Changes on the quality characteristics of stored semen during reproductive period of Hybrid toms

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In all animal species increasing age affects the reproductive performance. Since little is currently known about the effects of aging on the qualitative characteristics of turkey semen, the purpose of this study was to investigate the quality changes of the semen stored for 48 h at 5°C during the reproductive period of Hybrid toms. Hybrid turkey toms, 32-52 weeks of age, reared in a conditioned poultry house with artificial lighting (16 h light: 8 h dark) and free access to feed and water, were used. Semen was collected by dorso-abdominal massage and pooled (n=4 ejaculates/pool). In total 6 semen pools were analysed when Hybrid males were 32, 36, 40, 44, 48, 52 weeks of age. The pools were diluted with Beltsville Poultry Semen Extender (BPSE) and stored at 5°C for 48 h. The motility, viability and membrane integrity of sperm were measured at 3, 24 and 48 h of storage. The concentration of fresh semen was also evaluated. Toms ageing was accompanied by a significant decrease of the concentration, viability and motility (P<0.05) of fresh semen beginning from 44 weeks, while for membrane integrity of sperm (P<0.05) from 48 weeks. The deleterious effect of in vitro storage in turkey semen is well-known and it was also observed in our previous researches. The liquid storage of semen for 48 h significantly deteriorated the quality of semen but this occurred in a marked manner with the increasing of tom age. Indeed, at 44 weeks, a decrease of about 40-50% for all the quality characteristics of semen after 48 h of storage was observed. Our results suggest that the changes of quality semen in both fresh and stored semen markedly occurred at 44 weeks in Hybrid strain. Therefore, to improve the reproductive performance of ageing
males, it could be useful the addition of antioxidants to the tom diet or to the semen extender during the critical reproductive period.

**Keywords**: tom; semen quality; storage; aging

**Introduction**

Semen of the domestic turkey usually cannot be stored longer than 6 h without a loss of fertilizing capability (Thurston, 1995; Iaffalano et al., 2003). This problem is enhanced with animal aging (Douard et al., 2004; Iaffalano et al., 2005). Thus, investigations of factors determining the quality of fresh and stored semen would be beneficial to the turkey breeding industry since commercial production relies almost entirely on artificial insemination. Since little is currently known about the effects of aging on the qualitative characteristics of turkey semen, the purpose of this study was to investigate the quality changes of the semen stored for 48 h at 5°C during the reproductive period of Hybrid toms.

**Materials and methods**

Hybrid turkey toms 32-52 weeks of age were used and reared in a temperature regulated poultry house with artificial lighting (16 h light: 8 h dark) and free access to feed and water.

Semen was collected by dorso-abdominal massage (Burrows and Quinn, 1937) and pooled (n=4 ejaculates/pool). In total 6 semen pools were analysed when Hybrid males were 32, 36, 40, 44, 48, 52 weeks of age. Semen pool was thoroughly mixed, and diluted 1:1 with Beltsville Poultry Semen Extender (BPSE, Sexton, 1977). The total spermatozoa concentration was determined by using a Thoma-Zeiss chamber and then the semen was again diluted with BPSE to obtain a concentration of 2 x 10^9 spermatozoa/ml. The semen was placed in a beaker containing water at 25°C and stored at 5°C. The sperm mobility was assessed using an Accudenz® solution and determined the sperm motility test (SMT) that was expressed as O.D. (Donoghue et al., 1998) while a dual association of stains was used to assess sperm viability according to Donoghue et al., (1995). To determine membrane integrity, a hypo-osmotic H_2O test was used (Donoghue et al., 1996). Data were submitted to ANOVA (SPSS, 2003) and percentages were transformed into arc sine before analysis.
Results and discussion

The effects of aging on the qualitative characteristics of turkey semen are shown in Figures 1, 2, 3 and 4.

Figure 1. Effect of age on sperm concentration of Hybrid turkey toms. Values are means ± SE, n = 6. Means with different letters are significantly different (a-c = P < 0.05).

The tom ageing was accompanied by a significant decrease of the concentration, viability and motility (P<0.05) of fresh semen beginning from 44 weeks, while for membrane integrity of sperm (P<0.05) from 48 weeks. As expected, the cold storage of turkey semen for 48 h significantly worsened (P<0.01) sperm viability, hypo-osmotic membrane integrity, and the SMI of the turkey semen during the studied reproductive period, even if this occurred in a marked manner with the increasing of tom age. Indeed, at 44 weeks, a decrease of about 40-50% for the viability and motility after 48 h of storage was observed.
Figure 2. Effect of time of storage on Sperm Motility Index of Hybrid turkey toms at different age (32-52 weeks). The Sperm Motility Index is expressed as mean of Optical Density ± SE, n = 6. Means with different letters at the same time of storage are significantly different (a-c = P <0.05). Effect of time of storage: P <0.01.

Figure 3. Effect of time of storage on sperm viability of Hybrid turkey toms at different age (32-52 weeks). The sperm viability is expressed as percentage mean±SE, n = 6. Means with different letters at the same time of storage are significantly different (a-c = P <0.05). Effect of time of storage: P <0.01.
Our conclusion is not unique, since other researcher showed that the quality of fresh and stored semen was affected by age (Douard et al., 2003). Moreover, in our previous studies the quality of semen also decreased during the second part of the reproductive period but in different manner according the strain (Rosato et al., 2005). Douard et al. (2003) observed that the peroxidation in PUFAs of the n-3, n-9 and n-6 series due to the aging may be responsible of the changes of viability, motility and fertilizing ability of spermatozoa. Our results on one hand confirm that the semen quality was affected by age and on the other provide information about the period of the changes of semen quality in both fresh and stored semen of Hybrid strain that essentially occurred at 44 weeks. Therefore, to improve the reproductive performance of ageing males, it could be useful the addition of antioxidants to the tom diet or to the semen extender during the critical reproductive period.

References


SPSS/PC Statistics 12.0. SPSS Inc. Chicago, IL.