

## Chicken Eggs - A Source of Antibodies Alternative to Mammal

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Received June 22, 2018; Accepted July 28, 2018; Published November 30, 2018

### ABSTRACT

Antibodies presently available for research, diagnostic and therapies are mostly mammalian monoclonal or polyclonal antibodies. Traditionally, bigger animals such as horses, sheep, pigs, rabbits and chickens were used for the production of polyclonal antibodies, Chicken eggs present an ideal alternative antibody source to mammals, as the IgY in the chicken's blood is transported to the egg and accumulates in the egg yolk in large quantities. The yolks of eggs laid by immunized chicken have been recognized as an excellent source of polyclonal antibodies for over a decade. This simple noninvasive approach presents an appealing alternative to conventional polyclonal antibody production methods. This review offers summarized information about edible IgY and the use of the antibodies for aquaculture.

**Keywords:** Antibody, Immunization, Chicken eggs, Edible IgY

### INTRODUCTION

Aquaculture is an emerging industrial sector which requires continued research with scientific and technical developments and innovation. The aquatic environment contains plethora of obligate and opportunistic bacterial pathogens as well as beneficial and neutral bacterial strains. Bacterial infections are considered to be a major cause of mortality in fish hatcheries and bacteria most frequently associated with disease in farmed fish and shellfish are species.

Intensive use of antimicrobial agents in aquaculture provides a selective pressure creating reservoirs of drug-resistant bacteria and transferable resistance genes in fish pathogens and other bacteria in the aquatic environment [1]. From these reservoirs, resistance genes may disseminate by horizontal gene transfer and reach human pathogens, or drug-resistant pathogens from the aquatic environment may reach humans directly [2]. Horizontal gene transfer may occur in the aquaculture environment. Furthermore, fish farmers must confirm that fish are kept in the best state of health and welfare. The invention of novel drugs or the use of alternatives to antibiotics should also be encouraged.

Vaccination is commonly used to prevent disease outbreaks and is effective, but this kind of method does not seem to have effect to crustaceans because crustaceans lack a true adaptive immune response [3]. However, passive immunization using pathogen-specific antibodies raised in hens is a potential method against diseases, which especially

make crustaceans lacking a true adaptive immune response obtain specific antibody. Chicken eggs present an ideal alternative antibody source to mammals, as the IgY in the chicken's blood is transported to the egg and accumulates in the egg yolk in large quantities. The efficiency of egg yolk immunoglobulin (IgY) has been assessed for therapeutic application by passive immunization therapy through oral ingestion of IgY.

### IMPORTANCE OF IgY

During the past 20 years, the use of chickens instead of mammals for antibody production has increased. A major advantage of using birds is that the antibodies can be harvested from the egg yolk instead of serum, thus making blood sampling obsolete. In addition, the antibody productivity of an egg-laying hen is much greater than that of a similar sized mammal [4]. Purification of immunoglobulin from mammalian blood is time-consuming

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**Citation:** Kumaran T & Citarasu T. (2018) Screening of BCG Vaccine Efficacy among Healthy Vaccinated Adults in Khartoum, Sudan. *Adv Vaccines Vaccin Res*, 1(1): 16-19.

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and expensive. Today, hens are recognized as a convenient and inexpensive source of antibodies. It has been reported that the amount of immunoglobulin that can be yielded from one egg of an immunized hen is as much as that can be obtained from 300 ml of rabbit blood. The production of antigen-specific antibodies in egg yolk also has significant implications for nutraceutical and functional food development. However, the practical use of IgY in research and diagnostics is limited due to complex and time-consuming purification steps associated with the further purification of IgY [5].

## **IgY PRODUCTION METHODS**

### **Immunization**

Specific IgY development and production can be achieved by immunizing laying hens with the target antigen. However, the resulting immune response of the immunized hens cannot be very predictable. Mainly five factors influence this response: the antigen (dose and molecular weight), the type of adjuvant used, the route of application, the immunization frequency, and the interval between immunizations [6].

### **Antigen**

The immune response is triggered by contact of the organism with antigen, which is a structure that is recognized by the immune system as foreign ("non-self"). The dose of antigen influences significantly the immune response and the antibody titre that is evoked. Too much or too little antigen may induce suppression, sensitization, tolerance or other unwanted immunomodulation found that the injection of antigen concentrations ranging between 10 µg and 1 mg elicited good antibodies responses and this was also reported by other researchers [7].

### **Adjuvant**

The induction of high and sustainable egg yolk antibody titre reclaims the use of adjuvant. There are more than 100 known adjuvants, which differ in their chemical characteristics, their efficacy in stimulating the immune system, and their secondary side-effects. In mammals, the use of Freund's complete adjuvant leads systematically to severe inflammation at the injection site. In birds, the use of FCA does not seem to result in the same severe lesions as in mammals. The results of Gassmann et al. [8] suggest that chickens show higher resistance to tissue damaging potency of FCA than rabbits.

### **Route of application**

The most common route for antigen injection in hens for IgY production is the intramuscular route. Injection is usually performed in the breast muscle. Chicken can also be injected subcutaneously in the neck. With very young animals, it may be preferable to inject intramuscularly into the breast muscle, because subcutaneous injection is more

difficult to perform and can therefore cause more distress [6].

### **Immunization frequency**

The total number of immunizations required depends on the type and dose of the antigen as well as the adjuvant employed. At least two immunizations have to be given. Yolk antibody titres should be checked 14 days after the last immunization. The success of an immunization protocol depends also on the interval between the first and second and subsequent immunizations. Often reported interval is two to four weeks [9].

### **MERITS OF IgY**

IgY technology more popular and to convince the scientific community of its significant advantages. Chickens have the potential to be used to complete the spectrum of animals used for Ab production. The yolks of eggs laid by immunized chicken have been recognized as an excellent source of polyclonal antibodies for over a decade. IgY technology is a highly innovative and expanding branch of biotechnology which offers many advantages: it is produced by a non-invasive method which does not cause pain to animals or lead to their death, since it is based on the simple act of collecting eggs. Hens are inexpensive to keep than rabbits and, furthermore, the IgY production of a hen corresponds to that of a large mammal [6]. IgY Abs are used successfully in immunohistochemistry for detection of antigens of viral, bacterial, plant and animal origin, and also to assess the incidence of intestinal parasites in domestic animals and the contamination of foods with toxins or drugs Pichler et al. [10], 1998. Since lot of benefits of IgY technology and its universal application in both research and medicine, it is expected that IgY will play an increasing role in research.

### **INFLUENCE OF IgY ON GASTROINTESTINAL TRACT**

IgY can be used long term therapeutic agent without any negative effects on animal health. Because IgY are resistant to the gastric barrier, these are of particular interest for passive immunotherapy of gastrointestinal tract infections. Added to these, eggs being normal dietary element with no religious taboo issues so pose practically no risk of toxic side effects of IgY antibodies [11]. Most gastro intestinally absorbed proteins and peptides are believed to be hydrolyzed at the brush border or by cytoplasmic enzymes [12]. However, various bioactive proteins and peptides retain physiological activity when administrated orally. Wieddemann et al. [13] postulated that IgY performed well in the intestinal mucosa, because it could be recovered from the intestine after oral administration which showed good resistance to enzymatic activity and gastric acidic pH.

## LIST OF IgY DEVELOPED AGAINST MICROBIAL PATHOGENS IN AQUACULTURE

**Table 1** show the list of IgY antibodies which are created against the pathogenic micro-organisms in aquaculture.

**Table 1.** List of IgY against various pathogens.

S. No.	Target microbes	Aquatic Species	References
1	<i>Escherichia coli</i> , Rotaviruses, Coronavirus	Calves and piglets	Ebina [14]
2	<i>Vibrio anguillarum</i>	Rainbow trout	Akhlaghi [15]
3	<i>Yersinia ruckeri</i>	Rainbow trout	Lee et al. [16]
4	<i>Pseudomonas aeruginosa</i>	Humans	Carlander and Larsson [17]
5	<i>Edwardsiella tarda</i>	Japanese eels	Mine and Kovacs-Nolan [18]
6	<i>Salmonella enteritidis</i> and <i>Salmonella typhimurium</i>	Humans and chickens	Lee et al. [19]
7	<i>Helicobacter pylori</i>		Shin et al. [20]
8	White Spot Syndrome Virus	<i>Penaeus chinensis</i>	Kim et al. [21]
9	White Spot Syndrome Virus	<i>Penaeus monodon</i>	Kumaran et al. [22]
10	<i>Escherichia coli</i>	Dairy cows	Meenatchisundaram et al. [23]
11	<i>V. cholera</i>	Humans	Barati et al. [24]
12	<i>V. harveyi</i>	<i>Penaeus indicus</i>	Kumaran et al. [25]

## CONCLUSION

Today, there is no doubt that edible chicken IgY can be produced and used, with minor modifications, in similar ways to the use of mammalian Abs. It is to be expected that studies on the therapeutic or prophylactic use of IgY Abs will be intensified in future. In particular, because of the increasing resistance of microorganisms to antibiotics, research on all aspects related to the development of specific IgY against pathogenic microorganisms will have to be intensified. IgYs can be used in aquaculture, veterinary medicine and in human medicine. IgY technology is a fast developing field and we have tried to cover most of its aspects in this review. We are convinced that, once accepted and widely used, the technology will offer alternatives and solutions to science, to medicine and to society as a whole.

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