



Seroprevalence of leptospiral infection in cows in farms of Sarab, Iran

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Abstract

This research aims at studying sero infection of leptospira interrogans in farms of Sarab. For this purpose, 200 serum samples were randomly collected. Six kinds of alive leptospira antigens, ballum, canicola, grippityphosa, icterohaemorrhagia, hardjo, Pomona were isolated using MAT test. The results obtained from this test demonstrated that 23% of animals sera were serologically positive for different antigens and 67% samples were negative. Out of positive samples, 31.7% were pomona, 30.9% grippityphosa, 22.4% canicola, 8.8% icterohaemorrhagia and 6.2% ballum. In this study, none of the samples demonstrated positive reaction with hardjo serovar.

Keywords: Leptospirosis, Cow, Sarab, Iran

Introduction

Leptospirosis is an infectious zoonosis created by spiral spirochetes of leptospira (Hassani-Tabatabai and Firouzi, 2000). Leptospira are motile, spiral bacteria with two flagella. Although they are Gram-negative, they are not coloured with normal stains and are usually seen by dark-field microscopy. Silver staining and Victoria and Fontana blue stains are used to prove their existence (Quim et al., 2002). Leptospira are divided into genome species and serovars considering bacterial DNA and superficial antigens, respectively (Quim et al., 2002). More than 250 serovariation in 23 sero group belonging to 10 genome species have been known by now (Kaufmann et al., 2004).

The disease is widespread and has been found almost in all countries (Bhat et al., 1997). In every geographic area; special serotypes of leptospira are prevalent (Howard and Smith, 1999). Generally, distribution of the disease in animals varies from one country to another and even at different regions of a country. It depends on environmental and host factors (Bettelheim and Fogg, 2002). Reservoir hosts are of high importance in prevalence of disease, because most serovariations adapted with one or more hosts (Quim et al., 2002; Hickey and Demers, 2003). Rodents are the most common carriers of leptospira and wild animals are at the second position (Hirsh and Zee, 1999). But, sometimes, domestic animals are also regarded as reservoir (Bettelheim and Fogg, 2002). For instance, icterohaemorrhagia, grippityphosa, ballum have been isolated in rodents and rats, hardjo in cow, canicola in dogs and bratislava in pigs and horses (Hickey and

Demers, 2003). Serological reactions divide leptospira into two interrogans leptospira (pathogenic) and biflexa leptospira (non-pathogenic) species. Its pathogenic species result in developing diseases with different clinical symptoms in human and animal. For example, it appears as acute, sub acute and chronic in cows and is accompanied with symptoms including fever, acute haemolytic anemia, variations in milk, abortion, birth of weak calves, infertility, decrease in milk production and mastitis (Ratnam et al., 1994). It results in fever, icterus, muscle cramp, nervous system disorders, hepatomegaly and splenomegaly, renal disorders and ecchymosis (Ratnam et al., 1994). Microscopic agglutination test (MAT), ELISA, antibody fluorescent, complementary elements fixation test and polymerase chain reaction are used to diagnose contamination. Among them, use of MAT is more common (Bharadwaj et al., 2002).

This study aims at determining frequency of contamination by leptospira and also types of different serotypes found among cows of Sarab City.

Materials and Methods

In this study, 200 blood samples were randomly selected from different farms of Sarab. Venoject tube is used to take 10cc blood from animal's jugular vein. Additionally, some information was also collected regarding animals and the related farms. Sera was separated from the blood and stored in freezer at -20°C in laboratory. Then, samples were transferred to the laboratory and tested using MAT method. 1/50, 1/100, 1/200 and 1/400 dilutions were prepared from the

samples. In this study, six different serotypes of leptospira for icterohaemorrhagia, hardjo, Pomona, ballum, canicola, grippotyphosa were used. After compounding antigen and sera, they were studied using a dark-field microscopy. In case of observing agglutination in each of the samples, number of the sample was written in the results registering sheet while the amount of agglutination graded from 1+ to 4+.

1+ (+): 25% of leptospira reactors were agglutinated and 75% of them are motile and free.

2+ (++) : 50% of leptospira reactors were agglutinated and 50% of them are motile and free.

3+ (+++) : 75% of leptospira reactors were agglutinated and 25% of them are motile and free.

4+ (++++): Most of leptospira reactors were agglutinated (100%).

Negative: No agglutination was observed and leptospira reactors were seen as live and active under the dark-field microscopy. Samples with agglutination 1+ were regarded as negative and just samples 4+ were considered as positive ones. The rest cases were taken into account as suspicious.

Results and Discussion

Considering 200 tested sera, 46 cases demonstrated positive reaction and their relative frequency was estimated as 23%. The rest samples, i.e. 134 cases constituting 67% of all samples, were serologically negative (Fig. 1). Regarding all samples, Pomona with 31.7% and grippotyphosa with 30.9%, canicola with 22.4%, icterohaemorrhagia with 8.8% and ballum with 6.2% showed positive reaction. It should be mentioned that none of the samples had recognizable titer against hardjo serotype (graph 2).

Similar studies conducted at other regions of Iran confirm relatively high infection in cows. In the first wide study conducted in Iran in 1957, three thousand cows and sheep were serologically studied. In this study, 31% of cows and 17% of sheep were found as infected (Howard and Smith, 1999). In the study conducted on 735 serum samples of cows of Tehran suburbs in 1970, 31.2% of cases were positive (Moharrami, 1990). Rate of cows' infection in East Azerbaijan during 1994 was about 48.5% (Shoae, 1994).

Several serological studies regarding leptospirosis disease have been conducted all over the world which demonstrates high levels of infection in animals. In serological study of leptospira infection among dairy cows in Spain (1996-97), 18.33% out of 442 serum sample were positive (Guitian et al., 2001). The result outcome of another study conducted on dairy cows and beef flocks in this country in 1996 revealed infection of 8% in cows and 43% in cattle (Alonso-Andicoberry et al., 2001). In a similar study in Brazil, it was observed

that 46.9% of cows had positive serum reaction (Helio et al., 1999).

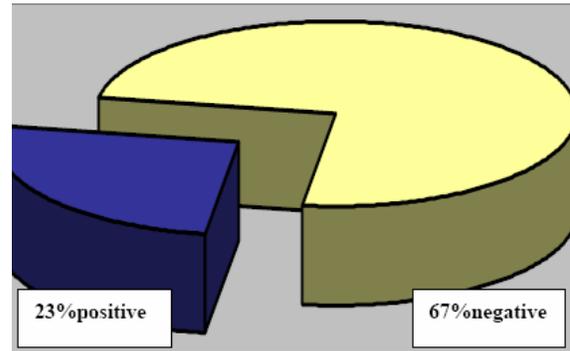


Fig. 1: Positive reactors and their relative frequency in the studied samples

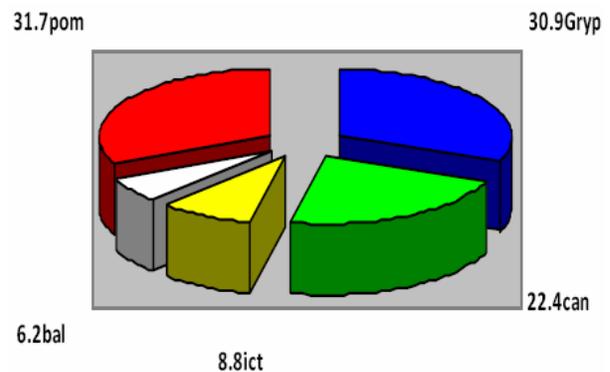


Fig. 2: Percent distribution of different leptospira antigen

Conclusion

Serological infection of leptospirosis in Sarab farms indicates endemic nature. The amount of loss imposed to animal husbandry of the region should be reduced through taking necessary healthy actions and promoting farmers' knowledge about this disease.

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