Analyses of apoptosis and DNA damage in bovine cumulus cells after in vitro maturation with different copper concentrations: consequences on early embryo development

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Summary

The aim of this study was to investigate the influence of copper (Cu) during in vitro maturation (IVM) on apoptosis and DNA integrity of cumulus cells (CC); and oocyte viability. Also, the role of CC in the transport of Cu during IVM was evaluated on oocyte developmental capacity. Damage of DNA was higher in CC matured without Cu (0 µg/dl Cu, \( P < 0.01 \)) with respect to cells treated with Cu for cumulus–oocyte complexes (COCs) exposed to 0, 20, 40, or 60 µg/dl Cu). The percentage of apoptotic cells was higher in CC matured without Cu than in CC matured with Cu. Cumulus expansion and viability of CC did not show differences in COC treated with 0, 20, 40, or 60 µg/dl Cu during IVM. After in vitro fertilization (IVF), cleavage rates were higher in COC and DO + CC (denuded oocytes + CC) with or without Cu than in DO. Independently of CC presence (COC, DO + CC or DO) the blastocyst rates were higher when 60 µg/dl Cu was added to IVM medium compared to medium alone. These results indicate that Cu supplementation to IVM medium: (i) decreased DNA damage and apoptosis in CC; (ii) did not modify oocyte viability and cumulus expansion; and (iii) improved subsequent embryo development up to blastocyst stage regardless of CC presence during IVM.

Keywords: Apoptosis, DNA damage, Early development, Minerals

Introduction

Hypocuprosis is the predominant deficiency that globally affects grazing cattle (Ramirez \textit{et al.}, 1998). National Animal Health Monitoring Service categorized 40.6% of United States beef cattle as copper deficient (Dargatz \textit{et al.}, 1999). Ramirez and colleagues (1998) reported similar values in Salado River Basin (Argentina), an area of 55793 km\(^2\) with 6.5 \( \times 10^6 \) beef cattle (Dillon, 1992). Copper deficiency is evidenced by different clinical signs such as pale coat; anemia; spontaneous fractures; poor capillary integrity; myocardial degeneration; hypomyelination; poor reproductive performance; reduce resistance to infectious disease; diarrhea, and generalized ill-health causing severe economic losses (Tessman \textit{et al.}, 2001).

The mammalian cumulus–oocyte complex (COC) and its extracellular matrix are involved in several