LH and FSH from the anterior pituitary gland, causing a

rapid elevation in plasma concentrations to a peak height in 30 to 60 minutes and return to preinjection levels within 4 hours (Zolman *et al.* 1974; Zaied *et al.* 1980), simulating endogenous LH and FSH surge that occurs during oestrus in cyclic cows. The ability of the pituitary to release LH in response to exogenous GnRH is influenced by several physiological factors and the modulating effect of gonadal steroids.

The present study evaluates the dynamic of hormonal modulation on LH and FSH response to GnRH in normal and oestradiol pretreated suckled cows in relation to post-partum interval, body weight, body condition score and ambient temperature at the time of treatment.

Materials and Methods

Experiments were carried out at the Glenlea Research Station of the University of

¹Assistant Director (AH), Visakha Co-op. Dairy (P.O.) Government Dairy Farm, VISAKHAPATNAM-530040, Andhra Pradesh.

Manitoba, Canada, during 1984. The experimental data on 16 spring-calved, mature crossbred anoestrous cows at 38 to 62 days postpartum in moderate body condition, were utilised in the present study.

The cows were treated with either a single intramuscular injection of 250 μ g GnRH (Factrel, Ayerst Laboratories, Montreal, Canada) (n=6) or 1 mg E_2 (Oestradiol-17 β , Sigma Chemical Co., St. Louis, USA) in arachis oil, followed 6 hours later by 250 μ g GnRH (n=10).

Blood samples were collected from indwelling jugular vein catheters at 2 h intervals before and after injection of E_2 and at intervals of 0.5 h for 8 h, 1 h till 12 h and 2 h until 24 h after administration of GnRH. The blood samples were allowed to clot at 4°C for several hours and the serum was removed and stored at -20°C until assay.

LH was quantified by a specific double-antibody RIA described by Howland (1972) using anti-ovine LH serum. Sensitivity was 0.13 ng/ml. The intra and inter-assay coefficient of variation was less than 8%.

FSH was determined by a specific double-antibody RIA procedure of Chang (1978). The mean sensitivity of the assay was 0.10 ng SDA-FSH-BP 3 eq/ml (28 \times NIH-FSH-S1). Intra and inter-assay coefficients of variation were 9 and 12% respectively.

A computer model was used to estimate the area under the response curve using values at least two standard deviations above the mean preinjection concentrations. The endocrine data were analysed by the split-plot analysis of variance (Gill and Hafs, 1971) due to repeated sampling of cows. The effects of ambient temperature, post-partum interval,

body weight and body condition score were studied by Student's 't' test.

Results and Discussion

Mean serum LH and FSH concentrations before and after treatment are shown in Table 1.

Effect of GnRH: Single injection of GnRH induced a surge release of LH immediately resulting in significant ($P<0.01$) elevation of serum LH levels at an interval of 160 ± 1.34 m post-treatment. The induced response lasted for 6.58 ± 0.44 h before reaching to basal levels with a total LH response of 44.0 ± 11.21 units.

Mean serum concentration of FSH was also elevated significantly ($P<0.01$) with a temporal pattern of FSH release similar to that seen for LH but with a smaller peak height, shorter duration and lesser total response. The pattern of LH and FSH release in response to GnRH was more or less similar to that reported by Zolman *et al.* (1974) in cattle.

Effect of oestradiol pretreatment: Injection of Oestradiol-17 β 6 h prior to GnRH produced a greater magnitude of LH response with higher mean values of peak height ($P<0.01$) duration and total LH response ($P<0.01$) as compared to that seen after GnRH alone. The interval to peak was shorter. This LH response was of the same order of magnitude of the endogenous peaks of 15 to 25 ng/ml and duration of 8 to 10 h found at natural oestrus (Chenault *et al.* 1975).

The present results are consistent with the earlier reports on the potentiating effects of oestradiol on Pituitary responsiveness to LH-RH in cows both *in vitro* (Padmanabhan and

Convey, 1978) and *in vivo* (Kesner *et al.* 1981), presumed to result from increased LH-RH receptors which normally were elevated at oestrus when pituitary responsiveness was highest and depressed when not in oestrus (Schonemann *et al.* 1985).

The FSH response, however, did not differ significantly from the response to single GnRH (Table 1) demonstrating that the sensitizing effect of oestradiol on pituitary was limited to the release of LH only but not to the release of FSH, corroborating the observation of Padmanabhan and Convey (1981) made for *in vitro* studies in cattle.

Effect of ambient temperature: The mean initial serum concentrations of LH and FSH for the cows treated at low ($<20^{\circ}\text{C}$) or high ($>20.1^{\circ}\text{C}$) ambient temperature were similar. A greater magnitude of LH surge with a higher peak height ($P<0.05$) and total response ($P<0.05$) but not FSH at lower, compared to higher ambient temperature was seen. The E_2 primed cows also responded better at lower temperature (Table 2).

The differential effects of temperature on LH and FSH release indicate a degree of specificity, LH being selectively affected. The results are congruous with the observation of Madan and Johnson (1973) who found lower plasma LH concentrations in cows exposed to hot environmental temperatures.

Effect of post partum interval: The mean serum LH and FSH for the cows in early (38-50 days) or late (51-62 days) post-partum did not show difference. The LH response to GnRH was marginally high in the late post-partum cows. The trend, however, was reversed in E_2 primed cows with a tendency towards greater LH release in early than in late post-partum cows ($P<0.10$). FSH response did not differ (Table 2).

Effect of body weight: The body weight of cows had no effect on mean serum LH and FSH concentrations before or after GnRH treatment. Pretreatment with E_2 elicited greater LH response ($P<0.10$) in heavier (>600 kg) than in lighter (<600 kg) cows.

Effect of body condition score: Whereas the mean LH before treatment was similar in cows with higher (>7) or lower (<6.9) scores (scored from 1 to 10), GnRH tended to release more LH in lean cows. In contrast, E_2 followed by GnRH resulted in greater ($P<0.10$) LH release in fatty cows. Such differences in FSH response were not evident (Table 2).

The degree of potentiation and the consequent LH release following E_2 priming in relation to different physiological conditions was highly variable displaying release patterns contrasting to that seen following single GnRH. Thus the combined treatment evoked greater LH response in cows in early post-partum ($P<0.10$) and in fatty cows ($P<0.10$), which is suggestive that these conditions are normally associated with diminished pituitary responsiveness and low LH-RH receptor content. Since the pituitary LH content in early or late post-partum and in lean or fatty cows was similar (Resby *et al.*, 1986) the E_2 stimulation might increase the LH-RH receptors and consequently the size of the pool of readily releasable LH in the pituitary.

In contrast, administration of saline (vehicle) or E_2 did not elicit and pituitary gonadotrophin response in anoestrous cows (Rao, 1985).

Acknowledgement

The award of visiting Fellowship of the Canadian International Development Agency and the Natural Science and Engineering Research Council, Canada to the senior author is gratefully acknowledged.

REFERENCES

- Chenault, J.R.; Thatcher, W.W.; Kalra, P.S.; Abrams, R.M. and Wilcox, C.J. (1975). Transitory changes in plasma progestins, estradiol and luteinizing hormone approaching ovulation in the bovine. *J. Dairy Sci.* **58**: 709.
- Cheng, K.W. (1978). Development and characterization of a homologous radioimmunoassay for bovine follicle stimulating hormone. *J. Endocr.* **77**: 185.
- Gill, J.L. and Hafs, H.D. (1971). Analysis of repeated measurements of animals. *J. Anim. Sci.* **33**: 331.
- Howland, B.E. (1972). Effect of restricted feed intake on LH levels in female rats. *J. Anim. Sci.* **34**: 445.
- Kesler, K.J.; Garverick, H.A.; Youngquist, R.S.; Elmore, R.G. and Bierschawl, C.J. (1977). Effect of days postpartum and endogenous reproductive hormones on GnRH- induced LH release in dairy cows. *J. Anim. Sci.* **45**: 797.
- Kesner, J.S.; Convey, E.M. and Anderson, C.R. (1981). Evidence that oestradiol induces and preovulatory LH surge in cattle by increasing pituitary sensitivity to LH-RH and then increasing LH-RH release. *Endocrinology* **108**: 1386.
- Madan, M.S. and Johnson, H.D. (1973). Environmental heat effects on bovine LH. *J. Dairy Sci.* **56**: 1420.
- Padmanabhan, V. and Convey, E.M. (1978). Does time of exposure of estradiol and LH-RH effect LH release from bovine pituitary cells. *Proc. Soc. Exp. Biol. Med.* **159**: 157.
- Padmanabhan, V. and Convey, E.M. (1981). Progesterone inhibits the ability of estradiol to increase basal and LH-RH- induced LH release from bovine pituitary cells in culture: Neither progesterone nor estradiol affects FSH hormone release. *Endocrinology* **109**: 1091.
- Rao, A.V.N. (1985) - Unpublished data.
- Resby, R.J.; Wagner, J.W.; Wettmann, R.P.; Giesert, R.D. and Lusby, K.S. (1986). Influence of body condition of beef cows on pituitary and ovarian function. *Ani. Sci. Rs. Report, Agrl. Exp. Sta. Okalahoma St. Uni. M.P.* **118**: 333.
- Schonemann, H.M.; Brown, J.L. and Reaves, J.J. (1985). LH-RH receptor, LH and FSH concentrations in anterior pituitaries of cycling, noncycling and early pregnant heifers. *J. Anim. Sci.* **60**: 532.
- Zaied, A.A.; Garverick, H.A.; Bierschawl, C.J.; Elmore, R.G.; Youngquist, R.S. and Sharp, A.J. (1980). Effect of ovarian activity and endogenous reproductive hormones on GnRH. *J. Anim. Sci.* **50**: 508.
- Zolman, J.; Convey, E.M.; Britt, J.H. and Hafs, H.D. (1974). Relationships between the luteinizing hormone responses to gonadotropin releasing hormone and endogenous steroids. *J. Anim. Sci.* **39**: 355.

Table 1: GnRH - Induced LH and FSH response data in anoestrous cows pretreated with and without Destrodiol- 17 β

Ex- di- st- ment	Mean basal concentration (ng/ml)	Mean interval to peak (m)	Mean peak height (ng/ml)	Mean duration of response (h)	Mean area under response curve (units)
LH					
GnRH	0.34 \pm 0.07 ^a	160 \pm 1.34 ^a	8.35 \pm 2.71 ^a	5.58 \pm 0.44 ^a	44 \pm 11.21 ^a
F ₂ + GnRH	0.35 \pm 0.06 ^a	126 \pm 0.56 ^a	21.40 \pm 4.88 ^b	7.65 \pm 0.18 ^a	100 \pm 15.05 ^b
FSH					
GnRH	0.36 \pm 0.05 ^a	130 \pm 1.02 ^a	1.81 \pm 0.17 ^a	4.70 \pm 0.14 ^a	9 \pm 2.12 ^a
F ₂ + GnRH	0.30 \pm 0.06 ^a	124 \pm 0.82 ^a	2.59 \pm 0.34 ^a	5.02 \pm 0.14 ^a	16 \pm 1.69 ^a

Values with different superscripts within the same column are different (P<0.01)

Table-2: Effect of ambient temperature, post-partum interval, body weight and body condition score on GnRH- Induced LH and FSH response in anoestrous cows pretreated with and without oestradiol- 17 β

	Serum 'LH'			Serum 'FSH'		
	Basal concentration (ng/ml)	Peak height (ng/ml)	Area under response curve (units)	Basal concentration (ng/ml)	Peak height (ng/ml)	Area under response curve (units)
<i>Ambient temp.</i>						
GnRH (<22 ⁰ C)	0.21 ^a	18.30 ^c	94 ^c	0.45 ^a	1.93 ^a	12 ^a
GnRH (>22.1 ⁰ C)	0.48 ^a	6.03 ^a	33 ^a	0.38 ^a	1.69 ^a	8 ^a
E ₂ +GnRH (<22 ⁰ C)	0.24 ^a	25.10 ^c	112 ^c	0.16 ^a	2.43 ^a	14 ^a
E ₂ +GnRH (>22.1 ⁰ C)	0.43 ^a	13.88 ^{ac}	65 ^{ac}	0.50 ^a	2.75 ^a	18 ^a
<i>Post-partum interval</i>						
GnRH (38-50 days)	0.24 ^a	7.73 ^a	39 ^a	0.34 ^a	1.76 ^a	9 ^a
GnRH (51-62 days)	0.44 ^a	8.97 ^a	48 ^a	0.49 ^a	1.86 ^a	10 ^a
E ₂ +GnRH (38-50 days)	0.35 ^a	32.90 ^b	151 ^b	0.27 ^a	2.63 ^a	17 ^a
E ₂ +GnRH (51-62 days)	0.34 ^a	13.73 ^a	66 ^a	0.39 ^a	2.55 ^a	15 ^a
<i>Body Weight</i>						
GnRH (> 600 kg)	0.17 ^a	10.10 ^a	56 ^a	0.39 ^a	1.95 ^a	12 ^a
GnRH (< 600 kg)	0.52 ^a	7.98 ^a	45 ^a	0.44 ^a	1.67 ^a	7 ^a
E ₂ +GnRH (> 600 kg)	0.31 ^a	28.20 ^b	123 ^b	0.17 ^a	2.51 ^a	15 ^a
E ₂ +GnRH (< 600 kg)	0.37 ^a	16.03 ^a	80 ^a	0.49 ^a	2.67 ^a	17 ^a
<i>Body Score</i>						
GnRH (>7)	0.24 ^a	7.45 ^a	38 ^a	0.38 ^a	1.46 ^a	7 ^a
GnRH (<6.9)	0.45 ^a	13.15 ^a	64 ^a	0.45 ^a	2.16 ^a	12 ^a
E ₂ +GnRH (>7)	0.48 ^a	30.22 ^b	140 ^b	0.22 ^a	2.50 ^a	15 ^a
E ₂ +GnRH (<6.9)	0.20 ^a	14.60 ^a	62 ^a	0.44 ^a	2.68 ^a	17 ^a

Values with different superscripts within the same column are different (ab-P<0.10, ac-P<0.05)

Effect Of GnRH Analogue (Buserelin) On Some Blood Constituents In Postpartum Crossbred Cows

S.B. BAGAL and M.S. KADU

Post Graduate Department of Animal Reproduction and Surgery, Punjabrao Krishi Vidyapeeth, Akola-444104

ABSTRACT

A significant low level of serum cholesterol was observed in treatment group at first post partum ovulation (143.59 ± 7.72 Vs 162.37 ± 14.39 mg per cent) and at detected oestrus (141.63 ± 7.64 Vs 173.20 ± 9.88 mg per cent) as compared to control group.

The blood glucose, serum calcium, inorganic phosphorus and haemoglobin at first ovulation in treatment and control groups were 45.17 ± 2.43 Vs 48.57 ± 1.15 mg percent, 9.19 ± 0.49 Vs 19.48 ± 0.57 mg per cent, 5.91 ± 0.47 Vs 5.90 ± 0.45 mg per cent and 10.40 ± 0.45 Vs 11.31 ± 0.59 mg per cent respectively and showed no significant difference. Similarly the levels of these parameters at first detected oestrus in these groups were 50.27 ± 1.98 Vs 48.06 ± 2.31 mg per cent, 9.64 ± 0.47 Vs 9.60 ± 0.34 per cent, 5.37 ± 0.38 Vs 5.51 ± 0.20 mg per cent and 12.10 ± 0.52 Vs 12.23 ± 0.36 mg per cent respectively and showed no significant difference. The serum total proteins also showed no significant difference between groups at first ovulation (6.74 ± 0.13 Vs 6.59 ± 0.07 gm per cent) and at detected oestrus (6.75 ± 0.12 Vs 6.65 ± 0.11 gm per cent). However, a significantly higher average level of total protein (6.49 Vs 6.79 gm per cent) prevailed in the treatment group.

* * *

The post-partum period is vulnerable to many disturbing influences like mal-nutrition and lactational stress which are reflected in

general metabolic activity and reproductive ability of an individual. Published investigations have linked specific nutritional deficiency to decreased reproductive efficiency based on their respective serum metabolic levels. In the recent approach to the management of sub-fertility in dairy animals, it is observed that GnRH administration resulted in preovulatory surge of LH, decreased the post-partum interval to first ovulation and established regular oestrus cycles. Therefore, the object of present investigation was to study the influence of administration of GnRH analogue on some blood constituents at post-partum reproductive events in crossbred cows.

Materials and Methods

In all 18 healthy crossbred cows free from parturient abnormalities were selected and divided into treatment group receiving 5 ml of GnRH analogue equivalent to 0.020 mg of Buserelin and control group receiving 5 ml of Normal Saline as placebo, on 14th day post-partum. Blood was collected on day 0, 7, 14, 21, 30, 45, 60, 75, 90 and 105 post-partum.

Blood glucose, serum calcium and inorganic phosphorus were estimated as per the methods previously adopted (Folin and Hu Wu, 1920; Gomorri, 1942; Trinder, 1960). Analytical studies in respect of serum cholesterol (Zak, 1954) Serum total proteins (Annino, 1976) and haemoglobin (Coes, 1967) were also carried out. As the blood collections were done at regular post-partum

intervals and did not necessarily coincide with the events like ovulation and oestrus, the level prevailing at the nearest regular interval was considered for comparison.

Results and Discussion

Serum cholesterol (SC): The SC level was lowest on the day of parturition in treatment (106.18 ± 5.37 mg per cent) and control (113.80 ± 12.66 mg per cent) group and gradually decreased upto day 45 and 30 in respective groups, thereafter showed a gradual decline upto 75th day and later resumed an increasing trend upto day 105 post-partum. These fluctuations were similar to earlier reports (Arave *et al.*, 1975; Jadhav *et al.*, 1977). The difference in SC level in two groups, however, was not significant. In the treatment group hyper-cholesteraemia was observed from day 45 to 75 which was the period indicating earlier resumption of the ovarian activity. It is evident that the function of the corpus luteum and conversion of cholesterol to progesterone was maximum at this stage resulting into increased turnover of cholesterol from plasma pool (Shima *et al.*, 1972).

The average SC levels at first post-partum ovulation and at detected oestrus were 143.50 ± 7.72 and 141.63 ± 7.64 mg per cent in treatment and 162.37 ± 14.39 and 173.20 ± 9.88 mg per cent in control group. The difference between the two groups at both the events was significant ($P < 0.01$). Thus the cows in treatment group showed the reproductive activity at a lower cholesterol level.

Blood Glucose (BG): The BG level showed fluctuations upto day 22 in control group but it was fairly constant in treatment group. It was slightly higher at first PP ovulation (48.57 ± 2.25 Vs 45.17 ± 2.93 mg per cent) in control

group and at detected oestrus (50.27 ± 1.98 Vs 48.06 ± 2.31 mg per cent) in treatment group. However, the difference in groups was not significant at both the events. The occurrence of ovulation in the treatment may be useful in inducing ovulatory activity in animals having comparatively lower energy intake.

Serum Calcium (SCa): In both group the SCa level was initially low but later gradually increased with slight fluctuations. In treatment group the level at first ovulation was relatively lower (9.19 ± 0.45 Vs 9.84 ± 0.57 mg per cent), whereas at detected oestrus it was almost similar in both groups (9.64 ± 0.47 Vs 9.60 ± 0.34 mg per cent) showing no influence of GnRH analogue on SCa level.

Serum Inorganic Phosphorus (SIP): The treatment group showed highest value (6.39 ± 0.44 mg per cent) on day 14 and the lowest value (4.88 ± 0.38 mg per cent) on day 105 post-partum. In this group the SIP level reduced after day 45, contrary to increase in control group after this period. The reduction in treatment group may be due to utilization of energy through the process of phosphorylation. There was no significant difference in SIP levels in both groups at first ovulation (5.91 ± 0.47 Vs 5.90 ± 0.45 mg per cent) and at detected oestrus (5.37 ± 0.38 Vs 5.51 ± 0.20 mg per cent).

Serum Total Proteins (STP): The STP showed increased values (6.49 ± 0.12 Vs 6.79 ± 0.14 gm per cent) due to treatment ($P < 0.01$). However, the effect due to treatment at first ovulation (6.74 ± 0.13 Vs 6.59 ± 0.07 gm per cent) and at detected oestrus (6.75 ± 0.12 Vs 6.56 ± 0.11 gm per cent) was not significant. Considering that the treatment helped in raising these levels, it can be concluded that even in animals having a lower protein intake GnRH treatment, probably helps in initiating

the reproductive activity during a post-partum period.

Haemoglobin (Hb): In both groups the lowest value of Hb was recorded on day 30 (10.62 ± 0.46 and 10.42 ± 0.52 gm per cent) followed by a trend of fluctuations. The Hb level at first ovulation (10.40 ± 0.54 Vs 11.31 ± 0.59 gm per cent) and at detected oestrus

(12.10 ± 0.52 Vs 12.23 ± 0.36 gm per cent) showed no significant difference in groups.

Thus the treatment of GnRH analogue did not significantly affect the blood biochemical constituents except for serum cholesterol and total proteins in post partum crossbred cows. The observations are suggestive of the trend and detailed investigation on this aspect.

REFERENCES

- Annino, J.S. (1976). Total proteins and albumin buret and dye binding methods. Clinical Chemistry, Principle and procedures, 4th Ed. Little, Browns and Company, Boston.
- Arave, C.W.; Miller, R.H. and Lamb, R.C. (1975). Genetic and environmental effect on serum cholesterol of dairy cattle of various ages. J. Dairy Sci. 58: 423-427.
- Coles, E.H. (1967). Veterinary clinical pathology W.B. Saunders Company, Philadelphia and London 11rd 107.
- Folin, O. and H. Wu. (1920). A simplified and improved method for determination of sugar. J. Biol. Chem. 41: 367-374.
- Gomorri G. (1942). Inorganic phosphorus, Gomorris method. J. Lab. Clin. Med. 27: 955.
- Jadhav, S.S.; Velhankar, D.P. and Deshpande, B.R. (1977). Variation in the serum cholesterol levels of Gir cows during pregnancy, parturition and puerperium. Indian Vet. J. 54(7): 536-540.
- Shima, C.B.; Bhattacharya, A.R.; Luktuke, S.N. and Pal, A.K. (1972). Studies on blood cholesterol in relation to oestrus cycle in bovines. F.A.O. Swedish followup seminar on Anim. Reprod. Bangalore.
- Trinder, P. (1960). Calcium, Trinder's method Analyst. 85: 880.
- Zak, B. (1954). Cholesterol, Ferric Chloride method Amer. J. Clin. Path. 24: 1307.

IJAR 9:2: 87-89:1988

Effects Of Various Concentrations Of Lugol's Iodine And Oxytetracycline On The Cytomorphology Of The Endometrium Of Repeat Breeding Cows

UMED SINGH, R.C. GUPTA, D.N. SHARMA¹ and A.K. SINHA²

Department of Veterinary Gynaecology & Obstetrics, College of Veterinary Sciences, Haryana Agricultural University, Hisar-125004

ABSTRACT

Uterine biopsy samples were collected from 30 repeat breeder cows just before infusion. 24 and 72 hours post infusions of 1:10 and 1:20 Lugol's solution, 1 g and 2 g

oxytetracycline, and normal saline solutions in 40 ml volumes, from each animal. The results revealed that the normal saline solution caused no specific changes in the endometrial histology, but all other

1. Associate Professor of Anatomy and Histology, H.A.U., Hisar.

2. Associate Professor of Gynaecology, B.A.U., Ranchi.

combinations incited acute inflammatory reactions. The changes were relatively milder, and more quickly reversible in case of 1:20 Lugol's solution, when compared to the other 3 combinations.

* * *

Lugol's solution and oxytetracycline are among the most widely used intrauterine infusions for the treatment of the repeat breeder cows. Absence of a comprehensive literature on the structural changes brought about by these drugs in the endometrium after infusion has prompted us to undertake this study.

Materials and Methods

Thirty repeat breeding cows (having failed to conceive even after at least 3 artificial inseminations) with normal estrous cycle and apparently normal genitalia were selected at random from the animal farm of the University. The animals were not treated with any intrauterine infusions during those cycles.

The animals were divided into six groups of five animals each. These animals received 40 ml of 1:10 Lugol's Solution (Stock Lugol's solution = 1:KI:Distilled water = 5:10:100) (Group A) ; 40 ml of 1:20 Lugol's solution (Group B) ; 1 G. oxytetracycline (Terramycin Liquid, each ml. containing 50 mg oxytetracycline; manufactured by Pfizer India Ltd., Bombay) (Group C) ; 2 G oxytetracycline (Group D) and normal saline solution (Group E) in a volume of 40 ml as intrauterine infusion. In group F no uterine infusions were given.

All the infusions were made during the standing estrus and no inseminations were done. From each of these animals uterine biopsies were obtained at the standing estrus before the infusion as per the procedure described by Sinha (1980). Subsequently the

biopsies were collected at 24 hours and 72 hours post infusion.

The biopsy samples were collected in chilled 10% neutral buffered formaline and preserved at 4°C in refrigerator. Tissues were prepared for paraffin microtomy and serial sections of 4-6 μ thickness were stained with hematoxylin and eosine, periodic acid schiff (PAS) and reticular stains (Luna, 1968).

Results and Discussion

The uterine mucus discharge at the standing estrus was clear and the endometrial biopsies were pink with little signs of haemorrhage. A fairly good amount of brownish discharge was observed till 72 hours post infusion of Lugol's solution and oxytetracycline. The uteri were comparatively thicker and harder than that of the standing estrus. The endometrial biopsies showed evidence of haemorrhage. The extent of hardness of uterus and the haemorrhage at the time of biopsies corresponded with the drug concentration in the infusion. The animal infused with normal saline solution showed no deviation from that of the non-infused (control) group. These changes might be due to endometrial irritation (Singh *et al.* 1972 and Seguin *et al.* 1974 a). In a few biopsy specimens at standing estrus the cytoplasm was vacuolated and the epithelium showed focal desquamation which was indicative of subclinical endometritis (Prabhakaran Nair and Raja, 1975, and Bhandari and Kaikini 1983).

The Intrauterine infusion with normal saline solution had no specific effect on the surface epithelium as reported earlier (Oxender and Seguin, 1976 and Seguin *et al.* 1974 b). But with the infusion of 1:10 Lugol's solution the surface epithelium was either completely denuded or necrosed, after 24

hours of infusion. Infusion of oxytetracycline at either doses (1 g and 2 g) caused denudation of surface epithelium and loss of cytoplasmic basophilia but much less than the strong Lugol's solution. Oxytetracycline at 2 g dosage was found to be more irritant than at 1 g dosage. At 72 hours post infusion the epithelial desquamation and loss of basophilia were increased marginally over that at 24 hours in all the treated groups (A-D). The Lugol's solution at 1:20 concentration revealed the least damaging effect on the epithelium among all the treatments. Necrosis and denudation of the surface epithelium at 24 hours post infusion with dilute iodine solution, oxytetracycline (Seguin *et al.*, 1974 a, b and Oxender and Seguin, 1976), 2% solution of nitrofurazone (Ginther and Meckley, 1972) and sodium carbonate (Singh *et al.*, 1972) have been reported.

The histology of normal saline solution infused biopsies corresponded to the control group at 24 hours post estrus. Infusion of the Lugol's solution as well as oxytetracycline caused severe, diffuse, subepithelial and periglandular haemorrhage and severe edema. Several subepithelial necrotic foci were surrounded by mononuclear and polymorphonuclear cells. The changes were more severe in the superficial zone than in the deeper zone. The intensity of irritant reaction, was much milder in 1:20 Lugol's solution infused biopsies when compared to 1 g and 2 g oxytetracycline and 1:10 Lugol's solution infused biopsies. After 72 hours of infusion, the subepithelial and periglandular haemorrhage and edema subsided in all the groups but the microscopic picture of the 1:20 Lugol's solution treated biopsy returned much closer to the normal histology as compared to other drug combinations.

REFERENCES

- Bhandari, R.M. and Kaikini, A.S. (1983). *Indian J. Anim. Sci.* **53**: 481-484.
- Ginther, O.J. and Meckley, P.E. (1972). *Vet. Med. Small Anim. Clin.* **67**: 751-754.
- Luna, L.G. (ed) (1968). *Manual of Histological Staining Methods of Armed Forces Institute of Pathology*. 3rd edn. McGraw Hill Book Co. New York. pp 38, 87, 113.
- Oxender, W.D. and Seguin, B.E. (1976). *J. Am. Vet. Med. Assoc.* **168**: 217-219.
- Prabhakaran Nair, K. and Raja, C.K.S.V. (1975). *Indian J. Anim. Sci.* **45**: 958-961.
- Seguin, D.E.; Morrow, D.A. and Louis, T.M. (1974 a). *Am. J. Vet. Res.* **35**: 57-61.
- Seguin, D.E.; Morrow, D.A. and Oxender, W.D. (1974 b). *J. Am. Vet. Med. Assoc.* **164**: 609-612.
- Singh, N.P.; Marion, G.B. and Gier, H.T. (1972). *Indian J. Anim. Sci.* **42**: 269-272.
- Sinha, A.K. (1980). Ph.D. Thesis, HAU, Hisar.

Studies On Factors Influencing Calving Interval In Nondescript Rural Cattle

S. KUMAR¹, S.K. AGARWAL², L.N. PURBEY³ and R. PRAKASH⁴

Indian Veterinary Research Institute, Izatnagar-243122

ABSTRACT

Calving interval of 99 non-descript rural cattle was studied in relation to season of conception, season of last calving, sex of calf, parity of animals and farmers status (land holding capacity). Season of conception and parity was found to have significant effect on intercalving period; whereas sex of calf, season of last calving and farmers status did not have any significant effect on intercalving period. The overall calving interval was found to be 574.25 ± 29.42 days (312-1010 days). The seasonwise distribution of calving was highest during winter followed by spring and wet summer seasons.

* * *

Infertility appears to be primarily a managemental problem in rural cattle as well as in a dairy herd. The present study deals with the effect of season of last calving, season of conception, sex of calf, parity and farmer status on calving interval.

Materials and Methods

The study was conducted on 99 cattle of non-descript origin at Operational Research Project of Indian Veterinary Research Institute at Rithoura Centre in Bareilly district of Uttar Pradesh. The details record viz., age, number of calving, month of last calving, month of conception, sex of calf horn, etc., of all the cases was maintained.

The animals were maintained by all the group of farmers ranging from large farmer to landless livestock owner under traditional system of animal husbandry practices under rural condition. The seasons were classified as winter, spring, dry summer, wet summer and autumn according to Ahuja (1958). The intercalving period was calculated and the data was analysed.

Results and Discussion

The calving interval in the non-descript cattle was found to be 574.25 ± 29.42 (312-1010) days. It was found that the season of conception had a significant effect on intercalving period (Table I). The intercalving period was significantly higher in the animals conceived during wet summer and autumn seasons. Season of last calving had no significant effect on intercalving period, but an extended intercalving period was observed in the animals calved during spring followed by winter season. Narayanakhedkar (1985) and Arora, *et al.* (1983) have reported that season of calving has no significant effect on the intercalving period in Holstein cattle. However, Dhillon, *et al.* (1970) and Mishra *et al.* (1977) are of opinion that season of calving had a significant effect on service period.

Parity level (number of calving) of the animals significantly affected the calving

1. Scientist S-1, Division of Extension

2. Scientist S-1, Division of L.P.R. (C&B)

3. Scientist S-3, Division of Animal Reproduction

4. Scientist S-1, Division of Livestock Economics and Statistics

interval. A decreasing trend in the length of calving interval was observed as the number of calvings increased. However, Sadana, *et al.* (1983) have reported that neither breed nor parity level had any significant effect on calving interval. No significant difference was observed on intercalving period due to sex of calf and also the status of farmers.

The seasonwise occurrence of calving in the present study was 34.12%, 22.06%, 9.13%, 21.41% and 13.10% respectively for winter, spring, dry summer, wet summer and autumn seasons. Maximum calving (34.12%) occurred during winter followed by spring (22.06%) and wet summer (21.41%). The findings of the present study reveals that more number of animals come in heat and conceive during spring and summer.

The extended intercalving period may be attributed mainly due to the improper nutrition, lack of management and also the negligence of livestock owners for proper sexual health care. As it was observed that the farmers status also did not have any significant effect on calving interval, it reflects that all the group of farmers irrespective of their land holding capacity rear their animals under traditional system of animal husbandry practices. Therefore, proper training and education to farmers for profitable animal husbandry practices is required.

Acknowledgement

The authors are thankful to Dr. P.N. Bhat, Director and Dr. O.N. Kunzru, Head, Division of Extension, Indian Veterinary Research Institute for providing facilities to conduct this study.

Table 1: Effect of season of conception and season of last Calving on intercalving period

Season	Intercalving Period (days)	
	According to season of conception	According to season of last calving
Winter	548.57 \pm 24.05 (361-661)	576.66 \pm 20.67 (393-822)
Spring	562.92 \pm 24.45 (345-828)	587.33 \pm 25.83 (320-824)
Dry Summer	528.87 \pm 27.38 (312-822)	559.56 \pm 45.44 (312-650)
Wet Summer	629.93 \pm 51.93** (366-1010)	571.90 \pm 42.37 (361-1010)
Autumn	662.36 \pm 65.47** (445-1009)	552.85 \pm 60.26 (366-1009)
Overall	574.25 \pm 29.42 (312-1010)	574.25 \pm 29.42 (312-1010)

**Significant at 1% level of significance (Figures under parenthesis indicate the range).

REFERENCES

- Ahuja, L.D. (1958). M.Sc. Ag. Thesis, Bombay University, Bombay.
Arora, D.N. and Sharma, J.G. (1983). Indian Vet. J. 60: 820-823.
Dhillon, J.S.; Acharya, R.M. and Tiwana, M.S. (1970). Anim. Prod. 12: 81.
Mishra, R.R.; Chauhan, R.S. and Bhatnagar, D.S. (1977). Indian J. Anim. Sci. 47: 418.
Narayankhedkar, S.C. (1973). M.V.Sc. Thesis Punjabrao Krishi Vidyapeeth, Akola-444104.
Sadana, D.K. and Basu, S.B. (1983). Indian J. Dairy Sci. 34(2): 121-124.

IJAR 9:2: 92-95:1988

Studies On Repeat Breeder Friesian Cows In Libya

K.M. BEN-HAJ, and S. ABDULMOLA

Faculty of Vet. Medicine, Alfateh University, P.O. Box 13662 Tripoli— Libya

ABSTRACT

Thirty-two swabs were taken from the external os uteri of sixty repeat breeding Friesian cows for microbiological examination. Antibiotic sensitivity test was carried out to select antibiotic of choice. Three types of micro-organism were isolated from all repeat breeder cows namely, *Proteus* spp., *E. coli* and *Anthraxoid*. In addition, *Staphylococcus aureus* and *Streptococcus bovis* were also isolated in cows with genital abnormality. Antibiotic sensitivity test showed that the isolated bacteria were sensitive to chloramphenicol, cephaloridine, gentamicin, colistin sulphate and neomycin, in order of sensitivity. Intrauterine therapy with antibiotic alone did not significantly improve the fertility rate, as only 12 out of 30 treated females became pregnant. This may be attributed to the specific pathological lesions of the genital tract. Seventy per cent of treated females (21 out of 30) conceived when treated with antibiotic followed by intrauterine infusion with Lugol's iodine in the next heat. No cow suffering from *Staphylococcus aureus* conceived.

Preliminary investigation for the incidence of reproductive disorders in Libya revealed that the phenomenon of repeat breeding accounts for over 67% of reproductive disorders (Baorgob *et al.*, 1982), although nutritional, management, hormonal imbalances and clinical or sub-clinical uterine infection are considered to be the major cause of infertility.

Concerning the microbiological causes of repeat breeding, there was general agreement that the bacteria were present in the genital tract (anterior vagina, cervix or in uterus) of normal cows and buffaloes (Gunter *et al.*, 1955; Gibbons *et al.*, 1959; Awad, 1972; Awad *et al.*, 1977). The bacteria in the normal female genitalia may become pathogenic and cause clinical or sub clinical signs of endometritis (Raghavan *et al.*, 1971 and Abo-El-Ata, 1973). This might occur when the resistance of the animal is lowered due to stress or any predisposing cause resulting in repeat breeding.

The aim of this investigation was to know the types of organisms prevalent and to select

the suitable treatment for repeat breeding cows in Libya.

Materials and Methods

The study was conducted on 60 repeat breeder cows that had at least three unsuccessful inseminations. These cows were randomly selected out of the herd of 660 Friesian cows of El-Gharian government farm, 25 km from Tripoli. Their age varied from 5-7 years. They were artificially inseminated. The farm was free from brucellosis and venereal diseases. According to rectal, vaginal, clinical examinations and the history, the animals were classified into two groups of 30 cows each. Group I included 30 cows which had normal oestrous cycles and apparent normal genitalia. Group II included 30 cows which had regular, near regular or irregular oestrous cycles and had palpable genital abnormality like cervicitis and/or endometritis.

A random 32 swabs (16 from each group) were taken near aseptically from the external os uteri and cultured on enriched media for the isolation and identification of the micro-organisms. Antibiotic sensitivity tests were carried out to select the drug of choice. Based on the antibiotic sensitivity test, two trials of treatment were instituted and all the animals included in this study were treated. In trial one, given to 30 cows (15 from each group randomly selected), 3 gm chloramphenicol in 30 ml of distilled water was infused intrauterine during oestrus, given twice at 24 hours interval. In trial two, rest 30 cows during oestrus (15 from each group), received interuterine infusion of 3 gm chloramphenicol in 30 ml distilled water followed by Lugol's solution (30 ml, 1/400) on the following day. The animals were not inseminated during the course of treatment.

In the next oestrus following treatment, the cows were inseminated with semen from known fertile bulls. Pregnancy was confirmed after two months in animals not reported in oestrus following insemination.

Results

Three types of micro-organisms were isolated from all samples of both the groups namely *Proteus* spp., *E. coli* and *Anthraxoids*. In addition, *Staphylococcus aureus* and *Streptococcus bovis* were isolated from 3 cases of group II (Table I).

All the isolated bacteria were found sensitive to chloramphenicol, gentamicin, colistin sulphate, neomycin in the order of sensitivity, while these were resistant to erythromycin, penicillin, streptomycin, tetracycline and linkamycin.

In trial one, cows receiving intrauterine infusion with chloramphenicol alone, failed to show significant improvement in conception rate in the cows of group II as only 20% (3 out of 15) cows became pregnant whereas 60% (9 out of 15) of cows from group I became pregnant, with an over all conception rate of 40% (12 out of 30).

In trial two, cows receiving Lugol's solution in addition to chloramphenicol intrauterine infusion, the over all conception rate was better as compared to trial one as 70% (21 out of 30) i.e. 12 out of 30 (80%) and 9 out of 30 animals (60%) of group I and group II conceived. However, the cows with *Staphylococcus aureus* and *Streptococcus bovis* infection did not conceive.

Discussion

Although the types of bacteria isolated (*Proteus* spp., *E. coli* and *Anthraxoids*) were those frequently reported as a cause of non-

specific genital infections, yet these organisms were usually saprophytic and occurred concurrently with other specific organisms. The isolation of *Staph. aureus* and *Strept. bovis* were, however, of significance as a cause of repeat breeding (Roberts, 1971; Laing, 1979 and El-Azab, 1980) which might render the genital tract unfavourable to sperm (Roberts, 1971; Awad, 1972; Abo-El-Ata, 1973; Awad *et al.*, 1977 and El-Azab, 1980) or affect implantation (Raghavan *et al.*, 1971). Kiesel and Daeres (1959) reported from a bacteriological study on the bovine genital tract that non-specific bacterial infection did not appear to have much significance as a cause of infertility. Also, several authors have reported that the uterus of repeat breeder cow might be free from micro-organisms (Murthy *et al.*, 1974 and Gupta *et al.*, 1983). On the other hand, Gunter *et al.* (1955) found bacterial growth in 95% of repeat breeders and 67% of regular breeder cows. The bacteria under unfavourable conditions, might become pathogenic showing clinical or sub-clinical signs of endometritis (Raghavan *et al.*, 1971; Abo-El-Ata, 1973 and Laing, 1979).

In the present study, *Proteus* spp., *E. coli*, *Staph. aureus* and *Strept. bovis* isolated from repeat breeding cows may be associated with infertility and initiating some pathogenic lesions. We believe that a variety of organisms might gain entrance into the uterus by contaminated instruments or catheters which may result in cervicitis and or clinical or sub-clinical endometritis thus leading to repeat breeding.

Bacteria isolated in the present study were highly sensitive to chloramphenicol. Similar

findings have also been reported by Awad and El-Hariri (1979) and El-Azab, (1980)

Intrauterine infusion with chloramphenicol alone showed overall lower response (40%) in the conception rate with better results (60%) in repeat breeders of group I having apparently normal genitalia. The poor results (20%) in group II animals with genital abnormality might be due to the fact that the infected uterus could not heal rapidly so as to enable normal fertility (Gupta *et al.*, 1983).

The response to the treatment of combination of chloramphenicol followed by Lugol's solution infusion was superior in both the groups (80% in group I and 60% in group II) with an overall conception rate of 70%. This may be due to the additional benefit of Lugol's solution which has an irritating effect on the uterus that leads to hyperaemia, increased mucus secretion and leucocytosis (Gupta *et al.*, 1983). Zemjanis (1980) also recommended the intrauterine infusion of Lugol's solution.

From the present study, it can be concluded that the additional infusion with Lugol's solution may cause increased leucocytosis, rapid healing of uterine mucosa thus leading to better conception rate. However, further work is needed to establish the uterine flora in apparently normal and endometritis cases and to establish anti-infectious intrauterine infusions in repeat breeder cows based on microbial isolation, drug sensitivity results and endometrial biopsy for the evaluation of chances of conception at the time of service.

REFERENCES

- Abo-El-Ata, M.M. (1973). Studies on chronic endometritis in cows and buffaloes in Egypt. M.D. Vet. Thesis, Cairo University.
- Awad, H.H. (1972). Studies on Puerperal infection in cattle and buffaloes. M.D. Vet. Thesis, Cairo University.
- Awad, H.H. and El Hariri, M.N.E. (1979). The bacteriology of the cervical mucus during estrus in repeat breeder cows and buffaloes and its sensitivity to different antibiotics in vitro. *Zagzig Vet. J.* 3.
- Awad, H.H.; Afiefy, M.M. and Ayoub, M.M. (1977). Genital discharge in an infertile Friesian herd. Their causes and treatment. *Assiut Vet. Med. J.* 4(8): 219-229.
- Boargoub, A.; Shalaby, A.S. and Ben-Haj (1982). Incidence of infertility in Libyan dairy farm. Under publication.
- El-Azab, M. (1980). Some studies on the repeat breeder cows and buffaloes M.D. Vet. Thesis, Zagazig University.
- El-Sawaf, S.A.; Farag, H. and Sameh, M. (1960). Bacteriological Survey of the flora of the buffalo uterus. *Vet. Med. J. Giza.* 7: 281-288.
- Gibbons, W.J.; Attleberger, M.H.; Kiesel, G.K. and Daores, W.H. (1959). The bacteriology of the cervical mucus in cattle. *Cornell Vet.* 49: 255.
- Gunter, J.J., Collins, W.J.; Owen, J.; Soreason, A.M.; Scales, J.W. and Alford, J.A. (1955). A survey of the bacteria in the reproductive tract of dairy animals and their relationship in infertility. *J. Vet. Research* 12: 282.
- Gupta, R.C.; Sinha, A.K. and Swamy, A.K. (1983). Studies on the efficacy of some post-service intrauterine infusions on the conception rate of repeat breeding cattle. *Theriogenology*, 20: 559-564.
- Kiesel, G.K. and Dacre, W.G. (1959). A study of infertility in cattle in Alabama. *Am. J. Vet. Res.* 20: 760.
- Laing, J.A. (1979). Fertility and infertility in the domestic animals. 3rd Edn. Baillier Tindall— Cassel, London.
- Luktuke, S.N. and Joshi, S.R. (1961). Intrauterine treatment of repeat breeders with antibiotics. *Ind. J. Vet. Sci.* 31: 10.
- Murthy, G.V.K.; Hanjiah, R.D. and Keshava Murthy, B.S. (1974). Bacterial flora of cervical mucus in repeat breeding bovines. *Indian Vet. J.* 51: 264-268.
- Raghavan, R.; Nilakantan, P.R. and Uppal, P.K. (1971). Studies on the bacteriology of the bovine genital tract. *Indian Vet. J.* 48: 779-783.
- Roberts, S.J. (1971). Veterinary obstetrics and genital diseases. 2nd Edn. Published by the author— Ithaca, New York.
- Zemjanis, R. (1980). Repeat breeding or conception failure in cattle. In: *Current Therapy in Theriogenology*. Ed. Morrow. A.D. W.B. Saunders Co., Philadelphia. p. 210.

Table I: Micro-organisms isolated from repeat breeder cows

Sr. No.	Micro-organisms	Group I n=16		Group II n=16	
		No.	%	No.	%
1.	<i>Proteus sp.</i>	6	37.5	6	37.5
2.	<i>E. coli</i>	6	37.5	6	37.5
3.	<i>Anthracoïds</i>	4	25.0	2	12.5
4.	<i>Staphylococcus aureus</i>	—	—	3	18.7
5.	<i>Streptococcus hōvis</i>	—	—	3	18.7

Blood-Serum Prolactin Levels In Red Dane, Swedish Red And Jersey Crossbreds During Various Phases Of Reproduction

C.V. DINDORKAR¹, A.S. KAIKIN², and A.R. SHETH³

Post-Graduate Institute, Punjabrao Krishi Vidyapeeth, Akola-444104.

Prolactin, also known as mammothrophin, plays a major role in regulating the growth and function of udder. It is now accepted as a hormone distinct from the growth hormone. Systematic information regarding the circulating levels of prolactin hormone in cross-breds during various phases of reproduction is not available. This study was therefore undertaken on 27 crossbred females, divided in 3 major groups according to the exotic inheritance viz., Red Dane (3), Swedish Red and White SRB (5) and Jersey (19). Jersey crossbreds were again sub-divided as per their genotypes into 3 sub-groups: Gir × Jersey (5), Gaolao × Jersey (6) and non-descript × Jersey (8) for studying the blood serum prolactin levels. All the crossbreds were of breedable age, regularly cycling and subjected to periodical gynaeco-clinical examination.

Blood serum samples were collected during pro-oestrus, oestrus, met-oestrus, dioestrus; early, middle and advanced pregnancy and post-partum (5 days after calving). Prolactin was estimated by radio-immuno-assay (RIA) technique as per

Greenwood *et al* (1963) and Midgley (1966), with slight modifications.

The average mean blood-serum prolactin values recorded are presented in Table-I.

Prolactin levels in advanced stage of pregnancy and post-partum stage could not be studied in the non-descript × Jersey crossbreds, since these were disposed off to farmers.

Our findings are in agreement with those of Gonzale *et al.* (1975) and Hale (1975), but are considerably higher in Red Dane and Swedish Red crossbred cows than in non-descript local cows (189.67 to 358.5 ng/ml) recorded by Pargaonkar (1978).

The variation in the prolactin levels during various phases of reproduction as well as between breeds studied, was non-significant.

Acknowledgement

Thanks are due to the Indian Council of Agricultural Research (ICAR), New Delhi for the funds provided under Ad-Hoc Scheme (1976-78) to the Principal Investigator Dr. A.S. Kaikini, Dean, Faculty of Veterinary Science, PKV, Akola.

1. Reader in Gynaecology, Nagpur Veterinary College, Nagpur-440006.

2. Emeritus Scientist (ICAR) PKV, Nagpur Veterinary College, Nagpur-440006.

3. Director, Institute for Research in Human Reproduction, (ICMR), Jehangir Merwanji Street, Parel, Bombay-400012.

Table 1: Mean blood serum prolactin values (ng/ml) in F1 cross-bred females of varying exotic inheritance during various phases of reproduction

Phase of reproduction	Red Dane (ng/ml)	Swedish Red (ng/ml)	Jersey (ng/ml)		
			Gir × Jersey	Gaolao × Jersey	Nondescript × Jersey
1. Pro-oestrus	222.66 ± 25.71	338.00 ± 124.98	285.16 ± 126.30	197.83 ± 111.68	211.28 ± 77.43
2. Ostrus	237.33 ± 61.45	364.20 ± 137.34	282.50 ± 137.34	335.16 ± 286.87	257.28 ± 77.43
3. Metoestrus	189.33 ± 65.73	304.00 ± 78.74	330.00 ± 106.33	147.00 ± 75.60	132.60 ± 72.45
4. Dioestrus	588.66 ± 362.46	359.20 ± 206.95	256.60 ± 217.90	168.80 ± 117.76	282.57 ± 215.73
5. Early pregnancy	225.00 ± 122.78	232.80 ± 41.82	219.50 ± 75.49	197.00 ± 86.76	173.57 ± 47.33
6. Middle pregnancy	209.33 ± 69.86	315.40 ± 43.46	310.00 ± 180.18	218.50 ± 126.92	230.42 ± 107.89
7. Advanced pregnancy	204.00 ± 33.94	534.00 - 500.63	132.33 ± 42.02	192.40 ± 131.51	
8. Post-partum	201.50 ± 4.94	280.00 ± 147.07	220.00 ± 110.64	202.40 ± 123.05	
9. Over-all average range	189.33 to 588.66	232.80 to 534.00	132.33 to 330.00	147.00 to 218.50	132.60 to 282.97

REFERENCES

- Gonzales, P.E.; Wiltbank, J.N. and Niswender, G.D. (1975). Puberty in beef heifers-I. The inter-relationship between pituitary, hypothalamic and ovarian hormones. *Veterinary Bulletin* 45(2): 856.
- Greenwood, F.C.; Hunter, W.M. and Glover, C.J.S. (1963). The preparation of I 125 labelled human growth hormone of high specific activity. *Biochem. J.* 89: 114.
- Hale, D.H. (1975). Studies on levels of luteinising hormone and prolactin in serum of cycling and anoestrus cow. *Veterinary Bulletin* 45(4): 296.
- Midgley, A.R. Jr. (1966). c.f. Pargaonkar, D.R. (1978).
- Pargaonkar, D.R. (1978). Studies on hormonal levels during various phases of reproduction in non-descript local cows — Ph.D. Thesis, Punjabrao Krishi Vidyapeeth, Akola-444104.

IJAR 9:2:98-100:1988

Pattern Of Conception And Calving In A Crossbred Herd

J.M. PATEL, S.V. SHAH, M.R. DHANGAR and M.M. TRIVEDI¹

Livestock Research Station, Gujarat Agricultural University Campus, Anand-388001.

The calving pattern has tremendous influence on efficiency and economy of milk production. For uniform milk production in the herd, the calving rate has to be maintained in a definite order. The present study was undertaken to study the pattern of conception and calving rate in Kankrej and its Jersey crosses.

The data on conception and calving were collected for the period 1981 to 1987 from the records of Livestock Research Station, Anand. The location of the station and managerial practices were as per the description given by Patel *et al.* (1987). The heat detection was carried out twice in a day. Inseminations were performed with frozen semen of crossbred and purebred exotic bulls. The animals were inseminated at random, irrespective of season or month as and when they came in heat. Analysis of data was done

after transforming them into arc sine percentage.

In Kankrej cattle, the maximum calvings were in the month of April (13.02%) followed by June (12.55%) and December (10.69%). The least calvings were in the month of July (4.36%) and August (5.56%). Similar trend has also been reported by Verma *et al.* (1983) in Haryana cattle. Distribution of calvings during different years also revealed similar trend.

In Jersey × Kankrej crossbreds, the calving pattern differed between different months. Pooled observations showed that the distribution of calving was quite uniform from November to January and February to May. Effects of months on calving pattern was nonsignificant. However, the peak in calving was noticed in October (13.87%) and September (11.29%). July and August were

the least calving months. Other workers (Verma *et al.*, 1983 and Pandey *et al.*, 1988) also observed less frequency of calvings in these months.

The number of breedable heifers added during April-May and December-January were maximum and it was the principal factor to change the distribution of calving among different years. The results of calving rate suggested that in Jersey-Kankrej crossbreds, December and January were the most favourable months for maximum conception. However the first A.I. conception rate indicated high conception rate during

November and April-May (Table 1). This shows that in Jersey crossbreds, reproduction is not affected adversely by high ambient temperature of summer, as expected. This may indicate that available shelter on the farm might be adequate enough to protect the animals against summer stress.

The results revealed that in Jersey × Kankrej crossbreds, the uniformity of calving is superior to that of Kankrej. Hence, for uniform and economical milk production, Jersey × Kankrej crossbreds will be highly suitable for the existing ecosystem of central Gujarat.

TABLE 1: Monthwise calving rate (%) and first insemination conception response in Kankrej and Jersey x Kankrej crossbred cows

	Calving rate (%)		First A.I. conception rate (%)	
	Kankrej	Jersey— Kankrej crossbreds	Kankrej	Jersey— Kankrej crossbreds
January	11.11	7.10	22.22	39.34
February	5.95	7.74	14.29	54.17
March	7.94	9.03	26.10	40.00
April	13.49	8.07	33.33	66.67
May	8.73	9.36	25.00	63.16
June	11.11	7.10	37.93	40.63
July	4.96	6.77	18.47	38.10
August	5.56	5.48	23.81	44.83
September	7.94	11.29	27.59	59.63
October	6.95	13.87	16.67	59.26
November	7.14	6.77	25.00	77.78
December	9.92	7.42	19.05	57.89
Total No. observations	252.00	310.00	125.00	189.00

$$\text{First A.I. conception Rate (\%)} = \frac{\text{No. animals pregnant with first A.I.}}{\text{Total No. of animals confirmed pregnant}}$$

REFERENCES

- Pandey, H.N.; Saxena, M.M.; Nivsarkar, A.E. and Nautiyal, L.P. (1988). Calving pattern and herd fertility in crossbred and Holstein Friesian cows. *Indian J. Anim. Prod. Mgmt.* 4(1): 5-8.
- Patel, J.M.; Patel, A.M.; Mansuri, M.N.; Patel, K.S. and Dave, A.D. (1987). Performance of Jersey x Kankrej F1 and Holstein x Kankrej F1 crossbred. *Indian J. Anim. Sci.* 51(8): 886-888.
- Verma, R.P.; Saxena, S.K. and Mishra, R.R. (1983). Effect of tropical climate on calving pattern in cattle. *Indian Vet. J.* 60: 107-110.

IJAR 9:2:100-103 : 1988

Incidence Of Certain Types Of Oestrus And Pathological Termination Of Pregnancy

S.N. LUKTUKEL¹, and L.N. PURKEY

Indian Veterinary Research Institute, Izatnagar - 243122 (U.P.)

ABSTRACT

Data pertaining to oestruses in heifers, cows and buffalo cows and normal and pathological termination of pregnancies in Haryana and Murrah females were analysed. It was found that 90.41% heats in heifers, 89.41% in cows and 90.70% heats in buffalo cows were fit for insemination out of 274, 1297 and 334 heats detected in the three respective categories. Similarly, 91.80%, 84.95% and 83.35% pregnancies terminated normally in heifers, cows and buffalo cows respectively.

In order to achieve economic production, it is necessary to increase the rate of efficiency of reproduction in farm animals. As such, the organised and rural herds must be under intensive sexual health control. Oestrus is the most vulnerable phase of reproduction, since according to the natural phenomena the sexual acceptability in livestock occurs during

this phase only and therefore appropriate time of service is important in relation to conception. In artificial breeding programme, it has been observed that every oestrus is not fit for service. Similarly, it has also been noted that every pregnancy may not terminate into normal parturition. In the present studies, the types of oestrus and pregnancy losses in bovines under organised farm conditions are reported.

Materials and Methods

The experimental Haryana and Murrah herds of IVRI, Izatnagar (U.P.), were under regular sexual health control and data for five years (1961-65) have been analysed to determine the types of oestrus and the rate of pregnancy losses. Oestrus was detected with the help of sexually active vasectomised bulls. Animals in heat were clinically examined for confirmation of oestrus and diagnosing gestational oestrus, if any. The quality of

1. Present address: 11, Paritosh Society, Ajwa Road, Baroda-390019.

REFERENCES

- Pandey, H.N.; Saxena, M.M.; Nivsarkar, A.E. and Nautiyal, L.P. (1988). Calving pattern and herd fertility in crossbred and Holstein Friesian cows. *Indian J. Anim. Prod. Mgmt.* 4(1): 5-8.
- Patel, J.M.; Patel, A.M.; Mansuri, M.N.; Patel, K.S. and Dave, A.D. (1987). Performance of Jersey x Kankrej F1 and Holstein x Kankrej F1 crossbred. *Indian J. Anim. Sci.* 51(8): 886-888.
- Verma, R.P.; Saxena, S.K. and Mishra, R.R. (1983). Effect of tropical climate on calving pattern in cattle. *Indian Vet. J.* 60: 107-110.

IJAR 9:2:100-103 : 1988

Incidence Of Certain Types Of Oestrus And Pathological Termination Of Pregnancy

S.N. LUKTUKE¹, and L.N. PURHEY

Indian Veterinary Research Institute, Izatnagar - 243122 (U.P.)

ABSTRACT

Data pertaining to oestruses in heifers, cows and buffalo cows and normal and pathological termination of pregnancies in Haryana and Murrah females were analysed. It was found that 90.41% heats in heifers, 89.41% in cows and 90.70% heats in buffalo cows were fit for insemination out of 274, 1297 and 334 heats detected in the three respective categories. Similarly, 91.80%, 84.95% and 83.35% pregnancies terminated normally in heifers, cows and buffalo cows respectively.

* * *

In order to achieve economic production, it is necessary to increase the rate of efficiency of reproduction in farm animals. As such, the organised and rural herds must be under intensive sexual health control. Oestrus is the most vulnerable phase of reproduction, since according to the natural phenomena the sexual acceptability in livestock occurs during

this phase only and therefore appropriate time of service is important in relation to conception. In artificial breeding programme, it has been observed that every oestrus is not fit for service. Similarly, it has also been noted that every pregnancy may not terminate into normal parturition. In the present studies, the types of oestrus and pregnancy losses in bovines under organised farm conditions are reported.

Materials and Methods

The experimental Haryana and Murrah herds of IVRI, Izatnagar (U.P.), were under regular sexual health control and data for five years (1961-65) have been analysed to determine the types of oestrus and the rate of pregnancy losses. Oestrus was detected with the help of sexually active vasectomised bulls. Animals in heat were clinically examined for confirmation of oestrus and diagnosing gestational oestrus, if any. The quality of

1. Present address: 11, Paritosh Society, Ajwa Road, Baroda-390019.

mucus discharge whether normal or pathological was also noted. Two months following parturition the cows were regularly investigated gynaecologically for the presence of active corpus luteum in all those cases which did not show oestrus and such corpus luteum was enucleated to induce oestrus. This was also done in heifers and buffalo heifers which had crossed their maturity age.

Pregnancy was clinically diagnosed as early as 42 days after service (around two oestrus cycle period) by rectal palpation of genitalia and were regularly examined further at least three times during the gestation period, at an interval of 2-3 months to determine the pregnancy losses, if any.

Results

The total number of oestrus observed were 274, 1297 and 334 in heifers, cows and buffalo cows respectively. Of these, 90.41%, 89.41% and 90.70% cases respectively were considered fit for artificial insemination (Table 1). Incidence of gestational oestrus was found to be 3.23, 6.32 and 5.78% in the three groups respectively. Active corpus luteum was detected in 3.65, 7.78 and 10.77% in heifers, cows and buffalo cows respectively and the oestrus was usually induced in 3-5 days. Early post-partum oestrus occurring within 35 days following parturition was detected in 4.54% cows and 1.50% buffaloes. Vaginal mucus discharge was pathological in 5.82% heats in heifers, 7.32% in cows and 2.02% in buffalo cows (Table 1).

Sixty one pregnancies in heifers, 359 in cows and 90 in buffalo cows were followed for their termination for a period of four years. Normal calvings occurred in 90.80%, 84.95% and 83.35% heifers, cows and buffalo cows respectively. In the remaining cases, the

incidences of abortion, stillbirth, early embryonal mortality and dystokia were recorded (Table 2). In addition, 1.95% cows died during pregnancy and 1.64%, 2.22% and 1.11% heifers, cows and buffalo cows respectively were disposed off during gestation. Rate of abortion was 1.64%, 2.54% and 4.44% respectively in the three types of animals. Occurrence of early embryonic mortality was 4.92%, 7.23% and 5.55% in the three respective categories (Table 2).

Discussion

From the present studies, it is observed that every heat manifested by the female is not fit for service. Service may have to be rendered in about 90% heats in cattle and buffaloes. Incidence of gestational oestrus was found to be higher in cows and buffaloes than in heifers. 3 to 6% oestruses were gestational and therefore, care has to be taken to diagnose pregnancy heat before artificial insemination is carried out. This fact has earlier been reported by Luktuke (1982).

Presence of corpus luteum in ovary was found in 7.78% cows and 10.77% buffaloes which did not manifest any heat symptoms after calving. This indicates the occurrence of weak and silent oestrus in both the species. Buffaloes particularly show weak intensity of oestrus and higher incidence of 'quiet ovulation' in comparison to cows. Mature heifers also show this phenomena. The incidence in the present study has been found to be 3.65% in heifers as indicated by the presence of active corpus luteum. Oestrus was induced in these animals by enucleating corpus luteum and the heat usually followed 3-5 days. At times there are complications of this treatment if the corpus luteum is not carefully removed from the ovary (Luktuke *et*

al., 1961). Treatment with Prostaglandins may be helpful in inducing oestrus and avoid complication in such cases. In *Bos taurus* females, incidences of death of the animals have also been reported following the enucleation of corpus luteum due to continuous bleeding from the ovary (Roberts, 1971).

Occurrence of early post-partum oestrus within 35 days was negligible (1.50%) in buffalo cows. It was 4.54% in cows. Insemination at this time would depend on completion of uterine involution, general health and the follicular development in the ovary leading to ovulation. Insemination however is generally advised in breeding practice after 45 days following parturition, for successful conception.

Incidence of pathological mucus discharge from genital tract during oestrus was high in cows (7.32%) as compared to buffalo cows (2.02%). Infections do ascend the genital tract at or soon after parturition, causing metritis. No inseminations are advised at such heats as pregnancy may not result. Intrauterine treatment with suitable antibiotics,

particularly based on the bacterial sensitivity tests, are very useful in such cases. Vasectomised bulls used for detection of heats may serve the female in heat and spread the infection.

As regards termination of pregnancies, it has been found that 83% to 91% pregnancies resulted in normal parturition. Repeated clinical examination during early gestation period helped in diagnosing embryonal losses and the incidence was found to be rather high (7.23%) in cows as compared to other two categories of animals (5%). Incidence of abortion was somewhat high (4.44%) in buffalo cows. All pregnancies diagnosed particularly at earlier stage thus do not result in normal parturition and this fact the breeder must note with a view to concentrate more on prenatal care and management of pregnant animals. Importance of sexual health control programme is stressed for improving breeding efficiency.

Acknowledgement

The authors are grateful to the Director IVRI, and the Head, Division of Animal Genetics, IVRI for the facilities provided for this work.

REFERENCES

- Luktuke, S.N. (1982). Fertility problems in bovines. *Advances in Research in Animal Production*. I.C.A.R. publication, Chapter III.
- Luktuke, S.N. and Joshi, S.R. (1961). Intra-uterine treatment of repeat breeders with antibiotics. *Indian J. Vet. Sci.* **34**(4): 10.
- Luktuke, S.N.; Joshi, S.R. and Roy, D.J. (1961). Induction of oestrus by enucleation of corpus luteum in bovines and studies on the fertility rate at induced and subsequent oestrus. *Indian J. Physiol. & Allied Sci.* **25**(4): 91.
- Roberts, S.J. (1971). *Veterinary Obstetrics and Genital Diseases (Theriogenology)*. Pub. Ann Arbor, Ithaca, USA.

TABLE 1: Incidences of different types of oestruses (%)

Item	Heifers	Cows	Buff. Cows
Total Oestruses	274	1297	334
Oestruses fit for Insemination	90.41	89.41	90.70
Gestational Oestrus	3.23	6.32	5.98
Early post-partum oestrus	—	4.54	1.50
Silent oestrus (in which heat induced)	3.65	7.78	10.77
Oestrus with pathological mucus discharge	5.82	7.32	2.02

Table 2: Incidence of normal and pathological termination of pregnancies (%)

Termination of pregnancies	Heifers	Cows	Buff. cows
Total pregnancies diagnosed	61	359	90
Normal calvings	91.80	84.95	83.35
Abortions	1.64	2.54	4.44
Stillbirths	—	—	1.11
Early embryonal mortality	4.92	7.23	5.55
Dystokia	—	1.95	4.44
Sold during gestation	1.64	2.22	1.11

Leptospiral Abortion In Cattle And Buffaloes

R.C. RAJASUNDARAM¹ and S. NEDUNCHELLIAN²

Scheme for Control of Abortion in Cattle, Erode-638001 Tamilnadu State.

Leptospirosis has been recognised as an important cause of abortion in cattle by many workers (Adinarayanan *et al.*, 1960; Sane and Deshpande, 1965; Somasundra Rao and Surendran, 1970; De *et al.*, 1983). The prevalence of leptospiral antibodies in aborted and repeat breeding cattle was reported by Venugopal *et al.* (1986).

The present work was undertaken to investigate the association of Leptospirosis in abortion amongst cattle and buffalo in Periar Dist. of Tamil Nadu.

During the year 1987-88 a total of 26 abortions (17 buffaloes and 9 cows) were examined. Of these, 4 abortions (2 buffaloes, 2 cows) were considered positive for Leptospirosis. Aborted foetuses and placenta were collected and examined. Piece of liver, spleen, kidney from aborted foetuses were collected in 10% formal saline. Sections were cut and stained with haemotoxylin and eosin and Levadit's stain. Blood serum from cows and buffaloes were collected for serological testing. Standard tube agglutination test for brucellosis and microscopic agglutination test (MAT) for leptospirosis were employed.

The abortion in buffaloes, occurred at 6-8 months of pregnancy and in cows between 3-5 months. All the aborted animals had retained their placenta. Gross lesions revealed necrotic placentitis. The characteristic feature of the aborted foetus was an oedematous umbilical cord which characteristically severed close to

the abdominal wall. The foetus showed evidence of jaundice with yellowish discolouration of the visible mucus membrane. Liver was pale and enlarged. The abdominal cavity contained yellowish ascitic fluid.

Histopathological examination of the tissues of both the foetuses revealed similar changes. The liver showed focal areas of necrosis around the central vein. Spleen and kidneys showed degenerative changes. Kidney sections stained by Levadit's method showed spiral bodies resembling *Leptospira* organisms.

Sera samples from both cows and buffaloes proved negative for brucellosis but gave positive reaction to Leptospirosis. The titre ranged from 1:320 to 1:640.

Abortions in cattle and buffaloes of organised farms are not infrequent. Majority of such cases often proved to be due to brucellosis. However, information on the association of *Leptospira* in abortions is scanty. In the present report, specific clinical diagnosis was established on the basis of individual serological findings, characteristic post mortem lesions and histopathological changes in the organs of the aborted foetuses. Of late, leptospiral abortions among cattle and buffalo have gained greater importance in India. Hence, a careful systematic study is necessary to determine the magnitude of leptospiral abortions in cattle and buffaloes.

1. Assistant Professor

2. Associate Professor and Head

REFERENCES

- Adinarayanan, N.; Jain, N.C.; Chandramani, N.K. and Hajela, S.K. (1960). Studies on Leptospirosis among Bovines in India. *Indian Vet. J.* 37:251-254.
- De, B.N.; Chatterjee, A.; Bidyanta, J.; Deb, S.K. and Sen, G.P. (1983). Studies on Leptospiral abortion in cattle. *Indian J. Anim. Health* 22(2): 143-145.
- Sane, C.R. and Deshpande, B.R. (1964). Record of detection of *Leptospira pomona* infection as the cause of Abortion in cows. *Indian Vet. J.* 42: 75-78.
- Somasundara Rao, G. and Surendran, N.S. (1970). Incidence and serotypes of bovine *Leptospira* in Andhra Pradesh. *Indian Vet. J.* 47: 296-298.
- Venugopal, K; Ratnam, S.; Raghavan, N. and Nachimuthu, K. (1986). Prevalance of leptospiral antibodies in aborted and repeat breeding cattle *Cheiron* 5(3): 80-82.

IJAR 9:2:105-109 : 1988

Bacteriological Studies On The Cervical Mucus Of Repeat Breeding Cross-Bred Cattle, Their Treatment And Conception Rate

R.N. SHARMA, B.K. SINGH and M.P. SINHA

Department of Gynaecology and Obstetrics, Ranchi Veterinary College, Birsa Agricultural University,
Ranchi — 834007, Bihar.

ABSTRACT

Cervical mucus samples from repeat breeding cows and 8 heifers 35 were subjected to bacteriological tests and various micro-organisms were isolated. The isolates were tested for drug sensitivity pattern using separate pasteur 'Bio-discs' for Gram Positive and Gram negative organisms. After bacteriological examination, the repeat breeder animals were treated with suitable drugs selected on the basis of sensitivity tests. After the completion of the schedule of treatment the animals were inseminated during the next heat period. Pregnancy diagnosis was done by rectal palpation 60 days after insemination. The overall conception rate in the treatment group was 77.14% as against 25.0% in the control.

Infertility in dairy cattle may result from infections, affecting various parts of the genital organs. Generally, nonspecific infection of the genitalia is considered to be the main cause of repeat breeding (Krishnamurthy *et al.*, 1974). Where there is an increase in the number of micro-organisms and/or in their virulence, cervicitis or endometritis of various degrees may result and may lead to embryonic death and repeat breeding problem (Easley *et al.*, 1951). Since there is very little information in respect of repeat breeding problem in cross-bred animals, this study was undertaken to isolate and identify various bacterial agents associated with repeat breeding, to conduct antibiogram and carryout treatment of affected animals with suitable drugs. An attempt was also made to determine the efficacy of treatment on the basis of conception rate.

*

*

*

Materials and Methods

The study was conducted on 43 repeat breeding crossbred cattle (35 cows and 8 heifers). The experimental animals were crosses of Holstein friesian \times local and Jersey \times local. From these animals cervical mucus was collected during the estrous period, with the help of a suitably designed thick glass pipette. Strict aseptic precautions were maintained during the experiment. After aspiration, the cervical mucus was inoculated into nutrient broth and incubated at 37°C for 24 hours, later streaked on nutrient agar and incubated at 37°C for 24 hours. Identification of different isolates was done on the basis of primary and secondary tests advocated by Cowan and Steel (1977).

Organisms identified were tested for drug sensitivity pattern by paper disc diffusion method (Bauer *et al.*, 1966) using standardised separate 'Bio-disc' for gram positive and gram negative bacteria.

The repeat breeding animals were treated with most suitable drugs selected on the basis of *in vitro* sensitivity tests. The experimental animals were divided in 5 treatment groups and one group was kept as control. In treatment group I which consisted of 10 animals, Chloramphenicol 1.25 gm (Tifocin injectable) was administered intrauterine (I/U) on alternate days for 5 times. Eight animals were included in group II where Kanamycin 0.5 gm (K-mycin water soluble powder) was administered I/U on alternate days for 5 times. The third group consisted of 7 animals where Sulfamoxazole 800 mg and Trimethoprim 160 mg contained in two Supristol tablets were given I/U for 5 times. Four animals in fourth group received Sulfamoxazole (800 mg) and Trimethoprim (160 mg) plus Chloramphenicol 1.25 gm I/U

for 5 times on alternate days. Group V consisted of 6 animals which were treated with 20 gms of Hostacycline water soluble powder I.U 5 times on alternate days. Since, specific information was not available about the schedule of treatment in respect of the medicines tried, the conventional procedure of alternate day I/U treatment was followed. The control group had 8 animals which received no treatment.

The cervical mucus from all the experimental animals were again collected at the time of subsequent heat after the completion of the schedule of treatment and cultured for bacteriological examination. During the same heat period the animal was inseminated. Pregnancy diagnosis was done by rectal palpation 60 days post insemination and the conception rate was calculated.

Results and Discussions

Altogether 48 isolates of different Bacteria were recovered from 43 mucus samples (Table I). The results of present study were by and large, in agreement with Roberts (1971), Mutiga (1978), Sharma *et al.* (1978) and Kodagali *et al.* (1980) who also isolated similar type of organisms from cervical mucus of repeat breeding cattle. In the case of cows, the occurrence of *Esch. Coli* and *Staph. aureus* was higher as compared to other organisms. In heifers, the occurrence of *Esch. Coli* was still higher. This finding is in agreement with Krishnamurthy *et al.* (1974) who also observed that repeat breeding problem in cows was mostly due to infection of uterus by nonspecific bacteria. *Klebsiella pneumoniae* has been rarely reported from the cases of repeat breeding cattle (Krishnamurthy *et al.*, 1974 and Sharma *et al.*, 1978) but they were isolated from 3 samples during this study.

The sensitivity patterns of pathogens revealed that none of the drugs tried was effective against all the isolates. However, most of the organisms were sensitive to Cotrimoxazole, Chloramphenicol and Kanamycin. The sensitivity patterns also revealed that none of the Gram positive organisms were sensitive to Penicillin and Ampicillin and that many of the strains of bacteria were resistant to higher antibiotics.

Bacteriological examinations of all the cervical mucus samples collected from repeat breeders after completion of treatment was done and it was seen that all the samples were completely free from any type of growth.

Conception rate was maximum (87.5%) in group II where kanamycin was used and

minimum (66.66%) in group V where hostacycline was administered. In the control group the conception rate was 25.0% (Table 2). Normal deviate test revealed significant difference in conception rates between group I and II and the control group. Overall conception rate was found to be 77.14% in the treatment group and 25.0% in the control group indicating that the effect of treatment was highly significant, more or less similar to the results of Khan and Luktuke (1967) and Sharma *et al.* (1978).

Acknowledgement

We thank Dr. H.R. Mishra, Dean of the College and Dr. T.S. Sharma, Professor of Microbiology for their help during the period of study.

REFERENCES

- Cowan, S.T. and Steel, K.J. (1977). Manual for the identification of Medical bacteria . 2nd edn. revised, Cambridge University Press.
- Easley, G.T.; Leonard, R.H. and Rotter, D.M. (1951). Bacteriological, pathological and clinical studies of reproductive tract of the Hereford bull semen. N. Am. Vet., 32: 258-284.
- Khan, B.U. and Luktuke, S.N. (1967). Studies on the incidence of repeat breeding in Haryana cows. Indian Vet. J. 44: 678-683.
- Kodagali, S.B.; Bhavsar, B.K.; Kavani, F.S.; Derasari, H.J.; Mansuri, A.N.; Dholkia, P.M.; Purohit, J.H. and Shah, N.M. (1980). Clinical trials with 'Furea Bolus' in endometritis and repeat breeder buffaloes. Indian Vet. J. 57: 945-950.
- Krishnamurthy, G.V.; Manjiah, R.D. and Keshvamurthy, B.S. (1974). Bacterial flora of cervical mucous in repeat breeding bovines. Indian Vet. J. 51: 264-268.
- Mutiga, E.R. (1978). Treatment of the repeat breeder cow syndrome in Kenya. Tropical Anim. Health Prod. 10: 223-228.
- Sharma, N.C.; Kulshreshtha, S.B.; Kaushik, S.N. and Katpatal, B.G. (1978). Study of repeat breeder cows and their treatment with special reference to drug sensitivity of infectious agents. Indian J. Anim. Sci. 48: 153-157.

Table 1 : Total number of isolates belonging to different groups of organisms from repeat breeders

Type of animals	<i>Str. pyogenes</i>	<i>Staph. aureus</i>	<i>Staph. epidermidis</i>	<i>M. luteus</i>	<i>C. pyogenes</i>	<i>Esch. coli</i>	<i>Kleb pneumoniae</i>	<i>Ps aeruginosa</i>	<i>Pr vulgaris</i>	<i>Paracol bactrum Hafniaahvei</i>	Total No of isolates
Crossbred cows n=35	6 15.38%	8 20.51%	2 5.13%	4 10.26%	1 2.56%	11 28.2%	3 7.69%	1 2.56%	2 5.13%	1 2.56%	39
Crossbred heifers n=8	1 11.11%	1 11.11%	-- --	-- --	-- --	5 55.56%	-- --	1 11.11%	1 11.11%	-- --	9
Total	7	9	2	4	1	16	3	2	3	1	48

Table 2: *In vivo* efficacy of drugs on different bacterial isolates and conception rate after treatment in repeat breeder animals

Treatment groups	No. of animals treated and inseminated	No. of animals pregnant	Percentage of conceptions
Chloramphenicol (T ₁)	10	8	80.00% ^a
Kanamycin (T ₂)	8	7	87.50% ^a
Supristol (T ₃)	7	5	71.42% ^{ab}
Supristol +			
Chloramphenicol (T ₄)	4	3	75.00% ^{ab}
Hostacyclin (T ₅)	6	4	66.66% ^{ab}
Control	8	2	25.00% ^b

Values bearing same superscript in row did not differ significantly.

IJAR 9:2:109-114 : 1988

Service Behaviour Of Bulls Of Different Breeds Under Uniform Management Conditions

M.R. BHOSREKAR, J.R. PUROHIT, A.B. PANDE and B.R. MANGURKAR

Bharatiya Agro-Industries Foundation, Uruli-Kanchan-412202 Maharashtra State.

ABSTRACT

Five adult bulls each of Holstein Friesian, Jersey, Cross-bred HF and Murrah-buffalo were selected for the study. No significant variation was noted between breeds for different items of service behaviour. There was also no seasonal variation in respect of service behaviour except for reaction time which showed highly significant variation between different seasons. Reaction time was longer in winter and shorter in summer irrespective of breed of bulls. Breed × Season interaction was also found significant for this parameter. Proper training of the bulls and suitable management practices help in optimal utilisation of bulls.

Service behaviour is one of the important measures for assessing reproductive soundness in bulls. Patterns of male sexual behaviour have been described by Hafez and Bouisoou (1975) which encompass sequential behavioural elements like sexual interest, erection, protrusion, mounting, body position, type of seeking, ejaculatory thrust and dismounting. Manifestations of these patterns in the bull varies from full to no expression. In the present communication an attempt was made to study these elements of behaviour through different seasons under similar management conditions for exotic, cross-bred and buffalo bulls.

Material and Methods

Five adult bulls each of Holstein Friesian (HF), Jersey, Cross-bred HF and Murrah

* * *

buffalo were included in the study. They were in regular semen collection schedule and their patterns were set. The collection of semen was done by artificial vagina technique using a live bull of respective breed and species as dummy.

The period of observation was divided into three seasons viz. summer (March to June), Rainy (July to October) and Winter (November to February). Management of bulls throughout the period of observation was under one man's control so variation on account of changes in management was minimised. The concentrate feed offered was one per cent of the body weight, while fodder offered was as per seasonal availability but ad libitum (Table 1).

Two types of AVs were used 30 cm for exotic and cross-bred bulls and 25 cm for buffalo bulls. The temperature pressure combination used was as per enclosed chart (Table 1). The semen collections were done between 5.00 A.M. to 8.30 A.M. twice a week for all the bulls included in the study. The components of behaviour like sexual interest, erection, protrusion, mounting, body position, type of seeking and ejaculatory thrust were quantified by giving scores from 0 to 4 according to the expression of pattern from nil to full.

Results and Discussions

It was seen that there was no significant difference between breeds for different components of service behaviour (Tables 2 and 3). This may be due to the fact that the bulls were already in regular semen collection programme and the patterns were set because of their proper training.

However, they showed seasonal variation significantly for reaction time ($P < 0.01$) Sharma *et al.* (1982) also observed significant seasonal variation for reaction time in Jersey bulls. Bhagoji (1985) recorded significant seasonal variation for reaction time in HF and Jersey cross-bred bulls. However, he also did not observe breed differences.

Bull to bull variation for all behavioural components was so significantly high in each breed that effects of breed and seasons were not seen (Tables 4 and 5). This also confirmed the earlier findings reported by Ali *et al.* (1981) who recorded highly significant variation between bulls for mounting behaviour, thrust intensity and reaction time.

Reaction time in the present findings was lowest in summer and highest in winter which is contrary to earlier reports for Egyptian buffalo bulls (Oloufa *et al.* 1959) which may be due to differences in climatic conditions, fodder availability and time of semen collection.

It is, therefore, concluded that proper training of the bulls helps in setting up uniform behavioural patterns without much variation between different breeds and species for getting optimum results. It is also important to have uniform management practices like least changes in attendants, bull pens and collection procedures and collector.

Acknowledgement

The authors wish to thank Dr. Manibhai Desai, President, BAIF for constant encouragement and permission to publish the results.

Table 1: Climatic variables, green fodder availability and A.V. conditions

Seasons	Atmospheric temperature °C		Relative humidity (%)	Rain fall (mm)	Fodder available	Artificial vagina	
	Maximum	Minimum				Temperature	Pressure mm
Summer (March to June)	35.1 (24.6 to 40.3)*	20.3 (17.4 to 23.4)	71.08	15.1	Green maize and silage of Sweet Sorghum and Maize. Lucerne to certain extent. Dry Sorghum Straw	46°C	35 to 40 for smaller and 50 for bigger
III Rainy (July to Oct.)	29.2 (22.7 to 34.5)	20.7 (14.2 to 23.8)	89.21	74.7	Subabul-Sweet Sorghum mixture. Green maize and Sweet Sorghum Subabul mixture and silage to some extent.	47°C	40 to 45 for smaller and 55-60 for bigger
Winter (Nov. to Feb.)	30.8 (26.8 to 35.3)	10.7 (5.2 to 12.9)	81.0	35.6	Green maize-Subabul mixture, Groundnut dry tops chaffed sugarcane.	49°C	40 to 45 for smaller and 55-60 for bigger

*The figures in parenthesis indicate the range of atmospheric temperature.

Table 2: Mean values of scores of service behavioural components for bulls of different breeds

Breed	Sexual interest	Erection	Protrusion	Mounting	Body position	Type of seeking	Thrust	Reaction time minutes
Holstein Friesian	2.54	2.41	2.37	2.55	2.40	2.37	2.82	2.99
Jersey	2.36	2.32	2.27	2.40	2.41	2.39	2.68	2.82
Cross-bred	2.37	2.26	2.29	2.42	2.43	2.36	2.64	3.01
Buffalo	2.45	2.41	2.41	2.52	2.44	2.48	2.68	3.09

Table 3 : Mean values of scores of service behavioural components for bulls of different seasons

	Sexual interest	Erection	Protrusion	Mounting	Body position	Type of seeking	Thrust	Reaction Time
Summer (March to June)	2.45	2.33	2.36	2.47	2.53	2.45	2.74	2.73
Rainy (July to October)	2.36	2.27	2.28	2.46	2.36	2.34	2.81	2.98
Winter (November to February)	2.48	2.45	2.36	2.46	2.36	2.41	2.58	3.22

Table 4: Mean values of scores of service behavioural components for bulls of different breeds for different seasons

Breed	Season	Components of service behaviour							
		Sexual interest	Erection	Pro-trusion	Mounting	Body position	Type of seeking	Thrust	Reaction (min.)
Holstein	Summer	2.55	2.41	2.50	2.49	2.52	2.41	2.88	2.77
Friesian	Rainy	2.41	2.26	2.26	2.50	2.37	2.29	2.92	3.02
	Winter	2.66	2.55	2.34	2.55	2.30	2.42	2.66	3.19
Jersey	Summer	2.50	2.23	2.25	2.40	2.57	2.46	2.82	2.24
	Rainy	2.21	2.29	2.23	2.40	2.25	2.32	2.77	2.94
	Winter	2.37	2.45	2.35	2.41	2.42	2.39	2.46	3.10
Cross-bred	Summer	2.35	2.21	2.26	2.55	2.55	2.33	2.59	2.94
	Rainy	2.34	2.17	2.21	2.41	2.41	2.37	2.73	3.01
	Winter	2.43	2.41	2.39	2.32	2.34	2.39	2.61	3.09
Murrah	Summer	2.41	2.45	2.45	2.46	2.48	2.62	2.66	2.81
Buffalo	Rainy	2.50	2.38	2.42	2.54	2.42	2.38	2.81	2.97
	Winter	2.46	2.39	2.36	2.57	2.40	2.45	2.57	3.49

Table 5: Analysis of variance for components of service behaviour of bulls of different breeds

Source of variance	df	Mean sum of squares							
		Sexual interest	Erection	Pro-trusion	Mounting	Body position	Type of seeking	Thrust time	Reaction time
Between breeds	3	3.37	2.37	2.00	1.76	0.16	1.36	2.58	5.99
Between seasons	2	2.40	5.07	1.41	0.04	5.82	2.15	8.68	36.3**
Interaction breed — season	6	1.39	0.99	1.04	0.93	0.80	0.66	1.14	3.61*
Between bulls within breed	16	3.34**	4.45**	5.41**	3.41**	2.28**	2.13**	2.50**	1.05
Residual	1832	0.47	0.44	0.36	0.46	0.41	0.39	0.57	1.21
Total	1859								

REFERENCES

- Ali, S.B.A.; Al Hakim, M.K. and Singh, B.P. (1961). Indian J. Anim. Hlth. 20(1): 9-13.
Bhagoji, G.P. (1985). Ph.D. Thesis, Gujarat Agricultural University, Anand.
Hafez, E.S.E. and Bouisouu (1975). "The behaviour of cattle" in Behaviour of Dometic Animals, 3rd Edn. Bailliere Tindall, London, U.K.
Oloufa, M.M.; Sayed, A.A. and Badreldin, A.L. (1959). Indian Dairy Sci. 12: 10-17.
Sharma, R.K.; Pareek, P.K. and Vyas, K.K. (1982). Indian J. Anim. Reprod. 2(1): 45.

IJAR 9:2:114-117 1988

Different Types Of Scroti And Preputial Sheaths In Crossbred Bulls*

K. BABU RAO¹, and A. RAMAMOCHAN RAO²

Department of Animal Reproduction and Gynaecology College of Veterinary Science, Tirupati-517502 (A.P.)

ABSTRACT

The different types of normal scroti observed in cross-bred bulls were oblong (23.53%), square (23.53%), elongated (14.71%), oval (14.71%), round (6.35%) and bifid (4.41%). Abnormal types of scroti were monorchid (8.32%), asymmetric (2.94%) and held close to abdomen (1.47%). Tight sheath (42.65%) was common, followed by medium sheath (33.82%), pendulous sheath (17.65%) and pendulous sheath with everted prepuce (5.38%).

* * *

Scrotum is considered as the most important organ having thermoregulatory mechanism. Different types of scroti are observed in bulls but certain types of scroti may cause disturbed spermatogenesis which may lead to reduction in fertility. Information on different types of scroti in normal fertile bulls is lacking except the

report of Mohanty *et al.* (1988), in Holstein bulls. Certain types of preputial sheaths like pendulous and pendulous with everted prepuce interfere with the breeding efficiency of the bull because they are more prone to infection and lacerations. The present paper deals with different types of scroti and preputial sheaths in crossbred bulls.

Material and Methods

Sixty-eight crossbred bulls aged 18 to 105 months maintained at A.I.C.R.P. (ICAR) on Cattle, Lam Farm, Guntur were examined critically for the type of scrotum and preputial sheath. Normal scroti were classified depending upon its shape into oblong, square, elongated, round, oval, bifid and abnormal types classified as monorchid, asymmetric and held close to abdomen. The preputial sheaths were classified into tight, medium, pendulous and pendulous with everted prepuce. Evaluation of semen was done as per

*Part of thesis submitted by the first author for the award of Ph.D. degree by Andhra Pradesh Agricultural University, Hyderabad.

1. Assistant Research Officer, Cattle Project, Lam Farm, Guntur-522034 (A.P.)

2. Dean of Post-Graduate Studies, A.P. Agricultural University, Rajendranagar, Hyderabad-500030

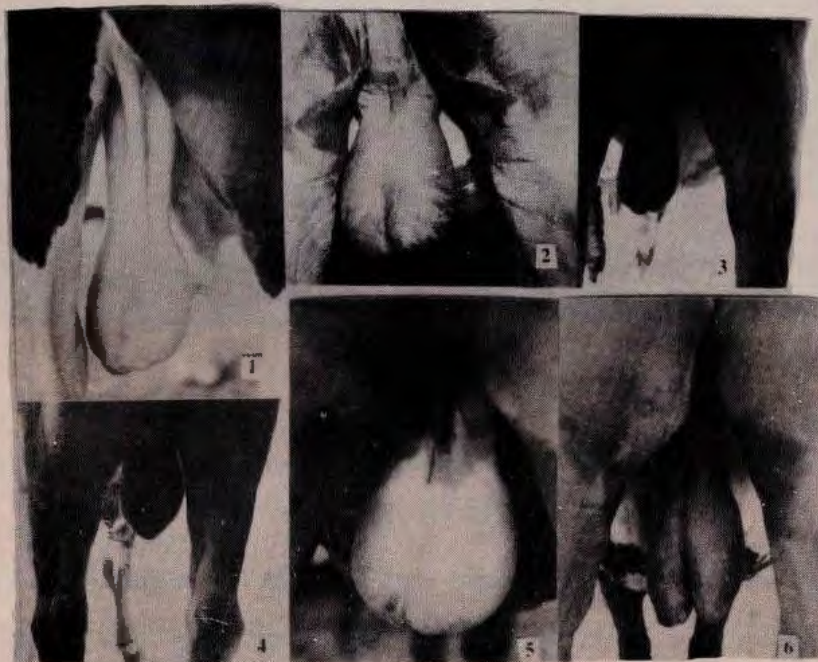
Rao (1971) over a period of one year. Statistical analysis was done as per the methods described by Snedecor and Cochran (1967).

Results and Discussions

Among different types of normal scrota observed in crossbred bulls in the present

normal scrota in this study are within the range stipulated for normal fertile crossbred bulls (Biswas *et al.*, 1976; Rao and Rao, 1978; Garg and Pandit, 1983). But the quality of semen was poor in bulls having abnormal type of scrota due to the presence of high percentage of morphologically abnormal sperms. The

Plate 1



1. Elongated scrotum
2. Square scrotum

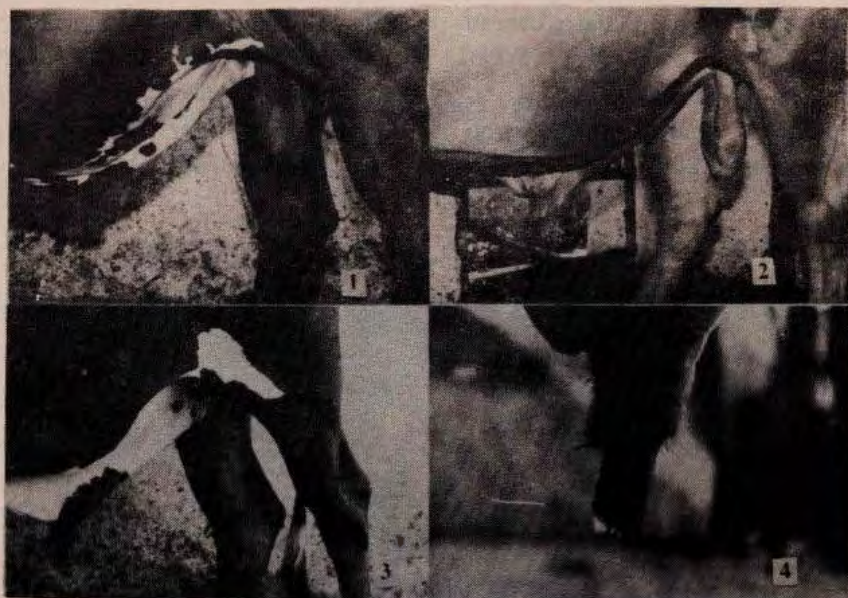
3. Oblong scrotum
4. Oval scrotum

5. Round scrotum
6. Bifid scrotum

study, square (23.53%) and oblong (23.53%) types were more common followed by elongated (14.71%), oval (14.71%), round (7.35%) and bifid (4.41%) (Plate 1). Abnormal types were monorchid (8.32%), asymmetric (2.94%) and held close to abdomen (1.47%) (Figs. 1 to 3). The mean values of various seminal attributes observed for different

difference in mean values among different normal scrota is insignificant.

Tight preputial sheath (42.65%) was found to be common in crossbred bulls followed by medium sheath (33.82%), pendulous sheath (17.65%) and pendulous sheath with everted prepuce (5.88%) (Plate 2).



- | | |
|----------------------------|--|
| 1. Tight preputial sheath | 3. Pendulous preputial sheath |
| 2. Medium preputial sheath | 4. Pendulous preputial sheath with everted prepuce |

Acknowledgements

The authors are thankful to the Senior

Scientist (AB), Cattle Project, Lam Farm,
Guntur (A.P.) for the facilities provided.

REFERENCES

- Biswas, J.C.; Raina, B.L.; Kumar R. and Bhat, P.N. (1976). Studies on live and abnormal spermatozoa in crossbred bulls. *Indian J. Anim. Hlth.* 15: 129.
- Garg, U.K. and Pandit, R.K. (1983). Studies on some seminal attributes in relation to fertility in crossbred bulls. *Indian J. Anim. Reprod.* 4: 26-29.
- Moahnty, D.N.; Dugwekar, Y.G. and Biswas, R.K. (1988). Testicular measurements of Holstein Friesian bulls as an indicator of reproductive performance. Paper presented at National Seminar on trends in Biotechnology to augment fertility and fecundity in farm animals. Madras, Jan. 1988.
- Rao, A.R. (1971). Changes in morphology of sperm during their passage through genital tract in bulls with normal and impaired spermatogenesis. Ph.D. thesis, Royal Vet. College, Stockholm, Sweden.
- Rao, T.L.N. and Rao, A.R. (1978). Semen characteristics of crossbred bulls. *Indian. Vet. J.* 55: 692.
- Snedecor, G.W. and Cochran, W.G. (1967). Statistical methods. 6th Edn. Oxford and IBH Publishing Co., Calcutta.



Fig. 1. Monorchid scrotum



Fig. 2. Asymmetric scrotum

IJAR 9:2:117-118; 1988

Types Of Preputial Sheath In Murrah Buffalo Bulls

A.R. RAO,* G. SOMASEKHARAM, and K. MOULI KRISHNA

Dept. of Animal Reproduction College of Veterinary Science, Tirupati-517502 Andhra Pradesh

The type of sheath in different breeds of cattle has been adequately studied (Sane *et al.*, 1982). The information pertaining to the preputial sheath of buffalo bulls is scanty. The present note deals with different types of preputial sheath observed in sixty Murrah Buffalo bulls aged 4-10 years stationed at two Frozen Semen stations in Andhra Pradesh.

The preputial sheath of Murrah bulls was classified as pendulous, medium and tight sheath. The medium type was found to be the most common variety (52.18%) followed by

pendulous (30.43%) and tight sheath (17.39%) (Fig. 1).

It was also observed that the type of sheath did not influence the semen quality. However, difficulty was encountered in collection of semen from bulls having pendulous sheath.

Pendulous sheath harbours ubiquitous microorganisms and is prone for lacerations (Sane *et al.*, loc. cit.). Since the preputial sheath is a heritable trait, breeding of bulls having pendulous sheath should be

*Presently: Dean of P.G. Studies, A.P. Agricultural University, Hyderabad-500030

discouraged. Efforts should be made to select bulls with tight sheath to overcome the problems mentioned above.

The incidence reported in this study may not represent the incidence in general population as the present study is based on bulls already selected for breeding purposes.

Fig. 1.

Photo plate showing different types of preputial sheath in Murrah Buffalo bulls.



1. Tight Sheath



2. Medium Sheath



3. Pendulous Sheath

REFERENCES

Sane, C.R.; Luktuke, S.N.; Kaikini, A.S.; Hukeri, V.B.; Deshpande, B.R.; Velhankar, D.P. and Kodagali, S.B. (1982). A text book "Reproduction in farm animals". Verghese Publishing House, Bombay-400014.

IJAR 9:2:118-122:1988

A Note On Training Of Boars For Collection Of Semen

M.K. TAMULI, and C.K. RAJKONWAR

Dept. of Gynaecology, Obstetrics and Artificial Insemination, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati-781022

ABSTRACT

Among the seven Landrace boars (11 to 12 months of age) taken for the study, six (85.71%) could be trained successfully over a dummy sow with the help of a tape recorder

by imitating the noisy sound of a teaser sow restrained in a service crate. The mean values of latency of mount, reaction time and ejaculation time were recorded as 52.1 sec., 3 min. 56.9 sec. and 5 min. 52.1 sec. respectively.

The mean values of semen, strained portion and gel mass were recorded as 235.8 ml, 172.0 ml and 63.8 ml respectively and per cent motility and the concentration of spermatozoa were recorded as 83.72 and 195.6 million per ml of semen respectively.

Artificial insemination in pigs is mostly confronted with the problems in training the boars for regular semen collection because of their varying idiosyncratic behaviour. Hence, a novel approach was made to overcome the problems encountered during training the boars for semen collection.

Seven Landrace boars at the age of 11 to 12 months were selected for the study. The boars were deprived from any natural service before training in the service crate (SC) and dummy sow (DS). One wooden SC was constructed on a portable platform (Fig. 1) and one iron frame of dummy back (Fig. 2) was constructed on which a dirty white canvas cushion was fixed and so placed tightly over

the SC that 15 cm of it protruded on the rear part. Both lateral and rear sides of the dummy back were then covered with a dirty white canvas cloth to give the shape of DS. This DS was fixed on the SC as and when needed.

At the outset of training, one teaser sow was forcibly restrained in the SC and semen was collected in the thermos flask for each boar over her for five consecutive days. For collection of semen, penis was grasped just one centimeter behind the tip with the neatly cleaned palm swabed with spirit alcohol and a rhythm of pressure was maintained by 'simple fist' method. Subsequently, the SC was modified into the DS and estrual discharge was smeared over the DS (Campbell and Lingam, 1965) and alternate exposures were made to the DS and teaser sow in the SC for ten trials in this method-I. A maximum period of fifteen minutes was allowed to each boar to remain near the DS for the positive response of mounting and ejaculation, but in method-I the response was totally nil. In method-II, the noisy sound produced by the teaser sow at the

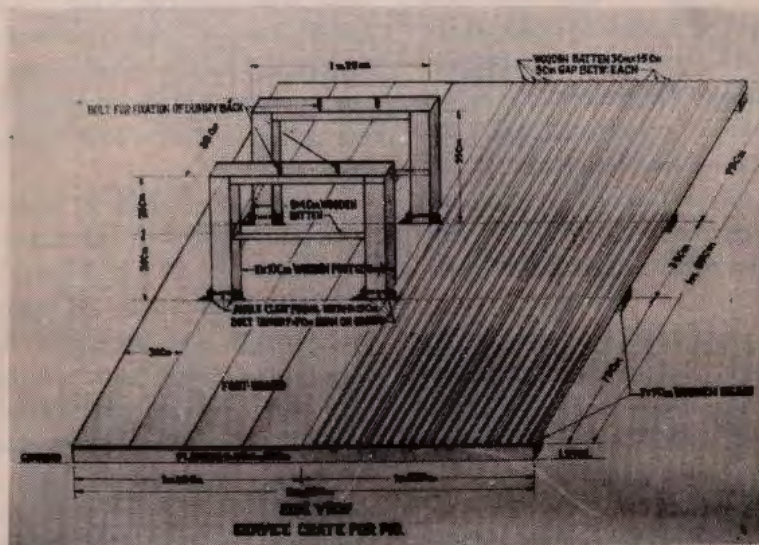
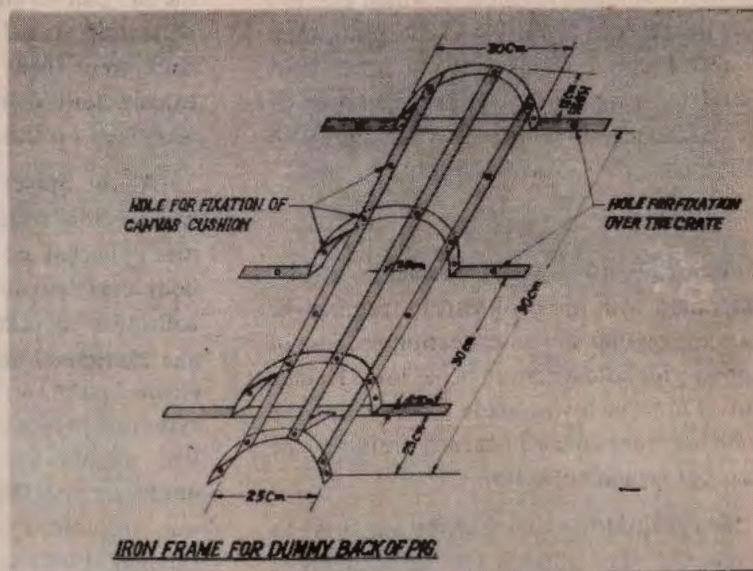


Fig. 1. Service crate (SC) for the Boar

Fig. 2.
Frame of dummy sow
(IS) back



time of semen collection from the boar over her, was recorded in the tape recorder and played near the DS when the boars were exposed for semen collection. As in method-I, alternate exposures to the DS and teaser sow were allowed to each boar for ten trials. On the very first day of trial with tape recorder, one of the boar had successfully mounted over the DS and ejaculated semen in 'simple fist' method. The rest of the boars subsequently mounted over the DS and gave successful ejaculation within the third day of trial resulting in success rate of 85.71%(6). one of the boars though mounting on DS, did not attain orgasm and hence semen could not be collected.

After training the boars with method-II, semen was collected over the DS (Fig. 3) from the six boars at three days interval. Latency to mount (Signoret, 1968), reaction time (Lindsay, 1969) and ejaculation time (Steinbach, 1972), volume of semen including volume of gel mass and strained portion, per cent motility and concentration of spermatozoa were recorded.

Courtship behaviour of smacking, biting and poking the head into the hind quarter were noted as in the case of natural service. These behaviours were also reported by Glover (1955). At the time of semen ejaculation, the tail twisted at its base clockwise and croup made a groove along with the intermittent constriction of anal sphincter muscle. Slight protrusion of the tongue and profuse salivation during the act of ejaculation were also noticed among the boars.

The mean values of latency to mount, reaction time and ejaculation time was recorded to be 52.1 ± 17.0 sec., 3 min. 56.9 ± 51.5 sec. and 5 min. 52.1 ± 19.5 sec. respectively. The latency to mount and the reaction time were found to be lower than that reported by Signoret (1968) and Steinbach (1972). The ejaculation time was found to be lower than that recorded by Steinbach (1972) and was inconsistent with the findings of Swierstra and Rahnefeld (1967). These discrepancies in findings of service behaviour might be due to the audio-visual condition



Fig. 3. Boar semen collection over a dummy sow (DS)

created by the tape recorder which played a positive role in the reflex stimulation of sexual excitement in boars.

The mean volumes of semen, gel mass and strained portion were found to be 235.8 ± 11.8 ml, 63.8 ± 4.1 ml and 172.0 ± 12.0 ml respectively and were within the range (Aamdal and Hogset, 1957; Turkheimer *et al.*, 1958; Campbell and Lingam, 1965). The mean value of per cent motility was found to be

83.72 ± 0.59 and was well within the range of normal fertile boar (Holst, 1949). The mean concentration of spermatozoa was found to be 195.6 ± 15.4 million per ml of semen and was higher than the value reported by Siler *et al.* (1977).

Acknowledgements

The help and assistance rendered by the Dean and Assoc. Director of Research is gratefully acknowledged.

REFERENCES

- Aamdal, J. and Hogset, I. (1957). Artificial insemination in swine. J. Amer. Vet. Med. Assoc. 131: 59-64.
- Campbell, E.A. and Lingam, S.A. (1965). Artificial insemination of pigs in Australia, I. Training of Boars and collection of semen samples. Aust. Vet. J. 47: 147-150.
- Glover, T.D. (1955). The semen of the Pig. Vet. Rec. 67: 36-40.
- Holst, S.J. (1949). Sterility in boars. Nord. Vet. Med. 1: 87-92.
- Signoret, J.P. (1968). In "The Behaviour of Domestic Animals" (Hafez, E.S.E. ed.) 3rd edn. Williams & Wilkins Co., Baltimore, U.S.A. pp. 305.

- Siler, R.; Pavlik, J.; Fulik, J. and Bazant, J. (1977). Phenotypic characters of semen in boars. *Zivocisna Vyroba*, 22: 687-696 (Anim. Breed. Abstr. 46: 5090).
- Steinbach, J. (1972). Bioclimatic influences on sexual activity in boars. VIIIth Int. Confer. Anim. Reprod. Artif. Insem. Munich Summaries, pp. 427.
- Swierstra, E.E. and Rahnefeld, G.W. (1967). Semen and testis characteristics in young yorkshire and Locombe boars. *J. Anim. Sci.* 26: 149-152.
- Turkheimer, A.R.; Young, D.C. and Foote, R.H. (1958). Technics of semen production in young boars. *Cornell Vet.* 48: 291-294.

IJAR 9:2:122-124: 1988

Efficacy Of Tris-Citric Acid And Sodium Citrate Buffers Against Cold Shock To Buffalo Spermatozoa

R.K. TULI, S.P. SINGAL and M.N. RAZDAN

Department of Animal Production Physiology, Haryana Agricultural University, Hisar-125004.

ABSTRACT

Sperm motility, live and abnormal spermatozoa and GOT enzyme were recorded before and after cold shock in buffalo semen, extended in tris-citric acid and sodium citrate buffers. The difference in percent motile spermatozoa before cold shock was not significant but after cold shock the percent motile spermatozoa were significantly ($P < 0.01$) higher in tris-citric acid than in sodium citrate buffer. The percent live spermatozoa were higher in Tris buffer and less in sodium citrate, the difference being significant ($P < 0.01$). However, after cold shock, these differences became non-significant. The extent of post shock sperm abnormalities too did not differ significantly either due to buffer or due to cold shock. The release of GOT before cold shock was significantly ($P < 0.01$) less in tris-citric acid than in sodium citrate buffer. Although, the total release of GOT after cold shock to semen extended in tris-citric acid buffer was

comparatively less, as compared to sodium citrate buffer, but the difference was not significant.

* * *

Some reports on the effect of cold shock on the livability and abnormalities of buffalo spermatozoa are available (Tiwari *et al.*, 1969; Roy *et al.*, 1976; and Chinnaiya and Ganguli, 1978). Tuli *et al.* (1982) studied the effect of cold shock on percent motile, live and abnormal spermatozoa of buffalo semen extended in tris yolk (TY) and citric acid whey (CAW) extenders. Roychauchary *et al.* (1974) studied the effect of cold shock on the release of GOT and GPT enzymes from bull and ram semen. This paper presents the observation on the effect of cold shock on motility, percent live and abnormal spermatozoa and release of GOT from buffalo bull semen extended in tris citric acid and sodium citrate buffer.

Materials and Methods

Ten semen ejaculates showing mass activity greater than plus three were collected

from four healthy Murrah buffalo bulls. Semen collection was made at weekly interval in the artificial vagina. The various seminal characteristics viz., volume, colour, consistency, mass activity and sperm concentration were recorded. One-half mililitre of each semen sample was taken in duplicate sets of test tubes, and total volume was made up to 5 mililitre by adding tris-citric acid in one set of tubes and 2.9 percent sodium citrate buffer in other set. Each extended semen sample was divided into two equal parts- one as control (before cold shock) and the other for cold shock treatment. A drop of semen from control sample was taken for recording motility and slides with eosin-nigrosin stain were prepared for counting percent live and abnormal spermatozoa. Rest of the control semen sample was centrifuged at 2000 rpm for 10 min at 5°C and the supernatant was used for estimation of GOT enzyme activity by the method of Yatzidis (1960). Cold shock was applied by keeping the tubes containing extended semen samples in crushed ice for 10 min. The motility, live and abnormal spermatozoa and GOT enzyme release was measured after giving cold shock, in the same way as before cold shock. The data were subjected to statistical analysis as per Snedecor and Cochran (1967).

Results and Discussion

The percent motile spermatozoa before cold shock were 59.00 ± 3.16 and 57.50 ± 3.54 per cent in tris-citric acid and sodium citrate buffer, respectively. Analysis of data revealed non-significant difference in motility before cold shock, but after cold shock, the percent motile spermatozoa were 28.00 ± 1.53 and 18.00 ± 2.13 in tris citric acid and sodium citrate buffer respectively, and differed significantly ($P < 0.01$).

The percent decrease in motility after cold shock was 31.00 and 39.50 in tris-citric acid and sodium citrate, respectively. The higher decrease in sperm motility in the present study may be due to the absence of egg yolk in both the buffers, because Tuli *et al.* (1982) have confirmed that the use of egg yolk in tris-citric acid buffer, prevents the impact of cold shock on sperm motility.

The percent live spermatozoa before cold shock were higher in tris-citric acid buffer (65.40 ± 3.05) than in sodium citrate (56.80 ± 3.20) and the difference was significant ($P < 0.01$). The percent live spermatozoa after cold shock were also higher (38.50 ± 4.38) in tris-citric acid buffer but the difference between buffers was non-significant. The percent decrease in live spermatozoa after cold shock was little less in tris-citric acid (26.90) than sodium citrate (28.90) Tiwari *et al* (1969) and Roy *et al* (1976) observed 44.80 and 68.10 percent loss in live spermatozoa after cold shock to "neat" buffalo semen. Tuli *et al* (1982) observed 22.16 and 16.16 percent decline in live spermatozoa after cold shock to buffalo semen extended in CAW and TY extender, respectively. There were non-significant differences in sperm abnormalities before and after cold shock to semen extended in both the buffers. The increase in sperm abnormalities after cold shock was 1.15 and 1.54 percent in tris-citric acid and sodium citrate buffer, respectively. These values were much lower than reported by Chinnaiya and Ganguli (1978), who observed 8.45 and 9.33 percent increase in abnormalities after cold shock treatment to first and second ejaculate of buffalo semen, respectively.

The release of GOT enzyme before cold shock was 74.50 ± 4.25 units/ml in tris-citric acid as compared to 110.50 ± 10.05 units in

sodium citrate and the difference was highly significant ($P < 0.01$). The release of GOT enzyme after cold shock was lower in tris-citric acid (111.00 ± 14.07 units) than sodium citrate (130.00 ± 7.15 units). This is in agreement with the findings of Roychaudhary *et al* (1974) who observed that release of GOT

and GPT enzymes was more after cold shock in bull and ram semen extended in sodium citrate than tris-citric acid buffer. It can be concluded from the present study that tris-citric acid buffer protects buffalo bull spermatozoa more efficiently against cold shock as compared to sodium citrate buffer.

REFERENCES

- Chinnaiya, G.P. and Ganguli, N.C. (1978). Cold shock resistance test in evaluating buffalo semen. *Indian J. Dairy Sci.* **31**: 284-286.
- Roy, D.J.; Deo, Sand Khan, B.V. (1976). Comparative studies on cold shock treatment to Haryana and Murrah bull semen. *Indian J. Animal Health.* **15**: 53-56.
- Roychaudhary, P.N.; Pareek, P.K. and Gowda, H.C. (1974). Effect of cold shock on GOT and GPT release from bull and ram spermatozoa. *Andrologia* **4**: 315-319.
- Snedecor, G.W. and Cochran, G.W. (1967). *Statistical methods* (6th Ed.) Oxford and IBH Publishing Co., New Delhi.
- Tiwari, R.B.L.; Abhi, H.L. and Sengupta, B.P. (1969). A note on the possibility of using cold shock resistance test on a dependable index for preliminary screening and grading of A.I. bulls under field conditions. *Indian Vet. J.* **46**: 424-426.
- Tuli, R.K.; Singh Mehar and Matharoo, J.S. (1982). Efficacy of tris yolk (TY) and citric acid whey (CAW) extenders against cold shock in buffalo semen. *Life Sci. Adv.* **1**: 43-45.
- Yatzidis, H. (1960). Serum transaminases. *Nature*, **186**: 79-80.

IJAR 9:2:124-131: 1988

Leakage Of Phosphatases From Buffalo Bulls Spermatozoa During Freezing: Effect Of Dilutors, Seasons, Bulls & Stages Of Freezing*

A.J. DHAMI and S.B. KODACALI

Department of Gynaecology and Obstetrics, Gujarat College of Veterinary Science and Animal Husbandry, Gujarat Agricultural University, Anand Campus, Anand-388001.

ABSTRACT

A study on the effects of semen extenders (TFYG, EYCG and LYG), seasons (hot, wet and cold), bulls and stages of freezing (pre-freeze and 7th day post-freeze) on the leakage of phosphatases from buffalo spermatozoa was

undertaken in 48 ejaculates. The overall mean AKP and ACP activities in pre-freeze seminal plasma averaged 68.02 ± 3.72 and 31.60 ± 1.74 KA units/100 ml, which increased highly significantly ($P < 0.01$) to 94.32 ± 4.50 and 53.22 ± 2.46 KA units/100 ml, respectively, in

* A part of the M.V.Sc. thesis submitted to the Gujarat Agril. Univ., Anand by the first author.

frozen-thawed samples. The release of alkaline phosphatase was highly significantly ($P<0.01$) influenced by the type of semen extenders used as well as between bulls under study. TFYG diluent was found to provide better protective action on the leakage of phosphatases during freezing. The seasonal influence on the leakage of both AKP and ACP was highly significant ($P<0.01$). Hot season had lowest AKP-ACP leakage as compared to wet and cold seasons. The interactions studied revealed that the bull \times season, period \times season and dilutor \times season interactions were significant ($P<0.05$) only for AKP activity. The levels of AKP and ACP showed negative relationship with each other as regards their release during freezing of semen. Fertility rate was significantly and negatively correlated with the AKP released pre-freeze (-0.572) and post-freeze (-0.628). Sperm concentration per ml was significantly and negatively correlated with AKP released pre-freeze (-0.633) and post-freeze (-0.623). These results indicated that higher the AKP leakage, lower the fertility rate of frozen semen.

* * *

Semen phosphatases play an important role in dephosphorylation in sperm metabolism. Alkaline and acid phosphatases (AKP-ACP) in semen reflect the functional state of accessory sex glands and metabolic activity of spermatozoa. Zvereva and Cuhrii (1971) reported increase in the conception rates in cows following inseminations with semen containing increasing order of AKP.

According to Ibrahim *et al* (1985), though the epididymis represents the main source of AKP and ACP in semen and ampullae share a great part in contributing these elements, at least some might be the results of leakage from sperm cells (Vorobev, 1980; Mahmoud *et al*,

1986). However, the studies on leakage of phosphatases during freezing of buffalo spermatozoa are scarce. Hence, the present study was undertaken to study the effect of deep freezing using various extenders on the leakage of phosphatases from Surti buffalo semen.

Materials and Methods

A study was conducted for a period of one year divided into three seasons viz., hot (March-June), wet (July-Oct.) and cold (Nov.-Feb.). Four healthy Surti buffalo bulls, aged 5 to 6 years, under a 5 day collection schedule and maintained under identical nutritional and managerial conditions at the Department of Gynaecology and Obstetrics, Gujarat Veterinary College, Anand were included in the study. Three extenders viz., tris fructose yolk glycerol (TFYG), egg yolk citrate glycerol (EYCG) and lactose yolk glycerol (LYG) were tried at 6% glycerol level.

Forty-eight semen ejaculates showing optimum quality, monthly once from each of 4 bulls, were subjected to ultra-low temperature freezing using split ejaculate technique with dilution rate of 1:10. Eight French medium straws were frozen from each of the three split samples in liquid nitrogen vapour for 10 minutes after 5 hrs of equilibration period at 5°C and were stored frozen for 7 days. Immediately after dilution and filling of straws, the remaining quantity of split samples (2-3 ml) was centrifuged at 1500 rpm for 15 minutes and the seminal plasma was maintained separately at 5°C in sterilized vials till enzymes assay was made. Similarly, the activity of these enzymes was determined in the seminal plasma of frozen-thawed samples. Thawing was done at 38°C temperature in water-bath for 15-20 seconds. The levels of acid and alkaline phosphatases

in the pre and post freeze seminal plasma were estimated by colourimetric method according to King and Armstrong (1934) within 2-4 hrs of collection or thawing. The data were analysed in 4 factors Factorial C.R.D. (Snedecor and Cochran, 1971).

Results and Discussions

The overall mean AKP and ACP enzymes activities in the prefreeze seminal plasma were 68.02 ± 3.72 and 31.60 ± 1.74 KA units/100 ml. These levels increased significantly to 94.32 ± 4.50 and 53.22 ± 2.46 KA units/100 ml, respectively, in the frozen-thawed samples. (Tables 1, 2 and Fig. 1)

The differences in the levels of both AKP and ACP enzymes between pre and post-freeze periods were highly significant ($P < 0.01$) (Table 3). These results closely agreed with the findings of Mahmoud *et al* (1986) who reported highly significant increase in the activity of both AKP and ACP in frozen-thawed bull seminal plasma after one day of freezing as compared to freshly diluted semen. The reason attributed was higher concentration of these enzymes in the spermatozoa of bulls than in the seminal plasma, thus leaking of enzymes from cells due to cold shock and damage while freezing. Vorobev (1980) observed the trend of significantly increasing and decreasing activity of AKP in the ram seminal plasma and spermatozoa immediately after dilution, equilibration, in cold shock samples and in frozen-thawed samples indicating leakage of sperm cell AKP enzyme into the seminal plasma. These findings thus, support the present findings in buffalo semen.

The effect of extenders on the leakage of AKP was observed to be highly significant ($P < 0.01$), but for ACP it was non-significant

(Table 3). The trend of mean AKP levels for split samples diluted in TFYG, EYCG and LYG diluents was in increasing order at both pre and post freeze stage, indicating low leakage in TFYG than in the other two diluents (Table 1). The levels of ACP between different diluents, though non-significantly different, showed greater and lesser sperm cell ACP leakage during freezing in EYCG and LYG diluents, respectively (Table 2). This indicated better protective action of LYG and the least of EYCG diluents on sperm cell ACP leakage during freezing.

Highly significant differences were observed between bulls for the levels of AKP enzymes in pre and post freeze seminal plasma. However, the bull effect for ACP activity was nonsignificant (Table 3). Variation between bulls regarding neat semen/seminal plasma levels of phosphatases have been reported by Abdou *et al* (1978). This may also be true in pre and post freeze seminal plasma.

The seasonal influence studied revealed highly significant effect on both AKP and ACP levels in pre and post freeze seminal plasma. Hot season was found to have lowest sperm cell damage as indicated by least increase in the percent post-freeze extracellular leakage of both AKP and ACP enzymes over pre-freeze levels, when compared with wet and cold seasons (Table 1 & 2). The seasonal variations in the AKP-ACP levels observed in the present study may be due to inherent variation in the semen quality and biochemical constituents in different months/seasons as reported by Nafornita *et al* (1977).

The interaction effects between bull \times season, period \times season and dilutor \times season

were statistically significant ($P < 0.05$) for the AKP levels only (Table 3). The alkaline phosphatase leakage at both pre and post freeze stages had significant and negative correlations with sperm concentration per ml (-0.633 and -0.623) and fertility of frozen semen (-0.572 and -0.628), while acid phosphatase leakage at pre and post freeze stage was positively but nonsignificantly correlated with post-thaw motility (+0.476 and +0.505). This indicated that higher the leakage of alkaline phosphatase, lower the fertility of

frozen semen. Bottor *et al* (1973) and Petkov *et al* (1980) found significant and positive correlation between seminal AKP and conception rates in cow-bulls, which is in accordance with the present findings in buffalo bulls.

Acknowledgement

We are grateful to Dr. M.K. Patel, Principal, Gujarat Veterinary College, Anand for his interest and the facilities provided for conduct of this work.

REFERENCES

- Abdou, M.S.S.; El-Guindi, M.N.; El-Menoufy, A.A. and Zaki, K. (1978). Enzymatic profile of the semen of bovine (*Bubalus bubalis* and *Bos taurus*). II Parallelism between acid and alkaline phosphatases and various measures of semen quality. Anim. Breed. Abstr. 48: 85.
- Bottor, T.; Gellne, R.; Bogdan, A.T.; Muntean, M.; Mates, N.; Kadar, L. and Salomir, O. (1973). Correlations of biochemical properties with fertility of buffalo spermatozoa from the selected A.I. centres at Cluj. Anim. Breed. Abstr. 41: 600.
- Ibrahmin, S.S.; El-Azab, A.I.; Rakha, A.M. and Soliman, F.A. (1985). The contribution of the different parts of the reproductive system to the ejaculates of the buffalo bulls. A brief communication-46, in Proceed. of *First World Buffalo Congress*, held at Cairo, Egypt on Dec. 27-31, 1985.
- King, E.J. and Armstrong, A.R. (1934). Can. Med. Assoc. J. 31: 37 c.f. Varley, H.C. (1963) in *Practical Clinical Biochemistry*, 3rd edn. Interscience Books, Inc. New York.
- Mahmoud, M.F.; Ragab, M.R.; Yousef, A.A.; Dowidar, M.F. and El-Sherbini, S. (1986). Comparative study of phosphatases activity of fresh and frozen semen of bovine. Indian. Vet. J. 63(5): 393-396.
- Nafornta, M.; Rosu, M.; Opera, I.; Negrea, M.; and Nicotae, M. (1977). Some physical and biochemical values and the activity of some enzymes in bull semen in winter. Anim. Breed. Abstr. 48: 1815.
- Petkov, Z.A.; Stoyanov, T.K.; Zagorski, D.I.; Tounchev, T.T. and Zlatarav, S.T. (1980). Studies into the correlations between the activity of alkaline phosphatase (AP) and biological indicators of bull semen. Anim. Breed. Abstr. 50: 1419.
- Snedecor, G.W. and Cochran, W.G. (1971). *Statistical Methods*. 6th edn. The Iowa State Univ. Press, Ames, Iowa, USA.
- Vorobev, N.N. (1980). Phosphatase activity in ram semen. Anim. Breed. Abstr. 50: 781.
- Zvereva, G.V. and Cuhrii, B.M. (1971). Phosphatase activity in bull semen and quality of spermatozoa. Anim. Breed. Abstr. 39: 3271.

Fig. 1. AVERAGE PRE AND POST FREEZE AKP & ACP LEVELS (KAU/100 ml) IN BUFFALO BULLS' SEMINAL PLASMA IN DIFFERENT DILUTORS & SEASONS.

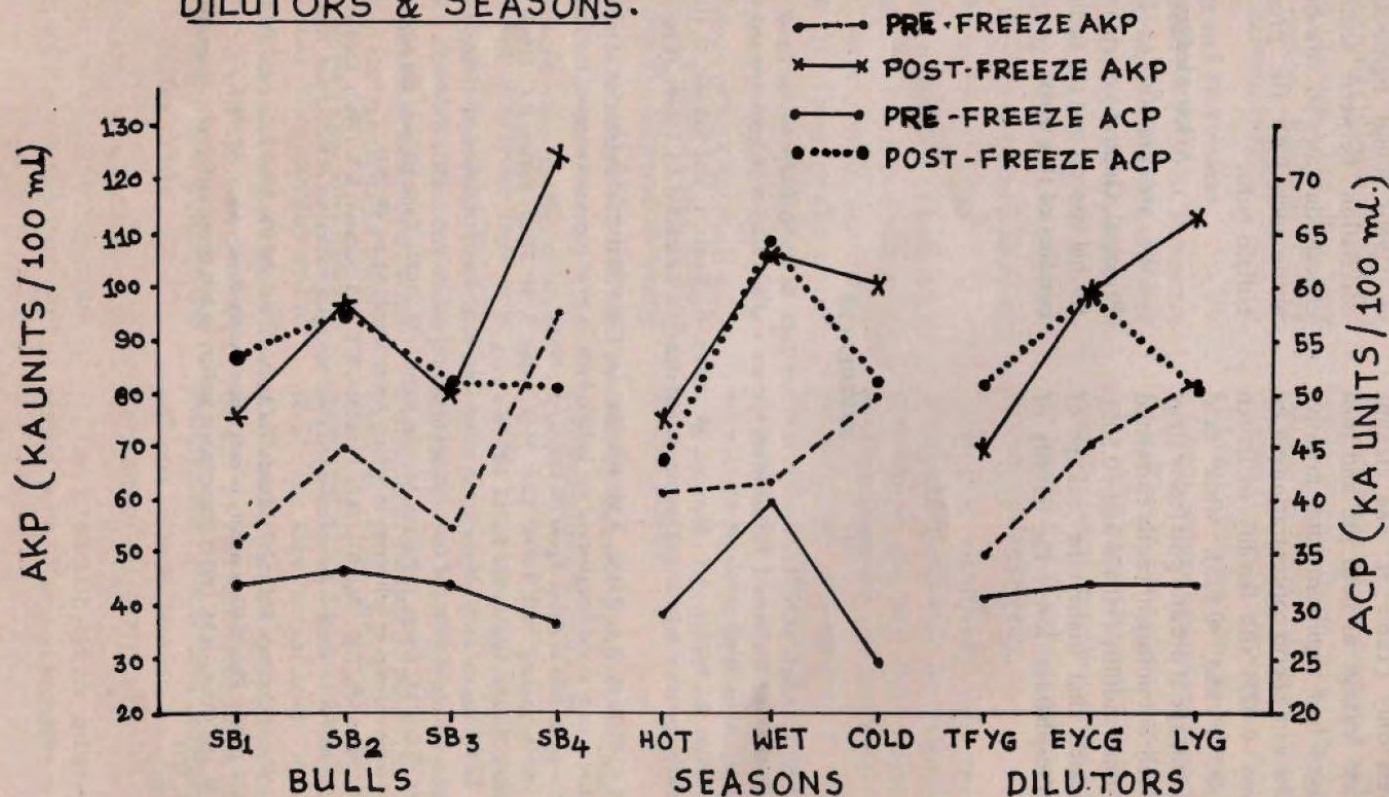


Table-1: Extracellular release of Acid Phosphatase (KAU/100 ml) during freezing of buffalo spermatozoa (in 1:10 dilution) in different seasons using three extenders (Mean \pm SE)

Bull Season	TFYG		EYCG		LYG		Overall Average	
	Pre-freeze	Post-freeze	Pre-freeze	Post-freeze	Pre-freeze	Post-freeze	Pre-freeze	Post-freeze
Bull								
SB ₁	39.58 \pm 7.50	59.16 \pm 8.73 (49.47%)	54.33 \pm 8.74	78.65 \pm 11.86 (44.76%)	62.54 \pm 9.49	91.09 \pm 14.34 (45.65%)	52.48 \pm 5.11	76.30 \pm 7.45 (45.39%)
SB ₂	54.77 \pm 9.56	72.30 \pm 8.45 (32.01%)	69.75 \pm 12.72	105.01 \pm 16.84 (50.55%)	84.85 \pm 14.73	109.84 \pm 14.82 (29.45%)	69.79 \pm 7.32	95.72 \pm 8.15 (37.15%)
SB ₃	40.51 \pm 9.57	57.34 \pm 10.28 (41.57%)	55.94 \pm 9.01	81.52 \pm 14.04 (45.73%)	67.18 \pm 9.57	101.83 \pm 13.23 (51.58%)	54.54 \pm 5.58	80.23 \pm 7.45 (47.10%)
SB ₄	63.24 \pm 9.06	90.28 \pm 12.21 (42.76%)	105.32 \pm 15.49	130.85 \pm 20.49 (24.24%)	117.19 \pm 17.62	153.99 \pm 17.72 (31.40%)	95.25 \pm 9.03	125.04 \pm 10.15 (31.28%)
Season								
Hot	53.84 \pm 6.92	65.83 \pm 7.23 (22.27%)	58.75 \pm 6.37	71.39 \pm 7.28 (21.51%)	71.95 \pm 9.62	86.67 \pm 10.86 (20.46%)	61.52 \pm 4.53	74.63 \pm 5.15 (21.31%)
Wet	37.50 \pm 7.07	69.99 \pm 11.11 (86.64%)	67.55 \pm 12.08	113.63 \pm 18.62 (68.22%)	84.79 \pm 14.50	138.56 \pm 17.00 (63.42%)	63.28 \pm 7.16	107.40 \pm 9.15 (69.72%)
Cold	57.24 \pm 9.16	73.49 \pm 8.85 (28.39%)	87.71 \pm 13.26	112.01 \pm 13.61 (27.70%)	92.82 \pm 12.64	117.33 \pm 10.94 (26.41%)	79.26 \pm 7.08	100.94 \pm 6.15 (27.35%)
Overall Average	49.53 \pm 4.57	69.77 \pm 5.22 (40.86%)	71.34 \pm 6.46	99.01 \pm 8.99 (38.79%)	83.19 \pm 7.15	114.19 \pm 8.10 (37.26%)	68.02 \pm 3.72	94.32 \pm 4.15 (38.67%)

Figures in parentheses indicate percent increase in the extracellular AKP levels in post-thaw seminal plasma over pre-freeze levels.

Table-2: Extracellular release of Alkaline Phosphatase (KAU/100 ml) during freezing of buffalo spermatozoa (in 1:10 dilution) in different seasons using three extenders (Mean \pm SE)

Bull Season	TFYG		EYCG		LYG		Overall Average	
	Pre-freeze	Post-freeze	Pre-freeze	Post-freeze	Pre-freeze	Post-freeze	Pre-freeze	Post-freeze
Bull								
SB ₁	33.01 \pm 6.37	55.02 \pm 7.24 (66.61%)	33.02 \pm 6.30	57.10 \pm 7.96 (75.65%)	30.78 \pm 5.47	48.42 \pm 8.03 (57.31%)	32.27 \pm 3.40	53.51 \pm 4.39 (65.82%)
SB ₂	31.02 \pm 4.49	50.48 \pm 5.56 (65.95%)	34.55 \pm 7.40	66.14 \pm 10.62 (91.54%)	35.03 \pm 7.17	56.16 \pm 11.31 (60.32%)	33.53 \pm 3.65	57.59 \pm 5.45 (71.76%)
SB ₃	30.32 \pm 4.41	48.78 \pm 7.36 (60.88%)	33.53 \pm 5.06	55.54 \pm 8.70 (65.64%)	32.09 \pm 7.79	49.47 \pm 10.84 (54.16%)	31.98 \pm 3.38	51.26 \pm 5.12 (60.29%)
SB ₄	29.79 \pm 5.68	50.33 \pm 5.67 (68.95%)	25.79 \pm 3.98	52.90 \pm 5.48 (105.12%)	30.24 \pm 8.51	48.33 \pm 12.52 (59.82%)	28.61 \pm 3.57	50.52 \pm 4.80 (76.58%)
Season								
Hot	31.37 \pm 5.19	45.51 \pm 5.39 (45.07%)	29.34 \pm 3.63	46.95 \pm 4.83 (60.02%)	28.19 \pm 4.54	39.07 \pm 5.74 (38.60%)	29.63 \pm 2.19	43.84 \pm 3.05 (47.96%)
Wet	34.73 \pm 5.09	55.04 \pm 5.98 (58.47%)	39.43 \pm 6.71	67.52 \pm 9.66 (71.24%)	46.15 \pm 8.11	70.49 \pm 12.06 (52.74%)	40.02 \pm 3.89	64.35 \pm 5.45 (60.79%)
Cold	27.00 \pm 2.70	52.90 \pm 5.13 (95.93%)	26.40 \pm 3.58	59.30 \pm 5.38 (124.62%)	21.76 \pm 3.55	42.33 \pm 6.37 (94.53%)	25.05 \pm 1.89	51.47 \pm 6.7 (105.47%)
Overall Average	31.03 \pm 2.57	51.15 \pm 3.17 (64.84%)	31.72 \pm 2.87	57.92 \pm 4.13 (82.60%)	32.03 \pm 3.58	50.60 \pm 5.25 (57.98%)	31.60 \pm 1.74	53.22 \pm 2.46 (68.42%)

Figures in parentheses indicate percent increase in the extracellular ACP levels in post thaw seminal plasma over preeeze levels.

Table - 3: Analysis of variance showing the effects of bulls, seasons, dilutors, stages of freezing (periods) and their interactions on the extracellular release of phosphatases from buffalo spermatozoa.

Sources of variation	D.F.	AKP		ACP	
		M.S.	'F'	M.S.	'F'
Bulls (B)	3	31527.820	18.067**	453.595	0.747 ^{NS}
Seasons (S)	2	12895.459	7.389**	6995.773	11.514 **
Dilutors (D)	2	37729.623	21.620**	419.625	0.691 ^{NS}
Periods (P)	1	49817.202	28.546**	33677.257	55.429 **
B × D	6	1267.471	0.726 ^{NS}	166.715	0.274 ^{NS}
B × S	6	3777.425	2.165*	1114.762	1.835 ^{NS}
B × P	3	113.279	0.065 ^{NS}	69.830	0.115 ^{NS}
D × S	4	4223.287	2.420*	1152.221	1.896 ^{NS}
D × P	2	727.946	0.417 ^{NS}	390.957	0.643 ^{NS}
S × P	2	6153.236	3.526*	1018.934	1.677 ^{NS}
Error	256	1745.160	—	607.579	—

** P< 0.01, * P<0.05, NS Non-significant.

IJAR 9:2:131-134 : 1988

Leakage Of Transaminases And Lactic Dehydrogenase In Chilled Semen Of Bucks Of Different Breeds

S.N. SINHA¹, S.K. SINGH and A.K. SINHA

Department of Gynaecology, Obstetrics and Artificial Insemination, Ranchi Veterinary College, Ranchi-834007

ABSTRACT

Activities of transaminases namely, aspartate aminotransferase (AST), alanine aminotransferase (ALT) and lactic dehydrogenase (LDH) were estimated in 132 chilled semen samples collected from bucks of different breeds. Of these, 36 semen samples were collected from Black Bengal bucks and 48 each from Jamnapari and Barbari bucks. Seminal plasma in all the semen samples

before dilution in Tris egg yolk fructose citric acid glycerol extender was removed by centrifugation. Assay of the activities of enzymes was made in diluted semen samples kept in refrigerator (4° to 8°C) for 24 hours. Mean values of activities of AST, ALT and LDH (units/10⁹ sperms) in Black Bengal bucks were found to be 39.39 ± 1.57, 26.81 ± 1.95 and 279.4 ± 3.57 respectively. Corresponding values in Jamnapari bucks

Present Address:

University Professor, Department of Gynaecology, Obstetrics and A.I., Bihar Veterinary College, Patna 800014.

were 27.96 ± 1.24 , 17.10 ± 0.87 and 253.69 ± 3.43 respectively and in Barbari bucks the values were 28.63 ± 1.35 , 18.54 ± 1.05 and 251.71 ± 2.73 respectively. Higher activities of the enzymes were found in Black Bengal bucks and the effect of breed on leakage of enzymes was found to be highly significant. There was no significant difference between bucks of Jamnapari and Barbari breeds but both the breeds were significantly different from that of Black Bengal breed in respect of leakage of AST, ALT and LDH.

* * *

Asparate Aminotransferase, alanine aminotransferase and lactic dehydrogenase are intracellular enzymes of spermatozoa and their leaching into the seminal plasma may indicate damage to the plasmalemma of the sperms. This also is expected to indicate variations in spermatozoal sensitivity to cryogenic stress and the assay of the activities of these enzymes has been used to evaluate the quality of semen (Flipse, 1960; Crabo *et al.*, 1971; Rao and Pandey, 1977; Jani *et al.*, 1983). This study was conducted to assess variations in sensitivity of the sperms of bucks of different breeds to the effects of chilling on the basis of leakage of certain intracellular enzymes.

Materials and Methods

A total of 11 bucks representing Jamnapari (4), Barbari (4) and Black Bengal (3) breeds were used in the study. Semen was collected using artificial vagina on twice a week schedule. Samples showing mass activity of +4 and above (Tomar, 1976) and volume of 0.8 ml and above were only included for enzymatic studies. Samples were prediluted in Tris buffer containing 8% glycerol and 10% egg yolk and were centrifuged at 3000 rpm for 15 minutes. The

supernatant was aspirated and the centrifugate was rediluted in Tris extender, but at this stage the concentration of egg yolk was raised to 20 percent. After dilution, penicillin G sodium and streptomycin at the rate of 1000 i.u. and 1000 microgram respectively per ml of diluted semen were added. In every sample the concentration of sperms in the centrifugate was ascertained by haemocytometer. After keeping the samples in refrigerator (4° to 8°C) for 24 hours, they were centrifuged at 1500 rpm for 15 minutes. The supernatant was aspirated and was collected in sterilised vials for measuring the activities of the enzymes. Activity of transaminases was measured according to Reitman and Frankel (1957) and that of LDH according to Cabaud *et al* (1958) as described by Oser (1965). Activities of transaminases and LDH expressed in units denoted amount of enzymes which catalysed the transformation of one micromole of substrate per minute when the enzymes were kept in particular concentration of the substrate at constant temperature and pH.

Results and Discussions

It is evident from the table that both for AST and ALT, the activity was more in Black Bengal bucks in comparison to Jamnapari and Barbari bucks (Table 1). At refrigerator temperature (4° to 8°C), the sperms are exposed to cold shock and the leakage of transaminases consequent upon membrane damage was evident. Degree of damage reflected through higher and lower values of transaminases in the seminal plasma/diluting fluid is ostensibly due to inherent quality of the sperms to withstand cooling. It is known that when mammalian sperms are cooled to 5°C , there occurs leakage of intracellular enzymes, potassium, lipoprotein, ATP and

other materials from the cells. The precise mechanism of cellular damage is unknown, but presumably changes occur at unequal rates on the surface and internal portions of spermatozoa during cooling and both physical and chemical damages result (Salisbury *et al.*, 1978). Leakage of transaminases in semen samples may be attributed to the effects of cooling and glycerol addition in the dilutor. Crabo *et al* (1971) observed that addition of glycerol to cooled diluted samples of boar semen significantly increased the level of extra cellular GOT and also relatively higher values of GOT were found in glycerolated semen samples in comparison to fresh samples by Pandit and Garg (1983). The effect of breed on the leakage of transaminases as was evident in this study may have been due to inherent differences in the structure of the plasma membrane of the sperms of different breeds. Appreciable difference in regard to AST release was noted between the sperms of Red Dane bulls (529.73 units) and crossbred bulls

(764.30 units), when they were subjected to cold shock (Bhatt and Chauhan, 1984).

The trend of leakage of LDH was similar to that of transaminases. Results of the study are comparable to that of Rao and Pandey (1977) who found that LDH activity was higher in the semen of Nali rams than than of Corriedale rams. Activity of LDH has been reported to vary with species and seasons as it has shown seasonal trend with increased activity during winter and spring (Singh and Sadhu, 1972; Dhanda and Razdan, 1983). Results of the study indicated that there are variations in spermatozoal sensitivity to chilling, leading to difference in the degree of damage of their plasmalemma.

Acknowledgements

The authors extend their most sincere thanks to Dr. H.R. Mishra, Dean and Principal, Ranchi Veterinary College, Ranchi for all the help and for providing necessary facilities for the study.

REFERENCES

- Bhatt, V.K. and Chauhan, R.A.S. (1984). Studies on certain biochemical aspects of semen of Red Dane and Crossbred bulls. *Indian Vet. J.* **61**: 434.
- Cabaud, Wroblewski and Ruggiero (1958). *Am. J. Clin. Pathol.* **30**: 234, Cited by Oser, B.L. (1965).
- Crabo, B.G.; Bower, R.E.; Brown, K.I.; Graham, E.F. and Pace, M.M. (1971). Extracellular glutamic oxaloacetic transaminase as a measure of membrane injury in spermatozoa during treatment. *Anim. Breed. Abstr.* **40** : 41.
- Dhanda, O.P. and Razdan, M.N. (1983). Dehydrogenase activity of buffalo spermatozoa in relationship to ambient temperature changes during various seasons *Trop. Vet. Anim. Sci. Res.* **1** : 237-239.
- Flipse, R.J. (1960). Metabolism of bovine semen. IX-Glutamic oxaloacetic and glutamic pyruvic transminase activity. *J. Dairy Sci.* **43**: 773-776.
- Jani, V.R.; Prabhu, G.A.; D-rashri, H.J.; Patel, M.M. and Kodagali, S.B. (1983). Release of transaminase from buffalo spermatozoa during deep freezing. *Indian J. Anim. Reprod.* **4**: 30-32.
- Oser, B.L. (1965). In *Hawk's Physiological chemistry*, 14th Ed. Tata McGraw Hill Publishing Co. Ltd., New Delhi.
- Pandit, R.K. and Garg, U.K. (1983). Freezability, GOT release and fertility of crossbred bull semen. *Indian J. Anim. Reprod.* **4**: 36-38.
- Rao, B.R. and Pandey, J.N. (1977). Studies on dehydrogenase activities of ram semen during different seasons. *Indian J. Anim. Sci.* **47**: 469-472.

- Reitman, S. and Frankel, S. (1957). A colorimetric method for the determination of serum glutamic oxaloacetic transaminase and serum glutamic pyruvic transaminase. *Amer. J. Clin. Path.* 28: 56-63.
- Salisbury, G.W.; Vandenmark, N.L. and Lodge, J.R. (1978). *Physiology of Reproduction and Artificial Insemination of Cattle*. 11nd Ed. W.H. Freeman and Company San Francisco.
- Singh, B. and Sadhu, D.P. (1972). Activities of lactic and succinic dehydrogenases of cattle and buffalo spermatozoa. *Indian J. Dairy Sci.* 25: 91-96.
- Tomar, N.S. (1976). *Artificial insemination and reproduction of cattle and buffaloes*. 2nd Ed. Saroj Prakashan, Allahabad.

Table 1: Mean values of AST, ALT and LDH activities in chilled semen samples in bucks of different breeds

Breed	No. of observations	AST units/ 10 ⁹ sperms	ALT units/ 10 ⁹ sperms	LDH units/ 10 ⁹ sperms
Black Bengal	36	39.39 ^a ± 1.57	26.81 ^a ± 1.95	279.4 ^a ± 3.57
Jamnapari	48	27.96 ^b ± 1.24	17.10 ^b ± 0.87	253.69 ^b ± 3.43
Barbari	48	28.63 ^b ± 1.35	18.54 ^b ± 1.05	251.71 ^b ± 2.73

Analysis of variance

(i) <i>Between breeds</i>			
Degrees of freedom	2	2	2
Mean squares	1617.46**	1081.07**	9411.94**
(ii) <i>Within breeds</i>			
Degrees of freedom	129	129	129
Mean squares	83.05	69.33	447.79

Values bearing same superscripts in a column do not differ significantly.

**Significant at P<0.01

IJAR 9:2:134-135 : 1988

Additives In Extender And Their Influence On The Quality Of Buck Semen

S.S. PRASAD, B.K. SINGH and M.P. SINGH

Department of Gynaecology, Ranchi Veterinary College, Ranchi-834007

For successful preservation of semen the selection of good dilutor as well as additive is essential. Almquist and Zaugg (1974)

reported that Penicillin plus Neomycin were satisfactory substitute for Penicillin plus streptomycin. Addition of tranquilizer in

semen dilutor has been advocated for prolonged survival of spermatozoa during storage (Fukuhara and Nishikawa, 1973) but it is not commonly used for that purpose. If consistently good results are obtained with the addition of tranquilizer in semen dilutor, it would become of more practical value in routine preservation of buck semen. The present work was therefore, conducted to estimate the effects of adding transquilizer in the dilutor on the keeping quality of buck semen.

Materials and Methods

Twelve breeding bucks (4 each of Barbari, Saanen and Jamnapari- Barbari crosses) between the age group of 2 and 3 years were used. One collection per buck per week in the morning hours was normally taken. Immediately after collection, semen samples were evaluated and then diluted in egg-yolk-citrate dilutor. Each diluted semen sample was divided into three equal parts. In one part, Benzyl Penicillin and Neomycin (Neomycin sulphate capsule, Unichem Laboratories) were added at the rate of 1000 I.U. and 1000 microgram per ml of diluted semen. In the second part, Penicillin, Neomycin and Largactil were added at the rate of 1000 I.U. 1000 microgram and 0.4 mg per ml of diluted semen. The third part was kept as control where no additive was added. All the semen samples were kept in the refrigerator and

examined at intervals of 24 hours upto 96 hours of preservation for live sperm and motility percentage.

Results and Discussions

The semen samples of all the breeds showed the highest live sperm and motility percentage of all hours of preservation where the dilutor contained tranquilizer along with antibiotics. In this dilutor group, the overall live sperm percentage of pooled data at 24, 48, 72 and 96 hours of preservation was 83.74, 70.36, 53.28 and 39.83 respectively. The corresponding values for sperm motility were 70.15, 56.87, 45.87 and 32.32 percent respectively. The control group showed the lowest live sperm and motility percentage.

When the data for all the hours and breeds were pooled, it was seen that the live sperm percentage (61.80) and motility percentage (51.33) were highest in the tranquilizer group. The corresponding values were 58.95 and 49.43 in the antibiotic group, whereas these values were 55.83 and 43.05 in the control group. Critical difference test also showed significant superiority of the tranquilizer group. This finding is in agreement with Maan (1958) and Fukuhara and Nishikawa (1973).

Acknowledgement

The authors acknowledge the Dean, College of Veterinary Science, Ranchi for providing necessary facilities.

REFERENCES

- Almquist, J.O. and Zaugg, N.L. (1974). Fertility of bovine semen in milk diluent containing combinations of Penicillin-neomycin and linomycin-spectinomycin. *J. Dairy Sci.* **57**: 1211-1213.
- Mann, T. (1958). *Proc. Soc. Study of Fertility.* **9**: 3.
- Fukuhara, R. and Nishikawa, Y. (1973). Effect of chlorpromazine hydrochloride on the viability and metabolism of goat and bull spermatozoa. *Japanese J. of Zootechnical Sci.* **44**: 338-342.

Cryoprotective Effect Of Different Concentrations Of Dimethyl Sulfoxide (DMSO) And Glycerol On Freezing Of Goat Embryos

INDRA MANI¹ and S.V. VADNERE

Department of Gynaecology and Obstetrics, College of Veterinary Sciences, G.B. Pant Univ. of Agri. and Tech. Pantnagar-263145 (U.P.)

ABSTRACT

Two concentrations each of DMSO (1.5M & 2.0M) and glycerol (1.0M and 1.33M) were tried to ascertain their cryoprotective effects on freezing of goat embryos. 2.0M DMSO and 1.0M glycerol gave better cryoprotection (75% and 100% respectively) to embryos than 1.5M DMSO (25%) and 1.33M (50%) glycerol.

* * *

With the success of embryo transfer (ET) technique in farm animals it has become imperative to longterm preserve the embryos for later use when the recipients are ready for transfer of embryos. By freezing and storage of embryos, the need to keep ready, large number of recipients, estrus synchronised for transfer of embryos will become unnecessary. By freezing the embryos, the genetic material could be preserved for future use, it could be transported for long distances between different countries and conservation of rare breeds and animals of superior germ plasm could be possible.

Short term preservation of embryos below 0°C was successfully done by Ferdows *et al* (1958) and Bilton and Moore (1976). Willadsen *et al* (1974) used 1.5M DMSO. Bilton and Moore (1976) used 2.0M DMSO or 1.0M glycerol and Ahmad and Maurya (1981) used 2.0M DMSO for freezing goat embryos with varying degree of success.

Materials and Methods

Twenty four embryos were collected by laparotomy of superovulated goats 72, 96 and 120 hours after the onset of oestrus by flushing fallopian tubes and uterine horns with the flushing medium TC-199 Medium, Hank's base (Centron Research Labs., Bombay) enriched with 20% goat serum with pH 7.2 at 37°C temperature. Dimethyl Sulphoxide (DMSO) and glycerol in two concentrations each 1.5M and 2.0M and 1.0M and 1.33M respectively were used as cryoprotectants.

The embryos (4 each for every treatment and control) were loaded in Cassou ministraws containing 0.24 ml modified Dulbecco's phosphate buffer saline enriched with 20% goat serum. The straws for DMSO treatment and the freezing medium were cooled to 5°C and the freezing medium was added to the straws over a period of 15 minutes in four equal fractions of 0.06 ml. The straws for glycerol concentrations were held at 37°C and the freezing medium containing 0.0, 2.0 and 2.60 M glycerol was added at 37°C in four equal fractions over a period of 15 minutes. The straws were sealed, cooled to 5°C, stored for 20 and 40 minutes respectively at 5°C and then frozen on liquid nitrogen vapours thus: Straws were cooled to -80°C at the rate 1.0°C/min. from 0°C to -50°C and 2°C/min. from -50°C to -80°C and then transferred to liquid nitrogen container and

stored for 3 days. The thawing of straws was done slowly, the embryos were washed thrice in a fresh culture medium and were cultured at 37°C for 24 hours. The control straws contained no cryoprotectant in the freezing medium.

Results and Discussions

Out of 16 embryos frozen (8 each, in DMSO and glycerol), 25% and 50% survived in 1.5M DMSO and 1.33M glycerol respectively. Out of the 8 embryos without any cryoprotectant, none survived.

DMSO concentration of 2.0M exerted better cryoprotective effect (75%) than 1.5M DMSO (25%) on the freezing of goat embryos, which was in agreement with the findings of Whittingham (1974), Whittingham and Adams (1974), Bilton and Moore (1976) and Willadsen *et al* (1976). The overall cryoprotective effect of glycerol was better than the overall effect of DMSO (41.00% vs 33.33%) for freezing of goat embryos.

1.0M glycerol gave better cryoprotection, than 1.33M glycerol (100% vs 50%), which was similar to the findings of Bilton and Moore (1976) who observed that 1.0M glycerol was the best concentration for freezing goat embryos. However, the findings of Whittingham *et al* (1972) who observed that 1.0 to 1.5M DMSO gave better cryoprotection than 1.0M glycerol, were at variance with the present findings.

The survival rate of embryos in the present investigation was much higher than the survival rate of only 20% obtained by Bilton and Moore (1976) and of 60% obtained by Ahmed and Maurya (1981).

It is therefore concluded that DMSO (2.0M) and glycerol (1.0M) could be used as cryoprotectants for freezing goat embryos in modified Dulbecco's phosphate buffer medium enriched with 20% goat serum and at pH 7.2 with better success.

REFERENCES

- Ahmed, T.; Maurya, S.N. (1981). Multiple ovulation in and recovery of eggs from goats. *Acta Veterinaria Academiae Scientiarum Hungaricae*, **29**(4): 471-475 (Fide: Anim. Breed. Abst. (1983) **53**: 681).
- Bilton, R.J.; Moore, N.W. (1976) *In vitro* culture, storage and transfer of goat embryos. *Aust. J. Biol. Sci.* **29**: 125-129.
- Ferdows, M.; Moore, C.L.; Dracy, A.E. (1958). Survival of rabbit ova stored at -79°C. *J. Dairy Sci.* **41**: 739.
- Whittingham, D.G. (1974). The viability of frozen thawed mouse blastocysts. *J. Reprod. Fert.* **37**: 159-162.
- Whittingham, D.G. and Adams, C.E. (1976). Low temperature preservation of rabbit embryos. *J. Reprod. Fert.* **47**: 269-274.
- Whittingham, D.G.; Leibo, S.B. and Mazur, P. (1972). Survival of mouse embryos frozen to -196°C and -269°C. *Science, U.S.A.* **178**(4059): 411-414.
- Willadsen, S.; Polge, C.; Rowson, L.E.A. and Moore, R.M. (1974). Preservation of sheep embryos in liquid nitrogen. *Cryobiology* **11**: 560.
- Willadsen, E.; Polge, C.; Rowson, L.E.A. and Moore, R.M. (1976). Deep freezing of sheep embryos. *J. Reprod. Fert.* **46**: 151-154.

SHORT COMMUNICATIONS

IJAR 9:2:138 : 1988

Spermine: Absence In Goat Semen

N.M. MARKANDEYA, D.R. PARGAONKAR and S.A. BAKSHI

Department of Gynaecology and Obstetrics, College of Veterinary and Animal Sciences, MAU, Parbhani-431402

Spermine is present in human semen, which acts as a bacteriostat and imparts Musk odour to human semen. Presence of spermine is used as a test in Medico-legal cases (White, 1968). Efforts were made to screen the semen of different breeds of buck for confirmation of presence or absence of spermine in their semen.

Materials and Methods

From the available lot of mature males from 'Goat Unit', College of Agriculture, M.A.U., Parbhani, semen was collected. Study included total fifteen bucks of different breeds viz., Osmanabadi, Saanen, Beetal and Jamanapari pure bred bucks with cross bred bucks of Saanen × Osmanabadi, Beetal ×

Osmanabadi and Alpine × Osmanabadi breeds.

Barberio's crystallographic analysis as cited by Mann (1974) was used to detect Spermine content in semen. Six semen samples of each buck were analysed.

Results and Discussions

Presence of Spermine in semen forms three sided crystals terminated at either ends in a point. Present study indicated absence of Spermine in all purebred and crossbred bucks.

Present findings are in agreement with Patil (1970), who analysed Malabari buck semen and reported absence of Spermine.

REFERENCES

- Mann, T. (1960). Biochemistry of semen and of the male reproductive tract. Pub. London: Methuen and Co. Ltd., New York: John Wiley & Sons.
Patil, R.V. (1970). M.V.Sc. Thesis submitted to University of Calicut, Kerala Veterinary College, Mannuthy, Trichur.
White, I.G. (1958). Biochemical aspects of mammalian semen. Anim. Breed Abst. 26(2): 109-123.

IJAR 9:2:138-139 : 1988

A Field Method For Collection Of Bovine Cervical Mucus For Microbiological Studies

Y.P.S. DABAS and S.N. MAURYA

College of Veterinary Sciences, G.B. Pant Univ. of Agri. and Tech. Pantnagar-263145 (U.P.)

This report describes a simple technique for the collection of cervical mucus for

microbial culture under field conditions. The equipments needed include an A.I. gun with

its sheath, a rubber connector and a 20 ml glass syringe. Insemination gun and rubber connector are wrapped in paper, sterilized in autoclave and stored in incubator till use. A small opening (1 cm long) is made at the back end of the factory sterilized polyethylene bag containing the sheaths to allow the withdrawal of one sheath at a time after insertion of A.I. gun full length through it. The polyethylene bag is kept in a clean and dry tray covered with a clean and dry towel.

The cow to be sampled is restrained properly in a chute. Taking routine sterile precautions, the vulvar lips are spread by an assistant while the insemination gun and assembled sheath are passed through the vagina. Rectally, the cervix and insemination gun are manipulated until the tip of the sheath is at the point from where the mucus is to be collected. The insemination gun is withdrawn leaving the sheath in situ (in the cervix/uterus) and stored in its box until next use. The glass syringe is attached to the back end of the

sheath with the help of the rubber connector and mucus is drawn into the sheath by retracting the syringe plunger. Since cervical mucus is quite viscid, the syringe plunger is slowly and maximally retracted and held in position for about 20 sec. The syringe is then folded back over the sheath at the rubber connector and the sheath is withdrawn. Immediately, both ends of the sheath are heat sealed using a cigarette lighter or match box. The sheath is whipped clean with a cotton swab soaked in 70% alcohol, kept in a polyethylene bag containing identification of the animal and is stored in refrigerator or crushed ice till its transport to the diagnostic laboratory. After receipt in the laboratory, the exterior of the sheath is again cleaned with 70% alcohol and dried. The sheath is cut aseptically near the centre of the column of mucus, under flame, and mucus is retrieved with a platinum loop. The technique is effective for collection of cervical/uterine mucus without contamination.

IJAR 9:2:139-141 : 1988

Post Partum Changes Of The Uterus And Ovaries In Relation To Uterine Microflora In Cows

K.C. DEKA, K.C. NATH and C.K. RAJKONWAR

Department of Gynaecology, Obstetrics and Artificial Insemination, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati-781022.

Infection of the uterus during early postpartum days affects uterine repairment soon after calving and delays uterine involution (Elliot *et al.*, 1968 and Gier and Marin, 1968) especially when the infection overcomes the resistance of the cow. Studies reported herein elucidate the effect of the uterine microflora on the postpartum changes of uterus and ovaries in cows.

Materials and Methods

Digital palpation of the uterus and the ovaries P.R. in 30 cows (Jersey, Red Dane and their crosses) of the University farm was carried out at 4 day intervals commencing from fourth day of calving till complete involution of the uterus. Diameter of the uterine horns and presence or absence of palpable follicle or corpus luteum in the

ovaries were recorded at each examination. Uterine discharge from each cow was collected as per Radoslavov (1975) at weekly intervals and analysed for microbial contaminants as described by Deka *et al* (1985). The cows were classified as microflora positive when either pathogenic or non-pathogenic microorganisms could be isolated from the uterus upto second week postpartum and microflora free when no organism could be isolated from the uterus. On each day of examination, the lochia was observed for quantity, colour and consistency as per Roberts (1971). Time of involution of the cervix and uterus, time of regression of pregnancy CL, time of development of palpable follicle and postpartum oestrous interval were recorded as per methods of Morrow *et al* (1966).

Results and Discussions

The quantity of lochia was recorded as free flowing in 66% cows on 4 days, scanty in 55-85% cows from 8 to 24 days and absent in 60-100% cows from 28 days parturition onwards in microflora-free cows. In the microflora positive group, it was medium in 40-60% cows upto 12 days and scanty in 50-70% cows from 16 to 40 days. In both the groups, lochial colour was reddish in 50-70% cows upto 16 days and clear in 43.75-100% cows thereafter. Consistency of lochia was recorded as watery in 60-70% cows on first 4 days and thick in 60-100% cows thereafter, in both the groups.

Time of involution of the cervix was 22.60 ± 1.56 and 28.40 ± 1.83 days in microflora free and positive group respectively. The cervix

was found to involute rapidly from 4 to 16 days in both groups.

Time of involution of gravid and nongravid horn was 25.60 ± 0.24 and 23.00 ± 1.26 days respectively in microflora free cows; while respective microflora positive cows were 29.60 ± 1.22 and 26.00 ± 1.29 days respectively. Uterine infection significantly ($P < 0.01$) delayed involution of the cervix. The delay in the involution of gravid and nongravid horn in microflora positive cows was also significant ($P < 0.05$) and highly significant respectively. This observation was similar to that observed by Elliot *et al* (1968) and Gier and Morion (1968) who reported that uterine infection acquired in the early part of puerperium affected uterine repairment and ultimately delayed return of the uterus to normal size. Arthur (1977) also observed association of nonspecific genital infections with delayed uterine involution. The rate of involution of uterus was rapid upto 16th day post partum in both the groups.

Time of regression of pregnancy C.L., time of development of palpable follicle and postpartum oestrous interval were 10.45, 12.00 and 59.60 days in microflora free and 10.00, 12.50 and 53.00 days in microflora positive group respectively. No significant difference between the two groups was observed in the time of regression of pregnancy C.L. and time of development of palpable follicle. On the other hand, postpartum oestrous interval was significantly greater ($P < 0.05$) in microflora free cows. Probably, the uterine infections were not severe enough to affect ovarian activities in the cows studied.

REFERENCES

- Arthur, G.H. (1977). *Veterinary Reproduction and Obstetrics*, 4th Ed. The English Language Book Society and Bailliere Tindal, London.
- Deka, K.C.; Rajkonwar, C.K.; Nath, K.C. and Bora, B.R. (1985). Studies on the uterine microflora of puerperal cows in normal and abnormal parturition. *Indian Vet. J.* 62: 265-267.
- Elliot, L.; Mc. Mohan, K.J.; Gier, H.T. and Marion, G.E. (1968). Uterus of the cow after parturition, Bacterial content. *Am. J. Vet. Res.* 29: 77-81.
- Gier, H.T. and Marion, G.B. (1968). Uterus of the cow after parturition. Involutional changes. *Am. J. Vet. Res.* 29: 83-96.
- Morrow, D.A.; Roberts, S.J.; McEntee, K. and Gray, H.G. (1966). Post partum ovarian activity and uterine involution in dairy cattle. *J.A.V.M.A.* 149: 1596-1609.
- Radoslavov, V. (1975). Quantitative changes in the uterine microflora during puerperium and at the time of conception in cows. *Veterinarnomeditsinski Neuki*, 12: 49-54 (*Vet. Bull.* 46: 1561).
- Roberts, S.J. (1971). *Veterinary Obstetrics and Genital Diseases*, 2nd Edn. Scientific Book Agency, Calcutta.

IJAR 9:2:141-142 : 1988

Incidence Of Mycotoxic Abortions In A Dairy Herd

B. RAMESHKUMAR, R.C. RAJASUNDARAM and A.C. SUBRAMANIAN

Livestock Research and Development Centre, Erode-638003 (Tamilnadu State)

ABSTRACT

Incidence of Mycotoxic abortions in a dairy herd is recorded.

* * *

Incidence of Mycotoxic abortions in bovines is not uncommon. Roberts (1971) recorded upto 10 percent abortions in a herd of cattle. In the present instance, incidence of five abortions over a period of three months due to Mycotoxic infection was recorded from a dairy unit in Periyar district of Tamil Nadu.

Case History and Observations

A well established dairy unit comprising of about 40 milch animals reported a series of abortions in 4 Buffaloes and one cow over a period of three months after rainy season. Abortions occurred during the last trimester of pregnancy in all the cases. The fetuses expelled were dead with mild degree of

autolysis. On post-mortem examination of the same, straw coloured serous fluid was present in their body cavity. No external skin lesions were seen.

Retained placenta which followed abortions, showed extensive superficial necrosis of the cotyledons. They were swollen, edematous and incarcerated. The necrotic areas showed a dull grey centre surrounded with haemorrhage. The cotyledons were firmly attached with the leathery chorion.

Serum samples collected from the affected animals did not reveal any antibody titre from common bacterial diseases such as Brucellosis and Leptospirosis. Vaginal Mucus test also did not reveal any titre against Vibriosis. Feed samples including the paddy straw and sorgum fodder fed to the animals were analysed for the presence of Mycotoxin. Aflatoxin B₁ produced by *Aspergillus* spp.

was identified at the levels of 22-56.5 mgms/kg of fodder by quantitative analysis.

Recovery of the affected animals was slow and prolonged and they showed signs of infertility and permanent sterility. Hence they were culled by the owner.

Discussion

The incidence of abortions occurred soon after a wet season as reported by Hugh-Jones and Austwick (1967) and in the present case incidence was as high as 10 percent of the herd. Hillman (1969) reported appearance of ringworm like skin patches externally in only

30 percent of Mycotic abortions with other fetuses normal. In the present case, no external skin lesions were seen. The lesions on the cotyledens were similar to those observed by Roberts (1971). The affected animals showed a prolonged recovery period and became sterile. Under these circumstances they were sold as sterile ones.

The absence of any other identifiable pathogen, the spate of sporadic abortions, the high levels of Mycotoxin in the incriminated fodder with highly suggestive pathological lesions on the placentomes, pointed to Mycotoxic abortion.

REFERENCES

- Hugh-Jones, M.E. and Austwick, P.K.C. (1967). Epidemiological studies in Bovine Mycotic Abortion I. The effect of climate to incidence. *Vet. Rec.* 81(12): 273.
Hillman, R.B. (1969). Bovine Mycotic placentitis in New York State. *Cornell. Vet.* 59(2): 269.
Roberts, S.J. (1971). *Veterinary obstetrics and Genital diseases (Therigenology)* Second Edn. CBS Publishers and Distributors, India.

IJAR 9:2:142-143

1988

A Genital Abnormality In Vaginal Passage In A Bannur Ewe

S.R. CHINCHKAR, D.P. VELHANKAR, B.R. DESHPANDE and V.L. DEOPURKAR

Department of Animal Reproduction, Bombay Veterinary College, Parel, Bombay-400012

Genital abnormality when present and potentially capable of interfering with reproductive functions, can be utilized as a basis for diagnostic, prognostic and therapeutic considerations.

While carrying out biometry of genital organs in Bannur ewes, one genital organ presented clinically detectable abnormality of vertical band stretching from roof of vagina to the floor of vagina. This vertical band was 0.9 cm in length and 0.5 cm in width which divided the vaginal passage into two channels.

It was otherwise normal for all practical purposes (Fig. 1).

Infertility or sterility in sheep and goats is much uncommon (Roberts, 1971). This is probably for two reasons; firstly, there has been little study on this topic and secondly the incidence of diseases and infertility is apparently low.

The abnormality as observed is likely to result in dystocia but may not interfere with successful conception. In the absence of any authentic data for its heritability, this could be

considered as individual developmental abnormality and when present can be successfully overcome by surgical intervention, if causing obstruction to the passage of foetus.

Fig. 1.

Presence of vertical band in vaginal passage from roof to vaginal floor in Bannur ewe.



REFERENCE

Roberts, S.J. (1971). Veterinary Obstetrics and genital diseases (Theriogenology) 2nd Edn. Scientific Book Agency, Calcutta-I

IJAR 9:2:143-144 : 1988

Agenesis Of Mullerian Duct System In A Gilt

P.K. SRIRAMAN¹, S.V. RAMACHANDRAIAH², K. SUBRAMANYAM NAIDU³ and K. KRISHNA REDDY⁴

College of Veterinary Science, A.P. Agricultural University, Tirupati-517502.

Segmental defects of the mullerian duct system are quite common in swine. In most of the cases, there is an arrested development of a portion of mullerian duct system and their different derivatives that produce the cranial

part of the vagina, cervix and uterus (Jubb and Kennedy, 1970).

The present report describes an unusual case of agenesis of mullerian duct system.

¹Veterinary Officer, AICRP on Pigs (ICAR)

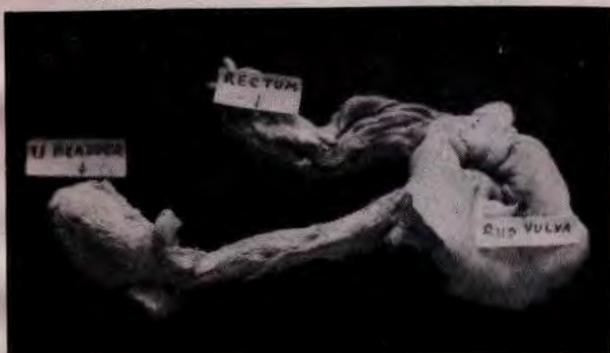
²Associate Professor of Animal Reproduction

³Assistant Professor of Animal Reproduction

⁴Scientist, AICRP on Pigs (ICAR)

Case History

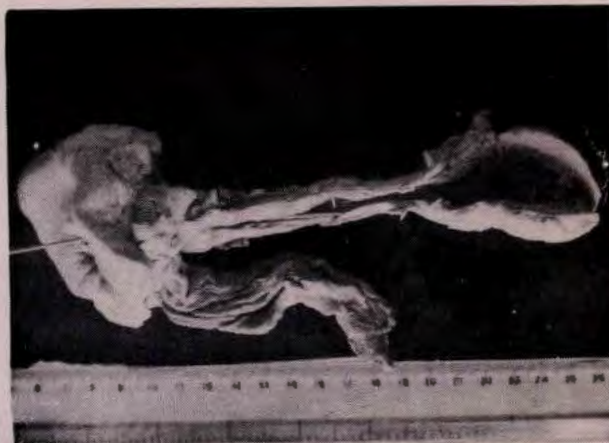
A crossbred gilt (Large white Yorkshire X Native), which was found to have a rudimentary vulva and not coming to heat, was slaughtered, uro-genitals removed and examined for any evidence of abnormality. After detailed study, it was found that the genital tract including the ovaries was missing, excepting the presence of rudimentary vulva (Fig. 1). Clitoris was also absent.



The urethra from the bladder opened straight into the vestibule. There was neither testicles nor ovaries, fallopian tubes, uterus, cervix and vagina. On exploration with a probe, it led directly into the bladder (Fig. 2) thereby indicating the agenesis of the mullerian duct system.

Discussion

Teige (1957) and Reed (1970) reported aplasia of mullerian duct in pigs. Reed (1970)



observed the presence of two testicles and Wolffian ducts in place of normal cervix, uterus, fallopian tube and ovary but in the present case, none of the segments could be noticed.

Most of the piglets born along with the present gilt, showed bodily defects like microphthalmia, anophthalmia, hydrocephalus, polycystic kidney etc. The sows had received high doses of antibiotics parenterally and aflatoxin in feed during early and mid gestation. Perhaps, this might have been the cause for the developmental defects.

Acknowledgements

Thanks are due to Indian Council of Agriculture Research, New Delhi for the facilities provided.

REFERENCES

- Jubb, K.V.F. and Kennedy, P.C. (1970). Pathology of domestic animals. 3rd Edn. Academic Press, New York, Pp. 492-494.
Reed, H.C.B. (1970). Incidence of breeding abnormalities in a random selection of large white gilts purchased for experimental purposes, 1970. Vet. Rec. 87: 778-781.
Teige, J. (1957). Congenital malformations of the mullerian duct and sinus urogenitalis in pigs. Nord. Vet. Med. 9: 609-629.

Pregnancy Duration In Jersey X Kankrej Crosses

A.M. PATEL

Livestock Research Station, College of Veterinary Science and Animal Husbandry, Anand-388001.

ABSTRACT

The present study comprised 315 normal calvings of Kankrej and Jersey X Kankrej F_1 cows. The average gestation period for Jersey X Kankrej F_1 and F_2 calves was 280.03 ± 0.45 and 281.89 ± 0.51 days respectively. Generation, weight of calf and sire had significant effect while sex of the calf, age of cow and season of calving had nonsignificant contribution towards the variation in pregnancy duration. The heritability value estimated to be 0.32 ($P < 0.05$).

A wide variation exists for duration of pregnancy, both in indigenous as well as exotic breeds of cattle (Mishra and Mishra, 1987). Thus, the knowledge of pregnancy duration of crossbreds helps in fixing the expected date of calving more exactly. The vigilant supervision at the time of parturition improves the health status and survivability of newborn and her dam.

Materials and Methods

Data on 315 normal single calvings of Kankrej and Jersey X Kankrej F_1 cross, recorded at Livestock Research Station, Veterinary College, Anand (1979 to 1986) were analysed to ascertain the gestation period and factors affecting it. The animals were maintained under isomanagerial condition. The year was divided into three seasons viz., summer (February to May); monsoon (June to September) and winter (October to January) seasons.

Results and Discussions

The average pregnancy duration in JK crosses was 281.03 ± 0.46 days, with a range from 276 to 298 days. The value is more towards the indigenous Kankrej dam (287.3 ± 9.9 days) than the pure Jersey (sire) breed (278.7 ± 10.5 days). This indicates a significant contribution of dam breed (Kankrej) for lengthening of the gestation period and in turn higher birth weight (22.76 ± 0.37 kg) and survival rate (95%) of crossbred calves (Anon., 1985). The pregnancy duration observed in this study is in agreement with the reports of Rao *et al.* (1984) for Jersey X Ongole crosses and Mishra and Mishra (1987) for Jersey X Red Sindhi and Jersey X Haryana crosses.

1. *Generation effect*: Jersey X Kankrej halfbreds (JKF_1) and *inter se* mated (JKF_2) calves carried in utero for 280.03 ± 0.45 and 281.89 ± 0.50 days respectively. The difference was statistically significant. The JKF_2 calves though carried for longer duration had significantly lower birth weight (21.47 ± 0.63 kg) than their JKF_1 counterparts (22.65 ± 0.38 kg). This is in agreement with the reports of slower prenatal growth (Tegegn *et al.*, 1981), and loss of heterosis (10.23%) for birth weight in *inter se* off-springs.

2. *Sex of the calf*: The frequency of male (170) to female (145) births was 53.95:46.05, showed a nonsignificant variation. Male foetuses were carried for longer time in utero

(281.64 ± 0.50) than their female counterparts (280.31 ± 0.46) in both the generations. However, the difference was significant in *inter se* mated JKF₂ calves only. The results are supported by the reports of Chandramohan and Bhat (1981). The incidence of prolonged gestation period (Busch *et al.*, 1985) was 8.25% of which, JKF₂ males contributed 68.38%, followed by JKF₂ female (19.29%), JKF₁ male (7.69%) and JKF₁ female (7.69%) calves.

3. *Birth weight of calf*: Irrespective of sex and generation, heavier calf was carried for longer time in utero. The correlation coefficient between birth weight and gestation period for JKF₁ male, JKF₁ female, JKF₂ male and JKF₂ female calves was 0.41, 0.56, 0.31 and 0.66 respectively. All values were statistically significant ($P < 0.01$).

4. *Heifers vs Cows*: Heifers carried (280.81 ± 0.86) the fetus for a little less period compared to cows. The difference was statistically nonsignificant. The correlation coefficient between age at first calving and gestation period was only +0.15. Das *et al* (1984) and Morales *et al* (1984) also did not observe any effect of parity or age on duration of pregnancy.

5. *Season of calving*: The maximum number of calvings were observed in summer (119) followed by winter (103) and monsoon (93). The variation due to seasons was nonsignificant and showed practically even distribution of calvings in different seasons.

Cows calved during summer season (281.62 ± 0.00) had maximum gestation period, followed by winter calvers and monsoon calvers. However, the difference was nonsignificant. Nonsignificant seasonal influence was also reported by Chandramohan and Bhat (1981) and Morales *et al* (1984).

6. *Paternal effect*: Paternal effect was observed to be significant. This study showed that, the calves of prepotent bulls had longer gestation period and in turn had higher birth weight (Newman *et al.*, 1985). The heritability estimate of pregnancy duration was 0.32 ($P < 0.05$). Durate *et al* (1985) also estimated 0.35 to 0.67 heritability values for gestation period and concluded that, it would be possible to reduce the incidence of stillbirth and dystokia by reducing the gestation period through selection of bulls.

REFERENCES

- Anonymous (1985). Annual Report, Livestock Research Station, Gujarat Veterinary College, GAU, Anand.
- Busch, W.; Gjahtow, E. and Willers, S. (1985). Prolonged gestation in cows. *Anim. Breed. Abstr.* **54**: 1564.
- Chandramohan, K.K. and Bhat, P.N. (1981). Note on factors affecting first gestation length in Sahiwal cattle. *Indian J. Anim. Sci.* **51**(8): 788-790.
- Das, S.; Baishya, N. and Rajkonwar, C.K. (1984). Studies on certain aspects of reproduction in Jersey cattle under farm conditions in Assam. *Indian Vet. J.* **61**(9): 774-780.
- Durate, W.J.; Leboute, E.M.; Martina, E.S. and Ribeiro, J.A.Q. (1985). Estimates of environmental effects and genetic parameters for gestation length, calfbirth weight and incidence of dystokia in a charolais herd. *Anim. Breed. Abstr.* **54**: 2747.
- Mishra, I. and Mishra, M. (1987). Lactation and reproduction performance of crossbred Jersey cows in humid conditions. *Indian J. Anim. Prod. & Mgmt.* **3**(2): 68-69.

- Morales, T.H.; Aguilar, C.J.A. and Hinojosa, C.S.A. (1984). Reproductive performance in a herd of Holstein Friesian in Chontatpa Tabasco. 2. Gestation period and calving interval. *Anim. Breed. Abstr.* 52: 5741.
- Newman, J.A.; Rahnefeld, G.W.; Tong, A.K.W. and Fredeen, H.T. (1985). Calving and pre-weaning performance of crossbred progeny of some foreign and domestic beef cattle breeds. *Canadian J. Anim. Sci.* 65(3): 587-593.
- Rao, K.B.; Sivaiah, K. and Rao, L.S. (1984). Some reproductive traits among three crossbred heifers. *Indian J. Anim. Sci.* 54(2): 201-203.
- Tegegn, A.; Galal, E.S.E. and Kebedo, B. (1981). A study on the reproduction of local zebu and F1 crossbred cows. *Anim. Breed. Abstr.* 53: 453.

INDIAN JOURNAL OF ANIMAL REPRODUCTION GUIDE LINES TO AUTHORS

1. The Journal is published twice a year as a Volume comprising of June and December issues.
2. Paper should be TYPE-WRITTEN and double spaced all throughout (including references and tables) on white, durable bond paper of size 22 cm x 28 cm, with a 4 cm margin at the top, bottom and left hand side. Articles including illustrations, should be sent in duplicate after a careful check-up of typographical errors.
3. Articles should not exceed Six typed pages. Short Communications/Research notes and clinical articles should be limited to two typed pages.
4. A recent issue of the Journal be consulted for the format of articles and methods of citation of references in the text as well as at the end of the article.
5. The Abstract and Introduction should be in brief. Review of Literature should be crisp and pertinent to the problem. The main emphasis in the text should be on the actual work done by the author(s). Details of Materials and Methods including experimental design and techniques used should be given. Where the methods are well known, the citation of standard work is sufficient. References should be reduced to the barest minimum.
6. Mean results with the relevant standard errors should be presented, rather than detailed data. The statistical methods used should be clearly stated. Tables should be minimum and fit in the normal layout of the page. All weights and measures should be in Metric units.
7. The Results and Discussion should be combined to avoid repetition. The Discussion should relate to the limitations or advantages of the author's actual work in comparison with that of others.
8. All articles are sent to Referees for scrutiny and author(s) should meet criticism by suitably revising the article. Block-making charges of Photographs, Graphs, Tables, Histograms and Line drawings appearing in the accepted articles shall have to be paid by the concerned author(s) in advance on receipt of the Bill from the Editor, I.J.A.R.
9. All efforts are made to acknowledge, process and accept the articles received for early publication. Authors are earnestly requested to become and enrol active paying subscribers of the Journal and help procure/book suitable advertisements for publication in the Journal to strengthen its financial resources.
10. Articles and all matters pertaining to the Journal be sent to the Editor, Indian Journal of Animal Reproduction, B-306, Ujwal Flats, Rahate Colony, Jail Road, Nagpur-440 022.

ISSAR NEWS

We are glad to inform that Dr. A. Ramamohana Rao, Dean Post Graduat Studies, APAU, Hyderabad and President, ISSAR representing India was elected to the Standing Committee of International Congress on Animal Reproduction held at Dublin, Ireland (U.K.) on 28th June 1988.

We congratulate him and wish him many more laurels in future.

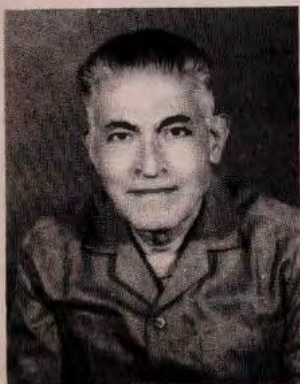
* *

*

* *

ICAR Awards of Emeritus Scientist

We are happy and proud to announce that the Indian Council of Agricultural Research, New Delhi have awarded the positions of Emeritus Scientist to :



Dr. A.S. Kaikini

Dr. A.S. Kaikini, Retd. Dean, Faculty of Veterinary Science, PKV, Head, University Department of Gynaecology & Surgery and Associate Dean, Nagpur Veterinary College, Nagpur and Editor, Indian Journal of Animal Reproduction w.e.f. 11-10-88. His research project is titled "Gynaeco-Clinical Studies in Sahiwal and Cross-bred cows".

Dr. S.B. Kodagali, Retd. Head, Department of Gynaecology & Obstetrics, GAU, Gujarat Veterinary College, Anand and Ex-Editor, India Journal of Animal Reproduction w.e.f. 15-11-88. His research project is titled "Clinical Andrological Studies of Breeding bulls (Buffalo and Cross-bred) in Gujarat State".



Dr. S.B. Kodagali

We congratulate them and wish them many healthy, long years of sustained service with valuable contributions in Animal Reproduction. We thank the ICAR for recognising their merits for these prestigious awards.

* *

*

* *

Dr. R.C. Gupta after a successful assignment in Libya has returned home and resumed duties as Professor and Head Deptt. of Gynaecology & Obstetrics, College of Veterinary Science, HAU, Hisar.

We welcome Dr. Gupta and wish him best of luck and health.

* *

*

* *

PROMOTIONS

Dr. C.K. Rajkonwar, Head Deptt. of Gynaecology & Obstetrics, College of Veterinary Science, Khanapara, Guwahati and Vice-President ISSAR is appointed Associate Dean of the new college of Veterinary Science, AAU, Azad-Lakhimpur (Assam).

Dr. S.R. Pattabiraman, Associate Prof. of Gynaecology, MVC, Madras and Treasurer, ISSAR is appointed Professor and Head, Department of Clinics, Madras Veterinary College, Madras.

Dr. D.P. Velhankar, Director of Clinics, Aarey Campus and Ex-Secretary, ISSAR is appointed Professor of Gynaecology and Obstetrics, Bombay Veterinary College, Bombay.

Dr. S.N. Sinha is appointed University Professor and Head, Deptt. of Gynaecology and Obstetrics, Rajendra Argi. University, Bihar Veterinary College, Patna.

Dr. M.S. Kadu is appointed Professor of Animal Reproduction and Offg. Head, Deptt. of Gynaecology and Surgery, PKV, Akola.

Dr. B. Munilal Dubay is appointed Professor and Head, Deptt. of Gynaecology and Obstetrics, UAS Veterinary College, Hebbal, Bangalore.

We congratulate them and wish them all a grand success.

, * *

*

* *

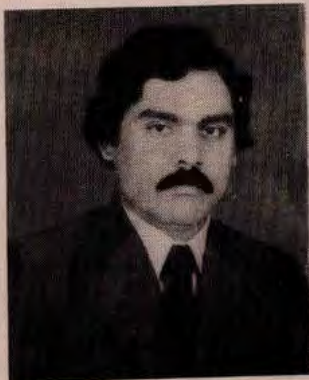
ISSAR Awards

We are glad to congratulate the following ISSAR Awardees :

(1) Nils Lagerlof Memorial Award

1986

Dr. C. Chandrahasan,
Assistant Prof. of Anatomy;
Dr. S.R. Pattabiraman,
Professor & Head, Deptt. of
Clinics, Treasurer, ISSAR
and Dr. V. Venkataswami,
Retd. Professor of Gynaecology,
Madras Veterinary college, Madras.



Dr. C. Chandrahasan



Dr. S.R. Pattabiraman



Dr. Shyam Zawar

1987 : : Dr. Shyam Zawar, Manager, Raymonds Embryo Research Centre, Gopalnagar, Bilaspur (M.P.).



Dr. A.J. Dhami

(2) Dr. G.B. Singh Memorial Young Scientist Award, 1987.

Dr. A.J. Dhami, Senior Research Assistant (Gynaecology) and Treasurer, ISSAR Gujarat Chapter, Gujarat Veterinary College, Anand; Ph.D. Scholar (ICAR), IVRI, Izatnagar. (U.P.).

(3) ISSAR Fellowship Awards :

Dr. C.P.N. Iyer, Head, Deptt. of Gynaecology & Obstetrics, College of Veterinary and Animal Sciences, Mannuthy, Trichur (Kerala).

Dr. H.C. Pant, Head Deptt. of Gynaecology & Obstetrics, College of Veterinary Science & Animal Husbandry, Mathura (UP).



Dr. S.K. Verma

Dr. S.K. Verma, Associate Professor of Gynaecology College of Veterinary Science, HAU, Hisar (Haryana).

We wish them all a brighter future.

From Secretary's Desk

Dear Members,

Through my appeals/circular letters, I have approached you and your response in the matters was also worth appreciating. On the eve of VII National Congress on Animal Reproduction, 3rd Executive Committee Meeting was held at Trichur on 18th August, 1988. There I submitted a brief report of the work done during the past few months. Summary of the same is as under:

1. As per the resolution adopted at the 1st Executive Council Meeting held at Hyderabad on in May 1987, all office bearers presumed to have taken over the charge and started functioning. However, the functioning could not be geared-up for want of important papers, files, records, etc. The ex-body took more than nine months to hand over the record. This resulted in communication gap in the early stages and also amounted to missing our Annual proposed conference at Srinagar.

Procedural details were planned for different Awards and reprints (in quadruplicate), nominations etc. were invited. Following were the Award Committees:

1. Prof. Nils Lagerlof Memorial Award (1986 & 1987):

Dr. B.R. Deshpande, Bombay	Chairman
Dr. B.R. Benjamin, IVRI, Izatnagar	Member
Dr. H.C. Pant, Mathura	Member
Dr. R.D. Sharma, Ludhiana	Member

2. Dr. G.B. Singh Memorial Award (1987):

This is newly instituted Award and is for the Young Scientist, who should be the first author of the paper and should also be below 35 years of age. The Award Committee constituted for Prof. Lagerlof Award was requested to work also for Dr. G.B. Singh Memorial Award, 1987.

3. ISSAR Fellowship Award Committee:

Dr. C.R. Sane, Bombay	Chairman
Dr. A.S. Kaikini, Nagpur	Member

The response for Prof. Lagerlof Memorial Award was found to be most encouraging as there were 16 and 17 entries for 1986 and 1987 Award, respectively. Response from the Young Scientists for Dr. G.B. Singh Memorial Award was not that encouraging as there were only 3 entries to be considered for the Award. Response for ISSAR Fellowship Award was also rather poor as there were only 4 nominations for the Award. The committees evaluated the papers nominations very critically and the winners of the different awards are as under:

(A) Prof. Nils Lagerlof Memorial Award, 1986:

Dr. C. Chandrahasan, Dr. S.R. Pattabhiraman and Dr. V. Venkataswami Deptt. of Gynaecology, Madras Vety. College, Madras, for their article, 'Studies on the effect of

glasswool column filtration on the quality of semen of cross-bred bulls" published in Indian Vet. J. 63: 913-918 (1986).

(B) Prof. Nils Lagerlof Memorial Award, 1987:

Dr. Shyam Zanwar: Manager, Embryo Research Centre, Gopalnagar, Bilaspur, MP for his article, "Embryo transfer in cattle" published in Indian J. Anim. Prod. and Management, 3: 161-165 (1987).

(C) Dr. G.B. Singh Memorial Award, 1987: (Young Scientist Award)

Dr. A.J. Dhami, (V.M. Mehta and S.B. Kodagali) Dept. of Gynaecology & Obstetrics, Gujarat Veterinary College, Anand for article, "Leakage of transaminases during freezing of buffalo bull semen: effect of dilutor, season, bulls and stages of freezing" published in Indian J. Anim. Sci. 57(12): 1272-1278 (1987).

(D) ISSAR Fellowship Award:

(i) Dr. C.P.N. Iyer, Head, Deptt. of Gynaec. & Obstetrics, College of Vety. & Animal Sciences, Mannuthy, Trichur.

(ii) Dr. H.C. Pant, Head, Deptt. of Gynaec. & Obst. College of Vety. Science and Animal Husbandry, Mathura.

(iii) Dr. S.K. Verma, Associate Prof. and Head, Deptt. of Gynaecology & Obstetrics, College of Veterinary Sciences, Haryana Agricultural University, Hissar.

I would like to thank profusely Chairman and members of different Award Committees for their excellent evaluation work and co-operation.

2. I approached the ICAR Authorities with a request for providing us an assistance for publication of IJAR and also for holding conference. I am happy to inform you that we have already received Rs. 7000/- for publication of IJAR for the year 1987 and another sanction of Rs. 7000/- may be received for publication of IJAR for the year 1986 also, shortly.

3. The important resolutions adopted at Executive Council Meeting held at Trichur are as under:

- 1) Resolved that new committees for Prof. Nils Lagerlof Memorial Award, Dr. G.B. Singh Memorial Award and ISSAR Fellowship Award be constituted. It was further resolved that the President and Secretary should also be the Members of ISSAR Fellowship Award Committee.
- 2) Resolved that the Annual Membership Rate be raised to Rs. 75/- from the present rate of Rs. 50/- after getting the necessary approval from the General Body meeting. Now the revised rates will be made applicable from the year 1989 as the General Body has approved the same.
- 3) Resolved that the State Chapter Secretaries will transfer the entire amount collected towards the Institutional membership Fees, without retaining 1/3rd of the amount, as is being done in case with Annual Subscription of Life-membership.
- 4) Resolved that the sustained membership be instituted and all the State Chapter Secretaries should try to enroll the maximum number from their respective States.

The rates prescribed for Sustained Membership are as under.

- (i) Rs. 2000/- wherein only the name of the donor will be inserted in 10 issues of IJAR.
- (ii) Rs. 5000/- Half page advertisement for 10 issues of IJAR.
- (iii) Rs. 10,000/- Full page advertisement for 10 issues of IJAR.

It was further resolved that 20 per cent of the amount thus collected be retained by the concerned State Chapter.

- 5) Resolved that, to provide incentive and to have healthy competition amongst the different States, the ISSAR should institute a Trophy (Shield/ Memento) for the Best Chapter in the country.
- 6) Resolved that a proforma be evolved to invite nominations for ISSAR fellowship Awards so that there can be proper and uniform evaluation in cases of Academic, Research and Administrative fields. It was further resolved that each State chapter should nominate only one Member for the Award considering his contribution to Science of Animal Reproduction as well as to the Society. There should be only two Fellowship Awards in each year in normal course and in exceptional cases the third one may also be considered for award.

Many of you have attended the VIIth National Symposium on Animal Reproduction held at Manuthy (Kerala State) and you will agree that it was a Grand Success. May I appeal to you all once again to try to enroll maximum number of annual subscribers, life-members, institutional members or Sustained Members from your respective State and secure/book maximum institutional subscribers and advertisements for our Journal (IJAR).

With Seasons Greetings and Wishing You ALL A HAPPY AND PROSPEROUS NEW YEAR.

Your's Sincerely

D.R. PARGAONKAR
Secretary, ISSAR

REPORT OF DR. S.R. PATTABIRAMAN, TREASURER — ISSAR. PRESENTED AT VIITH NATIONAL SYMPOSIUM AT TRICHUR — AUGUST, 1988

A report on the membership and finance of ISSAR for the period January 1987 to July 1988 besides the audit report for 1987 is presented.

MEMBERSHIP OF ISSAR

Annual Members : As on 1.8.1988 there are 931 members in ISSAR. Out of these, 221 are life members and remaining 710 are annual members. Among the annual members only 275 members, that is, less than 1/3 have paid their subscription for the years 1986, 87 and/or 1988. Nearly 185 members have paid their membership upto 1984-85 only and 250 members

have not paid their membership fee since 82-83 and 83-84. Changes in position, place of work and address and changes in the executive members of local chapter could be the reasons for this poor inflow of annual membership fees by way of renewal. To overcome this problem permanently the new executive body emphasises and encourages enrollment of lifemembers only. One way to achieve this is enroll the new members as life members instead of annual members. In addition, the defaulters who have not paid the annual membership for several years may be asked to become life members and the amount due from them as arrears may be waived in lieu of their becoming lifemembers. Now there is provision for payment of life membership of Rs. 500/- in 4 instalments within one year. To avoid unnecessary correspondence and reminders, the members should pay the instalments in time and the local chapter Secretary or Treasurer should monitor and help in regular payment of instalments by the members.

Life Members : There are 221 life members as on 1.8.88. The Directory of life members of ISSAR is printed to provide the name, address and the ISSAR number of the life members. On January 1987 when the new executive body took over the ISSAR Office, there were 116 life members. But during 1987 and upto July '88 in the past 19 months, 105 new members have been enrolled as life members with nearly 47% increase. This increase though encouraging, is not sufficient. It is not even 1/4th of the total strength of members. We must strive hard to increase the life membership to atleast half if not three fourth of the total strength, that is, the present number of 221 should increase to 500 and above. We have life members from 18 different States. Maharashtra, Rajasthan, Assam and Gujarat are the four States which have more than 30 life members each. Nearly 60% of the total number are from these four states. This is because of the special interest and effort taken by Dr. B.L. Bishnoi of Rajasthan Chapter, Dr. Dharmi, of Gujarat Chapter, Dr. Tamuli of Assam Chapter and Dr. Hukeri of Maharashtra Chapter. Response from Uttar Pradesh, Andhra Pradesh and Madhya Pradesh have been good and they have nearly 15-20 members. Karnataka, Orissa, Punjab, Haryana, Bihar and Tamil Nadu are the States with lots of potential to enroll more life members. It is strange to record that there are no life members from Kerala chapter.

Finance : Annual membership, Life membership and Institutional subscriptions are the main sources of finance to the Society. In the 2nd executive meeting held at Madras in January '88, it was decided that the lifemembership fee collected should be kept in fixed deposits and only the interest that accrue from such investments should be used for the activities of ISSAR, besides utilising the Annual subscription. Accordingly Rs. 60,000/- has been deposited in fixed deposit for 3 years period.

Institutional Subscription : This is different from the membership fee. Rs. 100/- per annum is collected from the institutions as subscription for the journal IJAR. Gujarat chapter has taken the lead in getting number of institutional subscriptions. In Tamil Nadu, Animal Husbandry Department has subscribed for the IJAR and included 40 institutions as subscriber for IJAR. The amount of institutional subscription besides the income by way of advertisement tariff are maintained by the Editor. The amount will be utilised for the purpose of printing and publishing the journal. As per the existing rules and regulations 1/3rd of the annual and life membership amount collected can be retained by the local chapter for its activities and remaining 2/3rd should be sent to the Main Office of ISSAR.

On the other hand amount collected by way of institutional subscription, advertisements, donations to funds etc., should be sent completely without deductions to the main office account.

Prof. Nils Lagerlof Memorial Fund : Rs. 10,000/- is in fixed deposit. This provision has been made by the previous executive body and no further contribution is collected for the fund, so far.

Dr. G.B. Singh Memorial Fund : So far, Rs. 11,000/- has been collected. Besides the amount collected at the National Conference at Assam in 1986, major contributions are Rs. 2005/- from Rajasthan Chapter through Dr. B.L. Bishnoi, Rs. 1450/- from Orissa Chapter through Dr. L.D. Mohanty and Rs. 1020/- through Dr. B.N. Mohanty, Rs. 870/- from Punjab and Rs. 700/- from Calcutta Chapter. This amount is kept in Fixed Deposit and the interest that accrue will be utilised for the award.

Dr. C.R. Sane Oration Fund : This fund is initiated recently with an objective to meet the expenditure in arranging the special lectures by eminent scientists. Members are requested to contribute generously for this fund.

The amount paid towards membership fees, subscriptions and also the contributions to the different funds should be sent by Bank draft payable to "Treasurer, ISSAR, Madras" and then mailed to Treasurer's address.

S.R. Pattabiraman
TREASURER, ISSAR

INDIAN SOCIETY FOR THE STUDY OF ANIMAL REPRODUCTION (ISSAR) MADRAS RECEIPTS AND PAYMENTS ACCOUNT FOR THE YEAR ENDED 31-12-1987

RECEIPTS		PAYMENTS	
To Opening Balances:		By Printing of IJAR Journals	33100.00
Cash on hand	518.47	IJAR Journal Office Expenses	2000.00
Cash with S.B.I. Bombay	61995.45	Postage	1085.40
Annual Membership Fee	7562.00	Printing & Stationery	361.35
Life Membership Fee	17377.00	Bank Charges	276.00
Advertisement	8250.00	Conveyance	455.90
Amount Received towards		Packing Charges of Journals	195.25
G.B. Singh Memorial Fund	4050.00	Audit Fees 1986	500.00
Interest earned on SB a/c	2892.45	President Office Expenses	500.00
		Secretary Office Expenses	500.00
		Closing Balances:	
		Cash on hand	312.65
		Cash with S.B.I., Madras	
		SB a/c	59148.05
		Cash with Bombay Office	4210.77
	<u>102645.37</u>		<u>102645.37</u>

INCOME AND EXPENDITURE ACCOUNT FOR THE YEAR ENDED 31-12-1987

EXPENDITURE		INCOME	
To Printing of IJAR Journals	33100.00	By Annual Membership Fee Received	7562.00
Bank Charges	276.00	Life Membership Fee Received	17377.00
Conveyance	546.90	Advertisements	8250.00
Postage	1763.15	Interest Earned on savings a/c	2892.45
Stationery	2592.60	Excess of Expenditure over	
Packing charges for journals	195.25	Income transferred to General	
Audit Fees 1986	500.00	Fund A/c	3392.45
Provision for Audit Fees 1987	500.00		
	<u>39473.90</u>		<u>39473.90</u>

BALANCE SHEET AS AT 31ST DECEMBER, 1987

LIABILITIES		ASSETS	
GENERAL FUND		CASH AND BANK BALANCES	
As per last Balance Sheet	74860.58	Cash on hand	312.65
Less: Excess of Expenditure over		Cash with S.B.I. Madras in SB a/c	59148.05
Income transferred from Income		Closing cash balance held with	
and Expenditure account	<u>3392.45</u>	Bombay Office	3969.59
	71468.13	Difference in cash balance brought	
ENDOWMENT FUNDS:		forward from 1986 to 1987 accounts	241.18
Nils Lagerlof Memorial Fund	10000.00	INVESTMENTS:	
G.B. Singh Memorial Fund	<u>4050.00</u>	In Fixed Deposit in S.B.I. towards	
	14050.00	Nils Lagerlof Memorial Fund	10000.00
AMOUNTS PAYABLE:		AMOUNTS RECEIVABLE:	
Audit Fees	500.00	I.C.A.R. Journal	7000.00
		Vlth National Congress	5000.00
		Interest Accrued & Due	<u>346.66</u>
	<u>86018.13</u>		12346.66
			<u>86018.13</u>

MADRAS 2. DATED 21-7-88

VIDE OUR REPORT OF EVEN DATE

A.R. Rao
President
ISSAR

D.R. Pargaonkar
Secretary
ISSAR

S.R. Pattabiraman
Treasurer
ISSAR

RAMANAN & Co.
Chartered Accountants