Review on the public health importance of leptospirosis and the role of rodent

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ABSTRACT

The study examined Leptospirosis, an infectious disease caused by pathogenic gram negative bacteria of genus Leptospira. It has a potential to affect all mammals including humans. It is a zoonotic bacterial disease which is transmitted by rodents and cause severe clinical disease in animals and humans. Rodent is one of the major reservoirs of the pathogen and lifelong renal carrier states observed on them. Rattus species of rodent has been identified as a major reservoir of icterohemorrhage serogroup of Leptospira. Water, soil, and food contaminated by urine of carriers/infected animals are the major sources of humans infection. The bacterium enters the body via intact mucous membrane or skin affected with lesion and scratches. In animals, it mainly causes reproductive loss through abortion, stillbirth, infertility and mastitis and milk drop syndrome. In humans, it can cause acute infectious disease with enlargement of spleen, jaundice, and nephritis. In humans, more than 500,000 estimated cases of severe leptospirosis are reported annually with the case fatality rate exceeding 10%. Few information are available In Ethiopia about the prevalence of leptospirosis in domestic animals, humans and wild reservoirs. Diagnosis of leptospirosis accomplished by direct detection of the organism, its component in the body fluid or tissue by isolation of leptospirosis in cultures or detection of specific antibody. Leptospirosis can be treated orally and intravenously. Human leptospirosis can be controlled by reducing its prevalence in wild and domestic animals. Focusing on infection sources should be as important as prevention and control measure. This review aims to discuss the public health importance of leptospirosis and to highlight the role of rodent in leptospirosis

Key words: Leptospirosis, rodent, zoonosis.

List of abbreviations: Ag, Antigen; CSF, Cerebrospinal fluid; DNA, Deoxyribonucleic Acid; ELISA, Enzyme-Linked Immuno Sorbent Assay; EMJH, Ellinghausen-McCullough-Johnson-Harris; IgG, Immunoglobulin G; IgM, Immunoglobulin M; Loa22, Leptospira Omp A-like lipoprotein; LPS, Lipopolysaccharide; MAT, Microscopic Agglutination Test; OmpA, Outer-membrane protein A; PCR, Polymerase Chain Reaction; TLR, Toll like Receptor.

INTRODUCTION

Leptospirosis is a re emerging bacterial disease and one of the most common zoonoses in the world. It causes severe clinical illness in animals and humans. It is caused by pathogenic species of spirochetes of the genus Leptospira that thrive directly within hosts and reservoirs (such as rodents), and indirectly within the environment (Bharti et al., 2003; Brown and Prescott, 2008). In many tropical and subtropical areas, leptospirosis is an endemic disease. In humans, more than 500,000 estimated cases of severe leptospirosis are reported annually, with case fatality rate exceeding 10% (WHO, 2003).

Leptospirosis was first explained by Adolf Weil when he reported it as an “acute infectious disease with enlargement of spleen, jaundice, and nephritis” in 1886 in Heidelberg. Later the most severe form of leptospirosis recognized as Weil disease. Leptospirae were first isolated in pure culture
by Japanese workers in 1914, but the first clear visual observation of *Leptospira* were made in 1907 by Stimson from a post mortem renal tissue sliced of human and in 1916 its presence in rats was noted (Levett, 2001).

Rodents are considered as the major reservoir of *Leptospira* serovars. They usually remain a carrier throughout their life. Lack of sanitation infrastructure such as open sewage systems and poor refuse collection services provide conditions for proliferation of rats, which are the main reservoir for leptospirosis in urban settings (Felzemburgh et al., 2014). Humans get infected when abraded skin or mucous membranes of the eyes, mouth, nose, vagina, come into contact with infected kidneys, urine or urine contaminated environments. Infection spread mainly through direct contact with rat urine or indirectly with water, soil or food contaminated with rat urine (Leibler et al., 2016).

Leptospirosis in animal is characterized by a broad range of clinical symptoms. It mainly causes reproductive losses through abortion, stillbirth, infertility and mastitis and milk drop syndrome (Radostits et al., 2007). In humans leptospirosis can cause headaches, fever, chills, sweats and myalgia. Other symptoms may include lethargy, aching joints, and long periods of sickness. Some highly pathogenic serovars may cause pulmonary hemorrhaging and death (Peter and Narasimha, 2011).

Diagnosis of leptospirosis may be accomplished by direct detection of the organism or its components in body fluid or tissues, by isolation of leptospires in cultures, or by detection of specific antibodies (Schreier et al., 2013). Leptospirosis is treated with high doses of intravenous penicillin in severe case. Human leptospirosis can be controlled by reducing its prevalence in wild and domestic animals. Prevention and control measures should be focused on the source of infection (Koutis, 2007).

The disease burden posed by leptospirosis in Africa has not yet been elucidated due to inadequate data (Costa et al., 2015). Focusing on animal reservoir as a source of infection is important for the disease control and prevention, however little attention has been given to study the reservoir animals. In Ethiopia, leptospirosis is a relatively unknown disease although already reported to occur in domestic animals.

Therefore the objectives of this seminar study are:

1. To review public health importance of Leptospirosis and
2. To highlight the role of rodent in the transmission of Leptospirosis.

**REVIEW ON LEPTOSPIROSIS**

**Etiology**

Leptospirosis is caused by spirochaetes from the genus *Leptospira* which currently contains 20 species containing nine pathogenic, six saprophytic, and five intermediate species (Sarah et al., 2014). These spirochaetes are finely coiled, thin, motile, obligate, slow-growing aerobes. Leptospires are gram-negative but silver staining and immune staining techniques which can offer better results and can be useful for post-mortem diagnosis using fixed or unfixed tissues (Smythe et al., 2013). The morphology of Leptospires is corkscrew-shaped bacteria, which differ from other spirochaetes by the presence of end hooks. Leptospires have two or more axial filaments that are responsible for the motion of the spirochete, and are visualized under dark field microscopy (Devishree, 2015).

The agents of leptospirosis are the pathogenic *L. interrogans*. Pathogenic leptospires are highly motile and invasive spirochetes that have the capacity to survive and grow in tissue by escaping natural defense mechanism. The spirochete *L. interrogans* has 20 serogroups and more than 280 serovars (Pal, 2007). The host for the agent of leptospirosis is divided into maintenance and incidental host. Maintenance or reservoir host is an animal infected with adapted serovar of the organism whereas incidental or accidental host is the exposure of susceptible animals to non host adapted serovars (Yadeta, 2016).

**Phylogeny of Leptospira**

Genus *Leptospira* is classified under Order spirochaetales, Family *Leptospiraceae*, Class Spirochaetes and it is divided into two species: *Leptospira interrogans* (*L. interrogans*), comprising of all pathogenic strains and *L. biflexa*, comprising of the saprophytic strains isolated from the environment (Sharma and Yadav, 2008) as shown in Figure 1 below.

**Pathogenesis**

The bacterium enters the body through intact mucous membranes (mouth, nose, eyes, vagina) or a skin with lesions and scratches (Adler and Moctezuma, 2010; Langston and Heuter, 2003). They multiply rapidly after entering the vascular system, spread and further replicate in many tissues including kidney, liver, spleen, central nervous system, eye and genital tract. The Extend of internal organ damage is available depending on the virulence of the organism and host susceptibility (Craig et al., 2006). In most infected animals, renal colonization occurs because the organism replicate and persists in renal tubular epithelial cells, even in the presence of serum neutralizing antibodies; the organism may be seen within the proximal tubular cells which coincides with the onset of shedding (Radostits et al., 2007).

The known virulence factors of *Leptospira* include Lipopolysaccharide (LPS) (a general virulence factor of Gram-negative bacteria), flagella, heme-oxygenase, the OmpA-like Loa22, and adhesion molecules. In addition,
Figure 1: The Order Spirochaetales contain genus *Leptospira* which is pathogenic to human. Source: [https://www.jfmed.uniba.sk/fileadmin/jlf/Pracoviska/ustav-mikrobiologie-aimunologie/spirochetes, 2018].

Hemolysins and sphingomyelinases may play a role during infection, although there are conflicting reports regarding their true contributions to overall virulence (Narayana et al., 2012). Human susceptibility to leptospirosis may be related to poor recognition of *Leptospira* LPS by the innate immune system (Werts et al., 2001). Human toll-like receptor (TLR), which responds to extremely low concentrations of gram-negative LPS (endotoxin), appears to be unable to bind *Leptospiral* LPS perhaps because of the unique methylated phosphate residue of its lipid A (Nahori et al., 2005).

The incubation period of leptospirosis depends on a dose, infectious strain and host but is averagely between 7-14 days. Antibodies become detectable 5-7 days after infection (Sykes et al., 2011). It takes about two weeks for the leptospires to reach the proximal tubular cells and the tubular lumen in the kidneys (Petrakovský et al., 2014). In some animals such as rodent: brown rat, despite an increased antibody titer, the bacteria can replicate and persist in the renal tubular cells. This may result in chronic shedding of leptospires in the urine for days to months and even years (Langston and Heuter, 2003).

**Leptospirosis in animals**

Leptospirosis has the capacity to infect all mammalian species, but cattle, sheep, goats, dogs, horses and pigs are more likely affected and cats are rarely affected (Levett, 2001). Leptospirosis can be transmitted from one carrier animal to another healthy animal through direct or indirect contact with urine or other body fluids that contain viable *Leptospira*. There are also other means of transmitting infection between farm animals via congenital or neonatal infection. A viable infected neonate can harbour the infection for several weeks after birth and can act as a source of infection. Semen of an infected bull may contain Leptospirae, so transmission by natural breeding or artificial insemination can occur but is uncommon (Radostits et al., 2007).

Leptospirosis is characterized by a broad range of clinical symptoms in livestock with minor difference between species affected: Clinical signs of acute or sub-acute disease are observed in the leptospirome phase and is characterized by septicemia, high fever, anorexia, petechiation of mucosa, depression and acute hemolytic anemia with a haemoglobinuria, jaundice and pallor of the mucosa (Petrakovský et al., 2014). Chronic infections are usually associated with reproductive losses through abortion, stillbirth, infertility and mastitis and milk drop syndrome. Abortion is common during the last trimester of pregnancy (Radostits et al., 2007). Although, other non-specific signs are common, anorexia, lethargy and vomiting were the three most common clinical signs in dogs with leptospirosis (Greenlee et al., 2004). In equine, severe forms of the disease is characterized by conjunctival
suffusion, jaundice, anemia, petechial haemorrhages on the mucosa and general depression. In foal’s renal failure and in pregnant mares, placentitis, abortion and stillbirths may also present (Verma et al., 2013).

Infertility and milk drop occurs only in pregnant or lactating cows because *Leptospira* organisms require pregnant uterus and lactating mammary gland to proliferate. Sudden drop in milk production may affect up to 50% of cows at one time and precipitate fall in the herds milk yield, the decline may last for up to 8 weeks but individual cow milk production will return to normal within 1-14 days (Radostits et al., 2006).

**Public health significance of leptospirosis**

Leptospirosis is one of the most common zoonosis in the world. In humans, more than 500,000 estimated cases of severe leptospirosis are reported annually, with case fatality rate exceeding 10% (Bharti et al., 2003; Tilahun et al., 2013).

**Mode of transmission in human**

The main sources of infection for the incidence of disease are urine of infected or carrier animals, contaminated surface water, mud, feed, soil, aborted fetuses and uterine discharges (Levett, 2001). From these sources, the organism enters the body via mucous membranes of the eyes, mouth, nose, vagina, or leptospires penetrate through small and sometimes invisible abrasions in the skin (Thayaparan et al., 2013).

Modes of transmission of leptospirosis are often categorized as direct or indirect depending on the immediate source of infection. Transmission is direct if the immediate source of infection is animal tissue, body fluids, urine, transplacental, or venereal, whereas immediate source of infection is an environment contaminated with urine of carrier animals and is indirect (Pal, 2012). Contact with rodents and water sources are significant factors, particularly in flood periods. Transmission between humans are very rare and it occurs through blood transfusion, organ transplantation, breast feeding and sexual intercourse (Johnson et al., 2004) as shown in Figure 2.

**Population at risk**

Leptospirosis affects risk groups that are exposed to animal reservoirs or contaminated environments, such as abattoir and sewage workers, salver workers, coal mines, plumbers, farm workers, trappers, veterinarians, pet shop owners, meat handlers, military personnel, laboratory workers, and workers in fishing industry (Katz et al., 2002). Recreational activities that increase the risk of leptospirosis are gardening and water sports such as canoeing, kayaking, swimming and white water rafting residents of some urban areas (Radostits et al., 2007). Household exposure with in
pet dogs, domesticated livestock, rainwater catchment systems, infestation by infected rodents. And also walking barefoot through surface water, skin lesions, contact with wild rodents, accidental laboratory exposure (Wasinski and Dutkowski, 2013). Men are more frequently diagnosed with leptospirosis compared with women and this has been traditionally attributed to the over representation of men in high-risk occupations (Pavli and Helena, 2008).

**The role of rodent**

Rodents play an important role in the transmission of leptospirosis. They may excrete up to 100 million leptospires in the urine and contaminate water, soil, and food by infected urine and become the major sources of infection to humans (Pal, 2007). It is thought that these reservoirs act as a source of infection for humans and domestic animals, which may then become a source of infection for other animals and humans. Rodent play a role in developing country may present reservoir of infection for humans (Guerra, 2009). In developed countries, contact with adopted wild rodents has also resulted in human disease. In urban and rural slum environments, rodents are the primary host responsible for transmitting leptospirosis to humans (Baer et al, 2010).

Many species of murine rodents have been recorded as carriers of this pathogen around the world. Due to their wide distribution and high abundance in rural areas with farmlands and in urban areas with high density of human population, feral and peridomestic rodents are believed to be the most important reservoirs (Gamage et al., 2014). This can be confirmed from the fact that several incidences of human and animal leptospirosis in over forty countries were attributed to the rodents (Mathias and Levett, 2002; Koutis, 2007).

Rats as reservoir, plays an important role for leptospirosis infection in humans. Although dogs, pigs, cows, horses, cats, rabbits, bats, squirrels, raccoons can also serve as a reservoir. However, Rats are the most potential vector and reservoir for *Leptospira* transmission to human beings among other. They are the main source of infections in urban environment (Sumanta et al., 2015). The distribution of leptospirosis in humans was concentrated in areas where rats were highly populated, as well as areas with unfavorable trash management and poor sanitary conditions. Leptospirosis has been associated with flooding and residents in inner cities where there is contact with rodent urine (Leilber et al., 2016).

Even though several strains and serovars are involved in human cases, Icterohaemorrhagiae is the most frequent serogroup, pointing to the importance of rodents as a major reservoir (Perez and Goarant, 2010). The commensal brown rat (*Rattus norvegicus*), which is closely linked to human activities, is believed to be the reservoir of it and transmitted to humans (Brown and Prescott, 2008). Through defective sewers, rats can enter people’s homes, factories; have contact with foodstuffs, etc., thus presenting a risk of transmitting *Leptospira* to humans (Mayer, 2004). Close proximity of rodents to humans and their food supplies, and high rodent densities would be expected to lead to potential disease epidemics, although data on actual disease prevalence in rodents and humans, and their environmental and socioeconomic correlates, are very sparse worldwide for both rural and urban communities as shown in Figure 3 (Singleton et al., 2003).

**Clinical sign in human**

The incubation period in humans is usually 7 to 12 days, with a range of 2 to 29 days (Ko et al, 2009). Human infections vary from asymptomatic to severe, while the classic presentation is a biphasic illness, leptospirosis infection.
can occur in many forms, which include mild, flu-like cases that may not be recognized as leptospirosis, as well as unusual syndrome or progressive fulminant illness without two distinct phases (Pavli and Helena, 2008). The first stage of the classical biphasic illness is called the acute or septicemic phase. It usually begins abruptly, and is characterized by nonspecific signs such as fever, chills, headache and conjunctival suffusion (Murray et al., 2002). Myalgia, which typically affects the back, thighs or calves, is often severe. Some patients also have other signs, such as weakness, photophobia, lymphadenopathy, abdominal pain, nausea, vomiting, a transient rash, sore throat, and coughing or chest pain. The phase last for 3 to 7 days (Peter and Narasimha, 2011).

The second stage is called the “immune phase” because anti leptospiral antibodies develop at this time followed by septicemic phase after 3-4 days (WHO, 2003; Hartskerel et al., 2006). Patients in the second stage of leptospirosis develop either anicteric or icteric disease. This syndrome is characterized by a severe headache, stiff neck and other meningeal symptoms, and typically lasts a few days. The anicteric form is more common and less severe (Bharti et al., 2003). Icteric leptospirosis is more severe also known as Weil disease. It occurs in 5–10% of all patients, is often rapidly progressive, and may be associated with multiorgan failure (Pavli and Helena, 2008). In this form, there may be no period of improvement between the septicemic and immune phases. The most commonly involved organ systems are the liver, kidneys and central nervous system. Jaundice can be severe and may give the skin an orange tone, and acute renal failure occurs in a significant number of cases. Some patients also have hemorrhages of varying severity, from petechiae and epistaxis to severe bleeding in the gastrointestinal tract and other organs. Some cases of icteric leptospirosis are fatal, and convalescence may be prolonged (Katz et al., 2002).

A severe pulmonary form of leptospirosis occurs in less than 5% of symptomatic patients, but has been an important cause of death in some recent outbreaks. It can be seen in both the anicteric and icteric forms, and usually appears on the 4th to 6th day of illness. It is characterized by pulmonary hemorrhage and edema, with dyspnea and hemoptysis, and can be rapidly fatal (Levett, 2001). Up to 10% of leptospirosis patients develop anterior or diffuse uveitis, a few weeks to a year after recovery. The severity of this condition varies, and the inflammation may be acute or recurrent. It usually has a good prognosis, when treated, and some cases may be self-limiting (Radostits et al., 2007).

Role of rodent in infection, transmission and maintenance of leptospirosis

Rodent as reservoir of leptospirosis

The primary reservoir hosts for most Leptospira serovars are wild mammals, particularly rodents. Rodents usually remain carrier throughout their life. Therefore they are considered as the major reservoir of infection (Levett, 2001). Rodents (Rat, Mice, Moles particularly Rattus norvegicus and Mus Musculus) are recognized as the most significant Mammal species maintaining and disseminating leptospires worldwide. The infections in them are thought to be asymptomatic (Jardine et al., 2011).

Rodents are reservoir hosts for a number of Leptospira serovars, including members of the serogroups Icterohaemorrhagiae, Grippotyphosa and Sejroe. In particular, rats are important hosts for Icterohaemorrhagiae and Copenhageni in the serogroup Icterohaemorrhagiae. Other domesticated and wild animals (examples are, skunks, raccoons, wild boars) are also thought to maintain pathogenic Leptospira (Bharti et al., 2003).

Source of infection and mode of transmission

Leptospira can survive in ponds, rivers, surface water, moist soil and mud as environmental temperature is warm. When these environmental temperatures are favorable and the pH is alkaline, these fragile organisms can survive for a week and are transmitted mostly by direct contact (Fentahun and Alemayehu, 2012). Rodents mainly acquire leptospirosis as pups, and maintain it as a chronic infection in the renal tubules, excreting bacteria in their urine throughout their life span, often in increasing amounts. Rats are reservoir with chronic leptospirosis infection. The infection is transmitted from rats to another through direct contact at a young age or older (Defaria et al., 2008).

Infected animals excrete Leptospira in the urine, and the primary route for further transmission of the infection is through contact with urine or water contaminated with urine of infected animals (Hartskeerl et al., 2006). Environmental transmission is possibly the major route for Leptospira infection in rodents. Infection at a very young age (>2 months) suggests that transmission may also occur in utero or to neonates, possibly through infectious milk, or by other routes within the nest including close contact with an infected mother via fomites or uro-gential cleansing of neonates by the dam (Costa, 2014).

Common species of rodent and Leptospira strain

Rattus species among the rodent have been identified as the main reservoir of the serogroup Icterohaemorrhagiae. Thus, the majority of human cases of leptospirosis are caused by the Icterohaemorrhagiae serogroup, which is the most frequently observed serogroup worldwide. Several rodent species were associated with the disease including Rattus rattus, Rattus norvegicus, Mus musculus, Bandicota bengalensis, Bandicota indica, and other (Zilber et al., 2016).

There were reports that some Leptospira species has specific susceptibility to particular rodents. Leptospira icterohaemorrhagiae serovar commonly infect Bandicota
Rodent as source of infection for domestic animal

Animal infections usually happen in areas with the common risk factors for leptospirosis, like moist and warm climates, as well as areas with high rodent infestation and poor sanitation (Ellis, 2015). Naïve animals become infected by contact of intact mucous membranes or abraded skin with infected urine or urine-contaminated soil, water, food, or bedding. Rodent passes the disease to domestic and wild animals directly or indirectly through urine contamination of shared water sources/bodies (Torgerson et al., 2015). Cats may be exposed as a result of rodent contact. Contact with rodents may also pose a risk to dogs to be infected (Sykes et al., 2011).

Factor that increase rodent infestation

Climate and season: winter is associated with increased rat related complaints as rat moves indoor to seek shelter. Increase growth rate in urban rat may be higher in winter, which may attributed to decreased competition (increased access to food) from an overall reduced population size during winter, and decreased decomposition of garbage from colder ambient temperature (Feng and Himsworth, 2014).

Environmental drivers interplaying by changes in climate, and certain anthropogenic factors, have been demonstrated to lead to incidences of leptospirosis in many parts of the world. For instance, higher than normal rainfall and extreme weather events are natural disasters that can cause a rise in rodent populations. This is due to the scattering of debris and garbage, interference of sewerage systems as well as the increased growth of vegetation leading to increased food availability. Flood waters that drive reservoir animals out of their habitats can lead them to move to human populations and increase human-animal contact. This can ephemerally lead to increased transmission of the disease (Lau et al., 2010). On the other hand, a decrease in rainfall can also increase the human-to-animal contact by reducing the water available on the surface as well as forcing rodents into human habitations in order to forage for food (Cook et al., 2008).

Availability of food and harborage: Availability of food source influences the abundance of rats. Improperly stored or disposed food and organic waste, disheveled gardens, and presence of domestic animal (dogs, cats, pets, livestock) in residences, gardens or city block are correlated with rat infestation. Availability harborage determines whether the population becomes established. Any structure that is easily accessible and abandoned may act as a source of infestation. Accessibility to shelter such as holes/crack in roof, wall, ceiling, and building foundation, access point near utility line and sewer systems, and particularly abandoned structures are associated with rat infestations. This is particularly the case with older aged housing and aging of community infrastructure such as defective drains. Defect in sewer system and inadequate sewer baiting also contribute to surface infestations (Michelle et al., 2016).

Higher housing density is possibly associated with urban rodent infestations because the dispersal and colonizan of one home can affect surrounding dwellings. Rats are more likely to disperse successfully over short distances. Impoverished neighborhoods are excessively affected by rat infestations. Suitable harborage and food source for rats tend to be most abundant in neighborhoods of lower socioeconomic status, where properties may be aging dilapidated and abandoned, and public services including waste disposal may be inadequate, leading to unhygienic environments that could be a source of food (Feng and Himsworth, 2014;Michelle et al., 2016).

Diagnosis of leptospirosis

Diagnosis of leptospirosis may be accomplished by direct detection of the organism or its components in body fluid or tissues, by isolation of leptospires in cultures, or by detection of specific antibodies (Hartskeerl et al., 2011; Schreier et al., 2013).

Direct microscopy

Leptospires may be seen on microscopic evaluation of blood, urine, CSF and peritoneal or pleural exudate during the first 10 days of the infection. Dark field microscopy is required as the leptospires are very small, however more than 10000 organisms/ml are required to be able to see them (Zuerner, 2010). The method of direct examination by using dark field microscopy is limited to urine because other body fluids contain artefacts similar to Leptospira organism and the method has both low sensitivity (40.2%) and specificity (61.5%) (Vijayachari et al., 2001). A range of staining methods has been applied to direct detection, including immune fluorescence staining, immune peroxidase staining, and silver staining. These methods are not widely used because they lack commercially available reagents and their relatively low sensitivity (Slack et al., 2007).
Isolation and identification

A sensitive technique for the isolation of *Leptospira* consists of the intraperitoneal inoculation of young guinea pig with fresh plasma or urine, within few days spirochetes become demonstrated in the peritoneal cavity. On the death of the animal haemorrhagic, lesions with spirochetes are found in many organs (Radostits et al., 2007).

*Leptospira* organisms could be isolated from body fluids, mainly urine. Nevertheless, tissue from dead animals gives a greater opportunity for a successful isolation, if the target tissue is not autolysed. Such target tissue is kidney, liver, lungs and brain. If the agent is suspected of abortions, isolation could be attempted from non autolysed abortion materials or tissue samples from a freshly aborted fetus. Isolation of the microorganism from fetal tissue (kidney, liver, lungs) confirms maternal infection (Adler and Moctezuma, 2010). Isolation requires expensive and properly prepared and kept culture media. Inoculated media are incubated at 28-30°C for several weeks or months (Ko et al., 2009).

Cultures are performed in albumin-poly sorbate media such as Ellinghausen-McCullough-Johnson-Harris (EMJH) medium, which is available commercially. Older media contained serum (WHO, 2003). Primary cultures are performed in semisolid medium, to which 5-flourouracil is usually added as a selective agent. Isolated leptospires are identified to serovar level by traditional serologic methods or by molecular methods, such as pulse field gel electrophoresis (Galloway and Levett, 2008).

Molecular technique

DNA amplification using PCR and DNA primers have become an excellent diagnostic tool for detecting the presence of *Leptospira* in animal tissues and fluids and it can be applied to blood, urine, CSF and tissue samples anti or post mortem (Levett, 2003). The chief advantage of PCR is the prospect of confirming the diagnosis during the early acute (leptospiremic) stage of the illness, before the appearance of immunoglobulin M (IgM) antibodies, when treatment is likely to have the greatest benefit. In fulminating cases, in which death occurs before seroconversion, PCR may be of great diagnostic value (Boonsilp et al., 2011).

Serological methods

Macroscopic and microscopic agglutination tests, complement fixation test and ELISA technique are used for the detection of leptospiarue in serum. The macroscopic agglutination examination is a screening test that uses dead Ag but suffers from specificity (Hirsh et al., 2004).

Microscopic Agglutination Test (MAT) is the standard method for the serological diagnosis of leptospirosis. To execute MAT, leptospires are grown in liquid media and used alive. Serum is mixed with this live liquid grown leptospires in order to test for agglutination. Agglutination indicates that the serum contains anti-leptospiral antibodies (Sykes et al., 2011). As the leptospires are thin and small, dark field microscopy is used to evaluate the agglutination. Agglutinating antibodies are most frequently IgM and to a lesser extent IgG. IgM concentrations fluctuate according to the presence of the organism. MAT has a high sensitivity and specificity but the difficulties are that live cultures of different serovars are necessary to carry out the method. It is also necessary that a trained person works with the samples and evaluates the result (Adler and Moctezuma, 2010).

Enzyme linked immuno sorbent assay (ELISA) test of leptospirosis can be performed either by using commercial kits or within house produced antigen. A broadly reactive so-called genus-specific antigen is generally used to detect IgM and sometimes also IgG antibodies against leptospiral antigen. The presence of IgM antibodies indicates current or recent leptospirosis (Brown and Prescott, 2008).

Common commercially-available *Leptospira* IgM ELISA is used to serologically detect acute leptospiral infections in patient serum samples. This ELISA works on the principle that any *Leptospira* IgM antibodies present in patient serum will bind to the leptospiral antigen attached to the polystyrene surface of the micro wells (Pal, 2007; Heymann, 2004). The ELISA test is more accurate than other tests and has much advantage from the point of view of laboratory practises. It has excellent diagnostic specificity and sensitivity, convenient technical feature including automation and can be used efficiently as serenity test for large number of serum samples (Hirsh et al., 2004).

Status of leptospirosis in Ethiopia

In Ethiopia, although climatic condition, socioeconomic and other factors are highly favorable for the occurrence and spreading of the disease, few information’s are available about leptospirosis in animals and humans. Rodents may be a significant reservoir of leptospirosis as they are in other areas of the world. In the case of human leptospirosis, there is a pilot study in Wonji Hospital. According to the study from a total of 59 febrile patients attending the outpatient department of Wonji Hospital, 47.46% of the patients were positive for leptospirosis and the occurrence of the disease was more common in males than females (Eshetu et al., 2004).

Seropositivity has been demonstrated in domestic animals working in Ethiopia by Moch et al. (1975), found incidences of 91.2% in horses, 70.7% in cows, 57.1% in pigs, 47.3% in goats, 43.4% in sheep, 15.4% in camels and 8.3% in dogs (Moch et al., 1975). A total of 184 out of 418
horses had antibody titres of 1:100 or greater to at least one of 16 serovars, demonstrating the presence of 16 serovars of *Leptospira* species in Central and Southern Ethiopian horses. This means, 44% of the sampled horses were seropositive to at least one serovar. The significant risk factor associated with *Leptospira* seropositive horses were drinking river water and the presence of dogs in adjacent neighboring properties. Dog had a protective effect against seropositivity to serovars Bratislava and Djasiman, which may be due to their ability to control rodents (Tsegay et al., 2016).

**Prevention and control**

Leptospirosis is treated with intravenous penicillin which is the first line antibiotic therapy in severe case. Oral antibiotics such as: amoxicillin, ampicillin, doxycycline or erythromycin used for the treatment of less severe cases. Other third generations cephalosporin’s, such as; ceftriaxone and cefotaxime and quinolone antibiotics also appear to be effective (Goris et al., 2013). Since some outbreaks have been associated with drinking of contaminated water, water purification should be implemented. Prevention and control measures should be focused on the source of infection (Koutis, 2007).

At present there are few effective prevention measures for leptospirosis. Currently, there is no human vaccine available against leptospirosis. Human leptospirosis can be controlled by reducing its prevalence in wild (rodents) and domestic animals (Suputtamongkol et al., 2004). Vaccination against leptospirosis in cattle and swine is generally used as an effective method for control of the disease. Most of the vaccines are formation inactivated bacterins containing one or more serotypes. The immune response is serotype specific; therefore protection is dependent on the use of bacterins containing serotype prevalent in the area. Regular serological testing in herds vaccinated annually can be used to monitor new infection (Sehgal et al., 2000).

Prevention of leptospirosis involves elimination of the carrier state, control of rodents in kennel, maintenance of environmental condition to discourage bacteria survival and isolation of infected animal. Draining or fencing of stagnant water may reduce transmission, limiting rodents and wild life contact with cattle and their feed and water is often difficult to accomplish but it reduces the potential for transmission of leptospirosis (Dhanze et al., 2013).

Rodent-vector control preferably through the use of slow acting rodenticides and improved hygiene may be some of the measures for diminishing the risk of leptospirosis transmission. Occupational hygiene (in sewers, farmers and other high risk groups) that includes the use of water proof shoes and gloves is fundamental for preventing human leptospirosis. Taking care of animal bite, drinking clean water, early treatment, prophylactic therapy, acquisition of information for people coming to high risk areas, is also important for preventing human leptospirosis (WHO, 2003; Massawe and Makundi, 2011).

**CONCLUSION AND RECOMMENDATIONS**

Leptospirosis is among the most important zoonotic bacterial diseases which can infect both animals and humans worldwide. The infection causes jaundice, kidney failure, hemorrhage and death to human in severe case. Rodent plays a significant role in the transmission of diseases as they are the main reservoir of leptospirosis. Availability of infested rodent, climate change and other anthropogenic activity leads to incidence of leptospirosis. Direct or indirect exposure to the urine of infected animal lead to infection in human and domestic animal. Contact with infected animals and their tissues, ingestion of contaminated surface water, sexual and transplacental transmissions are other modes of infection. Mortality in human with leptospirosis remains significant because of delay in diagnosis due to lack of diagnostic infrastructure. Reduction of prevalence in wild and domestic animals and implementing good sanitary measures, vaccination, and control of rodents are the most important control measures of the disease. In Ethiopia, very few studies are available regarding the prevalence of disease in rodent, domestic animal or human.

Therefore, based on the above conclusion, the following recommendations are suggested:

1. Further research will be required on the prevalence of leptospirosis in domestic animals, humans, and rodents in Ethiopia.

2. Awareness creation for risk group will be needed.

3. Implementation of proper hygiene, reduction of rodent infestation and prevent contamination from urine and animal body fluids will be important.

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REFERENCES


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