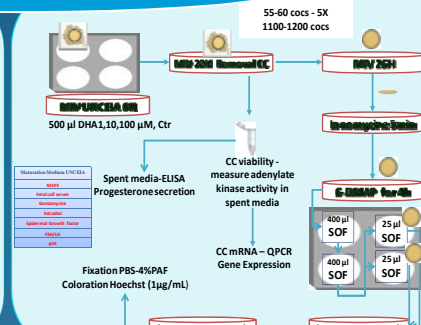
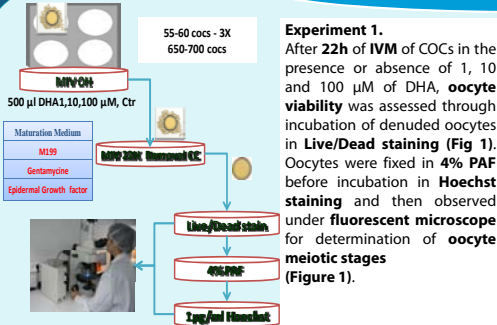


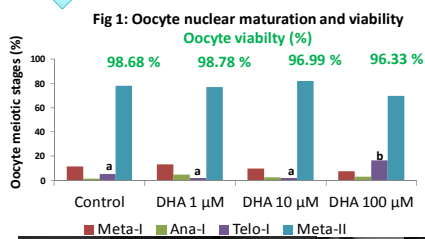
INTRODUCTION

Several studies reported that dietary n-3 polyunsaturated fatty acids such as fish oil rich in eicosapentaenoic acid (EPA, C20: 5 n-3) and **docosahexaenoic acid** (DHA, C22: 6 n-3) improved oocyte and blastocyst quality in bovine. Thus they tended to increase the number of large ovarian follicles and to improve the first service conception rate at 21 days after the artificial insemination when compared to control diet. DHA and EPA are considered more effective than α -linolenic acid (ALA; C18:3 n-3). *In vitro* maturation (IVM) studies showed that addition of palmitic, stearic and linoleic acid had a dose-dependent negative effect on bovine oocyte maturation and developmental competence, in contrast to oleic and ALA which improved bovine oocyte developmental competence. There is few information on the effects of **DHA** on bovine oocyte competence. **The objective of this study** was to investigate, in bovine, whether these positive **effects of DHA**, showed *in vivo*, are partly due to its direct effect on **oocyte maturation and developmental competence**. We also aimed to study the effects of DHA on **progesterone secretion** and **gene expression in cumulus cells** after IVM of oocyte-cumulus complexes (COCs) with a presence of DHA during IVM.

MATERIALS AND METHODS



RESULTS



There is no effects of 1, 10 and 100 μ M DHA on bovine oocyte nuclear maturation rate (Telo-I + Meta-II) and viability. However, 100 μ M DHA increased significantly the rate of Telophase-I compared with all other DHA-treated groups ($P < 0.05$).

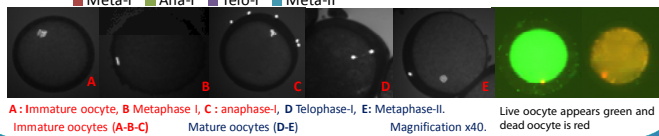


Fig 2: Cleavage rate at day 2 following Parthenogenetic activation

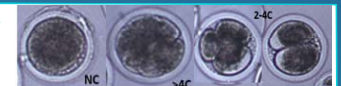
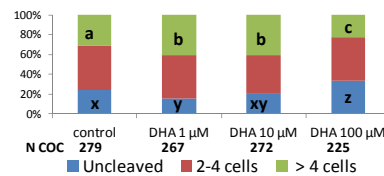


Fig 3: Embryo cells number at day 7 following Parthenogenetic activation

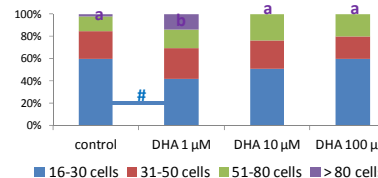


Fig 4: Expression of 3 reporter genes and 9 genes involved in different functions in CC removed from COCs after 22 H of bovine oocyte in vitro maturation

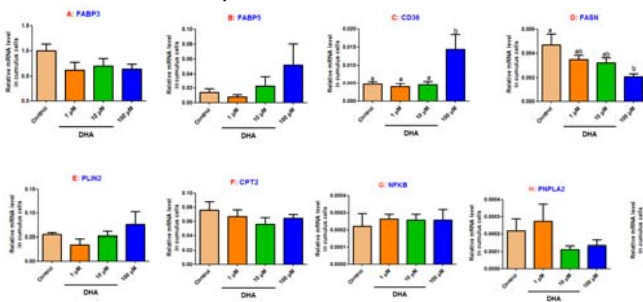
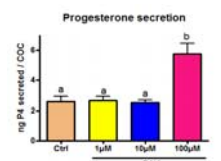



Fig 5: Progesterone secretion

DISCUSSION

These results suggest that treatment of 1, 10 and 100 μ M DHA during IVM did not affect bovine oocyte viability and nuclear maturation rate. However, the highest DHA concentration might delay the progression through meiosis, as showed by a significantly higher rate of oocytes still in Telo-1 after IVM with 100 μ M DHA compared with the control. 1 μ M DHA could improve the bovine oocyte developmental competence shown by increasing both the cleavage rate and the % of embryo with more cells at Day 7 after parthenogenetic activation. Expression pattern of all the lipid metabolism genes investigated did not differ in CC arising from oocytes matured with favorable concentration of DHA. Therefore, DHA effect on oocyte quality should be independent from lipid metabolism. However, high DHA concentration regulated **FASN** and **CD36** genes. An excessive FA amount in extracellular environment could up-regulate **CD36** expression in CC. Indeed, **CD36** is responsible of FA incorporation from extracellular environment, and CC protect maturing oocytes from increased free FA levels by massive intracellular lipid storage and this may safeguard the oocyte from their lipotoxic effects. Therefore, increase in extracellular FA concentration could up-regulate **CD36** expression in order to increase FA intracellular incorporation, thus protecting the oocyte. Consequently, increase in intracellular FA amount would down-regulate **FASN** expression, which is involved in lipogenesis. The higher progesterone level in the presence of 100 μ M DHA could have negative effects on oocyte developmental competence.

In conclusion, this present study suggests that a low dose of DHA (1 μ M) during IVM might improve oocyte developmental competence through possible effect on cytoplasm but not nuclear maturation. Also, we confirmed that a high dose of DHA (100 μ M) is deleterious for oocyte developmental potential. Specific role of DHA on the cytoplasmic maturation, structure and function of bovine oocytes, and the mechanisms through which the developmental potential is affected rests to be investigated.

