MiSAC matters
MICROBIOLOGY IN SCHOOLS ADVISORY COMMITTEE
50th Anniversary Articles

An ancient difference between a chicken and an ostrich uncovers a new way to fight influenza

Wendy Barclay and Jason Long

The influenza virus, like all viruses, is a parasite that relies on machinery from the host cell it invades to copy itself. This machinery is not there to help the virus,- it has a completely different function for the host, but the virus has evolved to 'borrow it'. The intricate relationship between a virus and its host has been refined over years of co-evolution, so that viruses adapted to infect one species may not infect another species if the cellular machinery they use is missing or different. If the two pieces of the jigsaw, one from virus and one from host, don't fit together like a key in a lock then the virus is blocked (**Figure 1**). This barrier protects us from being infected by viruses from animals.

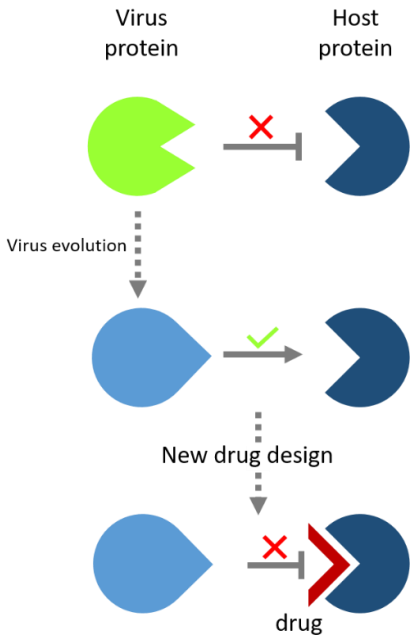

Figure 1. Viruses must adapt to use host proteins from different species. This 'lock and key' relationship can be very specific. If the virus enters a different species, the host protein (the lock) may have a different shape so the virus must adapt by mutating its protein (the key). If we discover the identity of the 'lock', we can design drugs that block the virus from using it and stop the virus in its tracks.

But the barrier can be breached. If an animal virus mutates in a way that allows it to use host machinery from a human cell, this can start a new pandemic as the mutated virus emerges into a human population that has no prior experience of being infected by it and therefore no protection. The paradigm for a pandemic is when an avian influenza virus ('bird 'flu') breaks the host range barrier and replicates in human cells. Birds are the natural reservoir for many different influenza viruses. Most avian influenza viruses do not replicate inside human cells due to incompatibilities with the human cell machinery. The virus has a polymerase enzyme that copies its RNA genome, that only works together with 'borrowed' host cell proteins. One of these cell proteins is ANP32A. There is an important difference between the ANP32 proteins of birds and those of mammals. During the evolution of flighted birds the ANP32A gene was partially duplicated, causing the avian ANP32A protein to be 33 amino acids longer. If a bird 'flu infects a mammal, it cannot use the shorter ANP32 orthologue. But we know there are influenza viruses that do infect mammals, including pigs, horses, dogs and of course humans, so how can they do that? Mutations in the virus polymerase enzyme can make it fit with the short mammalian ANP32 proteins, breaking the barrier and sometimes sparking pandemics such as the notorious Spanish influenza of 1918 that killed more than 50 million people worldwide. By comparing the sequence of virus genomes from birds and mammals, we can identify mutations that adapt the virus to mammals.

A strange but interesting observation helped confirm ANP32A as a key player in this story. Sometimes ostriches are infected by avian influenza viruses, but although ostriches are birds, the genomes of viruses from ostriches contain the

same polymerase mutations seen in mammals! This mystery is solved when one examines the ANP32A gene of the ostrich. Unlike the flighted birds, the DNA of non-flighted species did not duplicate, leaving the ANP32A protein short, as in mammals. To an influenza virus, an ostrich cell looks more like a mammalian cell than a bird cell (**Figure 2**).

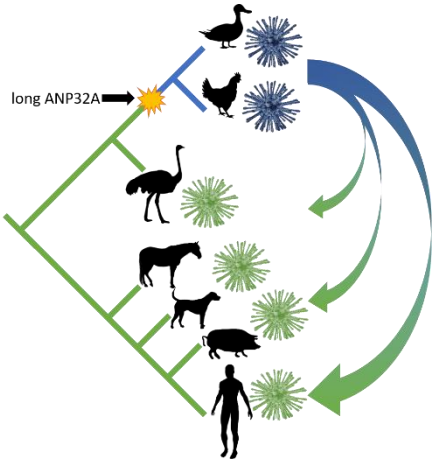

Figure 2. The evolution of ANP32 proteins in different species drives adaptation of influenza virus. This representation of a phylogenetic tree shows the close relationship between ANP32A proteins from different animals known to be infected by influenza viruses. The natural hosts for all influenza viruses are wild birds such as ducks and geese, indeed birds are the reservoir of many different types of influenza viruses today. During their evolution, flighted birds including ducks and chickens gained a longer form of ANP32A proteins (blue). Influenza viruses from ducks and chickens must mutate to successfully infect hosts, including ostriches, dogs, pigs, horses and humans, with the shorter form of ANP32A (green).

How can this information be of use? Influenza viruses cause up to half a million deaths every year. Antiviral drugs do exist but the current drugs directly target proteins of the virus, and because influenza virus can so readily mutate, viruses can easily evolve to become resistant. We can overcome this by designing new drugs that target the host factors the virus relies on. These do not change and so resistance is much less likely. The discovery that influenza virus relies on ANP32 to infect humans means we can search for new drugs that stop the virus accessing ANP32, protecting us from infection (**Figure 1**).

Even more worrying is the threat of a new influenza pandemic. The first step in a pandemic is the exposure of a human to an animal infected with an influenza virus. Influenza viruses in farmed chickens and pigs are of particular concern because some people come into regular close contact with these species, for example farmers or people buying live poultry at a market. In the future, we might be able to generate transgenic farmed animals that are resistant to influenza virus infection by changing their ANP32 proteins in such a way that the virus can no longer use them. Transgenic animals have their genes edited using techniques such as CRISPR, where specific mutations are introduced and become permanently part of the animals' genome. These animals could be bred to generate a new population of chicken or pigs resistant to influenza, safeguarding our food supply and at the same time protecting us against incursions of bird 'flu. We can't ever completely eradicate influenza viruses from the planet because they have a natural reservoir in wild birds that we can neither vaccinate nor genetically modify. But the major source of human infections with animal influenza viruses is from close contact with domestic animals and there we have a chance to control our exposure if we reduce the amount of influenza circulating in chicken and pigs. Reliance on ANP32 is an Achilles' heel of influenza that we can now take advantage of for our own benefit.

AUTHOR PROFILES

Wendy Barclay holds the Action Medical Research Chair in Virology at Imperial College London. She has studied influenza viruses for more than 25 years using molecular virological techniques to research the interaction between influenza viruses and their hosts. **Jason Long** is a postdoctoral fellow in Wendy's lab. He obtained his PhD from Imperial College London by studying avian influenza viruses in birds and the way they can adapt to infect mammals.

This article is one of a number invited as part of the MiSAC 50th anniversary celebrations. The articles are written by experts and are both up to date and relevant to microbiology in schools. MiSAC is grateful to all contributing authors.