



## **Polyclonal Antibodies - Immunoglobulin (IgY)**

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## **1.0 The Immune System**

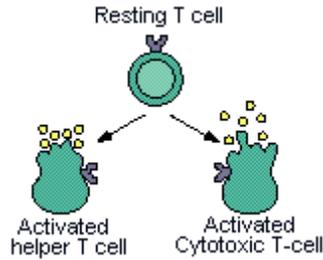
The human immune system is comprised of a complex network of lymphoid organs and highly specialized cells that protect the body against infection by recognizing and killing pathogens. This immune system includes lymphatic organs and vessels, lymph nodes, white blood cells, specialized cells and serum factors. The immune system utilizes both the lymphatic vessels and the blood circulatory system to transport white blood cells called lymphocytes through out the body (1).

The immune system can be classified as an innate or adaptive system. As a first line of defense against pathogens, we depend on innate immunity. Innate immunity is nonspecific and includes physical barriers such as skin, mucous membranes and secretions with antimicrobial activity including tears and mucous. Pathogens that cross this line of defense are often destroyed by phagocytes which are white blood cells that engulf and digest cellular debris and foreign agents by the process known as phagocytosis.

However, innate immunity is not sufficient to ward off all invaders. Thus, to allow for a stronger immune response and immunological memory, we have developed a specific, inducible system of defense called the **adaptive or acquired immune system**. A substance capable of inducing or triggering an immune response is called an **antigen**. The adaptive immune system is responsible for distinguishing between self and foreign antigens. The most common antigens are foreign proteins. Antigens are contained in toxins, bacteria, viruses, and foreign cells. The immune system recognizes and attacks foreign antigens via a binding process that signals the other parts of the immune system that a foreign antigen is present. Continued exposure to an antigen will result in an increase in the production of the antibodies to that antigen (1).

Lymphocytes, as previously mentioned, play an integral role in the adaptive immune system. There are two major types of lymphocytes known as B cells and T cells and they are derived from hematopoietic stem cells found in the bone marrow. To mature lymphocytes must migrate from the bone marrow to either the central lymphoid tissues for of the B cells or the thymus for the T cells. Both B or T lymphocytes have the ability to identify specific foreign invaders and in coordination with other immune components eliminate the foreign antigen. Antigen-activated T cells induce responses in other cells and destroy infected cells while antigen-activated B cells become plasma cells and secrete antibodies. Both cells carry receptor molecules that recognize specific targets.

T cells play an important role in monitoring the blood and lymph for pathogens, and destroying the cells they recognize as foreign invaders. They regulate the cell-mediated immune response which is produced by direct contact between T cells and foreign antigens on the surface of infected cells. T cells differentiate into two different types of T cells which are helper T cells and killer T cells (also known as cytotoxic T cells).

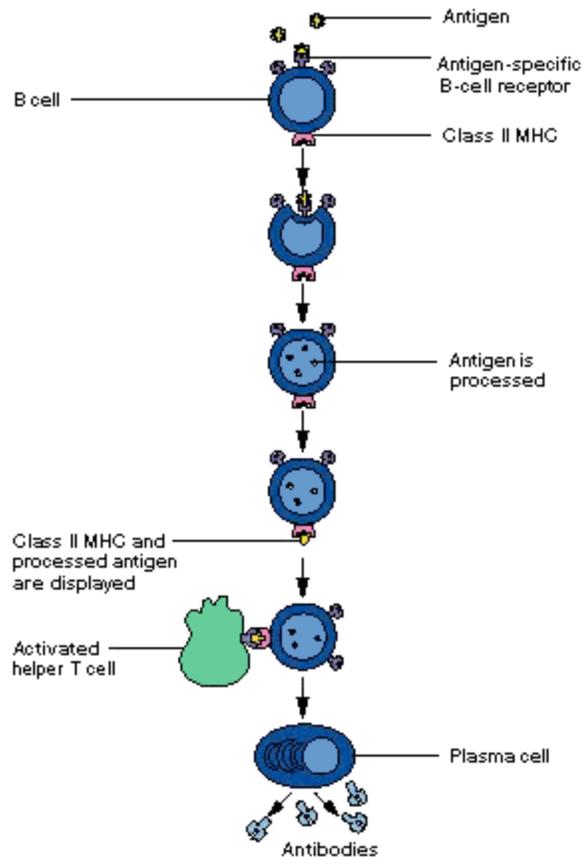


**Figure 1: T cells**

\*Derived from the National Institute of Allergy and Infectious Diseases

Helper T cells are responsible for alerting B cells to initiate antibody production and for activating other immune cells to influence which type of antibody is produced. Killer T cells primarily attack and destroy infected cells. When an antigen is encountered, a macrophage or dendritic cell (antigen-presenting cells in the skin, respiratory and gastrointestinal tracts) breaks down the antigen. The antigen fragments are displayed on the surface of the macrophage or the dendritic cell. T cells depend on special cell surface molecules known as the major histocompatibility complex (MHC) class I to assist them in recognizing antigen fragments. These molecules are displayed on the cell surface with the antigen fragments. A receptor on another Killer T cell recognizes the antigen-MHC class I complex and binds to it. The binding process and a helper T cell activate the killer T cell so that it can destroy the infected cell.

B cells, on the other hand, play a vital role in the humoral immune response which refers to the immune functions mediated by antibodies. In order to activate a B cell to produce antibodies, a foreign antigen triggers an immune response where a B cell binds a specific antigen to one of its receptors and displays the antigen fragments on the surface of the cell combined with an MHC class II protein. The whole complex binds to an activated T cell which stimulates the B cell to differentiate into plasma cells that produce antibodies.



**Figure 2: Activation of B cell to make antibody**

\*Derived from the National Institute of Allergy and Infectious Diseases

These antibodies recognize specific antigens and bind to them to mark them for destruction by other immune cells. They neutralize antigens by a lock and key interaction where the antibody receptor site called a paratope (lock) binds to the antigen binding site called an **epitope** (key). The antigen can be neutralized directly by the antibody as a part of the pathogen necessary for its growth or survival can be blocked by the binding interaction.

The immune system of humans is a complex network of numerous types of cells, proteins, tissues and organs. As part of this intricate immune response, our system is able to adapt over the course of time to recognize particular pathogens more proficiently. The process of adaptation creates immunological memories that ensure our bodies are effectively protected during future encounters with pathogens that have already been exposed to our system. Based on acquired immunity, the concept of vaccination was developed.

A vaccine is an antigenic preparation used to produce active immunity against a specific pathogen and its product resulting in prevention of the disease. **Vaccines** were developed to produce immunity against a disease by exposing the patient to an inactivated or dead strain of a microorganism so that the body can **produce protective antibodies** against the organism in

the vaccine. Primarily, the vaccination procedures and the subsequent antibodies would prevent the infection from occurring. Vaccines successfully prevent outbreaks in measles, chicken pox, polio, bubonic plague, influenza, and a number of other infections. A future development of vaccines is directed towards the cure and/or treatment of cancer.

## 2.0 Antibodies

Mammals have five major immunoglobulin classes which are IgA, IgD, IgE, IgG, and IgM. Although each immunoglobulin has different biological properties and functional differences, all immunoglobulins are made up of protein chains: two long chains called heavy chains and two shorter chains known as light chains. This creates a “Y” shaped image to the protein molecule (see diagram). The variations in the immunoglobulin molecules occur in the heavy chain. All classes of immunoglobulins can be produced as membrane bound receptors for antigen or as freely circulating secreted antibodies.

IgM is the first class of immunoglobulins to appear in the blood after injection of an antigen. In humans, the IgM molecules disappear approximately six months after the immune response. This immunoglobulin molecule has 10 combining sites that enable it to bind tightly to antigens that contain several epitopes. These first antibodies produced in an immune response have a higher affinity for antigen than those formed later.

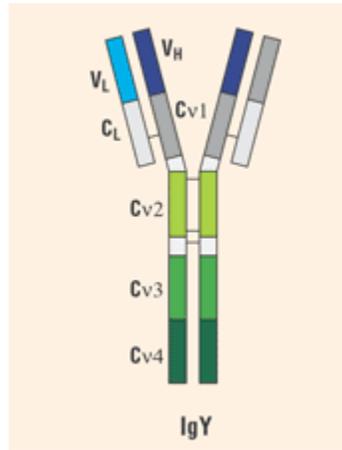
**IgG (IgY) antibodies are the most prevalent type** in the blood and tissues accounting for approximately **75% of the total immunoglobulins found in humans**. (IgG and IgY are initially the same antibody, except that IgY is found in the Yolk of an egg while IgG is found in mothers milk) It mediates the destruction of antigen it binds to in two ways. First IgG (IgY) assists in the destruction of foreign cells by phagocytosis. It is highly efficient in coating antigens which increases the rate at which the other cells of the immune system recognize and destroy them. Second, the binding of IgG to an antigen activates the complement system, which has secondary effects in stimulating the immune system and subsequent healing.

IgA antibodies are the primary immunoglobulins found in external secretions such as tears, sweat, saliva, milk and mucus of the intestinal and respiratory tract. IgA is crucial in the prevention of the disease prior to its access into the body. IgA, therefore, serves as the first line of defense against bacterial and viral infections.

IgE is produced by the B-cells and is generally present in the blood in low concentrations. It is associated with allergic reactions. Its primary function is to stimulate mast cells present in connective tissue to release vasoactive amines that cause dilation and increased permeability of the blood vessels. The IgE effector site has a high affinity to bind mast cells. Once the IgE molecules bound to mast cells are cross-linked with antigen, they enact the release the granules from these mast cells. These granules trigger an inflammatory reaction which may protect against parasitic infections.

### 3.0 Avian Antibodies

There are three types of immunoglobulins in chickens that are analogous to the mammalian immunoglobulin classes. These are IgA, IgM and IgY(5). When speaking of chicken antibodies, **the terms IgG and IgY are often used interchangeably**. Avian and mammalian IgA and IgM antibodies have similar molecular weights and morphology. IgY, the most abundant antibody in egg yolk, is found in chicken serum and is transferred to the chicken embryo from the mother through the egg yolk in high concentrations (2).



**Figure 3: IgY Antibody**

Chicken IgY exhibits some distinct structural and functional characteristics that differentiate it from mammalian IgG. A major difference of the IgY in hens is that it is the functional equivalent to both IgG and IgE found in mammals. It is classified as a primitive antibody and may actually be a precursor to mammalian antibodies. At first, avian antibodies were classified as IgG-like antibodies that are passed on to the egg yolk. Currently, IgY is classified as a distinct avian serum immunoglobulin having a different molecular weight than other mammalian antibodies (IgA, IgM, IgG, IgD and IgE).

Although structurally similar, the heavy chains of IgY are heavier and antigenically different from the IgG heavy chains. This leads to IgY having a higher molecular mass than IgG (180 and 150 kDa, respectively). IgY and IgG have different DNA sequences. IgY bears a closer resemblance to the sequence of mammalian IgE. Other differences between the two immunoglobulins are that IgG is more flexible than IgY because there is an absence of a hinge between the light and heavy chains of IgY.

Most biological activity functions of antibodies are triggered by the Fc region, which plays an important role as a receptor binding region and modulates immune cell activity. However, the major structural difference between IgG and IgY is in the Fc region. The IgG Fc region binds to Fc receptors and specific mammalian cells. This Fc region-Fc receptor complex is responsible for further immunological responses within the mammalian immune system. IgY does not bind to Fc receptors on cell surfaces and, therefore, has a different immunological effect (4).

#### 4.0 Why IgY?

Laying hens, as bioreactors for antibodies, are a highly productive and cost effective source. The ease of acquiring yolk and the amount of energy required to maintain chickens is far more efficient than maintaining large numbers of mammals. Eggs provide a daily source for large amounts of immunoglobulins which can be collected in an efficient and humane process.

**Immunoglobulins have applications** in disease control where they have been systemically used to successfully treat victims of **snake venom, rabies and respiratory syncytial virus (RSV)** (6). Other applications of immunoglobulins such as **passive protection against avian influenza and rotavirus have also been reported.**

As an alternative to mammalian IgG, Chicken antibodies have been produced against a variety of antigens as a therapeutic, research or diagnostic reagent. Their use and availability are an **ideal substitution for mammalian IgG and provides a breakthrough in passive immunological treatment of disease.** As mentioned below, most noteworthy is the potential use of **IgY to both prevent and treat diseases of the upper respiratory and gastrointestinal tracts.**

One example reports in a randomized placebo controlled clinical trial using IgY against **Rota virus** resulted in earlier clearance of rotavirus from the stools and an improvement in the diarrhea associated with this infection in children (7). Other studies in animals have indicated the efficacy of **IgY against parvovirus (8), coronavirus (9), Cryptosporidium parvum (10), Escherichia coli (11) and Salmonella (16) in gastrointestinal infections.**

In a clinical trial in Japan IgY successfully treated **Helicobacter pylori** infections with IgY as both a pure substance (12) and as an ingredient in food (13). These studies demonstrated a new way to treat susceptible populations by incorporating the therapeutic agent in a convenient medium.

Another area for the application of **IgY is as a supportive therapy** for common yet potentially life threatening diseases in the **immune-compromised host (14).** **HIV/AIDS** is an example where the use of IgY to control common commensals such as *Candida albicans* or other gastrointestinal organisms has been suggested as an alternative therapy to the use of **antibiotics or antifungal agents.** This could have wide spread application especially in areas where IgY could be added to food sources with little cultural or social impediments. Studies in Africa have suggested this use and indicate an enhanced quality of life for people living with HIV/AIDS (14).

Further studies also show **positive IgY effects** in the treatment of non-specific gastroenteritis and **Pseudomonas aeruginosa** infections in patients with cystic fibrosis (15).

**Potential applications of IgY antibodies may be to treat avian influenza, inflammatory bowel disease, gastroenteritis, fungal diseases and traveler's diarrhea complex. Other reports verify IgY's therapeutic efficacy in preventing dental caries (tooth decay) (16). IgY may also have relevance in treating acne.**

Additionally, IgY also shows a strong potential to be a useful tool in cancer research and analytical applications. In a number of research applications IgY has proven to be more effective than IgG or other mammalian immunoglobulins. It is suspected that as a result of evolutionary differences, IgY will bind to more epitomes on a mammalian protein than an equivalent mammalian antibody` (3). This is due to the chickens and its associated IgY's ability to differentiate and respond to mammalian proteins as foreign. Other applications may be the utilization of chicken antibodies in solid-phase immunometric assays in place of mammalian immunoglobulins. IgY has less cross-reactivity toward mammalian proteins and therefore can be used to reduce interference in clinical assays (4).

The summary of these studies is that the use of IgY for therapeutic or nutraceutical applications are many. Research into the most useful application is ongoing and promises substantial benefits to the sufferers of disease.

## 5 .0 Polyclonal Versus Monoclonal Antibodies

The decision regarding whether to use a PAb (Polyclonal) or MAb (Monoclonal) depends on a number of factors, the most important of which are its intended use and whether the antibody is readily available from commercial suppliers or researchers. PABs can be generated much more rapidly, at less expense, and with less technical skill than is required to produce MABs.

One can reasonably expect to obtain PABs within several months of initiating immunizations of the chicken, whereas the generation of hybridomas and subsequent production of MABs can take up to a year or longer in some cases, therefore requiring considerably more expense and time. The availability of an "off the shelf " reagent eliminates the issues of time and, frequently, cost.

The principal advantages of MABs are their homogeneity and consistency. The monospecificity provided by MABs is useful in evaluating changes in molecular conformation, protein-protein interactions, and phosphorylation states, and in identifying single members of protein families It also allows for the potential of structural analysis (e.g., x-ray crystallography or gene sequencing) to be determined for the antibody on a molecular level. However, the monospecificity of MABs may also limit their usefulness. Small changes in the structure of an epitope (e.g., as a consequence of genetic polymorphism, glycosylation, and denaturation) can markedly affect the function of a MAB. For that reason, MABs should be generated to the state of the antigen to which it will eventually need to bind. **In contrast, because PABs are eterogeneous and recognize a host of antigenic epitopes, the effect of change on a single or small number of epitopes is less likely to be significant. PABs are also more stable over a broad pH and salt concentration,** whereas MABs can be highly susceptible to

small changes in both. Another key advantage of MAbs is that once the desired hybridoma has been generated, MAbs can be generated as a constant and renewable resource.

PABs frequently have better specificity than MAbs because they are produced by a large number of B cell clones each generating antibodies to a specific epitope, and polyclonal sera are a composite of antibodies with unique specificities.

However, the concentration and purity levels of specific antibody are higher in MAbs. The concentration of specific antibody in polyclonal sera is typically 50 to 200  $\mu$ g/mL, and the range of total Ig concentration in sera is between 5 and 20 mg/mL. In comparison, MAbs generated as ascites or in specialized cell culture vessels are frequently 10-fold higher in concentration and of much higher purity. MAbs are not generally useful for assays that depend on antigen cross-linking (e.g., hemagglutination) unless dimeric or multimeric antigens or antigens bound to a solid phase are used. Additionally, they may not activate complement readily because activation requires the close proximity of Fc receptors. Modification of antibodies by covalently linking a fluorochrome or radionuclide may also alter antibody binding. This potential is less of a concern when using PABs, which recognize a host of epitopes, but it can be significant for MAbs if the change affects its monospecific binding site. Many of the disadvantages of MAbs can be overcome by pooling and using multiple MAbs of desired specificities. The pooled product is consistent over time and available in limitless quantity. However, it is frequently difficult, too expensive, and too time consuming to identify multiple MAbs of desired specificity.

## **6.0 Purpose of IGY**

IGY Immune Technologies & Life Sciences Inc. (IGY Inc) is a biotechnology company that specializes in the extraction, development and commercialization of chicken antibodies. IGY Inc has developed a proprietary and patented method for the extraction of IgY from egg yolk. The result is a product that has many therapeutic, medical and nutraceutical applications. By supplementing the body's own immune system IgY supports and enhances the ability to prevent disease.

IGY Immune Technologies & Life Sciences Inc. (IGY ITLS) acquired the IgY extraction technology from IRI Separation Technology Inc. who conducted thorough scientific investigation and research in developing a unique process for immunoglobulin extraction. A number of years have been spent perfecting the details of the process which has resulted in a higher purity product yielding greater quantities of immunoglobulins compared to the conventional processes. IGY Inc is now set to use advanced methods for IgY capture and purification, which will lead to the commercialization of the antibody products through the patented processes.

The IgY Extraction process is carried out in a Canadian Food Inspection Agency (CFIA) certified facility and consists of mixing chicken egg yolks with water – IgY Immunoglobulin molecules are water soluble and then separating the water (Water Soluble Fraction – WSF) from the egg yolk solids through a centrifuge process. The residual yolk – known as High Ratio Yolk is then

marketed as a commercial yolk product and the WSF is concentrated through Ultra Filtration before being passed through a chromatographic process using an ion exchange process, with specialty resins, that is selective to IgY Immunoglobulin protein molecules – the IgY protein molecules are then eluded from the exchange column and dried...Research has shown that when IgY protein molecules are stored at a fifty percent purity they have a much longer shelf life than when stored at higher purity levels. IGY Inc blends its IgY powder with egg white to achieve a fifty percent purity level.

Currently, IGY ITLS is probably the only company that has the capability to cost effectively produce and supply purified immunoglobulins at a commercial scale to meet the demands of the nutraceutical market. As the general population becomes more knowledgeable about health awareness issues, nutraceutical products are becoming increasingly beneficial in promoting and maintaining health. Enhancement of the immune system is one of the most cost-effective health investments and will become a progressively more central focus in dietary supplements. Few available nutritional supplements contain any immunoglobulins or have the potential to naturally boost our immune system to prevent or fight off disease.

IGY Inc is dedicated to exploring specific and general uses of antibodies to prevent disease complexes and specific diseases. As the health awareness paradigm shifts today from disease treatment to disease prevention, IGY Inc is indeed *helping prevent the need for a cure!*

## **7.0 IgY for therapeutical or prophylactic use in human medicine**

### **7.1 Treatment of intestinal infections in children**

The adherence ability of many viral and bacterial pathogens is a major prerequisite for the successful colonisation of a higher organism, especially with respect to the host's respiratory and intestinal mucosae. It has been shown that specific IgY Abs against Salmonella antigens are able to inhibit in vitro the adhesion of this bacterium to epithelial cells (Lee et al. 2002). Casswall (1999), Carlander et al. (2000), and Sarker et al. (2001) investigated the action of hyperimmune bovine colostrum (HBC) and IgY against human rotavirus isolated from infected children. The oral administration of IgY Abs resulted in a significant protective effect (Sarker et al. 2001). An anti-human rotavirus (strains Wa, RV5, RV3, ST3) IgY Ab was also effective, although to a lower extent than with HBC.

### **7.2 Treatment of Helicobacter pylori**

Therapeutic protection through IgY anti-Helicobacter pylori Abs has also been investigated in animals (Nomura et al. 2005) and humans (Shimamoto et al. 2002, Suzuki et al. 2004). Shin et al. (2003) were able to identify the immunodominant proteins of H. pylori. Antibodies with specificity against these proteins were more effective as a prophylactic reagent as compared to Abs directed against the whole bacterial lysate. Altogether, all studies demonstrated a curative effect of the anti-H. pylori Ab. In most cases no complete H. pylori eradication could be achieved. But in view of the increasing bacterial resistance the use of specific IgY Ab minimises the use of antibiotics. Horie et al. (2004) carried out a study with 42 volunteers to test the

protective effect of a drinking yogurt fortified with anti *H. pylori* urease IgY, obtaining a significant decrease in urea breath values of the treated group (fed with IgY-yogurt).

### **7.3 Use of IgY for treatment of colitis and celiac disease**

Worledge et al. (2000) demonstrated significant protective effects after oral application of specific IgY against tumour necrosis factor (TNF) in an experimental rat model for colitis. TNF is implicated in the pathogenesis of inflammatory bowel disease. The oral use of such Abs is considered to have fewer systemic side-effects than the intravenous infusion of a humanised murine anti-TNF monoclonal Ab (Infliximab, Centocor, Malvern, Pennsylvania, USA). Sunwoo and Sim (2004) reported on the use of IgY Ab against dietary gluten proteins which play a role in the autoimmune disorder of the celiac disease. The authors immunised chickens with gliadins and low- and high molecular glutenin. The resulting Ab can be used in different forms, such as table eggs, liquid and powdered eggs, and encapsulated nutraceuticals for treatment of celiac disease.

### **7.4 Treatment of cystic fibrosis**

Carlander et al. (2002) studied the benefits of IgY as a prophylactic tool against infectious diseases in patients with cystic fibrosis (CF), the most common fatal genetic disease of the Caucasian population in Europe and the USA. CF is caused by a mutation of the gene for a chloride channel protein, which results in the secretion of an abnormally thick mucus. This leads to secondary infections in the respiratory tract, caused by several bacterial species, one of which, *Pseudomonas aeruginosa*, infects virtually all CF patients. The researchers treated CF patients orally with an aqueous IgY anti-*P. aeruginosa* solution (70ml, 0.7mg/ml IgY), given as a mouth rinse in the evening. A high level of the specific chicken Abs could be demonstrated in the saliva via an ELISA, for approximately 8 hours after the treatment. Later, the IgY concentration gradually declined, and was completely undetectable in the saliva 16 hours after the treatment. These oral IgY treatments were successful in reducing chronic *P. aeruginosa* infections in CF patients, and thus resulted in a decrease in antibiotic prescriptions (Kollberg et al. 2003).

### **7.5 Prophylactic use of IgY in dental caries**

An effective local protection against plaque formation related to dental caries was achieved with anti-*Streptococcus mutans* IgY (Otake et al. 1991, Hamada and Kodoma 1996, Hatta et al. 1997, Chang et al. 1999, Smith et al. 2001). This passive protection was clearly shown with both SPF rats and human volunteers, following the use of either purified IgY or whole-egg powder. Active immunisation against *S. mutans* glucan-binding protein B (GBP-B,) under experimental conditions, induces good protection against experimental dental caries. This protection results from the continuous secretion of salivary Abs against GBP-B, which prevents the accumulation of *S. mutans* on the dental biofilm. The passive protection achieved by IgY is based on the same principle. In fact, the administration of IgY anti-*S. mutans* GBP-B via the diet and drinking water of experimentally infected rats caused a significant decrease in *S. mutans* aggregation on dental biofilms. In all these trials, a direct correlation was found between a given IgY dose and a

reduction in the incidence of dental caries (Smith et al. 2001). Furthermore, the decrease in the *S. mutans* infection rate did not require continuous IgY administration (Smith et al. 2001). Hatta et al. (1997) evaluated the efficacy of oral IgY anti-*S. mutans* rinses in human volunteers. This IgY inhibited *S. mutans* adherence to saliva-coated hydroxyapatite discs by 59%, while the control IgY from non-immunised hens only gave an 8% inhibition. All these results strongly support the efficacy of oral treatments with anti-*S. mutans* IgY as a new alternative for reducing dental plaque in humans. Zhou et al. (2003) investigated the protective effect of an anti *S. mutans* IgY spray in adult volunteers. There was no difference in dental plaque indexes between controls and IgY-spray group although a significant decrease in *S. mutans* colonies could be demonstrated in the test group after three weeks of IgY application.

### **7.6 Use of IgY as tool in context of bioterrorism**

To test the therapeutic use of IgY Abs LeClaire and colleagues (2002) produced IgY Abs against the highly toxic staphylococcal enterotoxin B (SEB). SEB is considered to be a potential biological warfare agent. Therefore, it exist an increasing necessity to develop vaccines and therapeutic approaches for intoxication with SEB. The authors demonstrated the prophylactic and therapeutic application of anti-SEB IgY. Complete protection of mice and rhesus monkeys against a lethal SEB aerosol challenge has been observed when applied twenty minutes before or four hours after challenge.

### **7.7 IgY as a tool in proteomics**

A new and an interesting field of the use of the IgY-technology is the proteomic analysis. A problem in separation of complex protein mixtures by 2D-electrophoresis is the predominance of high-abundant proteins like albumin which disturb the monitoring of low-abundant proteins. Low-abundant proteins can be of high importance for identification and monitoring of several human (and animal) diseases. Recently, it has been shown that IgY Abs directed against these high-abundant proteins are in fact useful tools for their removal. In addition, these Abs work more specific than matrices with affinity to albumin like for example blue sepharose (Hinerfeld et al. 2004, Ahmed and Rice 2005, Huang et al. 2005).

### **7.8 IgY for Pandemic Preparedness**

In a recent study completed by the International Vaccine Institute in Korea, the following extract from their published studies on H5N1 have been re-printed.

Pandemic influenza poses a serious threat to global health and the world economy. While vaccines are currently under development, passive immunization could offer an alternative strategy to prevent and treat influenza virus infection. Attempts to develop monoclonal antibodies (mAbs) have been made. However, passive immunization based on mAbs may require a cocktail of mAbs with broader specificity in order to provide full protection since mAbs are generally specific for single epitopes. Chicken immunoglobulins (IgY) found in egg yolk have been used mainly for treatment of infectious diseases of the gastrointestinal tract. Because the recent epidemic of highly pathogenic avian influenza virus (HPAIV) strain H5N1 has resulted in

serious economic losses to the poultry industry, many countries including Vietnam have introduced mass vaccination of poultry with H5N1 virus vaccines. We reasoned that IgY from consumable eggs available in supermarkets in Vietnam could provide protection against infections with HPAIV H5N1. (H. Nguyen et al Plos one, April 2010)

The I.V.I. study used H5N1-specific IgY that were prepared from eggs available in supermarkets in Vietnam by a rapid and simple water dilution method cross-protect against infections with HPAIV H5N1 and related H5N2 strains in mice. When administered intranasally before or after lethal infection, the IgY prevent the infection or significantly reduce viral replication resulting in complete recovery from the disease, respectively. In addition, further generated H1N1 virus-specific IgY by immunization of hens with inactivated H1N1 A/PR/8/34 as a model virus for the current pandemic H1N1/09 and found that such H1N1-specific IgY protect mice from lethal influenza virus infection.

The findings suggest that readily available H5N1-specific IgY offer an enormous source of valuable biological material to combat a potential H5N1 pandemic. In addition, their study provides a proof-of-concept for the approach using virus-specific IgY as affordable, safe, and effective alternative for the control of influenza outbreaks, including the current H1N1 pandemic.

## **Conclusion**

Since it is possible to produce antibodies in chicken against a vast array of antigens and epitopes, there exists scope for raising antibodies against any number of bacterial, viral, or biological antigens. 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, diagnostics, and immunotherapy in the future.

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