Influenza prophylaxis: a current medical issue

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Abstract: Influenza is a disease which returns every epidemic season in different degrees of severity, especially in high-risk groups. According to current data, 5–25% of the world population contract this disease and mortality because of influenza reaches 0.5–1 million people. However, this problem is still not dealt with appropriately and is even disregarded. In contrast to other viruses influenza cannot be eradicatedas a result of its changeability but post-influenza complications can be dealt with using prophylactic methods. Since May 9, 1977 there have been more and more fatal cases of human infections with bird flu. The danger of another outburst of a flu pandemic is imminent. According to World Health Organisation influenza is one of the priorities of public health – that is the reason of originating International Influenza Surveillance Programme. A basic method of protecting the population against influenza, which is also the cheapest, is vaccination of as many people in the population as possible, especially those from high-risk groups. Post-influenza complications result not only in illness but also in economic losses.

Key words: influenza, prophylaxis, surveillance, vaccinations

Influenza etiologic factor

Influenza virus is one of the Orthomyxoviridae family [1] and is divided into 3 types: A, B and C. It infects the epithelial cells of the nose, larynx, trachea and bronchi, injuring the epithelium of the respiratory system [2]. The most important characteristic of influenza virus is its heterogeneity. The diameter of a highly pleomorphic influenza virus molecule is 80–120 nm for the spherical forms, when the elongated ones are up to 1000 nm long. Molecular weight of the influenza virus molecule was estimated at 178–200 (±22) × 10^6 D [1]. Influenza virus is composed of the external lipid membrane and the internal core. Using electron microscopy "spikes" can be seen in the external membrane. They include the two most variable glycoproteins: hemaglutinin (HA) and neuraminidase (NA). Very immunogenic hemaglutinin is the basic virus antigen. It causes production of antibodies which inhibit its activity and neutralize the virus infectious potential. These antibodies are specific to each subtype (H1–16) and type (A, B or C). Neuraminidase is an enzyme which splits up the sialic acid, releasing newly formed viruses from the infected cell surface. It accounts for 7–11% of the virus proteins and plays a crucial role in the generation of immune response to influenza virus [2]. Ribonucleotide constitutes a genetic material containing 8 ribonucleic acid segments (RNA), nucleoproteins and a polymerase. The influenza virus core consists of ribonucleic complex, surrounded by M1 protein which comprises of virus genome (8 strands of single stranded RNA with negative polarity) and internal proteins, i.e. nucleoprotein and RNA polymerase. The protein M1 is a structural "matrix" protein that forms a capsid around the virus core. The protein M2 acts as an ion channel which controls inner pH. Based on the antigen differences of internal proteins, i.e. nucleoprotein and M1 protein, there are 3 types of influenza virus: A, B and C. Influenza virus type A occurs in birds and mammals, for example in pigs (H1, H3), horses (H3, H7), minks (H10N4), seals (H7N7), whales (H13N2, H13N9) and humans. Type B occurs in humans only, whereas type C viruses occur in humans and pigs. Additionally, among type A influenza viruses the subtypes are isolated, depending on the kind of HA and NA. There are 16 hemaglutinin subtypes (H1–H16) and 9 neuraminidase subtypes (N1–N9), and all of them occur in water birds. In humans a common occurrence of 3 subtypes of hemaglutinin has been determined so far: H1, H2 and H3, and also 2 neuraminidase subtypes: N1 and N2. However, recently there have been human infections by influenza virus strains which had occurred previously only in birds: A(H5N1) HPAI, A(H9N2) LPAI, A(H7N3) LPAI, A(H7N7) LPAI and A(H7N7) HPAI [1-3].

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Antigenic variation of influenza virus

There is a large antigenic variation in influenza viruses which concerns HA and NA. In type A influenza viruses the highest mutation rate is observed and slightly lower in type B, while type C viruses show a relatively high stability [2]. Changes, mentioned above, are named as antigenic displacement, inducing a seasonal flu epidemic, and as antigenic surge, inducing pandemics.

Antigenic displacement consists in point mutations: substitutions, deletions and insertions. Flu epidemics that appear every year result from accumulation of such alterations in the HA and NA encoding genes. The data regarding morbidity are delivered by family doctors to the Local or Provincial Sanitary and Epidemiological Station (PSES) [2].

The antigenic surge is, in turn, associated with genetic reasortation which is likely due to the sectional structure of the influenza virus genome. The following pandemics took place in the 20th century: so-called "The Spanish flu" of 1918–1919, induced by A(H1N1), caused the death of 100 million people; so-called "The Asian flu" in 1975, induced by A(H2N2), killed 1–4 million people; and so-called "The Hong Kong flu" in 1968, induced by A(H3N2), caused the death of 1–4 million people, where 5940 of them were inhibitants of Poland.

The break of the species barrier on 9 May 1997 in Hong Kong, where the avian flu virus became pathogenic or fatal in a few cases, is scientific evidence suggesting that direct transmission of the animal's influenza virus to the human organism without previous reasortation with influenza virus strain occurring in humans is possible [1].

Influenza surveillance

In 1947 the World Health Organization (WHO), being aware of consequences and risks related with influenza infections, established the Global Influenza Programme (during the 4th International Congress of Microbiology in Copenhagen). In the present constitution of the Programme Council are: the WHO International Collaborating Center for Reference on Research on Influenza and National Influenza Centers in the world [2]. The fundamental concept of this system is to integrate epidemiological and viral controls, so that the morbidity data will be associated with laboratory findings corroborating this pathogen infections. When based on clinical symptoms alone, it is difficult to differentiate the virus-induced influenza respiratory infection from other pathogenic microorganism infections [2,4,5]. The Polish National Influenza Center, one of 118 such centers in the world, plays a referential and coordination role for the whole nation. It participates in the WHO Global Influenza Programme as well as in the European Influenza Surveillance Scheme (EISS) which control is based on the SENTINEL system. In Poland, the SENTINEL system includes family doctors, the PSESs, and also more and more often the County Sanitary and Epidemiological Stations and the National Center for Influenza [6,7]. This system is aimed at the global monitoring of epidemiological and virusologic situation.

Clinical course of the disease

Influenza virus infection is droplet-transmitted. Usually through coughing or sneezing of an infected person, and sometimes also through direct contact with either the the patient, or infected surfaces [2]. In the influenza infection the upper respiratory tract epithelial cells are affected and the virus replication takes place within these cells. Neuraminidase through decreasing viscosity of the mucus which covers the mucous membranes of the respiratory system, causes receptor exposure on the cell surface and facilitates transmission the content of liquid enclosing viruses to the distal segments of the respiratory tract. In the upper respiratory tract influenza virus causes necrosis of ciliated and goblet cells in the mucous membranes in the trachea and bronchi and leads to denudation, which promotes bacterial pathogen invasion, i.e. *Staphylococcus aureus*, *Heamophious influenza* or *Streptococcus pneumoniae* [2]. In the course of the infection the parenchymatous pneumonia with epithelial cell necrosis in the bronchioli may develop, which leads to death. The presence of viremia is suspected because of the virus presence in a low titer within some organs, namely the heart, spleen, kidney, adrenal glands, meninges or the middle ear. Furthermore, in the autopsy material not only because of pneumonia, the presence of influenza virus is determined [2]. The replication cycle of influenza virus in the cell takes about 6–12 hours; during this time approximately 1000 new virions originate within one cell [8]. The influenza incubation period amounts to 1–4 days, on average 2 days. In adults virus excretion usually begins one day before the onset of symptoms and persists about 5 days afterwards. Children are contagious up to 10 days and in the case of small children the virus can be excreted even many days before the first signs of the disease. The virus excretion time in subjects with severe respiratory insufficiency can persist for a few weeks or months [8].

Influenza appears suddenly and is highly contagious. After incubation period there are:

- general symptoms – malaise, chills, skin hyperaesthesia, body temperature >37.8°C
- respiratorysymptoms – discharge from the nose, pharyngitis, hoarseness, chest pain, dry cough inducing vomiting
- other symptoms – headache, anorexia, myalgia, dizziness, diarrhea, abdominal pain, nausea and vomiting, sleepiness or drowsiness (in about 50% of children under 4 years of age, but only in 10% of children between 5 and 14 years).

Gastrointestinal symptoms, mostly nausea and vomiting, are rare in adults, although they are very common in children. Particularly in small children the first symptoms of influenza may sometimes resemble sepsis with a high body temperature, up to 40°C [2,8]. Twenty percent of children hospitalized for influenza develop febrile convulsions [2,8]. Furthermore, the middle ear inflammation in flu sick children is often identified. In the majority of people a cough and malaise may persist

for 2 weeks or even longer. In some patients flu can worsen another concomitant disease course (for example chronic lung or heart disease) and lead to bacterial secondary or flu primary pneumonia, or be part of a complex infection because of other viruses and bacteria [2,9].

Respiratory system signs are related with progressive respiratory tract epithelial cell necrosis where the virus is replicated, and the general signs – with the immune response, mainly cytokine secretion.

Laboratory work-up

Respiratory system infections are characterized by the fact that, on the one hand, a single pathogen can cause different clinical symptoms, but on the other hand, the same symptoms may be induced by various pathogens, including "pseudo flu" viruses, rhinoviruses, adenoviruses, respiratory syncytial virus, *Mycoplasma pneumoniae* or *Legionella sp*. The European Union emphasized the role of laboratory diagnostics towards influenza infection in the Commission's decision of March 19, 2002, where the definitions of cases demanding notification became established (Decision No 2119/98/EC of the European Parliament and the European Council registered under document number C [2002] 1043 [2002/253/EC]) [2].

A laboratory diagnosis of influenza is established in the presence of the following situations:

- influenza antigen or influenza virus specific RNA detection in the patient-collected material
- influenza virus isolation
- immune response identification through anti-type A or B influenza virus specific antibodies presence.

The material for the laboratory analysis can be collected in a form of nose smear, nasopharynx smear, pharynx rinsings, pharynx aspirate, bronchial rinsings, cerebrospinal fluid, middle ear exudate and biopsy material. In the case of basic laboratory analysis an immunofluorescent test, for single material collection, allows diagnosing as early as during 2 hours the following viruses: influenza type A and B, "pseudo flu" type 1, 2 and 3, respiratory syncytial virus and adenovirus. In Poland these analysis can be done in the National Influenza Center, the National Institute of Hygiene in Warsaw and in every PSES. Moreover, in the National Influenza Center there are performed many other analyses, for example influenza viruses isolation from the patient-collected material on the canine kidney cell or the Madin-Darby canine kidney cell (MDCK) culture and chicken embryos, or the virus genetic material detection by real time polymerase chain reaction (RT-PCR) [2,10]. Except for the diagnostic methods used in the laboratory, there are also fast diagnostic tests, so-called bedside tests, which allow obtaining the results after 10–15 minutes and therefore may be used by first contact doctors. However, sensitivity and specificity of such tests are lower than in the laboratory methods. A positive result of a fast diagnostic test must be verified and confirmed by one of the laboratory diagnostic methods [2,10].

Postinfluenza complications

The most exposed to the postinfluenza complications are the high risk morbidity patients. Commonly encountered influenza complications are:

- respiratory system complications – influenza lung inflammation and bronchitis, children bronchiolitis, secondary bacterial lung inflammation mainly induced by *Staphylococcus aureus*, *Streptococcus pneumoniae* and *Haemophilus influenzae*, exacerbation of the existing chronic diseases
- other systems complications – loss of transplant, middle ear inflammation, myocarditis and pericarditis, exacerbation of existing chronic diseases (for example congestive heart failure), febrile convulsions, toxic shock syndrome, Reye's syndrome, muscle inflammation and myoglobinuria which may lead to severe kidney failure, neurological complications (transverse myelitis, brain inflammation, meningitis), higher frequency of schizophrenia in the case of intrauterus inflammation during the pregnancy, meningococcal infections [1,2,4,8-10].

Post-infection complications may concern patients from each age group and every geographic region, but they cannot be confused with undesirable postvaccinal reactions. Moreover, in children the following postinfluenza complications are observed: auditory receptor dysfunction, partial loss of hearing (and even deafness), exacerbation of the asthma and mucoviscidosis, abdominal pain, gastrointestinal symptoms, diarrhea, vomiting (sometimes they mimic appendicitis), myalgia, myositis, neurological complications including Guillain-Barré's syndrome, transverse myelitis, brain and meningitis inflammation [2,8].

Epidemiology of the influenza

Influenza has been known as a highly contagious disease attacking the respiratory system since the ancient times. One of the first flu epidemics was described by Hippocrates in 412 BC [2].

In the temperate zones of the northern and southern hemispheres the influenza infections are seasonal and occur mostly in the winter months, whereas in the tropical countries they occur during the whole year, especially during the rain and the monsoon seasons. Currently, the flu infections affect 5–25% of the world population [3].

Influenza viruses induce epidemic infections and in some cases these epidemics may be global and be linked with particularly high mortality and morbidity. This type of an epidemic is called a flu pandemics which may occur when:

- flu virus strain with subtypes HA or NA different from those of the strains circulating among the people for many previous years appears as a result of sudden changes called antigenic surge
- people sensitive to a new strain will make a high percentage of the population; which means that either the lack of or low levels of antibodies for the new virus will be determined in the majority of the general population

– the new virus will manifest a high ability to transmission and induction of the disease
– new subtype of the virus will be the cause of high morbidity in more than one country [2,3,11].

Prophylaxis of the influenza

Available studies show that "avian-flu" pandemic appearance is highly probable [3,12]. Thus, the WHO and the European Union recommended that all nations of the world enlarge the amount of influenza-vaccinated people and engage in the epidemiological-virusologic surveillance. According to American data, the cost of influenza and its complications in the USA is 76–167 milliard dollars depending on an epidemic season. The results of clinical research on an immune response to influenza vaccinations in high-risk groups of children and adults combined with monitoring of virus morbidity and preparing educational-scientific brochures for doctors and patients, should convince people to influenza vaccinations – especially because of the high probability of influenza pandemic occurrence [13-22].

Vaccines and vaccinations

The discovery of human influenza virus in 1933 caused a very fast advance of research, which in 1941 resulted in the first permission for the production of influenza vaccine for the civil population. Currently, 2 types of vaccines are used for prophylaxis:
– inactivated vaccines
– live vaccines or attenuated vaccines [2,8].

At present, there are many types of inactivated vaccines: the whole virus vaccines, the split vaccines (with split virion), high purified subunit preparations (containing surface subunits – HA and NA), oil adjuvant vaccines, virosomal etc. Research works on safe adjuvants application are continued. These adjuvants will enhance antigen immunogenicity and at the same will make possible, for example, an injection of the smaller amount of antigen for a powerful immune response obtainment. In the process of laboratory analysis the following vaccines are tested: parenteral adjuvants, endometrium adjuvants, recombinant vaccines, DNA vaccines or other subunits of the influenza virus structure. At present, the strains for influenza vaccine production are obtained through amplifying on chicken embryos. Advanced research on the subunit influenza vaccine, using HA and NA reached by means of genetic recombination and expression within the insect cells by baculoviruses, is continued [2,7]. The vaccines produced in cell cultures, for example MDCK or Vero, are in the latest phase of investigations waiting for approval of the American Food and Drug Administration. In contrast to chicken embryos, the use of a tissue culture for amplification of vaccinal strains offers the opportunity of definitely faster vaccine production, which is especially important in influenza pandemic.

Live vaccines are obtained via attenuation of influenza virus by means of decreasing the temperature of replication. They are characterized by a lower cost of production, higher immunogenicity and a physiological way of application. In the United States they were registered and allowed to prophylaxis in 2003, but they have a number of contraindications. The vaccine can be applied to healthy individuals aged 5 to 49 years. Currently, the WHO Advisory Committee on Immunization Practices (ACIP) recommends the type split or subunit vaccines for influenza vaccination [2,8,10].

The inactivated influenza vaccines type split or subunit can be applied to patients from the age of 6 months, but children before <13 years are not allowed to receive the whole virion vaccines. Each 0.5 ml dose of vaccine contains 15 μg HA of three influenza virus strains: A(H1N1), A(H3N2) and B. The influenza vaccine antigen composition is assigned by the WHO and because of the high influenza virus variation it should be altered for every epidemic season. In Poland the influenza vaccinations have been appearing as the recommended vaccines in the Vaccination Protective Programme since 1994 [2].

The most important actualizations of the previous prescriptions for the flu vaccinations using type split or subunit of the inactivated vaccines, recommended by the ACIP in 2006 [8], include clinical indications for vaccinations, and the especially endangered on the influenza complication appearance high-risk groups are separated. They are:
– subjects after organ transplantation
– healthy 6- to 59-months-old children
– 6-months-old and follow-up patients of high-risk groups
– 50-years-old and follow-up people, because the amount of high-risk patients is increased in this group
– patients (adults and children) with chronic diseases of cardiovascular or respiratory system, including asthma
– patients with every disease which may cause the respiratory system dysfunction or removing mucus from the respiratory tracts deficiency, which may increase the choke risk (for example: cognitive dysfunctions, spinal cord injuries)
– diseases with convulsions or other neurologic muscular diseases
– adults and children which in the previous year demanded the regular physical examination by the doctor and were often hospitalized because of metabolic diseases (including diabetes), renal insufficiency, hemoglobinopathy or immune deficiencies (including caused by immunosuppressive treatment or HIV infection)
– children and young people (from 6-months-old to 18-years-old) protractedly treated with acetylsalicylic acid, which increases the risk of the Reye syndrome while influenza virus infected
– pregnant women
– old people from health care institutions and the institutions for chronically ill patients – despite their age.

From the epidemiological indications the vaccination of people which could transmit the influenza to high-risk patients, is recommended. For this purpose, there is advisable the vaccination of:

– doctors, nurses and other medical attendants of hospitals, outpatient departments and emergency ambulance service
– employees of the Elderly Houses and health care centers, which are in contact with guests or the sick (including children), and who guarantee home care for high-risk patients
– members of high-risk patient families (65-years-old and older, after transplantations, people suffering from AIDS and children before 2 years of age), babysitters
– public services workers, e.g. conductors, cashiers, policemen, the militaries, teachers, nursery school teachers, journalists, builders, shop and market assistants or craftsmen.

The influenza vaccination contraindications are: anaphylactic sensitivity to chicken egg white or other vaccine components (aminoglycozide antibiotics for instance), occurring of severe febrile states and Guillain-Barré's syndrome in a case history [8].

The influenza vaccinations are especially prescribed to high--risk patients which should be vaccinated before the beginning of the flu epidemic season. Since 2000 the ACIP has reported that people, who due to some reasons failed to make the vaccination before the season, can do this during the epidemic season, even if the influenza virus circulation in the population was investigated [2,7]. The inactivated vaccines prevent from flu infecting in about 70–90% of the healthy adults before 65 years of age and in children [2,8]. Moreover, they prevent postinfluenza complications and decrease the middle ear inflammation morbidity in children by about 30% [2,8]. The result of the influenza vaccination is a decrease in hospitalization due to lung inflammation and influenza, a decreased number of deaths because of influenza and its complications, as well as a decrease in the sickness absenteeism at work and school, the number of doctor visits, the amount of used antibiotics and over the counter medicines [8,23,24]. The studies performed by means of the meta-analysis showed that type split or subunit inactivated vaccines are immunologically equivalent [2].

Anti-influenza drugs

When the influenza symptoms occur, the antivirus drug incorporation to the treatment may be considered:
– the old generation: amantadine and rimantidine (prophylaxis and treatment)
– the new generation, zanamivir and oseltamivir (prophylaxis and treatment).

Amantadine and its methylated derivative – rimantidine – are the inhibitors of the influenza virus inner protein M2 that acts as an ion channel necessary to decrease pH inside the virion after its fusion with the cell. It enables RNA release and the start of replication. They are efficient only in type A influenza virus infections because type B influenza viruses do not have the protein M2. According to the ACIP 2006 indications in the United States, the application of the amantidine and rimantidine is not recommended, either for treatment or prophylaxis, due to a number of influenza virus strains resistant to these drugs.

New generation of antiinfluenza substances, zanamivir and oseltamivir, are the NA inhibitors and were registered for both treatment and prophylaxis. They are efficient for both, A and B, types of influenza viruses. They simulate the natural substrate for NA (sialic acid) and act by competition, having stronger affinity and specificity to NA than the proper sialic acid. Zanamivir (Relenza) was registered in Poland in April 2001 and is approved for treatment of 7-years-old patients and older and for prophylaxis of 5-years-old patients and older. It is used as inhalant powder. Oseltamivir (Tamiflu) was registered in Poland in May 2003 and is allowed to the treatment of 1-year-old children and prophylaxis. It is applied orally in the form of suspension for children (up to 12) or as capsules for adults. The efficiency of NA inhibitors for treatment is the greater the earlier they are applied after the severe symptoms appearance (not later than 36 hours) [7,8]. Furthermore, it should be emphasized that these drugs are effective only in the case of influenza virus infection. This dictates the need for the establishment of fast diagnostic tests for general practitioners [7,8]. Neither zanamivir nor oseltamivir impair the immune response to the flu vaccination and the natural injection as well [2,3,8]. To make the treatment with anti-influenza medicines efficient, the following requirements should be met: 1) the treatment should be begun as fast as possible, the best in 36 hours after the first symptoms occurring, 2) the influenza virus infection must be earlier confirmed by laboratory analyses – for two basic reasons: firstly, because both drugs work only in the case of the influenza virus infection, and secondly, because there is the necessity of the restriction of drug resistant influenza virus strains generation ability.

The use of oseltamivir to high-risk patients led to the expected therapeutical effect in Poland. These patients were not vaccinated and were admitted to hospital because of the following reasons: exertional dyspnea with the expectoration of bloody mucus and pus secretion, congenital myopathy, respiratory insufficiency, lung transplantation. Although in all patients poor outcomes of influenza were observed, the treatment succeeded [25]. Professor Robert Webster in 1999 said, "The world has received two new neuraminidase inhibitors. It is not right to use them as an influenza vaccine substitute but as an additional useful tool in fight against influenza" [2].

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