



# Effects of broiler egg storage on cell death

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## THE PROBLEM

Fertile eggs cold stored > 7 days



Embryos die & hatchability reduces



## OBJECTIVES

- Determine if cell death increases with broiler egg storage.
- Determine if egg storage changes gene expression.

## HYPOTHESES

- Egg storage will :
  - reduce viable embryonic cell numbers.
  - increase apoptotic & necrotic cells.
  - increase levels of apoptosis promoting genes (*Bax*, *Bak* and *Bok*) & decrease inhibitor genes (*Bcl-2* and *Bcl-xL*).

## OUR APPROACH

Stored broiler eggs for 4 d & 14 d at 18°C and 80% RH

### 1: METHOD 1: CELL DEATH

- Harvest chicken embryos (blastoderms) from yolk surface.
- Cleaned embryos under light microscope.
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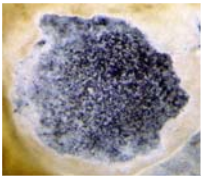


Fig.1 Photomicrograph of the embryo (60X). The chicken embryo contains up to 60,000 cells .

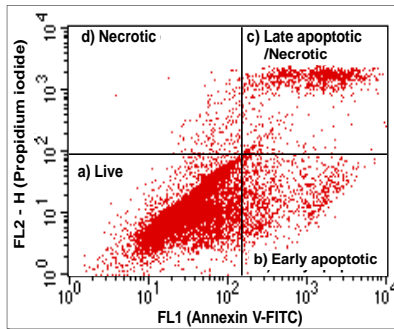


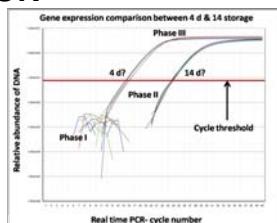
Fig. 2: Embryo separated into single cells and separated on flow cytometer into live, early apoptotic, necrotic & late apoptotic/necrotic cells.

### 2. METHOD 2: GENE EXPRESSION

- Total RNA extracted from blastoderms
- RNA used to synthesize DNA



c) Fig. 3 DNA levels measured in 4 d & 14 d treatments



## WHY ?

- Embryo mortality could be due to death of embryonic cells during egg storage (2).
- Cell death could be due to apoptosis (cell suicide) and/or necrosis (injury driven cell death) (2) ?
- In layer eggs, storage from 4 d to 14 d decreased viable cell numbers & increased apoptotic & necrotic cells (3).

## OUR OBSERVATIONS

Fig. 4: Effect of egg storage on broiler blastodermal Cells

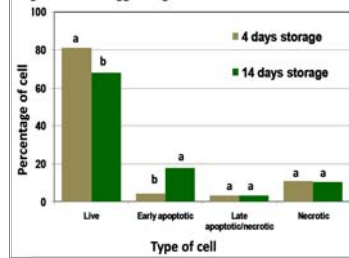
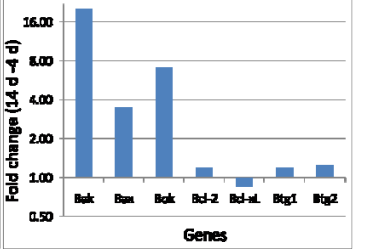


Fig. 5: Effects of egg storage on genes expression



## WHAT DO THE RESULTS MEAN?

- Storing eggs for 4 versus 14 d reduced live embryo cells numbers from 81% to 68 %.
- Storage up to 14 d increased percent early apoptotic cells.
- Increased percentage of dead embryo cells from 14 d stored eggs may have relationship with hatchability problems.
- Expression of genes that brings apoptosis (*Bak*, *Bax* & *Bok*) increased with increasing egg storage duration.
- Expression of genes that prevent apoptosis (*Bcl-2* & *Bcl-xL*) stayed unchanged or increased or decreased slightly.
- Genes that prevent cell division and induced apoptosis when expressed were increased slightly.
- The actions of these genes can be regulated to reduce apoptosis and increase hatchability in 14 d stored eggs.

## ACKNOWLEDGEMENTS



## REFERENCES

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