

# Zoonoses and Food Hygiene News

Vol. 15 No.1 January to March 2009

Government of Nepal, Registration Number: 148/049/050

***This Issue has been Supported by VLIR Project, Belgium and UESHP-III, IDRC, Canada***

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*Zoonoses and Food Hygiene News, published four times a year, provides a medium for disseminating technical information on matters related to zoonoses and food hygiene generated in the world, particularly in Nepal. The editors welcome submissions on these topics with appropriate illustrations and references. The views and opinions expressed in the News are those of the authors.*

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## **Mid-Term Review Workshop of VLIR Project**

Mid-Term Review Workshop of VLIR Project: **Impact Assessment and Control of Cysticercosis in the Indian Subcontinent Country: India/Nepal** was organized on March 1 to 8 March 2009 in Grand Hotel, Kathmandu, Nepal. Prof. Dr. Jozef Verduyck, Project Promoter, Prof. Dr. Pierre Dorny, Project-Co-Promoter, Prof. Dr. Vedantam Rajshekhar, Project Coordinator for India, Prof. Dr. Anna Oommen, Chief, Neurology Lab, CMC, Dr. Durga Datt Joshi, Project Coordinator for Nepal, NZFHRC, presented progress report of the project individually. Beside these PhD students Dr. Venkata Raghava and Mr. T. Jayaraman from India and Ms. Minu Sharma from Nepal presented their study research progress. At the end of the workshop following recommendations were made by the participants.

## **Recommendations**

- Training to NZFHRC staff at CMC Vellore.
- Quality control, retesting of former samples.
- Protocol environmental contaminations to be developed.
- To develop methods to control cysticercosis by mass treatment with niclosamide. Importance of criteria to evaluate/monitor programme: coproantigens, prevalence cysticercosis, use of sentinel pigs.
- Dissemination of information, produce extension material.
- Need for informations/scans of cysticercosis patients.
- Extend project for 6 months after the end of the project period.

## **UEHP-III Second Year Progress Report (August 2008 to March 2009)**

1. Preparation of reports for each of the six Act – six reports.
2. Preparation of short summary reports – five reports.
3. Survey (Preparation and knowledge) - 56 interviews.
4. Preparation of survey report.
5. Meeting with and preparation proposals for funding (funding strategy proposals)
6. Meeting with each executive committee of stakeholders groups to access their progress. There were 18 meetings held.
7. Assessment of stakeholders projects (4 projects)
8. Multistakeholders workshop (focus group discussion tools would be carried out)
9. Progress report of the stakeholders activities their continuity of their group work plan official status achievement, challenges etc. (1 joint progress report of all stakeholders).
10. UEHP III synthesis workshop with research team – 1 workshop is planned for May 2009.
11. Lobbying and policy influence strategy SWOT Analysis Workshop (1 workshop) finished.
12. Preparation of communication materials
13. Preparation of VCD< CD, Video etc
14. Annual Progress reviews workshop
15. Final report preparation (final project report)
16. Preparation of technical papers for journals
17. Preparation of technical papers prepared and presented in Chengdu China as IDRC outputs

## **Brucellosis in Milking Buffaloes of Bhaktapur Milk shed Areas**

*Y. P. Mishra, D. D. Joshi, S. P. Gautam, A. Aryal, K. Shahi*

### **Introduction**

Brucellosis is a bacterial zoonotic disease caused by various species of *Brucella* belonging to family Brucellaceae and order Eubacterales. *Brucella* are small, non-motile, nonsporing, intracellular, aerobic, gram-negative, coccobacilli.

Pyakural (1980) studied on *Brucella* agglutinins in human serum in Nepal. The seropositivity rates of human brucellosis were reported as 5.26% in Solukhumbu, 5.36% in Langtang, 2.56% in Malunga, 1.49% in Pokhara, 2.32% in Biratnagar, 3.63% in Bhairahawa, 1.65% in Kathmandu by plate test and 5.26%, 7.14%, 3.84%, 2.23%, 2.79%, 5.54%, 2.47% by tube agglutination test respectively.

Dhakal *et al.*, (2005) done the serological study in 56 slaughtered goats and 35 slaughtered buffaloes in the Chitwan district. Among them 5.36% of the goats and 2.86% of the buffaloes were found to be positive for the brucellosis.

DHS, (2003) indicated that in Nepal around 2-3 % of the cattle and buffaloes are seropositive. Jha, (2003) detected Brucellosis serologically from repeat breeder and aborted cows of cross bred and

exotic cattle. He reported 0.8% positive for Brucellosis out of 118 serum samples.

Shrestha, B., (2008) Seroprevalence of brucellosis in different species of meat animals of Nepal. Total 376 blood samples were collected randomly from buffalo 153. From Thankot slaughter house, 70 goats from IAAS, and 153 pigs from Itahari. The sero samples were investigated using card test. The incidence rate was found to be positives in Buffaloes all showed up to be negative reactors. While out of 70 sero samples of goat and 153 sera sample of pig by card tests 17.14% and 7.18% showed up to the positive reactors.

Rana, H. B., (2002) Sero epidemiological and animal Brucellosis in Surkhet district. Prevalence rate was found 20% positive and Bir hospital by collecting 200 human serum samples examined by card test 14% found positive. All the milk samples were analyzed by MRT. Out of 400 samples examined prevalence rate in buffalo 29% and cow 19%.

Joshi, (1983) studied the seroepidemiological prevalence of human and animal Brucellosis in Kathmandu. Sera samples of 1450 humans, 1069 cattle and buffalo and 247 sheep and goat were examined. The incidence rate was found to be 6.08 % ( 87 /1430) in humans, 8.7 % (93/1069) in cattle and buffalo and 3.64 % (9/247) in sheep and goat.

Joshi, (1984) made investigation on human Brucellosis in Kathmandu Valley serologically. The incidence on a total of 2117 human sera revealed that 57 serums were positive for Brucellosis indicating incidence rate of 2.7%.

A sero-prevalence of brucellosis conducted by Poudel (2006) in small ruminants of Nepal revealed 29.26% (12 out of 41) goats from Makwanpur; 5.12% (2 out of 39) from Chitwan and 5.4% (2 out of 37%) from Kathmandu, the positive one. Similarly, 14.28% (2 out of 14) sheep serum from Chitwan showed the positive results for brucellosis (Joshi et al 2005).

Lefkowitz *et al.*, (2003) done a study on Brucellosis in Yak, Naks, Chauries, and hilly cattle of Langtang valley by MRT (Milk Ring Test) revealed the prevalence to be 17.6% (13 out of 74).

Joshi (2000) studied animal brucellosis in Kathmandu and around the valley. The positive reaction was found to be 4.69% (31/660). Similarly he examined 4229 milk samples (single) of buffaloes and cows from DDC centers, milk vendors and villages in Kathmandu valley and around the valley viz, Panchkhal, Bhaktapur and Banepa and observed 1.25% positive by MRT. The incidence in cow was found to be 3.8% (2/53).

## Material and Methods

Bhaktapur district was selected as the study area. The selected area is a pocket area of buffaloes raising for mainly milk and meat production.

A total of 60 milk samples were collected from individual farmers when they came to the cooperative society to sell their milk and 60 blood samples were collected from the same animals by tracing back to farmers' herd.

## Methodology

A 5 ml Milk samples were collected in vial individual in Kharipati and Salaghari dairy society when farmer came to sell their milk. A 5ml blood was taken from the same animals by tracing back to their farms into vial from the jugular vein in standing position.

The examination was performed at the lab of National Zoonoses and Food Hygiene Research Center (NZFHRC) Chagal, Kathmandu, Laboratory.

5 ml sample was taken in test be the test be is performed by adding 30 to 50 micro liter of antigen (killed B.abortus 99) to 1-2 ml volume of whole milk. Then the sample and antigen was mixed normally. Then mixture was incubate at 37<sup>0</sup> C for 1 hour together with positive and negative control samples. A strong positive reaction was indicated by formation of a dark blue ring above a white milk column.

Rose Bengal test was performed according to the protocol of OIE< 1998 for mscreening purpose. The test was performed at National Zoonoses and Food Hygiene Research Centre (NZFHRC) Chagal, Kathmandu. Blood samples of all the 60 buffaloes were taken for the RBPT test. The agglutination rates were categorized into strong, milk and negative. Control positive and control negative samples were placed as provided by the company for comparison.

## Results

Out of 60 samples taken for milk ring test, 14 samples showed positive to brucellosis. It means 23.22% of sample revealed positive to the disease. The percentage positive case is higher i.e. 40% in Sanothimi VDC. Out of 60 samples taken for the RBPT Test, only 9 samples (i.e. 15%) revealed to positive to the RBPT reagent.

## Discussion

Main VDCs (Chitpot, Jhaukhel, Bageshwari, Chaling, Balkot, Sanothimi) of Bhaktapur district were selected which were having large number of buffaloes population (about 3666) and was a good example of semi urban community of Nepal where livestock rearing was not the primary aim of income generating.

## Recommendations

- The surveillance activities of both public health and animal health sector must be fully coordinated and integrated.
- Regular sero-monitoring or screening and control programme must be promoted in goat, cattle and swine raising areas.
- As it is a zoonotic disease, handling of the suspected or infected animals should be done with due precaution.

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## Serological Evidence of Visceral Leishmaniasis in Human Population of Siraha District

Rakesh Prasad Sah, D. D. Joshi, Arjun Aryal

### Introduction

Leishmaniasis is a vector-borne zoonotic disease caused by parasites of the genus *Leishmania*, a flagellate protozoan related to *Trypanosoma*. This disease is characterized by both diversity and complexity. It is caused by more than 20 *leishmanial species* and is transmitted to humans by ~30 different species of phlebotomine sandflies. Nepal, together with India, Bangladesh Sudan and Brazil constitutes the five countries of the world where more than 90% of VL occurs (WHO, 2002). In addition to VL, some cases of cutaneous Leishmaniasis and PKDL are also reported from Nepal. In Nepal the visceral leishmaniasis is caused by *L. donovani* and is transmitted by *Phlebotomus argentipes* (the vector). Visceral Leishmaniasis (VL) re-emerged in the Indian subcontinent in the mid-1970s after an almost complete absence in the previous fifteen or so years. The disease was first noted in Nepal in 1978 (Joshi 1987) and, since 1980, it has been reported regularly in increasing numbers (Joshi 1986; MOH, cited in Bern *et al* 2000). Asymptomatic carriers and PKDL patients are a particular source of infection for sandflies (WHO expert committee report, 1991). In Nepal Visceral Leishmaniasis is thought to be transmitted by *P. argentipes* whereas *P. papatasi* and *P. sergenti* are found to be responsible for Cutaneous Leishmaniasis in this region (Joshi *et al.*, 1990).

Joshi D.D., (1986) collected a total 1375 sandfly from Dhanusha district and 514 only identified from Dohar village of Singerjoda Panchayat of which the most abundant number of flies are *P.*

*papatasi* (73.4%), followed by *P. argentipes* (4.8%) and *P. sergenti* (17.65%). however, they were unable to identify a significant proportion (17.6%) of sand flies. Das (1998) studied about the vectors of KA and stated that sandflies are found near sinks, basement drains, crack and holes of walls, rat holes in the house and termite hills in the forests. According to Joshi AB, 283positives by rk39 and 284 positives by bone marrow test studied the visceral leishmaniasis in endemic district, Siraha.

The main objective was to document the prevalence of VL in Siraha district for baseline comparison.

### Methods

The blood samples of humans were collected from Siraha & Lahan hospital and the blood samples of dogs were collected from various kala-azar reported villages of Siraha districts from July to September, 2008. Human samples were tested by Formol-gel method as described by Monica Chessbrough, District lab Practices in Tropical Countries (2002). rk39 test method was also used for serology. The questionnaire survey was conducted on the patients from different villages came at hospitals of Siraha and Lahan having the history of fever for 3-5 days.

### Results

For the serological study of the visceral leishmaniasis in human population altogether 150 blood samples were collected from the patients came to hospital having intermittent fever for 3-5 days and were tested for the presence of raised levels of IgG and IgM against *L. donovani* via formol-gel test. Positive reacting serum samples were categorized according to their whitening and gelling as; strong positive, mild positive and negative. Under categorization, 16 samples (10.66%) found to be reacting as strong positive and 6 samples (4%) as mild positive. The appearance of two visible red bands means one control band (the test was valid) and one test band (rk39 antibody was present) which indicated that the test was positive. The mild positives are also considered as strong positive because of slight whitening and gelling within 30 minutes. Hence, altogether the sample prevalence found was 14.66%. And at 95% CI the population prevalence found ranging from 9.56-21.5%. Among the 70 patients, 48 were not using bednet and 22 were using bednet. And the examination of serum of these patients showed 12 positives among 48 and 2 positives among 22.

### Discussion

Our findings highlight the socio-economic factor and environmental factors associated with VL. Siraha district was a good example of rural district of Nepal where maximum percentage of people are facing the problem of poverty. Siraha is adjoined with boarder area of Bihar state of India which is endemic for kala-azar. The prevalence of visceral leishmaniasis in human population in the tested samples was 14.6% via formol-gel test which is higher than the prevalence obtained (ie.10.9%) via rk39 test kit by the data of hospitals of Siraha district in 2006/2007. The percentage prevalence of visceral leishmaniasis in males of study population was found more in male (17.39%) as compared to in females (10.34%). This finding revealed the finding of Joshi A.B., (2002) that the males are more prone to having VL than female due to their migratory outdoor work. The prevalence was found higher (25%) in patients not using bednet as protectant against bite of sandfly than in patients using the bednet (9.09%). This finding revealed the finding of Bern, 2000 that bednet is strongly protectant factor against VL.

**Conclusion**

This study indicates the leishmaniasis as great problem among lower community. As the study run in a small population, study revealed profound study of VL in human population and animal population very urgent. The incidence of Leishmaniasis in Nepal, mainly the visceral form, is increasing at a fast rate indicating that the control attempts are being ineffective. The disease has reemerged from the almost eradicated state of 1965-1970 (MOH, cited in Bern *et al.*, 2002), demanding for changes in the control strategies.

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**NEWS:**

**Animal Slaughterhouse and Meat Inspection Act 2055**

Three days workshop on "Synthesis Analysis Workshop of Urban Ecosystem Health Project Phase I, II and III" was organized by National Zoonoses and Food Hygiene Research Centre (NZFHRC) Chagal, Kathmandu with the support of International Development Research Centre (IDRC) Ottawa, Canada on June 5-7, 2009 in United World Trade Centre, Tripureshwor, Kathmandu, Nepal.

**K.D.M.A. Research Award:**

*Please kindly submit your research work paper on allergy for trust award consideration by the end of October 2009 to KDMART office Chagal, G.P.O. Box 1885, Kathmandu, Nepal, Phone: 4270667 and Fax 4272694. This award was established by Dr. D.D. Joshi in 2049 B.S. on the memory of his wife, the late Mrs. Kaushilya Devi Joshi. The award includes a grant of NCRs. 10,001 with certificate.*

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