

## PARASITIC CHALLENGES TO BOOMING DAIRY INDUSTRY OF PAKISTAN

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**ABSTRACT:** Livestock and dairy industry is an important part of Pakistan's economy, contributing about 11.5% in the national GDP i.e. about 55.1% of the agriculture value added. Now-a-days, Pakistan has a very booming dairy industry based on the import of high-producing milk breeds from advanced countries of the world. These imported dairy animals are facing number of challenges in Pakistan, including viral, bacterial, fungal and parasitic ones. Theileriosis, Babesiosis, Neosporosis, Fasciolosis, Trichuriasis and Tick-infestation are among those parasitic problems which are currently being faced by the imported dairy animals in the recently developed dairy farms of Pakistan. Improving diagnostic abilities for these infections will definitely contribute to the better intervention strategies against all these challenges, minimizing their economic impacts on the dairy industry and removing constraints on its flourishing.

**Key words:** Theileriosis, Babesiosis, Neosporosis, Fasciolosis, Trichuriasis, Tick-infestation

### INTRODUCTION

Livestock and dairy industry is an important part of Pakistan's economy. About 30-35 million rural based farmers are involved in raising livestock. The contribution of livestock and dairy industry in the national economy was 11.5% of national GDP which is approximately 55.1% of the agriculture value added. Gross value addition of livestock has reached Rs 1287 billion in 2008-2009. Bovines (35.6 million cattle and 31.7 million buffaloes) constitute a huge part of this livestock and dairy industry and serve as a major source of milk production (about 44 billion litres) and a huge source of meat production (about 1.5 million tons) to fulfil the national requirements[1]. Recently a new trend has arisen in Pakistan for livestock and dairy industry which is attracting huge investments in dairy farming and animal fattening. This would definitely contribute to further enhance the volume of this industry with an ultimate objective to have a competitive share in the international market for milk and meat. Keeping in view this trend, it has become an urgency to cope with all existing and potentially emerging challenges in this growing industry. Among these challenges are various infectious diseases caused by different viral, bacterial, parasitic and fungal pathogens capable of causing huge economic losses in terms of foetal deaths, abortions, loss of milk and meat production[2]. Among these challenges, the parasitic ones being faced currently by the dairy industry include Theileriosis, Babesiosis, Neosporosis, Fasciolosis, Trichuriasis as endoparasites and Tick-Infestation as ectoparasites[3-8].

Theileriosis, Babesiosis and Neosporosis are the diseases caused by protozoan parasites belonging to the genera Theileria, Babesia and Neospora. Theileria and Babesia are transmitted by tick vectors so the dairy farms having problems of ticks, would also be having problems of Theileria and Babesia in the animals at the farms. Theileriosis is caused by *Theileriaannulata* in large ruminants. The parasite multiplies in leucocytes after being transmitted by ticks. Theileria-induced rapid multiplication of leucocytes is apparent clinically in the form of swollen lymph nodes and are easily palpable particularly at pre-

scapular area[9]. Within few days, the parasite also infects red blood cells (RBCs) giving rise to piroplasms that are detectable in thin blood films stained with Giemsa or Field stain, under oil immersion lens of the microscope. Theilerialschizonts can also be observed in the stained thin blood films in case of Theileria spp. prevalent in Pakistan i.e. *Theileriaannulata*. The parasite is responsible for causing huge economic losses in the dairy industry in terms of high morbidity and treatment costs, loss of milk production and induction of abortion in pregnant animals[10, 11].

Babesiosis, the red-water disease, is caused by different species of genus Babesia. The organism is also transmitted by ticks and infects red blood cells (RBCs). The parasitic multiplication inside erythrocytes results into destruction of large number of RBCs. This results into haemoglobinaemia, jaundice and haemoglobinuria in the infected animals. Haemoglobinuria is visible clinically in the form of red-coloured urine. The name of the disease "Red-Water Disease" is also because of red-coloured urine. As ticks are involved in transmission, so the dairy farms having tick problem, will be having more cases of red-water disease due to Babesia[12, 13].

Microscopic examination of stained thin blood films is believed to help in the diagnosis of Theileriosis and Babesiosis. Many times, the only findings in the blood smear examinations are star-shaped RBCs with or without intracellular bodies, and sometimes RBCs with intra-cellular bodies. It becomes extremely difficult to differentiate among intra-erythrocytic parasites particularly between Theileria and Babesia except when theilerial-schizont like structures are visible in the smear[14]. This thing almost doubles the cost of treatment in dairy animals as in such type of cases both types of drugs are used to treat the animals. Recently, the blood examination of two cows that aborted in the last trimester (one cow aborted one month before parturition whereas the other one ten days before parturition) was performed in the Parasitology laboratory and it revealed presence of intra-erythrocytic bodies. Theilerialschizont like

structures were observed in the cow that aborted one month before parturition (Fig. 1).

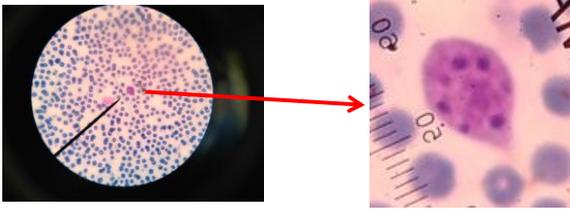


Figure 1: Showing Theileria schizont like structures

The cow was also having fever 104.8° F, salivation, slight redness at incisor 1 and 2 with slightly arched back. This cow responded well to the anti-theilerial treatment using Butalax whereas no schizonts were observed in the second case (Fig. 2).

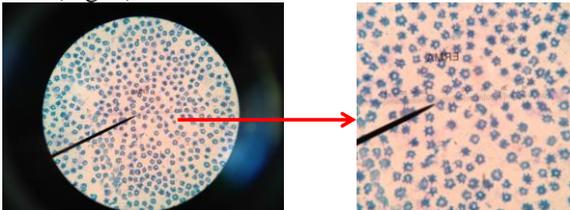


Figure 2: showing intraerythrocytic bodies

For the second cow, the recovery was observed after both treatments i.e. anti-babesial with Imizol and anti-theilerial with Butalax. Extensive search was conducted to find out ticks but no tick was observed. Still there is a question mark over the appearance of intra-erythrocytic bodies without the tick infestation in these cows.

Molecular techniques like PCR can really help at this moment to differentiate between Babesia and Theileria. For this purpose, there is a need to develop a multiplex PCR incorporating primers for all those parasites that can be present inside the RBCs[15].

Another important parasitic challenge is a protozoan disease neosporosis caused by *Neosporacanicum*. This parasite is transmitted originally to cattle via oocysts from dog (canine) faeces. Once infected bovines then maintain the infection by passage from mother to offspring. Only a minimal amount of work has been done in Pakistan to unravel the epidemiology of this parasite in dogs or bovines. Isolates of the parasite have not been obtained from clinical samples of aborted foetuses and biologically characterized. Recent studies demonstrating about 50% seroprevalence of this parasite among dairy animals [16, 17] is just like the tip of an iceberg indicating that a much larger problem may be underlying the tip. Because this disease is able to cause foetal mortality and abortion and has a 50% seroprevalence, it is logical to assume potential economic losses in hundreds of millions of dollars as 50% of the bovine population accounts for more than 25 million animals.

At present, what we need to do is that seroprevalence of *N. caninum* should be determined in dogs as well as in bovines while increasing our sample size to have more reliable data about the true picture of the disease prevalence. Neospora-infected dogs being the final hosts of this parasite[18-21] can easily contaminate the environment by shedding oocysts of

the parasite in the environment. Presence of dogs at the dairy farm creates a risk for this disease in the animals at the farm. In addition, the parasite is transmitted congenitally from infected dams to the calves and is therefore included among potentially major economic problems in booming dairy industry of Pakistan. The aborted foetuses and foetal membranes may act as a source of infection for dogs at the farm or in the wild, if not disposed off properly. The proper disposal means burning of these materials or at least deep burial in order to prevent access of canines like dogs, coyotes, wolves, dingoes that act as final host for this parasite[18, 20, 22-25]. The brain tissues of a maximum of aborted foetuses at the modern dairy farms should be examined through molecular techniques like polymerase chain reaction (PCR). The serological study of aborting cows should also be performed to rule out neosporosis infection. Regular faecal sample examination of dogs kept at the farm should be performed to rule out the presence of oocysts of *N. caninum*. No study regarding seroprevalence of *N. caninum* in canines nor parasite isolation attempts has been conducted in Pakistan. Few studies have done on seroprevalence of *N. caninum* in bovines on limited number of samples. As the bovine population in Pakistan exceeds 60 million, an enlarged sample will give us a more reliable picture of seroprevalence of *N. caninum*. Isolation of viable *N. caninum* and extraction of its DNA has never been performed in Pakistan. Our department is trying to achieve these objectives. International collaboration will be built up for this purpose in-shaa-Allah. Efforts should be made to better understand the parasite prevalence. The development of better diagnostic techniques is needed to better quantify the losses caused by *N. caninum*. Rapid and more reliable diagnostics would help us to devise more effective strategies against this parasitic pathogen for which there is no effective treatment. This will ultimately have a positive impact on bovine milk and meat production by reducing foetal deaths and abortions.

Fasciolosis is one of the most important endoparasitic infections affecting dairy animals and is caused by a trematode named as *Fasciola hepatica* or liverfluke belonging to the family Fasciolidae. Its prevalence is variable in different parts of world. In some European countries like Italy, France and Spain, it varies from 5% to about 30%[26, 27]. Similar level of prevalence has also been found in Pakistan[6]. The pathogenic effects of the parasite are more severe when parasite is immature and is undergoing migratory phase inside the final host i.e. dairy animals. The migration of the parasite in the liver parenchyma causes internal hemorrhages that are a major blow on the animal health and production. The more devastating thing is that microscopic examination of faecal sample will be negative for the presence of eggs. This is because the immature fluke will not be laying eggs. When the parasite matures and reaches gall bladder or bile duct, egg laying starts. At this stage, though eggs can be seen in faecal sample through microscopy but the parasite has already caused the major damage to the animal. In case of large number of migrating flukes through liver parenchyma,

there will be fibrosis of liver and most probably the animal will never return to its production potential. To intervene against this disease in the early phase of the infection, early diagnosis is required. Although control strategies against intermediate hosts of this fluke would help to minimize the spread of the infection but where animals are allowed to graze alongside natural water resources and are offered access to pond, river or canal water, it would become difficult to completely control the infections caused by flukes. For this purpose, there is a need of studies to be conducted on the ELISA designed to diagnose the immature stages of the fluke. Early diagnosis would definitely help to start the treatment of the animals before the extensive fibrosis occurs in the liver, thus preventing irreversible losses of production potential.

Trichuriasis is an infection caused by *Trichuris* species of nematodes. The parasite mainly infects the large intestine, caecum and rectum. The parasite has been found involved in the fatal infection of a Holstein heifer[28]. Similar post-mortem finding was observed when the post-mortem examination of an imported breed of dairy heifer i.e. Friesian was performed. The heifer died because of pneumonic signs within 20 hours after appearance of clinical signs. There was blood-containing frothy discharge from oral cavity. Inflammatory fluids were observed in the peritoneum and pericardium. Hemorrhages were present on cardiac muscles.

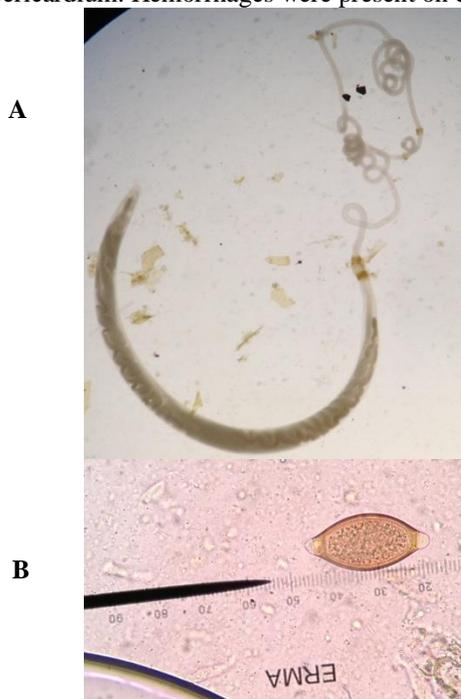


Figure 3: (A) Adult trichuris (B) Trichuris egg (photographs taken in the Parasitology Laboratory, UVAS, Lahore)

The examination of large intestine revealed large number of adult whip-worms (Fig. 3A) attached to the mucosa. The microscopic examination of the faeces also revealed the presence of double-plugged eggs of *Trichuris* species (Fig. 3B). Although lungs were found having interstitial pneumonia and frothy exudate in bronchi, the animal was not having fever whereas results of ELISA for the detection of IBR and BVD antibodies were negative. As the animal

died in acute phase the negative ELISA for antibodies does not rule out the presence of antigen for which antigen-detection ELISA should have been performed. The lack of serum sample did not allow us to perform that test.

A number of different species of ticks have been found infecting bovine population in Pakistan[14]. The most important thing is to prevent the colonization of tick in the dairy farm but once the ticks colonize the dairy farm then only solution is regular treatment of animals with acaricidal to get rid of ticks. In this condition, one should expect the outbreaks of babesiosis and theileriosis in the animals at the farm. For this purpose, timely diagnosis followed by correct treatment is very much necessary to minimize economic losses incurred in terms of loss of production and abortions at the dairy farms.

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