Immunobiology of Trematodes

Introduction

Dramatic and fast growth of science of immunology is taking place in recent past. Immunobiology of parasitic organisms and their immune reaction has received special attention. Immunology of trematodes has been subject of several valuabale works performed by workers like-

Bloom (1979)
Bloom et al.,(1979)
Bryceson (1980)
It was Terry (2018) who made a comprehensive review of all previous studies.

Immunological Problems of Trematode infection

Most of the work related to immunobiology of trematodes are largely concentrated around Schistosomiasis and fascioliasis. Not only this but most of the studies are in relation to adult than larval forms. The specific area that has received attention are-

1. Immunological response in vitro and in vivo
2. Attempts to develop vaccines
3. Evasion of immune response
4. Immunodiagnostics (Serodiagnosis)
5. Immunotaxonomy

These thrust areas are are directly related to the benefit of man and his live stock in general and at this time of global scenario when parasites, pathogens etc are used as biological war agents in particular.

Immunological Response During trematode infection

Immunological studies require antigen. As far as trematodes are concerned, antigens are derived from two sources-

1. **Somatic Antigen:**
   When various tissues and part of parasite body is used as antigen it is called smatic antigen. It may be endogenous, structural, bound, internal, surface or external.
2. **Metabolic Antigen:**
   When metabolic out come of the parasite (secretory/Excretory) is used as antigen it is called metabolic antigen

Most of the workers are of opinion that vertebrate response to trematode infection is not different from others. They do induce humoral and cellular responses.

Humoral response is characterised by appearance of specific antibodies in serum, body fluids which can combine directly with antigen of parasite. The antibodies produced against the antigen are from B lymphocytes like other pathogen. But transformation of B cells into plasma cell by trematode antigen is not understood fully.

Cellular Response

During trematode infection production of sensitized lymphocytes with specific receptors at the surface takes place. Like basic immune response these cells evolve from stem cells in the bone marrow which are subsequently processed in the thymus under the humoral influence. This processing eventually leads to the production of T cells, the effectors of cell mediated immunity (CMI). This is some times referred as delayed hypersensitivity. Some of these produced T cells work as T helper cell and co operate with B cells an antibody production. All these cells and antibodies are engaged in cellular traffic through out the body during treamtode infection

Schistosomiasis

Schistosomiasis is one of major parasitic disease of man in tropical and subtropical part of the globe. In these parts it can be an important infection of cattle. I endemic areas the prevalence is very high and people use to suffer for years together. This proves that during schistosome infection development of immunity is slow and inefficient. But experiments performed by Crompton and Crompton,(1986)and Butterworth and Hagan(,1987) proved that strong immunoprotection can be developed. Despite of this fact, Helminths have developed several means of escaping these immuneresponses. Recently, Maizels et al.2004 called them “masters of immunomodulation”. These immunomodulatory abilities enable the worm to persist in the host and can lead to interactions with inflammatory and immune mechanisms involved in other infections or to vaccines or in allergic and autoimmune diseases. The focus in this review is on pathogenic helminths of veterinary importance, especially in Ruminants medicine, and includes Fasciola spp. and gastrointestinal worms.

1. It was Smithers and Terry (1969) who first of all reported that inoculation of egg and infection with cercaria larva induces a substantial degree of resistance to challenge infection.
2. Like eggs when cercaria are placed in antisera (Serum from man or animals with patent or cured schistosome infection). The antibodies present in serum forms a thick coat around cercaraia larva. This is known as Cercarienhullen reaktion and is believed to result from antibody com

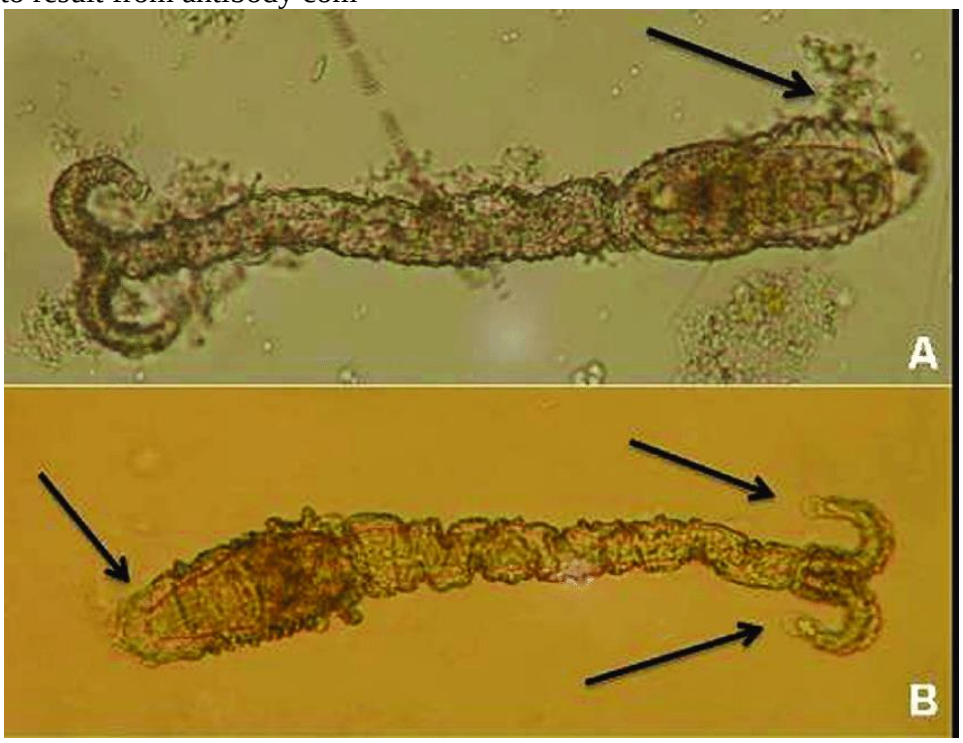

Fig.In the cercariae hullen reaction (CHR) technique, the cercariae of Schistosoma mansoni are placed in the patient serum or cerebrospinal fluid (CSF) for 24 to 48 h. If there are serum or CSF antibodies to the parasite, they react with the cercariae, manifesting itself by cercariae swelling, and the surface becomes wrinkled and with bubbles, that could be located in different areas. Cercariae Hullen Reaction (CHR) positive in CSF (arrows show wrinkled surface) (A). Cercariae Hullen Reaction (CHR) positive in serum (arrows show bubbles in the anterior and tail end of cercariae) (B)

The real nature of immune reaction against schistosomes has extensively been reviewd by Cox (1997). Most of the studies are based on experimental models (monkey and mouse). It was\ found that schistosomes use to mature in these hosts in 5 weeks post infection and adult worms use to live several months and produce eggs. These workers found that already infected host do not permit the establishment of subsequent /challenge infection.
This development of immunity in presence of active infection has been considered to be an example of "Concomitant Immunity". Initially it was thought that antibodies are chiefly involved in the process of schistosomula destroying process but not it is proved that it is much more complex and it involves-

1. Antibody
2. Complements
3. Complements activated through alternate pathway
4. Cells of immune system

Various cells which are involved in it are-

a. Macrophages (in presence of IgE)
b. Eosinophils (in presence of IgG and mast cells and their degranulation)
c. Esosinophils (in presence of complements)

Sher (2007) however observed that immune response varies from host to host and parasite to parasite. Moreover it also depends of variety of other factors like- age of the host, sex of the host, geographical range of the host, physiological status of the host etc.

British school of immunology has some evidence that activation alternate pathway and late complement activation are sufficient to kill the schistosomes in vitro without involment of antibodies and cells.

Butterworth et al (2014) observed that cells use to inflict considerable damage on schistosomula and eosinophils appears to be especially involved. This effect become more compound if anti-schistosome antibodies are present there.

Mechanism of attack

Several workers Butterworth et.al.(2014),Capron and Capron (2015),McLaren (2019) etc worked on the intricate mechanism of immune response and they found that-

a. It begins with the attachment of eosinophils on the surface of schistosomulum, followed by flattening and releasing their contents (granules).
b. These granules causes lesions on the tegument through which eosiniphils migrate into the worm and worm dies.
c. The granules release by eosinophils are unique molecules called as MBP (major basic protein). The histolytic enzymes of MBP are rich in arginine. In fact arginine forms the core of granules and account for 50% of the total protein contents of the granules.
d. The release of these granules on the surface of schistosomulum appears to be an antibody dependent reaction. It was found that IgG is involved.

Cox (2019) pointed out that eosinophil toxicity acts in three stages-

1. Activation of mast cells
2. Release of mediators from mast cells which act as signal to eosinophils
3. Reception of signals by eosinophils and degranulation

Since mast cells and Eosinophils has receptors for IgG, Complerments and IgG, thus it is clear that all this is a complex set of event.

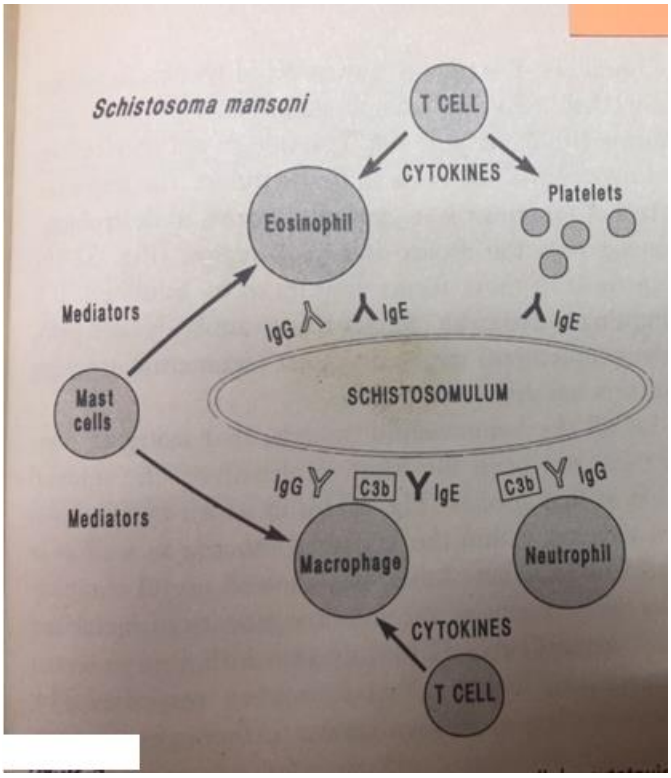

Fig. Antibodies and cells involved in development of immunity in case of Schistostomulum

Fascioliasis

Fascioliasis is a disease of ruminants caused by two major parasitic trematodes,*Fasciola hepatica* and *F.gigantica*. Over the past two decades human fascioliasis has gained notice as a disease of primary importance. Human fascioliasis is currently classified as a plant/food-borne trematode infection, commonly acquired by eating metacercaria encysted on leaves that are eaten as vegetables (Mas Coma et al.2014). There is a high prevalence of fascioliasis among herding communities in low income countries because of their constant close association with live stock.

Fascioliasis is primarily a disease of ruminants, although over the past two decades human fascioliasis has gained significance as an important disease in humans. Human fascioliasis is commonly characterized by a hypoendemic pattern, with low and stable levels of prevalence among a defined population, and generally shows a focal endemic distribution.

However, to date, there have been reports from every continent except the Antarctica, thereby showing a wide cosmopolitan distribution (Masa Coma et al 2014).

Distribution of Human Fascioliasis.

To date, human fascioliasis has been identified in many countries. The highest prevalence has been reported in Bolivia,Peru,Cuba,China, Spain, Nile Delta in Egypt, central areas of Vietnam, and Northern Iran . Bovine fascioliasis accounts for the majority of transmissions and is evenly spread around the world causing 29% of zoonoses . In South East Asia (SEA) and the Indian subcontinent, Cambodia, China, Vietnam, Singapore , Philippines, and India (Ramachandran et al,2012;Kumari et al.,2013 and Ghildial et al.,2014) have also recently reported rising case numbers of the disease that were previously unseen at country or regional health systems.

Immune Response

Some of the Important observation about immune response against fasciola are-

- Most of the studies have concluded that immune response against Faciaola varies greatly in different hosts
- Sheep and goat appear to be unable to resist secondary and subsequent infection.
- Cattle,rats,mice can develop some degree of immunity to challenge infection.
- This has been commonly agreed that like schistosomes fasciola also use to develop immunity by duel mechanism i.e., cellular and humoral

Metacercarial Precipitates

Cercariae hullen reaction can readily be demonstrated with the metacercaria and cercaria of fasciola as observed in case schistosomes. It was found that when metacercaria of fasciola is cultured in immune serum of rat, a precipitate use to appear on the tegument of metacercaria. This precipitate was analyses by Hughes et al,(1991) and it was found to be flukes' metabolic antigen and immunoglobulin of rat serum possibly IgG.

McLaren (2004) performed Ouchterlony diffusion and reported that one precipitation line use to appear which establishes that it is the single antigen and can be considered as functional antigen which can be used in vaccination studies.

Sandman and Howell (1980) studied that pattern of antibody formation in sheep and compared it with liver health enzymes viz.,GLDH (glutamate dehydrogenase) and GT (glutamate transferase) . They found that maximum production of antibodies use to take place when fluke use to migrate through liver and causes maximum damage which is evident from the levels of GLDH and GT. But as soon as it establish in the bile duct, damage is minimized and antibody production also declines.

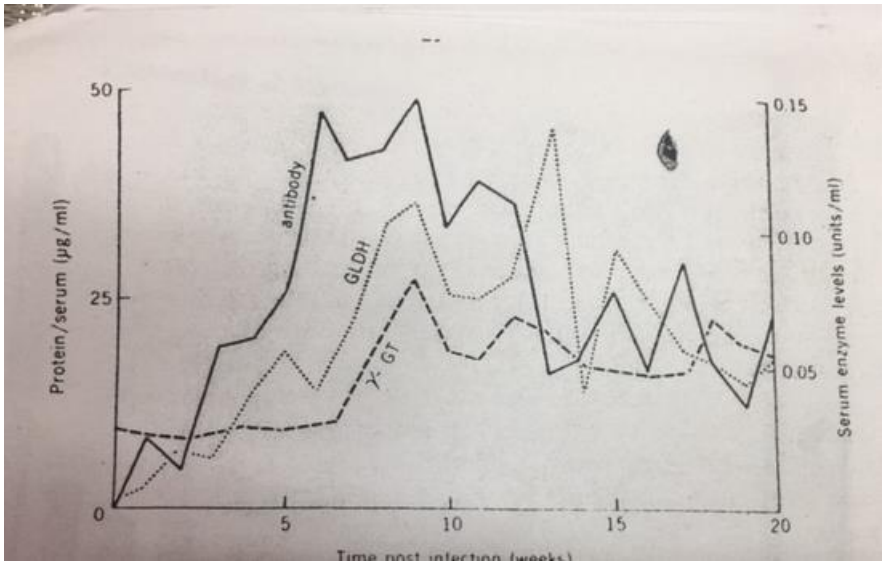

Fig.:Antibody production and level of GLDH and GT due to Infection of *Fascsiloa hepatica*

Hanna (2014) studied details of the tegument and found that in the tegument of Fasciola there are two types of tegument cells (Type I and Type II). The antobodies formed in the serum of infected rat and sheep appears to be the result from sequential expression of surface antigen originating from these cells (Fig.0

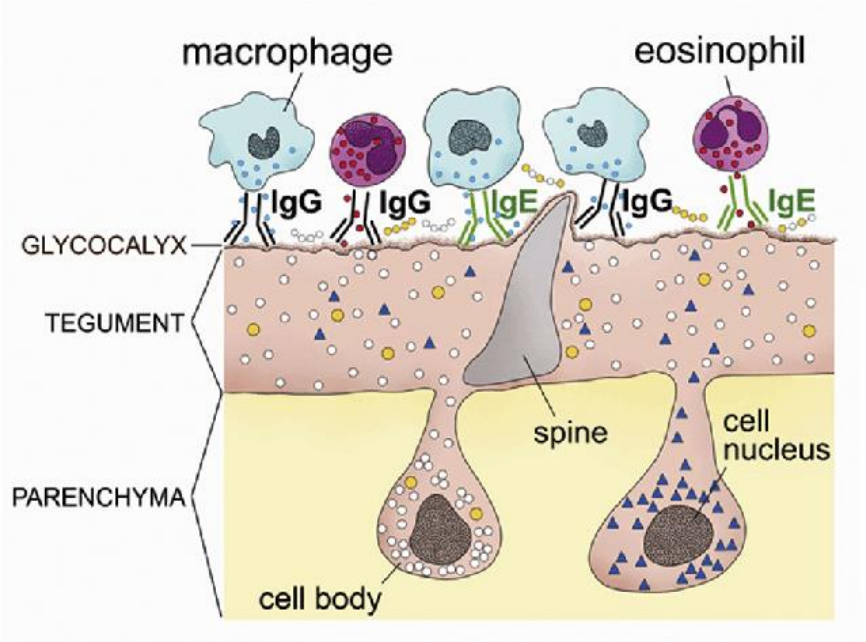

Fig. Tegument of F.hepatica showing types of tegument cells

He further pointed out that at metacercarial and at early juvenile state the tegument cells are are just one type. He designated them Type 0 and fount that it secretes only one type of secretory

body which appear on the surface, membrane bound, electron dense and spherical in out line measuring 0.21 micrometer in diameter. As soon as juvenile enters the liver, differentiation of Type 0 cell begins and it differentiates into Type I and Type II cells. However, some workers believe that Type Ii cells apparently arise independently from the embryonic cells of the parenchyma.

Type O and Type I bodies contain precursors of glycocalyx and use to continuously replace the sloufghed off glycocaly. However, Type II cells are immunogenic in nature and express antigen at the surface.

Immobilization of Miracidium

Kagan (1998) found that miracidia larva of fasciola become immobilized in immune rat serum containing antibodies. He also pointed out that maximum antibody titre appear in the rat serum after 2 weeks of infection, there after it starts declining. He was of opinion that it is parasitic adaptation-

1. At the time of entry and establishment in order to protect from various chemical hazards it use to discharge variety of substances, some of them are immunogenic as well.
2. But as soon as it establishes in the bile duct it minimizes the production and does the molecular mimicry to save it self from the host defence.

Detail Mechanism

The precise mechanism by which the immune response is mounted again fasciola are more or less similar to those of Schistosomes. Both cellular and humoral response is mounted.

Goose(2010) has provided an evidence thatactive and growing Fascila specimens protect themselves fromimmune defences of the host by producing secretory and excretory products which are toxic to Lymphocytes and other immunocompetent cells. He supported his hypothesis with following evedences-

1. Very young flukes do not produce effective quantities of S/E products and are supposed to be immunogenic stage of the life cycle.
2. Older and actively feeding worms produce good quantity of S/E and are non immunogenic satages.
3. Resistance developes early after initial infection.
4. Resistance wanes in long standing infection

Immunodiagnosis

Various immunological techniques are being used to detect the infection of treamtodes in general and fasciola in particular.These are broadly divided into two groups_

1. Intradermal (ID)
2. Serological

These includes-

a. Complement Fixation Test (CF)
b. Indirect haemoagglutination test (IHA)
c. Precipitin test (PT
d. Immunofluurescence test(IFA)
e. Immunoelectrophoresis test (IEP)
f. Defined antigen substrate sphere test (DASS)
g. Counter electrophoresis
h. ELISA
i. Double diffusion test