EVALUATION OF BOAR SEMEN EXTENDED IN BELT'S VILLE THAW SOLUTION

By
Kantharaj. S.

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

Master Of Veterinary Science

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University

Department of Animal Reproduction
COLLEGE OF VETERINARY AND ANIMAL SCIENCES
MANNUTHY, THRISSUR,
KERALA

2001

DECLARATION

I hereby declare that this thesis entitled **"EVALUATION OF BOAR SEMEN EXTENDED IN BELT'S VILLE THAW SOLUTION"** is a bonafide record of research work done by me during the course of research and that this thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society

8.10.01
Mannuthy

Kantharaj. S.

CERTIFICATE

Certified that this thesis, entitled **"EVALUATION OF BOAR SEMEN EXTENDED IN BELT'S VILLE THAW SOLUTION"** is a record of research work done independently by **Dr. Kantharaj. S.,** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to him.

Mannuthy
8-10-01

Dr. K.V. Athman,
Associate Professor and Head,
University Veterinary Hospital,
Kokkalai. Thrissur.

CERTIFICATE

We, the undersigned members of the Advisory Committee of **Dr. S. Kantharaj,** a candidate for the degree of Master of Veterinary Science in Animal Reproduction, agree that the thesis entitled **"EVALUATION OF BOAR SEMEN EXTENDED IN BELT'S VILLE THAW SOLUTION"** may be submitted by **Dr. S. Kantharaj,** in partial fulfillment of the requirement for the degree.

Dr. K.V. Athman,
(Chairman, Advisory Committee)
Associate Professor and Head,
University Veterinary Hospital,
Kokkalai.

Dr. T. Sreekumaran,
(Member, Advisory Committee)
Associate Professor and Head,
Department of Animal Reproduction.

Dr. P.P. Balakrishnan,
(Member, Advisory Committee)
Special Officer,
College of Veterinary
and Animal Sciences,
Pookote.

Dr. T.V. Viswanathan,
(Member, Advisory Committee)
Associate Professor and Head,
Centre for Pig Production and Research.

19/12/01
External Examiner

Dr. C. VEERAPONDIAN
Professor and Head,
Dept. of Obst. & Gynaec
Madras Veterinary College
Chennai: 7.

ACKNOWLEDGEMENTS

I wish to express my gratitude and indebtedness to Dr.K.V. Athman, Associate Professor and Head, University Veterinary Hospital, Kokkalai and Chairman of the Advisory committee for his inspiring professional guidance, critical comments and sustained encouragement which enabled the successful completion of this research programme.

I am extremely grateful to Dr. T. Sreekumaran, Associate Professor and Head of the Department of Animal Reproduction for his valuable suggestions and words of inspiration throughout the period of my study.

A great amount of indebtedness is also conveyed to Dr. P.P. Balakrishnan, Professor and Special officer, College of Veterinary and Animal Sciences, Pookote for his suggestions, support and assistance as a member of the advisory committee.

There are no words to pay my respect and gratitude to Dr. T.V. Viswanathan, Associate Professor and Head, Centre for Pig production and research, for his words of inspiration, kindness and professional guidance during the entire period of research.

My grateful thanks are due to Dr. Joseph Mathew, Dr. K.N. Aravinda Ghosh, (Associate Professors), Dr. Metilda Joseph, Dr. G. Ajitkumar and Dr. A.M. Vahida (Assistant professors).

I sincerely thank Dr. Joseph Mathew, Assistant Professor, Centre for Pig Production and Research and Dr. K.V. Raghunandanan, Director, Department of Animal Genetics and Breeding for their valuable suggestions and help rendered during the course of the study.

I am cordially obliged to Dr. C.P. Gopakumar and Dr. Shaji Antony (Pig Breeding Centre, Puthur) for their help and suggestions rendered during the training period.

I gratefully acknowledge the help rendered for statistical analysis by Dr. V. Narayanikutty and Smt. K.P. Shanthabai, (Department of Statistics).

I would like to express my sincere gratitude to my colleagues Dr. K. Lydia Priscilla, Dr. P. Senthil Kumar, Dr. P. Gopikrishnan, Dr. Geetha, Dr. Bibin Becha, Dr. Ranjini and Dr. Velayudhakumar for their incessant help and moral support rendered during the study.

A bouquet of thanks to Dr. P. Vijayakumar, Dr. S. Rajalakshmi, Dr. K. Rajkumar, Dr. K.M. Sakthivel, Dr. R. Sangeetha, Dr. A. Sabiha, Dr. Binsy and Dr. R. Lakshmi for their unstinted help and constant support.

My sincere thanks are due to Dr. S. Ramakrishnan and Dr.T.V. Raja for their whole hearted help and co-operation during the study.

I would like to record my sincere thanks to Dr. K.A. Arul Anand, Dr. N. Arunmozhi, Dr. M. Karthikeyan, Dr. M. Kalyan, Dr. M. Manigandan, Dr. S. Venky and Dr. S. Hariharan for their moral support, help and incessant encouragement throughout my study.

Let me thank Dr. M. Arumugam, Dr. S. Balasubramaniam, Dr. S. Balaji, Dr. P. Senthilkumar (Sur.), Dr. A. Gopinathan, Dr. A. Thirunavukkarasu, Dr. P. Senthilkumar (Pha.), Dr. Baskar, Dr. Venkatesakumar, Dr. G. Vasudevan, Dr. Kandaswamy, Dr. P. Murali, Dr. Madhu and Dr. Srinvasreddy for making my stay with them a memorable one.

My profound thanks to Dr. C. Paul Princely, Dr. D. Prasanna, Dr. M. Balasubramanian, Dr. N. Yuvaraj, Dr.Vimal Rajkumar, Dr. S. Rajendran and Dr. Bindya for their incessant help and moral support.

I am grateful to the Dean, College of Veterinary and Animal Sciences for the facilities provided for this investigation.

I am thankful to Kerala Agricultural University for awarding me the fellowship for the Post Graduate study.

I owe my sincere gratitude to the non-teaching staff of the Department of Animal Reproduction, Farm Supervisor and Labourers of the pig farm for their overwhelming help and support during the period of my study.

I appreciate with thanks, the care and skill with which the typing and compiling of thesis was done by Mr. T.S. Sumesh, M/s Delta Computers, Mannuthy.

I owe my sincere respect and special gratitude to my parents, sisters and brothers for their unfailing patience, moral support and encouragement throughout my carrier.

I bow my head to the almighty God, for being with me in all my endeavours.

Kantharaj. S.

Dedicated

To

My Beloved Parents

CONTENTS

Chapter No.	Title	Page No.
1.	INTRODUCTION	1
2.	REVIEW OF LITERATURE	4
3.	MATERIALS AND METHODS	32
4.	RESULTS	38
5.	DISCUSSION	54
6.	SUMMARY	67
	REFERENCES	70
	ABSTRACT	

LIST OF TABLES

Table No.	Title	Page No.
1	Mean semen characteristics of three LWY boars	46
2	Mean sperm abnormalities (per cent) of three LWY boars	47
3	Mean Protoplasmic droplets (per cent) in semen of three LYW boars	48
4	Mean motility (per cent) of boar semen preserved in BTS	48
5	Duration of oestrus, conception rate, farrowing rate, gestation length and litter size in naturally served and artificially inseminated gilts and sows	49

LIST OF FIGURES

Fig. No.	Title	Page No.
1.	Colour of boar semen	50
2.	Comparison of total abnormal sperms (per cent) among three boars	50
3.	Comparison of mean protoplasmic droplets among three boars	51
4.	Preservability of boars semen extended in BTS at various hours of preservation	51
5.	Comparison of conception rate among naturally served and artificially inseminated gilts and sows	52
6.	Comparison of gestation length among naturally served and artificially inseminated gilts and sows	52
7.	Comparison of litter size among naturally served and artificially inseminated gilts and sows	53

LIST OF PLATES

Plate No.	Title	Between pages
1.	Disposable AI pipette with semen receptacle, glass beaker and muslin cloth	37-38
2.	Disposable AI pipette with semen receptacle assembled	37-38
3.	Dummy sow	37-38
4.	Semen collection (Gloved hand technique)	37-38
5.	Artificial insemination (AI)	37-38
6.	Natural service (NS)	37-38
7.	Piglets born out of AI	37-38
8.	Sperm abnormality (Detached head)	37-38
9.	Sperm abnormality (Mid piece and tail abnormality)	37-38

Introduction

1. INTRODUCTION

The importance of animal protein as a protective food for human beings is well known. Milk and milk products, which constitute a rich source of animal protein, are not available in adequate quantities in developing countries due to poor productivity of the animals. The gap between demand and supply is increasing at a faster rate. Among livestock swine are credited to be prolific breeders, capable of converting cheap food materials into valuable animal protein at a faster rate than many farm animals. Thus pig is one of the animals that bridge the gap between requirement and availability of animal protein in our country. The productivity of the native stock is so low that they cannot be profitably employed. Therefore, efforts are being made to evolve suitable breeds capable of giving maximum returns under different agro-climatic conditions.

The study of artificial insemination was started as early as in 1927 (Rommele, 1927). Historically, the swine industry adopted the use of artificial insemination at a slower pace compared to dairy industry. However, this is about to change. The industry has begun to reexamine and realise the importance of artificial insemination as a breeding tool.

Artificial insemination in pigs has the primary objective of genetic improvement with maximum health control at a least cost (Walter *et al.*, 1984).

Artificial insemination when compared to natural service has got several advantages such as boar replacement, disease control, breeding stock improvement, improved conception rate and litter size. Apart from these advantages, artificial insemination is highly feasible and economical. Though this technique is very widely used for improvement of swine in countries like Denmark, Sweden, Norway, Britain and Japan, a beginning is yet to be made in our country.

The development of artificial insemination in any species depends to a large extent on developing semen preservation techniques in vitro for several days without causing a serious reduction in the fertilizing capacity of the sperms. The main problem limiting the development of artificial insemination is the inability to store semen for more than two or three days.

Relatively few experiments have been carried out on storage of boar semen. The report of Murty and Rao (1975) appears to be the first published work available from India on preservation of boar semen. There are many semen diluents

available; some of which requires sophisticated laboratory techniques to prepare and are used only by commercial artificial insemination laboratories. The most appropriate diluents for on-farm use are those that have simple, one stage mixing, such as Belt's ville thaw solution (BTS), Guelph, Kiev or Merck diluents. These can keep the sperms viable for three days. Various works on preservability of boar semen in BTS had been reported (Machaty *et al.*, 1992, Alexopoulos *et al.*, 1996 and Pandey and Singh, 1998). Good preservability and fertility of boar semen extended in BTS have been reported and is commercially used.

Reports on scientific works on artificial insemination in pigs are very scanty, especially in Kerala. Hence the present investigation has been carried out with the object of assessing the fertility and preservability of boar semen extended in BTS.

Review of Literature

2.REVIEW OF LITERATURE

2.1 Training

Foote *et al.* (1959) trained boars to serve artificial vagina at four months of age and found that most were disinterested, but four out of 22 produced ejaculates at approximately 18 weeks of age. Melrose (1963) found that the average age at the time of first ejaculation was six to seven months in Large white Yorkshire (LYW) boars, but semen of satisfactory quality was not obtained until one month later. Swierstra and Rahnefeld (1967) reported that the average age of Yorkshire boars at the start of semen collection were 8.9 months and 9.4 months in different years of study. Buisson and Signoret (1970) found that young LWY boars were trained for semen collection at the average age of 230 days. Swierstra (1973) could collect better quality semen from LWY boars of 21-31 weeks. Murty (1974) trained LWY boars for semen collection over a wooden dummy at the age of 11 to 12 months. Tamuli and Rajkonwar (1988) faced problems in training boars because of their varying idiosyncratic behaviour and observed that Landrace boars could be trained successfully for semen collection at the age of 11 to 12 months. Singleton (1997) stated that some boars of seven

months old were ready for collection, others may require another six to eight weeks.

Chamohoy *et al.* (1960) found that it took seven days for inexperienced boars to be trained to mount a dummy and ejaculate freely where as boars, which had previously experienced some sexual activity (natural mating), took an average of 14 days to be trained for ejaculation. Hughes and Varley (1980) stated that boars could be easily trained to mount simple dummy sow devices and ejaculated readily without sophisticated artificial vaginal devices. Tamuli and Rajkonwar (1988) recorded ejaculation over the dummy sow on the very first day in one Landrace boar when alternative exposure to the dummy sow and teaser sow was allowed, whereas the other three boars gave ejaculation within the third day of the trial. They also recorded the noisy sound produced by the teaser sow at the time of mounting of the boar over her and played it near the dummy sow when boars were trained for semen collection. Gordon (1997) suggested that dummies have an advantage over live mounts in providing stability and preventing possible disease transmission.

Niwa (1961) recommended that the training of boars for collection should be started when it is about 100 kg live weight. Swierstra and Rahnefeld (1967) found that the

average weights of Yorkshire boars at the beginning of the semen collection were 131 [illegible] and 121 kgs in two different periods. But Swierstra (1973) found attainment of 86-95 kg body weight at the start of semen collection in 21 to 31 weeks old LWY boars.

Maule (1962) instructed to construct the dummy sow as a padded framework of wood and metal on which the boar can mount and allows access to the penis for collection. Swierstra (1973) collected semen using a dummy sow constructed from a domestic water heater tank over which a one-inch thin layer of foam rubber and a canvas was covered. Tamuli and Rajkonwar (1988) fabricated a dummy sow using a portable wooden service crate after fixing one iron frame covered with a firm dirty white canvas cushion to give the shape of a dummy sow back for collection of semen.

Singleton (1997) stated that training was one of the most difficult and frustrating step in the artificial insemination process of pigs.

2.2 Semen collection

Stratman and self (1961) used a dummy sow and artificial vagina to collect semen from Landrace and Duroc boars. Murty (1974) could collect semen from LWY boars using

artificial vagina and dummy sow. Sreekumaran (1974) collected semen from LWY boars using an artificial vagina based on Norwegian type artificial vagina, which has a Y-shaped slit in the foam rubber piece, that was fastened over the entrance of vagina to provide a mechanical cleansing as the penis passed through it.

King and Macpherson (1973) collected semen from boars by using artificial vagina and the gloved hand technique of Hancock and Hovell (1959). In the gloved hand technique, by wearing a disposable vinyl glove, the spiral tip of the penis was firmly held after sliding through the partially closed fist for a few seconds. They recommended that gloved hand technique was satisfactory for routine collection of boar semen because it provided adequate stimulus to achieve optimum sperm output than artificial vagina.

Tamuli and Rajkonwar (1988) performed semen collection in boars by simple fist method in which the penis was grasped just one centimetre behind the tip with a neatly cleaned palm swabbed with spirit to maintain a rhythm of pressure.

Gordon (1997) stated that the satisfactory collection of boar semen could be normally achieved using the

gloved hand technique because the gloved hand readily adapts to the corkscrew shape of the boar's penis and provides the constant pressure necessary during the stimulatory and ejaculatory phases of the collection process.

2.2.1 Frequency of collection

Murty (1974) collected semen from LWY boars weekly by the artificial vagina method. Likewise Sreekumaran (1974) did weekly collection in LWY boars by artificial vagina method.

Semen was collected twice per week as the sperm rich fraction from Dutch Landrace (DL) and Dutch Large White (DLW) boars (Johnson *et al.*, 1979). Johnson *et al.* (1982) collected semen from Landrace boars by the gloved hand rubber sleeve method twice weekly. Machaty *et al.* (1992) conducted semen collection from five LWY boars twice weekly by the gloved hand method.

2.3 Semen Quality

2.3.1 Volume

Johari (1956) reported that the volume of semen ejaculated by a normal boar aged 9-18 months ranged from 30-165 ml. The gel portion was found to comprise of 30-40

per cent of the whole ejaculate. Aamdal and Hogset (1957) found that the mean total volume, strained volume and gel volume for mature boars as 261 ml, 202 ml and 59 ml respectively. Stratman and Self (1961) found that the average values for the total volume and strained volume in yearling Hampshire boars as 115 ml and 87.9 ml. Swierstra and Rahnefeld (1967) found that the mean values for total volume, strained volume and gel volume as 225 ml, 178 ml and 47 ml respectively. King and Macpherson (1973) studied the ejaculate characteristics for Yorkshire boars collected by gloved hand technique or artificial vagina method. The total volume, gel volume and gel free volume were 382 ± 219, 80 ± 49 and 302 ± 176 ml respectively for semen collected by gloved hand method. The corresponding figures were 336±173, 72 ± 45 and 264 ± 134 ml respectively for semen collected by artificial vagina method. Mean values for the total volume, strained volume and gel volume were 243.9 ml, 189.3 ml and 54.6 ml respectively in 11 to 12 months age LWY boars (Murty, 1974). The respective values obtained by Sreekumaran (1974) were 165.8 ml, 114.2 ml, 51.5 ml in 12 to 18 months old LWY boars. Kennedy and Wilkins (1984) found that the average semen volume for Yorkshire boars of nine to 11 months age was 81.5 ± 1.9 ml (Gloved hand technique). Tamuli and Rajkonwar (1988) found that the mean values for semen volume, strained volume and gel mass as 235.8 ± 11.8,

172 ± 12 and 63.8 ± 4.1 ml respectively in Landrace boars of 11 to 12 months age. Bhuyan *et al.* (1991) found mean values for total volume, filtered volume and gel mass as 191.65 ± 11.84, 156.71 ± 9.65 and 33.75 ± 2.5 ml respectively in Hampshire boars of nine to 12 months age as against 129.41 ± 6.42, 107.31 ± 5.46 and 20.81 ± 1.04 ml respectively in crossbred boars of nine to 12 months age. There was a significant difference in gel, filtered and total volume of semen collected by simple fist method in these Hampshire and crossbred boars. Arthur *et al.*(1996) stated that the semen volume of boars ranged from 125 to 500 ml with a mean of 250 ml. Ax *et al.*(2000) stated that the total semen volume of Yorkshire boars ranges from 240 to 250 ml.

Variations in semen volume of boar aged from one to two years depending upon the collection intervals were observed by Gerrits *et al.* (1962). When collections were made at four days interval they obtained a total volume of 286.3 ml, strained volume of 241.5 ml and gel volume of 44.8 ml. The corresponding figures when collection schedule was changed to every other day were 237.6 ml, 205.5 ml, and 32.1 ml respectively. When collections were made daily, the figures were 193.1 ml, 166.4 ml and 26.6 ml respectively. Swierstra (1973) studied the volume of collected semen in two ejaculates at 24 hours interval from Yorkshire and Lacombe boars. The

average semen volume was 180 ml for Yorkshire and 184 ml for Lacombe boars of 21 to 26 weeks age. Similarly, he also found that the semen volume increased to 194 ml and 199 ml respectively in another group of 27 to 31 weeks old boars.

Boar is unique among the domestic animals in that it ejaculates an enormous volume of semen not in relation to its body size because of the well developed Cowper's gland and seminal vesicles. In addition to the liquid portion, the seminal plasma contains a gelatinous material, which could at times make up more than half of the ejaculate (Mann, 1964).

2.3.2 Colour

Johari (1956) found that the semen of the boar was greyish white in colour. Melrose and Laing (1970) stated that the boar semen was greyish white and viscid. Roberts (1986) stated that the semen of boars with a low concentration of sperm cells would be pearly white to grey translucent colour with watery consistency.

2.3.3 Hydrogen ion concentration

Johari (1956) observed a pH range of 7.4 to 8.26 in boar semen. Foote *et al.* (1959) found that the average pH

value was 7.97, 7.74 and 7.56 in 18 – 33, 34 – 46 and 47 – 52 weeks old Yorkshire and Chester White boars respectively.

The mean pH value range of pre sperm, sperm rich and post sperm fraction of the semen were 7.0 to 7.4, 6.8 to 7.4 and 7.0 to 7.6 respectively (Hancock, 1959). Murty (1974) found that the average pH of the semen of LWY boars of 11 to 12 months old as 7.8 and also noted that the pH of the semen did not vary significantly between individual boars. Sreekumaran (1974) found that the average pH of LWY boar semen as 7.6 with a range of 7.3 to 7.9 and detected no significant variation in the pH of the semen between boars. He further observed that the pH was not correlated neither with motility nor with the concentration of the sperms in the ejaculate. Roberts (1986) stated that the boar semen would have normally a pH range of 7.0 to 7.8.

2.3.4 Motility

Johari (1956) found that 60 to 80 per cent of the sperms showed progressive movements in boar semen. Bane (1959) recorded that the initial sperm motility was in the range of 3.2 to 4.0 (zero to five scores) for boars in the age group of 150 to 284 days and noted improvement in sperm motility with increasing age. Similarly Lagerlof and

Carlquist (1961) observed that the initial motility of the spermatozoa of boars in the age group of 150 to 269 days varied from 2.4 to 4.0.

Initial motility indicates the viability of spermatozoa and fertility of the male; but it need not necessarily be a sign of fertilising capacity of sperms (Perry, 1960).

Stratman and self (1961) obtained mean sperm motility of 86.8 per cent with a range of 70 to 95. Buttle and Hancock (1965) observed that spermatozoa with knobbed defects of acrosome showed lower motility than normal sperms. Most authors agree that the number of actively motile spermatozoa should be about 65 to 75 per cent for best fertility (Roberts, 1986).

King and Macpherson (1973) recorded 79.4 per cent motile sperms in semen collected by gloved hand technique or artificial vagina method. Tamuli and Rajkonwar (1988) noted 83.7 per cent motility in semen of Landrace boars collected by simple fist method.

Murty (1974) recorded an average percentage of progressive motility of 80.9 in LWY boars and found a

significant difference in the percentage of motile sperms between boars. Sreekumaran (1974) found the mean percentage of motile sperms in the ejaculate ranged from 40 to 80 with a mean of 65.7. There was a significant variation in the motility of the semen sample obtained from different boars. Gibson and Johnson (1980) stated that young boars of eight to 12 months age had more than 85 per cent sperm motility in the ejaculate.

2.3.5 Live sperm count

Hancock (1959) stated that the estimates of percentage of dead spermatozoa were not reliable as far as boar semen sample is considered. Masek (1963) observed that the percentage of dead sperms in boar semen varied from 8.6 to 35.5. The percentage of live sperm in fertile boars was reported to vary from 91 to 100 (Buttle and Hancock, 1965) and 96 to 100 (Campbell and lingam, 1965). Murty (1974) reported 84.5 per cent of viable sperms in LWY boars, while Sreekumaran (1974) reported 88.8 percent of viable sperms. Kennedy and Wilkins (1984) found that the live sperm count for Yorkshire boars of nine to 11 months age was 61.91 ± 0.36 per cent (Gloved hand technique).

2.3.6 Sperm concentration

In mature boars mean concentration of sperms in millions per ml was reported to be as 245 (artificial vagina) by Ito *et al.* (1948), 275(artificial vagina) by Foote *et al.* (1959); 381 (Gloved hand technique) and 322 (artificial vagina) by King and Macpherson (1973); 297.3 (artificial vagina) by Murty (1974); 250 (artificial vagina) by Sreekumaran (1974) and 656 (gloved hand technique) by Kennedy and Wilkins (1984).

Bane (1959) noted improvement in sperm concentration with increasing age of boars. Similar result was reported by Lagerlof and Carlquist (1961) in growing boars. Roberts (1986) stated that the sperm concentration varies from 25 to 1000 millions per ml of semen with an average sperm concentration of 150 millions per ml.

2.3.7 Sperm abnormality

Milovanov (1934) reported that the normal boars might have as much as 30 per cent abnormal sperms in the ejaculate. According to Holst (1949) the semen of normal boars would not contain more than 14 per cent abnormal spermatozoa. Masek (1963) reported that the percentage of abnormal sperms in boar ejaculate ranged from 1.92 to 18.56

with a mean of 9.43. The mean percentage of sperm defects as reported by Hancock (1959) was malformed head three per cent, malformed mid-pieces 2.7 per cent, bent tails 4.5 per cent, coiled tails 0.9 per cent, headless 0.3 per cent, tailless 0.3 per cent, broken necks 0.1 per cent, neck bead (proximal) 11.8 per cent, and mid-piece beads (distal) 17.8 per cent. The mean percentage of sperm defects found by Murty (1974) were for head abnormality 2.5, mid-piece abnormality 0.47, abaxial mid-piece 16.1,malformed tails 1.9, proximal cytoplasmic droplet 0.91, distal cytoplasmic droplet 0.83 and tailless heads 1.1. The mean percentage of sperm defect found by Sreekumaran (1974) were malformed heads 1.6, malformed mid-piece 2.3, malformed tails 3.2, proximal cytoplasmic droplet 2.8 and distal cytoplasmic droplet 2.9 in Yorkshire boars. Bhuyan *et al.* (1991) reported that the mean percentage of head, mid-piece, tail and acrosomal abnormalities as 2.0, 17.7, 3.0 and 3.0 respectively in Hampshire and 1.3, 11.1, 3.3 and 3.3 in crossbred boars.

Bane (1959) and Lagerlof and carlquist (1961) observed that the percentage of abnormal sperm heads in the boar semen decreased with increase in age until full reproductive capacity was attained. The morphology of boar spermatozoa has been thoroughly investigated by Buttle and Hancock (1965). Roberts (1986) stated that the determination

of the number and type of abnormal sperm cells in an ejaculate should be used with other examinations, such as motility, concentration and live and dead sperm cell numbers for determining fertility in boars.

2.4 Preservation

2.4.1 Diluents and storage temperature

Pursel *et al.* (1973) found that boar semen diluted in BL1 extender maintained sperm fertilising capacity for 102 hours of in-vitro storage at 15°C. Johnson *et al.* (1982) conducted field trials both with Kiev and Belt's ville liquid extender and found that Kiev extender was better as a diluent for liquid semen than Belt's ville liquid extender. Aalbers *et al.* (1984) diluted boar semen with Belt's ville thawing solution (BTS) and stored at 16-18°C and used it within 24 hours or 25 to 48 hours after collection. Almlid *et al.* (1984) found that sperm motility was significantly higher for semen diluted in BTS or Zorleco diluent than for semen in Kiev diluent. Blichfeldt *et al.* (1988) found that there was no significant difference between the fertilising abilities of Kiev and BTS diluted semen on day one and two after collection. The semen diluted with BTS and used on third day was superior than semen diluted with Kiev diluent. Alexopoulos *et al.* (1996) diluted boar semen with BTS extender and left at room

temperature (18 to 24°C) for one hour and then stored at 17°C for a maximum of 120 hours. They also suggested that boar semen stored at 17°C for 72 hours could be used for insemination. Kim *et al.* (1998) used BTS without adding antibiotics in it as boar semen extender and performed artificial insemination within an interval of 12 hours after maintaining at 16-18°C. Pandey and Singh (1998) preserved boar semen up to 72 hours at 18°C after extension with BTS, Kiev or BL-1 dilutors and found a decline in sperm quality with increase in preservation time.

Vijayakumaran and Iyer (1980) conducted preservation studies of boar semen in Glucose-Glycine-EDTA-Sodium bicarbonate-Citrate diluent (GGEBC), Kiew II and Kiew I diluents and found all of them were superior in maintaining semen quality both at 15°C and 5°C. Tamuli *et al.* (1986) diluted boar semen with five diluents such as Glucose Potassium Sodium tartarate Sodium citrate dihydrate Edate (GPSE-I), Glucose-Potassium Sodium citrate dihydrate Edate (GPSE-II), Glucose Potassium Sodium tartarate Sodium citrate dihydrate Edate (GPSE-III), Egg yolk Glucose Sodium bicarbonate (EYGS) and BL-I at 15°C and observed that the preservability was highest in GPSE-I diluent upto 96 hours of preservation where minimum 50 per cent liveability was maintained.

Cheng (1988) studied the preservation of boar semen and stated that Zorlesco extender containing polyvinyl alcohol instead of Bovine serum albumin was an effective extender for the maintenance of vigorous sperm motility, acrosomal morphology and fertilising capacity of boar spermatozoa in a 15°C *in vitro* storage system. Rao *et al.* (1991) preserved boar semen in four diluents such as Egg yolk Glucose- glycine citrate (EYGC), Egg yolk-Glucose-sodium bicarbonate diluent (EYGB), Tris diluent, Glucose Potassium Sodium tartarate Edate (GPSE) and found that the keeping quality was best in GPSE diluent and poorest in EYGC diluent at 15°C even after 96 hours of preservation. They also noted that the overall progressive motility in all the diluents were superior at 15°C storage.

2.4.2 Dilution rate

Dziuk (1958) found that sperm motility was considerably lower when one part of boar semen was mixed with one part of diluent than one part of semen to two, four or eight parts of diluent. Kalev and Zagorski (1964) compared the spermatozoal survival in semen samples diluted with 10 diluents at the rate of 1:1, 1:4 and 1:9. Ooi (1970) collected semen from Large White, Berkshire and Landrace boars and diluted the semen at the rate of 1:3 and 1:5 with modified IVT

diluent. No significant variations between the dilution rates were observed. Murty (1974) diluted boar semen at 1:2 ratio with various diluents and found that the spermatozoal motility was best maintained in EYGB and EYGC. Bamba and Cran (1985) diluted one part of boar semen with BTS at the rate of 1:8. Tamuli *et al.* (1986) diluted the boar semen with GPSE-I, GPSE-II, GPSE-III, EYGS and BL-I at the ratio of 1:2 for dilution and found that preservability was maintained highest in GPSE-I for upto 96 hours of preservation where minimum 50 per cent livability was restored. Rao *et al.* (1991) used a dilution rate of 1:2 for dilution of boar semen. Machaty *et al.* (1992) diluted boar semen with BTS and Modified Kiev (MK) diluents at the ratio of 1:2 and 1:5 and found that the farrowing rate was significantly higher for BTS diluted semen.

2.4.3 Preservability at various hours of preservation

Murty (1974) estimated the progressive motility of the diluted boar semen at 37°C temperature after 12, 24,48, 72 and 96 hours of storage at five and 15°C. The percentage of motile sperms was significantly higher ($p < 0.01$) at 12 hours followed by 24, 48, 72 and 96 hours of storage. Cheng (1988) reported that the percentage of motile spermatozoa in Zorlesco extender containing 0.1 per cent polyvinyl alcohol was greater

across storage days than spermatozoa preserved in the Kiev extender. Zorpvo extender maintained good spermatozoal motility for upto nine days at 15°C storage. Rao *et al.* (1991) found that the percentage of progressively motile sperms were significantly higher ($p<0.01$) in EYGC, EYGB, Tris, GPSE diluents kept at 15°C, than at 5°C. The best results were obtained in GPSE followed by Tris, EYGB and EYGC diluents at 15°C.

Pursel *et al.* (1974) found that BL-1 failed to maintain the optimum preservability during various preservation hours. Aalbers *et al.* (1983) found that the sperm motility was 41 per cent for boar semen diluted in BTS and stored at 18°C for 72 hours. Tamuli *et al.* (1986) found that the mean preservability of boar semen diluted in BL-1 as 76.72, 67.92. 50.17, 33.50 and 16.75 at zero, 24, 48, 72 and 96 hours of preservation respectively. Alexopoulos *et al.* (1996) found that the degree of motility loss using BTS extender during preservation was lower than that reported for Kiev extender and higher than that reported for extenders containing bovine serum albumin. They concluded that the preservability of boar semen was decreased significantly from 48 hours to 120 hours of storage in BTS.

2.5 Artificial insemination

2.5.1 Heat Detection

Willingness to stand for service, which appears during actual heat, can in the absence of boar, be determined by the so-called 'standing reflex'. Presence of standing reflex is considered as the most important sign of heat in artificial insemination of swine (Madden 1959). Signoret and Buisson (1961) demonstrated that both the sound and smell of the boar could make the signs of oestrus much more definite. Bennett and O'Hagan (1964) detected oestrus in pigs as the mounting reflex when their snouts were sprayed with preputial fluid or semen. Buisson and Signoret (1970) indicated that the presence or the absence of standing reaction in the female at the time of insemination was very important and reported a decline of 20-30 percent in farrowing rate similar to Madden (1959) and Buisson and Dauzieur (1959). Murty (1974) detected oestrus in gilts by observing standing reflex along with the presence of swollen oedematous and reddened vulval lips. Roberts (1986) stated that swelling and slight reddening of the vulva in sows and gilts persists for two to three days of proestrus, two to three days of oestrus and for about two days of post oestrus with maximum swelling generally seen at the onset of oestrus. Cheng (1988) recorded behavioural oestrus in gilts by the back pressure test twice daily in the presence of

vasectomised boar. Revell and Glossop (1989) selected sows for insemination following a positive reaction to the backpressure test. Machaty *et al.* (1992) detected standing oestrus in gilts by the classical back pressure test.

2.5.2 Observations of oestrus

According to Niwa (1961) and Boender (1966) the duration of oestrus was on an average of 55 hours for gilts and 70 hours for sows. From the observations on the first oestrous cycle of 52 Dutch Landrace, Bednjfsontwikkeilng (1977) found that the average duration of oestrus as 40 hours (15-70 hours). Clark *et al.* (1986) noted that 55 per cent gilts showed onset of oestrus by 0600 hour whereas 24 per cent and 21 per cent of gilts showed onset of oestrus by 1800 h and 2400 hours respectively. Mohanty and Nayak (1986) observed that duration of oestrus in LWY and LWY X local averaged 72.4 and 82.3 hours respectively. Roberts (1986) stated that the duration of oestrum was one to four days, with an average of two to three days or about 60 hours. Anderson (1993) recorded that the duration of oestrus was 40 to 60 hours in sows. Hmar (1993) recorded that the average duration of first oestrus and second oestrus in heavy and light weaners of LWY pigs as 42.75 and 40.91 and 58.88 and 54.18 hours respectively. Stevernik *et al.* (1999) noted that the duration of oestrus

ranged from 36.6 to 60.6 hours in various farms and found that the reproductive performance would be better on farms with longer duration of oestrus. Almeida *et al.* (2000) detected that the average duration of oestrus as 52.6 hours with a range of 30-72 hours in Camborough X Canabrid terminal line gilts.

2.5.3 Method of Insemination

A plastic tube (Diameter 7-8 mm) with an inflatable cuff posterior to the tip (Aamdal and Hogset, 1957) or simple stiff rubber tubes with or without an enlargement at its tip (Polge, 1956 and Niwa *et al.*, 1959) have been used. Alternatively, Melrose and O'Hagan (1961) used a stiff rubber tube with a series of spiral ridges at its tip such that the spiral ridges locked firmly into the cervix. Simunic (1964) used a synthetic rubber catheter with a bulb close to the cranial end.

Dziuk and Henshaw (1958) performed inseminations in sows and gilts using a modified plastic pipette whose diameter was half inch larger over the first two inches of the end inserted into the vagina. Podany (1964) performed artificial insemination in gilts and sows using a plastic inseminating apparatus.

Melrose (1963) stated that insemination methods in pigs are mainly aimed at passing a small bore catheter (diameter 4mm) through the cervix to allow deposition of semen directly into the uterus or, alternatively, to use a large catheter (diameter 12mm) which can be firmly lodged into the cervical folds for preventing back flow of semen into vagina.

Artificial insemination in pigs was performed using Melrose catheter by Murty (1974), Johnson *et al.* (1982) and Cheng (1988).

2.5.4 Volume and concentration

Wiggins *et al.* (1951) reported that the minimum quantity of semen required for insemination was 50 ml to get successful results. Polge and Rowson (1956) suggested that 100 ml of semen in gilts and 200 ml in sows were sufficient to get satisfactory results in artificial insemination. Aamdal (1966) considered that 2000 to 4000 millions, as the quantity of sperm required to obtain satisfactory fertility rate in artificial insemination. Buisson and Signoret (1970) found an increase in farrowing rate by raising the volume of the semen inseminated from 50 to 200 ml. Ooi (1970) used an insemination dose of 50 ml and obtained a farrowing percentage of 62.7 to 68.6 for day one semen extended with

IVT and the corresponding figures of 66.2 to 80.3 for day two semen. Johnson *et al.* (1982) used an insemination volume of 100 ml containing three billion sperms for inseminating pigs. Cheng (1988) inseminated gilts with 50 ml of semen extended in Zorlesco containing 2.5 x 10^9 spermatozoa. Machaty *et al.* (1992) used 70 ml of diluted boar semen extended with BTS to inseminate sows and noted an increased farrowing rate than with MK diluent. Gordon (1997) suggested that porcine artificial insemination using liquid semen required 2-3 x 10^9 sperm cells per dose of 70 to 80 ml. Kim *et al.* (1998) used varying concentrations of sperm cells per insemination dose of 80ml semen diluted with BTS and found no significant difference in the farrowing rate and litter size. Mercat *et al.* (1999) found that the reduction in the sperm number in the insemination dose had no effect on both fertility and prolificacy. Kuo *et al.* (2000) indicated that there was no significant difference in any reproductive performance between gilts or sows inseminated with 3 x 10^9 or 5 x 10^9 sperms per 80 ml of diluted semen ($p>0.05$).

2.5.5 Time of insemination

Stratman and Self (1960) conducted insemination in gilts within four hours after onset of oestrus. Stratman and Self (1961) bred sows either naturally or artificially

approximately from the time they were first detected in oestrus. Melrose (1963) indicated that for maximum fertility, the correct time of insemination would be 10 to 30 hours after the sow will first stand for the boar i.e., late in the first or early in the second day of heat. Simunic and Abram (1968) reported that the conception rate and litter size was increased by double insemination after the onset of oestrus. Murty (1974) inseminated gilts twice during oestrus. The first insemination was done 24 hours after the onset of oestrus and the same was repeated 12 hours later. Johnson *et al.* (1982) inseminated sows and gilts once, 12 to 24 hours after detection of standing oestrus. Cheng (1988) inseminated gilts twice a day at an interval of 12 hours during the behavioural oestrus. Revell and Glossop (1989) did double inseminations in sows at 24 hours apart. Machaty *et al.* (1992) inseminated sows and gilts only once, 12 to 24 hours after detection of standing oestrus. Alexopoulos *et al.* (1996) inseminated sows twice at 12 and 24 hours after detection of standing oestrus. Kim *et al.* (1998) performed insemination in pigs with liquid semen twice a day with an interval of 12 hours. Munoz *et al.* (1999) concluded that the number of artificial insemination (AI) had significant effects on fecundity of adult sows with two AI having a better performance than three AI.

2.5.6 Conception rate

Hancock (1958) found that the conception rate after artificial insemination was inferior to those obtained with natural service. On the contrary, Stratman and Self (1961) reported that the conception rate for natural service was 58.3 per cent, which was lower than 83.3 per cent obtained in artificial insemination. Simunic and Abram (1968) reported that the conception rate to first insemination was 79.22 to 87.44 per cent in various farms. Cole and Cupps (1969) suggested that the conception rate in artificial insemination appeared to be equal to natural service. Silaeva (1970) obtained 98 per cent conception rate with semen stored upto three days in a glucose-chelate-citrate-sulphate diluent at 16 to 20°C. Murty (1974) obtained 66.0 per cent conception rate among artificially inseminated gilts. Hughes and Cole (1975) reported that the conception rate in pubertal gilts was 92.4 per cent. Kim *et al.* (1998) obtained 87.8 per cent non-return rate at the numbers of $2x10^9$ sperm cells per insemination dose. Kuo *et al.* (2000) could not find any significant difference in the conception rate between gilts or sows inseminated with $3x10^9$ or $5x10^9$ sperm cells.

Pay and Davies (1973) observed that the conception rate (72.5 per cent) among gilts bred at pubertal

oestrus was significantly lower than those bred at subsequent oestrus. Libal and Wahistrom (1976) and Macpherson *et al.* (1977) reported that the conception rate among gilts bred at first oestrus (64.0 to 69.6 per cent) was lower than that of those mated on third oestrus (83.0 to 86.0 per cent). Young and King (1981) reported a higher conception rate among gilts bred at third oestrus.

2.5.7 Farrowing rate

Corteel *et al.* (1964) reported that the farrowing rate was higher for gilts and sows inseminated with carbonated diluent than the non-carbonated diluent. Silaeva (1970) obtained 70 per cent farrowing rate for semen diluted in glucose-chelate-citrate-sulphate diluent. Johnson *et al.* (1982) reported that the farrowing rate among gilts and sows inseminated with semen diluted and stored for three days in BL-1 or Kiev extender was 66.1 and 61.3 per cent respectively. Performing inseminations with BTS diluted semen one and two days after collection, Aalbers *et al.* (1984) found that the farrowing rate was 80 and 79 per cent respectively. Machaty *et al.* (1992) reported that all first inseminations with BTS diluted semen containing $5x10^9$ spermatozoa and used on day four gave a farrowing rate significantly higher ($p<0.05$) than MK diluted semen (74.5 vs 54.2 per cent). Kim *et al.* (1998)

claimed that the farrowing rate was the highest (84.5 per cent) at the numbers of $2.5x10^9$ sperm cells per dose but it was not statistically significant. Kuo *et al.* (2000) found that gilts or sows inseminated with homospermic semen showed significantly higher farrowing rate.

2.5.8 Gestation length

Murty (1974) found that the mean gestation length for artificially inseminated gilts was 114 days. Roberts (1986) cited that the gestation period in swine varied from 111 to 116 days. Kao *et al.* (1996) reported that the gestation period was exactly 114 days for both artificially inseminated and naturally served sows and the gestation period was not significantly affected by breed, parity or mating. Sharda (2000) reported that gestation length was influenced by breed, number of foetuses, foetal pituitary and adrenal corticoids.

2.5.9 Litter size

Stratman and Self (1961) reported that the mean litter size was 7.8 piglets in natural service compared to 12.2 piglets in artificial insemination. Whereas, Lindstrom (1966) observed a significant difference in the litter size after natural service and artificial insemination. The litter size in artificial

insemination was 10.8 compared to 11.6 in natural service. Murty (1974) obtained a mean litter size of 8.2 for artificially inseminated gilts. Johnson *et al.* (1982) found that the litter size among sows and gilts inseminated with semen diluted and stored for three days in BL-1 or Kiev extender as 10.2 and 9.1 respectively. Fecundity for BTS diluted semen stored for one, two or three days was better than that of semen diluted with Kiev, Zorlesco and Modena extenders (Aalbers *et al.*, 1983; Johnson and Aalbers, 1984). Machaty *et al.* (1992) found that sows and gilts inseminated with BTS semen had a greater total number of piglets born alive per litter than sows and gilts inseminated with MK semen (9.5 Vs 8.9, $p<0.05$). Kim *et al.* (1998) showed that the litter size was not influenced by reduction of motile sperm cells in artificial insemination from $3x10^9$ cells to $1.5x10^9$ cells per dose. Kuo *et al.* (2000) indicated that there was no significant difference in litter size between gilts or sows inseminated with $3x10^9$ or $5x10^9$ sperms ($p>0.05$).

Materials and Methods

3. MATERIALS AND METHODS

Twenty four Large White Yorkshire (LWY) gilts and twenty four sows and six boars belonging to the Centre for Pig Production and Research (CPPR), Kerala Agricultural University, Mannuthy were utilised for the study. The age of the boars varied from eight to twelve months and weighed around 100 Kg. Healthy sows of same parity and gilts belonging to same age groups and under identical conditions were selected for the study.

Out of six boars, three boars were used for natural service and the other three for artificial insemination. Twelve sows and 12 gilts were used for natural service (Group-I) and the remaining 12 gilts and 12 sows for artificial insemination (Group-II) using semen extended in Belt's ville thaw solution.

3.1 Collection of semen

Three out of six boars were trained to mount a wooden dummy covered with a rexine material (Plate 3) for collection of semen. All the three boars were trained for a minimum of 10 minutes per day before feeding for a training

period of 10 days. A total of 52 semen collections were made from these three boars at weekly intervals using gloved hand technique (Plate 4). As soon as the boar mounted the dummy, the spiral tip of the penis was firmly held after it had been allowed to slide through the partially closed fist (King and Macpherson, 1973). The gel portion of the ejaculate was filtered at the time of collection by covering the collection beaker with two to three layers of muslin cloth. The beaker containing the semen was kept at 37°C for further evaluation.

3.2 Semen quality

3.2.1 Volume of semen

The volume of the semen was measured by means of a measuring cylinder. The quantity of gelatinous material and gel free semen was measured separately.

3.2.2 Colour

The colour of the semen was judged with naked eye, holding the measuring cylinder against a white background.

3.2.3 Hydrogen ion concentration

A pH indicator paper strip[1] was used to determine the pH of semen. The paper was dipped in the semen sample and the change in colour was compared with standard pH colours.

3.2.4 Motility

Immediately after collection of semen, initial motility was assessed by examining a drop of undiluted semen on a slide at 37°C. It was graded from zero to hundred depending upon the percentage of progressive motile sperms, in increments of 10.

3.2.5 Sperm concentration

The total number of spermatozoa per cubic millimetre was counted, using a haemocytometer and expressed in millions per ml.

3.2.6 Live sperm count and sperm abnormality

The method described by Blom (1950) was adopted for enumerating live and dead spermatozoa and sperm

[1] Range from 2.0 to 10.5, Indicator paper strips of Merck*.

abnormality. A drop of fresh semen was mixed with a drop of two per cent eosin and four or five drops of 10 per cent nigrosin on a cover slide, taking care not to damage the sperms. Spermatozoa, which were stained deep pink, were considered as dead. Partially stained or unstained sperms were counted as live sperms. Percentage of live and dead spermatozoa in the semen sample was ascertained by counting spermatozoa in various fields. The same slide was used to study the sperm abnormalities.

3.3 Preservation

Twenty good quality ejaculates collected by Gloved hand technique were utilized for preservation studies in Belt's ville thaw solution (BTS). The composition of BTS used is as follows:

D- Glucose	37.15 g
Tri-sodium citrate	6.00 g
EDTA disodium salt	1.25 g
Sodium hydrogen carbonate	1.25 g
Potassium chloride	0.75 g
Sodium penicillin	0.60 g
Streptomycin sulphate	1.00 g
Glass distilled water upto	1 litre

The gel free semen was diluted in 1:2 ratio with the diluent and the progressive motility was assessed immediately after dilution (zero hour). The semen sample was gradually cooled to room temperature and then kept at 15°C in a temperature controlled refrigerated centrifuge. The progressive motility was estimated at 37°C by examining a drop of diluted semen at 24, 48,72 and 96 hours of storage.

3.4 Artificial insemination

Oestrus was detected in gilts and sows daily by observing standing reflex in conjunction with the presence of swollen oedematous and reddened vulval lips. While detecting oestrus, the animals were approached from their rear part and pressure was applied over the back of the animal by the two hands.

Animals in Group-I, which were sexually receptive, were mated (Plate 6) once within twelve hours after the onset of oestrus and a second mating with the same boar at 18 to 24 hours later.

Animals in Group-II were inseminated with semen extended in Belt's ville thaw solution (Plate 5). The vulval lips were thoroughly washed and wiped. The disposable AI pipette

(Plate 1) was held at the middle to prevent contamination at both ends. The pipette was passed in an anterior-dorsal direction such that it prevents the entry of the spongy tip into the bladder. Once the pipette had reached the cervix, some resistance was felt. Then the pipette was directed through cervix with gentle pressure until the cervical lock prevents the pipette from becoming dislodged from the cervix even if the female moves. At this stage the semen receptacle was attached and squeezed slowly to flow out semen gently into the uterus. Animals in Group-II were re-inseminated at 18 to 24 hours later.

A comparative study on the conception rate, farrowing rate, gestation length and litter size was made and analysed statistically (Snedecor and Cochran, 1967[56]).

Plate 1

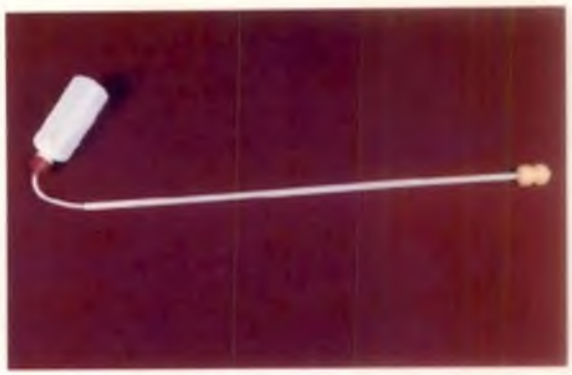


Plate 2

Plate 3

Plate 4

Plate 5

Plate 6

Plate 7

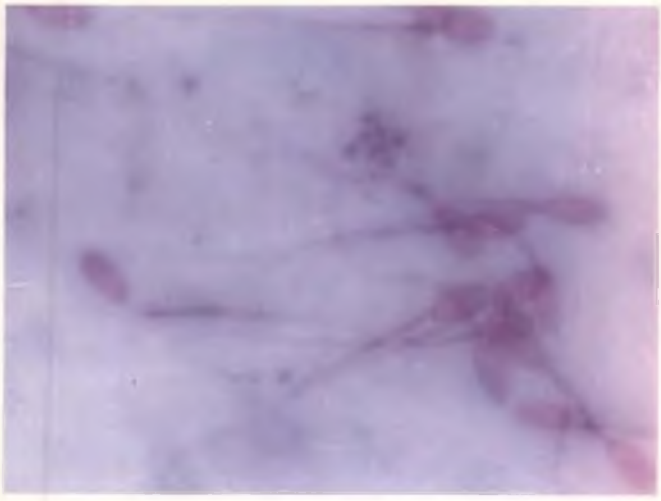


Plate 8

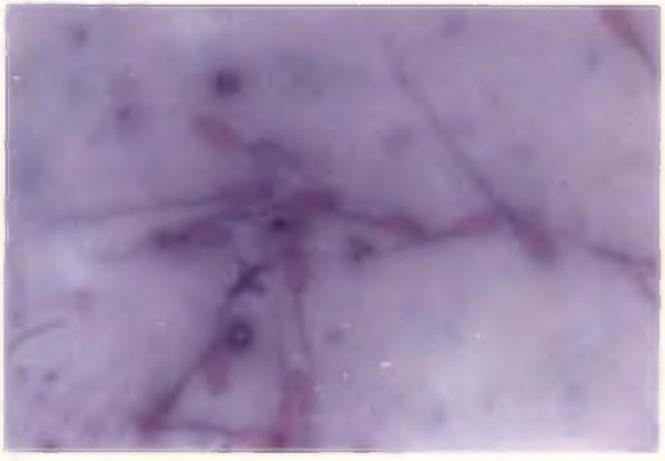


Plate 9

Results

4. RESULTS

4.1 Semen characteristics

Semen characteristics of 52 ejaculates collected by Gloved hand technique from three Large White Yorkshire (LWY) boars are presented in Tables 1, 2 and 3.

4.1.1 Volume

4.1.1.1 Total volume

The average total volume of ejaculate was found to be 209.50 ± 4.63 ml with a range of 155 to 265 ml (Table 1). The total volume of semen did not vary significantly between individual boars.

4.1.1.2 Gel free volume

The average gel free volume of semen was found to be 168.11±4.15 ml with a range of 120 to 225 ml (Table 1). There was no significant variation in the gel free volume of different boars.

4.1.1.3. Gel volume

Gel constituted an average of 41.38±0.79 ml with a range of 30 to 56 ml (Table 1). There was no significant variation in the gel volume of different boars.

4.1.2 Colour

The colour of gel free portion of boar semen was observed to vary from thin milky to thick milky. Out of the 52 ejaculates presently studied 24 samples were milky and eight thick milky in appearance. Twenty samples appeared to be thin milky (Fig.1).

4.1.3 Motility

The percentage of motile sperms in the ejaculate ranged from 40 to 80 with a mean of 71.73 ± 1.19 (Table 1). Analysis of the data revealed that there was no significant variation in the value of motile sperms in the semen samples obtained from different boars.

4.1.4 Hydrogen ion concentration

The average pH of semen was found to be 7.72±0.02 with a range of 7.5 to 7.9 (Table 1). The pH of the semen did not vary significantly between individual boars.

4.1.5 Sperm concentration

The concentration of spermatozoa in boar ejaculate varied from 261 to 329 millions per ml with a mean value of 288.28±2.73 millions per ml (Table 1). Variations in the sperm concentration between boars was not significant. It was observed that there was a negative correlation between sperm concentration and total volume of ejaculate.

4.1.6 Live sperm count

The percentage of live sperms varied from 81 to 92 with a mean value of 87.53±0.56 (Table 1). Analysis of the data revealed no significant variation in the percentage of live sperm count between boars.

4.1.7 Sperm morphology

4.1.7.1 Total abnormalities of sperm

The total abnormality of sperms ranged from 1.67 to 10.67 per cent with a mean of 6.88±0.40 per cent (Table 2).

Analysis of the data revealed no significant variation in the percentage of total abnormalities of sperms between boars. Also there was no significant correlation between total abnormality of sperm and live sperm count.

4.1.7.2 Head abnormalities

The percentage of total head abnormalities varied from 0.33 to 4.67 with a mean value of 2.59±0.19 (Table 2). Analysis of the data showed no significant variation in the percentage of head abnormalities between boars.

4.1.7.3 Mid piece abnormalities

The mean percentage of mid piece abnormalities observed was 2.33±0.16 with a range of 0.90 to 4.0 (Table 2). There was no significant variation in the mid piece abnormalities between boars.

4.1.7.4 Tail abnormalities

The mean percentage of tail abnormalities was found to be 2.10±0.18 with a range of 0.90 to 4 (Table 2). No significant variation in the percentage of tail abnormalities between boars was observed.

4.1.8 Protoplasmic droplets

4.1.8.1 Proximal protoplasmic droplet

The percentage of spermatozoa with proximal protoplasmic droplet ranged from zero to 3.30 with an average of 1.25±0.05 (Table 3). The variation in the percentage of proximal protoplasmic droplet between boars was found to be significant ($p < 0.05$) (Fig.3).

4.1.8.2 Distal protoplasmic droplet

The percentage of spermatozoa with distal protoplasmic droplet was found to vary from zero to 2.10 with an average of 1.13±0.06 (Table 3). Analysis of the data showed significant variation in the percentage of sperms with distal protoplasmic droplet between boars ($p<0.05$) (Fig.3).

4.2 Preservation

The mean percentage of progressively motile sperms preserved in BTS and stored at 15°C is given in (Table 4) and shown in (Fig. 4).

The mean percentage of progressively motile sperms stored in BTS at zero hour was 82 and decreased to

74.50, 68, 61.50, 53 per cent after an interval of 24, 48, 72 and 96 hours of preservation respectively (Fig.4). The variation in the preservability of boar spermatozoa was found to be non significant between individual boars, whereas there was a highly significant variation in the preservability of the spermatozoa between various hours of preservation ($p < 0.01$).

4.3 Artificial insemination

The data regarding duration of oestrus, conception rate, farrowing rate, gestation length and litter size of naturally served and artificially inseminated gilts and sows are presented in Table 5.

4.3.1 Duration of oestrus

The average duration of oestrus in naturally served gilts and sows were 42.0 ± 1.81 (36-48) and 59.0 ± 2.74 (48-72) hours respectively (Table 5). The corresponding figures for artificially inseminated gilts and sows where 41.0 ± 1.78 (36-48) and 60.0 ± 2.94 (48-72) hours respectively. Analysis of the data revealed a significant variation in the duration of oestrus between gilts and sows ($p < 0.05$). The method of mating (natural service or AI) had no significant effect on the duration of oestrus between the treatment groups.

4.3.2 Conception rate

The conception rates for the naturally served gilts and sows were 75 and 91.6 per cent respectively. The corresponding figures for artificially inseminated gilts and sows were 75 and 83.3 per cent respectively (Table 5).

4.3.3 Farrowing rate

The farrowing rate for both naturally served and artificially inseminated gilts and sows was 100 per cent (Table 5).

4.3.4 Gestation length

The mean gestation length for naturally served gilts and sows were 113.8±0.27 and 113.8±0.57 days (Table 5). Analysis of the data revealed no significant difference in the gestation length between gilts and sows.

The mean gestation length for artificially inseminated gilts and sows were 113.2 ± 0.96 and 115.3 ± 0.50 days (Table 5). Analysis of the data revealed a significant difference in the gestation length between gilts and sows ($p < 0.05$) (Fig.6).

4.3.5 Litter size

The average litter size for naturally served gilts and sows were 7.3 ± 0.63 and 6.9 ± 0.75. The corresponding figures for artificially inseminated gilts and sows were 8.2 ± 0.70 and 9.3 ± 0.37 (Table 5). Analysis of the data revealed a significant variation in litter size between naturally served and artificially inseminated sows ($p<0.05$) (Fig.7).

Table 1. Mean semen characteristics of three LWY boars

Boar No.	No of ejaculates	Total volume (ml)	Gel free volume (ml)	Gel volume (ml)	PH	Initial sperm motility (%)	Sperm concentration (millions/ml)	Live sperm count (%)
1/753	23	212.52 ± 7.07* (155-265)	171.52 ± 6.54* (120-225)	41.0 ± 1.08 * (30- 52)	7.73 ± 0.02* (7.5-7.9)	69.56 ± 2.22* (40-80)	295.04 ± 4.12* (262-329)	87.58 ± 0.98* (81-91.1)
1/754	15	197.06 ± 5.22* (160-252)	157.20 ± 5.00* (120–210)	39.86 ± 0.97* (30-45)	7.71 ± 0.04* (7.4-7.9)	74.00 ± 1.30* (70-80)	282.53 ± 4.73* (261-324)	87.42 ± 0.86* (82.6-91)
1/752	14	217.85 ± 11.0* (161-262)	174.21 ± 9.44* (125-214)	43.64 ± 2.02* (32-56)	7.71 ± 0.03* (7.5-7.9)	72.85 ± 1.94* (60-80)	283.35 ± 4.97* (258-314)	87.61 ± 1.12* (80.3-92)
Overall average		209.50 ± 4.63	168.11 ± 4.15	41.38 ± 0.79	7.72 ± 0.02	71.73 ± 1.19	288.28 ± 2.73	87.53 ± 0.56

* Non significant

Table 2 Mean sperm abnormalities (per cent) of three LWY boars.

Boar No.	No of ejaculates	Head abnormality	Mid piece abnormality	Tail abnormality	Total abnormal sperms
1/753	10	2.40±0.37* (0.60-4.33)	1.79±0.24* (0.90-3.0)	2.14±0.31* (0.90-3.67)	5.94±0.76* (1.67-9.00)
1/754	10	2.63±0.31* (0.33-4.0)	2.60±0.28* (1.0-4.0)	2.09±0.34* (0.67-4.0)	7.34±0.67* (3.00-9.34)
1/752	10	2.73±0.31* (1.67-4.67)	2.60±0.24* (1.67-4.0)	2.07±0.29* (1.34-4.0)	7.37±0.58* (5.00-10.67)
Overall average		2.59±0.19	2.33±0.16	2.10±0.18	6.88±0.40

* Non significant

Table 3. Mean Protoplasmic droplets (per cent) in semen of three LYW boars

Boar No.	No of ejaculates	Proximal protoplasmic droplet	Distal protoplasmic droplet
1/753	10	1.43±0.08[a] (0.60-3.30)	1.33±0.10[a] (0.30-3.60)
1/754	10	1.16±0.04[b] (0.67-1.34)	0.98±0.08[b] (0.00-1.67)
1/752	10	1.17±0.07[b] (0.00-1.67)	1.07±0.09[b] (0.00-2.00)
Overall average		1.25±0.05	1.13±0.06

Means bearing different superscript within the same column differ significantly ($p<0.05$)

Table 4 Mean motility (per cent) of boar semen preserved in BTS.

Boar No:	No of ejaculates	Zero hour	24 hours	48 hours	72 hours	96 hours
1/753	6	83.33±2.10 (80-90)	75.00±2.23 (70-80)	68.33±3.07 (60-80)	63.33±2.10 (60-70)	55.00±2.23 (50-60)
1/754	6	80.00±0.00 (80)	73.33±2.10 (70-80)	66.66±2.10 (60-70)	58.33±1.66 (50-60)	50.00±2.58 (40-60)
1/752	8	82.50±1.63 (80-90)	75.00±1.89 (70-80)	68.75±2.26 (60-80)	62.50±1.63 (60-70)	53.75±2.63 (40-60)
Overall average		82.00±0.92[a]	74.50±1.14[b]	68.00±1.38[c]	61.50±1.09[d]	53.00±1.47[e]

Means bearing different superscript within the same row differ significantly ($p<0.01$)

Table 5 Duration of oestrus, conception rate, farrowing rate, gestation length and litter size in naturally served and artificially inseminated gilts and sows

Parameter	Gilt (24)		Sow (24)	
	Natural service (12)	Artificial insemination (12)	Natural service (12)	Artificial insemination (12)
Duration of oestrus	42.0 ± 1.81[a] (36-48)	41.0±1.78[a] (36-48)	59.0±2.74[b] (48-72)	60.0±2.94[b] (48-72)
Conception rate	75.0	75.0	91.6	83.3
Farrowing rate	100.0	100.0	100.0	100.0
Gestation length (days)	113.8±0.27[a] (112-115)	113.2±0.96[a] (110-115)	113.8±0.57[a] (111-117)	115.3±0.50[b] (113-118)
Litter size	7.3±0.63[a] (4-10)	8.2±0.70[a] (5-12)	6.9±0.75[a] (3-10)	9.3±0.37[b] (8-11)

Means bearing different superscript within the same row differ significantly ($p<0.05$)

Fig 1. Colour of boar semen

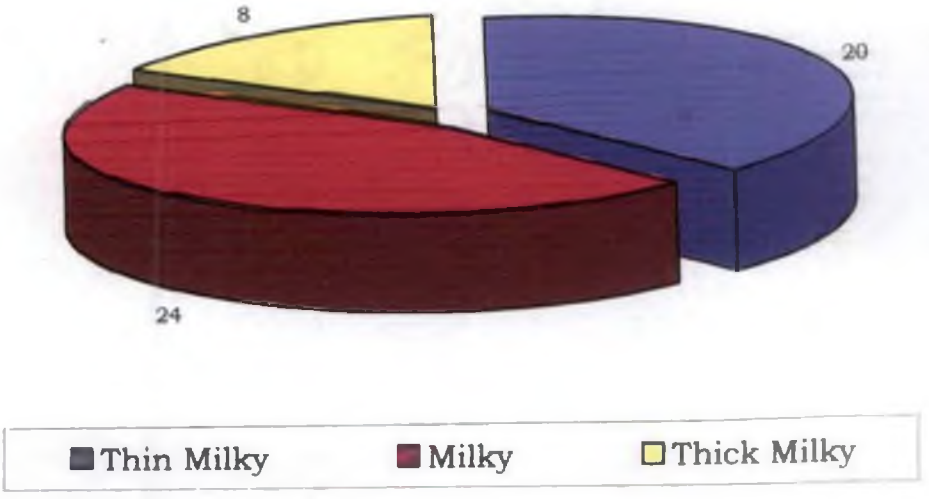


Fig 2. Comparison of total abnormal sperms (per cent) among three boars

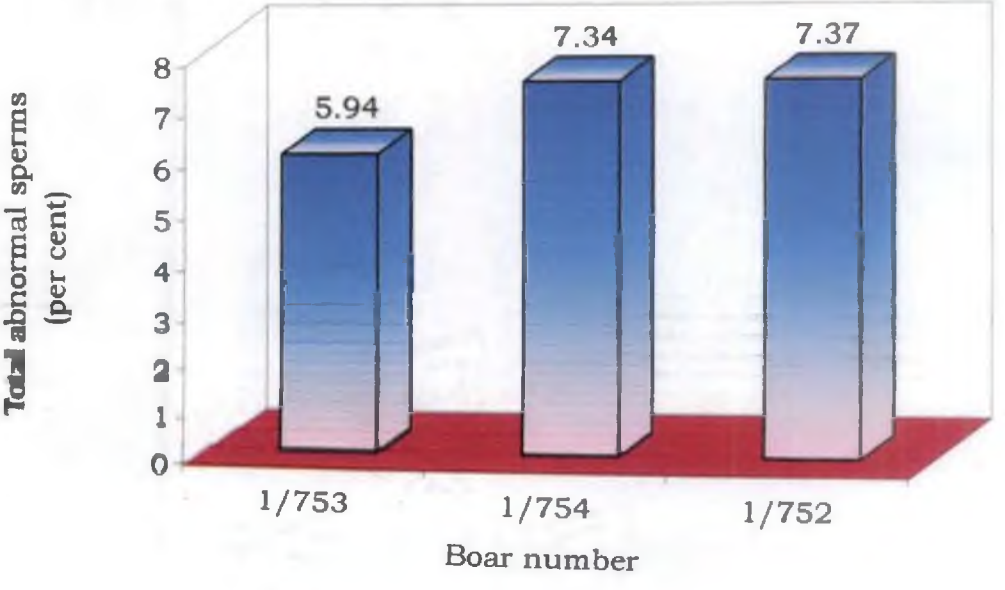


Fig 3. Comparison of mean protoplasmic droplets (per cent) among three boars

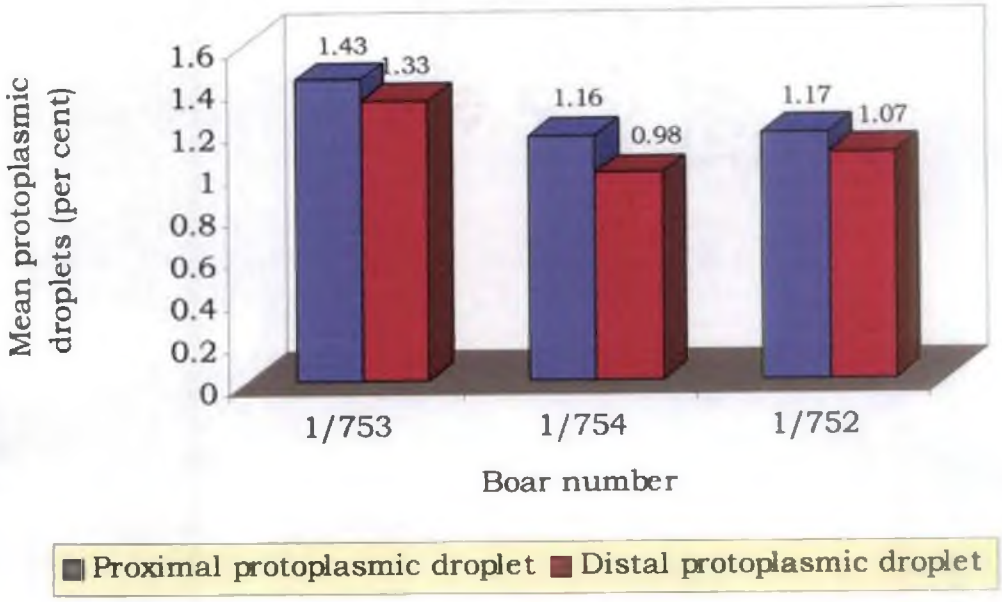


Fig 4. Preservability of boar semen extended in BTS at various hours of preservation

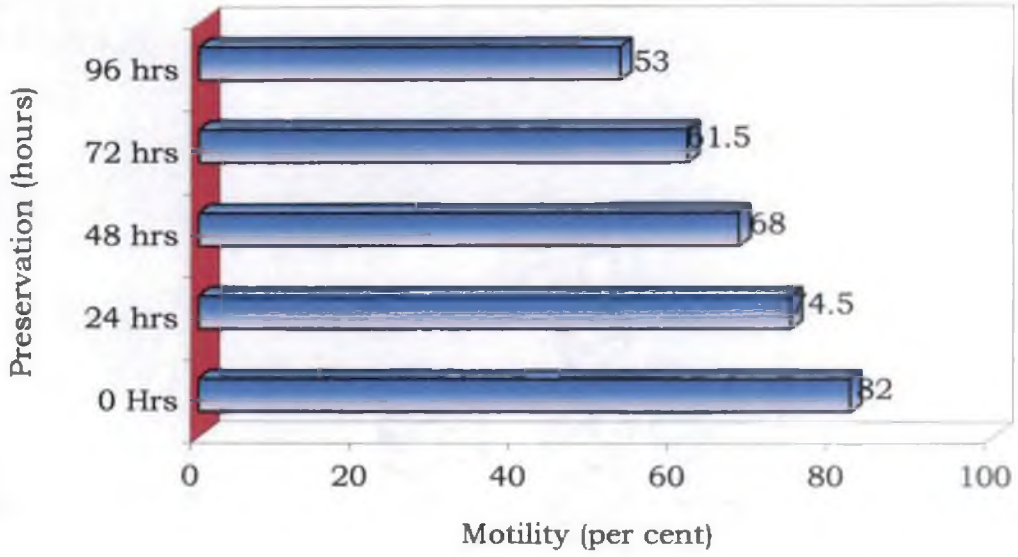


Fig 5. Comparison of conception rate among naturally served and artificially inseminated gilts and sows

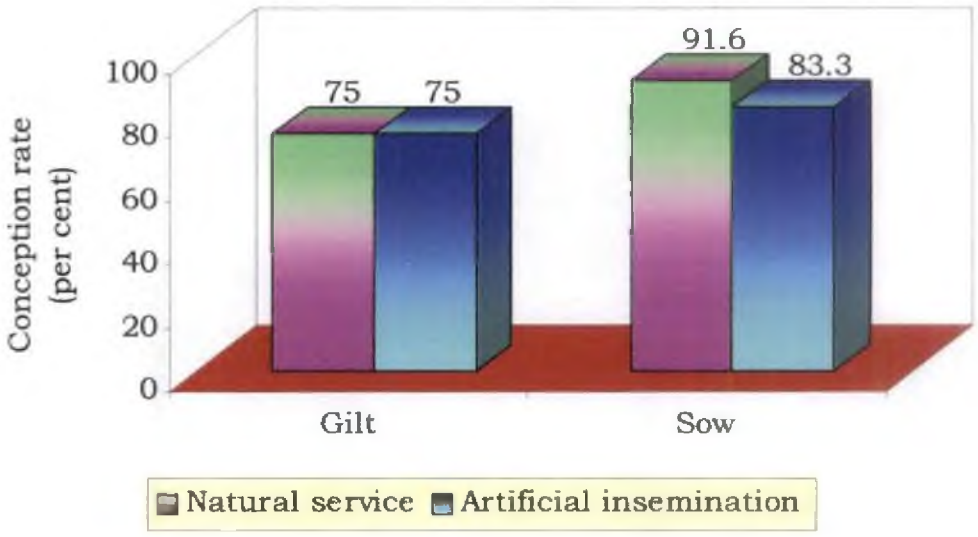


Fig 6. Compariosn of gestation length among naturally served and artificially inseminated gilts and sows

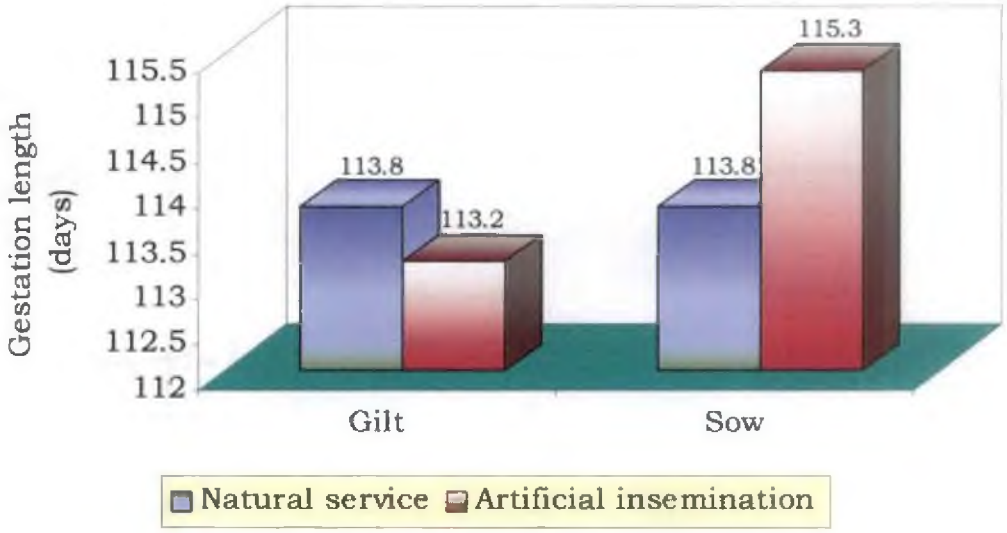
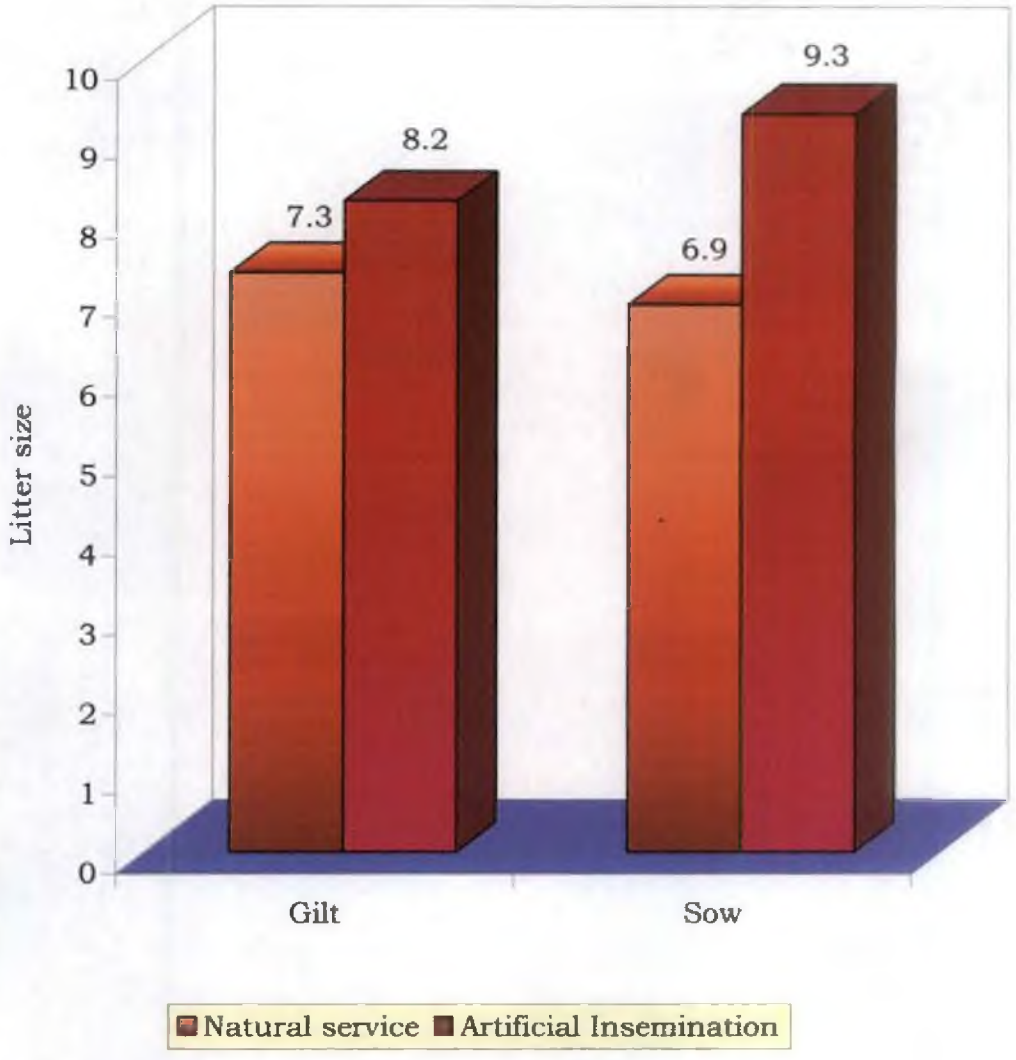


Fig 7. Comparison of litter size among naturally served and artificially inseminated gilts and sows



Discussion

5. DISCUSSION

5.1 Semen Quality

5.1.1 Semen volume

Boar is unique among the domestic animals that in relation to the body size it ejaculates an enormous volume of semen. Boar semen is emitted in three distinct fractions usually designated as pre-sperm, sperm rich and post sperm fractions. The first fraction has the highest proportion of fluid while the last is rich in gel (Mann, 1964).

The mean total volume of the ejaculate in the present study was found to be 209.50±4.63 ml. This value was found to be much lower than the values reported by the earlier workers (Aamdal and Hogset, 1957; King and Macpherson, 1973 and Murty, 1974). The findingwas in agreement with the values obtained by Swierstra and Rahnefeld (1967). However, Sreekumaran (1974) and Kennedy and Wilkins (1984) reported a much lower value for Yorkshire boars.

The average gel free volume was found to be 168.11 ± 4.15 ml. This valuewas found to be in close agreement with the findings of Swierstra and Rahnefeld (1967) and

Murty (1974). However, King and Macpherson (1973) obtained a higher value of 302 ml as the gel free volume. On the contrary, Sreekumaran (1974) obtained a lower value of 114.2 ml.

In the present study the gel volume was found to range from 30 to 56 ml with an average of 41.38 ± 0.79 ml. This constituted 19.7 per cent of the total ejaculate. This was essentially in keeping with the findings of Swierstra and Rahnefeld (1967) and King and Macpherson (1973) who reported that the gel portion comprised of 20.8 and 20.9 per cent of the total ejaculate respectively.

Analysis of the data revealed that there was no significant variation in the total volume, gel free volume and gel volume between individual boars. This might be due to the similarities in the age of boars and less number of observations taken for statistical analysis.

The variation in the values for semen volume reported by earlier workers might be due to the differences in the age of the boars, the method of the collection and the frequency of collection.

5.1.2 Colour

Of the 52 samples studied, 24 samples were noted to be milky, eight thick milky and 20 samples thin milky. These findings are similar to the values reported by Sreekumaran (1974) for colour of boar semen.

Johari (1956) found boar semen to be greyish white in colour. Melrose and Laing (1970) stated that the boar semen was greyish white and viscid. Roberts (1986) stated that the semen of boars with a low concentration of sperm cells would be pearly white to grey translucent colour with watery consistency.

5.1.3 Hydrogen ion concentration

The average pH of boar semen was found to be 7.72 ± 0.02 with a range of 7.5 to 7.9. This is in accordance with the reports in this regard (Johari, 1956; Foote *et al.*, 1959; Hancock, 1959; Murty, 1974 and Roberts, 1986). The pH of the semen did not vary significantly between boars as reported by Sreekumaran (1974).

5.1.4 Motility

It has been demonstrated that spermatozoa are propelled by lashing of the tail, a movement initiated by proximal centriole in the neck which forces the sperm head forward and causes it to rotate on its longitudinal axis. The average percentage of motile sperms in the present study of 71.7 per cent was in keeping with the values reported by earlier workers (Johari, 1956; Stratman and Self, 1961; King and Macpherson, 1973 and Roberts, 1986). However, Sreekumaran (1974) reported much lower value of 65.7 per cent than those in the present study. Analysis of the data showed no significant variation in the sperm motility between boars, which is contradictory to the findings of Murty (1974) and Sreekumaran (1974).

5.1.5 Sperm concentration

The value of 288.2 millions per ml being the average sperm concentration in the present investigation was nearer to the value reported by Foote *et al.* (1959). However, King and Macpherson (1973) and Kennedy and Wilkins (1984) recorded much higher values for mean sperm concentration.

In boars the sperm concentration decreased when the total volume of ejaculate increased (Ito *et al.*, 1948). The present study revealed a similar negative correlation between these two characteristics.

5.1.6 Live sperm count

The value of 87.5 per cent obtained for the percentage of live spermatozoa differed considerably from those recorded by Buttle and Hancock (1965), Campbell and Lingam (1965) and Kennedy and Wilkins (1984). The present value however is close to the values reported by Murty (1974) and Sreekumaran (1974).

5.1.7 Sperm abnormality

5.1.7.1 Total abnormalities

The total abnormality of sperms was in the range of 1.67 to 10.67 per cent with a mean of 6.88 ± 0.40 per cent. According to Holst (1949), the semen of normal boar would not contain more than 14 per cent abnormal spermatozoa. Masek (1963) reported that the percentage of abnormal sperms ranged from 1.92 to 18.56 with a mean of 9.43. Milovanov (1934) reported that normal boar might have 30 per cent abnormal sperms in the ejaculate.

The present data revealed that there was no significant variation in the occurrence of sperm abnormality in the ejaculate of different boars.

5.1.7.2 Head abnormalities

The percentage of head abnormality of sperms in the boar semen was found to vary from 0.33 to 4.67 with a mean of 2.59±0.19, which was close to the value of three per cent reported by Hancock (1959) for fertile boars and very close to the value of 2.5 per cent reported by Murty (1974). However, Sreekumaran (1974) reported a lower value of 1.66 per cent for normal boars. The value reported in the present study was well within the range suggested for normal boars. The data on analysis showed that there was no significant variation in the percentage of head abnormalities between boars.

5.1.7.3 Mid piece abnormalities

The incidence of mid piece abnormality of sperms varied from 0.90 to four per cent with a mean of 2.33 ± 0.16 per cent, which was close to the values reported by Hancock (1959) and Sreekumaran (1974). However, Murty (1974) reported much lower value of 0.47 per cent as mean mid piece

abnormality. No significant variation in the mid piece abnormality between boars could be detected.

5.1.7.4 Tail abnormalities

The tail abnormalities met with during the course of the present study were coiled tail, bent tail, broken tail, headless tail and detached tails. The total tail abnormalities ranged from 0.90 to four per cent with a mean of 2.10 ± 0.18. This was in close agreement with the value reported by Murty (1974). But Hancock (1959) and Sreekumaran (1974) reported a much higher value for the total tail abnormalities. The tail abnormalities did not vary significantly between boars.

5.1.8 Protoplasmic Droplet

5.1.8.1 Proximal protoplasmic droplet

The values reported for proximal protoplasmic droplet in fertile boars by Hancock (1959) and Sreekumaran (1974) were much higher than the value of 1.25 per cent recorded in the present study. However the mean value of 0.91 per cent for proximal protoplasmic droplet recorded by Murty (1974) was in close agreement with the value obtained in the present study.

The variation in percentage of proximal protoplasmic droplet between boars was found to be significant.

5.1.8.2 Distal protoplasmic droplet

The percentage of spermatozoa with distal protoplasmic droplet was in the range of zero to 2.10 with a mean of 1.13±0.06. This was in close agreement with the values reported by Murty (1974). However Hancock (1959) and Sreekumaran (1974) reported a much higher values.

Analysis of the data revealed that there was a significant variation in the percentage of sperms with distal protoplasmic droplet.

5.2 Preservation

The mean percentage of progressively motile sperms for semen diluted in BTS and preserved at 15°C was 82 per cent at zero hour and declined to 74.50, 68, 61.50, 52 per cent after an interval of 24, 48, 72 and 96 hours of preservation respectively. The findings of the present study was higher than the value reported by Aalbers *et al.* (1983)

who found that the sperm motility was 41 per cent at 72 hours of storage at 18°C.

The differences in the sperm motility may partly be due to the breed differences and different storage temperatures.

The results of the present study revealed that spermatozoal motility was best maintained upto 72 hours of storage (61.50 per cent) for semen diluted in BTS and stored at 15°C. These findings were similar to the works of Alexopoulos *et al.* (1996) who found that the preservability was best maintained upto 48 hours for semen diluted in BTS.

Analysis of the data showed that there was no significant variation in the preservability of spermatozoa between boars, whereas there was a highly significant variation ($p<0.01$) in the preservability of boar semen at various hours of preservation.

5.3 Artificial insemination

5.3.1 Duration of oestrus

The average duration of oestrus in naturally served gilts and sows were 42.0 ± 1.81 and 59.0 ± 2.74 hours

respectively. Thiswas in close agreement with the findings of Bednjfsontwikkeiling (1977). However, Niwa (1961) and Boender (1966) reported higher values for duration of oestrus in gilts and sows.

The average duration of oestrus in artificially inseminated gilts and sows were 41.0 ± 1.78 and 60.0 ± 2.94 hours respectively.

Analysis of the data revealed a significant variation ($p<0.05$) in the duration of oestrus between gilts and sows.

5.3.2 Conception rate

The conception rate for naturally served gilts and sows were 75 and 91.6 per cent. The corresponding figures for artificially inseminated gilts and sows were 75 and 83.3 per cent.

The lower conception rate for naturally served gilts was similar to the findings of previous workers (Pay and Davies, 1973; Libal and Wahistrom, 1976 and Macpherson *et al.*, 1977).

The conception rates among naturally served and artificially inseminated gilts and sows were almost equal. Cole and Cupps (1969) suggested that the conception rate in artificial insemination appeared to be equal to natural service. Whereas, Hancock (1958) found that the conception rate in artificial insemination was inferior to those obtained with natural service.

The high conception rate for artificial insemination obtained in the present study, which was nearer to that obtained with natural service, might be attributed to the effect of diluent and proper timing of insemination.

5.3.3 Farrowing rate

The farrowing rate for naturally served and artificially inseminated gilts and sows was 100 per cent. This was similar to the findings of Aalbers et al (1984) and Kim *et al.* (1998). However, Silaeva (1970) found a lower farrowing rate with semen diluted in Glucose- Chelate- Citrate- Sulphate diluent.

Most of the above workers reported 50 to 70 per cent farrowing rates on first service using fertile semen inseminated correctly at the proper stage of oestrus.

5.3.4 Gestation length

The gestation length in naturally served gilts and sows were 113.8 and 113.8 days. The corresponding figures for artificially inseminated gilts and sows were 113.2 and 115.3 days. These values were in keeping with the findings of Kao *et al.* (1996) who reported that the gestation length for naturally served and artificially inseminated sows was exactly 114 days.

Analysis of the data revealed that there was a significant variation ($p<0.05$) in the gestation length between artificially inseminated gilts and sows. This might be due to the differences in the number of foetuses carried upto term in gilts and sows.

The method of mating (natural service or artificial insemination) had no significant effect on the gestation length of gilts and sows.

5.3.5 Litter size

The average litter size in naturally served gilts and sows were 7.3 and 6.9. This is in close agreement with the findings of Stratman and Self (1961).

The average litter size for artificially inseminated gilts was 8.2. This was in agreement with the findings of Murty (1974). The average litter size for artificially inseminated sows was 9.3. The value obtained in the present study was similar to the findings of Machaty *et al.* (1992) who reported that sows inseminated with BTS diluted semen had a greater total number of piglets born than sows inseminated with MK semen.

Analysis of the data revealed that there was a significant variation in litter size between naturally served and artificially inseminated sows.

The variation in litter size between these treatment groups might be due to the inherent difficulties involved in establishing right time for insemination.

The conception rate following artificial insemination was almost equal to that obtained in natural service. The farrowing rate for BTS extended semen was 100 per cent. The litter size for artificially inseminated sows was more than naturally served sows. From the above results, it could be concluded that artificial insemination could well be practiced under commercial farming conditions.

Summary

6.SUMMARY

Twenty four each Large White Yorkshire (LWY) gilts and sows and six LWY boars were selected for the study. Out of six boars, three boars were used for natural service and the rest maintained for artificial insemination purpose.

A total of 52 ejaculates were collected at weekly intervals by the Gloved hand technique from three LWY boars of eight to twelve months age.

Mean values for the different semen characteristics are the following: total volume of the ejaculate 209.50 ml, gel free volume 168.11 ml, gel volume 41.38 ml, colour thin milky to thick milky, pH 7.70, motility 71.73 per cent, live sperm count 87.53 per cent, sperm concentration 288.2 millions per ml, total sperm abnormality 6.88 per cent; head abnormalities 2.59 per cent, mid piece abnormalities 2.33 per cent, tail abnormalities 2.10 per cent, proximal protoplasmic droplet 1.25 per cent and distal protoplasmic droplet 1.13 per cent.

Among boars no significant differences were found for most of the semen characteristics except proximal and

distal protoplasmic droplets.There was a negative correlation between total volume of ejaculate and sperm concentration. No significant correlation between total abnormality of sperm and live sperm count could be detected.

A total of twenty ejaculates from these boars were used for preservation studies. Belt's ville thaw solution (BTS) was used as the semen diluent. The gel free semen was mixed with the diluent at 1:2 ratio and the preservability was recorded after storage at 15°C upto 96 hours. The preservability (motility in percentage) was 82.0 per cent at zero hours of preservation. The mean percentage of progressively motile sperms decreased to 74.5, 68.0, 61.50 and 53.0 per cent at intervals of 24, 48, 72 and 96 hours of preservation respectively. No significant variation in the preservability of spermatozoa could be detected between boars, where as, there was a highly significant variation ($p<0.01$) in the preservability of the spermatozoa between hours of preservation.

Twelve Large White Yorkshire gilts and twelve sows (Group-I) and three boars were used for natural service. Each gilt and sow was mated twice during the oestrus. Twelve gilts and 12 sows (Group-II) were artificially inseminated twice during the oestrus with the semen diluted in BTS.

The mean duration of oestrus in naturally served gilts and sows were 42.0 and 59.0 hours respectively. The corresponding figures for artificially inseminated gilts and sows were 41.0 and 60.0 hours respectively. A significant variation ($p < 0.05$) in the duration of oestrus between gilts and sows were observed. The conception rates for naturally served gilts and sows were 75.0 and 91.6 per cent respectively. The corresponding figures for artificially inseminated gilts and sows were 75.0 and 83.3 per cent respectively. The farrowing rate was 100 per cent for both naturally served and artificially inseminated gilts and sows. The mean gestation length for both naturally served gilts and sows was 113.8 days. The corresponding figures for artificially inseminated gilts and sows were 113.2 and 115.3 days respectively. The litter size for naturally served gilts and sows was 7.3 and 6.9 respectively. The corresponding figures for artificially inseminated gilts and sows were 8.2 and 9.3 respectively. There was a significant variation ($p < 0.05$) in the litter size between naturally served and artificially inseminated sows.

Overall results suggest that artificial insemination could be performed under commercial farming conditions for effective disease control and increased productivity.

References

REFERENCES

Aalbers, J.G., Johnson, L.A., Rademaker, J.H.M. and Grooten, H.J.G. (1984). Use of boar spermatozoa for artificial insemination: Fertility and morphology of semen diluted in BTS and used for insemination within 24 hours (or) 25 to 48 hours after collection. *Proc. 10th Int. Congr. Anim. Reproduction and AI Urbana.* IL. **2**(180): 1-3.

Aalbers, J.G. Rademaker, J.H.M., Grooten, H.J.G. and Johnson, L.A. (1983). Fecundity of boar semen stored in BTS, Kiev, Zorlesco and Modena extenders under field conditions. *J. Anim. Sci.* **57**(Suppl. 1): 314-315.

Aamdal, J. (1966). Artificial insemination in pigs in Norway. *Wld. Review Anim. Prod.* Special Issue, II, 64.

Aamdal, J. and Hogset, I. (1957). Artificial insemination in swine. *J. Amer. Vet. Med. Assoc.* **131**: 59-64.

Alexopoulos, C., Boscos, C., Saratsis, P. Saoulidis, C. and Kyriakis, S. (1996). The effect of storage time and number of spermatozoa per insemination dose on semen characteristics and fertilising capacity of boar semen diluted with Belts ville thaw solution (BTS) extender. *A. Sci.* **62**:599-604.

*Almeida, F.R.C.L., Novak, S., Foxcraft, G.R. (2000). The time of ovulation in relation to oestrus duration in gilts. *Theriogenology*. **53**(7): 1389-1396.

*Almlid, T., Blichfeldt, T. and Stavne, S.E. (1984). Trials with different diluting fluids for boar AI semen. *10th Int. Congr. Anim. Reprod. AI* [USA]. Vol. II, Cited in *Anim. Breed. Abstr.* (1985). **53** (5): 400. Abstr. No. 2977.

Anderson, L.L. (1993). *Pig.* In *Reproduction in farm animals,* Hafez, E.S.E. (ed.), 6th ed., Lea and Febiger, Philadelphia. pp. 343-360.

Arthur, G.H., Noakes, D.E., Pearson, H. and Parkinson, T.J. (1996). *Veterinary reproduction and obstetrics.* 7th ed., WB Saunders company limited, Philadelphia. p: 622.

Ax, R.L., Dally, M., Didion, B.A., Lenz, R.W., Love, C.C., Varner, D.D., Hafez, B. and Bellin, M.E. (2000). *Semen Evaluation.* In *Reproduction in farm animals,* Hafez, E.S.E. and Hafez, B. (7th ed.). Lippincott Williams and Wilkins. pp. 365-375.

Bamba, K. and Cran, D.G. (1985). Effect of rapid warming of boar semen on sperm morphology and physiology. *J. Reprod. Fert.* **75**(1): 133-138.

Bane, A. (1959). Some results of studies on the semen of young boars and on artificial insemination in pigs. *Anim. Zootech.* (Paris) **8**: 15-20.

*Bednjfsontwikkeling (1977). Behaviour of gilts during oestrus. *Anim. Breed. Abstr.* **45** (6): 2865.

*Bennett, G.H. and O' Hagan, C. (1964). Factors influencing the success of artificial insemination of pigs. *5th Int. Congr. Anim. Reprod. AI* [Trento] 4: 481 –487. Cited in *Anim. Breed. Abstr.* (1965). **33** (2): 266. Abstr. No. 1463.

Bhuyan, D., Borgohain, B.N., Ahmed, K., Sarmah, B.C. and Chakravarthy, P. (1991). Physico-Morphological and Biochemical characteristics of semen of Hampshire and crossbred boars. *Indian J. Anim. Reprod.* **12** (1): 90-91.

*Blichfeldt, T., Almlid, T. and Stavne, S.E. (1988). Liquid preservation of boar semen in Kiev or BTS. A field comparison. *11th Int. Congr. Anim. Reprod. AI* [Dublin]. Cited in *Anim. Breed. Abstr.* (1989). **57**(8): 717 Abstr. No. 5657.

*Blom, E. (1950). A rapid staining method using Eosin – Nigrosin to distinguish between live and dead spermatozoa. *Nord. Vet. Med.* **2**: 58.

*Boender, J. (1966). The development of artificial insemination in pigs in Netherlands and the storage of boar semen. *Wld. Rev. Anim. Prod.* II. Special issue, 29. Cited in Roberts (1986).

*Buisson, D.M.D.F. and Dauzier, L.C. (1959). *Ann. Zootech.* **8**(suppl): 81. Cited in Buisson and Signoret (1970).

Buisson, D.M.D.F. and Signoret, J.P. (1970). Reproductive physiology and artificial insemination in pigs. *Vet. Rec.* **87**: 562-568.

Buttle, H.R.L. and Hancock, J.L. (1965). Sterile boars with knobbed spermatozoa. *J. Agri. Sci.* **65**: 255-260.

Campbell, E.A and Lingam, S.A. (1965). Artificial insemination of pigs in Australia. I. Training of boars and collection of samples. *Aus. Vet. J.* **41**: 147.

*Chamohoy, L.L., Abilay, J.P. and Paled, D.A. (1960). *Vet. Rec.* **78**(5): 159-167. Cited in Hughes, P.E. and Varley, M.A. (1980).

Cheng, W.T.K. (1988). Preservation of boar semen at 15°C. *J. Chinese Soc. Vet. Sci.* **14**: 339-350.

Clark, J.R., Komkov, A., Wiginton, L.W. and Tribble, L.F. (1986). Time of onset of oestrus in gilts. *Theriogenology.* **26**(5): 639-642.

Cole, H.H. and Cupps, P.T. (1969). *Reproduction in domestic animals.* 2nd ed., Academic press, New York and London. pp. 122.

*Corteel, J.M., Buisson, D.M.D.F. and Signoret, J.P. (1964). The modified French method of artificial insemination of pigs: recent results. *5th Int. Congr. Anim. Reprod. A.I.* (Trento) **4**:661-666. Cited in *Anim. Breed Abstr.* (1965). **33**(2): 267.

Dziuk, P.J. (1958). Dilution and storage of boar semen. *J. Anim. Sci.* **17**: 548-553.

Dziuk, P.J. and Henshaw, G. (1958). Fertility of boar semen artificially inseminated following *in vitro* storage. *J. Anim. Sci.* **17**: 554-558.

Foote, R.H., Young, D.C., Turkheimer, A.R. and Bratton, R.W. (1959). Collection, preservation and artificial insemination of boar semen. *Ann. Zootech.* 27-30.

*Gerrits, R.J., Graham, E.F. and Cole, C.L. (1962). *J. Anim. Sci.* **21**: 1022.

Gibson, C.D. and Johnson, R.G. (1980). *Evaluation of the boar for breeding soundness.* In Current Therapy in Theriogenology, Morrow, D.A. (ed.), W.B. Saunders company, Philadelphia. pp. 1034-1037.

Gordon, I (1997). *Controlled reproduction in the pig.* CAB International. University Press, Cambridge.pp. 35-39.

*Hancock, J.L. (1958). The fertility of natural and of artificial matings in the pig. *Proc. Soc. Stud. Fertil.* **9**: 146.

Hancock, J.L. (1959). Semen and testis characteristics and sexual behaviour of boars. *J. Agri. Sci.* (Cambridge). **53**: 313-326.

Hancock, J.L. and Hovell, G.L.R. (1959). The collection of boar semen. *Vet. Rec.* **71**: 664.

Hmar, L. (1993). Effect of weight at weaning and plane of feeding on the onset of puberty in gilts. M.V.Sc. Thesis, College of Veterinary and Animal Sciences, Mannuthy. pp. 78.

*Holst, S.J. (1949). The semen of sterile boars. *Proc. 14th Int. Vet. Congr. III. Sect. 4(C): 118.* Cited in Roberts (1986).

Hughes, P.E. and Cole, D.J.A. (1975). Reproduction in the gilts. I. The influence of age and weight at puberty on ovulation rate and embryo survival in the gilt. *Anim. Prod.* **21**: 183-189.

Hughes, P.E. and Varley, M.A. (1980). *Reproduction in the pig.* Butterworth and Co. (Publishers) Ltd. pp. 212.

*Ito, S., Niwa, T. and Kudo, A. (1948). Studies on the artificial insemination in swine. I. On the method of collection of semen and the condition of ejaculation. *Zoot. Explt. Sta. Res. Bull.*, Japan, **55**:1.

Johari, M.P. (1956). The Gel Mass in the semen of the boar. *Vet. Rec.* **68**: 158-163.

*Johnson, L.A. and Aalbers, J.G. (1984). Artificial Insemination of Swine: Fertility using several liquid semen diluents. *Proc. 8th Int. Pig. Vet. Soc. Congr.*, Ghent, Belgium. Cited in *Anim. Breed. Abstr.* (1985), **53** (2)

Johnson, L.A., Aalbers, J.G., Willems, C.M.T., Rademaker, J.H.M. and Rexroad, Jr.C.E. (1982). Use of boar spermatozoa for artificial insemination. III. Fecundity of boar spermatozoa stored in Belts Ville liquid and Kiev extenders for three days at 18°C. *J. Anim. Sci.* **54**: 132-136.

Johnson, L.A. Aalbers, J.G., Willems, C.M.T. and Sybesma, W. (1979). Effectiveness of fresh and frozen boar semen under practical conditions. *J. Anim. Sci.* **49**(suppl. 1): 306.

*Kalev, G. and Zagorski, D. (1964). Some diluents for boar semen. *5th Int. Congr. Anim. Reprod. A.I* (Trento), IV: 673-681. Cited in *Anim. Breed. Abstr.* (1965) **33**(2): 267. Abstr. No. 1469.

Kao, Z.C., Lai, Y.Y., Chang, H.L. and Wu, M.C. (1996). Gestation period and litter size of artificially inseminated sows. *J. Chinese Soc. Anim. Sci.* **25**(4): 403-416.

Kennedy, B.W. and Wilkins, J.N. (1984). Boar, breed and environmental factors influencing semen characteristics of boars used in artificial insemination. *Can. J. Anim. Sci.* **64**: 833-843.

Kim, I.C., Lee, J.H., Kim, H.J., Choi, D.Y., Son, D.S. and Park, C.S. (1998). Effect of number of motile sperm cells per dose in the artificial insemination of liquid boar semen. *Proc. VIIIth Wld. Conf. Anim. Prod*: 852-853.

King, G.J. and Macpherson, J.W. (1973). A comparison of two methods for boar semen collection. *J. Anim. Sci.* **36**(3): 563-565.

Kuo, Y.H., Huang, S.Y. and Lee, K.H. (2000). Effect of sperm number and semen type on sows reproductive performance in sub-tropical areas. *Asian-Aus. J. Anim. Sci.* **13**(1): 6-9.

*Lagerlof, N. and Carlquist, H. (1961). The semen of boars of the Yorkshire breed between the ages of five and nine months. *Proc. IV. Int. Congr. Anim. Reprod.* (Hague). IV: 818-821.

Libal, G.W. and Wahistrom, R.C. (1976). Effect of early breeding on gilt reproduction. *J. Anim. Sci.* **42**: 1359.

*Lindstrom, U. (1966). Unpublished data. *Wld. Rev. Anim. Prod.* Special issue, II. pp.72.

Machaty, Z., Takacs, T. and Gathy, I. (1992). Fertilizing capacity of boar semen diluted with Belts ville TS (BTS) and Modified Kiev (MK) extenders in relation to storage time and number of spermatozoa per insemination dose. *Anim. Reprod. Sci.* **29**: 289-295.

Macpherson, R.M., Hovell, F.D. and Jone, D.A.S. (1977). Performance of sows mated at puberty or second or third oestrus and carcass assessment of once bred gilt. *Anim. Prod.* **24**: 333-342.

*Madden, D.H.L. (1959). Progress in the artificial insemination of swine – some factors influencing fertility levels in its field application. *Vet. Rec.* **71**: 227.

Mann, T. (1964). *The Biochemistry of semen and of the male reproductive tract.* 2nd ed. Methuen and co. Ltd. London. Johnwiley and Sons Inc., New York. pp. 94.

*Masek, N. (1963). Some indices characteristics of the quality of boar ejaculates. *Anim. Breed. Abstr.* **32**. Abstr. No. 3201.

Maule, J.P. (1962). *The semen of animals and artificial insemination.* Commonwealth agricultural bureaux, Central press, Great Britain. pp. 263-277.

Melrose, D.R. (1963). Artificial insemination in the pig. A review of its development. *Br. Vet. J.* **119**: 532-543.

Melrose, D.R. and Laing, F.A. (1970). *Fertility and infertility in domestic animals.* Edited by Laing, Bailliere, Tindall and Cox, London. pp. 60

*Melrose, D.R. and O'Hagan, C. (1961). Proc. IVth *Int. Congr. Anim. Reprod.* (The Hague) **4**: 855.

*Mercat, M.J., Floch, M., Pellois, H. and Runavot, J.P. (1999). Consequences of a reduction in the number of spermatozoa per insemination dose on reproductive performance in the sow. *Jounee's de la recherché porcine en france.* 31: 53-58. Cited in *Anim. Breed Abstr.* (2000). **68**(10): 887. Abstr. No. 6084.

*Milovanov, V.K. (1934). [Cited by Polge (1956)] Probl. zivotn. 4:31. Anim. Breed. Abstr. 1:112

Mohanty, S. and Nayak, J.B. (1986). Reproductive performance of Large White Yorkshire pigs and their crosses with indigenous pig in hot-humid climate of Orissa. A note. *Indian J. Anim. Prod. Mngmt.* **2**(3): 134-137.

*Munoz, M.B., Mansilla, M.A. and Soto, G.G. (1999). Effect of number of inseminations and oxytocin treatment on fertility and prolificacy of adult sows. *Advance in production Animal.* **24**(1/2): 83-89.

Murty, R.P. (1974). Physical characteristics of boar semen, preservation and artificial insemination in swine. M.Sc. Thesis, Andhra Pradesh Agricultural University. pp. 8-10, 15-24, 38-46.

Murty, R.P. and Rao, R.A. (1975). Preservation of boar semen. *Indian Vet. J.* **52**: 415 –420.

*Niwa, T. (1961). Researches and practices in the artificial insemination of pigs. *Proc.* IV *Int. Congr. Anim. Reprod.* The Hague, **1**:83. Cited in Roberts (1971).

*Niwa, T., Mizuho, A. and Soejima, A. (1959). *Bull. Nat. Inst. Agri. Sci.* (Chiba). **18**: 45.

*Ooi, S. (1970). Motility and fertility studies with boar semen stored in Illini Variable Temperature (I.V.T) diluent. *Kajian Vet.* 2: 165-170. Cited in *Anim. Breed Abstr* (1972). **40**(2): 336. Abstr. No. 2167.

Pandey, R. P. and Singh, B.K. (1998). Fecundity of preserved boar semen with Bts, Kiev and BL-1 Dilutor. *Indian J. Anim. Reprod.* **19**(1): 66-67.

Pay, M.G. and Davies, F.E. (1973). Growth, feed consumption and litter production of female pigs mated at puberty and at low body weight. *J. Anim. Sci.* **47**: 780.

Perry, E. J. (1960). *The Artificial Insemination of farm Animals.* 3rd ed., Rutgers University Press, Brunswick, New York. pp. 258-270.

*Podany, J. (1964). A contribution to the artificial insemination of the pig. *5th Int. Congr. Anim. Reprod. A.I.* (Trento) **4**: 562-565. Cited in *Anim. Breed Abstr* (1965). **33**(2): 268. Abstr. No. 1473.

Polge, C. (1956). Artificial insemination in pigs. *Vet. Rec.* **68**: 62.

Polge, C. and Rowson, L.E. (1956). The practical application of artificial insemination in pig breeding. *Vet. Rec.* **68**: 952.

Pursel, V.G., Johnson, L.A. and Schulman, L.L. (1974). Acrosome morphology of boar spermatozoa during *in vitro* aging. *J. Anim. Sci.* **38**: 113-116.

Pursel, V.G., Schulman, L.L. and Johnson, L.A. (1973). Effect of holding time on storage of boar spermatozoa at 5°C. *J. Anim. Sci.* **37**: 785-789.

Rao, V.B., Venkatamunichetty, A., Ramachandraiah, S.V. and Sreeraman, P.K. (1991). Preservation of native Boar Semen. *Indian J. Anim. Reprod.* **12**(2): 148-150.

Revell, S.G. and Glossop, C.E. (1989). A long-time ambient temperature diluent for boar semen. *Anim. Prod.* **48**: 579-584.

Roberts, S.J. (1986). *Veterinary obstetrics and Genital diseases (Theriogenology).* 3rd ed., Edward Brothers, Inc. Ann Arbor, Michigan. pp. 872-893.

*Rommele, O. (1927). Biologische und physiologische untersuchungen am sperma und am seheidensekret des Rinder im Hinblick auf die kunstliche besamung. *Zool. Jahrb. Abt.* Cited in Vijayakumaran and Iyer (1980).

Sharda, D.P. (2000). *Swine production.* Indian Council of Agricultural Research. M/s Press, New Delhi. pp. 99

*Signoret, J.P. and Buisson, D.M.D.F. (1961). Study of the behaviour of sows in oestrus. *Proc. IV^th Int. Congr. Anim. Reprod.* II: 171.

*Silaeva, M.V. (1970). Diluent for extension and storage of boar semen. *Veterinariya, Mork.* **47**.(12). 81-83. Cited in *Anim. Breed. Abstr.* (1971). **39**(2): 334. Abstr. No. 2207.

*Simunic, B. (1964). A practical catheter for inseminating sows. *Vet. Glasn.* **18**: 813-815. Cited in *Anim. Breed Abstr* (1965). **33**(2): 269. Abstr. No. 1481.

*Simunic, B. and Abram, K. (1968). Artificial insemination of pigs in Yugoslavia. *Vet. Glasn.* **22**: 429-433. Cited in *Anim. Breed Abstr* (1968). **36**(4): 641. Abstr. No. 3906.

Singleton, W. (1997). Training Boars. *www.ansc. purdue. edu.* pp. 1-2.

Snedecor, G.W. and Cochran, W.G. (1967). *Statistical Methods.* 6th ed., Iowa state university press, Ames, Iowa. pp. 237 - 328.

Sreekumaran, T. (1974). Studies on boar semen. M.Sc. Thesis, University of Calicut. pp. 6-43, 72-80.

*Stevernik, D.W.B., Soede, N.M. and Kemp, B. (1999). Registration of oestrus duration can help to improve insemination strategies at commercial pig farms. *Reproduction in Domestic animals*. Cited in *Anim. Breed. Abstr.* (2000). **68**(1): 46 Abstr. No. 276.

Stratman, F.W. and Self, H.L. (1960). Effect of semen volume and number of sperm on fertility and embryo survival in artificially inseminated gilts. *J. Anim. Sci.* **19**(4): 1081-1088.

Stratman, F.W. and Self, H.L. (1961). Comparison of natural mating with artificial insemination and influence of semen volume and sperm numbers on conception, embryo survival and litter size in sows. *J. Anim. Sci.* **20** (4): 708-711.

Swierstra, E.E. (1973). Influence of breed, age and ejaculation frequency on boar semen composition. *Can. J. Anim. Sci.* **53**: 43.

Swierstra, E.E. and Rahnefeld, G.W. (1967). Semen and testis characteristics in young Yorkshire and Lacombe boars. *J. Anim. Sci.* **26**(1): 149-157.

Tamuli, M.K. and Rajkonwar, C.K. (1988). A note on training of boars for collection of semen. *Indian. J. Anim. Reprod.* **9**(2): 118-122.

Tamuli, M.K., Rajkonwar, C.K. and Goswami, B.K. (1986). Preservation of boar semen. *Indian J. Anim. Reprod.* **7**(2): 70-72.

Vijayakumaran, V. and Iyer, N.C.P. (1980). Extenders for preservation of boar semen. *Kerala J. Vet. Sci.* **11**(2): 215-220.

*Walters, J.R., Hooper, P.M. and Green, C.G. (1984). The utilization of artificial insemination in commercial pig improvement. E.A.A.P., *Wld. Rev. Anim. Reprod.* **22**(2). Cited in Pandey and Singh (1998).

Wiggins, E.L., Grummer, R.H. and Casida, L.E. (1951). Minimal volume of semen and number of sperm for fertility in artificially inseminated swine. *J. Anim. Sci.* **10**: 138.

Young, L.G. and King, G.J. (1981). Reproductive performance of gilt bred on third vs. first oestrus. *J. Anim. Sci.* **53**: 19.

* Reference not consulted in original

EVALUATION OF BOAR SEMEN EXTENDED IN BELT'S VILLE THAW SOLUTION

By
Kantharaj. S.

ABSTRACT OF A THESIS
submitted in partial fulfilment of the
requirement for the degree of

Master Of Veterinary Science

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University

Department of Animal Reproduction
COLLEGE OF VETERINARY AND ANIMAL SCIENCES
MANNUTHY, THRISSUR,
KERALA

2001

ABSTRACT

An experiment was conducted to study the effect of natural service and artificial insemination on reproductive performance of pigs and to assess the fertility and preservability of boar semen extended in Belt's ville thaw solution.

Twenty-four Large White Yorkshire (LWY) gilts, twenty-four sows and six LWY boars were selected for the study. Out of six boars, three boars were used for natural service and the other three maintained for artificial insemination purpose.

A total of 52 ejaculates were collected from boars maintained for artificial insemination purpose at weekly intervals by the gloved hand technique. Among these boars, no significant differences were found for most of the semen characteristics except proximal and distal protoplasmic droplets, which were significantly different between boars ($P<0.05$). Negative correlation between total volume of ejaculate and sperm concentration was observed. No significant correlation between total sperm abnormalities and live sperm count could be detected.

Twenty ejaculates from these boars were used for preservation studies. No significant variation in the preservability of spermatozoa could be detected between

boars, whereas, there was a highly significant difference ($p<0.01$) in the preservability of spermatozoa between hours of preservation.

Twelve LWY gilts and twelve sows (Group-I) and three boars were used for natural service. Each gilt and sow was mated twice during the oestrus. Twelve gilts and twelve sows (Group-II) were artificially inseminated twice during the oestrus with the semen diluted in BTS.

There was a significant difference ($P<0.05$) in the duration of oestrus between gilts and sows. There was a marginal difference in the conception rates between naturally served and artificially inseminated gilts and sows. The farrowing rate was 100 per cent for both naturally served and artificially inseminated gilts and sows. There was a significant difference in the gestation length between artificially inseminated gilts and sows. There was a significant variation in the litter size between naturally served and artificially inseminated gilts and sows.

It can be concluded from the study that artificial insemination in pigs could well be performed under commercial farming conditions for effective disease control and increased productivity.