Immunologic Concepts Relating to the Bovine Respiratory System

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The science of immunology has moved from the area of serology to the study of cells and antibodies within the animal during the last 15 years. Information gleaned from studies during this period has provided us with a better basis for understanding the reason for bacterial and viral diseases. A summary of some of these advances and an indication of where this knowledge has assisted us in gaining a better understanding of defense against disease processes will be reviewed. Consideration of the antigens associated with the microorganisms, the organs and cells involved in the immune responses and means of resistance or interaction within the host resulting in disease will be discussed.

Microorganisms, whether they be bacteria, viruses or fungi, contain many antigens. For instance, bacteria and fungi may possess antigens in 1) exotoxins or endotoxins, 2) the cell wall and 3) the various enzymes and euchromatin present within the cell. Similarly, viruses contain numerous antigenic compounds. Antigenic sites which may consist of as few as eight to ten amino acids are in 1) the protein coat, 2) the many internal enzymes and 3) the nucleoprotein within each virus. Furthermore, as viruses enter cells and begin replication, they cause the cell to produce new enzymes and structural proteins which are released into the extracellular spaces or incorporated into the cell membranes. These substances are antigenic and recognized by the host immune system as foreign substances. There is good evidence that many neoplasms caused by viruses have new antigenic sites on the cell surface as a result of viral infections. The host's immune system then recognizes these new antigens as foreign and responds against the tumor cells.

The bone marrow, thymus, lymph nodes, spleen and gut-associated lymphoid tissue are the organs which harbor most of the cells involved in immunologic responses. Stem cells in the bone marrow give rise to lymphocytes and monocytes. In fetal and embryonic life these cells may be found in the liver, placenta or occasionally in the spleen. Monocytes enter the circulation and migrate from the vessels into various lymphoid organs and interstitial tissues of the body. Subpopulations of lymphocytes (B cells) derived from stem cell precursors in the bone marrow may travel via the blood and localize in the cortex or medullary cords of lymph nodes, or in periarterial sheaths and splenic cords of the spleen. These cells are the precursors of the immunoglobulin (antibody) producing cells. Another population of lymphocytes arising from stem cells in the bone marrow migrate to the thymus where they differentiate into T-lymphocytes. These lymphocytes, probably through the influence of a thymic hormone, are committed to serve as the carriers of cellular immunity, i.e., delayed hypersensitivity, and allograft rejections. As these cells leave the thymus they tend to localize in the paracortical areas of lymph nodes and the lymphocytic mantles around germinal centers in lymph nodes and spleen (1).

Evidence at the present indicates that there are three cell types, namely macrophages, lymphocytes and plasma cells, which are important in immune responses. Macrophages are important because they trap particulate matter which contain antigens (2). The antigenic substance remains closely associated with the macrophage cell membrane allowing other cells such as lymphocytes to come in contact with the antigens. B-lymphocytes with specific antigen receptor sites on the cell membrane become attached to the complementary antigen of the microbe (3). Specific complementary antigen-binding T-lymphocytes become closely associated with the above cells and as a result of this interaction, both the bone marrow and the thymic derived lymphoid cells undergo a series of cell divisions, giving rise to clones of cells (4). These clones, or daughter cells, are committed to interact only with the specific antigen which triggered the initial response. The bone marrow-derived lymphocytes will differentiate into plasma cells, the cells which produce circulating antibodies with activity specifically for the inciting antigen. The daughter population of T-lymphocytes are concerned with delayed hypersensitivity and with controlling the magnitude of the response to the specific antigen inciting the initial response. With few exceptions, immunologic responses result in the above cell-cell interactions and the formation of both humoral (antibody) and cellular (delayed hypersensitivity) responses.

The basic immunoglobulin molecule which contains antibody activity consists of four polypeptide chains with a molecular weight of 160,000. Two of the

polypeptide chains are heavy or H chains and have a molecular weight of approximately 60,000 apiece. The remaining two chains known as light or L chains have a molecular weight of approximately 22,000 apiece. These chains are held together by a series of disulfide bonds. There are two antigen combining sites located at the poles of the heavy and light chains. Each site consists of a portion of a heavy and light chain. The opposite end of the immunoglobulin molecule which consists only of the heavy chains functions as the portion of the molecule to which complement may attach and/or the molecule in some instances may attach to the surface of cells such as neutrophils, macrophages, basophils or mast cells.

Four different immunoglobulin classes have been recognized to date in the bovine (5). The four classes of immunoglobulins defined at present are immunoglobulins (Ig) G, M, A and E. The characteristic differences in immunoglobulin classes are based on the composition of the heavy chains. IgG, the prototype immunoglobulin, contains two antigen-combining sites and has a molecular weight of 160,-000. It is the most prevalent immunoglobulin in the blood and interstitial fluids.

In the bovine there are two subclasses of IgG, that is IgG_1 and IgG_2. IgG_1 is found in small quantities in secretions. This immunoglobulin fixes the complex series of serum proteins known as complement thereby making this molecule much more effective in lysis, phagocytosis and neutralization of microorganisms. In addition, IgG is selectively secreted into milk and colostrum by cells in the udder. This immunoglobulin subclass becomes the most important one in transferring the cow's antibodies to the newborn calf. In contrast, the other subclass, IgG_2, is not secreted into secretions nor does it fix complement. It is important for controlling and overcoming infections caused by Corynebacterium, etc.

IgM is a polymer of five molecules with ten antigen-combining sites and a molecular weight of 900,000. Because of the large size of this immunoglobulin it remains within the intact blood vascular system. The initial antibody response is characteristically IgM which later resides when the IgG levels rise. Certain diseases such as anaplasmosis are associated with high levels of IgM for a prolonged time (6,7).

IgA is the subclass of globulins which is often associated with secretions (5). The secretory immunoglobulin consists of two molecules with four antigen-combining sites and a secretory piece. The molecular weight is approximately 390,000. Secretory IgA is produced by plasma cells in the mucous membranes and glands lining the respiratory, gastrointestinal and genital tracts. The immunoglobulins then move through the epithelial cells lining the lumen of these organs where they acquire the secretory piece. The immunoglobulin then moves into the secretions where it serves an important role in "local immunity." The secretory piece appears to have an affinity for mucus and holds the IgA antibodies in the mucous blanket. IgA antibodies have virus-neutralizing properties and undoubtedly play an important role in regulating other microbial agents.

IgE immunoglobulin is produced by plasma cells located in the respiratory and gastrointestinal mucous membranes. It consists of a single molecule and has a molecular weight of approximately 200,000. IgE has an affinity for the cytoplasmic membranes of basophils and mast cells. Once the appropriate antigen comes in contact with the IgE molecules on the surface of these cells, the reaction initiates a set of biochemical events in these cells which culminates in the release of vasoactive substances, i.e., serotonin or histamine. In some species, IgE is the immunoglobulin responsible for acute allergic responses such as anaphylaxis.

Immunization has been used successfully to prevent diseases. Many vaccines, such as blackleg vaccine, have proven to be a very effective means of controlling and preventing this disease. Successful immunization procedures are dependent upon 1) deriving the appropriate antigens from micro-organisms, 2) the nature of immunizing agents, and 3) the routes of immunization which are most suitable for the desired protection.

As mentioned earlier, bacteria, fungi and viruses contain a wide variety of antigens to which the immune system responds. Many of the antigens in micro-organisms are of little consequence as far as protection is concerned. Specific protection is associated with those antibodies which inactivate toxins or prevent viruses from attaching and invading cells. In order for a vaccine to be effective, it is important that it contains appropriate amounts of the antigen(s) necessary for protection.

Most of the immunizing agents are either killed or inactivated or they are modified live agents. Killed or inactivated vaccines are most effective when used to immunize against toxins such as those associated with tetanus and blackleg. The problems associated with these vaccines include 1) limited numbers of antigens, 2) alteration of antigens by inactivating substances, 3) the addition of appropriate adjuvants to stimulate and maintain a response, and 4) difficulty in stimulating secretory antibodies. In contrast, the use of modified live micro-organisms often stimulate a more vigorous immune response resulting from 1) larger numbers of replicating organisms, 2) greater diversity of antigens, and 3) the persistence of antigens which assist in maintaining immunity. The greatest disadvantages are related to the possibility of spreading infections and potential vaccine breaks.

Effective immunization with killed or inactivated vaccines is difficult to achieve with one vaccination. Adjuvants incorporated into vaccines may provide a long slow release of antigens resulting in a long-standing immunity. An alternative way of attaining good immunity is by giving periodic boosters at 2-3 week intervals. Although immunity obtained from the kill-

ed or inactivated antigens may appear to be transient, particularly as measured by antibody responses, theoretically the individual should be able to develop a rapid secondary response upon exposure to antigen. The memory T and B lymphocytes would quickly replicate leading to some form of resistance.

The routes used to immunize individuals will often have a bearing on the effectiveness of the vaccine. Systemic immunity is easily attained through parenteral injection of the vaccine. This type of immunization results in cellular and humoral immunity, the latter being most often associated with IgG and IgM immunoglobulin classes. The immunity afforded by this route of vaccination is very effective in controlling bacterial, fungal and viral infections which normally invade cells within the body. In contrast, this type of immunity offers modest protection against agents which normally invade mucosal surfaces.

Local immunity is that type of resistance associated with mucosal and glandular secretions. IgA is the predominant and most effective immunoglobulin involved in local protection. Usually, antigens must be introduced on or around mucous membranes for successful immunization. The plasma cells producing the immunoglobulins tend to localize in the lamina propria of the mucous membranes. Although IgA is found in small quantities in serum, most of it is secreted onto mucous membranes where it serves as part of the first line of defense. IgA does not fix complement through the conventional pathway; however, it does so through an alternate pathway. This then provides a very efficient way of assisting in the destruction of invading microorganisms.

A low percentage (usually less than 1%) of animals may develop a fulminating disease as a result of the vaccination. This may occur in individuals that have genetic or acquired defect in their immune systems. These animals may have a combined deficiency with both B and T cells or the deficiency may be selective, involving one or more immunoglobulin classes or cells. Examples of immune deficiency states which have been recognized in cattle include 1) T-lymphocyte deficiency in Black Pied Danish cattle and 2) a selective IgG_2 deficiency in Red Danish cattle. Introduction of certain live micro-organisms into these animals will lead to a fulminating infection and eventually death (8,9).

Summary

Research within the last 15 years has allowed us to appreciate that many of the phenomena which occur in test tubes in the laboratories also occur within the animal. The recognition of subpopulations of lymphocytes with different functions and the various biological characteristics of immunoglobulins provide a better understanding of the types of immunity one attempts to attain when employing vaccines to prevent disease. Additional research is needed to improve our understanding of resistance to infections and for providing more effective vaccines than are currently available.

References

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Questions

1. What types of antibodies are most important for protection against toxins? viruses?
2. Which class of immunoglobulin is most important for protection in the intestinal and respiratory system?
3. Which immunoglobulin class plays an important role in anaphylaxis by triggering a release of histamine or histamine-like substances?
4. Why are parenterally administered vaccines not always effective for providing good local, i.e., respiratory, immunity?
5. What condition may lead to a fulminating viral infection after immunization with a modified live virus vaccine in a small number of animals?