Urban Echinococcosis in Health Transition Nepal

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Conversion Table from Nepali year B.S. to English year A.D.

]	Nepali Month Conversion Table			
Nepali Month	Equivalent			
Baishak	middle of April to middle of May			
Jestha	middle of May to middle of June			
Asadh	middle of June to middle of July			
Srawan	middle of July to middle of August			
Bhadra	middle of August to middle of September			
Aswin	middle of September to middle of October			
Kartik	middle of October to middle of November			
Mangsir	middle of November to middle of December			
Poush	middle December of to middle of January			
Magh	middle of January to middle of February			
Falgun	middle of February to middle of March			
Chaitra	middle of March to middle of April			

Nepali Year Conversion Table				
Nepali Year (Begins in Mid April)	Equivalent			
B.S.	A.D.			
2045	1988/89			
2046	1989/90			
2047	1990/91			
2048	1991/92			
2049	1992/93			
2050	1993/94			
2051	1994/95			
2052	1995/96			
2053	1996/97			
2054	1997/98			
2055	1998/99			
2056	1999/2000			

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Preface and Acknowledgment

Echinococcosis/Hydatidosis is a global disease occurring from arctic to tropical zones. The causative agent is *Echinococcus* species which are *E. granulosus*, *E. multilocularis*, *E. oligarthrus* and *E. vogeli*. There are a number of subspecies, types and paratypes which are taxonomically different. In this study the main concentration was on *Echinococcus granulosus* urban cycle as well the traditional pastoral/sylvatic cycle. In this study the following persons and institutions were main the cooperative partners in this project.

- 1. Dr. Don de Savigny, Chief, Health Science Division, IDRC, Canada
- 3. Dr. David Waltner-Toews, Project Co-coordinator and Associate Professor of Population Medicine, University of Guelph, Canada
- 4. Dr. Peter M. Schantz, Chief Division of Parasitology, Centers for Disease Control and Prevention (CDC), Atlanta, GA, USA
- 5. Dr. Philip S. Craig, University of Salford, UK

His Majesty's Government of Nepal (HMG/N) Ministry of Agriculture, Ministry of Health, National Planning Commission and Ministry of Finance and Social Welfare Council have agreed to implement this project with the main support of IDRC and supplementary support by the Danish International Development Agency (DANIDA). The project agreement was signed December 11, 1992. The project was for a period of three years culminating with the National Seminar on January 23-24, 1996. The study reveals that this disease is prevalent in both animal and human communities not only in Kathmandu, but other urban and rural areas of Nepal because of a lack of organized sanitary animal slaughtering facilities, meat inspection and intervention programs. The study shows that there is a need for greater public awareness of the disease and precautions against health risks for both producers and consumers of meat and all those who live in the environment associated with dogs.

This research was supported by the International Development Research Centre (IDRC) in Ottawa and the National Zoonoses and Food Hygiene Research Centre (NZFHRC) in Kathmandu. Special thanks is due to Dr. Don de Savigny from IDRC, who actively facilitated this work. We are also grateful to Dr. P. S. Craig, Dept. of Biological Sciences, University of Salford, Salford, M5 4WT, England and his laboratory staff for providing technical training in ELISA testing for D.R. Bhatta. Special thanks to the laboratory staff at the Centers for Disease Control and Prevention, Atlanta, GA, USA for providing reference laboratory testing. The study of existing slaughtering practices of butchers and meat sellers referred to was funded by the Danish International Development Assistance (DANIDA). Consulting services in lab and epidemiology were provided by Dr. Peter M. Schantz, CDC, Atlanta, GA, USA and Dr. David Waltner-Toews, Univ. of Guelph, Ontario, Canada. Dr. Dominique Baronet was instrumental in organizing the initial stages of the community health survey especially in the dog ecology section. Finally, we wish to thank the following staff members from the NZFHRC: Mr. Alan R. Wald, MPH (Consultant), Mr. C. K. Gurung, Mr. P.R. Bista, Mr. D. B. Thapa, Mr. D. R. Bhatta, Dr. P. B. Chand, Mrs. Mary ann Acharya, Mr. Bobi Thapa, Ms. Meena Dahal and Mr. S. P. Bhandari.

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Special Thanks goes to the Chairmen from Wards 19 and 20 and Dr. B.R. Gautaum, Medical Officer Kathmandu Municipality, Mr. S. Ojha, Nepal Television Hospital Directors, Physicians and Surgeons, Veterinary Hospitals, Veterinary Surgeons from private veterinary clinics.

Urban Echinococcosis in Health Transition, Nepal

1. Epidemiological Background

In Kathmandu, the capital city of Nepal, farm livestock is slaughtered each day by the river, either in small courtyards or on the ground floors of houses. The resulting fresh meat is then distributed between small family businesses which can be found clustered in certain areas of the city. Most of the slaughtered animals are brought from outside the Kathmandu valley and many come from outside the country; the only meat animal raised and left to wander freely in the city or along the river banks is the pig. There is little in the way of washing or waste disposal facilities where the animals are killed and carcass waste is often left in areas accessible to scavenging birds, pigs and dogs.

Echinococcosis was first investigated in Nepal (Joshi, 1973) when echinococcal cysts were found in buffalo, goats, sheep and pigs slaughtered in Kathmandu. A later, preliminary study on echinococcosis in Kathmandu (Joshi, 1984) indicated that there had been 47 cases of echinococcosis amongst the 30,792 operations performed in the city's three hospitals between 1985 and 1990. Of these 47 patients, 26 were male and 21 female and most had cysts in the liver (55%) or lungs (43%). There was no active screening or case finding procedure for *Echinococcus* infection in Nepal at the time; all 47 cases were found at a late stage or during surgery for other purposes. Ten of the cases were fatal.

Five per cent (153/3065) of the water buffalo, 3% (55/1783) of the goat, 8% (12/150) of the sheep and 7% (10/143) of the pig carcasses examined in 77 small abattoirs in Kathmandu between May and September 1991 also carried hydatid cysts (Joshi, 1985a) Thirty (10%) of 291 canine stools collected in the vicinity of the

abattoirs and examined by stool concentration methods were found positive for taeniid eggs, but adult *E. granulosus* were not recorded (Joshi, 1985b).

Cystic echinococcosis is a zoonotic infection caused by the cestode parasite *Echinococcus granulosus*. The parasite's life cycle involves two hosts: dogs and other canids are the definitive hosts to the intestinal tapeworm stage and a variety of domestic livestock and wild ungulate species are the intermediate hosts to the larval cystic stages (see figures 1-5). Cystic echinococcosis occurs in humans when they accidentally ingest the tapeworm's eggs through fecal oral contact with infected dogs. Larval (hydatid) cysts develop in the liver, lungs and other organs causing illness requiring surgery or prolonged chemotherapy. *Echinococcus* occurs worldwide. The disease has been controlled successfully in Iceland, New Zealand, Cyprus and parts of Australia and South America, however, it remains an important pubic health problem in rural areas of many countries where it is endemic.

Anecdotal historical evidence suggests that cystic echinococcosis is an important and widespread zoonosis in Nepal. Interviews with surgeons in the capital city Kathmandu indicate that the disease is a common cause of surgery. The national economy is largely based on the rearing of livestock of many species under conditions which are favorable for the transmission of *Echinococcus granulosus* in the dog livestock species cycle (see figures 1-5).

With IDRC support studies by the National Zoonoses and Food Hygiene Consulting Centre (NZFHRC) has documented the prevalence and transmission of echinococcosis in Nepal. To determine the incidence of diagnosis of human disease the surgical records of three hospitals in Kathmandu were reviewed for cases of

cystic echinococcosis diagnosed and treated during the 6 year period 1985-1990. Among 30,792 surgical procedures performed during those years, 47 patients (26 males and 21 females) were operated on for cystic echinococcosis in the liver (55%), lungs (43%) or other organs (4%). Ten cases (21.3%) were fatal. Although the annual incidence of surgical cases calculated from these figures, 0.3

cases per million population per year is relatively low in comparison with those in some other endemic countries, it is probably a vast underestimate of the national problem since the data were collected from only three hospitals in the capital city and the general population especially in rural areas has limited access to medical care. Furthermore, in the absence of diagnostic procedure, most cases were either severe, late stage cases (as evidenced by the very case fatality rate), or were detected during surgery for other purposes.

To document the occurrence of echinococcosis cyst infection in domestic livestock, the infection was identified and recorded in animals slaughtered at 77 small scale abattoirs in Kathmandu from May to September 1991. Numbers of animals slaughtered and rates of echinococcal cyst infection in different species were as follows: water buffalo 5% (153/3065); goats 3% (55/1783); sheep 8% (12/150); and pigs 7% (10/143). The geographical origin of slaughtered animals was incomplete, however, the limited data indicated that infected animals had originated from many areas of Nepal, Tibet, China and India. The 77 abattoirs varied greatly in construction and sanitary facilities, however, most were generally deficient. Dogs were invariably present during slaughter and only 58% of the abattoirs (45/77) were constructed so as to exclude entry of dogs. Slaughtered carcasses were usually processed on the floor

or open ground and parts of the carcass, especially organ meat considered "unfit for human consumption" were frequently given to dogs. Offal was disposed directly into a local river at 42% (32/77) of abattoirs, thus further insuring their availability to dogs. Thirty (10%) of 291 canine stools collected in the vicinity of the abattoirs and examined by stool concentration methods were positive for taeniid eggs. Although it was not possible to confirm the identification of the taeniid infections in dogs as *Echinococcus* species, the situation suggests probable high rates of infection.

The limited data suggest that the zoonosis is widely distributed in Kathmandu and elsewhere in Nepal. Dogs are frequently kept in homes and dog/human contact is close among most of the national ethnic groups, thus favoring zoonotic transmission of the parasite. The deficiencies in the abattoirs and slaughtering procedures in Kathmandu indicate possibilities for considerable transmission of the disease in the urban setting. The present research was proposed to quantify the prevalence of echinococcosis in human and animal hosts, to determine the socio-cultural and environmental determinants of disease transmission and to design and evaluate control strategies.

Among patients with liver diseases, 41 clinically suspected hydatid cases were admitted to Bir and Kanti hospitals between Jan. 1962 Dec. 1966 are described. Most patients were female, in the 20-40 year old age group, and from the hills area; the reason for this being that women more often tend the animals hosting the cestode *Echinococcus granulosus* (Sharma *et al.*, 1967)

A total of 43 cases of hydatid disease observed during 5 years and of which 42 cases suffered from hydatid cyst of the liver is presented. Age group of the patients ranged from 21 to 41 years of

age. Surgical procedure was adopted for seating the patients. This type of disease is common in the western hilly district of Nepal. The disease can be eradicated by health education, imposition of a dog tax and destroying stray dogs and burning or burying the dead sheep (Panel Report, 1967).

Typically hydatid disease is only diagnosed after five to twenty years but significant exceptions to this have been noted. A six year old girl presented at Kanti Children's Hospital, Kathmandu, because of abdominal mass and haemoptysis. Alimentary system examination revealed an enlarged liver, and a case of hepatic hydatid cyst was diagnosed. Operative findings showed large cysts and 100 ml of clear hydatid fluid (Shrestha *et al.*, 1988). A seven year old boy with huge bilateral hydatid cysts of lung was operated

with bilateral anterolateral thoracotomy in one stage and both cysts were removed in one sitting. There was no intraoperative and postoperative complications. Therefore, bilateral thoracotomy for removal of bilateral hydatid cyst may be considered a safe procedure and it also decreases cost and duration of hospital stay of the patient (Sayami *et al.*, 1994).

There was a case of pulmonary hydatid disease where an over enthusiastic investigative procedure, namely bronchoscopic biopsy was done. Histopathological the diagnosis misled the physicians and the patient was referred elsewhere for treatment which could have been done at the original clinic. Hydatid disease is encountered quite often in daily practice. Often the disease is diagnosed readily by either a plain radiograph or ultrasonogram. (Pathak *et al.*, 1989)

A retrospective study over period of nearly 5 years of 42 cases of hydatid liver disease due to *Echinococcus granulosus* was done. Four different ultrasonic characteristics of hydatid disease were described and as well as criteria for diagnosing the solid of hydatid disease which had not been described before. (Shrestha, 1989).

Four patients with unusual presentations of Hydatid disease of the liver were studied. Diagnosis by imaging modalities are the most reliable and case management with a combination of preoperative Albendazole and surgery is suggested. (Shrestha *et al.*, 1991).

Baseline epidemiological surveys on the prevalence of echinococcosis/hydatidosis in humans of Kathmandu were conducted from July to December 1983. Over five years (1979-1983), a total of 27188 cases were operated for many reasons of which 76 were for hydatidosis in Bir Hospital, Kanti Hospital and Shanta Bhawan Hospital (Patan Hospital) in Kathmandu Valley. Out of 76 cases, 46 were female and 30 male. There were 57 cases who had cysts in the liver and 19 cases had cysts in the lungs.

Out of 76 operated, 59 were cured and 17 died due to anaphylactic shock. The average death rate was 22% and prevalence was 0.28 surgical case per 10,000 general surgery cases. It was observed that *E. granulosus* is controllable in its domestic animal cycle by applying strong rabies control and meat inspection programmes bringing almost immediate benefits for human health (Joshi 1985c).

Following these initial studies, a larger project was designed and an agreement was made with IDRC on December 11, 1992 to investigate the possibility of *Echinococcus* transmission from the livestock slaughtered in the city to the people of Kathmandu.

2. Objectives

2.1 General Objectives

To identify the socio-cultural, occupational and environmental determinants of urban echinococcosis in Nepal particularly in Kathmandu and to design, implement and test an integrated intervention strategy with the view to health policy recommendations for sustainable disease prevention and control.

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2.2 Specific Objectives

a) **Human Disease**. To determine the demographic, socioeconomic, and environmental features of urban human cystic echinococcosis with a view to identifying risk factors and quantifying socio-economic impact of the disease.

b) **Canine Infection**. To determine the incidence of echinococcosis in individually and community-owned dogs; to determine risk factors for infection in dogs; to identify characteristics of human-canid-environment interactions predisposing transmission to people; and to identify an appropriate intervention to interrupt infection of dogs and transmission to people.

c) **Occupational Determinants of Transmission.** To determine the species and origin of slaughter animals contributing to urban cystic echinococcosis in Kathmandu; to identify the religious, legal, cultural and empirical characteristics of slaughter practices and meat distribution which may be most efficiently and effectively modified to interrupt the transmission cycle.

d) **Intervention**. Based on the results of research on risk factors for infection in people and dogs, and on the occupational determinants,

an integrated intervention program for echinococcosis was designed. This program included such things as mass treatment of dogs, modifications in slaughtering facilities, community-based education programs, and changes in consumer, producer and butcher behaviors. This program was evaluated based on the baseline prevalence and incidence data collected to meet objectives a), b) and c).

2.3 Outputs, Users and Beneficiaries

Outputs additional to the above objectives will include increased public awareness of the disease and its health risks for both producers and consumers of meat and all those who live in environments associated with dogs. It is anticipated that improved practices with regard to sanitation and food hygiene and new legislation regarding meat inspection codes was put in place. A diagnostic capability was established and continues after the end of the research program. This research study directly benefits the risk population and dog owners. This was a great help to exchange the ideas among the scientific community of the Canadian scientists and Nepalese scientists. This research also significantly contributed to the strengthening of National Zoonoses and Food Hygiene Consulting Centre.

3. Project Summary

The prevalence, incidence and risk factors of infection by *Echinococcus granulosus* were studied in the domestic and street dogs of Kathmandu, Nepal. Using ELISA coproantigen test as a screening method, the highest prevalence (5/88=5.7%) was seen in domestic dogs from an area in the city used for slaughtering livestock.

A prevalence of 1.8% (3/171) was found in domestic dogs seen at eight veterinary clinics distributed around the city. Although none of the 73 street dogs sampled in the ELISA screening survey was positive, three of 20 street dogs killed with poison as part of the city's dog control programme harboured from one to five adult worms. As none of 99 dogs treated with an anthelmintic was found re-infected 3 months later, it was impossible to calculate accurately the incidence of infection over a 3- month period. Information about the feeding, sleeping and roaming practices of the dogs was also gathered using questionnaires and direct observations.

Three hospitals of Kathmandu (Bir Hospital, Kanti Children's Hospital and Tribhuvan University Teaching Hospital), Patan Hospital and one hospital from Pokhara were contacted and all major surgery cases, done between 1985 and 1995, were reviewed to find the percentage of hydatid cases found (Progress Reports of 1992, 1993 and 1994).

Twenty-three samples from active cases of hydatid disease were received from March to December 1995 from the hospitals and private nursing homes. The most specimens (n=9) came from Bir Hospital, the largest hospital in Nepal. Tribhuvan University Teaching hospital sent five and Scheer Memorial Hospital, four. There were six specimens testing negative for echinococcal antibodies and seventeen positive. Among the positive, nine (53%) were male and eight (47%) were female. The age group 35 and older had the highest number of positive (14%) among both genders.

Sixty-four (18%) samples were positive out of 348. All 20 (12%) positive results in ward 19 came from 19 households. In ward 20, 44 (23%) came from 26 households. The difference between the

number of household showing a positive ELISA screening test result, 19 (35%) in ward 19 and 26 (41%) in ward 20, was found not to be significant. But ward 20 had 11 households with 2 or more positive individuals compare to only one in ward 19. Among the age groups, all gender and wards combined, 9 (12%) were found in below 15, 21 (26%) in 15-24, 15 (23%) in 25-34 and 19 (15%) over 34 years old. However, the confirmatory immunoblot tests by the CDC showed no confirmed result positive in this group.

The community survey study was carried in wards 19 and 20 of Kathmandu where considerable butchering and meat selling occurs. In focusing on the household as the unit of analysis, we have assumed that households have some degree of control over exposure of their members to *Echinococcus eggs*, and that household practices are not overwhelmed by general contamination in the community.

4. Methodology

4.1 Canine Study

4.1.1 Sampling procedure

The dog populations of Kathmandu were divided into three categories of risk for echinococcosis (high, medium and low), based largely on their closeness to the so-called "target area" where most livestock was slaughtered (the nearer the higher the risk) but also on whether they were domestic (i.e. owned) or street (the street dogs being assumed to be at higher risk because of their greater access to the offal and other waste from the slaughtering).

HIGH RISK DOGS

Dogs actually in the target area, a sub-section of Ward 19, one of the 35 wards into which the city is split, were considered to be at highest risk. This small area, of about 200 X 250 m, contained 19 slaughtering places (ten enclosed courtyards/buildings and nine riverside sites), had poor waste disposal, a high concentration of dogs (assessed by eye) and, usefully for the study, well delimited boundaries. Plastic loops (Butler) were used to take faecal samples from the rectums of those dogs in the target area that were estimated to be aged >2 months; all these dogs were then tagged, given a colour coded collar, a rabies vaccination (if estimated to be aged >4 months) and a curative dose of praziguantel (Droncit; Bayer), photographed and released. Sampling forms, including data on the owner (if any), collar colour, tag number, photograph, sex, estimated age, number of pups (where appropriate), general health status and sampling date, where completed for all dogs. Questionnaires (see Annex 1) on feeding practices, sleeping habits, defecation sites and freedom of movement of their dog(s) were completed by the household owning each domestic dog.

MEDIUM RISK DOGS

Domestic dogs in areas neighboring the target area were considered at medium risk. Publicity distributed in the remainder of Ward 19 and in Ward 20 offered free anti-rabies vaccination and praziquantel for any dog brought to the ward office by its owner. A questionnaire and sampling form was completed for each owner and the dogs were sampled, treated, tagged, collared and photographed like the high-risk dogs. Street dogs were also caught in the non-target areas of Ward 19 and Wards 15 and 20 and treated in the same way.

LOW RISK DOGS

Dogs taken to eight veterinary clinics in Kathmandu or Patan (a city separated from Kathmandu by the Bagmati river) for routine procedures were presumed to be at low risk of acquiring the parasite. In returned for supplies of injectable praziquantel, veterinarians collected faecal samples from all the dogs >3 months of age and filled out one questionnaire per dog sampled.

4.1.2 Follow-up

Three months after the first survey, the owners of the dogs that had been treated in the target area were offered photographs of themselves with their dog(s) in exchange for second faecal samples from their dogs. Street dogs from the target area were also re-sampled if their identity could be confirmed. Deaths and collar losses were also recorded.

4.1.3 Poisoned dogs and Identification of Adult Parasites:

As a part of a municipal dog-control programme, the Sanitary Department of the city of Kathmandu periodically kills street dogs by feeding them bait laced with strychnine sulphate. The bait is put out after midnight, and dead dogs are collected the next morning and taken to a landfill site for burial. In September 1993, arrangements were made with the city authorities for researchers to take faecal samples and the small intestines from the poisoned dogs prior to their burial, up to 9 hours post-mortem. The faecal samples were kept at room temperature in 5% formal saline. Each intestine was identified with a tag matching the number on the faecal sample vial and injected with 10% formalin before being immersed in the same solution. After fixation for 55-60 days, the intestines were 13

opened longitudinally and examined in detail for the presence of *E. granulosus* adults as well as nematodes and other cestodes. Any dubious worm-like material was examined by light microscopy. *Echinococcus* adults were fixed in 70% ethanol and stained with aceto-carmine.

4.1.4 Coproantigen ELISA Test

All faecal samples were tested using a coproantigen ELISA based on hyperimmune rabbit serum raised against adult E. granulosus (proglottis) somatic antigen. The protocol used was generally that described by Allen et al. (1992) but the preparation of the faecal supernatant fractions and reading of the plates differs slightly. The supernatant fractions were prepared by mixing one g faecal material with 1.5 ml of 5% formol-saline containing 0.3% Tween 20 (Sigma). At this stage, the samples were often kept at room temperature for several days before being processed further. Once at the laboratory, the samples were shaken vigorously by hand and centrifuged at 2000 g for 20 min. The supernatant was stored in 1.5 ml aliquots at 4°C until the test was performed. To compare two methods of reading, a section of plates was read visually by a single person in Kathmandu, and at 450 nm wavelength in Salford, U.K. Faecal samples from 17 dogs originating from Kathmandu but found free of E. granulosus post-mortem served a negative controls; these were also tested in Salford to establish the cut-off level to use on the samples collected in Kathmandu.

4.1.5 Dog Observation

On each of 8 days in February 1993 and six in April 1993, the target area was walked for 2 h. The time(s) when a dog was observed during the period and its location, activity, collar colour and tag number were recorded. The time of day each observation period was begun was varied to cover all 12 hours of daylight.

4.1.6 Statistics

The data from questionnaires and sampling forms collected in wards 19 and 20 and the veterinary clinics were recorded using Epi-Info software (USD, Stone Mountain, GA) and then transferred to Quattro-Pro (Borland) for tabulation. SAS software (SAS Institute, Cary, NC, USA) was then used to perform Student's *t-tests* and χ^2 tests.

4.2 Household survey

4.2.1 Sampling procedure

The ward chairmen from all 35 wards in Kathmandu were contacted and asked to provide the names of ten households which owned dogs in their ward. These 350 households were visited and the self-designated head of the household was asked to answer questions about dog(s) management in their home. The questionnaire (see Annex 7) was also used to collect human population data.

4.2.2 Statistics

The data from questionnaires collected in 35 wards of Kathmandu were recorded using Epi Info 6 software (Centers for Disease Control and Prevention, Atlanta, GA, USA, 1994).

4.3 Human study

4.3.1 Serum Collection

4.3.1.1 Serum from Blood Bank

Blood samples were collected from Nepal Red Cross over 3 months period. The samples were brought the same day to the NZFHRC, the serum separated and frozen at -20°C until the test was performed.

4.3.1.2 Serum from Hospitals

Over a 3 months period, blood samples were collected from Patan Hospital, Tribhuvan University Teaching Hospital, Public Health Laboratory, Teku and Infectious Disease Hospital, Teku. The samples came from patients admitted for various reasons. These were brought to the NZFHRC on the sampling day and serum separated and frozen at -20°C until the test was performed.

4.3.2 Community Study

4.3.2.1 Epidemiologic Design

The sampling frame comprised all the households in two wards of Kathmandu where considerable butchering and meat selling occurs. In focusing on the household as the unit of analysis, we have assumed that households have some degree of control over exposure of their members to Echinococcus eggs, and that household practices are not overwhelmed by general contamination in the community. A complete list of all the households in each ward was obtained, houses were numbered, and a simple random sample, using a list of computer-generated random numbers, was selected. Based on an expected prevalence of infected households of 10%, and a desire to be within 5% of the true prevalence, 95% of the time, a sample size of 150 (82 for Ward 19 and 68 for Ward 20) was deemed adequate (Martin et al., 1987). Families were to have lived in the house for at least five years in order to be eligible, and a set of rules was devised for the field workers to select alternates should it not be possible to interview the pre-selected family. At this first visit, field workers asked six questions about occupation, family size, and dog ownership (see Annex 3); the selfdesignated head of the family was then given a numbered identification card and all family members five years old or older were asked to attend a temporary health clinic which would be arranged in their ward.

Beside random sampling, a purposive sampling was done on the butchers and meat sellers of both wards to collect as much information as possible on those households. They were located and given numbered identification cards (see Annex 2).

4.3.2.2 Health clinics

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Prior to the dates of the health clinics, ward political leaders also encouraged residents to attend these clinics. Clinics were held three days per week in each of the two wards for two months. At the time of the clinic, all agreeable family members from the randomly selected families over the age of five years were given a physical exam by a physician, blood was taken for serological examination and a sampling form was filled out (see Annex 4). A community survey questionnaire about various household practices was administered to the head of the household (see Annex 5). Members of families who were not part of the random sample were examined, but no blood was taken. Free medicine and health advice were given to all those who were ill.

4.3.2.3 Blood Sample Collection

Human blood samples were collected in special 0.6 ml vials during the clinics and were immediately stored in cold boxes. After few hours, these were brought to the NZFHRC's laboratory to be spun and each serum stored in 0.5 ml vials was kept at -20 °C until the test was performed, usually once per week.

4.3.3 Hospital Hydatid Cases

4.3.3.1.1 Recording of Old Hydatid Cases

Three hospitals of Kathmandu (Bir Hospital, Kanti Children's Hospital and Tribhuvan University Teaching Hospital), Patan

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Hospital and one hospital from Pokhara were contacted and all major surgery cases, done between 1985 and 1991, were reviewed to find the number of cystic cases found. Gender, age, location of cyst, outcome of operation and total number of major surgeries were also recorded (see Annex 6).

4.3.3.1.2 Recording of New Active Hydatid Cases

Active and suspected cases were also reviewed with surgeons of seven hospitals and clinics of the valley (Kanti Hospital, Sheer Hospital, Tribhuvan University Teaching Hospital, Bir Hospital, Rokpa Clinic, Baudha, Kathmandu Nursing Home and Rama Diagnostic Clinic). Besides getting a free *Echinococcus* serology screening test, some patients were also screened by ultrasound and/or X-rays. Age, gender and location of cysts were recorded.

4.3.3.2 Follow-up

Human cases admitted in the four hospitals of the valley were followed. Nine patients were successfully operated on at Bir Hospital and the hydatid fluid from four patients was collected for analysis.

4.3.4 Serum tests

An *Echinococcus* serology screening (ELISA) test was first carried on blood samples collected from blood bank, community study and hospitals. All wells were read at 450 nm and samples with OD greater than 0.5 were considered positive. Though the test has a reported sensitivity of 100%, there is a significant cross reactivity reported with cysticercosis infections. Therefore, the samples were sent to CDC in Atlanta to be reassayed with a more specific immunoblot test. See package insert for the LMD ELISA Test kit (Carlsbad, Calif., USA) for specific method.

4.4 Slaughtered Animal Examination

Regular slaughtered animal examination was done for the period of two years in the eight municipalities of the country with the collaboration of Danish Meat Trade College Denmark. The cyst were observed, involved organs were examined *e.g.*, liver, lungs, kidney and others.

4.5 National Survey of Slaughtering and Meat Production

The final report of the study which was carried out by the Danish Meat Trade College (Denmark) and the National Zoonoses and Food Hygiene Research Centre has completed and has been received by His Majesty's Government of Nepal (HMG/N) for further action. The Meat Inspection Act has not yet been passed by the Parliament of HMG/N, it has already been processed by the concerned ministries including the Ministry of Agriculture, Ministry of Health and has been submitted to the Ministry of Law and Justice for their comments and approval prior to submission to parliament. A copy of the final report was given to Dr. de Savigny of IDRC.

5. Results

5.1 Canine study

5.1.1 Dog population data

Most (465) of the 539 dogs sampled were domestic, being owned by 400 households; samples from nine of these were lost or destroyed (see Table 1). The remaining 74 dogs were street dogs, for which no owner was identified. Most (129) of the 174 dogs

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presented to the veterinary clinics came from Kathmandu; only 45 came from Patan. The sex ratio of the sampled dogs differed significantly between the area ($p \le 0.001$), the proportion of females being relatively low in wards 19 and 20 (30.8%) and the veterinary clinics

(30.5%) compared with the 54.3% in the target area and 48.6% in the street sample in general. The proportion of young dogs (≤ 6 months) was not associated with sex or sampling area. There was no significant difference in the age of male dogs and bitches for any of the areas. There were no statistically significant differences between sampling areas in the proportion of bitches aged > 6 months that had whelped in the previous 6 months.

Table 1	Information	of the dogs	sampled in	Kathmandu in 1993
		or ere erego		

Area Sampled	No. of House holds			Dogs Sampled			
		Ν	Sex (% female)	Age		Whelped within 6 months (% of females)	
				$(\% \le 6 \text{ months})$	(Months)*		
Domestic Dogs,Target area	66	92	54.3	12.2	40.0±37.7	30.2	
Domestic Dogs, rest of ward 19 and ward 20	165	199	30.8	20.2	32.0±31.2	23.4	
Veterinary Clinics	169	174	30.5	21.7	34.9±34.9	10.8	
Street Dogs, wards 15, 19 and 20	NA	74	48.6	NA	NA	25.0	

* Values are means \pm S. D.

NA, Not applicable

The proportion of owners feeding any raw meat, offal, cysts or bones to their dogs was significantly associated with the area sampled ($p \le 0.001$) (see Table 2), being highest in the target area (61.2%), lower in the rest of the Ward 19 and 20 (41.7%) and still lower in the veterinary clinics (15%). Dogs sleeping indoors generally slept on the floor, their owner's beds or sofas or had their own beds. Dogs defecating inside the house did so on the room, floors or roof or in the hallways. In all areas, domestic dogs spent more time indoors than in the courtyards or street or both (see Table 3).

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Table 2. Feeding, Sleeping and Defecating Habits of Domestic DogsStudied in Kathmandu in 1993

Area Sampled	Feeding (% of owners feeding raw meat, offal, bones or cysts)	Dogs (%) sleeping in:		d	Dogs (% efecating	%) g in:	
		street	court yard	hous e	street	court yard	house
Target area	61.2	13.8	6.2	98.5	81.5	1.5	27.7
Rest of Ward 19+20	41.7	0.6	3.7	95.1	35.0	44.8	22.0
Veterinary Clinics	15.0	0.6	23.6	85.1	13.8	79.9	14.9

Table 3.	Roaming	Behaviour	of domest	tic dogs	Kathmano	du in	1993
			01 40 111 00	ere erogo		~~~ ~~~	

Area Sampled	No. of dogs	Time (hours) spent in: *				Time (hours) spent in: *		
		street	courtyard	house				
Target area	63	5.7±7.3	2.7±6.5	1.9±5.2	13.4±9.1			
Rest of Ward 19+20	165	0.5±1.9	0.9 ± 3.2	1.4 ± 3.4	21.1 ± 4.8			
Veterinary Clinics	169	0.6±1.9	8.2 ± 8.4	0.6 ± 3.4	14.9 ± 8.5			

Values are the means of \pm S.D.

5.1.2 Coproantigen ELISA Test

The mean optical density (OD) for the negative controls was the mean reading of the duplicate wells for each control. The cut-off level used (0.072) was this mean plus three S.D.

In order to measure the agreement between the visual and automated readings of the ELISA results, a kappa value (Martin *et al.*, 1987) was calculated for 342 wells read by both methods. Relative to the results of the automated readings, the visual readings yielded nine true positives, 20 false positives, 2 false negatives and 311 true negatives (kappa = 0.387)

The automated readings indicated 12 (1.7%) coproantigen-positive samples out of the 696 tested. Four of the 67 samples for which no questionnaires or sampling forms were available were found positive; three of these were from stools collected on the ground in the target area and one was found from a dog killed by the municipality and found to be infected at necropsy (used as a positive control). Of the 629 samples for which questionnaires and/or sampling forms existed (Table 4), 530 were first samples and 99 were samples taken 3 months post-treatment. Only eight (1.5%) of the first samples were coproantigen positive, five from domestic dogs in the target area (5.7% of samples from these dogs) and three (1.8%) from the veterinary clinics. These last three dogs all came from ward 6, although only 4.8% of all the dogs sampled from veterinary clinics came from this ward. No other samples were positive; all street dogs and domestic dogs in the non-target areas of ward 19 & 20 and the samples taken from dogs 3 months post-treatment were coproantigen-negative. Of the eight coproantigen- positive dogs for which data existed, two were reported never to have been fed any kind of raw food by their owners.

The other six dogs were reported to have been fed raw cysts. Although, in those areas where infections were detected, prevalence of infection was higher in dogs fed raw food than in those not fed raw food (12.2% v. 0% in the target area and 3.8% v. 1.4% in the veterinary clinics) these differences were not statistically different (p=0.10). Of the eight dogs infected, five, all from the target area, were said to have access to the the street (for 2-22 h/day) and were also observed on the streets, whereas the other three, all from veterinary clinics, were said only to have access to the courtyard adjoining the owners' houses (for 4-24 h/day). Of 92 domestic dogs originally observed in the target area, 11 had died three months later (giving a mortality rate of 12.2%/3 months) and three dogs were lost to follow-up. Six of the 44 street dogs originally observed in the same area had died 3 months later (15.8% mortality/3 months) and 12 were lost to follow-up.

Table 4. Coproantigen ELISA results for dogs sample in Kathmandu in1993

Sampling area	Pre-treatme	nt	Three months post-treatment		
	No. tested	No. positive	No. tested	No. positive	
Domestic dogs Target Area Rest of Ward 19+20	88	5	76	0	
	198	0	NA	NA	
Veterinary Clinics	171	3	NA	NA	
Street dogs Wards 15, 19 and 20	73	0	23	0	

NA, Not applicable

5.1.3 Adult Parasites

Most of the 20 dogs examined post mortem were infected with hookworm (85%) and/or *Dipylidium caninum* (85%). Some carried *Toxocara* spp. (35%), *Taenia* spp. (15%) and/or *Echinococcus granulosus* (15%). The identification of adult *Echinococcus granulosus* found in three of the dogs was based on gross morphology,size, shape of the uterus and the posterior position of the genital pore. This is the first recorded observation of adult *Echinococcus granulosus* in Nepal. Only the faecal sample of one of the three *Echinococcus* infected dogs from Kathmandu was coproantigen positive and no more than five worms were recovered from this dog. Only one worm was found in the intestine of each of the other two dogs.

5.1.4 Dog observations

In February 1993, 390 observations of dog behaviour were made over 8 days and in April 1993 174 observations were made over 6 days. The behaviour was divided into 12 categories: defecating, eating, feeding pups, fighting, grooming, barking, playing, copulating, testing, roaming, sleeping and delimited territory. Sleeping and resting occupied 58.4% of the dogs' diurnal activity in February and 52% in April. Roaming occupied 24.4% and 28.9% in the same time periods. Little time was actually spent eating (4.6% in February and 6.9% in April). Fighting and copulating were not observed during the two study periods, although both were observed a few times in March 1993. Dogs had a limited roaming range and were usually observed in the same location each day. Dogs living in the small streets roamed far less (<50 m) than the dogs living on the riverside (≤ 200 m).

5.2 Household Survey (Dog Ecology)

5.2.1 Household Population Data

Responses were tabulated from 350 dog-owning households who were contacted in the 35 wards of Kathmandu. Most (70%) had no fence or walls. Others (30%) had some kind of enclosure. These households used mostly public dumps which were cleaned at least once a week (see Table 5).

Table 5.	Percentage	Distribution	ot	Individuals	ın	Households	by	Age
Group								

Age Group	Number	Percentage
< 5 Years	243	9.2
5 - 10 Years	299	11.3
11 - 17 Years	410	15.6
18 - 50 Years	1381	52.6
> 50 Years	299	11.3
Total	2632	100.0

The total population covered by the *household with dog(s) study* was 2632 individuals. About two thirds of the people were above 18 years of age.

5.2.2 Dog Population Data

There were 396 dogs from 350 households (1.13 dog/household). Most households 319 (91%) had only one dog, 21(6%) had two, 7 (2%) had three, 1 (0.3%) had four and 2 (0.6%) had five dogs. These dogs were generally fed by households (77%) some family leftover (60%), commercial food, butchers' waste or were left to find their food from neighbours or in garbage.

They were either owned by the whole household (59%) or by only one person in the household (41%), and the owner was an adult 99% of the time. These dogs were most commonly bought or traded (54%), received as gift (35%) or the offspring of an owned bitch (11%). They were either pets or guard dogs. At night, 156 (44%) of dogs were kept inside the house, others were kept free in the courtyard (21%) or in dog kennel (35%).

Table 6. Information on the dogs in the households surveyed in 1994.

Gender	Number of dogs	Age in months (mean+S.D.)	Dogs below 6 months (%)	Pregnant females (%)	Lactating females (%)
Female	104 (26.3%)	43.6 + 37.4	13 (12.5%)	11 (10.6%)	9 (8.7%)
Male	292 (73.7%)	43.3 + 31.2	25 (8.6%)	n.a.	n.a.

There were 73.7% of males among households surveyed (see Table 6). There was no significant difference in the age of male dogs and bitches and the proportion of males and females below 6 months was not found significant. There was 10.6% of females lactating and 8.7% of females pregnant.

5.3 Human study

5.3.1 Laboratory testing

Eight hundred and thirty-one sera were tested in Nepal using a commercially produced enzyme-linked immunosorbent assay (ELISA) (LMD Laboratories, Carlsbad, CA) and an automated reader at 450 nm.

Because of difficulties with test results were encountered in Nepal, all sera were re-tested using the same ELISA test at the Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia, USA. A total of 115 (14%) screened ELISA positive with an optical density (O.D.) greater than 0.5 (see Table 7). Confirmatory immunoblot testing of the ELISA positive specimens from the original 363 specimens from the study area of wards 19 and 20 was done by the CDC. The results were *no* confirmation of any positive ELISA results among the project area specimens (wards 19 and 20).

Sample source	Tested in Nepal (total samples)	Positive samples in Nepal (%)	Tested in CDC, Atlanta (total number)	Positive samples in Atlanta (%)
Community study (wards 19 and 20)	363	356 (98%)	348	64 (18%)
Hospitals and Blood bank	468	262 (56%)	457	51 (11%)

Table 7. Comparison of results from screening ELISA test performed in Nepal and CDC, Atlanta

5.3.2 Serum Bank

The prevalence of individuals sero-positive by the ELISA screening test for *E. granulosus* in anonymous blood samples from area hospitals (Patan Hospital, Public Health Lab and Infectious Disease Hospital, Teku, Kathmandu) and the blood bank (Nepal Red Cross) was 27 (11.7%) of 230 for the hospitals and 24 (10.6%) of 227 for the blood bank.

5.3.3 Community study

5.3.3.1 Demographic Information

There were 1000 households listed for Ward 19 and 1100 for Ward 20. Of the 185 households selected, 42 (23%) were households which were in the meat business (butchers, sellers or both). These represented 29 (27%) of 106 households in ward 19 and 13 (17%) of 79 households in ward 20. Sera and/or questionnaire information were obtained from 136 households, 33 (24%) were in the meat business. Of 136 households, 121 had from 1 to 13 blood samples taken. The 363 blood samples collected represented 30% of the number of people recorded from 185 households (see Table 8). Out of 121 households, 19 (16%) were in meat business and 64 (18%) of total blood samples were drawn from those 19 households. Table 8 shows the proportion of people and households initially selected. Ward 20 had and overall better yield of sampled people (36%) and households (81%) compare to ward 19, which had 25% of people and 54% of households sampled.

Table 8. Proportion of People and Households in Surveyed and
Sampled Population Compared to Population Initially
Contacted, by Occupation and by Wards

ward	Occupat ion	Contact People	ed population Households	Surveyed People	population households	Sampled p People	opulation households
19	Meat business	231	29	189 (82%) 21 (72%)	29 (13%)	8 (28%)
	Others	472	77	294 (62%) 50 (65%)	144 (31%)	49 (64%)
20	Meat business	97	13	92 (95%)	12 (92%)	35 (36%)	11 (85%)
	Others	428	66	347 (81%) 53 (80%)	155 (36%)	53 (80%)
Total	All	1228	185	944 (77%) 136 (74%)	363 (30%)	121 (65%)

The age distribution of 944 people from 136 surveyed households is shown in table 9. Table 10 shows the age distribution of 363 sampled people from 121 households. Total of age groups for both wards as well as gender by wards are compared to same proportions in surveyed households. Males of ward 19 were slightly less sampled than all others. They represented 65 (18%) of 363 people compare to 239 (25%) of 944 people in surveyed households. Both age group below 15 years and over 34 were slightly more sampled than their representing proportion in surveyed population. 22% instead of 19% for below 15 and 37% instead of 31% in over 34. The age group of 15-24 and 25-34 had proportions of 23% and 18% respectively compare to a proportion of 25% in surveyed population.

Table 9. Age Distribution (all age over 4 years old) within Surveyed Households (n=944).

Ward	Gender	< 15	15 - 24	25 - 34	> 34	Total
19	F	60	73	54	79	266 (28%)
	М	49	59	59	72	239 (25%)
20	F	38	50	51	76	215 (23%)
	М	36	51	67	70	224 (24%)
Total	M & F	183 (19%)	233 (25%)	231 (25%)	297 (31%)	944 (100%)

Table 10. Age distribution (all age over 4 years old) of sampled individuals (n=363).

Ward	Gender	< 15	15 - 24	25 - 34	> 34	Total (%)
19	F	20	32	17	39	108 (30%)
	М	18	13	12	22	65 (18%)
20	F	14	23	26	37	100 (28%)
	М	27	15	12	36	90 (25%)
Total	M & F	79 (22%)	83 (23%)	67 (18%)	134 (37%)	363 (100%)

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Most households (98%) included at least one literate person (defined as being able to read a local newspaper). In both wards, the literacy among the surveyed households was found to be the highest in the age group 15 to 24 years (see Table 11).

AGE	WARD Literate	19 e Illitera	ate % Literacy	WARE Literat	020 te Illite	rate % Literacy
5 - 14	93	16	85%	68	5	93%
15 - 24	115	4	97%	96	4	96%
25 - 34	95	14	87%	109	9	92%
35 +	80	80	50%	94	55	63%

 Table 11. Literacy within Surveyed Population Households (n=944).

5.3.3.2 Infection in Sampled Population

A total of 363 human serum samples were collected from ward 19 (n=173) and 20 (n=190). ELISA optical density (OD) values of greater than or equal to 0.5 were considered to be positive. All the samples were tested in the centre's lab and 348 samples were sent to the Centers for Disease Control and Prevention (CDC) laboratory in Atlanta, Georgia, USA for confirmation. Of the 15 samples rejected, 13 were from ward 19 and 2 from ward 20.

Table 12 shows the summary of the positive results for ELISA screening test. Sixty-four (18%) samples were positive out of 348. All 20 (12%) positive results in ward 19 came from 19 households. In ward 20, 44 (23%) came from 26 households. The difference between the number of households showing a positive ELISA screening test result, 19 (35%) in ward 19 and 26 (41%) in ward 20, was found not to be significant. But ward 20 had 11 households

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with 2 or more positive individuals compared to only one in ward 19. Among the age groups, all gender and wards combined, 9 (12%) were found in below 15, 21 (26%) in 15-24, 15 (23%) in 25-34 and 19 (15%) over 34 years old. Other more common diseases such as cysticercosis (known to be endemic in Nepal) can cause false positive results on the ELISA screening test for anti-*Echinococcus* antibodies. None of screening positive test results were confirmed by immunoblot testing.

 Table 12. Positive ELISA Screening Test Results by Gender, Age

 Group and Ward (n=348)

Ward	Gender	AGE < 15 ELISA +	15 - 24 ELISA +	25 - 34 ELISA +	> 34 ELISA +	Total ELISA +
19	F	0/18	5/31	3/16	5/35	13/100 (13%)
	М	3/16	3/12	1/12	0/20	7/60 (12%)
20	F	1/14	10/23	7/26	5/36	23/99 (23%)
	М	5/27	3/14	4/12	9/36	21/89 (24%)
Total	M & F	9/75 (12%)	21/80 (26%)	15/66 (23%)	19/127 (15%)	64/348 (18%)

5.3.3.3 Dog Care Practices

Table 13 shows the proportion of households owning at least one dog. Total proportions were 28% for the initially contacted 185 households, 23% for the surveyed households and 29% for the ELISA positive households. Among this last group, ward 20 had a slightly higher number of households owning dogs (35%) compare to ward 19 (21%), but the difference was not found significant.

Table 13. P	roportion o	f Households	Owning at 1	Least One Dog.

Ward	Contacted population (Intro. quest.)	Surveyed households (Comm. survey quest.)	ELISA positive households	
19	30/106 (28%)	16/71 (23%)	4/19 (21%)	
20	21/79 (27%)	15/65 (23%)	9/26 (35%)	
total	51/185 (28%)	31/136 (23%)	13/45 (29%)	

Selected dog-care practices of study households are listed in table 14. Of the 31 households (23%) who owned dogs, 25 (80%) had one dog, 5 had two, and one had four. All dog-owning households fed their dogs table scraps, and 84% fed cooked foods of various sorts; 29% also fed raw meat or organs. Of the 12 that allowed their dog to defecate in the house, 7 disposed of the feces in the garbage, 3 in the garden, and one each in the street or "other" (unspecified). Of the 29 whose dog slept in the house, 17 had a special place for the dog, and the remaining 12 allowed the dogs to sleep on the floor (3), in people's beds (3), on a sofa (2) or elsewhere (4).

Table 14. Household Management of Dogs within Surveyed Households (n=31).

wa rd	Dogs fed raw meat and/or organs	Dogs sleep in the house	Dogs defecate inside the house	Dogs allowed in food preparation area	Dogs allowed in dining area	Dogs allowed into the street
19	5/16 (31%)	16/16 (100%)	4/16 (25%)	5/16 (31%)	5/16 (31%)	12/16 (75%)
20	4/15 (27%)	13/15 (87%)	8/15 (53%)	5/15 (33%)	4/15 (27%)	7/15 (47%)

5.3.3.4 Dietary Practices and Health Knowledge

Selected dietary practices and indications of knowledge of disease transmission are listed in Table 15. All study households ate meat. Most households ate buffalo (94%), sheep or goat (99%), or poultry (92%); only 25% of households reported eating pork, and 73% reported eating "other" meat. Of those who ate raw meat, most ate it because they liked the taste (80%) or for religious reasons (28%). When cysts were seen, they occurred most commonly in buffalo meat (94%); Only one or two households reported seeing cysts in other meats.

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Table 15. Household Activities and Knowledge related to Possible *E. granulosus* Transmission (n=136)

	Eat raw meat	Have heard of disease from dogs	Have heard of disease from raw meat	Have seen cysts in organs	Have heard of hydatid disease
ward 19*	37/50 (74%)	22/50(44%)	4/50 (8%)	5/50 (10%) ¹	1/50 (2%)
ward 20 [*]	31/53 (58%)	35/53(66%)	12/52 (23%)	9/48 (19%) ¹	0/53 (0%)
butchers of both wards	21/33 (64%)	20/33(61%)	7/33 (21%)	8/33 (55%) ¹	3/33 (9%)

* excluding butchers' households

¹ significant at p=.05 level

Just over half of the households were aware that one could get diseases from dogs. The most commonly cited diseases were rabies (88%) and intestinal infections (5%). Less than a quarter of total households (17%) knew that one could acquire diseases from raw meat. Most of these (18 of 23) cited some form of stomach ailment or diarrhea.

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There was a significant difference between households which had seen cysts in organs, being relatively low (10%) in ward 19 and 20 (19%) compare to 55% for households which were in the meat business (either meat selling or butchering). Only a total of 4 households had heard of hydatid disease. Finally, respondents were asked what they thought were the most important public health problems in their ward. These are listed in table 16.

Table 16. Perceived Health Problems in Study Area by Wards (n=136).

List of public health problems	ward 19	ward 20
Lack of good drinking water	22 (31%)	22 (34%)
Lack of toilets	3 (4%)	0
Slaughterhouse waste	0	6 (9%)
Street dogs	3 (4%)	2 (3%)
Food spoilage	2 (3%)	0
Poor draining of sewage	12 (17%)	6 (9%)
Poor garbage pick-up	29 (41%)	28 (43%)
Other	0	1 (2%)

5.3.3.5 Risk factors for Testing ELISA Screening Test Positive

None of the risk factors listed in table 17 were significant at the household level.

Risk	Odds ratio (OR)	Confidence interval (CI)	P value at .05 level
Having at least one dog	1.84	0.69 <or <4.88<="" td=""><td>0.17</td></or>	0.17
Feeding raw meat to their dog	0.62	0.04 <or <6.61<="" td=""><td>0.62</td></or>	0.62
Allowing dog to defecate inside the house	2.53	0.41 <or <17.79<="" td=""><td>0.23</td></or>	0.23
Allowing dog to enter the food preparation area	1.00	0.14 <or <7.28<="" td=""><td>1.0</td></or>	1.0
Allowing dog to enter the eating area	0.69	0.08 <or <5.34<="" td=""><td>0.66</td></or>	0.66
Allowing dog to sleep inside the house	-	-	0.24
Eating raw meat	0.77	0.33 <or <1.83<="" td=""><td>0.52</td></or>	0.52
Being from a family of butchers	1.02	0.31 <or <3.18<="" td=""><td>0.97</td></or>	0.97

Table 17. Risk factors for Households to Have at Least One ELISA Positive Among Sampled Population (n=118)

5.3.4 Hospital Cases

Table 18 shows the review of major surgery cases from four hospitals of Kathmandu valley and one from Pokhara. These were collected for various periods, depending on hospital, but were all recorded between 1985 and 1995.

Table 18. Operated patients in four hospitals of Kathmandu between2042 and 2051 (1985-1995).

HOSPITAL (years)	CAS ES	% MAL E	AGE (mean±S.D)	% in lungs	% in liver	% death	%cystic /operated cases
Bir 2042-2047	19	10 (53%)	34 ± 17	7 (37%)	3 (16%)	3 (16%)	19/9650 (.2%)
Teaching Hospital 2042-2047	20	13 (65%)	40 ± 16	12 (60%)	8 (40%)	3 (15%)	20/17563 (.1%)
Kanti 2046- 2047	2	1 (50%)	8 ± 1	1 (50%)	1 (50%)	0	2/593 (.3%)
Patan 2046-2050	8	3 (38%)	28 ± 15	n.a.	n.a.	n.a.	n.a.
Pokhara 2046-2051	10	3 (30%)	34 ± 16	n.a.	n.a.	1 (11%)	n.a.

n.a. = not available

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Hydatid cases in Bir Hospital:

Total general operated cases including hydatidosis during the year 1985-90 in Bir Hospital are presented in table 19. Total percentage of hydatid cases over the major operated cases is about 0.2 percent which is alarming.

Year	Major operated cases	Hydatidosis	(%)
1985	1934	2	0.10
1986	1439	5	0.34
1987	1360	6	0.44
1988	1445	1	0.07
1989	1647	3	0.18
1990	1825	2	0.11
1991	3455	6	0.2
1992	2916	2	0.1
1993	3899	5	0.1
Total	19920	32	0.2

Table 19. General Operated Cases Including Hydatidosis in Bir Hospital During the Year 1985-1990:

Age and gender distribution, results after operation, organ affected with hydatid cyst and cases from the different districts. The total reported cases were 19 and there were no differences in gender distribution. Out of 19 operated cases 3 were died. Thirteen patients had cyst in the liver and others had in lungs. The cases were reported from Kathmandu, Sarlahi, Gorkha, Kanchanpur, Kaski, Bhojpur, Lamjung, Sindhupalchok, Syangja and Salvan districts of Nepal and from Tibet (see Table 20).

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Table 20. Age and gender distribution and affected organ of hydatid cases in Bir Hospital During the Years 1985-1990:

		Hydatid	operated	cases		
Year	Age	Sex	Result	Organ affected	Reported From	
1985	м ⁶	F Cure	Cure Liver	Liver	Sarlahi	2
1986	18 28 22 46 29	M F M M	Cure Cure Cure Cure Cure	Liver Liver Lung Lung Liver	Gorkha Kanchanpur Neukha, KTM Kaski ndrachowk	
1987	30 33 58 33 450	M F M F M	Cure Death Death Cure Cure	Lung Liver Lung Liver Lung	Tibet Bhojpur Lamjung Bhojpur Sindhupalchok	
1988 1989	15 16 49	M F F	Death Cure Cure	Liver Liver Liver	Syangja Pokhara Salyan	
1990	65 65 33	F F F	Cure Cure Cure	Liver Liver Gu Liver/Lung/K	Jocħhe, KTM ucchatole,KTM idney Dallu,KTM	

Hydatid Cases in Teaching Hospital:

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Total hydatid operated cases in Teaching Hospital during the year 1985 to 1993 are presented in table 21. The percentage of hydatid cases is 1.43 out of total general operated cases.

Year	Total Major operated cases	Hydatidosis	(%)
1985	442	4	0.90
1986	1650	1	0.06
1987	3319	2	0.06
1988	3720	6	0.16
1989	4220	4	0.09
1990	4212	3	0.07
1991	4715	2	0.04
1992	5062	1	0.01
1993	4798	2	0.042
Total	32138	25	1.43

 Table 21. Total General Operated Cases Including Hydatidosis in

 Tribhuvan University Teaching Hospital During the Years 1985-1993:

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Age, gender, morbidity and mortality, organs affected and patients address are presented in table 22. Of the total cases, thirteen were male and seven female. Three patients died in hospital. Twelve patients had cysts in liver and eight in lungs. Of the total, seven cases were from Kathmandu and thirteen were from Nawalparasi, Gorkha, Surkhet, Parsa, Sindhuli, Lalitpur, Rupandehi, Kaski, Bhojpur, Morang, Makawanpur and Palpa districts.

Table 22. Age and Gender Distribution of Hydatidosis cases fromTeaching Hospital of the Year 1985-1990:

Year 1985	Age Sex	Result	Organ	Reported From
	30 F 41 F 50 F 28 M	Cure Cure Cure Cure	Left lung Liver Lung Lung	Kathmandu Kathmandu Kathmandu Kathmandu
1986	50 M	Cure	Lung	Nawalparasi
1987	329 499 166 57 516 F MM F MM F S 7 6 F	Cure Cure Cure Cure Cure Death Death	Liver Liver Lung Liver Liver Lung Lung Lung (R)	Gorkha Surkhet Birganj Sindhuli Kathmandu Patan Butwal Pokhara
1988	56 M 68 M 63 M 23 M	Cure Death Cure Cure	Lung Liver Lung (R) Lung	Bhojpur Kathmandu Lalitpur Birathagar
1989	20 F 31 M 43 M	Cure Cure Cure	Liver Liver Lung	Makawanpur Palpa Kathmandu

Hydatid cases in Kanti Children's Hospital:

General and hydatid operated cases in Kanti Children's Hospital during the years 1985 to 1993 are presented in table 23. Of the total operated cases 1.37% were hydatid cases. There was equal distribution of cases between males and females.

Year	Major operated cases	Hydatidosis	(%)
1985	494	NA	NA
1986	594	1	0.17
1987	1185	3	0.25
1988	259	1	0.39
1989	593	2	0.34
1990	454	1	0.22
1991	425	NA	NA
1992	1194	NA	NA
1993	761	NA	NA
Total	5959	8	1.37

Total 23 General and Hydatid Operated Cases in Kanti Children's Hospital during the year 1985-1993:

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NA=Not available

Hydatid Cases in Western Regional Hospital Pokhara:

During the years 1985 to 1993 ten cases were operated for hydatid cyst in western Regional Hospital Pokhara. Out of which seven were female and male cases. Only one of the patient died in the hospital (see Table 24).

Table 24. Inpatient Surgical	Cases of Echinococcosis/Hydatidosis	in
Western Regional Hospital, Po	okhara During the Years 1985-1993	

Age	Sex	Туре	Recovered/Death
14	F	Hydatid Cyst	Recovered
25	F	Hydatid Cyst	Recovered
37	М	Hydatid Cyst - R Liver	Recovered
18	F	Hydatid Cyst	Recovered
39	F	Hydatid Cyst	Recovered
48	М	Hydatid Cyst	Recovered
29	F	Hydatid Cyst	Recovered
36	F	Hydatid Cyst	Recovered
68	F	Hydatid Cyst	Death
22	Μ	Hydatid Cyst	Unknown

Hydatid Cases in Patan Hospital (UMN), Lalitpur:

Clinical data were obtained from the Patan Hospital of Lalitpur district. Eight cases were reported during the years 1990-1994. Five were females and three males. No death were reported among cases (see Table 25).

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Fiscal Year	Age	Sex	Operation
1993-94	16	F	Excision
1993-94	28	F	Laparotomy + erusion (sic) of hydatid cyst
1992-93	54	F	Excision of Cyst/Cholecystectomy
1992-93	39	F	NA
1991-92	34	F	Laparotomy
1991-92	7	М	Infection & removal of hydatid cyst
1990-91	19	М	NA
1990-91	30	М	Laparotomy

Table 25. Patan	Hospital Inpatient Surgical Cases of Echinococcosis	foi
the Fiscal Years	1990-1994	

NA= not available

Geographical distribution of human hydatid cases in Nepal

In the recent study the echinococcosis/hydatidosis has been reported from Kathmandu, Dhading, Kailali, Bhaktapur, Bhojpur, Tanahu, Kabhre, Dolakha, Bharatpur and Makawanpur districts (see Table 26). A majority of the cases nine (39%) were reported from Kathmandu. Three (12%) were from Kabhre district. Most of the cases were reported from districts near Kathmandu Valley. The reported cases and their distribution indicates the disease may be prevalent in other remote districts with limited access to health services. There are seventy-five districts in Nepal, most have limited health services and physical infrastructure.

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Table 26. Geographical Distributi	on of Human	Hydatid	Cases in	Nepal
by District				

After the completion of training programme in different hospitals including Tribhuvan University (TU) Teaching Hospital, Scheer Memorial Hospital, Banepa, Bir Hospital, Patan Hospital and Kanti Children's Hospital serum samples from suspected human cases of hydatid disease were sent to the National Zoonoses and Food Hygiene Research Centre (NZFHRC) lab.

Name of district	Total number of cases	%
Kathmandu	9	39
Dhading	1	4
Kailali	2	8
Bhaktapur	2	8
Bhojpur	1	4
Tanahu	1	4
Kabhre	3	12
Dolakha	1	4
Bharatpur	2	8
Makawanpur	1	4
Total	23	100

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Twenty-three samples were received from March to September 1995 from the hospitals and private nursing homes. Detailed results are listed in table 27. The most specimens (n=9) came from Bir Hospital, the largest hospital in Nepal. TU Teaching hospital sent five and Scheer Memorial, four. There were six specimens testing negative for echinococcal antibodies and seventeen positive. Among the positive, nine (53%) were male and eight (47%) were female. The age group 35 and older had the highest number of positive (13,76%) among both genders.

 Table 27. Suspected Cases from Seven Hospitals and Private Clinics of

 Kathmandu valley, tested by ELISA for Echinococcal Antibodies

Health Institution	Suspecte d number of cases	Percen tage of males	Age (mean)	Number of ELISA positive	Number of X-ray positive	Number of ultrasound positive
Kanti Hospital	2	50%	9	1	n.a.	n.a.
Scheer Hospital	4	50%	47	4	n.a.	1
Tribhuvan Teaching H.	5	60%	41	2	1	3
Bir Hospital	9	67%	39	8	2	2
Rokpa Clinic	1	0	26	1	n.a.	n.a.
Kathmandu Nursing Home	1	0	46	1	n.a.	n.a.
Rama Diagnostic Clinic	1	0	56	0	n.a.	n.a.

n.a. = not available

5.3.5 Epidemiological Cycle

Echinococcosis is associated with the presence of infected dogs and canids. This study showed presence of infected dogs, positive human cases, high infection rate of hydatid cyst in different slaughtered animals. The findings revealed that disease does exist in Nepal in both pastoral and urban cycles (see figure 1 to 5).

5.4 Slaughtering and Meat Production

Animals Brought for Slaughtering in Kathmandu

Buffalo:

Mostly buffaloes (male and female) both calves and adults are brought to kathmandu from adjoining districts of Kathmandu Valley such as Sindhupalchok, Kabhre, Dolkha, Rasuwa, Nuwakot, Dhading, Makawanpur, Gorkha, Tanahu and Chitwan. Besides there districts buffaloes are also brought from Nawalparasi, Rupandehi, Kapilvastu, Dang, Bake, and Bardia districts by trucks. About 40% of the buffaloes are brought almost daily from Bihar, Uttar Pradesh and Panjab states of India to Kathmandu in trucks for slaughtering.

Sheep and Goats:

Sheep and Goats are brought to Kathmandu mostly from adjoining districts of Kathmandu Valley. During Vijaya Dasain (annual fall festival) sheep and goats are also brought from Tibet.

Pigs:

Pigs are reared mostly in Kathmandu Valley. Except during 1993/94, because of foot and mouth disease and anthrax outbreak in pigs of Kathmandu, butchers had to import from India and Tarai areas of Nepal. Normally pigs are reared and slaughtered in Kathmandu.

Distribution of Hydatid Cysts Infection in Buffalos, Sheep, Goats, Pigs and *Echinococcus granulosus* in Dogs:

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During the epidemiological surveillance period (1993-1995), slaughtered animals examined in Kathmandu have been found infected with hydatid cysts (single and/or multiple). Adult female water buffaloes had the highest infection rate. Adult female sheep and goats had a higher rate than castrated and non-castrated sheep and goats. Adult male and female pigs were found infected with hydatid cysts but were in few in number. In buffaloes, sheep and goats the infection by Echinococcus granulosus eggs occurred primarily outside Kathmandu The infection in dogs of Kathmandu results from the ingested hydatid cysts which were discarded during slaughtering. The infected dogs (definitive host) harbor the adult parasite E. granulosus and their feces containing the parasite eggs are deposited in pasture land, kitchen gardens, sewer water, rivers, ponds, streets, playgrounds, house courtyards and the interior of houses. The eggs are then ingested by the intermediate hosts (animals and humans). In case of the pig, the pig-dog-pig life cycle of *E. granulosus* is completed inside Kathmandu Valley, whereas buffalo-dog-buffalo, sheep-dog-sheep, goat-dog-goat cycles do not generally exist.

A total 451 dogs both street and domestic dogs stools were examined using coproantigen ELISA test. Eleven (2.4%) were positive for *Echinococcus granulosus*. Three out of 20 street dogs killed (poisoned) by Kathmandu municipality were found positive for adult *E. granulosus* parasite in their small intestine during post-mortem.

A total number of 18,805 slaughtered animals were examined during the year 1993 to 1995. Among them 1,200 (18%) buffalo,

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240 (9%) sheep, 350 (4%) goat and 110 (9%) pig were positive for cysts. These cysts were classified according the organ involvement. Lung and liver were found equally affected and occasionally the same animal had cysts in both places (see Table 28).

Animal species	Total examined	Hydatidosis cyst found (%)	Hydatid cyst positive organs					
			Liver	%	Lun gs	%	Both liver & lungs	%
Buffalo	6550	1200 (18)	600	50	400	33	200	17
Sheep	2540	240 (9)	120	50	100	42	20	8
Goat	8460	350 (4)	200	57	140	40	10	3
Pig	1255	110 (9)	60	55	40	36	10	9
Total	18805	1990 (10)	980	52	680	36	240	12

 Table 28. Total Animals Examined for Hydatid Cysts During the

 Period 1993-1995 in Slaughtering Areas of Kathmandu:

Slaughtering Practices in Eight Selected Municipalities in Nepal

The final report of the study which was carried out by the Danish Meat Trade College (Denmark) and the National Zoonoses and Food Hygiene Research Centre has completed and has been received by His Majesty's Government of Nepal (HMG/N) for further action. The Meat Inspection Act has not yet been passed by the Parliament of HMG/N, it has already been processed by the concerned ministries including the Ministry of Agriculture, Ministry of Health and has been submitted to the Ministry of Law and Justice for their comments and approval prior to submission to parliament. A copy of the final report was given to Dr. de Savigny.

5.5 Intervention programmes

5.5.1. Upgrading of Slaughtering Practices

Slaughterhouses were built in the following municipality and wards: Those slaughterhouses were used by the people of Humat Tole, Ward 20, Kathmandu Municipality, Bhaktapur Municipality, Nepalganj Municipality, and Pokhara Municipality.

5.5.2 Training

National Zoonoses and Food Hygiene Research Centre trained butchers and meat sellers on hygienic meat production and marketing in general and echinococcosis/hydatidosis in particular.

Butchers and Meat sellers training was conducted in Ward No. 20 and 19 on April 11 and 28, 1995 respectively.,

April 26, 1995 training was given to the chief physicians, surgeons and

On May 10, 1995, training was given in Bir Hospital about Echinococcosis / Hydatidosis for 17 Physicians, Surgeons and post graduate students of Bir Hospital.

laboratory technicians at Kanti Children's Hospital, Kathmandu

On June 1 training was held in Patan Mission Hospital with 23 Physicians, Surgeon and Technicians.

On June 2, 1996 training was done for 17 Veterinarians at Veterinary Complex Tripureshwor.

June 5, 1996 training was given at the TU Teaching Hospital for 16 Physician, Surgeons and post graduate students.

Special training program was also organized by the centre for physicians, surgeons and veterinarians of Kathmandu Valley about the epidemiological, diagnostic and prevention and control of echinococcosis/hydatidosis in both animals and humans.

5.6 Training Abroad

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In January 1995, Mr. Dwij R. Bhatta, MSc, the microbiologist from National Zoonoses and Food Hygiene Research Centre has spent a month at the laboratory of Prof. P. S. Craig, University of Salford, England. He studied new ELISA techniques for the detection of the coproantigen of *Echinococcus granulosus* in dogs as well as human serum antibodies.

In 1994/1995 Dr. Harish Joshi received training at Mahidol University, Bangkok, Thailand leading to his Master of Public Health (Oral Health) DEGREE specializing in epidemiology. 51

In 1993/1994, Dominique Baronet obtained Master of Science degree in Veterinary Epidemiology at the University of Guelph, Ontario, Canada, with field work done in this project in Kathmandu.

5.7 Video production

Using a small Sony video camera, Hi-8 mm and 8 mm tapes were recorded on dog sampling activities, butchering and dog behaviour. About ten 90 minutes tapes were recorded between January 1993 and June 1994. Filming was also done during the training of butchers and meat sellers.

A video on the canine research was produced in the Fall 1995. Narration and script were done by Dr. D. Baronet and Dr. D. D. Joshi from tapes recorded between January 93 and June 94. Editing was completed by Nepal Television (NTV), both in Nepali and English versions, and broadcasted on television for public viewing. A second video was produced by Danish Group on slaughtering practices in Nepal.

5.8 National Seminar on Echinococcosis/Hydatidosis

A National Seminar on Echinococcosis/Hydatidosis took place in Kathmandu on January 23-24, 1996 was jointly organized by National Zoonoses and Food Hygiene Research Centre (NZFHRC) Tahachal, Kathmandu, Nepal and International Development Research Centre, Ottawa, Canada. Dr. Peter M. Schantz of the CDC, Atlanta, GA, USA, Dr. Bertha Mo, representative of IDRC, Dr. David Waltner-Toews of the University of Guelph, Ontario, Canada attended. Altogether their were seventy participants and twenty-one technical papers were presented. Abstracts were distributed at the time of the seminar and the proceedings including the full papers is being prepared for distribution in summer 1996.

6. Discussion

6.1 Canine

Recent screening methods for Echinococcus have included killing stray dogs (Macpherson et al., 1985; Craig et al., 1992., Ming et al., 1992) and use of the taeniafuge arecoline hydrobromide (Pappaionaou et al., 1984; Chi et al., 1990). Use of arecoline can be hazardous and not very sensitive; results using arecoline were shown to underestimate the real prevalence by about 10-fold (Wachira et al., 1990). An Echinococcus coproantigen ELISA offers the potential for indirect diagnosis of canine echinococcosis. The genus specificity of this test in natural canine Echinococcus infections was reported to be 96.5% (Allan et al., 1992) or 98% (Deplazes et al., 1992). However, the sensitivity of the test appears to be variable. Using anit-E. granulosus excretory/secretory antigens, Deplazes et al. (1992) only recorded an overall sensitivity of 56% (10/17), as burdens of <70 worms could not be detected. However, use of anti-proglottis somatic capture antibodies, as in the present study, gave a sensitivity of 88% (8/9) for burdens of ≥ 15 worms (Allan *et al.*, 1992), i.e. a higher sensitivity than specific serum antibody tests (Craig et al., 1994).

Although the coproantigen ELISA used on the 20 samples collected at postmortem gave no false positive, it did give two false negatives. Very few worms were, however, found in the three infected intestines and because of the time between death and fixation, breakdown of some worm tissue may have occurred. It was not possible, in the current study, to estimate sensitivity relative to worm burden; a positive correlation has been observed by counting the worms purged from tested dogs (Craig *et al.*, 1994).

Allan *et al.* (1992) found a lower cut-off value (0.013) for negative control samples stored with 5% formol-saline than for those stored frozen. Since all the samples collected in Kathmandu had been stored with 5% formol-saline, the cut-off level was calculated using only the 17 negative controls found in Kathmandu and stored in conditions similar to the other Kathmandu samples.

The kappa value, calculated between the visual reading performed in Kathmandu and the automated reading performed in Salford, indicates an agreement of 38.7% beyond what one would expect based merely on chance. The poor-to-moderate level of agreement reflects a high false positive rate in the visual readings.

Difference between sex ratios of dogs brought in for vaccination and those observed in the field have been observed previously, and may reflect a higher value place on male dogs (D.D. Joshi, 1985abc). This would bean important consideration in any public health initiatives which rely on the control of the dog populations. Based on an ELISA cut-off level of 0.072, the prevalence of *Echinococcus* coproantigen was highest in the domestic dogs from the target area (5.7%), followed by the dogs presented to the veterinary clinics (1.8%). The absence of faecal-antigen positives in the street dogs indicates that very little infective material may be available to them. It would be useful to measure specific serum antibodies level in order to determine overall exposure to Echinococcus in the dog population (Gasser et al., 1993). During the present study, fertile cysts were observed in the offal from old buffalo; these were not discarded but were usually sent to the market and sold to customers along with other meat. In the target area, four of the five coproantigen positive dogs were living beside a slaughtering place where old buffalo cows were killed. Since the killing of buffalo cows is prohibited in Nepal, only buffalo bulls are

killed in open sites by the river, where the street dogs have free access. These bulls are usually slaughtered at a younger age than the cows and are therefore less likely to be infected. This may explain the absence of current infections in the street dogs.

The lack of statistically significant association between dogs fed raw food and the coproantigen positives was probably due to the small number of infected dogs. Since no street dog seemed to be infected, there may be less risk of infection in having access to the street than in being fed potentially infective material. As none of the treated dogs became infected within three months, the infection pressure is, however, apparently low. In Turkana, Kenya, prevalence was shown to return to predosing level (>50%) within six months (Wachira *et al.*, 1990). Also, based on observations of the 20 killed dogs, worm burdens seem to be generally low in Kathmandu.

Observing the daytime behaviour of the dogs provided useful information. Street dogs usually adopted a territory which was shared with other street dogs and the domestic dogs which had access to the street. During the day, a dog would often be found in the same location. Some of the local people were observed feeding leftovers to street dogs and these dogs were generally well tolerated and appreciated by the people for guarding temples and private residences at night. Any dog control initiatives which are contemplated by the public health authorities will need to take territoriality and community "ownership" into account.

6.2 Human

There are several test which have been used to diagnose echinococcosis in humans, these included the Casoni intradermal test, the complement fixation test, the indirect haemagglutination test, the agar gel diffusion test, the latex agglutination test, and more recently, the enzyme linked immunosorbent essay (ELISA), the indirect immunofluorescence antibody test and immunoprecipitation tests, such as immunoblotting. While the ELISA, used as a screening method in this project, is using a crude antigen preparation (LMD Laboratories), the immunoblotting is using more specific antigen proteins. The *Echinococcus* ELISA screening test has a reported 100% sensitivity but because of lower specificity, a false positive rate of 10 to 15% is typical (LMD Laboratories). This is mainly because of cross reactivity between echinococcosis and cysticercosis (*Taenia solium*), though filariasis, fascioliasis and schistosomiasis are recognized tissue helminth infections that may produce false positive results (Craig *et al.*, 1986).

In Turkana, the highest prevalence for echinococcosis among human population is found. Depending on the tests used, it will vary between 9.4% with IHA alone (French and Ingera, 1984) and 2.1%, using a combination of IHA, ELISA, and IFA tests (Romig *et al.*, 1985). It has also the highest incidence of hydatid surgical cases: 198/100,000 people per annum (Eckert *et al.*, 1984).

The prevalence found in the blood samples from the community study (18%) as well as the blood bank and hospitals (11%), were obtained using the ELISA screening test. But since a false positive rate of 10 to 15% is typical using the ELISA technique, it is not surprising that the confirmatory immunoblot tests done by the CDC in Atlanta showed *no* confirmed positives among the samples from

the study area (wards 19 and 20). Nevertheless, the number of clinical cases reported from the hospitals of the valley, 23 cases between March and December for the year 1995, as well as those found in the hospital records between 1985 and 1995 indicates there is infection occurring in Nepal. Furthermore, the presence of dogs infected with the parasite and the management of dogs (sleeping in the houses, defecating in the houses,...) raises suspicion that there are infections from the dogs in the city, though none of the risk factors analysed were significant.

In order to draw as many people as possible to Health Clinics (held in both wards every week for two months), one female motivator from each ward was hired. Among other tasks, they were expected to encourage household members to attend the clinics. The ward specific rates of attendance show lower proportions in ward 19 than in ward 20. The female motivator of ward 19 was a student, around 20 years old, while the female motivator of ward 20 was a mother of two children and was clearly more outgoing with higher self confidence. The selection of motivators is an important aspect in community research such as this one. Older and more established people will usually get greater attention from community members and should be preferred when possible.

7. Conclusion

The present survey identified the requirements for slaughter/abattoir facilities in Nepal. Based upon the findings the main conclusions and recommendations are (Joshi *et al.*, 1995ab):

- to pass the National Meat Act as soon as possible.
- to establish slaughterhouses in all municipalities and a biogas plant adjacent to each slaughterhouses.

- training and education program for construction maintenance and waste treatment.
- A training program for supervisors, veterinarians, public health authorities, butchers and meat sellers.
- to establish a national coordination committee.
- A course for selected veterinarians and medical officers should be planned to take place in the Veterinary Dept. of the Danish Meat Trade College in Roskilde, Denmark.

8. Recommendations from National Seminar 1996

The following recommendations were made by the seminar participants:

- A. National Epidemiological Surveillance (human and animal) to be carried out regularly for:
 - A.1 Epidemiological data collection
 - A.2 Sero-surveillance/monitoring
 - A.3 Epidemiological network system

B. Diagnostic Techniques (Human) to be carried out in the country by applying:

- B.1 ELISA Technique
- B.2 Immuno-blot Technique
- B.3 Ultra sound and other imaging Techniques (X-ray, MRI, Computerized Tomography *etc.*).

C. Diagnostic Techniques (animals) to be carried out by applying:

- C.1 Stool examination
- C.2 Coproantigen ELISA Technique
- C.3 Hydatid cyst examination in slaughtered animals
- D. Medical Treatment of Echinococcosis/Hydatidosis (Human) should be provided:
 - D.1 Pre-operative chemotherapy under supervision
 - D.2 Surgery if necessary
 - D.3 Post-operative chemotherapy Chemotherapeutic agents; (Albendazole 10 ml/kg Body wt. x 30 days is better choice)

E. Chemotherapy in Animals should be provided:

Broad spectrum prophylactic dewormer at regular interval, direct in pet dogs and on bait in stray or street dogs Praziquantal/Albendazole/Mebendazole **Chemotherapy in dogs**: Praziquantal is the drug of choice intermediate hosts, no chemotherapy Treat-cysts/destroy)

F. Intervention Programme for Disease Control should be Implemented:

- F.1 Certificate dewormer at the entry point if not deworm
- F.2 Sterilization of dog population mass companion by govt support

- F.3 Registration and licensing policy
- F.4 national Coordination Committee and sub-committee for registration, parasitic and other zoonoses
- F.5 Meat Inspection Act Meat inspection Veterinary unit in each Municipalities
- F.6 model Slaughter house with basic facilities in each Municipalities

G. Training and Education to be Conducted:

- G.1 Orientation training to health personnel
- G.2 Patient and family
- G.3 Public health Education (Zoonotic Importance) in the school curriculum
- G.4 Mass awareness through mass media
- H. National Reference Centre for Zoonoses will be National Zoonoses and Food Hygiene Research Centre which is a National Centre for Zoonotic Diseases in Nepal
- I. Support and Coordination with National and International Organization for Zoonotic Disease Surveillance and Control to be Explored by the Referral Centre for Zoonoses.

Human Study

Blood Bank Survey Community Survey Case Investigation

Dog Study

Coproantigen Testing of Dog Stool Veterinary Clinic Collaboration Sampling of Dogs for *Echinococcus* parasite Dog-Human Interaction Study

Occupational Study

National Slaughterhouse Survey

Training

Consultant Visits

- IDRC, Ottawa, Canada
- IDRC Regional Office, New Delhi
- University of Guelph, Ontario, Canada
- University of Salford, Salford, England
- Centers for Disease Control and Prevention (CDC), Atlanta, GA,

USA

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Laboratory Training Training in Epidemiology

9. Project Administration Plan

The following activities were carried out over a three year and three month period jointly by National Zoonoses and Food Hygiene Research Centre and the University of Guelph with the support of IDRC. The University of Salford and the Centers of Disease Control and Prevention (CDC) were the collaborating institutions in this study.

Intervention Studies

Upgrading Slaughterhouse Facilities Education Programmes Slaughterhouse Survey Household Survey Sampling of Dogs (Followup)

Project Activities

Data Analysis and Report Writing National Seminar Final Report Writing Submission of Report to Appropriate Authorities Project Continuation Planning

10. Project Finance Management

The total project budget was divided into three sections. One operated by IDRC headquarters in Ottawa, another by University of Guelph and the last by National Zoonoses and Food Hygiene Research Centre.

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1. Ward # Tole <A >

2.	Total number in house {totpeople}	##
	Number {under 5}	##
	Number {5-10}	##
	Number $\{11-17\}$	##
	Number $\{18-50\}$	##
	{Number 50} and over}	##

- 3. Type of {dwelling} #
 - 1 traditional single family house
 - 2 farm house
 - 3 modern single family house
 - 4 multi-apartment building
 - 5 apartment above commercial area
 - 6 tent
- 4. Type of dog restraint #
 - 1. no fence or wall
 - 2. fence or wall but does not restrain dogs
 - 3. fence or wall, completely restrains dogs
- 5. Garbage Disposal: #
 - 1 private disposal in public dump
 - 2 private disposal in other places
 - 3 municipal pickup service more often than weekly
 - 4 municipal pickup service less often than weekly
 - 5 solid waste management project pickup daily

- 66
- 6. Own any {livestock} kept on site? <Y>

Dog Ownership:

7.	{total dogs}	##
	{adult male}	##
	adult female	##

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Annexes

Annex 1 Dog Ecology Survey Questionnaire, 1994

Research Study Number ####

Household Information:

{puppies M&F}

8. How many litters did your bitch have in past twelve months? ## {littersborn}

9. How many dogs did you acquire in the past twelve months (other than by birth) {dog acquired} ##

10. How many dogs did you get rid of in the past twelve months? {doggetrid} ##

11. Do dogs other than yours eat at your home? {dogeathome} $\langle Y \rangle$

- If yes: a Fed by your household {fedhouse} <Y>
 - b Eat at your garbage container {fedgarbage} <Y>
 - c {Scavenge} your premises <Y>

12. Are there {unknown dogs} in your neighborhood?<Y>If yes: a {Always present} in community?<Y>b Number of dogs {numunkdog}##

13. In the past twelve months have members of your family been bit by dogs?

a	{By your own dog	<y></y>
b	{Neighbors dog}	<y></y>
c	{Community dog}	<y></y>
d	{Strange dog}	<y></y>

Individual Dog Information:

14. Who is the owner of the dogs? #

- 1 head of household
- 2 male adult
- 3 female adult
- 4 child
- 5 whole household

15 What dog breeds do you have?

a {Native breed} <Y> b {Cross breed} <Y>

16 What type of dogs do you have and what are the average ages?

21		<i>U</i>	\sim
а	{t Male adult}	<y></y>	
	{a Male adult}		##
b	{t female adult}	<y></y>	
	{a female adult}	##	
c	{t female lactating}	<y></y>	
	{a female lactating}	##	
d	{t female pregnant}	<y></y>	
	{a female pregnant}	##	
e	{t male puppy}	<y></y>	
	{a male puppy}	##	
f	{t female puppy}	<y></y>	
	{a female puppy}	##	

17. How have you acquired your currently owned dogs?

а	offspring of your own bitch	{fbitch}	<y></y>
b	bought from neighbor {bfn}		<y></y>
c	bought from outside	{bfo}	<y></y>
d	gift from neighbor	{gfn}	<y></y>
e	gift from outside	{gfo}	<y></y>

18. At what age was the most recent dog acquired? {agedogacq}#.#

19. How do you use your dog?

u	(Ouurung) of promises	11
b	{Hunting}	<y></y>
с	{Pet}	<y></y>
d	{Herding}	<y></y>
e	{meat source}	<y></y>
f	{other use}	<y></y>

20. Location of dog:

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a Confined to premises only day {confon} <Y>

 $\langle V \rangle$

- b Confined to premises only night $\{conon\} < Y>$
- c Confined to premises day and night {confdn} <Y>

21. Leashing of dog:

- a Leashed on premises only day {lod} <Y>
- b Leashed on premises only night {lon} <Y>
- c Leashed on premises day and night {ldn} <Y>

22. Percentage of time dog is:

а	{Indoor}s #	#
b	{Yard leashed} #	#
с	{Yard free} #	#
23. Type of do	og shelter	
a	{dog kennel}	<y></y>
b	{owner's house}	<y></y>
с	{free in yard}	<y></y>
24. Dog is fed	by:	
a	Householder members	{fedbyhm}
b	Neighbors {fedbyn}	
с	Finds its own food {fed	byfind}
25. Type of fc	ood eaten:	
a	commercial {eatdog for	od} <y></y>
b	family garbage {eatfg}	<y></y>
с	butcher waste {eatbw}	<y></y>
d	street garbage {eatsg}	<y></y>
e	small rodents {eatsr}	<y></y>
26 Who inter	acts with the dog (play)?	

26. Who interacts with the dog (play)?

a		17
b	{DP adult}	<y></y>
c	{DP child}	<y></y>
d	{DP strangers}	<y></y>
e	{DP nobody}	<y></y>

27. Dogs Vaccinations:

a	{rabies}	<y></y>
b	{distemper}	<y></y>

c canine {hepatitis} <Y> d {leptospirosis} <Y>

If rabies vaccinated, how long ago {yrrabies} in years ##.#

- 28. How many litters did the {bitch prod}uce during her life? ##
- 29. How long ago was her last {whelping} (years) ##.#

30. Information on last litter:

а	Number puppies {born live}	##
b	Number {still alive} and with the househol	ld ##
с	Number of puppies {died due to d}isease	##
d	Number of puppies {killed by	##
e	Number of puppies {killed by hu}mans	##
f	Number of puppies {given away} or sold	##
g	Number of puppies {abandoned}	##

31. How many {litters in pa}st twelve months? ##.

<Y>

<Y> <Y> 71

Annex 4 Task Description for Clinic

Annex 2 ID Card for Clinic for Selected Household Members

English translation of Nepali language ID card.

NZFHRC/IDRC Clinic ID Card

Date of start: 2051/2/18 (June 1, 1994)

Clinic Hours

In Ward No. 19 - Wednesday, Thursday and Friday every week In Ward No. 20 - Sunday, Monday and Tuesday every week

Time: 4 pm to 6 pm

Research Study Number:

Annex 3 Introductory Questionnaire for Community Survey

Research Study Number:

Name of head of household
 Address: Ward # _____ Block (Nepali Alphabet)

House Number

3. Does anyone in this family work in the meat business? Yes / No

4. If yes, what is their job?

1) Butcher

2) Meat seller

3) Other meat job (what is it?)

5. How many people over 5 years old are there in this family?

6. Does your family own any dogs? Yes/No

Greeting team (2)

material:

- list with study numbers and '*' corresponding to the number of people ≥ 5 yrs in each household (same as number of cards distributed), and one column for checking when questionnaires have been filled.

- health forms pre numbered, stacked in increasing order

- pens

tasks:

- (1) greet people, take cards, remove the same number of '*' as there are people from the same household coming to the clinic (for example: there are 5 people from household study number 7, 3 are coming together, so you remove 3 * from the list. You now know there are two people still to come from that household) and check the questionnaire column if done.

- (1) find the corresponding health form with the same survey number, accompany one person to interviewers (if questionnaire not filled) and others for health examination. Provide study number to interviewers.

Interviewers (2)

material:

- 200 questionnaires, pens and hard pads.

- cysts in jars

tasks:

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- (2) put study number on questionnaire provided by greeting team and proceed with questions. Keep questionnaires and send person for health examination.

Health examiners (2)

material:

- -thermometers, stethoscope, bandages, alcohol, betadine, deworming medicine, antibiotics
- prescription paper (with heading from center), pens
- health forms brought by one of greeting team

tasks:

- (2) check patients, fill health forms; fill prescription forms and give medicine when needed.

Blood collection (2)

material:

- needles and blood collectors (tubes), alcohol, cotton wool, bandages, jar to collect used needles.

- stickers and pens

- box with ice pads and rack to put samples

tasks:

- (2) collect blood from patients, put stickers on tubes corresponding to each patient, store samples in cold box. Return health forms to greeting team.

Research Study Number

(General family questionnaire, administered at the time of the clinic to someone familiar with food preparation practices in the family.)

1. Name of head of household

2. Name of person answering

3. Address: Ward _____ Block

4. Family members

Age Group	Male	Female	Total	Literate
6-14				
15-20				
21-40				
over 41				

75

Note: literate means can read a newspaper

5. Does your family own any dogs? Yes/ no

If yes, do questions 6 - 15; if no, go directly to question 16.

Annex 5 Community Survey - Urban Echinococcosis in Kathmandu

6. How many dogs do you own now?

7. Years Dogs owned?

8. What do you feed your dog?

Kitchen leftovers (cooked)
 Special dog food (cooked)
 Cooked meat
 Raw meat and/or organs
 Cysts from raw meat
 Other (specify)

9. Where does your dog usually defecate?

1). Inside the house

2). Outside the house

3). Don't know

10. If the dog usually defecates in the house, where do you dispose of the dog stool?

1). Garbage container

2). Kitchen garden

- 3). On the street
- 4). Other (where?)

11. Does your dog ever come into the place where you are preparing food? Yes/No $% \left(\frac{1}{2}\right) =0$

12. Does your dog ever come into the place where food is being eaten? Yes/No

13. Does your dog ever go out in the street? Yes/No

14. Does your dog sleep inside the house? Yes/No

15. If Yes, where does the dog sleep? 1) Dog's own place 2) On the floor 3) On people's beds 4) On sofa seat 5) Other (specify) 16. Does anyone in your family eat meat? Yes/No If yes, answer questions 17-23: If no, go to question 24. 17. What kinds of meat are eaten in your family? 1). Buffalo 2). Sheep or goat 3). Pig 4). Poultry 5). Other (name)

18. Is meat ever eaten raw/half cooked in your family? Yes/No

19. If raw meat is eaten, why is this?

1) Like the taste
 2) Believe it is good for you
 3) For religious reasons
 4) Other reasons (specify)

Show the person a hydatid cyst.

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20. Have you ever seen one of these cysts in meat you have handled? Yes/No

If yes to above question, then do questions 21, 22, and 23; if no, go directly to question 24:

21. From which animals have you seen such cysts? (can be more than one answer):

- 1). Buffalo
- 2). Sheep or goat
- 3). Pig
- 4). Other (name)

22. In what kind of meat have you seen such cysts?

1). Liver

25. If yes, where did you hear about it?

- 2). Lungs
- 3). Other (name)

23. If you find such a cyst, do you usually

Eat it
 Feed it to your dog
 Feed it to a community dog
 Throw it into the garbage
 Other (what?)

24. Have you ever heard of hydatid disease in people? Yes/No

26. If yes, where do you think it comes from?

27. Do you think that people can get any sicknesses from dogs? Yes/No

28. If yes, what kind of sickness :

29. Do you think that people can get any sicknesses from handling raw meat? Yes/No

30. If yes, what kind of sickness :

31. Which of the following do you think are the most important public health problems in your area? (*Interviewer should read the whole list before answer is given*)

- 1). Lack of good drinking water
- 2). Lack of toilets
- 3). Slaughterhouse waste
- 4). Street dogs
- 5). Food is spoiled
- 6). Poor draining of sewage
- 7). Poor garbage pick-up
- 8). Other (specify):

Other notes by the interviewer (*e.g.*, comments from the person being interviewed).

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Annex 6 Checklist for Collection of Data From Hospitals

a. Bir Hospitalb. Kanti Children's Hospitalc. Teaching Hospitald. Patan Hospital (United Mission)

1.1. Hospital Cases

a. Operation theatre cases of echinococcosis/hydatidosis during the last five years, all age and sex groups.

b. Cases diagnosed radiologically.

c. Seroepidemiological, immunodiagnostic cases.

d. Clinical cases.

1.2 Case Record Sheets (from patient or guardian)

1. Name of patient

2. Age

3. Sex

4. Address

5. Date of Admission

6. Date of Operation

7. Date of Discharge

8. Cysts found in: a. liver b. lungs c. brain d. others. 9. Operation a. successful b. patient death 10. Type of cyst found a. single b. multiple c. ruptured 11. Total volume of cyst a. < 1/2 liter b. 1/2 to 1 litre c. > 1 liter 12. Food habit of the patient a. vegetarian b. non-vegetarian If vegetarian, he/she eats raw vegetable salad? Y/N 13. Does the patient keep dogs at home? Y/N If yes, a. Do they feed the dog raw meat? Y/N b. Do they tie up the dog? Y/N If no. a. Do they let the dog loose in the street or in meat slaughtering or marketing areas? Y/N 14. Type of Occupation a. agriculture b. service c. business d. labour e. others

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1.3 Case review form for the Surgeon

1. Have you ever operated on a case of echinococcosis/hydatidosis? Y/N

If yes, how many cases					
	Age Group				
Male	< 15 years	> 15 years _			
Female	< 15 years	> 15 years _			

2. Which organ was affected?

Number of cases ____

a. liver	
b. lung	
c. lung & liver	
d. others	

3. Types of cyst

a. Single b. multiple

4. Prognosis

a. successfulb. patient died

- 5. What are the clinical signs your observed before the operation:
 a. enlargement of stomach
 b. differentee in meaningtion
 - b. difficulty in respiration
 - c. Very lean and thin/emaciated
 - d. fever/nausea/vomiting

a. immunodiagnosisb. radiographyc. clinical symptomsd. autopsye. CT scanningf. Others

1.4 Morbidity and Mortality Record Morbidity and Mortality Record Record of Operated Human Cases of Hydatidosis

Name of Hospital

Date of Operation	Name of Patient	Address	Occupation	Age	Sex	Cured ?	Operated on or Died

Date

Signature of Surveyor

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6. Diagnostic procedures:

Annex 7. Household Survey Questionnaire (Dog Ecology) 1995

Research Study Number ####

Household Information:

1. Ward ## Tole <A > 2. Total number in house {totpeople} ## Number {under 5} ## Number {5-10} ## Number {11-17} ## Number {18-50} ## {Number 50} and over} ##

3. Type of {dwelling} #

- 1. traditional single family house
- 2. farm house
- 3. modern single family house
- 4. multi-apartment building
- apartment above commercial area
 tent

4. Type of dog restraint #

1. no fence or wall

- 2. fence or wall but does not restrain dogs
- 3. fence or wall, completely restrains dogs

5. Garbage Disposal:

private disposal in public dump
 private disposal in other places
 municipal pickup service more often than weekly
 municipal pickup service less often than weekly
 solid waste management project pickup daily

#

6. Own any {livestock} kept on site? <Y>

DOG Ownership 7. {Total dogs} ## {adult male} ## {adult female ## {puppies M&F} ##

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8. How many litters did your bitch have in past twelve months? {littersborn} ##

9. How many dogs did you acquire in the past twelve months (other than by birth) {dog acquired}##

10. How many dogs did you get rid of in the past twelve months? {doggetrid} ##

11. Do dogs other than yours eat at your home? {dogeathome} <Y> If yes: a Fed by your household {fedhouse} <Y> b Eat at your garbage container {fedgarbage} <Y> c {Scavenge} your premises <Y>

12. Are there {unknown dogs} in your neighborhood? <Y> If yes: a {Always present} in community? <Y> b Number of dogs {numunkdog} ##

13. In the past twelve months have members of your family been bit by dogs?

a {By your own dog <Y> b {Neighbors dog} <Y> c {Community dog} <Y> d {Strange dog} <Y> Individual Dog Information:

14. Who is the owns the dogs?

- head of household 1
- 2 male adult
- female adult 3
- 4 child
- whole household 5

15 What dog breeds do you have?

- {Native breed} <Y> a
- {Cross breed} <Y> b

16 What type of dogs do you have and what are the average ages?

#

a {t Male adult}	<y></y>
{a Male adult}	##
b {t female adult}	<y></y>
{a female adult}	##
c {t female lactating}	<y></y>
{a female lactating}	##
d {t female pregnant}	<y></y>
{a female pregnant}	##
e {t male puppy}	<y></y>
{a male puppy}	##
f {t female puppy}	<y></y>
{a female puppy}	##
17. How have you acqu	ired your currently owned dogs?

a offspring of your own	bitch {fbitch	} <y></y>
b bought from neighbor	{bfn}	<y></y>
c bought from outside	{bfo}	<y></y>
d gift from neighbor	$\{gfn\}$	<y></y>
e gift from outside	{gfo}	<y></y>

18. At what age was the most recent dog acquired? {agedogacq} #.#

19. How do you use your dog?		
a {Guarding} of premises		<y></y>
b {Hunting}		<y></y>
c {Pet}		<y></y>
d {Herding}	<y></y>	
e {meat source}	<y></y>	
f {other use}		<y></y>

20. Location of dog:

a Confined to premises only day {confon}	<y></y>
b Confined to premises only night {conon}	<y></y>
c Confined to premises day and night {confdn}	<y></y>

21. Leashing of dog: a Leashed on premises only day {lod} <Y> b Leashed on premises only night {lon} <Y> c Leashed on premises day and night {ldn} <Y>

22. Percentage of time dog is: a {Indoor} -~ : ##

a {	{Indoor}		##
b ·	{Yard leashed}	##	

c {Yard free} ##

23. Type of dog shelter

a	{dog kennel}	<y></y>	
b	{owner's house}		<y></y>

- $\langle Y \rangle$
- c {free in yard}

24. Dog is fed by: a Householder members {fedbyhm} <Y> b Neighbors {fedbyn} <Y> c Finds its own food {fedbyfind} <Y>

- 25. Type of food eaten:
- a commercial {eatdog food} <Y>
- b family garbage {eatfg} <Y>
- c butcher waste $\{eatbw\}$ $\langle Y \rangle$
- d street garbage {eatsg} <Y>
- e small rodents {eatsr} $\langle Y \rangle$

26. Who interacts with the dog (play)?

- a {DP owner} $\langle Y \rangle$
- b {DP adult} $\langle Y \rangle$
- c {DP child} $\langle Y \rangle$
- d {DP strangers} <Y>
- e {DP nobody} $\langle Y \rangle$

27. Dogs Vaccinations:

- a {rabies} <Y>
- b {distemper} <Y>
- c canine {hepatitis} <Y> d {leptospirosis} <Y>

If rabies vaccinated, how long ago {yrrabies} in years ##.#

- 28. How many litters did the {bitch prod}uce during her life? ##
- 29. How long ago was her last {whelping} (years) ##.#

30 Information on last litter a Number puppies {born live} ## Number {still alive} and with the household ## b Number of puppies {died due to d}isease ## с ## d Number of puppies {killed by bi}tch e Number of puppies {killed by hu}mans ## f Number of puppies {given away} or sold ## ## Number of puppies {abandoned} g 31 How many {litters in pa}st twelve months? ##

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Figure 1 Comparative Morphology of Adult *Echinococcus* species

A: Echinococcus vogeli B: Echinococcus granulosus

C: Echinococcus oligarthrus

D: Echinococcus multilocularis

Arrows indicate position of genital pores

Figure 2 Life Cycle of Echinococcus granulosus



Figure 3. Life Cycle of Echinococcus granulosus

Figure 4. Life Cycle of *Echinococcus granulosus* Dog/Sheep

(1) Adult worm, (2) Protoglottid with eggs. (3) Eggs, (4) Ungulates,
 (4a) Accidental infection of man, (5) Infected liver with metacestodes,
 (5a) Fertile metacestodes with protoscolices

Source: Eckert J, Gemmell M A, Matyas Z (eds.) (1984). Guidelines for Surveillance, Prevention and Control of Echinococcosis/Hydatidosis. WHO. Geneva VPH/81.28.

Source: Modified after Pierkarkski, 1973. Courtesy Department of Parasitology, University of Zürich

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Map of Project Area

Figure 5. Life Cycle of *Echinococcus granulosus* Dog/Water Buffalo

(1) Adult worm, (2) Protoglottid with eggs. (3) Eggs, (4) Ungulates, (4a) Accidental infection of man, (5) Infected liver with metacestodes, (5a) Fertile metacestodes with protoscolices

Source: Modified after Pierkarkski, 1973. Courtesy Department of Parasitology, University of Zürich

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