Production of IgY Antibodies from Egg Yolk of Immunized Chickens

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Abstract
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. This simple non invasive approach presents an appealing alternative to conventional polyclonal antibody production methods. This mini review offers production of edible antibodies used for passive immunization.

Keywords: Immunoglobulin Y; Antibody; Chicken eggs; Immunization

Introduction
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 [1]. Purification of immunoglobulin from mammalian blood is time-consuming 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.

Antibodies
Antibodies are produced by the immune system of an animal in a specific response to a challenge by an immunogen. Immunogens (antigens) are molecules which can induce a specific immune response and are usually foreign proteins or carbohydrates or sometimes lipids and nucleic acids. Antibodies are secreted from plasma cells which have differentiated from B lymphocytes after appropriate stimulation by the foreign immunogen. Chicken egg yolk antibody (IgY) has received much attention in recent years because it can be easily prepared in high concentration and is both affordable and safe [2]. IgY is successfully used in medical immune testing, diagnosis, hetero grafts and therapy. The use of chicken IgY in a double antibody sandwich ELISA for detecting African horse sickness virus by Du-Plessis et al. [3]. New vaccine technology has led to vaccines containing highly purified antigens with improved tolerability and safety profiles, but the immune response they induce is suboptimal without the help of adjuvants. Gottstein and Hemmeler [4] reported Chickens store high contents of IgY in the yolk and are considered to be efficient antibody producers.

IgY production
Immunization of the hens: 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 interval between immunizations and the frequency of immunization [5].

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 titer that is evoked. Too much or too little antigen may induce suppression, sensitization, tolerance or other unwanted immune modulation. Found that the injection of antigen concentrations ranging between 10μg and 1mg elicited good antibodies responses and this was also reported by other researchers [6].

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 [5].

Immunization frequency and interval between immunizations: The total number of immunizations required depend 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 [7].

**Isolation and purification methods for IgY**

Several methods were described in the 1950s for purifying IgY based on the strategy of separation of proteins (levitins) from lipoproteins (lipovitellins) and the rest of the yolk lipids using extraction with organic solvents with rather low yields of antibody. However, purification methods based on organic solvents like chloroform remain in use. Other methods are based on affinity chromatography or on dilution of the yolk followed by a freezing-thawing process after which the process consists of ion exchange chromatography and salt precipitations often combining a number of salts like for e.g. polyethylene glycol (PEG), dextran sulfate, dextran blue, sodium sulfate, ammonium sulfate, caprylic acid and sodium citrate. Water dilution method found to be superior in terms of ease of use and large scale production of IgY. This is simple rapid and efficient means of purifying IgY with high activity [8].

**Applications of IgY in biomedical research and in human and veterinary medicine**

Powdered whole eggs or yolks have been used as an inexpensive alternative for the IgY treatment of enteric diseases in veterinary medicine Kumaran et al. [9]. The most famous example of a successful therapeutic/prophylactic use of IgY is the treatment of calves and piglets with specific Abs against Escherichia coli, rotaviruses and coronaviruses. Studies using both animal models and trials in field herds have been carried out. These studies confirmed that treatment of diarrhea in calves and piglets with specific egg yolk Abs has achieved significant prophylactic and therapeutic benefits. Another effect of IgY binding to bacterial surface antigens is a marked impairment of bacterial attachment to the intestinal mucosa Lee et al. [10]. Thus, therapeutic IgY administration could reduce the clinical use of antibiotics, and so could lower the risk that bacteria will develop antibiotic resistance.

**Conclusion**

IgY technology more popular and to convince the scientific community of its significant advantages. The significant potential of avian antibodies for further use in immunodiagnosics and identification of disease markers, immunotherapy and the treatment and prevention of disease is expected. 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.

**References**


