Chapter 6
Pandemic Influenza A Virus (pH1N1)

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Abstract The latest flu-pandemic caused by influenza A (H1N1) pdm09 (pH1N1) has taken several hundred lives. Influenza virus contributes to respiratory diseases that lead to the nasal secretions, barking cough, decreased appetite, etc., human beings serve to be the dead-end hosts for the virus. The seasonal reassortment and regeneration of virus contribute to chronic infections, which cannot be treated and leads to drug-resistant strains and antigenic shift that is involved in viral entry, spread and tissue tropism. Various antiviral drugs and vaccines are undergoing clinical trials to fight against the virus. For the treatment of infection antiviral drugs like zanamivir and oseltamivir are given to the patients within 48 h of symptom initiation. The main objective nowadays is the search for alternative vaccines that can effectively combat the reasserted virus. Therefore, this article emphasizes on the availability of vaccines and antiviral drugs which can be used to prevent viral infections during the severe outbreaks.

Keywords Flu-pandemic · Influenza virus · Antiviral agent · Vaccines

Abbreviations

ARIs	Acute respiratory infections
DC-Chol/DPPC	Cationic liposomes comprising cationic compound neutral phospholipids
IFITM3	Interferon-inducible trans-membrane protein family membranes 3

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6.1 Prologue

The worldwide recurring outbreaks of influenza, leads the significant morbidity and mortality with variable severity. Influenza virus leads to 2,90,000–6,50,000 fatalities annually and 3 to 5 million serious cases of infections globally (Omoto et al. 2018). Moreover, epidemics are triggered by newly emerging viruses with catastrophic global impact. Influenza is one of the leading causes of lung infections specially acute respiratory infections (ARIs) in humans as compared to the other viruses such as respiratory syncytial virus, adenovirus, rhinovirus, enterovirus, etc. (Dziąbowska et al. 2018). Influenza virus disease contributes to clinical signs such as fever, sore throat, headache, body pain, myositis, emphysema, joint pain, fatigue, secondary renal failure, pneumatoceles and diarrhoea resulting in severe fatality. Pigs are the major influenza A reservoirs, which act as a middle host both for transmission of the interspecies and for the genetic reassortment of viruses. Therefore, pigs may be potential source of risk to human and avian influenza viruses and pandemics too. Co-infection was observed in pigs by both human and avian influenza viruses (Neumann and Kawaoka 2015). The important reservoirs for influenza viruses are waterfowls and wild boars. Influenza viruses can transmit their distinct alleles to new mammalian hosts, such as the recent appearance of influenza in bats in Central America (Venkatesh et al. 2018).

Influenza is also known as “mother of all pandemics”. Among all subtypes, influenza A has been associated with the major worldwide outbreaks (Paules and Subbarao 2017). In the last century, four major outbreaks of influenza have been reported: swine influenza (H1N1) in 2009, Hong Kong influenza (H3N2) in 1968, Asian flu (H2N2) in 1957 and Spanish flu (H1N1) in 1918 (Gagnon et al. 2018). Although the annual vaccination is recommended and antiviral drugs are available, but both have several limitations. The various drawbacks associated with the vaccines are: slow manufacturing and short duration of protection, antigenic changes over time and lack of cross-reactivity and poor immunogenicity in certain populations. Similarly, antiviral agents are also associated with high drug resistance due to reassortment and regeneration of the virus (Sherman et al. 2019).

6.2 Epidemiology

The H1N1 epidemic firstly appeared in Spain in 1918, resulting in millions of fatalities. Several outbreaks have been recorded later due to influenza virus in 1968, 1998, 2009, etc. Recently, the World Health Organization (WHO) GISRS/NIC laboratories tested more than 46,002 specimens from 13 to 26 May 2019 (data as of 07-06-2019); for influenza virus detection, 5285 were found to be positive, of which 3157 (59.7%) were typed as influenza A and 2128 (40.3%) as influenza B. Of the sub-typed influenza A viruses, 620 (30.5%) were influenza A (H1N1) pdm09 and 1414 (69.5%) were influenza A (H3N2). Of the characterized B viruses, 34 (3%) belongs to the B Yamagata lineage and 1104 (97%) to the B-Victoria lineage (Fig. 6.1). Summary of influenza virus detections from influenza laboratory

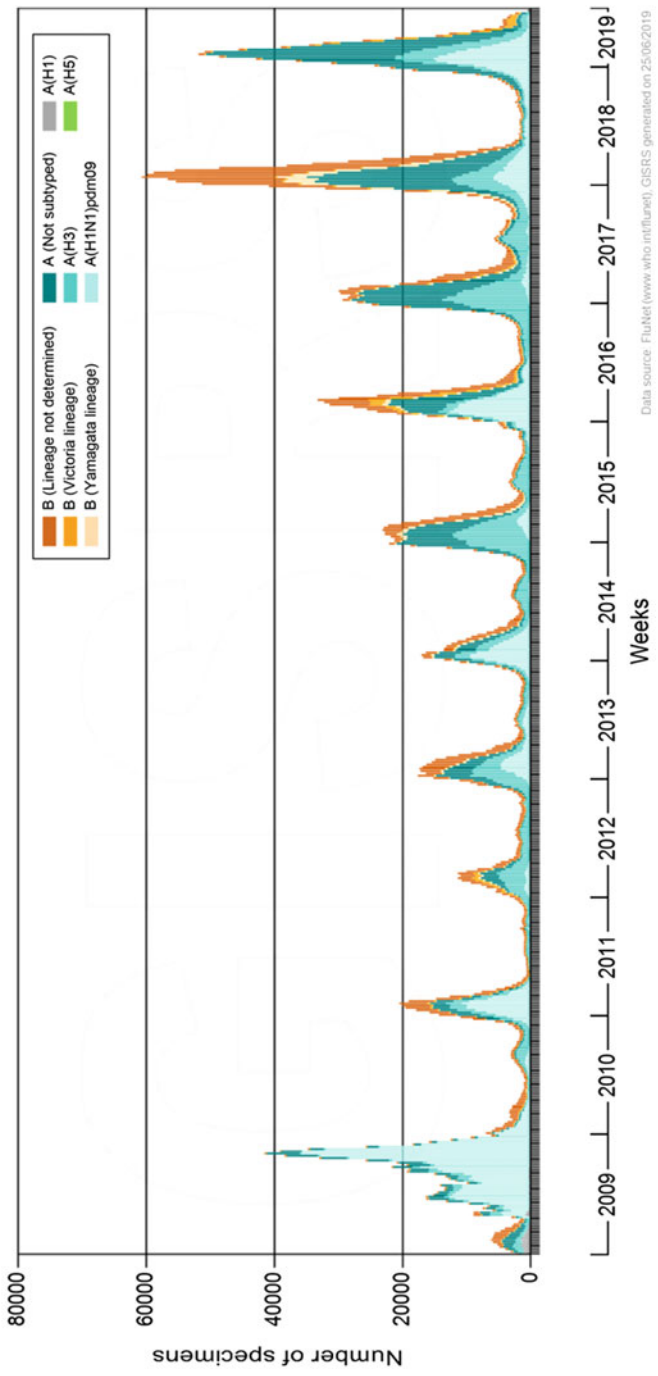

Fig. 6.1 Global circulation of influenza viruses 2009–2019

surveillance information in India during 2009–2019 (as on 26 May 2019; source WHO) has been presented in (Fig. 6.2). Antigenic drift results in mutation in the genome of the virus making it efficient in pathogenesis. The adult population is 90% more common in fatalities, suggesting an increase in immune system dysregulation in conjunction with age (Flu Net 2019).

6.3 Structure of Influenza Viruses

Influenza A virus has a diameter of about 80–120 nm and belongs to the family *Orthomyxoviridae*. The virus genome size is ~13.5 kb (Saxena et al. 2009). The influenza RNA genome is divided in 8 segmented negative sense RNA strands, encoding 11 distinct proteins, i.e. envelope proteins (HA and NA), matrix proteins (M1and M2), non-structural proteins (NS1 and NS2) and viral RNA polymerases (PB2, PB1, PB1-F2, PB and PA) are important for replication of viruses and pathogenesis (Fig. 6.3) (Saxena et al. 2012). Influenza viral infections take place due to the evolution of new viral strains arising from the reassortment of haemagglutinin (HA) and neuraminidase (NA) viral proteins. Both the proteins are mainly responsible for the pathogenesis of the virus by facilitating its internalization and replication in host cells. Modification of viral genome and cellular adaption is the key feature of the virus which leads to severe infections. The significant viral changes in the new host systems rely on the host cell tropism, distribution and viral infection (de Silva et al. 2012).

6.4 Molecular Mechanisms and Ramification

Various influenza viruses have distinct antigenic features for binding to the host's sialyl moiety. Some strains are capable of binding to both glycan bonds, which makes them more virulent and causes intestinal diseases such as diarrhoea (Schrauwen and Fouchier 2014). Mutations in the HA virus region may change the host binding affinity of the virus via distinct strain antigenic shift which contributes to the differential pathogenesis. Various studies have been performed to decipher the internalization mechanism of the virus. HA protein enables the binding of host sialic acid receptor with virus, and this binding complex internalized into the host cell via endocytosis (Sriwilaijaroen and Suzuki 2012). After the entry of virus into the host cells, viral mRNA releases and viral genomic ssRNA synthesis starts by using host cellular machinery. Then it is assembled, matured and eventually produced by budding from host cell membranes in progeny virions (Long et al. 2019).

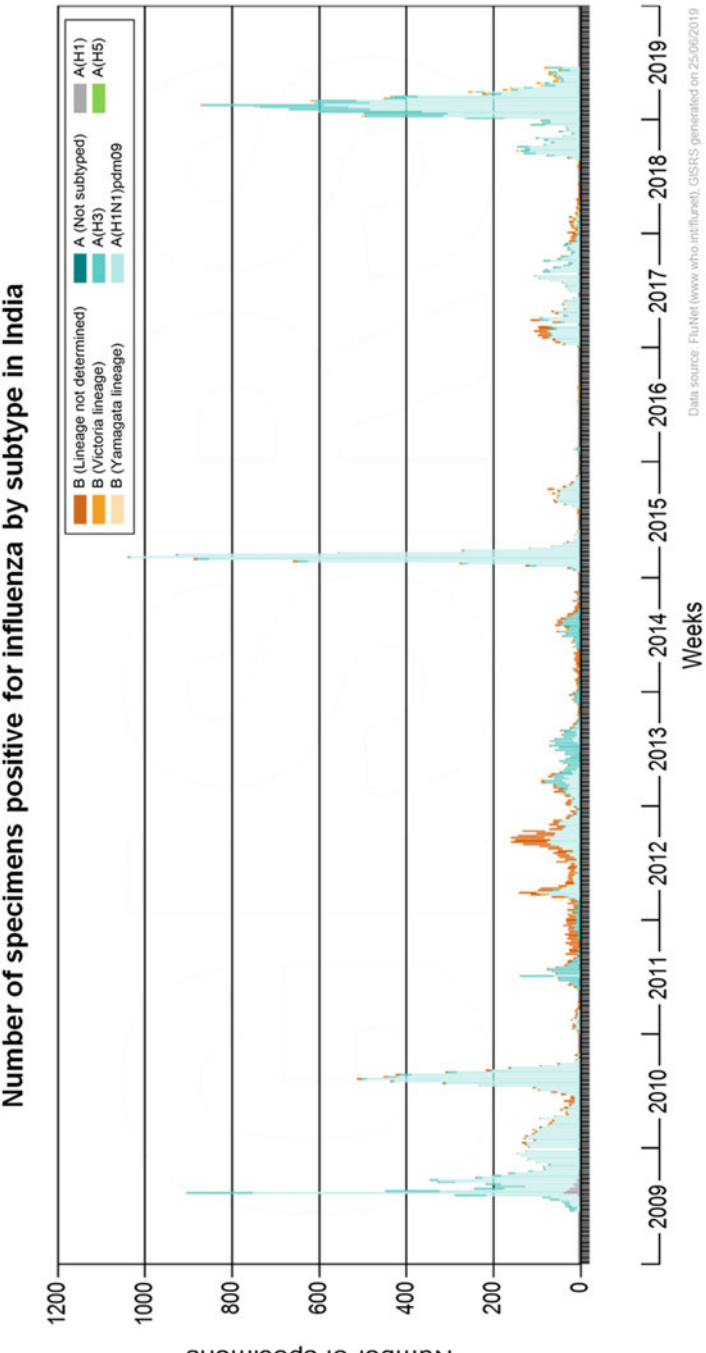

Fig. 6.2 Circulation of influenza viruses in India (2009–2019)

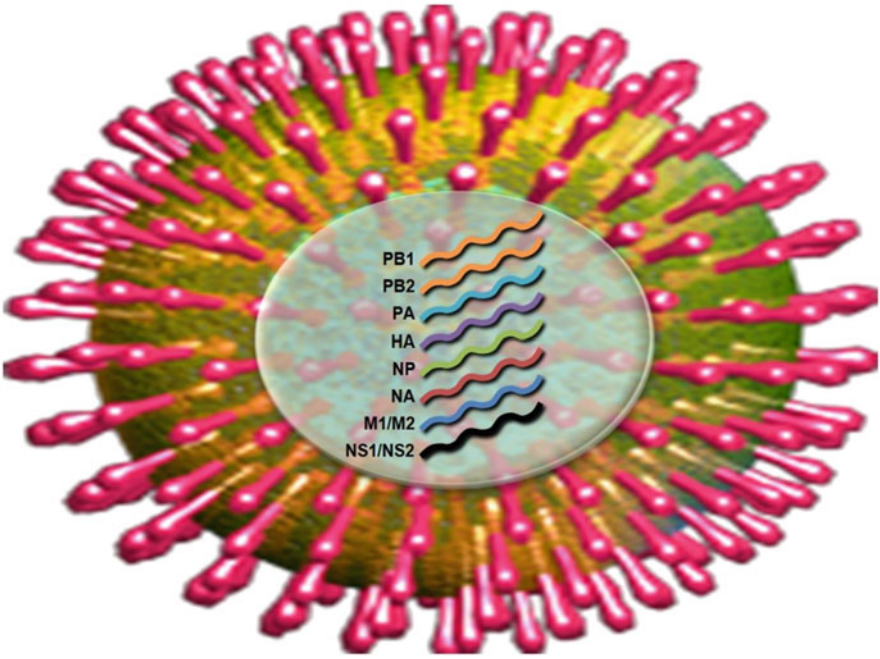

Fig. 6.3 Structure of influenza virus. Influenza virus is a segmented RNA virus possessing eight (single) segmented negative sense RNA strands. Segmented genome encodes eight structural proteins and at least two non-structural proteins. Envelope proteins (HA and NA), viral RNA polymerases (PB2, PB1, PB1-F2, PB and PA), matrix proteins (M1and M2) and non-structural proteins (NS1 and NS2)

6.5 Treatment

There are presently two treatment options suggested for influenza: vaccination and use of antiviral drugs. Antivirals can be used either to avoid or manage individuals who have become infected with influenza. For the successful management of infection, antiviral treatment should begin within 48 h of first symptoms (Barik 2012). NA inhibitors (zanamivir and oseltamivir) and M2 ion channel blockers (rimantadine and amantadine) are presently accessible for the treatment of influenza. NA inhibitors block the viral infection by inhibiting the release of new viral particles from the host cells. The antagonists of M2 ion channel are used to inhibit the viral replication by preventing the virus nucleus being uncoated within the host cell (Duwe 2017). Interferon-inducible trans-membrane protein family membranes 3 (IFITM3) is shown to be a prospective candidate for restricting influenza infections demonstrated by recent studies (Anafu et al. 2013). A broad-spectrum antiviral agent is urgently needed because influenza strains are resistant to present antiviral drugs (Table 6.1).

Table 6.1 Antiviral medications recommended for treatment of influenza (adapted from CDC)

Antiviral agent	Effective against	Recommended dose for adults	Adverse drug reactions
Oral oseltamivir	Influenza A and B	75 mg twice daily	Nausea, vomiting, headache. Postmarketing reports of serious skin reactions and sporadic, transient neuropsychiatric events
Inhaled zanamivir	Influenza A and B	10 mg (two 5-mg inhalations) twice daily	Risk of bronchospasm, especially in the setting of underlying airways disease, sinusitis and dizziness. Postmarketing reports of serious skin reactions and sporadic, transient neuropsychiatric events
Intravenous peramivir	Influenza A and B4	(13 years and older) one 600 mg dose, via intravenous infusion for a minimum of 15 min	Diarrhoea. Postmarketing reports of serious skin reactions and sporadic, transient neuropsychiatric events
Oral baloxavir	Influenza A and B6	(12 years and older) 40 to <80 kg: One 40 mg dose; >80 kg: One 80 mg dose	None more common than placebo in clinical trials
Amantadine	Influenza A	Capsule/tablet, syrup; 100 mg amantadine hydrochloride, twice a day	Mostly discontinued due to resistance; may be recalled in future epidemics
Laninamivir	Influenza A, B (for example, H1N1, H3N2)	Single inhalation (20 or 40 mg)	Similar to oseltamivir. Approved in Japan, but not yet in the USA

6.6 Prevention

Vaccines are the most efficient way of avoiding infectious diseases. There are safe and efficient vaccines that have been used for over 60 years against influenza (Rémy et al. 2015). Developing and producing new influenza vaccines are vital elements for the health care professionals in case of an extensive seasonal and pandemic response to influenza (Buckland 2015). It is necessary to develop potential candidate for vaccines strategy against the H1N1 sequence-based target. The current H1N1 vaccines were designed based on HA and NA has shown to be ineffective due to the antigenic shift and virus reassortment. A universal vaccine against influenza can be developed by targeting the potent epitopes of viral nucleoproteins (Sautto et al. 2018). Microneedle-based vaccines are the most recent and promising strategy, where the microneedles coated with inactivated influenza virus provides lifelong protection, via inducing humoral and cellular immune responses (Song et al. 2010). In combination with the subunit vaccinations, adjuvants are also essential to cause adequate immediate responses. DC-Chol/DPPC (cationic liposomes comprising cationic compound neutral phospholipids) have demonstrated potent

immunogenicity against H1N1 due to the physicochemical property of cationic liposomes which are needed for effective adjuvanticity in subunit vaccines (Barnier Quer et al. 2012). MF59—an adjuvant vaccine—is also more immunogenic, providing constant virus protection. Adjuvant monovalent vaccines are based on nanoparticles that use poly(d), l-lactic-co-glycolic acid (PLGA) and toll-like receptors (TLR) that provide effective protection against H1N1 infection (Pati et al. 2018). According to the immunization practices advisory committee on 20 June 2018, the general efficacy of the 2017–2018 flu vaccine against influenza A and B viruses is estimated at 40%. Similarly, as per the CDC reports 2018, it is suggested that the available vaccines only have 25% protection against A (H3N2), 49% protection against influenza B and 65% protection against A (H1N1) against various influenza strains (Centers for Disease Control and Prevention (CDC) 2019).

6.7 Conclusions and Future Perspectives

Influenza has been causing human morbidity and mortality through the routine seasonal spread and worldwide pandemics for a long time. H1N1 swine flu is a subtype of influenza A virus which is mainly responsible for upper and lower respiratory tract infections in humans. Combined with the assortment of its various genomic sections and mutation rate of the viral RNA genome encourages antigenic variety and new subtypes, enabling the virus to escape vaccines and become resistant to antiviral drugs. Therefore, the novel anti-influenza therapies with new targets have high significance during the future influenza outbreaks. Further, various control measures like using hygienic masks from protection of infected aerosols, keeping infected individuals under medical guidance and isolating them from immunocompromised non-infected individuals and preventing the mass gathering of infected individuals can minimize the risk of disease transmission.

The emergence of various influenza strains has been reported globally due to the rapid globalization, climate shifts and reassortment of viral strains. Therefore, a global alert scheme that can forecast the likelihood of a pandemic in future by its statistical analysis is highly essential. The implementation of best practices in patient care, prevention, diagnosis and selection of antiviral drugs may help to combat the viral infection. The sequence-based drug targeting approaches should be modified while developing newer drugs or vaccines. Similarly, to manage the emerging and re-emerging viral infections, the harmonization among the researchers, doctors, policymakers, virologists, drug designers and the local population is necessary.

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References

Anafu AA, Bowen CH, Chin CR, Brass AL, Holm GH (2013) Interferon-inducible transmembrane protein 3 (IFITM3) restricts reovirus cell entry. J Biol Chem 288(24):17261–17271

Barik S (2012) New treatments for influenza. BMC Med 10:1–15

Barnier Quer C, Elsharkawy A, Romeijn S, Kros A, Jiskoot W (2012) Cationic liposomes as adjuvants for influenza hemagglutinin: more than charge alone. Eur J Pharm Biopharm 81 (2):294–302

Buckland BC (2015) The development and manufacture of influenza vaccines. Hum Vaccin Immunother 11(6):1357–1360

Centers for Disease Control and Prevention (CDC) (2019) Summary of the 2017–2018 influenza season. Accessed 20 June 2019. https://www.cdc.gov/

de Silva UC, Tanaka H, Nakamura S, Goto N, Yasunaga T (2012) A comprehensive analysis of reassortment in influenza A virus. Biol Open 1(4):385–390

Duwe S (2017) Influenza viruses - antiviral therapy and resistance. GMS Infect Dis 5:1–10

Dziąbowska K, Czaczyk E, Nidzworski D (2018) Detection methods of human and animal influenza virus-current trends. Biosensors (Basel) 8(4):1–24

Flu Net (2019) Influenza update n° 343. World Health Organization (WHO). Accessed 24 June 2019. https://www.who.int/influenza/surveillance_monitoring/updates/2019_06_10_surveillance_update_343.pdf?ua=1

Gagnon A, Acosta E, Hallman S, Bourbeau R, Dillon LY, Ouellette N, Earn DJD, Herring DA, Inwood K, Madrenas J, Miller MS (2018) Pandemic paradox: early life H2N2 pandemic influenza infection enhanced susceptibility to death during the 2009 H1N1 pandemic. mBio 9 (1):1–15

Long JS, Mistry B, Haslam SM, Barclay WS (2019) Host and viral determinants of influenza A virus species specificity. Nat Rev Microbiol 17(2):67–81

Neumann G, Kawaoka Y (2015) Transmission of influenza A viruses. Virology 479-480:234–246

Omoto S, Speranzini V, Hashimoto T, Noshi T, Yamaguchi H, Kawai M, Kawaguchi K, Uehara T, Shishido T, Naito A, Cusack S (2018) Characterization of influenza virus variants induced by treatment with the endonuclease inhibitor baloxavir marboxil. Sci Rep 8(1):1–15

Pati R, Shevtsov M, Sonawane A (2018) Nanoparticle vaccines against infectious diseases. Front Immunol 9:1–16

Paules C, Subbarao K (2017) Influenza. Lancet 390(10095):697–708

Rémy V, Zöllner Y, Heckmann U (2015) Vaccination: the cornerstone of an efficient healthcare system. J Mark Access Health Policy 3:1–6

Sautto GA, Kirchenbaum GA, Ross TM (2018) Towards a universal influenza vaccine: different approaches for one goal. Virol J 15(1):1–12

Saxena SK, Mishra N, Saxena R, Saxena S (2009) Swine flu: influenza A/H1N1 2009: the unseen and unsaid. Future Microbiol 4(8):945–947

Saxena SK, Kotikalapudi R, Tiwari S, Muvva C (2012) Influenza A (H1N1) pdm09 virus: therapeutics and challenges. Futur Virol 7(10):947–950

Schrauwen EJ, Fouchier RA (2014) Host adaptation and transmission of influenza A viruses in mammals. Emerg Microbes Infect 3(2):1–10

Sherman AC, Mehta A, Dickert NW, Anderson EJ, Rouphael N (2019) The future of flu: a review of the human challenge model and systems biology for advancement of influenza vaccinology. Front Cell Infect Microbiol 9:1–9

Song JM, Kim YC, Lipatov AS, Pearton M, Davis CT, Yoo DG, Park KM, Chen LM, Quan FS, Birchall JC, Donis RO, Prausnitz MR, Compans RW, Kang SM (2010) Microneedle delivery of H5N1 influenza virus-like particles to the skin induces long-lasting B- and T-cell responses in mice. Clin Vaccine Immunol 17(9):1381–1389

Sriwilaijaroen N, Suzuki Y (2012) Molecular basis of the structure and function of H1 hemagglutinin of influenza virus. Proc Jpn Acad Ser B Phys Biol Sci 88(6):226–249

Venkatesh D, Poen MJ, Bestebroer TM, Scheuer RD, Vuong O, Chkhaidze M, Machablishvili A, Mamuchadze J, Ninua L, Fedorova NB, Halpin RA, Lin X, Ransier A, Stockwell TB, Wentworth DE, Kriti D, Dutta J, van Bakel H, Puranik A, Slomka MJ, Essen S, Brown IH, Fouchier RAM, Lewis NS (2018) Avian influenza viruses in wild birds: virus evolution in a multihost ecosystem. J Virol 92(15):1–20