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## **Preventing Vaccine Failure in Poultry Flocks**

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#### Abstract

Poultry sector is very useful for humans in terms of production of food items like meat and eggs. Pakistan has a developing poultry sector and is the second important sector after the textile industry. The poultry sector is encountered with many challenges; among them is the high incidence of disease outbreaks that result in colossal economic losses. The diseases of commercial and rural poultry include Newcastle disease (ND), infectious bursal disease (IBD), fowl pox, Marek's disease, infectious bronchitis (IB), avian influenza, hydropericardium syndrome, etc. The disease outbreaks have also occurred in vaccinated flocks. Better understanding of the causes of vaccine failure will result in identifying prophylactic measures regarding disease outbreaks in poultry flocks. This chapter overviews the common causes of vaccine failure and further highlights the procedures for successful immunization.

Keywords: immunization, vaccine failure, poultry

## 1. Introduction

Poultry sector is the source of animal proteins in the form of meat and eggs. The total strength poultry in Pakistan is 1210 million. The poultry sector provides 1,391,000 tons of meat and 18,037 million eggs annually. The contribution of poultry sector in agriculture and livestock sectors is 7.5 and 12.7%, respectively, while its contribution in total GDP is 1.4. The annual growth rate of poultry sector is 5–10% in the country. The poultry meat contributes 32.7% of total meat production in the country [1]. The demand of poultry meat has increased over the years due to the increasing demand of quality food in the form of meat and eggs.

Poultry birds being living creatures are prone to infections. Diseases are a cause of high economic losses to poultry farmers [2]. In developing countries, poultry diseases are a cause

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of very large economic losses to poultry industry [3, 4]. Among bacterial, viral, parasitic and fungal diseases, the outbreak of viral diseases can cause havoc to the poultry industry causing reduced meat and egg production. The important viral diseases of poultry include Newcastle disease (ND), avian influenza, infectious bursal disease (IBD), infectious bronchitis (IB), etc. The high prevalence of diseases creates major constraints in the development of poultry sector. Immunization is the use of a biological preparation in the form of vaccine for enhanced immunity and prophylactic measures against specific diseases [5]. The process of injecting the vaccine in the body is known as vaccination. Proper vaccination can prevent losses due to diseases in poultry flocks [6]. Mostly, the vaccines are carried out against viral diseases but vaccines against salmonella, mycoplasma and coryza infections are also available. The vaccines against parasitic infections like coccidiosis are also being tested in different countries.

Vaccination is one of the most important tools for preventing diseases and in reducing the economic losses of the poultry producers [2]. Vaccination comprises the use of attenuated, killed, or recombinant organisms for stimulation of the body's immune response that recognizes the injected organism as a foreign antigen, resulting in clearing the antigen and developing memory cells in the body. Vaccination is the cheapest, reliable, effective, economical, affordable and suitable alternate for prevention of diseases in poultry flocks [5].

Live vaccines comprise a virulent virus whose pathogenicity has been weakened through consecutive cultures in living cells but the virus maintains its immunogenic antigenicity for stimulating the body's immune response; this whole process is commonly known as attenuation [7]. Commonly used live vaccines against diseases of poultry are Newcastle disease, infectious bronchitis, infectious bursal disease, etc. [8].

Killed vaccines comprise viruses whose pathogenicity has been inactivated through the use of physical and chemical means, but the protein coat structure has been maintained, which acts immunogenic. The viruses are physically inactivated by the use of ultraviolet radiations and heat and through chemical means by the use of formalin [9]. Killed vaccines against Newcastle disease and Avian Influenza are being used and have an advantage of providing long term immunity to flocks.

Vaccine failure is the consequence of the inability of the chicken to develop adequate immunity after immunization or susceptibility of bird to field outbreak after administration of vaccine [3]. High rates of 53.5% of vaccination failures have been recorded in vaccinated poultry flocks. Rates of 25.6, 25.6 and 2.3% of vaccine failure in Newcastle disease, infectious bursal disease (Gumboro) and fowl pox, respectively, have been recorded [2]. The common breaches in transportation, handling, storage and administration of vaccines are responsible for high rates of vaccine failure in poultry flocks in developing countries [2].

## 2. Causes of vaccine failure

The causes of vaccine failure can be categorized into two major factors: antigen factor and host response.

#### 2.1. Antigen factors

The protective vaccine antigen is of prime importance in the production of effective vaccine. The vaccines available in the market may have the following shortcomings resulting in vaccine failure.

#### 2.1.1. Improper formulation of vaccine

The vaccines are manufactured in a processing plant where the titer of antigen of specific virus or bacteria may not be maintained properly; as a result, the inoculums may not initiate protective immune response in birds. The titer of antigen in the vial of vaccine may be low which results in low immunity level in birds. The dose–response relationship among the virus content, serological response and clinical protection has been reported [10]. Virus concentration has a significant effect on immunogenicity of vaccines [11]. The inadequate procedure of formulation of vaccine and lack of standard procedures of vaccine formulation result in the production of nonpotent vaccine.

#### 2.1.2. Nonusage of local antigens

Some of the viral diseases of poultry like infectious bursal disease and salmonella have many serotypes. Some of the serotypes are prevalent in one area, while others are prevalent in other areas. The local disease causing agents in any area are of prime importance for vaccine manufacturing. The strains of viruses differ from area to area. The local serotypes and locally isolated antigens are considered the most suitable immunogens for formulating vaccines. The nonusage of local vaccine antigens may result in disease outbreaks [2]. The foreign vaccine may be made from serotypes that are different from field strain [12]. Moreover, vaccination with foreign vaccine may not provide immunity to birds if the field strain is of higher virulence and of a different nature [13].

#### 2.1.3. Improper storage temperature

After the formulation of the vaccine, its storage is of utmost importance. The freeze-dried vaccines require freezing temperatures, while lyophilized vaccines may be stored at 4°C, and during transportation the low temperature might not be properly maintained. The Marek's disease vaccine is stored in liquid nitrogen at very low temperatures, while live vaccines of ND, IBD, IB, etc. are stored at 4–8°C. The oil-based vaccines may be stored below 8°C. In the poultry sector, almost all the vaccines available are thermolabile in nature. The maintenance of proper cold chains and storage temperature is a prerequisite for optimal potency of vaccines. The shortage of electricity, weak, nonfunctional, obsolete and repaired storage equipment, high temperature during transport, refrigerators without thermometers, etc. are the common problems of vaccine storage of developing countries like Malaysia, India, Tanzania and Pakistan [14–18]. Data have been recorded about use of vaccines after purchase from the market in Nigeria, and it has been found that 16% of farmers do not perform vaccination on the date of purchase of vaccine and 7% of farmers store the vaccine on the shelf without proper preservation, thus resulting in vaccine failure [2].

#### 2.1.4. Exposure to direct sunlight

It has been documented that vaccines are transported like ordinary drugs [2]. Direct sunlight has UV radiations which are lethal for live viruses. The exposure of vaccine to direct sunlight results in the killing of antigens present in the vial, and as a result, the number of viral antigens is reduced in the vaccine and the vaccine may become ineffective.

#### 2.1.5. Use of expired vaccines

The potency of vaccines is maintained to a certain period of time, provided that the transportation and storage temperature is properly maintained. The use of vaccines after the date of expiry may not result in optimal immune response and can also result in vaccine failure.

#### 2.1.6. Mutation of viruses

Some of the viruses like the influenza virus are of a mutating nature and as a result pose a serious threat regarding the effectiveness of vaccine against certain diseases.

#### 2.2. Host factors

The poultry birds to be vaccinated against diseases may not respond effectively against vaccines due to the following shortcomings, thus resulting in vaccine failure.

#### 2.2.1. Stress on birds

Stress is a condition of vulnerable homeostasis and is affected by management and environment factors. Birds normally have limited resources in the body for growth, response to environment changes and maintain a defense system for diseases. The stress on birds can be due to a number of factors including cold stress, heat stress, high humidity, transportation stress, intensive farming, high stocking density, overcrowding, low per bird space, decreased ventilation, poor litter conditions, accumulation of bad smell in sheds and poultry houses, off feeding, water deprivation, poor management, bad sanitary conditions, very wet or extremely dry litter, dusty environment, parasitism, nutritional deficiency, fever, and so on. In these cases, there can also be vaccine failure in livestock. The poultry birds are sensitive to both cold and warm weather [3]. Heat stress is an important factor of economic loss for the producer [19], while cold stress modifies the immune response of broilers [20]. The symptoms of stress in birds include panting, increased thirst, reduced appetite, reduced egg production, decreased weight gain, small sized eggs, thin egg shells, reduced growth, prostration, etc. All the factors including management conditions, substandard hygienic conditions, etc. contribute to the possible causes of high economic losses by leading to vaccine failure [21].

#### 2.2.2. Concurrent disease

It is highly important that the vaccination should be done in healthy birds. The vaccination in sick and diseased birds may not provide fruitful results; rather, vaccine reaction may occur leading to extra stress and an increased morbidity and mortality rate. Moreover, any other

disease condition may also contribute to vaccine failure. When the birds are morbid due to the same disease for which vaccination had been done, then there will also be vaccine failure because the antibodies produced against the pathogenic agent will neutralize the antigen of vaccine and a reaction may take place in the body of birds and vaccination may worsen the condition of disease.

#### 2.2.3. Immunosuppressive diseases

Certain diseases are immune-suppressive in poultry flocks like mycotoxicosis, infectious bursal diseases (Gumboro), chicken infectious anemia, Marek's disease, etc. These immune suppressive diseases may also lead to vaccine failure. The fungal toxins present in poultry feed have a bad effect on the feed conversion, growth, health and immune status. The fungal toxins cause the following effects: carcinogenic, allergic, hypersensitivity and depression. The common age of infection of infectious bursal disease (Gumboro) in poultry flock is at 3rd to 7th week of age. The bursa is a lymphoid organ in poultry where maturation of B cells takes place in poultry. The infection of IBD during this stage of age may lead to permanent damage to bursa; as a result, the maturation of B Cells may not take place in birds throughout their life span and thus the birds remain prone to vaccine failure during the rest of their lives.

#### 2.2.4. Immaturity of birds

The receptors for some antigens develop in the body with advancing age. Some of receptors of virus develop as early as with the hatch of a chick. The receptors of diseases like Newcastle disease, infectious bronchitis, etc. develop at a very early age while the receptors of diseases like infectious bursa disease, fowl pox, etc. develop late in the body. Vaccination at a very early age before the development of certain receptors may also result in vaccine failure. The age of the bird is very important at the time of vaccination.

#### 2.2.5. Interaction with maternal antibodies

The antibodies of certain viral diseases are transmitted through eggs. As the breeder/parent flocks of poultry are routinely vaccinated against viral diseases which are prevalent in the area, the newly hatched chicks have maternal antibodies in their blood and these can interact with vaccine antigens. The antibodies against ND virus and IBD virus are transmitted in eggs and provide protection to the newly hatched chicks during the first week of birth. High maternal antibodies interfere with multiplication of live vaccines and reduce the level of immunity production in the chicks. The use of live vaccines during the first week of birth in chicks against diseases whose maternal antibodies still persist in the body of the chick will result in neutralizing of antigen and active immunity may not be provided by the vaccine [22].

#### 2.2.6. Improper route of administration

The vaccines have specific routes for their administration in the body of the bird, that is, through oral, subcutaneous (S/C), intramuscular (I/M), wing web (W/W), drinking water (D/W), eye dropping (E/D), spray, etc. Not following f specific recommended routes of

vaccination may result in vaccine failure in poultry flocks. The fault of administering the vaccine also results in vaccine failure [3, 4].

#### 2.2.7. Inadequate dosage

If the optimum dose of vaccine had not been injected in the bird, then there is also failure of vaccine. Overdosage may lead to reaction, and underdosage can lead to vaccine failure. There are certain factors which cause reduction in optimal vaccine dosage, that is, use of chlorinated water for vaccination, use of water having antimicrobial contents, etc. Moreover, in the case of injecting vaccine to more number of birds than recommended by the company or manufacturer, the low dose will be available to the whole the flock and thus may be prone to low vaccine titers and vaccine failure.

#### 2.2.8. Lack of booster dose

Some of the vaccines require a booster dose for successful immunization. The booster dose is required after 10–20 days of the initial dose. The initial dose is required for priming of vaccine while the booster is required for maximum protection against antigen. The lack of booster dose results in low antibody titers, resulting in vaccine failure.

#### 2.2.9. Wrong timing of vaccination

Mostly the vaccines should be done early in the morning or later if it is during summer. The birds feel comfortable during cold hours of the day. As a result, a good response is obtained after vaccination. Otherwise, the chances of vaccine failures are increased in the case of vaccinating birds during the hot hours of the day.

#### 2.2.10. Climatic factors

The climate variation is a change in climatic parameters (temperature, rainfall, humidity and soil moisture) [23]. Climate change affects both living and nonliving creatures, thus contributing to the health of poultry flocks and may lead to vaccine failure and disease outbreaks.

### 3. Preventing vaccine failure

The following procedures can prevent vaccine failure in livestock and poultry flocks.

#### 3.1. Vaccine factors

#### 3.1.1. Proper formulation of vaccine

The vaccines must be properly formulated. The dosage of vaccinal antigen and properly processed vaccines provide good results and prevent vaccine failure. The record of all batches of vaccines and their standard tests of vaccine potency may be maintained. Moreover, the titer of antigen should be optimal so that the proper immunity level may be provided by the vaccine.

#### 3.1.2. Use of local strains of viruses

For maximum immune protection, the local strains of antigens must be used for manufacturing of vaccine. The local disease causing agents of any area are specific targeted pathogens and antigens from local disease outbreaks and provide maximum protection against local disease causing organisms.

#### 3.1.3. Adequate procedure of vaccine formulation

The viruses used for vaccine production are harvested in live cells like chicken embryo. The bacteria used for vaccine production are culture in growth media like nutrient agar, etc. Similarly, the procedures for live attenuated, killed inactive vaccines, subunit vaccines differ from antigen to antigen. The adequate procedure for vaccine formulation will result in a maximum immune response from the antigen and hence a successful immune response.

#### 3.1.4. Proper storage and cold chain temperature

Vaccines are to be manufactured in a plant and then after stored and transported to remote areas. Temperature has direct effect on the efficacy of vaccine [24]. The vaccines lose their potency with the passage of time; hence, they require proper cold temperatures to remain stable and viable for long periods of time. The proper storage and cold chain temperature of vaccine is of utmost importance; the vaccines must be stored below 4°C. The storage of food items, chemotherapeutic agents, specimens for pathological examinations, tissue samples for laboratory findings along with vaccine should be avoided [25]. During transportation, the maintenance of cold chain is a challenge for developing countries. A number of factors create hurdles in maintaining cold chain systems including loss of electric power, substandard refrigeration system, overchilling, etc. Moreover, the extra chilling of oil-based vaccines results in crystal formation of adjuvant material of vaccine like aluminum salts, etc. resulting in reduced potency of vaccines. The thermostable vaccines can be stored at 2-8°C and has more significance where cold chain temperature is not maintained and is less expensive [24]. Thermostable vaccines have some resistance to cold and hot environments, while freeze-dried vaccines should be preserved and stored at low temperatures in the refrigerator at 4°C and even during the transport of vaccine the cooling/ice blocks should be used to maintain low temperatures during transportation of vaccine. Freezing and thawing must be avoided. The vaccines must only be brought out of the refrigerator/freezer at the time of use at the farm. The live vaccines in poultry flocks must be used within 2 h of its reconstitution. Once they have been reconstituted, they drop their potency rapidly. The reconstituted vaccines should be used as early as possible and unused vaccines may be stored in the refrigerator for a maximum of 6 h; after that period the vaccines should be discarded.

The use of thermostable vaccines can be an alternative to overcome the difficulties related to cold chain and storage temperature [26]. The thermostable vaccines can maintain their

potency and vaccinal activity for 1 year at 2–8°C and for 3 months up to 28°C in dried form [27]. Routes including intraocular, intranasal, paternal (injection) and oral (drinking water and feed) can be used for administration of thermostable vaccines [28, 29].

#### 3.1.5. Avoiding exposure to direct sunlight

Direct exposure of sunlight results in the killing of antigen present in the vial; as a result, the titer of vaccine antigens are reduced in the vaccine and it may become ineffective. During formulation of solution for oral or parental vaccines, direct exposure to sunlight should be avoided and for oral vaccines the cap of the vaccine vial should be opened inside water. The vaccines should be mixed in drinking water in a room or in a shady place; moreover, during the transportation of vaccine, black or colored bags and cartons should be used to prevent sunlight affecting the vaccine.

#### 3.1.6. Avoiding use of expired vaccines

The date of expiry mentioned on the vial of vaccine should be checked before opening the vaccine vial. The expired vaccines should be discarded or returned to the manufacturers. At places where vaccines are frequently used, it should be a practice to purchase a fresh stock of vaccines. Some oil-based vaccines have a very narrow range of shelf life of 3–6 months. While some lyophilized live vaccines have longer shelf life of 1–2 years, provided the vaccines are stored at proper temperatures. The use of expired vaccines should be avoided [25].

#### 3.1.7. Use of adjuvant

Adjuvants are substances that are added in the vaccine to increase the bioavailability of vaccine. In the poultry sector, the oral route of vaccination is followed for most of the vaccines. The oral route of vaccine delivery is difficult due to barriers in the gastrointestinal tract. In order to overcome this challenge, the antigen must be protected from such an environment and the immune response must be activated. This challenge can be overcome by the use of adjutants. Adjuvants improve the safety of the vaccine and have a potential effect on inducing mucosal immune response [30]. The use of adjuvants provides good results for live vaccines. The adjuvants enhance the availability of vaccine and act as a sticking agent for vaccine and the mucous membranes of the body.

#### 3.1.8. Use of stabilizers

Stabilizers are substances which are added in a vaccine to increase the shelf life of the vaccine. The stabilizers like Vac-Safe (Intervet), Vital Blue, etc. can be used for oral live vaccines like ND, IBD, IB, etc. of poultry. The skimmed milk at the rate of 2 g/L can also be added as a suitable alternative stabilizer [31].

#### 3.1.9. Manufacturer guidelines

The company's manufacturing guidelines provide valuable information regarding vaccine efficacy, usage, storage and route. The guidelines are: (1) open the vaccine vial in water and (2) use one complete vial after opening it. The vaccines must be utilized as early as possible

after its reconstitution in diluents, etc. Once the vaccine is reconstituted, the time limit is set. Vaccines must be used within 2 h of their reconstitution during winter and within 1 h of their reconstitution during summer. IB vaccines lose potency after 1 h of their reconstitution, while pox vaccines lose 50% of their potency after 1 h of their reconstitution [32].

#### 3.1.10. Use of immune boosters

There are many substances that have been used in poultry for immune stimulation. Some of them are vitamin E, selenium and levamisole [33, 34]. The selenium supplementation has effect on enhancing humoral immune response in chicks [35, 36]. The selenium supplementation increased natural resistance of increasing response of organisms to antigenic stimuli [37, 38]. The increased humoral antibody titers are observed when selenium is used in feed [39].

#### 3.1.11. Booster dose

Some of the vaccines require a booster dose for successful immunization [2]. The booster dose is required after 10–20 days of the initial dose. The initial dose is required for priming of vaccine, while the booster is required for maximum protection against antigen. The lack of booster dose results in low antibody titers. As a result, vaccine failure may result. It has been documented that the priming with live attenuated vaccine followed by booster of killed vaccine and second booster with live vaccine provides best protection against Newcastle disease [40]. However, subsequent inoculums are also required at regular intervals.

#### 3.2. Host factors

An effective vaccine response may be obtained if the bird is healthy. The following recommendations/guidelines can overcome the shortcomings regarding prevention of vaccine failure.

#### 3.2.1. Stress-free birds

All types of stresses mentioned earlier should be avoided before administration of vaccines to poultry birds. The temperature of the environment and sheds should be normal before vaccination. Moreover, the birds should be in a good physical condition before administration of vaccines. Stress suppresses the chicken's immune response, and during these conditions of stress, birds should not be vaccinated [41]. The stress on birds can be minimized by the use of vitamins and minerals in drinking water before, during and after vaccination [13].

#### 3.2.2. Deworming before vaccination

The adult birds may be dewormed before vaccination at least 15 days before injection of vaccine; moreover, diseased birds should be treated properly and be given vaccines after recovery. Only healthy flocks should be vaccinated.

#### 3.2.3. Monitoring of subclinical infections

Some of the diseases in poultry have subclinical infections, like coccidiosis. The birds apparently seem health, but subclinical infections persist in birds over long periods of time, which have previously been infected with coccidiosis infections. On the day of vaccination, the birds should be closely monitored. The apparent health of flock should be analyzed. Moreover, the color and consistency of fecal droppings, abnormal sounds from birds, respiratory distress, etc. should be evaluated. After being satisfied with the proper health status of the bids, the concerned staff may be allowed to vaccinate the flock.

#### 3.2.4. Balanced feed

Nutrition plays a significant role in the development and function of the immune system [42]. The commercial feed offered to poultry should be analyzed regularly and the level of toxins be checked on a regular basis. Especially in summer and humid environment conditions, the fungus grows on feed ingredients and fungal metabolites gain entry into the body of poultry, and as a result, they cause immune-suppression, decreased growth, hypersensitivity and decreased feed intake.

#### 3.2.5. Maturity of bird

In poultry birds, age is considered for the vaccination of bird; receptors for different pathogens develop in the body of poultry bird at specific ages, so the vaccination is done keeping in view the age of the bird, that is, ND + IB vaccine is done on the first day of birth. Similarly, the Marek's disease vaccine is done immediately after hatching of chicks in the hatchery machine. The IBD (infectious bursal disease/Gumboro) vaccine is done at 10–12 days of birth and booster is given after 10 days. In broiler birds, the hydropericardium syndrome (HPS)/ Angara vaccine is done at 21–23 days of birth. So, the age of the bird is very important for vaccination. The domestic/rural poultry requires injection of ND after every 2–2.5 months.

#### 3.2.6. Consideration of maternal antibodies

As the breeder/parent flocks of poultry are routinely vaccinated against viral diseases which are prevalent in the area, the newly hatched chicks have maternal antibodies in their blood. It is suggested that the bird should be a minimum 11 days old at the time of administration of IBD vaccine and 7 days old at the time of administration of ND vaccine [31].

#### 3.2.7. Proper vaccination schedule

In Pakistan, the outbreak of diseases like infectious bronchitis and avian influenza occurs in birds during winter; for that purpose, the birds must be vaccinated prior to winter, so that proper antibody titer may be reached in birds before exposure to the disease causing virus or bacteria in birds. To avoid any economic loss, a record may be maintained and a strict vaccination schedule according to disease prevalence in the area must be followed in poultry flocks [43].

#### 3.2.8. Preparation of flocks for vaccination

The poultry flocks are properly prepared for administration of oral vaccines. The birds are offered feed and are kept off water for 2 h before administration of vaccines. The drinkers

are properly washed and the vaccine is given to birds. The number of drinkers is increased in order to ensure that all the birds drink vaccine water. The water should be provided to birds in such a way that birds drink all the vaccine water within 2 h. The birds are regularly moved during this process so that all the birds drink water containing the vaccine virus. The stabilizers and coloring agents can be added in the vaccine. A tinge of the coloring agent can also be noticed on the beak of birds which indicates the drinking of water.

#### 3.2.9. Host resistance

Certain genes are discovered which have genetic resistance against viral diseases of poultry [44]. The breeding for disease resistance may provide good long-term solutions for disease control [45]. However, the emergence of new genetic groups and mutations require new vaccine practices for successful immunization [46].

#### 3.2.10. Vitamin and mineral supplementation

Vitamin and mineral supplements help to develop immune response by acting on the immune cell or by changing metabolic or endocrine functions [47]; as a result, the antibodies are produced in the body at a faster rate and a protective level of antibodies is gained in a shorter time. Vitamin E and selenium have a role in modulating the immune response and have shown good results in preventing vaccine failure. Research conducted shows that vitamin E may enhance immune response to antigens in cockerels but excessive vitamin E may depress specific immune responses [48]. Administration of excess vitamins, amino acids, minerals and their combinations enhance the disease resistance by stimulating humoral and cellular immunity and phagocytosis [49]. Optimal vitamin nutrition is required for optimal immune response and disease resistance. The addition of higher levels of vitamin A, C, E and Selenium ensures better immune response of birds to vaccination and reduces the chances of vaccination failure in broiler poultry flocks [50]. Studies have suggested that the nutrient levels that are adequate for growth and feed efficiency may not be adequate for normal immunity for maximizing the resistance to disease [51, 52].

#### 3.2.11. Continuous surveillance

The regular and continuous surveillance of prevalent diseases should be conducted in order to collect data on the disease pattern. For this purpose, the blood/serum and fecal samples may be collected and sent to the laboratory for diagnosis of disease. Moreover, the tracheal and cloacal swabs can also be sent to the laboratory for isolation of pathogens. The antibodies titer against injected vaccines may be got routinely checked for maintaining optimum titer of antibody against the disease.

#### 3.2.12. General precautions

Considerations regarding the use of live and killed vaccines should also be kept in mind during vaccination. The live vaccines may cause vaccine reactions and injection of killed vaccines may cause local tissue reactions. Therefore, only an expert professional or qualified veterinary assistant should be allowed to vaccinate the poultry birds. In general, vaccination should be done during early hours of the day or late hours after noon during summer. Vaccination during hot hours of the day may not give good results. Moreover, after transportation of birds, the birds should be given proper rest before vaccination [2].

#### 4. Conclusion

Vaccination may be considered as insurance against diseases. A successful vaccination program is dependent on many factors including vaccine handling, quality and nature of vaccine, use of local antigens, immunogenic response inside the body of the bird and following manufacturers' instructions. Although the disease outbreaks against specific diseases in nonvaccinated flocks cause very high economic losses, the severity of disease outbreaks in properly vaccinated flocks is low. The potential threat of disease outbreaks even in vaccinated flocks cannot be avoided 100%, but the losses can be minimized through thoughtful consideration of success of vaccination program for poultry flocks. In Pakistan, there is a high incidence and prevalence of contagious diseases of poultry and vaccination is the only tool to prevent birds from diseases. Through preventing vaccine failure, the productivity of food items like meat and eggs can be increased in the country, and shortage of animal protein can be overcome and thus per capita availability of eggs and meat can be increased. Moreover, the poultry sector can play a better role in the economy of the country by decreasing economic losses due to vaccine failure, thus increasing annual share in GDP value and becoming a major contributor of the agricultural sector of the country.

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