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Review Article

Animal Virus Evolution under Vaccine Pressure: From Immune Escape Mechanisms to Next-Generation Vaccine Design

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Abstract: This review examines the critical challenge of evolving animal virus resistance to vaccines, a phenomenon threatening veterinary medicine, global food security, and public health. While effective for controlling bacterial diseases, vaccination against rapidly mutating RNA viruses often imposes strong selective pressures, driving the emergence of antigenic variants that evade host immunity. These vaccines escape mutants, evidenced in viruses like canine parvovirus (CPV), avian influenza (AIV), and foot-and-mouth disease (FMD), lead to outbreaks in vaccinated populations and complicate disease management. The paper elucidates the mechanisms behind this resistance, primarily genetic mutations in antigenic sites and sophisticated immune evasion strategies, which enable viruses to circumvent neutralization by vaccine-induced antibodies. It critically evaluates the limitations of current vaccine technologies, including inactivated, live-attenuated, and subunit vaccines, noting that imperfect immunity can inadvertently promote the selection of resistant strains. To address this ongoing evolutionary arms race, the authors advocate for a paradigm shift in vaccine design. They propose leveraging advanced technologies such as viral vector platforms, structural biology, and predictive modeling to develop next-generation vaccines targeting conserved epitopes. Furthermore, the review emphasizes the necessity of global surveillance programs to monitor viral evolution in real-time and calls for a collaborative, interdisciplinary approach to create more robust and future-proof vaccination strategies, thereby mitigating the risk of vaccine-driven viral and ensuring long term efficacy.

Keywords: Animal Viruses, Vaccine Escape, Viral Evolution, Immune Evasion and Next-Generation Vaccines.

1. INTRODUCTION

Effective vaccination strategies have led to the successful control and eradication of many infectious diseases in livestock. However, rapid evolution (antigenic drift) of viruses often renders vaccines available less effective, resulting in disease outbreaks even in vaccinated populations. Some viral diseases in livestock, such as foot-and-mouth disease (FMD) and classical swine fever (CSF), highlight how vigilant surveillance and development of proactive vaccines can mitigate the emergence of new viral variants. Designing vaccines to prevent zoonotic diseases is daunting because the animal viruses that threaten human health are poorly immunogenic, genetically diverse (quasispecies), and naturally evolve in environments that cannot be readily sampled *(Elrashedy et al., 2025)*

Advancements in rapidly improving viral genome sequence data pre-and post-outbreak, coupled with sophisticated reverse genetics techniques and structural biology information on antigenic determinants on virions, provide opportunities to design more effective vaccines against such viruses. Several recent studies provide examples of employing this approach to control emerging threats of viruses of livestock origin that also infect humans. However, there is a need for a broader collaborative approach to research on refining and applying these or similar approaches by virologists, immunologists and structural biologists globally and under more stringent measures against convenient, continuing emergence of new zoonotic viruses (*J. King et al., 2022*).

RNA viruses are defined by a rapid accumulation of mutations leading to a heterogeneous swarm of individual variants (quasispecies) that co-exist in an infected host. The speed at which antigenic variants can emerge through the mutation of vaccine strains is highlighted by the evolution of resistances of animal viruses to vaccination. A number of virologists have studied the evolution of viruses in the context of vaccination. Examples are mentioned where neutralizing antibodies (NAbs) or T cell epitopes elicited by previous vaccination or infection exerted strong selection pressures on viruses, which in turn generated resistant variants, allowing re-infection of previously immune hosts, despite the mutations occurring in highly conserved genome or proteome regions (*C Layman et al., 2021).*

2. Overview of Animal Viruses

Animal diseases have parasite infection as well as viral infection, in this paper the discussion will be limited to viral diseases. Domestic animals are used for many purposes, and a lot of them are kept as pets only. They are helpful for humans to alleviate stress and are trained to do various tasks (*Brun, 2015*). Some are harvested for their products, meat, skin and dairy products, and generally involved in agriculture or some economic sectors. Aquaculture is the fastest growing sector, and animal viruses have not been an important concern until recently. With the globalization of animal products, the awareness and surveillance of these diseases have significantly increased, and the health maintenance and pre emptied control of animal populations are becoming more important. Due to the rapid evolution of domestic animals, in particular poultry, offspring species of disease viruses recognizing the hosts' incompatibility with the vaccine strains have continued to evolve, and as a result, there are now some residual virulent strains at a considerable rate in a vaccinated flock (*Salaheldin et al., 2017).*

The emergences of residual virulent strains have been revealed in many cases, and difficulties against further control of these diseases have been reported. However, despite the extensive investigations, knowledge of the basic mechanisms of emergence remains lacking, and there are not so provoking reports at the molecular level. Globally, animal industry and aquaculture businesses play important roles in conventional or organic food supplies and latest biopharmaceutics. Concurrently, animal viruses cause numerous diseases, which are irretrievable loss of economic productivity, food supply, or safety to humans through zoonosis; perceived public health risks are bona fide consequences. In consideration of local situations and insurability, policymakers implement vaccination programs, and vaccines are designed with the hope that infected hosts develop protective immunity (*Lee NH et al., 2012*).

3. Vaccine Development History

It is appropriate at this point to consider the history of vaccine development for animal viruses, particularly focusing on vaccines influenced by contemporary knowledge of pathogenesis of the diseases. Vaccination against these diseases dates back more than a century. Rabies virus vaccines were developed in 1885 by Louis Pasteur. Rinderpest virus vaccines were developed in 1914 by Waldemar Haffkine, who was inspired by Pasteur's work on rabies. Fowl plague virus vaccines were developed in 1878 following observations by Pasteur. The mid-20th century saw the introduction of vaccines for foot and mouth disease, avian infectious bronchitis, and an important vaccine for canine distemper, all based on interval passage in embryonated eggs or cell cultures. With the advent of molecular biology in the mid-1970s, attempts to produce vaccines using recombinant DNA technology began. Vaccine vectors based on vaccinia, fowl pox and adenoviruses were developed which used the polyhedron gene, which encoded the viral lipid protein, as insertion sites for genes of other viruses. Remarkably, a similar approach was adopted for the development of human vaccines against smallpox, hepatitis B virus and HIV-1. More recently, commercially available sub-unit vaccines have been produced, using protein sub-units derived from several viruses, including parvoviruses, hepatitis B virus and FMDV (*A Plotkin, 2005*).

4. Mechanisms of Viral Resistance

Thanks to advances in molecular biology, mechanics of resistance of many endogenous viruses, the switch from productive reproduction in a pre-viral ancestor to dormant reproduction in the pre-cellular ancestor of viruses, or the permanence of newer viruses, have been studied to manageable parameters. Despite the hindsight of retro-alphavirus common ancestry the switch to endogenous alphavirus status is described as poorly understood. Viral genomes can pop up quickly in pre-cells or cells from pre-cellular cycles of events. Note that the discussion of the switch to cytoplasmically replicated birth control on DNA-monomer vectors has been somewhat accumulated. Infection and hyperinfection have also been postulated to be linked, but much of the data would appear anecdotal at best. Resistance of the chronically infected cell is universal against synthesising viruses (*B. Bang, 2008).*

Observations from fish cells that have been treated to lose infectious progeny suggest a two-component ratchet. It may also be possible to create too strong a selective pressure when used as a therapy. Contractile cells with three septa like bacterial T4s can infect and/or reproduce profusely in 12 h, making laboratory-induced resistance unlikely to be robust in the wild, and if ABC bentonite cap-shaped viruses eluded detection (*Kariithi et al.,2018*)

4.1. Genetic Mutations

Genetic changes have played a significant role in the evolution of many viruses, enabling them to evade vaccine-induced immunity by altering their immunological properties. Although it is often difficult to determine which mutations contribute to antigenic changes, the function of many of the viral proteins has been elucidated. As viruses evolve, their sinonasal and pharyngeal replication sites range from avian to swine to human, resulting in increased virulence in humans. The virus must adapt to the specific receptors and innate defenses in each new host. Changes in viruses that were previously adapted to one host to arise in those with a different innate immune system have been noted (*B. Bang, 2008*). Exploring the genetic basis of this adaptation is complicated because many mutations are necessary for virulence and many of these changes are correlated. Nevertheless, recent developments in high-throughput sequencing technology have resulted in the finding of sequences that correlate with virulence. A microarray was developed for the characterization of the BDV genome, in contrast to the available methods for many other viruses. It turned out to be a useful tool for investigating the variability of the BDV genome. Based on its characterization, new genotypes could be established and tools for the determination of genetic variants and other genetic changes provided (*S. Novella et al., 1995).*

4.2. Immune Evasion Strategies

As noted in the reports, viruses have evolved a host of strategies to evade host antiviral immune responses at all levels. There is a complex relationship between host population immunity and the emergence of new viral variants. Importantly, protective immunity can wane for both humans and animals. For example, the use of vaccines such as the ones against canine parvovirus offer good protection against severe disease, but do not prevent infection and shedding of the virus. From a virologist’s viewpoint, the virus must be faced in terms of its composition, replication strategies, and interaction with host factors, especially those that regulate the surface proteins. In addition, a co-evolution model of host and virus matched in time, environment, maintenance strategy and other factors is important. Viruses employ different mechanisms to escape neutralization by the host immune system (*Jie Wen Tay et al., 2022*).

Plaque reduction neutralization tests use infectious virus displays and secreted viruses, either with cells in suspension culture, or plaque assays on 2D or 3D culture. Virus neutralization on cells in one (or more) compartments will be a continuing theme in virology. Cell-to-cell transmission will involve membrane fusion in the cytoplasm. Viruses such as Papilloma. To infect nearby cells, viruses like Papilloma, HIV, and others use a range of biological mechanisms that are remarkably complex. Pragmatic viral transmissibility (T) mutations are thought to be an evolutionary route for evading host immunity and spreading to other hosts (and evading medications). The Bunya virus's gene alterations for enhanced T, virulence, and pathogenicity serve as one example. Viruses may attempt to finally evade the demands of vaccinations, immunological memory, or monoclonal antibodies, depending on host conditions and the period in evolutionary cues. On the virus's surface, these pigmentation spots may be widespread or confined. They might be for distinct antibodies, resulting in various patterns of recognition, access, and neutralization. Individual mutants may abruptly take over a community, but they may also be Classic examples are provided by rye cessation of smallpox vaccination as a punctuated equilibrium (*Guimond et al., 2022)*

5. Case Studies of Resistance

Resistant strains of viruses have advanced against vaccine strains derived from attenuated viruses and against vaccines based on inactivated viruses. Examples come from studies of organisms that are routinely used for research and have a long history of laboratory domestication. Studies are most advanced with canine parvovirus (*K. Irwin et al., 2016*). The emergence of resistance was foreshadowed by the development of resistant strains of feline panleukopenia virus, which had an almost identical amino acid sequence. A single mutation in the canine parvovirus vaccine strain is responsible for the resistance of field isolates. Under selective pressure from the vaccine, this strain became extinct in less than a year. The inactivated vaccine against vesicular stomatitis virus was in widespread use for 30 years, but a virus resistant to neutralization by the vaccine appeared in the field. The resistant strains had a cumulative difference of 21 amino acid substitutions from the original serotype, and out of eight variants tested, none could be neutralized by the original sera (*K. Irwin et al., 2016).*

Two inactivated fowl plague virus vaccines produced by different laboratories elicited different immune responses in the same flock of chickens. The underlying changes in the viruses were identified as major antigenic determinants and were the product of divergent evolution. It is clear that animal viruses can develop resistance to vaccine strains quite rapidly, often with simple mutations of two to three nucleotides causing major changes in viral properties. The evolution of these variants is documented. In the case of viruses like vesicular stomatitis virus and fowl plague virus, laboratory studies have provided some insights into the mechanisms of evolution. However, detailed studies of the resistance mechanisms adopted by canine parvovirus variants are few in number. These studies have profound implications for understanding resistance in as yet untouched viruses, especially those inflicting massive mortality upon naive populations like the human immunodeficiency virus (*Voorhees et al., 2019)*

5.1. Avian Influenza

Vaccination has been shown to be an effective method of controlling the virus's spread, particularly in chickens (*Michael Becker, 2015*). Also, the H5N2 strains have altered the HA gene and predicted a global pandemic potential. Hence, it is important to study the evolution of AIV in vaccinated chicken flocks. A large-scale and long-term AIV vaccination program instituted in Mexico in 1995 is analyzed here. (*Escorcia et al., 2008*).

5.2. Canine Parvovirus

Vaccination is one of the most relevant measures adopted worldwide to control CPV, and the great majority of commercial vaccines currently used were developed during the early 1980s. First CPV-2 vaccines were prepared using either strains of CPV-2 itself or CPV-2a isolates, which are still used in most P.I. and live-attenuated vaccines. Despite the wide use of these vaccines for nearly three decades, CVA-iCPV strains re-emerged during the late 1990s, found in pups dying of canine parvoviral enteritis. Furthermore, with the introduction of CPV-2c in the United Kingdom since 2008, including after the wide use of type 2a and 2b vaccines, severe CPV-2c infections have been recently reported also in vaccinated dogs. *(Decaro & Buonavoglia, 2012*)

Concerns about the real efficacy of CPV-2 vaccines against the antigenic variants currently circulating in the field have been based mainly on in vitro cross-neutralization assays. However, either the data obtained are irrelevant in vivo, as suggested by the residual prevalence of vaccine type strains in vaccinated pups, or vaccine failures occurred thus allowing clinical disease to be detected. The relevant aspect of disease control is that the majority of CPV infections occur in puppies when MDA wane and at the time when the animals became susceptible to any strain of the virus. Most of the vaccine companies are actually and continuously updating the sequence of the CPV used in their vaccines, but not all these new strains have yet been extensively tested in field conditions. Given the current level of vaccination of the canine population, it would be of utmost relevance to understand the mechanisms behind the emergence of the antigenic variant strains in areas of high vaccination coverage. Continuous evolution of CPV through point mutations in the viral genome presently has important implications for viral diagnostics, vaccine production, epidemiologic investigations, and the assessment of the influence of host-derived factors on immunity and viral pathogenesis. The emergence of new antigenic variants may require a revision of vaccination programs and the update of the viral strains in commercially available products (*Decaro et al., 2020*)

5.3. Feline Immunodeficiency Virus

There are two groups of lentiviruses that infect domestic, wild or semi-wild feline species in veterinary circumstances: the feline immunodeficiency virus (FIV) and the feline foamy virus (FFV). Both biological agents are the subject of active research, although with a very different emphasis. Studies of FIV are directed at understanding the biology and evolution of a virus that is similar but not homologous to the human immunodeficiency virus (HIV), which is one of the etiological agents of AIDS in human beings. FIV-induced disease in cats resembles that of AIDS and AIDS-related complex in human beings both virologically and clinically, raising concerns regarding a possible interspecies transmission of FIV, and the social impact of such a transmission should it occur. FIV-induced disease in cats is a notifiable disease in the EU (*Hosie et al., 2002*).

6. Current Vaccination Strategies

Disease controlling vaccines cannot completely wipe-out viruses from the infected animal population. If a virus is allowed to circulate in a population where vaccinated and unvaccinated host individuals coexist, if the vaccine does not induce sterilizing immunity, as it is the case with many veterinary vaccines, viruses with an altered antigenic profile might be gradually selected (*Domingo, 2016*). These events in the case of vaccines used in veterinary medicine may alter the cell tropism and host range of viruses, increasing the possibilities of zoonotic transmission to humans. For human viruses, an obvious example is the selection and spread of a vaccine-escape mutant of the mumps virus in a population where the majority of children had received the mumps vaccine Further evidence for vaccine escape mutants is available for hepatitis A and B viruses. (*Domingo, 2016*).

Vaccines rarely provide protection for all vaccinated individuals. Many mechanisms play a role: differences in vaccine acceptance, immunosuppression in vaccine recipients, insufficient time between vaccination and exposure to viral pathogens, antigenic differences between vaccine strains and circulating viruses, and inappropriate use of vaccines. Vaccination can sometimes promote the selection of antigenic variants that escape humoral immune responses. Some yellow fever vaccinees may develop post-vaccinal viscerotropic disease. Intensively used vaccinia virus vaccines may promote the selection of antigenic variants that do escape from the humoral immune response. In aquaculture, vaccination against infectious salmon anemia (ISA) using inactivated vaccines may promote the selection of viruses with modified tropism involving a shift from a mainly erythrocytic virus toward a broader tropism targeting endothelial cells. Attenuated vaccine strains may revert toward virulent counterparts that do readily infect the vaccinated population. Reversion of live-attenuated vaccine viruses into virulent forms is a cause of disease derived from the evolutionary potential of viruses (*Roukens et al.,2011*).

6.1. Inactivated Vaccines

Viruses have been inactivated by attachment to aluminum oxide. Five viruses representing five families of viral pathogens were purified by differential centrifugation. Viruses attached to pretreated aluminum oxide were inactivated by either heating at 100 °C or exposure to phenolic disinfectants such as ethylphenol and paravuytylphenol. Heating at 100 °C inactivated Encephalomyocarditis, Foot-and-mouth, Influenza, and Inoroviruses to their detection limits, but mumps virus required heating for five times longer. Repeated sevenfold washes were required for removal of contaminating virus from purified, disinfectant-treated aluminum oxide which was not required for aluminum oxide purified by other methods (*Tanneru et al., 2014*). Types of inactivated poliovirus vaccines, IPV and bOPV, were similar except for the types of attenuated viral strains. The IPV vaccine was manufactured using the Mahoney and MEF1 strains for types 1 and 3, respectively. The Sabin strains were used for the bOPV vaccine. Both IPV and bOPV vaccines were lyophilized and stored at −20 °C. Their storage temperatures were monitored continuously by digital data loggers with a lower limit of −30 °C and an upper limit of 45 °C. Observations of a long-term frozen lot of IPV samples revealed that a gradual temperature increase at −18 °C had occurred, which was of concern with regard to the stability of the vaccine (*Das et al., 2020*).

6.2. Live Attenuated Vaccines

Recurrently, vaccination failure can be attributed to an insufficient response from vaccine-induced antibodies. The FDA has approved three live oral vaccines against cholera. Two vaccines against typhoid fever and a total of three recombinant vaccines for hepatitis B with CE-deleted vectors. Similar vaccines against adenovirus are in development. Newly developed subunit genetic vaccines are being introduced on the market, including the Simian Virus-40 vaccine against prostate cancer, and vaccines against malignant melanoma and viruses. Several viruses already licensed as live vaccines can delay, but not prevent, a viral infection. Live, virulent sera and vaccines induce a high probability of vaccine induced as well as vaccine escape mutants, a phenomenon rarely observed in Subunit vaccines, and almost absent in DNA and protein vaccines. Concerns about their use have prompted calls for a ban on live vaccines. Virus vector vaccines might be thought to have a chance of being better and safer than live vaccines. After all, vector vaccines are only using specific genes of the virus for expression or replication. This advantage is counterbalanced by their own hurdles. The innate immune response in the vector host can prematurely destroy the vector and hence the transgene cultivars. The vector itself is often immunogenic, limiting the repetitive enabling use. Second generation vectors have incorporated transgenes of viruses like hepatitis B virus to reduce immunogenicity but at TCOs of conserved antigens, rendering them essentially an expanded SET. Multiple and different vectors need to be used, and combination vectors are needed to circumvent the absence of antigen-specific T celularity and subsequent cellular entry and antibody responses (*S. Lauring et al., 2010).*

6.3. Subunit Vaccines

Subunit vaccines, either proteins or polysaccharides, produced from known virulence determinants of the pathogen have been employed successfully for many years to immunize animals against disease, although these prototypes are not free of disadvantages. In some cases, such as aerogenic infection with avian respiratory tract viruses or chronic respiratory disease caused by infection with mycoplasmas, no subunit vaccines are commercially available (*Babiuk & Potter, 2002*). In other cases, most notably pseudorabies virus infections in cattle, the immunogenicity of purified viral glycoproteins has proven insufficient to protect against virulent challenge. A good target for the future vaccine design, which will better satisfy and comply with the legislation relating to the manufacture of vaccines and veterinary medicinal product as a whole, wouldbe protein or polysaccharide subunit vaccines, formulated in a suitable adjuvant. The safety and efficacy of such products will depend on the tenor and quality of the production process, and quality control by polymerase chain reaction (PCR) or alternatively by reverse transcription-PCR (RT-PCR) should be sufficient to prevent the possibility of multiple incorporation of the vaccine virus in the genetically engineered live virus vector used for vaccine production (*Manukyan et al., 2019*).

7. Emerging Technologies in Vaccine Development

Vaccination has been the cornerstone for the control of several endemic and zoonotic viral diseases; however, viruses evolve rapidly in response to vaccination, which often leads to mismatches between vaccine strains and circulating field strains. This shortcoming has been evidenced very clearly in the case of the viral diseases caused by RNA viruses such as influenza or West Nile virus where mismatched vaccines fail to induce adequate immunity. The emergence of vaccine-resistant viruses has drawn great attention during past outbreaks of animal diseases such as classical swine fever or avian Influenza. The close monitoring of vaccine–virus co-evolution is crucial to mitigate the emergence and the wide spread of porcine reproductive and respiratory syndrome viruses, which are highly genetically and antigenically diverse and evolve considerably faster than current vaccinations in pigs (*Brun, 2015*).

Rapid antigenic evolution of viruses in both vaccinees and non-vaccinees presents a challenge to vaccine strain selection and prompts a shift from strain-based to element-based or system-based vaccine design. The ultimate goal of vaccine design is to create a prolonged, lasting, and stronger immune response. Vaccinated animals exhibit complex immune responses

against vaccinal viruses and generating antibodies or CTLs against a large array of viral epitopes. A deep characterization of broadly neutralizing antibodies or protective T-Cell epitopes will help design next-generation vaccines. Understanding how neutralization is achieved may allow one to construct next-generation or mixture vaccines capable of evoking similar immune responses as those observed in the natural cases. Tuning several variables such as the size of peptides and the position and number of mutations will aid determine CTL epitopes that are effective in evoking immune responses capable of recognizing both vaccine viruses and viruses that escape vaccination (*Valkenburg et al., 2013).*

With the integration of next-generation sequencing, deep learning, structural biology, and drug modeling, a new, deep understanding of antigenic drift and antigenic evolution is growing. An increasing number of viral pathogens have been sequenced, expanding the potential for hunting conserved epitopes or performing unbiased screening of possible peptide libraries. The recently developed approaches allow significant improvements in the identification of CTL epitopes. Memory B cells are likely the sources of highly mutated bNAbs. To meet the challenge of more complex virus populations, a strategy to target B-cell memory is needed, which is much difficult to the classical vaccine design (*Castelli et al., 2013*).

7.1. Viral Vector Vaccines

Different viral vectors comprising replicating or non-replicating viruses represent potential vaccine platforms. Viral vectors which are common in both veterinary and human vaccines include Venezuelan equine encephalitis virus (VEEV), flavivirus, alphavirus, poliovirus, vesicular stomatitis virus (VSV), etc. Compiled data from studies performed with viral vectors demonstrate that most of the above vectors have been successfully employed in animal vaccine research and many of these are in advanced clinical stages for use in human clinical trials or are in clinical use. The safety and efficacy profiles of the various vectors also display differences. Non-replicating viruses tend to elicit lower immunogenicity than their replicating counterparts. Nevertheless, newer strategies are able to exploit the ability of non-replicating viruses to target antigen to dendritic cells which improves immunogenicity (*Robert-Guroff, 2007*). Interestingly, since pox viruses were controlled using vaccination, the research and developments of pox virus vaccines have declined in the last two decades compared to the previously favored viral vectors. In contrast, the recently emerged viral vectors are promising candidates to be evaluated for human and animal vaccination to replace the older vaccine strategies. A common strategy for vaccine development against various infectious agents is to introduce immunogenic viral surface proteins into viral expression vectors for verification of antigen expression in vertebrate cells, and by immunization studies in animal models to evaluate immune responses as well as potential protection against lethal doses of pathogenic infectious agents (*Lundstrom, 2020*).

7.2. Impact of Zinc Methionine and Vaccine on Broiler Performance

The trial was done to determine if there was a positive effect by the supplement, Zinc Methionine, on the performance of the body in the ROSS 308 strain broilers vaccinated to prevent Newcastle disease (ND)There was enhanced growth performance in the group supplemented with Zinc Methionine compared to the control group. We make the recommendation for inclusion in the diet alongside the giving of the vaccines (killed and eye drop) (*Abd and Ali.,2024*)

8. Challenges in Vaccine Efficacy

While vaccination is highly effective in eliminating many animals' viral infections, it has been hampered by a number of unresolved issues. Vaccines that do not completely eliminate wild-type virus have been shown to select for naturally modified virus variants that elude vaccine-induced immunity much like other immunological escape mechanisms in cancers and other diseases. Furthermore, newer methods of bio vaccination using genetically modified viruses capable of expressing heterologous vaccine antigens raise concerns over post-vaccination emergence of virulent vaccine variants (*Barnett & J. Civitello, 2020).*

Imperfect vaccines can thus be thought of in terms of their effects on the host's resistance phenotypes in a population vaccination context. Three possible resistance effects are anti-infection, antidisease, and ant transmission. Vaccines which provide durable levels of anti-infection effectiveness are often the most powerful at altering epidemic dynamics and pathogen evolution. Antidisease effects are often short-lived, as pathogens adapt to tolerance-related host changes much like drug tolerance. Vaccines that confer primarily anti transmission effects are generally less effective in averting outbreaks than anti-infection vaccines but can nonetheless be effective at controlling interspecies pathogen spillover events (*C Layman et al., 2021*).

8.1. Vaccine Coverage and Herd Immunity

The vaccination rate is critical for vaccine control to be successful. The minimum coverage needed to eliminate an infection or disease, the vaccination threshold, can be defined in a variety of ways. Since unvaccinated hosts are assumed to remain susceptible, the basic reproduction ratio R0 must be less than one if the infection is to be extirpated. Policymakers have been counseled to aim for a coverage threshold equal to the inverse of this ratio, but this is somewhat misleading (*Barnett & J. Civitello, 2020*).

Vaccine coverage can be lower or R0 can still fall below one due to natural mortality or a return to a highly virulent host pathogen strain. Because R0 = R0u

× (1 - c), policymakers could optimize their efforts and seek to lower R0u or nudge c closer to one. B16 serves as a case study in how changes in vaccine dynamics and host demography made vaccination ineffective. Feral pig populations in Florida grew rapidly in the latter half of the twentieth century. In contemporaneously modeling this epidemic with swine fever, only short lags to extinction were predicted unless vaccination coverage was increased to greater than 90 percent. A combination reluctance and fierce competition raccoon rabies model is presented. For example, vaccination coverage of 60 percent in raccoons and *Canis latrans* was sufficient to eliminate the rabies epidemic, but with Africanized honeybees and with rabies in bats, the spread of the insect pest and consequently the raccoon rabies persisted even with 80 percent coverage. Most pathogens mutate, but in non-overlapping generation models sex-linked pathogens' fitness dynamics can be counter intuitive. Gonorrhea in zebras changes types too. Pre-existing immunity generally fails to regulate parasite-driven virulence evolution whereas vaccines can be paramount, which is timelier because antibiotic resistance is widespread. R0 estimates for Africanized honeybees indicate this strain of honeybee is highly resistant to Varroa, but it can operate near the critical vaccination threshold. Willow trees infected with *Melampsora* need only be covered as seedlings. Vaccines also shape shifts in the phylogeny of pathogens across the community according to the proportions of the populations mass vaccinated. Confirming previous predictions, it is shown that vaccine effectiveness matters less than the patterns of their distribution (*Bitsouni et al., 2019*).

8.2. Public Perception and Compliance

Questions about vaccine efficacy and human behavior often are in the news. Viral outbreaks and vaccination issues are occurring daily in the media worldwide. Science and scientists are involved to varying degrees. Such controversies are of great interest and have implications for development and the use of vaccines against publicly feared viruses or those concerning human health, livestock, and crops. The concern is orderly understanding of the public perception of vaccine "success" against the emergence of variant viruses with animal virus–transmitted vaccines. A search on social media in the last 60 days contained numerous examples of vaccines and underlying viruses of concern to the public (*C Layman et al., 2021*).

Questions about the estimate of how well vaccines block the emergence of viral variants were addressed, and the design of potential "best" future vaccines with declining public concern that directly impact public health was discussed. While there are many aspects to this story, the main primary concern is how the public perceives the success, service life, and application of vaccines they have already taken against emerging variant viruses. There is much evidence that no vaccine is 100% effective. Hence, it is a question of degrees or scales. If this rate is sufficiently high, such a vaccine application is considered a successful, protective, preventive, usable, and long-lasting vaccine (*O. Njoga et al., 2022*).

9.1. Adjuvant Technologies

Adjuvants are defined as agents incorporated into the vaccine formulation to amplify the immune response. They are considered as a strategic alternative approach to improve vaccine effectiveness in the elderly and are being studied across diverse diseases (*Jung et al., 2024*). Veterinary vaccines have been used for several years and play an important role in promoting animal health, welfare, food production, and public health. They are a cost-effective method to protect animals from disease, improve food production, and reduce the need for antibiotics in food and companion animals. Furthermore, vaccines are essential in preventing transmission of zoonotic disease to humans. Interestingly, there are unique challenges with respect to animal vaccines including the need for stability, low cost, ease of administration, etc., which require new solutions in vaccine development. The process for development and licensing of animal vaccines is much faster than that for human vaccines. As a result, many vaccine technologies and adjuvants are first tried on animals before being used in humans. Almost a century ago, Freund introduced a combination of mineral oils and bacterial cell components (Freund's complete adjuvant) for the improvement of vaccine-induced immune responses (*Garg et al., 2017*).

9.2. Novel Delivery Systems

Recent studies on exosome-derived vaccine formulations depict a great decline in under-vaccinated populations. However, the advantages do not guarantee an easy implementation for a robust vaccination scheme. The wide variability of functionalities is both another source for possible adverse side effects and a challenge for publicly regulated production systems. The fate of bioactive cargo, such as siRNA, could be key for an effective anti-viral activity but extremely difficult to thoroughly quantify (*Montaner-Tarbes et al., 2021*).

In spite of using exosomes as vaccine platforms, most of the vaccines where exosome-based vaccine platforms offered improved performance offer an end temperature probably too high for cold-chain free situations. Moreover, when having access to the working conditions like temperature, pH, or time between mixing and injection that enhance the immune response, the working conditions become a crucial aspect in vaccine implementation in public health vaccination schemes. These aspects need to be carefully studied and new methodologies developed before transferring the promising exosome-based vaccines from immunological perspectives to public health as new tools to ameliorate vaccination action against animal viruses with evolving resistance (*Zhang et al., 2024*).

10.1. Approval Processes

Regardless of the country or veterinary vaccine licensing region, the approval process for veterinary vaccines generally proceeds through the following steps. Information on the requirements for submitting applications and the approval process is available from both the USDA Center for Veterinary Biologics and the European Medicines Agency (*A Roth, 2011*). Open communication and continued collaboration between all stakeholders are paramount to achieving progress toward regulatory and manufacturer acceptance of alternative, non-animal methods. Continued efforts are needed to promote early and extensive discussions between manufacturers, OMCLs, academia, and regulators. Equally important is the need for international harmonization of regulatory requirements to help accelerate acceptance and remove the hurdle of different regions having different expectations. From a scientific and technical perspective, many opportunities are available thanks to new and evolving vaccine platform technologies, which make it possible to use in vitro testing entirely. Newer vaccines are better characterized, and robust GMP and consistency of production are now standard in the veterinary industry, allowing QC strategies to be developed from the start. The consensus among webinar and workshop participants was that progress toward implementation of 3Rs is still hindered by regulatory hurdles, including the diversity of regulatory requirements and lack of consistency of expectations from different agencies. Resources are another significant hurdle and further exchange and co-operation between manufacturers would be beneficial. Large-scale international validations of new methods demand sufficient resources from all parties (*Viviani et al., 2023*).

10.2. Ethical Implications of Vaccine Research

There are certain ethical concerns with vaccine research. To reduce unforeseen dangers to wildlife or domesticated animals, a variety of vaccine platforms, such as genetically engineered viral vectors or laboratory-bred intracellular parasites implicated in the reducing sensitivity to allergens, should be carefully investigated. When combined with fitness costs, vaccine efficacy can have contradictory impacts on virulence. Conflicts over vaccine evolution like these highlight a difficulty in creating vaccinations that work. The difficulty is in reducing vaccine evolvability through engineering or supporting immunization schedules while permitting enough evolution to increase efficacy (*C Layman et al., 2021*).

Several issues are concerned with prevention of spillovers and design of animal population vaccines against zoonotic viruses. Cryptic infections could allow spreading of wildlife vaccines yet minimize infection risks to domesticated species. These potential conflicts should be explored for zoonotic viruses associated with serious outbreaks or pandemic potential. Transmission of attenuated vaccines may establish subclinical infections, and thus weaker immunity and shorter durations of immunity than direct vaccination. When these vaccinations are employed in widely dispersable strains likely to mix with other strains, this weaker immunity might accelerate the spread of the wild-type pathogen. Even if immunity was sufficiently strong and long-lasting to keep the wild-type pathogen at bay, a newly introduced strain of wild-type pathogen might proliferate aggressively before becoming attenuated (*J. Bull et al., 2017*).

A sustained all-in vaccination event allows only small populations to remain unvaccinated. Would it be possible, for a specific vaccine, to ensure that individuals vaccinated existence? Would a safety mechanism that suppresses engineered traits only after dilution of vaccine individuals with wild-types strains be feasible? How to ensure that an engineered vaccine virus recedes as quickly as possible after all-in vaccination? It might be possible to engineer vaccines that be transmitted only once (or some finite but small number of times) but cannot transmit indefinitely (*Nuismer et al., 2019*).

11.1. Predictive Modeling of Virus Evolution

Viral pathogens are obligate intracellular parasites that depend on host cellular machinery to replicate. However, as cellular mechanisms evolved to hinder viral infections, different organisms highlighted their own immune systems capable of recognizing and destroying non-self-elements, as viruses emerge as authors of a “red queen” story between infection and immunity (*Marchi et al., 2019*). Host immune systems exert a constant selective pressure on evolving viral populations. The evolutionary dynamics of RNA viruses presenting a high mutation rate lead to a long-term evolution shaped by the fitness landscape of the host immune system. This landscape results from the fitness of various viral phenotypes against the immune antibodies produced by the host. Diverse population behaviors appear in time as the virus host dynamics proceed through a mutually profound modification of both components. These behaviors affect the prediction of emergence and reemergence of pandemics qualitatively and quantitatively. The most extensively studied long-range interactions appear in the context of the evolution of antigenic viruses. Such viruses are subject to selective pressure from the immune memory. This pressure arises from the production of long-lived, neutralizing antibodies. Binding to a newly emerged strain that diverged from the original valence one can raise a memory reaction. However, even though the new strain is not recognized as the immediate previous state with a different set of receptors, partial recognition is allowed to maintain continuity in the antigenic structure of different isolates (*C Layman et al., 2021*).

11.2. Global Surveillance Programs

Vaccination against viruses remains an effective and considerably economical option of protecting animals from vaccine-preventable diseases

and safeguarding food security. Despite such advantages, the emergence of resistant viral variants that can escape vaccine-induced immunity remains a formidable problem. Vaccinated populations of cattle, pigs, and poultry have been impacted by overwhelming outbreaks of viral diseases caused by viruses against which vaccines are in use. Such outbreaks have even been caused by circulating viral variants that emerge post-vaccination. Here, a theoretical framework is proposed to elucidate the potential dynamics of evolution of viruses against vaccination (*Pal Yadav et al., 2019*). The framework uses a simple scenario of a susceptible vaccineinduced immunity infected network structure comprising multi-strains of a virus, a type of vaccination, and randomly sampled population of host animals. Upon assuming reasonable number of approximations, mathematical equations governing major anticipated evolutionary outcomes of the network systems are derived and analyzed using analytical and numerical techniques. An example of the outcomes of a single strain vaccine applied to a multi-strain virus is discussed in details. A vaccination model, two model-systems, and corresponding analysis methods are analytically and numerically described. The results are expected to elucidate various facets of viral evolution against vaccination and help understand dimensions of the problem. In light of these results, it is interesting to scrutinize viral evolution in the context of vaccination in real and experimental systems. The veterinary vaccine and veterinary epidemiology communities can benefit immensely from developing combined attend of critical issues posed by the prospects/limits of vaccination-induced viral evolution (*Monette & J. Mouland, 2018*).

12. CONCLUSION

Evolution is an important strategy for the long-term success of any biological entity. However, in viruses, evolution is instinctively associated with the emergence of escape mutants that resist antiviral agents. Such mechanisms are well known in both the human immunodeficiency virus and in the influenza viruses responsible for seasonal and pandemic diseases in humans. There is increasing concern that a similar fate may befall other viruses, including veterinary pathogens. Vaccination is the best, most effective, and easiest means of control, but such control may be compromised if vaccines are unable to influence the evolution of the virulent strains (*J. King et al., 2022*).

In addition to experimental studies, vaccine failure is a very real concern for avian influenza vaccine use since field strains have emerged that are not neutralized by post-vaccination sera or fail to be restricted by class I major histocompatibility complex-mediated presentation. For the pestivirus border disease virus, this has led to the realization that excessive sequence variability is a serious impediment to vaccine development, particularly of recombinant vaccines. These problems, and the increasing understanding of how viruses adapt to cellular and species barriers, highlight areas where vaccines may need to evolve in parallel with the target viruses. In some cases, this may require systemic and altered strain vaccines (*Swayne et al., 2015*).

13. Conflict of Interest: Research does not conflict with any party.

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