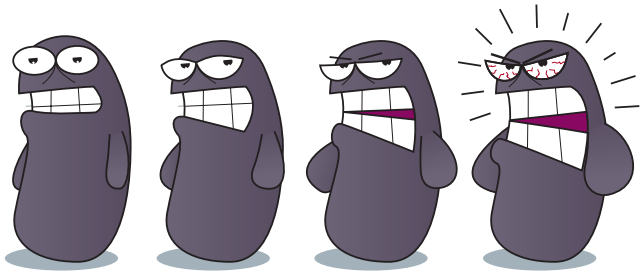

VIRAL EVOLUTION

Pandemic flu can go from bad to worse

> more virulent pandemic influenza strains can indeed emerge owing to mutations and re-assortment with seasonal viruses.

Despite the rapid worldwide spread of the current pandemic influenza virus, no variants of increased pathogenicity have so far been identified from patients, which suggests a stable viral phenotype. However, it is not clear whether this situation may change in the near future. Now, Richard Webby and colleagues show that the emergence of more virulent strains of pandemic influenza can indeed occur by re-assortment with a seasonal influenza virus and by spontaneous mutations.

The influenza virus genome consists of eight separate segments of RNA. Therefore, when a cell is infected by two different viral genotypes, offspring can combine segments from the two infecting viruses, producing new genetic combinations. To explore possible scenarios of influenza virus evolution, Webby *et al.* co-infected normal cells from human bronchial epithelium with three different ratios of a seasonal strain (A/New Jersey/15/07) and a pandemic strain (A/Tennessee/1-560/09). After ten successive passages, they genotyped 25 clones from each of the three groups and sequenced the genomes of six of these clones for each group. In all co-infections, the pandemic genotype predominated in the viral offspring. However, when the seasonal strain dominated in the initial co-infection mixture, most of the progeny carried the seasonal haemagglutinin (HA) gene in a pandemic background, as well as a mutation (D87N) in polymerase basic protein 2 (PB2). In the other two groups, the viral offspring often harboured two mutations, K154Q in HA and L295P in polymerase acidic protein (PA).

Two representative strains of the new variants were selected for further analysis: G1 (carrying mutations in HA and PA) and G2 (with the seasonal-strain HA and the mutation in PB2). Interestingly, G1 and G2 displayed higher replication levels than the parental pandemic and seasonal strains, both in cell culture and in ferrets (a common animal model for influenza studies). To explore possible roles of the observed mutations in the G1 and G2 strains, the authors assayed the viral polymerase activities and receptor specificities and found that, when compared with the parental strains, G1 exhibited higher polymerase activity at 37 °C, whereas G2 showed higher polymerase activity at 33 °C, the temperature of the mammalian airway. Furthermore, G1 displayed a unique pattern of binding to α2,6-sialyl receptors.

This study demonstrates that more virulent pandemic influenza strains can indeed emerge owing to mutations and re-assortment with seasonal viruses — some of which have become resistant to common antiviral drugs. The authors suggest that these adaptive changes identified in the G1 and G2 variants could be used as potential markers of increased pathogenicity during the surveillance of pandemic influenza viruses.

Cesar Sanchez

ORIGINAL RESEARCH PAPER Ilyushina, N. A. *et al.* Does pandemic A/H1N1 virus have the potential to become more pathogenic? *mBio* **1**, e00249-10 (2010).