Innate vs. Adaptive

The avian immune system is divided into two types of immunity – innate and adaptive. Innate immunity can be thought of as the most basic tools the system has to fight off infection. These include physical and chemical barriers, blood proteins and phagocytic cells. The skin, mucosal epithelium, and gastric secretions are all examples of the various physical and chemical barriers pathogens have to evade. Complement is a serum protein that works with antibodies in order to lyse certain target cells. Several blood cells have phagocytic functions, meaning they engulf and remove pathogens, including macrophages, heterophils, thrombocytes, and natural killer cells. Innate immunity is considered the first line of defense and lacks specificity, protecting against multiple types of pathogens (2,3).

Adaptive immunity takes over when innate immunity fails to stop an invading pathogen. Adaptive immunity involves targeted recognition of specific molecular features on the surface of a pathogen, resulting in a series of events intended to eliminate that pathogen and establish protection to subsequent challenges (3). This specific protection can be provided by either passive immunity or active immunity. Passive immunity consists of the maternal antibodies that are present at hatch, providing protection against various pathogens the hen was exposed to or vaccinated against. Active immunity is the immunity that the bird develops through exposure to pathogens, either by natural infection or vaccination, and can be further divided to humoral and cell-mediated immunity.

Certain types of antigens or modifications of antigens will preferentially lead to development of either cell-mediated response or humoral immunity against the antigen (3). Antigen presenting cells, like macrophages, process and present an antigen to lymphocytes, both B and T lymphocytes (22). Those two lymphocyte types are the main cells responsible for humoral and cell-mediated immunity.

In order to understand disease processes and vaccination, a firm understanding of the basics of the avian immune system is required. The avian immune system is composed of several lines of defense to prevent pathogen entry and infection. This review was developed to re-introduce veterinarians in the field and poultry production personnel to the complexities of this important system.

(A): Chicken Stratch, LLC. Dacula, GA. USA. (B): Merial Select. Gainesville, GA. USA
Humoral Immunity

Antibodies are the functional unit of humoral immunity. They are secreted by plasma cells, a type of B lymphocyte. When on the surface of a B cell, these molecules are immunoglobulins, following secretion they are termed antibodies. Antibodies are found in body fluids and tissue spaces and are most effective in eliminating extracellular pathogens.

They react to surface proteins on bacteria, parasites or viruses, attaching to specific molecular features on the pathogen. Three classes or isotypes of immunoglobulins are found in the avian immune system: IgM, IgY (IgG) and IgA (12). Table 1 (see above) demonstrates a few important characteristics of the three known immunoglobulins: When an antibody interacts with the antigen, they activate or enhance effector mechanisms to assist in elimination of the pathogen (see Figure 1). These mechanisms include activation of the classical complement pathway directly on the antigen; preparation of the antigen for phagocytosis through opsonization, agglutination or precipitation; neutralization of the antigen by preventing cellular entry (3).
Additionally, antibodies can bind to antigens that are expressed on the outer surface of infected cells, triggering cytotoxic cells to eliminate infected or neoplastic cells in a process known as antibody-dependent cellular cytotoxicity (3). T-cells are the primary cells active in CMI, composed of several different cell types discussed later. This portion of the immune system functions through a direct effector, an activated T cell, and its target cell contact (17).
examples of actions by cell-mediated responses include activation of macrophages, cell lysis by cytotoxic T lymphocytes and natural killer cells, all mediated by cytokines released by T helper cells or other cells. Cytokines are chemical messengers that coordinate the interactions between immune cells as one of their extensive functions. Cytokines are crucial stimulators of the initiation and maintenance of the immune response and play a role as effector molecules themselves to impact the duration and strength of the response (10). B lymphocytes originate in the lymphoid follicles of the bursa. In the bursa follicles, progenitor cells undergo a process known as gene conversion (see figure 2), creating a multitude of daughter cells each capable of recognizing individual
Lymphocyte Classes

Two types of avian lymphocytes are present – B lymphocytes and T lymphocytes. The letter associated with each type represents its site of differentiation – B in the Bursa of Fabricius and T from the thymus (1). Each type plays different roles; B lymphocytes are more associated with humoral immunity, while T cells are the main players in cell-mediated immunity.

antigens (19). The cells will then mature, proliferate, and differentiate to form either plasma cells or memory cells. These two types of cells will produce antibodies that function to agglutinate or neutralize antigens and are the basis for maternal protection.

T lymphocytes are the antigen specific cells in the CMI response, capable of recognizing a wide range of pathogens. T lymphocytes are further characterized by their role, cell surface markers and T cell receptors. All T cells express a CD3 complex on their cell surface, independent of the T cell receptor present. T helper cells are typically identified by CD4 surface markers, serving primarily a regulatory role in adaptive immunity, both cell-mediated and humoral. T helper cells function to activate macrophages by secretion of cytokines and stimulate B cell growth and differentiation. Cytotoxic T lymphocytes can be identified by typically having CD8 on their surface and are important in lysis of virus infected cells and tumor cells (3).

When either lymphocyte class is stimulated by antigen, proliferation and differentiation occurs into effector and memory cells. Memory cells are recalled when the same antigen is encountered again, quickly differentiating into effector cells to promptly remove the antigen. Production of a wide variety of antigen-specific memory cells is the basis of disease protection and vaccination concepts (3,21).

Non-Specific Immune Cells

Several other cell types play important roles in innate and adaptive immunity. The most significant are natural killer cells, macrophages and heterophils. Natural killer (NK) cells are non-lymphocyte, non-macrophage cytotoxic cells that are important for lysis of virus infected cells and tumor cells. These cells are important for completing the antibody-dependent cellular cytotoxicity described earlier. NK cells can be found in the spleen, peripheral blood, thymus, bursa and intestine. These cells are distinguished from cytotoxic T cells since they do not carry antigen specific recognition molecules on their surface, making them part of innate immunity (3).
Important and unique functions of monocytes and macrophages included chemotaxis, phagocytosis, killing of bacteria and tumor cells and cytokine production. Chemotaxis is the migration of cells toward an inflammatory gradient and in response to various chemotactic signals derived from bacterial products or products of the immune reaction (17).

Macrophages are a part of the mononuclear phagocytic system and play roles in both innate and adaptive immunity. Macrophages are tissue forms of blood monocytes, which begin differentiation in the bone marrow (16,18). Macrophages engulf bacteria or dying red blood cells through phagocytosis. Once internalized in a phagosome, the macrophage contains lysosomes that will fuse with the phagosome to kill the bacteria. After degradation has occurred, macrophages will present bacterial peptides to immune cells, B or T lymphocytes, serving another role as antigen presenting cells. The ability of macrophages and lymphocytes to stimulate each other’s functions provides an important amplification mechanism for specific immunity (3).

Heterophils are considered to be equivalent to mammalian neutrophils, as they are highly efficient in phagocytosis and bacterial killing. These cells respond rapidly to chemotactic stimuli and are often the first cells found in inflammatory responses. Heterophils are effector cells of humoral immunity and appear to be the main effector cells in both antibody-dependent and natural cytotoxicity (7).

Lymphoid Organs

Various organs function to differentiate avian immune cells, either as primary lymphoid organs or secondary lymphoid organs. The thymus, Bursa of Fabricius, and bone marrow are considered to be the primary avian lymphoid organs. The secondary lymphoid organs are the spleen, mucosal associated lymphoid tissues, diffuse lymphoid tissues and lymph nodes, and germinal centers (15).

The thymus is a flat, multiple lobed organ, located in the neck, in close association with the vagus nerve and jugular veins. This is the primary location for development of T lymphocytes. T lymphocytes complete maturation as they move from the cortex to the medulla of the thymus, entering general circulation through medullary vessels (see image 1). The thymus also contains a population of B lymphocytes, approximately 5-20%, with the percentage being age-dependent (15).

The Bursa of Fabricius is an organ unique to birds and is the sole site of B cell maturation and differentiation (6). The bursa is a blind sac on the dorsal surface of the cloaca (see image 2), lined internally with major and minor longitudinal folds, containing the bursal follicles. Approximately 8,000-12,000 follicles are found in the bursa, consisting of a medulla and a cortex, with the cortex containing large numbers of closely packed lymphocytes (15,21).
The lymphocytes are exposed to external antigens through cloacal drinking, with antigen presentation by specialized phagocytic epithelial cells to the developing lymphocytes (5). Within each follicle, the lymphocytes are undergoing the process of gene conversion to generate antigenic diversity (19). Once mature, B lymphocytes will enter the circulatory system and peripheral lymphoid organs, ready to encounter the specific antigen they are programmed to recognize. By sending the specific B lymphocytes into the periphery, the immune system maintains its ability to respond to antigens and produce humoral immunity once the bursa naturally regresses at around 14-20 weeks (15).
The spleen, a lymphocyte predominante organ, is a major site of antigen processing and antibody production in mature birds.

In the developing embryo, the spleen is a major site of granulocyte production; it then shifts to a defensive organ containing many specialized lymphocyte accumulations and macrophages important for antigen processing (15).

Mucosal associated lymphoid tissues are found in numerous locations, such as the gastrointestinal (GI) tract, respiratory tract, and the head. The gut-associated lymphoid tissue (GALT) includes aggregated lymphoid nodules, such as the cecal tonsils, Meckel’s diverticulum, Peyer’s patches, and a recently described esophageal tonsil (14). Collections of lymphocytes can be found throughout the GI tract in the intraepithelium (IEL) and lamina propria (LPL) layers of the walls of the intestine. These collections are mostly T cells, playing a role in antigen recognition and signal transduction (12).

The cecal tonsil is the largest of the GALT centers, containing both T and B lymphocytes. The cecal tonsil can serve as an alternative location for B cell differentiation and plays a role in antibody production and cell-mediated immune functions. The cecal tonsil is considered a prime site for virus isolation for diseases past the acute stage.
of infection, especially important in isolating infectious bronchitis. Peyer’s patches are dense lymphoid cell accumulations that can occur in various locations along the GI tract, with the most consistent location being at the ileocecal junction. These patches appear to be the major inductive site for IgA responses to pathogenic organisms and undigested antigens (12). Meckel’s diverticulum, the yolk sac remnant on the small intestine, becomes a highly developed lymphoid tissue containing germinal centers or B cells and macrophages (10).

The respiratory tract contains tracheal and bronchial associate lymphoid tissues. Along the trachea, a small degree of lymphoid infiltration in the lamina propria is normal, with the amount dependent on age and antigenic stimulation. Commercial chickens in good health have well developed lymphoid tissue in the trachea due to vaccination and environmental antigenic stimulation (15). Bronchial associate lymphoid tissue (BALT) is composed of both B and T lymphocytes, forming distinct nodules along the primary bronchus and secondary bronchus. The BALT also contains lymphoepithelium, dendritic cells, macrophages, heterophils and when stimulated plasma cells and germinal centers (20). In the avian respiratory tract, heterophils are an important component of defense as bronchial tree macrophages are relatively scarce (7,20).

Within the head several lymphoid tissues exist and are collectively referred to as head associated lymphoid tissues (HALT). HALT includes the Harderian gland, lacrimal gland, conjunctival associated lymphoid tissue, lymphoid cell infiltrates of the lamina propria of the nasal cavity and other structures in the larynx and nasopharynx. The Harderian gland is located behind the eyeball within the orbit and is the major secondary lymphoid organ of the HALT. The Harderian gland contains large numbers of plasma cells, with 80-90% of the cell population being B cells. With such a large plasma cell population, this gland serves as the major antibody secreting accessory gland of the lacrimal apparatus and is important in development of vaccinal immunity (15). Antibody levels in tears have been demonstrated to be specific and locally produced for respiratory pathogens such as infectious bronchitis virus (4).
Tools to measure immune responses

Classical immunology tests to measure the cell-mediated immune response are time-consuming and cumbersome, limiting their use to research purposes.

Examining the cell-mediated or humoral immune response can by accomplished by using chemical or surgical means to replicate T or B cell depletion. Mitogen assays are used to examine transformation of lymphoid cells following exposure to non-specific mitogens – substances that induce cell division. Recently through the ability to produce monoclonal antibodies to avian cytokines, a potential diagnostic test has been reported that appears to reflect the CMI response to a pathogen. An enzyme linked immunosorbant assay (ELISA) test has been developed to detect chicken interferon-γ after antigen recall stimulation or vaccination. By selecting a cytokine common to all immune responses, this ELISA has the potential to be utilized for a variety of pathogens (11). This test could become a future diagnostic tool available to more accurate reflect CMI responses to poultry diseases.

Diagnostic tools routinely used to determine disease exposure and vaccine response measure antibody levels produced in response to a particular antigen. The simplest of these tests include the Mycoplasma plate agglutination tests, when a clumping reaction is observed when antibodies are present in serum reflective of active infection. Traditional serological diagnostic tests like hemagglutination inhibition (HI) and ELISA measure antibody responses to a wide assortment of avian pathogens.

New molecular techniques are being used to further the understanding of the avian immune system and develop potential diagnostic tests. Monoclonal antibodies have been developed to recognize avian cell populations (8) and have been used to examine the roles of cell types in their various locations within the bird. Cytokines important for T cell function have been sequenced and cloned, with recombinant cytokines available for detection, quantification, and neutralization of cytokine production (3).
References

AVIAN IMMUNE SYSTEM