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Universal Influenza Vaccines: Mechanisms of Broad Protection

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Abstract The effectiveness of seasonal influenza vaccines varies with changes in virus strains, posing challenges for controlling influenza outbreaks. In recent years, the development of a universal influenza vaccine (UIV) capable of providing broad protection has become a focal point in influenza research. This study explores the potential mechanisms by which a universal influenza vaccine can achieve broad protection, including targeting conserved antigens, inducing broadly neutralizing antibodies (bnAbs), activating T-cell responses, and innate immune responses. By reviewing existing literature and clinical trial data, the study analyzes the key mechanisms in the development of universal influenza vaccines, such as targeting the hemagglutinin (HA) stem domain, neuraminidase, and M2 protein, as well as the role of broadly neutralizing antibodies and T-cell responses. The research indicates that vaccine strategies targeting conserved antigens and broadly neutralizing antibodies show significant broad protective effects in animal models. Additionally, T-cell-mediated immune responses also play a crucial role in preventing influenza infection. The successful development of a universal influenza vaccine would eliminate the dependency on strain-specific vaccines, provide long-term and effective protection against multiple influenza strains, significantly reduce influenza morbidity and mortality, alleviate the burden on healthcare systems, and have a profound impact on global public health security.
Keywords Universal influenza vaccine; Broadly neutralizing antibodies; T-cell response; Conserved antigens; Broad protection mechanisms

1 Introduction

Influenza viruses are significant respiratory pathogens that pose a substantial public health burden worldwide, causing up to 650,000 deaths annually (Freyn et al., 2020). These viruses are responsible for seasonal epidemics and occasional pandemics, leading to widespread morbidity and mortality (Vogel and Manicassamy, 2020). The constant evolution of influenza viruses through antigenic drift and shift complicates the control and prevention of influenza outbreaks (Nachbagauer et al., 2021).

Current seasonal influenza vaccines primarily target the highly variable hemagglutinin (HA) and neuraminidase (NA) proteins of the virus, necessitating annual reformulation to match the predicted circulating strains (Nabel and Fauci, 2010; Lo et al., 2021). This approach has several limitations, including imperfect strain prediction, lengthy vaccine production times, and variable vaccine efficacy (Pica and Palese, 2013; Memoli et al., 2020). Additionally, the emergence of unexpected pandemic strains further challenges the effectiveness of seasonal vaccines (Pica and Palese, 2013).

Given the limitations of seasonal vaccines, there is a critical need for universal influenza vaccines (UIVs) that provide broad and durable protection against diverse influenza virus strains (Jang and Seong, 2019; Vogel and Manicassamy, 2020). UIVs aim to target conserved viral antigens, such as the HA stalk, matrix 2 (M2) protein, and nucleoprotein (NP), to elicit long-lasting immune responses that are effective against multiple influenza subtypes (Krammer et al., 2013; Freyn et al., 2020; Lo et al., 2021). The development of UIVs is a priority in influenza research, as they have the potential to eliminate the need for annual vaccine updates and improve pandemic preparedness (Vogel and Manicassamy, 2020).

This study explores the mechanisms by which universal influenza vaccines (UIVs) provide broad protection. By reviewing the latest advancements in UIV research, including new vaccine designs and immunization strategies,

this study offers a comprehensive overview of the current state of UIV development. It also discusses the challenges and prospects of achieving a truly universal influenza vaccine, focusing on the potential for broad and long-lasting immunity. Understanding and applying mechanisms of broad protection is crucial for developing vaccines with durability and wide-ranging protective capabilities. The development of such vaccines could not only reduce the need for annual vaccine updates but also enhance preparedness for unexpected influenza outbreaks.

2 Need for Universal Influenza Vaccines

Influenza viruses pose a significant public health challenge due to their high mutation rates and the resulting antigenic drift and shift, which necessitate frequent updates to seasonal vaccines. Current influenza vaccines primarily target the hemagglutinin (HA) and neuraminidase (NA) proteins, which are prone to rapid evolution. This leads to a mismatch between the vaccine strains and circulating viruses, reducing vaccine efficacy and leaving populations vulnerable to outbreaks (Jang and Seong, 2019; Nachbagauer et al., 2021; Lo et al., 2021). The unpredictability of influenza virus strains and the lengthy process of vaccine production further complicate timely and effective immunization efforts (Freyn et al., 2020; Vogel and Manicassamy, 2020).

The development of a universal influenza vaccine (UIV) aims to address these challenges by providing broad and long-lasting protection against diverse influenza strains. Such a vaccine would target conserved viral antigens, reducing the need for annual reformulation and offering a more robust defense against both seasonal and pandemic influenza viruses. The National Institute of Allergy and Infectious Diseases (NIAID) has prioritized the development of UIVs to mitigate the substantial public health burden associated with influenza (Scorza et al., 2016; Vogel and Manicassamy, 2020).

2.1 Limitations of existing vaccines

Current influenza vaccines have several limitations that hinder their effectiveness. One major issue is the reliance on strain-specific neutralizing antibodies against the variable globular head domain of the HA protein. This specificity necessitates frequent updates to the vaccine composition to match the circulating strains, which is a time-consuming and often imprecise process (Jazayeri and Poh, 2019; Nachbagauer et al., 2021). Additionally, the efficacy of these vaccines can vary widely from year to year, depending on the accuracy of strain predictions and the degree of antigenic match (Bernstein et al., 2019; Vogel and Manicassamy, 2020).

Moreover, existing vaccines primarily induce humoral immune responses, with limited activation of cell-mediated immunity against conserved internal viral proteins. This narrow focus on surface antigens leaves gaps in protection, particularly against drift and shift variants. The need for annual vaccination and the potential for significant mismatches between vaccine and circulating strains underscore the urgent need for more broadly protective vaccine strategies (Vemula et al., 2017; Jazayeri and Poh, 2019).

2.2 Benefits of a universal vaccine

A universal influenza vaccine would offer several significant benefits over current seasonal vaccines. By targeting conserved viral antigens, such as the HA stalk, neuraminidase, and internal proteins like nucleoprotein (NP) and matrix 2 (M2), a UIV could provide broad and durable protection against a wide range of influenza strains, including those that have undergone antigenic drift or shift (Freyn et al., 2020; Nachbagauer et al., 2021; Lo et al., 2021). This broad-spectrum protection would reduce the need for annual vaccine updates and improve overall vaccine efficacy (Jang and Seong, 2019; Vogel and Manicassamy, 2020).

Furthermore, a UIV could enhance pandemic preparedness by providing a baseline level of immunity against emerging influenza strains, potentially mitigating the impact of future pandemics. The development of novel vaccine platforms, such as nucleoside-modified mRNA and chimeric HA-based vaccines, has shown promise in eliciting strong and long-lasting immune responses in preclinical and clinical studies (Figure 1) (Bernstein et al., 2019; Freyn et al., 2020; Nachbagauer et al., 2021). These advancements highlight the potential of UIVs to revolutionize influenza prevention and control, offering a more reliable and comprehensive approach to combating this persistent public health threat (Jazayeri and Poh, 2019).

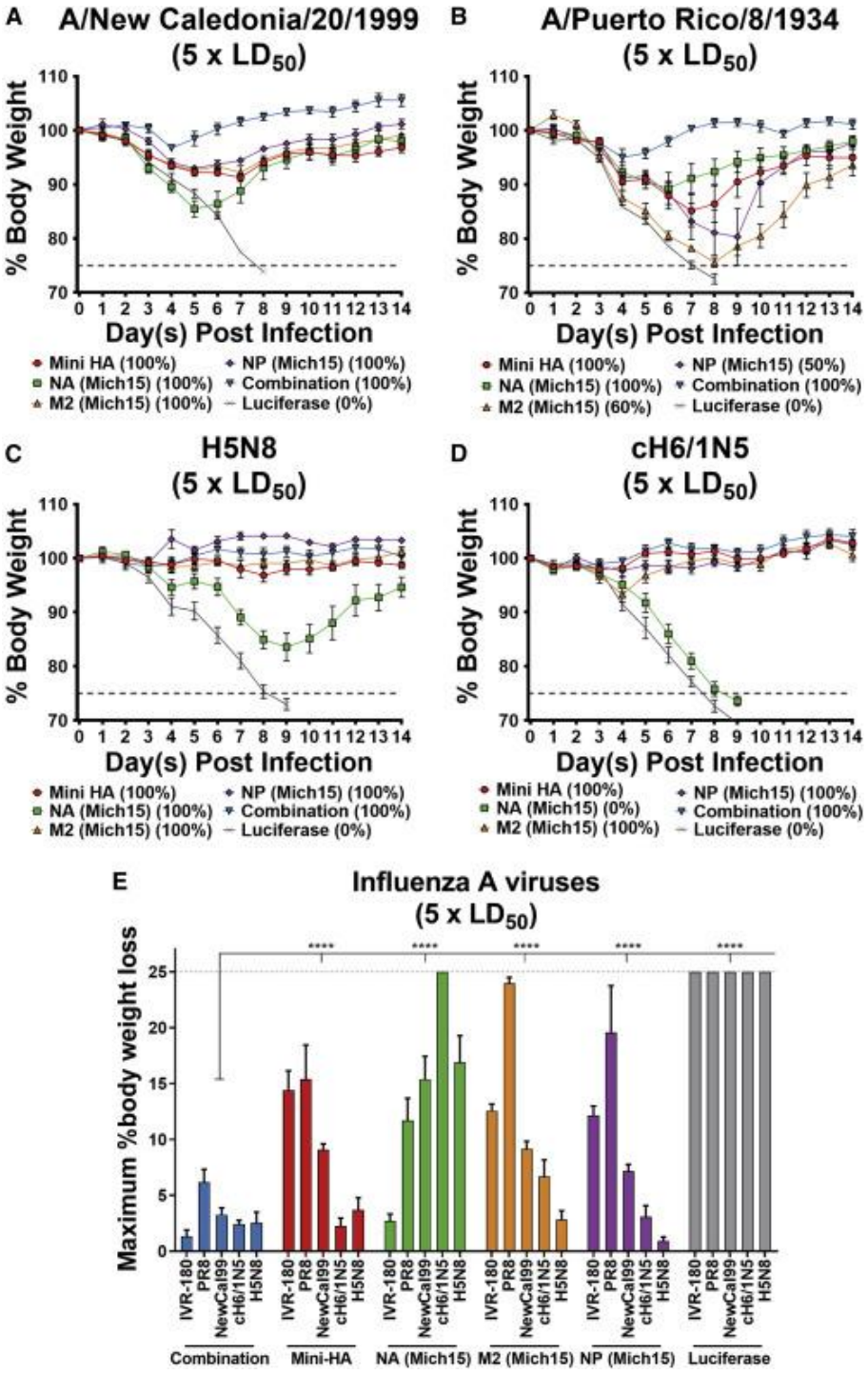

Figure 1 Protection of mice against heterologous challenge by a single immunization with nucleoside-modified mrna encoding influenza virus antigens (Adapted from Freyn et al., 2020)

Image Caption: The figure shows the body weight change curves of mice 28 days after a single intradermal injection of 20 μg mRNA-LNPs (nucleoside-modified mRNA nanoparticles). Mice in different groups were exposed to 5 times the lethal dose (LD50) of the influenza virus: A) A/New Caledonia/20/1999 H1N1 virus, B) A/Puerto Rico/8/1934 H1N1 virus, C) H5N8 virus, D) cH6/1N5 virus. E summarizes the maximum weight loss in each group during the different virus challenge experiments; each group shows the mean and standard error of the mean (SEM) (Adapted from Freyn et al., 2020)

Freyn et al. (2020) evaluated the protective efficacy of a nucleoside-modified mRNA-based vaccine encoding antigens from multiple influenza viruses in mice. Through a single vaccine injection, mice exhibited significant weight retention and survival rates when exposed to various influenza viruses. The experimental results demonstrated that the vaccine effectively mitigated weight loss, particularly under high-dose viral exposure. This

study suggests that nucleoside-modified mRNA vaccines hold promise as an effective means of combating heterologous influenza virus infections, contributing to the development of broad-spectrum influenza vaccines.

3 Mechanisms of Broad Protection

3.1 Targeting conserved antigens

The hemagglutinin (HA) stalk domain is a highly conserved region of the influenza virus, making it an attractive target for universal influenza vaccines. Traditional influenza vaccines primarily target the variable head domain of HA, which necessitates frequent updates to match circulating strains. However, the conserved nature of the HA stalk allows for the development of vaccines that provide broad protection against multiple influenza strains. Studies have shown that vaccines targeting the HA stalk can induce broadly neutralizing antibodies that offer protection against diverse influenza viruses (Krammer and Palese, 2019; Zost et al., 2019; Nachbagauer et al., 2021). For instance, a chimeric HA-based vaccine has demonstrated the ability to elicit strong and durable immune responses targeting the HA stalk, suggesting its potential as a universal influenza vaccine candidate (Nachbagauer et al., 2021). Additionally, nanoparticle-based vaccines incorporating HA stem epitopes have shown promise in eliciting cross-protective immune responses in animal models (Qiao et al., 2022).

Neuraminidase (NA) is another conserved antigen that has been relatively underutilized in traditional influenza vaccines. NA plays a crucial role in the viral life cycle by facilitating the release of newly formed viral particles from infected cells. Antibodies targeting NA can inhibit this process, thereby reducing viral spread. Recent research has highlighted the potential of NA as a target for universal influenza vaccines. For example, studies have demonstrated that NA-specific antibodies can provide broad protection against multiple influenza strains, including those with antigenic variations in HA (Kim et al., 2017; Eichelberger and Monto, 2019). Furthermore, vaccines incorporating NA have shown enhanced immune responses and protection in animal models, suggesting that NA should be considered in the formulation of future universal influenza vaccines (Eichelberger and Monto, 2019; Wang et al., 2021).

The matrix 2 (M2) protein is a small, highly conserved ion channel protein found in the influenza virus. The extracellular domain of M2 (M2e) is particularly conserved across different influenza strains, making it an attractive target for universal vaccine development. Vaccines targeting M2e have been shown to induce robust immune responses and provide broad protection against various influenza viruses (Bernasconi et al., 2018; Lo et al., 2021). For instance, a study demonstrated that a single intranasal dose of a recombinant adenovirus expressing M2e provided long-lasting immune protection in mice against both group 1 and group 2 influenza A viruses, as well as influenza B viruses (Lo et al., 2021). Additionally, nanoparticle-based vaccines incorporating M2e have shown enhanced immune responses and protection in animal models, further supporting the potential of M2e as a target for universal influenza vaccines (Bernasconi et al., 2018; Wang et al., 2021).

3.2 Broadly neutralizing antibodies (bnAbs)

Broadly neutralizing antibodies (bnAbs) have emerged as a promising avenue for achieving broad protection against diverse strains of influenza viruses. These antibodies target conserved regions of viral proteins, primarily hemagglutinin (HA) and neuraminidase (NA), which are less prone to mutation compared to other viral epitopes.

Research has identified several bnAbs that target the conserved stem domain of the HA protein, which is shared across multiple influenza subtypes. For instance, Wei et al. demonstrated that a prime-boost vaccination strategy involving HA DNA and a seasonal vaccine elicited bnAbs in mice, ferrets, and nonhuman primates. These antibodies were directed against the conserved HA stem region and provided protection against a wide range of H1N1 strains (Wei et al., 2010). Similarly, studies have shown that bnAbs can target multiple strains of influenza A viruses by binding to highly conserved epitopes on the HA protein (Ekiert and Wilson, 2012; Feng et al., 2018).

In addition to HA, neuraminidase (NA) has also been identified as a target for bnAbs. Stadlbauer et al. isolated human monoclonal antibodies from an H3N2-infected donor that exhibited broad protection against both influenza A and B viruses. These antibodies neutralized the virus by binding to the active site of the NA enzyme, thereby inhibiting its activity (Stadlbauer et al., 2019).

The protective efficacy of bnAbs is mediated through several mechanisms. Primarily, these antibodies neutralize the virus by blocking the interaction between viral proteins and host cell receptors. For example, bnAbs targeting the HA stem region prevent the conformational changes required for viral fusion and entry into host cells (Wei et al., 2010; Cho and Wrammert, 2016). Beyond neutralization, bnAbs also exploit Fc-dependent effector functions to confer protection. These functions include antibody-dependent cellular cytotoxicity (ADCC) and antibody-dependent cellular phagocytosis (ADCP), which involve the recruitment of immune effector cells such as natural killer (NK) cells and macrophages. For instance, a study on nonneutralizing antibodies specific for influenza B virus hemagglutinin demonstrated that Fc-dependent effector functions, such as ADCC, were crucial for protection in mice (Arunkumar et al., 2019). Similarly, research has shown that Fc-Fcγ receptor interactions are essential for the in vivo protective efficacy of bnAbs against influenza, regardless of their specific epitope (DiLillo et al., 2016).

3.3 T-Cell responses

Cytotoxic T-lymphocytes (CTLs) play a crucial role in the immune response against influenza viruses by targeting and eliminating infected cells. CTLs recognize viral peptides presented by MHC class I molecules on the surface of infected cells, leading to the destruction of these cells and limiting viral replication. The induction of virus-specific CTLs is a key component of broad-protective influenza vaccines, as these cells can target conserved epitopes shared among different influenza subtypes, providing cross-protection. For instance, studies have shown that CTLs contribute significantly to protective immunity against various influenza strains, including H5N1, by targeting conserved viral epitopes (Rimmelzwaan et al., 2007). Additionally, recombinant vaccines incorporating conserved T-cell epitopes have demonstrated the ability to induce functional CTL responses and protect against both influenza and other viral infections (Isakova-Sivak et al., 2020).

Cross-reactive T-cell epitopes are critical for the development of universal influenza vaccines. These epitopes are conserved regions of viral proteins that can be recognized by T-cells across different influenza subtypes, enabling the immune system to mount a response against a wide range of influenza viruses. Research has highlighted the importance of these conserved epitopes in eliciting broad and robust T-cell responses. For example, vaccines targeting conserved epitopes of the influenza virus hemagglutinin (HA) stem have been shown to induce cross-protective immune responses, providing complete protection against lethal challenges with diverse influenza strains (Qiao et al., 2022). Furthermore, the use of minigene vaccines encoding CTL epitopes from influenza A virus has demonstrated the feasibility of inducing specific and directed CTL responses, although additional strategies may be required to achieve full protection.

3.4 Innate immune responses

Natural killer (NK) cells are a vital component of the innate immune response against influenza infection. NK cells can recognize and kill infected cells without prior sensitization, providing an early defense mechanism. They are activated by cytokines and can produce antiviral cytokines such as IFN-γ, which enhances the overall immune response. Studies have shown that NK cells play a significant role in controlling influenza virus replication and spread, contributing to the early containment of the infection (Oftung et al., 2022). The activation of NK cells by influenza vaccines can enhance the overall efficacy of the vaccine by providing immediate protection and shaping subsequent adaptive immune responses.

Pattern recognition receptors (PRRs) are crucial for the detection of influenza viruses and the initiation of innate immune responses. PRRs, such as Toll-like receptors (TLRs) and RIG-I-like receptors (RLRs), recognize viral components and trigger signaling pathways that lead to the production of type I interferons and other pro-inflammatory cytokines. This activation of PRRs is essential for the recruitment and activation of various immune cells, including NK cells and T-cells. The role of PRRs in influenza immunity has been well-documented, with studies demonstrating that the activation of these receptors is necessary for effective antiviral responses and the development of adaptive immunity (Jansen et al., 2019; Oftung et al., 2022). Enhancing PRR activation through vaccine adjuvants or other strategies can improve the efficacy of influenza vaccines by promoting robust and broad immune responses.

4 Strategies for Universal Vaccine Development

4.1 Viral vector-based vaccines

Viral vector-based vaccines utilize modified viruses to deliver genetic material encoding influenza antigens to host cells, thereby inducing an immune response. One promising approach involves the use of recombinant adenoviruses (rAd) expressing conserved influenza antigens such as nucleoprotein (NP) and matrix 2 (M2). Studies have shown that a single intranasal dose of rAd expressing these antigens can induce long-lasting immune responses, with antibody and T-cell responses persisting for over a year without the need for boosting. This approach has demonstrated broad protection against both group 1 and 2 influenza A viruses, as well as influenza B viruses, in animal models (Lo et al., 2021). Another study combined chimeric hemagglutinin constructs with viral vectors expressing NP and matrix protein 1 (M1), resulting in enhanced protection against diverse influenza strains, including H3N2, H10N8, by inducing robust antibody and T-cell responses (Figure 2) (Arunkumar et al., 2019).

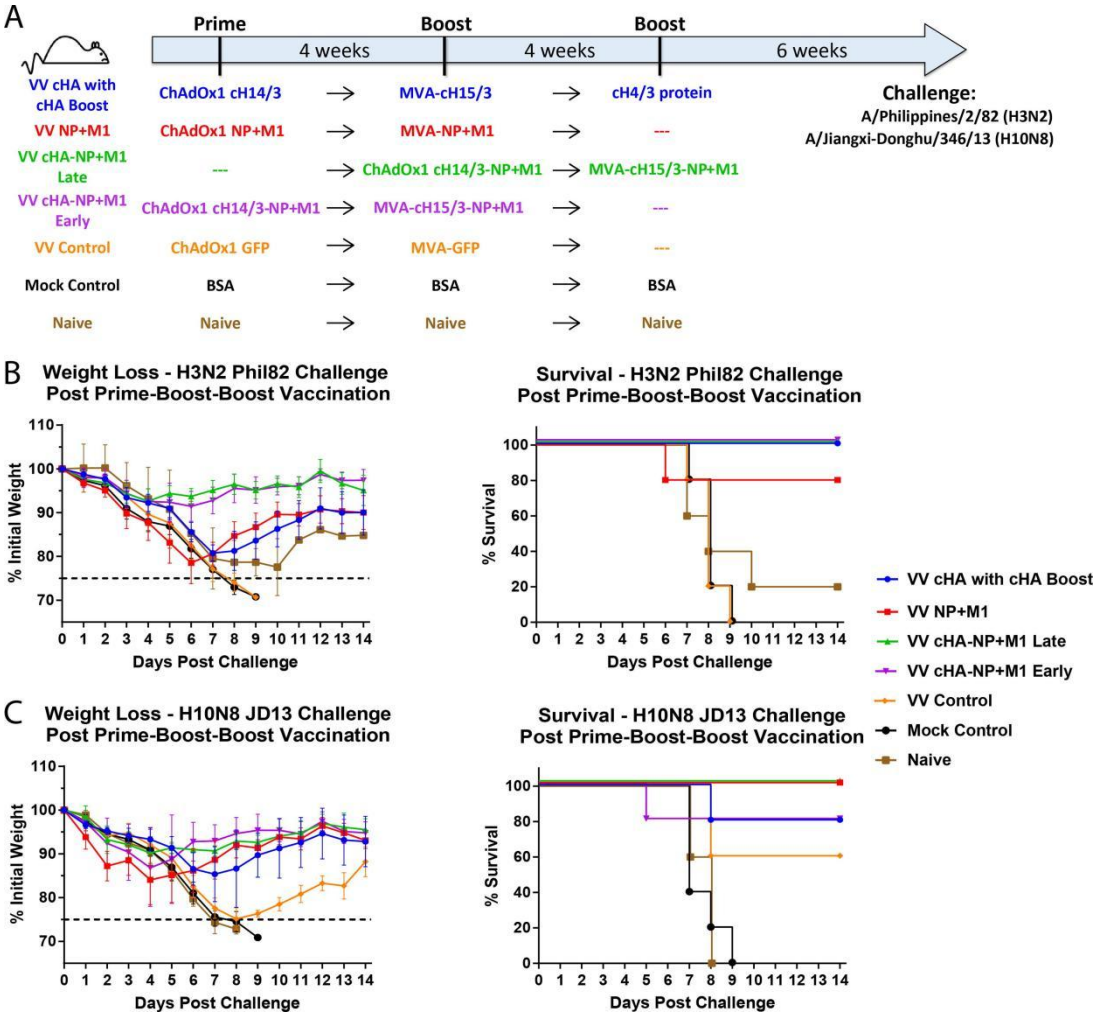

Figure 2 Evaluation of the protective efficacy of viral vector vaccines against H3N2 and H10N8 virus challenges (Adapted from Arunkumar et al., 2019)

Image Caption: (A) shows the prime, boost, and second boost vaccination regimen for mice administered with viral vector vaccines, with doses given four weeks apart; (B) and (C) display the weight changes and survival rates of mice following challenges with H3N2 and H10N8 viruses, respectively. The vaccination groups include vaccines containing HA antigen, NP+M1 vaccine, early and late combined vaccine groups, and a control group. The experiments measured weight changes and 14-day survival rates post-infection for each group of mice (Adapted from Arunkumar et al., 2019)

Additionally, the use of viral vectors such as Chimpanzee Adenovirus Oxford 1 and Modified Vaccinia Ankara virus has been explored. These vectors express conserved internal influenza virus antigens, aiming to induce

T-cell responses. Combining these vectors with chimeric hemagglutinin constructs has shown promising results in providing broad protection against influenza virus challenges in mice (Arunkumar et al., 2019). This strategy highlights the potential of viral vector-based vaccines to induce durable and broad immune responses, making them a viable candidate for universal influenza vaccines.

4.2 Recombinant protein vaccines

Recombinant protein vaccines focus on using conserved influenza antigens to elicit immune responses. One approach involves the use of fusion proteins containing multiple copies of the ectodomain of matrix protein 2 (M2e), a highly conserved antigen. Incorporating these fusion proteins into nanoparticle-based delivery systems has shown enhanced immune protection against live influenza virus challenges. For instance, a study demonstrated that a fusion protein with three copies of M2e, when incorporated into porous maltodextrin nanoparticles, provided strong and broadly protective immunity against heterosubtypic influenza virus infections (Bernasconi et al., 2018).

Another promising strategy involves the use of recombinant outer membrane vesicles (OMVs) engineered to display M2e antigens. These OMVs have been shown to elicit strong IgG titers and provide 100% survival against lethal influenza challenges in mice. The protection was largely driven by antibody-mediated immunity, indicating the potential of OMVs as a platform for universal influenza vaccine development (Rappazzo et al., 2016). Additionally, recombinant baculovirus vaccines expressing multiple M2e copies and adjuvants have demonstrated cross-clade protection against H5N1 influenza viruses, further supporting the efficacy of recombinant protein vaccines in providing broad protection (Zhang et al., 2016).

4.3 mRNA vaccines

mRNA vaccines represent a novel approach to universal influenza vaccine development. These vaccines utilize lipid nanoparticle-encapsulated, nucleoside-modified mRNA to deliver genetic instructions for the production of conserved influenza antigens within host cells. A study demonstrated that mRNA vaccines encoding a combination of conserved influenza antigens, such as hemagglutinin stalk, neuraminidase, matrix-2 ion channel, and nucleoprotein, induced strong and broad immune responses in mice. The immunity conferred by these vaccines provided protection from pandemic H1N1 virus challenges at high doses, highlighting their potential for broad protection (Freyn et al., 2018).

The use of mRNA vaccines offers several advantages, including rapid development and production, as well as the ability to induce both humoral and cellular immune responses. The broad protective potential of mRNA vaccines has been confirmed by challenges with a panel of group 1 influenza A viruses, supporting their advancement as universal influenza vaccine candidates (Freyn et al., 2018). This innovative approach leverages the flexibility and efficacy of mRNA technology to address the challenges posed by rapidly evolving influenza viruses.

4.4 Adjuvants and delivery systems

Adjuvants and delivery systems play a crucial role in enhancing the efficacy of universal influenza vaccines. One example is the use of Advax-SM adjuvant in combination with an M2-based influenza vaccine. This formulation has shown protective efficacy in both maternal and neonatal immunization models, providing protection against diverse influenza A strains. The adjuvant enhanced the immune response, leading to high antibody levels and significant reduction in lung virus load in immunized pups (Sakala et al., 2021).

Another innovative approach involves the use of recombinant fusion proteins linking influenza M2e to adjuvants such as Onchocerca volvulus activation associated protein-1 (ASP-1). These fusion proteins have demonstrated strong humoral and cellular immune responses, providing significant cross-clade protection against divergent H5N1 viruses. The use of adjuvants like ASP-1 enhances the immunogenicity of the vaccine, making it a promising candidate for universal influenza vaccine development. Additionally, the incorporation of adjuvants such as LTB in recombinant baculovirus vaccines has shown improved survival and decreased lung virus shedding in mice, further supporting the role of adjuvants in enhancing vaccine efficacy (Zhang et al., 2016).

5 Preclinical and Clinical Studies

5.1 Animal models and immunogenicity

Preclinical studies have demonstrated the potential of universal influenza vaccines to provide broad and long-lasting protection against diverse influenza strains. For instance, a study using a recombinant adenovirus (rAd) expressing influenza A nucleoprotein (A/NP) and matrix 2 (M2) in mice showed that a single intranasal dose could induce antibody and T-cell responses that persisted for over a year without boosting. This vaccine provided broad protection against both group 1 and 2 influenza A viruses, as well as influenza B viruses, even a year after vaccination (Lo et al., 2021). Another study highlighted the importance of targeting conserved antigens, such as the hemagglutinin (HA) stalk, to elicit broadly cross-reactive antibodies. This approach has shown promising results in animal models, suggesting that it could be a viable strategy for developing universal influenza vaccines (Jang and Seong, 2019).

In addition to the rAd-based vaccine, other preclinical studies have explored different vaccine platforms and formulations. For example, a plant-derived virus-like particle (VLP) vaccine candidate was tested in both young and older adults. The vaccine induced significant homologous and heterologous antibody responses, as well as antigen-specific CD4+ T cells producing interferon-gamma (IFN-γ), interleukin-2 (IL-2), and tumor necrosis factor-alpha (TNF-α). These findings support the potential of VLP-based vaccines to provide broad and durable protection against influenza (Pillet et al., 2019).

5.2 Phase I/II clinical trials

Phase I and II clinical trials have been crucial in evaluating the safety and immunogenicity of universal influenza vaccine candidates in humans. A randomized, placebo-controlled phase I trial tested a chimeric hemagglutinin-based vaccine in healthy adults. The vaccine was found to be safe and induced a broad, strong, and durable immune response targeting the conserved HA stalk domain. This suggests that chimeric hemagglutinins could be developed as universal vaccines that protect broadly against influenza viruses (Nachbagauer et al., 2021). Another phase I study tested a similar chimeric HA-based vaccine regimen, which included both live-attenuated and inactivated vaccines. The results showed that the vaccine regimens were tolerable and elicited cross-reactive serum IgG antibodies targeting the HA stalk domain, providing proof-of-principle for this approach in humans (Bernstein et al., 2019).

In a phase II trial, the immunogenicity and safety of an oral influenza vaccine, VXA-A1.1, were evaluated using a human influenza challenge model. The vaccine was well tolerated and generated protective immunity against virus shedding, similar to a licensed intramuscular inactivated influenza vaccine (IIV). These results represent a significant step forward in developing a safe and effective oral influenza vaccine (Liebowitz et al., 2020). Another phase II trial tested the FLU-v vaccine, composed of synthetic peptides with conserved epitopes from influenza A and B strains. The trial aimed to evaluate the vaccine's ability to elicit both cell-mediated and humoral immunity, providing broad protection against multiple influenza strains.

5.3 Phase III clinical trials

Phase III clinical trials are essential for confirming the efficacy and safety of universal influenza vaccine candidates in larger populations. A systematic review of universal influenza vaccines in clinical trials during the 2010-2019 decade identified three vaccines currently in phase III trials. These vaccines have the potential to provide significant improvements over seasonal influenza vaccines by offering broader and more durable protection (Corder et al., 2020). One such phase III trial evaluated the immunogenicity and safety of an inactivated quadrivalent influenza vaccine (IIV4) in a healthy population aged 3 years and older. The vaccine demonstrated good immunogenicity and safety, adding protection against an additional influenza B strain without increasing safety concerns (Chu et al., 2020).

Another phase III trial examined the safety, immunogenicity, and lot-to-lot consistency of an IIV4 candidate in children, adolescents, and adults. The vaccine was well tolerated, induced robust antibody responses to all four influenza strains, and met all European Medicines Agency (EMA) immunogenicity criteria for adults. These

findings support the potential of IIV4 to offer broader protection against seasonal influenza compared to trivalent vaccines (Cadorna-Carlos et al., 2015). The ongoing phase III trials of these universal influenza vaccine candidates are expected to provide critical data on their efficacy and safety, paving the way for their potential approval and widespread use.

6 Challenges and Future Directions

6.1 Addressing antigenic diversity

One of the primary challenges in developing a universal influenza vaccine is the antigenic diversity of the virus. Influenza viruses exhibit high genetic variability, particularly in the hemagglutinin (HA) and neuraminidase (NA) proteins, which are the main targets of current vaccines. This variability necessitates the annual reformulation of vaccines to match the predicted circulating strains, a process that is both time-consuming and often imperfect (Coughlan and Palese, 2018; Isakova-Sivak et al., 2021; Lo et al., 2021). The development of vaccines targeting highly conserved regions of the virus, such as the HA stalk or the matrix 2 (M2) protein, has shown promise in preclinical studies. These approaches aim to elicit broadly neutralizing antibodies that can provide cross-protection against diverse influenza strains (Pica and Palese, 2013; Zhao and Xu, 2018; Zhang et al., 2019).

Despite these advancements, significant hurdles remain. The immune response to conserved antigens must be robust enough to confer protection against a wide array of influenza viruses, including those with pandemic potential. Recent studies have highlighted the importance of inducing a balanced immune response that includes both antibody and T-cell mediated immunity to achieve broad protection (Jang and Seong, 2019; Jang and Seong, 2020). Future research should focus on optimizing vaccine formulations to enhance the immunogenicity of conserved antigens and evaluating their efficacy in diverse populations through extensive clinical trials (Isakova-Sivak et al., 2021).

6.2 Overcoming technical and logistical barriers

The production and distribution of universal influenza vaccines face several technical and logistical challenges. Traditional vaccine manufacturing methods, such as egg-based production, are not only time-consuming but also susceptible to issues like egg supply shortages and the risk of egg-adapted mutations that can reduce vaccine efficacy (Pica and Palese, 2013; Lo et al., 2021). Alternative production platforms, including cell-based and recombinant technologies, offer potential solutions by enabling faster and more scalable vaccine production. These methods also allow for the incorporation of novel antigens and adjuvants to enhance the breadth and durability of the immune response (Jang and Seong, 2019).

Logistical challenges extend beyond production to include the distribution and administration of vaccines. Ensuring global access to universal influenza vaccines requires robust supply chains and infrastructure, particularly in low- and middle-income countries. Additionally, public health strategies must address vaccine hesitancy and ensure high vaccination coverage to achieve herd immunity (Coughlan and Palese, 2018; Jang and Seong, 2020). Future efforts should focus on developing cost-effective production methods, improving vaccine stability and storage, and implementing comprehensive vaccination programs that can rapidly respond to emerging influenza threats (Zhao and Xu, 2018; Zhang et al., 2019).

6.3 Future research directions

Future research in the field of universal influenza vaccines should prioritize several key areas. First, there is a need for a deeper understanding of the immune correlates of protection. Identifying the specific immune responses that confer broad and long-lasting protection against influenza will guide the design of more effective vaccines (Jang and Seong, 2019; Isakova-Sivak et al., 2021). This includes studying the roles of different antibody isotypes, T-cell subsets, and innate immune responses in mediating protection (Pica and Palese, 2013; Jang and Seong, 2020).

Second, the development of novel vaccine platforms and delivery methods should be explored. Mucosal vaccines, for example, have shown promise in eliciting strong local immune responses in the respiratory tract, which is the primary site of influenza infection (Coughlan and Palese, 2018; Lo et al., 2021). Additionally, prime-boost

strategies that combine different vaccine modalities, such as live attenuated and subunit vaccines, may enhance the breadth and durability of the immune response (Carter et al., 2016; Jang and Seong, 2019). Finally, conducting large-scale clinical trials in diverse populations is essential to evaluate the safety and efficacy of universal influenza vaccines and to understand how preexisting immunity influences vaccine responses (Zhao and Xu, 2018).

7 Concluding Remarks

The pursuit of a universal influenza vaccine has yielded promising results across various studies. Several approaches have demonstrated the potential for broad and long-lasting immune protection. For instance, a single intranasal dose of recombinant adenoviruses expressing influenza A nucleoprotein and matrix 2 has shown to provide durable immune responses and protection against diverse influenza A and B virus strains in animal models. Similarly, chimeric hemagglutinin-based vaccines have been found to induce broad and durable immunity in human trials, targeting the conserved stalk domain of hemagglutinin. Additionally, nucleoside-modified mRNA vaccines encapsulated in lipid nanoparticles have shown substantial breadth and potency in eliciting immune responses in mice. Other strategies, such as the use of computationally optimized broadly reactive antigens and quadrivalent nanoparticle vaccines, have also demonstrated broad protection against multiple influenza strains.

The development of universal influenza vaccines holds significant implications for public health. Current seasonal vaccines require frequent updates and may not always match circulating strains, leading to reduced effectiveness. Universal vaccines targeting conserved antigens could provide consistent and broad protection, reducing the need for annual reformulation and improving pandemic preparedness. The ability to induce long-lasting immunity would also alleviate the burden of repeated vaccinations, potentially increasing vaccination coverage and compliance. Moreover, the broad protection offered by these vaccines could mitigate the impact of antigenic drift and shift, thereby reducing morbidity and mortality associated with influenza outbreaks.

The future of influenza vaccination lies in the continued development and refinement of universal vaccine candidates. Advances in vaccine technology, such as mRNA and nanoparticle platforms, offer promising avenues for achieving broad and durable protection. Future research should focus on optimizing these platforms for human use, ensuring safety, efficacy, and scalability. Additionally, understanding the mechanisms of immune response and the role of preexisting immunity will be crucial in designing effective universal vaccines. Collaborative efforts between researchers, public health organizations, and policymakers will be essential to bring these vaccines to market and integrate them into existing immunization programs. Ultimately, the goal is to achieve a universal influenza vaccine that provides comprehensive protection against all influenza strains, significantly reducing the global burden of influenza.

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Conflict of Interest Disclosure

The author affirms that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

References

Arunkumar G., Ioannou A., Wohlbold T., Meade P., Aslam S., Amanat F., Ayllon J., García-Sastre A., and Krammer F., 2019, Broadly Cross-Reactive, Nonneutralizing Antibodies against Influenza B Virus Hemagglutinin Demonstrate Effector Function-Dependent Protection against Lethal Viral Challenge in Mice, Journal of Virology, 93:6.
https://doi.org/10.1128/JVI.01696-18
PMid:30626682 PMCid:PMC6401450

Arunkumar G., McMahon M., Pavot V., Aramouni M., Ioannou A., Lambe T., Gilbert S., and Krammer F., 2019, Vaccination with viral vectors expressing NP, M1 and chimeric hemagglutinin induces broad protection against influenza virus challenge in mice. Vaccine, 37: 5567-5577.
https://doi.org/10.1016/j.vaccine.2019.07.095
PMid:31399277 PMCid:PMC6717082

Bernasconi V., Bernocchi B., Ye L., Lê M., Omokanye A., Carpentier R., Schön K., Saelens X., Staeheli P., Betbeder D., and Lycke N., 2018, Porous nanoparticles with self-adjuvanting M2e-fusion protein and recombinant hemagglutinin provide strong and broadly protective immunity against influenza virus infections, Frontiers in Immunology, 9: 2060.
https://doi.org/10.3389/fimmu.2018.02060
PMid:30271406 PMCid:PMC6146233

Bernstein D., Guptill J., Naficy A., Nachbagauer R., Berlanda-Scorza F., Feser J., Wilson P., Solórzano A., Wielen M., Walter E., Albrecht R., Buschle K., Chen Y., Claeys C., Dickey M., Dugan H., Ermler M., Freeman D., Gao M., Gast C., Guthmiller J., Hai R., Henry C., Lan L., McNeal M., Palm A., Shaw, D., Stamper C., Sun W., Sutton V., Tepora M., Wahid R., Wenzel H., Wohlbold T., Innis,B., García-Sastre A., Palese P., and Krammer F., 2019, Immunogenicity of chimeric haemagglutinin-based, universal influenza virus vaccine candidates: interim results of a randomised, placebo-controlled, phase 1 clinical trial, The Lancet. Infectious Diseases, 20: 80-91.
https://doi.org/10.1016/S1473-3099(19)30393-7
PMid:31630990

Cadorna-Carlos J., Nolan T., Borja-Tabora C., Santos J., Montalbán M., Looze F., Eizenberg P., Hall S., Dupuy M., Hutagalung Y., Pépin S., and Saville M, 2015, Safety, immunogenicity, and lot-to-lot consistency of a quadrivalent inactivated influenza vaccine in children, adolescents, and adults: A randomized, controlled, phase III trial, Vaccine, 33(21): 2485-2492.
https://doi.org/10.1016/j.vaccine.2015.03.065
PMid:25843270

Carter D., Darby C., Lefoley B., Crevar C., Alefantis T., Oomen R., Anderson,S., Strugnell T., Cortés-Garcia G., Vogel T., Parrington M., Kleanthous H., and Ross T., 2016, Design and Characterization of a Computationally Optimized Broadly Reactive Hemagglutinin Vaccine for H1N1 Influenza Viruses. Journal of Virology, 90: 4720-4734.
https://doi.org/10.1128/JVI.03152-15
PMid:26912624 PMCid:PMC4836330

Cho A., and Wrammert J., 2016, Implications of broadly neutralizing antibodies in the development of a universal influenza vaccine.. Current opinion in virology, 17: 110-115.
https://doi.org/10.1016/j.coviro.2016.03.002
PMid:27031684 PMCid:PMC4940123

Chu K., Xu K., Tang R., Tian X., Hu J., Yang T., Li C., Hu Y., and Zeng, G., 2020, Immunogenicity and safety of an inactivated quadrivalent influenza vaccine: a randomized, double-blind, controlled phase III study in healthy population aged≥ 3 years, Vaccine, 38(37): 5940-5946.
https://doi.org/10.1016/j.vaccine.2020.06.071
PMid:32732142

Corder B.N., Bullard B.L., Poland G.A., and Weaver E.A., 2020, A decade in review: a systematic review of universal influenza vaccines in clinical trials during the 2010 decade, Viruses, 12(10): 1186.
https://doi.org/10.3390/v12101186
PMid:33092070 PMCid:PMC7589362

Coughlan L., and Palese P., 2018, Overcoming barriers in the path to a universal influenza virus vaccine, Cell Host and Microbe, 24(1): 18-24.
https://doi.org/10.1016/j.chom.2018.06.016
PMid:30001520

DiLillo D.J., Palese P., Wilson P.C., and Ravetch J.V., 2016, Broadly neutralizing anti-influenza antibodies require Fc receptor engagement for in vivo protection, The Journal of Clinical Investigation, 126(2): 605-610.
https://doi.org/10.1172/JCI84428
PMid:26731473 PMCid:PMC4731186

Eichelberger M.C., and Monto A.S., 2019, Neuraminidase, the forgotten surface antigen, emerges as an influenza vaccine target for broadened protection, The Journal of Infectious Diseases, 219(Supplement_1): S75-S80.
https://doi.org/10.1093/infdis/jiz017
PMid:30715357 PMCid:PMC7325326

Ekiert D.C., and Wilson I.A., 2012, Broadly neutralizing antibodies against influenza virus and prospects for universal therapies, Current Opinion in Virology, 2(2): 134-141.
https://doi.org/10.1016/j.coviro.2012.02.005
PMid:22482710 PMCid:PMC3368890

Feng D., Liu M., Zhang X., and Xu J., 2018, Research progress in broad neutralizing antibodies and universal vaccines against influenza A virus, Chinese Journal of Microbiology and Immunology, 782-789.

Freyn A.W., da Silva J.R., Rosado V.C., Bliss C.M., Pine M., Mui B.L., Tam Y., Madden T., Ferreira L., Weissman D., Krammer F., Coughlan L., Palese P., Pardi N., and Nachbagauer R., 2020, A multi-targeting, nucleoside-modified mRNA influenza virus vaccine provides broad protection in mice, Molecular Therapy, 28(7): 1569-1584.
https://doi.org/10.1016/j.ymthe.2020.04.018
PMid:32359470 PMCid:PMC7335735

Isakova-Sivak I., Matyushenko V., Stepanova E., Matushkina A., Kotomina T., Mezhenskaya D., Prokopenko P., Kudryavtsev I., Kopeykin P., Sivak K., and Rudenko L., 2020, Recombinant live attenuated influenza vaccine viruses carrying conserved T cell epitopes of human adenoviruses induce functional cytotoxic T cell responses and protect mice against both infections, Vaccines, 8(2): 196.
https://doi.org/10.3390/vaccines8020196
PMid:32344618 PMCid:PMC7349758

Isakova-Sivak I., Stepanova E., Mezhenskaya D., Matyushenko V., Prokopenko P., Sychev I., Wong P., and Rudenko L., 2021, Influenza vaccine: progress in a vaccine that elicits a broad immune response, Expert Review of Vaccines, 20(9): 1097-1112.
https://doi.org/10.1080/14760584.2021.1964961
PMid:34348561

Jang Y.H., and Seong B.L., 2019, The quest for a truly universal influenza vaccine, Frontiers in Cellular and Infection Microbiology, 9: 344.
https://doi.org/10.3389/fcimb.2019.00344
PMid:31649895 PMCid:PMC6795694

Jang Y.H., and Seong B.L., 2020, Call for a paradigm shift in the design of universal influenza vaccines by harnessing multiple correlates of protection, Expert Opinion on Drug Discovery, 15(12): 1441-1455.
https://doi.org/10.1080/17460441.2020.1801629
PMid:32783765

Jansen J.M., Gerlach T., Elbahesh H., Rimmelzwaan G.F., and Saletti G., 2019, Influenza virus-specific CD4+ and CD8+ T cell-mediated immunity induced by infection and vaccination, Journal of Clinical Virology, 119: 44-52.
https://doi.org/10.1016/j.jcv.2019.08.009
PMid:31491709

Jazayeri S.D., and Poh C.L., 2019, Development of universal influenza vaccines targeting conserved viral proteins, Vaccines, 7(4): 169.
https://doi.org/10.3390/vaccines7040169
PMid:31683888 PMCid:PMC6963725

Kim Y.J., Ko E.J., Kim M.C., Lee Y.N., Kim K.H., Jung Y.J., and Kang S.M., 2017, Roles of antibodies to influenza A virus hemagglutinin, neuraminidase, and M2e in conferring cross protection, Biochemical and Biophysical Research Communications, 493(1): 393-398.
https://doi.org/10.1016/j.bbrc.2017.09.011
PMid:28887040 PMCid:PMC5636684

Krammer F., and Palese P., 2019, Universal influenza virus vaccines that target the conserved hemagglutinin stalk and conserved sites in the head domain, The Journal of Infectious Diseases, 219(Supplement_1): S62-S67.
https://doi.org/10.1093/infdis/jiy711
PMid:30715353 PMCid:PMC6452318

Krammer F., Pica N., Hai R., Margine I., and Palese P., 2013, Chimeric hemagglutinin influenza virus vaccine constructs elicit broadly protective stalk-specific antibodies, Journal of Virology, 87(12): 6542-6550.
https://doi.org/10.1128/JVI.00641-13
PMid:23576508 PMCid:PMC3676110

Liebowitz D., Gottlieb K., Kolhatkar N.S., Garg S.J., Asher J.M., Nazareno J., Kim K., McIlwain D., and Tucker S.N., 2020, Efficacy, immunogenicity, and safety of an oral influenza vaccine: a placebo-controlled and active-controlled phase 2 human challenge study, The Lancet Infectious Diseases, 20(4): 435-444.
https://doi.org/10.1016/S1473-3099(19)30584-5
PMid:31978354

Lo C.Y., Misplon J.A., Li X., Price G.E., Ye Z., and Epstein S.L., 2021, Universal influenza vaccine based on conserved antigens provides long-term durability of immune responses and durable broad protection against diverse challenge virus strains in mice, Vaccine, 39(33): 4628-4640.
https://doi.org/10.1016/j.vaccine.2021.06.072
PMid:34226103

Memoli M.J., Han A., Walters K.A., Czajkowski L., Reed S., Athota R., Rosas L., Cervantes-Medina A., Park J., Morens D., Kash J., and Taubenberger J.K., 2020, Influenza a reinfection in sequential human challenge: implications for protective immunity and "universal" vaccine development, Clinical Infectious Diseases, 70(5): 748-753.
https://doi.org/10.1093/cid/ciz281
PMid:30953061 PMCid:PMC7319262

Nabel G.J., and Fauci A.S., 2010, Induction of unnatural immunity: prospects for a broadly protective universal influenza vaccine, Nature Medicine, 16(12): 1389-1391.
https://doi.org/10.1038/nm1210-1389
PMid:21135852

Nachbagauer R., Feser J., Naficy A., Bernstein D.I., Guptill J., Walter E.B., Berlanda-Scorza F., Stadlbauer D., Wilson P., Aydillo T., Behzadi M., Bhavsar D., Bliss C., Capuano C., Carreño J., Chromikova V., Claeys C., Coughlan L., Freyn A., Gast C., Javier A., Jiang K., Mariottini C., McMahon M., McNeal M., Solórzano A., Strohmeier S., Sun W., Wielen M., Innis B., García-Sastre A., Palese P., and Krammer F., 2021, A chimeric hemagglutinin-based universal influenza virus vaccine approach induces broad and long-lasting immunity in a randomized, placebo-controlled phase I trial, Nature Medicine, 27(1): 106-114.
https://doi.org/10.1038/s41591-020-1118-7
PMid:33288923

Oftung F., Næss L.M., Laake I., Stoloff G., and Pleguezuelos O., 2022, FLU-v, a broad-spectrum influenza vaccine, induces cross-reactive cellular immune responses in humans measured by dual IFN-γ and granzyme B ELISpot assay, Vaccines, 10(9): 1528.
https://doi.org/10.3390/vaccines10091528
PMid:36146606 PMCid:PMC9505334

Pica N., and Palese P., 2013, Toward a universal influenza virus vaccine: prospects and challenges, Annual Review of Medicine, 64(1): 189-202.
https://doi.org/10.1146/annurev-med-120611-145115
PMid:23327522

Pillet S., Couillard J., Trépanier S., Poulin J.F., Yassine-Diab B., Guy B., Ward B., and Landry N., 2019, Immunogenicity and safety of a quadrivalent plant-derived virus like particle influenza vaccine candidate-two randomized Phase II clinical trials in 18 to 49 and≥ 50 years old adults, PloS one, 14(6): e0216533.
https://doi.org/10.1371/journal.pone.0216533
PMid:31166987 PMCid:PMC6550445

Qiao Y., Li S., Jin S., Pan Y., Shi Y., Kong W., and Shan Y., 2022, A self-assembling nanoparticle vaccine targeting the conserved epitope of influenza virus hemagglutinin stem elicits a cross-protective immune response, Nanoscale, 14(8): 3250-3260.
https://doi.org/10.1039/D1NR08460G
PMid:35157751

Rappazzo C.G., Watkins H.C., Guarino C.M., Chau A., Lopez J.L., DeLisa M.P., Leifer C., Whittaker G., and Putnam D., 2016, Recombinant M2e outer membrane vesicle vaccines protect against lethal influenza A challenge in BALB/c mice, Vaccine, 34(10): 1252-1258.
https://doi.org/10.1016/j.vaccine.2016.01.028
PMid:26827663

Rimmelzwaan G.F., Fouchier R.A., and Osterhaus A.D., 2007, Influenza virus-specific cytotoxic T lymphocytes: a correlate of protection and a basis for vaccine development, Current Opinion in Biotechnology, 18(6): 529-536.
https://doi.org/10.1016/j.copbio.2007.11.002
PMid:18083548

Sakala I.G., Honda-Okubo Y., Li L., Baldwin J., and Petrovsky N., 2021, A M2 protein-based universal influenza vaccine containing Advax-SM adjuvant provides newborn protection via maternal or neonatal immunization, Vaccine, 39(36): 5162-5172.
https://doi.org/10.1016/j.vaccine.2021.07.037
PMid:34362601

Scorza F.B., Tsvetnitsky V., and Donnelly J.J., 2016, Universal influenza vaccines: shifting to better vaccines, Vaccine, 34(26): 2926-2933.
https://doi.org/10.1016/j.vaccine.2016.03.085
PMid:27038130 PMCid:PMC4899887

Stadlbauer D., Zhu X., McMahon M., Turner J.S., Wohlbold T.J., Schmitz A.J., Strohmeier S., Yu W., Nachbagauer R., Mudd P., Wilson I., Ellebedy A., and Krammer F., 2019, Broadly protective human antibodies that target the active site of influenza virus neuraminidase, Science, 366(6464): 499-504.
https://doi.org/10.1126/science.aay0678
PMid:31649200 PMCid:PMC7105897

Vemula S.V., Sayedahmed E.E., Sambhara S., and Mittal S.K., 2017, Vaccine approaches conferring cross-protection against influenza viruses, Expert Review of Vaccines, 16(11): 1141-1154.
https://doi.org/10.1080/14760584.2017.1379396
PMid:28925296 PMCid:PMC6005355

Vogel O.A., and Manicassamy B., 2020, Broadly protective strategies against influenza viruses: universal vaccines and therapeutics, Frontiers in Microbiology, 11: 135.
https://doi.org/10.3389/fmicb.2020.00135
PMid:32117155 PMCid:PMC7020694

Wang Y., Li S., Dong C., Ma Y., Song Y., Zhu W., Kim J., Deng L., Denning T., Kang S., Prausnitz M., and Wang B.Z., 2021, Skin vaccination with dissolvable microneedle patches incorporating influenza neuraminidase and flagellin protein nanoparticles induces broad immune protection against multiple influenza viruses, ACS Applied Bio Materials, 4(6): 4953-4961.
https://doi.org/10.1021/acsabm.1c00240
PMid:34179728 PMCid:PMC8232372

Wei C.J., Boyington J.C., McTamney P.M., Kong W.P., Pearce M.B., Xu L., Andersen H., Rao S., Tumpey T., Yang Z., and Nabel G.J., 2010, Induction of broadly neutralizing H1N1 influenza antibodies by vaccination, Science, 329(5995): 1060-1064.
https://doi.org/10.1126/science.1192517
PMid:20647428

Zhang J., Fan H.Y., Zhang Z., Zhang J., Zhang J., Huang J.N., Ye Y., and Liao M., 2016, Recombinant baculovirus vaccine containing multiple M2e and adjuvant LTB induces T cell dependent, cross-clade protection against H5N1 influenza virus in mice, Vaccine, 34(5): 622-629.
https://doi.org/10.1016/j.vaccine.2015.12.039
PMid:26724200

Zhang Y., Xu C., Zhang H., Liu G.D., Xue C., and Cao Y., 2019, Targeting hemagglutinin: approaches for broad protection against the influenza a virus, Viruses, 11(5): 405.
https://doi.org/10.3390/v11050405
PMid:31052339 PMCid:PMC6563292

Zhao C., and Xu J., 2018, Toward universal influenza virus vaccines: from natural infection to vaccination strategy, Current Opinion in Immunology, 53: 1-6.
https://doi.org/10.1016/j.coi.2018.03.020
PMid:29604600

Zost S.J., Wu N.C., Hensley S.E., and Wilson I.A., 2019, Immunodominance and antigenic variation of influenza virus hemagglutinin: implications for design of universal vaccine immunogens, The Journal of Infectious Diseases, 219(Supplement_1): S38-S45.
https://doi.org/10.1093/infdis/jiy696
PMid:30535315 PMCid:PMC6452323