CHAPTER I

INTRODUCTION

Intestinal parasitosis continues to be one of the major causes of public health problems in the world, particularly in developing countries. It has been estimated to affect 3.5 billion people globally and 450 million are thought to be ill as a result of these infections, the majority being children (WHO, 2000). Approximately 10% of the world's population is suffering from amoebiasis. *A. lumbricoides*, hookworm and *T. trichiura* have been estimated to infect 250 million, 151 million and 45 million people, respectively and thousands of deaths are associated to these infections (WHO, 1997).

Nepal is a landlocked and underdeveloped country located in South Asia. Intestinal parasitosis is one of the major public health problems in Nepal (Rai SK, Asian Parasitology, 2004). The reported prevalence varies considerably approaching nearly one hundred percent in some areas (Rai & Gurung, 1986, Estevez et al., 1983, Rai et al., 2000). Polyparasitism is common in rural areas (Rai et al., 2001). Intestinal worm infection alone ranks fourth in "top-ten-diseases" in Nepal (MOHP 2008). Intestinal parasitosis has been associated different morbidities associated with malnutrition, anemia and others particularly in children and pregnant women including mortality. This is attributed to low socio-economic, educational and poor hygienic conditions of the people (Rai et al., 1998, Rai SK, Asian Parasitology 2005; Rai et al., 2001).

Sukumbasi denotes the people living without land ownership (*Sukumbasi* Ayog, 2000). There are many *Sukumbasi* (living without land ownership) *Basti* (area) in the Kathmandu Valley mostly along the Bagmati River. One of the *Sukumbasi Basti* was Dirgayu Tole, Shantinagar, Ward No-34, Kathmandu. The children of this area were the study subjects. There are 500 huts in the *Sukumbasi Basti* with around 3000 population. The occupations of the *Sukumbasi* people vary; some work as labour, driver, carpenter, business etc. The people were of different castes- *Tibeto-Burman, Indo-Aryan* and *Dalits*. The children go to nearby high school. They play in the river side and often swim in the Bagmati River during summer season. Hence, they are likely to be infected with intestinal parasites. However, no such data are available. This study, therefore, was done to see the prevalence of intestinal parasitosis in children of a *Sukumbasi Basti* in the Kathmandu Valley. This would assist to assess the health and socio-economic status of the children. Furthermore, it would help in determination of mode of outbreak of the newly emerged parasites with their clinical and epidemiological aspects. Moreover, the findings would aid the researchers in getting insight of the health status of *Sukumbasi* children and help concerned body in crafting appropriate plans and policies on health.

CHAPTER II

2. OBJECTIVES

2.1 General objective

I. To find out the prevalence of intestinal parasite among children of "*Sukumbasi Basti*" in Kathmandu Valley.

2.2 Specific objectives:

I. To perform stool examination for parasites detection.
II. To describe the distribution of parasitic infection with sex and age of the children.
III. To know the sanitary behavior of the children in relation to the infection.
IV. To correlate the infection among the children with their socio-economic status.
V. To assess the prevalence of parasitic infection after deworming in the past six months.

CHAPTER III

3. REVIEW OF LITERATURE

3.1 Global Scenario

According to WHO, 2000; most of the illness in the world is caused by inadequate sanitation, unsafe water and unavailability of water, intestinal parasitosis being one of them. Both the protozoa and helminthes are responsible for the intestinal infection leading to many cases and deaths, particularly in underdeveloped countries.

The prevalence of *B. hominis* was evaluated in the community of Campo Verde, a district of Pitanga. Samples of faeces from children and adults were collected and submitted to the techniques of direct wet mount, flotation in zinc sulphate solution, tube sedimentation; sedimentation in formalin-ether and staining by Kinyoun and iron hematoxylin methods (70.7%) showed protozoa and/or helminthes in stool samples. The most prevalent species were *E. nana* (33.7%), *B. hominis* (26.5%), *G. lamblia* (18.2%), *E. coli* (17.1%), *A. lumbricoides* (16.6%), *I. butschlii* (9.4%), and ancylostomatidae (7.7%) (Nascimento et al., 2005).

The prevalence of *C. cayetanensis* was studied among apparently healthy persons in Peru. One group included those consulting private physician and next included people from marginal area. They had common complain of abdominal pain and diarrhoea. The incidence rate has been found to be (41.6%) and (7.3%), respectively among the two groups. He reported higher rate among young and elder adults up to 60 year-old (Burstein et al., 2005). A study conducted in a community of rural Coted’Ivoire found the prevalence of hookworm, *E. histolytica/E. dispar* and *S. mansoni* to be (45.0%), (42.2%), and (39.8%), respectively. Three-quarters of the population harbored multiparasites (Raso et al., 2004).

A national survey of the prevalence of intestinal parasitic infections in the Islamic Republic of Iran was made by Sayyari et al., 2005. Altogether 45,128 stool samples were analyzed by formalin-ether precipitation. Among them, intestinal parasitic infections were found (19.1%) in female. *G. lamblia* (10.9%), *A. lumbricoides* (1.5%), *E. histolyica* (1.0%) and *E. vermacularis* (0.5%) were common parasites. The infection rate was highest in the 2-14 years age group (25.5%).

Bitkowska et al., 2004 examined stool of group of 7 years old children of Poland for the presence of intestinal parasite. The study was based on the examination of single fecal specimen and a cellophane swab. The studies include 31,504 children. The most frequently encountered parasites in the examination included: *E. vermacularis* (15.0%), *A. lumbricoides* (0.83%), *G. intestinalis* (0.96%), *E. coli* (0.60%) and *T. trichuria* (0.12%). The overall percentage of the infected children was 15.4%. The number of infected among children inhabiting countryside (19%) was significantly higher than among those from the towns (10.4%).

The prevalence of intestinal parasite was determined for 1,370 children in Khan Younis Governorate, Gaza strip. The age of the children ranged from 6 to 11 years. For stool samples inspection, direct smear microscopy, flotation and sedimentation techniques were used. The general prevalence of intestinal parasites was 34.2%. Different types of intestinal parasites were detected during this survey. *A. lumbricoides* seemed to be the most common parasite (12.8%), whereas *G. lamblia* had a prevalence of (8.0%), *E. histolytica (*7.0%), *E. coli* (3.6%), *T. trichiura (*1.6%) and *H. nana* (1.0%). The prevalence of enterobiasis was determined using a scotch tape preparation. A total of 20.9% of the children examined were infected and there was sex variation in the prevalence of enterobiasis (Astal, 2004).

The evaluation of the impact of drug treatment on infection by *A. lumbricoides*, *T. trichiura* and hookworms in a rural community from the sugar-cane zone of Pernambuco, Brazil was performed; Individual diagnosis was based on eight slides (four by the Kato-Katz method and four by the Hoffman method) per survey. Infected subjects were assigned to two groups for treatment with either albendazole or metrobendazole. Prevalence of infection fell significantly ($p < 0.05$) one month after treatment. *A. lumbricoides* (from 47.7% to 6.6%), *T. trichiura (*from 45.7% to 31.8%) and Hookworm (from 47.7% to 24.5%). One year after treatment, infections by *T. trichiura* and hookworm remained significantly below pre-control levels (Zani et al., 2004).

The study was conducted Oliveira et al., 2003 in a rural area of Uberlandia, State of minas Gerais, Brazil in which 65.5% of the total population were found to be infected with intestinal parasites among which 45.1% were children and 54.5% adults. Within this study group 66.7% were mono infected, 17.6% bi-infected and 15.7% polyinfected. 47.0% individual was infected with protozoa, 29.4% helminthes and 23.6% with both. According to sex, the positively rate for intestinal parasites was 41.0% for male and 24.4% for female. Regarding to age group, high positive rate (29.6%) was found in children of age 1-15 years followed by 16-30 years (20.6%), 31-45 years (5.1%), 46-60 years (6.4%) and above 60 (3.8%). Among the parasites, *H. nana* was the most frequent helminthes (14.1%) and *G. lamblia* (11.5%) the major protozoa. High positive rate of 6.4% was detected both for hookworm and *S. stercolaris. T. trichiura* was found in 5.1% of the study.

Miller et al., 2003 examined the presence of intestinal protozoa and helminthes infections and their association with clinical signs and symptoms in children in Trujillo, Venezuela. Conventional microscopic method (thick-smear, saline and iodine solution) was used to identify parasites in stool samples in of 301 children attending day care centre. A subgroup of 45 children was evaluated clinically and parasitological five times during a 1-month period using conventional method and the Kinyong acid fast for Cryptosporidium identification. The point prevalence of protozoa infection was 21.0% for *G. duodenalis*, 1.0% for *E. histoltica/dispar*, 4.0% for *E. coli*, 16.0% for *B. hominis*, and 89% for *C.* p*arvum*. Prevalence of helminthes was 11% for *A. lumbricoides*, 10.0%

for *T. trichuria*, 0.3% for *S. stercolaris*, and 1.3% for *H .nana*. Over a 1 month time frame, new infection were observed at a rate of 11.0% for *G. duodenalis* 4.0% for *E. histolytica* ,7.0% for *A. lumbricoides* , 11.0% for *T. trichuria*, 0% for *S. stercolaris*, and 2% for *H. nana* . Intestinal parasitic infections contribute significantly to the enteric disease burden experienced by this group of children.

A study of helminthic infection in Vientiance Provience of Lao PDR found the positive rate (63.3). The single, double, triple and multiple infection rates were 64.0%, 28.0%, 7.0% and 1.0% respectively. *A .lumbricoides* (40.0%) was the commenest helminthes followed by *T. trichuria* (28.0%) and hookworm (8%). The study had found that women have 1.25 times more chance of being infected with *A. lumbricoides* than others ($p>0.005$).The monoinfection rate of the *T. trichuria* was found to be only 8.6% while its infection along with *A. lumbricoides* was 23.5% (Phetsouvannh et al., 2001).

Intestinal parasitosis alone affects almost 3.5 billion people worldwide and due to these infection 450 million are suffered from various kind of illness, the majority being children (WHO, 2000). According to WHO, 1997; globally *A. lumbricoides* infect 250 million, hookworm infect 151 million and *T. trichuria* infect 45 million people, respectively. Annually, each of these parasites has been responsible for the deaths of 65,000, 60,000 and 70,000 people, respectively (WHO, 2000).

Paul and Gnanmani et al., 1998 carried out a study to determine the prevalence and intensity of intestinal helminthes infections among the children belonging to lower socio-economic status. Stool samples collected were processed by modified formalin ethyl acetate sedimentation technique.177 children were infected with one or more of intestinal parasites as *A. lumbricoides*, *T. trichiura* and hookworm. The overall prevalence of infection was 82.0%. *A. lumbricoides* was the most common infection with prevalence of 75.0% followed by *T. trichiura* (66.0%) and hookworm (9.0%).

A study of prevalence and intensity of soil-transmitted helminthes among pre-school children aged 0 to 7 years from an Orang Asli village of Malasiya. The overall prevalence of soil transmitted helminthes (STH) infections was 56.0%. The predominant helminthes found was *A. lumbricoides* while the commonest type of infection was a mixed infection with *A. lumbricoides* and *T. trichiura.* The prevelace rates of *Ascaris, Trichuris* and hookworm infection were 47.5%, 33.9% and 6.2% respectively. The prevalence of helminthiasis (STH) shows an-age dependent relationship, with the lowest prevalence in 0-< 1year age group and highest in the 6-< 7 year age group (Zukifli et al., 1999).

Knightlinger et al., 1998 surveyed 633 children, within age group 4-10 years living in Southern Madagascar. The study revealed the maximum (93.0%) prevalence rate of *A. lumbricoides* followed by *T. trichiura* and hookworm by 55.0% and 27.0%, respectively.

Mafiana et al., 1998 conducted an investigation to determine the prevalence of STH parasites in children in Ogun state, Nigeria. Fecal examination of 1,060 children reveled the prevalence of (64.0%) for *A. lumbricoides*, (21.9%) for *T. trichiura* and (14.5%) for hookworm.

Saito et al., 1996 surveyed for intestinal parasites by using thin smear and floating method for fecal examination in residents in Caazapa Department, Paraguay. Out of 608 samples of residents in Boqueron, a community of Caazap Department, 343 (56.55%) were found positive. The most prevalent parasite was *N. americanas* (27.0%) followed by *E. coli*.

Sugnan et al., 1996 carried out of survey among the rural and urban settler and two urban groups *viz*, Nicobares and Onges, of Andaman and Nicobar is island in 1996.The survey was conducted between preschool school aged children and adults. Among the preschool children, Nicobares showed highest overall prevalence rate (80.5%) followed by urban (46.7%) and rural (38.6%) preschool children. *A. lubricoides* was the commonest form of parasite encountered in all the groups of preschool children, followed by *T. trichiura*. The school age children among rural settles showed an overall prevalence rate of 61.1% which was significantly higher than that among the rural preschool children. The Nicobarese and Onage adults showed significantly higher overall prevalence rates (72.2%) and (71.1% vs. 48.6%) compared to rural adults. In all among trichuriasis and giardiasis were common than ascariasis.

Milano et al., 1995 have evaluated the importance of enteroparasitosis in young urban population. Serial stool sample were analyzed and was also studied. Environmental data were also collected via semi-structured surveys. Soil samples were tested to determine the degree of soil contamination. The following species were identified such as *B. hominis, E. vermicularis, cocidius* sp.*, G. intestinalis,* hookworms*, S. stercolaris, T. trichiura, A. lumbricoides, E. coli, E. nana,* and *Taenia* sp. Children infection prevalence was 73.5%. The frequency of enteroparasitosis was largest in the population from 3 to 8 years. The homes of the children analyzed were brick houses with tin roof and access to tap water. Of these 79.5%, houses had bedrooms. The remaining used outdoor latrines. In 95.5% of these houses, the residents lived with one or more dogs and cats. The stool collected from nine houses was contaminated with infectious forms of *T. canis* and *Anlyomideous*. The relationship between parasitosis and latrines and overcrowding were verified. Five cases of malnutrition were detected (4.4%).

A community-based study was conducted among randomly selected 300 children aged less than five years selected from three camps of the police force in Khartom, Sudan. The commonest infestations were giardiasis (21.1%), taeniasis (10.45%) and enterobiasis (7.4%). Non pathogenic *E. coli, E. histolytica* and *T. saginata* were detected (2.7%), (0.7%) and (1.7%) of stools specimen, respectively. Children aged between 3 years and above were the most affected group and the infection rate was highest among the illiterate, overcrowded and large sized families. Malnourished children comprised (9.4%) of the study group was but there was no significant association between and malnutrition and the overall prevalence of intestinal infestations, although *G. lamblia* significantly affected the malnourished group (Karrar and Rahim, 1995).

Gbakima in 1994 assessed the prevalence of intestinal and urogenital parasites in Moyamba District, South-central Sierra Leone. Stool and urine sample were submitted by 1106 individuals and examined by the iron-haematoxylin staining and the formalin-ether concentration techniques for fecal sample and centrifugation method for the urine sample. The overall parasitic infection rate was 61.7% while 5.9% of the population had multiple infections. *E. histolytica* in infection rate was 12.3% and most infected individual passing cysts. *G. lamblia* and *Trichomonas vaginalis* infection rates were 10.0% and 0.4%, respectively. Among the helminthic infection, *A. lumbricoides* was the most commonly observed (13.7%), followed by hookworm (12.1%), *T. trichiura* (9.3%), *S. stercolaris* (7.7%) and tapeworm (2.6%). The high parasitic infection rate (61.7%) and the frequency of multiple infections indicate an interrelationship of environmental factors which support transmission rather than single factor.

Lee et al., 1994 examined fecal specimens of a total 52,522 patients by formalin-ether sedimentation or direct smear method. The overall egg positive rate of helminthes was 6.5%. The egg positive rate for each species of helminthes was *C. sinensis* (3.2%), *T. trichuria* (2.0%), *A. lumbricoides* (0.2%), *Trichostrongylous orientalis* (0.1%), *Taenia* sp. (0.05%), *H. nana* (0.03%) , hookworms (0.03%), *P. westermani* (0.02%), *E. chinostoma* sp.(0.03%), *E. vermacularis* (0.02%), *S. stercolaris* (0.1%) and *Diphyllobothrium latum* (0.004%). In comparison to various data, *A. lumbricoides* and *T. trichuria* has been found in decreasing ratio.

Anderson et al., 1993 carried out the research on the distribution of intestinal helminth infections in a rural village in Guatemala. Fecal egg count scores were used to investigate the distribution and abundance of intestinal helminthes in the population of a rural village. Prevalence of the major helminthes was 41.0% with *A. lumbricoides*, 60.0% with *T. trichiura* and 50.0% with *N. americannus*. Infected females had higher burdens of *T. trichiura* than infected males in all age classes of the population; there were no other effects of the host gender. Analysis of associations between parasites within hosts reveled strong correlations between *A. lumbrcoides* and *T. trichiura*. Individuals with heavy infections with *A. lumbricoides* and *T. trichiura* showed highly significant aggregation within households. Association between a variety of household features and heavy infections with *A. lumbricoides* and *T. trichiura* were described.

Study conducted by Cachin-Bonilla et al., 1992 showed that prevalence of intestinal helminthes parasites were assessed in a suburban community of Maracaibo, Venezuela by examination of stool from 342 individual, using iron hematoxylin stained fecal smears and formalin-ether concentration. The overall parasitic infection rate was 80.4% and 65.8% of the population had multiple infections. *T. trichiura* (71.9%) and *A. lumbricoides* (54.0%) were the most common parasites, particularly in school children. The high rates of parasitic and multiple infections reflect the low socio-economic status of the community studied.

In a survey in Indonesia, 419 stool samples were examined by using direct smear, flotation and formalin ether concentration technique. Five nematodes and seven protozoan parasites were detected while trematodes and cestodes infection were not observed. STH infections were predominant. Among the younger inhabitants aged less than 15, positives rates of *A. lumbricoides*, *T. trichiura* and hookworm infections were almost same (45.7%), (45.3%), and (47.7%), respectively. Among the people aged 15 or more, positive rate of hookworm infection (89.4%) was much higher than *A. lumbricoides* and *T. trichuria* infections (19.3%) and (26.1%), respectively (Hasegawa et al., 1992).

A survey conducted by Marnell et al., 1992 among the refugee in Juba, Sudan, involving 241 fecal samples revealed 66.0% of the population harbored intestinal helminthes. The most commonly found infection was hookworm (36.0%), followed by *S. mansoni* (26.0%), *S. stercolaris* (20.0%), *H. nana* (11.0%), *A. lumbricoides* (1.2%), *T. trichiura*

(0.8%) and *Taenia* sp. (0.4%). Among examinee, 42.0% had single infection, 21.0% had double and 3.0% had multiple infections. Parasitic prevalence and intensities were analyzed in relation to age, sex, relation and occupation. Females were less infected (50.0%) than Christians (68.0%) and agriculturists (90.0%) were the most infected occupational group.

Obiamiwe and Nmorst, 1991 examined 862 fecal samples from male and female donors of nine age groups (1-90 years) in three geographical zones, after examination of concentration from formal-ether concentration technique. They found the overall percentage incidence of six parasites which were encountered were as follows, *E. coli* (19.9%), *E. histolytica* (3.9%), *G. lamblia* (1.4%), hookworm (29.4%), *A. lumbricoides* (38.2%), *T. trichuria* (7.3%).

Stool specimens from 1,282 children between the age of 5 and 13 years attending ten primary schools for boys in Saudi Arabia, were examined for the presence of intestinal parasites. Of these, 313 (24.4%) were found to be infected with one or more species of 11 intestinal protozoa and helminthes. The most common pathogenic protozoa being *G. lamblia* (10.9%) followed by *E. histolytica* (4.1%). The non pathogenic protozoa, *E. coli* had the highest prevalence rate (11.3%) in the children stools. *H. nana* was the commonest intestinal helminth (3.0%). Other intestinal helminthes, including *A. lumbricoides, T. trichiura, S. mansoni, Dicrocoelium dendriticum* were detected to a lesser extent. The distributions of the common intestinal infections among the children surveyed were also analyzed according to age and multiplicity of infection. Prevalence of *E. histolytica* was found to increase with age whereas *Giardia* infections were less common among older children (Omar et al., 1991).

Chandiwana et al., 1989 surveyed to access the prevalence and intensities of hookworm and roundworm infection in 15 farm-worker communities in Zimbabwe with poor living conditions. Examinations of 1,635 fecal samples showed that hookworm were the commonest helminthes (61.7%). They were followed by *A. lumbricoides* and *T.*

trichiura. Age prevalence and age intensity profiles for hookworm showed that infection increased with age, with a peak in the adult age groups. *A. lumbricoides* prevalence was relatively high in children but declined in adults.

In cockle Province, Republic of Panama, Robertson et al., 1989 carried out a survey of intestinal helminthes in children by microscopic examination (modified Kato-katz technique) of stool samples from 661 children attending primary school. The overall prevalence of *A. lumbricoides*, hookworm and *T. trichiura* infection were found (18.2%), (12.0%) and (27.5%) respectively. There were significant differences between the infection prevalence values for children attending the different school, but with not respect to age or sex. Positive association was detected between particular pairs of infection and there were most evident with *T. trichiura.*

Haswell et al., 1989 studied the distribution of *A. lumbricoides* within the community and found significant variation in the intensity of infection between household in the community. The number of family members living in the house strongly influenced the mean *Ascaris* burden and proportion of relatively heavy infection within adults and children. This finding suggest that the density of people in the house positively influences the frequency of exposure to infective stages of *Ascaris*, which in turn plays a major role in determining which individual will harbor heavy infection. A comparative examination of hypothetical treatment strategies suggest that for *Ascaris* infection in this community, targeting age groups with antihelminthic treatment would probably be more cost-effective in the long term in reducing the abundance of this parasite than selective treatment of individually identified heavy infection.

Fagberno-Beyioku and Oyerindo, 1987 examined microscopically the stool samples from 1,659 children, aged 15 years and below in metropolitan Lagos and showed a (71.9%) and (68.3%) infection with *T. trichuria* and *A. lumbricoides* respectively. While the infection rate with hookworm was 22.5%. Infection with more than one parasite was also very common. Those children whose household had no toilet facilities had the lowest infection frequency with the intestinal parasite, while those who used buckets as their means of fecal disposal had highest rates.

According to Wiesenthal et al., 1980 165 Meo Laotians stool sample were screened for intestinal parasites. One hundred thirty- nine had at least one parasite detected. Hookworm was detected most frequently, followed by *G. lamblia*, *T. trichiura,* and *A. lumbricoides*. Hookworm and overall infection were more frequent in persons 4 years of age and older, while giardiasis, ascariasis, and trichuriasis were the most common in the 4-14 years age group. Most infection were helminthes and of no public health consequences in the United States. However, giardiasis was seven times as prevalent in refugee children as in the general US population, posing a potential public health risk in child- care setting.

Survey for intestinal parasites was conducted in Napsan on the island of Palawan, Philippines. A total of 353 stool specimens were obtained from 155 males and 198 females ranging in age from one and half months to 70 years. *A. lumbricoides* (34.8%), Hookworm (34.8%) and *T. trichiura* (25.2%) were the most common helminthes encountered, while *E*. coli (27.7%) and *E. nana* (22.2%) was the common protozoan (Oberst and Alquiza, 1987).

Study conducted at South Kalimantan Province of Indonesia; by Cross et al., 1975 to1998, different intestinal parasitic infections were detected (97.0%) of the people. Those parasites most frequently found were *T. trichiura* (83.0%), *A. lumbricoides* (79.0%), and hookworm (65.0%), followed by *E. coli* (37.0%), *E. nana* (12.0%), *E. histolytica* (12.0%), *G. lamblia* (5.0%), *E. hartmanni* (2.0.0%), *C. mesnili* (2.0%). Other parasites found were *E. vermicularis, S. stercoralis, Capillaria* sp., *Echinostoma* sp., *H. diminuta and T. hominis*. *G. lamblia* was found more often in younger people. Hookworm occurred more frequently in males and *A. lumbricoides* in females.

3.2 SAARC countries scenario

The health risks of wastewater use in agriculture were investigated by Ensink et al., 2005 in the city of Faisalabad, Pakistan, by means of a cross-sectional study. The study showed an increased risk of intestinal nematode infection and hookworm infection, in particular, in wastewater farmers (OR = 31.4, 95% CI 4.1-243) and their children (OR = 5.7, 95% CI 2.1-16) when compared with farming households using regular (non-wastewater) irrigation water.

The study was designed to examine stool specimens of irritable bowel syndrome (IBS) patients for *B. hominis.* One hundred fifty patients were enrolled, 95 IBS cases and 55 controls. The 95 cases (51 males and 44 females) had a mean +/- SD age of 37.8 +/- 13.2 years. Stool microscopy was positive for *B. hominis* in 32% (30 of 95) of the cases and 7% (4 of 55) of the controls (P = 0.001). Stool culture was positive in 46% (44 of 95) of the cases and 7% (4 of 55) of the controls (P < 0.001. Stool culture has a higher positive yield for *B. hominis* than stool microscopy (Yakoob et al., 2004).

The study was undertaken to measure the impact of periodic deworming with albendazole on growth status and incidence of diarrhea in children aged 2-5 years in an urban setting in India. . The two study groups received two doses of albendazole (400 mg) or placebo six months apart. Mean weight increased significantly in the albendazole group compared to the control group at three months, six months and nine months following treatment (P<0.01, P<0.01 and P<0.001, respectively). The albendazole group also experienced fewer episodes of diarrhoea than their control counterparts (relative risk 1.3, 95% CI 1.07-1.53) with a 28% reduction (Sur et al., 2005).

Sayyari et al., 2005 in a national survey of intestinal parasitosis in Iran, intestinal parasitic infection rate was detected as 19.3% (19.7% male, 19.1% female). In the study, *G. lamblia* (10.9%), *A. lumbricoides* (1.5%), *E. histolytica* (1.0%) and *E. vermicularis* (0.5%) were the most common parasites. The infection rate was highest in the 2–14 years age group (25.5%) and in rural residents (23.7%). The prevalence rate of *T.*

saginata, T. colubriformis, T. trichiura and *A. duodenale* were 0.2%, 0.2%, 0.1% and less than 0.1% respectively. The total prevalence of intestinal parasite among people of age group 40-69 was 15.0% and greater than 69 years was 11.6%. The prevalence of individual parasites in 40-69 years age group was *G. lamblia* (7.3%), *A. lumbricoides* (1.5%), *E. histolytica* (1.1%) and *E. vermicularis* (0.2%). The prevalence of individual parasites was *G. lamblia* (5.0%), *A. lumbricoides* (1.2%), *E. vermicularis* (0.1%) and *E. histolytica* (0.7%) in age group.

Study conducted in the union Territory, Chandigarh, India estimates the prevalence of intestinal parasitic infections in different population groups. The prevalence of intestinal parasitic infections was found to be 14.6% with highest prevalence of 19% from the slum area. Children were the most commonly affected group (18%) with those from slums showing the highest prevalence (24.6%). The most common parasite was Giardia (5.5%) followed by *H. nana* and *A. lumbricoides* (2.8% and 2.7%, respectively), (Khurana et al., 2005).

Singh et al., 2004 studied parasitic infection among primary school-going children between the age group of 5 to 10 in the urban and rural areas of Manipur. A total of 248 (24.5%) were positive for various helminthes. Among the positive cases, 110 (26.3%) were from the urban area (city) and 138 (23.4%) from the rural areas of Manipur. Maximum number of parasitic infection occurred in the age group of 5 to 6 years (27.0%) in both sexes. Among the parasites, *A. lumbricoides* was the commonest (19.6%) followed by *T. trichiura* (2.18%), *H. nana* (0.99%), tapeworm (0.19%), hookworm (0.09%), *S. stercoralis* (0.09%), *E. vermicularis* (0.09%).

Stool samples were collected 3 to 6 months post-treatment to study the rate of reinfection. The cure rates for *A. lumbricoides, T. trichiura* and hookworms were 70.8%, 68.7% and 93.0%, respectively. Re-infection rates after 3 months of successful treatment were 19.6% for *A. lumbricoides,* 30.9% for *T. trichiura* and 11.3% for hookworms. Six months post-treatment, the prevalence of re-infection was highest with *T. trichiura* (43.6%); followed by *A. lumbricoides* (35.3%). The rate of reinfection with hookworms was lower (11.3%) six months post-treatment. The rates of re-infection with *A. lumbricoides and T. trichiura* were higher in children below 15 years of age, compared with adults. Hookworm reinfection was higher in the adult age group (15 to 39 years). The rates of new infection in previously uninfected subjects were lower compared with the rates for re-infection (Narain et al., 2004).

Allen et al., 2004 surveyed in the western region of Bhutan to assess the prevalence and intensity of soil-transmitted helminth (STH) infections after 15 years of school deworming in the country. Stool samples were collected from each child as well as nutritional indicators and general information on each school. The survey found a cumulative prevalence of 16.5% STH (4.8% in schools treated in the last three months and 24% in the untreated schools). An unexpected finding was that the tapeworm infection rate of 6.7%. These results indicate a high reinfection rate in this area. WHO recommends 50% prevalence as the threshold for the establishment of community intervention.

Several reports of patients with cysticercosis from many countries in Asia such as India, China, Indonesia, Thailand, Korea, Taiwan and Nepal are a clear indicator of the wide prevalence of *T. solium*, cysticercosis and taeniosis in these and other Asian countries. It is also a major cause of epilepsy in Bali (Indonesia), Vietnam and possibly China and Nepal. Seroprevalence studies indicate high rates of exposure to the parasite in several countries (Vietnam, China, Korea and Bali (Indonesia)) with rates ranging from 0.02 to 12.6%. An astonishingly high rate of taeniosis of 50.0% was reported from an area in Nepal populated by pig rearing farmers. Undoubtedly, cysticercosis is a major public health problem in several Asian countries affecting several million people by not only causing neurological morbidity but also imposing economic hardship on impoverished populations. (Rajshekhar et al., 2003).

A. lumbricoides infestation (ALI) is one of the most common helmintic diseases of the gastrointestinal tract, and may cause severe surgical complications, especially in children. A case of a 5-years old Pakistan girl treated in Italy for acute abdomen in which Ali was detected during surgical exploration (Mosiello et al., 2003).

This study was carried out at the northern part of Bangladesh to determine the impact of sanitary latrine use and of health education on intestinal parasites in school-aged children. The children were between 5 and 13 years of age and stool samples revealed that more than half (53%) of the study sample was still infected with one or more intestinal parasites even after 4 years of intervention. Ascariasis was found to have the highest prevalence rate (36.2%) and hookworm the lowest (10.7%). Intestinal parasite infection was significantly lower ($P < 0.05$) among those who used a sanitary latrine and received health education (Hosain et al., 2003).

The study was conducted in Konkor, Gadap, District East, Karachi to determine the prevalence of intestinal parasitic infection. Out of 263 residents 185 tested for intestinal parasites and 88 (47.5%) had pathogenic parasites. The distributions of parasites were *G. lamblia* (50%) and *E. histolytica* (48.86%). Statistically none of the socio demographic variables were associated except education and age group (Siddiqui et al., 2002).

Gilgen et al., 2001 conducted a randomized clinical intervention trial over 24 weeks on a tea estate in north-east Bangladesh to investigate the effect of iron supplementation and antihelmintic treatment on the labour productivity of adult female tea pluckers. A total of 553 full-time tea pluckers, not pregnant and not breastfeeding, were randomly assigned to one of the four intervention groups: group 1 received iron supplementation on a weekly basis, group 2 received antihelmintic treatment at the beginning and half-way through the trial (week 12), group 3 received both iron supplementation as group 1 and antihelmintic treatment as group 2, and group 4 was a control group and received placebos. However, there was a negative association for all three worms (*A. lumbricoides, T. trichiura* and hookworms) between the intensity of helminth infections (eggs/ faeces) and all measures of labour productivity. Lower haemoglobin values and anaemia (< 120 g/l Hb) were both associated with lower labour productivity and more days sick and absent. Taller women with greater arm circumference were able to pluck more green leaves, earn higher wages and were absent less often.

In developing countries many enteric infections are caused by acid-sensitive pathogens. Study was conducted in 185 Bangladeshi men admitted to hospital for the treatment of enteric infection. Patients with dysentery (amoebiasis, n = 24 and shigellosis, n = 19) and culture-negative diarrhoea (n = 69) had similar mean gastric acid levels (basal, 3-5 mmol/h; stimulated, 11-17 mmol/h), which remained stable in those patients studied throughout 12 weeks of convalescence. Gastric acid levels were not associated with *G. duodenalis* or *S. stercoralis* co-infection, fever, use of tobacco, or chewing betel nut. Factors which impair gastric acid secretion may predispose to diarrhoeal disease in developing countries (Evans et al., 1997).

A study conducted in Srilanka, showed examination of total 192 stool samples from the adult and the pre-school children) was done. *E. histolytica* was not seen in any of the samples; *Giardia* cysts and Cryptosporidium oocysts were seen in 3 and 1 sample respectively from the pre-school children. The overall prevalence of geohelminth infections was 21.3% among the adults and 24.5% among the children. *A. lumbricoides* was the predominant species in both populations (De silva et al., 1994).

Stool samples from 880 residents in an urban slum in Dhaka, Bangladesh, were collected and examined for intestinal parasites. Information on many potential risk factors for infection was obtained by questionnaire from a respondent in each household studied. In a crude univariate analysis of the data, several of the factors were found to be significantly associated with S. *stercoralis* infection. Most of these factors were co-variate with one another and with poverty generally (Hall et al., 1994).

Stool survey was carried out in some of the villages of Dadraul and Bhawal Khera PHC's of district Shahjahanpur (Uttar Pradesh). Among them, 29.2 % were found positive for one or the other intestinal parasite. *A. lumbricoides* superseded all the parasites by showing a positivity of 17.8%. Other parasites found were Hookworm, *H.*

nana, Tapeworm, *T. trichiura, E. vermicularis*, *E. coli* and *G. lamblia*. Parasitic load was slightly higher in females (33.5%) than males (28.1%). The highest positivity was encountered in the age groups between 6 to 14 years (Virk et al., 1994).

Analysis of egg and worm counts of *Ascaris* recorded at various intervals following a mass antihelmintic treatment programme in a South Indian fishing community was done. Three indices of infection in the community are compared, namely the prevalence and intensity of egg output (at 2, 6 and 11 months following treatment) and the number of worms expelled following an 11 month period of reinfection. Detailed examination of these measurements revealed significant associations with patient sex and age. Although 85.0% of both males and females harbored *Ascaris* initially, the prevalence following 11 months reinfection was decreased, due to a significantly lower proportion of males being reinfected. By the 11th month of reinfection, the age-intensity profiles of egg output were similar to those observed at initial treatment in the older age groups (10 years and above) and in male children (less than 10 years) (Elkins et al., 1988).

3.3 National Scenario

Rai et al., 2007 was carried out a retrospective study to see the prevalence of intestinal parasitosis among the patients visiting a health care centre in Kathmandu Valley. Of the total 1,316 subjects included, 395 (30.0%) showed some kind of parasites. Females had significantly higher positive rate (34.0%) compared with males (27.5%). Highest positive rate (32.3%) was found in the age group 16-30 years followed by the age group 31-45 (30.8%) and others ($p>0.05$). Most of the patients (89.9%) were infected with single parasites. Protozoan parasites were more common than helminthes. Of the helminth parasites detected, *A. lumbricoides* was the commonest one followed by others. Among the protozoa, *E. histolytica* topped the list.

The study was carried out among the elderly people in Kathmandu Valley to assess the prevalence of intestinal parasitosis in them. Stool samples were collected from 235 elderly people. The samples were examined by formal ether sedimentation and

Sheather's sucrose floatation followed by Kinyoun's modified Ziehl-Neelsen staining. The overall prevalence of intestinal parasites was found to be 41.7% out of which 30.6% had multiple parasitisms. The government elderly home had significantly higher parasitic prevalence (50.8%) followed by the rural community (46.8%) and the private elderly home (21.2%). Males (43.8%) had slightly high infection rate than females (40.4%). There was equal infection rate with protozoa (25.8%) and helminths (27.0%). *T. trichiura* (39.4%) and *E. histolytica* (19.7%) were the commonest helminth and protozoa respectively (Shakya et al., 2006).

Adhikari et al., 2004 was found 34% prevalence of intestinal helminthic infection among school children in Kathmandu Valley. Such infection was found equally among males and female population. *T. trichiura* was the most common parasite among the study subjects followed by *A. lumbricoides*, hookworm, *H. nana* and *S. stercoralis*. It was observed that the rate of helminthic infection among *Dalits, Indo-Aryans* and *Tibeto-Burman* does not differ significantly.

Rai et al., 2005 studied the predisposing factors of enteric parasitic infections in school children in a rural area in Kathmandu Valley, Nepal. Fecal samples were examined by formal-ether concentration technique. A total of 71.2% children had parasite. Altogether nine types of parasite were recovered; *T. trichiura,* was the commonest one followed by hookworm and others. Among the protozoa, *E. coli* was commonest followed by *E. histolytica,* and others. Boys had relatively higher prevalence compared with girls. Highest infection rate was observed in the children aged ≥ 15 years. Infection rate was found higher in *Dalit* children compared with *Tibeto-Burman* and *Indo-Aryan*. Prevalence of parasitic infection among children with history of talking antihelminthic drugs within past ix months was found significantly lower compared to those not taking drugs.

Intestinal parasitic infestation continues to be of public health importance in many tropical and subtropical countries for their high prevalence and effects on the morbidity in the population. This analysis was aimed to find out the intestinal protozoal parasitic profile in pre-school and school-going children visiting the hospit*al* with gastrointestinal illness in western Nepal. *G. lamblia* was the most prevalent pathogenic protozoan intestinal parasite (73.4%), followed by *E. histolytica* (24.4%). Interestingly, newer opportunistic pathogens like *C. cayetanensis* (1.0%) and *Cryptosporidium* sp. (1.0%) were detected from immunocompromised children below 2 years of age (Easow et al., 2005).

Kimura et al., 2005 studied diarrhoeal diseases associated with *C. cayetanensis* in Nepal and Lao PDR. *C. cayetanensis* was detected by direct microscopy using ultraviolet and differential interference contrast microscopy. The overall positive rate in Nepal was 9.2% (128/1397). A higher positive rate was observed in children aged 10 years and under (11.1%) and was lowest in the age group of 51-60 years (3.1%). A significantly higher positive rate was observed in the summer (rainy season) (12.6%) with the lowest prevalence in the spring (dry season) (1.8%) ($P<0.05$). The positive rate was closely associated with rainfall (ml/month). Only one of the total 686 samples (0.1%) from Lao PDR was found to be positive for *Cyclospora* oocysts.

The study was conducted to represents the status of intestinal parasitosis in public schoolchildren (1 to 10 classes) in a rural area of the Kathmandu Valley to their habits, including factors predisposing to parasitic infections. Stool samples from the children were examined. The overall prevalence of parasitosis was 66.6% (395/533). Altogether, nine types of parasites were recovered. The recovery rate of helminthes was higher (76.9%) than protozoa (23.1%). *T. trichiura* was the most common helminthes detected, followed by hookworm, *A. lumbricoides* and others. *E. coli* was the most common protozoan parasite, followed by *E. histolytica, G. lamblia* and others (Sharma et al., 2004).

Intestinal parasitological survey was conducted to clarify the distribution of intestinal parasites in Nepal and Lao Peoples' Democratic Republic (Lao PDR) from 2001 to 2003.The stool specimens were examined using the formalin-ether sedimentation (FES) and sucrose centrifugal flotation (SCF) techniques. Nine species (3 Nematoda, 1

Cestoda, and 5 Protozoa) of parasites were recovered from Nepal, whereas seven species (3 Nematoda, 1 Trematoda, and 3 Protozoa) from Lao PDR. Out of which (14.4%) was the most common in Nepal, and was *O. viverrini* (29.8%) in Lao PDR. Infection rates were markedly different among age groups in both countries; higher rates were observed in age groups of 10-29 years than in 0-9 years group (Takemasa et al., 2004).

Intestinal parasites were detected in diarrhoeal stool samples collected from individuals aged 1 to 68 years (males: 239 and females: 157) in Nepal. Parasites were detected by employing the formal-ether sedimentation technique. Of a total of 396 fecal samples investigated, 193 (49.0 %) were positive for some kind of parasite. Altogether, 15 species of parasites were detected. *G. intestinalis* topped the list of protozoa, whereas *T. trichiura* was the most frequently detected among helminth parasites. Of the 193 positive samples, 109 (56.0 %) had single parasite infections, whereas 84 (43.0 %) had multiple infections with a maximum of five species. Of the total positive, 45 (23.0%) had both protozoa and helminths whereas 37 (19.0%) had only protozoa. Females (52.0%) and children (15 years and under) (52%) had a marginally higher prevalence compared with males (46.0%) and adults (45.0%), respectively ($p > 0.05$) (Uga et al., 2004).

Ishiyama et al., 2001 had reported the parasite prevalence of 72.4% among school children in Kathmandu. *Indo-Aryans* were found to have marginally high rate of infection. Among which (46.9%) had multiple infections. *T. trichiura* (30.4%) was found to be the commonest helminth and *G. lamblia* (17.0%), the commonest protozoan. No bacterial enteropathogens were reported in the study population.

Rai et al., 2001 studied the intestinal parasitic infection in rural hilly area of Western Nepal, Achham district. The stool test revealed (76.4%) prevalence of intestinal parasites in the children of the district. A total of nine species of parasites were detected, of which, *A. lumbricoides* was the commonest one followed by hookworm and others.

Rai et al., 2000 intestinal helminth infection and its effect on vitamin A, retinol and B carotene, was studied in Okharpauwa Village Development Committee (VDC) (Nuwakot district) and 79 inhabitants (mainly adults) of Boya VDC (Bhojpur District)] subjects living at an altitude of 2000 m. Most common helminth detected was *A. lumbricoides* followed by *T. trichiura* in Okharpauwa VDC and by Hookworm in Boya VDC, respectively. Mixed helminth infections are were relatively low (7.3% in Okharpauwa VDC and 11.1% in Boya VDC). The retinol and B-carotene were estimated by high performance liquid chromatography (HPLC). The retinol concentration in helminth eggs positive children significantly increased after one month of anti-helminthic (albendazole) treatment ($P>0.05$). No significant difference in serum retinol concentration was observed among helminth eggs positive and negative inhabitants of Boya VDC ($P>0.05$) but in B-carotene level ($P<0.05$).

Rai and Rai, 1999 a hospital-based study in Kathmandu showed ascariasis as major casuses of public health problem in Nepal though the extent of ascariasis-associated morbidity and mortality has not been investigated yet. In some rural areas, over (75.0 %) people are infected with this parasite.

The parasitic infection rate of (50.0%) has been reported by the studies conducted in the Nepalese communities from 1979-1995 by different organizations. *A. lumbricoides* and *G. lamblia* topped the list respectively among helminthes and protozoans. Similarly, the hospital records showed the infection rate of 30 to 40 % (Chhetri, 1997).The burden of parasitic infection including other infections also reflected in the list of top ten diseases of Nepal.

According to Rai et al., 1997 the health and sanitary status of Boya Village was studied. The number of households having latrine increased significantly ($p<0.05$) in one year period but without significant impact on the reduction of intestinal helminth infection ($p>0.05$). *A. lumbricoides* was the commonest intestinal parasite followed by hookworm and others. Public piped line water was provided to (32.6%) households while remaining (48.4%) and (19.0%) were using natural tap and *kuwa* water, respectively. No association between the type of water source and gastro-enteritis was observed. There was poor hygienic condition. Majority of complaints were gastrointestinal.

Sherchand et al., 1997 carried out a stool survey on intestinal parasites and its transmission factors in rural village of Dhanusha Districts, Southern Nepal. Out of 604 children aged between 0-9 years examined, 363 (60.1%) were found positive for one or more intestinal parasites. Hookworm infection superseded all the parasites by showing a positivity of (11.6%). Other parasites found were *A. lumbricoides, T. trichura, Oxyuris vermicularies, S. stercolaris,* tapeworm, *H. nana, E. histolytica, E. coli, G. lamblia, C. parvum, C. cayetanensis, I. belli, O. viverrini, S. mansoni* and *I. butschlii*. The parasitic load was found slightly higher in female children (58.1%) compared to male children (41.9%).

The study was conducted on intestinal parasites from the Kathmandu area of Nepal was done in children and adults. The total parasite load was 28.1% and 38.8%, respectively, whereas children and adults with abdominal discomfort had a load of 62.7% and 67.8%. The prevalence of nematodes in the 4 groups was significantly higher in those with abdominal discomfort, particularly of hookworm, *Enterobius*, and *Ascaris*. *H. nana* was the most common tapeworm, and with the highest incidence in patients with abdominal complaints. *T. solium* and *T. saginata* were only found in the two adult groups, but with low prevalence rates. The highest incidence of *Cryptosporidium* was found in both groups with abdominal discomfort, notably among children. The presence of *Giardia* was highest among the sick children, many "healthy" carriers among both children and adults were noted. *E. histolytica* and *E. dispar* had a surprisingly low prevalence in all 4 groups. *B. hominis* was most common among adults with abdominal complaints (2.8%). *Trichomonas* sp. was also more common in this group, in which of 34 positive specimen, 28 were from women (Sherchand et al., 1996).

Stool samples were investigated for the prevalence of intestinal parasitic infection in Tribhuvan University Teaching Hospital, in Kathmandu. Among the various types of protozoan parasites, the most common was *G. lamblia* followed by *E. histolytica* (Rai et al., 1995).

A study conducted in Jiri revealed that the prevalence of whipworm, roundworm and hookworm among *Jirel* were 18.1%, 25.3% and 73.5%, respectively. Similarly, prevalence of whipworm, roundworm and hookworm among *Sherpa* were 11.2%, 23.6% and 46.1%, respectively. Similarly, the prevalence of whipworm, roundworm and hookworm among *Hindu* of age group 45 above were 7.1%, 26.2% and 59.5% respectively. The study found the increasement in multiple helminthic infections with increasement in age (Blangero et al., 1993).

CHAPTER IV

4. MATERIALS AND METHODS

4. Materials 1

A list of materials, chemicals, equipments, reagents for the study is presented in Appendix 1.

4.2 Methods

4.2.1 Study area

The laboratory investigation was carried out at Department of Microbiology, Nepal Medical College, Teaching Hospital Atterkhel, Jorpati, Kathmandu. A part of work was also done in National Institute of Tropical Medicine and Public Health Research (NITMPHR), Maharajgunj, Chakrapath, Sankhamarg, Kathmandu. The study period was from August 2008 to December 2008. The stool samples were collected from the *Sukumbasi* children of Shantinagar, Ward no-34, Dirgayu Tole, Kathmandu.

4.2.2 Samples collections

Each *Sukumbasi* children were given the brief description about the importance of the examination of stool to detect the parasite. They were advised not to contaminate the stool with water and urine. The containers were labeled with patient's name, code number, date and time of collection. During the process of specimen collection from each child, a questionnaire accompanying the queries about their clinical history, hygienic practice and nutritional behavior was filled. Labeled dry, clean disinfectant free wide mouthed plastic container was distributed and asked them to bring about 20 gms stool sample next morning.

4.2.3 Transportation of the samples

The collected stool samples were brought to the laboratory and immediately fixed with 10% formal saline mixing with equal part of formal saline and stool. Then, processing was done at NITMPHR.

4.2.4 Laboratory processing of the samples (Rai et al., 1996)

Each stool sample was processed in 2 steps as macroscopic examination and microscopic examination.

4.2.4.1 Macroscopic examination

The direct visualization of each sample was done for the color, consistency, presence of mucus, blood, and adult worm or worm segment.

Color

Based on the color, the stool specimen were categorized into groups i.e. normal color of stool (yellowish brown) and abnormal color of stool (muddy, black, pale etc.)

Consistency

Based on consistency stool specimen were classified as formed, semi-formed and loose.

Blood and mucus

The stool specimens were observed whether it contains blood and mucus or not.

Adult worms and segments

The stool specimens were observed whether it contains adult worms and segments or not. The adult worms of *A. lumbricoibes* and *E. vermicularis* are often seen in the specimen. Tapeworm segments may be occasionally seen in stool specimen.

4.2.4.2 Microscopic examination

Microscopic examination was carried out for the detection and identification of protozoal cysts, oocysts, trophozoites and heminthic eggs or larva.

Microscopical examination was done by saline and iodine wet mount. The slides were observed under low power (10x) followed by high power (40x) of the microscope.

While performing wet mount, all the samples were subjected to concentration. There are various floatation and sedimentation techniques of concentration.

Formal-ether sedimentation method leading to saline/Iodine wet mount

The technique performed as follow:

1. Further 3-4 ml of 10% formal saline was added in a preserved sample and then shaken well.

2. The suspension was sieved through cotton gauge in a funnel into a 15 ml centrifuge tube.

3. 3-4 ml of ether was added and shaken vigorously for 5 minutes.

4. The tube was immediately centrifuged at 1000 rpm for 10 minutes

5. Four layers of suspension were obtained in the tube after centrifugation.

    a. A small amount of sediment at the bottom of the tube containing the parasite.

    b. A layer of formalin on the top of the sediment.

    c. A plug of fecal debris on the top of formalin layer.

d. A layer of diethyl ether at the top.

e. The plug of debris formed between diethyl ether and formalin was removed by rotating the tip of the applicator along the inner wall of the tube.

6. The supernatant layers of suspension were discarded and the sediment was examined by saline and iodine wet mount.

Saline wet mount

It was used to detect helminthes eggs, larvae and the protozoa cysts. A drop of normal saline was taken on a clean glass slide; a drop of sediment from the above process was mixed with it and observed under microscope after covering with a cover slip.

Iodine wet mount

This was mainly used for detecting protozoa cysts; however helminthes eggs were also stained and could be detected. Iodine stained cysts showed pale refractile nuclei, yellowish cytoplasm and brown glycogen material. A drop of 5 times diluted Lugol's iodine was taken on a slide and a drop of sediment from above process was mixed with it. The preparation was covered with a cover slip and observed microscope.

4.2.5 Recording of the results

After laboratory processing of the samples the result obtained was recorded in thesis log book. Then it was recorded in computer.

4.4.4 Report distribution

The report distribution was done as the result was obtained after laboratory processing of the samples. Each volunteer with positive cases were given antiparasitic drug along with the report. The complete dose of antiparasitic drugs distributed were albendazole, metronidazole, niclosamide and trimethoprim sulfamethoxazole according to the parasites detected.

4.4.5 Statistical analysis

Chi-square test was applied for statistical analysis of results using Win Pepi software program (PEPI-for-Windows): computer programs for epidemiologists, 2004. Association of intestinal infections with different variables was tested. Results were considered significant if P values were less than 0.05.

CHAPTER V

5 Results

Parasitosis among children of *Sukumbasi Basti* was carried out by examine 279 stool sample. Altogether 121 samples were found positive for parasites.

5.1 Parasites detection

Of the 121 positive cases, 97 single parasites and 24 multiple parasites were detected (Table-1).

Table 1: Types of parasites detected

Type of parasites	Total n	%
Single Parasite	**97**	**80.1**
i) Protozoa	63	52.1
Multiple Parasites	**24**	**19.8**
ii) Helminthes	34	28.0
i) Protozoa	16	13.2
ii) Helminthes	0	0
iii) Protozoa and Helminthes	8	6.6
Total	**121**	**100**

5.2 Distribution of parasites

Among 145 parasites, 39 were helminthes and 106 were protozoan (Table-2).

Table 2: Types and frequency of parasites detected

Type of parasites	Total (n=121)	%
Helminthes	**39**	**26.9**
i) *T. trichiura*	12	8.3
ii) *A. lumbricoides*	10	6.9
iii) Hookworm	6	4.1
iv) *H. nana*	7	4.8
v) *E. vermicularis*	4	2.7
Protozoans	**106**	**73.1**
i) *G. lamblia*	48	33.1
ii) *E. histolytica*	30	20.7
iii) *B. hominis*	6	4.1
iv) *E. coli*	5	3.4
v) *C. cayetanensis*	12	8.3
vi) *E. hartmani*	5	3.4
Total Parasites	**145**	**100**

5.3 Demographic pattern of result

Out of 279 stool samples, the numbers of positive cases in males and females were 73 and 48 respectively. Statistically, there is no significant difference between sex and occurrence of the parasitic infection (p=0.07) (Table-3). In age group, the highest positive cases were in ≤ 10 years. The occurrence of parasitic infection with age group was statistically insignificant (p=0.44) (Table-4). On the basis of family size, the highest percentages (50.0%) of positive cases were in > 5 members (Table-5). Among different ethnic groups, the highest percentages (55.0%) of positive cases were in *Tibeto-Burman* (Table-6)

Table 3: Gender wise prevalence of parasitic infection

	Total n	Pos. n	%	p value
Male	151	73	48.3	**0.07**
Female	128	48	37.5	
Total	**279**	**121**	**43.3**	

Table 4: Age wise distribution of parasitic infection

Age (yrs)	Total n	Pos. n	%	p value
≤10	154	70	45.4	**0.44**
>10	125	51	40.8	
Total	**279**	**121**	**43.3**	

Table 5: prevalence of parasites by family size

Family size	Total n	Pos. n	%	p value
≤5	185	74	40.0	**0.11**
>5	94	47	50.0	
Total	**279**	**121**	**43.3**	

Table 6: Prevalence of parasites in different ethnic group

Ethnic group	Total n	Pos. n	%	p value
Dalits	35	17	51.5	**0.01**
Tibeto-Burman	140	77	55.0	
Indo-Aryan	106	27	25.4	
Total	**279**	**121**	**43.3**	

5.4 Socio-economic behavior

The prevalence rate of parasitic infection was higher in the people not having toilet (62.1%) than those having toilet (41.2%). There is significant difference between the toilet using habit and occurrence of the parasitic infection (p=0.03) (Table-7). The prevalence of parasitic infection was found lowest in the people who cut their nails (37.8%) and the highest those not cutting their nails (69.3%). Statistically there is significant difference between nail cutting habit and occurrence of the parasitic infection (p<0.01) (Table-8). The highest prevalence was in those consuming tap water (45%) than those taking well water. Statistically, there was no significant difference between the source of water and occurrence of the parasitic infection (p=0.35) (Table-9). The highest prevalence of parasitic infection was in those using untreated water (49.5%). Statistically, there was significant difference between type of water used and the occurrence of parasitic infection (p=0.004) (Table-10). The highest prevalence rate of parasitic infection was in children from parents with labour as occupation (52.5%). Statistically there was significant difference between the occupation of parent and occurrence of the parasitic infection (p=0.02) (Table-11).

Table 7: Prevalence of parasites in relation to presence of toilet

Toilet	Total n	Pos. n	%	p value
Yes	250	103	41.2	**0.03**
No	29	18	62.1	
Total	**279**	**121**	**43.3**	

Table 8: Prevalence of Parasitic infection according to nail cutting habit

Nail cut	Total n	Pos. n	%	p value
Yes	230	87	37.8	**<0.01**
No	49	34	69.4	
Total	**279**	**121**	**43.3**	

Table 9: Prevalence of parasitic infection according to the source of drinking water

Source of water	Total n	Pos. n	%	p value
Tap	209	94	45	**0.35**
Well	70	27	38.6	
Total	**279**	**121**	**43.4**	

Table 10: Prevalence of parasitic infection according to the type of water used for drinking

Type of water	Total n	Pos. n	%	p value
Treated	95	30	31.5	**0.004**
Untreated	184	91	49.5	
Total	**279**	**121**	**43.36**	

Table 11: Prevalence of parasitic infection according to parents' occupation

Parents occupation	Total n	Pos. n	%	p value
Business	32	9	28.1	**0.02**
Service	105	47	44.8	
Labour	99	52	52.5	
Others	43	13	30.2	
Total	**279**	**121**	**43.3**	

5.5 Parasitic infection in relation to anti-helminthic drug intake in the past six months

The highest prevalence of parasitic infection was in children not taking anti-helminthic drugs (48.2%). Statistically, there was significant difference between use of anti-helminthic drug and occurrence of the parasitic infection (p=0.002) (Table-12).

Table 12: Prevalence of parasitic infection in relation to anti-helminthic drug intake in the past six months

Anti-helminthic drug	Total n	Pos. n	%	p value
Yes	59	15	25.4	**0.002**
No	220	106	48.2	
Total	**279**	**121**	**43.3**	

CHAPTER VI

6. DISCUSSION AND CONCLUSION

6.1 DISCUSSION

Sukumbasi community being a community of people without land ownership has no proper sanitary and waste disposal system. Moreover, they live in very congested settlements mostly of temporary in nature. Therefore, it is likely to have higher rate of intestinal parasitic infection particularly among children. However, present study showed not very high (43.3%) positive rate of intestinal parasitosis. To the best of our knowledge, no such study on intestinal parasitic infection was available from Nepal. Hence, there are no such data for comparison.

The percentage of monoparasitism were higher than multiparasitism in this study. The finding was in harmony with other previous results (Saldiva et al., 1999; Phetsouvannh et al., 2001 and Singh et al., 1993). Obiamiwe and Nmorst, 1991 also observed higher multiparasitism rate. The rate of monoparasitism and multiparasitism were independent of the gender of the elderly people; however, Rai et al., 2000a has reported higher rate of multiparasitism among males in rural regions of Nepal. Monoparasitism were higher in children of *Sukumbasi Basti* due to high intake of deworming medicine.

In stool samples, 11 species of intestinal parasites were detected, 5 helminths and 6 protozoa. Among them *T. trichiura* (8.3%) was the most common parasite. *T. trichiura* was detected the most among the parasites from Nepal (Ishiyama et al., 2001; Uga et al., 2004; Sharma et al., 2004; Rai et al., 2005; Sherchand and Cross, 2007). It would be due to ineffective deworming with single dose of antihelminthic drug particularly in case of heavy infections. The high prevalence of trichiuriasis is because of its special mode of attachment to caecal mucosa, longer life span and refractory reaction to most antihelminhic drugs; resulting chronic infection in the intestine.

The rate of protozoal infection was higher than helminthic infection in the study. This was consistent with the previous report from Meo Laotians (Wiesenthal et al., 1980; Milao et al., 1995). The higher rate of protozoal infection might be due to presence of land contaminated with faecal matter resulted due to open defecation, swimming in Bagmati River, collecting mud from the river and use of contaminated drinking water.
The study by Shakya et al., 2006 has also shown higher protozoal infection rate than the helminthes in elder people. However, other studies in Nepal among general population have found higher prevalence of helminthic infection (Estevez et al., 1983; Nepal and Palfy, 1980; Rai and Gurung, 1986; Rai et al., 1995; Sherchand et al., 1996; Sharma et al., 2004).

Ascaris prevalence rate was marginally reduced than that of *Trichuris*. However, *A. lumbricoides* has been reported as the predominant parasites in the country by Nepal and Palfy, 1980; Sherchand et al., 1996; Rai et al., 2004 and Ishiyama et al., 2003 which was not consistent with this result. This might be due to use of antihelminthic drugs. The geographical variation may be another factor for lower prevalence of *A. lumbricoides* (Adhikari et al., 2006).

The prevalence of hookworm infection was low (4.13%). Higher rate of infection has been reported earlier from Nepal (Rai et al., 1997; Sherchand et al., 1997) which is different from the result. The reason for low prevalence of hookworm might be due to use of shoes and slippers during most of their time that prevent skin penetration by larvae. The increasing use of toilet in the study area might also be the reason behind it. Ingestion of the filariform larvae present in the soil, breast milk from mother to infants and transplacental is *H. nana* was detected 4.8% and this was in agreement with report given previously by other researchers (Sherchand et al., 2001; 2007). It was reported to be 3.3% in Southern Nepal (Sherchand and Cross, 2004) and 4.9% in Kathmandu valley (Sharma et al., 2004). Again, *H. nana* was also reported as commonest tapeworm in the Kathmandu Valley (Sherchand et al., 1996). The transmission rarely occurred from the ingestion of food contaminated with fleas harbouring the cysticercoid larvae.

The prevalence rate of *E. coli* (3.4%) was found to be slightly lower than that of *E. histolytica*. The similar result has been reported by Oberst and Alquiza, 1987 in Phillipines and Saito et al., 1996 in Paraguya. The prevalence rate of *B. hominis* was 6.0%. It might be *due* to the autolysis of *B. hominis* cysts during the time lapsed between sample collection and examination. *B. hominis* has been reported earlier in people with abdominal complaints (Sherchand et al., 1996) and in general population (Gianotti, 1990; Rai et al**.**, 2001). However, Uga et al., 2004 in a similar study did not report *B. hominis*.

Similarly, the prevalence of C*yclospora* (8.3) was higher than *E. coli*. It might be due to utilization of contaminated water containing fecal materials in washing vegetables that long been proposed as a possible route for transmission of *Cyclospora* (Cannor and Shlim et al., 1995). Vegetables provide an optimal environment for survival of parasites prior to host ingestion. Cabbage, lettuce and mustard leaves are contaminated with *Cyclospora* causing food borne transmission (Sherchand and Cross, 2001).

The study revealed low prevalence (43.3%) of intestinal parasitosis among *Sukumbasi* community. The result was in agreement with previous report from Nepal (Sherchand et al., 1996). The prevalence of parasitic infection was found (44.0%) in rural village of Chitwan District (Yong et al., 2002). However, Ishiyama et al., 2003 found the prevalence (27.0%) in a remote village western Nepal. The main reason for low prevalence would due to deworming with antihelminthic drug and facility of toilet in *Sukumbasi Basti* of Shanti nagar.

The parasitic infection rate was slightly higher in males (48.3%) than in females (37.5%). The result was in agreement with the reports from Nepal (Rai and Gurung, 1986 and Ishiyama et al., 2001). It might be due the reason that the males usually work in farms and in dumping sides of Bagmati River while females are limited to household work. However, sex was independent predictor with parasitic infection (P=0.069). This indicated that both males and females could be infected with parasites. On the other hand, the result was in contrast with several reports (Phetsovannh et al., 2001; Rai et al., 1995; 2004; Knightlinger et al., 1995).

The parasitic infection rate was higher in children having age ≤ 10 than >10. The result was in agreement with the report of Rai et al., (2002). However, the result was in contrast with the reports of Rai et al., 1994 and Sherchand et al., 1996. This could be due to childish activity like eating, swimming, playing on Bagmati River and dumping sites, walking with bare foot, which causes larvae to penetrate into skin.

Parasitic prevalence in the *Sukumbasi Basti* living in the large family members consisting of more than five members had higher percentage of infection than those living in small size of 5 or less than 5. The over crowdiness, poor sanitation and lack of proper care might be the reason behind high infection. This finding was similar to Rai et al., 2005. The result however was inconsistent with the study of Karrer and Rahim, 1995.

In this study, incidence rate of parasites were higher in *Tibeto-Burman* (55.0%) followed by Dalit (51.5%) and the least in *Indo-Aryan* (25.4%). The result was dissimilar with Rai et al., (2002) and Ishiyama et al., (2003). This might be due to the lower socioeconomic, poor health, sanitation and illiteracy.

The parasitic prevalence rate was higher among people not having toilet (62.1%) compared with people having toilet (41.20%). The result was consistent with the study conducted by Rai et al., 2002b; Sorensen et al., 1996 and Toma 1999. The lack of toilet affects the environmental sanitation resulting the incidence of soil-transmitted helminthes. Open defecation around houses, fields, roads, and playgrounds increase the chance of parasitic infection. A marginally higher positive rate was seen having pit latrine than having modern toilet (Rai et al., 2002; Ishiyama et al., 2001). It could be due to pit type of toilet or improper flushing system leading to distribution of maximum parasites. The association of parasitic infection with presence/absence of toilet was statistically significant (P=0.032).

In this study, higher parasitic infection rate was found in those children who had not cut their nails (69.4%) and found the least who cut their nails (37.8%). The finger nail is one of the many sources of infection and may be the important one in Nepal. The children are commonly infected due to their unsanitary habit and poor knowledge of health as the children suck their fingers and play any where in the house and kitchen garden. Similar parasitic positive result was found in the study conducted by Yodmani et al., 1983; Ismid and Rukmono, 1983.

This study showed that the children drinking water from different sources had different incidence rate of parasites. Children drinking water from *kuwa* (shallow well) had marginally lower prevalence rate (38.6%) than who used tap water (45.0%). Here, the tap water might be highly contaminated by sewage as the pipe line is laid along the sewer. On the other hand, the water from shallow well is comparatively safe to drink as soil acts as a self-filtration. However, the association between incidence rate of parasites and sources of water was statistically insignificant (P=0.35).

The high prevalence (31.5%) of parasitic infection was observed in children those had taken unboiled water. This was in agreement with the previous reports by Ishiyama et al., 2001 and Oda Y and Sherchand JB, 2002. This might be due to contamination of drinking water.

The higher infection rate (52.5%) was observed in children belonging to labour family and least in the business family (28.1%). The finding was consistent with the report of Rai et al., 2005. Moreover, similar trends of infection have been reported by Ishiyama et al., 2003. The reason might be due to low socio-economic status, illiteracy, lack of awareness toward the health and could not give time to look after their children.

The parasitic prevalence was higher in children (48.2%) who had not taken anthelminthic drug in past 6 months than those who had taken anthelminthic drug (25.4%). The association was statistically significant (0.002). Similar findings were also reported by Bundy et al., 1987; Albonico et al., 1999; Rai et al., 2005). This clearly indicates the importance of deworming. The prevalence of helminthic infection was reduced remarkably and there was dramatic reduction in infection intensity after deworming programme (WHO, 2002).

6.2 CONCLUSION

The intestinal parasites were detected from *Sukumbasi* children by concentration method. The parasites identified were *T. trichiura, A. lumbricoides,* hookworm, *H. nana, E. vermicularis, G. lamblia, E. histolytica, B. hominis, C. cayetanensis* and *E. hartmani*. The parasites could infect both sex and of all age group in children. The parasitic infection was lower in children maintaining personal hygiene and having good socio-economic status. Additionally, the prevalence of parasitosis was greatly decreased after deworming in the past six months. Therefore, good sanitary practices and timely administration of drugs can reduce the prevalence of parasitic infections in the children.

CHAPTER VII

7. SUMMARY AND RECOMMENDATIONS

7.1 Summary

1. Stool samples were collected from 279 children in Shantinagar, Ward No-34, *Sukumbasi Basti*, Dirgayu Tole during the study period.

2. The prevalence rate of parasitic infection in male children (48.3%) was higher than female children (37.5%).

3. Among the study subjects, 80.2% had monoparasitism and 19.8% had multiparasitism.

4. *T. trichiura* (8.3%) was the commonest helminth and *G. lamblia* (33.1%) was the commonest protozoa. Other parasites detected were *A. lumbricoides*, hookworm, *H. nana, E. vermicularis, E. histolytica, E. coli, B. hominis, C. cayetanensis* and *E. hartmani.*

5. The children of age group ≤ 10 were the most affected (45.5%) with the parasites than > 10 years old (40.8%).

6. The prevalence of parasitic infection rate in family size above 5 was higher (50%) than below or equal to 5 (40%).

7. The prevalence rate of parasitic infection was higher in *Tibeto-Burman* (55%) than that of Dalit (51.5%). The least was in *Indo-Aryan* (25.4%).

8. The parasitic infection rate was lower (41.2%) in children having toilet in their house than not having toilet (62.1%).

9. The parasitic infestation rate was lower in children who had nail cutting habit (37.8%) than those not cutting nail (69.4%).

10. The children drinking tape water (45%) were highly infected by parasites than those having *kuwa* water (38.6%).

11. The children who had used anti-parasitic drug in the past 6 months had lower prevalence rate (25.4%) than those not having drug (48.2%).

12. The highest parasitic infection rate of children was from labour family (52.5%) followed by service (44.8%), others (30.2%) and business (28.1%).

13. The children having unboiled water (49.5%) were highly infected by parasites than having boiled water (31.6%).

7.2 Recommendations

1. *T. trichiura* was detected in high frequency among other parasites therefore; proper course of drug should be administered.

2. The children of either sex or age were infected by parasites so; health promotion activities should be initiated irrespective of their sex and age.

3. The children not maintaining personal hygiene were highly infected with parasites therefore; sanitary practices should be done strictly along with awareness programmes.

4. The prevalence of protozoa was higher than helminthes indicating contamination of water so proper management of water supply should be done.

5. The children who had taken anti-parasitic drugs in the past six months were less infected by parasites so periodic administration of drugs is highly recommended to reduce infestation.

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APPENDIX -1

Materials and Chemicals used

1. Chemicals and reagents

Sodium chloride	Qualigens, India
Basic Fushin	Qualigens, India
Ethanol	Bengal, India
Diethyl ether	Qualigens, India
Formaldehyde	Qualigens, India
Iodine crystals	Loba Chemic, India
Sulphuric acid	Qualigens, India
Methanol	Loba Chemic, India
Malachite Green	Qualigens, India
Sucrose Crystals	Qualigens, India
2.5% Potassium Dichromate	Qualigens, India

2. Materials

Test tube	Borosil, India
Conical Flask	Borosil, India
Beaker	Borosil, India
Measuring cylinder	Borosil, India
Glass slide and cover slips	Borosil,India
Droppers	Borosil, India
Pipettes	Borosil, India
Glass rod	Borosil, India
Test tube stand	Borosil, India

3. Equipments

Microscope	Olympus (Japan)
Refrigerator	LG, Korea
Centrifuge	Remi, India

APPENDIX-2

MICROBIOLOGICAL PROFILE

Serial No: Date:

Name: Age:

Gender: Educational status:

Height: Weight:

Patient's Clinical

History:………………………………………………………………………………………

………………………………………………………………………………………

Questionnaire:

1. How many members are there in your family? ………………….
2. Which is the source of water you use to drink? I. Tap II. Kuwa III. River
3. Which type of water do you drink? I. Boiled II. Non Boiled
4. Do you wash your hands before meal? Yes / No
5. Do you have toilet in your house? Yes / No
6. Do you wash your hands after toilet? Yes / No
7. Do you cut your nail regularly? Yes / No
8. Did you suffer from diarrhea recently? Yes / No
9. Have you taken Antihelmenthic drug recently? Yes / No

Report of stool examination:

Macroscopic examination: Colour: Consistency: Blood and Mucus:	***Microscopic Examination(Findings):*** Saline mount/Iodine mount: Concentration Technique
Treatment:	

Authorised Signature………..