Investigating the zoonotic risk of human leptospirosis among febrile patients in Nepal

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Introduction

The incidence and prevalence of human leptospirosis are believed to be consistently high in Nepal. However, due to a general lack of awareness of the disease and appropriate diagnostic facilities within the country, this disease remains under-reported and under-diagnosed. As a result, despite being a recognized problem, very little is known about the extent of leptospirosis in human and animal populations.

Some serovars and their main animal reservoirs have been described, but it is unknown which of these contribute the most to cases of human infection. Our study aimed to address this issue by determining the distribution and aetiology of leptospirosis in Nepalese human and animal populations. To do we aimed to collect serum samples from febrile human patients with suspected leptospirosis and from the animals present in their households, and then screen these samples for the most common *Leptospira* serovars in the country.

Objectives

- Describe the prevalence of *Leptospira* serovars in febrile patients in Kaski and neighbouring districts.
- Describe the seroprevalence and distribution of selected *Leptospira* serovars in domestic animal species (ruminantspets) and rodents present in the immediate environment of febrile patients diagnosed with leptospirosis (cases) and febrile patients not diagnosed with leptospirosis (controls).
- Evaluate associations between *Leptospira* serovars in human cases and in various mammalian species in-contact with or in the environment of human cases.
- Determine major sources of infection and risk factors for clinical leptospirosis in humans in Kaski and neighbouring districts.
- Determine the prevalence of risk factors for human cases of leptospirosis in the population of Kaski and neighbouring districts.
- Use the results of the study to develop initial guidelines for a leptospirosis control policy in Nepal.

Methods

A case control study was designed in which febrile patients above 18 years of age with suspected leptospirosis were recruited from eight major healthcare institutions (state owned and private) in Kaski district in April through September 2013. At the stage of patient enrollment, three to five ml blood sample was collected via venepuncture and processed to separate the serum. In addition, patients were interviewed for basic demographic information, contact details, and clinical symptoms using a closed questionnaire form.

Between three to ten weeks after enrollment, patients were visited in their households and a second blood sample was collected from them. During this house visit, blood samples were also collected through venepuncture from the accessible domestic animals owned by the patient, such as cows, buffaloes, goats, dogs, and cats above six months of age. In addition, blood samples were collected from any live rodent species that had been trapped by the patients in their households. The rodent blood samples were collected directly from heart and the rodents were then euthanised. During the house visit, each patient was interviewed for further information on possible risk factors associated with leptospirosis.

All blood samples were processed for serum separation. Serum specimen of each patient and animal was divided into two parts before storing at -80ÐC. The first part of serum was used for microscopic agglutination test (MAT) for leptospirosis and the reminder as a back-up specimen. MAT assays were performed in Mahidol University, Thailand.

Figure 1. Collecting a convalescent blood sample from a previously febrile patient during a follow-up visit to their household.

Preliminary results

Acute blood samples were collected from 295 febrile patients, of which 243 were followed up with a household visit and convalescent blood samples were obtained from 241 of these. The following results exclude the lost-to-follow-up patients.

The mean age of patients was 37 (median 34, Q1–Q3= 25–49). Approximately 67% (n=165) cases came from four municipality areas, the rest came from 38 different village development committees within five districts, including Kaski. The proportion of female patients (n=126) was slightly higher than males (n=117). The majority of patients were engaged in subsistence agriculture (n=65) as their major occupation for income, followed by students (n=35) and home makers (n=28). A high proportion (42%, n= 103) of patients worked in rice fields.

Approximately 44% patients (n=106) owned an animal, and 12% (n=13) of those owning an animal reported a recent history of abortion in at least one of their animals. About half (n=104) of the cases handled animal faecal materials of animals regularly, of which 98% (n=102) did so barehanded. Furthermore, 21% (n=51) patients had tasted cow's urine recently. The proportion of cases owing a dog or a cat was 18%.

More than 85% (n=209) patients noticed rodent or rodent droppings in their households regularly, among which 57% (n=120) used a rat-control method. Using a rodent trap was the most commonly used rodent control method.

All households owned a toilet with a vast majority of them owing a flushing toilet (n= 237). Additionally, 20% (n=48) owned a bio-gas plant to extract methane for cooking and lighting from livestock dung and urine. While a closed source of water (tap water) was the most common source of drinking water (80%, n=195), approximately 65% (n=157) used at least one method for water purification. The use of commercial clay candles was the most popular used method for water purification (n=95). The proportion of patients who used soap and water to wash their hands each time was fairly low (35%, n=86).

The MAT results for the human and animal serum samples collected in this study are not yet available. Due to a short study period of less than a year, seasonal incidence of leptospirosis could not be analysed. More than 90% of cases in this study were recruited from a single hospital, which could have introduced some selection bias into the study. We tried to compensate for this through a high follow-up proportion of patients regardless of their challenging geographical locations.

Figure 2. Collecting a blood sample from the jugular vein of a buffalo belonging to one of the human cases during a follow-up visit to their household.

Lessons learned

This study was undertaken as a One Health collaboration between human health and animal health sectors in Nepal, and a successful working relationship was established between these two at the local level during project implementation. This success was reflected in terms of jointly planning the study, and in the sharing of resources to implement project activities such as recruitment of cases and conducting the household visits.

Furthermore, several capacity building events such as training workshops were organised during the course of this study for both the human and the animal health employees working at different levels within the government and within the non-government sectors. This provided excellent opportunities for people to share their experience and generate ideas for collaborative activities in the future.

Recommendations

Based on our experience from this study we recommend that Ministry of Health and Population and Ministry of Agriculture in Nepal should work more closely in the future. Such collaboration should come through planning research studies jointly, improving existing laboratory diagnostic capabilities and sharing information and resources. This effort would not only help to strengthen the one-health spirit but also control zoonotic diseases that have been recognized as important public health issues.

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