Nematicidal Activity of Lantana Camara L. for Control of Root-Knot Nematodes

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Abstract

Various concentrations of aqueous leaf extract of Lantana camara L. were assessed against second stage juveniles (J₂) of Meloidogyne spp. (Goeldi, 1982) for its nematicidal potency in vitro conditions. Study showed 50% concentration of Lantana camara leaf extract at 48 hrs of incubation period and above showed effective in immobilizing second stage of larvae (J₂) of Meloidogyne spp. The standard concentration ‘S’ (100%) of leaf extract was found to be highly nematostatic, 98.66% of nematode were found dead in 48 hrs. Similarly, 57.66% of nematode juveniles were found dead when applied 50% concentration in 48 hrs. Mean number of (J₂) dead at 100% concentration for three time period was statistically significant highest at 48 hrs. So far, 50% concentration in 48 hrs and above was appropriate for controlling the root-knot nematode which seems as an alternative to chemical pesticides.

Keywords: Concentration, Leaf extract, Meloidogyne, Nematostatic, Second stage juveniles (J₂)

Introduction

One of the most damaging groups of plant nematodes is the root-knot nematode (Meloidogyne spp.). It is an obligate root parasite of more than 200 plant species including vegetable, horticulture and woody plants (Hussey 1985). Root-knot nematode, Meloidogyne spp. (Goeldi, 1982) (Tylenchida: Heteroderidae) is a major plant-parasitic nematode species affecting the quantity and quality of the crop production in many annual and perennial crops. Infected plants show typical symptoms including root galling, stunting and nutrient deficiency, particularly nitrogen deficiency (Siddiqui et al. 2001). In Nepal, the root-knot nematode is considered as the major problem for many agriculture crops (Manandhar and Amatya 1988, Keshari 2004). Therefore, the control of root-knot nematodes is very important to enhance plant productivity. Since, the chemical pesticides cause hazard to the biodiversity, therefore, the use of botanical pesticides can be better option to control root-knot nematodes.

Materials and Methods

Study Area

The proposed study area for the research was Kirtipur Municipality of the Kathmandu district. Kirtipur is one of the recently urbanized cities of Kathmandu valley located to South-west of the central Kathmandu. It extends from 27° 41’ 36” – 27° 38’ 37” N to 85° 18’ 00” – 85° 14’ 64” E and has 1300 to 1402 meter of altitudinal range from sea level (Fig. 1).
Figure 1. Map of Nepal and study area.

Plant material

*Lantana camara* L. used for the experiment was collected from natural habitats of Kirtipur. The study was conducted in laboratory of Central Department of Biotechnology and Central Department of Zoology. The study was conducted from December 2012 to September 2013.

Preparation and storage of aqueous leaf extract

Fresh mature healthy leaves of *Lantana camara* were washed and air dried using an oven at 28°C. The dried leaves were ground using a Classic Maraja Electric Blinder. A 250 gm of fine powder was dissolved in 1000 ml sterilized DH₂O (1gm/4 ml basis). Aqueous suspension was allowed to soak on Stvart Orbital Shaker at room temperature for 24 hrs for the extraction of active ingredients and then passed through two folds of muslin cloth followed by filtration through Whatman no.1 filter paper. The filtrate obtained so far was centrifuged at 2400 rpm for 10 min and clear supernatant was stored at 4°C in a plastic container as standard solution ‘S’. Other different concentrations i.e. 10%, 50%, and 100% respectively were prepared by adding required amount of sterilized DH₂O for laboratory experiments.

Source of root-knot nematode (*Meloidogyne* spp.)

Root-knot nematode, *Meloidogyne* spp. was collected from the roots of heavily infected tomato plants. Cultures of *Meloidogyne* spp. were maintained on tomato roots in the greenhouse at the Central Department of Botany, Kirtipur. Egg masses of *Meloidogyne* spp. were hand-picked using sterilized forceps from heavily infected tomato roots, washed in distilled water and incubated at 28± 2°C for 24 hrs for hatching. The hatched juveniles were collected after placing the juvenile suspension through a coarse sieves (8 cm in diameter) containing tissue paper and kept in the petridish with water just deep enough to contact the tissue paper to collect second stage juveniles, so called *(J₂)*.

In vitro experiment

The aqueous leaf extract prepared as above was evaluated for nematicidal activity against second stage juvenile *(J₂)* of *Meloidogyne* spp. under laboratory conditions in order to assess the larval mortality. For
In this experiment, 100 freshly hatched J2 larvae were transferred to 2.5 cm in diameter plastic petridish containing 10 ml of different concentrations of leaf extract i.e. 10%, 50%, and 100% respectively. The petridish with 10 ml of distilled water (without plant extract) was considered as a control. All the petridishes were maintained at 25± 2°C in an incubator. After 12 hrs, 24 hrs and 48 hrs of incubation, mobile and immobile J2 larvae were counted under stereoscopic microscopic in order to record larval mortality. Immobilized larvae were confirmed by using needle as dead larvae failed to respond to stimulation with a needle. Each treatment was replicated three times.

**Statistical analysis**

On the basis of laboratory experiment, the data were recorded as larval mortality (dead or alive). All the data were analyzed according to analysis of variance (ANOVA) using SPSS 17.0 program.

**Results**

The percentage of mortality of juveniles of root-knot nematode (100 juveniles used for each experiment) differed with different concentration (S = 100%, S/2 = 50%, and S/10 = 10%) and duration of treatment (12 hrs, 24 hrs, and 48 hrs). Among all the treatment, 100% of leaf extract proved highly toxic to juveniles followed by 50% and 10%.

The percentage mortality of J2 of *Meloidogyne* spp. was 82.6%, 36.6% and 3.3 in 100%, 50%, 10% concentrations of leaf extract respectively as compared to control after 12 hrs. In other way, out of 100 J2 used in each experiment 17.4%, 63.4% and 96.7% larvae were remained active in 100%, 50% and 10% concentration of leaf extract of *L. camara* as compared to control. Among all the treatment, 100% of leaf extract proved highly toxic to juveniles followed by 50% and 10%. Similarly, the percentage mortality of J2 was 89.3%, 50.0% and 16.0% in 100%, 50%, 10% concentrations of leaf extract, respectively as compared to control after 24 hrs. In terms of mobilization of larvae, out of 100 larvae used in each experiment 10.7%, 50% and 84% larvae were remain active in 100%, 50% and 10% concentration of leaf extract. Likewise, the percentage mortality of J2 was 98.6%, 57.6 and 28.6% in 100%, 50%, 10% concentration respectively as compared to control after 48 hrs. In terms of mobilization of J2 larvae, out of 100 larvae, 1.4%, 42.4% and 71.4% larvae were found active in 100%, 50% and 10% concentration of leaf extract of *L. camara*. In general, percentage mortality was proportionally correlated with the concentrations and exposure periods of extract. The highest mortality (98.6%) was recorded in 100% concentration of leaf extract at 48 hrs of exposure period followed by 89.6% in 24 hrs and 82.6% in 12 hrs (Table 1).

**Table 1.** Effect of different concentration of plant extract of *L. camara* on larval mortality after 12, 24 and 48 hrs

<table>
<thead>
<tr>
<th>Plant</th>
<th>Incubation period (hrs)</th>
<th>% of mortality of nematode in different concentrations</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lantana camara</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12 hrs</td>
<td>S (100%)</td>
<td>S/2 (50%)</td>
</tr>
<tr>
<td>24 hrs</td>
<td>82.6</td>
<td>36.6</td>
</tr>
<tr>
<td>48 hrs</td>
<td>89.3</td>
<td>50.0</td>
</tr>
<tr>
<td></td>
<td>98.6</td>
<td>57.6</td>
</tr>
</tbody>
</table>

Each value is the mean of three replicates.

Number of second stage juveniles used = 100 (for each treatment)
Result in table 2 also showed that effect of various concentrations of leaf extracts of *Lantana camara* on larval mortality over exposure time i.e 12 hrs, 24 hrs and 48 hrs. In general, percentage of larval mortality increased with the increase in exposure period but decrease with dilutions. Comparison of treatment mean regarding period of leaf extract of *Lantana camara* indicated that test plant gave the maximum mortality at 100% concentration at 48 hrs of exposure showing each treatment significantly different than other. Mortality in control treatment was negligible.

**Table 2.** Mean number of larval mortality in different concentration at 12, 24 and 48 hrs

<table>
<thead>
<tr>
<th>Plant</th>
<th>Incubation period (hrs)</th>
<th>Concentrations</th>
<th>LSD (0.05)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lantana Camara</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12 hrs</td>
<td>S (100%)</td>
<td>S/2 (50%)</td>
<td>0.39</td>
</tr>
<tr>
<td></td>
<td>8.26⁺</td>
<td>3.67ᵇ</td>
<td></td>
</tr>
<tr>
<td>24 hrs</td>
<td></td>
<td>S/10 (10%)</td>
<td>0.53</td>
</tr>
<tr>
<td></td>
<td>8.93⁺</td>
<td>5.00ᵇ</td>
<td></td>
</tr>
<tr>
<td>48 hrs</td>
<td></td>
<td></td>
<td>0.45</td>
</tr>
<tr>
<td></td>
<td>9.86⁺</td>
<td>5.77ᵇ</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.86ᶜ</td>
<td></td>
</tr>
</tbody>
</table>

Degree of freedom (df) = 58  
T-value = 2.0017

**Discussion**

In the present studies, various concentrations of aqueous leaf extract of *L. camara* were assessed for nematicidal activity in the laboratory conditions. All the treatments exhibited natural nematicidal potential to varying degree. The result showed significant juvenile mortality potential of plant extract against *Meloidogyne* juveniles. The nematicidal effect of leaf extract on juvenile mortality of *Meloidogyne* spp. was concentration dependent i.e. the juvenile mortality decreases with increased extract concentration as the efficacy of the plant extract depends on the concentration and duration of exposure of juveniles to the extract (Mahmood et al. 1997). Among all the concentrations of leaf extract of *L. camara* tested, 100% concentration at 48 hrs was found effective in controlling *Meloidogyne* juveniles. This result agrees with the result obtained by (Akhtar and Mahmood 1994) who reported that water extracts from leaves and root of Mexican marigold and leaves of *Lantana* reduced the hatching of *M incognita* eggs significantly. The nematicidal activity of *L. camara* against juveniles of *Meloidogyne* spp. has also been reported by many authors (Begum et al. 2008, Qamar et al. 2005, Shaukat and Siddiqui 2001). The mortality of juveniles might be due to nematicidal chemicals present in the leaf extract as *L. camara* contains camaric acid and olenolic acids which may have larvicidal or ovicidal properties.

The findings of the present investigation are not conclusive. Further studies should be conducted in greenhouse and field conditions to assess the nematicidal activity. In comparison to the other countries, very limited work on nematicidal treatment is done in Nepal despite the fact that *Meloidogyne* spp. cause serious problem limiting the plant productivity of many crops. This work will hopefully fill the gap in this research.

**Conclusion**

It was found that 50% concentration of *L. camara* leaf extract at 48 hrs and above was found deleterious to root-knot nematode. This finding could be important from the point of view of controlling the root-knot nematode without the use of chemical pesticides in view of environmental pollution likely to cause. The control of *Meloidogyne* spp. by the leaf extracts used in this study might be probably based on a complex mode of action involving multiple mechanisms. Therefore, further studies are needed to characterize the active compounds in the test plant that are nematicidal and possessing complex modes of action.
Acknowledgements

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References


Prevalence of Pulmonary Tuberculosis in Jutpani VDC, Chitwan, Nepal

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Abstract

Tuberculosis (TB) is one of the major public health problems in the developing countries like Nepal. The present study was carried out in Jutpani VDC of Chitwan Nepal from July to December 2012 with the objective to determine the situation of Tuberculosis in Jutpani VDC, Chitwan. During the study period, a total of 600 sputum samples from 200 TB suspected patients were collected and examined in the Laboratory of Jutpani Primary Health Center (PHC). Diagnosis was made after staining the sputum smear by Z-N staining method and examined under light microscope. Out of 200 TB suspected patients, 18(9%) were found to be AFB positive. Among patients diagnosed as having pulmonary Tuberculosis (PTB), males (55.56%) were found to be infected more than females (44.44%) which was found to be statistically insignificant ($\chi^2$ cal. = 0.4524, P<0.01). Highest prevalence of TB infection (36.89%) was found to be in the age group of 30-40 years. A questionnaire survey of 57 Pulmonary Tuberculosis (PTB) patients was done to assess their knowledge, attitude and preventive practices for TB. Out of 57 PTB patients interviewed, majority of the patients had positive attitude but the knowledge regarding cause, transmission, prevention of the tuberculosis was not adequate.

Key words: AFB, Mycobacterium tuberculosis, PHC, PTB, Z-N stain

Introduction

Tuberculosis (TB) is the world's most serious public health problem particularly in developing and under developing countries. It is a disease of great antiquity and contributing to more morbidity and mortality than any other bacterial infection (Grange et al. 1998). It causes ill-health among millions of people each year and ranks as the second leading cause of death from an infectious disease worldwide, after the human immunodeficiency virus (HIV) (WHO 2012a). Approximately one-third of the world's population is infected from TB (Miller and Schieffelbein 1998). There were almost 9 million new cases and 1.4 million TB deaths (9,90,000 among HIV-negative people and 4,30,000 HIV-associated TB deaths) in 2011 (WHO 2012b). So, TB remains one of the deadliest threats to public health.

TB is an infectious disease caused by the bacillus Mycobacterium tuberculosis that spreads to others via aerosol route. TB typically affects the lungs (pulmonary TB) but can affect other sites as well (extra-pulmonary TB) (Kumar et al. 2007). It is estimated that 6 out of 10 adults in Nepal are infected with TB and 80,000 Nepalese people have active TB disease and every year, 40,000 people develop active TB, of whom 20,000 have infectious pulmonary disease (DoHS 2004/2005).
Materials and Methods

This research work was carried out in Jutpani VDC of Chitwan Nepal from July to December 2012. The study was designed to assess the situation of TB in the study area. A total of 200 TB suspected patients visiting Jutpani PHC with the clinical history of two or more week’s continuous cough, fever, and marked weight loss were included for prevalence study. Sputum samples of three consecutive days from total of peoples were collected and smear examined for Acid Fast bacilli (AFB) at the laboratory of Jutpani PHC and diagnosed after staining by Z-N method. A questionnaire survey of 57 smear positive TB patients was done to assess their Knowledge, Attitude and Preventive Practice (KAP) towards TB.

Results

Out of 200 TB suspected patients, 18(9%) were found smear positive. Out of 18(9%) smear positive patients, 10(55.56%) were male and 8(44.44%) were female and highest prevalence of smear positivity (38.89%) was found in the age group of 30-40. Table 1 shows the age and sex wise prevalence of PTB in Jutpani VDC. Out of 18(9%) smear positive cases, highest prevalence of tuberculosis was found in the ward number four (5/18) of the VDC which constitutes 27.78%, followed by the ward number one (4/18) which constitute 22.22% (Table 2). Among the 57 pulmonary TB patients interviewed, 46(80.7%) believed that blood in sputum, 41(71.9%) believed evening rise in fever, 16(28.1%) believed weakness and loss of weight and 12(21.1%) believed continue cough for more than 3 weeks were the symptoms of TB. Similarly, among 57 TB patients of Jutpani VDC, 47(82.5%) believed that TB is a communicable disease and majority of them, 30(52.6%) believed that TB can be prevented by avoiding the personal contact with the TB patient and few of them 25(43.9%) believed that It can be prevented from transmission by providing public awareness about the TB. Out of 57 patients, 41(71.9%) believed that consuming excessive alcohol is the cause of TB and 40(70.1%) believed that smoking is the cause TB. Only 10(17.5%) peoples knew *Mycobacterium tuberculosis* is the causative organism of TB. Similarly, majority of the respondent 41(71.9%) agree that TB can be prevented but only 2(3.5%) strongly disagree. More than half of the respondents 33(57.6%) agreed to the statement that if one family member is infected, other family members will also develop TB.

Table 1. Age and sex wise prevalence of PTB

<table>
<thead>
<tr>
<th>Age in years</th>
<th>Male Patients</th>
<th>Female Patients</th>
<th>Total slide number</th>
<th>Slide Positivity Rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total slide</td>
<td>Positive slides</td>
<td>Total slide</td>
<td>Positive slides</td>
</tr>
<tr>
<td>&lt;10</td>
<td>4</td>
<td>0</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>10-20</td>
<td>17</td>
<td>2</td>
<td>17</td>
<td>0</td>
</tr>
<tr>
<td>20-30</td>
<td>13</td>
<td>1</td>
<td>27</td>
<td>3</td>
</tr>
<tr>
<td>30-40</td>
<td>13</td>
<td>3</td>
<td>23</td>
<td>4</td>
</tr>
<tr>
<td>40-50</td>
<td>12</td>
<td>1</td>
<td>13</td>
<td>0</td>
</tr>
<tr>
<td>50-60</td>
<td>20</td>
<td>1</td>
<td>14</td>
<td>1</td>
</tr>
<tr>
<td>60-70</td>
<td>9</td>
<td>1</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>70-80</td>
<td>4</td>
<td>0</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>80-90</td>
<td>4</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>96</td>
<td>10</td>
<td>104</td>
<td>8</td>
</tr>
</tbody>
</table>

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Table 2. Ward and Sex wise prevalence of total TB positive patients

<table>
<thead>
<tr>
<th>Ward No.</th>
<th>Male Patients</th>
<th>Female patients</th>
<th>Total slide number</th>
<th>Total positive slides</th>
<th>Slide positivity Rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total slide</td>
<td>Positive slides</td>
<td>Total slide</td>
<td>Positive slides</td>
<td></td>
</tr>
<tr>
<td>One</td>
<td>17</td>
<td>2</td>
<td>24</td>
<td>2</td>
<td>41</td>
</tr>
<tr>
<td>Two</td>
<td>6</td>
<td>0</td>
<td>6</td>
<td>0</td>
<td>12</td>
</tr>
<tr>
<td>Three</td>
<td>8</td>
<td>1</td>
<td>4</td>
<td>0</td>
<td>12</td>
</tr>
<tr>
<td>Four</td>
<td>28</td>
<td>3</td>
<td>34</td>
<td>2</td>
<td>62</td>
</tr>
<tr>
<td>Five</td>
<td>4</td>
<td>1</td>
<td>7</td>
<td>1</td>
<td>11</td>
</tr>
<tr>
<td>Six</td>
<td>1</td>
<td>0</td>
<td>4</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Seven</td>
<td>2</td>
<td>0</td>
<td>4</td>
<td>1</td>
<td>6</td>
</tr>
<tr>
<td>Eight</td>
<td>19</td>
<td>2</td>
<td>14</td>
<td>1</td>
<td>33</td>
</tr>
<tr>
<td>Nine</td>
<td>11</td>
<td>1</td>
<td>7</td>
<td>0</td>
<td>18</td>
</tr>
<tr>
<td>Total</td>
<td>10</td>
<td>8</td>
<td>200</td>
<td>18</td>
<td>9%</td>
</tr>
</tbody>
</table>

Discussion and Conclusion

Out of 200 TB suspected patients visiting the Jutpani PHC, 18(9%) samples were found to be positive for acid fast bacilli (AFB). Age wise observation of the smear positivity showed that the highest prevalence of TB 38.89% (7/18) was found to be in the age group of 30-40. A similar study conducted by Dhungana 2002 at United Mission Hospital Tansen (UMHT) and Dhungana 2004 in TUTH had found the highest prevalence of TB among age group of 20-30 years. The highest prevalence of TB among this age group might be due to the exposure of young people to different environment during their work and activities that will make their health more prone to infection by TB organisms. Highest prevalence of TB in the age group 30-40 is because people of this age group are exposed to the outer environment as well as due to high work load and wide range of mobility. So people of this age group are more prone to the infection with TB organisms.

Sex wise prevalence of TB showed that out of 18 smear positive cases, 10(55.54%) male and 8(44.46%) female. Similar higher prevalence of tuberculosis in males compared to females has been reported by other earlier studies carried out in Gorkha district (Smith et al. 1994), in histopathological studies at TUTH (Shrestha 1989). Present study revealed high prevalence of TB in the month of Asar 33.33% (6/18) followed by Shrawan and Bhadra which had the similar prevalence of 22.22% (4/18). The reason of high prevalence of TB in Asar might be due to high incidence of cough and cold which may help in the transmission of PTB during coughing in the month due to seasonal change. Moreover a study done in Chitwan by Dhital (2007) in CMCTH found that Asar, Shrawan and Bhadra had more incidence of upper respiratory tract infection than other months of the year which support the finding of the present study. Present study revealed that the highest prevalence of TB among illiterate people (28.1%). Similarly highest prevalence of PTB was found among farmers (49.1%) followed by people in service (22.80%) in relation to the occupation. This showed that occupation of the individuals also plays a major role in the prevalence of TB. Lifson et al. (1999) stated that the risk of TB is greater in areas of residence characterized by crowding, poverty and lower education. In another study, National Tuberculosis Center

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also found that majorities of the respondents (67.8%) were employed in the agriculture sector – as farmers (NTC 2009). Among the 57 TB patients interviewed, majority of them, 30(52.6%) believed that TB can be prevented by avoiding the personal contact with the TB patients. About 40% of the respondents in Sindhupalchowk, one of the hilly district believed that the infection could be prevented by covering mouth and nose while coughing and sneezing (NTC 2009). But present study showed that only 7(12.3%) believe on personal protection like using mask could prevent TB transmission. It showed that the knowledge about TB prevention is poor among the TB patients of this VDC as compared to the people of Sidhupalchowk district.

Similarly, 41(71.9%) believed that consuming excessive alcohol was the cause of TB and 40(70.1%) believed that smoking was the cause TB. Only 10(17.5%) peoples knew Mycobacterium tuberculosis is the causative organism of TB in this VDC. Similar study conducted in Rajasthan showed only 1.6% had knowledge of tuberculosis causative agent (Yadav et al. 2006) which is very less than the present study where 26.3 % have the knowledge about the causative organism of TB as Mycobacterium tuberculosis. Present study revealed that 94.7% (54) patients under DOTS said that they had general knowledge about TB. But study carried out in Gorkha as reported earlier, 88% (Sharma 2008) and in Patan Hospital 75.86% (Joshi 2004) indicated impressive knowledge of TB as maximum participants knew TB as the infectious diseases. This showed that people in Chitwan have comparatively poor knowledge about the infectious nature of TB. This may be one of the main reasons of high prevalence of TB among the study population.

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Population status of Spotted Deer in Baghmara Buffer Zone Community Forest, Chitwan National Park, Nepal

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Abstract

The population status of spotted deer was studied in Baghmara Buffer Zone Community Forest (BBZCF) during different four seasons of 2012-13. Population status was determined by direct count method at the time of its active movement in the morning and early evening. A total of 255 individuals of spotted deer were counted with a crude density of 118.6 deer/km². Male to female sex ratio varied seasonally (summer-1: 2.2, rainy-1: 2.22, autumn-1: 2.15 and in winter-1: 2.50). The average sex ratio of males to females was computed as 44.47:100. The largest herd of 41 individuals was observed in summer. The average herd size was 13.49. We used Kruskal-Wallis Rank Sum test to find the significant difference in mean population distribution due to seasons and blocks. There was no significant difference in mean population distribution due to seasons ($\chi^2=0.7624$, df=3, $P=0.8584$) but significant differences was found in mean population distribution due to blocks ($\chi^2=52.7534$, df=13, $P=0.000000097$).

Key words: BBZCF, direct count, herd, mean population, sex ratio

Introduction

The spotted deer or Chital (Axis axis) is one of the most beautiful deer of Nepal and a favourite object of sport because of its lovely spotted coat (Shrestha 2003). Spotted deer is classified as a member of order Artiodactyla under the family Cervidae (Mitchell 1982). Ellerman and Morrison-Scott (1964) recognize two sub species of Axis axis. Axis axis axis is native to Nepal, India and Srilanka and Axis axis ceylonensis is native to Srilanka.

The spotted deer is a medium sized deer standing about 35-38 inches at the shoulder and the coat is rufous brown with white spots (Schaller 1967). Spotted deer appears like a small elk or wapiti in outline. Does are slightly lighter in color than bucks especially over the face and neck (Graf and Nichols 1966). The average weight of males is 71 kg and the female is 50 kg in Chitwan National Park (CNP) (Mishra 1982). Only males have antlers and are functional during the mating season (Rajaram 2004).

Axis deer are found at lower elevations below ~915 m (Ables 1977) and rarely found above an altitude of 1000 m in its native Asian habitat, including Nepal. They usually prefer ecotonal region between the forests and the grass patch (Mitra 1990). It is widely distributed cervids in the Indian sub-continent (Srinivasulu 2001). In Nepal, spotted deer occur throughout the Siwalik Hills and adjoining plains. Spotted deer preferred habitat is Tarai plain while may reach to the basin of inner mountains. They are found scattered in lower number in the mid-hill, warmer valleys and forest areas (Chalise 2001, 2013).
It is distributed in Bardia National Park (BNP), CNP, Sukla-Phanta Wildlife Reserve (SWR), Parsa Wildlife Reserve (PWR) and Koshi Tappu Wildlife Reserve (Majupuria and Majupuria 2006). Spotted deer is considered as least concern as per the IUCN categories as it occurs over a very wide range with large populations (Chalise 2013, Duckworth et al. 2008).

**Materials and Methods**

This study was carried out in Baghmara Buffer Zone Community Forest, BBZCF (Location 27°34.78’-27°35.53’ Northern Latitude and 84°28.43’-84°29.40’ Eastern Longitude), Buffer Zone area of CNP, Chitwan, Nepal. The forest covers 215 ha area at 170 meter elevation (Sharma et al. 2011).

Population status was determined by direct count method assisted by binocular of 10x50 mm and age sex categorized. Hence to reduce biasness, total population was estimated by using statistical theorem. Observation was conducted concentrated mostly at the time of its active movement in the morning (6.00–9.00) and early evening (15.00–18.00). Counting was done in four seasons—Summer (April), Rainy (July), Autumn (November) of 2012 and winter (February) of 2013. The number of Spotted Deer was confirmed by repeated count in one observation session. During the total count, the exact total numbers were different at the same place in different replicates. The whole study area was divided into 14 micro habitats. The number of spotted deer was confirmed by repeated observation in one observation session. During the total count, the exact total numbers were different at the same place in different replicates. Hence to reduce biasness, estimated population was calculated by \(N = 2n_k - n_{k-1}\) formula and count was derived from first and second maximum counting such as:

Here, \(N = 2n_k - n_{k-1}\)……………………………………

Where,
\[
N = \text{estimated population.}
\]
\[
n_k = \text{highest Value of observed Population}
\]
\[
n_{k-1} = \text{second highest value of observed Population.}
\]

The approximate upper confidence limit \((N_u)\) at 100(1-\(a\)) % is obtained by;
\[
N_u = n_k + (1-a)/a (n_k - n_{k-1})……………………………………2
\]

Where,
\[
N_u = \text{upper confidence limit of population}
\]
\[
a = \text{the level of test}
\]

Similarly at given confidence limit the lower limit of population is given by
\[
N_L = N_k
\]

Hence, the range of total number \(N\) is given by
\[
N_L < N < N_U
\]

**Data Analysis**

For direct count method

R – Software (R Console Version 2.15.2) was used for the statistical analysis. It is a non parametric test and alternative to ANOVA. Since the data was not found normal, hence this test was performed. Kruskal–
Wallis Rank Sum Test was used to find the significant difference in the mean population distribution of spotted deer in different habitat due to four seasons and in different seasons due to blocks.

**Results**

There were five different micro-habitats identified inside BBZCF. Habitat of Spotted deer was explored by using ecological quadrate method. Within each transect four quadrates of 25x25 m² for tree species, 5x5 m² for shrub species and 1x1 m² for herbs were made (Sharma et al. 2011). There were altogether 55 transects within 14 blocks. These micro-habitats were further divided into 14 blocks to make the study easier. Ten of the 14 blocks were used by deer frequently and taken for population counting (Table 1). The total number of spotted deer counted was 255 of different age and sex. Among 255 individuals of spotted deer, 137 (53.73%) were females, 61 (23.92%) were males and 57 (22.35%) were young. The area of BBZCF is only of 2.15 km². So the crude density is estimated to be 118.6 deer/km².

**Table 1. Population status of spotted deer in different blocks of BBZCF in 2012-13.**

<table>
<thead>
<tr>
<th>Habitat used</th>
<th>Blocks</th>
<th>Population of spotted deer observed</th>
<th>Estimated population N= 2n_k – n_{k-1}</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Maximum (n_k)</td>
<td>Second Maximum (n_{k-1})</td>
</tr>
<tr>
<td>Bhellar-Padle (Trewia nudiflora Albizia lucidior) forest</td>
<td>A</td>
<td>25</td>
<td>23</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>13</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>H</td>
<td>28</td>
<td>24</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>7</td>
<td>6</td>
</tr>
<tr>
<td>Bhellar forest (Trewia nudiflora)</td>
<td>E</td>
<td>27</td>
<td>26</td>
</tr>
<tr>
<td></td>
<td>L</td>
<td>38</td>
<td>36</td>
</tr>
<tr>
<td></td>
<td>M</td>
<td>8</td>
<td>6</td>
</tr>
<tr>
<td>Padke (Albizia lucidior) forest</td>
<td>K</td>
<td>45</td>
<td>42</td>
</tr>
<tr>
<td>Sisso (Dalbergia sissoo) forest</td>
<td>B</td>
<td>7</td>
<td>5</td>
</tr>
<tr>
<td>Grassland</td>
<td>J</td>
<td>57</td>
<td>56</td>
</tr>
<tr>
<td><strong>Total counted</strong></td>
<td></td>
<td>255</td>
<td>234</td>
</tr>
</tbody>
</table>

In Bhellar-Padle forest, at 95% and 99% confidence level, the upper limits are as highest as 104 and 424 respectively in block Hand the least as 26 and 106 respectively in block N (Table 2). At 90% confidence level, the upper limit is highest as 72 in Padke forest (block K). Similarly, the lower limits are found to be highest as 57 in grassland (block J) and least as 7 in Sisso forest (block B) and Bhellar-Padle forest (block N).

The census in different seasons varied for the spotted deer in BBZCF. The total number of spotted deer counted was 237 in summer season. Out of them, 58 (24.47%) were males, 129 (54.43%) were females and 50 (21.09%) were young. The male to female sex ratio was 1: 2.2 and the Young to female sex ratio was 1: 2.58.

The total number of spotted deer counted was 203 in rainy season. Out of them, 49 (24.13%) were males, 109 (53.69%) were females and 45 (22.16%) were young (Table 3). The male to female sex ratio was to be 1: 2.22 and the young to female sex ratio was 1: 2.4.
The total number of spotted deer counted was 226 in Autumn season. Out of them, 54 (23.89%) were males, 114 (50.44%) were females and 58 (25.66%) were young. The male to female sex ratio was 1: 2.15 and the young to female sex ratio was computed to be 1: 1.96.

**Table 2.** Range of population of spotted deer in different blocks in BBZCF in 2012-13

<table>
<thead>
<tr>
<th>Habitat</th>
<th>Blocks</th>
<th>Nᵢ = nᵢ</th>
<th>Range of Estimated Population (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Levels of Test</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.10 (90%)</td>
</tr>
<tr>
<td>Bhellar-Padke (Trewia nudiflora Albizia lucidior) forest</td>
<td>A</td>
<td>25</td>
<td>25 &lt; N &lt; 43</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>13</td>
<td>13 &lt; N &lt; 40</td>
</tr>
<tr>
<td></td>
<td>H</td>
<td>28</td>
<td>28 &lt; N &lt; 64</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>7</td>
<td>7 &lt; N &lt; 16</td>
</tr>
<tr>
<td>Bhellar forest (Trewia nudiflora)</td>
<td>E</td>
<td>27</td>
<td>27 &lt; N &lt; 36</td>
</tr>
<tr>
<td></td>
<td>L</td>
<td>38</td>
<td>38 &lt; N &lt; 56</td>
</tr>
<tr>
<td></td>
<td>M</td>
<td>8</td>
<td>8 &lt; N &lt; 26</td>
</tr>
<tr>
<td>Padke (Albizia lucidior) forest</td>
<td>K</td>
<td>45</td>
<td>45 &lt; N &lt; 72</td>
</tr>
<tr>
<td>Sisso forest</td>
<td>B</td>
<td>7</td>
<td>7 &lt; N &lt; 25</td>
</tr>
<tr>
<td>Grassland</td>
<td>J</td>
<td>57</td>
<td>57 &lt; N &lt; 66</td>
</tr>
</tbody>
</table>

The total number of spotted deer counted was 232 in winter season. Out of them, 51 (21.98%) were males, 128 (55.17%) were females, 53 (22.84%) were young. The male to female sex ratio was 1: 2.50 and the young to female sex ratio was 1: 2.41.

**Table 3.** Number of spotted deer counted in different seasons in BBZCF

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Seasons of year</th>
<th>Young</th>
<th>Males</th>
<th>Females</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Summer</td>
<td>50</td>
<td>58</td>
<td>129</td>
<td>237</td>
</tr>
<tr>
<td>2.</td>
<td>Rainy</td>
<td>45</td>
<td>49</td>
<td>109</td>
<td>203</td>
</tr>
<tr>
<td>3.</td>
<td>Autumn</td>
<td>58</td>
<td>54</td>
<td>114</td>
<td>226</td>
</tr>
<tr>
<td>4.</td>
<td>Winter</td>
<td>53</td>
<td>51</td>
<td>128</td>
<td>232</td>
</tr>
<tr>
<td></td>
<td>Average Total in a year</td>
<td>51.5</td>
<td>53.0</td>
<td>120</td>
<td>224.5</td>
</tr>
</tbody>
</table>

The average herd size was 13.49 individuals computed from 3,832 individuals in 284 herds during the study period. The largest herd size of 41 individuals (mixed herd) was observed in habitat J. There was no significant difference in mean population distribution due to seasons (P-value > 0.05) but significant differences was found in mean population distribution due to habitat (P-value < 0.05) (Table 4).

**Table 4.** Kruskal Wallis test of different seasons and habitat

<table>
<thead>
<tr>
<th>Variables</th>
<th>X² – value</th>
<th>d.f</th>
<th>P-value</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seasons</td>
<td>0.7624</td>
<td>3</td>
<td>0.8584</td>
<td>No Significant</td>
</tr>
<tr>
<td>Habitat</td>
<td>52.7534</td>
<td>13</td>
<td>0.000000997</td>
<td>Significant</td>
</tr>
</tbody>
</table>
Discussion

Sharma et al. (2012) recorded a total of 182 individuals of spotted deer in BBZCF but we recorded 255 individuals of spotted deer in BBZCF. Seidensticker (1976) recorded 629 individuals of spotted deer in CNP with density of 17.7 deer/km². This study computed crude density of 118.6 deer/km². Raman et al. (1996) recorded density of 212.3 deer/km² in Guindy National Park, India. Spotted Deer prefers ecotonal region between the forests and the grass patch and avoid hilly terrain (Mitra 1990). Due to the availability of such type of habitat and resources in BBZCF, the difference in the study time, multiple observations, direct count method, small study area were possible reasons for higher population of spotted deer and high density estimate during this study.

Bedi (1985) counted about 600 spotted deer in a single herd in Corbett National Park, Kurt (1990) explained occurrence of 800 or more members of spotted deer in a herd in India and Sri Lanka. However, such large herds were not observed in this study area. Commonly, herds of 4 to 18 individuals were observed in this study and the largest herd of 41 individuals. The mean size of mixed herd was 13.49. Danger from the predators and the climatic factors could be the possible reasons behind their presence in small herds. The sex ratio of male to female was 0.59:1 in Sri Lanka (De Silva and De Silva 2001). Seidensticker (1976) found sex ratio as 115:100 in CNP. The average sex ratio of male to female was found to be 44.47:100 in BBZCF (seasonal basis). The sex ratio is found to be low in comparison to Sri Lanka and CNP. Illegal and selective hunting, unequal natality and mortality rates of male young etc may be the possible factors for the unequal sex ratio.

Conclusion

A total of 255 individuals of spotted deer were counted from different habitat and the crude density was computed to be 118.6 deer/km². The highest population of spotted deer was counted in grassland (block J) and the lowest in sissoo dominated forest (block B). The average sex ratio was found to be 44.47 bucks to 100 does with more females than males in all the seasons. The highest sex ratio of 47.39 bucks to 100 does was found in Autumn season. The largest herd of 41 individuals was observed in grassland (block J) in summer. The average mixed herd size was computed as 13.49 individuals. Kruskal-Wallis test revealed that there was no significant difference (P>0.05) in the mean population distribution of deer in different habitat due to four seasons and there was a significant difference (P<0.05) in the mean population distribution of deer in different seasons due to different habitat.

Acknowledgements

We would like to thank Central Department of Zoology, TU, for approval of research, DNPWC and BBZCF for granting the permission to carry out this research work in Baghmara. We would like to express our sincere thanks to Mr Hari B Acharya, Mr Manoj Choudhary Mr Chandra Lama, and all the staff of BBZCF for their valuable information, continuous support, encouragements and suggestions during the research.

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Pesticide Use Practices Among Tomato Growers in Kavre District, Nepal

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Abstract
Common practices to manage the pests of tomato were explored in three VDCs (Panchkhal, Nala and Mahadevsthan) of Kavre District from January to April 2011. The data were collected by direct observation and also by using semi-structured questionnaires. Altogether 48 tomato farms were observed during the study period that included 16 farms in each VDC. Farmers were found to be relied on pesticides from bedding of plant till harvesting to control pests. Farmers were found positive towards the pesticide use despite their familiarity on ill-effects of pesticides. Personal safety measures during application of pesticides have not been followed. Scanty knowledge, labor intensiveness and lack of coordination between the farmers were the major factors which prevented the adoption of eco-friendly alternative pest management methods.

Key words: Aphis, Bemisia, Helicoverpa, IPM, White grub

Introduction
The tomato (Lycopersicon esculentum Mill.) is herbaceous fruiting plant that survives in diverse environmental conditions (Rice et al. 1987). Tomato fruit is considered to be fairly rich in vitamins of high cash value and used in our daily life for various purposes but farmers are using pesticides to kill pests and diseases which attack them for better production (Knezevic and Serdar 2008). Over 1000 compounds are applied to agricultural crops in order to control objectionable moulds, insects and weeds (Ortelli et al. 2006). Pesticides' striking effort in preventing crop loss and controlling vectors of diseases have led to their acceptance and expanded use throughout the world (Sharpdan and Peter 2005). However, the potent chemicals for killing pests have elevated anxiety that they are agents of human diseases and environmental pollution. It has been observed that their long term, low dose exposure is increasingly linked to human health effects such as immune-suppression, hormone disruption, diminished intelligence, reproductive abnormalities and cancer (Wiles et al. 1998). Pesticide residues in food are global problems (Abinash and Singh 2009). According to WHO, developing countries use about twenty-five percent of the pesticides in the world and the use is in increasing trend (WHO 2003). This intrinsically dangerous technology is being promoted in a setting without technical and human resources to control it properly.

Agriculture is one of the most prevalent types of employment in the world. Nearly 50 percent of the world labor is employed in agriculture and they carry significant risk for development of pesticide risk (Das et al. 2001). Chemical pesticides for the first time were commenced in Nepal in 1952 when Paris green, Gammexane and Nicotine sulphate were imported from USA solely for malaria control program (Koirala et al. 2009). The sequential order of different groups of pesticides introduced in Nepal is: 1950s – organochlorines; 1960s – organophosphates; 1970s – carbamates; 1980s – synthetic pyrethroids (Manandhar 2007).
There is no clear drift of import and use of pesticides. Also there is possibility of an open border trading of commonly used pesticides and some of the banned pesticides such as DDT and BHC. It is somewhat difficult to document the amount of illegal trading and thus the size of such trading has not been reflected in the public data so far. In recent years, there are different pesticides used erratically, which is pretty common in Nepal. Endosulfan is used in ponds, streams and rivers for killing fishes and also used to attain polished appearance in vegetables such as tomato, brinjal, mustard leaves (Manandhar 2007).

Nepal has a marvelous opportunity for producing tomato as diverse agro-eco-zone favors both season and off-season varieties and farmers are encouraged to produce, so production and productivity has been increasing significantly for the last decade. Nepal has potential for fresh vegetable (tomato) and processed products in the international market as well. Reports suggested that use of pesticides in vegetables in Nepal is higher than other food products (Koirala et al. 2009). Therefore, this study aims to know the current situation of pesticide used in tomato and to deal the knowledge, practice among tomato growers and use of pesticides in Kavre district, Nepal.

Materials and Methods

Insect pests of tomato and their management practices were explored in three VDCs of Kavre District (Panchkhal, Nala and Mahadevsthan). These are major tomato production areas in the district so were chosen for this study which was conducted from January to April 2011. The data were collected by direct observation and also by using semi-structured questionnaires. Altogether 48 tomato farms were observed during the study that included 16 farms in each VDC. Tomato field was visited for direct observation of the plants and insect pests. The information regarding pesticide use was collected from farmers, agro-vet dealers, government agencies and related associations.

Results

Opinion of farmers on application of pesticide in the field

Majority of farmers (79.2%) of the study area had positive opinion towards the use of pesticide. The farmers’ views on the increased use of pesticides in the field include proper and safe for health and environment, increases crop production and reduces loss due to pest damage. They further mentioned that the knowledge, awareness and training about pesticides is a must. About 20.8% farmers were with negative opinion on the use of pesticides. They mentioned that the use of pesticides must be decreased in the field and biofriendly alternatives must be implemented to prevent health and environmental hazard.

Figure 1. Opinion of farmers of using pesticides in the field

<table>
<thead>
<tr>
<th>Opinion of farmers of using pesticides in the field</th>
</tr>
</thead>
<tbody>
<tr>
<td>Should be Increased: 20.80%</td>
</tr>
<tr>
<td>Should be Increased: 79.20%</td>
</tr>
</tbody>
</table>
Use of Pesticides for Pest Control

A total of 48 farmers and their fields were surveyed during the study. All of respondents (100%) were reported to use pesticides in tomatoes as pest control measure. It indicates that there is a high frequency of pesticides use in the vegetables which pose higher risk to vegetable growers and consumers.

Table 1. Pesticide users among tomato growers

<table>
<thead>
<tr>
<th>Pesticide user among tomato growers</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes- use pesticides</td>
<td>100%</td>
</tr>
<tr>
<td>No- do not use pesticides</td>
<td>0%</td>
</tr>
</tbody>
</table>

Knowledge and practices regarding use of pesticides

During study 80% farmers were found to use pesticides to increase productivity, 18.5% of them used pesticides to kill pest and 1.5% used to improve color. About 24.8% determine amount of pesticides to be used according to the information given in the label, 60.2% determine through retailer where they purchase the pesticide and 15% determine with their local agricultural expert. About 95.8% farmers bought pesticides from authorized shop or retailers whereas 4.2% from non-authorized shops.

Reasons of using Pesticides in the tomato field

- To kill pest: 19%
- To improve color: 2%
- To increase productivity: 80%

Safety precautions, personal protective equipment and personal hygiene during application of pesticides

Majority of respondent farmers (91.6%) read the label in the bottle or package where as 8.3% didn’t read the label. About 70.8% mixed the pesticides with hands wearing hand gloves, 4.2% mixed with bare hands and 25% mixed with stick and wearing hand gloves. During study, we found that none of them eat, drink or smoke while spraying pesticides in the field. Most of them, 50% washed the pesticide bottle or pesticide sprayer in the river, pond or well, whereas 45.8% washed in a distant place far from pond, river or well and 4.2% wash in tap at home. None of them display a signboard or red flag or empty bottles in the sprayed area after an application in order to inform others and also none of them keep the pesticides in the same place where they keep their food.
Table 2. Use of Personal Protective Equipments

<table>
<thead>
<tr>
<th>Items</th>
<th>Yes (%)</th>
<th>No (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shoes</td>
<td>100%</td>
<td>-</td>
</tr>
<tr>
<td>Hat/Head cover</td>
<td>33.4%</td>
<td>66.6%</td>
</tr>
<tr>
<td>Glass</td>
<td>4.2%</td>
<td>95.8%</td>
</tr>
<tr>
<td>Full sleeves shirt/trouser</td>
<td>100%</td>
<td>-</td>
</tr>
<tr>
<td>Gloves</td>
<td>4.2%</td>
<td>95.8%</td>
</tr>
<tr>
<td>Mask</td>
<td>-</td>
<td>100%</td>
</tr>
<tr>
<td>Change the clothes</td>
<td>100%</td>
<td>-</td>
</tr>
<tr>
<td>Take a bath</td>
<td>25%</td>
<td>75%</td>
</tr>
<tr>
<td>Clean hands and foots</td>
<td>75%</td>
<td>25%</td>
</tr>
</tbody>
</table>

Disposal techniques

About 58.3% of the farmers disposed the pesticide bottles or packets by throwing in the field, 29.2% bury in the field and 12.5% disposed in other ways like burning or use for household purpose after washing or throw them in to the dumping sites. If pesticide was spilled in some places, all of them were found that they clean the places with water only but they did not use any cleaning agent. During the application of pesticides, 87.5% had experience of pesticide spoil in their body parts and 12.5% had no such experience.

Health Problems due to use of pesticides

All the farmers (100%) knew about the adverse effects of pesticides to human health. Out of them 75% knew that pesticide use can cause short term health effect and only 25% knew that pesticides can also cause long term health effects. The different types of health hazards experienced by the farmers due to the use of pesticides were eye irritation, vomiting, skin irritation, dizziness, headache, diarrhea and shortness to breathe. The symptoms of health effects they suffered due to the use or exposure of pesticides were determined by the health worker (37.5%), self by the farmers (50%) and neighbor (12.5%) but not by the doctor.

Knowledge about Integrated Pest Management (IPM)

During the study, 96% farmers were found to be familiar with IPM where as 4% of them were unfamiliar. The reasons behind not using the IPM techniques despite their familiarity include pesticides are cheaper (62.5%), use of IPM is highly labor intensive (98.4%), not practiced by neighbor (12.5%) and lack of IPM training (16.6%).
Discussion

As the farming of tomato were done totally by the use of pesticides. Farmers used pesticides to increase productivity and to kill the pests so that more profit and high yield can be achieved. Even though they bought the pesticides directly from shop or agro-vet dealer and used that according to the label in containers or according to shopkeeper. Lack of knowledge about pesticides, its composition and its formulation made more misuse of pesticides. Palikhe (2002) documented that misuse and overuse of pesticides, particularly among commercial farmers, posed a health risk to the public and have numerous cases caused serious poisoning. The illegitimate used was due to unawareness of toxicity, aggressive marketing by dealers and profit interests. Many farmers did not understand the instructions written on the pesticide labels. The harmful effects of pesticides have been experienced by farmers and their families. There was a higher risk of presence of pesticides residue in vegetables which poses higher health risk to vegetable growers as well as consumers.

The adoption of safety measures during and after pesticide application was very important factor for preventing against harmful impacts of pesticide. The various safety options could be used as gloves, masks, long sleeved cloth, glass, long boots etc. They did not like to use safety measure as they thought that they disturb their work and uncomfortable to work in the field. For example, they thought that wearing a mask makes breathing difficult and they didn't have habit of wearing (Palikhe 2002). This might put them in the risk of pesticides that the health of these people was not secured of the disease caused by the inhalation, ingestion and absorption via skin (Karmacharya 2012). Personal protective devices were also found used less because of lack of awareness, unavailable and feeling discomfort by using them. From the field study, as expressed by farmers themselves, it was difficult for them to change their behavior in which they were adopted. Due to unsafe practices, farmers were more vulnerable to expose with toxic pesticides and were in higher health risks as there has been too much use of pesticides with too little or no protection. This result is also supported by Shrestha (2001).

Farmers knew about health effects and they suffered by different kind of symptoms. Most of the farmers knew about short term effects of pesticides where as only some knew about long term effects of pesticides (Shrestha et al. 2010). The different health problems they suffered were eye irritation, skin irritation, dizziness, shortness to breathe, vomiting, headache and diarrhea. They even diagnosed their health effects direct by themselves or by their neighbors but very few by the help of health workers. They rarely
consulted health personnel and took treatment in health centers, and they used their home treatment by themselves, which was comparable with the research work carried out by Koirala et al. (2009).

Pesticides application in the field is not only the solution of pests control and high yield of crops besides these farmers need awareness programs, training and proper guidance for application of pesticides. Other alternative methods of pests management is the demand of today. Shrestha (2001) reported that overuse of synthetic pesticides has also resulted in pest resistance to pesticides, resurgence of pests, elimination of natural enemies and disruption of ecosystems. Although the agricultural policies during the last few decades promoting higher input of chemicals have resulted in higher yields and more food, they have also resulted in poisoning, health related poverty and environmental degradation. There is, therefore, a need for alternative pest control measures for both commercial farmers currently overusing pesticides and food insecure subsistence farmers living at the mercy of pests. A healthy, effective and lasting mechanism for plant protection is required for food security, food safety, poverty reduction and rural development (Shrestha et al. 2010).

Acknowledgements

Authors are grateful to Mr Pramod Koirala, Senior Food Research officer, Department of Food Technology and Quality Control for his support and suggestions.

References


Study of Ornamental Fish in Lalitpur, Nepal

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Abstract

This work has been done from 2068 to 2069. The main theme of the survey was to find out the Aquarium fish interest among people, highest and lowest price of different fishes, feeding criteria, socio economic condition of aquarium fish trade. Survey points out that few shops are in Lalitpur. Gold fish varieties are famous among the people. Only artificial feed was given to the fishes. Increasing number of shops and satisfactory response regarding this business shows better socio economic condition regarding this sector.

Key words: Bio filter, Guppy, Parrot fish, Pearl scale, Sucker head

Introduction

Keeping a home aquarium has now become a very popular past time all over the world. It is a hobby and it appeals to young as well as old. It is very exiting to watch gentle and colourful fishes. Aquarium also adds fascinating colors to a drawing room. Besides, maintaining aquarium has also opened a flourishing business in all cities. Together all countries of the European Union are the largest market for ornamental fishes; however, the United States (US) is the single largest importer of ornamental fish in the world (Chapman 2000). Pet industry surveys have estimated the aquarium industry worth over Dollar 1,000 million USD (Cato and Brown 2003)

In cold water of mountain the Cyprinids, Schizothorax and Schizothoraichthys dominate. In high hill and hilly region they are joined by Tor, Neolissochelius, Barilius, Glyptothorax, Botia and Clupisoma (Rajbansi 1976). Zebra fish, Medaka and Xiphophorus fishes are grouped as aquarium fishes (Yang, Chen and Lan 1994).

Shop imports large varieties of ornamental fish and the other requirement like heater, filter water vibrator, medicines, fish food, purifier, net, aquarium cleaner decorating materials such as coloured stones, gravel, corals, wood, rocks and aquarium plants from India, Thailand, Hongkong and Srilanka (Amatya and Gurung 2005).

Materials and Methods

Weekly visits were made up to two months at initial period followed frequent visits later in different ornamental fish shops at Lalitpur to collect information about the fish marketing system, types of fishes available; their cost price, sizes of aquarium used, source of aquarium fish supply, socio economic condition aquarium fish markets.
Likewise, frequent visits in different houses were made for information collection regarding ornamental fishes and to know about people’s interest towards this recreation. The data were collected by direct observation, through questionnaire and by phone, using a writing pad, a pen, and a camera.

Results

There are around 30 aquarium shops in Kathmandu Valley including all four districts. In Lalitpur, Kupundol, Jawalakhel, Lagankhel & other place of this district aquarium fish shops are available. During visit to shops it showed that more than 40 types of aquarium fishes as shown in the Table 1. Out of these fishes most of them are imported from foreign country and very less native species were seen in the market.

Table 1. Fish prices at Lalitpur

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Name of Fish</th>
<th>Price (NRs/2pc)</th>
<th>S. No.</th>
<th>Name of Fish</th>
<th>Price (NRs/2pc)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Brown gold fish</td>
<td>300</td>
<td>16</td>
<td>Serpa tetra</td>
<td>125</td>
</tr>
<tr>
<td>2</td>
<td>Lion head</td>
<td>3000</td>
<td>17</td>
<td>Guppy</td>
<td>200</td>
</tr>
<tr>
<td>3</td>
<td>Bubble eye</td>
<td>200</td>
<td>18</td>
<td>Balloon molly</td>
<td>150</td>
</tr>
<tr>
<td>4</td>
<td>Black mor gold</td>
<td>1000</td>
<td>19</td>
<td>Silver shark</td>
<td>200</td>
</tr>
<tr>
<td>5</td>
<td>Red oranda</td>
<td>275</td>
<td>20</td>
<td>Tiger shark</td>
<td>150</td>
</tr>
<tr>
<td>6</td>
<td>Suban kin</td>
<td>375</td>
<td>21</td>
<td>Rainbow shark</td>
<td>200</td>
</tr>
<tr>
<td>7</td>
<td>Ray kin gold</td>
<td>2600</td>
<td>22</td>
<td>White shark</td>
<td>150</td>
</tr>
<tr>
<td>8</td>
<td>White gold</td>
<td>300</td>
<td>23</td>
<td>Red tail black shark</td>
<td>200</td>
</tr>
<tr>
<td>9</td>
<td>Mix gold</td>
<td>525</td>
<td>24</td>
<td>Green terror</td>
<td>800</td>
</tr>
<tr>
<td>10</td>
<td>Red carp oranda</td>
<td>275</td>
<td>25</td>
<td>Dollar fish</td>
<td>200</td>
</tr>
<tr>
<td>11</td>
<td>Bensent tetra</td>
<td>150</td>
<td>26</td>
<td>Oscar</td>
<td>1500</td>
</tr>
<tr>
<td>12</td>
<td>Angel fish</td>
<td>1150</td>
<td>27</td>
<td>Flower horn</td>
<td>3000</td>
</tr>
<tr>
<td>13</td>
<td>Green tiger barb</td>
<td>550</td>
<td>28</td>
<td>Texax</td>
<td>250</td>
</tr>
<tr>
<td>14</td>
<td>Red eye sword tail</td>
<td>150</td>
<td>29</td>
<td>Tinfoil barb</td>
<td>350</td>
</tr>
<tr>
<td>15</td>
<td>Platy</td>
<td>100</td>
<td>30</td>
<td>Zebra fish</td>
<td>400</td>
</tr>
</tbody>
</table>

Among these fish Parrot Fish has highest price which reaches upto Rs. 20,000 per two pieces of large size available where as Gupppi has lowest price which ranges from Rs. 15 to 35 per two pieces. Concerning size of the fish, Guppi is the smallest one which has about one inch length and Shark is the largest one having length of about one & half feet.

Only artificial feed are given to aquarium fishes, in which Tokyo, six seven, nine star optimum, C.P., Etc are famous ones. While talking about aquarium filter, three types of filter are seen i.e. physical filter, bio filter and over head filter. For decorative purpose stones, different types of statues, artificial fishes, turtle, snakes, artificial plants etc are used. In shops and few houses also natural plants like hydrilla, cocomba, amazon etc are seen.

People showed more interest in keeping gold fish varieties due to their different coloration and shape. They keep different varieties of gold fishes like Red Oranda, Black oranda, Subun kin, Calico gold, Bubble eyes, Pearl scale etc. besides they also shows their interest in fishes like Oscar, Texax, green terror, Pirana, Sweaper, Angle fish, Parrot fish and Sharks etc. Fishes like Guppy and Tetra are famous among children due to their small size, active nature and low cost. As well as Gold fishes like Ray kin,
red Oranda, Bubble eyes are also liked by children. Decorative items like stones gravels, colorful scenery artificial fish turtle artificial plants kept in aquarium gives extra ordinary look to tank, which also helps people to increase their interest in aquarium fish keeping.

**Conclusion and Recommendations**

By visiting different shops in the Lallitpur suggest that more than 40 types of aquarium fishes are available. Only glass tank is made locally but it is imported from different countries. Shopkeepers in group use to import so most of them represent them self as a wholesalers. Shopkeeper not pleased with government and scholars of related field. The government agencies, NGO, INGO can play supportive role in the developing fish marketing and other related things. In this way number of increasing aquarium shops year by year in the valley is positive & satisfactory point regarding aquarium fish trade and also indicates the public demand. For this, government sector should put forward artificial breeding programs in extensive way so that fish can made available at reliable prices & mass killing of fish events can be lowered.

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**Figure 1.** Parrot Fish  
**Figure 2.** Guppy  
**Figure 3.** Red cap Oranda
References


Habitat Preference of the Greater One-Horned Rhinoceros (*Rhinoceros unicornis* Linn.) in the Baghmara Buffer Zone Community Forest, Chitwan National Park, Nepal

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**Abstract**

The Greater One-Horned Rhinoceros (*Rhinoceros unicornis* Linn.) is listed in Appendix I of the Convention on International Trade in Endangered Species of wild fauna and flora (CITES). A year round (October 2010 to September 2011) research was conducted in the Baghmara Buffer Zone Community Forest (BBZCF), located in the central lowland of Nepal, covering an area of 215 hectares, to collect information on the habitat types of the study area and the habitat preference of the Greater One-Horned Rhinoceros in that area. A total of four nested quadrates (n=4) that were already laid on transects traversing east to west were randomly selected to collect information about the habitat types and the habitat preference of the Greater One-Horned Rhinoceros in the study area. The four nested quadrates, representing the major habitat types of the study area, were laid in four different sites of the Community Forest, Bandevi area (Forest habitat type), Kholsa forest area (Riverbed Forest habitat type), Macchan area (Grassland habitat type) and Riverside forest area near the Budi Rapti River (Riverside Forest habitat type). On the basis of the observed frequency of the animal locations and the availability of their indirect signs studied in the four habitat types of the study area, it was found that the Greater One-Horned Rhinoceros preferred the Grassland habitat type the most in the winter and the monsoon seasons; and likewise the Forest habitat type in the summer season as their habitats.

**Key words:** Bandevi, BBZCF, Grassland, Kholsa, Machan

**Introduction**

Habitat is a complex mixture of plant communities, water, weather, animals and other environmental features that provide food and cover that an organism needs. Habitat is the home of the wildlife. When the habitat factors are in a good supply, they contribute to the well being of the wildlife and if any one of these factors is in a short supply, the wildlife number and location in that habitat are limited. Habitat may be selected for cover availability, forage quality and quantity and resting or den sites, each of which may vary seasonally. The preference of habitat by an organism is the consequence of habitat selection and this result in the disproportional use of some resources over the others.

This research work was conducted in the Baghmara Buffer Zone Community Forest, which has an advantage of having direct land-linkage with Chitwan National Park, the original habitat of the Greater One-Horned Rhinoceros. The Greater One-Horned Rhinoceros is listed in Appendix I of the Convention on International Trade in Endangered Species of wild fauna and flora (CITES). The species was
categorized under IUCN criteria as Endangered for a long period until the relative recovery of a single population in India (Kaziranga National Park) resulted in down-listing to vulnerable category on the IUCN Red List of Threatened species (IUCN 2008). The Government of Nepal has regarded the Greater One-Horned Rhinoceros in the list of protected animals and has been conducting various conservation programs in collaborations with different conservation partners (Chalise 2008).

The Greater One-Horned Rhinoceros (*Rhinoceros unicornis* Linn.), belonging to the family Rhinocerotidae, is a large mammal primarily found in the parts of north-eastern India and in protected areas in the Tarai of Nepal. This heavily built species is the second largest living rhino next to the White Rhino. The adult male rhinos weigh up to 2100 kilogram and stand up to 186 centimeter at the shoulder. While the adult female rhinos reach weights of about 1600 kilogram and heights of 160 centimeter. The rhino skin, which is thick and silver-brown in colour, becomes pinkish near the large skin folds that cover its body. The upper legs and shoulders are covered in wart-like bumps. The rhino’s single horn, present in both males and females (but not on newborn young), is pure keratin and starts to show about six years and is normally between 15 and 45 centimeter long (Chalise 2008).

The rhinos are mostly solitary creatures, with the exception of mothers and calves and breeding pairs, although they sometimes congregate at bathing areas. Mothers will stay close to their calves for up to four years after their birth; sometimes allowing an older calf to continue to accompany her once a newborn calf arrives. Being a solitary and primitive herbivorous mammal, the rhino shows distinct behavioral characteristics. Laurie (1978, 1982) has done remarkable studies on the behavioral activities (both diurnal and nocturnal), which covered feeding behavior, drinking behavior, aggressive behavior, non-breeding play behavior and reproductive behavior, etc. Activity takes place mostly at night, early in the morning and in the late afternoon (Laurie 1978), but in the middle of the day, the rhinos are commonly seen resting in the shade or mud; and wallowing and bathing in the lakes, rivers and pools. The rhinos are excellent swimmers and they can run at speeds up to 55 kilometer/hour for short periods of time. They also have excellent senses of hearing and smell, but relatively poor eyesight.

The rhino is a grazer. The rhino feeds mainly on grasses; and also fruits, leaves, branches of trees and shrubs; and cultivated crops (Jnawali 1989). Feeding occurs during the morning and evening. When feeding on tall grasses, the rhino curls its prehensile upper lip around the grass stems, bends the stems over and bites off, and chews the top (Laurie 1978). Drinking takes place on a daily basis. Drinking normally lasts only a minute or two and mineral licks are visited regularly (Laurie 1978). The rhino requires special habitat conditions including wallows, grasslands, woodlands and forest cover. The rhino spends about 8 hours/day in wallows or streams during the period of high humidity (August-September) but they spend at least an hour/day wallowing in December and January (Laurie 1978). The Asiatic rhino species prefer to reside in alluvial flood-plain vegetation of sub-tropical climate where water and green grasses are available all year round. Their phylogeny, ecology and nutritional energetic have evolved around these grassland ecosystems. The other preferred habitat includes marshy lowland Sal forest, wooded grassland and riverbeds. In Nepal, many rhinos now live within blocks of the suitable rhino habitat in Chitwan National Park. The rhinos occurred in highest densities along the flood-plain grasslands and riverine forests bordering the Rapti, Narayani, Reu and Dhunge rivers, suggesting that the flood-plain grasslands dominated by 4-6 meter tall *Saccharum spontaneum* are the single critical habitat (Dinerstein and Price 1991).
Materials and Methods

Study area

The Baghmara Buffer Zone Community Forest is situated on the North-East boundary of Chitwan National Park in Bachhauli Village Development Committee of Chitwan district, covering an area of 215 hectares. It lies between 27°34’78” to 27°35’53” northern latitude and 84°28’43” to 84°29’40” eastern longitude, in the sub-tropical region of lowland Nepal. The Baghmara Buffer Zone Community Forest borders the cultivated land to the east, Budhi Rapti to the west and the north, and Badreni village and Rapti River to the south.

The climate of the study area is sub-tropical with a summer monsoon from mid June to late September and a relatively dry winter. Humidity is high all-year round. Winter lasts from October to the end of February. January is the coldest month, with temperatures falling almost to freezing-point, especially when it rains. March to early June is the traditional hot months, with temperatures rising progressively to a peak in May. During April, despite the heat of the day, the nights can be quite cold. Heavy flooding occurs during the monsoon.

The dominant species of the Community Forest are Simal (Bombax ceiba), Bhellar (Trewia nudiflora) and Padke (Albizia julibrissin). The Community Forest that was once degraded and deprived of wildlife has now become a vibrant habitat for over 20 species of mammals and 162 species of birds. This Community Forest has harbored the Rhino (Rhinoceros unicornis), Spotted Deer (Axis axis), Sambhar Deer (Cervus unicolor), Barking Deer (Muntiacus muntjak), Hog Deer (Axis porcinus), Wild Boar (Sus scrofa), Rhesus Monkey (Macaca mulatta) and the Royal Bengal Tiger (Panthera tigris) (Pant 2003, Sharma et al 2013).

Figure 1. Location and Physical Parts of Chitwan National Park in Nepal.
Method

A year round (October 2010 to September 2011) research was conducted in the Baghmara Buffer Zone Community Forest to collect information on the habitat types and the habitat preference of the Greater One-Horned Rhinoceros in the study area. A total of four nested quadrates (n= 4) that were already laid on transects traversing east to west were randomly selected to collect information about the habitat types and indirect signs of the Greater One-Horned Rhinoceros in the study area. Nested quadrates of 400 square meters (20 meter x 20 meter), 25 square meter (5 meter x 5 meter) and 1 square meter (1 meter x 1 meter) were used to collect data on tree, under-story and ground vegetation respectively.

Habitat Utilization Pattern

All the sightings of the rhinos and their every indirect signs in the different habitat types of the study area, with respect to the age-sex composition and the habitat types were recorded to study the habitat utilization pattern. During the investigation, the vegetation types of the areas wherever the animals were observed and their every activity were recorded, as well as, any indirect signs, such as, fresh wallow uses, footprints, dung heaps and feeding signs, if observed were also noted down as indirect evidences of the rhino visit in the habitat. The indirect signs of the animal were also collected from 400 square meter, 25 square meter and 1 square meter quadrates of the four study sites of the Community Forest. A total of 25 quadrates were surveyed in the Baghmara Buffer Zone Community Forest (Table 1).

Table 1. Total Number of Quadrates Surveyed in Different Habitat Types of the Baghmara Buffer Zone Community Forest in 2011.

<table>
<thead>
<tr>
<th>Habitat Types</th>
<th>Number Of Quadrates Surveyed</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Tree</td>
<td>Under-story</td>
</tr>
<tr>
<td>Forest</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Riverbed Forest</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Grassland</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Riverside Forest</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>3</strong></td>
<td><strong>6</strong></td>
</tr>
</tbody>
</table>

Habitat Preference

Habitat preference was evaluated by comparing the frequency distribution of the animal locations on the different habitat types within the study area. Here, the observed frequency of the animal locations included not only the different habitat types which the rhinos were noticed using in each season; but also all those habitat types in which the animal signs were also found. Those signs were the indirect evidences which showed the physical presence of the animals and the habitats were used by the animals. So, all the habitat types which the rhinos were observed using and where the rhino signs were found were considered as the preferred habitat types by the animal.

In calculating the Important Value Index (IVI), the percentage values of the relative density, relative frequency and relative dominance were summed up together and the value was designated as the Importance Value Index (IVI) of the species (Krebs 1989). Relative Density was the study of numerical strength of a species in relation to the total number of individuals of all the species. Relative Frequency
was the degree of dispersion of individual species in an area in relation to the number of all the species occurred. Relative Dominance of trees was determined by calculating the basal area. Tree basal area was the cross-sectional area of a tree’s trunk at 1.3 meter off the ground, which was approximately chest-height.

Prominence Values (PV) of individual species in under-story or ground layers was calculated by multiplying the percent cover of individual species and square root of its frequency (Dinerstein 1979).

**Results**

The study revealed that the rhinos utilized four major habitat types in the study area, Forest habitat type, Riverbed Forest habitat type, Grassland habitat type and Riverside Forest habitat type.

Among the four nested quadrates studied, three contained 105 individuals of trees of six species. The Important Value Index (IVI) of Bhellar (*Trewia nudiflora*) (110.534 percentage) was found to be the maximum, followed by that of Padke (*Albizia julibrissin*) (103.059 percentage) and then that of Dhamin (*Dysoxylum binectariferum*) (33.853 percentage) and Aureli Kanda (*Caesalpinia decapetala*) (19.538 percentage) and Kalo Bilaune (*Antidesma acuminatum*) (16.508 percentage) and Ban Belauti (*Psidium guajava*) (16.508 percentage) respectively. In the under-story layer of the forest habitat type, Bhati (*Clerodendrum viscosum*) was found to be the most prominent species (PV= 582.521 percentage). In the ground vegetation of the forest habitat type, Pipla species was found to be the most prominent species (PV= 244.524 percentage). Similarly, in the under-story layer of the riverbed forest habitat type, Bilaune (*Maesa species*) was found to be the most prominent species (PV= 265.149 percentage) and Himalayan Maiden Hair Fern (*Adiantum venustum*) was found to be the most prominent species (PV= 183.712 percentage) in the ground vegetation of the riverbed forest habitat type. Likewise, in the grassland habitat type, Chepte Jhar species was found to be the most prominent species (PV= 105.399 percentage).

**Habitat Use by the Rhino in the Baghmara Buffer Zone Community Forest in Different Season of the Year**

The study showed that the observed rhinos used different habitat types in different seasons in the Baghmara Buffer Zone Community Forest. In general, all the individual rhinos intensively used the forest habitat type and the grassland habitat type, which was then followed by the riverbed forest habitat type. The riverside forest habitat type was used in low frequency. Similarly, the individual rhinos used the grassland habitat type the most, followed by the riverbed forest habitat type and then the forest habitat type in the winter season. The forest habitat type was extremely used by the individual rhinos in the summer season. Likewise, the individual rhinos used the grassland habitat type frequently followed by the forest habitat type in the monsoon season.
Table 2. The Observed Frequency of Habitat Use by the Observed Rhinos in the Different Seasons in the Baghmara Buffer Zone Community Forest (October 2010 to September 2011).

<table>
<thead>
<tr>
<th>Name Of Animal</th>
<th>Forest</th>
<th>Riverbed Forest</th>
<th>Grassland</th>
<th>Riverside Forest</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adult Female (Winter)</td>
<td>2</td>
<td>4</td>
<td>6</td>
<td>-</td>
<td>12</td>
</tr>
<tr>
<td>Adult Female (Summer)</td>
<td>5</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>5</td>
</tr>
<tr>
<td>Adult Female (Monsoon)</td>
<td>4</td>
<td>-</td>
<td>5</td>
<td>-</td>
<td>9</td>
</tr>
<tr>
<td>Adult Male (Monsoon)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Calf Male (Winter)</td>
<td>2</td>
<td>4</td>
<td>6</td>
<td>-</td>
<td>12</td>
</tr>
<tr>
<td>Calf Male (Summer)</td>
<td>5</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>5</td>
</tr>
<tr>
<td>Calf Female (Monsoon)</td>
<td>4</td>
<td>-</td>
<td>5</td>
<td>-</td>
<td>9</td>
</tr>
<tr>
<td>Unidentified *</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>-</td>
<td>3</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>24</strong></td>
<td><strong>9</strong></td>
<td><strong>23</strong></td>
<td><strong>1</strong></td>
<td><strong>57</strong></td>
</tr>
<tr>
<td>Percentage (%)</td>
<td>42.11%</td>
<td>15.79%</td>
<td>40.35%</td>
<td>1.75%</td>
<td>100%</td>
</tr>
</tbody>
</table>

(* Indirect signs, such as, dung heaps, fresh wallow uses and footprints of the observed rhinos recorded.)

Figure 2. The Observed Frequency of Habitat Use by the Observed Rhinos in Three Different Seasons in 2011.

Habitat Preference

In the Baghmara Buffer Zone Community Forest, the observed rhinos showed a marked preference towards the forest habitat type, the grassland habitat type and the riverbed forest habitat type, whereas, the riverside forest habitat type was significantly avoided (Table-2). More precisely, in the winter season, the observed rhinos preferred the grassland habitat type the most, which was then followed by the riverbed forest habitat type and then the forest habitat type. There was no sign of utilization of the riverside forest habitat type in the winter season. While in the summer season, the forest habitat type was extensively preferred by the observed rhinos. Also, an indirect sign of fresh wallow use in the riverbed forest habitat type too indicated the preference of that habitat type by the rhinos in the summer season. Similarly, the
grassland habitat type and then the forest habitat type were much preferred by the observed rhinos in the monsoon season. The riverside forest habitat type was preferred in low frequency by the observed rhinos in the monsoon season, whereas, the riverbed forest habitat type was completely avoided.

**Discussion**

The present study showed that in the three out of the four nested quadrates studied, Bhellar (Trewia nudiflora) (IVI=110.534 percentage) had a higher or major controlling influence by its number and its size in the tree canopy layer, followed by Padke (Albizia julibrissin) (IVI= 103.059 percentage) then by Dhamin (Dysoxylum binectariferum) (IVI= 33.853 percentage) then by Aureli Kanda (Caesalpinia decapetala) (IVI= 19.538 percentage) then by Kalo Bilaune (Antidesma acuminatum) (IVI= 16.508 percentage) and by Ban Belauti (Psidium guajava) (IVI=16.508 percentage) respectively. The study showed that Bhellar (Trewia nudiflora) which was one of the main planted species in the study area happened to be an important tree species in the three nested quadrates and Padke (Albizia julibrissin) which was not the planted species in the study area appeared to be the second most important tree species in the tree canopy layer. It shows that the soil of the study area is suitable for the succession of the tree species. The number of the tree species in the tree canopy layer in the forest, the riverbed forest and the riverside forest habitat types was six and Bhellar (Trewia nudiflora), the most important tree species in the canopy layer, offers the favourite fruits for the rhinos, and hence, offers a good habitat for the rhinos.

In the grassland habitat type, Chepte Jhar species (PV= 105.399 percentage) was found to be the most prominent species, followed by Dubo (Cynodon dactylon) (PV= 75.434 percentage). The species diversity in the grassland habitat type appeared to be rich in all the seasons and hence, provided a suitable habitat for the rhinos throughout the year.

The present study revealed that the rhinos exhibited seasonal differences in the habitat use and preference and intensively used and preferred the forest habitat type and the grassland habitat type, followed by the riverbed forest habitat type, in general. Whereas, the riverside forest habitat type was used and preferred in low frequency throughout. Furthermore, in detail, in the winter season, the grassland habitat type was preferred the most by the rhinos, followed by the riverbed forest habitat type and then the forest habitat type. Moreover, the present study also revealed that the forest habitat type was extremely preferred by the rhinos in the summer season and the grassland habitat type was preferred more followed by the forest habitat type in the monsoon season. The difference observed in the frequency distribution of the rhino locations in the different habitat types of the study area can be presumably explained by the fact that in the natural habitats the resources, such as, food, water and cover are not distributed uniformly. Also, the considerable seasonal variations in the availability of the food in the study area may have resulted in the movements of the rhinos between the different habitat types.

The major preference for the grassland habitat type in the winter season may have been due to the occurrence of the preferred food species of the rhinos, such as, the new flush of Tinpate Jhar (Trifolium repens), Dubo (Cynodon dactylon) after the periodic burning in the habitat type, the easy access to the sun basking in the chilly winter days and the availability of the open land for different activities like running, playing. Likewise, the extreme preference of the forest habitat type by the rhinos during the summer season may be explained by the fact that the forest provided the rhinos with their preferred fodder plant species, such as, young leaves of Bilaune (Maesa species), Bhati (Clerodendrum viscosum); cover from the intense summer heat and most importantly, the several, small water holes formed at places due to the summer rain in the forest allowed the animals for wallowing during the daytime. Majority of the summer time was spent in wallowing in different wallow sites in the different forest habitat types by the rhinos. The abundant presence of the preferred food species of the rhinos in the grassland habitat type, during the
monsoon season, further significantly contributed to the habitat preference of the rhinos. Some of the preferred food species of the rhinos found in the grassland habitat type of the study area during the monsoon season were, Bhati (*Clerodendrum viscosum*), Siru Jhar (*Imperata cylindrica*), Dubo (*Cynodon dactylon*), Gandhe (*Ageratum conyzoides*), Neuro (*Dryopteris cochleata*), Namle Jhar (*Coix species*), Kuro (*Chrysopogon aciculatus*), Kans (*Saccharum spontaneum*), Bansur Jhar; of which, during the study, the rhinos were found frequently feeding on the young leaves and stem of Bhati (*Clerodendrum viscosum*), Siru Jhar (*Imperata cylindrica*), Dubo (*Cynodon dactylon*), Gandhe (*Ageratum conyzoides*) and Neuro (*Dryopteris cochleata*).

**Conclusion**

The rhinos use four main habitat types, namely, the forest habitat type, the riverbed forest habitat type, the grassland habitat type and the riverside forest habitat type in the study area; where food, water and cover are available. Throughout the year, with the considerable seasonal variations in the availability of food and shelter in the study area, the rhinos move between the different habitats types, thus, resulting in the disproportional use of one resource over the other. In the Baghmara Buffer Zone Community Forest, the rhinos exhibit seasonal differences in the habitat preferences and there is a statistically significant difference in the use of different habitat types by the rhinos between the different seasons of the year as a whole. The rhinos prefer the grassland habitat type the most in the winter season, the forest habitat type in the summer season and the grassland habitat type in the monsoon season. The forest and the short grassland habitat types of the Baghmara Buffer Zone Community Forest, although is still in the succession stage, provide a suitable habitats for the rhinos.

**Acknowledgements**

We express our gratitude to the Central Department of Zoology for providing the opportunity to study, the Department of National Parks and Wildlife Conservation, Nepal (DNPWC) for providing the research permission and the Baghmara Buffer Zone Community Forest (BBZCF) for providing the research permit. J.K. Lama, Field Technician, is also highly acknowledged for his kind support.

**References**


Arthropod Pests of Pear (*Pyrus pyrifolia*, Nakai) in Central Horticulture Centre, Kirtipur, Kathmandu, Nepal

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Abstract

Arthropod pests cause serious threats to pear orchards, resulting heavy reduction in their yield. The present study focused on the arthropod pests of Pear (*Pyrus pyrifolia*, Nakai), in the Pear orchard of Central Horticulture Centre, Kirtipur, Kathmandu. It was carried out in two seasons: Spring and Summer of 2012. The pests were collected through different methods such as hand picking, knock down process, beating process, sweeping and use of aspirator, depending upon the size of pests. The statistical analyses were performed at 95% confidence level in R-Software (R-Console version2.15.2).

Altogether, 12 species of pests belonging to 11 families were observed during the study. The results showed that the pest's population was independent to months ($X^2=7.663$, df=5, $P=0.175$), seasons ($X^2=0.188$, df=1, $P=0.664$), and sites ($F=0.446$, df=2, $P=0.652$), however, population density of pests was higher in June (409) and summer (966). Spider mite (*Tetranychus* spp.), Aphids (*Aphis* spp.), Thrips (*Scirtothrips* spp.) Tortoise beetle (*Metrona* spp.) and Fruitfly (*Dacus* spp.) were the common pest species and the most dominant one was the Spider mite (*Tetranychus* spp.), occurring almost in all months, seasons and sites. The overall diversity index of pear pests was high, i.e, 0.800, however regarding individual species diversity index and dominance index, Spider mite (*Tetranuchus* spp.) had higher diversity indices, followed by Tortoise beetle (*Metronia* spp.), Fruit fly (*Dacus* spp.) and the least was observed in Click Beetle (*Agriotes* spp.). Although the pest population is independent to months and seasons, the present study showed that pear plants were more affected by pests during hot and warm season.

Keywords: Click beetle, Fruit fly, Pear orchard, Spider mite, Tortoise beetle

Introduction

Pear is commonly a medium sized tree or shrub of genus *Pyrus* (Riegel 2006), which generally reaches height of 10 m to 17 m. The leaves of this plant are alternately arranged, simple and are 2-12 cm long. The flowers are white, rarely tinted yellow or pink and 2-4 cm in diameter and have five petals. The inflorescence is corymbose with five to seven flowers (Riegel 2006). It is one of the very important deciduous fruit plants of Nepal (Devkota 1999) which is also named as Pomaceous fruit. There are over 5,000 varieties of pears cultivated in temperate climate worldwide (Herbst 2001), of which the most important species for commercial production are European Pear (*Pyrus communis*) and Asian Pear (*Pyrus pyrifolia*), which is also known as Japanese pear or Oriental Pear (Beutel 1990 and Riegel 2006).
In Nepal two kinds of pears are grown: Asian pear (*Pyrus pyrifolia*) and European pear (*Pyrus communis*) (Devkota 1990), former is also called oriental pear which is locally named as Naspati. Asian pears have a distinct pear like taste and a crisp texture. These pears ripen on the trees and they do not need cold storage period while European Pear are not rounder in shape and less crunchier than Asian Pear (Beutel 1990). The Asian Pear includes Japanese Pears, Chinese Pears, etc. The Japanese Pear includes different varieties like Shinsu, Shunko, Kosui, Hosui, etc. while European Pears are Barlett and Comice.

There are many threats in the cultivation of Pears. Generally, Pears are attacked by various kinds of pests such as Mites, Aphids, Thrips and Scale insects (CHC Annual Report 2011/12). Mites are known to migrate from host plants to other plants and vice-versa (Banerjee 1971) and are known as an important vector of plant viruses too (Gupta 1985, 1990). They may cause various types of direct damages to plants such as loss of chlorophyll, stunted growth, burnt appearance of leaves, heavy leaf fall and reduction in yield (Dhoria 1999). Generally, the Spider mite (*Tetranychus*) is susceptible to orchard plants. They suck sap from leaves, flowers and buds resulting in discoloration, appearance of silvery patches and drying of affected plants (Banerjee 1971).

Aphids are another recognized pests of Pear, which are sap sucking in nature (Sharma 2000). They are found in flowers, leaves and stem of Pear and are known to have transmitted more than 100 plant viruses (Sharma 2005). Similarly, Pear thrips and Scale insects are other common pests of Pear plant. Young thrips feed almost entirely on tender foliage and fruits (Daniel 1904), while scale insects make small, narrow or circular flat holes on stem and fruits of Pear (Evans 1984).

**Materials and Methods**

**Study area**

Central Horticulture Centre lies in Kirtipur Municipality, Kathmandu, which is 5 km south from Kathmandu valley. Geographically, it lies between 27° 30’ N latitude to 85° 15’ E longitude, at an altitude of 1,320 m asl (CHC Annual Report 2011/12).

The station has mild temperature reaches up to 32°C during summer and falls below -3°C during winter. The average rainfall during monsoon season is 1,025 ml (CHC Annual Report 2011/12).
Figure 1. Map of Central Horticultural Centre, showing pear orchard (study area). **Study design**

The total area covered by CHC is 20 hectares, of which 11 hectares is occupied by 21 different fruit orchards, including Pears (*Pyrus pyrifolia*, Nakai) (CHC Annual Report 2011/12). Within this orchard area, three different sites: A, B and C were selected which are 20 m apart from one another. They cover an area of 42,831.25 sq. feet, 7,820.65 sq. feet and 33,594 sq. feet respectively. From each site, 10 different Pear plants were selected randomly and were marked. Then they were checked for the insect pests, by observing all parts of the plants such as stem, twig, leaf, blossoms, fruits, flowers and roots for two different seasons. They were observed through naked eyes and by hand lens, if needed. The sites were visited twice a month in the intervals of 15 days.

**Pest collection and preservation**

The pests which were large enough to be seen readily with naked eyes were collected by hand picking method with the help of forceps, and were put in vials containing 70% ethanol as preservative. Specimens like aphids, which occur in clusters were collected by using soft brush soaked in ethanol and also by knockdown process. Thrips and mites were also collected in similar way. Sweep net was used for collecting flying insect pests. Also, hanging vials with pheromones (Batocera compositae lure) was used for pest like Fruit flies. On the other hand, beating process and aspirator was used for the collection of small and light insects.

The general objective of this research was to study the arthropod pests of Pear (*Pyrus pyrifolia*, Nakai) in Central Horticulture Centre, Kirtipur.

**Data analysis**

One-way Analysis of Variance (ANOVA) was used to test the significant difference in number of pests observed in different sites. But, to test the significant difference in pest’s number due to
months and seasons, Kruskal-Wallis Rank Sum test was used. It is a non parametric test which is alternative to ANOVA. Since the data was not found normal, hence this test was performed. The statistical analyses were performed at 95% confidence level in R-Software (R-Console version 2.15.2).

On the other hand, the Shannon diversity index (H̅) was calculated by using the formula
\[
\text{Shanon index of diversity (H̅) = } -\sum (ni/N) \log(ni/N) \quad \text{(Odum 1996)}
\]
Where, ni = Importance value for each species  
N = Total of importance value

Similarly, the index of dominance (c) was calculated as
\[
\text{Index of dominance (c) = } \sum (ni/N)^2 \quad \text{(Odum 1996)}
\]
Where, ni = Importance value for each species  
N = Total of importance value

**Results**

**Pests of pear**

Altogether 12 species of pear pests belonging to 11 families were observed in the Central Horticulture Centre, Kirtipur during the study. Among them, Family Chrysomelidae was represented by two species, whereas other families by single species (Table 1). The classification is based on Borror and Delong (1971).

**Table 1. Pest species with their families**

<table>
<thead>
<tr>
<th>S.N</th>
<th>Pests</th>
<th>Family</th>
<th>Scientific Name</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Aphid</td>
<td>Aphidae</td>
<td>Aphis spp.</td>
</tr>
<tr>
<td>2</td>
<td>Roaches</td>
<td>Blattodae</td>
<td>Unidentified</td>
</tr>
<tr>
<td>3</td>
<td>Tortoise Beetle</td>
<td>Chrysomelidae</td>
<td>Metriona spp.</td>
</tr>
<tr>
<td>4</td>
<td>Leaf Beetle</td>
<td>Chrysomelidae</td>
<td>Basilepta spp.</td>
</tr>
<tr>
<td>5</td>
<td>Snout Beetle</td>
<td>Curculionidae</td>
<td>Unidentified</td>
</tr>
<tr>
<td>6</td>
<td>San Jose Scale</td>
<td>Diaspidae</td>
<td>Quadraspidiotus spp.</td>
</tr>
<tr>
<td>7</td>
<td>Click Beetle</td>
<td>Elatridae</td>
<td>Agriotes spp.</td>
</tr>
<tr>
<td>8</td>
<td>Brown Marmorated Stink Bug</td>
<td>Pentatomidae</td>
<td>Halyomorpha spp.</td>
</tr>
<tr>
<td>9</td>
<td>Fruit fly</td>
<td>Tephritidae</td>
<td>Dacus spp.</td>
</tr>
<tr>
<td>10</td>
<td>Spider mite</td>
<td>Tetranychidae</td>
<td>Tetranychus spp.</td>
</tr>
<tr>
<td>11</td>
<td>Thrips</td>
<td>Thripidae</td>
<td>Scirtothrips spp.</td>
</tr>
<tr>
<td>12</td>
<td>Hornet</td>
<td>Vespidae</td>
<td>Vespa spp.</td>
</tr>
</tbody>
</table>

**4.2 Species diversity of pests**

Altogether 1,652 individuals of pests belonging to 12 species were observed during the study. Among them, Spider mite (*Tetranychus* spp.) was the most common pest of Pear. Similarly, the other frequently
occurring pests were Aphids (*Aphis* spp.), Thrips (*Scirtothrips* spp.), Tortoise Beetle (*Metriona* spp.) and fruitfly (*Dacus* spp.) (Figure 2). Click Beetle (*Agriotes* spp.) was the least common species in Pear Orchard of CHC during the study.

![Figure 2. Pest species and their number.](image)

**Table 2.** Species diversity of pests

<table>
<thead>
<tr>
<th>S.N</th>
<th>Pests</th>
<th>ni</th>
<th>ni/N</th>
<th>log ni/N</th>
<th>Shannon Index</th>
<th>Species Dominance</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Tetranychus</em> spp.</td>
<td>643</td>
<td>0.389225</td>
<td>-0.409799</td>
<td>0.159504117</td>
<td>0.151496242</td>
</tr>
<tr>
<td>2</td>
<td><em>Aphis</em> spp.</td>
<td>150</td>
<td>0.090799</td>
<td>-1.041919</td>
<td>0.094605216</td>
<td>0.008244464</td>
</tr>
<tr>
<td>3</td>
<td><em>Scirtothrips</em> spp.</td>
<td>132</td>
<td>0.079903</td>
<td>-1.097436</td>
<td>0.0876886</td>
<td>0.006384513</td>
</tr>
<tr>
<td>4</td>
<td><em>Metriona</em> spp.</td>
<td>308</td>
<td>0.186441</td>
<td>-0.7294593</td>
<td>0.136000891</td>
<td>0.034760126</td>
</tr>
<tr>
<td>5</td>
<td><em>Dacus</em> spp.</td>
<td>182</td>
<td>0.110169</td>
<td>-0.9579387</td>
<td>0.105535615</td>
<td>0.012137317</td>
</tr>
<tr>
<td>6</td>
<td><em>Halyomorpha</em> spp.</td>
<td>7</td>
<td>0.004237</td>
<td>-2.372912</td>
<td>0.010054712</td>
<td>1.79546E0-05</td>
</tr>
<tr>
<td>7</td>
<td><em>Quadraspidoitus</em> spp.</td>
<td>55</td>
<td>0.033293</td>
<td>-1.4776474</td>
<td>0.049195281</td>
<td>0.001108422</td>
</tr>
<tr>
<td>8</td>
<td><em>Agriotes</em> spp.</td>
<td>5</td>
<td>0.003027</td>
<td>-2.51904</td>
<td>0.007624213</td>
<td>9.16052E-06</td>
</tr>
<tr>
<td>9</td>
<td><em>Basilepta</em> spp.</td>
<td>95</td>
<td>0.057506</td>
<td>-1.2402864</td>
<td>0.071323978</td>
<td>0.003306946</td>
</tr>
<tr>
<td>10</td>
<td>Roaches (Unidentified)</td>
<td>13</td>
<td>0.007869</td>
<td>-2.1040667</td>
<td>0.016557426</td>
<td>6.19251E-05</td>
</tr>
<tr>
<td>11</td>
<td><em>Vespa</em> spp.</td>
<td>46</td>
<td>0.027845</td>
<td>-1.5552522</td>
<td>0.043306054</td>
<td>0.000775346</td>
</tr>
<tr>
<td>12</td>
<td>Snout Beetle (Unidentified)</td>
<td>16</td>
<td>0.009685</td>
<td>-2.0138901</td>
<td>0.019504988</td>
<td>9.38037E-05</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td><strong>1652</strong></td>
<td></td>
<td></td>
<td><strong>0.800901092</strong></td>
<td></td>
</tr>
</tbody>
</table>
The overall diversity index of pear pests was high, i.e., 0.800, however, regarding individual species diversity index, Spider mite (*Tetranychus* spp.) had higher diversity index (0.1595), followed by Tortoise Beetle (*Metriona* spp.), Fruitfly (*Dacus* spp.) and so on. Among 12 species of Pear pests, Click Beetle (*Agriotes* spp.) had the lowest diversity index (0.0076). Similarly, the index of dominance was also higher in Spider mite (*Tetranychus* spp.), which is 0.1514, followed by Tortoise beetle (*Metriona* spp.), Fruitfly (*Dacus* spp.) and so on. The Click Beetle (*Agriotes* spp.) was the lowest dominant pest species having index of dominance only 9.16052E-06 (Table 2).

### 4.3 Monthly variation in number of pests

Kruskal Wallis test revealed that there was no significant difference ($X^2=7.666$, df=5, $P=0.175$) in pest number due to different months. However, their number gradually increases from March to June, peaked on this month and then decreasing onwards in the subsequent months. Clearly, the highest number of pests was observed in June (409) and the lowest in March (94) (Figure 2). Similarly, species richness was also high in June and also in July too. Out of 12 species recorded, the same number of species, i.e. 11 was found in June and July, which was the highest number of species recorded among six months (Figure 2). Regarding specific pests, Aphids (*Aphis* spp.) and Thrips (*Scirtothrips* spp.) were dominant in March and April, whereas, Spider mite was the most dominant pest species in the subsequent months; in fact, there was more or less same number of Spider mite (*Tetranychus* spp.) and Tortoise beetle (*Metriona* spp.) in July (Figure 3). In May and June, Spider mite (*Tetranychus* spp.) alone damaged more than 50 per cent of total damage, done by overall pests in Pear orchard of CHC.

![Figure 3](image-url)  
**Figure 3.** Monthly variation in species and number of pests.

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4.4 Seasonal variation and number of pests

With variation in seasons, the number of Pear pests did not vary. Kruskal Wallis test showed that there was no significant difference ($X^2=0.188$, df=1, P=0.664) in number of pests in Pear plants due to different seasons. However, summer (966) had higher number of pests than the spring (686) (Table 3). But, among 12 pear pests recorded, summer had more number of species (11) than spring (9). Click Beetle (*Agriotes* spp.), Roaches (Unidentified) and Hornet (*Vespa* spp.) were absent in spring whereas, Thrips (*Scirtothrips* spp.) was not observed in summer during the study. In overall, Spider mite (*Tetranychus* spp.) was the most dominant pest species in both seasons.

**Figure 4.** Monthly variation and species dominance.
Table 3. Pests of Pear in two seasons

<table>
<thead>
<tr>
<th>SN</th>
<th>Pests</th>
<th>Season</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Spring</td>
<td>Summer</td>
<td>Total</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Spider mite (Tetranychus spp.)</td>
<td>196</td>
<td>447</td>
<td>643</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Aphids (Aphis spp.)</td>
<td>109</td>
<td>41</td>
<td>150</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Thrips (Scirtothrips spp.)</td>
<td>132</td>
<td>0</td>
<td>132</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Tortoise Beetle (Metriona spp.)</td>
<td>124</td>
<td>184</td>
<td>308</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Fruitfly (Dacus spp.)</td>
<td>32</td>
<td>150</td>
<td>182</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>Brown Marmorated Stink Bug (Halyomorpha spp.)</td>
<td>4</td>
<td>3</td>
<td>7</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>San Jose Scale (Quadraspidiotus spp.)</td>
<td>21</td>
<td>34</td>
<td>55</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>Click Beetle (Agriotes spp.)</td>
<td>0</td>
<td>5</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>Leaf Beetle (Basilepta spp.)</td>
<td>59</td>
<td>36</td>
<td>95</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Roaches (Unidentified)</td>
<td>0</td>
<td>13</td>
<td>13</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>Hornet (Vespa spp.)</td>
<td>0</td>
<td>46</td>
<td>46</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>Snout Beetle (Unidentified)</td>
<td>9</td>
<td>7</td>
<td>16</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>686</td>
<td>966</td>
<td>1,652</td>
<td></td>
</tr>
</tbody>
</table>

4.5 Variation in number of pests in different sites

There was no variation in number of pests occurring in different sites. One-way ANOVA revealed that there was no significant difference (F=0.446, df=2, p=0.652) between number of pest species and sites. More or less same number of pests was observed in all sites. Spider mite (Tetranychus spp.), Aphids (Aphis spp.), Thrips (Scirtothrips spp.), Tortoise beetle (Metriona spp.) and Fruitfly (Dacus spp.) were the most common pest species, occurring in all sites. The median value of pest’s number was more than 80 in all sites.

Figure 5. Box plot showing number of pests observed in three different sites (A, B and C). The dark line in the box plot represents the median or mid value and its arm represents the quartile value.
Discussion

Species diversity is the ratio between number of species and “importance values” (number, biomass, productivity, and so on) of individuals (Odum 1996) in which it tends to be low in physically controlled ecosystem and high in biologically controlled ecosystems. Species Diversity Index (SDI) is the most frequently used tool by many scientists for measuring the species diversity (Parrotta et al. 1997, Whitford 1997). It is a numerical measurement of species and can express the diversity within communities and often used to compare the diversity of the species. In Central Horticulture Centre (CHC), the SDI of overall pear pests was high (0.800), which shows stability in pest’s ecosystem. But regarding individual diversity indices, both SDI and index of dominance were high in Spider mite (Tetranychus spp.) comprising 0.1595 and 0.1514 respectively (Table 2). On the other hand, Click Beetle (Agriotes spp.) was the least dominating pest species having dominance index only 9.16052E-06. As this study was carried out in spring and summer months, this might be cause for high number of Spider mite (Tetranychus spp.) during the study because the mite population was positively correlated with high temperature and low relative humidity (Putatunda and Tagore 2000, Mohamed Osman and Mahmoud 2008). Therefore, its SDI and dominance index were relatively high among other pests of pear.

Although the pests’ population was statistically independent to months, however, the high number of pests was observed in June (409). It showed gradual increment from March to June, was peaked on June and their number gradually dwindled in the subsequent months (Figure 3). This result is in agreement with the findings of Dhooria and Bhutani (1983) and Putatunda and Tagore (2000), in which the pest population peaked during May to September and there was negligible population during December to March, as a result, their population was peaked in summer months. As temperature is generally high in summer months (May and June), this might be another cause for high density of pests in these months because the life cycle of most of the insect pests became functional or more active during hotter days (Pedigo 2002). Indeed, eggs overwinter and hatching occurs after onset of spring, i.e. when the environment became warm. Regarding specific pests in the present study, the density of Spider mite (Tetranychus spp.) was highest which is followed by Tortoise beetle (Metriona spp.), Fruitfly (Dacus spp.), Aphids (Aphis spp.), Thrips (Scirtothrips spp.) and so on (Figure 2). The annual report of CHC Annual Report (2011/12) also mentioned Spider mite (Tetranychus spp.) as a pest of pear, including other species such as Hornet (Vespa spp.), Thrips (Scirtothrips spp.) and Fruitfly (Dacus spp.) which resembles with our study.

There was no statistical variation in pest population due to seasons, but more number of pests was observed in summer (966) than in spring (686). Temperature is one of the most important factors that have great effect on pests (insects) developmental rates primarily because of their poikilothermic adaptation. As environmental conditions become more and more warm, their biological processes and hence the life cycle proceeds more well (Pedigo 2002). Therefore, due to higher temperature in summer months, pest population generally becomes higher. This was observed during our study as well. Regarding specific pests, Spider mite (Tetranychus spp.) alone dominated the both seasons in present study, but its number was higher in summer which is similar to the study of Portor (2007) and Murray and Alston (2011), where they argued that Spider mites reproduce during hot and dry conditions of summer and thus their density was high in this period. In contrast, Krewer and Bertrand (2012) discussed that Red Spider mites attack pears during bloom in great proportion, i.e. at spring which contradicts with our findings. This may be because we had not gone to species level, as perhaps the species of Spider mites were different.
Similarly, Alston and Reding (2011) revealed that pear is the most sensitive to mite feeding than other stone fruits. Chhillar and Kumar (2000) also argued that phytophagous mites occur almost in all ornamental crops, including pear, reducing sharp decrease in yield and serving as a vector of several plant diseases. The annual report of CHC (2011/12) also mentioned that there was a noticeable decrease in the yield of pear due to damage done by mites. Mite feeding may cause leaf curling, leaf burning and defoliation (Gupta 1990), that lead to decrease in total yields. During our visit too, leaf curling and burning in some pear plants were observed. This was more noticed in June and July; because perhaps their population is at peak in these months which directly correlates to nature of damage.

The study further showed that Thrips (Scirtothrips spp.) were more dominant during first two spring months (Table 3). Booth (2007) discussed that pear is one of the host trees of Thrips, in which it overwinters in the soil and the adult Thrips emerge from soil in early spring and begin feeding just as the buds begin to expand, which is similar to our results. Therefore, this might be a cause for high density of Thrips during spring months. Gardescu (2008) also discussed that adult Thrips emerge from their underground cells in spring when soil temperature become warm, therefore, their population seem higher in spring than other season of a year.

On the other hand, there was no significant difference in the number of pests in different sites (Site A, B and C) of pear orchard of CHC (Figure 5). In all sites, Spider mite (Tetranychus spp.), Aphids (Aphis spp.), Thrips (Scirtothrips spp.), Tortoise beetle (Metriona spp.) and Fruitfly (Dacus spp.) are the common pests that were recorded mostly. The three sites are no more than 20 m from one another, so that any insect pest whether flying or non-flying could easily migrate from one to other. Similarly, the physical location of these sites is also similar; all are facing towards east and somehow sloppy in nature. There did not exist any physical or topographical difference among these sites, though they occupy different area. As a result, the pest composition seemed similar in all sites.

Results showed that some pests such as Fruit fly (Dacus spp.), Hornet (Vespa spp.) and Tortoise beetle (Metriona spp.) were more in number during fruiting season of pear. The annual report of CHC (2011/12) also mentioned Hornet (Vespa spp.) and Fruitfly (Dacus spp.) as a destructive pests of pear during fruiting period. This study seemed close to this annual report of CHC, where the density of Hornet (Vespa spp.), Fruitfly (Dacus spp.) and beetles were comparatively high in summer, a fruiting time of pear. As these species prefer fruits than other parts of plants, hence this might be a cause for these species to occur more in fruiting trees of pear.

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Parasitic burden in Red panda (*Ailurus fulgens* Cuvier, 1825) of Illam district community forest, Nepal.

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Abstract

Parasitic diseases pose significant conservation threat in conservation of Red panda. In order to assess the parasitic burden in Red panda of Illam district, Nepal, 14 faecal samples were aseptically collected from community forest of Illam using line transect method following the GPS location. The samples were examined using standard concentration techniques. All the collected samples were found to be positive for both protozoan and helminth parasites. The recorded protozoan parasites includes *Eimeria* sp., *Entamoeba* sp., and *Balantidium* sp. with 64.28%, 57.14% and 14.28% proportion respectively. Similarly the proportion of seven helminthes parasites revealed *Oxyuris* (100%), *Baylisascaris* (57.14%), *Trichostrongylus* (50%), *Strongyloides* (50%), *Trichuris* (42.8%), *Crenosoma* (42.85%) and Hookworm (35.7%). The most of the samples (78.52%) revealed either multiple parasitic infection or triple infection (21.42%) with low to moderate intensity of infection. In conclusion, The parasitic burden in Red Pandas of Illam, community forest is very high hence urgently needs to address this threat in Red panda conservation action plan.

Introduction

The Red Panda (*Ailurus fulgens* Cuvier, 1825) is the one living species of family aeluridae (Shrestha 2015) and commonly called Cat bear, Panda bear, Lesser Panda etc. There are two sub species of the genera that is *Ailurus fulgens fulgens* and *Ailurus fulgens styani*. *Ailurus fulgens fulgens* is smaller and lighter than *Ailurus fulgens styani*. Red Panda is an endemic sepsis of Himalayan mountain region (Roberts and Gittleman 1984) which threatened with extinction worldwide (Wang et al. 2008). It inhabits in the temperate zone at low temperatures (Yonzon 1989) in the countries of the Himalayan Mountain Range which includes Nepal, India, Bhutan, Myanmar and China (Roberts and Gittleman 1984).

The Red Panda resides in evergreen, deciduous, and mixed forests with dense bamboo covered (Roberts and Gittleman 1984, Wei et al. 1999, Choudhury 2001, Pradhan et al. 2001). Despite being a member of the order Carnivora, Red Panda is a specialized herbivore with a low nutrient diet. More than 86% of its diet includes ringal bamboo (Pradhan et al. 2001, Yonzon & Hunter 1991), which has forced the animal to inhabit in narrow range of forest types and restricted geographic area (Yonzon 1989, Choudhury 2001, Pradhan et al. 2001b, Sharma and Belant 2009). Because of the specialized diet and narrow range of habitat, Red Panda has been considered as an indicator species of ecosystem health in eastern Himalayan broadleaved and conifer forests (Yonzon et al. 2000).

Red Panda was classifies as vulnerable status by IUCN, that suggest a likely extinction globally if conservation measures are not initiated soon (Wang et al. 2008) and by Convention on International Trade.
in Endangered Species of Wild Fauna and Flora (CITES) in its Appendix I (species threatened with extinction which are or may be affected by trade). In the present days, the population of Red Pandas are declining day by day and occurs with a patchy distribution due to habitat fragmentation, loss of foraging habitat, human and livestock disturbances, poaching, and disease (Yonzon and Hunter 1991b, Wei et al. 1999, Choudhury 2001, Patterson-Kane et al. 2009, Sharma and Belant 2009, Dorji et al. 2012, Sharma et al. 2014). Parasitic diseases is posing still neglected but significant conservation threat of Red panda (Zhang et al. 2007, Thomas 2002) worldwide including Nepal. The most of the Parasitic infection negatively impact on body weight gain, quality of reproduction due to loss of appetite, nutrient uptake and utilization (Gross et al. 1999) and even death (Rao and Acharjyo 1984, Hansen and Perry 1994). Since the Red panda population is very less and in declining trend in Illam community forest, the present study was designed to assess the parasitic burden in Red panda.

Materials and Methods

The study was carried out at Maimajhuwa, Mabu, Jamuna and Jogmai VDCs of Illam. It was designed to cover all the Red Pandas habitat of the community forest of Illam. Using GIS system line transect grids were selected from the elevation ranged 2200-4800m with the grid size 1.7 × 1.7 (2.89) Km² while in each grid two transects were made. First transect was 500m apart from starting point of grid and second transect was 700m apart from first transect. Samples were collected from transects and also by opportunistically from the study area. Altogether 14 faecal samples were collected from the study area and preserved in 2.5% Potassium dicromate. The faecal samples were examined in the laboratory of CDZ by both direct smear and concentration methods (floatation and sedimentation) for detection of intestinal parasites as well as stoll’s count for intensity of parasites of Red Panda. The size of the eggs, oocyst and cyst measured using oculomicrometer. The identification of the parasitic oocyst, cyst, egg and larva were done on the basis of shape and size along with published literature (Barutzki and Schaper 2009, Bhir 1998, Villeneure 2013, Brianti et al. 2012).

Results

All the samples of Red Panda collected from study area were found positive for parasitic infection. From the examination 10 different genera of parasites (Three protozoan and seven helminth) were observed. Among the protozoan parasites coccidian; Eimeria sp. showed the highest prevalence (64.28%) followed by Entamoeba sp. (57.14%) and Balantidium sp. (14.28%). Study revealed the existence of several species of coccidian parasites, which were grouped into two broad groups; Eimeria with micropyle and without micropyle. The Red Panda were found to be almost equally infected with both groups of this parasites (Table: 1).

<table>
<thead>
<tr>
<th>S.N</th>
<th>Class</th>
<th>Name of Parasite</th>
<th>Prevalence Rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Sarcodina</td>
<td>Entamoeba sp.</td>
<td>8(57.14%)</td>
</tr>
<tr>
<td>2.</td>
<td>Sporozoa</td>
<td>Eimeria with micropyle</td>
<td>9(64.28%)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Eimeria without micropyle</td>
<td>8(57.14%)</td>
</tr>
<tr>
<td>3.</td>
<td>Litostomatea</td>
<td>Balantidium sp.</td>
<td>2(14.28%)</td>
</tr>
</tbody>
</table>

Helminth parasites observed in the Red Panda of Illam community forest includes only nematode parasites, while cestode and trematode were not observed. Oxyuris sp. (100%) was the most predominant parasites among the seven genera of nematode parasites followed by Baylisascaris sp. (57.14%).
Trichostrongylus sp. (50%), Strongyloides sp. (50%), Crenosoma sp. (42.85%), Trichuris sp. (42.85%) and Hookworm (35.71%).

**Figure 1.** Prevalence of nematode in Red Panda

Out of 14 samples, 78.57% were found to be co-infected by different genera, 21.42% by triple genera while single infection and double co-infection were absent (Fig: 2).

Heavy parasitic infection was considered in those samples which has six or more ova or oocyst observed per field. Maximum five samples of Red Panda showed heavily infected with coccidian parasite; *Eimeria*. While two each samples showed high intensity of *Oxyuris*, *Trichostrongylus* and *Baylisascaris*.

**Figure 2.** Mixed infection on Red panda
Table 2. Intensity of infection of intestinal parasite in Red Panda

<table>
<thead>
<tr>
<th>S.N.</th>
<th>Class</th>
<th>Name of Parasite</th>
<th>+</th>
<th>++</th>
<th>+++</th>
<th>++++</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Sarcodina</td>
<td>Entamoeba sp.</td>
<td>3</td>
<td>4</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>2.</td>
<td>Sporozoa</td>
<td>Eimeria with micropyle</td>
<td>-</td>
<td>1</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Eimeria without micropyle</td>
<td>1</td>
<td>4</td>
<td>2</td>
<td>-</td>
</tr>
<tr>
<td>3.</td>
<td>Litostomatea</td>
<td>Balantidium sp.</td>
<td>2</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>4.</td>
<td>Nematoda</td>
<td>Oxyuris sp.</td>
<td>2</td>
<td>6</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td>5.</td>
<td></td>
<td>Strongyloides sp.</td>
<td>4</td>
<td>2</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>6.</td>
<td></td>
<td>Hook worm</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>7.</td>
<td></td>
<td>Crenosoma sp.</td>
<td>3</td>
<td>1</td>
<td>2</td>
<td>-</td>
</tr>
<tr>
<td>8.</td>
<td></td>
<td>Trichostrongylus sp.</td>
<td>2</td>
<td>1</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>9.</td>
<td></td>
<td>Baylisascaris sp.</td>
<td>3</td>
<td>2</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>10.</td>
<td></td>
<td>Trichuris sp.</td>
<td>4</td>
<td>1</td>
<td>1</td>
<td>-</td>
</tr>
</tbody>
</table>

**Discussion and Conclusion**

Wild life disease research in Nepal is very much limited particularly in Red panda. Available data (Lama et al. 2015, Shrestha 2015) emphasized that parasitic burden is becoming a major conservation threat in Rolpa and Rara National Park, Mugu. The small number of Red panda has been found to be distributed in Community forest of Illam district. A total of 14 faecal samples of Red Panda were collected from the community forest of Illam and examined by concentration methods. All the samples (100%) were found to be positive for both protozoan and helminthes parasites. This prevalence rate of Red Panda was almost similar as compared to 93.02% reported in Red panda from Rara National Park (RNP) (Shrestha 2015) and 100% in Kothi Bhir community area (KBCA), Rolpa (Lama et al. 2015). But higher than the reports of Bartelsen et al. (2010) and Pradhan et al. (2011) which showed 35% and 46.66% parasitic infection from European zoo and Darjeeling, India respectively.

From the economic and sanitary point of view, coccidian parasites are the most prevalent among protozoa. *Eimeria* is the most common coccidian parasites among wildlife and livestock. The prevalence of *Eimeria* with micropyle and without micropyle in Illam almost similar with *Eimeria* reported from RNP (Shrestha 2015). High prevalence of *Eimeria* infection has been also reported from Raccoons of America (Dubey et al. 2000, Wright and Gompper 2005 and Foster et al. 2004).

Besides the coccidian parasites, the Red Panda were found to be infected with two other protozoan parasites, *Entamoeba* sp. and *Balantidium* sp. Amoebic dysentery, an intestinal disease caused by infection with the protozoan parasite *Entamoeba* sp. is an important disease of man and animals throughout the world. *Entamoeba* sp. had also been reported from Red Panda of RNP (Shrestha 2015). In both RNP and Illam, Red Panda were infected with more than 50% by *Entamoeba* species.

*Balantidium coli* is the ciliate zoonotics protozoan parasites. Non-human primates have been considered the most important reservoirs for human infection (Nakauchi 1999). *Balantidium* sp. has been reported from different animals by Nakauchi (1999) but this is the first case to report the *Balantidium* sp. in Red Panda in the global context with prevalence rate 14.28%. The first report on isolation and maintenance of *B. coli* was done by Barrett and Yarbroug (1921) in animals. *B. coli* are a ciliated and a normal inhabitant of intestine of wild and domestic animals, probably capable of becoming somewhat pathogenic under favorable condition. It has been identified by Varadharajan and Kandasamy (2000) from India. The
infection of *B. coli* may be due to the contamination of water or food with cyst in the grazing area (Schuster and Ramirez 2008).

Generally wild animals become infected with nematode, cestode and trematode helminth parasites. To compared the life cycle of cestode and trematode, a suitable intermediate host is required but not for most of the nematode parasites. Interestingly, Red Panda of Illam were found infected with only nematode parasites but livestock were infected by cestode and trematode too.

However the trematode, *Ogmocotyle ailuri* was previously described from Red Panda at a zoo in the America (Price 1954 and 1960). *O. ailuri* also isolated from the small intestine of Taiwanese monkey (*Macaca cyclopis*) (Yoshimura et al. 1996) and Japanises monkey (*Macaca fasciculata*) (Iwaki et al. 2012). Another trematode, *Heterobiharzia americana* also recorded in Archer and Wichita countries of north contra Texa and overall prevalence was 47.2% (Kelley 2010) and other trematodes *Alaria* sp., *Digenea* sp. and *Eurytrema procyonis* were observed in Raccoon (Wright and Gompper 2005). Prevalence of trematode were found 13% in Red Panda from KBCA (Lama et al. 2015) but genera was unidentified. Absent of trematodes in present study might be due to absent of suitable intermediate host in Illam. Trematode infection was most common among livestock (Bandyopadhaya et al. 2010, Yadav and Tando 1989, Byanju et al. 2011, Kanyari et al. 2009, Chouwdhary et al.1993, Pathak 2011).

Cestode infection in Red Panda (Lama et al. 2015) had shown from KBCA similarly the Red Panda of RNP have been reported to be infected with *Moniezia*, a common herbivore cestode parasite but none of the Red Panda samples collected from Illam were positive. However three genera of cestode had been reported from Raccoon in Archer and Wichita countries of North Central Taxas including *Atrioenia procyonis*, *Mesocestoides spp.*, *Taenia pisiformis* (Kelley and Horner 2008). Altogether seven genera of nematodes in Red Panda were observed from Illam community forest. Among them *Oxyuris* sp. showed the 100% prevalence which was highest than 58.14% recorded by Shrestha (2015). The highest prevalence rate of *Oxyuris* may be due to the cool climate of the area. Cool climate is suitable for the development of *Oxyuris* larva.

*Baylisascaris* is an important intestinal nematode of Red Panda as well as Raccoon. This parasite had been recorded from Spain in white-headed lemurs (*Eulemur albifrons*) (Martinez et al. 2015), North America (Kazacos 2001), Germany (Bauer et al. 2011). In this study the prevalence of *Baylisascaris* was found 57.14% which was higher than 38.88% and 13.04% prevalence rate of *Baylisascaris* reported in Red Panda from Rara National Park, Mugu, Nepal (Shrestha 2015) and Kothi Bhir Community area, Rolpa, Nepal (Lama et al. 2015) respectively. *Baylisascaris* is found in Red Panda, Gient Panda, Raccoon, Cat, Dog etc. Nematodes like, *B. procyonis*, *Capillaris acrophili*, *C. plica*, *C. procyonis*, *C. putorii* and *Placoconus lotoris* had been reported in Raccoons from Southern New York (Wright and Gompper 2005). Similarly, *B. procyonis* was also reported in Raccoons from Western North Carolina (Hernandez et al. 2012). The highest prevalence rate were recorded from North-Eastern, mid-western, mid-Atlantic, some western states (California, Washington, Oregon and Coloradol and some region of Taxas (Kazacos 2001, Long et al. 2006, Chavez et al. 2012).

During the study, *Trichostrongylus* sp. was recorded for first time from Red Panda in the global context. The prevalence of *Trichostrongylus* sp. and *Strongyloides* sp. in Red Panda was found 41.46% and 50% respectively. *Strongyloides* sp. had been recorded in Red Panda by Shrestha (2015) and prevalence was low compared to present study. The parasite was already recorded in American Raccoons from New York (Wright and Gompper 2005).
Trichuris is another common nematode parasite prevalent in Red Panda. The parasite has been reported from RNP (Shrestha 2015) and KBCA (Lama et al. 2015). The prevalence of Trichuris was 42.85% which was higher than 4.65% and 26.08% recorded by Shrestha (2015) and Lama et al. (2015) respectively. High worm load may cause growth retardation, anemia and hemorrhagic diarrhea (Hale and Stewart 1979).

Crenosoma spp. is the Metastrongylus lungworm infecting wild and domesticated canids in Europe (Morgan et al. 2005, Traversa et al. 2010). Recently, emergence of this parasite was observed in several European countries (Traversa et al. 2010) due to population increase and urbanization of Red foxes (Vulpes vulpes) (Deplazes et al. 2004) which is the major reservoir hosts of this parasite in Europe. The prevalence of Crenosoma was found 42.85% in Red Panda which was almost similar 34.88% by Shrestha 2015 from Rara National Park, Mugu, Nepal and higher than 4.3% from European zoos (Bertelsen et al. 2010). Hook Worm infection has been reported from Red Panda of RNP (Shrestha 2015) with prevalence rate 44.19% which was almost similar with present study which revealed 35.7%. Hookworms are cosmopolitan in distribution (Bowman et al. 2003) and can be transmitted orally but also by cutaneous penetration and cause high mortality in animals and human (Hotez et al. 2004).

Angiostrongylus vasorum was recorded in Red Panda from different countries. It is a most important lungworm which causes pathogenic pneumonia to Red Panda. A. vasorum was recorded from Denmark, U.K and European zoos by Bolt et al.(1992), Janet et al. (2009), Bertelsen et al. (2010 ) respectively. Angiostrongylus sp. was also reported in Nepal (Lama et al. 2015 and Shrestha 2015) but during this study A. vasorum was not recorded. Aeurostrongyloid spp. have been reported in carnivora by different researchers in global and national context. In the present study, none of these nematodes were isolated in the faecal matter of Red Panda of Illam community forest, Nepal.

Among 14 samples, multiple infection was found to be highest (78.57%) in Red Panda followed by double (21.42%) which was similar with Shrestha (2015). No single infection was observed during study. The intensity of different parasites in Red Panda of Illam community forest were observed in this study. According to result maximum number of Red Pandas were found to be infected with light infection. Some of the faecal samples of Red Panda found to be positive for heavy infection by Eimeria, Oxyuris, Trichostrongylus and Baylisascaris. The heavy infection indicates symptomatic condition causing serious diseases in Red Panda. In general, the parasitic burden in Red panda of Community forest of Illam showed very high posing an important conservation threat.

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Free living soil Nematodes as Ecological Indicators

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Abstract

Free living Nematode communities used as bioindicators of soil health because their composition well correlates with, two critical ecological processes i.e. nitrogen cycling and decomposition in soil. Nematodes indices withstand statistical rigor better than abundances, proportions, or ratios of trophic groups. Nematodes indices respond to a variety of land-management practices, based largely on life history characteristics of families.

Key words: soil, bioindicator, nematodes indices, monitoring, trophic diversity

Introduction

The importance of soil organisms for maintaining soil functionality and regulating processes that support ecosystem services, like nutrient and water retention, carbon storage and pest resistance, is widely recognized (Wall 2012 and Mulder et al. 2011). Among soil biota nematodes are among the most thoroughly studied groups as they combine several unique characteristics. They occur in high diversity and density in every soil and sediment type (Treonis and Wall 2005 and Wu et al. 2011) and are very diverse in terms of trophic preferences, represent all trophic groups and levels in the soil food web (Bongers and Ferris 1999 and Yeates 2003) and have variable life history strategies (Bongers 1990). They are involved in fundamental ecological processes like decomposition and nutrient cycling (de Goede and Bongers 1994) and their functional composition is indicative of the major channels of matter and energy transfer across the decomposition pathways. Nematodes have a permeable cuticle, which allows them to respond with a range of reactions to pollutants (Wasilewska 1994). Some nematodes have resistant stages such as cryptobiotic forms or cysts that allow them to survive inactively during unfavorable environmental conditions for growth. However, some nematode taxa such as Dorylaimidae have no resistant stages, which may make them more sensitive to environmental change (Bongers 1999). Thus, nematode community structure is a sensitive indicator of the soil status and environmental disturbances (Bongers and Bongers 1998) and nematodes are among the most preferred bioindicators of soil condition (Bongers and Ferris 1999, Zhao and Neher 2005). The composition of soil nematode communities is influenced by environmental variables such as vegetation, soil type, season, soil moisture level and soil organic matter content (Goralczyk 1998). In many cases there are strongly related with specific microhabitats or with specific hosts in the case of parasitic nematodes (Pinto et al. 2006).

Feeding groups (Trophic groups)

The allocation of nematodes to feeding groups is an effective method to condense information. Yeates et al. (1993) published a synthesis in which the following groups were distinguished: 1. Plant feeding (Sedentary parasites, Migratory endoparasites, Semi-endoparasites, Ectoparasites i.e. Epidermal cell and root hair feeders, Algal and moss feeders, Feeders on plant tissues eg- Tylenchidae, Psilenchidae, Atylenchidae; Tylenchus, Laimaphelenchus, Anguiniidae). 2. Bacterial feeding group i.e. cephalobidae,
Plectidae etc 3. Fungal feeding group eg. Aphelenchidae - Aphelenchoides 4. Predators groups eg-Mononchidae, Longidoridae 5.Omnivorous (both predators as well as plant feeders)eg- Qudsianematidae, Thornematidae etc.

**Colonizer persister (C-P)**

Based on life strategy Bongers (1990) allocated terrestrial and freshwater nematodes on a continuum from colonizers to persisters (r- to K-strategists) followed by a similar proposal for marine nematodes (Bongers et al., 1991). The following groups can be distinguished on the colonizer-persister (cp) scale. These are used to indicate the soil status.

cp-1 group: Nematodes with a short generation time, producing many small eggs resulting in an explosive population growth under food-rich conditions, for example, bacteria blooming in enriched soils. They are relatively tolerant to pollution-induced stress. for example, Rhabditidae. Obviously they have a high metabolic activity. These enrichment opportunists show a phoretic relation with insects and other vectors and are only active under transient conditions of high microbial activity, they form

cp-2 groups: Nematodes with a short generation time and a high reproduction rate, but do not form dauerlarvae. They occur under food-rich as well as food-poor conditions and are very tolerant to pollutants and other disturbances. This group is composed of the smaller tylenchids, mainly feeding on epidermal cells; The fungal feeding aphelenchoids and anguinids and the bacterial feeding cephalobids, plectids and monhysterids.

cp-3 groups: They have a longer generation time and are relatively sensitive to disturbances. This group is composed of the bacterial feeding teratocephalids, the Araeolaimida and Chromadorida; the larger tylenchid nematodes that feed on deeper cell layers in the roots; the diphtherophorids, assumed to feed on fungi, and the carnivorous tripylids.

cp-4 groups: Small dorylaimids and large non-dorylaimids. These nematodes are characterized by a long generation time, permeable cuticle and sensitivity to pollutants. The non-carnivorous nematodes in this group are relatively sessile, whereas carnivorous have to move. This group is composed of larger carnivores, the bacterial feeding Alaimidae and Bathyodontidae, the smaller Dorylaimid nematodes and the plant feeding trichodorids.

cp-5 groups: Large dorylaimid nematodes with a long life span and low reproduction rate; both probably a corollary of low metabolic activity, highly sensitive to disturbance.

**Nematode community indices**

For the evaluation of soil conditions, well documented nematological indices have been developed for this purpose (Bongers 1990, 1993, Bongers and Ferris 1999, Ferris et al. 2001, Verschoor et al. 2001, Yeates 2003, 2007) and successfully applied to monitor land use changes, management effects, environmental disturbance and pollution among others (Tsiafouli et al. 2007; Vonk et al. 2013). According to Neher et al. (2005), nematode indices are cost-effective, easy to calibrate and interpret.

**Nematological indices**

**Maturity index (MI):** Bonger (1990) proposed maturity index for free-living taxa. It is viewed as a measure of disturbance, with smaller values being indicative of a more disturbed environment and larger values characteristic of a less disturbed environment. Yeates’ (1994) proposed modification of the index based on merging free-living and plant parasitic nematodes (PPI) for the summed of (SMI).
**Enrichment index (EI).** Enrichment index (Ferris et al. 2001) is based on the expected responsiveness of the opportunistic guilds (bacterivorous nematodes with c-p1) to organic resources enrichment. The enrichment-opportunistic bacterivorous nematode guild includes species in the families Rhabditidae, Panagrolaimidae and Diplogasteridae (Bongers and Ferris 1999, Ferris et al. 2001). Therefore, EI describes whether the soil environment is nutrient enriched (high EI) or depleted (low EI). Food webs become enriched when disturbance occurs. Resources become available due to external input, organism mortality, turnover, or favorable shifts in the environment.

**Structure index (SI):** The SI (Ferris et al. 2001) represents an aggregation of functional guilds with c-p values ranging from 3-5 and describes whether the soil ecosystem is structured with greater trophic links (high SI) or degraded (low SI) with fewer trophic links. Similarly, Plotting of EI vs. SI provides a model framework of nematode faunal analysis as an indicator of the likely conditions of the soil food web.

Channel index (CI). The CI (Ferris et al. 2001) is a percentage of fungivores among the total fungivores and c-p 1 bacterivorous. It indicates predominant decomposition channels in the soil food web. A high CI (> 50 %) indicates fungal decomposition channels whereas low CI (< 50 %) suggests bacterial decomposition channels. The CI also provides a means of tracking succession between fungivorous and bacterivorous nematodes as organic resources are supplied and depleted in agricultural systems. Decomposition rates of readily degraded material in bacterial pathways are expected to be faster than that in fungal pathways where materials may be more complex. Due to the similarity of C/N ratios of fungi and fungivorous nematodes, mineralization rates in fungal channels should be slower than those in bacterial channels.

**Basal index (BI):** The BI is based on the proportion of bacteria-feeding and fungi-feeding nematodes with a c-p value of 2. It also indicates predominant decomposition channels in the soil food web.

**Nematode Metabolic footprints (MF) indices:** These are the more recent set of indices (Ferris, 2010) that refer to carbon utilization by nematodes. The metabolic footprint of component taxa, is the sum of the lifetime amount of C partitioned into growth, egg production and respiration which is the amount of C utilized in metabolic activity. MF gives insight to the nature but also the magnitude of soil functions. MF are calculated for each trophic group (bacterivore MF, fungivore MF, herbivore MF, omnivore MF and predatory MF).

**References**


Autoimmune Diseases: An Introduction

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Abstract

The immune system recognizes and eliminates foreign agents, and protects the host against infection. Autoimmunity is a natural phenomenon where self-reactive antibodies and autoimmune cells are present in all individuals. A combination of genetic predisposition and environmental factors contribute to the development of autoimmune diseases. Autoantibodies attack structures within individuals that produce them. Autoimmunity is a major cause for a number of serious and fatal diseases. Presence of one autoimmune disease increases the chance for simultaneously developing other autoimmune diseases in the same person.

Key words: Antibodies, Immune system, Molecular mimicry, polygenic, self-reactive

Introduction

An immune system is a complex network of special cells and organs that defend the body from pathogens, such as viruses, bacteria, and parasites, and keeps us in a healthy state. The immune system has a unique ability to differentiate between self and non-self, i.e., it destroys the non-self proteins without attacking body’s own (self) proteins. However, autoimmunity occurs to some degree in all individuals. When the immune system fails to differentiate between ‘self’ and ‘non self’, it can attack body’s own proteins, thereby causing autoimmune disorders. Patients with autoimmune diseases frequently have unusual antibodies circulating in their blood that target their own body tissues. Autoimmune diseases can affect any part of the body, and have a myriad of clinical manifestations that make diagnosis extremely difficult. Most autoimmune diseases disproportionately affect women. For diseases such as thyroiditis, scleroderma, lupus, and Sjögren’s syndrome, more than 85 percent of patients are female.

Autoimmune disease is an illness that develops when an individual’s immune system attacks its own body organs or tissues. The exact cause of autoimmune diseases is unknown, although some individuals are genetically more susceptible to developing autoimmune diseases than others. This mostly happens when certain genes start showing abnormalities. These genes could be of antibodies, T-cell receptors or major histocompatibility complex (MHC). In certain types of autoimmune diseases, such as rheumatic fever, a bacteria or virus may trigger an immune response and the antibodies or T-cells attack normal cells because the normal cellshave parts similar to that of an infecting germ (molecular mimicry).

Most autoimmune diseases are thought to be polygenic, involving more than one gene. Evidence from clinical reports suggests that patients with a family history of autoimmune diseases are more likely to develop an autoimmune disease. For example, patients with the autoimmune thyroid diseases, Graves’ disease or Hashimoto’s thyroiditis, have a family history of those diseases. Patients with autoimmune
thyroid disease are also more likely to develop other autoimmune diseases such as systemic lupus erythematosus (lupus), pernicious anemia, type I diabetes or Addison disease.

External environmental factors such as hormones, diet, pesticides, drugs, toxins and/or infections are important in determining whether or not an individual will develop autoimmune diseases. Environmental agents are able to amplify autoimmunity in genetically susceptible individuals and to break tolerance in genetically resistant individuals, thereby increasing the risk of developing autoimmune disease. C-reactive proteins (CRPs), produced by the liver, are an essential part of the immune system. The levels of CRPs are elevated in almost all autoimmune disorders. Since the whole immune system dysfunctions, a person suffering with an autoimmune disorder will show increased susceptibility to any bacterial infections.

Most autoimmune diseases are more common in women than in men. Conservative estimates indicate that nearly 80% of individuals with autoimmune diseases are women. Exceptions include diabetes mellitus, ankylosing spondylitis and inflammatory heart disease, which occur more frequently in men. Autoimmune diseases are among the tenth leading causes of death among women, in all age groups up to 65, in USA. No data is available for Nepal. It is predicted that estrogen in women may influence the immune system to predispose them to autoimmune diseases. Furthermore, presence of one autoimmune disease increases the chance for simultaneously developing other autoimmune diseases in the same person.

**Some Common Autoimmune Diseases**

**Acute disseminated encephalomyelitis (ADEM)** (or acute demyelinating encephalomyelitis): ADEM is a rare autoimmune disease marked by a sudden and widespread attack of inflammation in the brain and spinal cord due to viral infection. ADEM also attacks nerves of the central nervous system and damages their myelin insulation, thereby destroying the white matter. ADEM's symptoms resemble that of multiple sclerosis (MS), therefore the disease is classified as a multiple sclerosis borderline disease. However, ADEM has several features that are different from MS. Unlike MS, ADEM occurs mostly in children and is marked by rapid fever. ADEM leads to loss of consciousness, coma and death, which is very rare in MS (except for severe MS cases).

**Multiple Sclerosis:** Multiple sclerosis (MS), also known as disseminated sclerosis or encephalomyelitis disseminata, is the most common autoimmune disorder affecting the central nervous system. The name *multiple sclerosis* refers to scars (sclerae—better known as plaques or lesions) in the white matter of the brain and spinal cord. It is a demyelinating disease in which the insulating cover of nerve cells in the brain and spinal cord are damaged. This damage disrupts the ability to communicate, resulting in a wide range of signs and symptoms, including physical, mental, and sometimes psychiatric problems. There is no known cure for multiple sclerosis.

The cause of MS is still unknown. Scientists believe that the disease is triggered by unidentified environmental factor(s) in a person who is genetically predisposed to MS. In multiple sclerosis (MS), myelin sheaths around nerve fibers of the central nervous system (CNS) are damaged. Such damage interferes with transmission of nerve signals between the brain, spinal cord and the rest of the body. The myelin sheath is to a nerve cell what wire insulation is to a wire. Both help to conduct electrical signals better. Disrupted nerve signals result in walking problems, numbness, and balance issues. Depending on where the damage occurs, symptoms of MS vary from one person to another, and vary over time for any given individual.
**Ankylosing spondylitis:** It is an inflammatory disease that can cause some of the spine vertebrae to fuse together. This fusing makes the spine less flexible and can result in a hunched-forward posture. If ribs are affected, it may cause difficulty in taking deep breaths. Ankylosing spondylitis mostly affects men. There is no cure for ankylosing spondylitis, but treatments can decrease pain and lessen symptoms.

**Hashimoto's thyroiditis (HT):** Hashimoto’s thyroiditis is an autoimmune disease that affects the thyroid. Thyroid is an important endocrine gland located in front of the neck. It secretes thyroxine (T4) and triiodothyronine (T3) hormones that regulate growth and many important reactions that use energy. Thyroid hormone levels are controlled by thyroid stimulating hormone (TSH), which is secreted by the pituitary gland. With Hashimoto’s disease, the immune system makes auto-antibodies that damage thyroid cells and interfere with their ability to make thyroid hormone. Over time, thyroid damage can lead to low thyroid hormone levels. This is called an underactive thyroid or hypothyroidism. An underactive thyroid slows down various functions of the body such as heart rate, brain function, and the rate your body turns food into energy. Hashimoto’s disease is the most common cause of an underactive thyroid. An enlarged thyroid, called a goiter, is often the first sign of Hashimoto’s disease. The goiter may cause the front of the neck to look swollen. If large, it may cause a feeling of fullness in the throat or make it hard to swallow. It rarely causes pain. Hashimoto’s disease is more common in women than in men.

**Systemic lupus erythematosus (SLE):** SLE is an autoimmune disease that attacks the brain, kidneys, joints, skin, heart, lungs and other organs in the body. People with this problem sometimes have a characteristic ‘malar rash’ on their face. Auto-antibodies produced in this disease attack a vast array of tissue antigens such as DNA, histones, RBCs, platelets, leukocytes and clotting factors. Interaction of these auto-antibodies with their specific antigens produces various disease symptoms. Ultraviolet radiation from sun exposure can exacerbate disease in patients with systemic lupus erythematosus.

**Graves' disease (GD):** Graves' disease is an autoimmune disease that affects the thyroid, a small gland situated in front of the neck. It makes hormones called T3 and T4 that regulate the body’s usage of energy. Production of thyroid hormones is regulated by the thyroid-stimulating hormone (TSH), which is secreted by the pituitary (a pea-sized gland in the brain). In Graves' disease, the immune system makes auto-antibodies that act like TSH, causing the thyroid to make more thyroid hormone than the body needs. This is called an overactive thyroid or hyperthyroidism. An overactive thyroid speeds up every function of the body such as heart rate and metabolism. Graves' disease is one cause of hyperthyroidism. Unlike other causes of an overactive thyroid, Graves' disease can also cause eye changes. For some people with Graves' disease, tissues behind the eyes become inflamed and swollen, causing one or both eyeballs to bulge out of the head and sometimes affects vision.

Both men and women can get Graves’ disease, but women are 10 times more affected than men. Graves’ disease occurs in people of all ages, but most often starts in between ages 20s and 30s. People who get Graves' disease often have family members with thyroid or other autoimmune diseases.

**Myasthenia gravis (MG):** Myasthenia gravis, also known as Goldflam disease, is an autoimmune neuromuscular disease characterized by varying degrees of weakness of the skeletal (voluntary) muscles. In Myasthenia gravis, literally meaning 'grave muscle weakness,' the muscles become easily tired and weak because there is a problem with transmission of nerve impulses to muscles. It occurs when normal communication between nerve and muscle is interrupted at the neuromuscular junction - the place where nerve cells connect to the muscles they control. Normally when impulses travel down
the nerve, the nerve endings release a neurotransmitter called acetylcholine. Acetylcholine travels from the neuromuscular junction and binds to acetylcholine receptors to activate and generate a muscle contraction. In MG, circulating abnormal antibodies block, alter, or destroy acetylcholine receptors at the post-synaptic neuromuscular junction, thereby preventing muscle contraction. These antibodies are produced by the body's own immune system. In MG, certain muscles that control eye and eyelid movement, facial expression, chewing, talking, swallowing and breathing are often, but not always, affected. Muscles that control neck and limb movements may also be affected, however, cardiac muscles are not affected. The disease is treated with immunosuppressant or cholinesterase inhibitors. Although there is no cure, treatment is effective in alleviating symptoms of arm/leg weakness, double vision, drooping eyelids, speech difficulties, chewing, swallowing and breathing. In fact, many people with MG become completely free of symptoms after treatment. It is more common in women under 40 years and men over 60. However, it may affect individuals of any age.

**Diabetes type 1:** Diabetes type 1 is one of the most common autoimmune disorders in many countries of the world including Nepal. It is an insulin-dependent diabetes. Insulin hormone, secreted by beta cells of Islets of Langerhans in the pancreas, controls blood glucose level. The immune system targets and ultimately destroys these insulin producing beta cells, causing insulin deficiency. At the same time, B cells make antibodies against beta cell proteins. When the number of beta cells is reduced by approximately 80%, the body is unable to secrete enough insulin, thereby causing an elevated blood glucose level. High blood sugar, clinically diagnosed as diabetes, can damage eyes, kidneys, nerves, gums and teeth. But, the most serious problem caused by diabetes is heart disease.

**Rheumatoid arthritis (RA):** Rheumatoid arthritis is a common autoimmune disorder in which the auto-antibodies called rheumatoid factors attack joint lining throughout the body. Body joints such as knee or finger joints are attacked and destroyed. People with rheumatoid arthritis have joint pain and face difficulty while moving.

**Goodpasture syndrome** (GS): Goodpasture's disease (also known as Goodpasture Syndrome, anti-glomerular basement membrane disease, anti-GBM disease) is an autoimmune disorder. It was first described by Ernest Goodpasture in 1919. The GS syndrome results when the immune system attacks a particular molecule, the Goodpasture antigen, found in the kidneys and the lungs. Persons with this syndrome develop antibodies that attack a protein called collagen found in tiny air sacs in the lungs and in filtering units (glomeruli) of the kidneys. These antibodies are called anti-glomerular basement membrane antibodies. The glomerular basement membrane, part of the kidneys, helps filter waste and extra fluid from the blood. Anti-glomerular basement membrane antibodies are antibodies against this membrane. They damage the basement membrane and cause rapid destruction of kidneys. Sometimes this disorder is triggered by a viral respiratory infection or by breathing in hydrocarbon solvents. In such cases, the immune system may attack organs or tissues because it mistakes them for these viruses or foreign chemicals.

**Antiphospholipid Syndrome (APS):** Antiphospholipid Syndrome, also known as Antiphospholipid Antibody Syndrome (APLS), Hughes Syndrome, or Sticky Blood, is an immune disorder in which the person's immune system produces antibodies called antiphospholipid antibodies. The abnormal antibodies attack proteins and fats in the blood, specifically phospholipids. Such attacks make the blood excessively sticky and results in a significantly higher risk of developing blood clots.

In APS, thromboses (abnormal blood clots) can develop in any blood vessel of the body, but it is most frequent in the veins and arteries (arterial and venous thrombosis) of the lower limbs. If a blood clot forms in the vessels of the brain, blood flow is impaired and can lead to stroke. Approximately 1 in
every 3 patients with APS has heart valve abnormalities. Patients most commonly develop DVT (deep vein thrombosis) - blood clots that form in the legs; however, clots may also form in the kidneys, lungs and other organs.

Guillain-Barre Syndrome: The immune system attacks nerves that connect the brain and spinal cord with rest of the body. Damage to these nerves make it difficult to transmit signals, as a result, the muscles have trouble responding to the brain. Guillain-Barré syndrome has been associated with a number of bacterial and viral infections.

Vitiligo: Vitiligo, a common autoimmune disorder, is characterized by patchy loss of pigment in the skin and hair. The immune system destroys cells that give skin its color. It can also affect tissue inside the mouth and nose. Vitiligo often clusters in families, and is frequently seen in individuals who have multiple autoimmune diseases.

Addison’s disease: Addison's disease is a disorder that occurs when outer layer of the adrenal glands produce insufficient amounts of cortisol hormone. This occurs when the body's immune system mistakenly attacks the adrenal glands. Addison's disease is also called “adrenal insufficiency” or hypocortisolism. Cortisol helps the body respond to stress, regulates body's use of protein, carbohydrates and fat, maintains blood pressure and cardiovascular function, and controls inflammation. Addison's disease can be life threatening, and can occur in all age groups and affect both sexes.

Aldosterone regulates the amount of salt and water in the body, which is the main way to regulate blood volume and to control blood pressure. When aldosterone levels are low, kidneys cannot keep salt and water levels in balance, thereby causing low blood pressure.

Risk Factors

Autoimmune diseases are a leading cause of death and disability. Some autoimmune diseases are rare, while others, such as Hashimoto's disease, are common. Autoimmune diseases can affect anyone, but people who might be at greater risk are as follows:

- **Women of childbearing age**: More women than men have autoimmune diseases, which often start during their childbearing years.
- **People with family history**: Some autoimmune diseases run in families, such as lupus and multiple sclerosis. It is also common for different types of autoimmune diseases to affect different members of a single family. Inheriting certain genes can make it more likely to get an autoimmune disease. But, a combination of genes and other factors may trigger the onset of a disease.
- **People and their environment**: Certain events or environmental exposures may cause some autoimmune diseases or make them worse. Sunlight, chemicals called solvents, and viral and bacterial infections are linked to many autoimmune diseases.
- **People of certain races or ethnic backgrounds**: Some autoimmune diseases are more common among certain groups of people. For instance, type 1 diabetes is more common in white people. Lupus is more severe in African-American and Hispanic people.

Although each disease is unique, many share common symptoms such as fatigue, dizziness and low-grade fever. For many autoimmune diseases, symptoms come and go, and can be mild or severe. When
symptoms do not appear for some time, it's called remission. Flares are the sudden and severe onset of symptoms.

**Treatments**

There are many types of medicines used to treat autoimmune diseases. Here is a list of available treatments for autoimmune diseases:

- **Relieve symptoms**: Drugs like aspirin and ibuprofen can be used for mild symptoms. Severe symptoms may need prescription drugs to help relieve pain, swelling, depression, anxiety, sleep problems, fatigue, or rashes.

- **Replace vital substances the body can no longer make on its own**: Some autoimmune diseases, like type 1 diabetes mellitus and thyroid disease, can affect the body's ability to make substances that are essential for its function. For example, pancreatic islet cells of type 1 diabetes mellitus patients cannot produce insulin, therefore patients can take insulin to regulate blood sugar. Similarly, patients with autoimmune thyroiditis can be treated with thyroid hormones to restore thyroid hormone levels. These methods do not halt the autoimmune process, although patients may undergo remission while receiving symptom-based treatment. In most cases, however, patients must depend on replacement therapy throughout his or her lifetime. Sometimes a damaged organ can be replaced by transplantation. For example, scientists are now testing the effectiveness of islet cell transplant as a treatment for diabetes.

- **Use of immunosuppressants**: Some immunosuppressive drugs can suppress auto-immune response and ameliorate disease manifestations. However, because these drugs also reduce the individual’s resistance to infection, they must be used with great caution. Additionally, they often have adverse side effects. Such treatments are most often used for debilitating diseases such as lupus and rheumatoid arthritis. These treatments provide non-specific suppression of the immune system, and do not distinguish between a pathologic autoimmune response and a protective immune response. Immunosuppressive drugs such as azathioprine, cyclosporin and corticosteroids are often given to reduce severity of autoimmune disorders. Azathioprine (Imuran) is not only used to prevent organ rejection in kidney transplants, but also in treatment of rheumatoid arthritis. Cyclosporin (Sandimmune, Neoral) is used for multiple sclerosis, diabetes and myesthenia gravis. Mycopehnolate is used to prevent kidney problems associated with lupus erythematosus.

- **Plasmapheresis**: Plasmapheresis is a process in which blood is separated into cells and plasma (liquid) by centrifugation. The plasma components such as autoantibodies, which can cause autoimmune disease, are removed.

- **T-cell vaccination**: Vaccination using T-cells prevent activation of autoreactive T-cells. It is an effective way of treating autoimmune diseases.

- **Monoclonal antibodies**: Monoclonal antibodies decrease T or B cells, act on activated T cells, inhibit particular cytokine mediators of inflammation, or block recruitment and localization of lymphocytes to the target organ. Although these targeted approaches usually have fewer side effects, they may increase a patient’s vulnerability to infection, and therefore must be used with caution.

- **Stem cells**: Adult hematopoietic stem cell transplantation can be done. Stem cell therapies might allow replacement or repair of damaged organs.

**Prevention**
A person with an autoimmune disease can follow these steps to stay healthy:

- **Eat healthy and well-balanced meals:** Diet should include fruits and vegetables, whole grains, fat-free or low-fat milk products, and lean source of proteins. Saturated fat, *trans* fat, cholesterol, salt, and added sugars must be limited. Healthy and well-balanced meals provide all the required nutrients for the body.

- **Get regular physical activity:** A gradual and gentle exercise program often works well for people with long-lasting muscle and joint pain. Yoga or tai chi exercises may be helpful.

- **Get enough rest:** Rest allows body tissues and joints to repair. Sleeping is a great way to help both body and mind. If you don't get enough sleep, your stress level and your symptoms could get worse. Most people need at least 7 to 9 hours of sleep each day to feel well-rested. Better sleep helps to tackle problems better and lowers risk for illness.

- **Reduce stress:** Stress and anxiety can also trigger symptoms of some autoimmune diseases. So, lead a simple life and cope with daily stressors. Meditation, self-hypnosis, and guided imagery are simple relaxation techniques that might help to reduce stress and lessen pain.

**Conclusion**

Fundamentally, all autoimmune diseases are a consequence of impaired immune function that results from interactions of genetic and environmental factors. These diseases pose a significant challenge to the field of immunology. They are different from other diseases, as our own protein becomes our foe. The treatment of autoimmune diseases is equally challenging as the degree of severity and symptoms vary from person to person. There is no cure for autoimmune diseases, but treatments can decrease pain and lessen symptoms. Furthermore, most autoimmune diseases are chronic in nature, requiring a lifetime of care. It is necessary to understand the mechanisms behind deregulation of immune responses to develop better therapies for treatment and possibly even for prevention of autoimmune diseases.

**References**


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**Abstract**

Goat (*Capra* sp.), an important source of meat is greatly affected by the helminth parasites. The current study aimed to determine the seasonal prevalence of intestinal helminthes parasites using sedimentation and flotation techniques. The overall prevalence of helminthes parasite found to be 58 (54.71%) and 86 (81.13%) for the winter and summer respectively. Fecal samples collected during winter season revealed the presence of trematodes (12.26%), cestodes (6.66%), and nematodes (35.84%) and other 6.31%. Similarly the fecal samples collected during summer season revealed presence of helminth eggs including trematodes (17.92%), cestodes (10.37%) and nematodes (52.83%) nematodes. The prevalence percentages of identified genera of trematodes were *Dicrocoelium* sp. (3.47%), *Fasciola* sp. (15.97%) and *Paramphistomum* sp. (2.77%). The difference in the prevalence of different genera of trematodes during winter and summer were not found statistically insignificant ($\chi^2= 1.325$, p 0.250). Among cestodes the only one genus identified with were *Moniezia* sp. (2.77%) and *Taenia* sp. (9.72%). The difference in the prevalence of different genera of cestodes during winter and summer were not found statistically insignificant ($\chi^2= 2.186$, p 0.139).

Similarly the identified nematode genera included *Toxocara* sp. (16.66%), *Strongyl* sp. (1.38%), *Bunostomum* (2.77%), *Capillaria* (4.86%), *Chabertia* (4.16%), *Cooperia* (2.08%), *Heamonchus* (3.47%), *Oesophagostomum* (8.33%), *Nematodirus* (0.69%), *Ostertagia* (1.38%), *Strongyloides* (6.25%), *Trichuris* (5.55%), *Trichostrongylus* (4.86%) and others (4.86%). Mixed infection was observed in out of a total of 134 (63.20%) mixed infection 54 (50.94%) and 80 (75.47%) samples showed helminthes eggs during winter and summer season were respectively detected. The difference in the prevalence of helminth parasites during winter and summer seasons were found statistically significant ($\chi^2=6.193$, P<0.013, d. f. = 1)

**Key words**: Helminth, Trematodes, Cestodes, Nematodes, Parasite, Prevalence, Sedimentation, Flotation.

**Introduction**

Goat (*Capra* sp.) belongs to family bovine and subfamily caprinae, is one of the important oldest domesticated small ruminant, raised mainly for meat, dairy, and manure. The animal is widely distributed all over the world. The national production is unable to fulfill the need of Nepal; hence significant numbers of goats are imported from neighboring countries. About 74% of goats are brought from India and 26% from different parts of Nepal in the Kathmandu market (3\(^{rd}\) NASc Conv., 1998/1999). Livestock...
farming is an integral part of the farming system and goats contribute substantially in the livestock sector of Nepal.

Goats are very much prone to the infection of gastrointestinal helminthes parasites. The parasites cause more than 20 percent loss in the production and productivity of animal. It is very important to know the status helminthes in various geoclimatic conditions in order to design an appropriate prevention and control strategies. Therefore present study aimed at the determination of the status of gastrointestinal helminthes in buffaloes.

Materials and Methods

The study area “Shivraj Municipality-13” Balanagar is the one of the village of Kapilvastu district of Nepal, where people kept many domestics animals including goats mainly for meat and income generation purpose. It’s about 330 km west for the capital city Kathmandu. The goats are supplied from this place to slaughter house of Khasibazar, Chandrauta, Kapilvastu.

The present study was carried out in May/June (summer) 2012 and December/January (winter) 2012-2013. The site of collection of samples was Shivraj Municipality-13, Kapilvastu, Lumbini.

The study was aimed to determine the seasonal prevalence of helminth parasites in goats. A total of 212 fecal samples including 106 each collected during winter and summer season were collected from the study area and preserved in 2.5% potassium dichromate. The samples were tested at the laboratory of Central Veterinary Hospital, Kathmandu using both sedimentation and flotation method. The eggs of different genera of helminthes were identified according to their size, shapes and other morphological features as observed in microscope.

Results

Out of total 212 fecal samples tested, 144 (67.92%) samples showed the presence of helminth eggs with flotation and sedimentation technique. The seasonal difference of negative samples were found statistically insignificant ($\chi^2= 3.118$, $p=0.077$).

Among the positive samples, the trematode, cestode and nematode eggs were detected in 32 (22.22%), 18 (12.50%) and 94 (65.27%) samples. Out of a total of 106 fecal samples, the helminth eggs were detected in 58 (54.71%) and 86 (81.13%) samples collected during winter and summer season respectively.

A total of 17 helminthes genera identified included 3 trematodes, 2 cestodes and 12 nematodes. The prevalence of each of those helminth parasites has been given in Table 1. The prevalence of helminth was found more during summer 81.13% than in winter 54.71%. The seasonal prevalence of different genera of helminth parasites were found statistically insignificant ($\chi^2= 3.118$, $p=0.077$).

Out of 106 samples tested, 32 (22.22%) samples including 19 (17.92%) and 13 (12.26%) samples tested during summer and winter respectively showed trematode eggs. The 13 trematode positive samples of winter season, included *Fasciola* (11 or 10.37%) and *Dicrocoelium* (2 or 1.88%) sp. Similarly 19 trematode positive samples of summer season include *Fasciola* sp.(12 or 11.32%), *Paramphistomum* sp. (4 or 3.77%) and *Dicrocoelium* sp. (3 or 2.83%). Prevalence of *Fasciola* (15.97%) was found to be the highest followed by *Dicrocoelium* (3.47%) and *Paramphistomum* (2.72%). The seasonal prevalence of various trematodes were found statistically insignificant ($\chi^2= 1.325$, $p=0.25$).

Out of 106 samples tested during summer and winter, altogether 18 (12.50%), including 8 (7.54%) summer and 6 (5.66%) winter samples, were found positive for cestodes. The genera of *Taenia* and
Moniezia were identified in 4 (3.77%) samples. The seasonal prevalence of various cestodes were found statistically insignificant ($\chi^2 = 2.186$, p = 0.139).

Out of the total samples 94 (65.27%) samples showed nematode eggs including Toxocara, Bunostomum, Oesophagostomum, Chabertia, Capillaria, Heamonchus, Cooperia, Strongyloides, Trichuris, Trichostrongylus and Ostertagia. (Table 1). The highest and lowest prevalence was shown by Toxocara (16.66%) and Ostertagia (1.38%) sp. respectively. 6.31% samples showed nematodes that could not be identified. The seasonal prevalence of various nematodes were found statistically insignificant ($\chi^2 = 6.193$, P<0.013, d. f. =1).

Table 1. Prevalence of various helminth genera

<table>
<thead>
<tr>
<th>S.N</th>
<th>Class</th>
<th>Genera of helminth</th>
<th>Prevalence Percentage</th>
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<tbody>
<tr>
<td></td>
<td>Trematoda</td>
<td>Fasciola</td>
<td>15.97%</td>
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<tr>
<td></td>
<td>(Total positive: 32)</td>
<td>Dicrocoelium</td>
<td>3.47%</td>
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<tr>
<td></td>
<td>Cestoda</td>
<td>Paramphistomum</td>
<td>2.77%</td>
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<tr>
<td></td>
<td>(Total positive: 18)</td>
<td>Taenia sp</td>
<td>9.72%</td>
</tr>
<tr>
<td></td>
<td>Nematoda</td>
<td>Toxocara</td>
<td>16.66%</td>
</tr>
<tr>
<td></td>
<td>(Total positive: 94)</td>
<td>Bunostomum</td>
<td>2.77%</td>
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<td></td>
<td></td>
<td>Chabertia</td>
<td>4.16%</td>
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<td></td>
<td></td>
<td>Capillaria</td>
<td>4.86%</td>
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<td></td>
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<td>Oesophagostomum</td>
<td>8.33%</td>
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<td></td>
<td></td>
<td>Heamonchus</td>
<td>3.47%</td>
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<td>Cooperia</td>
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<td>Strongyloides</td>
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<td></td>
<td></td>
<td>Trichuris</td>
<td>5.55%</td>
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<td></td>
<td></td>
<td>Trichostrongylus</td>
<td>4.86%</td>
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<td></td>
<td></td>
<td>Ostertagia</td>
<td>1.38%</td>
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<td></td>
<td></td>
<td>Others</td>
<td>6.31%</td>
</tr>
</tbody>
</table>

A higher rate of mixed infection was found during summer season (134 or 63.20%) as compared to that of winter (54 or 50.94%) samples.

Light infection (+) was found in 5 Fasciola positive samples whereas mild (++) infection was shown by 5 Toxocara positive samples. Moderate infection (+++) was shown by 4 Toxocara positive samples and heavy infection (++++) was shown by 1 Taenia, 1 Fasciola and 1 Toxocara positive samples.

Discussion and Conclusion

Out of total 212 samples tested, 86 (81.13%) and 58 (54.71%) samples showed the presence of helminthes eggs in summer and winter season respectively indicating that the parasites were infecting the goats throughout the years.

Present study showed a prevalence of trematode (22.22%), cestode (11.08%) and nematode (65.27%). Opara et al. (2005) reported a prevalence of trematode (78.4%), cestodes (13%) and nematode (8.7%).
respectively in Nigeria. Presence of suitable temperature and moisture and availability of intermediate host could be the reason behind higher prevalence of certain helminth parasites during summer season.

Present study showed a higher prevalence of nematodes (65.27%), cestodes (11.08%), and trematodes (22.22%) which somehow corroborated to the findings of Ijaz et al. (2008) who showed highest infection rate of nematodes (42.67%) followed by trematodes (16.67%) and cestodes (4%) in Lahore, Pakistan.

A research by Wanjala et al. (2002) in the month of May/June and August /September showed 52% infection of helminths in small ruminants in pastoral community in Kenya.

In the present study, 3 genera of trematodes, 2 genera of cestodes and 14 genera of nematodes were identified. Among trematodes *Fasciola*, was common during both winter and summer season but *Dicrocoelium* was found only during winter season and *Paramphistomum* in summer only. Present study exhibited 10.37% and 11.32% prevalence of fascioliasis during winter and summer season respectively. The increase in the prevalence during summer may be due to increase in humidity and availability of favorable temperature. High prevalence of *Fasciola* was reported from Surkhet among goats (Ghimire 1987) followed by 58% from Chitwan district (Dhakal and Kharel 1988), 31.25% infection from Dhanusa district (Jaiswal 2006). Similarly, the prevalence of *Dicrocoelium* was reported to be 1.88%. that corroborated with the earlier finding of Jithendran (1997). Present study recorded a 2.77% prevalence of *Paramphistomum* in summer samples.

Among cestodes, *Moniezia* was found only in summer and *Taenia* found in both seasons. *Moniezia* has been reported from Kathmandu and Surkhet district among buffaloes, sheep, goat and cattle (Ghimire 1987). In the present study, *Moniezia* has been reported only in summer only.

The findings of present study regarding the prevalence of nematodes included *Toxocara, Bunostomum, Chabertia, Capillaria, Oesophagostomum, Heamonchus, Cooperia, Strongyloides, Trichuris, Trichostrongylus* and *Ostertagia*. However, *Bunostomum* and *Ostertagia* were not observed during winter season. Mankir (2007) reported highest prevalence rate of *Haemonchus* followed by *Trichostrongylus* in goats and sheep in Ethiopia. The difference in the result could be due to the variation in weather conditions and humidity in atmosphere.

Yadav et al. (2005) reported the highest prevalence of *Heamonchus, Trichostrongylus, Bunostomum, Oesophagostomum* and *Strongyloides*.

In the current study, the rate of mixed infection was also observed. Among 144 (67.92%) positive samples, 134 (63.20%) samples were found to have mixed infection. During winter and summer 54 (50.94%) and 80 (75.47%) mixed infection had been shown out of 106 samples. In the current study, the rate of mixed infection was also observed. Among 144 (67.92%) positive samples, 134 (63.20%) samples were found to have mixed infection.

**Acknowledgements**

We are greatly thankful to all persons who helped to complete this study. Special thanks to Anju Bhusal.

**References**


ISOLATION OF PLANT PARASITIC NEMATODES IN PEAR ORCHARD AT CHHAIMALE (PHARPING) VDC OF KATHMANDU

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2- Ashok Bahadur Bam, Central Department of zoology, T.U, Kirtipur, ashokbam@gmail.com

Abstract

Plant-parasitic nematodes are of considerable importance worldwide and their devastating effects on crops have major economic and social impacts. They depend on plants and feed on all parts of the plant including roots, stems, leaves, flowers and seeds. The majority of species of plant-parasitic nematodes live on or around plant roots. Nematode damage of the plant’s root system also provides an opportunity for other plant pathogens to invade the root and thus further weakens the plant. A study was carried out to isolate and identify different genera of plant parasitic nematodes from the rhizospheres of pear plants in one of the major pear producing area of Nepal, Chhaimale VDC of Kathmandu district.

Altogether seven species of root nematodes were isolated from soil samples collected: Tylenchorhynchus sp., Longidorus sp., Helicotylenchus sp., Hoplolaimus sp., Aphelenchus sp., Mononchus sp. and Diplogaster sp. Among isolated species of nematodes Tylenchorhynchus sp., Longidorus sp., Helicotylenchus sp., and Hoplolaimus sp. are parasitic, Aphelenchus sp. is mycophagical and Mononchus sp. and Diplogaster sp. are predatory in nature. Number of nematodes were not significantly different in different season and at different locations.

Keywords: Isolation, Root nematodes, Pear

Introduction

Nematodes (nema= thread in Gr.) are pseudocoelomate, unsegmented worm-like animals, commonly described as filiform or thread-like. At present, nematodes are generally regarded as a separate phylum, the Nematoda or Nemata (De Ley and Blaxter, 2004). They are either free-living or parasites of plants and animals. Of the known nematode species, approximately 50% are free-living species found in soil or freshwater, 25% are marine (found in sea water), 15% are animal parasites, and 10% are known plant parasitic nematodes (Pokharel and Larsen, 2007). Nematodes have evolved to fill almost every conceivable niche on earth that contains some amount of water. They are highly diverse. There are a number of plant parasitic nematodes which depend of root system of plants for feeding. The feeding system damages the plant’s root system and reduces the ability to absorb water and nutrients. Typical nematode damage symptoms are a reduction of root mass, a distortion of root structure and/or enlargement of the roots. Nematode damage of the plant’s root system also provides an opportunity for other plant pathogens to invade the root and thus further weakens the plant. The aboveground symptoms of nematode damage to roots are relatively nondescript, including nutrient deficiency, incipient wilt, stunting, poor yield and sometimes plant death (Lambert and Bekal, 2002).

There are a lot of nematodes parasitic to fruit plants, there is certainly a considerable number of nematodes parasitic to pear plants. These nematodes attack different parts of plant and lowers the production significantly if infested heavily and thus is a limiting factor to production. In order to constrain or even banish this limiting factor in agricultural production, it is vital to identify accurately the nematode pests and to understand their biology. This information may useful to control them and consequently...
increase production. There have been a number of studies done of the nematode diversity of different parts of Nepal but no work has been carried out to study the nematodes associated with pear plants. Present study is thus first of its kind. Present study was thus focused to study the prevalence of nematodes in pear orchards of Chhaimale VDC of Kathmandu district which is a pocket area of pear production in Nepal.

The variety of pear grown in this area is Sand Pear (Pyrus pyrifolia) commonly called as Pharping Naspati in Nepali. It is a suitable variety to be cultivated as cash crop in Nepal and Himalayan states of India. It can be cultivated in mid hills between about 1500 and 2500 meters above sea level. This variety needs comparatively low chilling hours (<1000 hours) and are well adapted in warmer temperate region of Nepal.

Materials and Methods

Present study was carried out in Chhaimale VDC which is the southern most VDC of Kathmandu district. The main agricultural crops in this VDC are maize and millet while major horticultural product are Pear, Cucumber, oil seed, vegetables and Peach. The sample collection was carried out for 6 months divided into two seasons. First study was carried out in summer from March 2012 to May 2012 and second study was carried out in autumn from August 2012 to October 2012.

Soil samples were collected from the rhizosphere of 30 mature pear plants selected randomly at study site with 6 samples from each ward. The soil sample was collected from the rhizosphere of each selected plant from nearly to five feet away from the main trunk of the tree and about 20 – 30 cm below the surface of the soil. The soil was dug by iron rod and hole was made. From this hole about 30 cm depth, about 500 gm of soil with rhizosphere was collected in polythene bag. The samples were analyzed for presence of parasitic and non-parasitic nematodes. The collected soil samples were processed by Cobb’s (1918) modified sieving and decantation technique followed by Baermann funnel technique (Baermann, 1917). For the processing and fixation of collected samples of nematodes mainly two chemicals were used prepared in laboratory. The chemicals used were TAF solution for fixation and Glycerol-ethanol solution for processing. Permanent slides were prepared from the nematodes collected by fixation and collection. The slides were observed under the low power and high power magnification (i.e. 10x, 20x and 40x) and the characters were compared with the taxonomy key of Mai and Lyon, 1975 and arranged by Baldwin, 1990. Data analysis was done using Chi-square test and ANOVA with the help of SPSS V 16.0.

Result and Discussion

Nematode Prevalence

Preparation of permanent sample was done from randomly sampled nematodes one from each sample. Thus, altogether 60 samples of nematodes were studied by preparing slides from the rhizosphere collected from study site among which 30 were from summer and 30 from autumn. Prevalence of different nematode species collected during study period in two seasons is shown in table 1.

<table>
<thead>
<tr>
<th>S. No</th>
<th>Nematodes species</th>
<th>Number in summer sample</th>
<th>Percentage</th>
<th>Number in autumn sample</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Tylenchorhynchus</em> sp.</td>
<td>12</td>
<td>40.00</td>
<td>6</td>
<td>20.00</td>
</tr>
<tr>
<td>2</td>
<td><em>Longidorus</em> sp.</td>
<td>5</td>
<td>16.67</td>
<td>7</td>
<td>23.33</td>
</tr>
<tr>
<td>3</td>
<td><em>Helicotylenchus</em> sp.</td>
<td>7</td>
<td>23.33</td>
<td>4</td>
<td>13.33</td>
</tr>
</tbody>
</table>

Table 1: Nematode species identified in two seasons in pear plants of Chhaimmale
There was significant difference ($\chi^2_{\text{cal}} > \chi^2_{\text{tab}}$) between the total number of nematode species in samples studied in summer. In summer, number of Tylenchorhynchus sp. was highest (40%) followed by Helicotylenchus sp. (23.3%), Longidorus sp. (16.7%), Mononchus sp. (10%) and Hoplolaimus sp. (3.3%), Aphelenchus sp. (3.3%) and Diplogaster sp. (3.3%) in as compared to other species. Whereas total number of different species of nematodes in soil samples collected in autumn also showed no significant difference ($\chi^2_{\text{cal}} < \chi^2_{\text{tab}}$).

There was significant difference ($\chi^2_{\text{cal}} > \chi^2_{\text{tab}}$) among the total number of soil nematodes studied in slides in both seasons. Tylenchorhynchus sp. which is a plant parasitic nematode was recorded in highest number (30%) followed by Longidorus sp. (20%), Helicotylenchus sp. (18%), Mononchus sp. (17%), Hoplolaimus sp. (7%), Aphelenchus sp. (5%) and Diplogaster sp. (3%) in lowest number which is a free living nematode.

Different species of Tylenchorhynchus were found to be parasitic on different types of plants (McSorley, 2013; Zaaki and Mantoo, 2003; Ismail et al., 2002; Upreti, 2000; Pokharel and Regmi, 2000; Golden et al., 1987). Upreti (2000) and Pokharel and Regmi (2000) have recorded Tylenchorhynchus sp. in rice and wheat crops of lowland as well as uplands of Nepal. Zaki and Mantoo (2003) have recorded Tylenchorhynchus sp. in different fruit trees (Cherry, Walnut, Apple, Apricot, Plum and Pear) in Kashmir valley along with other 9 parasitic species of nematodes. Though this nematode is recorded in cereal crops in Nepal, no record of Tylenchorhynchus sp. in fruit trees of Nepal has been found yet. Thus, this may be the first record of Tylenchorhynchus sp. in fruit trees in Nepal. Longidorus sp. also known as needle nematodes are also reported to affect trees as they are responsible for severe decline symptoms in trees (Hashim, 1983) when infested heavily. No record of Longidorus sp. has been reported in Nepal.

Present study recorded relatively higher infestation of Longidorus sp. in pear tree rhizosphere in summer as well as autumn samples. Pokharel and Regmi (2000) and Upreti (2000) have reported this species from rice and wheat fields in Nepal but no report of this genera has been found so far in trees, thus, this may be the first report of Helicotylenchus sp. in pear trees in Nepal. Pokharel and Regmi (2000) and Upreti (2000) have recorded Hoplolaimus sp. from fields with rice and wheat crop systems in Nepal while McSorley (2013) reported that this species is prominent in forest as compared to fields. This study also found Hoplolaimus sp. in rhizosphere of pear trees in both summer and autumn season but in comparatively lower density.

Aphelenchus species are common soil inhabiting nematode feeding on fungi (Barker and Darling, 1965). Aphelenchus avenae have been used successfully as a potential biological control agent for root rot fungi (Barnes et al., 1981). These nematodes may be beneficial rather than harmful if they are of mycophagic species. A detail study at species level is necessary to know about the species and its economic importance. Mononchus nematodes are non-parasitic predatory nematodes that feed on protists as well as other small nematodes (Mankau and Mankau, 1963). There are no records of occurrence of Mononchus sp. in cultivated plants in Nepal thus, this may be first report about occurrence of Mononchus sp. in cultivated fruit tree in Nepal. Diplogaster sp. are one of the less studied predaceous nematodes which may

<table>
<thead>
<tr>
<th></th>
<th>Hoplolaimus sp.</th>
<th>1</th>
<th>3.33</th>
<th>3</th>
<th>10.00</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>Aphelenchus sp.</td>
<td>1</td>
<td>3.33</td>
<td>2</td>
<td>6.67</td>
</tr>
<tr>
<td>6</td>
<td>Mononchus sp.</td>
<td>3</td>
<td>10.00</td>
<td>7</td>
<td>23.33</td>
</tr>
<tr>
<td>7</td>
<td>Diplogaster sp.</td>
<td>1</td>
<td>3.33</td>
<td>1</td>
<td>3.33</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>30</td>
<td>100</td>
<td>30</td>
<td>100</td>
</tr>
</tbody>
</table>

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be beneficial to cultivars. Present study also identified Diplogaster sp. from the specimens collected. This shows that Diplogaster species may be present there for predation on other species of nematodes present in rhizosphere of pear.

**Prevalence of nematodes in different wards**

There were six samples collected from each of 5 wards in study area in both summer and autumn season. Number of nematodes found in samples of 500 gm soil from each site in both seasons is shown in figure 1.

There was no significant difference ($P > 0.05$) in number of soil nematodes in samples collected from pear orchards of different wards in Chhaimale in summer season. Similarly, there was no significant difference ($P > 0.05$) in number of soil nematodes per 500 gm sample from different wards during autumn season among different wards in Chhaimale VDC. Figure 2 shows the average number of nematodes per 500 gm soil sample in two seasons in different wards of study area.

Present study found that number of nematodes per 500 gm soil sample ranged from 4-19 with an average of 9.18±1.54. Aksary et al. (2011) reported that at least 30 nematodes/100gm soil may cause any significant suppress in growth of plant. Some nematodes may cause a damage only when they are present in a larger number i.e. upto 5000 nematodes/100gm soil. Compared with these data, number of nematodes recorded in soil of present study may not be able to cause any significant damage to pear plants. Also, the presence of predaceous nematodes such as Mononchus and Diplogaster may be account for relatively lower density of nematodes in rhizosphere of pear plants.

**Conclusion**

Diversity of nematodes in rhizosphere of pear plants from the pear orchards in Chhaimale VDC of Kathmandu have been studied. Altogether 7 species of nematodes have been recorded during study period of two seasons: Summer and Autumn of 2012. Seven species recorded during study were: Tylencorhynchus, Longidorus, Helicotylenchus, Hoplolaimus, Aphelenchus, Mononchus and Diplogaster. Among the recorded species Tylencorhynchus, Longidorus, Helicotylenchus and Hoplolaimus are parasitic in nature either endoparasite, semi-endoparasite or ectoparasite while Aphelenchus is a mycophagic nematode feeding on fungi and Mononchus and Diplogaster are predatory an wide range of micro-
organisms including some nematodes also. There was no significant difference in occurrence of nematodes in two seasons: summer (pre-monsoon) and autumn (post-monsoon). Number of *Tylenchorhynchus* was significantly higher in summer as compared to other nematodes while in autumn occurrence of *Longidorus* and *Mononchus* was higher relatively. In overall, number of *Tylenchorhynchus* was highest in all sites and *Diplogaster* was lowest. Present study found that number of nematodes per 500 gm soil sample ranged from 4-19 with an average of 9.18±1.54. There was no significant difference in number of soil nematodes in samples collected from pear orchards of different wards in Chhaimale during whole study period.

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We would like to express our sincere gratitude to Head of the Department Prof. Ranjana Gupta and Central Department of Zoology, Kirtipur for providing a chance to carry out this research and for providing laboratory facilities and equipments. We would like to express our sincere thanks to Prof. Arvind kumar Keshari, Associate Prof. Dr. Mahendra Maharjan, Central Department of Zoology and Dr. Suraj Baidhaya, NARC, Khumaltar, for the continuous guidance, critical suggestions, regular inspiration and deep concern throughout the field work without the help of whom this research work could not have been carried out successfully. Finally we would like to appreciate all our friends and colleagues who co-operated during all the works of this research work.

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MCSorley, R. 2013. Effect of forest and field habitat features on abundance of genera in nematode assemblages. Nematology, 15: 947-95


AGONISTIC INTERACTION BETWEEN Rhesus Monkey AND HUMAN AT SWAYAMBHU AND PASHUPATI AREA, NEPAL

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ABSTRACT The study was designed using Scan Sampling and Ad libitum recording to investigate the interaction between Rhesus monkey and Humans in Pashupatinath Temple Area and Swayambhunath Stupa Area for a total of 250 hours (8 hours per day; from 9:00 a.m.-5:00 p.m.). Interactions at Swayambhu were occurring high in midday (1p.m. to 2 p.m.) and at Pashupati interactions were occurring high in morning (10 a.m. to 11 a.m.) and evening (4 p.m. to 5 p.m.). Monkey interacted more for the context of food while humans interacted for recreation purpose. Biting was observed only in Swayambhu area. Agonistic behaviour by human was 44% at Pashupati and 34.7% at Swayambhu and Agonistic monkey behaviour was 23.1% at Swayambhu while 22.4% at Pashupati. Living in commensalism with human agonistic behaviour of monkey was high in response to human behaviour rather than through its initiation and also monkeys’ have devised passive behavior strategy during presence of food. Threat shown by monkey at both places tends to increase in absence of food. Female monkey individuals residing in Swayambhu initiate more encounters (58%) than of Pashupati (28%) area while the overall encounter was accounted for male individual. Female monkey individuals were likely to start an encounter at Swayambhu preferring agonistic behaviour during encounter than male individuals while at Pashupati male monkey individual were likely to prefer agonistic behaviour.

INTRODUCTION

Different types of urban habitat support different kinds of wildlife. Urban wildlife, also referred as a synanthrope, has well adapted and suited themselves with human made environment and lives side by side in association with human element which also often lead to extreme case of conflict. With increased population and fragmentation of habitat, human-wildlife conflict is certainly bound to occur. The phrase ‘human–wildlife conflict management’ is being applied to situations that involve any negative interactions between humans and wildlife (Messmer 2000). Urban wildlife such as Rhesus Macaque (Macaca mulatta Zimmermann, 1780), is exposed to stress condition of human presence creates unlike wild population. Fragmentation in habitat through deforestation has pushed primates into areas of human settlement and agricultural lands leading to conflict (Marsh 2003). These wildlife populations adapt and modify their behaviour according to urban stresses (Ditchkoff et al. 2006).

In Nepal, six species of monkey are recorded with major three species the Rhesus, Assamese spp. and Langur spp. (Chalise 2008). Rhesus monkey has been observed living in close association with humans (Chalise 2008a). The rhesus in Kathmandu is localized basically in religious areas, such as Pashupati, Swayambhunath, Nilbarahi, Thapathali, Sankhu-Bajrajogini etc. Population of approximately 400 monkeys in seven-eight social groups live at Swayambhu, near around in Pashupatinath population of 300 monkeys live, fragmented population of about 40 Rhesus monkeys reside in the Thapathali area and about 100 monkeys in Sankhu-Bajrajogini (Chalise 1998, 2013). They come in close contact with humans for sharing resources such as space and for food as well. In the temple areas such as Pashupati and Swayambhu monkeys are frequently snatching away possessions of humans. The monkeys are not harmed by humans in Nepal due to their religious belief and are hence under some sort of protection. Rhesus Macaque, lives in large group showing continuous interaction among them (Chalise 2008). There exist a type of commensalism interaction between human and monkey.

Agonistic behaviour is defined as “Any activity related to fighting whether aggression or conciliation and retreat” (Scott and Fredericson 1951). Study of agonistic behaviour focuses on behaviour such as threat, aggression and submission, which are found in successional pattern from the beginning to the end of the interactions (McGlone 1986). Such behaviour is studied collectively or any one form of the behaviour is studied alone. In urban areas due to artificial feeding the aggressive competitive behaviour within monkey is also seen to increase (Southwick et al. 1976). Behaviour study of animal and specially primates was systematically initiated since the 70s (Altman 1974). Aggression were generally higher per male monkey than per female by analyzing aggressive acts like threats, chases, attacks, fights directed at other species (principally humans) of Kathmandu (Teas et al. 1982).

Presence of wildlife in urban area had always been of high concern. Moreover when there is presence of wildlife such as Monkey in religious areas their interaction and conflict with humans occurs in regular basis. Monkey has been long observed to create a lot nuisance disturbing human and damaging their property. Study on urban wildlife is minimal and there has been a long gap on works related to conflict between monkey and human.

MATERIALS AND METHODS

From preliminary observation a troop of monkey were selected and the route the troops took to move was marked in study area at Swayambhu (27.7116-27.7177 latitude north and 85.2841-85.2952
longitude East) and Pashupati (27.7041-27.7152 latitude North and 85.3461-85.375 longitude East). The study area, comprising of isolated population of Rhesus Macaque adapted to urban environment, was widely intermixed with human settlement with traces of forest area. Study was carried out over a time period of 250 hours (8 hours a day) making observation points in each area; one where visitors spent time performing various recreational activities and the other place was in the core temple area. Encounters were recorded when both interactor; humans and monkeys, were within the two metre distance. Interaction between rhesus and human related to agonistic encounter was recorded using Scan sampling, along with some ad libitum recording, method useful for direct observation to record rare but fairly obvious behaviours (Altman 1974). Observational study of Hinde and Rowell (1962), Sade (1973) on behavioural description of threat, attack, and fear was combined with personal field recording for data acquisition. Field data were recorded moving constantly and periodically around the designated study site from 9:00 a.m. in the morning to 5:00 p.m. for total of 250 hours.

Cross tabulation method was used in IBM SPSS statistics 21 to measure association between categorical variables such as “Encounter”, “Food” and “Sex”. Each variable was further categorised into two levels. Encounter was categorized into aggression and passive. Food was categorized into present and absent. Sex was categorized into male and female. Interacting individuals were classified into two types, human and monkey. Sex of each type was further separated into male and female. Each association was observed under “P” value. “P” value less than 0.05 lead to rejection of null hypothesis under each model of association.

RESULTS

Observation of Human and Rhesus monkey interaction at two places; Swayambhunath Stupa Area and Pashupatinath Temple Area showed that there were more interactions in Swayambhu area (769) than Pashupati area (510) (i.e. nearly about 3:2 hourly interactions) where Humans initiated most encounter at both sites and the initiation among monkey was seen to be high in Swayambhu while comparing to Pashupati. In both study place monkeys were more interactive for food (78% at Swayambhu and 73% at Pashupati) and humans for recreation purpose (78% at Swayambhu and 75% at Pashupati). Share in encounter by age group is listed in Table 1.

Table 1. Share of encounters according to age group by human and monkey, 2014/015.

<table>
<thead>
<tr>
<th></th>
<th>Monkey</th>
<th>Human</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Adult</td>
<td>Young</td>
</tr>
<tr>
<td>Swayambhu</td>
<td>250</td>
<td>71</td>
</tr>
<tr>
<td>Pashupati</td>
<td>108</td>
<td>51</td>
</tr>
</tbody>
</table>

Hourly interaction in Fig.1 reveals that at Swayambhu interaction gradually increased over time with highest interaction during 1:00-2:00 p.m. (136 encounters) and then decreased until 5:00 p.m. in contrast at Pashupati interaction increased becoming highest at 10:00-11:00 a.m. (83 encounters) and then decreased throughout the day finally increasing from 4:00-5:00 p.m. again (86 encounters).
Fig 1. Hourly encounter at Pashupati (Top) and Swayambhu (Bottom)

Behavioural Aspect

In Swayambhu

23.1% of the total encounter accounted for agonistic monkey behaviour; 11.4% threat, 7.1% attack/charge, 1.5% biting, 3.1% chase. 52.8% of the behaviour was passive behaviour and 24.1% accounted for joyful physical contact initiated by monkey at Swayambhu. Human responses were shown through behaviour such as; chase (30%), watch (23%), flee (20%), retreat (13%), feeding (10%), and leave (4%).

In Human behaviour, 31.2% was allocated for feeding, 27.1% recreational activity of taking photo, 34.7% for agonistic (28.9% for threat and 5.8% chase), 2.5% touch, 2.2% moving close, 2.2% just watch. Passive responsive behaviour of monkey such as eating, flee, no interest, watch, leave, move close, and physical contact accounted to be 27.8%, 18.6%, 17.9%, 6.5%, 1.6%, 2%, 1.6% respectively while agonistic response such as threat (14.6%), attack 5.2%, stare (3.1%) and bite was (1.1%) (i.e. agonistic responsive behaviour was 24%). Furthermore, look within the behaviour of monkey reaction showed that monkeys’ response was high in only two type of human action i.e. taking photo and showing threat.

In Swayambhu, 78.1% of encounter was in presence of food and 21.9% in absence of food. When food was absent, threat was highest with 42.3%, physical contact, 28.2%, charge 14.1%, passive 7%, bite 5.6% and chase was lowest with 2.8% and When food was present, high occurring behaviour was passive behaviour with 65.6%, physical contact 22.9%, charge 5.1%, chase 3.2%, threat, 2.8% and bite was lowest with 0.4%.
Threat in absence of food was the most common and high form of aggression (42.3%) than in presence of food (2.8%) whereas in presence of food passive form of behaviour was high and common. Biting was high in absence of food (5.6%) than in presence (0.4%), chasing by monkey was high in food absence than presence but attacking (14.1%) was more pronounced in absence of food than in presence with 5.1%

**In Pashupati**

Monkey initiation of agonistic behaviour recorded to be 22.4% at Pashupati; 11.2% threat, 10.6% attack, 0.6% chasing. Passive behaviour accounted for 70.2% and physical contact (p.c.) 7.5%.

In Pashupati, humans’ threat behaviour was recorded to be about 37%, feeding monkey 33%, taking photo 11%, chase 7%, touching 7%, moving close 3% and watch 2%. Agonistic human behaviour was 44% (with threat and chase). In responsive monkey behaviour, agonistic response was 23.5% (attack 4% and threat 19.5%) whereas non-agonistic behaviour such as eating, watching, leaving, fleeing resulted to be 75.1% while physical contact 1.9%.

In Pashupati 72.7% of encounter was in presence of food and 27.3%. In absence of food, threat had high share with 34.1%. 29.5% was accounted for passive behaviour, 20.5% physical contact and lowest was attack with 15.9% and within food presence, passive behaviour was dominant with 85.5%, attack 8.5%, threat and physical contact was 2.6% each and chase was least with 0.9%.

Threat in absence of food was the most common and high form of aggression (34.1%) than in presence of food (2.6%) whereas biting, chasing by monkey was not observed in absence of food but attacking (15.9%) was more pronounced in absence of food than in presence with 8.5%.

**Association between variables in interactions**

There was significant association (p<0.05) between Type of initiator and Sex of individual of initiator, Monkey behaviour and availability of food, Type of initiator and availability of food, Type of Encounter and Sex of monkey individual at both area. In addition, Food availability and Type of Encounter is significant at Pashupati but significant association does not exist (p>0.05) between Food availability and Sex of monkey individual in both area, Food availability and Type of Encounter at Swayambhu and Type of Encounter and Sex of monkey individual at Pashupati.

Agonistic behaviour shown by monkey in response to human behaviour was higher (24% at Swayambhu and 23.5% at Pashupati) rather than the initiation of behaviour (23.1% at Swayambhu and 22.4% at Pashupati).

In case of monkeys, female individual residing in Swayambhu were more interactive than male individual and in humans males were more interactive than female. Encounter at Swayambhu and Pashupati showed female monkey at Swayambhu is more interactive than female monkey at Pashupati and monkey from Swayambhu area (41.9%) was more interactive than from Pashupati area (31.2%) where as human from Pasupati area (68.8%) shows high form of interaction than from Swayambhu (58.1%).

At both study place, Swayambhu and Pashupati, interaction between Human and Monkey were mostly passive encounter and most encounter at both places were in absence of food.

**DISCUSSION**

Swayambhu and Pashupati area is religious and natural habitat for rhesus monkey but due to urbanization these area is now mixed with high human presence. Rhesus and Human interaction is
common event in these locations. Humans have made this place a recreational spot for them. Rhesus monkey beside natural food source have adequate amount of food source these days as human are being major source for artificial food. Rhesus adapts to human population by increasing commensalism and thievery (Southwick and Siddqi 1994). Rhesus monkeys have well established its territory in the two studied area in association with urban elements that human has created. Once troop establishes its territory competition for food, space, and high density results in increase of aggressive behaviour (Ciani 1986).

In Pashupati interactions are high during morning and evening whereas in Swayambhu encounters occur during midday. There are more interactions in Swayambhu area than Pashupati area among monkey and human and adult monkey/humans are highly involved in the encounters. In both places monkey initiated interaction has been occurring in presence of food. But female monkey individual in absence and presence are likely to interact most frequently than male individual at Swayambhu whereas unlike Swayambhu male monkey individual at Pashupati are likely to interact most frequently than female. But male and female monkey individual are likely to interact with human in presence of food rather than in absence.

It is well observed now that from both study place agonistic behaviour between human and monkey occurs in absence of food rather than presence and human initiated aggression are likely to occur than monkey. At both places in absence of food human more frequently initiates aggressive encounter than monkey. But in presence of food monkey at Swayambhu are more prone to start aggression also at Pashupati human causes more aggression in presence of food as well.

CONCLUSION

Six type of behaviour is shown by monkey in form of passive and aggressive behaviour i.e. attack, bite, chase, physical contact, threat while humans showed seven types of behaviour; chase, feed, threat, watch, touch, take photo, move close. Monkeys were tolerant to everyday vendors and interacted less often with them. Interaction at Pashupati and Swayambhu follows different time pattern. Monkeys have developed a type of passive strategy around human whenever food is available. Field observations in natural setting provide the basic information needed on behavioural analysis for potential risk assessment from a particular species to humans. So, these kinds of behavioural study under naturalistic conditions helps in sound management and develop conservation strategies to resolve the human-wildlife conflict.

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