Abstract

Have you ever heard of Newcastle disease? It’s a dangerous viral infection among chickens and wild birds. It rarely infects people, but it is a threat to the global poultry industry. This is especially true in low and middle-income countries where it’s harder for farmers to protect their chickens. One strategy to combat Newcastle disease is to select for breeds of chicken which are less likely to get the disease. But which breeds resist Newcastle disease and what enables this resistance? To find out we compared the innate immune responses of different breeds of chicken. We looked at which ones were more likely to get infected. Our results revealed the higher expression of five common genes among these breeds. We also found that some versions of the breeds (known as sublines) express some unique genes. These genes might be the key to new breeding strategies that fight Newcastle disease.

Introduction

Do you like eating eggs for breakfast? In order for you to have eggs, farmers look after the chickens that lay them. Like people though, chickens can catch diseases. Perhaps you have heard of avian flu? Another similar threat to chickens is Newcastle disease. This viral infection spreads very quickly amongst chickens (and wild birds too), often causing death. The disease is a major threat to the global poultry industry, especially in countries where there is little veterinary care.

So what can farmers do? Vaccination is an option but usually only in high-income countries. Previous research has found that some breeds of chicken are less likely to get infected with Newcastle disease. Raising these kinds of chickens could save the farmers a lot of trouble.

But how come certain breeds of chicken are able to resist Newcastle disease? We thought that it was these breeds’ immune systems, more specifically – their innate immune response. To find out if we were right, we compared the innate immune responses of different breeds of chickens – Leghorn and Fayoumi (Fig. 1).
Methods

We wanted to compare two sublines (versions) of each of these breeds — a total of four sublines. The Leghorn sublines are known as Ghs6 and Ghs13, and the Fayoumi sublines are known as M5.1 and M15.2.

We inoculated (injected) embryonated (fertilized) eggs from each subline with the virus (Fig. 2). We injected other eggs with sterile solution as our control. We incubated the eggs for three days and then took lung tissue from the embryos before they could hatch.

We extracted RNA from the tissue, and then transcribed it to DNA. To look for different innate immune genes we made lots of copies of particular parts of DNA through a process called real-time PCR. By copying bits of DNA millions of times in tiny wells on a plate, we can start to see and identify the key genes. We then analyzed and compared the expressed innate immune genes. We also estimated the concentration of the virus (called viral load) in each subline.

Results

- We found five expressed genes that were common in all the sublines.
- M5.1 Fayoumi showed the largest number of unique genes, whereas the Ghs6 Leghorn had no unique genes (Fig. 3).
- The immune response differed between breeds and sublines:
  - The Fayoumi breeds had a stronger innate immune response than the Leghorn breeds.
  - The viral load was lowest in M15.2 Fayoumi subline and highest in Ghs6 Leghorn subline. Meaning that M15.2 Fayoumi had the best resistance to the disease.

Why RNA and not DNA? — If we want to look for the existence of genes we could look at the DNA. But the fact that the genes are there doesn’t automatically mean they are active (that they are expressed as traits in the organism). When a gene is active the cell transcribes DNA into RNA. This is why we extracted RNA — we wanted to look for activated (expressed) genes.
Conclusion

Newcastle disease barely ever harms humans. However, chickens or their meat and eggs can sometimes infect you with dangerous diseases. To keep yourself safe, always keep eggs and meat refrigerated. Cook them to a safe temperature. Wash your hands with soap after handling raw eggs or meat.

Check your understanding

1. If Newcastle disease doesn’t pose a problem to human health, why is it a problem at all?

2. If a virus enters your body and tries to infect you what part of the immunity does it trigger first?

3. Why did we research the innate immune response to Newcastle disease for four sublines of chicken?

4. Which chicken subline was the most resistant to the virus?

Discussion

All sublines expressed a set of five common genes in response to the Newcastle disease virus. We found differences in gene expression in response to Newcastle Disease amongst the breeds and sublines. This is good news because it suggests scientists and geneticists could use this information for selective breeding to help farmers. They could breed the chickens which have the strongest response against Newcastle disease.

Researchers need to investigate further though. Are there similar responses in chickens descended from these breeds but mixed with other breeds? Would the results differ if the virus was not a lab one but a wild one? Or would they differ if we inoculated hatched chickens instead of eggs? These questions still need answering but our research provides a strong foundation for future efforts to protect chickens.

Figure 3:
Venn diagram demonstrating the common and uncommon expressed innate immune genes in each subline of chicken.

How many expressed innate immune genes are common to all four sublines? Which subline expresses the highest number of unique innate immune genes?
Glossary of Key Terms

**Breed** – group of domestic animals which have common characteristics and usually are very similar genetically.

**Control** – the part of the scientific experiment that is subject to all the same experimental conditions except for the treatment variable (here: for instance presence or absence of virus). This provides us with a baseline so that we can compare the results of our experiment against it.

**Embryonated eggs** – fertilized eggs where the embryo is in an advanced stage of development and scientists can use them to culture and research viruses and the responses to those viruses in the embryos.

**Expression (gene expression)** – the process where the cell reads information from the DNA and turns it into a useful product: RNA and then a protein. This means the gene becomes a trait, or characteristic, in the living organism. For example, you may carry the DNA for blue and brown eyes, but if you have brown eyes, it is the genes for brown eyes that have been expressed.

**Incubation** – keeping eggs at a certain temperature.

**Innate immunity** – defense mechanisms of an organism that is not specific to the object (virus, bacteria) which is threatening that organism. This is opposed to the acquired immune system, which is specific to the harmful object. For instance: a virus enters the body – the innate immune system is triggered immediately, while the acquired immune system learns how to fight the specific virus over time.

**Inoculation** – scientists give a pathogen (or sometimes just a part of it) to a living organism. This is done in order to observe the effect of an infection or for vaccination.

**Newcastle disease** – a viral disease among chickens and wild birds, it’s very contagious and often deadly to the birds. It doesn’t usually harm humans.

**Poultry** – domestic birds like chickens, ducks, turkeys, geese, used for the production of meat or eggs.

**Real-time PCR** – real-time polymerase chain reaction - a method for creating multiple copies of a specific part of the genetic material. By making a lot of copies, we can visualize them and eventually identify them.

**Select / selective breeding** – the process by which humans use animal breeding to selectively develop particular characteristics. E.g., Farmers may want chickens that produce more meat. They can breed the biggest male and female chickens generation after generation to get bigger offspring.

**Subline** – a subset of a breed.

**Transcription** – the process where the cell reads the DNA and turns it into RNA.

**Viral load** – this tells us how many viruses are in a certain volume, like a milliliter of blood for example. Generally, if it’s high – it means that the virus is happy and the infection is worse.

REFERENCES

