Response of Chicken-Originated Cells to Virus Mimic

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Summary and Implications

We successfully cultured fibroblast cells and bone marrow-derived cells from White Leghorn and Fayoumi breed chickens. Treatment with a viral-mimic molecule demonstrated breed and cell-type-specific differences in cell survival. Thus, we established a cell-based assay system that may be further explored as a substitute for challenging live animals with viruses to determine factors influencing their response. This could form a cost-effective and welfare-friendly method to determine viral responses.

Introduction

Cell-based systems can be effective tools to understand the chickens' response to pathogens. It is necessary to assess various cell types of the birds to fully understand the response of the whole bird against disease. Few studies have evaluated chicken cell lines to reflect host responses to virus infection. The aim of this study was to establish primary culture methods of cells, which were derived from bone marrow and fibroblasts, and to evaluate breed and cell-type responses to a viral genome-like substance.

Materials and Methods

Cell culture: Cells were cultured from chickens of an egg-laying breed (White Leghorn) and a scavenger-type breed (Fayoumi), which are relatively susceptible and resistant to diseases, respectively. Primary fibroblasts (CEFs) were collected from 5-day-old embryos and cultured in medium with fetal bovine serum (FBS) and antibiotics. Bone marrow cells (BMs) were collected from freshly hatched chicks and cultured in medium with FBS, chicken serum, and antibiotics. Cells were incubated at 39 °C in 5% CO_2.

Poly(IC) treatment by lipofection: Cultured cells were lipofected with 10, 50, and 100 ng/ml poly(IC) to mimic a viral infection.

Measurement of cell viability: Cell viability was determined by counting live and dead cells after tryptophan blue staining.

Statistical analysis: Means and standard deviations of the experimental values were calculated using Microsoft Excel. The statistical significance of differences in the cell number and cell viability between before and after the treatments were assessed using T-test and One-way analysis of variance, followed by Tukey's test (Prism 5.01, GraphPad, San Diego, CA, USA).f

Results and Discussion

We successfully cultured fibroblasts and bone marrow-derived cells from White Leghorn and Fayoumi breed chickens, through passage 5 (Fig 1).

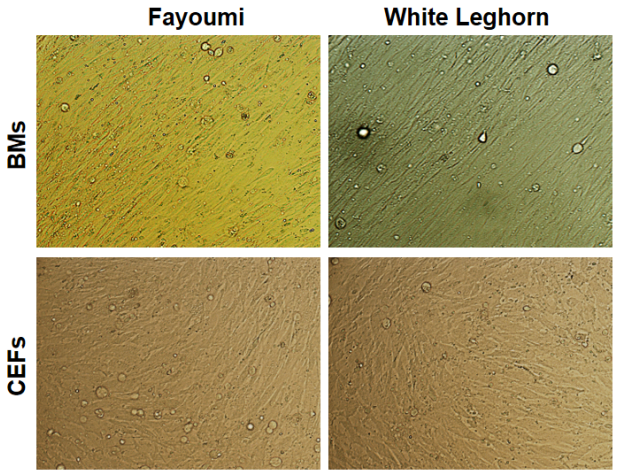

Figure 1. Successful culture of primary cells. Primary bone marrow-derived cells (BMs) and chicken embryonic fibroblasts (CEFs) are cultured from Fayoumi and White Leghorn breeds.

Lipofection without poly(IC) did not have influence on 24-hour cell survival of CEFs or BMs of either breed (Fig 2).

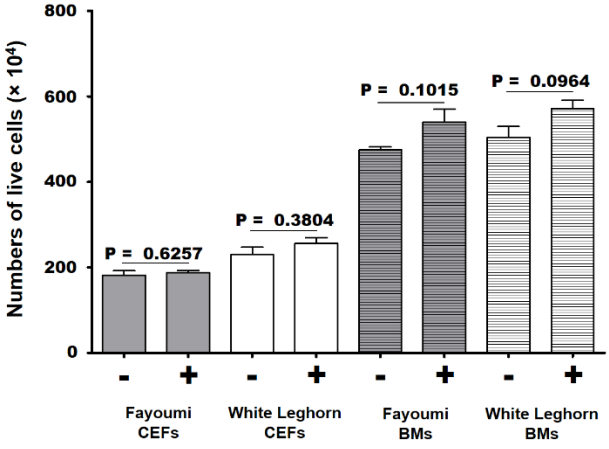

Figure 2. Lipofection does not reduce cell viability. Cell viabilities are compared between non-treated control and at 24 hours after lipofection. -, non-lipofection; +, lipofection; CEFs, chicken embryonic fibroblasts; BMs, bone marrow-derived cells.

There was a tendency for a negative correlation of cell survival with poly(IC) dose in lipofected cells of both types and both breeds. The BMs of Fayoumi origin were more sensitive (had lower cell survival) to poly(IC) treatment than those of White Leghorn origin. Fibroblast survival was lower than BMs after poly(IC) treatment (Fig 3).

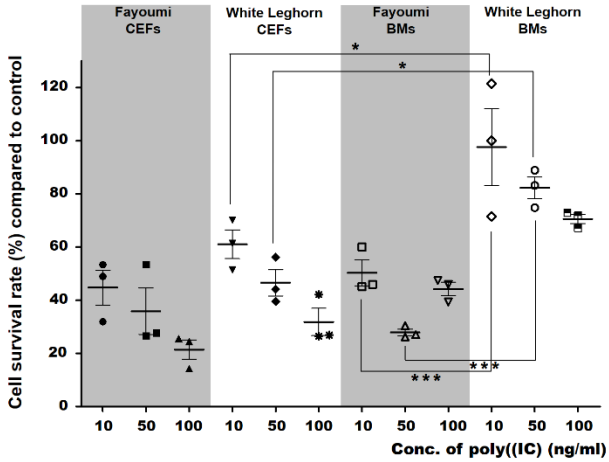

Figure 3. Effect of poly(IC) treatment. Cell viabilities with 10, 50, and 100 ng/ml poly(IC) lipofection are measured at 24 hours after the treatment and compared to non-lipofected controls. CEFs, chicken embryonic fibroblasts; BMs, bone marrow-derived cells. *, P < 0.05; ***, P < 0.001.

Conclusions

In this study, we demonstrated that chicken primary cells respond to poly(IC) in a tissue (cell type) and breed-specific manner. This knowledge enhances the understanding of cellular mechanisms that chickens may use to naturally fight disease. Cell-culture-based assays will avoid the expensive, laborious and welfare-unfriendly practice of challenging live birds with pathogens.

Acknowledgement

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