

The effect of the age of eggs and age of the diluent used on the quality of frozen semen*

G. VEERAIAH¹, K. BABU RAO², K. VENUGOPAL NAIDU³

Department of Animal Reproduction & Gynaecology,
College of Veterinary Science, Tirupathi - 517 502.

ABSTRACT

A significant ($P < 0.05$) and progressive decline was recorded in percentage of post thaw motility (52.78 to 24.72); Live sperms (54.06 to 26.67) and acrosomal integrity (86.89 to 69.08) in diluents constituted with increased age of eggs (from fresh to two weeks). Similarly significant ($P < 0.05$) lower values were recorded in two weeks aged diluents in respect to percentage of post thaw motility (28.89), Live sperms (29.11) and acrosomal integrity (58.89) than fresh diluent, four day and one week old diluents.

—x—x—x—

The most commonly used diluent for bull semen contains Tris, Citric acid, Fructose, Glycerol and Egg yolk. The Yolk affords protection to sperm cells during freezing and thawing, but yolk from old eggs and old diluents may be detrimental to semen quality (Willington, 1988). Therefore eggs purchased from market are considered undesirable because they are of unknown age. Hence the present study was carried out to determine the effect of the age of the eggs used to constitute the diluent and age of the diluent on the Post thaw motility, Live sperms and acrosomal integrity.

MATERIALS AND METHODS

The present study was carried out at Cattle Project, Lam Farm, Guntur on Ongole Bull semen. Eggs and diluents were stored at refrigerator temperature. The yolk from fresh eggs and eggs stored for one week

and two weeks were used for constituting the diluents. Freshly constituted diluent and diluents constituted and stored for four days, one week and two weeks were utilised for freezing of semen. Eighteen ejaculates from six bulls were divided into two portions after initial evaluation. One portion was used to study the effect of the age of eggs and another portion was used to study the effect of the age of diluents on frozen semen quality. After initial evaluation the semen was diluted with respective diluents by keeping 40 millions of sperms per milliliter at 37°C and cooled to 5°C over about an hour in cold handling cabinet. Then the semen was packed in 0.5 ml. straws and equilibrated for 5 hours before freezing in liquid nitrogen vapours for 8 minutes and then stored in liquid nitrogen. Post thaw motility, live sperms and acrosomal integrity were assessed after 24 hours of freezing by adopting the standard procedures, by thawing the straws at 37°C for 30 seconds. The data was analysed statistically as per Snedecor and Cochran (1967).

* Part of M.V.Sc. Thesis submitted to A.P. Agricultural University, Hyderabad by the first author.

PRESENT ADDRESS:

1. Vety. Asst. Surgeon, Vety. Dispensary, Chodavaram, W.G. Dist., A.P.
2. Research Officer, (Animal Reproduction) Cattle Project, Lam Farm, Guntur.
3. Associate Professor, College of Veterinary Science, Tirupathi.

RESULTS AND DISCUSSION

The effect of the age of eggs and age of the diluents on post thaw motility, Live sperms and acrosomal integrity was presented in Table-1. A significant ($P < 0.05$) and progressive decline in mean percentage of post thaw motility from 52.78 to 24.72, live sperms from 54.06 to 26.67 and acrosomal integrity from 86.89 to 67.06 was observed as age of stored eggs used for constituting the dilutor increased. Significant difference was also observed in between the age of the eggs in respect to the means of above mentioned parameters. Similar results were obtained with ageing of diluent containing yolk and in between ageing of diluents used. The results obtained in the present study in respect to the age of eggs and also the age of diluent were in agreement with the result of Willington (1988). The

decrease in mean percentage of post thaw motility, live sperms and acrosomal integrity in respect to the aged eggs and the aged diluents might be due to the lack of protective action of lecithin during extension, freezing and thawing of semen, since the denature and breakdown of lecithin occurs due to prolonged storage of eggs and diluent at 5°C (Thompson 1944). The observations in the present study indicates that the eggs used in the preparation of dilutor and the dilutor should be as fresh as possible for semen extension and freezing to get high post thaw motility, live sperms and acrosomal integrity.

Acknowledgements: The authors are thankful to Senior Scientist (Animal Breeding) Cattle Project, Lam Farm, Guntur for providing the necessary facilities to undertake this study.

Table 1. Effect of the age of eggs and the age of diluent on frozen semen quality.

Post thaw Semen Characters	Age of Eggs			Age of Diluent			
	Fresh	1 Week	2 Week	Fresh	Four Days	1 Week	2 Week
motility (%)	52.78 ^a ±1.99	32.78 ^b ±1.72	24.72 ^c ±1.39	52.78 ^a ±1.98	43.89 ^b ±2.24	36.11 ^a ±1.91	28.89 ^d ±2.06
Live sperms (%)	54.06 ^a ±2.06	35.72 ^b ±1.66	26.67 ^c ±1.13	54.06 ^a ±2.06	45.44 ^b ±1.96	37.39 ^c ±2.17	29.11 ^d ±1.41
Acrosomal integrity (%)	86.89 ^a ±0.61	73.83 ^b ±1.13	67.06 ^c ±1.57	86.89 ^a ±0.61	76.44 ^b ±0.79	66.72 ^c ±1.41	58.89 ^d ±1.67

Means bearing different superscripts differ significantly ($P < 0.05$).

REFERENCES

- Snedecor Gand Cochran WG (1967) Statistical Methods 6th Edn., The Iowa State University Press, Ames, IOWA, U.S.A.
- Thompson WL (1944) Bull. Univ. Pittsburgh. 40: 311-317 cited in the Avain Egg Romanoff AL and Romahoff AJ 1949 John Wiley & Sons, INC, New York, Champman & Hall Limited, London, P.517, 518.
- Willington JA (1986) The effect on frozen semen quality of age of eggs used in UHG/Egg yolk/glycerol diluent and the effect of the age of this diluent. In 11th International Congress on Animal Reproduction and Artificial Insemination, University College Dublin, Ireland June 26-30-1988 Volume 3. Brief Communications. cited Animal Breeding Abstracts 57(8): 5563.