In vitro buffalo embryo production under heat stress: Role of granulosa cells co-culture as a monolayer
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Abstract
Effect of heat shock and co-culture of monolayer granulosa cells (GCs) on in vitro embryo developmental competence in Egyptian buffalo was the aim of the present study. Cumulus oocytes complexes (COCs) were collected from ovaries (n= 448) of cyclically slaughtered buffalo cows. Good quality immature oocytes (n= 1512) were subjected to in vitro maturation and fertilization. Post 18–22 h of fertilization, presumptive embryos were randomly assigned into four groups: (G1) No heat shock (38.5°C), (G2) Heat shock (40.5°C) and (G3) Co-culture with GCs monolayer and heat shock or (G4) Co-culture with GCs monolayer and no heat shock. Heat-shocked embryos were exposed to temperature of 40.5°C for the first two hours of culture before exposing to 38.5°C afterward up to the end of culture period (8 days). The data were analyzed by General Linear Univariate model using SPSS.

Oocyte expansion rate was 90.8±1.1%. This percentage is higher than nuclear maturation rate (oocytes at Telophase and Metaphase II) as detected by aceto-orcein staining by about 17%. Cleavage rate as recorded at day 3 post fertilization (day 0) was higher (p≤0.05) for G1 and G4 (71.1±10.5% and 70.5±7.9 with GCs, respectively). On the other hand, cleavage rate was dramatically decreased (P ≤0.05) for embryos of G2 (43.7±7.0%) compared with that of G3 (80.2±7.0%). In addition, embryos of G3 showed approximately the same rate of developed embryos (Morula and blastocyst stages at day 8 of culture) as of G1 (50.9±5.3 and 51.7±7.9%, respectively). In conclusion, using co-culture of GCs as a monolayer enhances cleavage rate and subsequent embryo development of buffalo embryos under heat stress condition.

Keywords: in vitro embryo production, Heat stress, Co-culture, Buffalo