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Title: Cyclodextrins or cholesterol-loaded-cyclodextrins? A better choice for improved cryosurvival of chicken spermatozoa

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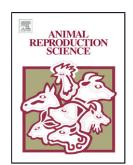
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## ACCEPTED MANUSCRIPT

#### Cyclodextrins or cholesterol-loaded-cyclodextrins?

#### A better choice for improved cryosurvival of chicken spermatozoa

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### Highlights

- cyclodextrins (HBCD, MBCD) and cholesterol-loaded cyclodextrins (HCLC, MCLC) were tested for improved cryosurvival of chicken spermatozoa
- CLCs did not improve chicken sperm quality after cryopreservation.
- 2 mg HBCD increased sperm motility and mitochondrial activity up to 3 hrs of post-thaw storage
- MBCD was detrimental to chicken spermatozoa.

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#### **Abstract**

This study was designed to test if treating chicken sperm with i) the cyclodextrins 2-hydroxypropyl-β-cyclodextrin (HBCD) and methyl-β-cyclodextrin (MBCD) alone improve fresh, liquid-stored and cryopreserved semen quality, or ii) cholesterol-loaded cyclodextrins (CLCs): 2-hydroxypropyl-β-cyclodextrin loaded with cholesterol (HCLC) and methyl-β-cyclodextrin loaded with cholesterol (MCLC) enhance chicken semen quality for application to assisted reproductive technologies. Three consecutive experiments were performed with different concentrations of additives: Exp. 1: 1, 2, 4 mg of HBCD and MBCD in fresh and liquid stored semen; Exp. 2: 1, 2, 4 mg of HCLC and MCLC in fresh and liquid stored semen; and Exp. 3: 1, 2 mg of HBCD, HCLC, 1 mg MBCD, MCLC in cryopreserved and post-thaw storage semen. Sperm motility parameters were assessed by CASA system and comprehensive sperm characteristics were evaluated by flow cytometry. Supplementation with 4 mg HBCD, MBCD, HCLC and MCLC

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